CHAPTER 4

RESULTS

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Results.

In the present study the effects of stimulation of opioid receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation to repeated stimulation by supramaximal shocks at a rate of 1/sec were studied.

GROUP I :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in normal krebs solution.

Control experiments (n = 5)

The results are shown in table (1). It can be observed that the mean height of the direct contraction at the onset of the experiment was 5.2=1.03 cm. The mean height of the indirect contraction at the onset of the experiment was 4.2=1.32cm. The percentage ratio between the indirect and direct response (I/D%) was 79.8=11.36.

After quarter of an hour rest in normal krebs solution the mean values of the direct and indirect responses became 4.6=1.04cm and 3.6= 0.83cm respectively. The I/D% was 78.3=8.19. As observed from table (1) there was a significant decrease in the direct response ($P < 0.01$) and N.S. decrease in the indirect. The I/D% was not changed.

After repeated stimulation for 1/2h (at the end of experiment) the mean

values of the three studied parameters (direct, indirect & I/D%) were 2±0.77cm, 1±0.5cm and 50±11.95 respectively. There was a significant decrease in the three parameters studied compared with the corresponding values at onset ($P < 0.0005$, < 0.0005 & < 0.00005 respectively) (table (1)).

Figure (1) represents one of the experiment done in this study group.

The effect of morphine.

At is to be recalled that the doses of morphine tried were 0.2mg%, 0.5mg% and 1.0mg%.

Table (2) represents morphine subgroup at a dose 0.2mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 6.0±0.6 cm. The mean height of the indirect contraction at the onset of the experiment was 4.0±0.72cm. The I/D% was 66.0±6.76.

After quarter of an hour rest in normal krebs solution in presence of morphine (0.2mg%) the mean values of the direct and indirect responses became 5±0.92cm and 3.7±0.64cm respectively. The I/D% was 74.0±5.33.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 1.3±0.51cm, 0.5±0.27cm and 38.5±18.55 respectively.

Figure (2) represents one of the experiments included in morphine subgroup at a dose 0.2mg%.

Table (3) represents morphine subgroup at a dose 0.5mg% . It included

five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.9±0.87 cm. The mean height of the indirect contraction at the onset of the experiment was 2.7±0.72cm. The I/D% was 69.2±5.0.

After quarter of an hour rest in normal krebs solution and morphine (0.5mg%) the mean values of the direct and indirect responses became 3.4±0.75cm and 2.6±0.5cm respectively. The I/D% was 76.5±6.23. As seen from this table the direct response was significantly decreased ($P<0.01$) compared with the corresponding value at onset, the I/D% was increased ($P<0.02$) and the indirect response was not changed.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 1.9±0.45cm, 0.8±0.46cm and 42.1±20.89 respectively.

Figure (3) represents one of the experiment included in morphine subgroup at a dose 0.5mg%.

Table (4) represents morphine subgroup at a dose 1.0mg%. It included seven experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.1±0.84 cm. The mean height of the indirect contraction at the onset of the experiment was 2.4±0.56cm. The I/D% was 79.2±12.79.

After quarter of an hour rest in normal krebs solution and morphine (1.0mg%) the mean values of the direct and indirect responses became 3.0±0.59cm and 2.4±0.51cm respectively. The I/D% was 80±5.21. As seen from

this table these values are not different from the values at the onset of the experiment.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 1.5±0.35cm, 0.5±0.28cm and 33.3±17.08 respectively.

Figure (4) represents one of the experiments included in morphine subgroup at a dose 1.0mg%.

Table (5) represents a comparison between the mean values of the three studied parameters in the control group carried out in normal krebs solution and the corresponding values in morphine subgroups. As observed, there was no significant difference in the direct and indirect response between morphine (0.2mg%) and control at the onset of the experiment. However, the I/D% was less than the corresponding value in the control group ($P<0.05$).

As also observed both the direct and indirect responses in morphine subgroup at a dose 0.5mg% were non significantly less than the corresponding values in the control group. Both the direct and indirect responses in morphine subgroup at a dose 1.0mg% were significantly less than the corresponding values in the control group ($P<0.005$ & <0.01).

As seen from this table, the investigated dose of morphine (0.2mg%) shows no significant difference in the direct and indirect responses as compared with the control group after 15min. rest and at the end of the experiment i.e., morphine at a dose 0.2mg% had no effect on the response of the isolated rat phrenic diaphragm preparation bathed in normal krebs solution.

Table (1) : The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirec		
1	3.9	2.6	66.7	2.9	2.3	79.3	1.0	0.4	40.0	$\frac{1}{2}$ h
2	4.8	3.6	75.0	4.5	3.6	80.0	1.9	0.8	42.1	
3	5.8	5.6	96.6	4.9	4.4	89.8	2.3	1.6	69.6	
4	5.0	3.8	76.0	5.2	3.5	67.3	3.1	1.4	45.2	
5	6.6	5.6	84.8	5.6	4.2	75.0	1.8	0.7	38.9	
Mean	5.2	4.2	79.8	4.6	3.6	78.3	2.0	1.0	50	
S.D.	1.026	1.323	11.36	1.043	0.833	8.186	0.766	0.502	11.95	
S.E.	0.459	0.592	5.081	0.466	0.368	3.661	0.343	0.225	5.342	
T				3.39	2.17	1.2	6.73	6.21	8.49	
P <				0.01*	N.S.	N.S.	.0005*	.0005*	.00005*	

* Significant decrease compared with the corresponding value at the onset of the experiment.

I = indirect response
D = direct response

Table (2) : The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	5.6	3.3	58.9	4.6	3.3	71.7	1.0	0.5	50.0	$\frac{1}{2}$ h
2	6.0	3.7	61.7	5.0	3.4	68.0	1.0	0.3	30.0	
3	6.3	4.0	63.5	5.0	3.7	74.0	1.2	0.1	8.3	
4	6.9	5.2	75.4	6.5	4.8	73.8	2.2	0.8	36.4	
5	5.4	3.8	70.4	4.0	3.3	82.5	1.1	0.6	54.5	
Mean	6.0	4.0	66.0	5.0	3.7	74.0	1.3	0.5	38.5	
S.D.	0.596	0.718	6.762	0.923	0.636	5.329	0.510	0.274	18.55	
S.E.	0.267	0.321	3.024	0.413	0.285	2.383	0.228	0.122	8.297	

Table (3): The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.0	3.0	75.0	3.5	3.0	85.7	2.2	1.5	68.2	$\frac{1}{2}$ h
2	3.0	2.0	66.7	2.7	2.2	81.5	1.8	0.5	27.8	
3	3.2	2.2	68.8	3.0	2.4	80.0	1.6	0.9	56.3	
4	4.0	2.5	62.5	3.0	2.2	73.3	1.3	0.5	38.5	
5	5.2	3.8	73.1	4.6	3.3	71.7	2.4	0.4	16.7	
Mean	3.9	2.7	69.2	3.4	2.6	76.5	1.9	0.8	42.1	
S.D.	0.868	0.721	5.004	0.752	0.502	6.228	0.447	0.458	20.89	
S.E.	0.388	0.323	2.238	0.336	0.225	2.785	0.200	0.205	9.342	
T				3.58	1.24	3.28				
P <				0.01*	N.S.	0.02				

* Significant difference compared with the corresponding value at the onset of the experiment.

Table (4) : The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (1.0mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.1	1.9	90.5	2.4	1.9	79.2	1.0	0.5	50.0	$\frac{1}{2}$ h
2	4.1	2.2	53.7	3.4	2.6	76.5	1.4	0.6	42.9	
3	3.0	2.8	93.3	2.8	2.4	85.7	1.4	0.6	42.9	
4	2.3	1.8	78.3	2.3	1.6	69.6	1.1	0.3	27.3	
5	2.5	2.0	80.0	2.9	2.4	82.8	1.9	0.6	31.6	
6	3.6	2.9	80.6	3.6	2.9	80.6	1.7	0.1	5.9	
7	4.1	3.2	78.0	3.8	3.0	78.9	1.8	1.0	55.6	
Mean	3.1	2.4	79.2	3.0	2.4	80.0	1.5	0.5	33.3	
S.D.	0.843	0.557	12.79	0.586	0.507	5.211	0.346	0.283	17.08	
S.E.	0.319	0.210	4.834	0.221	0.191	1.970	0.131	0.107	6.455	
T				1.11	0.0	0.27				
P <				N.S.	N.S.	N.S.				

Table (5): Comparison between the mean values ± S.D. of the three studied parameters in the control group carried out in normal krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		5.2±1.03	4.2±1.32	79.8±11.36	4.6±1.04	3.6±0.83	78.3±8.2	2 ±0.77	1.0±0.5	50±11.95
Morphine (0.2mg%)		6±0.6	4±0.72	66±6.76	5±0.92	3.7±0.64	74 ±5.33	1.3±0.51	0.5±0.27	38.5±18.6
	T	1.51	0.3	2.33	0.64	0.22	0.98	1.7	1.95	1.17
	P <	N.S.	N.S.	0.05	N.S.	N.S.	N.S.	N.S.	N.S	N.S.
Morphine (0.5mg%)		3.9±0.87	2.7±0.72	69.2±5.0	3.4±0.75	2.6±0.5	76.5±6.2	1.9±0.45	0.8±0.46	42.1±20.89
	T	2.16 (↑)	2.33 (↑)	1.91	2.09	2.32	0.39	0.25	0.66	0.73
	P<	N.S.	N.S.	N.S.	N.S.	0.05	N.S.	N.S.	N.S.	N.S.
Morphine (1.0mg%)		3.1±0.84	2.4±0.56	79.2±12.79	3±0.59	2.4±0.51	80±5.21	1.5±0.35	0.5±0.28	33.3±17.08
	T	3.9	3.27	0.08	3.41	3.15	0.44	1.54	2.21	1.87
	P<	0.005	0.01	N.S.	0.01	0.02	N.S.	N.S.	N.S.	N.S.

Fig. (1) : Control Experiment Carried out in Normal Krebs Solution.

