RESULTS

RESULTS

I. SEMEN EXAMINATION:

A. MACROSCOPIC EXAMINATION:

The volumes of the whole ejaculates ranged from 2.9 - 5.8 ml with a mean value of 4.30 ± 1.04 ml. The mean value of the volume of the first fractions of split ejaculates was 1.99 ± 0.45 ml with a range of 1.4 - 2.7 ml. The mean value of the volume of the second fractions of split ejaculates was 2.21 ± 0.45 ml with a range of 1.6 - 3.1 ml (Table 1).

All ejaculates were greyish-white in color and had normal odor and viscosity.

B. MIROSCOPIC EXAMINATION:

1. COUNT:

The concentration of sperms in whole ejaculates ranged from 61-109 millions/ml with a mean value of 81.92 ± 13.4 millions/ml. The mean value of sperm concentration in the first split fractions was 144.12 ± 25.05 millions/ml with a range of 98-215 millions/ml. It was significantly more than that of the whole ejaculates. The mean value in the second split fractions was 29.34 ± 11.98 millions/ml with a range of 10-56 millions/ml. It was less significantly than that of the whole ejaculates (Tables 1 & 2).

2. MOTILTY:

The percentage of motile sperms in whole ejaculates ranged from 60-95% with a mean value of 73.8 ± 11.45%. The mean value in the first split fractions was 83.8 ± 8.6% with a range of 70-95%. It was significantly higher than that of the whole ejaculates. The mean value in the second split fractions was 47.7 ± 9.7% with a range of 35-65%. It was significantly less than that of the whole ejaculates. (Tables 1 & 2).

Table (1): Showing the volume of semen ejaculates, count, percent of motile sperms and percent of normal forms of sperms in the whole ejaculates and the first and second split fractions.

WHOLE EJACULATES					SPLIT EJACULATES								
No. of ejaculate					No. of ejaculate	1st fraction				2nd fraction			
	Vol	Ct	Mt	Mr		Vol	Ct	Mt	Mr	Vol	Ct	Mt	Mr
1	4.1	89	75	75	51	2.6	143	75	80	2.9	25	60	85
2	3.0	95	90	90	52	1.7	126	90	66	2.2	23	50	75
3	5.4	63	65	70	53	1.4	145	95	60	1.6	21	40	65
4	3.4	109	85	75	54	2.5	155	85	85	1.5	15	40	60
5	5.6	80	60	85	55	2.7	105	70	90	2.7	51	60	65
6	5.8	70	60	90	56	2.3	163	80	65	2.1	35	45	90
7	3.2	102	90	80	57	2.6	112	75	60	3.0	53	65	75
8	5.2	83	60	65	58	2.5	123	70	85	3.1	31	65	70
9	4.3	96	70	80	59	1.9	172	90	80	2.5	43	55	85
10	3.1	85	75	75	60	2.7	134	75	85	2.1	28	45	65
11	5.0	71	80	70	61	1.5	132	95	80	2.4	42	55	70
12	2.9	95	95	65	62	2.1	148	85	70	2.8	30	60	60
13	3.1	98	80	80	63	1.4	125	95	75	2.8	20	55	70
14	3.4	78	75	80	64	2.1	135	80	70	2.4	29	55	90
15	3.0	105	95	75	65	1.7	163	90	65	2.0	30	45	70
16	4.2	81	75	90	66	2.5	106	85	75	2.4	22	55	75
17	5.1	70	60	75	67	1.7	122	90	85	2.8	24	60	75
18	5.8	75	60	85	68	1.5	152	95	70	2.9	42	65	80
19	4.8	89	70	65	69	2.1	164	85	80	2.3	26	50	65
20	5.4	65	65	80	70	1.6	131	90	70	2.5	56	55	65
21	3.0	75	80	85	71	2.0	151	85	85	1.6	18	35	60
22	3.2	96	75	70	72	1.5	133	95	65	1.9	22	45	70
23	5.6	63	60	90	73	2.3	122	85	60	2.4	36	50	85
24	3.9	92	75	65	74	2.6	131	80	90	2.7	52	55	75
25	4.4	85	70	80	75	1.8	160	90	70	1.7	42	30	60
26	5.7	69	60	85	76	2.4	145	75	90	1.8	12	45	75
27	5.3	72	65	75	77	1.4	215	95	75	2.3	25	50	65
28	2.9	81	95	85	78	2.4	142	85	90	1.8	32	45	65
29	3.5	75	80	60	79	1.5	178	95	60	2.0	48	40	80
30	3.2	78	90	75	80	1.6	136	95	65	1.9	38	40	75
31	3.1	107	70	60	81	2.6	98	75	65	2.5	12	50	80
32	4.0	89	70	85	82	2.7	116	70	60	2.1	43	45	85
33	5.7	69	60	60	83	2.3	144	80	70	1.8	28	35	85
34	5.5	77	65	75	84	2.0	129	85	85	1.7	13	35	80
35	3.2	98	85	60	85	1.4	159	95	75	1.8	19	35	65
36	3.0	76	95	75	86	1.6	138	85	60	1.9	12	40	70
37	5.6	64	60	75	87	2.1	152	85	80	2.0	27	45	70
38	5.1	85	65	90	88	2.4	120	80	85	3.0	31	60	65
39	3.4	92	90	65	89	1.7	123	85	80	2.6	41	55	60
40	5.0	62	65	70	90	1.5	169	95	85	1.8	31	40	60
41	5.6	68	65	75	91	1.7	175	90	90	1.6	24	35	70
42	4.0	61	70	90	92	1.5	162	90	75	1.7	10	35	65
43	5.7	65	60	85	93	1.6	163	90	70	2.1	40	45	85
44	3.5	103	85	75	94	1.8	192	85	85	2.0	18	40	90
45	4.7	91	75	60	95	2.5	144	75	65	2.7	27	55	75
46	4.3	87	90	65	96	1.7	105	90	75	2.4	22	45	90
47	4.2	96	70	70	97	1.6	201	95	80	1.6	21	40	80
48	5.8	63	60	90	98	2.4	140	75	85	2.5	42	55	70
49	4.0	76	75	65	99	2.6	129	75	60	2.2	23	45	75
50	3.3	82	80	85	100	1.4	178	95	65	1.6	12	35	60
Mean	4.30	81.92	73.8	75.9	Mean	1.99	144.12	83.8	74.8	2.21	29.34	47.7	72.9
S.D.	1.04	13.4	11.45	9.51	S.D.	0.45	25.05	8.6	9.95	0.45	11.98	9.7	9.26

Vol: Volume of each semen ejaculate in ml.
Ct: Count of sperms in millions/ml
Mr: Percent of normal forms of sperms.
Mt: Percent of motile sperms.

Table (2-a): Showing the comparison between sperm count, percent of motile sperms and percent of normal forms of sperms in *the whole non-washed* and *first split fraction specimens.*

	Whole non-Washed Specimens.	First Split Fraction Specimens.	t	p	Signifi-cance.
Sperm count	81.92 ± 13.40	144.12 ± 25.05	15.481	<0.001	+++
Motile sperms%	73.80 ± 11.45	83.80 ± 8.60	4.936	<0.001	+++
Normal forms%.	75.90 ± 9.51	74.80 ± 9.95	0.565	<0.05	-

Table (2-b): Showing the comparison between sperm count, percent of motile sperms and percent of normal forms of sperms in *the whole non-washed* and *second split fraction specimens.*

	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Signifi-cance.
Sperm count	81.92 ± 13.40	29.34 ± 11.98	20.686	<0.001	+++
Motile sperms%	73.80 ± 11.45	47.70 ± 9.27	12.525	<0.001	+++
Normal forms%.	75.90 ± 9.51	72.90 ± 9.26	1.598	<0.05	-

Table (2-c): Showing the comparison between sperm count, percent of motile sperms and percent of normal forms of sperms in *the first split fraction and second split fraction specimens.*

	First Split Fraction Specimens.	Second Split Fraction Specimens.	t	p	Signifi-cance.
Sperm count	144.12 ± 25.05	29.34 ± 11.98	29.225	<0.001	+++
Motile spems%	83.80 ± 8.60	47.70 ± 9.27	20.182	<0.001	+++
Normal forms%	74.80 ± 9.95	72.90 ± 9.26	0.988	<0.05	-

+++ =highly significant - = insignificant.

3. HISTOLOGICAL EXAMINATION:

The percentage of normal forms of sperms in whole ejaculates was normal ranged from 60% to 90% with a mean value of 75.9 ± 9.51%. The mean value of the percentage of the normal forms of sperms in the first split fractions was 74.8 ± 9.95% with a range of 60-90%, while the mean value in the second split fractions was 72.9 ± 9.26% with a range of 60-90%. Both values did not differ significantly from that of the whole ejaculates (Tables 1 & 2).

Abnormal aggregation of sperms {more than 10 cells} were not observed.

The presence of immature testicular cells was withen the normal limits {2%}.

The presence of the other cells rather than sperms like leukocytes, erythtrocytes and macrophages was withen the normal limits {0-2 / H.P.F}.

4. HISTOCHEMICAL EXAMINATION:

I. ADENOSINE TRIPHOSPHATASE:

(A) WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 3)

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was noticed in the mid-piece and tail. No reaction was seen in other parts of spermatozoa (Fig. 1).

The percentage of spermatozoa giving a strong reaction for adenosine triphosphatase ranged from 19% to 39 % with a mean value of 30.3 ± 5.95 %. Those with a moderate reaction ranged from 16% to 39 % with a mean value of 26.2 ± 7.47 %. Weakly stained sperms ranged from 12% to 35 % with a mean value of 24.4 ± 7.31 %. Sperms with a negative reaction ranged from 12% to 27 % with a mean value of 19.1 ± 5.34 %.

Table (3): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in whole non-washed specimens.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	29	36	20	15	11	24	21	27	28	21	18	30	23	29	31	17	14	28	41	41	2	6	16	76
2	35	26	19	20	12	35	26	22	17	22	24	25	27	24	32	15	21	20	44	42	0	2	8	90
3	26	24	29	21	13	31	27	20	22	23	14	16	33	37	33	12	13	17	58	43	1	7	14	78
4	39	30	19	12	14	28	21	31	20	24	31	23	17	29	34	11	9	27	53	44	4	17	19	60
5	26	16	35	23	15	35	33	17	15	25	25	15	36	24	35	22	29	15	34	45	3	13	17	67
6	30	18	25	27	16	29	22	20	29	26	12	17	32	39	36	20	28	20	32	46	2	15	18	65
7	37	39	12	12	17	15	14	35	36	27	17	24	29	30	37	8	10	29	53	47	3	6	11	80
8	33	24	24	19	18	18	25	35	22	28	19	16	28	37	38	19	18	23	40	48	1	5	13	81
9	29	29	26	16	19	25	21	24	30	29	26	22	21	31	39	18	17	24	41	49	3	13	19	65
10	19	20	35	26	20	29	20	21	30	30	29	24	23	24	40	10	18	17	55	50	3	16	15	66
Mean	30.3	26.2	24.4	19.1	Mean	26.9	23.0	25.2	24.9	Mean	21.5	21.2	26.9	30.4	Mean	15.2	17.7	22.0	45.1	Mean	2.2	10.0	15.0	72.8
S.D.	5.95	7.47	7.31	5.34	S.D.	6.59	5.08	6.49	6.69	S.D.	6.42	4.96	5.92	5.66	S.D.	4.73	6.80	4.97	9.10	S.D.	1.23	5.35	3.59	9.53

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

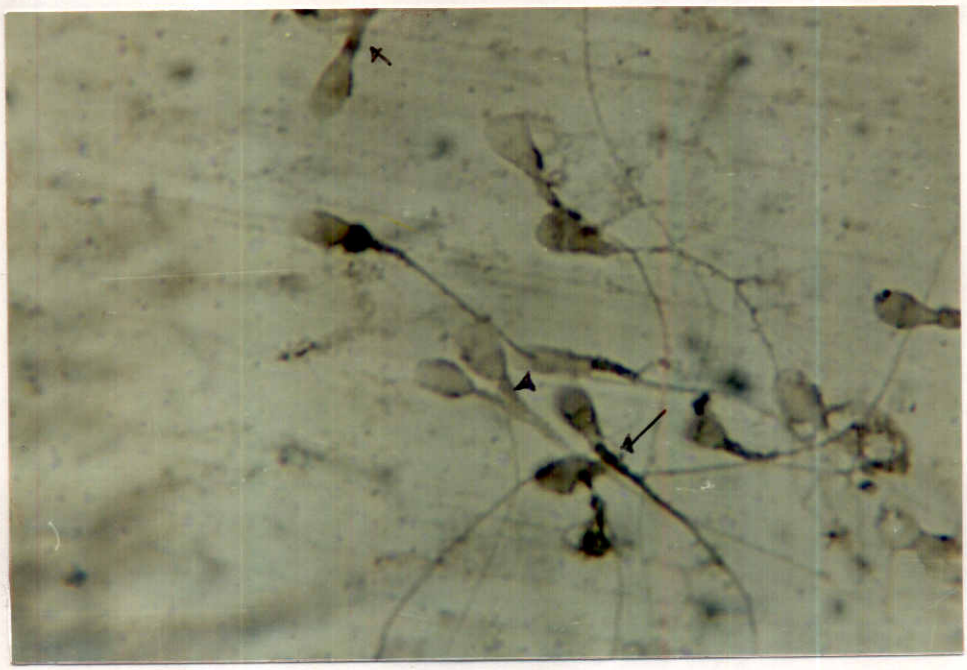

Fig. (1) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **adenosine triphosphatase.**

Note the strong (long arrow), moderate (short arrow), and weak (arrow head) reactions in the sperms.

(Modified method of Tunell and Hart for ATPase. Proj: 10 Obj: 100)

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction for adenosine triphosphatase ranged from 15% to 35 % with a mean value of 26.9 ± 6.59 %. Moderately stained sperms ranged from 14% to 33 % with a mean value of 23 ± 5.08 %. Those with a weak reaction ranged from 17% to 35 % with a mean value of 25.2 ± 6.49 %. Sperms with a negative reaction ranged from 15% to 36 % with a mean value of 24.9 ± 6.69 %.

c. Specimens examined after two hours:

A positive adenosine triphosphatase reaction was also detected in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction for adenosine triphosphatase ranged from 12% to 31 % with a mean value of 21.5 ± 6.42 %. Those with a moderate reaction ranged from 15% to 30 % with a mean value of 21.2 ± 4.96 %. Sperms with a weak reaction ranged from 17% to 36 % with a mean value of 26.9 ± 5.92 %. Sperms with a negative reaction ranged from 24% to 39 % with a mean value of 30.4 ± 5.66 %.

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged for adenosine triphosphatase from 8% to 22 % with a mean value of 15.2 ± 4.73 %. Moderately stained sperms ranged from 9% to 29 % with a mean value of 17.7 ± 6.8 %. Those with a weak reaction ranged from 15% to 29 % with a mean value of 22 ± 4.97 %. Sperms with a negative reaction ranged from 32% to 58 % with a mean value of 45.1 ± 9.1 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction for adenosine triphosphatase ranged from 0% to 4 % with a mean value of 2.2 ± 1.23 %. Those with

a moderate reaction ranged from 2% to 17 % with a mean value of 10 ± 5.35 %. Weakly stained sperms ranged from 8% to 19 % with a mean value of 15 ± 3.59 %. Sperms with a negative reaction ranged from 60% to 90 % with a mean value of 72.8 ± 9.53 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in whole non-washed specimens and the period of incubation [r = - 0.887; P < 0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in whole non-washed specimens and the period of incubation [r = - 0.694; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in whole non-washed specimens and the period of incubation [r = - 0.549; P < 0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in whole non-washed specimens and the period of incubation [r = 0.938; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) Washed specimens: (Table 4)

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 15% to 41 % with a mean value of 28.5 ± 8.05 %. It did not differ statistically from

Table (4): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in washed specimens.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
1	41	26	19	14	11	19	22	29	30	21	15	29	17	39	31	1	9	28	62	41	0	1	9	90
2	27	27	23	23	12	23	29	22	26	22	14	25	20	41	32	7	20	16	57	42	0	1	6	93
3	26	21	24	29	13	19	26	26	29	23	2	11	26	61	33	1	10	25	64	43	0	0	9	91
4	38	33	11	18	14	18	22	27	33	24	13	23	26	38	34	2	12	21	65	44	1	2	8	89
5	15	23	30	32	15	23	27	26	24	25	8	21	24	47	35	6	18	15	61	45	0	2	10	88
6	18	28	30	24	16	16	13	31	40	26	3	13	25	59	36	3	9	21	67	46	1	1	13	85
7	33	34	16	17	17	11	16	35	38	27	6	11	29	54	37	0	9	21	70	47	0	0	8	92
8	31	26	22	21	18	17	19	27	37	28	9	12	27	52	38	5	19	17	59	48	0	0	10	90
9	30	25	26	19	19	28	25	27	20	29	7	16	20	57	39	2	11	21	66	49	0	2	10	88
10	26	23	25	26	20	13	15	34	38	30	10	17	25	48	40	0	8	19	73	50	0	2	9	89
Mean	28.5	26.6	22.6	22.3	Mean	18.7	21.4	28.4	31.5	Mean	8.7	17.8	23.9	49.6	Mean	2.7	12.5	20.4	64.4	Mean	0.2	1.1	9.2	89.5
S.D.	8.05	4.20	5.97	5.62	S.D.	5.01	5.48	3.95	6.80	S.D.	4.42	6.39	3.73	8.36	S.D.	2.50	4.65	3.98	4.90	S.D.	0.42	0.88	1.81	2.27

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (5): Showing the comparison between the *adenosine triphosphatase* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Signific-ance.
Strong reaction	*0*	30.3 ± 5.95	28.5 ± 8.05	0.569	<0.05	-
	1 hour	26.9 ± 6.59	18.7 ± 5.01	3.132	<0.05	+
	2 hours	21.5 ± 6.42	8.7 ± 4.42	5.194	<0.001	+++
	4 hours	15.2 ± 4.73	2.7 ± 2.50	7.387	<0.001	+++
	8 hours	2.2 ± 1.23	0.2 ± 0.42	4.867	<0.001	+++
Moderate reaction	*0*	26.2 ± 7.47	26.6 ± 4.20	0.148	<0.05	-
	1 hour	23.0 ± 5.08	21.4 ± 5.48	0.677	<0.05	-
	2 hours	21.2 ± 4.96	17.8 ± 6.39	1.329	<0.05	-
	4 hours	17.7 ± 6.8	12.5 ± 4.65	1.996	<0.05	-
	8 hours	10.0 ± 5.35	1.1 ± 0.88	5.188	<0.001	+++
Weak reaction	*0*	24.4 ± 7.31	22.6 ± 5.97	0.603	<0.05	-
	1 hour	25.2 ± 6.49	28.4 ± 3.95	1.331	<0.05	-
	2 hours	26.9 ± 5.92	23.9 ± 3.73	1.357	<0.05	-
	4 hours	22.0 ± 4.97	20.4 ± 3.98	0.795	<0.05	-
	8 hours	15.0 ± 3.59	9.2 ± 1.81	4.560	<0.001	+++
No reaction	*0*	19.1 ± 5.34	22.3 ± 5.62	1.305	<0.05	-
	1 hour	24.9 ± 6.69	31.5 ± 6.80	2.187	<0.05	+
	2 hours	30.4 ± 5.66	49.6 ± 8.36	6.016	<0.001	+++
	4 hours	45.1 ± 9.10	64.4 ± 4.90	5.905	<0.001	+++
	8 hours	72.8 ± 9.53	89.5 ± 2.27	5.390	<0.001	+++

+++ = highly significant + = significant - = non-significant

that of the strongly stained sperms in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 5).

Those with a moderate reaction ranged from 21% to 34 % with a mean value of 26.6 ± 4.2 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 5).

Weakly stained sperms ranged from 11% to 30 % with a mean value of 22.6 ± 5.97 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 5).

Sperms with a negative reaction ranged from 14% to 32 % with a mean value of 22.3 ± 5.62 %. They did not also differ statistically from the negatively stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 5).

b. Specimens examined after one hour:

A similar adenosine triphosphatase reaction was noticed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 11% to 28 % with a mean value of 18.7 ± 5.01 %. This was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 5).

Moderately stained sperms ranged from 13% to 29 % with a mean value of 21.4 ± 5.48 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 5).

Those with a weak reaction ranged from 22% to 35 % with a mean value of 28.4 ± 3.95 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 5).

Sperms with a negative reaction ranged from 20% to 40 % with a mean value of 31.5 ± 6.8 %. They were significantly more than that of the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 5).

c. Specimens examined after two hours:

A positive reaction was also detected in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 2% to 15 % with a mean value of 8.7 ± 4.42 %. Statistically, it was highly significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.001$] (Table 5).

Those with a moderate reaction ranged from 11% to 29 % with a mean value of 17.8 ± 6.39 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 5).

Sperms with a weak reaction ranged from 17% to 29 % with a mean value of 23.9 ± 3.73 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 5).

Sperms with a negative reaction ranged from 38% to 61 % with a mean value of 49.6 ± 8.36 %. This was highly significantly more than that of the negatively stained

sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.001$] (Table 5).

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive adenosine triphosphatase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 7 % with a mean value of 2.7 ± 2.5 %. They were highly significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 5).

Moderately stained sperms ranged from 8% to 20 % with a mean value of 12.5 ± 4.65 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 5).

Those with a weak reaction ranged from 15% to 28 % with a mean value of 20.4 ± 3.98 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 5).

Sperms with a negative reaction ranged from 57% to 73 % with a mean value of 64.4 ± 4.9 %. Statistically, they were highly significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 5).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.2 ± 0.42 %. It was highly significantly less than that of the

strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 5).

Those with a moderate reaction ranged from 0% to 2 % with a mean value of 1.1 ± 0.88 %. They were highly significantly less than that of the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 5).

Weakly stained sperms ranged from 6% to 13 % with a mean value of 9.2 ± 1.01 %. They were highly significantly less than that of the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 5).

Sperms with a negative reaction ranged from 85% to 93 % with a mean value of 89.5 ± 2.27 %. Statistically, they were highly significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 5).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the washed specimens and the period of incubation [$r = - 0.789$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the washed specimens and the period of incubation [$r = - 0.885$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the washed specimens and the

period of incubation [r = - 0.774; P <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the washed specimens and the period of incubation [r = 0.955; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 6).

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 18% to 36 % with a mean value of 26.8 ± 6.68 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 7).

Those with a moderate reaction ranged from 20% to 38 % with a mean value of 29 ± 5.94 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined washed specimens [P < 0.05] (Table 7).

Weakly stained sperms ranged from 17% to 35 % with a mean value of 26.1 ± 6.72 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined washed specimens [P < 0.05] (Table 7).

Sperms with a negative reaction ranged from 9% to 27 % with a mean value of 18.1 ± 5.57 %. They did not also differ statistically from the negatively stained sperms for adenosine triphosphatase in the immediately examined washed specimens [P < 0.05] (Table 7).

Table (6): Showing the percentage of sperms with different grades of *Adenosine Triphosphatase* reaction in *Washed* specimens to which calcium was added.

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
1	25	29	27	19	11	17	22	30	31	21	17	27	21	35	31	1	9	23	67	41	0	3	13	84
2	36	38	17	9	12	26	30	25	19	22	14	28	22	36	32	5	21	24	50	42	0	0	8	92
3	22	24	33	21	13	25	28	29	18	23	5	16	30	49	33	3	11	30	56	43	0	2	10	88
4	34	35	17	14	14	20	28	25	27	24	16	23	25	36	34	6	17	17	60	44	0	5	15	80
5	18	20	35	27	15	26	29	22	23	25	11	18	26	45	35	6	20	21	53	45	0	4	12	84
6	19	26	29	26	16	19	28	28	25	26	7	15	27	51	36	4	14	20	62	46	1	2	16	81
7	28	30	25	17	17	16	20	30	34	27	8	18	26	48	37	1	7	24	68	47	0	1	6	93
8	21	29	32	18	18	24	20	31	25	28	10	20	25	45	38	7	13	24	56	48	0	0	10	90
9	33	36	18	13	19	23	31	22	24	29	17	22	20	41	39	5	14	16	65	49	0	3	11	86
10	32	23	28	17	20	15	19	33	33	30	9	28	18	45	40	2	8	26	64	50	0	2	10	88
Mean	26.8	29.0	26.1	18.1	Mean	21.1	25.5	27.5	25.9	Mean	11.4	21.5	24.0	43.1	Mean	4.0	13.4	22.5	60.1	Mean	0.1	2.2	11.1	86.6
S.D.	6.68	5.94	6.72	5.57	S.D.	4.23	4.67	3.81	5.45	S.D.	4.35	4.90	3.65	5.80	S.D.	2.16	4.84	4.17	6.14	S.D.	0.32	1.62	3.03	4.40

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (7): Showing the comparison between the *adenosine triphosphatase* reaction in the sperms of *washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Calcium	t	p	Significance.
Strong reaction	*0*	28.5 ± 8.05	26.8 ± 6.68	0.514	<0.05	-
	1 hour	18.7 ± 5.01	21.1 ± 4.23	1.157	<0.05	-
	2 hours	8.7 ± 4.42	11.4 ± 4.35	1.376	<0.05	-
	4 hours	2.7 ± 2.50	4.0 ± 2.16	1.245	<0.05	-
	8 hours	0.2 ± 0.42	0.1 ± 0.32	0.600	<0.05	-
Moderate reaction	*0*	26.6 ± 4.20	29.0 ± 5.94	1.043	<0.05	-
	1 hour	21.4 ± 5.48	25.5 ± 4.67	1.800	<0.05	-
	2 hours	17.8 ± 6.39	21.5 ± 4.90	1.452	<0.05	-
	4 hours	12.5 ± 4.65	13.4 ± 4.84	0.424	<0.05	-
	8 hours	1.1 ± 0.88	2.2 ± 1.62	1.890	<0.05	-
Weak reaction	*0*	22.6 ± 5.97	26.1 ± 6.72	1.231	<0.05	-
	1 hour	28.4 ± 3.95	27.5 ± 3.81	0.519	<0.05	-
	2 hours	23.9 ± 3.73	24.0 ± 3.65	0.060	<0.05	-
	4 hours	20.4 ± 3.98	22.5 ± 4.17	1.152	<0.05	-
	8 hours	9.2 ± 1.81	11.1 ± 3.03	1.699	<0.05	-
No reaction	*0*	22.3 ± 5.62	18.1 ± 5.57	1.679	<0.05	-
	1 hour	31.5 ± 6.80	25.9 ± 5.45	2.032	<0.05	-
	2 hours	49.6 ± 8.36	43.1 ± 5.80	2.021	<0.05	-
	4 hours	64.4 ± 4.90	60.1 ± 6.14	1.731	<0.05	-
	8 hours	89.5 ± 2.27	86.6 ± 4.40	1.851	<0.05	-

- = non-significant

b. Specimens examined after one hour:

A similar location of adenosine triphosphatase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 15-26 % with a mean value of 21.1 ± 4.23 %. This did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 7).

Moderately stained sperms ranged from 19% to 31 % with a mean value of 25.5 ± 4.67 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 7).

Those with a weak reaction ranged from 22% to 33 % with a mean value of 27.5 ± 3.81 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 7).

Sperms with a negative reaction ranged from 18% to 34 % with a mean value of 25.9 ± 5.45 %. They did not differ statistically from the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 7).

c. Specimens examined after two hours:

A positive adenosine triphosphatase reaction was also detected in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 5% to 17 % with a mean value of 11.4 ± 4.35 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 7).

Those with a moderate reaction ranged from 15% to 28 % with a mean value of 21.5 ± 4.9 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 7).

Sperms with a weak reaction ranged from 18% to 30 % with a mean value of 24 ± 3.65 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 7).

Sperms with a negative reaction ranged from 35% to 51 % with a mean value of 43.1 ± 5.8 %. This did not differ statistically from that of negatively stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [P <0.05] (Table 7).

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 1% to 7 % with a mean value of 4 ± 2.16 %. They did not differ statistically from the strongly stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [P <0.05] (Table 7).

Moderately stained sperms ranged from 7% to 21 % with a mean value of 13.4 ± 4.84 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [P < 0.05] (Table 7).

Those with a weak reaction ranged from 16% to 30 % with a mean value of 22.5 ± 4.17 %. They did not also differ statistically from the weakly stained sperms for

adenosine triphosphatase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 7).

Sperms with a negative reaction ranged from 50% to 68 % with a mean value of 60.1 ± 6.14 %. They did not differ statistically from the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 7).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.1 ± 0.32 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 7).

Those with a moderate reaction ranged from 0% to 5 % with a mean value of 2.2 ± 1.62 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 7).

Weakly stained sperms ranged from 6% to 16 % with a mean value of 11.1 ± 3.03 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 7).

Sperms with a negative reaction ranged from 80% to 93 % with a mean value of 86.6 ± 4.4 %. They did not differ statistically from the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 7).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the washed specimens to which calcium was added and the period of incubation [r = - 0.848; P < 0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the washed specimens to which calcium was added and the period of incubation [r = - 0.906; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the washed specimens to which calcium was added and the period of incubation [r = - 0.778; p <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the washed specimens to which calcium was added and the period of incubation [r = 0.966; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Washed specimens to which ascorbic acid was added: (Table 8)

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 24% to 38 % with a mean value of 31 ± 5.03 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 9).

Those with a moderate reaction ranged from 20% to 34 % with a mean value of 26.5 ± 4.7 %. They did not differ statistically from the moderately stained sperms for

Table (8): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in the washed specimens to which ascorbic acid was added.*

No.of Ejaculate	Immediate				No.of Ejaculate	1 hour				No.of Ejaculate	2 hours				No.of Ejaculate	4 hours				No.of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	38	34	14	14	11	24	21	33	22	21	15	22	31	32	31	4	21	22	53	41	3	4	10	83
2	33	27	23	17	12	27	28	34	11	22	20	23	28	29	32	6	26	27	41	42	0	2	6	92
3	25	20	31	24	13	23	33	25	19	23	9	16	24	51	33	4	15	16	65	43	2	5	5	88
4	35	30	19	16	14	23	21	24	32	24	21	22	26	31	34	7	15	12	66	44	3	9	11	77
5	27	21	30	22	15	31	30	22	17	25	16	24	27	33	35	8	25	26	41	45	1	8	9	82
6	24	21	29	26	16	21	31	22	26	26	8	17	24	51	36	5	24	23	48	46	4	9	11	76
7	36	31	16	17	17	15	26	36	23	27	10	20	23	47	37	4	13	18	65	47	1	3	7	89
8	35	28	18	19	18	18	24	31	27	28	11	23	22	44	38	7	20	25	48	48	0	3	5	92
9	30	28	21	21	19	26	27	23	24	29	14	19	28	39	39	4	20	21	55	49	3	6	15	76
10	27	25	24	24	20	15	20	35	30	30	13	22	24	41	40	3	16	18	63	50	2	7	7	84
Mean	31.0	26.5	22.5	20.0	Mean	22.3	26.1	28.5	23.1	Mean	13.7	20.8	25.7	39.8	Mean	5.2	19.5	20.8	54.5	Mean	1.9	5.6	8.6	83.9
S.D.	5.03	4.70	5.99	4.00	S.D.	5.19	4.53	5.80	6.26	S.D.	4.42	2.70	2.79	8.32	S.D.	1.69	4.60	4.78	9.87	S.D.	1.37	2.59	3.20	6.24

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (9): Showing the comparison between *the adenosine triphosphatase reaction in the sperms of washed specimens and washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	28.5 ± 8.05	31.0 ± 5.03	0.833	<0.05	-
	1 hour	18.7 ± 5.01	22.3 ± 5.19	1.578	<0.05	-
	2 hours	8.7 ± 4.42	13.7 ± 4.42	2.528	<0.05	+
	4 hours	2.7 ± 2.50	5.2 ± 1.69	2.624	<0.05	+
	8 hours	0.2 ± 0.42	1.9 ± 1.37	3.750	<0.05	+
Moderate reaction	*0*	26.6 ± 4.20	26.5 ± 4.7	0.050	<0.05	-
	1 hour	21.4 ± 5.48	26.1 ± 4.53	2.089	<0.05	-
	2 hours	17.8 ± 6.39	20.8 ± 2.70	1.367	<0.05	-
	4 hours	12.5 ± 4.65	19.5 ± 4.60	3.385	<0.05	+
	8 hours	1.1 ± 0.88	5.6 ± 2.59	5.204	<0.001	+++
Weak reaction	*0*	22.6 ± 5.97	22.5 ± 5.99	0.037	<0.05	-
	1 hour	28.4 ± 3.95	28.5 ± 5.80	0.045	<0.05	-
	2 hours	23.9 ± 3.73	25.7 ± 2.79	1.223	<0.05	-
	4 hours	20.4 ± 3.98	20.8 ± 4.78	0.203	<0.05	-
	8 hours	9.2 ± 1.81	8.6 ± 3.2	0.515	<0.05	-
No reaction	*0*	22.3 ± 5.62	20.0 ± 4.00	1.055	<0.05	-
	1 hour	31.5 ± 6.80	23.1 ± 6.26	2.873	<0.05	+
	2 hours	49.6 ± 8.36	39.8 ± 8.32	2.628	<0.05	+
	4 hours	64.4 ± 4.90	54.5 ± 9.87	2.841	<0.05	+
	8 hours	89.5 ± 2.27	83.9 ± 6.24	2.665	<0.05	+

+++ = highly significant + = significant - = non-significant

adenosine triphosphatase in the immediately examined washed specimens [$P < 0.05$] (Table 9).

Weakly stained sperms ranged from 14% to 31 % with a mean value of 22.5 ± 5.99 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined washed specimens [$P < 0.05$] (Table 9).

Sperms with a negative reaction ranged from 14% to 26 % with a mean value of 20 ± 4 %. They did not also differ statistically from the negatively stained sperms for adenosine triphosphatase in the immediately examined washed specimens [$P < 0.05$] (Table 9).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece and tail. The percentage of spermatozoa giving a strong adenosine triphosphatase reaction ranged from 15% to 31 % with a mean value of 22.3 ± 5.19 %. Also, this did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 9).

Moderately stained sperms ranged from 20% to 33 % with a mean value of 26.1 ± 4.53 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 9).

Those with a weak reaction ranged from 22% to 36 % with a mean value of 28.5 ± 5.8 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 9).

Sperms with a negative reaction ranged from 11% to 32 % with a mean value of 23.1 ± 6.26 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 9).

c. Specimens examined after two hours:

A similar location of adenosine triphosphatase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 8% to 21 % with a mean value of 13.7 ± 4.42 %. It was significantly more than that of the strongly stained sperms in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 9).

Those with a moderate reaction ranged from 16% to 24 % with a mean value of 20.8 ± 2.7 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 9).

Sperms with a weak reaction ranged from 22% to 31 % with a mean value of 25.7 ± 2.79 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 9).

Sperms with a negative reaction ranged from 29% to 51 % with a mean value of 39.8 ± 8.32 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 9).

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 3% to 8 % with a mean value of 5.2 ± 1.69 %. This was significantly more than that of the strongly stained sperms for

adenosine triphosphatase in the washed specimens examined after four hours of incubation [P <0.05] (Table 9).

Moderately stained sperms reaction ranged from 13% to 26 % with a mean value of 19.5 ± 4.6 %. They were significantly more than that of the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [P < 0.05] (Table 9).

Those with a weak reaction ranged from 12% to 27 % with a mean value of 20.8 ± 4.78 %. They did not differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [P < 0.05] (Table 9).

Sperms with a negative reaction ranged from 41% to 66 % with a mean value of 54.5 ± 9.87 %. This was significantly less than that of the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after four hours of incubation [P <0.05] (Table 9).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 1.9 ± 1.37 %. It was significantly more than that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [P <0.05] (Table 9).

Those with a moderate reaction ranged from 2% to 9 % with a mean value of 5.6 ± 2.59 %. They were highly significantly more than that of the moderately stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [P < 0.001] (Table 9).

Weakly stained sperms ranged from 5% to 15 % with a mean value of 8.6 ± 3.2 %. They did not differ statistically from the weakly stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 9).

Sperms with a negative reaction ranged from 76% to 92 % with a mean value of 83.9 ± 6.24 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 9).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.849$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.878$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.748$; $P < 0.001$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = 0.952$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(B) SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 10).

a. Immediately examined specimens:

Also, only the mid-piece and tail were the sites of positive adenosine triphosphatase reaction. The percentage of spermatozoa giving a strong reaction ranged from 29% to 44 % with a mean value of 36.3 ± 5.52 %. They were significantly more than the strongly stained sperms in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 11).

Moderately stained sperms ranged from 16% to 38 % with a mean value of 25.6 ± 6.8 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 11).

Those with a weak reaction ranged from 11% to 34 % with a mean value of 24.2 ± 7.38 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 11).

Sperms with a negative reaction ranged from 8% to 21 % with a mean value of 13.9 ± 4.38 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 11).

b. Specimens examined after one hour:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 25% to 43 % with a mean value of 33.9 ± 6.24 %. It was significantly more than that of the strongly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 11).

Table (10): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in the first split fraction specimens.*

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
51	33	27	25	15	61	29	24	24	23	71	26	29	23	22	81	20	23	25	32	91	6	14	22	58
52	25	23	29	13	62	41	34	16	9	72	41	20	21	18	82	15	12	26	47	92	5	13	20	62
53	44	36	11	9	63	37	20	23	20	73	24	18	25	33	83	17	23	23	37	93	4	14	18	64
54	41	25	22	12	64	36	24	21	19	74	20	18	32	30	84	25	20	19	36	94	6	18	22	54
55	29	16	34	21	65	38	26	24	12	75	32	27	27	14	85	28	25	19	28	95	3	13	16	68
56	37	25	24	14	66	25	20	29	26	76	33	30	21	16	86	21	20	18	41	96	3	14	16	67
57	30	19	33	18	67	27	22	34	17	77	36	26	21	17	87	23	22	27	28	97	6	15	23	56
58	31	23	27	19	68	28	21	35	16	78	22	20	27	31	88	13	19	20	48	98	1	13	13	73
59	40	38	14	8	69	43	25	21	11	79	35	17	21	27	89	16	19	25	40	99	4	14	12	70
60	43	24	23	10	70	35	22	25	18	70	21	23	26	30	80	26	22	26	26	100	3	11	16	70
Mean	36.3	25.6	24.2	13.9	Mean	33.9	23.8	25.2	17.1	Mean	29.0	22.8	24.4	23.8	Mean	20.4	20.5	22.8	36.3	Mean	4.1	13.9	17.8	64.2
S.D.	5.52	6.80	7.38	4.38	S.D.	6.24	4.13	5.92	5.34	S.D.	7.32	4.87	3.69	7.18	S.D.	5.08	3.57	3.46	7.82	S.D.	1.66	1.79	3.85	6.51

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (11): Showing the comparison between *the adenosine triphosphatase* reaction in the sperms of *whole non-washed* and *first split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Significance.
Strong reaction	0	30.3 ± 5.95	36.3 ± 5.52	2.339	<0.05	+
	1 hour	26.9 ± 6.59	33.9 ± 6.24	2.438	<0.05	+
	2 hours	21.5 ± 6.42	29.0 ± 7.32	2.437	<0.05	+
	4 hours	15.2 ± 4.73	20.4 ± 5.08	2.362	<0.05	+
	8 hours	2.2 ± 1.23	4.1 ± 1.66	2.905	<0.05	+
Moderate reaction	0	26.2 ± 7.47	25.6 ± 6.80	0.188	<0.05	-
	1 hour	23.0 ± 5.08	23.8 ± 4.13	0.387	<0.05	-
	2 hours	21.2 ± 4.96	22.8 ± 4.87	0.728	<0.05	-
	4 hours	17.7 ± 6.8	20.5 ± 3.57	1.153	<0.05	-
	8 hours	10.0 ± 5.35	13.9 ± 1.79	2.184	<0.05	+
Weak reaction	0	24.4 ± 7.31	24.2 ± 7.38	0.061	<0.05	-
	1 hour	25.2 ± 6.49	25.2 ± 5.92	0.000	<0.05	-
	2 hours	26.9 ± 5.92	24.4 ± 3.69	1.134	<0.05	-
	4 hours	22.0 ± 4.97	22.8 ± 3.46	0.418	<0.05	-
	8 hours	15.0 ± 3.59	17.8 ± 3.85	1.681	<0.05	-
No reaction	0	19.1 ± 5.34	13.9 ± 4.38	2.380	<0.05	+
	1 hour	24.9 ± 6.69	17.1 ± 5.34	2.881	<0.05	+
	2 hours	30.4 ± 5.66	23.8 ± 7.18	2.283	<0.05	+
	4 hours	45.1 ± 9.10	36.3 ± 7.82	2.320	<0.05	+
	8 hours	72.8 ± 9.53	64.2 ± 6.51	2.356	<0.05	+

+ = significant - = non-significant

Those with a moderate reaction ranged from 20% to 34 % with a mean value of 23.8 ± 4.13 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 11).

Weakly stained sperms ranged from 16% to 35 % with a mean value of 25.2 ± 5.92 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 11).

Sperms with a negative reaction ranged from 9% to 26 % with a mean value of 17.1 ± 5.34 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 11).

c. Specimens examined after two hours:

A similar location of adenosine triphosphatase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 20% to 41 % with a mean value of 29 ± 7.32 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 11).

Those with a moderate reaction ranged from 17% to 30 % with a mean value of 22.8 ± 4.87 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 11).

Sperms with a weak reaction ranged from 21% to 32 % with a mean value of 24.4 ± 3.69 %. They did not also differ statistically from the weakly stained sperms for

adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 11).

Sperms with a negative reaction ranged from 14% to 33 % with a mean value of 23.8 ± 7.18 %. This was significantly less than that of negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 11).

d. Specimens examined after four hours:

A positive reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 13% to 28 % with a mean value of 20.4 ± 5.08 %. It was significantly more than that of the strongly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 11).

Those with a moderate reaction ranged from 12% to 25 % with a mean value of 20.5 ± 3.57 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 11).

Weakly stained sperms ranged from 18% to 27 % with a mean value of 22.8 ± 3.46 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 11).

Sperms with a negative reaction ranged from 26% to 48 % with a mean value of 36.3 ± 7.82 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 11).

c. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 1% to 6 % with a mean value of 4.1 ± 1.66 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 11).

Those with a moderate reaction ranged from 11% to 18 % with a mean value of 13.9 ± 1.79 %. They were significantly more than that of the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 11).

Weakly stained sperms ranged from 12% to 23 % with a mean value of 17.8 ± 3.85 %. They did not differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 11).

Sperms with a negative reaction ranged from 54% to 73 % with a mean value of 64.2 ± 6.51 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 11).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the first split fractions and the period of incubation [$r = - 0.912$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens

and the period of incubation [r = - 0.684; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the first split fraction specimens and the period of incubation [r = - 0.456; $P < 0.001$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the first split fractions and the period of incubation [r = 0.947; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added:

(Table 12).

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 27% to 48 % with a mean value of 37.1 ± 7.87 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined first split fraction specimens [$P < 0.05$] (Table 13).

Those with a moderate reaction ranged from 21% to 33 % with a mean value of 27.2 ± 4.26 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 13).

Weakly stained sperms ranged from 11% to 33 % with a mean value of 22.8 ± 7.79 %. They did not also differ statistically from the weakly stained sperms for

Table (12): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in the first split fraction specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	42	26	21	11	61	27	23	32	18	71	26	28	24	22	81	27	24	25	24	91	8	17	22	53
52	31	28	27	14	62	36	31	20	13	72	38	31	18	13	82	20	18	24	38	92	6	16	15	63
53	48	33	11	8	63	32	26	29	13	73	22	20	32	26	83	30	22	21	27	93	7	17	21	55
54	44	27	18	11	64	35	26	25	14	74	26	20	29	25	84	30	28	18	24	94	10	26	14	50
55	27	22	33	18	65	40	33	16	11	75	37	24	24	15	85	27	23	25	25	95	4	14	20	62
56	41	23	23	13	66	26	21	34	19	76	30	26	25	19	86	21	25	22	32	96	8	15	12	65
57	30	29	26	15	67	29	29	26	16	77	41	31	19	9	87	24	23	24	29	97	9	23	14	54
58	29	21	33	17	68	21	22	36	21	78	24	24	28	24	88	22	22	28	28	98	6	17	16	61
59	46	33	11	10	69	39	29	19	13	79	33	26	24	17	89	23	18	20	39	99	4	12	20	64
60	33	30	25	12	70	29	27	27	17	70	27	25	25	23	80	31	24	19	26	100	6	19	23	52
Mean	37.1	27.2	22.8	12.9	Mean	31.4	26.7	26.4	15.5	Mean	30.4	25.5	24.8	19.3	Mean	25.5	22.7	22.6	29.2	Mean	6.8	17.6	17.7	57.9
S.D.	7.87	4.26	7.79	3.14	S.D.	6.10	3.92	6.62	3.21	S.D.	6.52	3.84	4.24	5.68	S.D.	4.03	3.02	3.13	5.47	S.D.	1.99	4.17	3.92	5.63

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (13): Showing the comparison between the *adenosine triphosphatase reaction in the sperms of first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	36.3 ± 5.52	37.1 ± 7.87	0.263	<0.05	-
	1 hour	33.9 ± 6.24	31.4 ± 6.10	0.906	<0.05	-
	2 hours	29.0 ± 7.32	30.4 ± 6.52	0.452	<0.05	-
	4 hours	20.4 ± 5.08	25.5 ± 4.03	2.486	<0.05	+
	8 hours	4.1 ± 1.66	6.8 ± 1.99	3.293	<0.05	+
Moderate reaction	*0*	25.6 ± 6.80	27.2 ± 4.26	0.630	<0.05	-
	1 hour	23.8 ± 4.13	26.7 ± 3.92	1.611	<0.05	-
	2 hours	22.8 ± 4.87	25.5 ± 3.84	1.377	<0.05	-
	4 hours	20.5 ± 3.57	22.7 ± 3.02	1.489	<0.05	-
	8 hours	13.9 ± 1.79	17.6 ± 4.17	2.579	<0.05	+
Weak reaction	*0*	24.2 ± 7.38	22.8 ± 7.79	0.413	<0.05	-
	1 hour	25.2 ± 5.92	26.4 ± 6.62	0.427	<0.05	-
	2 hours	24.4 ± 3.69	24.8 ± 4.24	0.225	<0.05	-
	4 hours	22.8 ± 3.46	22.6 ± 13.3	0.136	<0.05	-
	8 hours	17.8 ± 3.85	17.7 ± 3.92	0.058	<0.05	-
No reaction	*0*	13.9 ± 4.38	12.9 ± 3.14	0.586	<0.05	-
	1 hour	17.1 ± 5.34	15.5 ± 3.21	0.812	<0.05	-
	2 hours	23.8 ± 7.18	19.3 ± 5.68	1.555	<0.05	-
	4 hours	36.3 ± 7.82	29.2 ± 5.47	2.353	<0.05	+
	8 hours	64.2 ± 6.51	57.9 ± 5.63	2.315	<0.05	+

+ = significant - = non-significant

adenosine triphosphatase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 13).

Sperms with a negative reaction ranged from 8% to 18 % with a mean value of 12.9 ± 3.14 %. They did not also differ statistically from the negatively stained sperms for adenosine triphosphatase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 13).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece and tail. The percentage of spermatozoa giving a strong adenosine triphosphatase reaction ranged from 21% to 40 % with a mean value of 31.4 ± 6.1 %. Also, this did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 13).

Moderately stained sperms ranged from 21% to 33 % with a mean value of 26.7 ± 3.92 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 13).

Those with a weak reaction ranged from 16% to 36 % with a mean value of 26.4 ± 6.62 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 13).

Sperms with a negative reaction ranged from 11% to 21 % with a mean value of 15.5 ± 3.21 %. They did not differ statistically from the negatively stained sperms for adenosine triphosphatase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 13).

c. Specimens examined after two hours:

A similar location of adenosine triphosphatase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 22% to 41 % with a mean value of 30.4 ± 6.52 %. Also, it did not differ statistically from that of the strongly stained sperms for adenosine triphosphatase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 13).

Those with a moderate reaction ranged from 20% to 31 % with a mean value of 25.5 ± 3.84 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 13).

Sperms with a weak reaction ranged from 18% to 32 % with a mean value of 24.8 ± 4.24 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 13).

Sperms with a negative reaction ranged from 9% to 26 % with a mean value of 19.3 ± 5.68 %. They did not differ statistically from the negatively stained sperms for adenosine triphosphatase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 13).

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 20% to 31 % with a mean value of 25.5 ± 4.03 %. This was significantly more than that of the strongly stained sperms for adenosine triphosphatase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 13).

Moderately stained sperms ranged from 18% to 28 % with a mean value of 22.7 ± 3.02 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 13).

Those with a weak reaction ranged from 18% to 28 % with a mean value of 22.6 ± 3.13 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 13).

Sperms with a negative reaction ranged from 24% to 39 % with a mean value of 29.2 ± 5.47 %. This was significantly less than that of negatively stained sperms for adenosine triphosphatase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 13).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 4% to 10 % with a mean value of 6.8 ± 1.99 %. It was significantly more than that of the strongly stained sperms in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 13).

Those with a moderate reaction ranged from 12% to 26 % with a mean value of 17.6 ± 4.17 %. They were significantly more than that of the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 13).

Weakly stained sperms ranged from 12% to 23 % with a mean value of 17.7 ± 3.92 %. They did not differ statistically from the weakly stained sperms for

adenosine triphosphatase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 13).

Sperms with a negative reaction ranged from 50% to 65 % with a mean value of 57.9 ± 5.63 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 13).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the first split fractions to which ascorbic acid was added and the period of incubation [$r = - 0.875$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.691$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.416$; $P < 0.05$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the first split fractions to which ascorbic acid was added and the period of incubation [$r = 0.953$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 14).

a. Immediately examined specimens:

Also, only the mid-piece and tail were the sites of positive adenosine triphosphatase reaction. The percentage of spermatozoa giving a strong reaction ranged from 12% to 29 % with a mean value of 22.2 ± 6.29 %. They were significantly less than the strongly stained sperms in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 15).

Moderately stained sperms ranged from 15% to 30 % with a mean value of 22.9 ± 5.47 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 15).

Those with a weak reaction ranged from 18% to 30 % with a mean value of 24.4 ± 4.88 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 15).

Sperms with a negative reaction ranged from 21% to 41 % with a mean value of 30.5 ± 6.9 %. They were highly significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined immediately [$P < 0.001$] (Table 15).

b. Specimens examined after one hour:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 9% to 23 % with a mean value of 16.8 ± 4.47 %. It was highly significantly less than that of the strongly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.001$] (Table 15).

Table (14): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in the second split fraction specimens.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	18	26	21	35	61	21	29	18	32	71	7	12	28	53	81	5	14	16	65	91	0	2	7	91
52	15	19	30	36	62	17	21	24	38	72	10	12	33	45	82	8	22	24	46	92	1	7	15	77
53	26	21	24	29	63	15	16	21	48	73	18	27	17	38	83	7	20	21	52	93	1	6	10	83
54	17	15	29	39	64	14	14	23	49	74	16	28	19	37	84	6	15	22	57	94	0	4	7	89
55	29	27	19	25	65	9	12	27	52	75	13	16	29	42	85	4	17	15	64	95	2	6	15	77
56	26	28	22	24	66	20	17	26	37	76	9	26	30	35	86	3	12	19	66	96	3	8	13	76
57	29	28	18	25	67	23	31	16	30	77	12	10	29	49	87	2	16	15	67	97	0	3	9	88
58	28	30	21	21	68	21	22	22	35	78	14	17	24	45	88	10	14	26	50	98	1	6	7	86
59	12	17	30	41	69	12	12	25	51	79	15	21	21	43	89	9	13	27	51	99	0	6	9	85
60	22	18	30	30	70	16	17	24	43	70	14	13	27	46	80	7	16	22	55	100	2	7	17	74
Mean	22.2	22.9	24.4	30.5	Mean	16.8	19.1	22.6	41.5	Mean	12.8	18.2	25.7	43.3	Mean	6.1	15.9	20.7	57.3	Mean	1.0	5.5	10.9	82.6
S.D.	6.29	5.47	4.88	6.90	S.D.	4.47	6.64	3.47	8.15	S.D.	3.36	6.83	5.23	5.56	S.D.	2.60	3.11	4.37	7.66	S.D.	1.05	1.90	3.78	6.13

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (15): Showing the comparison between the *adenosine triphosphatase* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Significance.
Strong reaction	*0*	30.3 ± 5.95	22.2 ± 6.29	2.961	<0.05	+
	1 hour	26.9 ± 6.59	16.8 ± 4.47	4.012	<0.001	+++
	2 hours	21.5 ± 6.42	12.8 ± 3.36	3.799	<0.05	+
	4 hours	15.2 ± 4.73	6.1 ± 2.60	5.328	<0.001	+++
	8 hours	2.2 ± 1.23	1.0 ± 1.05	2.343	<0.05	+
Moderate reaction	*0*	26.2 ± 7.47	22.9 ± 5.47	1.128	<0.05	-
	1 hour	23.0 ± 5.08	19.1 ± 6.64	1.475	<0.05	-
	2 hours	21.2 ± 4.96	18.2 ± 6.83	1.124	<0.05	-
	4 hours	17.7 ± 6.80	15.9 ± 3.11	0.761	<0.05	-
	8 hours	10.0 ± 5.35	5.5 ± 1.90	2.505	<0.05	+
Weak reaction	*0*	24.4 ± 7.31	24.4 ± 4.88	0.000	<0.05	-
	1 hour	25.2 ± 6.49	22.6 ± 3.47	1.117	<0.05	-
	2 hours	26.9 ± 5.92	25.7 ± 5.23	0.481	<0.05	-
	4 hours	22.0 ± 4.97	20.7 ± 4.37	0.621	<0.05	-
	8 hours	15.0 ± 3.59	10.9 ± 3.78	2.486	<0.05	+
No reaction	*0*	19.1 ± 5.34	30.5 ± 6.90	4.131	<0.001	+++
	1 hour	24.9 ± 6.69	41.5 ± 8.15	4.977	<0.001	+++
	2 hours	30.4 ± 5.66	43.3 ± 5.56	5.142	<0.001	+++
	4 hours	45.1 ± 9.10	57.3 ± 7.66	3.244	<0.05	+
	8 hours	72.8 ± 9.53	82.6 ± 6.13	2.734	<0.05	+

- = non-significant + = significant +++ = highly significant

Those with a moderate reaction ranged from 12% to 31 % with a mean value of 19.1 ± 6.64 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 15).

Weakly stained sperms ranged from 16% to 27 % with a mean value of 22.6 ± 3.47 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 15).

Sperms with a negative reaction ranged from 30% to 52 % with a mean value of 41.5 ± 8.15 %. They were highly significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after one hour of incubation [$P < 0.001$] (Table 15).

c. Specimens examined after two hours:

A positive adenosine triphosphatase reaction was also detected in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 7% to 18 % with a mean value of 12.8 ± 3.36 %. It was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 15).

Those with a moderate reaction ranged from 10% to 28 % with a mean value of 18.2 ± 6.83 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 15).

Sperms with a weak reaction ranged from 17% to 33 % with a mean value of 25.7 ± 5.23 %. They did not also differ statistically from the weakly stained sperms for

adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 15).

Sperms with a negative reaction ranged from 35% to 53 % with a mean value of 43.3 ± 5.56 %. This was highly significantly more than that of negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after two hours of incubation [$P < 0.001$] (Table 15).

d. Specimens examined after four hours:

A similar location of adenosine triphosphatase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 2% to 10 % with a mean value of 6.1 ± 2.6 %. It was highly significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 15).

Those with a moderate reaction ranged from 12% to 22 % with a mean value of 15.9 ± 3.11 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 15).

Weakly stained sperms ranged from 15% to 27 % with a mean value of 20.7 ± 4.37 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 15).

Sperms with a negative reaction ranged from 46% to 67 % with a mean value of 57.3 ± 7.66 %. They were significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 15).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1 ± 1.05 %. It was significantly less than that of the strongly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 15).

Those with a moderate reaction ranged from 2% to 8 % with a mean value of 5.5 ± 1.9 %. They were significantly less than that of the moderately stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 15).

Weakly stained sperms ranged from 7% to 17 % with a mean value of 10.9 ± 3.78 %. They were significantly less than that of the weakly stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 15).

Sperms with a negative reaction ranged from 74% to 91 % with a mean value of 82.6 ± 6.13 %. They were significantly more than the negatively stained sperms for adenosine triphosphatase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 15).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the second split fractions and the period of incubation [$r = - 0.856$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the second split fraction

specimens and the period of incubation [r = - 0.756; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens and the period of incubation [r = - 0.726; P < 0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the second split fractions and the period of incubation [r = 0.935; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added: (Table 16).

a. Immediately examined specimens:

A positive adenosine triphosphatase reaction was also observed in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 20% to 38 % with a mean value of 29.2 ± 6.37 %. It was significantly more than that of the strongly stained sperms in the immediately examined second split fraction specimens [P < 0.05] (Table 17).

Those with a moderate reaction ranged from 21% to 32 % with a mean value of 26.7 ± 4.22 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the immediately examined second split fraction specimens [P < 0.05] (Table 17).

Weakly stained sperms ranged from 15% to 28 % with a mean value of 20.3 ± 4.32 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the immediately examined second split fraction specimens [P < 0.05] (Table 17).

Table (16): Showing the percentage of sperms with different grades of *adenosine triphosphatase reaction in the second split fraction specimens to which calcium was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	38	30	15	17	61	27	18	24	31	71	13	15	27	45	81	5	14	23	58	91	1	5	13	81
52	33	25	21	21	62	15	18	23	44	72	11	18	28	43	82	12	21	31	36	92	4	10	14	72
53	21	25	26	28	63	18	21	22	39	73	20	31	21	28	83	12	16	26	46	93	3	9	13	75
54	20	21	28	31	64	29	25	23	23	74	22	26	23	29	84	7	20	25	48	94	0	9	9	82
55	28	32	20	20	65	23	20	17	40	75	15	16	20	49	85	6	19	22	53	95	7	10	16	67
56	26	26	18	30	66	28	19	25	28	76	19	22	30	29	86	10	18	16	56	96	5	11	19	65
57	35	32	17	16	67	30	27	19	24	77	15	20	25	40	87	6	20	16	58	97	2	7	12	79
58	36	31	15	18	68	28	21	26	25	78	20	22	27	31	88	13	21	31	35	98	2	9	11	78
59	31	22	22	25	69	17	20	19	44	79	18	24	27	31	89	13	22	24	41	99	6	9	25	60
60	24	23	21	32	70	21	22	20	37	70	16	21	29	34	80	11	16	33	40	100	4	8	11	77
Mean	29.2	26.7	20.3	23.8	Mean	23.6	21.1	21.8	33.5	Mean	16.9	21.5	25.7	35.9	Mean	9.5	18.7	24.7	47.1	Mean	3.4	8.7	14.3	73.6
S.D.	6.37	4.22	4.32	6.14	S.D.	5.54	2.92	2.94	8.26	S.D.	3.48	4.77	3.37	7.68	S.D.	3.17	2.63	5.89	8.89	S.D.	2.22	1.70	4.69	7.40

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (17): Showing the comparison between the *adenosine triphosphatase* reaction in the sperms of *second split fraction specimens and second split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Significance.
Strong reaction	*0*	22.2 ± 6.29	29.2 ± 6.37	2.473	<0.05	+
	1 hour	16.8 ± 4.47	23.6 ± 5.54	3.021	<0.05	+
	2 hours	12.8 ± 3.36	16.9 ± 3.48	2.681	<0.05	+
	4 hours	6.1 ± 2.60	9.5 ± 3.17	2.621	<0.05	+
	8 hours	1.0 ± 1.05	3.4 ± 2.22	3.087	<0.05	+
Moderate reaction	*0*	22.9 ± 5.47	26.7 ± 4.22	1.741	<0.05	-
	1 hour	19.1 ± 6.64	21.1 ± 2.92	0.872	<0.05	-
	2 hours	18.2 ± 6.83	21.5 ± 4.77	1.253	<0.05	-
	4 hours	15.9 ± 3.11	18.7 ± 2.63	2.176	<0.05	+
	8 hours	5.5 ± 1.90	8.7 ± 1.70	3.966	<0.001	+++
Weak reaction	*0*	24.4 ± 4.88	20.3 ± 4.32	1.989	<0.05	-
	1 hour	22.6 ± 3.47	21.8 ± 2.94	0.557	<0.05	-
	2 hours	25.7 ± 5.23	25.7 ± 3.37	0.000	<0.05	-
	4 hours	20.7 ± 4.37	24.7 ± 5.89	1.725	<0.05	-
	8 hours	10.9 ± 3.78	14.3 ± 4.69	1.784	<0.05	-
No reaction	*0*	30.5 ± 6.90	23.8 ± 6.14	2.293	<0.05	+
	1 hour	41.5 ± 8.15	33.5 ± 8.26	2.179	<0.05	+
	2 hours	43.3 ± 5.56	35.9 ± 7.68	2.468	<0.05	+
	4 hours	57.3 ± 7.66	47.1 ± 8.89	2.749	<0.05	+
	8 hours	82.6 ± 6.13	73.6 ± 7.40	2.692	<0.05	+

+++ = highly significant + = significant - = non-significant

Sperms with a negative reaction ranged from 16% to 32 % with a mean value of 23.8 ± 6.14 %. They were significantly less than the negatively stained sperms for adenosine triphosphatase in the immediately examined second split fraction specimens [P <0.05] (Table 17).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece and tail. The percentage of spermatozoa giving a strong adenosine triphosphatase reaction ranged from 15% to 30 % with a mean value of 23.6 ± 5.54 %. This was significantly more than that of the strongly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after one hour of incubation [P <0.05] (Table 17).

Moderately stained sperms ranged from 18% to 27 % with a mean value of 21.1 ± 2.92 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 17).

Those with a weak reaction ranged from 17% to 26 % with a mean value of 21.8 ± 2.94 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 17).

Sperms with a negative reaction ranged from 23% to 44 % with a mean value of 33.5 ± 8.26 %. They were significantly less than negatively stained sperms for adenosine triphosphatase in the second split fraction specimens examined after one hour of incubation [P <0.05] (Table 17).

c. Specimens examined after two hours:

A positive reaction was also detected in the mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 11% to 22 % with a mean value of 16.9 ± 3.48 %. It was significantly more than that of the strongly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 17).

Those with a moderate reaction ranged from 15% to 31 % with a mean value of 21.5 ± 4.77 %. They did not differ statistically from the moderately stained sperms for adenosine triphosphatase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 17).

Sperms with a weak reaction ranged from 20% to 30 % with a mean value of 25.7 ± 3.37 %. They did not also differ statistically from the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 17).

Sperms with a negative reaction ranged from 28% to 49 % with a mean value of 35.9 ± 7.68 %. This was significantly less than that of negatively stained sperms for adenosine triphosphatase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 17).

d. Specimens examined after four hours:

Also, only the mid-piece and tail were the sites of positive adenosine triphosphatase reaction. The percentage of spermatozoa giving a strong reaction ranged from 5% to 13 % with a mean value of 9.5 ± 3.17 %. They were significantly more than the strongly stained sperms in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 17).

Moderately stained sperms ranged from 14% to 22 % with a mean value of 18.7 ± 2.63 %. They were significantly more than that of the moderately stained sperms for adenosine triphosphatase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 17).

Those with a weak reaction ranged from 16% to 33 % with a mean value of 24.7 ± 5.89 %. They did not differ statistically from the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 17).

Sperms with a negative reaction ranged from 35% to 58 % with a mean value of 47.1 ± 8.89 %. They were significantly less than negatively stained sperms for adenosine triphosphatase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 17).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of adenosine triphosphatase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 7 % with a mean value of 3.4 ± 2.22 %. It was significantly more than that of the strongly stained sperms in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 17).

Those with a moderate reaction ranged from 5% to 11 % with a mean value of 8.7 ± 1.7 %. They were highly significantly more than that of the moderately stained sperms for adenosine triphosphatase in the second split fraction specimens examined after eight hours of incubation [$P < 0.001$] (Table 17).

Weakly stained sperms ranged from 9% to 25 % with a mean value of 14.3 ± 4.69 %. They did not differ statistically from the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 17).

Sperms with a negative reaction ranged from 60% to 82 % with a mean value of 73.6 ± 7.4 %. They were significantly less than negatively stained sperms for adenosine triphosphatase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 17).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for adenosine triphosphatase in the second split fractions to which calcium was added and the period of incubation [$r = - 0.867$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for adenosine triphosphatase in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.854$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A significantly negative correlation was found between the percent of the weakly stained sperms for adenosine triphosphatase in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.416$; $P < 0.05$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significant positive correlation was found between the percent of the negatively stained sperms for adenosine triphosphatase in the second split fractions to which calcium was added and the period of incubation [$r = 0.915$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

II. LACTATE DEHYDROGENASE:

A. WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 18)

a. Immediately examined specimens:

A positive reaction for lactate dehydrogenase was noticed in the mid-piece. No reaction was seen in other parts of spermatozoa (Fig. 2).

The percentage of spermatozoa giving a strong reaction ranged from 17% to 28 % with a mean value of 21.5 ± 3.5 %. Those with a moderate reaction ranged from 30% to 55 % with a mean value of 43.5 ± 9.26 %. Weakly stained sperms ranged from 10% to 29 % with a mean value of 17.5 ± 6.79 %. Sperms with a negative reaction ranged from 7% to 28 % with a mean value of 17.5 ± 6.47 %.

b. Specimens examined after one hour:

A similar lactate dehydrogenase reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 17% to 26 % with a mean value of 20.4 ± 2.95 %. Moderately stained sperms ranged from 28% to 53 % with a mean value of 41.8 ± 8.6 %. Those with a weak reaction ranged from 14% to 29 % with a mean value of 19.1 ± 4.84 %. Sperms with a negative reaction ranged from 11% to 28 % with a mean value of 18.7 ± 6.7 %.

c. Specimens examined after two hours:

A positive lactate dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 8% to 22 % with a mean value of 15.9 ± 5.43 %. Those with a moderate reaction ranged from 22% to 44 % with a mean value of 32.6 ± 7.79 %. Sperms with a weak reaction ranged from 18% to 35 % with a mean value of 25.4 ± 5.93 %. Sperms with a negative reaction ranged from 17% to 35 % with a mean value of 26.1 ± 6.35 %.

Table (18): Showing the percentage of sperms with different grades of *lactate dehydrogenase in the whole non-washed specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	22	50	16	12	11	20	42	25	13	21	19	44	20	17	31	12	13	30	45	41	1	5	13	81
2	21	43	16	20	12	22	49	15	14	22	21	41	18	20	32	15	24	31	30	42	2	3	10	85
3	17	36	23	24	13	23	43	17	17	23	14	26	27	33	33	14	13	36	37	43	1	11	13	75
4	28	55	10	7	14	17	35	20	28	24	22	42	18	18	34	18	23	26	33	44	7	11	19	63
5	22	36	23	19	15	26	49	14	11	25	10	36	25	29	35	16	22	19	43	45	2	14	18	66
6	17	32	29	22	16	18	53	15	14	26	9	26	34	31	36	21	21	20	38	46	5	11	21	63
7	23	52	11	14	17	18	28	29	25	27	8	22	35	35	37	10	13	26	51	47	0	4	11	85
8	22	50	10	18	18	22	31	21	26	28	21	31	25	23	38	20	24	20	36	48	3	5	12	80
9	25	51	13	11	19	21	50	16	13	29	20	26	28	26	39	12	15	33	40	49	5	14	18	63
10	18	30	24	28	20	17	38	19	26	30	15	32	24	29	40	13	14	24	49	50	4	8	15	73
Mean	21.5	43.5	17.5	17.5	Mean	20.4	41.8	19.1	18.7	Mean	15.9	32.6	25.4	26.1	Mean	15.1	18.2	26.5	40.2	Mean	3.0	8.6	15.0	73.4
S.D.	3.50	9.26	6.79	6.47	S.D.	2.95	8.60	4.84	6.70	S.D.	5.43	7.79	5.93	6.35	S.D.	3.63	4.96	5.89	6.78	S.D.	2.21	4.14	3.77	9.14

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

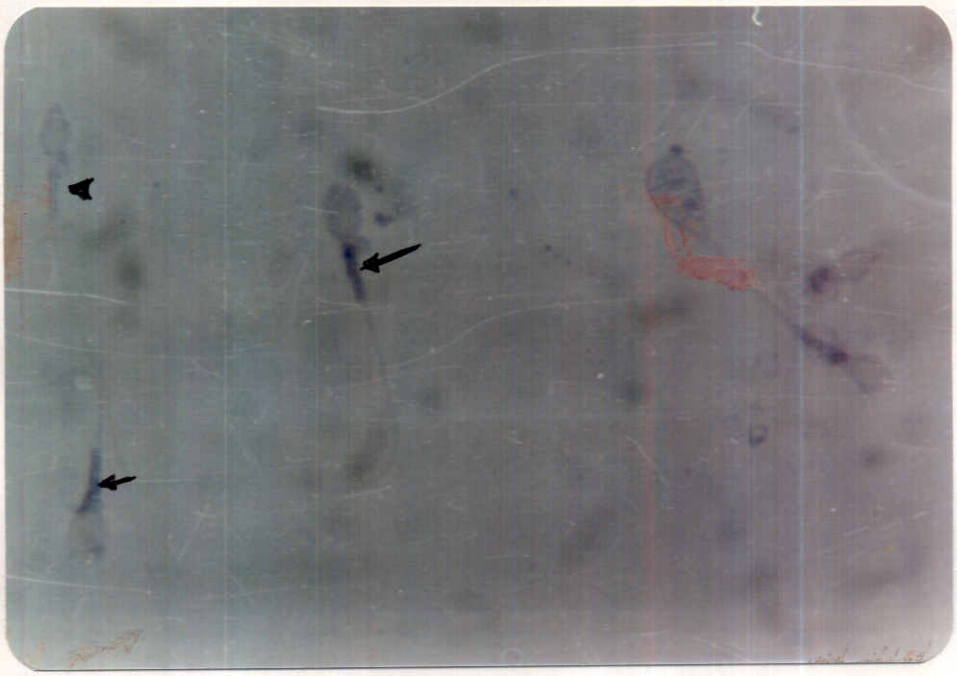

Fig. (2) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **lactate dehydrogenase.**

Note the strong (long arrow), moderate (short arrow), and weak (arrow head) reactions in the midpiece of sperms.

(NBT method. Proj: 10 Obj: 100)

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 10% to 21 % with a mean value of 15.1 ± 3.63 %. Moderately stained sperms ranged from 13% to 24 % with a mean value of 18.2 ± 4.96 %. Those with a weak reaction ranged from 19% to 36 % with a mean value of 26.5 ± 5.89 %. Sperms with a negative reaction ranged from 30% to 51 % with a mean value of 40.2 ± 6.78 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the site of lactate dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 7 % with a mean value of 3 ± 2.21 %. Those with a moderate reaction ranged from 3% to 14 % with a mean value of 8.6 ± 4.14 %. Weakly stained sperms ranged from 10% to 21 % with a mean value of 15 ± 3.77 %. Sperms with a negative reaction ranged from 63% to 85 % with a mean value of 73.4 ± 9.14 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens and the period of incubation [r = - 0.864; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens and the period of incubation [r = - 0.863; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens and the period of incubation [r = - 0.149; P <0.05].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens and the period of incubation [r = 0.943; p <0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) Washed specimens: (Table 19)

a. Immediately examined specimens:

A positive lactate dehydrogenase reaction was also observed only in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 14% to 25 % with a mean value of 18.8 ± 2.82 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined whole non-washed specimens [P <0.05] (Table 20).

Those with a moderate reaction ranged from 30% to 51 % with a mean value of 41.7 ± 8.01 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 20).

Weakly stained sperms ranged from 14% to 31 % with a mean value of 22.8 ± 5.35 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 20).

Sperms with a negative reaction ranged from 10% to 25 % with a mean value of 16.7 ± 4.69 %. They did not also differ statistically from the negatively stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P <0.05] (Table 20).

Table (19): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the washed specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	17	48	19	16	11	14	36	26	24	21	13	33	23	31	31	0	7	20	73	41	0	0	11	89
2	19	44	24	13	12	16	47	20	17	22	12	31	25	32	32	2	12	15	71	42	0	0	7	93
3	14	30	31	25	13	17	47	17	19	23	2	18	35	45	33	1	7	20	72	43	0	0	5	95
4	25	51	14	10	14	12	37	25	26	24	11	30	32	27	34	2	11	14	73	44	1	2	12	85
5	19	32	27	22	15	16	48	18	18	25	10	28	30	32	35	3	14	14	69	45	0	1	4	95
6	17	35	28	20	16	14	44	18	24	26	5	13	34	48	36	1	11	19	69	46	0	1	10	89
7	20	41	22	17	17	7	34	28	31	27	6	20	31	43	37	0	5	20	75	47	0	0	7	93
8	18	50	20	12	18	8	29	29	34	28	7	26	24	43	38	3	13	14	70	48	0	0	5	95
9	20	36	26	18	19	21	32	22	25	29	5	26	29	40	39	2	9	15	74	49	0	1	9	90
10	19	50	17	14	20	9	31	30	30	30	9	27	26	38	40	0	3	17	80	50	0	1	10	89
Mean	18.8	41.7	22.8	16.7	Mean	13.4	38.5	23.3	24.8	Mean	8.0	25.2	28.9	37.9	Mean	1.4	9.2	16.8	72.6	Mean	0.1	0.6	8.0	91.3
S.D.	2.82	8.01	5.35	4.69	S.D.	4.43	7.32	4.92	5.71	S.D.	3.56	6.30	4.23	7.03	S.D.	1.17	3.61	2.70	3.31	S.D.	0.32	0.70	2.79	3.40

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (20): Showing the comparison between the *lactate dehydrogenase* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Signific-ance.
Strong reaction	*0*	21.5 ± 3.50	18.8 ± 2.82	1.898	<0.05	-
	1 hour	20.4 ± 2.95	13.4 ± 4.43	4.160	<0.001	+++
	2 hours	15.9 ± 5.43	8.0 ± 3.56	3.850	<0.05	+
	4 hours	15.1 ± 3.63	1.4 ± 1.17	11.343	<0.001	+++
	8 hours	3.0 ± 2.21	0.1 ± 0.32	4.106	<0.001	+++
Moderate reaction	*0*	43.5 ± 9.26	41.7 ± 8.01	0.465	<0.05	-
	1 hour	41.8 ± 8.60	38.5 ± 7.32	0.924	<0.05	-
	2 hours	32.6 ± 7.79	25.2 ± 6.30	2.335	<0.05	+
	4 hours	18.2 ± 4.96	9.2 ± 3.61	4.636	<0.001	+++
	8 hours	8.6 ± 4.14	0.6 ± 0.70	6.023	<0.001	+++
Weak reaction	*0*	17.5 ± 6.79	22.8 ± 5.35	1.940	<0.05	-
	1 hour	19.1 ± 4.84	23.3 ± 4.92	1.924	<0.05	-
	2 hours	25.4 ± 5.93	28.9 ± 4.23	1.520	<0.05	-
	4 hours	26.5 ± 5.89	16.8 ± 2.70	4.733	<0.001	+++
	8 hours	15.0 ± 3.77	8.0 ± 2.79	4.719	<0.001	+++
No reaction	*0*	17.5 ± 6.47	16.7 ± 4.69	0.317	<0.05	-
	1 hour	18.7 ± 6.70	24.8 ± 5.71	2.191	<0.05	+
	2 hours	26.1 ± 6.35	37.9 ± 7.03	3.939	<0.001	+++
	4 hours	40.2 ± 6.78	72.6 ± 3.31	13.584	<0.001	+++
	8 hours	73.4 ± 9.14	91.3 ± 3.40	5.802	<0.001	+++

+++ = highly significant + = significant - = non-significant

b. Specimens examined after one hour:

A similar reaction for lactate dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 7% to 21 % with a mean value of 13.4 ± 4.43 %. This was highly significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [P <0.001] (Table 20).

Moderately stained sperms ranged from 29% to 48 % with a mean value of 38.5 ± 7.32 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 20).

Those with a weak reaction ranged from 17% to 30 % with a mean value of 23.3 ± 4.92 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 20).

Sperms with a negative reaction ranged from 17% to 34 % with a mean value of 24.8 ± 5.71 %. They were significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P <0.05] (Table 20).

c. Specimens examined after two hours:

A positive reaction was also detected only in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 2% to 13 % with a mean value of 8 ± 3.56 %. It was significantly less than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 20).

Those with a moderate reaction ranged from 13% to 33 % with a mean value of 25.2 ± 6.3 %. They were significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 20).

Sperms with a weak reaction ranged from 23% to 35 % with a mean value of 28.9 ± 4.23 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 20).

Sperms with a negative reaction ranged from 27% to 48 % with a mean value of 37.9 ± 7.03 %. This was highly significantly more than that of the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P <0.001] (Table 20).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive lactate dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1.4 ± 1.17 %. They were highly significantly less than the strongly stained sperms for in the whole non-washed specimens examined after four hours of incubation [P <0.001] (Table 20).

Moderately stained sperms ranged from 3% to 14 % with a mean value of 9.2 ± 3.61 %. They were highly significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.001] (Table 20).

Those with a weak reaction ranged from 14% to 20 % with a mean value of 16.8 ± 2.7 %. They were highly significantly less than that of the weakly stained sperms

for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.001] (Table 20).

Sperms with a negative reaction ranged from 69% to 80 % with a mean value of 72.6 ± 3.31 %. They were highly significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.001] (Table 20).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.1 ± 0.32 %. It was highly significantly less than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P <0.001] (Table 20).

Those with a moderate reaction ranged from 0% to 2 % with a mean value of 0.6 ± 0.7 %. They were highly significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.001] (Table 20).

Weakly stained sperms ranged from 4% to 12 % with a mean value of 8 ± 2.79 %. They were highly significantly less than that of the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.001] (Table 20).

Sperms with a negative reaction ranged from 85% to 95 % with a mean value of 91.3 ± 3.4 %. They were highly significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P <0.001] (Table 20).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the washed specimens and the period of incubation [r = - 0.831; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the washed specimens and the period of incubation [r = - 0.896; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the washed specimens and the period of incubation [r = - 0.759; P <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the washed specimens and the period of incubation [r = 0.954; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 21).

a. Immediately examined specimens:

A positive reaction for lactate dehydrogenase was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 14% to 26 % with a mean value of 20 ± 4.22 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P <0.05] (Table 22).

Those with a moderate reaction ranged from 35% to 54 % with a mean value of 44.3 ± 5.89 %. They did not differ statistically from the moderately stained sperms for

Table (21): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the washed specimens to which calcium was added.*

No. of Ejac-ulate	Immediate +++	++	+	-	No. of Ejac-ulate	1 hour +++	++	+	-	No. of Ejac-ulate	2 hours +++	++	+	-	No. of Ejac-ulate	4 hours +++	++	+	-	No. of Ejac-ulate	8 hours +++	++	+	-
1	22	46	22	10	11	14	40	25	21	21	18	29	23	30	31	0	7	19	74	41	1	0	12	87
2	25	47	20	8	12	22	37	18	23	22	15	37	22	26	32	4	12	16	68	42	0	0	6	94
3	15	38	32	15	13	18	51	19	12	23	6	21	33	40	33	0	8	20	72	43	0	1	13	86
4	26	50	15	9	14	16	38	27	19	24	15	29	25	31	34	2	11	23	64	44	2	5	11	82
5	17	41	23	19	15	22	45	17	16	25	12	30	27	31	35	5	16	13	66	45	0	4	10	86
6	14	35	30	21	16	17	41	28	14	26	6	24	32	38	36	4	9	25	62	46	0	3	10	87
7	20	46	23	11	17	14	34	25	27	27	10	24	31	35	37	0	7	17	76	47	0	0	5	95
8	17	47	18	18	18	12	35	28	25	28	7	27	27	39	38	3	13	15	69	48	0	1	7	92
9	24	54	12	10	19	18	52	12	18	29	10	29	25	36	39	1	15	26	58	49	1	0	7	92
10	20	39	25	16	20	12	38	24	26	30	9	31	29	31	40	0	6	21	73	50	1	1	15	83
Mean	20.0	44.3	22.0	13.7	Mean	16.5	41.1	22.3	20.1	Mean	10.8	28.1	27.4	33.7	Mean	1.9	10.4	19.5	68.2	Mean	0.5	1.5	9.6	88.4
S.D.	4.22	5.89	6.18	4.67	S.D.	3.63	6.30	5.46	5.17	S.D.	4.13	4.46	3.78	4.57	S.D.	1.97	3.53	4.33	5.75	S.D.	0.71	1.84	3.27	4.55

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (22): Showing the comparison between the *lactate dehydrogenase* reaction in the sperms of *washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incuba-tion.	Washed Specimens.	Washed Specimens + Calcium	t	p	Signific-ance.
Strong reaction	*0*	18.8 ± 2.82	20.0 ± 4.22	0.748	<0.05	-
	1 hour	13.4 ± 4.43	16.5 ± 3.63	1.713	<0.05	-
	2 hours	8.0 ± 3.56	10.8 ± 4.13	1.624	<0.05	-
	4 hours	1.4 ± 1.17	1.9 ± 1.97	0.690	<0.05	-
	8 hours	0.1 ± 0.32	0.5 ± 0.71	1.633	<0.05	-
Moderate reaction	*0*	41.7 ± 8.01	44.3 ± 5.89	0.827	<0.05	-
	1 hour	38.5 ± 7.32	41.1 ± 6.30	0.851	<0.05	-
	2 hours	25.2 ± 6.30	28.1 ± 4.46	1.188	<0.05	-
	4 hours	9.2 ± 3.61	10.4 ± 3.53	0.751	<0.05	-
	8 hours	0.6 ± 0.70	1.5 ± 1.84	1.445	<0.05	-
Weak reaction	*0*	22.8 ± 5.35	22.0 ± 6.18	0.309	<0.05	-
	1 hour	23.3 ± 4.92	22.3 ± 5.46	0.430	<0.05	-
	2 hours	28.9 ± 4.23	27.4 ± 3.78	0.837	<0.05	-
	4 hours	16.8 ± 2.70	19.5 ± 4.33	1.674	<0.05	-
	8 hours	8.0 ± 2.79	9.6 ± 3.27	1.177	<0.05	-
No reaction	*0*	16.7 ± 4.69	13.7 ± 4.67	1.434	<0.05	-
	1 hour	24.8 ± 5.71	20.1 ± 5.17	1.929	<0.05	-
	2 hours	37.9 ± 7.03	33.7 ± 4.57	1.584	<0.05	-
	4 hours	72.6 ± 3.31	68.2 ± 5.75	2.097	<0.05	-
	8 hours	91.3 ± 3.40	88.4 ± 4.55	1.614	<0.05	-

- = non-significant

lactate dehydrogenase in the immediately examined washed specimens [$P < 0.05$] (Table 22).

Weakly stained sperms ranged from 12% to 32 % with a mean value of 22 ± 6.18 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined washed specimens [$P < 0.05$] (Table 22).

Sperms with a negative reaction ranged from 8% to 21 % with a mean value of 13.7 ± 4.67 %. They did not also differ statistically from the negatively stained sperms for lactate dehydrogenase in the immediately examined washed specimens [$P < 0.05$] (Table 22).

b. Specimens examined after one hour:

A similar location of reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 12% to 22 % with a mean value of 16.5 ± 3.63 %. This did not differ statistically from that of the strongly stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 22).

Moderately stained sperms ranged from 34% to 52 % with a mean value of 41.1 ± 6.3 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 22).

Those with a weak reaction ranged from 12% to 28 % with a mean value of 22.3 ± 5.46 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 22).

Sperms with a negative reaction ranged from 12% to 27 % with a mean value of 20.1 ± 5.17 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [P <0.05] (Table 22).

c. Specimens examined after two hours:

A positive lactate dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 6% to 18 % with a mean value of 10.8 ± 4.13 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after two hours of incubation [P <0.05] (Table 22).

Those with a moderate reaction ranged from 21% to 37 % with a mean value of 28.1 ± 4.46 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 22).

Sperms with a weak reaction ranged from 22% to 33 % with a mean value of 27.4 ± 3.78 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 22).

Sperms with a negative reaction ranged from 26% to 40 % with a mean value of 33.7 ± 4.57 %. This did not differ statistically from that of the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after two hours of incubation [P <0.05] (Table 22).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 5 % with a mean value of

1.9 ± 1.97 %. They did not differ statistically from the strongly stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [P <0.05] (Table 22).

Moderately stained sperms ranged from 6% to 16 % with a mean value of 10.4 ± 3.53 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [P < 0.05] (Table 22).

Those with a weak reaction ranged from 13% to 26 % with a mean value of 19.5 ± 4.33 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimen examined after four hours of incubation [P < 0.05] (Table 22).

Sperms with a negative reaction ranged from 58% to 76 % with a mean value of 68.2 ± 5.75 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [P <0.05] (Table 22).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of lactate dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 2 % with a mean value of 0.5 ± 0.71 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [P <0.05] (Table 22).

Those with a moderate reaction ranged from 0% to 5 % with a mean value of 1.5 ± 1.84 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [P < 0.05] (Table 22).

Weakly stained sperms ranged from 5% to 15 % with a mean value of 9.6 ± 3.27 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 22).

Sperms with a negative reaction ranged from 82% to 95 % with a mean value of 88.4 ± 4.55 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 22).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the washed specimens to which calcium was added and the period of incubation [$r = - 0.845$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the washed specimens to which calcium was added and the period of incubation [$r = - 0.918$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the washed specimens to which calcium was added and the period of incubation [$r = - 0.759$; $P < 0.001$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the washed specimens to which calcium was added and the period of incubation [$r = 0.957$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(4) Washed specimens to which ascorbic acid was added: (Table 23)

a. Immediately examined specimens:

A positive lactate dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 13% to 24 % with a mean value of 19.3 ± 3.89 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P <0.05] (Table 24).

Those with a moderate reaction ranged from 32% to 53 % with a mean value of 42.1 ± 8.1 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 24).

Weakly stained sperms ranged from 14% to 34 % with a mean value of 25.3 ± 6.07 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 24).

Sperms with a negative reaction ranged from 5% to 20 % with a mean value of 13.3 ± 4.74 %. They did not also differ statistically from the negatively stained sperms for lactate dehydrogenase in the immediately examined washed specimens [P <0.05] (Table 24).

b. Specimens examined after one hour:

A similar reaction for lactate dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 20 % with a mean value of 15.6 ± 2.88 %. Also, this did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [P <0.05] (Table 24).