Fig.(2) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of Morphine (0.2mg%).

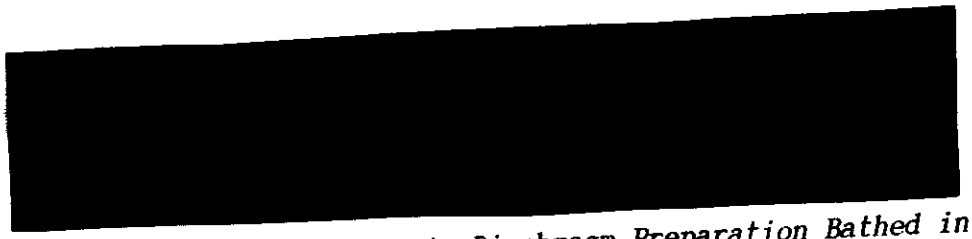

Fig.(3) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of Morphine (0.5mg%).

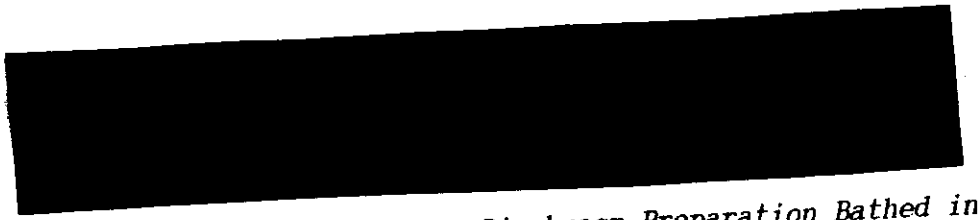

Fig.(4) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of Morphine (1.0mg%).

The effect of naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in normal krebs solution.

It is to be recalled that the doses of naloxone tried were 0.01mg%, 0.02mg% and 0.04mg%.

Table (6) and figure (5) represent naloxone subgroup at a dose 0.01mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.8±0.82cm. The mean height of the indirect contraction at the onset of the experiment was 2.7±0.38cm. The I/D% was 72.6±10.54. After quarter of an hour rest in normal krebs solution in presence of naloxone (0.01mg%), the mean values of the direct and indirect responses became 3.6±1.65cm and 1.7±0.26cm respectively. The I/D% was 56±26.15. As seen from this table the indirect response was significantly decreased (P<0.005) compared with the corresponding value at the onset, the I/D% was non significantly decreased; the direct response was not affected.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 2.6±1.14 cm, 0.2±0.12cm and 8.5 ±4.09 respectively.

Table (7) shows the subgroup done to study the effect of naloxone at a dose 0.02mg% on the response of rat diaphragm preparation. It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 5±1.16cm. The mean height of the indirect contraction at the onset of the experiment was 3.4±0.59cm. The I/D% was 69.5±10.03.

After quarter of an hour rest in normal krebs solution and naloxone (0.02mg%), the mean values of the direct and indirect responses became 4.4±1.41cm and 3.0±0.61cm respectively. The I/D% was 70.8±15.05%.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 1.8±0.58 cm, 0.4±0.2cm and 18.2±8.24% respectively.

Figure (6) represents one of the experiments included in the naloxone subgroup at a dose 0.02mg%.

Table (8) and figure (7) represent naloxone subgroup at a dose 0.04mg%. It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.7±1.44cm. The mean height of the indirect contraction at the onset of the experiment was 2.7±0.93cm. The I/D% was 73.1±6.87%.

After quarter of an hour rest in normal krebs solution the mean values of the direct and indirect responses became 2.7±1.02cm and 2.2±0.93cm respectively. The I/D% was 81.7±15.21%. As seen from this table both the direct and indirect responses were significantly decreased ($P<0.05$ & <0.001 respectively) compared with the corresponding values at the onset, the I/D% parameter was not changed.

After repeated stimulation for 1/2h (at the end of experiment) the mean values of the three studied parameters (direct, indirect & I/D%) were 1.5±0.91 cm, 0.4±0.27cm and 22.7±13.92% respectively.

Table (9) represents a comparison between the mean values of the three

studied parameters in the control group carried out in normal krebs solution and the corresponding values in naloxone subgroups. As observed both direct and indirect parameters in naloxone subgroup at a dose 0.01mg% were significantly less than the corresponding values in the control ($P<0.05$) at the onset. As also observed from table (9), there was no significant difference in the three studied parameters (direct, indirect & I/D%) between naloxone subgroup at a dose 0.02mg% and the control at the onset of the experiment.

Using naloxone at a dose 0.04mg% as seen from table (9) there was no significant difference in the direct response between naloxone subgroup (0.04mg%) and control at the onset. However, as observed the indirect response in naloxone subgroup at this dose was non significantly less than the corresponding value in control at the onset.

The investigated dose of naloxone (0.02mg%) shows no significant difference in the three studied parameters compared with the control after 15 min. rest bathing in normal krebs solution. At the end of the experiment, there was no significant difference in the direct response between naloxone (0.02mg%) and control. As observed naloxone (0.02mg%) decreased the indirect response ($P<0.05$) at the end, the I/D% was significantly decreased ($P<0.001$) (table 9). It is thus concluded that naloxone at a dose 0.02mg% decreased the indirect contraction of the diaphragm bathed in normal krebs solution at the end of experiment.

As also observed naloxone at the large dose (0.04mg%) decreased the direct contraction ($P<0.02$) after 15 min. rest as compared with the control group. However, at the end of the experiment this dose had no such effect.

Table (6): The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.5	3.2	71.1	3.0	1.5	50.0	1.7	0.2	11.8	$\frac{1}{2}$ h
2	3.7	3.0	81.1	5.5	1.6	29.1	3.2	0.2	6.3	
3	4.8	2.7	56.3	5.2	1.8	34.6	3.8	0.1	2.6	
4	2.9	2.4	82.8	1.9	1.5	78.9	1.1	0.1	9.1	
5	3.2	2.3	71.9	2.4	2.1	87.5	3.2	0.4	12.5	
Mean	3.8	2.7	72.6	3.6	1.7	56.0	2.6	0.2	8.5	
S.D.	0.817	0.384	10.54	1.648	0.255	26.15	1.142	0.123	4.089	
S.E.	0.365	0.172	4.716	0.737	0.114	11.69	0.511	0.055	1.829	
T				0.803	3.91	2.09				
P <				N.S.	0.005*	N.S.				

* Significant decrease compared with the corresponding value at the onset of the experiment.

Table (7): The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	5.3	3.6	67.9	4.3	2.7	62.8	2.3	0.4	17.4	$\frac{1}{2}$ h
2	6.2	4.2	67.7	5.6	4.0	71.4	1.9	0.3	15.8	
3	4.0	3.3	82.5	3.0	2.6	86.7	1.8	0.5	27.8	
4	3.5	2.6	74.3	3.0	2.5	83.3	0.8	0.05	6.3	
5	5.8	3.2	55.2	6.0	3.0	50.0	2.1	0.5	23.8	
Mean	5.0	3.4	69.5	4.4	3.0	70.8	1.8	0.4	18.2	
S.D.	1.16	0.585	10.03	1.408	0.612	15.05	0.581	0.195	8.239	
S.E.	0.52	0.262	4.485	0.6297	0.274	6.731	0.259	0.087	3.685	

Table (8): The response of rat phrenic diaphragm preparation bathed in normal krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.04mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.7	2.5	67.6	3.2	2.4	75.0	1.6	0.6	37.5	$\frac{1}{2}$ h
2	4.9	3.1	63.3	2.6	2.8	107.7	2.7	0.6	22.2	
3	2.4	2.0	83.3	1.7	1.3	76.5	0.7	0.2	28.6	
4	5.4	4.0	74.1	4.2	3.4	81.0	2.0	0.5	25.0	
5	2.2	1.7	77.3	1.9	1.3	68.4	0.5	0.0	00.0	
Mean	3.7	2.7	73.1	2.7	2.2	81.7	1.5	0.4	22.7	
S.D.	1.438	0.931	6.873	1.019	0.930	15.21	0.914	0.2693	13.92	
S.E.	0.643	0.417	3.074	0.456	0.415	6.802	0.409	0.1204	6.224	
T				2.79	4.67	1.16				
P <				0.05*	0.001*	N.S.				

* Significant decrease compared with the corresponding value at the onset of the experiment.

Table (9): Comparison between the mean values ± S.D. of the three studied parameters in the control group carried out in normal krebs solution and the corresponding values in Naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		5.2±1.03	4.2±1.32	79.8±11.36	4.6±1.04	3.6±0.83	78.3±8.19	2.0±0.77	1.0±0.5	50.0±11.95
Naloxone (0.01mg%)		3.8±0.82	2.7±0.38	72.6±10.54	3.6±1.65	1.7±0.26	56±26.14	2.6±1.14	0.2±0.12	8.5±4.09
	T	2.39	2.43	1.04	1.15	4.94	1.82	0.98	3.46	4.35
	P<	0.05	0.05	N.S.	N.S.	0.001	N.S.	N.S.	0.01	0.002
Naloxone (0.02mg%)		5 = 1.16	3.4±0.59	69.5±10.03	4.4±1.41	3±0.61	70.8±15.05	1.8±0.58	0.4±0.2	18.2±8.23
	T	0.29	1.24	1.52	0.26	1.31	0.98	0.47	2.49	4.9
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	0.05*	0.001*
Naloxone (0.04mg%)		3.7±1.44	2.7±0.93	73.1±6.87	2.7±1.02	2.2±0.93	81.7±15.21	1.5±0.91	0.4±0.27	22.7±13.92
	T	1.9	2.07 (↑)	1.13	2.91	2.52	0.44	0.94	2.35	3.45
	P<	N.S.	N.S.	N.S.	0.02*	0.05	N.S.	N.S.	0.05	0.01

* Significant decrease compared with the control.