Table (23): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the washed specimens to which ascorbic acid was added.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	23	35	25	17	11	13	38	29	20	21	19	32	26	23	31	3	13	14	70	41	1	3	9	87
2	24	53	14	9	12	16	45	22	17	22	21	32	23	24	32	7	17	18	58	42	0	1	5	94
3	13	37	30	20	13	20	41	19	20	23	8	22	32	38	33	4	15	12	69	43	1	3	10	86
4	21	47	19	13	14	17	30	30	23	24	19	32	29	20	34	3	11	20	66	44	2	8	14	76
5	15	34	33	18	15	16	49	22	13	25	16	29	33	22	35	5	19	20	56	45	1	5	13	81
6	17	32	34	17	16	18	37	24	21	26	9	20	34	37	36	6	16	11	67	46	2	7	12	79
7	20	52	23	5	17	15	30	30	25	27	8	23	34	35	37	5	14	10	71	47	0	2	12	86
8	16	51	24	9	18	12	30	35	23	28	10	30	18	42	38	6	17	21	56	48	0	2	7	91
9	24	37	26	13	19	18	34	28	20	29	9	29	31	31	39	7	15	21	57	49	1	5	13	81
10	20	43	25	12	20	11	33	30	26	30	13	28	27	32	40	1	13	19	67	50	2	6	9	83
Mean	19.3	42.1	25.3	13.3	Mean	15.6	36.7	26.9	20.8	Mean	13.2	27.7	28.7	30.4	Mean	4.7	15.0	16.6	63.7	Mean	1.0	4.2	10.4	84.4
S.D.	3.89	8.10	6.07	4.74	S.D.	2.88	6.63	4.93	3.82	S.D.	5.12	4.45	5.25	7.71	S.D.	1.95	2.36	4.38	6.18	S.D.	0.82	2.35	2.91	5.50

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (24): Showing the comparison between the *lactate dehydrogenase* reaction in the sperms of *washed specimens* and *washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incuba-tion.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Signific-ance.
Strong reaction	*0*	18.8 ± 2.82	19.3 ± 3.89	0.329	<0.05	-
	1 hour	13.4 ± 4.43	15.6 ± 2.88	1.318	<0.05	-
	2 hours	8.0 ± 3.56	13.2 ± 5.12	2.638	<0.05	+
	4 hours	1.4 ± 1.17	4.7 ± 1.95	4.591	<0.001	+++
	8 hours	0.1 ± 0.32	1.0 ± 0.82	3.250	<0.05	+
Moderate reaction	*0*	41.7 ± 8.01	42.1 ± 8.10	0.111	<0.05	-
	1 hour	38.5 ± 7.32	36.7 ± 6.63	0.576	<0.05	-
	2 hours	25.2 ± 6.30	27.7 ± 4.45	1.025	<0.05	-
	4 hours	9.2 ± 3.61	15.0 ± 2.36	4.250	<0.001	+++
	8 hours	0.6 ± 0.70	4.2 ± 2.35	4.648	<0.001	+++
Weak reaction	*0*	22.8 ± 5.35	25.3 ± 6.07	0.977	<0.05	-
	1 hour	23.3 ± 4.92	26.9 ± 4.93	1.634	<0.05	-
	2 hours	28.9 ± 4.23	28.7 ± 5.25	0.094	<0.05	-
	4 hours	16.8 ± 2.70	16.6 ± 4.38	0.123	<0.05	-
	8 hours	8.0 ± 2.79	10.4 ± 2.91	1.882	<0.05	-
No reaction	*0*	16.7 ± 4.69	13.3 ± 4.74	1.612	<0.05	-
	1 hour	24.8 ± 5.71	20.8 ± 3.82	1.840	<0.05	-
	2 hours	37.9 ± 7.03	30.4 ± 7.71	2.274	<0.05	+
	4 hours	72.6 ± 3.31	63.7 ± 6.18	4.014	<0.05	+
	8 hours	91.3 ± 3.40	84.4 ± 5.50	3.374	<0.05	+

+++ = highly significant + = significant - = non-significant

Moderately stained sperms ranged from 30% to 49 % with a mean value of 36.7 ± 6.63 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [P < 0.05] (Table 24).

Those with a weak reaction ranged from 19% to 35 % with a mean value of 26.9 ± 4.93 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [P < 0.05] (Table 24).

Sperms with a negative reaction ranged from 13% to 26 % with a mean value of 20.8 ± 3.82 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after one hour of incubation [P <0.05] (Table 24).

c. Specimens examined after two hours:

A similar location of lactate dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 8% to 21 % with a mean value of 13.2 ± 5.12 %. It was significantly more than that of the strongly stained sperms in the washed specimens examined after two hours of incubation [P <0.05] (Table 24).

Those with a moderate reaction ranged from 20% to 32 % with a mean value of 27.7 ± 4.45 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 24).

Sperms with a weak reaction ranged from 18% to 34 % with a mean value of 28.7 ± 5.25 %. They did not also differ statistically from the weakly stained sperms for

lactate dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 24).

Sperms with a negative reaction ranged from 20% to 42 % with a mean value of 30.4 ± 7.71 %. They was significantly less than the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 24).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 1% to 7 % with a mean value of 4.7 ± 1.95 %. This was highly significantly more than that of the strongly stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.001$] (Table 24).

Moderately stained sperms ranged from 11% to 19 % with a mean value of 15 ± 2.36 %. They were highly significantly more than that of the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.001$] (Table 24).

Those with a weak reaction ranged from 10% to 21 % with a mean value of 16.6 ± 4.38 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 24).

Sperms with a negative reaction ranged from 56% to 71 % with a mean value of 63.7 ± 6.18 %. This was significantly less than that of the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 24).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of lactate dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 2 % with a mean value of 1 ± 0.82 %. It was significantly more than that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [P <0.05] (Table 24).

Those with a moderate reaction ranged from 1% to 8 % with a mean value of 4.2 ± 2.35 %. They were highly significantly more than that of the moderately stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [P < 0.001] (Table 24).

Weakly stained sperms ranged from 5 % to 14 % with a mean value of 10.4 ± 2.91 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [P < 0.05] (Table 24).

Sperms with a negative reaction ranged from 76% to 94 % with a mean value of 84.4 ± 5.5 %. They were significantly less than the negatively stained sperms for lactate dehydrogenase in the washed specimens examined after eight hours of incubation [P <0.05] (Table 24).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [r = - 0.871; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the washed specimens to which

ascorbic acid was added and the period of incubation [r = - 0.914; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [r = - 0.762; P <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [r = 0.956; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

B. SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 25)

a. Immediately examined specimens:

Also, only the mid-piece was the site of positive lactate dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 18% to 33 % with a mean value of 24.5 ± 4.6 %. They did not differ statistically from the strongly stained sperms in the whole non-washed specimens examined immediately [P <0.05] (Table 26).

Moderately stained sperms ranged from 42% to 56 % with a mean value of 48 ± 5.58 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 26).

Those with a weak reaction ranged from 7% to 21 % with a mean value of 14.2 ± 4.64 %. They did not also differ statistically from the weakly stained sperms for

Table (25a): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the first split fraction specimens.*

No. of Ejac-ulate	Immediate				No. of Ejac-ulate	1 hour				No. of Ejac-ulate	2 hours				No. of Ejac-ulate	4 hours				No. of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	22	51	14	13	61	20	41	24	15	71	23	42	21	14	81	23	32	23	22	91	7	15	21	57
52	23	48	14	15	62	23	54	16	7	72	21	49	17	13	82	15	19	21	45	92	4	13	11	72
53	30	56	9	5	63	26	44	21	9	73	20	36	24	20	83	20	29	26	25	93	6	13	19	62
54	33	43	13	11	64	28	45	21	6	74	19	35	27	19	84	19	25	25	31	94	8	22	14	56
55	18	42	19	21	65	28	58	9	5	75	25	42	20	13	85	18	36	27	19	95	3	11	12	74
56	26	55	7	12	66	21	40	26	13	76	15	47	22	16	86	18	23	21	38	96	4	12	13	71
57	19	44	20	17	67	23	46	17	14	77	24	53	13	10	87	19	28	26	27	97	8	17	21	54
58	23	42	21	14	68	20	35	31	14	78	17	38	24	21	88	16	21	22	41	98	3	7	18	72
59	26	54	11	9	69	24	55	14	7	79	18	45	20	17	89	18	24	21	37	99	3	6	18	73
60	25	45	14	16	70	25	46	17	12	70	24	32	20	24	80	21	35	26	18	100	7	13	19	61
Mean	24.5	48.0	14.2	13.3	Mean	23.8	46.4	19.6	10.2	Mean	20.6	41.9	20.8	16.7	Mean	18.7	27.2	23.8	30.3	Mean	5.3	12.9	16.6	65.2
S.D.	4.60	5.58	4.64	4.45	S.D.	2.97	7.26	6.36	3.79	S.D.	3.37	6.71	3.91	4.32	S.D.	2.31	5.81	2.44	9.56	S.D.	2.11	4.61	3.75	7.96

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (26): Showing the comparison between the *lactate dehydrogenase reaction in the sperms of whole non-washed and first split fraction specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Signific-ance.
Strong reaction	*0*	21.5 ± 3.50	24.5 ± 4.60	1.640	<0.05	-
	1 hour	20.4 ± 2.95	23.8 ± 2.97	2.566	<0.05	+
	2 hours	15.9 ± 5.43	20.6 ± 3.37	2.327	<0.05	+
	4 hours	15.1 ± 3.63	18.7 ± 2.31	2.643	<0.05	+
	8 hours	3.0 ± 2.21	5.3 ± 2.11	2.379	<0.05	+
Moderate reaction	*0*	43.5 ± 9.26	48.0 ± 5.58	1.316	<0.05	-
	1 hour	41.8 ± 8.60	46.4 ± 7.26	1.293	<0.05	-
	2 hours	32.6 ± 7.79	41.9 ± 6.71	2.861	<0.05	+
	4 hours	18.2 ± 4.96	27.2 ± 5.81	3.726	<0.05	+
	8 hours	8.6 ± 4.14	12.9 ± 4.61	2.195	<0.05	+
Weak reaction	*0*	17.5 ± 6.79	14.2 ± 4.64	1.277	<0.05	-
	1 hour	19.1 ± 4.84	19.6 ± 6.36	0.198	<0.05	-
	2 hours	25.4 ± 5.93	20.8 ± 3.91	2.048	<0.05	-
	4 hours	26.5 ± 5.89	23.8 ± 2.44	1.339	<0.05	-
	8 hours	15.0 ± 3.77	16.6 ± 3.75	0.952	<0.05	-
No reaction	*0*	17.5 ± 6.47	13.3 ± 4.45	1.692	<0.05	-
	1 hour	18.7 ± 6.70	10.2 ± 3.79	3.491	<0.05	+
	2 hours	26.1 ± 6.35	16.7 ± 4.32	3.870	<0.05	+
	4 hours	40.2 ± 6.78	30.3 ± 9.56	2.672	<0.05	+
	8 hours	73.4 ± 9.14	65.2 ± 7.96	2.140	<0.05	+

+ = significant - = non-significant

lactate dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 26).

Sperms with a negative reaction ranged from 5% to 21 % with a mean value of 13.3 ± 4.45 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 26).

b. Specimens examined after one hour:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 20% to 28 % with a mean value of 23.8 ± 2.97 %. It was significantly more than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 26).

Those with a moderate reaction ranged from 35% to 58 % with a mean value of 46.4 ± 7.26 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 26).

Weakly stained sperms ranged from 9% to 31 % with a mean value of 19.6 ± 6.36 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 26).

Sperms with a negative reaction ranged from 5% to 15 % with a mean value of 10.2 ± 3.79 %. They were significantly less than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 26).

c. Specimens examined after two hours:

A similar location of lactate dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 15% to 25 % with a mean value of 20.6 ± 3.37 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 26).

Those with a moderate reaction ranged from 32% to 53 % with a mean value of 41.9 ± 6.71 %. They were significantly more than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 26).

Sperms with a weak reaction ranged from 13% to 27 % with a mean value of 20.8 ± 3.91 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 26).

Sperms with a negative reaction ranged from 10% to 24 % with a mean value of 16.7 ± 4.32 %. This was significantly less than that of negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 26).

d. Specimens examined after four hours:

A positive reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 15% to 23 % with a mean value of 18.7 ± 2.31 %. It was significantly more than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 26).

Those with a moderate reaction ranged from 19% to 36 % with a mean value of 27.2 ± 5.81 %. They were significantly more than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 26).

Weakly stained sperms ranged from 21% to 27 % with a mean value of 23.8 ± 2.44 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 26).

Sperms with a negative reaction ranged from 18% to 45 % with a mean value of 30.3 ± 9.56 %. They were significantly less than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.05] (Table 26).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of lactate dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 3% to 8 % with a mean value of 5.3 ± 2.11 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [P <0.05] (Table 26).

Those with a moderate reaction ranged from 6% to 22 % with a mean value of 12.9 ± 4.61 %. They were significantly more than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 26).

Weakly stained sperms ranged from 11% to 21 % with a mean value of 16.6 ± 3.75 %. They did not differ statistically from the weakly stained sperms for lactate

dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 26).

Sperms with a negative reaction ranged from 54% to 74 % with a mean value of 65.2 ± 7.96 %. They was significantly less than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after 8 hours of incubation [$P < 0.05$] (Table 26).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the first split fraction specimens and the period of incubation [$r = - 0.898$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens and the period of incubation [$r = - 0.908$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant positive correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the first split fraction specimens and the period of incubation [$r = 0.056$; $P < 0.05$].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the first split fraction specimens and the period of incubation [$r = 0.935$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added: (Table 27)

a. Immediately examined specimens:

A positive reaction for lactate dehydrogenase was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 18% to 30 % with a mean value of 23.2 ± 3.52 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined first split fraction specimens [P <0.05] (Table 28).

Those with a moderate reaction ranged from 34% to 55 % with a mean value of 46.2 ± 7.67 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined first split fraction specimens [P < 0.05] (Table 28).

Weakly stained sperms ranged from 9% to 27 % with a mean value of 15.6 ± 5.91 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined first split fraction specimens [P < 0.05] (Table 28).

Sperms with a negative reaction ranged from 9% to 22 % with a mean value of 15 ± 4.59 %. They did not also differ statistically from the negatively stained sperms for lactate dehydrogenase in the immediately examined first split fraction specimens [P <0.05] (Table 28).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 19% to 29 % with a mean value of 24.4 ± 3.27 %. Also, this did not differ statistically from that of the strongly stained sperms for lactate dehydrogenase in the first split fraction specimens examined after one hour of incubation [P <0.05] (Table 28).

Table (27): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the first split fraction specimens to which ascorbic acid was added.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	23	39	18	20	61	21	36	26	17	71	27	44	15	14	81	24	34	26	16	91	6	11	22	61
52	23	47	16	14	62	26	51	15	8	72	28	38	22	12	82	20	30	20	30	92	7	13	16	64
53	24	55	11	10	63	23	46	22	9	73	17	29	32	22	83	18	31	28	23	93	9	13	21	57
54	27	51	9	13	64	28	38	23	11	74	16	39	25	20	84	22	41	21	16	94	10	17	27	46
55	21	53	13	13	65	26	53	16	5	75	21	41	24	14	85	25	44	15	16	95	8	11	22	59
56	23	52	13	12	66	19	37	31	13	76	25	36	24	15	86	21	34	22	23	96	9	15	21	55
57	18	34	27	21	67	22	36	28	14	77	26	41	23	10	87	24	32	23	21	97	12	18	13	57
58	19	36	23	22	68	23	30	30	17	78	18	43	21	18	88	18	22	31	29	98	4	6	22	68
59	30	52	9	9	69	29	48	16	7	79	27	45	16	12	89	21	23	28	28	99	7	12	23	58
60	24	43	17	16	70	27	39	24	10	70	19	32	32	17	80	26	43	17	14	100	5	10	22	63
Mean	23.2	46.2	15.6	15.0	Mean	24.4	41.4	23.1	11.1	Mean	22.4	38.8	23.4	15.4	Mean	21.9	33.4	23.1	21.6	Mean	7.7	12.6	20.9	58.8
S.D.	3.52	7.67	5.91	4.59	S.D.	3.27	7.57	5.88	4.09	S.D.	4.67	5.20	5.62	3.81	S.D.	2.81	7.60	5.13	5.99	S.D.	2.41	3.50	3.84	5.96

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (28): Showing the comparison between the *lactate dehydrogenase* reaction in the sperms of *first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incuba-tion.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	24.5 ± 4.60	23.2 ± 3.52	0.710	<0.05	-
	1 hour	23.8 ± 2.97	24.4 ± 3.27	0.429	<0.05	-
	2 hours	20.6 ± 3.37	22.4 ± 4.67	0.988	<0.05	-
	4 hours	18.7 ± 2.31	21.9 ± 2.81	2.783	<0.05	+
	8 hours	5.3 ± 2.11	7.7 ± 2.41	2.371	<0.05	+
Moderate reaction	*0*	48.0 ± 5.58	46.2 ± 7.67	0.600	<0.05	-
	1 hour	46.4 ± 7.26	41.4 ± 7.57	1.507	<0.05	-
	2 hours	41.9 ± 6.71	38.8 ± 5.20	1.155	<0.05	-
	4 hours	27.2 ± 5.81	33.4 ± 7.60	2.049	<0.05	-
	8 hours	12.9 ± 4.61	12.6 ± 3.50	0.164	<0.05	-
Weak reaction	*0*	14.2 ± 4.64	15.6 ± 5.91	0.589	<0.05	-
	1 hour	19.6 ± 6.36	23.1 ± 5.88	1.278	<0.05	-
	2 hours	20.8 ± 3.91	23.4 ± 5.62	1.201	<0.05	-
	4 hours	23.8 ± 2.44	23.1 ± 5.13	0.390	<0.05	-
	8 hours	16.6 ± 3.75	20.9 ± 3.84	2.533	<0.05	+
No reaction	*0*	13.3 ± 4.45	15.0 ± 4.59	0.841	<0.05	-
	1 hour	10.2 ± 3.79	11.1 ± 4.09	0.510	<0.05	-
	2 hours	16.7 ± 4.32	15.4 ± 3.81	0.714	<0.05	-
	4 hours	30.3 ± 9.56	21.6 ± 5.99	2.440	<0.05	+
	8 hours	65.2 ± 7.96	58.8 ± 5.96	2.036	<0.05	+

+ = significant +++ = highly significant

Moderately stained sperms ranged from 30% to 53 % with a mean value of 41.4 ± 7.57 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens examined after one hour of incubation [P < 0.05] (Table 28).

Those with a weak reaction ranged from 15% to 31 % with a mean value of 23.1 ± 5.88 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the first split fraction specimens examined after one hour of incubation [P < 0.05] (Table 28).

Sperms with a negative reaction ranged from 5% to 17 % with a mean value of 11.1 ± 4.09 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the first split fraction specimens examined after 1 hour of incubation [P <0.05] (Table 28).

c. Specimens examined after two hours:

A similar location of lactate dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 16% to 28 % with a mean value of 22.4 ± 4.67 %. Also, it did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after two hours of incubation [P <0.05] (Table 28).

Those with a moderate reaction ranged from 29% to 45 % with a mean value of 38.8 ± 5.2 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens examined after two hours of incubation [P < 0.05] (Table 28).

Sperms with a weak reaction ranged from 15% to 32 % with a mean value of 23.4 ± 5.62 %. They did not also differ statistically from the weakly stained sperms for

lactate dehydrogenase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 28).

Sperms with a negative reaction ranged from 10% to 22 % with a mean value of 15.4 ± 3.81 %. They did not differ statistically from the negatively stained sperms for lactate dehydrogenase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 28).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 18% to 26 % with a mean value of 21.9 ± 2.81 %. This was significantly more than that of the strongly stained sperms for lactate dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 28).

Moderately stained sperms ranged from 22% to 44 % with a mean value of 33.4 ± 7.6 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 28).

Those with a weak reaction ranged from 15% to 31 % with a mean value of 23.1 ± 5.13 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 28).

Sperms with a negative reaction ranged from 14% to 30 % with a mean value of 21.6 ± 5.99 %. This was significantly less than that of negatively stained sperms for lactate dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 28).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of lactate dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 4% to 12 % with a mean value of 7.7 ± 2.41 %. It was significantly more than that of the strongly stained sperms in the first split fraction specimens examined after eight hours of incubation [P <0.05] (Table 28).

Those with a moderate reaction ranged from 6% to 18 % with a mean value of 12.6 ± 3.5 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens examined after eight hours of incubation [P < 0.05] (Table 28).

Weakly stained sperms ranged from 13% to 27 % with a mean value of 20.9 ± 3.84 %. They were significantly more than that of the weakly stained sperms for lactate dehydrogenase in the first split fraction specimens examined after eight hours of incubation [P < 0.05] (Table 28).

Sperms with a negative reaction ranged from 46% to 68 % with a mean value of 58.8 ± 5.96 %. They were significantly less than the negatively stained sperms for lactate dehydrogenase in the first split fraction specimens examined after eight hours of incubation [P <0.05] (Table 28).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = - 0.816; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the first split fraction specimens to

which ascorbic acid was added and the period of incubation [r = - 0.875; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant positive correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = 0.135; p <0.05].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = 0.907; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 29)

a. Immediately examined specimens:

Also, only the mid-piece was the site of positive reaction for lactate dehydrogenase. The percentage of spermatozoa giving a strong reaction ranged from 12% to 21 % with a mean value of 17.8 ± 3.29 %. They were significantly less than the strongly stained sperms in the immediately examined whole non-washed specimens [P <0.05] (Table 30).

Moderately stained sperms ranged from 29% to 48 % with a mean value of 37.8 ± 7.11 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 30).

Those with a weak reaction ranged from 13% to 34 % with a mean value of 22.1 ± 7.4 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 30).

Table (29): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the second split fraction specimens.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
51	21	46	13	20	61	15	39	21	25	71	7	12	25	56	81	6	11	26	57	91	0	3	4	93
52	18	35	24	23	62	12	29	22	37	72	11	18	23	48	82	14	13	38	35	92	1	4	2	93
53	12	29	32	27	63	13	31	26	30	73	16	21	25	38	83	10	12	32	46	93	1	5	8	86
54	13	29	34	24	64	8	25	24	43	74	17	28	20	35	84	4	8	25	63	94	0	1	2	97
55	21	40	17	22	65	8	28	25	39	75	6	20	22	52	85	6	12	26	56	95	1	7	9	83
56	20	34	22	24	66	13	32	24	31	76	8	17	18	57	86	9	10	30	51	96	2	7	11	80
57	19	46	15	20	67	21	37	14	28	77	9	15	25	51	87	2	8	27	63	97	0	1	1	98
58	21	48	14	17	68	24	39	17	20	78	10	25	24	41	88	12	11	36	41	98	1	6	9	84
59	17	39	23	21	69	7	22	23	48	79	12	23	23	42	89	13	12	37	38	99	0	4	3	93
60	16	32	27	25	70	14	38	18	30	70	13	20	22	45	80	5	10	24	61	100	0	5	10	85
Mean	17.8	37.8	22.1	22.3	Mean	13.5	32.0	21.4	33.1	Mean	10.9	19.9	22.7	46.5	Mean	8.1	10.7	30.1	51.1	Mean	0.6	4.3	5.9	89.2
S.D.	3.29	7.11	7.40	2.91	S.D.	5.52	6.09	3.89	8.54	S.D.	3.67	4.72	2.31	7.53	S.D.	4.09	1.70	5.32	10.54	S.D.	0.70	2.16	3.84	6.32

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (30): Showing the comparison between the *lactate dehydrogenase* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Signific-ance.
Strong reaction	*0*	21.5 ± 3.50	17.8 ± 3.29	2.433	<0.05	+
	1 hour	20.4 ± 2.95	13.5 ± 5.52	3.485	<0.05	+
	2 hours	15.9 ± 5.43	10.9 ± 3.67	2.415	<0.05	+
	4 hours	15.1 ± 3.63	8.1 ± 4.09	4.043	<0.001	+++
	8 hours	3.0 ± 2.21	0.6 ± 0.70	3.273	<0.05	+
Moderate reaction	*0*	43.5 ± 9.26	37.8 ± 7.11	1.543	<0.05	-
	1 hour	41.8 ± 8.60	32.0 ± 6.09	2.941	<0.05	+
	2 hours	32.6 ± 7.79	19.9 ± 4.72	4.407	<0.001	+++
	4 hours	18.2 ± 4.96	10.7 ± 1.70	4.521	<0.001	+++
	8 hours	8.6 ± 4.14	4.3 ± 2.16	2.910	<0.05	+
Weak reaction	*0*	17.5 ± 6.79	22.1 ± 7.40	1.449	<0.05	-
	1 hour	19.1 ± 4.84	21.4 ± 3.89	1.171	<0.05	-
	2 hours	25.4 ± 5.93	22.7 ± 2.31	1.342	<0.05	-
	4 hours	26.5 ± 5.89	30.1 ± 5.32	1.434	<0.05	-
	8 hours	15.0 ± 3.77	5.9 ± 3.84	5.345	<0.001	+++
No reaction	*0*	17.5 ± 6.47	22.3 ± 2.91	2.141	<0.05	+
	1 hour	18.7 ± 6.70	33.1 ± 8.54	4.194	<0.001	+++
	2 hours	26.1 ± 6.35	46.5 ± 7.53	6.549	<0.001	+++
	4 hours	40.2 ± 6.78	51.1 ± 10.54	2.752	<0.05	+
	8 hours	73.4 ± 9.14	89.2 ± 6.32	4.495	<0.001	+++

+++ = highly significant + = significant - = non-significant

Sperms with a negative reaction ranged from 17% to 27 % with a mean value of 22.3 ± 2.91 %. They were significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined immediately [P <0.05] (Table 30).

b. Specimens examined after one hour:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 7% to 24 % with a mean value of 13.5 ± 5.52 %. It was significantly less than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P <0.05] (Table 30).

Those with a moderate reaction ranged from 22% to 39 % with a mean value of 32 ± 6.09 %. They were significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 30).

Weakly stained sperms ranged from 14% to 26 % with a mean value of 21.4 ± 3.89 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 30).

Sperms with a negative reaction ranged from 20% to 48 % with a mean value of 33.1 ± 8.54 %. They were highly significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P <0.001] (Table 30).

c. Specimens examined after two hours:

A positive lactate dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 6% to 17 % with a mean value of 10.9 ± 3.67 %. It was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 30).

Those with a moderate reaction ranged from 12% to 28 % with a mean value of 19.9 ± 4.72 %. They were highly significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P < 0.001] (Table 30).

Sperms with a weak reaction ranged from 18% to 25 % with a mean value of 22.7 ± 2.31 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 30).

Sperms with a negative reaction ranged from 35% to 57 % with a mean value of 46.5 ± 7.53 %. This was highly significantly more than that of negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P <0.001] (Table 30).

d. Specimens examined after four hours:

A similar location of reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 2% to 14 % with a mean value of 8.1 ± 4.09 %. It was highly significantly less than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.001] (Table 30).

Those with a moderate reaction ranged from 8% to 13 % with a mean value of 10.7 ± 1.7 %. They were highly significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 30).

Weakly stained sperms ranged from 24% to 38 % with a mean value of 30.1 ± 5.32 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 30).

Sperms with a negative reaction ranged from 35% to 63 % with a mean value of 51.1 ± 10.54 %. They were significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 30).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 2 % with a mean value of 0.6 ± 0.7 %. It was significantly less than that of the strongly stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 30).

Those with a moderate reaction ranged from 1% to 7 % with a mean value of 4.3 ± 2.16 %. They were significantly less than that of the moderately stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 30).

Weakly stained sperms ranged from 1% to 11 % with a mean value of 5.9 ± 3.84 %. They were highly significantly less than that of the weakly stained sperms

for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 30).

Sperms with a negative reaction ranged from 80% to 98 % with a mean value of 89.2 ± 6.32 %. They were highly significantly more than the negatively stained sperms for lactate dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 30).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the second split fraction specimens and the period of incubation [$r = - 0.835$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens and the period of incubation [$r = - 0.872$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the second split fraction specimens and the period of incubation [$r = - 0.557$; $P < 0.001$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the second split fraction specimens and the period of incubation [$r = 0.940$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added: (Table 31)

a. Immediately examined specimens:

A positive reaction for lactate dehydrogenase was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 12% to 27 % with a mean value of 19 ± 4.69 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined second split fraction specimens [P <0.05] (Table 32).

Those with a moderate reaction ranged from 27% to 47 % with a mean value of 36.8 ± 6.86 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the immediately examined second split fraction specimens [P < 0.05] (Table 32).

Weakly stained sperms ranged from 11% to 29 % with a mean value of 19.9 ± 5.0 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the immediately examined second split fraction specimens [P < 0.05] (Table 32).

Sperms with a negative reaction ranged from 16% to 32 % with a mean value of 24.3 ± 5.66 %. They did not also differ statistically from the negatively stained sperms for lactate dehydrogenase in the immediately examined second split fraction specimens [P <0.05] (Table 32).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 24 % with a mean value of 17 ± 4.27 %. It did not differ statistically from that of the strongly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after one hour of incubation [P <0.05] (Table 32).

Table (31): Showing the percentage of sperms with different grades of *lactate dehydrogenase reaction in the second split fraction specimens to which calcium was added.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
51	18	39	22	21	61	17	34	24	25	71	10	19	25	46	81	6	17	32	45	91	0	6	14	80
52	20	32	20	28	62	11	23	32	34	72	11	23	21	45	82	18	21	25	36	92	2	9	14	75
53	16	31	22	31	63	15	28	27	30	73	21	31	20	28	83	16	17	33	34	93	2	12	13	73
54	12	27	29	32	64	22	39	20	19	74	22	35	19	24	84	10	17	24	49	94	1	6	8	85
55	21	46	13	20	65	15	33	26	26	75	18	28	21	33	85	11	13	23	53	95	3	13	13	71
56	27	35	19	19	66	18	36	24	22	76	10	19	26	45	86	9	14	32	45	96	2	11	19	68
57	17	41	21	21	67	24	37	19	20	77	13	26	17	44	87	7	12	32	49	97	1	6	9	84
58	26	47	11	16	68	21	45	17	17	78	21	37	16	26	88	17	23	22	38	98	1	12	20	67
59	15	40	20	25	69	12	27	30	31	79	16	27	21	36	89	11	22	30	37	99	2	7	7	84
60	18	30	22	30	70	15	31	27	27	70	15	26	19	40	80	16	15	33	36	100	1	9	8	82
Mean	19.0	36.8	19.9	24.3	Mean	17.0	33.3	24.6	25.1	Mean	15.7	27.1	20.5	36.7	Mean	12.1	17.1	28.6	42.2	Mean	1.5	9.1	12.5	76.9
S.D.	4.69	6.86	5.00	5.66	S.D.	4.27	6.41	4.81	5.59	S.D.	4.67	6.03	3.14	8.53	S.D.	4.33	3.81	4.53	6.78	S.D.	0.85	2.77	4.55	6.94

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (32): Showing the comparison between the *lactate dehydrogenase reaction in the sperms of second split fraction specimens and second split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incuba-tion.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Significance.
Strong reaction	*0*	17.8 ± 3.29	19.0 ± 4.69	0.662	<0.05	-
	1 hour	13.5 ± 5.52	17.0 ± 4.27	1.586	<0.05	-
	2 hours	10.9 ± 3.67	15.7 ± 4.67	2.558	<0.05	+
	4 hours	8.1 ± 4.09	12.1 ± 4.33	2.122	<0.05	+
	8 hours	0.6 ± 0.70	1.5 ± 0.85	2.586	<0.05	+
Moderate reaction	*0*	37.8 ± 7.11	36.8 ± 6.86	0.320	<0.05	-
	1 hour	32.0 ± 6.09	33.3 ± 6.41	0.465	<0.05	-
	2 hours	19.9 ± 4.72	27.1 ± 6.03	2.973	<0.05	+
	4 hours	10.7 ± 1.70	17.1 ± 3.81	4.446	<0.001	+++
	8 hours	4.3 ± 2.16	9.1 ± 2.77	4.322	<0.001	+++
Weak reaction	*0*	22.1 ± 7.40	19.9 ± 5.00	0.779	<0.05	-
	1 hour	21.4 ± 3.89	24.6 ± 4.81	1.635	<0.05	-
	2 hours	22.7 ± 2.31	20.5 ± 3.14	1.786	<0.05	-
	4 hours	30.1 ± 5.32	28.6 ± 4.53	0.679	<0.05	-
	8 hours	5.9 ± 3.84	12.5 ± 4.55	3.504	<0.05	+
No reaction	*0*	22.3 ± 2.91	24.3 ± 5.66	0.994	<0.05	-
	1 hour	33.1 ± 8.54	25.1 ± 5.59	2.478	<0.05	+
	2 hours	46.5 ± 7.53	36.7 ± 8.53	2.724	<0.05	+
	4 hours	51.1 ± 10.54	42.2 ± 6.78	2.247	<0.05	+
	8 hours	89.2 ± 6.32	76.9 ± 6.94	4.145	<0.001	+++

+++ = highly significant + = significant - = non-significant

Moderately stained sperms ranged from 23% to 45 % with a mean value of 33.3 ± 6.41 %. They did not differ statistically from the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 32).

Those with a weak reaction ranged from 17% to 32 % with a mean value of 24.6 ± 4.81 %. They did not also differ statistically from the weakly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 32).

Sperms with a negative reaction ranged from 17% to 34 % with a mean value of 25.1 ± 5.59 %. They were significantly less than negatively stained sperms for lactate dehydrogenase in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 32).

c. Specimens examined after two hours:

A similar location of reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 10% to 22 % with a mean value of 15.7 ± 4.67 %. It was significantly more than that of the strongly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 32).

Those with a moderate reaction ranged from 19% to 37 % with a mean value of 27.1 ± 6.03 %. They were significantly more than that of the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 32).

Sperms with a weak reaction ranged from 16% to 26 % with a mean value of 20.5 ± 3.14 %. They did not differ statistically from the weakly stained sperms for lactate

dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 32).

Sperms with a negative reaction ranged from 24% to 46 % with a mean value of 36.7 ± 8.53 %. This was significantly less than that of negatively stained sperms for lactate dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 32).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive lactate dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 6% to 18 % with a mean value of 12.1 ± 4.33 %. They were significantly more than the strongly stained sperms in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 32).

Moderately stained sperms ranged from 12% to 23 % with a mean value of 17.1 ± 3.81 %. They were highly significantly more than that of the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.001$] (Table 32).

Those with a weak reaction ranged from 22% to 33 % with a mean value of 28.6 ± 4.53 %. They did not differ statistically from the weakly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 32).

Sperms with a negative reaction ranged from 34% to 53 % with a mean value of 42.2 ± 6.78 %. They were significantly less than negatively stained sperms for lactate dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 32).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1.5 ± 0.85 %. It was significantly more than that of the strongly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after eight hours of incubation [P <0.05] (Table 32).

Those with a moderate reaction ranged from 6% to 13 % with a mean value of 9.1 ± 2.77 %. They were highly significantly more than that of the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens examined after eight hours of incubation [P < 0.001] (Table 32).

Weakly stained sperms ranged from 7% to 20 % with a mean value of 12.5 ± 4.55 %. They were significantly more than that of the weakly stained sperms for lactate dehydrogenase in the second split fraction specimens examined after eight hours of incubation [P < 0.05] (Table 32).

Sperms with a negative reaction ranged from 67% to 85 % with a mean value of 76.9 ± 6.94 %. They were highly significantly less than negatively stained sperms for lactate dehydrogenase in the second split fraction specimens examined after eight hours of incubation [P <0.001] (Table 32).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for lactate dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = - 0.845; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for lactate dehydrogenase in the second split fraction specimens

to which calcium was added and the period of incubation [r = - 0.871; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A significantly negative correlation was found between the percent of the weakly stained sperms for lactate dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = - 0.396; P <0.05]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for lactate dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = 0.929; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

III. SUCCINIC DEHYDROGENASE:

(A) WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 33)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was noticed in the mid-piece. No reaction was seen in other parts of spermatozoa (Fig. 3).

The percentage of spermatozoa giving a strong reaction ranged from 16% to 31 % with a mean value of 23.1 ± 4.43 %. Those with a moderate reaction ranged from 36% to 57 % with a mean value of 46.8 ± 6.39 %. Weakly stained sperms ranged from 7% to 24 % with a mean value of 15.4 ± 5.93 %. Sperms with a negative reaction ranged from 8% to 21 % with a mean value of 14.7 ± 3.95 %.

b. Specimens examined after one hour:

A similar reaction for succinic dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 17% to 27 % with a

Table (33): Showing the percentage of sperms with different grades of *succinic dehydrogenase reaction in the whole non-washed specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	25	46	12	17	11	19	35	25	21	21	24	44	17	15	31	12	24	31	33	41	6	15	20	59
2	16	49	18	17	12	22	52	12	14	22	19	44	19	18	32	11	31	28	30	42	0	2	11	87
3	19	47	17	17	13	24	39	17	20	23	13	33	27	27	33	12	24	35	29	43	4	8	14	74
4	27	52	9	12	14	22	26	24	28	24	21	40	21	18	34	8	34	25	33	44	6	16	23	55
5	21	38	23	18	15	27	49	16	8	25	20	42	22	16	35	16	37	23	24	45	6	9	17	68
6	19	36	24	21	16	21	47	16	16	26	18	33	24	25	36	19	39	20	22	46	3	14	23	60
7	31	51	10	8	17	17	26	27	30	27	11	27	35	27	37	6	27	31	36	47	1	1	12	86
8	23	43	20	14	18	17	44	23	16	28	15	39	22	24	38	18	30	31	21	48	2	3	16	79
9	25	57	7	11	19	24	39	20	17	29	19	50	18	13	39	13	33	32	22	49	5	14	22	59
10	25	49	14	12	20	24	30	26	20	30	16	32	25	27	40	12	35	24	29	50	7	12	19	62
Mean	23.1	46.8	15.4	14.7	Mean	21.7	38.7	20.6	19.0	Mean	17.6	38.4	23.0	21.0	Mean	12.7	31.4	28.0	27.9	Mean	4.0	9.4	17.7	68.9
S.D.	4.43	6.39	5.93	3.95	S.D.	3.27	9.38	5.13	6.46	S.D.	3.89	7.01	5.25	5.54	S.D.	4.08	5.19	4.78	5.34	S.D.	2.40	5.70	4.42	11.86

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

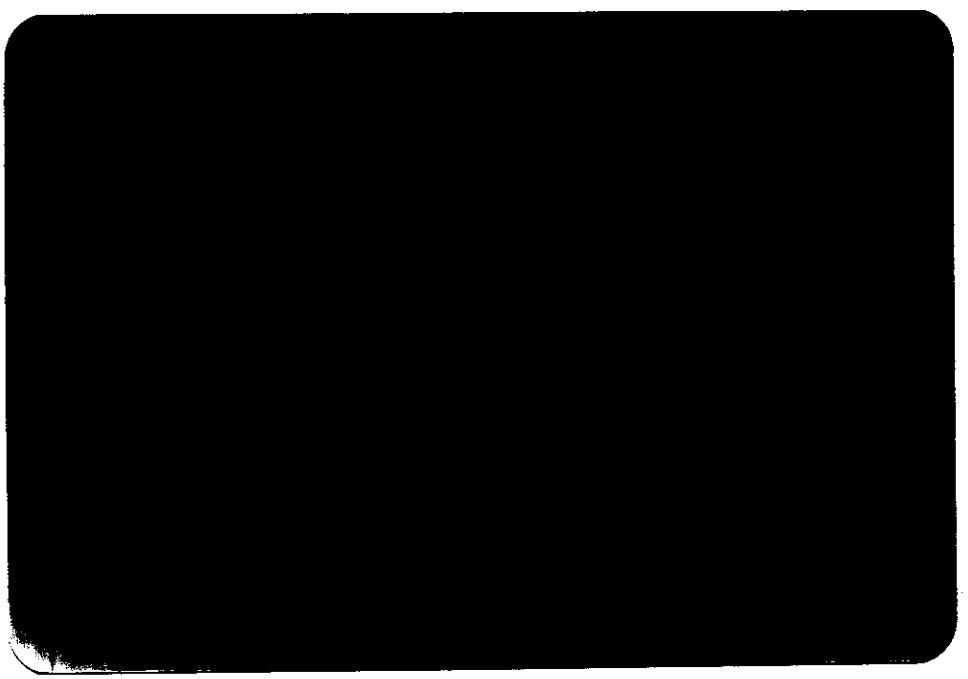

Fig. (3) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **succinic dehydrogenase.**

Note the strong (long arrow), moderate (short arrow), and weak (arrow head) reactions in the midpiece of sperms.

(NBT method. Proj: 10 Obj: 100)

mean value of 21.7 ± 3.27 %. Moderately stained sperms ranged from 26% to 52 % with a mean value of 38.7 ± 9.38 %. Those with a weak reaction ranged from 12% to 27 % with a mean value of 20.6 ± 5.13 %. Sperms with a negative reaction ranged from 8% to 30 % with a mean value of 19 ± 6.46 %.

c. Specimens examined after two hours:

A positive succinic dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 24 % with a mean value of 17.6 ± 3.89 %. Those with a moderate reaction ranged from 27% to 50 % with a mean value of 38.4 ± 7.01 %. Sperms with a weak reaction ranged from 17% to 35 % with a mean value of 23 ± 5.25 %. Sperms with a negative reaction ranged from 13% to 27 % with a mean value of 21 ± 5.54 %.

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive reaction for succinic dehydrogenase. The percentage of spermatozoa giving a strong reaction ranged from 6% to 19 % with a mean value of 12.7 ± 4.08 %. Moderately stained sperms ranged from 24% to 39 % with a mean value of 31.4 ± 5.19 %. Those with a weak reaction ranged from 20% to 35 % with a mean value of 28 ± 4.78 %. Sperms with a negative reaction ranged from 21% to 36 % with a mean value of 27.9 ± 5.34 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 7 % with a mean value of 4 ± 2.4 %. Those with a moderate reaction ranged from 1% to 16 % with a mean value of 9.4 ± 5.7 %. Weakly stained sperms ranged from 11% to 23 % with a mean value of 17.7 ± 4.42 %. Sperms with a negative reaction ranged from 55% to 87 % with a mean value of 68.9 ± 11.86 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens and the period of incubation [r = - 0.889; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens and the period of incubation [r = - 0.878; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant positive correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens and the period of incubation [r = 0.066; P <0.05].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens and the period of incubation [r = 0.911; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) Washed specimens: (Table 34)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed only in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 20% to 30 % with a mean value of 24.6 ± 3.66 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 35).

Those with a moderate reaction ranged from 29% to 53 % with a mean value of 42.6 ± 8.18 %. They did not differ statistically from the moderately stained sperms for

Table (34): Showing the percentage of sperms with different grades of *succinic dehydrogenase reaction in the washed specimens.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	24	41	14	21	11	11	48	22	19	21	15	40	26	19	31	0	12	34	54	41	0	1	5	94
2	21	33	16	10	12	19	46	21	14	22	12	39	25	24	32	7	22	34	37	42	0	0	4	96
3	20	29	24	27	13	26	49	17	8	23	6	28	34	32	33	2	19	32	47	43	0	1	6	93
4	30	51	10	9	14	22	29	32	17	24	14	36	27	23	34	2	19	36	43	44	1	3	10	86
5	22	40	17	21	15	26	50	16	8	25	17	28	33	22	35	8	26	31	35	45	1	2	11	86
6	24	34	21	21	16	23	33	22	22	26	7	34	27	32	36	5	23	35	37	46	1	1	9	89
7	28	50	14	8	17	16	26	36	22	27	8	24	35	33	37	1	16	43	40	47	0	1	4	95
8	21	49	20	10	18	13	47	23	23	28	6	37	26	31	38	5	24	37	34	48	0	1	3	96
9	27	35	20	18	19	17	42	24	17	29	15	29	24	32	39	3	20	38	39	49	0	1	8	91
10	29	44	14	13	20	17	30	30	23	30	11	37	27	25	40	1	11	45	43	50	0	2	8	90
Mean	24.6	42.6	17.0	15.8	Mean	19.0	39.4	24.3	17.3	Mean	11.1	33.2	28.4	27.3	Mean	3.4	19.2	36.5	40.9	Mean	0.3	1.3	6.8	91.6
S.D.	3.66	8.18	4.22	6.61	S.D.	5.16	9.12	6.45	5.74	S.D.	4.12	5.51	4.01	5.21	S.D.	2.72	4.96	4.50	6.10	S.D.	0.48	0.82	2.78	3.81

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (35): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Significance.
Strong reaction	*0*	23.1 ± 4.43	24.6 ± 3.66	0.825	<0.05	-
	1 hour	21.7 ± 3.27	19.0 ± 5.16	1.397	<0.05	-
	2 hours	17.6 ± 3.89	11.1 ± 4.12	3.625	<0.05	+
	4 hours	12.7 ± 4.08	3.4 ± 2.72	5.996	<0.001	+++
	8 hours	4.0 ± 2.40	0.3 ± 0.48	4.772	<0.001	+++
Moderate reaction	*0*	46.8 ± 6.39	42.6 ± 8.18	1.279	<0.05	-
	1 hour	38.7 ± 9.38	39.4 ± 9.12	0.169	<0.05	-
	2 hours	38.4 ± 7.01	33.2 ± 5.51	1.844	<0.05	-
	4 hours	31.4 ± 5.19	19.2 ± 4.96	5.373	<0.001	+++
	8 hours	9.4 ± 5.70	1.3 ± 0.82	4.448	<0.001	+++
Weak reaction	*0*	15.4 ± 5.93	17.0 ± 4.22	0.695	<0.05	-
	1 hour	20.6 ± 5.13	24.3 ± 6.45	1.421	<0.05	-
	2 hours	23.0 ± 5.25	28.4 ± 4.01	2.586	<0.05	+
	4 hours	28.0 ± 4.78	36.5 ± 4.50	4.091	<0.001	+++
	8 hours	17.7 ± 4.42	6.8 ± 2.78	6.597	<0.001	+++
No reaction	*0*	14.7 ± 3.95	15.8 ± 6.61	0.452	<0.05	-
	1 hour	19.0 ± 6.46	17.3 ± 5.74	0.622	<0.05	-
	2 hours	21.0 ± 5.54	27.3 ± 5.21	2.621	<0.05	+
	4 hours	27.9 ± 5.34	40.9 ± 6.10	5.070	<0.001	+++
	8 hours	68.9 ± 11.86	91.6 ± 3.81	5.765	<0.001	+++

- = non-significant + = significant +++ = highly significant

succinic dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 35).

Weakly stained sperms ranged from 10% to 24 % with a mean value of 17 ± 4.22 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 35).

Sperms with a negative reaction ranged from 8% to 27 % with a mean value of 15.8 ± 6.61 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 35).

b. Specimens examined after one hour:

A similar reaction for succinic dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 26 % with a mean value of 19 ± 5.16 %. This did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 35).

Moderately stained sperms ranged from 26% to 50 % with a mean value of 39.4 ± 9.12 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 35).

Those with a weak reaction ranged from 16% to 36 % with a mean value of 24.3 ± 6.45 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 35).

Those with a moderate reaction ranged from 24% to 40 % with a mean value of 33.2 ± 5.51 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 35).

Sperms with a weak reaction ranged from 24% to 35 % with a mean value of 28.4 ± 4.01 %. They were significantly more than that of the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 35).

Sperms with a negative reaction ranged from 19% to 33 % with a mean value of 27.3 ± 5.21 %. This was significantly more than that of negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 35).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 8 % with a

mean value of 3.4 ± 2.72 %. They were highly significantly less than the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.001] (Table 35).

Moderately stained sperms ranged from 11% to 26 % with a mean value of 19.2 ± 4.96 %. They were highly significantly less than that of the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.001] (Table 35).

Those with a weak reaction ranged from 31% to 45 % with a mean value of 36.5 ± 4.5 %. They were highly significantly more than that of the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.001] (Table 35).

Sperms with a negative reaction ranged from 34% to 54 % with a mean value of 40.9 ± 6.1 %. They were highly significantly more than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.001] (Table 35).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.3 ± 0.48 %. It was highly significantly less than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P <0.001] (Table 35).

Those with a moderate reaction ranged from 0% to 3 % with a mean value of 1.3 ± 0.82 %. They were highly significantly less than that of the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.001] (Table 35).

Weakly stained sperms ranged from 3% to 11 % with a mean value of 6.8 ± 2.78 %. They were highly significantly less than that of the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 35).

Sperms with a negative reaction ranged from 86% to 96 % with a mean value of 91.6 ± 3.81 %. They were highly significantly more than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 35).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase for succinic dehydrogenase in the washed specimens and the period of incubation [$r = - 0.854$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the washed specimens and the period of incubation [$r = - 0.924$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A significantly negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the washed specimens and the period of incubation [$r = - 0.370$; $P < 0.05$]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the washed specimens and the period of incubation [$r = 0.967$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 36).

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 17% to 27 % with a mean value of 22 ± 3.46 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P <0.05] (Table 37).

Those with a moderate reaction ranged from 40% to 61 % with a mean value of 48.9 ± 6.14 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 37).

Weakly stained sperms ranged from 8% to 24 % with a mean value of 16.2 ± 5.41 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 37).

Sperms with a negative reaction ranged from 7% to 18 % with a mean value of 12.9 ± 3.73 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P <0.05] (Table 37).

b. Specimens examined after one hour:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 16% to 28 % with a mean value of 20.3 ± 4.22 %. This did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [P <0.05] (Table 37).

Table (36): Showing the percentage of sperms with different grades of *succinic dehydrogenase reaction in the washed specimens to which calcium was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	23	46	18	13	11	17	34	27	22	21	16	43	20	21	31	3	16	28	53	41	0	2	12	86
2	26	48	16	10	12	28	42	16	14	22	16	42	18	24	32	7	24	28	41	42	0	1	6	93
3	19	40	24	17	13	26	51	11	12	23	7	30	35	28	33	3	17	37	43	43	0	3	7	90
4	25	56	9	10	14	20	34	20	26	24	19	41	24	16	34	4	22	29	45	44	1	4	11	84
5	21	49	12	18	15	24	42	19	15	25	13	35	26	26	35	5	29	36	30	45	0	4	11	85
6	19	44	23	14	16	17	47	17	19	26	8	34	31	27	36	6	14	43	37	46	1	3	10	86
7	24	61	8	7	17	16	39	22	23	27	8	27	32	33	37	5	16	29	50	47	0	0	6	94
8	17	50	17	16	18	19	35	25	21	28	15	35	28	22	38	6	27	31	36	48	0	1	7	92
9	27	44	20	9	19	19	48	17	16	29	19	40	21	20	39	7	22	34	37	49	0	2	9	89
10	19	51	15	15	20	17	38	21	24	30	12	43	24	21	40	2	15	32	51	50	0	3	8	89
Mean	22.0	48.9	16.2	12.9	Mean	20.3	41.0	19.5	19.2	Mean	13.3	37.0	25.9	23.8	Mean	4.8	20.2	32.7	42.3	Mean	0.2	2.3	8.7	88.8
S.D.	3.46	6.14	5.41	3.73	S.D.	4.22	6.09	4.62	4.73	S.D.	4.47	5.66	5.57	4.85	S.D.	1.75	5.33	4.85	7.50	S.D.	0.42	1.34	2.21	3.49

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (37): Showing the comparison between the *succinic dehydrogenase reaction in the sperms of washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Calcium	t	p	Significance.
Strong reaction	*0*	24.6 ± 3.66	22.0 ± 3.46	1.632	<0.05	-
	1 hour	19.0 ± 5.16	20.3 ± 4.22	0.617	<0.05	-
	2 hours	11.1 ± 4.12	13.3 ± 4.47	1.144	<0.05	-
	4 hours	3.4 ± 2.72	4.8 ± 1.75	1.370	<0.05	-
	8 hours	0.3 ± 0.48	0.2 ± 0.42	0.493	<0.05	-
Moderate reaction	*0*	42.6 ± 8.18	48.9 ± 6.14	1.948	<0.05	-
	1 hour	39.4 ± 9.12	41.0 ± 6.09	0.461	<0.05	-
	2 hours	33.2 ± 5.51	37.0 ± 5.66	1.521	<0.05	-
	4 hours	19.2 ± 4.96	20.2 ± 5.33	0.434	<0.05	-
	8 hours	1.3 ± 0.82	2.3 ± 1.34	2.014	<0.05	-
Weak reaction	*0*	17.0 ± 4.22	16.2 ± 5.41	0.369	<0.05	-
	1 hour	24.3 ± 6.45	19.5 ± 4.62	1.913	<0.05	-
	2 hours	28.4 ± 4.01	25.9 ± 5.57	1.153	<0.05	-
	4 hours	36.5 ± 4.50	32.7 ± 4.85	1.815	<0.05	-
	8 hours	6.8 ± 2.78	8.7 ± 2.21	1.690	<0.05	-
No reaction	*0*	15.8 ± 6.61	12.9 ± 3.73	1.208	<0.05	-
	1 hour	17.3 ± 5.74	19.2 ± 4.73	0.808	<0.05	-
	2 hours	27.3 ± 5.21	23.8 ± 4.85	1.555	<0.05	-
	4 hours	40.9 ± 6.10	42.3 ± 7.50	0.458	<0.05	-
	8 hours	91.6 ± 3.81	88.8 ± 3.49	1.715	<0.05	-

- = non-significant

Moderately stained sperms ranged from 34% to 51 % with a mean value of 41 ± 6.09 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 37).

Those with a weak reaction ranged from 11% to 27 % with a mean value of 19.5 ± 4.62 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 37).

Sperms with a negative reaction ranged from 12% to 26 % with a mean value of 19.2 ± 4.73 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 37).

c. Specimens examined after two hours:

A positive succinic dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 7% to 19 % with a mean value of 13.3 ± 4.47 %. Also, it did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 37).

Those with a moderate reaction ranged from 27% to 43 % with a mean value of 37 ± 5.66 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 37).

Sperms with a weak reaction ranged from 18% to 35 % with a mean value of 25.9 ± 5.57 %. They did not also differ statistically from the weakly stained sperms for

succinic dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 37).

Sperms with a negative reaction ranged from 16% to 33 % with a mean value of 23.8 ± 4.85 %. This did not differ statistically from that of negatively stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 37).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 2% to 7 % with a mean value of 4.8 ± 1.75 %. They did not differ statistically from the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 37).

Moderately stained sperms ranged from 14% to 29 % with a mean value of 20.2 ± 5.33 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 37).

Those with a weak reaction ranged from 28% to 43 % with a mean value of 32.7 ± 4.85 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 37).

Sperms with a negative reaction ranged from 30% to 53 % with a mean value of 42.3 ± 7.5 %. Also, they did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 37).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.2 ± 0.42 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [P <0.05] (Table 37).

Those with a moderate reaction ranged from 0% to 4 % with a mean value of 2.3 ± 1.34 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [P < 0.05] (Table 37).

Weakly stained sperms ranged from 6% to 12 % with a mean value of 8.7 ± 2.21 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [P < 0.05] (Table 37).

Sperms with a negative reaction ranged from 84% to 94 % with a mean value of 88.8 ± 3.49 %. They did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [P <0.05] (Table 37).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the washed specimens to which calcium was added and the period of incubation [r = - 0.892; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the washed specimens to which

calcium was added and the period of incubation [r = - 0.951; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A significantly negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the washed specimens to which calcium was added and the period of incubation [r = - 0.283; P <0.05]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the washed specimens to which calcium was added and the period of incubation [r = 0.979; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Washed specimens to which ascorbic acid was added: (Table 38)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 16% to 28 % with a mean value of 22.6 ± 3.47 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P <0.05] (Table 39).

Those with a moderate reaction ranged from 36% to 52 % with a mean value of 45.2 ± 5.67 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 39).

Weakly stained sperms ranged from 13% to 24 % with a mean value of 18.2 ± 4.13 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P < 0.05] (Table 39).

Table (38): Showing the percentage of sperms with different grades of *succinic dehydrogenase reaction in the washed specimens to which ascorbic acid was added.*

No.of Ejaculate	Immediate				No.of Ejaculate	1 hour				No.of Ejaculate	2 hours				No.of Ejaculate	4 hours				No.of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	22	48	16	14	11	17	27	27	29	21	20	47	18	15	31	7	25	32	36	41	2	5	17	76
2	23	49	15	13	12	21	50	18	11	22	19	44	20	17	32	8	32	30	30	42	0	2	9	89
3	16	40	23	21	13	25	54	11	10	23	10	39	28	23	33	7	25	37	31	43	3	3	13	81
4	28	46	15	11	14	16	30	22	32	24	18	42	21	19	34	10	22	31	37	44	4	8	21	67
5	21	37	24	18	15	25	41	19	15	25	16	37	26	21	35	13	28	32	27	45	1	5	19	75
6	22	36	23	19	16	24	47	14	15	26	10	33	31	26	36	11	23	37	29	46	3	6	20	71
7	25	52	14	9	17	19	38	21	22	27	17	29	27	27	37	5	21	29	45	47	0	3	12	85
8	24	48	20	8	18	18	31	23	28	28	16	30	23	31	38	11	31	34	24	48	0	4	17	79
9	19	45	19	17	19	23	49	15	13	29	12	43	26	19	39	7	28	34	31	49	2	3	11	84
10	26	51	13	10	20	18	39	26	17	30	20	41	21	18	40	4	18	37	41	50	3	7	18	72
Mean	22.6	45.2	18.2	14.0	Mean	20.6	40.6	19.6	19.2	Mean	15.8	38.5	24.1	21.6	Mean	8.3	25.3	33.3	33.1	Mean	1.8	4.6	15.7	77.9
S.D.	3.47	5.67	4.13	4.55	S.D.	3.44	9.30	5.21	8.00	S.D.	3.85	6.11	4.12	5.06	S.D.	2.87	4.47	2.98	6.52	S.D.	1.48	1.96	4.14	6.95

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (39): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *washed specimens* and *washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	24.6 ± 3.66	22.6 ± 3.47	1.254	<0.05	-
	1 hour	19.0 ± 5.16	20.6 ± 3.44	0.816	<0.05	-
	2 hours	11.1 ± 4.12	15.8 ± 3.85	2.634	<0.05	+
	4 hours	3.4 ± 2.72	8.3 ± 2.87	3.922	<0.05	+
	8 hours	0.3 ± 0.48	1.8 ± 1.48	3.055	<0.05	+
Moderate reaction	*0*	42.6 ± 8.18	45.2 ± 5,67	0.826	<0.05	-
	1 hour	39.4 ± 9.12	40.6 ± 9.30	0.291	<0.05	-
	2 hours	33.2 ± 5.51	38.5 ± 6.11	2.036	<0.05	+
	4 hours	19.2 ± 4.96	25.3 ± 4.47	2.887	<0.05	+
	8 hours	1.3 ± 0.82	4.6 ± 1.96	4.919	<0.001	+++
Weak reaction	*0*	17.0 ± 4.22	18.2 ± 4.13	0.643	<0.05	-
	1 hour	24.3 ± 6.45	19.6 ± 5.21	1.793	<0.05	-
	2 hours	28.4 ± 4.01	24.1 ± 4.12	2.366	<0.05	+
	4 hours	36.5 ± 4.50	33.3 ± 2.98	1.873	<0.05	-
	8 hours	6.8 ± 2.78	15.7 ± 4.14	5.645	<0.001	+++
No reaction	*0*	15.8 ± 6.61	14.0 ± 4.55	0.709	<0.05	-
	1 hour	17.3 ± 5.74	19.2 ± 8.00	0.611	<0.05	-
	2 hours	27.3 ± 5.21	21.6 ± 5.06	2.482	<0.05	+
	4 hours	40.9 ± 6.10	33.1 ± 6.52	2.762	<0.05	+
	8 hours	91.6 ± 3.81	77.9 ± 6.95	5.466	<0.001	+++

+++ = highly significant + = significant - = non-significant

Sperms with a negative reaction ranged from 8% to 21 % with a mean value of 14 ± 4.55 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the immediately examined washed specimens [P <0.05] (Table 39).

b. Specimens examined after one hour:

A similar reaction for succinic dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 16% to 25 % with a mean value of 20.6 ± 3.44 %. Also, this did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [P <0.05] (Table 39).

Moderately stained sperms ranged from 27% to 54 % with a mean value of 40.6 ± 9.3 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [P < 0.05] (Table 39).

Those with a weak reaction ranged from 11% to 27 % with a mean value of 19.6 ± 5.21 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [P < 0.05] (Table 39).

Sperms with a negative reaction ranged from 10% to 32 % with a mean value of 19.2 ± 8 %. They did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after one hour of incubation [P <0.05] (Table 39).

c. Specimens examined after two hours:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 10% to 20 % with a mean value of 15.8 ± 3.85 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [P <0.05] (Table 39).

Those with a moderate reaction ranged from 29% to 47 % with a mean value of 38.5 ± 6.11 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 39).

Sperms with a weak reaction ranged from 18% to 31 % with a mean value of 24.1 ± 4.12 %. They were significantly less than that of the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [P < 0.05] (Table 39).

Sperms with a negative reaction ranged from 15% to 31 % with a mean value of 21.6 ± 5.06 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after two hours of incubation [P <0.05] (Table 39).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 4% to 13 % with a mean value of 8.3 ± 2.87 %. This was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [P <0.05] (Table 39).

Moderately stained sperms ranged from 18% to 32 % with a mean value of 25.3 ± 4.47 %. They were significantly more than that of the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 39).

Those with a weak reaction ranged from 29% to 37 % with a mean value of 33.3 ± 2.98 %. They did not differ statistically from the weakly stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 39).

Sperms with a negative reaction ranged from 24% to 45 % with a mean value of 33.1 ± 6.52 %. This was significantly less than that of negatively stained sperms for succinic dehydrogenase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 39).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 1.8 ± 1.48 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 39).

Those with a moderate reaction ranged from 2% to 8 % with a mean value of 4.6 ± 1.96 %. They were highly significantly more than that of the moderately stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 39).

Weakly stained sperms ranged from 9% to 21 % with a mean value of 15.7 ± 4.14 %. They were highly significantly more than that of the weakly stained

sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 39).

Sperms with a negative reaction ranged from 67% to 89 % with a mean value of 77.9 ± 6.95 %. They were highly significantly less than the negatively stained sperms for succinic dehydrogenase in the washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 39).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.914$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.929$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.059$; $P < 0.05$].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = 0.943$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(B) SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 40)

a. Immediately examined specimens:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 25% to 35 % with a mean value of 29.1 ± 3.28 %. They were significantly more than the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 41).

Moderately stained sperms ranged from 32% to 53 % with a mean value of 41.5 ± 7.14 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 41).

Those with a weak reaction ranged from 12% to 27 % with a mean value of 18.5 ± 5.17 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 41).

Sperms with a negative reaction ranged from 5% to 16 % with a mean value of 10.9 ± 4.12 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 41).

b. Specimens examined after one hour:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 20% to 28 % with a mean value of 25 ± 2.62 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 41).

Table (40): Showing the percentage of sperms with different grades of *succinic dehydrogenase in the first split fraction specimens.*

No.of Ejaculate	Immediate +++	++	+	-	No.of Ejaculate	1 hour +++	++	+	-	No.of Ejaculate	2 hours +++	++	+	-	No.of Ejaculate	4 hours +++	++	+	-	No.of Ejaculate	8 hours +++	++	+	-
51	25	32	27	16	61	21	34	26	19	71	22	44	23	11	81	16	42	27	15	91	6	23	22	49
52	32	48	14	6	62	28	47	16	9	72	23	46	21	10	82	11	31	30	28	92	5	16	12	67
53	31	38	19	12	63	25	34	23	18	73	19	28	33	20	83	17	35	26	24	93	10	16	14	60
54	30	53	12	5	64	26	47	20	7	74	15	33	33	19	84	20	40	23	17	94	7	24	21	48
55	28	34	23	15	65	27	54	11	8	75	23	39	26	8	85	17	35	25	23	95	3	19	14	64
56	26	47	19	8	66	26	41	23	10	76	23	37	25	15	86	19	36	23	22	96	10	19	17	54
57	25	35	25	15	67	24	48	17	11	77	24	47	18	11	87	17	29	34	20	97	9	20	18	53
58	28	46	15	11	68	20	31	29	20	78	17	33	30	20	88	13	26	34	27	98	5	14	14	67
59	35	45	13	7	69	26	43	22	9	79	20	43	22	15	89	13	30	29	28	99	6	18	19	57
60	31	37	18	14	70	27	39	22	12	70	21	48	21	10	80	20	39	23	18	100	4	13	14	69
Mean	29.1	41.5	18.5	10.9	Mean	25.0	41.8	20.9	12.3	Mean	21.1	39.8	25.2	13.9	Mean	16.3	34.1	27.4	22.2	Mean	6.5	18.2	16.5	58.8
S.D.	3.28	7.14	5.17	4.12	S.D.	2.62	7.38	5.17	4.85	S.D.	3.51	6.88	5.25	4.53	S.D.	3.09	5.22	4.25	4.66	S.D.	2.46	3.58	3.41	7.74

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (41): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *whole non-washed* and *first split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Significance.
Strong reaction	*0*	23.1 ± 4.43	29.1 ± 3.28	3.440	<0.05	+
	1 hour	21.7 ± 3.27	25.0 ± 2.62	2.490	<0.05	+
	2 hours	17.6 ± 3.89	21.1 ± 3.51	2.111	<0.05	+
	4 hours	12.7 ± 4.08	16.3 ± 3.09	2.222	<0.05	+
	8 hours	4.0 ± 2.40	6.5 ± 2.46	2.298	<0.05	+
Moderate reaction	*0*	46.8 ± 6.39	41.5 ± 7.14	1.749	<0.05	-
	1 hour	38.7 ± 9.38	41.8 ± 7.38	0.822	<0.05	-
	2 hours	38.4 ± 7.01	39.8 ± 6.88	0.451	<0.05	-
	4 hours	31.4 ± 5.19	34.1 ± 5.22	1.160	<0.05	-
	8 hours	9.4 ± 5 70	18.2 ± 3.58	4.133	<0.001	+++
Weak reaction	*0*	15.4 ± 5.93	18.5 ± 5.17	1.246	<0.05	-
	1 hour	20.6 ± 5.13	20.9 ± 5.17	0.130	<0.05	-
	2 hours	23.0 ± 5.25	25.2 ± 5.25	0.938	<0.05	-
	4 hours	28.0 ± 4.78	27.4 ± 4.25	0.297	<0.05	-
	8 hours	17.7 ± 4.42	16.5 ± 3.41	0.680	<0.05	-
No reaction	*0*	14.7 ± 3.95	10.9 ± 4.12	2.106	<0.05	+
	1 hour	19.0 ± 6.46	12.3 ± 4.85	2.621	<0.05	+
	2 hours	21.0 ± 5.54	13.9 ± 4.53	3.137	<0.05	+
	4 hours	27.9 ± 5.34	22.2 ± 4.66	2.542	<0.05	+
	8 hours	68.9 ± 11.86	58.8 ± 7.74	2.256	<0.05	+

+++ = highly significant + = significant - = non-significant

Those with a moderate reaction ranged from 31% to 54 % with a mean value of 41.8 ± 7.38 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 41).

Weakly stained sperms ranged from 11% to 29 % with a mean value of 20.9 ± 5.17 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 41).

Sperms with a negative reaction ranged from 7% to 20 % with a mean value of 12.3 ± 4.85 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 41).

c. Specimens examined after two hours:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 15% to 27 % with a mean value of 21.1 ± 3.51 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 41).

Those with a moderate reaction ranged from 28% to 48 % with a mean value of 39.8 ± 6.88 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 41).

Sperms with a weak reaction ranged from 18% to 33 % with a mean value of 25.2 ± 5.25 %. They did not also differ statistically from the weakly stained sperms for

succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 41).

Sperms with a negative reaction ranged from 8% to 20 % with a mean value of 13.9 ± 4.53 %. This was significantly less than that of negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 41).

d. Specimens examined after four hours:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 20 % with a mean value of 16.3 ± 3.09 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 41).

Those with a moderate reaction ranged from 26% to 42 % with a mean value of 34.1 ± 5.22 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 41).

Weakly stained sperms ranged from 23% to 34 % with a mean value of 27.4 ± 4.25 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 41).

Sperms with a negative reaction ranged from 15% to 28 % with a mean value of 22.2 ± 4.66 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 41).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 3% to 10 % with a mean value of 6.5 ± 2.46 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 41).

Those with a moderate reaction ranged from 13% to 24 % with a mean value of 18.2 ± 3.58 %. They were highly significantly more than that of the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 41).

Weakly stained sperms ranged from 12% to 22 % with a mean value of 16.5 ± 3.41 %. They did not differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 41).

Sperms with a negative reaction ranged from 48% to 69 % with a mean value of 58.8 ± 7.74 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 41).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens and the period of incubation [$r = - 0.933$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the first split fraction specimens

and the period of incubation [r = - 0.815; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens and the period of incubation [r = - 0.148; P <0.05].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the first split fraction specimens and the period of incubation [r = 0.925; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added: (Table 42)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 21% to 31 % with a mean value of 27.4 ± 3.6 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the immediately examined first split fraction specimens [P <0.05] (Table 43).

Those with a moderate reaction ranged from 36% to 54 % with a mean value of 45.2 ± 6.68 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined first split fraction specimens [P < 0.05] (Table 43).

Weakly stained sperms ranged from 12% to 24 % with a mean value of 18.1 ± 4.15 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined first split fraction specimens [P < 0.05] (Table 43).

Table (42): Showing the percentage of sperms with different grades of *succinic dehydrogenase reaction in the first split fraction specimens to which ascorbic acid was added.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	23	37	24	16	61	23	37	23	17	71	25	45	19	11	81	25	42	23	10	91	9	21	24	46
52	31	46	17	6	62	30	51	12	7	72	29	40	23	8	82	16	35	24	25	92	10	18	21	51
53	25	53	15	7	63	26	44	18	12	73	24	45	18	13	83	22	39	27	12	93	7	18	18	57
54	29	43	20	8	64	31	52	8	9	74	21	33	29	17	84	21	45	24	10	94	13	26	24	37
55	25	48	18	9	65	25	39	22	14	75	23	43	19	15	85	20	38	24	18	95	5	18	23	54
56	30	54	12	4	66	26	41	20	13	76	27	48	16	9	86	18	27	34	21	96	11	17	23	49
57	29	36	20	15	67	23	40	21	16	77	27	48	18	7	87	22	44	20	14	97	12	25	17	46
58	21	40	23	16	68	23	29	27	21	78	17	34	30	19	88	17	31	29	23	98	7	19	18	56
59	31	53	12	4	69	24	46	19	11	79	21	42	24	13	89	21	40	28	11	99	11	16	20	53
60	30	42	20	8	70	27	51	13	9	70	22	46	22	10	80	18	48	22	12	100	6	18	15	61
Mean	27.4	45.2	18.1	9.3	Mean	25.8	43.0	18.3	12.9	Mean	23.6	42.4	21.8	12.2	Mean	20.0	38.9	25.5	15.6	Mean	9.1	19.6	20.3	51.0
S.D.	3.60	6.68	4.15	4.69	S.D.	2.86	7.30	5.74	4.25	S.D.	3.57	5.32	4.76	3.94	S.D.	2.75	6.47	4.06	5.68	S.D.	2.73	3.37	3.20	6.86

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (43): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incuba-tion.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Signific-ance.
Strong reaction	*0*	29.1 ± 3.28	27.4 ± 3.60	1.104	<0.05	-
	1 hour	25.0 ± 2.62	25.8 ± 2.86	0.652	<0.05	-
	2 hours	21.1 ± 3.51	23.6 ± 3.57	1.580	<0.05	-
	4 hours	16.3 ± 3.09	20.0 ± 2.75	2.828	<0.05	+
	8 hours	6.5 ± 2.46	9.1 ± 2.73	2.239	<0.05	+
Moderate reaction	*0*	41.5 ± 7.14	45.2 ± 6.68	1.197	<0.05	-
	1 hour	41.8 ± 7.38	43.0 ± 7.30	0.366	<0.05	-
	2 hours	39.8 ± 6.88	42.4 ± 5.32	0.946	<0.05	-
	4 hours	34.1 ± 5.22	38.9 ± 6.47	1.826	<0.05	-
	8 hours	18.2 ± 3.58	19.6 ± 3.37	0.900	<0.05	-
Weak reaction	*0*	18.5 ± 5.17	18.1 ± 4.15	0.191	<0.05	-
	1 hour	20.9 ± 5.17	18.3 ± 5.74	1.064	<0.05	-
	2 hours	25.2 ± 5.25	21.8 ± 4.76	1.519	<0.05	-
	4 hours	27.4 ± 4.25	25.5 ± 4.06	1.022	<0.05	-
	8 hours	16.5 ± 3.41	20.3 ± 3.20	2.571	<0.05	+
No reaction	*0*	10.9 ± 4.12	9.3 ± 4.69	0.810	<0.05	-
	1 hour	12.3 ± 4.85	12.9 ± 4.25	0.294	<0.05	-
	2 hours	13.9 ± 4.53	12.2 ± 3.94	0.895	<0.05	-
	4 hours	22.2 ± 4.66	15.6 ± 5.68	2.840	<0.05	+
	8 hours	58.8 ± 7.74	51.0 ± 6.86	2.384	<0.05	+

+ = significant - = non-significant

Sperms with a negative reaction ranged from 4% to 16 % with a mean value of 9.3 ± 4.69 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the immediately examined first split fraction specimens [P <0.05] (Table 43).

b. Specimens examined after one hour:

A similar reaction for succinic dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 23% to 31 % with a mean value of 25.8 ± 2.86 %. Also, this did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after one hour of incubation [P <0.05] (Table 43).

Moderately stained sperms ranged from 29% to 52 % with a mean value of 43 ± 7.3 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the first split fraction specimens examined after one hour of incubation [P < 0.05] (Table 43).

Those with a weak reaction ranged from 8% to 27 % with a mean value of 18.3 ± 5.74 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after one hour of incubation [P < 0.05] (Table 43).

Sperms with a negative reaction ranged from 7% to 21 % with a mean value of 12.9 ± 4.25 %. They did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the first split fraction specimens examined after one hour of incubation [P <0.05] (Table 43).

c. Specimens examined after two hours:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 17% to 29 % with a mean value of 23.6 ± 3.57 %. Also, it did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after two hours of incubation [P <0.05] (Table 43).

Those with a moderate reaction ranged from 33% to 48 % with a mean value of 42.4 ± 5.32 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the first split fraction specimens examined after two hours of incubation [P < 0.05] (Table 43).

Sperms with a weak reaction ranged from 16% to 30 % with a mean value of 21.8 ± 4.76 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after two hours of incubation [P < 0.05] (Table 43).

Sperms with a negative reaction ranged from 7% to 19 % with a mean value of 12.2 ± 3.94 %. They did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the first split fraction specimens examined after two hours of incubation [P <0.05] (Table 43).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 16% to 25 % with a mean value of 20 ± 2.75 %. This was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after four hours of incubation [P <0.05] (Table 43).

Moderately stained sperms ranged from 27% to 48 % with a mean value of 38.9 ± 6.47 %. They did not differ statistically from the moderately stained sperms for

succinic dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 43).

Those with a weak reaction ranged from 20% to 34 % with a mean value of 25.5 ± 4.06 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 43).

Sperms with a negative reaction ranged from 10% to 25 % with a mean value of 15.6 ± 5.68 %. This was significantly less than that of negatively stained sperms for succinic dehydrogenase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 43).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 5% to 13 % with a mean value of 9.1 ± 2.73 %. It was significantly more than that of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 43).

Those with a moderate reaction ranged from 16% to 26 % with a mean value of 19.6 ± 3.37 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 43).

Weakly stained sperms ranged from 15% to 24 % with a mean value of 20.3 ± 3.2 %. They were significantly more than that of the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 43).

Sperms with a negative reaction ranged from 37% to 61 % with a mean value of 51 ± 6.86 %. They were significantly less than the negatively stained sperms for succinic dehydrogenase in the first split fraction specimens examined after eight hours of incubation [P <0.05] (Table 43).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = - 0.906; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = - 0.823; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant positive correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = 0.201; P <0.05].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [r = 0.888; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 44)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 13% to 22 % with a mean value of 18.9 ± 3.14 %. It did not differ statistically from that of the strongly

Table (44): Showing the percentage of spermes with different grades of *succinic dehydrogenase in the second split fraction specimens.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
51	22	36	21	21	61	20	36	20	24	71	7	26	29	38	81	5	24	27	44	91	0	6	8	86
52	19	46	19	16	62	16	24	29	31	72	9	25	28	38	82	12	38	18	32	92	3	14	15	68
53	17	36	26	21	63	16	32	22	30	73	15	43	19	23	83	8	34	30	28	93	2	5	10	83
54	18	35	24	23	64	23	35	24	18	74	13	41	20	26	84	3	20	34	43	94	1	6	7	86
55	22	47	15	16	65	18	26	29	27	75	12	37	23	28	85	12	27	30	31	95	3	13	13	71
56	20	46	17	17	66	17	41	19	23	76	13	32	28	27	86	10	30	24	36	96	3	10	14	73
57	22	42	20	16	67	21	46	15	18	77	10	25	32	33	87	4	29	26	41	97	0	5	6	89
58	21	52	13	14	68	21	40	18	21	78	16	40	17	27	88	15	39	22	24	98	0	9	16	75
59	13	36	26	25	69	13	28	29	30	79	15	42	17	26	89	11	32	28	29	99	2	7	12	79
60	15	45	18	22	70	18	30	30	22	70	9	32	28	31	80	6	26	28	40	100	1	3	9	87
Mean	18.9	42.1	19.9	19.1	Mean	18.3	33.8	23.5	24.4	Mean	11.9	34.3	24.1	29.7	Mean	8.6	29.9	26.7	34.8	Mean	1.5	7.8	11.0	79.7
S.D.	3.14	5.99	4.43	3.73	S.D.	2.98	7.10	5.48	4.88	S.D.	3.03	7.24	5.55	5.17	S.D.	4.01	6.03	4.52	6.97	S.D.	1.27	3.61	3.50	7.53

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (45): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Wasned Specimens.	Second Solit Fraction Specimens.	t	p	Significance.
Strong reaction	*0*	23.1 ± 4.43	18.9 ± 3.14	2.444	<0.05	+
	1 hour	21.7 ± 3.27	18.3 ± 2.98	2.430	<0.05	+
	2 hours	17.6 ± 3.89	11.9 ± 3.03	3.652	<0.05	+
	4 hours	12.7 ± 4.08	8.6 ± 4.01	2.267	<0.05	+
	8 hours	4.0 ± 2.40	1.5 ± 1.27	2.908	<0.05	+
Moderate reaction	*0*	46.8 ± 6.39	42.1 ± 5.99	1.697	<0.05	-
	1 hour	38.7 ± 9.38	33.8 ± 7.10	1.317	<0.05	-
	2 hours	38.4 ± 7.01	34.3 ± 7.24	1.286	<0.05	-
	4 hours	31.4 ± 5.19	29.9 ± 6.03	0.596	<0.05	-
	8 hours	9.4 ± 5.70	7.8 ± 3.61	0.750	<0.05	-
Weak reaction	*0*	15.4 ± 5.93	19.9 ± 4.43	1.922	<0.05	-
	1 hour	20.6 ± 5.13	23.5 ± 5.48	1.122	<0.05	-
	2 hours	23.0 ± 5.25	24.1 ± 5.55	0.456	<0.05	-
	4 hours	28.0 ± 4.78	26.7 ± 4.52	0.624	<0.05	-
	8 hours	17.7 ± 4.42	11.0 ± 3.50	3.758	<0.05	+
No reaction	*0*	14.7 ± 3.95	19.1 ± 3.73	2.564	<0.05	+
	1 hour	19.0 ± 6.46	24.4 ± 4.88	2.108	<0.05	+
	2 hours	21.0 ± 5.54	29.7 ± 5.17	3.633	<0.05	+
	4 hours	27.9 ± 5.34	34.8 ± 6.97	2.484	<0.05	+
	8 hours	68.9 ± 11.86	79.7 ± 7.53	2.432	<0.05	+

+ = significant - = non-significant

stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [P <0.05] (Table 45).

Those with a moderate reaction ranged from 35% to 52 % with a mean value of 42.1 ± 5.99 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 45).

Weakly stained sperms ranged from 13% to 26 % with a mean value of 19.9 ± 4.43 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined whole non-washed specimens [P < 0.05] (Table 45).

Sperms with a negative reaction ranged from 14% to 25 % with a mean value of 19.1 ± 3.73 %. They were significantly more than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined immediately [P <0.05] (Table 45).

b. Specimens examined after one hour:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 13% to 23 % with a mean value of 18.3 ± 2.98 %. It was significantly less than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P <0.05] (Table 45).

Those with a moderate reaction ranged from 24% to 46 % with a mean value of 33.8 ± 7.1 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 45).

Weakly stained sperms ranged from 15% to 30 % with a mean value of 23.5 ± 5.48 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 45).

Sperms with a negative reaction ranged from 18% to 31 % with a mean value of 24.4 ± 4.88 %. They were significantly more than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 45).

c. Specimens examined after two hours:

A positive succinic dehydrogenase reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 7% to 16 % with a mean value of 11.9 ± 3.03 %. It was significantly less than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 45).

Those with a moderate reaction ranged from 25% to 43 % with a mean value of 34.3 ± 7.24 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 45).

Sperms with a weak reaction ranged from 17% to 32 % with a mean value of 24.1 ± 5.55 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 45).

Sperms with a negative reaction ranged from 23% to 38 % with a mean value of 29.7 ± 5.17 %. This was significantly more than that of negatively stained sperms for

succinic dehydrogenase in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 45).

d. Specimens examined after four hours:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 3% to 15 % with a mean value of 8.6 ± 4.01 %. It was significantly less than that of the strongly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.05] (Table 45).

Those with a moderate reaction ranged from 20% to 39 % with a mean value of 29.9 ± 6.03 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 45).

Weakly stained sperms ranged from 18% to 34 % with a mean value of 26.7 ± 4.52 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 45).

Sperms with a negative reaction ranged from 24% to 44 % with a mean value of 34.8 ± 6.97 %. They were significantly more than the negatively stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after four hours of incubation [P <0.05] (Table 45).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1.5 ± 1.27 %. It was significantly less than that of the strongly

stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P <0.05] (Table 45).

Those with a moderate reaction ranged from 3% to 14 % with a mean value of 7.8 ± 3.61 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 45).

Weakly stained sperms ranged from 6% to 16 % with a mean value of 11 ± 3.5 %. They were significantly less than that of the weakly stained sperms for succinic dehydrogenase in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 45).

Sperms with a negative reaction ranged from 68% to 89 % with a mean value of 79.7 ± 7.53 %. They were significantly more than the negatively stained sperms for succinic dehydrogenase in whole non-washed specimens examined after eight hours of incubation [P <0.05] (Table 45).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens and the period of incubation [r = - 0.891; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens and the period of incubation [r = - 0.869; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the second split fraction specimens

and the period of incubation [r = - 0.490; P <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the second split fraction specimens and the period of incubation [r = 0.938; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added: (Table 46)

a. Immediately examined specimens:

A positive succinic dehydrogenase reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 16% to 25 % with a mean value of 20.4 ± 3.17 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the immediately examined second split fraction specimens [P <0.05] (Table 47).

Those with a moderate reaction ranged from 31% to 51 % with a mean value of 40.8 ± 6.12 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the immediately examined second split fraction specimens [P < 0.05] (Table 47).

Weakly stained sperms ranged from 13% to 31 % with a mean value of 20.8 ± 6.32 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the immediately examined second split fraction specimens [P < 0.05] (Table 47).

Sperms with a negative reaction ranged from 14% to 24 % with a mean value of 18 ± 2.91 %. They did not also differ statistically from the negatively stained sperms for succinic dehydrogenase in the immediately examined second split fraction specimens [P <0.05] (Table 47).

Table (46): Showing the percentage of sperms with different grades of *succinic dehydrogenase in the second split fraction specimens to which calcium was added.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	20	43	22	15	61	24	38	18	20	71	10	24	34	32	81	3	21	29	47	91	0	8	10	82
52	24	42	16	18	62	16	23	28	33	72	8	29	30	33	82	15	37	21	27	92	4	12	17	67
53	19	31	26	24	63	18	39	19	24	73	17	42	21	20	83	11	30	26	33	93	1	8	19	72
54	16	34	31	19	64	13	28	25	34	74	16	39	25	20	84	2	30	20	48	94	0	5	12	83
55	25	46	13	16	65	16	23	29	32	75	15	36	28	21	85	5	34	21	40	95	4	15	18	63
56	23	44	15	18	66	16	36	24	24	76	17	34	26	23	86	5	33	19	43	96	2	12	10	76
57	21	41	21	17	67	21	35	23	21	77	10	26	36	28	87	7	30	25	38	97	0	8	9	83
58	22	51	13	14	68	24	39	17	20	78	13	35	27	25	88	13	41	16	30	98	3	5	21	71
59	18	34	27	21	69	13	27	26	34	79	9	38	29	24	89	12	36	23	29	99	0	7	17	76
60	16	42	24	18	70	19	38	16	27	70	10	30	32	28	80	2	24	29	45	100	3	9	8	80
Mean	20.4	40.8	20.8	18.0	Mean	18.0	32.6	22.5	26.9	Mean	12.5	33.3	28.8	25.4	Mean	7.5	31.6	22.9	38.0	Mean	1.7	8.9	14.1	75.3
S.D.	3.17	6.12	6.32	2.91	S.D.	4.00	6.62	4.70	5.88	S.D.	3.50	5.87	4.44	4.72	S.D.	4.86	5.99	4.31	7.82	S.D.	1.70	3.21	4.77	6.96

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (47): Showing the comparison between the *succinic dehydrogenase* reaction in the sperms of *second split fraction specimens and second split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Significance.
Strong reaction	*0*	18.9 ± 3.14	20.4 ± 3.17	1.063	<0.05	-
	1 hour	18.3 ± 2.98	18.0 ± 4.00	0.190	<0.05	-
	2 hours	11.9 ± 3.03	12.5 ± 3.50	0.409	<0.05	-
	4 hours	8.6 ± 4.01	7.5 ± 4.86	0.552	<0.05	-
	8 hours	1.5 ± 1.27	1.7 ± 1.70	0.298	<0.05	-
Moderate reaction	*0*	42.1 ± 5.99	40.8 ± 6.12	0.480	<0.05	-
	1 hour	33.8 ± 7.10	32.6 ± 6.62	0.391	<0.05	-
	2 hours	34.3 ± 7.24	33.3 ± 5.87	0.339	<0.05	-
	4 hours	29.9 ± 6.03	31.6 ± 5.99	0.633	<0.05	-
	8 hours	7.8 ± 3.61	8.9 ± 3.21	0.719	<0.05	-
Weak reaction	*0*	19.9 ± 4.43	20.8 ± 6.32	0.369	<0.05	-
	1 hour	23.5 ± 5.48	22.5 ± 4.70	0.438	<0.05	-
	2 hours	24.1 ± 5.55	28.8 ± 4.44	2.092	<0.05	-
	4 hours	26.7 ± 4.52	22.9 ± 4.31	1.924	<0.05	-
	8 hours	11.0 ± 3.50	14.1 ± 4.77	1.657	<0.05	-
No reaction	*0*	19.1 ± 3.73	18.0 ± 2.91	0.736	<0.05	-
	1 hour	24.4 ± 4.88	26.9 ± 5.88	1.035	<0.05	-
	2 hours	29.7 ± 5.17	25.4 ± 4.72	1.944	<0.05	-
	4 hours	34.8 ± 6.97	38.0 ± 7.82	0.966	<0.05	-
	8 hours	79.7 ± 7.53	75.3 ± 6.96	1.357	<0.05	-

- = non-significant

b. Specimens examined after one hour:

A similar reaction for succinic dehydrogenase was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 13% to 24 % with a mean value of 18 ± 4 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after one hour of incubation [P <0.05] (Table 47).

Moderately stained sperms ranged from 23% to 39 % with a mean value of 32.6 ± 6.62 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 47).

Those with a weak reaction ranged from 16% to 29 % with a mean value of 22.5 ± 4.7 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 47).

Sperms with a negative reaction ranged from 20% to 34 % with a mean value of 24.9 ± 5.02 %. They did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the second split fraction specimens examined after one hour of incubation [P <0.05] (Table 47).

c. Specimens examined after two hours:

A similar location of succinic dehydrogenase reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 8% to 17 % with a mean value of 12.5 ± 3.5 %. Also, it did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after two hours of incubation [P <0.05] (Table 47).

Those with a moderate reaction ranged from 24% to 42 % with a mean value of 33.3 ± 5.87 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 47).

Sperms with a weak reaction ranged from 21% to 36 % with a mean value of 28.8 ± 4.44 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 47).

Sperms with a negative reaction ranged from 20% to 33 % with a mean value of 25.4 ± 4.72 %. This did not differ statistically from that of negatively stained sperms for succinic dehydrogenase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 47).

d. Specimens examined after four hours:

Also, only the mid-piece was the site of positive succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 2% to 15 % with a mean value of 7.5 ± 4.86 %. They did not differ statistically from the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 47).

Moderately stained sperms ranged from 21% to 41 % with a mean value of 31.6 ± 5.99 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 47).

Those with a weak reaction ranged from 16% to 29 % with a mean value of 22.9 ± 4.31 %. They did not also differ statistically from the weakly stained sperms for

succinic dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 47).

Sperms with a negative reaction ranged from 27% to 48 % with a mean value of 38 ± 7.82 %. Also, they did not differ statistically from negatively stained sperms for succinic dehydrogenase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 47).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of succinic dehydrogenase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 1.7 ± 1.7 %. It did not differ statistically from that of the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 47).

Those with a moderate reaction ranged from 5% to 15 % with a mean value of 8.9 ± 3.21 %. They did not differ statistically from the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 47).

Weakly stained sperms ranged from 8% to 21 % with a mean value of 14.1 ± 4.77 %. They did not also differ statistically from the weakly stained sperms for succinic dehydrogenase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 47).