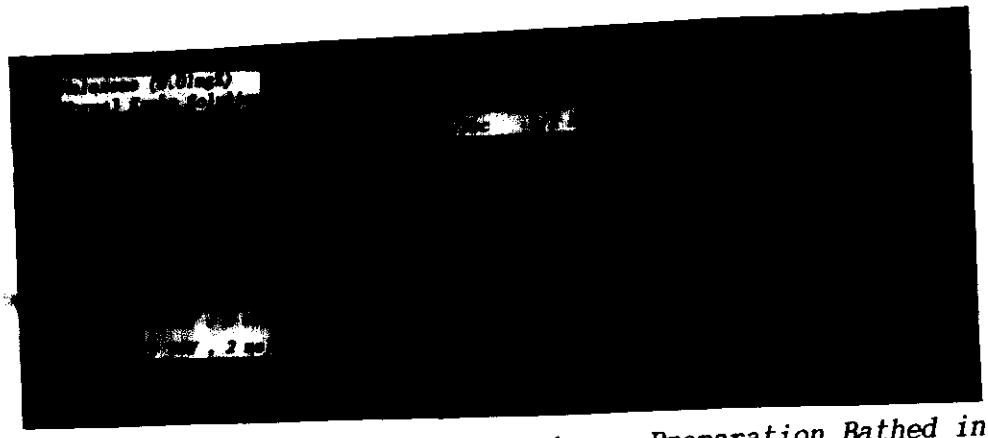

Fig.(5) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of naloxone (0.01mg%).

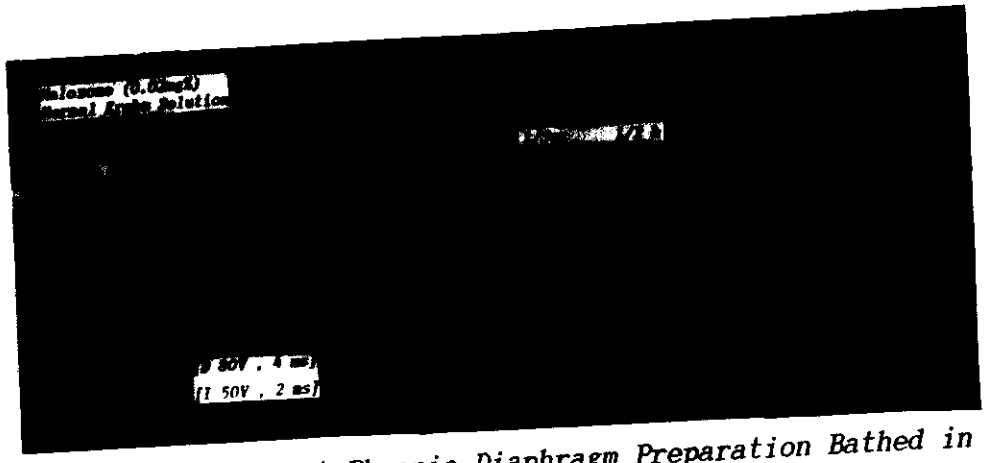

Fig.(6) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of Naloxone (0.02mg%).

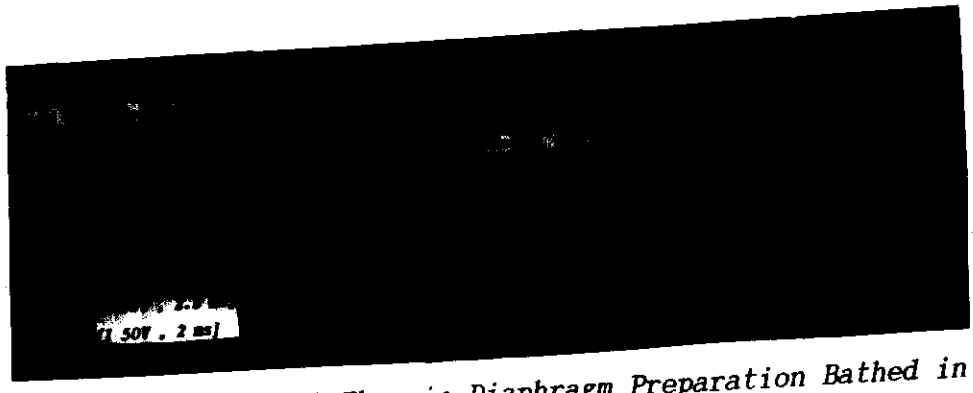

Fig.(7) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Normal Krebs Solution in Presence of Naloxone (0.04mg%).

GROUP II (A):

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in various calcium ions concentrations in the krebs solution.

According to Ca^{2+} concentrations in the krebs, this group included

Group II(A)-1 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in calcium free krebs solution.

Group II(A)-2 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in half normal concentration of calcium in krebs solution.

Group II(A)-3 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in bathed in double normal concentration of calcium in krebs solution

Group II(A)-4 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the

GROUP II (A):

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in various calcium ions concentrations in the krebs solution.

According to Ca^{2+} concentrations in the krebs, this group included

Group II(A)-1 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in calcium free krebs solution.

Group II(A)-2 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in half normal concentration of calcium in krebs solution.

Group II(A)-3 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in bathed in double normal concentration of calcium in krebs solution

Group II(A)-4 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the

response of the isolated rat phrenic diaphragm preparation bathed in bathed in 4 times normal concentration of calcium in krebs solution.

Each of the above groups included a control group, morphine subgroups and naloxone subgroups.

Group II(A)-1 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in calcium free krebs solution.

Control experiments (n = 7)

The results are shown in table (10). It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.1±1.17 cm. The mean height of the indirect contraction at the onset of the experiment was 3±0.95cm. The percentage ratio between the indirect and direct response (I/D%) was 73.1±4.97%. After quarter of an hour rest in calcium free krebs solution the mean values of the direct and indirect responses became 3.7±0.84 cm and 1±0.22cm respectively. The I/D% was 29.1±11.29%. As seen from this table both the indirect and the I/D% parameters were significantly decreased (P<0.0005 & <0.00001 respectivly) compared with the corresponding values at the onset.

At the end of experiment the mean values of the direct, indirect and I/D% were 2.6±1.1cm, 0.06±0.05cm & 1.8±1.77% respectively. As observed from this table there was complete neuromuscular block (CNMB) in three experiments after 20, 19 and 10 min. repeated stimulation.

Figure (8) represents one of the experiment included in this group.

The effect of morphine on the response of the isolated rat phrenic diaphragm preparation bathed in calcium free krebs solution.

The doses of morphine used were 0.2mg% and 0.5mg%.

Table (11) and figure (9) represent morphine subgroup at a dose 0.2mg% . It included six experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.3±1.59cm. The mean height of the indirect contraction at the onset of the experiment was 2.8±1.2cm. The I/D% was 63.4±9.03%.

After quarter of an hour rest in calcium free krebs solution and morphine (0.2mg%) the mean values of the direct and indirect responses became 3.4±1.46cm and 1.2±0.38cm respectively. The I/D% was 37±10.67%.

After repeated stimulation (at the end of experiment) the mean values of the direct, indirect and I/D% were 2.4±0.54cm, 0.09±0.07cm and 3.5±2.03% respectively. As observed from this table there was CNMB in only one experiment after 24 min. of repeated stimulation.

Table (12) represents the response of rat diaphragm bathed in calcium free solution in presence of morphine at the higher dose (0.5mg%). It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.3±1.75cm. The mean height of the indirect contraction at the onset of the experiment was 3.0±1.23cm. The I/D% was 70.4±6.03%.

After quarter of an hour rest in calcium free krebs solution and morphine (0.5mg%) the mean values of the direct and indirect responses became 3.8±1.45cm and 1.±0.66cm respectively. The I/D% was 31.7±26.14%.

After repeated stimulation (at the end of experiment) the mean values of the direct, indirect and I/D% were 2.6±1.7cm, 0.06±0.09cm and 1.4±2.16% respectively. As observed from this table there was CNMB in three experiments after 23, 12 and 28 min. repeated stimulation. Figure (10) represents one of the experiment included in this subgroup.

Table (13) shows a comparison between the mean values of the direct, indirect and I/D% in the control group carried out in calcium free krebs solution and the corresponding values in morphine subgroups. As observed, there was no significant difference in the direct and indirect response between morphine subgroups and control at the onset of the experiment. However, the I/D% in morphine subgroup at a dose 0.2mg% was significantly less than the corresponding in the control ($P<0.05$). There was no significant difference in the I/D% parameter between morphine subgroup at the large dose (0.5mg%) and the control at onset.

As observed from table (13), there was no significant difference in the studied parameters between morphine subgroups and control after 15 min. rest and at the end of the experiment. It is thus concluded that morphine at the used doses (0.2mg% & 0.5mg%) showed no observed effect on the response of the rat phrenic diaphragm preparation bathed in calcium free krebs solution.

Table (10) : The response of the isolated rat phrenic diaphragm preparation bathed in Ca^{2+} free krebs to repeated stimulation at a rate 1/sec .

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	4.5	3.2	71.1	4.6	1.0	21.7	3.2	0.1	3.1	30
2	3.9	2.7	69.2	3.6	0.6	16.7	2.3	0.1	4.3	30
3	3.5	2.6	74.3	3.1	1.1	35.5	1.6	0.0	0.0	20
4	3.0	2.0	66.7	2.8	1.3	46.4	1.2	0.0	0.0	19
5	2.6	2.0	76.9	2.8	1.1	39.3	2.0	0.0	0.0	10
6	5.4	4.4	81.5	4.6	0.9	19.6	3.7	0.1	2.7	30
7	5.7	4.1	71.9	4.5	1.1	24.4	4.1	0.1	2.4	30
Mean	4.1	3.0	73.1	3.7	1.0	29.1	2.6	0.06	1.8	24
S.D.	1.174	0.954	4.967	0.842	0.220	11.2 9	1.098	0.0535	1.77	
S.E.	0.445	0.361	1.878	0.318	0.083	4.26 8	0.415	0.0202	0.670	
P<					0.0005*	.00001*				

* Significant decrease compared with the corresponding value at the onset.