Sperms with a negative reaction ranged from 63% to 83 % with a mean value of 75.3 ± 6.96 %. Also, they did not differ statistically from the negatively stained sperms for succinic dehydrogenase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 47).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for succinic dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = - 0.868; P <0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for succinic dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = - 0.850; P <0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the weakly stained sperms for succinic dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = - 0.455; P <0.001]. This means that the percent of such weakly stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for succinic dehydrogenase in the second split fraction specimens to which calcium was added and the period of incubation [r = 0.944; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

VI. NON-SPECIFIC ESTERASE:

(A) WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 48)

a. Immediately examined specimens:

A positive reaction for non-specific esterase was observed in the equatorial segment, post-acrosomal region, mid-piece and tail (Fig. 4).

The percentage of spermatozoa giving a strong reaction ranged from 21% to 42 % with a mean value of 32 ± 6.58 %. Those with a moderate reaction

Table (48): Showing the percentage of sperms with different grades of *non-specific esterase in the whole non-washed specimens.*

No.of Ejaculate	Immediate				No.of Ejaculate	1 hour				No.of Ejaculate	2 hours				No.of Ejaculate	4 hours				No.of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	35	31	20	14	11	35	25	22	18	21	26	16	22	36	31	13	27	27	33	41	19	10	35	36
2	33	39	12	16	12	27	30	23	20	22	25	27	23	25	32	23	26	30	21	42	8	18	30	44
3	36	29	14	21	13	22	35	20	23	23	30	25	27	18	33	19	20	32	29	43	20	9	32	39
4	27	45	13	15	14	25	39	17	19	24	24	23	27	26	34	25	27	25	23	44	13	25	26	36
5	21	46	11	22	15	31	18	25	26	25	24	14	28	34	35	16	30	24	30	45	11	22	31	36
6	39	32	9	20	16	26	41	17	16	26	29	18	24	29	36	16	36	17	31	46	6	33	23	38
7	30	37	21	12	17	35	33	20	12	27	28	20	29	23	37	26	23	29	22	47	13	28	25	34
8	24	45	17	14	18	36	20	12	32	28	21	35	25	19	38	15	33	18	34	48	15	30	27	28
9	42	26	15	17	19	33	22	14	31	29	27	30	26	17	39	22	32	20	26	49	9	20	28	43
10	33	28	21	18	20	34	23	13	30	30	22	33	28	17	40	28	21	24	27	50	14	15	33	38
Mean	32.0	35.8	15.3	16.9	Mean	30.4	28.6	18.3	22.7	Mean	25.6	24.1	25.9	24.4	Mean	20.3	27.5	24.6	27.6	Mean	12.8	21.0	29.0	37.2
S.D.	6.58	7.64	4.30	3.31	S.D.	4.99	8.15	4.42	6.85	S.D.	2.95	7.19	2.33	6.93	S.D.	5.21	5.28	5.08	4.58	S.D.	4.52	8.18	3.83	4.52

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Fig. (4) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **non-specific esterase.**

Note the strong (long arrow) and moderate (short arrow) reactions in the equatorial segment, post-acrosomal region, midpiece and tail of sperms.

(Alpha naphthyl acetate method. Proj: 10 Obj: 100)

ranged from 26% to 46 % with a mean value of 35.8 ± 7.64 %. Weakly stained sperms ranged from 9% to 21 % with a mean value of 15.3 ± 4.3 %. Sperms with a negative reaction ranged from 12% to 22 % with a mean value of 16.9 ± 3.31 %.

b. Specimens examined after one hour:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 22% to 36 % with a mean value of 30.4 ± 4.99 %. Moderately stained sperms ranged from 18% to 41 % with a mean value of 28.6 ± 8.15 %. Those with a weak reaction ranged from 12% to 25 % with a mean value of 18.3 ± 4.42 %. Sperms with a negative reaction ranged from 12% to 32 % with a mean value of 22.7 ± 6.85 %.

c. Specimens examined after two hours:

A positive reaction was also detected in the same sites. The percentage of spermatozoa giving a strong reaction ranged from 21% to 30 % with a mean value of 25.6 ± 2.95 %. Those with a moderate reaction ranged from 14% to 35 % with a mean value of 24.1 ± 7.19 %. Sperms with a weak reaction ranged from 22% to 29 % with a mean value of 25.9 ± 2.33 %. Sperms with a negative reaction ranged from 17% to 36 % with a mean value of 24.4 ± 6.93 %.

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 13% to 28 % with a mean value of 20.3 ± 5.21 %. Moderately stained sperms ranged from 20% to 36 % with a mean value of 27.5 ± 5.28 %. Those with a weak reaction ranged from 17% to 32 % with a mean value of 24.6 ± 5.08 %. Sperms with a negative reaction ranged from 21% to 34 % with a mean value of 27.6 ± 4.58 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 6% to 20 % with a mean value of 12.8 ± 4.52 %. Those with a moderate reaction ranged from

9% to 33 % with a mean value of 21 ± 8.18 %. Weakly stained sperms ranged from 23% to 35 % with a mean value of 29 ± 3.83 %. Sperms with a negative reaction ranged from 28% to 44 % with a mean value of 37.2 ± 4.52 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the whole non-washed specimens and the period of incubation [r = - 0.819; P < 0.001]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the whole non-washed specimens and the period of incubation [r = - 0.463; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the whole non-washed specimens and the period of incubation [r = 0.679; P < 0.001]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the whole non-washed specimens and the period of incubation [r = 0.780; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) Washed specimens: (Table 49)

a. Immediately examined specimens:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 22% to 38 % with a mean value of 30 ± 6.06 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined whole non-washed specimens [P <0.05] (Table 50).

Table (49): Showing the percentage of sperms with different grades of *non-specific esterase in the washed specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	38	36	14	12	11	31	34	13	22	21	26	13	30	31	31	20	25	27	28	41	5	22	31	42
2	22	40	12	26	12	23	33	27	17	22	19	27	21	33	32	6	33	24	37	42	8	15	28	49
3	28	34	21	17	13	35	29	23	13	23	30	16	29	25	33	9	28	32	31	43	1	32	20	47
4	30	29	19	22	14	37	17	20	26	24	31	23	23	23	34	13	25	28	34	44	10	18	26	46
5	22	37	23	18	15	38	20	22	20	25	22	24	25	29	35	21	18	33	28	45	6	23	33	38
6	24	38	17	21	16	33	29	13	25	26	24	19	31	26	36	19	20	25	36	46	12	21	24	43
7	35	25	25	15	17	25	31	21	23	27	29	17	26	28	37	12	31	24	33	47	9	20	22	49
8	30	22	24	24	18	26	36	14	24	28	16	30	20	34	38	20	21	29	30	48	3	27	30	40
9	33	31	16	20	19	32	35	15	18	29	24	20	27	29	39	8	22	30	40	49	7	25	24	44
10	38	28	15	19	20	42	28	16	14	30	18	31	24	27	40	21	18	26	35	50	13	16	21	50
Mean	30.0	32.0	18.6	19.4	Mean	32.2	29.2	18.4	20.2	Mean	23.9	22.0	25.6	28.5	Mean	14.9	24.1	27.8	33.2	Mean	7.4	21.9	25.9	44.8
S.D.	6.06	5.96	4.50	4.17	S.D.	6.12	6.29	4.86	4.57	S.D.	5.20	6.06	3.72	3.47	S.D.	5.93	5.26	3.19	3.97	S.D.	3.81	5.17	4.46	4.08

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (50): Showing the comparison between the *non-specific estrase* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Signific-ance.
Strong reaction.	*0*	32.0 ± 6.58	30.0 ± 6.06	0.707	<0.05	-
	1 hour	30.4 ± 4.99	32.2 ± 6.12	0.720	<0.05	-
	2 hours	25.6 ± 2.95	23.9 ± 5.20	0.900	<0.05	-
	4 hours	20.3 ± 5.21	14.9 ± 5.93	2.263	<0.05	+
	8 hours	12.8 ± 4.52	7.4 ± 3.81	2.891	<0.05	+
Moderate reaction	*0*	35.8 ± 7.46	32.0 ± 5.96	1.240	<0.05	-
	1 hour	28.6 ± 8.15	29.2 ± 6.29	1.184	<0.05	-
	2 hours	24.1 ± 7.19	22.0 ± 6.06	0.707	<0.05	-
	4 hours	27.5 ± 5.28	24.1 ± 5.26	1.443	<0.05	-
	8 hours	21.0 ± 8.18	21.9 ± 5.17	0.294	<0.05	-
Weak reaction	*0*	15.3 ± 4.30	18.6 ± 5.50	1.677	<0.05	-
	1 hour	18.3 ± 4.42	18.4 ± 4.86	0.048	<0.05	-
	2 hours	25.9 ± 2.33	25.6 ± 3.72	0.216	<0.05	-
	4 hours	24.6 ± 5.08	27.8 ± 3.19	1.687	<0.05	-
	8 hours	29.0 ± 3.83	25.9 ± 4.46	1.668	<0.05	-
No reaction.	*0*	16.9 ± 3.31	19.4 ± 4.17	1.484	<0.05	-
	1 hour	22.7 ± 6.85	20.2 ± 4.57	0.961	<0.05	-
	2 hours	24.4 ± 6.93	28.5 ± 3.47	1.672	<0.05	-
	4 hours	27.6 ± 4.58	33.2 ± 3.97	2.925	<0.05	+
	8 hours	37.2 ± 4.52	44.8 ± 4.08	3.950	<0.001	+++

- = non-significant + = significant +++ = highly significant

Those with a moderate reaction ranged from 22% to 40 % with a mean value of 32 ± 5.96 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [P < 0.05] (Table 50).

Weakly stained sperms ranged from 12% to 25 % with a mean value of 18.6 ± 4.5 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [P < 0.05] (Table 50).

Sperms with a negative reaction ranged from 12% to 26 % with a mean value of 19.4 ± 4.17 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [P < 0.05] (Table 50).

b. Specimens examined after one hour:

A similar reaction was noticed in the same sites. The percentage of spermatozoa giving a strong reaction ranged from 23% to 42 % with a mean value of 32.2 ± 6.12 %. This did not differ statistically from that of the strongly stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 50).

Moderately stained sperms ranged from 17% to 36 % with a mean value of 29.2 ± 6.29 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 50).

Those with a weak reaction ranged from 13% to 27 % with a mean value of 18.4 ± 4.86 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 50).

Sperms with a negative reaction ranged from 13% to 26 % with a mean value of 20.2 ± 4.57 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [P <0.05] (Table 50).

c. Specimens examined after two hours:

Similar locations of non-specific esterase reaction were detected. The percentage of spermatozoa giving a strong reaction ranged from 16% to 31 % with a mean value of 23.9 ± 5.2 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 50).

Those with a moderate reaction ranged from 13% to 31 % with a mean value of 22 ± 6.06 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 50).

Sperms with a weak reaction ranged from 20% to 31 % with a mean value of 25.6 ± 3.72 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 50).

Sperms with a negative reaction ranged from 23% to 34 % with a mean value of 28.5 ± 3.47 %. This did not also differ statistically from that of negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 50).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 6% to 21 % with a mean value of 14.9 ± 5.93 %. They were significantly less than the strongly stained sperms for in

the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 50).

Moderately stained sperms ranged from 18% to 33 % with a mean value of 24.1 ± 5.26 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 50).

Those with a weak reaction ranged from 24% to 33 % with a mean value of 27.8 ± 3.19 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 50).

Sperms with a negative reaction ranged from 28% to 40 % with a mean value of 33.2 ± 3.97 %. They were significantly more than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 50).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 1% to 13 % with a mean value of 7.4 ± 3.81 %. It was significantly less than that of the strongly stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 50).

Those with a moderate reaction ranged from 15% to 32 % with a mean value of 21.9 ± 5.17 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 50).

Weakly stained sperms ranged from 20% to 33 % with a mean value of 25.9 ± 4.46 %. They did not also differ statistically from the weakly stained sperms

for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 50).

Sperms with a negative reaction ranged from 38% to 50 % with a mean value of 44.8 ± 4.08 %. They were highly significantly more than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 50).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the washed specimens and the period of incubation [$r = - 0.837$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the washed specimens and the period of incubation [$r = - 0.444$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the washed specimens and the period of incubation [$r = 0.493$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the washed specimens and the period of incubation [$r = 0.910$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 51).

a. Immediately examined specimens:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 23% to 34 % with a mean value of

Table (51): Showing the percentage of sperms with different grades of *non-specific esterase in the washed specimens to which calcium was added.*

No.of Ejaculate	Immediate				No.of Ejaculate	1 hour				No.of Ejaculate	2 hours				No.of Ejaculate	4 hours				No.of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	26	42	19	13	11	26	30	17	27	21	21	23	28	28	31	9	35	31	25	41	12	20	25	43
2	34	34	14	18	12	38	26	18	18	22	32	18	21	29	32	17	29	22	32	42	2	23	33	42
3	29	40	16	15	13	21	45	25	9	23	28	25	24	23	33	12	17	34	37	43	11	22	28	39
4	26	30	21	23	14	25	30	20	25	24	30	30	22	18	34	11	25	29	35	44	12	21	32	35
5	23	32	23	22	15	41	32	14	13	25	22	31	28	19	35	13	24	36	27	45	1	20	30	49
6	24	40	25	11	16	32	34	17	17	26	18	26	27	29	36	15	22	39	24	46	8	29	26	37
7	27	37	22	14	17	30	27	22	21	27	33	21	25	21	37	10	37	27	26	47	3	18	28	51
8	30	44	17	9	18	34	41	15	10	28	20	18	29	33	38	8	35	24	33	48	8	17	29	46
9	32	36	15	17	19	20	38	19	23	29	27	24	23	26	39	12	32	28	28	49	3	29	31	37
10	24	35	21	20	20	23	30	23	24	30	19	29	27	25	40	16	17	33	34	50	3	30	27	40
Mean	27.5	37.0	19.3	16.2	Mean	29.0	33.3	19.0	18.7	Mean	25.0	24.5	25.4	25.1	Mean	12.3	27.3	30.3	30.1	Mean	6.3	22.9	28.9	41.9
S.D.	3.66	4.47	3.68	4.64	S.D.	7.20	6.20	3.53	6.38	S.D.	5.64	4.65	2.80	4.84	S.D.	2.98	7.41	5.33	4.63	S.D.	4.37	4.77	2.60	5.36

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (52): Showing the comparison between the *non-specific estrase* reaction in the sperms of *washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Calcium	t	p	Significance.
Strong reaction	*0*	30.0 ± 6.06	27.5 ± 3.66	1.117	<0.05	-
	1 hour	32.2 ± 6.12	29.0 ± 7.20	1.071	<0.05	-
	2 hours	23.9 ± 5.20	25.0 ± 5.64	0.454	<0.05	-
	4 hours	14.9 ± 5.93	12.3 ± 2.98	1.238	<0.05	-
	8 hours	7.4 ± 3.81	6.3 ± 4.37	0.600	<0.05	-
Moderate reaction	*0*	32.0 ± 5.96	37.0 ± 4.47	1.697	<0.05	-
	1 hour	29.2 ± 6.29	33.3 ± 6.2	1.468	<0.05	-
	2 hours	22.0 ± 6.06	24.5 ± 4.65	1.036	<0.05	-
	4 hours	24.1 ± 5.26	27.3 ± 7.41	1.114	<0.05	-
	8 hours	21.9 ± 5.17	22.9 ± 4.77	0.449	<0.05	-
Weak reaction	*0*	18.6 ± 5.50	19.3 ± 3.68	0.924	<0.05	-
	1 hour	18.4 ± 4.86	19.0 ± 3.53	0.316	<0.05	-
	2 hours	25.6 ± 3.72	25.4 ± 2.80	0.136	<0.05	-
	4 hours	27.8 ± 3.19	30.3 ± 5.33	1.272	<0.05	-
	8 hours	25.9 ± 4.46	28.9 ± 2.60	1.838	<0.05	-
No reaction	*0*	19.4 ± 4.17	16.2 ± 4.64	1.623	<0.05	-
	1 hour	20.2 ± 4.57	18.7 ± 6.38	0.605	<0.05	-
	2 hours	28.5 ± 3.47	25.1 ± 4.84	1.805	<0.05	-
	4 hours	33.2 ± 3.97	30.1 ± 4.63	1.608	<0.05	-
	8 hours	44.8 ± 4.08	41.9 ± 5.36	1.361	<0.05	-

- = insignificant

27.5 ± 3.66 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 52).

Those with a moderate reaction ranged from 30% to 44 % with a mean value of 37 ± 4.47 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined washed specimens [P < 0.05] (Table 52).

Weakly stained sperms ranged from 14% to 25 % with a mean value of 19.3 ± 3.68 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined washed specimens [P < 0.05] (Table 52).

Sperms with a negative reaction ranged from 9% to 23 % with a mean value of 16.2 ± 4.64 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the immediately examined washed specimens [P < 0.05] (Table 52).

b. Specimens examined after one hour:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 20% to 41 % with a mean value of 29 ± 7.2 %. This did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [P < 0.05] (Table 52).

Moderately stained sperms ranged from 26% to 45 % with a mean value of 33.3 ± 6.2 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after one hour of incubation [P < 0.05] (Table 52).

Those with a weak reaction ranged from 14% to 25 % with a mean value of 19 ± 3.53 %. They did not also differ statistically from the weakly stained sperms for

non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 52).

Sperms with a negative reaction ranged from 9% to 27 % with a mean value of 18.7 ± 6.38 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 52).

c. Specimens examined after two hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 18% to 33 % with a mean value of 25 ± 5.64 %. Also, it did not differ statistically from that of the strongly stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 52).

Those with a moderate reaction ranged from 18% to 31 % with a mean value of 24.5 ± 4.65 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 52).

Sperms with a weak reaction ranged from 21% to 29 % with a mean value of 25.4 ± 2.8 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 52).

Sperms with a negative reaction ranged from 18% to 33 % with a mean value of 25.1 ± 4.84 %. This did not differ statistically from that of negatively stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 52).

d. Specimens examined after four hours:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa

giving a strong reaction ranged from 8% to 17 % with a mean value of 12.3 ± 2.98 %. They did not differ statistically from the strongly stained sperms in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 52).

Moderately stained sperms ranged from 17% to 37 % with a mean value of 27.3 ± 7.41 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 52).

Those with a weak reaction ranged from 22% to 39 % with a mean value of 30.3 ± 5.33 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 52).

Sperms with a negative reaction ranged from 24% to 37 % with a mean value of 30.1 ± 4.63 %. Also, they did not differ statistically from the negatively stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 52).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 1% to 12 % with a mean value of 6.3 ± 4.37 %. It did not differ statistically from that of the strongly stained sperms for in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 52).

Those with a moderate reaction ranged from 17% to 30 % with a mean value of 22.9 ± 4.77 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 52).

Weakly stained sperms ranged from 25% to 33 % with a mean value of 28.9 ± 2.6 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 52).

Sperms with a negative reaction ranged from 35% to 51 % with a mean value of 41.9 ± 5.36 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 52).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the washed specimens to which calcium was added and the period of incubation [$r = - 0.843$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the washed specimens to which calcium was added and the period of incubation [$r = - 0.565$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the washed specimens to which calcium was added and the period of incubation [$r = 0.463$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the washed specimens to which calcium was added and the period of incubation [$r = 0.873$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(4) **Washed specimens to which ascorbic acid was added:** (Table 53)

a. Immediately examined specimens:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 19% to 40 % with a mean value of 28.9 ± 6.81 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [$P < 0.05$] (Table 54).

Those with a moderate reaction ranged from 22% to 44 % with a mean value of 33.2 ± 7.18 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined washed specimens [$P < 0.05$] (Table 54).

Weakly stained sperms ranged from 10% to 25 % with a mean value of 17.2 ± 5.07 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens immediately examined [$P < 0.05$] (Table 54).

Sperms with a negative reaction ranged from 13% to 28 % with a mean value of 20.7 ± 5.27 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the immediately examined washed specimens [$P < 0.05$] (Table 54).

b. Specimens examined after one hour:

A similar reaction was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 15% to 38 % with a mean value of 29.1 ± 7.23 %. Also, this did not differ statistically from that of the strongly stained sperms for non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 54).

Moderately stained sperms ranged from 23% to 40 % with a mean value of 31 ± 6.25 %. They did not differ statistically from the moderately stained sperms for

Table (53): Showing the percentage of sperms with different grades of *non-specific esterase reaction in the washed specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	34	41	10	15	11	25	35	14	26	21	20	32	27	21	31	21	30	20	29	41	15	14	27	44
2	27	30	16	27	12	15	40	15	30	22	18	37	20	25	32	22	27	24	27	42	6	20	38	36
3	40	22	25	13	13	22	38	17	23	23	38	17	25	20	33	17	24	26	33	43	13	9	35	43
4	37	25	23	15	14	38	23	24	15	24	31	15	24	30	34	32	17	31	20	44	12	13	36	39
5	27	35	15	23	15	36	25	22	17	25	36	16	26	22	35	17	33	22	28	45	16	26	24	34
6	33	31	18	18	16	27	37	17	19	26	22	27	23	28	36	19	38	19	24	46	6	27	29	38
7	26	28	21	25	17	34	27	23	16	27	17	39	18	26	37	14	35	25	26	47	8	23	40	29
8	19	39	20	22	18	36	24	13	27	28	38	17	26	19	38	29	21	15	35	48	14	16	35	35
9	22	37	13	28	19	30	29	21	20	29	29	21	19	31	39	15	35	18	32	49	15	15	25	45
10	24	44	11	21	20	28	32	12	28	30	21	30	22	27	40	29	21	29	21	50	11	21	37	31
Mean	28.9	33.2	17.2	20.7	Mean	29.1	31.0	17.8	22.1	Mean	27.0	25.1	23.0	24.9	Mean	21.5	28.1	22.9	27.5	Mean	11.6	18.4	32.6	37.4
S.D.	6.81	7.18	5.07	5.27	S.D.	7.23	6.25	4.39	5.43	S.D.	8.39	9.09	3.16	4.23	S.D.	6.40	7.17	5.04	4.97	S.D.	3.75	5.93	5.80	5.44

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (54): Showing the comparison between the *non-specific estrase* reaction in the sperms of *washed specimens* and *washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	30.0 ± 6.06	28.9 ± 6.81	0.382	<0.05	-
	1 hour	32.2 ± 6.12	29.1 ± 7.23	1.034	<0.05	-
	2 hours	23.9 ± 5.20	27.0 ± 8.39	0.993	<0.05	-
	4 hours	14.9 ± 5.93	21.5 ± 6.40	2.392	<0.05	+
	8 hours	7.4 ± 3.81	11.6 ± 3.75	2.486	<0.05	+
Moderate reaction	*0*	32.0 ± 5.96	33.2 ± 7.18	0.407	<0.05	-
	1 hour	29.2 ± 6.29	31.0 ± 6.25	0.642	<0.05	-
	2 hours	22.0 ± 6.06	25.1 ± 9.09	0.898	<0.05	-
	4 hours	24.1 ± 5.26	28.1 ± 7.17	1.422	<0.05	-
	8 hours	21.9 ± 5.17	18.4 ± 5.93	1.407	<0.05	-
Weak reaction	*0*	18.6 ± 5.50	17.2 ± 5.07	0.653	<0.05	-
	1 hour	18.4 ± 4.86	17.8 ± 4.39	0.290	<0.05	-
	2 hours	25.6 ± 3.72	23.0 ± 3.16	1.685	<0.05	-
	4 hours	27.8 ± 3.19	22.9 ± 5.04	2.597	<0.05	+
	8 hours	25.9 ± 4.46	32.6 ± 5.80	2.897	<0.05	+
No reaction	*0*	19.4 ± 4.17	20.7 ± 5.27	0.612	<0.05	-
	1 hour	20.2 ± 4.57	22.1 ± 5.43	0.847	<0.05	-
	2 hours	28.5 ± 3.47	24.9 ± 4.23	2.081	<0.05	-
	4 hours	33.2 ± 3.97	27.5 ± 4.97	2.834	<0.05	+
	8 hours	44.8 ± 4.08	37.4 ± 5.44	3.442	<0.05	+

+ = significant - = non-significant

non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 54).

Those with a weak reaction ranged from 12% to 24 % with a mean value of 17.8 ± 4.39 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 54).

Sperms with a negative reaction ranged from 15% to 30 % with a mean value of 22.1 ± 5.43 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 54).

c. Specimens examined after two hours:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 17% to 38 % with a mean value of 27 ± 8.39 %. Also, it did not differ statistically from that of the strongly stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 54).

Those with a moderate reaction ranged from 15% to 39 % with a mean value of 25.1 ± 9.09 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 54).

Sperms with a weak reaction ranged from 18% to 27 % with a mean value of 23 ± 3.16 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 54).

Sperms with a negative reaction ranged from 19% to 31 % with a mean value of 24.9 ± 4.23 %. They did not differ statistically from the negatively stained sperms

for non-specific esterase in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 54).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 14% to 32 % with a mean value of 21.5 ± 6.4 %. This was significantly more than that of the strongly stained sperms for in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 54).

Moderately stained sperms ranged from 17% to 38 % with a mean value of 28.1 ± 7.17 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 54).

Those with a weak reaction ranged from 15% to 31 % with a mean value of 22.9 ± 5.04 %. They were significantly less than that of the weakly stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 54).

Sperms with a negative reaction ranged from 20% to 35 % with a mean value of 27.5 ± 4.97 %. This was significantly less than that of negatively stained sperms for non-specific esterase in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 54).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 6% to 16 % with a mean value of 11.6 ± 3.75 %. It was significantly more than that of the strongly stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 54).

Those with a moderate reaction ranged from 9% to 27 % with a mean value of 18.4 ± 5.93 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 54).

Weakly stained sperms ranged from 24% to 40 % with a mean value of 32.6 ± 5.8 %. They were significantly more than that of the weakly stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 54).

Sperms with a negative reaction ranged from 29% to 45 % with a mean value of 37.4 ± 5.44 %. They were significantly less than the negatively stained sperms for non-specific esterase in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 54).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.713$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.550$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the washed specimens to which ascorbic acid was added and the period of incubation [$r = 0.750$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the washed specimens to which

ascorbic acid was added and the period of incubation [r = 0.771; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(B) SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 55)

a. Immediately examined specimens:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 22% to 42 % with a mean value of 31.2 ± 6.97 %. They did not differ statistically from the strongly stained sperms in the whole non-washed specimens examined immediately [P < 0.05] (Table 56).

Moderately stained sperms ranged from 25% to 49 % with a mean value of 35.7 ± 7.76 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [P < 0.05] (Table 56).

Those with a weak reaction ranged from 13% to 23 % with a mean value of 18.7 ± 3.5 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [P < 0.05] (Table 56).

Sperms with a negative reaction ranged from 10% to 18 % with a mean value of 14.4 ± 2.8 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined immediately [P < 0.05] (Table 56).

b. Specimens examined after one hour:

No difference was noticed as regards the sites of non-specific esterase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 24% to 42 % with a mean value of 32.9 ± 6.3 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 56).

Table (55): Showing the percentage of sperms with different grades of *non-specific esterase reaction in the first split fraction specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	35	29	23	13	61	35	27	17	21	71	33	25	23	19	81	29	18	29	24	91	20	19	33	28
52	25	45	16	14	62	40	22	12	26	72	25	26	29	20	82	21	32	30	17	92	13	24	28	35
53	42	25	22	11	63	24	35	27	14	73	31	28	19	22	83	28	22	32	18	93	18	8	36	38
54	34	33	21	12	64	26	32	23	19	74	32	18	25	25	84	18	34	24	24	94	20	12	34	34
55	32	36	15	17	65	25	33	26	16	75	33	21	23	23	85	32	13	30	25	95	21	13	39	27
56	26	38	18	18	66	42	17	28	13	76	30	34	19	17	86	20	30	23	27	96	12	24	29	35
57	33	34	17	16	67	31	28	21	20	77	24	31	27	18	87	25	32	26	17	97	23	11	36	30
58	22	49	13	16	68	37	27	14	22	78	27	26	28	19	88	22	27	28	23	98	13	20	30	37
59	40	27	23	10	69	33	21	22	24	79	25	32	21	22	89	30	15	25	30	99	17	23	31	29
60	23	41	19	17	70	36	22	24	18	70	28	24	26	22	80	26	26	31	17	100	14	25	28	33
Mean	31.2	35.7	18.7	14.4	Mean	32.9	26.4	21.4	19.3	Mean	28.8	26.5	24.0	20.7	Mean	25.1	24.9	27.8	22.2	Mean	17.1	17.9	32.4	32.6
S.D.	6.97	7.76	3.50	2.80	S.D.	6.30	5.85	5.46	4.19	S.D.	3.46	4.95	3.59	2.50	S.D.	4.70	7.53	3.12	4.69	S.D.	3.90	6.33	3.81	3.86

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (56): Showing the comparison between the *non-specific estrase* reaction in the sperms of *whole non-washed* and *first split fraction specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Signific-ance.
Strong reaction	*0*	32.0 ± 6.58	31.2 ± 6.97	0.264	<0.05	-
	1 hour	30.4 ± 4.99	32.9 ± 6.30	0.984	<0.05	-
	2 hours	25.6 ± 2.95	28.8 ± 3.46	2.226	<0.05	+
	4 hours	20.3 ± 5.21	25.1 ± 4.70	2.164	<0.05	+
	8 hours	12.8 ± 4.52	17.1 ± 3.90	2.279	<0.05	+
Moderate reaction	*0*	35.8 ± 7.46	35.7 ± 7.76	0.029	<0.05	-
	1 hour	28.6 ± 8.15	26.4 ± 5.85	0.693	<0.05	-
	2 hours	24.1 ± 7.19	26.5 ± 4.95	0.870	<0.05	-
	4 hours	27.5 ± 5.28	24.9 ± 7.53	0.894	<0.05	-
	8 hours	21.0 ± 8.18	17.9 ± 6.33	0.948	<0.05	-
Weak reaction	*0*	15.3 ± 4.30	18.7 ± 3.50	1.941	<0.05	-
	1 hour	18.3 ± 4.42	21.4 ± 5.46	1.395	<0.05	-
	2 hours	25.9 ± 2.33	24.0 ± 3.59	1.404	<0.05	-
	4 hours	24.6 ± 5.08	27.8 ± 3.12	1.697	<0.05	-
	8 hours	29.0 ± 3.83	32.4 ± 3.81	1.991	<0.05	-
No reaction	*0*	16.9 ± 3.31	14.4 ± 2.80	1.823	<0.05	-
	1 hour	22.7 ± 6.85	19.3 ± 4.19	1.339	<0.05	-
	2 hours	24.4 ± 6.93	20.7 ± 2.50	1.588	<0.05	-
	4 hours	27.6 ± 4.58	22.2 ± 4.69	2.608	<0.05	+
	8 hours	37.2 ± 4.52	32.6 ± 3.86	2.447	<0.05	+

+ = significant - = non-significant

Those with a moderate reaction ranged from 17% to 35 % with a mean value of 26.4 ± 5.85 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 56).

Weakly stained sperms ranged from 12% to 28 % with a mean value of 21.4 ± 5.46 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 56).

Sperms with a negative reaction ranged from 13% to 26 % with a mean value of 19.3 ± 4.19 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 56).

c. Specimens examined after two hours:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 24% to 33 % with a mean value of 28.8 ± 3.46 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 56).

Those with a moderate reaction ranged from 18% to 34 % with a mean value of 26.5 ± 4.95 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 56).

Sperms with a weak reaction ranged from 19% to 29 % with a mean value of 24 ± 3.59 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 56).

Sperms with a negative reaction ranged from 17% to 25 % with a mean value of 20.7 ± 2.5 %. This did not differ significantly from that of the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 56).

d. Specimens examined after four hours:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 18% to 32 % with a mean value of 25.1 ± 4.7 %. It was significantly more than that of the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 56).

Those with a moderate reaction ranged from 13% to 34 % with a mean value of 24.9 ± 7.53 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 56).

Weakly stained sperms ranged from 23% to 32 % with a mean value of 27.8 ± 3.12 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 56).

Sperms with a negative reaction ranged from 17% to 30 % with a mean value of 22.2 ± 4.69 %. they was significantly less than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 56).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of non-specific esterase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 12% to 23 % with a mean value of 17.1 ± 3.9 %. It was significantly more than

that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 56).

Those with a moderate reaction ranged from 8% to 25 % with a mean value of 17.9 ± 6.33 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 56).

Weakly stained sperms ranged from 28% to 39 % with a mean value of 32.4 ± 3.81 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 56).

Sperms with a negative reaction ranged from 27% to 38 % with a mean value of 32.6 ± 3.86 %. They were significantly less than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 56).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the first split fraction specimens and the period of incubation [$r = - 0.832$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the first split fraction specimens and the period of incubation [$r = - 0.604$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the first split fraction specimens and the period of incubation [$r = 0.773$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the first split fraction specimens and the period of incubation [$r = 0.843$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added: (Table 57)

a. Immediately examined specimens:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 21% to 37 % with a mean value of 28.7 ± 5.46 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined first split fraction specimens [$P < 0.05$] (Table 58).

Those with a moderate reaction ranged from 22% to 46 % with a mean value of 33.9 ± 7.99 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 58).

Weakly stained sperms ranged from 16% to 28 % with a mean value of 21.3 ± 3.83 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 58).

Sperms with a negative reaction ranged from 7% to 27 % with a mean value of 16.1 ± 6.94 %. They did not also differ statistically from the negatively stained sperms for non-specific esterase in the immediately examined first split fraction specimens [$P < 0.05$] (Table 58).

b. Specimens examined after one hour:

A similar non-specific esterase reaction was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 22% to 42 % with a mean value of 30.3 ± 6.22 %.

Table (57): Showing the percentage of sperms with different grades of *non-specific esterase reaction in the first split fraction specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	22	46	25	7	61	29	32	21	18	71	29	27	19	25	81	25	17	29	29	91	25	27	24	24
52	30	33	21	16	62	36	31	23	10	72	32	26	26	16	82	23	24	27	26	92	18	32	19	31
53	21	24	28	27	63	33	21	25	21	73	25	27	28	20	83	28	37	18	17	93	23	16	27	34
54	24	43	18	15	64	25	36	22	17	74	37	20	16	27	84	26	20	29	25	94	25	20	25	30
55	31	40	22	7	65	22	28	24	26	75	31	34	20	15	85	31	24	22	23	95	30	17	30	23
56	37	28	24	11	66	28	27	26	19	76	23	36	32	9	86	32	22	25	21	96	17	32	20	31
57	28	35	19	18	67	31	34	23	12	77	28	35	19	18	87	29	26	27	18	97	29	18	27	26
58	33	30	23	14	68	23	39	15	23	78	33	22	22	23	88	36	18	24	22	98	18	28	21	33
59	35	22	17	26	69	42	18	26	14	79	35	33	21	11	89	24	34	23	19	99	22	31	22	25
60	26	38	16	20	70	34	20	22	24	70	26	20	30	24	80	31	29	26	14	100	19	33	19	29
Mean	28.7	33.9	21.3	16.1	Mean	30.3	28.6	22.7	18.4	Mean	29.9	28.0	23.3	18.8	Mean	28.5	25.1	25.0	21.4	Mean	22.6	25.4	23.4	28.6
S.D.	5.46	7.99	3.83	6.94	S.D.	6.22	7.12	3.20	5.27	S.D.	4.51	6.18	5.36	6.07	S.D.	4.09	6.59	3.40	4.55	S.D.	4.65	6.90	3.81	3.86

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (58): Showing the comparison between the *non-specific estrase* reaction in the sperms of *first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	31.2 ± 6.97	28.7 ± 5.46	0.893	<0.05	-
	1 hour	32.9 ± 6.30	30.3 ± 6.22	0.929	<0.05	-
	2 hours	28.8 ± 3.46	29.9 ± 4.51	0.612	<0.05	-
	4 hours	25.1 ± 4.70	28.5 ± 4.09	1.726	<0.05	-
	8 hours	17.1 ± 3.90	22.6 ± 4.65	2.867	<0.05	+
Moderate reaction	*0*	35.7 ± 7.76	33.9 ± 7.99	0.511	<0.05	-
	1 hour	26.4 ± 5.85	28.6 ± 7.12	0.755	<0.05	-
	2 hours	26.5 ± 4.95	28.0 ± 6.18	0,599	<0.05	-
	4 hours	24.9 ± 7.53	25.1 ± 6.59	0.063	<0.05	-
	8 hours	17.9 ± 6.33	25.4 ± 6.90	2.533	<0.05	+
Weak reaction	*0*	18.7 ± 3.50	21.3 ± 3.83	1.585	<0.05	-
	1 hour	21.4 ± 5.46	22.7 ± 3.20	0.650	<0.05	-
	2 hours	24.0 ± 3.59	23.3 ± 5.36	0.343	<0.05	-
	4 hours	27.8 ± 3.12	25.0 ± 3.40	1.919	<0.05	-
	8 hours	32.4 ± 3.81	23.4 ± 3.81	5.287	<0.001	+++
No reaction	*0*	14.4 ± 2.80	16.1 ± 6.94	0.719	<0.05	-
	1 hour	19.3 ± 4.19	18.4 ± 5.27	0.422	<0.05	-
	2 hours	20.7 ± 2.50	18.8 ± 6.07	0.915	<0.05	-
	4 hours	22.2 ± 4.69	21.4 ± 4.55	0.387	<0.05	-
	8 hours	32.6 ± 3.86	28.6 ± 3.86	2.315	<0.05	+

+++ = highly significant + = significant - = non-significant

Also, this did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 58).

Moderately stained sperms ranged from 18% to 39 % with a mean value of 28.6 ± 7.12 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 58).

Those with a weak reaction ranged from 15% to 26 % with a mean value of 22.7 ± 3.2 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 58).

Sperms with a negative reaction ranged from 10% to 26 % with a mean value of 18.4 ± 5.27 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 58).

c. Specimens examined after two hours:

Similar locations of reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 23% to 37 % with a mean value of 29.9 ± 4.51 %. Also, it did not differ statistically from that of the strongly stained sperms for non-specific esterase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 58).

Those with a moderate non-specific esterase reaction ranged from 20% to 36 % with a mean value of 28 ± 6.18 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 58).

Sperms with a weak reaction ranged from 16% to 32 % with a mean value of 23.3 ± 5.36 %. They did not also differ statistically from the weakly stained sperms

for non-specific esterase in the first split fraction specimens examined after two hours of incubation [$P \leq 0.05$] (Table 58).

Sperms with a negative reaction ranged from 9% to 27 % with a mean value of 18.8 ± 6.07 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 58).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 23% to 36 % with a mean value of 28.5 ± 4.09 %. This did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 58).

Moderately stained sperms ranged from 17% to 37 % with a mean value of 25.1 ± 6.59 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 58).

Those with a weak reaction ranged from 18% to 29 % with a mean value of 25 ± 3.4 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 58).

Sperms with a negative reaction ranged from 14% to 29 % with a mean value of 21.4 ± 4.55 %. This did not also differ statistically from that of negatively stained sperms for non-specific esterase in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 58).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of non-specific esterase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from

17% to 30 % with a mean value of 22.6 ± 4.65 %. It was significantly more than that of the strongly stained sperms for non-specific esterase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 58).

Those with a moderate reaction ranged from 16% to 33 % with a mean value of 25.4 ± 6.9 %. They were significantly more than that of the moderately stained sperms for non-specific esterase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 58).

Weakly stained sperms ranged from 19% to 30 % with a mean value of 23.4 ± 3.81 %. They were significantly less than that of the weakly stained sperms for non-specific esterase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 58).

Sperms with a negative reaction ranged from 23% to 34 % with a mean value of 28.6 ± 3.86 %. They were significantly less than the negatively stained sperms for non-specific esterase in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 58).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.448$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.335$; $P < 0.05$]. This means that the percent of such moderately stained sperms decreased with time.

A non-significant positive correlation was found between the percent of the weakly stained sperms for non-specific esterase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.167$; $P < 0.05$].

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.636$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 59)

a. Immediately examined specimens:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 22% to 35 % with a mean value of 28 ± 4.32 %. It did not differ statistically from the strongly stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 60).

Moderately stained sperms ranged from 28% to 50 % with a mean value of 38.6 ± 7.43 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 60).

Those with a weak reaction ranged from 13% to 21 % with a mean value of 18 ± 2.71 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 60).

Sperms with a negative reaction ranged from 9% to 23 % with a mean value of 15.4 ± 4.06 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 60).

b. Specimens examined after one hour:

No difference was noticed as regards the sites of non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 18% to 42 % with a mean value of 28.5 ± 8.28 %. It did not also differ statistically from that of the

Table (59): Showing the percentage of sperms with different grades of *non-specific esterase reaction in the second split fraction specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	26	43	15	16	61	26	34	22	18	71	13	26	26	35	81	13	33	23	31	91	12	28	24	36
52	35	28	19	18	62	31	31	23	15	72	27	20	23	30	82	11	36	25	28	92	4	20	34	42
53	28	44	18	10	63	19	44	16	21	73	30	17	16	37	83	19	26	24	31	93	6	25	30	39
54	22	50	16	12	64	30	29	22	19	74	20	32	22	26	84	20	22	26	32	94	10	11	30	49
55	31	32	13	24	65	42	22	26	10	75	18	35	24	23	85	17	16	30	37	95	10	15	28	47
56	24	41	20	15	66	27	34	17	22	76	24	27	28	21	86	4	34	28	34	96	5	16	39	40
57	33	29	21	17	67	32	20	24	24	77	19	32	17	32	87	11	21	31	37	97	8	15	27	50
58	23	46	20	11	68	20	37	17	26	78	22	30	19	29	88	13	27	27	33	98	11	12	36	41
59	30	36	17	17	69	40	26	18	16	79	2	35	29	24	89	22	17	25	36	99	7	13	41	39
60	28	37	21	14	70	18	32	21	29	70	15	34	26	25	80	18	15	29	38	100	11	17	35	37
Mean	28.0	38.6	18.0	15.4	Mean	28.5	30.9	20.6	20.0	Mean	20.0	28.8	23.0	28.2	Mean	14.8	24.7	26.8	33.7	Mean	8.4	17.2	32.4	42.0
S.D.	4.32	7.43	2.71	4.06	S.D.	8.28	7.11	3.41	5.62	S.D.	5.89	6.27	4.50	5.31	S.D.	5.41	7.75	2.66	3.27	S.D.	2.80	5.57	5.48	4.97

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (60): Showing the comparison between the *non-specific estrase* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Significance.
Strong reaction	*0*	32.0 ± 6.58	28.0 ± 4.32	1.606	<0.05	-
	1 hour	30.4 ± 4.99	28.5 ± 8.28	0.622	<0.05	-
	2 hours	25.6 ± 2.95	20.0 ± 5.89	2.689	<0.05	+
	4 hours	20.3 ± 5.21	14.8 ± 5.41	2.316	<0.05	+
	8 hours	12.8 ± 4.52	8.4 ± 2.80	2.619	<0.05	+
Moderate reaction	*0*	35.8 ± 7.46	38.6 ± 7.43	0.831	<0.05	-
	1 hour	28.6 ± 8.15	30.9 ± 7.11	0.672	<0.05	-
	2 hours	24.1 ± 7.19	28.8 ± 6.27	1.559	<0.05	-
	4 hours	27.5 ± 5.28	24.7 ± 7.75	0.945	<0.05	-
	8 hours	21.0 ± 8.18	17.2 ± 5.57	1.214	<0.05	-
Weak reaction	*0*	15.3 ± 4.30	18.0 ± 2.71	1.681	<0.05	-
	1 hour	18.3 ± 4.42	20.6 ± 3.41	1.303	<0.05	-
	2 hours	25.9 ± 2.33	23.0 ± 4.5	1.811	<0.05	-
	4 hours	24.6 ± 5.08	26.8 ± 2.66	1.213	<0.05	-
	8 hours	29.0 ± 3.83	32.4 ± 5.48	1.608	<0.05	-
No reaction	*0*	16.9 ± 3.31	15.4 ± 4.06	0.905	<0.05	-
	1 hour	22.7 ± 6.85	20.0 ± 5.62	0.964	<0.05	-
	2 hours	24.4 ± 6.93	28.2 ± 5.31	1.376	<0.05	-
	4 hours	27.6 ± 4.58	33.7 ± 3.27	3.431	<0.05	+
	8 hours	37.2 ± 4.52	42.0 ± 4.97	2.261	<0.05	+

+ = significant - = non-significant

strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 60).

Those with a moderate reaction ranged from 20% to 44 % with a mean value of 30.9 ± 7.11 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 60).

Weakly stained sperms ranged from 16% to 26 % with a mean value of 20.6 ± 3.41 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 60).

Sperms with a negative reaction ranged from 10% to 29 % with a mean value of 20 ± 5.62 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 60).

c. Specimens examined after two hours:

A positive reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 12% to 30 % with a mean value of 20 ± 5.89 %. It was significantly less than that of the strongly stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 60).

Those with a moderate reaction ranged from 17% to 35 % with a mean value of 28.8 ± 6.27 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 60).

Sperms with a weak reaction ranged from 16% to 29 % with a mean value of 23 ± 4.5 %. They did not also differ statistically from the weakly stained sperms for

non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 60).

Sperms with a negative reaction ranged from 21% to 37 % with a mean value of 28.2 ± 5.31 %. This did not differ statistically from that of negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 60).

d. Specimens examined after four hours:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 4% to 22 % with a mean value of 14.8 ± 5.41 %. It was significantly less than that of the strongly stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 60).

Those with a moderate reaction ranged from 15% to 36 % with a mean value of 24.7 ± 7.75 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 60).

Weakly stained sperms ranged from 23% to 31 % with a mean value of 26.8 ± 2.66 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 60).

Sperms with a negative reaction ranged from 28% to 38 % with a mean value of 33.7 ± 3.27 %. They was significantly more than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 60).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of non-specific esterase reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from

4% to 12 % with a mean value of 8.4 ± 2.8 %. It was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 60).

Those with a moderate reaction ranged from 11% to 28 % with a mean value of 17.2 ± 5.57 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 60).

Weakly stained sperms ranged from 24% to 41 % with a mean value of 32.4 ± 5.48 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 60).

Sperms with a negative reaction ranged from 36% to 50 % with a mean value of 42 ± 4.97 %. They were significantly more than the negatively stained sperms for non-specific esterase in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 60).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the second split fraction specimens and the period of incubation [$r = - 0.785$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the second split fraction specimens and the period of incubation [$r = - 0.704$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the second split fraction specimens and the period of incubation [$r = 0.798$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the second split fraction specimens and the period of incubation [$r = 0.871$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added:

(Table 61)

a. Immediately examined specimens:

A positive non-specific esterase reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 22% to 40 % with a mean value of 31 ± 5.75 %. It did not differ statistically from the strongly stained sperms in the immediately examined second split fraction specimens [$P < 0.05$] (Table 62).

Moderately stained sperms ranged from 24% to 44 % with a mean value of 33.9 ± 6.98 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the immediately examined second split fraction specimens [$P < 0.05$] (Table 62).

Those with a weak reaction ranged from 13% to 22 % with a mean value of 17.2 ± 3.12 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the immediately examined second split fraction specimens [$P < 0.05$] (Table 62).

Sperms with a negative reaction ranged from 14% to 22 % with a mean value of 17.9 ± 3.00 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the second split fraction specimens examined immediately [$P < 0.05$] (Table 62).

b. Specimens examined after one hour:

A similar reaction was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from

Table (61): Showing the percentage of sperms with different grades of *non-specific esterase reaction in the second split fraction specimens to which calcium was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	32	33	13	22	61	29	34	18	19	71	28	20	17	35	81	18	17	29	36	91	6	21	32	41
52	26	42	17	15	62	28	26	26	20	72	17	31	28	24	82	16	24	27	33	92	1	27	29	43
53	33	30	15	22	63	26	36	24	14	73	18	33	29	20	83	18	22	24	36	93	9	18	35	38
54	31	36	14	19	64	34	21	17	28	74	21	28	26	25	84	7	30	26	37	94	12	13	38	37
55	22	44	16	18	65	35	19	20	26	75	29	18	15	38	85	17	24	28	31	95	2	23	33	42
56	37	27	15	21	66	24	37	22	17	76	18	27	26	29	86	3	35	30	32	96	10	10	37	43
57	25	39	20	16	67	38	19	11	32	77	25	22	21	32	87	14	24	24	38	97	11	17	31	41
58	40	24	22	14	68	40	20	18	22	78	21	25	14	40	88	7	30	28	35	98	0	24	39	37
59	28	38	19	15	69	33	22	20	25	79	17	29	27	27	89	9	29	25	37	99	10	17	33	40
60	36	26	21	17	70	20	39	17	24	70	22	24	18	36	80	4	36	26	34	100	5	19	37	39
Mean	31.0	33.9	17.2	17.9	Mean	30.7	27.3	19.3	22.7	Mean	21.6	25.7	22.1	30.6	Mean	11.3	27.1	26.7	34.9	Mean	6.6	18.9	34.4	40.1
S.D.	5.75	6.98	3.12	3.00	S.D.	6.38	8.25	4.19	5.40	S.D.	4.43	4.81	5.74	6.64	S.D.	5.93	5.95	2.06	2.33	S.D.	4.43	5.11	3.31	2.28

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (62): Showing the comparison between *non-specific estrase* reaction in the sperms of *second split fraction specimens and second split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Significance.
Strong reaction	*0*	28.0 ± 4.32	31.0 ± 5.75	1.318	<0.05	-
	1 hour	28.5 ± 8.28	30.7 ± 6.38	0.666	<0.05	-
	2 hours	20.0 ± 5.89	21.6 ± 4.43	0.687	<0.05	-
	4 hours	14.8 ± 5.41	11.3 ± 5.93	1.379	<0.05	-
	8 hours	8.4 ± 2.80	6.6 ± 4.43	1.087	<0.05	-
Moderate reaction	*0*	38.6 ± 7.43	33.9 ± 6.98	1.458	<0.05	-
	1 hour	30.9 ± 7.11	27.3 ± 8.25	1.046	<0.05	-
	2 hours	28.8 ± 6.27	25.7 ± 4.81	1.241	<0.05	-
	4 hours	24.7 ± 7.75	27.1 ± 5.95	0.777	<0.05	-
	8 hours	17.2 ± 5.57	18.9 ± 5.11	0.711	<0.05	-
Weak reaction	*0*	18.0 ± 2.71	17.2 ± 3.12	0.612	<0.05	-
	1 hour	20.6 ± 3.41	19.3 ± 4.19	0.761	<0.05	-
	2 hours	23.0 ± 4.5	22.1 ± 5.74	0.391	<0.05	-
	4 hours	26.8 ± 2.66	26.7 ± 2.06	0.094	<0.05	-
	8 hours	32.4 ± 5.48	34.4 ± 3.31	0.988	<0.05	-
No reaction	*0*	15.4 ± 4.06	17.9 ± 3.00	1.566	<0.05	-
	1 hour	20.0 ± 5.62	22.7 ± 5.40	1.096	<0.05	-
	2 hours	28.2 ± 5.31	30.6 ± 6.64	0.893	<0.05	-
	4 hours	33.7 ± 3.27	34.9 ± 2.33	0.945	<0.05	-
	8 hours	42.0 ± 4.97	40.1 ± 2.28	1.099	<0.05	-

- = non-significant

20% to 40 % with a mean value of 30.7 ± 6.38 %. It did not also differ statistically from that of the strongly stained sperms for non-specific esterase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 62).

Those with a moderate reaction ranged from 19% to 39 % with a mean value of 27.3 ± 8.25 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 62).

Weakly stained sperms ranged from 11% to 26 % with a mean value of 19.3 ± 4.19 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 62).

Sperms with a negative reaction ranged from 14% to 32 % with a mean value of 22.7 ± 5.40 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 62).

c. Specimens examined after two hours:

Similar locations of non-specific esterase reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 17% to 29 % with a mean value of 21.6 ± 4.43 %. Also it did not differ statistically from that of the strongly stained sperms in the second split fraction specimens examined after two hours of incubation [P < 0.05] (Table 62).

Those with a moderate reaction ranged from 18% to 33 % with a mean value of 25.7 ± 4.81 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the second split fraction specimens examined after two hours of incubation [P < 0.05] (Table 62).

Sperms with a weak reaction ranged from 14% to 29 % with a mean value of 22.1 ± 5.74%. They did not also differ statistically from the weakly stained sperms

for non-specific esterase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 62).

Sperms with a negative reaction ranged from 20% to 40 % with a mean value of 30.6 ± 6.64 %. This did not differ statistically from that of negatively stained sperms for non-specific esterase in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 62).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 3% to 18 % with a mean value of 11.3 ± 5.93 %. Also it did not differ statistically from that of the strongly stained sperms for non-specific esterase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 62).

Those with a moderate reaction ranged from 17% to 36 % with a mean value of 27.1 ± 5.95 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 62).

Weakly stained sperms ranged from 24% to 30 % with a mean value of 26.7 ± 2.06 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 62).

Sperms with a negative reaction ranged from 31% to 38 % with a mean value of 34.9 ± 2.33 %. Also, they did not differ statistically from that of the strongly stained sperms for non-specific esterase in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 62).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of non-specific esterase reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 12 % with a mean value of 6.6 ± 4.43%. It they did not differ statistically from that of the strongly stained sperms in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 62).

Those with a moderate reaction ranged from 10% to 27 % with a mean value of 18.9 ± 5.11 %. They did not differ statistically from the moderately stained sperms for non-specific esterase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 62).

Weakly stained sperms ranged from 29% to 39 % with a mean value of 34.4 ± 3.31 %. They did not also differ statistically from the weakly stained sperms for non-specific esterase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 62).

Sperms with a negative reaction ranged from 37% to 43 % with a mean value of 40.1 ± 2.28 %. They did not differ statistically from the negatively stained sperms for non-specific esterase in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 62).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for non-specific esterase in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.832$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for non-specific esterase in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.556$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the second split fraction specimens to which calcium was added and the period of incubation [$r = 0.855$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for non-specific esterase in the second split fraction specimens to which calcium was added and the period of incubation [$r = 0.829$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

IV. PERIODIC ACID SCHIFF REACTION:

A. WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 63)

a. Immediately examined specimens:

A positive PAS reaction was noticed in the mid-piece. No reaction was seen in other parts of spermatozoa (Fig. 5).

The percentage of spermatozoa giving a strong PAS reaction ranged from 11% to 24 % with a mean value of 17 ± 3.83 %. Those with a moderate reaction ranged from 40% to 58 % with a mean value of 47.8 ± 6.73 %. Weakly stained sperms ranged from 16% to 30 % with a mean value of 24.1 ± 4.58 %. Sperms with a negative reaction ranged from 3% to 17 % with a mean value of 11.1 ± 5.43 %.

b. Specimens examined after one hour:

A similar PAS reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 9% to 17 % with a mean value of 12.8 ± 2.57 %. Moderately stained sperms ranged from 37% to 60 % with a mean value of 49 ± 8.98 %. Those with a weak reaction ranged from 16% to 36 % with a mean value of 26.7 ± 6.8 %. Sperms with a negative reaction ranged from 6% to 19 % with a mean value of 11.5 ± 4.12 %.

Table (63): Showing the percentage of sperms with different grades of *PAS in the whole non-washed specimens.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	24	51	16	9	11	17	38	33	12	21	9	33	43	15	31	7	43	31	19	41	0	24	37	39
2	14	42	29	15	12	10	44	32	14	22	10	31	42	17	32	2	29	41	28	42	1	27	46	26
3	17	43	24	16	13	9	38	34	19	23	18	45	25	12	33	6	34	38	22	43	1	26	36	37
4	19	42	27	12	14	14	53	21	12	24	13	35	37	15	34	4	37	40	19	44	0	17	38	45
5	21	56	20	3	15	11	37	36	16	25	8	40	36	16	35	3	26	41	30	45	0	21	41	38
6	19	55	22	4	16	13	60	20	7	26	11	39	40	10	36	2	34	40	24	46	0	18	39	43
7	16	58	21	5	17	16	59	16	9	27	13	51	25	11	37	6	42	33	19	47	1	22	45	32
8	15	48	23	14	18	14	51	23	12	28	9	44	39	8	38	3	34	35	28	48	1	26	44	29
9	11	43	30	16	19	13	54	25	8	29	7	39	36	18	39	4	41	27	28	49	0	26	33	41
10	14	40	29	17	20	11	56	27	6	30	10	28	42	20	40	7	39	39	15	50	0	21	34	45
Mean	17.0	47.8	24.1	11.1	Mean	12.8	49.0	26.7	11.5	Mean	10.8	38.5	36.5	14.2	Mean	4.4	35.9	36.5	23.2	Mean	0.4	22.8	39.3	37.5
S.D.	3.83	6.73	4.58	5.43	S.D.	2.57	8.98	6.80	4.12	S.D.	3.19	7.00	6.55	3.82	S.D.	1.96	5.59	4.81	5.14	S.D.	0.52	3.55	4.57	6.60

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

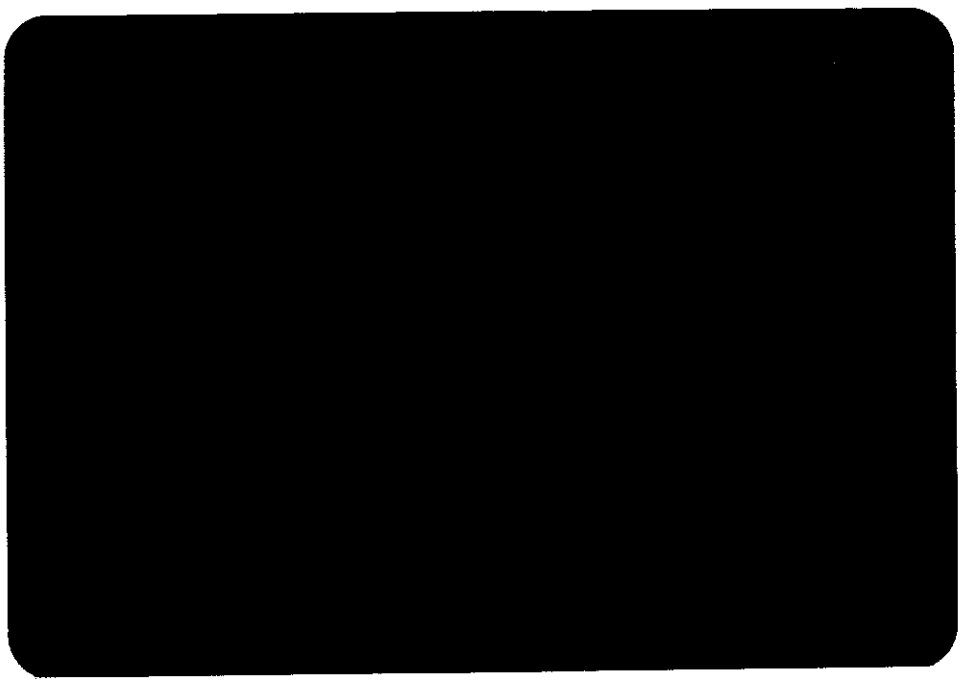

Fig. (5) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **carbohydrates.**

Note the strong (long arrow), moderate (short arrow), and weak (arrow head) reactions in the midpiece of sperms.

(PAS technique. Proj: 10 Obj: 100)

c. Specimens examined after two hours:

A positive PAS reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 7% to 18 % with a mean value of 10.8 ± 3.19 %. Those with a moderate reaction ranged from 28% to 51 % with a mean value of 38.5 ± 7.0 %. Sperms with a weak reaction ranged from 25% to 43 % with a mean value of 36.5± 3.82 %. Sperms with a negative reaction ranged from 8% to 20 % with a mean value of 14.2 ± 3.82 %.

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 2% to 7 % with a mean value of 4.4 ± 1.96 %. Moderately stained sperms ranged from 26% to 43 % with a mean value of 35.9 ± 5.59 %. Those with a weak reaction ranged from 27% to 41 % with a mean value of 36.5 ± 4.81 %. Sperms with a negative reaction ranged from 15% to 30 % with a mean value of 23.2 ± 5.14 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.4 ± 0.52 %. Those with a moderate reaction ranged from 17% to 27 % with a mean value of 22.8 ± 3.55 %. Weakly stained sperms ranged from 33% to 46 % with a mean value of 39.3 ± 4.57 %. Sperms with a negative reaction ranged from 26% to 45 % with a mean value of 37.5 ± 6.6 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the whole non-washed specimens and the period of incubation [$r = -0.886$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the whole non-washed specimens and the period of incubation [r = - 0.809; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the whole non-washed specimens and the period of incubation [r = 0.625; P < 0.001]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the whole non-washed specimens and the period of incubation [r = 0.892; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) Washed specimens: (Table 64)

a. Immediately examined specimens:

A positive PAS reaction was also observed only in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 22 % with a mean value of 15.4 ± 3.89 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined whole non-washed specimens [P <0.05] (Table 65).

Those with a moderate reaction ranged from 36% to 58 % with a mean value of 48.2 ± 8.63 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined whole non-washed specimens [P < 0.05] (Table 65).

Weakly stained sperms ranged from 15% to 30 % with a mean value of 23.2 ± 4.29 %. They did not also differ statistically from the weakly stained sperms for

Table (64): Showing the percentage of sperms with different grades of *PAS in the washed specimens.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	13	57	23	7	11	12	60	15	13	21	2	35	39	24	31	3	42	31	24	41	1	16	37	46
2	15	49	22	15	12	8	49	21	22	22	5	33	38	24	32	0	29	41	30	42	2	22	40	36
3	12	55	22	11	13	7	39	32	22	23	11	48	25	16	33	1	26	41	32	43	1	21	34	44
4	11	58	15	16	14	10	42	31	17	24	9	34	37	20	34	4	38	30	28	44	0	14	34	52
5	20	36	28	16	15	6	44	28	22	25	8	40	34	18	35	1	30	38	31	45	1	16	30	53
6	22	39	21	18	16	11	51	21	17	26	9	44	36	11	36	2	34	38	26	46	1	13	39	47
7	11	57	20	12	17	12	55	23	10	27	11	48	29	12	37	3	37	30	30	47	1	22	32	45
8	18	37	30	15	18	9	42	25	24	28	9	32	41	18	38	1	23	41	35	48	1	19	31	49
9	14	50	25	11	19	12	35	34	19	29	3	31	42	24	39	3	31	38	28	49	0	18	34	48
10	18	45	26	11	20	11	58	21	10	30	5	33	45	17	40	3	38	36	23	50	0	16	36	48
Mean	15.4	48.2	23.2	13.2	Mean	9.8	47.5	25.1	17.6	Mean	7.2	37.8	36.6	18.4	Mean	2.1	32.8	36.4	28.7	Mean	0.8	17.7	34.7	46.8
S.D.	3.89	8.63	4.29	3.33	S.D.	2.20	8.42	6.03	5.15	S.D.	3.22	6.66	6.02	4.72	S.D.	1.29	6.01	4.50	3.68	S.D.	0.63	3.23	3.30	4.73

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (65): Showing the comparison between the *P.A.S.* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Signific-ance.
Strong reaction	*0*	17.0 ± 3.83	15.4 ± 3.89	0.927	<0.05	-
	1 hour	12.8 ± 2.57	9.8 ± 2.20	2.802	<0.05	+
	2 hours	10.8 ± 3.19	7.2 ± 3.22	2.910	<0.05	+
	4 hours	4.4 ± 1.96	2.1 ± 1.29	3.108	<0.05	+
	8 hours	0.4 ± 0.52	0.8 ± 0.63	1.549	<0.05	-
Moderate reaction	*0*	47.8 ± 6.73	48.2 ± 8.63	0.116	<0.05	-
	1 hour	49.0 ± 8.98	47.5 ± 8.42	0.385	<0.05	-
	2 hours	38.5 ± 7.00	37.8 ± 6.66	0.229	<0.05	-
	4 hours	35.9 ± 5.59	32.8 ± 6.01	1.194	<0.05	-
	8 hours	22.8 ± 3.55	17.7 ± 3.23	3.357	<0.05	+
Weak reaction	*0*	24.1 ± 4.58	23.2 ± 4.29	0.454	<0.05	-
	1 hour	26.7 ± 6.8	25.1 ± 6.03	0.557	<0.05	-
	2 hours	36.5 ± 6.55	36.6 ± 6.02	0.036	<0.05	-
	4 hours	36.5 ± 4.81	36.4 ± 4.50	0.048	<0.05	-
	8 hours	39.3 ± 4.57	34.7 ± 3.30	2.580	<0.05	+
No reaction	*0*	11.1 ± 5.43	13.2 ± 3.33	1.044	<0.05	-
	1 hour	11.5 ± 4.12	17.6 ± 5.15	2.927	<0.05	+
	2 hours	14.2 ± 3.82	18.4 ± 4.72	2.187	<0.05	+
	4 hours	23.2 ± 5.14	28.7 ± 3.68	2.751	<0.05	+
	8 hours	37.5 ± 6.60	46.8 ± 4.73	3.620	<0.05	+

+ = significant - = non-significant

PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 65).

Sperms with a negative reaction ranged from 7% to 18 % with a mean value of 13.2 ± 3.33 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 65).

b. Specimens examined after one hour:

A similar PAS reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 6% to 12 % with a mean value of 9.8 ± 2.2 %. This was less significantly than that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 65).

Moderately stained sperms ranged from 35% to 60 % with a mean value of 47.5 ± 8.42 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 65).

Those with a weak reaction ranged from 15% to 34 % with a mean value of 25.1 ± 6.03 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 65).

Sperms with a negative reaction ranged from 10% to 24 % with a mean value of 17.6 ± 5.15 %. They were significantly more than the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 65).

c. Specimens examined after two hours:

A positive PAS reaction was also detected only in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 2% to 11 % with a mean value of 7.2 ± 3.22 %. It was significantly less than that of the strongly stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 65).

Those with a moderate reaction ranged from 31% to 48 % with a mean value of 37.8 ± 6.66 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 65).

Sperms with a weak reaction ranged from 25% to 45 % with a mean value of 36.6 ± 6.02 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 65).

Sperms with a negative reaction ranged from 11% to 24 % with a mean value of 18.4 ± 4.72 %. This was significantly more than that of negatively stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 65).

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 2.1 ± 1.29 %. They were significantly less than the strongly stained sperms for in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 65).

Moderately stained sperms ranged from 23% to 42 % with a mean value of 32.8 ± 6.01 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 65).

Those with a weak reaction ranged from 30% to 41 % with a mean value of 36.4 ± 4.5 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 65).

Sperms with a negative reaction ranged from 23% to 35 % with a mean value of 28.7 ± 3.68 %. They were significantly more than the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P <0.05] (Table 65).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 2 % with a mean value of 0.8 ± 0.63 %. It did not differ statistically from the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 65).

Those with a moderate reaction ranged from 13% to 22 % with a mean value of 17.7 ± 3.23 %. They were significantly less than that of the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 65).

Weakly stained sperms ranged from 30% to 40 % with a mean value of 34.7 ± 3.3 %. They were significantly less than that of the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 65).

Sperms with a negative reaction ranged from 36% to 53 % with a mean value of 46.8 ± 4.73 %. They were significantly more than the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 65).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the washed specimens and the period of incubation [$r = - 0.811$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the washed specimens and the period of incubation [$r = - 0.852$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the washed specimens and the period of incubation [$r = 0.506$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the washed specimens and the period of incubation [$r = 0.941$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 66).

a. Immediately examined specimens:

A positive PAS reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 13% to 23 % with a mean value of

Table (66): Showing the percentage of sperms with different grades of *PAS reaction in the washed specimens to which calcium was added.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
1	20	44	23	13	11	15	57	19	9	21	3	39	33	25	31	2	39	26	33	41	0	18	34	48
2	17	37	34	12	12	11	46	24	19	22	4	35	38	23	32	1	32	43	24	42	1	18	36	45
3	21	42	30	7	13	10	37	30	23	23	7	49	23	21	33	2	33	29	36	43	1	19	27	53
4	19	39	25	17	14	8	52	24	16	24	5	39	32	24	34	2	34	32	32	44	0	12	32	56
5	20	52	19	9	15	7	37	31	25	25	7	33	39	21	35	0	28	43	29	45	0	13	33	54
6	23	52	12	13	16	9	34	36	21	26	7	50	23	20	36	0	31	34	35	46	0	12	32	56
7	13	54	20	13	17	16	54	21	9	27	9	46	28	17	37	2	36	32	30	47	1	17	38	44
8	14	52	23	11	18	17	47	26	10	28	6	47	31	16	38	1	27	36	36	48	1	17	42	40
9	13	44	33	10	19	11	41	31	17	29	3	37	35	25	39	1	34	36	29	49	0	12	43	45
10	15	48	29	8	20	19	39	31	11	30	4	38	36	22	40	3	35	26	36	50	0	13	36	51
Mean	17.5	46.4	24.8	11.3	Mean	12.3	44.4	27.3	16.0	Mean	5.5	41.3	31.8	21.4	Mean	1.4	32.9	33.7	32.0	Mean	0.4	15.1	35.3	49.2
S.D.	3.60	6.04	6.86	2.95	S.D.	4.14	8.00	5.33	6.00	S.D.	2.01	6.13	5.67	3.10	S.D.	0.97	3.60	6.06	4.00	S.D.	0.52	2.92	4.83	5.59

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (67): Showing the comparison between the *P.A.S.* reaction in the sperms of *washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incuba-tion.	Washed Specimens.	Washed Specimens + Calcium	t	p	Significance.
Strong reaction	*0*	15.4 ± 3.89	17.5 ± 3.60	1.253	<0.05	-
	1 hour	9.8 ± 2.20	12.3 ± 4.14	1.687	<0.05	-
	2 hours	7.2 ± 3.22	5.5 ± 2.01	1.414	<0.05	-
	4 hours	2.1 ± 1.29	1.4 ± 0.97	1.376	<0.05	-
	8 hours	0.8 ± 0.63	0.4 ± 0.52	1.549	<0.05	-
Moderate reaction	*0*	48.2 ± 8.63	46.4 ± 6.04	0.541	<0.05	-
	1 hour	47.5 ± 8.42	44.4 ± 8.00	0.844	<0.05	-
	2 hours	37.8 ± 6.66	41.3 ± 6.13	1.223	<0.05	-
	4 hours	32.8 ± 6.01	32.9 ± 3.60	0.045	<0.05	-
	8 hours	17.7 ± 3.23	15.1 ± 2.92	1.886	<0.05	-
Weak reaction	*0*	23.2 ± 4.29	24.8 ± 6.86	0.625	<0.05	-
	1 hour	25.1 ± 6.03	27.3 ± 5.33	0.864	<0.05	-
	2 hours	36.6 ± 6.02	31.8 ± 5.67	1.835	<0.05	-
	4 hours	36.4 ± 4.50	33.7 ± 6.06	1.132	<0.05	-
	8 hours	34.7 ± 3.30	35.3 ± 4.83	0.324	<0.05	-
No reaction	*0*	13.2 ± 3.33	11.3 ± 2.95	1.352	<0.05	-
	1 hour	17.6 ± 5.15	16.0 ± 6.00	0.640	<0.05	-
	2 hours	18.4 ± 4.72	21.4 ± 3.10	1.681	<0.05	-
	4 hours	28.7 ± 3.68	32.0 ± 4.00	1.919	<0.05	-
	8 hours	46.8 ± 4.73	49.2 ± 5.59	1.036	<0.05	-

- = non-significant

17.5 ± 3.6 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 67).

Those with a moderate reaction ranged from 37% to 54 % with a mean value of 46.4 ± 6.04 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined washed specimens [P < 0.05] (Table 67).

Weakly stained sperms ranged from 12% to 34 % with a mean value of 24.8 ± 6.86 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens immediately examined [P < 0.05] (Table 67).

Sperms with a negative reaction ranged from 7% to 17 % with a mean value of 11.3 ± 2.95 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the immediately examined washed specimens [P < 0.05] (Table 67).

b. Specimens examined after one hour:

A similar location of PAS reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 7% to 19 % with a mean value of 12.3 ± 4.14 %. This did not also differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [P < 0.05] (Table 67).

Moderately stained sperms ranged from 34% to 57 % with a mean value of 44.4 ± 8.0 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the washed specimens examined after one hour of incubation [P < 0.05] (Table 67).

Those with a weak reaction ranged from 19% to 36 % with a mean value of 27.3 ± 5.33 %. They did not also differ statistically from the weakly stained sperms for

PAS reaction in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 67).

Sperms with a negative reaction ranged from 9% to 25 % with a mean value of 16 ± 6.0 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 67).

c. Specimens examined after two hours:

A positive PAS reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 3% to 9 % with a mean value of 5.5 ± 2.01 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 67).

Those with a moderate reaction ranged from 33% to 50 % with a mean value of 41.3 ± 6.13 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 67).

Sperms with a weak reaction ranged from 23% to 39 % with a mean value of 31.8 ± 5.67 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 67).

Sperms with a negative reaction ranged from 16% to 25 % with a mean value of 21.4 ± 3.1 %. This did not differ statistically from that of negatively stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 67).

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1.4 ± 0.97 %. They did not differ statistically from the strongly stained sperms in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 67).

Moderately stained sperms ranged from 27% to 39 % with a mean value of 32.9 ± 3.6 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 67).

Those with a weak reaction ranged from 26% to 43 % with a mean value of 33.7 ± 6.06 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 67).

Sperms with a negative reaction ranged from 24% to 36 % with a mean value of 32 ± 4.0 %. They did not differ statistically from the negatively stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 67).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.4 ± 0.52 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 67).

Those with a moderate reaction ranged from 12% to 19 % with a mean value of 15.1 ± 2.92 %. They did not differ statistically from the moderately stained sperms for

reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 67).

Weakly stained sperms ranged from 27% to 43 % with a mean value of 35.3 ± 4.83 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 67).

Sperms with a negative reaction ranged from 40% to 56 % with a mean value of 49.2 ± 5.59 %. They did not differ statistically from the negatively stained sperms for PAS reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 67).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the washed specimens to which calcium was added and the period of incubation [$r = -0.798$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the washed specimens to which calcium was added and the period of incubation [$r = -0.901$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the washed specimens to which calcium was added and the period of incubation [$r = 0.517$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the washed specimens to which calcium was

added and the period of incubation [r = 0.952; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Washed specimens to which ascorbic acid was added: (Table 68)

a. Immediately examined specimens:

A positive PAS reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 8% to 19 % with a mean value of 13.1 ± 4.36 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 69).

Those with a moderate reaction ranged from 39% to 65 % with a mean value of 50.4 ± 7.95 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined washed specimens [P < 0.05] (Table 69).

Weakly stained sperms ranged from 14% to 35 % with a mean value of 25.7 ± 7.42 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the immediately examined washed specimens [P < 0.05] (Table 69).

Sperms with a negative reaction ranged from 5% to 16 % with a mean value of 10.8 ± 3.65 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the immediately examined washed specimens [P < 0.05] (Table 69).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 8% to 16 % with a mean value of 12.2 ± 3.19 %. Also, this did not differ statistically from that of the strongly stained sperms for reaction in the washed specimens examined after one hour of incubation [P < 0.05] (Table 69).

Table (68): Showing the percentage of sperms with different grades of *PAS reaction in the washed specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
1	14	39	33	9	11	16	48	25	11	21	2	38	35	25	31	3	26	32	39	41	2	16	30	52
2	9	42	35	14	12	9	48	24	19	22	4	35	36	25	32	1	16	44	39	42	4	17	36	43
3	12	61	14	7	13	13	41	26	20	23	9	48	24	19	33	3	28	37	32	43	2	15	32	51
4	9	48	28	15	14	14	61	13	12	24	4	44	31	21	34	2	21	42	35	44	0	10	31	59
5	14	51	25	10	15	10	36	32	22	25	7	39	35	19	35	0	16	44	40	45	1	11	33	55
6	18	52	17	13	16	8	48	26	18	26	10	43	29	18	36	0	23	40	37	46	0	10	32	58
7	10	65	20	5	17	16	51	18	15	27	8	41	32	19	37	4	32	28	36	47	3	15	38	44
8	16	45	28	11	18	13	51	20	16	28	5	46	24	25	38	0	15	46	39	48	1	17	36	46
9	8	49	35	8	19	8	44	28	20	29	6	35	37	22	39	3	29	32	36	49	1	16	26	57
10	10	52	22	16	20	15	55	19	11	30	8	36	35	21	40	2	31	36	31	50	1	13	25	61
Mean	13.1	50.4	25.7	10.8	Mean	12.2	48.3	23.1	16.4	Mean	6.3	40.5	31.8	21.4	Mean	1.8	23.7	38.1	36.4	Mean	1.5	14.0	31.9	52.6
S.D.	4.36	7.95	7.42	3.65	S.D.	3.19	7.02	5.57	4.03	S.D.	2.54	4.65	4.78	2.76	S.D.	1.48	6.46	6.08	3.06	S.D.	1.27	2.79	4.20	6.48

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (69): Showing the comparison between the *P.A.S.* reaction in the sperms *of washed specimens* and *washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Significance.
Strong reaction	0	15.4 ± 3.89	13.1 ± 4.36	1.245	<0.05	-
	1 hour	9.8 ± 2.20	12.2 ± 3.19	1.958	<0.05	-
	2 hours	7.2 ± 3.22	6.3 ± 2.54	0.693	<0.05	-
	4 hours	2.1 ± 1.29	1.8 ± 1.48	0.485	<0.05	-
	8 hours	0.8 ± 0.63	1.5 ± 1.27	1.561	<0.05	-
Moderate reaction	0	48.2 ± 8.63	50.4 ± 7.95	0.593	<0.05	-
	1 hour	47.5 ± 8.42	48.3 ± 7.02	0.231	<0.05	-
	2 hours	37.8 ± 6.66	40.5 ± 4.65	1.051	<0.05	-
	4 hours	32.8 ± 6.01	23.7 ± 6.46	3.259	<0.05	+
	8 hours	17.7 ± 3.23	14.0 ± 2.79	2.740	<0.05	+
Weak reaction	0	23.2 ± 4.29	25.7 ± 7.42	0.922	<0.05	-
	1 hour	25.1 ± 6.03	23.1 ± 5.57	0.771	<0.05	-
	2 hours	36.6 ± 6.02	31.8 ± 4.78	1.974	<0.05	-
	4 hours	36.4 ± 4.50	38.1 ± 6.08	0.711	<0.05	-
	8 hours	34.7 ± 3.30	31.9 ± 4.20	1.657	<0.05	-
No reaction.	0	13.2 ± 3.33	10.8 ± 3.65	1.538	<0.05	-
	1 hour	17.6 ± 5.15	16.4 ± 4.03	0.580	<0.05	-
	2 hours	18.4 ± 4.72	21.4 ± 2.76	1.736	<0.05	-
	4 hours	28.7 ± 3.68	36.4 ± 3.06	5.084	<0.001	+++
	8 hours	46.8 ± 4.73	52.6 ± 6.48	2.285	<0.05	+

+++ = highly significant + = significant - = non-significant

Moderately stained sperms ranged from 36% to 61 % with a mean value of 48.3 ± 7.02 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the washed specimens examined after one hour of incubation [P < 0.05] (Table 69).

Those with a weak reaction ranged from 13% to 32 % with a mean value of 23.1 ± 5.57 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after one hour of incubation [P < 0.05] (Table 69).

Sperms with a negative reaction ranged from 11% to 22 % with a mean value of 16.4 ± 4.03 %. They did not differ statistically from the negatively stained sperms for PAS reaction in washed specimens examined after one hour of incubation [P <0.05] (Table 69).

c. Specimens examined after two hours:

A similar location of PAS reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 2% to 10 % with a mean value of 6.3 ± 2.54 %. Also, it did not differ statistically from that of the strongly stained sperms in the washed specimens examined after two hours of incubation [P < 0.05] (Table 69).

Those with a moderate reaction ranged from 35% to 48 % with a mean value of 40.5 ± 4.65 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [P < 0.05] (Table 69).

Sperms with a weak reaction ranged from 24% to 37 % with a mean value of 31.8 ± 4.78 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [P < 0.05] (Table 69).

Sperms with a negative reaction ranged from 18% to 25 % with a mean value of 21.4 ± 2.76 %. They did not differ statistically from the negatively stained sperms for PAS reaction in the washed specimens examined after two hours of incubation [P <0.05] (Table 69).

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 1.8 ± 1.48 %. This did not differ statistically from that of the strongly stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [P < 0.05] (Table 69).

Moderately stained sperms ranged from 15% to 32 % with a mean value of 23.7 ± 6.46 %. They were highly significantly less than the moderately stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [P < 0.05] (Table 69).

Those with a weak reaction ranged from 28% to 46 % with a mean value of 38.1 ± 6.08 %. They did not differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [P < 0.05] (Table 69).

Sperms with a negative reaction ranged from 31% to 40 % with a mean value of 36.4 ± 3.06 %. This was highly significantly more than that of negatively stained sperms for PAS reaction in the washed specimens examined after four hours of incubation [P <0.001] (Table 69).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 1.5 ± 1.27 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 69).

Those with a moderate reaction ranged from 10% to 17 % with a mean value of 14 ± 2.79 %. They were significantly less than that of the moderately stained sperms for PAS reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 69).

Weakly stained sperms ranged from 25% to 38 % with a mean value of 31.9 ± 4.2 %. They did not differ statistically from the weakly stained sperms for PAS reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 69).

Sperms with a negative reaction ranged from 43% to 61 % with a mean value of 52.6 ± 6.48 %. They were significantly more than the negatively stained sperms for PAS reaction in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 69).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.765$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.897$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the washed specimens to which ascorbic acid was added and the period of incubation [r = 0.398; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the washed specimens to which ascorbic acid was added and the period of incubation [r = 0.960; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(B) SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 70)

a. Immediately examined specimens:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 10% to 22 % with a mean value of 13.9 ± 3.84 %. They did not differ statistically from the strongly stained sperms in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 71).

Moderately stained sperms ranged from 39% to 68 % with a mean value of 53.9 ± 9.56 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 71).

Those with a weak reaction ranged from 16% to 30 % with a mean value of 21.9 ± 3.87 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 71).

Table (70): Showing the percentage of sperms with different grades of *PAS reaction in the first split fraction specimens.*

No.of Ejaculate	Immediate +++	++	+	-	No.of Ejaculate	1 hour +++	++	+	-	No.of Ejaculate	2 hours +++	++	+	-	No.of Ejaculate	4 hours +++	++	+	-	No.of Ejaculate	8 hours +++	++	+	-
51	12	58	24	6	61	15	52	22	11	71	2	33	44	21	81	1	19	47	33	91	0	11	40	49
52	17	53	23	7	62	5	42	34	19	72	5	31	36	28	82	3	35	37	25	92	1	13	46	40
53	15	60	20	5	63	7	56	21	16	73	12	40	28	20	83	2	21	43	34	93	1	12	38	49
54	17	46	23	14	64	9	50	24	17	74	11	52	25	12	84	2	26	35	37	94	0	10	47	43
55	13	39	30	18	65	6	42	30	22	75	6	41	35	18	85	0	24	45	31	95	1	15	47	37
56	22	42	18	18	66	10	43	34	13	76	7	36	39	18	86	3	25	49	23	96	0	12	45	43
57	10	50	24	16	67	11	54	17	18	77	3	33	35	29	87	2	26	45	27	97	0	11	45	44
58	10	59	21	10	68	14	45	27	14	78	9	50	28	13	88	4	36	34	26	98	1	13	40	46
59	12	64	20	4	69	5	51	25	19	79	6	50	32	12	89	3	33	36	28	99	0	11	43	46
60	11	68	16	5	70	6	46	33	15	70	8	35	38	19	80	0	28	34	38	100	1	14	40	45
Mean	13.9	53.9	21.9	10.3	Mean	8.8	48.1	26.7	16.4	Mean	6.9	40.1	34.0	19.0	Mean	2.0	27.3	40.5	30.2	Mean	0.5	12.2	43.1	44.2
S.D.	3.84	9.56	3.87	5.68	S.D.	3.65	5.15	5.93	3.27	S.D.	3.21	7.92	5.81	5.98	S.D.	1.33	5.74	5.85	5.18	S.D.	0.53	1.55	3.35	3.74

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (71): Showing the comparison between *the P.A.S.* reaction in the sperms of *whole non-washed* and *first split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Significance.
Strong reaction	*0*	17.0 ± 3.83	13.9 ± 3.84	1.807	<0.05	-
	1 hour	12.8 ± 2.57	8.8 ± 3.65	2.835	<0.05	+
	2 hours	10.8 ± 3.19	6.9 ± 3.21	2.724	<0.05	+
	4 hours	4.4 ± 1.96	2.0 ± 1.33	3.207	<0.05	+
	8 hours	0.4 ± 0.52	0.5 ± 0.53	0.429	<0.05	-
Moderate reaction	*0*	47.8 ± 6.73	53.9 ± 9.56	1.650	<0.05	-
	1 hour	49.0 ± 8.98	48.1 ± 5.15	0.275	<0.05	-
	2 hours	38.5 ± 7.00	40.1 ± 7.92	0.779	<0.05	-
	4 hours	35.9 ± 5.59	27.3 ± 5.74	3.397	<0.05	+
	8 hours	22.8 ± 3.55	12.2 ± 1.55	8.649	<0.001	+++
Weak reaction	*0*	24.1 ± 4.58	21.9 ± 3.87	1.160	<0.05	-
	1 hour	26.7 ± 6.8	26.7 ± 5.93	0.000	<0.05	-
	2 hours	36.5 ± 6.55	34.0 ± 5.81	0.903	<0.05	-
	4 hours	36.5 ± 4.81	40.5 ± 5.85	1.669	<0.05	-
	8 hours	39.3 ± 4.57	43.1 ± 3.35	2.121	<0.05	+
No reaction	*0*	11.1 ± 5.43	10.3 ± 5.68	0.322	<0.05	-
	1 hour	11.5 ± 4.12	16.4 ± 3.27	2.947	<0.05	+
	2 hours	14.2 ± 3.82	19.0 ± 5.98	2.138	<0.05	+
	4 hours	23.2 ± 5.14	30.2 ± 5.18	3.034	<0.05	+
	8 hours	37.5 ± 6.60	44.2 ± 3.74	2.793	<0.05	+

+++ = highly significant + = significant - = non-significant

Sperms with a negative reaction ranged from 4% to 18 % with a mean value of 10.3 ± 5.68 %. Also, they did not differ statistically from the negatively stained sperms for PAS reaction in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 71).

b. Specimens examined after one hour:

No difference was noticed as regards the site of PAS reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 5% to 15 % with a mean value of 8.8 ± 3.65 %. It was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 71).

Those with a moderate reaction ranged from 42% to 56 % with a mean value of 48.1 ± 5.15 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 71).

Weakly stained sperms ranged from 17% to 34 % with a mean value of 26.7 ± 5.93 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 71).

Sperms with a negative reaction ranged from 11% to 22 % with a mean value of 16.4 ± 3.27 %. They were significantly more than the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 71).

c. Specimens examined after two hours:

A similar location of PAS reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 2% to 12 % with a mean value of 6.9 ± 3.21 %. It

was significantly less than that of the strongly stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 71).

Those with a moderate reaction ranged from 31% to 52 % with a mean value of 40.1 ± 7.92 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 71).

Sperms with a weak reaction ranged from 25% to 44 % with a mean value of 34 ± 5.81 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 71).

Sperms with a negative reaction ranged from 12% to 29 % with a mean value of 19 ± 5.98 %. This was significantly more than that of negatively stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 71).

d. Specimens examined after four hours:

A positive PAS reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 0% to 4 % with a mean value of 2 ± 1.33 %. It was significantly less than that of the strongly stained sperms in whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 71).

Those with a moderate reaction ranged from 19% to 36 % with a mean value of 27.3 ± 5.74 %. They were significantly less than that of the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 71).

Weakly stained sperms ranged from 34% to 49 % with a mean value of 40.5 ± 5.85 %. They did not differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 71).

Sperms with a negative reaction ranged from 23% to 38 % with a mean value of 30.2 ± 5.18 %. They were significantly more than the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P <0.05] (Table 71).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.5 ± 0.53 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 71).

Those with a moderate reaction ranged from 10% to 15 % with a mean value of 12.2 ± 1.55 %. They were highly significantly less than that of the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [P < 0.001] (Table 71).

Weakly stained sperms ranged from 38% to 47 % with a mean value of 43.1 ± 3.55 %. They were significantly more than that of the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [P < 0.05] (Table 71).

Sperms with a negative reaction ranged from 37% to 49 % with a mean value of 44.2 ± 3.74 %. They were significantly more than the negatively stained sperms for PAS

reaction in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 71).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the first split fraction specimens and the period of incubation [$r = -0.789$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the first split fraction specimens and the period of incubation [$r = -0.913$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the first split fraction specimens and the period of incubation [$r = 0.777$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the first split fraction specimens and the period of incubation [$r = 0.928$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added: (Table 72)

a. Immediately examined specimens:

A positive PAS reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 11% to 22 % with a mean value of 16.3 ± 3.77 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined first split fraction specimens [$P < 0.05$] (Table 73).