Table (11) : The response of the isolated rat phrenic diaphragm preparation bathed in Ca^{2+} free krebs to repeated stimulation at a rate 1/sec in presence of morphine(0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.5	1.6	64.0	2.0	1.1	55.0	2.2	0.1	4.5	30
2	5.1	3.8	74.5	4.7	1.3	27.7	2.5	0.1	4.0	30
3	4.5	3.2	71.1	2.9	1.3	44.8	2.5	0.1	4.0	30
4	6.9	4.4	63.8	5.7	1.8	31.6	3.4	0.2	5.9	30
5	3.0	1.7	56.7	2.4	0.8	33.3	2.0	0.05	2.5	30
6	3.8	1.9	50.0	2.7	0.8	29.6	1.9	0.0	0.0	24
Mean	4.3	2.8	63.4	3.4	1.2	37.0	2.4	0.09	3.5	29
S.D.	1.589	1.198	9.027	1.462	0.377	10.67	0.542	0.0665	2.03	
S.E.	0.649	0.489	3.685	0.597	0.154	4.355	0.221	0.0271	0.827	

Table (12) : The response of the isolated rat phrenic diaphragm preparation bathed in Ca^{2+} free krebs to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.3	1.6	69.6	2.1	0.9	42.9	1.2	0.0	0.0	23
2	2.4	1.8	75.0	2.9	2.1	72.4	1.4	0.0	0.0	12
3	5.7	4.1	71.9	5.3	0.8	15.1	4.1	0.2	4.9	30
4	5.6	4.2	75.0	5.3	1.0	18.9	4.7	0.1	2.1	30
5	5.3	3.2	60.4	3.3	0.3	9.1	1.4	0.0	0.0	28
Mean	4.3	3.0	70.4	3.8	1.0	31.7	2.6	0.06	1.4	24.6
S.D.	1.751	1.234	6.025	1.453	0.661	26.14	1.696	0.0894	2.16	
S.E.	0.783	0.552	2.695	0.65	0.296	11.69	0.758	0.04	0.965	

Table (13): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in calcium free krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		4.1±1.17	3.0±0.95	73.1±4.97	3.7±0.84	1.0±0.22	29.1±11.29	2.6±1.1	0.06±0.05	1.8 ±1.77	24
Morphine (0.2 mg%)		4.3±1.59	2.8±1.2	63.4±9.03	3.4±1.46	1.2±0.38	37±10.67	2.4±0.54	0.09±0.07	3.5±2.03	29
	T	0.26	0.34	2.45	0.46	1.19	1.29	0.40	0.09	1.62	
	P<	N.S.	N.S.	0.05	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Morphine (0.5 mg%)		4.3±1.75	3.0±1.23	70.4±6.03	3.8±1.45	1±0.66	31.7±26.14	2.6±1.7	0.06±0.09	1.4±2.16	24.6
	T	0.24	0.00	0.85	0.15	0.00	0.24	0.0	0.0	0.35	
	P<	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	

Fig. (8) : Control Experiment Carried out in Ca^{2+} Free Krebs Solution.

Fig.(9) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Ca^{2+} Free Krebs Solution in Presence of Morphine (0.2mg%).

Effect of Naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in calcium free krebs solution.

The doses of naloxone used were 0.01mg% and 0.02mg%

Table (14) represents naloxone subgroup at a dose 0.01mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.7±1.81cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±1.22cm. The I/D% was 68.4±2.59%. After quarter of an hour rest in calcium free krebs solution and naloxone (0.01mg%), the mean values of the direct and indirect responses became 3.2±1.01cm and 1.1±0.43cm respectively. The I/D% was 33.4±9.77%. As observed from this table both the indirect response and I/D% were significantly decreased (P<0.01 & <0.00005 respectivly) compared with the corresponding values at the onset.

After repeated stimulation at a rate 1/sec for 1/2h (at the end of experiment) the mean values of the direct, indirect and I/D% were 2.6±1.01cm, 0.2±0.19cm and 6.1±4.89% respectively. As observed from this table there was CNMB in one experiment after 11 min. of repeated stimulation.

Figure (11) represents one of the experiment included in the present group.

Table (15) and figure (12) represent naloxone subgroup at a dose 0.02mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.6±1.45cm. The mean height of the indirect contraction at the onset of the experiment was 2.7±1.24cm. The I/D% was 72.4±4.08%.

After quarter of an hour rest in calcium free krebs solution and naloxone (0.02mg%) the mean values of the direct and indirect responses became 3.2±1.22cm and 1.±0.16cm respectively. The I/D% was 31.8±9.08%. After repeated stimulation (at the end of experiment) the mean values of the direct, indirect and I/D% were 2.6±1.02cm, 0.04±0.05cm and 1.1±1.45% respectively. As observed from this table there was CNMB in three experiments after 12, 24 and 27 min repeated stimulation. The mean value of CNMB was 21min.

Table (16) shows a comparison between the mean values of the three studied parameters (direct, indirect & I/D%) in the control group carried out in calcium free krebs solution and the corresponding values in naloxone subgroups. As observed, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control at the onset of the experiment, after 15 min. rest and at the end. i.e, naloxone at the used doses (0.01mg% & 0.02mg%) had no observed effect on the response of the rat phrenic diaphragm preparation bathed in calcium free krebs solution.

Table (14) : The response of the isolated rat phrenic diaphragm preparation bathed in Ca^{2+} free krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.1	1.4	66.7	2.4	0.5	20.8	1.8	0.0	0.0	11
2	2.7	1.9	70.4	2.5	1.2	48.0	1.5	0.1	6.7	30
3	6.6	4.5	68.2	4.6	1.6	34.8	3.9	0.5	12.8	30
4	4.3	2.8	65.1	4.0	1.3	32.5	3.3	0.1	3.0	30
5	2.8	2.0	71.4	2.6	0.8	30.8	2.5	0.2	8.0	30
Mean	3.7	2.5	68.4	3.2	1.1	33.4	2.6	0.2	6.1	26.2
S.D.	1.813	1.216	2.59	1.0112	0.433	9.765	1.005	0.1936	4.89	
S.E.	0.811	0.544	1.158	0.452	0.1936	4.367	0.449	0.0866	2.187	
T					3.6	8.878				
P<					0.01*	.00005*				

* Significant decrease compared with the corresponding value at the onset.

Table (15) : The response of the isolated rat phrenic diaphragm preparation bathed in Ca^{2+} free krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.0	2.1	70.0	2.4	0.8	33.3	1.8	0.0	0.0	12
2	6.0	4.7	78.3	5.3	1.1	20.8	3.7	0.1	2.7	30
3	4.0	3.0	75.0	3.4	1.0	29.4	3.8	0.1	2.6	30
4	2.3	1.6	69.6	2.4	1.1	45.8	1.9	0.0	0.0	24
5	2.9	2.0	69.0	2.7	0.8	29.6	2.0	0.0	0.0	27
Mean	3.6	2.7	72.4	3.2	1.0	31.8	2.6	0.04	1.1	24.6
S.D.	1.454	1.24	4.084	1.2227	0.1581	9.081	1.017	0.0548	1.453	
S.E.	0.6504	0.5545	1.827	0.547	0.0707	4.061	0.455	0.0245	0.65	

Table (16) : *Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in calcium free krebs solution and the corresponding values in naloxone subgroups.*

Group		At Onset			After 15 Min. Rest			At End			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		4.1±1.17	3.0±0.95	73.1±4.97	3.7±0.84	1.0±0.22	29.1±11.29	2.6±1.1	0.06±0.05	1.8 ±1.77	24
Naloxone (0.01mg%)		3.7±1.81	2.5±1.22	68.4±2.59	3.2±1.01	1.1±0.43	33.4±9.77	2.6±1.01	0.2±0.19	6.1±4.89	26.2
	T	0.47	0.80	1.92	0.94	0.53	0.69	0.00	1.85	2.17	
	P <	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Naloxone (0.02mg%)		3.6±1.45	2.7±1.24	72.4±4.08	3.2±1.22	1±0.16	31.8±9.08	2.6±1.02	0.04±0.05	1.1±1.45	24.6
	T	0.66	0.48	0.26	0.84	0.00	0.44	0.0	0.63	0.72	
	P <	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	

Fig.(11) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Ca^{2+} Free Krebs Solution in Presence of Naloxone (0.01mg%).

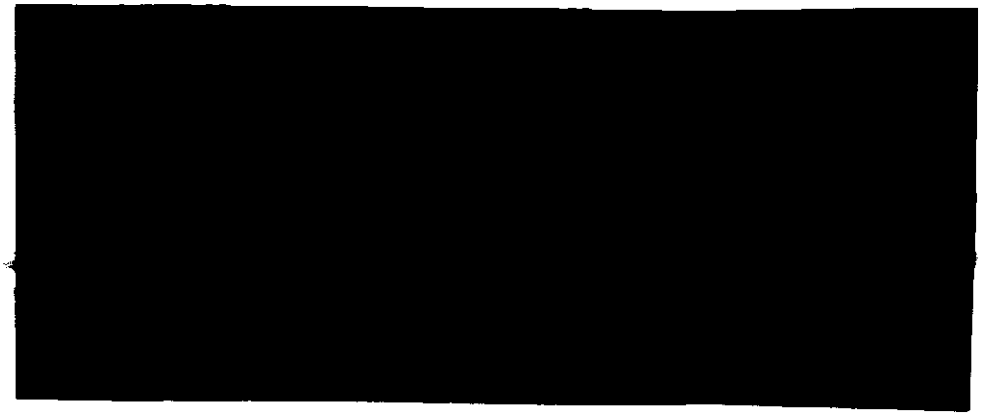

Fig.(12) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Ca^{2+} Free Krebs Solution in Presence of Naloxone (0.02mg%).

Group II(A)-2 :

The effects of stimulation of opioid receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium concentration in krebs solution.

Control experiments (n = 6)

The results are shown in table (17). It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.5±0.97 cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±0.88cm. The percentage ratio between the indirect and direct response (I/D%) was 72.1±5.09%. After quarter of an hour rest in calcium half krebs solution the mean values of the direct and indirect responses became 3.4±1.12cm and 2.4±0.93cm respectively. The I/D% was 68.2±10.99%, showing no significant difference compared with the corresponding values at the onset of the experiment.