Table (72): Showing the percentage of sperms with different grades of *PAS reaction in the first split fraction specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	29	56	18	7	61	16	40	28	16	71	4	29	41	26	81	0	20	30	50	91	0	13	37	50
52	25	51	23	11	62	8	43	25	24	72	5	31	37	27	82	2	31	30	37	92	1	14	38	47
53	13	54	20	13	63	10	42	28	20	73	12	34	31	23	83	1	28	29	42	93	0	17	29	54
54	11	41	27	21	64	12	39	28	21	74	9	39	36	16	84	0	23	34	43	94	0	10	35	55
55	22	44	11	23	65	9	38	29	24	75	7	41	30	22	85	2	20	40	38	95	1	17	38	44
56	14	43	25	18	66	14	55	15	16	76	4	38	37	21	86	3	27	37	33	96	0	11	35	54
57	12	50	25	13	67	17	51	19	13	77	6	31	40	23	87	0	17	40	43	97	0	10	36	54
58	20	55	17	8	68	14	56	17	13	78	12	46	27	15	88	2	25	39	34	98	0	16	42	42
59	19	43	18	20	69	8	47	22	23	79	8	39	34	19	89	0	26	42	32	99	0	11	40	49
60	18	42	21	19	70	6	43	22	29	70	6	35	32	27	80	1	19	37	43	100	1	12	43	44
Mean	16.3	47.9	20.5	15.3	Mean	11.4	45.4	23.3	19.9	Mean	7.3	36.3	34.5	21.9	Mean	1.1	23.6	35.8	39.5	Mean	0.3	13.1	37.3	49.3
S.D.	3.77	5.90	4.77	5.64	S.D.	3.75	6.55	5.08	5.30	S.D.	2.95	5.27	4.50	4.25	S.D.	1.10	4.53	4.76	5.68	S.D.	0.48	2.77	4.00	4.88

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (73): Showing the comparison between the *P.A.S.* reaction in the sperms of *first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	13.9 ± 3.84	16.3 ± 3.77	1.409	<0.05	-
	1 hour	8.8 ± 3.65	11.4 ± 3.75	1.573	<0.05	-
	2 hours	6.9 ± 3.21	7.3 ± 2.95	0.290	<0.05	-
	4 hours	2.0 ± 1.33	1.1 ± 1.10	1.646	<0.05	-
	8 hours	0.5 ± 0.53	0.3 ± 0.48	0.885	<0.05	-
Moderate reaction	*0*	53.9 ± 9.56	47.9 ± 5.90	1.689	<0.05	-
	1 hour	48.1 ± 5.15	45.4 ± 6.55	1.024	<0.05	-
	2 hours	40.1 ± 7.92	36.3 ± 5.27	1.263	<0.05	-
	4 hours	27.3 ± 5.74	23.6 ± 4.53	1.601	<0.05	-
	8 hours	12.2 ± 1.55	13.1 ± 2.77	0.898	<0.05	-
Weak reaction	*0*	21.9 ± 3.87	20.5 ± 4.77	0.721	<0.05	-
	1 hour	26.7 ± 5.93	23.3 ± 5.08	1.378	<0.05	-
	2 hours	34.0 ± 5.81	34.5 ± 4.50	0.215	<0.05	-
	4 hours	40.5 ± 5.85	35.8 ± 4.76	1.970	<0.05	-
	8 hours	43.1 ± 3.35	37.3 ± 4.00	3.515	<0.05	+
No reaction	*0*	10.3 ± 5.68	15.3 ± 5.64	1.976	<0.05	-
	1 hour	16.4 ± 3.27	19.9 ± 5.30	1.777	<0.05	-
	2 hours	19.0 ± 5.98	21.9 ± 4.25	1.250	<0.05	-
	4 hours	30.2 ± 5.18	39.5 ± 5.68	3.825	<0.05	+
	8 hours	44.2 ± 3.74	49.3 ± 4.88	2.625	<0.05	+

+ = significant - = non-significant

Those with a moderate reaction ranged from 41% to 56 % with a mean value of 47.9 ± 5.9 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined first split fraction specimens [$P < 0.05$] (Table 73).

Weakly stained sperms ranged from 11% to 27 % with a mean value of 20.5 ± 4.77 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the immediately examined first split fraction specimens [$P < 0.05$] (Table 73).

Sperms with a negative reaction ranged from 7% to 23 % with a mean value of 15.3 ± 5.64 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the immediately examined first split fraction specimens [$P < 0.05$] (Table 73).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 6% to 17 % with a mean value of 11.4 ± 3.75 %. Also, this did not differ statistically from that of the strongly stained sperms for PAS reaction in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 73).

Moderately stained sperms ranged from 38% to 56 % with a mean value of 45.4 ± 6.55 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 73).

Those with a weak reaction ranged from 15% to 29 % with a mean value of 23.3 ± 5.08 %. They did not also differ statistically from the weakly stained sperms for

PAS reaction in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 73).

Sperms with a negative reaction ranged from 13% to 29 % with a mean value of 19.9 ± 5.3 %. They did not differ statistically from the negatively stained sperms for PAS reaction in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 73).

c. Specimens examined after two hours:

A similar location of PAS reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 4% to 12 % with a mean value of 7.3 ± 2.95 %. Also, it did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 73).

Those with a moderate reaction ranged from 29% to 46 % with a mean value of 36.3 ± 5.27 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 73).

Sperms with a weak reaction ranged from 27% to 41 % with a mean value of 34.5 ± 4.5 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 73).

Sperms with a negative reaction ranged from 15% to 27 % with a mean value of 21.9 ± 4.25 %. They did not differ statistically from the negatively stained sperms for PAS reaction in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 73).

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 3 % with a mean value of 1.1 ± 1.1 %. This did not differ statistically from that of the strongly stained sperms for in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 73).

Moderately stained sperms ranged from 17% to 31 % with a mean value of 23.6 ± 4.53 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 73).

Those with a weak reaction ranged from 29% to 42 % with a mean value of 35.8 ± 4.76 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 73).

Sperms with a negative reaction ranged from 32% to 50 % with a mean value of 39.5 ± 5.68 %. This was significantly more than that of negatively stained sperms for PAS reaction in the first split fraction specimens examined after four hours of incubation [$P \leq 0.05$] (Table 73).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.3 ± 0.48 %. Also, it did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 73).

Those with a moderate reaction ranged from 10% to 17 % with a mean value of 13.1 ± 2.77 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 73).

Weakly stained sperms ranged from 29% to 43 % with a mean value of 37.3 ± 4.0 %. They were significantly less than that of the weakly stained sperms for PAS reaction in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 73).

Sperms with a negative reaction ranged from 42% to 55 % with a mean value of 49.3 ± 4.88 %. They were significantly more than the negatively stained sperms for PAS reaction in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 73).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = -0.815$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = -0.913$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.685$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.909$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 74)

a. Immediately examined specimens:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 11% to 21 % with a mean value of 15.4 ± 3.47 %. They did not differ statistically from the strongly stained sperms in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 75).

Moderately stained sperms ranged from 36% to 60 % with a mean value of 47.8 ± 8.0%. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 75).

Those with a weak reaction ranged from 14% to 31 % with a mean value of 23.4 ± 6.0 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 75).

Sperms with a negative reaction ranged from 7% to 21 % with a mean value of 13.4 ± 4.72 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 75).

b. Specimens examined after one hour:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 6% to 16 % with a mean value of

Table (74): Showing the percentage of sperms with different grades of *PAS reaction in the second split fraction specimens.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
51	17	48	23	14	61	7	40	33	20	71	17	51	22	10	81	10	40	32	18	91	1	25	44	30
52	12	45	27	16	62	16	56	19	9	72	14	52	24	10	82	4	22	43	31	92	0	14	36	50
53	21	50	18	11	63	11	48	27	14	73	8	34	40	18	83	3	29	38	30	93	0	18	41	41
54	20	54	19	7	64	13	52	22	13	74	6	33	40	21	84	8	41	31	20	94	1	24	43	32
55	11	49	29	20	65	14	51	28	7	75	9	41	33	17	85	6	31	44	19	95	0	15	36	49
56	14	36	29	21	66	10	42	29	19	76	7	39	36	18	86	5	37	33	25	96	1	15	35	49
57	14	57	17	12	67	6	42	36	16	77	15	45	31	9	87	9	40	33	18	97	1	18	50	31
58	15	39	31	15	68	7	49	32	12	78	8	32	40	20	88	4	28	35	33	98	0	27	35	38
59	12	51	27	10	69	15	59	18	8	79	6	39	39	16	89	3	32	34	31	99	1	19	42	38
60	18	60	14	8	70	8	55	18	19	70	14	51	27	8	80	4	29	38	29	100	1	23	41	35
Mean	15.4	47.8	23.4	13.4	Mean	10.7	49.4	26.2	13.7	Mean	10.4	41.7	33.2	14.7	Mean	5.6	32.9	36.1	25.4	Mean	0.6	19.8	40.3	39.3
S.D.	3.47	8.00	6.00	4.72	S.D.	3.65	6.47	6.60	4.76	S.D.	4.14	7.70	6.94	4.92	S.D.	2.55	6.33	4.53	6.10	S.D.	0.52	4.64	4.85	7.72

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (75): Showing the comparison between the *P.A.S.* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incuba-tion.	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Significance.
Strong reaction	*0*	17.0 ± 3.83	15.4 ± 3.47	0.979	<0.05	-
	1 hour	12.8 ± 2.57	10.7 ± 3.65	1.486	<0.05	-
	2 hours	10.8 ± 3.19	10.4 ± 4.14	0.242	<0.05	-
	4 hours	4.4 ± 1.96	5.6 ± 2.55	1.182	<0.05	-
	8 hours	0.4 ± 0.52	0.6 ± 0.52	0.866	<0.05	-
Moderate reaction	*0*	47.8 ± 6.73	47.8 ± 8.00	0.000	<0.05	-
	1 hour	49.0 ± 8.98	49.4 ± 6.47	0.114	<0.05	-
	2 hours	38.5 ± 7.00	41.7 ± 7.70	0.972	<0.05	-
	4 hours	35.9 ± 5.59	32.9 ± 6.33	1.123	<0.05	-
	8 hours	22.8 ± 3.55	19.8 ± 4.64	1.624	<0.05	-
Weak reaction	*0*	24.1 ± 4.58	23.4 ± 6.00	0.293	<0.05	-
	1 hour	26.7 ± 6.8	26.2 ± 6.60	0.167	<0.05	-
	2 hours	36.5 ± 6.55	33.2 ± 6.94	1.093	<0.05	-
	4 hours	36.5 ± 4.81	36.1 ± 4.53	0.191	<0.05	-
	8 hours	39.3 ± 4.57	40.3 ± 4.85	0.474	<0.05	-
No reaction	*0*	11.1 ± 5.43	13.4 ± 4.72	1.012	<0.05	-
	1 hour	11.5 ± 4.12	13.7 ± 4.76	1.105	<0.05	-
	2 hours	14.2 ± 3.82	14.7 ± 4.92	0.254	<0.05	-
	4 hours	23.2 ± 5.14	25.4 ± 6.10	0.873	<0.05	-
	8 hours	37.5 ± 6.60	39.3 ± 7.72	0.560	<0.05	-

- = non-significant

10.7 ± 3.65 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 75).

Those with a moderate reaction ranged from 40% to 59 % with a mean value of 49.4 ± 6.47 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 75).

Weakly stained sperms ranged from 18% to 36 % with a mean value of 26.2 ± 6.6 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 75).

Sperms with a negative reaction ranged from 7% to 20 % with a mean value of 13.7 ± 4.76 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after one hour of incubation [P <0.05] (Table 75).

c. Specimens examined after two hours:

A positive PAS reaction was also detected in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 6% to 17 % with a mean value of 10.4 ± 4.14 %. Also, it did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 75).

Those with a moderate reaction ranged from 32% to 52 % with a mean value of 41.7 ± 7.7 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 75).

Sperms with a weak reaction ranged from 22% to 40 % with a mean value of 33.2 ± 6.94 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P < 0.05] (Table 75).

Sperms with a negative reaction ranged from 8% to 21 % with a mean value of 14.7 ± 4.92 %. This did not differ statistically from that of negatively stained sperms for PAS reaction in the whole non-washed specimens examined after two hours of incubation [P <0.05] (Table 75).

d. Specimens examined after four hours:

A similar location of PAS reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 3% to 10 % with a mean value of 5.6 ± 2.55 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 75).

Those with a moderate reaction ranged from 22% to 41 % with a mean value of 32.9 ± 6.33 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 75).

Weakly stained sperms ranged from 31% to 44 % with a mean value of 36.1 ± 4.53 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after four hours of incubation [P < 0.05] (Table 75).

Sperms with a negative reaction ranged from 18% to 33 % with a mean value of 25.4 ± 6.1 %. Also, they did not differ statistically from the negatively stained sperms

for PAS reaction in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 75).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.6 ± 0.52 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 75).

Those with a moderate reaction ranged from 14% to 27 % with a mean value of 19.8 ± 4.64 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 75).

Weakly stained sperms ranged from 35% to 50 % with a mean value of 40.3 ± 4.85 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 75).

Sperms with a negative reaction ranged from 30% to 50 % with a mean value of 39.3 ± 7.72 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 75).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the second split fraction specimens and the period of incubation [$r = -0.839$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the second split fraction specimens and the period of incubation [r = - 0.849; P < 0.001]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the second split fraction specimens and the period of incubation [r = 0.692; P < 0.001]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the second split fraction specimens and the period of incubation [r = 0.863; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added: (Table 76)

a. Immediately examined specimens:

A positive PAS reaction was also observed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 7% to 20 % with a mean value of 13.6 ± 4.25 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined second split fraction specimens [P < 0.05] (Table 77).

Those with a moderate reaction ranged from 33% to 52 % with a mean value of 43.2 ± 6.34 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the immediately examined second split fraction specimens [P < 0.05] (Table 77).

Weakly stained sperms ranged from 21% to 36 % with a mean value of 28.1 ± 5.49 %. They did not also differ statistically from the weakly stained sperms for

Table (76): Showing the percentage of sperms with different grades of *PAS reaction in the second split fraction specimens to which calcium was added*

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
51	17	40	26	17	61	9	46	25	20	71	11	50	26	13	81	5	38	27	30	91	1	18	35	46
52	13	48	21	18	62	12	51	24	13	72	9	46	27	18	82	1	23	41	35	92	0	17	30	53
53	14	47	23	16	63	9	52	22	17	73	3	37	42	18	83	0	29	42	29	93	0	13	36	51
54	20	37	29	14	64	15	36	32	17	74	3	33	40	24	84	4	40	31	25	94	1	12	43	44
55	11	33	35	21	65	15	56	16	13	75	5	36	37	22	85	3	32	39	26	95	0	15	33	52
56	15	36	26	23	66	6	44	31	19	76	5	40	31	24	86	2	27	37	34	96	0	12	37	51
57	11	46	36	7	67	7	42	26	25	77	10	48	29	13	87	4	27	41	28	97	1	17	44	38
58	7	44	35	14	68	4	41	31	24	78	3	39	36	22	88	2	23	37	38	98	0	14	38	48
59	19	49	23	9	69	13	48	23	16	79	6	47	27	20	89	1	21	44	34	99	0	18	39	43
60	9	52	27	12	70	6	43	30	21	70	12	46	26	16	80	1	32	40	27	100	0	17	37	46
Mean	13.6	43.2	28.1	15.1	Mean	9.6	45.9	26.0	18.5	Mean	6.7	42.2	32.1	19.0	Mean	2.3	29.2	37.9	30.6	Mean	0.3	15.3	37.2	47.2
S.D.	4.25	6.34	5.49	5.00	S.D.	3.95	5.95	5.08	4.12	S.D.	3.50	5.88	6.12	4.11	S.D.	1.64	6.36	5.24	4.38	S.D.	0.48	2.41	4.21	4.73

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (77): Showing the comparison between the *P.A.S.* reaction in the sperms of *second split fraction specimens and second split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Significance.
Strong reaction	*0*	15.4 ± 3.47	13.6 ± 4.25	1.038	<0.05	-
	1 hour	10.7 ± 3.65	9.6 ± 3.95	0.647	<0.05	-
	2 hours	10.4 ± 4.14	6.7 ± 3.50	2.158	<0.05	+
	4 hours	5.6 ± 2.55	2.3 ± 1.64	3.447	<0.05	+
	8 hours	0.6 ± 0.52	0.3 ± 0.48	1.342	<0.05	-
Moderate reaction	*0*	47.8 ± 8.00	43.2 ± 6.34	1.426	<0.05	-
	1 hour	49.4 ± 6.47	45.9 ± 5.95	1.259	<0.05	-
	2 hours	41.7 ± 7.70	42.2 ± 5.88	0.163	<0.05	-
	4 hours	32.9 ± 6.33	29.2 ± 6.36	1.304	<0.05	-
	8 hours	19.8 ± 4.64	15.3 ± 2.41	2.724	<0.05	+
Weak reaction	*0*	23.4 ± 6.00	28.1 ± 5.49	1.828	<0.05	-
	1 hour	26.2 ± 6.60	26.0 ± 5.08	0.076	<0.05	-
	2 hours	33.2 ± 6.94	32.1 ± 6.12	0.376	<0.05	-
	4 hours	36.1 ± 4.53	37.9 ± 5.24	0.822	<0.05	-
	8 hours	40.3 ± 4.85	37.2 ± 4.21	1.525	<0.05	-
No reaction	*0*	13.4 ± 4.72	15.1 ± 5.00	0.782	<0.05	-
	1 hour	13.7 ± 4.76	18.5 ± 4.12	2.411	<0.05	+
	2 hours	14.7 ± 4.92	19.0 ± 4.11	2.121	<0.05	+
	4 hours	25.4 ± 6.10	30.6 ± 4.38	2.191	<0.05	+
	8 hours	39.3 ± 7.72	47.2 ± 4.73	2.759	<0.05	+

+ = significant - = non-significant

PAS reaction in the second split fraction specimens immediately examined [$P < 0.05$] (Table 77).

Sperms with a negative reaction ranged from 7% to 23 % with a mean value of 15.1 ± 5.0 %. They did not also differ statistically from the negatively stained sperms for PAS reaction in the immediately examined second split fraction specimens [$P < 0.05$] (Table 77).

b. Specimens examined after one hour:

A similar reaction was noticed in the mid-piece. The percentage of spermatozoa giving a strong reaction ranged from 4% to 15 % with a mean value of 9.6 ± 3.95 %. It did not differ statistically from that of the strongly stained sperms for PAS reaction in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 77).

Moderately stained sperms ranged from 36% to 56 % with a mean value of 45.9 ± 5.95 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 77).

Those with a weak reaction ranged from 16% to 32 % with a mean value of 26 ± 5.08 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 77).

Sperms with a negative reaction ranged from 13% to 25 % with a mean value of 18.5 ± 4.12 %. They was significantly more than the negatively stained sperms for PAS reaction in the second split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 77).

c. Specimens examined after two hours:

A similar location of reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 3% to 12 % with a mean value of 6.7 ± 3.5 %. It was significantly less than that of the strongly stained sperms for PAS reaction in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 77).

Those with a moderate reaction ranged from 33% to 50 % with a mean value of 42.2 ± 5.88 %. They did not differ statistically from the moderately stained sperms for PAS reaction in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 77).

Sperms with a weak reaction ranged from 26% to 42 % with a mean value of 32.1 ± 6.12 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 77).

Sperms with a negative reaction ranged from 13% to 24 % with a mean value of 19 ± 4.11 %. This was significantly more than that of negatively stained sperms for PAS reaction in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 77).

d. Specimens examined after four hours:

Also, only the mid-piece was site of positive PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 5 % with a mean value of 2.3 ± 1.64 %. They were significantly less than the strongly stained sperms in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 77).

Moderately stained sperms ranged from 21% to 40 % with a mean value of 29.2 ± 6.36 %. They did not differ statistically from the moderately stained sperms for

PAS reaction in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 77).

Those with a weak reaction ranged from 27% to 44 % with a mean value of 37.9 ± 5.24 %. They did not also differ statistically from the weakly stained sperms for PAS reaction in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 77).

Sperms with a negative reaction ranged from 25% to 38 % with a mean value of 30.6 ± 4.38 %. They were significantly more than negatively stained sperms for PAS reaction in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 77).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of PAS reaction. The percentage of spermatozoa giving a strong reaction ranged from 0% to 1 % with a mean value of 0.3 ± 0.48 %. It did not differ statistically from that of the strongly stained sperms in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 77).

Those with a moderate reaction ranged from 12% to 18 % with a mean value of 15.3 ± 2.41 %. They were significantly less than that of the moderately stained sperms for PAS reaction in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 77).

Weakly stained sperms ranged from 30% to 44 % with a mean value of 37.2 ± 4.21 %. They did not differ statistically from the weakly stained sperms for PAS reaction in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 77).

Sperms with a negative reaction ranged from 38% to 53 % with a mean value of 47.2 ± 4.73 %. They were significantly more than negatively stained sperms for PAS reaction in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 77).

A highly significantly negative correlation was found between the percent of the strongly stained sperms for PAS reaction in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.786$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms for PAS reaction in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.883$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms for PAS reaction in the second split fraction specimens to which calcium was added and the period of incubation [$r = 0.570$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms for PAS reaction in the second split fraction specimens to which calcium was added and the period of incubation [$r = 0.933$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

V. SUDAN BLACK B.:

(A) WHOLE EJACULATE SPECIMENS:

(1) Whole non-washed specimens: (Table 78)

a. Immediately examined specimens:

A positive Sudan black B. reaction was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail (Fig. 6).

Table (78): Showing the percentage of sperms with different grades of *Sudan black B, in the whole non-washed specimens.*

No. of Ejac-ulate	Immediate				No. of Ejac-ulate	1 hour				No. of Ejac-ulate	2 hours				No. of Ejac-ulate	4 hours				No. of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	28	50	8	14	11	26	47	16	11	21	23	42	18	17	31	32	24	23	21	41	14	23	43	20
2	38	33	11	18	12	43	33	9	15	22	34	33	23	10	32	26	29	28	17	42	15	30	36	19
3	24	53	7	16	13	41	35	15	9	23	39	26	19	16	33	23	32	22	23	43	18	19	36	27
4	49	28	6	17	14	40	31	11	18	24	18	46	25	11	34	28	32	29	11	44	13	25	46	16
5	33	32	17	18	15	23	44	17	16	25	39	26	17	18	35	35	30	20	15	45	19	28	35	18
6	37	31	16	16	16	28	44	18	10	26	35	28	27	10	36	24	37	27	12	46	10	27	37	26
7	42	38	13	7	17	41	35	10	14	27	28	36	18	18	37	31	29	21	19	47	20	18	45	17
8	40	42	10	8	18	45	28	14	13	28	24	41	16	19	38	20	32	26	22	48	18	22	39	21
9	33	46	11	10	19	38	33	13	16	29	30	33	20	17	39	27	25	28	20	49	14	31	36	19
10	39	43	9	9	20	28	41	19	12	30	35	31	24	10	40	25	30	24	21	50	21	21	38	20
Mean	36.3	39.6	10.8	13.3	Mean	35.3	37.1	14.2	13.4	Mean	30.5	34.2	20.7	14.6	Mean	27.1	30.0	24.8	18.1	Mean	16.2	24.4	39.1	20.3
S.D.	7.15	8.55	3.65	4.35	S.D.	8.11	6.42	3.43	2.91	S.D.	7.14	6.96	3.77	3.84	S.D.	4.53	3.71	3.22	4.20	S.D.	3.52	4.53	4.07	3.59

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

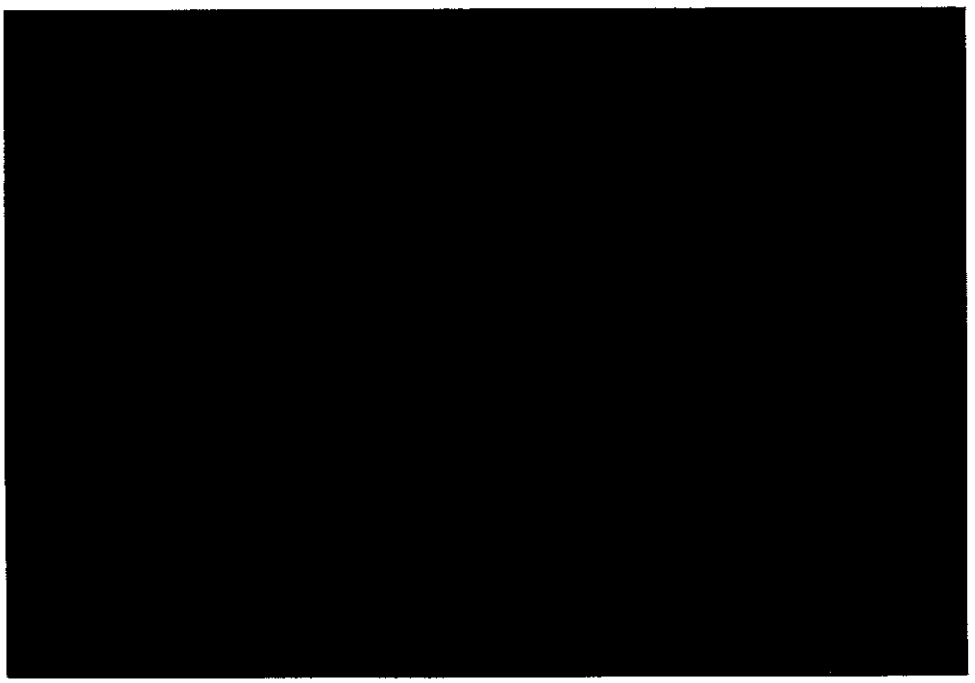

Fig. (6) : A photomicrograph of spermatozoa in a whole-non washed ejaculate stained immediately for **lipids.**

Note the strong (long arrow), moderate (short arrow), and weak (arrow head) reactions in the equatorial segment, post-acrosomal region, midpiece and tail of the sperms.

(Sudan black B. stain. Proj: 10 Obj: 100)

The percentage of spermatozoa giving a strong reaction ranged from 24% to 49 % with a mean value of 36.3 ± 7.15 %. Those with a moderate reaction ranged from 28% to 53 % with a mean value of 39.6 ± 8.55 %. Weakly stained sperms ranged from 6% to 17 % with a mean value of 10.8 ± 3.65 %. Sperms with a negative reaction ranged from 7% to 18 % with a mean value of 13.3 ± 4.35 %.

b. Specimens examined after one hour:

Similar locations of reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 23% to 45 % with a mean value of 35.3 ± 8.11 %. Moderately stained sperms ranged from 28% to 47 % with a mean value of 37.1 ± 6.42 %. Those with a weak reaction ranged from 9% to 19 % with a mean value of 14.2 ± 3.43 %. Sperms with a negative reaction ranged from 9% to 18 % with a mean value of 13.4 ± 2.91 %.

c. Specimens examined after two hours:

A positive Sudan black B. reaction was also detected in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 18% to 39 % with a mean value of 30.5 ± 7.14 %. Those with a moderate reaction ranged from 26% to 46 % with a mean value of 34.2 ± 6.96 %. Sperms with a weak reaction ranged from 16% to 27 % with a mean value of 20.7 ± 3.77 %. Sperms with a negative reaction ranged from 10% to 19 % with a mean value of 14.6 ± 3.84 %.

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 20% to 35 % with a mean value of 27.1 ± 4.53 %. Moderately stained sperms ranged from 24% to 37 % with a mean value of 30 ± 3.71 %. Those with a weak reaction ranged from 20% to 29 % with a mean value of 24.8 ± 3.22 %. Sperms with a negative reaction ranged from 11% to 23 % with a mean value of 18.1 ± 4.2 %.

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 10% to 21 % with a mean value of 16.2 ± 3.52 %. Those with a moderate reaction ranged from 18% to 31 % with a mean value of 24.4 ± 4.53 %. Weakly stained sperms ranged from 35% to 46 % with a mean value of 39.1 ± 4.07 %. Sperms with a negative reaction ranged from 16% to 27 % with a mean value of 20.3 ± 3.59 %.

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the whole non-washed specimens and the period of incubation [$r = - 0.765$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the whole non-washed specimens and the period of incubation [$r = - 0.663$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the whole non-washed specimens and the period of incubation [$r = 0.938$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the whole non-washed specimens and the period of incubation [$r = 0.593$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(2) **Washed specimens:** (Table 79)

a. Immediately examined specimens:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 30% to 51 % with a mean value of 40 ± 7.82 %. It did

Table (79): Showing the percentage of sperms with different grades of *Sudan black B.* in the washed specimens.

No.of Ejaculate	Immediate +++	++	+	-	No.of Ejaculate	1 hour +++	++	+	-	No.of Ejaculate	2 hours +++	++	+	-	No.of Ejaculate	4 hours +++	++	+	-	No.of Ejaculate	8 hours +++	++	+	-
1	35	41	15	9	11	44	31	12	13	21	37	26	20	17	31	9	26	34	31	41	1	21	42	36
2	44	35	9	12	12	33	34	11	22	22	26	24	27	23	32	10	31	29	30	42	1	17	50	32
3	30	43	14	13	13	26	44	18	12	23	15	44	22	19	33	13	25	30	32	43	8	13	39	40
4	51	22	10	17	14	41	37	10	12	24	27	29	19	25	34	19	21	32	28	44	3	23	43	31
5	31	46	13	10	15	29	44	14	13	25	23	42	25	10	35	11	31	31	27	45	6	15	51	28
6	49	26	16	9	16	36	37	13	14	26	28	33	23	16	36	23	26	26	25	46	5	19	46	30
7	40	34	11	15	17	36	35	18	11	27	33	25	16	26	37	17	20	33	30	47	13	13	40	34
8	46	31	12	11	18	28	46	16	10	28	29	36	22	13	38	22	19	35	24	48	7	12	44	37
9	31	44	17	8	19	24	43	12	21	29	16	42	21	21	39	22	16	32	30	49	15	12	41	32
10	43	29	16	12	20	28	38	15	19	30	36	31	18	15	40	15	21	38	26	50	2	17	42	39
Mean	40.0	35.1	13.3	11.6	Mean	32.5	38.9	13.9	14.7	Mean	27.0	33.2	21.3	18.5	Mean	16.1	23.6	32.0	28.3	Mean	6.1	16.2	43.8	33.9
S.D.	7.82	8.20	2.75	2.84	S.D.	6.64	5.04	2.81	4.32	S.D.	7.48	7.50	3.27	5.25	S.D.	5.28	5.04	3.33	2.71	S.D.	4.84	3.88	4.05	3.98

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (80): Showing the comparison between the *Sudan black B.* reaction in the sperms of *whole non-washed* and *washed specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	Washed Specimens.	t	p	Significance.
Strong reaction	*0*	36.3 ± 7.15	40.0 ± 7.82	1.104	<0.05	-
	1 hour	35.3 ± 8.11	32.5 ± 6.64	0.845	<0.05	-
	2 hours	30.5 ± 7.14	27.0 ± 7.48	1.070	<0.05	-
	4 hours	27.1 ± 4.53	16.1 ± 5.28	4.999	<0.001	+++
	8 hours	16.2 ± 3.52	6.1 ± 4.84	5.336	<0.001	+++
Moderate reaction	*0*	39.6 ± 8.55	35.1 ± 8.2	1.201	<0.05	-
	1 hour	37.1 ± 6.42	38.9 ± 5.04	0.697	<0.05	-
	2 hours	34.2 ± 6.96	33.2 ± 7.50	0.309	<0.05	-
	4 hours	30.0 ± 3.71	23.6 ± 5.04	3.234	<0.05	+
	8 hours	24.4 ± 4.53	16.2 ± 3.88	4.349	<0.001	+++
Weak reaction	*0*	10.8 ± 3.65	13.3 ± 2.75	1.731	<0.05	-
	1 hour	14.2 ± 3.43	13.9 ± 2.81	0.214	<0.05	-
	2 hours	20.7 ± 3.77	21.3 ± 3.27	0.380	<0.05	-
	4 hours	24.8 ± 3.22	32.0 ± 3.33	4.909	<0.001	+++
	8 hours	39.1 ± 4.07	43.8 ± 4.05	2.589	<0.05	+
No reaction	*0*	13.3 ± 4.35	11.6 ± 2.84	1.036	<0.05	-
	1 hour	13.4 ± 2.91	14.7 ± 4.32	0.789	<0.05	-
	2 hours	14.6 ± 3.84	18.5 ± 5.25	1.896	<0.05	-
	4 hours	18.1 ± 4.20	28.3 ± 2.71	6.451	<0.001	+++
	8 hours	20.3 ± 3.59	33.9 ± 3.98	8.017	<0.001	+++

+++ = highly significant + = significant - = non-significant

not differ statistically from that of the strongly stained sperms in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 80).

Those with a moderate reaction ranged from 22% to 46 % with a mean value of 35.1 ± 8.2 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 80).

Weakly stained sperms ranged from 9% to 17 % with a mean value of 13.3 ± 2.75 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 80).

Sperms with a negative reaction ranged from 8% to 17 % with a mean value of 11.6 ± 2.84 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 80).

b. Specimens examined after one hour:

A similar reaction for Sudan black B. was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 24% to 44 % with a mean value of 32.5 ± 6.64 %. This did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 80).

Moderately stained sperms ranged from 31% to 46 % with a mean value of 38.9 ± 5.04 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 80).

Those with a weak reaction ranged from 10% to 18 % with a mean value of 13.9 ± 2.81 %. They did not also differ statistically from the weakly stained sperms

by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 80).

Sperms with a negative reaction ranged from 10% to 22 % with a mean value of 14.7 ± 4.32 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 80).

c. Specimens examined after two hours:

Similar locations of reaction were detected. The percentage of spermatozoa giving a strong reaction ranged from 15% to 37 % with a mean value of 27 ± 7.48 %. It did not differ statistically from that of the strongly stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 80).

Those with a moderate reaction ranged from 24% to 44 % with a mean value of 33.2 ± 7.5 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 80).

Sperms with a weak reaction ranged from 16% to 27 % with a mean value of 21.3 ± 3.27 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 80).

Sperms with a negative reaction ranged from 10% to 26 % with a mean value of 18.5 ± 5.25 %. This did not also differ statistically from that of negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 80).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive Sudan black B. reaction. The percentage of spermatozoa giving a

strong reaction ranged from 9% to 23 % with a mean value of 16.1 ± 5.28 %. They were highly significantly less than the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 80).

Moderately stained sperms ranged from 16% to 31 % with a mean value of 23.6 ± 5.04 %. They were significantly less than that of the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 80).

Those with a weak reaction ranged from 26% to 38 % with a mean value of 32 ± 3.33 %. They were highly significantly more than that of the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 80).

Sperms with a negative reaction ranged from 24% to 32 % with a mean value of 28.3 ± 2.71 %. They were highly significantly more than the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.001$] (Table 80).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 1% to 15 % with a mean value of 6.1 ± 4.84 %. It was highly significantly less than that of the strongly stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 80).

Those with a moderate reaction ranged from 12% to 23 % with a mean value of 16.2 ± 3.88 %. They were highly significantly less than that of the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 80).

Weakly stained sperms ranged from 39% to 51 % with a mean value of 43.8 ± 4.05 %. They were significantly more than that of the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 80).

Sperms with a negative reaction ranged from 28% to 40 % with a mean value of 33.9 ± 3.98 %. They were highly significantly more than the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 80).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the washed specimens and the period of incubation [$r = - 0.867$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the washed specimens and the period of incubation [$r = - 0.778$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the washed specimens and the period of incubation [$r = 0.952$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the washed specimens and the period of incubation [$r = 0.885$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Washed specimens to which calcium was added: (Table 81).

a. Immediately examined specimens:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 25% to 46 % with a mean value of 34.7 ± 6.82 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [$P < 0.05$] (Table 82).

Those with a moderate reaction ranged from 28% to 50 % with a mean value of 39.5 ± 7.93 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined washed specimens [$P < 0.05$] (Table 82).

Weakly stained sperms ranged from 8% to 19 % with a mean value of 12.5 ± 3.75 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined washed specimens [$P < 0.05$] (Table 82).

Sperms with a negative reaction ranged from 9% to 17 % with a mean value of 13.3 ± 2.91 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the immediately examined washed specimens [$P < 0.05$] (Table 82).

b. Specimens examined after one hour:

A similar location of Sudan black B. reaction was noticed. The percentage of spermatozoa giving a strong reaction ranged from 29% to 45 % with a mean value of 36.1 ± 5.78 %. This did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 82).

Moderately stained sperms ranged from 25% to 43 % with a mean value of 36.2 ± 5.49 %. They did not differ statistically from the moderately stained sperms

Table (81): Showing the percentage of sperms with different grades of *Sudan black B. reaction in the washed specimens to which calcium was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
1	40	35	10	11	11	40	30	21	9	21	17	42	20	21	31	8	29	30	33	41	10	16	41	33
2	42	28	13	17	12	31	41	11	17	22	14	47	17	22	32	18	23	35	24	42	6	22	43	29
3	32	40	19	9	13	45	34	14	7	23	35	26	28	11	33	9	31	28	32	43	6	18	42	34
4	26	50	12	12	14	34	39	14	13	24	31	21	25	23	34	10	28	29	33	44	3	21	39	37
5	36	37	11	16	15	29	43	22	6	25	21	45	26	8	35	12	29	29	30	45	2	19	44	35
6	46	29	8	17	16	41	37	12	10	26	28	36	29	7	36	19	29	26	26	46	12	16	45	27
7	30	50	10	10	17	30	37	18	15	27	30	31	21	18	37	17	24	30	29	47	10	14	47	29
8	33	46	9	12	18	42	25	19	14	28	26	33	22	19	38	18	25	28	29	48	2	19	46	33
9	25	43	18	14	19	38	35	15	12	29	24	41	21	14	39	10	19	34	37	49	7	26	40	27
10	37	33	15	15	20	31	41	20	8	30	31	30	24	15	40	15	19	31	35	50	12	20	40	28
Mean	34.7	39.5	12.5	13.3	Mean	36.1	36.2	16.6	11.1	Mean	25.7	35.2	23.3	15.8	Mean	13.6	25.6	30.0	30.8	Mean	7.0	19.1	42.7	31.2
S.D.	6.82	7.93	3.75	2.91	S.D.	5.78	5.49	3.89	3.67	S.D.	6.70	8.51	3.77	5.75	S.D.	4.25	4.30	2.75	3.99	S.D.	3.89	3.45	2.75	3.61

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (82): Showing the comparison between the *Sudan black B.* reaction in the sperms of *washed specimens* and *washed specimens to which calcium was added.*

Type of Reaction.	Time of Incubation.	Washed Specimens.	Washed Specimens + Calcium	t	p	Significance.
Strong reaction	*0*	40.0 ± 7.82	34.7 ± 6.82	1.616	<0.05	-
	1 hour	32.5 ± 6.64	36.1 ± 5.78	1.293	<0.05	-
	2 hours	27.0 ± 7.48	25.7 ± 6.70	0.409	<0.05	-
	4 hours	16.1 ± 5.28	13.6 ± 4.25	1.167	<0.05	-
	8 hours	6.1 ± 4.84	7.0 ± 3.89	0.458	<0.05	-
Moderate reaction	*0*	35.1 ± 8.2	39.5 ± 7.93	1.220	<0.05	-
	1 hour	38.9 ± 5.04	36.2 ± 5.49	1.145	<0.05	-
	2 hours	33.2 ± 7.50	35.2 ± 8.51	0.558	<0.05	-
	4 hours	23.6 ± 5.04	25.6 ± 4.30	0.955	<0.05	-
	8 hours	16.2 ± 3.88	19.1 ± 3.45	1.767	<0.05	-
Weak reaction	*0*	13.3 ± 2.75	12.5 ± 3.75	0.544	<0.05	-
	1 hour	13.9 ± 2.81	16.6 ± 3.89	1.779	<0.05	-
	2 hours	21.3 ± 3.27	23.3 ± 3.77	1.267	<0.05	-
	4 hours	32.0 ± 3.33	30.0 ± 2.75	1.464	<0.05	-
	8 hours	43.8 ± 4.05	42.7 ± 2.75	0.711	<0.05	-
No reaction	*0*	11.6 ± 2.84	13.3 ± 2.91	1.323	<0.05	-
	1 hour	14.7 ± 4.32	11.1 ± 3.67	2.009	<0.05	-
	2 hours	18.5 ± 5.25	15.8 ± 5.75	1.096	<0.05	-
	4 hours	28.3 ± 2.71	30.8 ± 3.99	1.638	<0.05	-
	8 hours	33.9 ± 3.98	31.2 ± 3.61	1.587	<0.05	-

- = non-significant

by Sudan black B. in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 82).

Those with a weak reaction ranged from 11% to 22 % with a mean value of 16.6 ± 3.89 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 82).

Sperms with a negative reaction ranged from 6% to 17 % with a mean value of 11.1 ± 3.67 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the washed specimens examined after one hour of incubation [$P < 0.05$] (Table 82).

c. Specimens examined after two hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 14% to 35 % with a mean value of 25.7 ± 6.7 %. Also, it did not differ statistically from that of the strongly stained sperms by Sudan black B. in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 82).

Those with a moderate reaction ranged from 21% to 47 % with a mean value of 35.2 ± 8.51 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 82).

Sperms with a weak reaction ranged from 17% to 29 % with a mean value of 23.3 ± 3.77 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 82).

Sperms with a negative reaction ranged from 7% to 23 % with a mean value of 15.8 ± 5.75 %. This did not differ statistically from that of negatively stained

sperms Sudan black B. in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 82).

d. Specimens examined after four hours:

A positive reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 8% to 19 % with a mean value of 13.6 ± 4.25 %. They did not differ statistically from the strongly stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 82).

Moderately stained sperms ranged from 19% to 31 % with a mean value of 25.6 ± 4.30 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 82).

Those with a weak reaction ranged from 26% to 35 % with a mean value of 30 ± 2.75 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 82).

Sperms with a negative reaction ranged from 24% to 37 % with a mean value of 30.8 ± 3.99 %. Also, they did not differ statistically from the negatively stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 82).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of Sudan black B. reaction. The percentage of spermatozoa giving a strong reaction ranged from 2% to 12 % with a mean value of 7 ± 3.89 %. It did not differ statistically from that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 82).

calcium was added and the period of incubation [r = 0.804; P <0.001]. This means that the percent of such negatively stained sperms increased with time.

(4) Washed specimens to which ascorbic acid was added: (Table 83)

a. Immediately examined specimens:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 25% to 49 % with a mean value of 36.7 ± 7.75 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined washed specimens [P < 0.05] (Table 84).

Those with a moderate reaction ranged from 24% to 46 % with a mean value of 36.9 ± 6.92 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined washed specimens [P < 0.05] (Table 84).

Weakly stained sperms ranged from 8% to 20 % with a mean value of 13.1 ± 3.51 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined washed specimens [P < 0.05] (Table 84).

Sperms with a negative reaction ranged from 8% to 17 % with a mean value of 13.3 ± 3.2 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the immediately examined washed specimens [P < 0.05] (Table 84).

b. Specimens examined after one hour:

A similar reaction for Sudan black B. was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 30% to 44 % with a mean value of 36.5 ± 4.72 %. Also, this did not differ statistically from that of the strongly stained sperms in the washed specimens examined after one hour of incubation [P < 0.05] (Table 84).