At the end of experiment (after repeated stimulation for 1/2h) the mean values of the direct, indirect and I/D% were 1.7±0.47cm, 0.3±0.17cm and 16.6±12.14% respectively.

Figure (13) represents one of the experiment included in this subgroup.

The effect of morphine on the response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium concentration in krebs solution.

The doses of morphine used were 0.2mg%, 0.5mg% and 1.0mg%.

Table (18) represents morphine subgroup at a dose 0.2mg% (7 experiments). It can be observed that at the onset of the experiment, the mean height of the direct contraction was 4.1±0.66cm, the mean height of the indirect contraction at the onset of the experiment was 2.8±0.44cm. The I/D% was 68.2±4.58%.

After quarter of an hour rest in calcium half krebs solution and morphine (0.2mg%) the mean values of the direct and indirect responses became 3.9±0.77cm and 2.4±0.61cm respectively. The I/D% was 61.5±10.65%.

After repeated stimulation for 1/2h the mean values of the direct response, indirect response and I/D% were 2.1±0.5cm, 0.40±0.2cm and 21.0 ± 15.45% respectively.

Figure (14) represents one of the experiment included in this group.

Table (19) and figure (15) represent morphine subgroup at a dose 0.5mg% (7 experiments). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.7±0.45cm. The mean height of the indirect contraction at the onset of the experiment was 2.6±0.22cm. The I/D% was 70.8±4.56%. After quarter of an hour rest in a solution containing half the normal concentration of calcium and morphine (0.5mg%) the mean values of the direct and indirect responses became 3.7±0.62cm and 2.5±0.30cm respectively. The I/D% was 68.1 ± 9.5%. After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% became 2.2±0.4cm, 0.60±0.35cm and 29.5±17.77% respectively.

Table (20) represents the subgroup done to study the effect of morphine at a dose 1.0mg% (7 experiments). It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.4±1.14cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±0.95cm. The I/D% was 71.8±6.83%. After quarter of an hour rest in presence of 1/2 the calcium in krebs solution and morphine (1.0mg%) the mean values of the direct and indirect responses became 3.5±1.28cm and 2.4±1.07cm respectively. The I/D% was 66.7±7.8%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.6±0.68cm, 0.40±0.39cm and 24.8±16.14% respectively.

Figure (16) represents one of the experiment included in this subgroup.

A comparison between the mean values of the three studied parameters (direct, indirect & I/D%) in the control group carried out in calcium half krebs solution and the corresponding values in morphine subgroups is shown in table (21).

As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment, after 15 min. rest and at the end i.e, morphine at the three used doses (0.2mg%, 0.5mg% & 1.0mg%) had no observed effect on the response of the rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution.

Table (17) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset (after rest)		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	5.3	4.2	79.2	5.6	4.2	75.0	1.9	0.5	26.3	
2	2.5	1.8	72.0	2.6	2.0	76.9	1.1	0.4	36.4	
3	3.4	2.6	76.5	3.6	2.1	58.3	1.7	0.1	5.9	1/2h
4	3.3	2.1	63.6	2.9	2.0	69.0	1.5	0.1	6.7	
5	2.9	1.9	65.5	3.1	1.6	51.6	1.6	0.2	12.5	
6	3.3	2.5	75.8	2.8	2.2	78.6	2.5	0.3	12.0	
Mean	3.5	2.5	72.1	3.4	2.4	68.2	1.7	0.3	16.6	
S.D.	0.9685	0.8843	5.085	1.1153	0.9306	10.99	0.467	0.1673	12.14	
S.E.	0.395	0.361	2.076	0.4553	0.38	4.488	0.191	0.0683	4.955	

Table (18) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.2	2.2	68.8	2.7	1.7	63.0	1.3	0.6	46.2	
2	3.3	2.1	63.6	3.2	2.0	62.5	2.1	0.3	14.3	
3	5.1	3.1	60.8	5.0	3.0	60.0	2.5	0.1	4.0	1/2h
4	4.3	2.9	67.4	3.7	1.9	51.4	2.0	0.2	10.0	
5	4.4	3.2	72.7	4.2	2.1	50.0	2.0	0.4	20.0	
6	4.2	3.0	71.4	4.4	2.7	61.4	2.8	0.4	14.3	
7	4.0	2.9	72.5	4.0	3.3	82.5	1.7	0.65	38.2	
Mean	4.1	2.8	68.2	3.9	2.4	61.5	2.1	0.4	21	
S.D.	0.658	0.4397	4.581	0.767	0.6124	10.65	0.495	0.2	15.45	
S.E.	0.2488	0.1662	1.731	0.2899	0.2315	4.026	0.187	0.0756	5.838	

Table (19) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.9	2.9	74.4	4.7	2.8	59.6	2.9	0.5	17.2	1/2h
2	3.9	2.6	66.7	4.3	2.9	67.4	2.5	1.0	40.0	
3	3.4	2.6	76.5	2.8	2.4	85.7	2.1	0.5	23.8	
4	4.4	2.8	63.6	3.7	2.1	56.8	2.1	0.6	28.6	
5	3.6	2.6	72.2	3.4	2.2	64.7	1.7	0.8	47.0	
6	3.9	2.7	69.2	3.8	2.7	71.1	1.9	0.0	00.0	
7	3.0	2.2	73.3	3.5	2.5	71.4	2.0	1.0	50.0	
Mean	3.7	2.6	70.8	3.7	2.5	68.1	2.2	0.6	29.5	
S.D.	0.4472	0.2236	4.561	0.6205	0.3028	9.501	0.404	0.3512	17.77	
S.E.	0.169	0.0845	1.724	0.2345	0.1144	3.591	0.153	0.1327	6.717	

Table (20) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (1.0mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.2	2.4	75.0	2.8	2.1	75.0	0.7	0.1	14.3	1/2h
2	2.1	1.4	66.7	2.0	1.3	65.0	1.1	0.3	27.3	
3	2.5	1.8	72.0	3.0	1.6	53.3	1.3	0.1	7.7	
4	4.0	2.4	60.0	3.8	2.5	65.8	2.2	1.0	45.5	
5	5.6	4.4	78.6	6.1	4.6	75.4	2.5	0.9	36.0	
6	3.1	2.2	71.0	3.4	2.1	61.8	1.3	0.5	38.5	
7	3.4	2.7	79.4	3.7	2.6	70.3	2.2	0.1	4.5	
Mean	3.4	2.5	71.8	3.5	2.4	66.7	1.6	0.4	24.8	
S.D.	1.1424	0.9539	6.833	1.2839	1.0739	7.798	0.6795	0.3873	16.14	
S.E.	0.4318	0.3606	2.583	0.4853	0.4059	2.947	0.2568	0.1468	6.1	

Table (21): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in half normal calcium in krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		3.5±0.97	2.5±0.88	72.1±5.09	3.4±1.12	2.4±0.93	68.2±10.99	1.7±0.47	0.3 ±0.17	16.6±12.14
Morphine (0.2 mg%)		4.1±0.66	2.8±0.44	68.2±4.58	3.9±0.77	2.4±0.61	61.5±10.65	2.1±0.5	0.40±0.2	21 ±15.45
	T	1.33	0.79	1.46	0.95	0.0	1.11	1.86	0.97	0.56
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Morphine (0.5 mg%)		3.7±0.45	2.6±0.22	70.8±4.56	3.7±0.62	2.5±0.3	68.1±9.5	2.2±0.4	0.6 ±0.35	29.5±17.77
	T	0.49	0.29	0.49	0.61	0.27	0.02	2.07	1.9	1.49
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Morphine (1.0 mg%)		3.4±1.14	2.5±0.95	71.8±6.83	3.5±1.28	2.4±1.07	66.7±7.8	1.6±0.68	0.4 ±0.38	24.8±16.14
	T	0.17	0.00	0.09	0.15	0.00	0.29	0.3	0.58	1.02
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

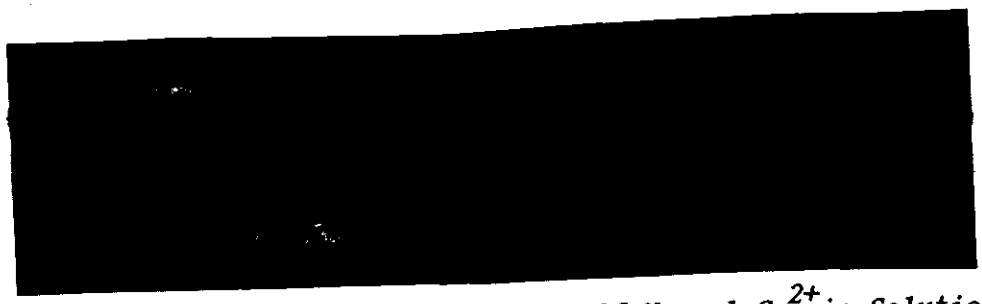

Fig. (13) : Control Experiment Carried out in Half Normal Ca^{2+} in Solution.

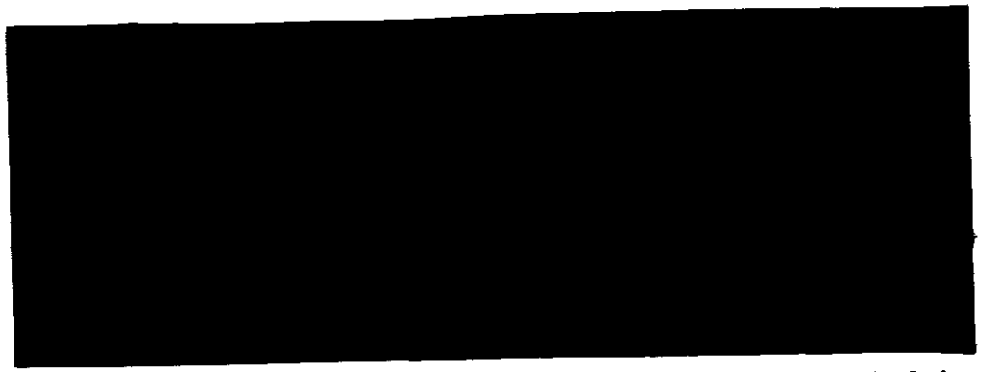

Fig.(14) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.2mg%).

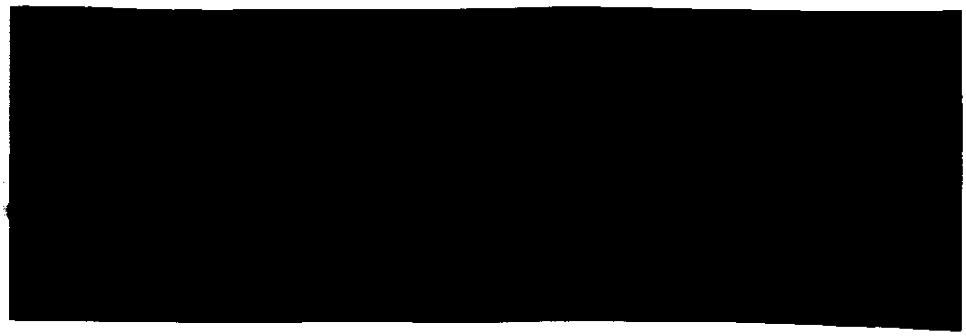

Fig.(15) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.5mg%).

Fig.(16) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal Ca^{2+} in Krebs Solution in Presence of Morphine (1.0mg%).

Effect of naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium concentration in krebs solution

The doses of naloxone used were 0.01mg% and 0.02mg%

Table (22) and figure (17) represent naloxone subgroup at a dose 0.01mg% (5 experiments). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.3±0.73cm. The mean height of the indirect contraction at the onset of the experiment was 2.3±0.57cm. The I/D% was 70.7±6.05%. After quarter of an hour rest in a solution containing half the normal concentration of calcium and naloxone (0.01mg%) the mean values of the direct and indirect responses became 3.6±0.10cm and 2.4±0.55cm respectively. The I/D% was 69.5±14.16%. After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 2.2±1.03cm, 0.3±0.32cm and 11.7±12.08% respectively.

Table (23) represents the subgroup done to study the effect of naloxone at a dose 0.02mg%. It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.7±1.18cm. The mean height of the indirect contraction at the onset of the experiment was 2.6±0.97cm. The I/D% was 70.1±7.44%. After quarter of an hour rest in half normal calcium in krebs solution in presence of naloxone (0.02mg), the mean values of the direct and indirect responses became 3.7±1.37cm and 2.8±1.11cm respectively. The I/D% was 75.6±10.58%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.7±0.79cm, 0.50±0.3cm and 28.2±17.06% respectively.

Figure (18) represents one of the experiments included in this group.

Table (24) represents a comparison between the mean values of the three studied parameters (direct, indirect & I/D%) in the control group carried out

in half calcium of krebs solution and the corresponding values in naloxone subgroups.

As observed, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control at the onset of the experiment, after 15 min. rest and at the end i.e, naloxone at the two used doses (0.01mg% & 0.02mg%) had no observed effect on the response of the rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution.

Table (22) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.9	2.8	71.8	5.3	2.2	41.5	3.6	0.4	11.1	
2	2.2	1.7	77.3	2.7	1.8	66.7	1.5	0.1	6.7	$\frac{1}{2}$ h
3	3.0	2.0	66.7	3.2	2.3	71.9	0.9	0.0	0.0	
4	4.0	3.0	75.0	3.3	3.3	100.0	2.3	0.2	8.7	
5	3.2	2.0	62.5	3.7	2.5	67.6	2.5	0.8	32.0	
Mean	3.3	2.3	70.7	3.6	2.4	69.5	2.2	0.3	11.7	
S.D.	0.7348	0.5657	6.052	0.995	0.5545	14.16	1.029	0.3162	12.08	
S.E.	0.3286	0.253	2.707	0.445	0.248	6.330	0.461	0.1414	5.401	

Table (23) : The response of the isolated rat phrenic diaphragm preparation bathed in half normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.2	1.6	72.7	2.1	1.7	81.0	1.0	0.4	40.0	
2	3.5	2.2	62.9	3.7	3.3	89.2	1.5	0.6	40.0	$\frac{1}{2}$ h
3	3.9	2.4	61.5	4.0	2.5	62.5	1.9	0.7	36.8	
4	3.5	2.7	77.1	2.8	1.9	67.9	1.0	0.0	0.0	
5	5.5	4.2	76.4	5.7	4.4	77.2	2.9	0.7	24.1	
Mean	3.7	2.6	70.1	3.7	2.8	75.6	1.7	0.5	28.2	
S.D.	1.1843	0.9708	7.437	1.3657	1.1091	10.58	0.791	0.2958	17.06	
S.E.	0.5296	0.4342	3.326	0.6107	0.496	4.731	0.354	0.1323	7.631	

Table (24): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in half normal calcium in krebs solution and the corresponding values in naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		3.5±0.97	2.5±0.88	72.1±5.09	3.4±1.12	2.4±0.93	68.2±10.99	1.7±0.47	0.3 ±0.17	16.6±12.14
Naloxone (0.01mg%)		3.3±0.73	2.3±0.57	70.7±6.05	3.6±0.1	2.4±0.55	69.5±14.16	2.2±1.03	0.30±0.32	11.7±12.08
	T	0.38	0.43	0.42	0.31	0.0	0.17	1.07	0.00	0.67
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Naloxone (0.02mg%)		3.7±1.18	2.6±0.97	70.1±7.43	3.7±1.37	2.8±1.11	75.6±10.58	1.7±0.79	0.5 ±0.30	28.2±17.06
	T	0.31	0.18	0.53	0.40	0.65	1.13	0.00	1.42	1.32
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

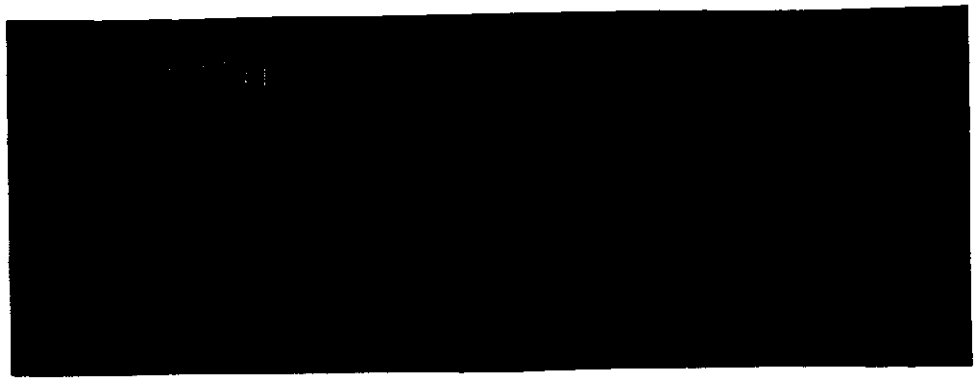

Fig.(17) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.01mg%).

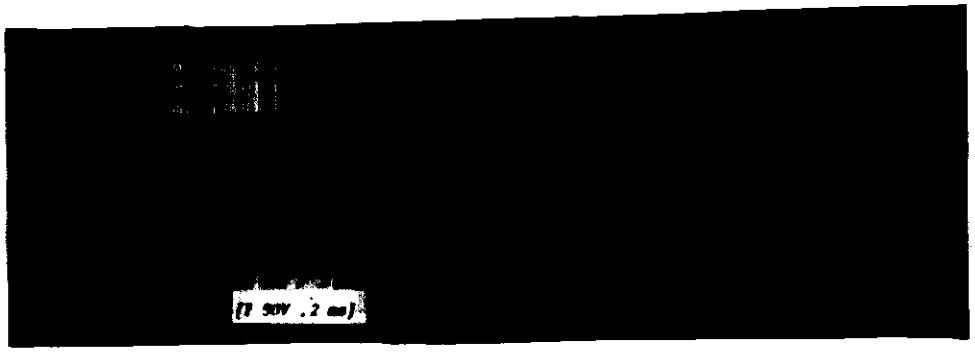

Fig.(18) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.02mg%).

Group II(A)-3 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in double the normal calcium in krebs solution.

Control experiments (n = 5)

The results are shown in table (25). It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.2±1.29 cm. The mean height of the indirect contraction at the onset of the experiment was 3±0.9cm. The percentage ratio between the indirect and direct response (I/D%) was 72.6±6.79%. After quarter of an hour rest in calcium double krebs solution the mean values of the direct and indirect responses became 2.8±0.92cm and 2.3±0.85cm respectively. The I/D% was 84.4±13.62%.

At the end of experiment, after repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.7±0.99cm, 0.5±0.52cm and 26.7±15.75 respectively.

Figure (19) represents one of the experiment included in this subgroup.

The effect of morphine.

The doses of morphine used were 0.2mg%, 0.5mg% and 1.0mg%.

Table (26) and figure (20) represent morphine subgroup at a dose 0.2mg% (5 experiments). It can be observed that the mean height of the direct contraction at the onset of the experiment was 4±0.51cm. The mean height of the indirect contraction at the onset of the experiment was 3±0.44cm. The I/D% was 75±2.77%.

After quarter of an hour rest in presence of double the normal calcium in krebs solution and morphine (0.2mg%), the mean values of the direct and indirect responses became 3.2±0.58cm and 2.7±1cm respectively. The I/D% was 84.6±22.21%.

After repeated stimulation for 1/2h the mean values of the direct, indirect and I/D% were 1.3±0.75cm, 0.4±0.4cm and 30.8±10.72% respectively.

Table (27) represents morphine subgroup at a dose 0.5mg% (5 experiments). It can be observed that, the mean height of the direct response the onset of the experiment was 3.4±0.78cm. The mean height of the indirect response at the onset of the experiment was 2.5±0.55cm. The I/D% was 72.6±7.35%.