Table (83): Showing the percentage of sperms with different grades of *Sudan black B. reaction in the washed specimens to which ascorbic acid was added.*

No.of Ejac-ulate	Immediate +++	++	+	-	No.of Ejac-ulate	1 hour +++	++	+	-	No.of Ejac-ulate	2 hours +++	++	+	-	No.of Ejac-ulate	4 hours +++	++	+	-	No.of Ejac-ulate	8 hours +++	++	+	-
1	43	33	8	16	11	39	35	14	12	21	38	27	15	20	31	27	27	23	23	41	10	27	37	26
2	28	44	20	8	12	30	33	18	19	22	39	29	17	15	32	17	36	21	26	42	18	21	36	25
3	45	31	9	15	13	33	40	23	4	23	31	36	16	17	33	29	19	22	30	43	6	31	34	29
4	36	42	12	10	14	36	27	20	17	24	26	38	18	18	34	12	33	31	24	44	8	30	40	22
5	49	24	11	16	15	44	36	12	8	25	25	41	23	11	35	16	36	30	18	45	15	29	36	20
6	31	40	14	15	16	30	47	17	6	26	30	31	19	20	36	30	24	26	20	46	19	27	35	19
7	40	31	16	13	17	38	37	15	10	27	36	29	22	13	37	23	30	27	20	47	8	30	41	21
8	32	40	14	14	18	34	35	11	20	28	25	32	20	23	38	21	32	25	22	48	7	30	39	24
9	25	46	12	17	19	41	32	19	8	29	28	40	20	12	39	24	26	22	28	49	11	25	39	25
10	38	38	15	9	20	40	27	22	11	30	32	33	21	14	40	19	25	29	27	50	13	21	38	28
Mean	36.7	36.9	13.1	13.3	Mean	36.5	34.9	17.1	11.5	Mean	31.0	33.6	19.1	16.3	Mean	21.8	28.8	25.6	23.8	Mean	11.5	27.1	37.5	23.9
S.D.	7.75	6.92	3.51	3.20	S.D.	4.72	5.92	4.07	5.50	S.D.	5.23	4.90	2.60	3.95	S.D.	5.90	5.55	3.60	3.91	S.D.	4.60	3.70	2.27	3.35

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (84): Showing the comparison between the *Sudan black B.* reaction in the sperms of *washed specimens* and *washed specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incuba-tion.	Washed Specimens.	Washed Specimens + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	40.0 ± 7.82	36.7 ± 7.75	0.948	<0.05	-
	1 hour	32.5 ± 6.64	36.5 ± 4.72	1.553	<0.05	-
	2 hours	27.0 ± 7.48	31.0 ± 5.23	1.386	<0.05	-
	4 hours	16.1 ± 5.28	21.8 ± 5.90	2.276	<0.05	+
	8 hours	6.1 ± 4.84	11.5 ± 4.60	2.557	<0.05	+
Moderate reaction	*0*	35.1 ± 8.2	36.9 ± 6.92	0.531	<0.05	-
	1 hour	38.9 ± 5.04	34.9 ± 5.92	1.627	<0.05	-
	2 hours	33.2 ± 7.50	33.6 ± 4.90	0.141	<0.05	-
	4 hours	23.6 ± 5.04	28.8 ± 5.55	2.193	<0.05	+
	8 hours	16.2 ± 3.88	27.1 ± 3.70	6.432	<0.001	+++
Weak reaction	*0*	13.3 ± 2.75	13.1 ± 3.51	0.142	<0.05	-
	1 hour	13.9 ± 2.81	17.1 ± 4.07	2.048	<0.05	-
	2 hours	21.3 ± 3.27	19.1 ± 2.60	1.666	<0.05	-
	4 hours	32.0 ± 3.33	25.6 ± 3.60	4.127	<0.001	+++
	8 hours	43.8 ± 4.05	37.5 ± 2.27	4.290	<0.001	+++
No reaction	*0*	11.6 ± 2.84	13.3 ± 3.20	1.257	<0.05	-
	1 hour	14.7 ± 4.32	11.5 ± 5.50	1.446	<0.05	-
	2 hours	18.5 ± 5.25	16.3 ± 3.95	1.059	<0.05	-
	4 hours	28.3 ± 2.71	23.8 ± 3.91	2.991	<0.05	+
	8 hours	33.9 ± 3.98	23.9 ± 3.35	6.076	<0.001	+++

+++ = highly significant + = significant - = non-significant

Moderately stained sperms ranged from 27% to 47 % with a mean value of 34.9 ± 5.92 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the washed specimens examined after one hour of incubation [P < 0.05] (Table 84).

Those with a weak reaction ranged from 11% to 23 % with a mean value of 17.1 ± 4.07 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the washed specimens examined after one hour of incubation [P < 0.05] (Table 84).

Sperms with a negative reaction ranged from 4% to 20 % with a mean value of 11.5 ± 5.5 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the washed specimens examined after one hour of incubation [P < 0.05] (Table 84).

c. Specimens examined after two hours:

Similar locations of Sudan black B. reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 25% to 39 % with a mean value of 31 ± 5.23 %. Also, it did not differ statistically from that of the strongly stained sperms in the washed specimens examined after two hours of incubation [P < 0.05] (Table 84).

Those with a moderate reaction ranged from 27% to 41 % with a mean value of 33.6 ± 4.9 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the washed specimens examined after two hours of incubation [P < 0.05] (Table 84).

Sperms with a weak reaction ranged from 15% to 23 % with a mean value of 19.1 ± 2.6 %. They did not also differ statistically from the weakly stained sperms Sudan black B. in the washed specimens examined after two hours of incubation [P < 0.05] (Table 84).

Sperms with a negative reaction ranged from 11% to 23 % with a mean value of 16.3 ± 3.95 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the washed specimens examined after two hours of incubation [$P < 0.05$] (Table 84).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 12% to 30 % with a mean value of 21.8 ± 5.9 %. This was significantly more than that of the strongly stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 84).

Moderately stained sperms ranged from 19% to 36 % with a mean value of 28.8 ± 5.55 %. They were significantly more than that of the moderately stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 84).

Those with a weak reaction ranged from 21% to 31 % with a mean value of 25.6 ± 3.6 %. They were highly significantly less than that of the weakly stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.001$] (Table 84).

Sperms with a negative reaction ranged from 18% to 30 % with a mean value of 23.8 ± 3.91 %. This was significantly less than that of negatively stained sperms by Sudan black B. in the washed specimens examined after four hours of incubation [$P < 0.05$] (Table 84).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of Sudan black B. reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 6% to 19 % with a mean value of 11.5 ± 4.6 %. Statistically, it was significantly more than that of the strongly stained sperms in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 84).

Those with a moderate reaction ranged from 21% to 31 % with a mean value of 27.1 ± 3.7 %. They were highly significantly more than that of the moderately stained sperms by Sudan black B. in the washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 84).

Weakly stained sperms ranged from 34% to 41 % with a mean value of 37.5 ± 2.27 %. They were significantly less than that of the weakly stained sperms by Sudan black B. in the washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 84).

Sperms with a negative reaction ranged from 19% to 29 % with a mean value of 23.9 ± 3.35 %. They were highly significantly less than the negatively stained sperms by Sudan black B. in the washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 84).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.859$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the washed specimens to which ascorbic acid was added and the period of incubation [$r = - 0.550$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the washed specimens to which ascorbic acid was added and the period of incubation [$r = 0.939$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the washed specimens to which

ascorbic acid was added and the period of incubation [r = 0.700; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(B) SPLIT EJACULATE SPECIMENS:

(1) First split fraction specimens: (Table 85)

a. Immediately examined specimens:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive Sudan black B. reaction. The percentage of spermatozoa giving a strong reaction ranged from 25% to 46 % with a mean value of 39.2 ± 6.2 %. They did not differ statistically from the strongly stained sperms in the whole non-washed specimens examined immediately [P < 0.05] (Table 86).

Moderately stained sperms ranged from 26% to 48 % with a mean value of 35.4 ± 6.64 %. They did not differ statistically from the moderately stained sperms Sudan black B. in the immediately examined whole non-washed specimens [P < 0.05] (Table 86).

Those with a weak reaction ranged from 11% to 16 % with a mean value of 13.4 ± 1.84 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [P < 0.05] (Table 86).

Sperms with a negative reaction ranged from 7% to 18 % with a mean value of 12 ± 4.03 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined immediately [P < 0.05] (Table 86).

b. Specimens examined after one hour:

No difference was noticed as regards the sites of Sudan black B. reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 25% to 46 % with a mean value of 33.7 ± 6.33 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [P < 0.05] (Table 86).

Table (85) : Showing the percentage of sperms with different grades of *Sudan black B. reaction in the first split fraction specimens.*

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
51	43	28	15	14	61	46	25	14	15	71	18	43	18	21	81	18	39	23	20	91	13	15	43	29
52	36	35	12	17	62	34	24	20	22	72	35	28	21	16	82	32	15	29	24	92	16	16	40	28
53	45	26	11	18	63	40	27	19	14	73	16	45	17	22	83	31	23	28	18	93	10	18	47	25
54	46	30	14	10	64	25	45	12	18	74	34	26	22	18	84	20	31	25	24	94	9	17	41	33
55	40	32	12	16	65	33	37	23	7	75	29	31	26	14	85	30	24	30	16	95	7	23	39	31
56	37	41	15	7	66	36	35	16	13	76	31	35	23	11	86	14	30	31	25	96	14	14	48	24
57	44	37	11	8	67	28	38	15	19	77	39	21	25	15	87	30	21	27	22	97	7	19	44	30
58	25	48	15	12	68	29	29	21	21	78	33	27	19	21	88	23	31	25	21	98	15	17	36	32
59	40	39	13	8	69	29	49	13	9	79	27	39	20	14	89	26	30	24	20	99	12	23	38	27
60	36	38	16	10	70	37	42	10	11	70	21	42	24	13	80	16	41	26	17	100	8	20	46	26
Mean	39.2	35.4	13.4	12.0	Mean	33.7	35.1	16.3	14.9	Mean	28.3	33.7	21.5	16.5	Mean	24.0	28.5	26.8	20.7	Mean	11.1	18.2	42.2	28.5
S.D.	6.20	6.64	1.84	4.03	S.D.	6.33	8.69	4.27	5.07	S.D.	7.70	8.29	3.03	3.81	S.D.	6.72	7.98	2.66	3.09	S.D.	3.35	3.08	4.05	3.03

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (86): Showing the comparison between the *Sudan black B.* reaction in the sperms of *whole non-washed* and *first split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	First Split Specimens.	t	p	Significance.
Strong reaction	*0*	36.3 ± 7.15	39.2 ± 6.20	0.969	<0.05	-
	1 hour	35.3 ± 8.11	33.7 ± 6.33	0.492	<0.05	-
	2 hours	30.5 ± 7.14	28.3 ± 7.70	0.663	<0.05	-
	4 hours	37.1 ± 4.53	24.0 ± 6.72	1.210	<0.05	-
	8 hours	16.2 ± 3.52	11.1 ± 3.35	3.319	<0.05	+
Moderate reaction	*0*	39.6 ± 8.55	35.4 ± 6.64	1.227	<0.05	-
	1 hour	37.1 ± 6.42	35.1 ± 8.69	0.586	<0.05	-
	2 hours	34.2 ± 6.96	33.7 ± 8.29	1.146	<0.05	-
	4 hours	30.0 ± 3.71	28.5 ± 7.98	0.539	<0.05	-
	8 hours	24.4 ± 4.53	18.2 ± 3.08	3.580	<0.05	+
Weak reaction	*0*	10.8 ± 3.65	13.4 ± 1.84	2.014	<0.05	-
	1 hour	14.2 ± 3.43	16.3 ± 4.27	1.213	<0.05	-
	2 hours	20.7 ± 3.77	21.5 ± 3.03	0.523	<0.05	-
	4 hours	24.8 ± 3.22	26.8 ± 2.66	1.513	<0.05	-
	8 hours	39.1 ± 4.07	42.2 ± 4.05	1.708	<0.05	-
No reaction	*0*	13.3 ± 4.35	12.0 ± 4.03	0.694	<0.05	-
	1 hour	13.4 ± 2.91	14.9 ± 5.07	0.812	<0.05	-
	2 hours	14.6 ± 3.84	16.5 ± 3.81	1.112	<0.05	-
	4 hours	18.1 ± 4.20	20.7 ± 3.09	1.576	<0.05	-
	8 hours	20.3 ± 3.59	28.5 ± 3.03	5.520	<0.001	+++

+++ = highly significant + = significant - = non-significant

Those with a moderate reaction ranged from 24% to 49 % with a mean value of 35.1 ± 8.69 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 86).

Weakly stained sperms ranged from 10% to 23 % with a mean value of 16.3 ± 4.27 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 86).

Sperms with a negative reaction ranged from 7% to 22 % with a mean value of 14.9 ± 5.07 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 86).

c. Specimens examined after two hours:

Similar locations of Sudan black B. reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 16% to 39 % with a mean value of 28.3 ± 7.7 %. Also, it did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 86).

Those with a moderate reaction ranged from 21% to 45 % with a mean value of 33.7 ± 8.29 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 86).

Sperms with a weak reaction ranged from 17% to 26 % with a mean value of 21.5 ± 3.03 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 86).

Sperms with a negative reaction ranged from 11% to 22 % with a mean value of 16.5 ± 3.81 %. This did not differ statistically from that of the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 86).

d. Specimens examined after four hours:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 14% to 32 % with a mean value of 24 ± 6.72 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 86).

Those with a moderate reaction ranged from 15% to 41 % with a mean value of 28.5 ± 7.98 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 86).

Weakly stained sperms ranged from 23% to 31 % with a mean value of 26.8 ± 2.66 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 86).

Sperms with a negative reaction ranged from 16% to 25 % with a mean value of 20.7 ± 3.09 %. Also, they did not differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 86).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of Sudan black B. reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from 7% to 16 % with a mean value of 11.1 ± 3.35 %. It was significantly less than that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 86).

Those with a moderate reaction ranged from 14% to 23 % with a mean value of 18.2 ± 3.08 %. They were significantly less than that of the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 86).

Weakly stained sperms ranged from 36% to 48 % with a mean value of 42.2 ± 4.05 %. They did not differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 86).

Sperms with a negative reaction ranged from 24% to 33 % with a mean value of 28.5 ± 3.03 %. They were highly significantly more than the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.001$] (Table 86).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the first split fraction specimens and the period of incubation [$r = - 0.842$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the first split fraction specimens and the period of incubation [$r = - 0.679$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the first split fraction specimens and the period of incubation [$r = 0.954$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the first split fraction specimens and the

period of incubation [r = 0.840; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

(2) First split fraction specimens to which ascorbic acid was added: (Table 87)

a. Immediately examined specimens:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 29% to 43 % with a mean value of 36.8 ± 4.71 %. It did not differ statistically from that of the strongly stained sperms in the immediately examined first split fraction specimens [P < 0.05] (Table 88).

Those with a moderate reaction ranged from 32% to 46 % with a mean value of 38.4 ± 5.06 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined first split fraction specimens [P < 0.05] (Table 88).

Weakly stained sperms ranged from 9% to 18 % with a mean value of 14.2 ± 3.19 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined first split fraction specimens [P < 0.05] (Table 88).

Sperms with a negative reaction ranged from 5% to 17 % with a mean value of 10.6 ± 4.12 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the immediately examined first split fraction specimens [P < 0.05] (Table 88).

b. Specimens examined after one hour:

A similar reaction for Sudan black B. was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 23% to 47 % with a mean value of 35.4 ± 8.28 %. Also, this did not differ statistically from that of the strongly stained sperms in the first

Table (87): Showing the percentage of sperms with different grades of *Sudan black B. reaction in the first split fraction specimens to which ascorbic acid was added.*

No. of Ejaculate	Immediate				No. of Ejaculate	1 hour				No. of Ejaculate	2 hours				No. of Ejaculate	4 hours				No. of Ejaculate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	33	39	18	10	61	31	46	11	12	71	38	31	19	12	81	33	24	29	14	91	23	29	28	20
52	39	41	15	5	62	42	38	10	10	72	35	28	22	15	82	40	12	24	24	92	27	34	25	14
53	35	42	9	14	63	34	41	19	6	73	26	40	17	17	83	26	28	26	20	93	12	36	29	23
54	42	34	18	6	64	47	24	15	14	74	39	28	18	15	84	36	24	25	15	94	24	33	25	18
55	34	40	14	12	65	29	48	12	11	75	33	33	21	13	85	30	33	24	13	95	11	43	31	15
56	43	33	17	7	66	36	38	18	8	76	23	41	20	16	86	28	29	28	15	96	22	32	33	13
57	33	46	12	9	67	26	42	17	15	77	22	41	23	14	87	31	21	25	23	97	16	41	27	16
58	29	44	10	17	68	46	31	16	7	78	28	43	17	12	88	20	32	27	21	98	9	40	30	21
59	38	33	13	16	69	23	47	13	17	79	25	34	24	17	89	24	30	30	16	99	18	36	26	20
60	42	32	16	10	70	40	36	12	10	70	30	36	19	15	80	18	31	32	19	100	8	38	32	22
Mean	36.8	38.4	14.2	10.6	Mean	35.4	39.3	14.3	11.0	Mean	29.9	35.5	20.0	14.6	Mean	28.6	26.4	27.0	18.0	Mean	17.0	36.2	28.6	18.2
S.D.	4.78	5.06	3.19	4.12	S.D.	8.28	7.44	3.13	3.56	S.D.	6.12	5.56	2.45	1.84	S.D.	6.88	6.38	2.71	3.92	S.D.	6.82	4.37	2.88	3.52

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (88): Showing the comparison between the *Sudan black B.* reaction in the sperms of *first split fraction specimens and first split fraction specimens to which ascorbic acid was added.*

Type of Reaction.	Time of Incubation.	First Split Fraction Specimens.	First Split Fractions + Ascorbic Acid	t	p	Significance.
Strong reaction	*0*	39.2 ± 6.20	36.8 ± 4.71	0.975	<0.05	-
	1 hour	33.7 ± 6.33	35.4 ± 8.28	0.516	<0.05	-
	2 hours	28.3 ± 7.70	29.9 ± 6.12	0.514	<0.05	-
	4 hours	24.0 ± 6.72	28.6 ± 6.88	1.513	<0.05	-
	8 hours	11.1 ± 3.35	17.0 ± 6.82	2.457	<0.05	+
Moderate reaction	*0*	35.4 ± 6.64	38.4 ± 5.06	1.137	<0.05	-
	1 hour	35.1 ± 8.69	39.3 ± 7.44	1.161	<0.05	-
	2 hours	33.7 ± 8.29	35.5 ± 5.56	0.570	<0.05	-
	4 hours	28.5 ± 7.98	26.4 ± 6.38	0.650	<0.05	-
	8 hours	18.2 ± 3.08	36.2 ± 4.37	10.648	<0.001	+++
Weak reaction	*0*	13.4 ± 1.84	14.2 ± 3.19	0.687	<0.05	-
	1 hour	16.3 ± 4.27	14.3 ± 3.13	1.195	<0.05	-
	2 hours	21.5 ± 3.03	20.0 ± 2.45	1.218	<0.05	-
	4 hours	26.8 ± 2.66	27.0 ± 2.71	0.167	<0.05	-
	8 hours	42.2 ± 4.05	28.6 ± 2.88	8.659	<0.001	-
No reaction	*0*	12.0 ± 4.03	10.6 ± 4.12	0.769	<0.05	-
	1 hour	13.9 ± 5.07	11.0 ± 3.56	1.992	<0.05	-
	2 hours	16.5 ± 3.81	14.6 ± 1.84	1.421	<0.05	-
	4 hours	20.7 ± 3.09	18.0 ± 3.92	1.711	<0.05	-
	8 hours	28.5 ± 3.03	18.2 ± 3.52	7.013	<0.001	+++

- = non-significant + = significant +++ = highly significant

split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 88).

Moderately stained sperms ranged from 24% to 48 % with a mean value of 39.3 ± 7.44 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 88).

Those with a weak reaction ranged from 10% to 19 % with a mean value of 14.3 ± 3.13 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 88).

Sperms with a negative reaction ranged from 6% to 17 % with a mean value of 11 ± 3.56 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the first split fraction specimens examined after one hour of incubation [$P < 0.05$] (Table 88).

c. Specimens examined after two hours:

Similar locations of Sudan black B. reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 22% to 39 % with a mean value of 29.9 ± 6.12 %. Also, it did not differ statistically from that of the strongly stained sperms in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 88).

Those with a moderate reaction ranged from 28% to 43 % with a mean value of 35.5 ± 5.56 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 88).

Sperms with a weak reaction ranged from 17% to 24 % with a mean value of 20 ± 2.45 %. They did not also differ statistically from the weakly stained sperms by

Sudan black B. in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 88).

Sperms with a negative reaction ranged from 12% to 17 % with a mean value of 14.6 ± 1.84 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the first split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 88).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 18% to 40 % with a mean value of 28.6 ± 6.88 %. This did not differ statistically from that of the strongly stained sperms by Sudan black B. in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 88).

Moderately stained sperms ranged from 12% to 33 % with a mean value of 26.4 ± 6.38 %. They did not differ statistically from the moderately stained sperms Sudan black B. in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 88).

Those with a weak reaction ranged from 24% to 32 % with a mean value of 27 ± 2.71 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 88).

Sperms with a negative reaction ranged from 13% to 24 % with a mean value of 18 ± 3.92 %. This did not also differ statistically from that of negatively stained sperms by Sudan black B. in the first split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 88).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of Sudan black B. reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from

8% to 27 % with a mean value of 17 ± 6.82 %. It was significantly more than that of the strongly stained sperms in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 88).

Those with a moderate reaction ranged from 29% to 43 % with a mean value of 36.2 ± 4.37 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 88).

Weakly stained sperms ranged from 25% to 33 % with a mean value of 28.6 ± 2.88 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the first split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 88).

Sperms with a negative reaction ranged from 13% to 23 % with a mean value of 18.2 ± 3.52 %. They were highly significantly less than the negatively stained sperms by Sudan black B. in the first split fraction specimens examined after eight hours of incubation [$P < 0.001$] (Table 88).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.731$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A non-significant negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = - 0.215$; $P < 0.05$].

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.834$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the first split fraction specimens to which ascorbic acid was added and the period of incubation [$r = 0.618$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(3) Second split fraction specimens: (Table 89)

a. Immediately examined specimens:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were sites of positive Sudan black B. reaction. The percentage of spermatozoa giving a strong reaction ranged from 33% to 54 % with a mean value of 41.1 ± 7.84 %. They did not differ statistically from the strongly stained sperms in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 90).

Moderately stained sperms ranged from 31% to 43 % with a mean value of 36.4 ± 4.17 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 90).

Those with a weak reaction ranged from 10% to 15 % with a mean value of 13.2 ± 1.55 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined whole non-washed specimens [$P < 0.05$] (Table 90).

Sperms with a negative reaction ranged from 2% to 15 % with a mean value of 9.3 ± 4.4 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined immediately [$P < 0.05$] (Table 90).

b. Specimens examined after one hour:

No difference was noticed as regards the site of Sudan black B. reaction. The percentage of spermatozoa giving a strong reaction ranged from 28% to 39 % with a mean value of 33.6 ± 3.89 %. It did not also differ statistically from that of the

Table (89): Showing the percentage of sperms with different grades of *Sudan black B. reaction in the second split fraction specimens.*

No. of Ejaculate	Immediate +++	++	+	-	No. of Ejaculate	1 hour +++	++	+	-	No. of Ejaculate	2 hours +++	++	+	-	No. of Ejaculate	4 hours +++	++	+	-	No. of Ejaculate	8 hours +++	++	+	-
S1	37	39	15	9	s1	31	36	16	17	71	35	23	23	19	B1	31	31	19	19	91	14	23	37	26
S2	41	34	10	15	s2	35	35	19	11	72	36	27	19	18	B2	14	39	25	22	92	11	26	34	29
S3	50	33	12	5	s3	28	37	17	18	73	41	31	13	15	B3	17	41	18	24	93	9	34	36	21
S4	34	41	13	12	s4	31	41	13	15	74	23	39	25	13	B4	27	30	31	12	94	16	21	45	18
S5	48	31	14	7	s5	39	38	14	9	75	24	44	16	16	B5	26	28	22	24	95	9	25	38	28
S6	46	34	15	5	s6	37	39	20	4	76	27	34	19	20	B6	22	33	29	16	96	19	18	44	19
S7	33	43	13	11	s7	29	43	18	10	77	41	22	20	17	B7	20	25	27	28	97	18	19	38	25
S8	54	32	12	2	s8	36	34	15	15	78	33	33	20	14	B8	18	39	23	20	98	18	20	40	22
S9	33	40	14	13	s9	38	39	15	8	79	28	38	18	16	B9	25	20	30	25	99	8	25	43	24
S0	35	37	14	14	s0	32	42	19	7	70	24	42	13	21	B0	31	34	18	17	100	7	25	45	23
Mean	41.1	36.4	13.2	9.3	Mean	33.6	38.4	16.6	11.4	Mean	31.2	33.3	18.6	16.9	Mean	23.1	32.0	24.2	20.7	Mean	12.9	23.6	40.0	23.5
S.D.	7.64	4.17	1.55	4.40	S.D.	3.89	2.99	2.37	4.65	S.D.	6.92	7.63	3.86	2.60	S.D.	5.86	6.65	4.96	4.83	S.D.	4.63	4.62	4.00	3.63

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (90): Showing the comparison between the *Sudan black B.* reaction in the sperms of *whole non-washed* and *second split fraction specimens.*

Type of Reaction.	Time of Incubation.	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Significance.
Strong reaction	*0*	36.3 ± 7.15	41.1 ± 7.84	1.431	<0.05	-
	1 hour	35.3 ± 8.11	33.6 ± 3.89	0.598	<0.05	-
	2 hours	30.5 ± 7.14	31.2 ± 6.92	0.223	<0.05	-
	4 hours	27.1 ± 4.53	23.1 ± 5.86	1.708	<0.05	-
	8 hours	16.2 ± 3.52	12.9 ± 4.63	1.794	<0.05	-
Moderate reaction	*0*	39.6 ± 8.55	36.4 ± 4.17	1.064	<0.05	-
	1 hour	37.1 ± 6.42	38.4 ± 2.99	0.581	<0.05	-
	2 hours	34.2 ± 6.96	33.3 ± 7.63	0.276	<0.05	-
	4 hours	30.0 ± 3.71	32.0 ± 6.65	0.831	<0.05	-
	8 hours	24.4 ± 4.53	23.6 ± 4.62	0.391	<0.05	-
Weak reaction	*0*	10.8 ± 3.65	13.2 ± 1.55	1.916	<0.05	-
	1 hour	14.2 ± 3.43	16.6 ± 2.37	1.823	<0.05	-
	2 hours	20.7 ± 3.77	18.6 ± 3.86	1.230	<0.05	-
	4 hours	24.8 ± 3.22	24.2 ± 4.96	0.321	<0.05	-
	8 hours	39.1 ± 4.07	40.0 ± 4.00	0.499	<0.05	-
No reaction	*0*	13.3 ± 4.35	9.3 ± 4.40	2.045	<0.05	-
	1 hour	13.4 ± 2.91	11.4 ± 4.65	1.153	<0.05	-
	2 hours	14.6 ± 3.84	16.9 ± 2.60	1.569	<0.05	-
	4 hours	18.1 ± 4.20	20.7 ± 4.83	1.284	<0.05	-
	8 hours	20.3 ± 3.59	23.5 ± 3.63	1.982	<0.05	-

- = non-significant

strongly stained sperms in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 90).

Those with a moderate reaction ranged from 34% to 43 % with a mean value of 38.4 ± 2.99 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 90).

Weakly stained sperms ranged from 13% to 20 % with a mean value of 16.6 ± 2.37 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 90).

Sperms with a negative reaction ranged from 4% to 18 % with a mean value of 11.4 ± 4.65 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after one hour of incubation [$P < 0.05$] (Table 90).

c. Specimens examined after two hours:

A positive Sudan black B. reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 23% to 41 % with a mean value of 31.2 ± 6.92 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 90).

Those with a moderate reaction ranged from 22% to 44 % with a mean value of 33.3 ± 7.63 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 90).

Sperms with a weak reaction ranged from 13% to 25 % with a mean value of 18.6 ± 3.86 %. They did not also differ statistically from the weakly stained sperms

by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 90).

Sperms with a negative reaction ranged from 13% to 21 % with a mean value of 16.9 ± 2.6 %. Also, this did not differ statistically from that of negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after two hours of incubation [$P < 0.05$] (Table 90).

d. Specimens examined after four hours:

Similar locations of reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 14% to 31 % with a mean value of 23.1 ± 5.86 %. It did not differ statistically from that of the strongly stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 90).

Those with a moderate reaction ranged from 20% to 41 % with a mean value of 32 ± 6.65 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 90).

Weakly stained sperms ranged from 18% to 31 % with a mean value of 24.2 ± 4.96 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 90).

Sperms with a negative reaction ranged from 12% to 28 % with a mean value of 20.7 ± 4.83 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after four hours of incubation [$P < 0.05$] (Table 90).

e. Specimens examined after eight hours:

No difference was noticed as regards the site of Sudan black B. reaction. Anyhow, the percentage of spermatozoa giving a strong reaction ranged from

7% to 19 % with a mean value of 12.9 ± 4.63 %. It did not differ statistically from that of the strongly stained sperms in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 90).

Those with a moderate reaction ranged from 18% to 34 % with a mean value of 23.6 ± 4.62 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 90).

Weakly stained sperms ranged from 34% to 45 % with a mean value of 40 ± 4.0 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 90).

Sperms with a negative reaction ranged from 18% to 29 % with a mean value of 23.5 ± 3.63 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the whole non-washed specimens examined after eight hours of incubation [$P < 0.05$] (Table 90).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the second split fraction specimens and the period of incubation [$r = - 0.847$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the second split fraction specimens and the period of incubation [$r = - 0.673$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms by Sudan black B. in the second split fraction specimens and the period of incubation [$r = 0.936$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the second split fraction specimens and the period of incubation [$r = 0.752$; $P < 0.001$]. This means that the percent of such negatively stained sperms increased with time.

(4) Second split fraction specimens to which calcium was added: (Table 91)

a. Immediately examined specimens:

A positive reaction was also observed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a strong reaction ranged from 31% to 44 % with a mean value of 36.2 ± 4.29 %. It did not differ statistically from that of the strongly stained sperms by Sudan black B. in the immediately examined second split fraction specimens [$P < 0.05$] (Table 92).

Those with a moderate reaction ranged from 33% to 45 % with a mean value of 39.9 ± 4.23 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the immediately examined second split fraction specimens [$P < 0.05$] (Table 92).

Weakly stained sperms ranged from 5% to 17 % with a mean value of 11.5 ± 4.12 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the immediately examined second split fraction specimens [$P < 0.05$] (Table 92).

Sperms with a negative reaction ranged from 8% to 17 % with a mean value of 12.4 ± 3.06 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the immediately examined second split fraction specimens [$P < 0.05$] (Table 92).

b. Specimens examined after one hour:

A similar reaction for Sudan black B. was noticed in the equatorial segment, post-acrosomal region, mid-piece and tail. The percentage of spermatozoa giving a

Table (91) Showing the percentage of sperms with different grades of *Sudan black B. reaction in the second split fraction specimens to which calcium was added.*

No.of Ejac-ulate	Immediate				No.of Ejac-ulate	1 hour				No.of Ejac-ulate	2 hours				No.of Ejac-ulate	4 hours				No.of Ejac-ulate	8 hours			
	+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-		+++	++	+	-
51	44	34	12	10	61	41	37	11	11	71	35	31	19	15	81	38	16	32	14	91	15	27	40	18
52	32	41	16	11	62	34	41	16	9	72	21	43	24	12	82	22	31	27	20	92	19	28	37	16
53	34	43	10	13	63	33	38	13	16	73	24	38	19	19	83	20	35	25	20	93	14	25	32	29
54	38	39	8	15	64	37	43	17	3	74	32	34	24	10	84	28	29	24	19	94	9	35	36	20
55	42	33	17	8	65	29	45	9	17	75	35	27	21	17	85	34	18	30	18	95	17	23	41	19
56	36	42	8	14	66	39	36	19	6	76	37	29	23	11	86	32	24	29	15	96	8	35	44	13
57	34	45	5	16	67	29	40	18	13	77	20	42	20	18	87	26	28	25	21	97	15	25	43	17
58	38	36	9	17	68	35	35	12	18	78	21	44	23	12	88	20	38	23	19	98	7	32	33	28
59	31	44	16	9	69	32	43	20	5	79	36	30	19	15	89	24	35	28	13	99	19	18	39	24
60	33	42	14	11	70	43	39	10	8	70	17	46	23	14	80	32	22	29	17	100	17	28	34	21
Mean	36.2	39.9	11.5	12.4	Mean	35.2	39.7	14.5	10.6	Mean	27.8	36.4	21.5	14.3	Mean	27.6	27.6	27.2	17.6	Mean	14.0	27.6	37.9	20.5
S.D.	4.29	4.23	4.12	3.06	S.D.	4.78	3.30	3.98	5.27	S.D.	7.87	7.04	2.12	3.06	S.D.	6.24	7.47	2.90	2.76	S.D.	4.47	5.34	4.18	5.15

+++ = Strong reaction ++ = Moderate reaction + = Weak reaction - = negative reaction

Table (92): Showing the comparison between the *Sudan black B.* reaction in the sperms of *second split fraction specimens and econd split fraction specimens to which calcium was added.*

Type of Reaction.	Time of Incuba-tion.	Second Split Fraction Specimens.	Second Split Fractions + Calcium.	t	p	Signific-ance.
Strong reaction	*0*	41.1 ± 7.84	36.2 ± 4.29	1.734	<0.05	-
	1 hour	33.6 ± 3.89	35.2 ± 4.78	0.821	<0.05	-
	2 hours	31.2 ± 6.92	27.8 ± 7.87	1.026	<0.05	-
	4 hours	23.1 ± 5.86	27.6 ± 6.24	1.663	<0.05	-
	8 hours	12.9 ± 4.63	14.0 ± 4.47	0.541	<0.05	-
Moderate reaction	*0*	36.4 ± 4.17	39.9 ± 4.23	1.864	<0.05	-
	1 hour	38.4 ± 2.99	39.7 ± 3.30	0.923	<0.05	-
	2 hours	33.3 ± 7.63	36.4 ± 7.04	0.944	<0.05	-
	4 hours	32.0 ± 6.65	27.6 ± 7.47	1.391	<0.05	-
	8 hours	23.6 ± 4.62	27.6 ± 5.34	1.791	<0.05	-
Weak reaction	*0*	13.2 ± 1.55	11.5 ± 4.12	1.222	<0.05	-
	1 hour	16.6 ± 2.37	14.5 ± 3.98	1.434	<0.05	-
	2 hours	18.6 ± 3.86	21.5 ± 2.12	2.080	<0.05	-
	4 hours	24.2 ± 4.96	27.2 ± 2.90	1.651	<0.05	-
	8 hours	40.0 ± 4.00	37.9 ± 4.18	1.149	<0.05	-
No reaction	*0*	9.3 ± 4.40	12.4 ± 3.06	1.829	<0.05	-
	1 hour	11.4 ± 4.65	10.6 ± 5.27	0.360	<0.05	-
	2 hours	16.9 ± 2.60	14.3 ± 3.06	2.048	<0.05	-
	4 hours	20.7 ± 4.83	17.6 ± 2.76	1.762	<0.05	-
	8 hours	23.5 ± 3.63	20.5 ± 5.15	1.506	<0.05	-

- = non-significant

strong reaction ranged from 29% to 43 % with a mean value of 35.2 ± 4.78 %. It did not differ statistically from that of the strongly stained sperms in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 92).

Moderately stained sperms ranged from 35% to 45 % with a mean value of 39.7 ± 3.3 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 92).

Those with a weak reaction ranged from 9% to 20 % with a mean value of 14.5 ± 3.98 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 92).

Sperms with a negative reaction ranged from 3% to 18 % with a mean value of 10.6 ± 5.27 %. They did not differ statistically from the negatively stained sperms by Sudan black B. in the second split fraction specimens examined after one hour of incubation [P < 0.05] (Table 92).

c. Specimens examined after two hours:

Similar locations of reaction were noticed. The percentage of spermatozoa giving a strong reaction ranged from 17% to 37 % with a mean value of 27.8 ± 7.87 %. Also, it did not differ statistically from that of the strongly stained sperms by Sudan black B. in the second split fraction specimens examined after two hours of incubation [P < 0.05] (Table 92).

Those with a moderate reaction ranged from 27% to 46 % with a mean value of 36.4 ± 7.04 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the second split fraction specimens examined after two hours of incubation [P < 0.05] (Table 92).

Sperms with a weak reaction ranged from 19% to 24 % with a mean value of 21.5 ± 2.12 %. They did not also differ statistically from the weakly stained sperms

by Sudan black B. in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 92).

Sperms with a negative reaction ranged from 10% to 19 % with a mean value of 14.3 ± 3.06 %. This did not differ statistically from that of negatively stained sperms by Sudan black B. in the second split fraction specimens examined after two hours of incubation [$P < 0.05$] (Table 92).

d. Specimens examined after four hours:

Also, the equatorial segment, post-acrosomal region, mid-piece and tail were the sites of positive reaction. The percentage of spermatozoa giving a strong reaction ranged from 20% to 38 % with a mean value of 27.6 ± 6.24 %. They did not differ statistically from the strongly stained sperms by Sudan black B. in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 92).

Moderately stained sperms ranged from 16% to 38 % with a mean value of 27.6 ± 7.47 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 92).

Those with a weak reaction ranged from 23% to 32 % with a mean value of 27.2 ± 2.9 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 92).

Sperms with a negative reaction ranged from 13% to 21 % with a mean value of 17.6 ± 2.76 %. Also, they did not differ statistically from negatively stained sperms Sudan black B. in the second split fraction specimens examined after four hours of incubation [$P < 0.05$] (Table 92).

e. Specimens examined after eight hours:

No difference was noticed as regards the sites of Sudan black B. reaction. The percentage of spermatozoa giving a strong reaction ranged from 7% to 19 % with a

mean value of 14 ± 4.47 %. It did not differ statistically from that of the strongly stained sperms in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 92).

Those with a moderate reaction ranged from 18% to 35 % with a mean value of 27.6 ± 5.34 %. They did not differ statistically from the moderately stained sperms by Sudan black B. in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 92).

Weakly stained sperms ranged from 32% to 44 % with a mean value of 37.9 ± 4.18 %. They did not also differ statistically from the weakly stained sperms by Sudan black B. in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 92).

Sperms with a negative reaction ranged from 13% to 29 % with a mean value of 20.5 ± 5.15 %. They did not also differ statistically from the negatively stained sperms by Sudan black B. in the second split fraction specimens examined after eight hours of incubation [$P < 0.05$] (Table 92).

A highly significantly negative correlation was found between the percent of the strongly stained sperms by Sudan black B. in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.804$; $P < 0.001$]. This means that the percent of such strongly stained sperms decreased with time.

A highly significantly negative correlation was found between the percent of the moderately stained sperms by Sudan black B. in the second split fraction specimens to which calcium was added and the period of incubation [$r = - 0.637$; $P < 0.001$]. This means that the percent of such moderately stained sperms decreased with time.

A highly significantly positive correlation was found between the percent of the weakly stained sperms Sudan black B. in the second split fraction specimens to which calcium was added and the period of incubation [$r = 0.929$; $P < 0.001$]. This means that the percent of such weakly stained sperms increased with time.

A highly significantly positive correlation was found between the percent of the negatively stained sperms by Sudan black B. in the second split fraction specimens to which calcium was added and the period of incubation [r = 0.642; P < 0.001]. This means that the percent of such negatively stained sperms increased with time.

II. BIOCHEMICAL RESULTS:

A. ZINC:

The concentration of zinc in whole ejaculates ranged from 85-238 µg/ml with a mean value of 163.82 ± 42.01 µg/ml. The mean value in the first split fractions is 240.12 ± 30.15 µg/ml with a range of 187-301 µg/ml. It was highly significantly higher than that of the whole ejaculates. The mean value in the second split fractions was 83.18 ± 21.92 µg/ml with a range of 53-129 µg/ml. This was highly significantly less than that of the whole ejaculates. (Tables 93 & 94).

B. FRUCTOSE:

The concentration of fructose in whole ejaculates ranged from 1.16-5.12 mg/ml with a mean value of 3.01 ± 1.05 mg/ml. The mean value in the first split fractions was 1.2 ± 0.46 mg/ml with a range of 0.49-2.21 mg/ml. This was highly significantly less than that of the whole ejaculates. The mean value in the second split fractions was 5.25 ± 1.21 mg/ml with a range of 3.14-7.78 mg/ml. It was highly significantly less than that of the whole ejaculates (Tables 93 & 94).

Table 93: The results of biochemical analysis.

WHOLE EJACULATES			SPLIT EJACULATES				
				1st fraction		2nd fraction	
No. of ejaculate	Zn	Fr	No. of ejaculate	Zn	Fr	Zn	Fr
1	175	2.83	51	211	1.6	87	3.24
2	193	2.45	52	268	0.86	89	5.43
3	122	4.91	53	278	0.98	64	5.63
4	230	1.67	54	281	0.76	53	5.87
5	143	3.22	55	210	1.24	96	3.89
6	139	4.51	56	269	0.92	93	4.98
7	210	3.6	57	223	1.86	122	3.56
8	162	4.23	58	206	1.78	125	4.67
9	180	3.26	59	294	0.49	53	6.43
10	176	2.24	60	229	1.03	78	4.87
11	146	2.51	61	197	1.88	96	4.56
12	210	2.07	62	272	0.84	50	7.13
13	225	2.67	63	221	1.54	76	5.72
14	162	3.64	64	246	1.37	73	5.56
15	238	1.46	65	253	0.98	69	6.54
16	190	2.85	66	205	1.32	82	4.89
17	133	3.05	67	216	1.67	115	5.17
18	155	4,12	68	201	2.03	99	3.56
19	205	2.89	69	221	0.65	55	6.83
20	125	4.67	70	233	1.24	96	6.12
21	196	1.72	71	263	0.91	45	6.78
22	188	2.91	72	266	0.68	54	7.45
23	85	4.12	73	215	1.56	96	4.34
24	173	2.66	74	199	2.21	123	3.67
25	164	3.54	75	251	0.83	68	5.21
26	93	3.16	76	254	0.87	86	5.87
27	110	4.88	77	273	0.61	59	5.32
28	143	2.66	78	187	0.92	87	4.81
29	192	1.28	79	226	1.72	71	4.12
30	183	2.24	80	231	1.12	65	4.56
31	122	3.61	81	278	1.12	54	6.69
32	216	3.14	82	203	2.13	106	3.32
33	152	4.78	83	249	1.26	97	3.56
34	164	3.45	84	269	0.67	73	7.78
35	221	2.8	85	245	0.59	86	5.76
36	215	1.93	86	236	1.3	93	6.2
37	123	5.12	87	260	1.43	77	5.67
38	216	1.54	88	201	1.79	129	3.78
39	193	1.16	89	227	1.33	118	4.25
40	115	4.02	90	275	0.82	99	4.98
41	126	3.42	91	264	0.71	68	4.82
42	93	1.99	92	222	1.02	96	5.19
43	125	2.63	93	232	0.99	79	5.23
44	202	1.45	94	298	0.63	62	7.66
45	143	2.31	95	203	1.63	118	3.19
46	184	1.87	96	271	0.98	94	6.13
47	100	4.5	97	301	0.56	56	6.87
48	95	3.76	98	223	1.28	84	3.93
49	161	2.08	99	235	1.88	74	4.95
50	173	2.73	100	215	1.46	71	5.77
Mean	163.82	3.01	Mean	240.12	1.2	83.18	5.25
S.D.	42.01	1.05	S.D.	30.15	0.46	21.92	1.21

Zn: Zinc concentration µg/ml. Fr: Fructose concentration mg/ml.

Table (94-a): Showing the comparison between the biochemical results in *the whole non-washed* and *first split fraction specimens.*

	Whole non-Washed Specimens.	First Split Fraction Specimens.	t	p	Signifi-cance.
Zinc Conc.	163.82 ± 41.01	240.12 ± 30.15	10.60	<0.001	+++
Fructose Conc.	3.01 ± 1.05	1.20 ± 0.46	11.108	<0.001	+++

Table (94-b): Showing the comparison between the biochemical results in *the whole non-washed* and *second split fraction specimens.*

	Whole non-Washed Specimens.	Second Split Fraction Specimens.	t	p	Signifi-cance.
Zinc Conc.	163.82 ± 41.01	83.18 ± 21.92	12.264	<0.001	+++
Fructose Conc.	3.01 ± 1.05	5.25 ± 1.21	9.890	<0.001	+++

Table (94-c): Showing the comparison between the biochemical results in *the first split fraction and second split fraction specimens.*

	First Split Fraction Specimens.	Second Split Fraction Specimens.	t	p	Signifi-cance.
Zinc Conc.	240.12 ± 30.15	83.18 ± 21.92	29.770	<0.001	+++
Fructose Conc.	1.20 ± 0.46	5.25 ± 1.21	22.134	<0.001	+++

+++ =highly significant