After quarter of an hour rest in presence of double the normal calcium in krebs solution and morphine (0.5mg%), the mean values of the direct and indirect responses became 2.5±0.64cm and 2.1±0.93cm respectively. The I/D% was 83.1±17.91%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.1±0.52cm, 0.5±0.36cm and 38.7±15.33% respectively.

Figure (21) represents one of the experiment done in this group.

Table (28) represents morphine subgroup at a dose 1.0mg% (5 experiments). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.4±1.51cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±1.26cm. The I/D% was 73.5±5.35%.

After quarter of an hour rest in presence of double the calcium concentration in krebs solution and morphine (1.0mg%), the mean values of the direct and indirect responses became 2.7±1.16cm and 2.2±1.12cm respectively. The I/D% was 80±12.96%.

After repeated stimulation for 1/2h, the mean values of the direct,

indirect and I/D% were 1.3±0.72cm, 0.4±0.48cm and 29.7±23.33% respectively.

Figure (22) represents one of the experiment done in this group.

Table (29) represents a comparison between the mean values of the three studied parameters (direct, indirect & I/D%) in the control group carried out in calcium double krebs solution and the corresponding values in morphine subgroups.

As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment, after 15 min. rest and at the end i.e, morphine at the three used doses (0.2mg%, 0.5mg% & 1.0mg%) had no observed effect on the response of the rat phrenic diaphragm preparation bathed in presence of double the calcium in krebs solution.

Table (25) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.6	3.5	76.1	3.4	3.5	102.9	2.3	0.2	8.7	$\frac{1}{2}$ h
2	5.1	3.1	60.8	1.9	1.5	78.9	0.8	031	37.6	
3	2.2	1.7	77.3	1.8	1.7	94.4	0.8	0.1	12.5	
4	3.7	2.7	73.0	2.8	2.0	71.4	1.7	0.5	29.4	
5	5.4	4.1	75.9	3.9	2.9	74.4	3.1	1.4	45.2	
Mean	4.2	3.0	72.6	2.8	2.3	84.4	1.7	0.5	26.7	
S.D.	1.2903	0.90	6.79	0.92	0.85	13.62	0.99	0.52	15.75	
S.E.	0.577	0.403	3.038	0.407	0.380	6.089	0.444	0.235	7.042	

Table (26) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec. in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.0	2.9	72.5	3.8	4.4	115.8	1.0	0.2	20.0	$\frac{1}{2}$ h
2	3.3	2.4	72.7	2.3	2.0	87.0	0.6	0.1	16.7	
3	4.2	3.2	76.2	3.2	2.1	65.6	1.1	0.3	27.3	
4	3.8	3.0	78.9	3.1	2.9	93.5	1.3	0.4	30.8	
5	4.7	3.6	76.6	3.6	2.2	61.1	2.6	1.1	42.3	
Mean	4.0	3.0	75.0	3.2	2.7	84.6	1.3	0.4	30.8	
S.D.	0.5148	0.4387	2.773	0.5788	1.0037	22.21	0.75	0.397	10.72	
S.E.	0.2302	0.1962	1.24	0.2588	0.4489	9.405	0.34	0.178	4.796	

Table (27) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec. in presence of morphine(0.5mg%).

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.9	3.2	82.1	3.3	3.7	112.1	1.2	0.4	33.3	
2	4.2	2.7	64.3	2.4	2.1	87.5	1.8	1.0	55.6	$\frac{1}{2}$ h
3	2.3	1.8	78.3	1.7	1.3	76.5	0.4	0.1	25.0	
4	3.8	2.6	68.4	3.0	2.0	66.7	1.1	0.6	54.5	
5	3.0	2.1	70.0	2.2	1.6	72.7	0.8	0.2	25.0	
Mean	3.4	2.5	72.6	2.5	2.1	83.1	1.1	0.5	38.7	
S.D.	0.7778	0.5454	7.349	0.6384	0.9301	17.91	0.52	0.361	15.33	
S.E.	0.3479	0.2439	3.287	0.2855	0.4159	8.01	0.232	0.1612	6.855	

Table (28) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec. in presence of morphine(1.0mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.9	1.9	65.5	3.2	1.9	59.4	0.9	0.1	11.1	
2	1.8	1.3	72.2	1.3	1.0	76.9	0.6	0.0	00.0	$\frac{1}{2}$ h
3	2.3	1.7	73.9	1.7	1.4	82.4	0.8	0.4	50.0	
4	5.0	4.0	80.0	3.8	3.4	89.5	1.7	0.6	35.3	
5	5.0	3.8	76.0	3.7	3.4	91.9	2.3	1.2	52.2	
Mean	3.4	2.5	73.5	2.7	2.2	80.0	1.3	0.4	29.7	
S.D.	1.5116	1.2629	5.347	1.164	1.1236	12.96	0.718	0.482	23.33	
S.E.	0.676	0.565	2.391	0.5218	0.503	5.795	0.321	0.2156	10.44	

Table (29): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in double normal calcium in krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		4.2±1.29	3.0±0.9	72.6±6.79	2.8±0.92	2.3±0.85	84.4±13.62	1.7±0.99	0.5 ±0.52	26.7±15.75
Morphine (0.2 mg%)		4.0±0.51	3.0±0.44	75.0±2.77	3.2±0.58	2.7±1.0	84.6±22.21	1.3±0.75	0.4 ±0.4	30.8±10.72
	T	0.32	0.0	0.37	0.82	0.68	0.02	0.72	0.34	1.25
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Morphine (0.5 mg%)		3.4±0.78	2.5±0.55	72.6±7.35	2.5±0.64	2.1±0.93	83.1±17.91	1.1±0.52	0.5 ±0.36	38.7±15.33
	T	0.3	1.06	0.0	0.6	0.35	0.13	1.2	0.0	1.22
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Morphine (1.0 mg%)		3.4±1.51	2.5±1.26	73.5±5.35	2.7±1.16	2.2±1.12	80.0±12.96	1.3±0.72	0.4 ±0.48	29.7±23.33
	T	0.9	0.72	0.23	0.15	0.16	0.52	0.73	0.31	0.24
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

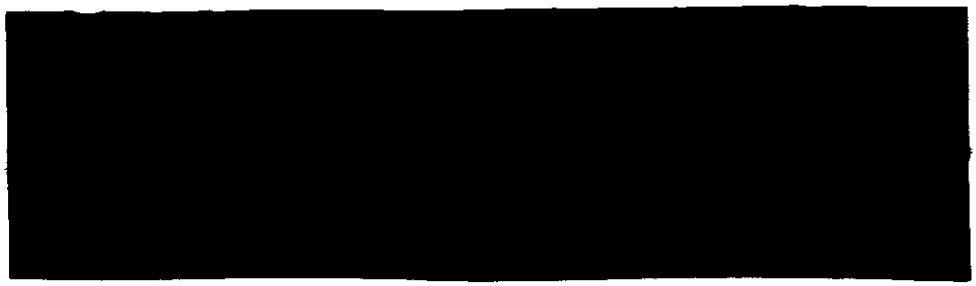

Fig. (19) : Control Experiment Carried out in Double Normal Ca^{2+} in Solution.

Fig.(20) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.2mg%).

Fig.(21) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.5mg%).

Fig.(22) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal Ca^{2+} in Krebs Solution in Presence of Morphine (1.0mg%).

Effect of Naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in double the normal calcium in krebs solution.

The doses of naloxone tried were 0.01mg% and 0.02mg%

Table (30) shows naloxone subgroup at a dose 0.01mg% (five experiments). It can be noted that the mean height of the direct response at the onset of the experiment was 3.6±1.16cm, while the indirect response was 2.5±0.81cm. The I/D% was 70.3±9.66%. After quarter of an hour rest in kreds solution containing double the calcium concentration and naloxone (0.01mg%), the mean values of the direct and indirect responses became 2.7±1.14cm and 2±0.7cm respectively. The I/D% was 74.7±7.42%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.7±0.98cm, 0.8±0.7cm and 34.2±25.88% respectively.

Figure (23) represents one of the experiments included in this subgroup.

Table (31) and figure (24) show the response of rat diaphragm bathed in double normal calcium in the krebs solution in presence of naloxone at a dose 0.02mg% (five experiments). It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.2±0.85cm. The mean height of the indirect contraction at the onset of the experiment was 2.3±0.53cm. The I/D% was 73.8±5.57%. After quarter of an hour rest, the mean values of the direct and indirect responses became 2.7±0.69cm and 2.2±0.51cm respectively. The I/D% was 84.2±15.77%.

At the end of the experiment, after repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.3±0.57cm, 0.4±0.31cm and 29.6±23.49% respectively.

Table (32) shows a comparison between the mean values of the three parameters studied (direct, indirect & I/D%) in the groups carried out in absence of naloxone and in its presence.

As observed, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control at the onset of the experiment, after 15 min. rest and at the end i.e, naloxone at the two used doses (0.01mg% & 0.02mg%) had no effect on the response of the rat phrenic diaphragm preparation bathed in double calcium concentration in krebs solution.

Table (30) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.5	2.0	80.0	1.2	0.9	75.0	0.7	0.0	0.0	
2	4.2	3.1	73.8	3.1	2.4	77.4	2.3	1.2	52.2	$\frac{1}{2}$ h
3	5.2	3.4	65.4	4.2	2.6	61.9	3.0	1.6	53.3	
4	2.5	1.4	56.0	2.0	1.6	80.0	0.8	0.1	12.5	
5	3.4	2.6	76.5	2.9	2.3	79.3	1.7	0.9	52.9	
Mean	3.6	2.5	70.3	2.7	2.0	74.7	1.7	0.8	34.2	
S.D.	1.1597	0.8124	9.660	1.1391	0.7036	7.424	0.982	0.696	25.88	
S.E.	0.5187	0.3633	4.32	0.5094	0.3146	3.320	0.439	0.3114	11.57	

Table (31) : The response of rat phrenic diaphragm preparation bathed in double normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.0	2.1	70.0	2.3	1.5	65.2	1.3	0.2	15.4	
2	1.9	1.5	78.9	2.2	2.2	100.0	0.6	0.2	33.3	$\frac{1}{2}$ h
3	3.1	2.5	80.6	2.0	2.0	100.0	1.2	0.2	16.7	
4	4.1	2.8	68.3	3.3	2.4	72.7	2.2	0.3	13.6	
5	3.8	2.7	71.1	3.5	2.9	82.9	1.3	0.9	69.2	
Mean	3.2	2.3	73.8	2.7	2.2	84.2	1.3	0.4	29.6	
S.D.	0.853	0.532	5.573	0.689	0.505	15.77	0.572	0.308	23.49	
S.E.	0.381	0.238	2.492	0.308	0.226	7.051	0.256	0.138	10.50	

Table (32): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in double normal calcium in krebs solution and the corresponding values in naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		4.2±1.29	3.0±0.9	72.6±6.79	2.8±0.92	2.3±0.85	84.4±13.62	1.7±0.99	0.5 ±0.52	26.7±15.75
Naloxone (0.01mg%)		3.6±1.16	2.5±0.81	70.3±9.66	2.7±1.14	2.0±0.7	74.7±7.42	1.7±0.98	0.8 ±0.7	34.2±25.88
	T	0.77	0.92	0.44	0.15	0.61	1.4	0.0	0.77	0.55
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Naloxone (0.02mg%)		3.2±0.85	2.3±0.53	73.8±5.57	2.7±0.69	2.2±0.51	84.2±15.77	1.3±0.57	0.4 ±0.31	29.6±23.49
	T	1.45	1.5	0.31	0.19	0.23	0.02	0.78	0.37	0.23
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

Fig.(23) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.01mg%).

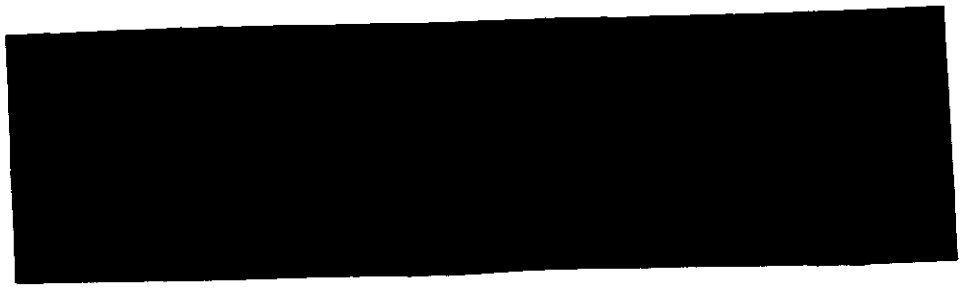

Fig.(24) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.02mg%).

Group II(A)-4 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing 4 times the normal calcium of krebs solution.

Control experiments (n = 6)

These experiments were carried out to show the response of the rat diaphragm bathed in 4 times the normal calcium in solution. The results are shown in table (33). It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.1±0.78 cm. The mean height of the indirect contraction at the onset of the experiment was 3.1±0.52cm. The percentage ratio between the indirect and direct response (I/D%) was 75.6±11.92%. After quarter of an hour rest, the mean values of the direct and indirect responses became 2.5±0.35cm and 2.1±0.67cm respectively. The I/D% was 84±17.38%. At the end of experiment, after repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1±0.12cm, 0.5±0.16cm and 50±18.8% respectively.

Figure (24) represents one of the experiment included in this group.

The effect of morphine in presence of 4 times the normal calcium concentration of krebs solution.

The doses of morphine tried were 0.2mg%, 0.5mg% and 1.0mg%.

Table (34) shows the effect of morphine at a dose 0.2mg% . It included six experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.4±1.67cm. The mean height of the indirect contraction at the onset of the experiment was 3.1±1.24cm. The I/D% was 69.3±11.77%.

After quarter of an hour rest, the mean values of the direct and indirect responses became 2.5±1.14cm and 2.5±0.85cm respectively. The I/D% was 106.3± 31.36%. As shown from this table, the direct response was significantly decreased (P<0.0002) and I/D% was increased (P<0.05) compared with the corresponding values at the onset.

After repeated stimulation at a rate of 1/sec (at the end of experiment), the mean values of the direct, indirect and I/D% were 0.62±0.32cm, 0.18±0.23cm and 23.5±28.07% respectively. As observed from this table there were CNMB in two experiments after 20 min. and 25 min. repeated stimulation.

Figure (26) represents one of the experiment included in this subgroup.

Table (35) and figure (27) show the effects of morphine at a dose 0.5mg% . It included eight experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.2±1.01cm. The mean height of the indirect contraction at the onset of the experiment was 2.9±0.72cm. The I/D% was 70.7±9.35%.

After quarter of an hour rest in presence of 4 times the normal concentration of calcium in krebs solution and morphine (0.5mg%), the mean values of the direct and indirect responses became 2.2±0.61cm and 2.1±0.69cm respectively. The I/D% was 94.9±14.86%. As seen, the direct response was significantly decreased (P<0.00001) and I/D% was increased (P<0.0005) compared with the corresponding values at the onset.

After repeated stimulation (at the end of experiment), the mean values of the direct, indirect and I/D% were 0.75±0.36cm, 0.19±0.2cm and 29.5±29.84% respectively. As observed from this table there were CNMB in three experiments after 25min., 25min and 12min. repeated stimulation, the mean time for CNMB was 20.7 min.

Table (36) shows the effect of morphine at a dose 1.0mg% (five experiments). It can be observed that, the mean height of the direct contraction at the

onset of the experiment was 3.7±1.18cm. The mean height of the indirect contraction at the onset of the experiment was 2.8±0.76cm. The I/D% was 75 ±4.64%.

After quarter of an hour rest, the mean values of the direct and indirect responses became 2.2±0.64cm and 2±0.76cm respectively. The I/D% was 95.2± 29.87%.As seen, the direct response was significantly decreased (P<0.001) and I/D% was increased (P<0.05) compared with the corresponding values at the onset.

At the end of experiment, the mean values of the direct, indirect and I/D% were 0.8±0.39cm, 0.2±0.19cm and 20.3±14.3% respectively. As observed from this table, there was CNMB in one experiment after 25 min. repeated stimulation.

Figure (28) represents one of the experiment done in this group.

Table (37) compares the mean values of the three studied parameters (direct, indirect & I/D%) in the all the groups carried out in presence of 4 times the normal concentration of calcium in krebs solution, in absence and in the presence of morphine. As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment.

After 15 min. rest, as observed from table (37), there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control i.e, morphine at the three used doses had no effect on the response of the rat phrenic diaphragm preparation after 15 min. rest bathed in calcium 4 times krebs solution.

At the end of the experiment (table 37), the mean value of the direct contraction in morphine subgroup at the smaller dose (0.2mg%) was 0.62±0.32 cm, the corresponding value in the control was 1.0±0.12cm. Morphine at this dose (0.2mg%) decreased the direct contraction (P<0.02). However, the larger two doses of morphine had no effect on the same parameter.

The mean values of the indirect response in morphine subgroups at the three doses (0.2mg%, 0.5mg% & 1.0 mg%) at the end of the experiment was significantly decreased (P<0.05, <0.02 & 0.02) compared with the experiment done in absence of morphine. Also, the I/D% was significantly decreased (P<0.02) at the dose 1.0 mg%.

As observed from table (37'), there was no significant difference between morphine subgroups carried out in 4 times calcium concentration of normal krebs solution at the onset of experiment. There was no significant difference between morphine subgroups in the three studied parameters (direct, indirect & I/D%) after 15 min. rest and at the end of the experiment.

Table (33) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.4	2.2	64.7	2.2	1.7	77.3	0.9	0.5	55.6	
2	4.2	3.2	76.2	2.2	1.4	63.3	1.0	0.2	20.0	$\frac{1}{2}$ h
3	3.4	2.8	82.4	2.1	2.0	95.2	1.1	0.4	36.4	
4	4.3	3.7	86.0	2.8	3.1	110.7	0.9	0.6	66.7	
5	3.9	3.4	87.2	2.8	2.8	100.0	1.0	0.6	60.0	
6	5.5	3.2	58.2	2.8	1.8	64.3	1.2	0.4	33.3	
Mean	4.1	3.1	75.6	2.5	2.1	84.0	1.0	0.5	50.0	
S.D.	0.778	0.523	11.92	0.349	0.669	17.38	0.118	0.1612	18.8	
S.E.	0.318	0.214	4.869	0.143	0.273	7.095	0.048	0.0658	7.677	

Table (34) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.7	1.7	63.0	1.6	2.4	150.0	0.2	0.0	0.0	20
2	6.5	4.4	67.7	3.7	3.1	83.8	1.0	0.1	10.0	30
3	3.7	3.3	89.2	1.6	2.5	156.3	0.3	0.0	0.0	25
4	5.8	4.4	75.9	3.0	3.3	110.0	0.9	0.6	66.7	30
5	5.2	2.9	55.8	3.8	2.6	68.4	0.7	0.1	14.3	30
6	2.5	1.6	64.0	1.3	0.9	69.2	0.6	0.3	50.0	30
Mean	4.4	3.1	69.3	2.5	2.5	106.3	0.62	0.18	23.5	
S.D.	1.673	1.239	11.77	1.135	0.846	31.36	0.319	0.232	28.07	
S.E.	0.683	0.506	4.803	0.463	0.345	12.8	0.13	0.095	11.46	
T				5.94		2.74				
P<				0.0002*		0.05*				

* Significant difference copmared with the corresponding values at the onset.

Table (35) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.1	2.1	67.7	1.3	1.2	92.3	0.3	0.0	0.0	25
2	4.7	3.9	83.0	3.0	3.3	110.0	0.7	0.6	85.7	30
3	4.3	3.2	74.4	2.8	2.4	85.7	0.6	0.2	33.3	30
4	3.0	2.2	73.3	1.7	1.4	82.4	0.6	0.2	33.3	30
5	3.2	2.0	62.5	1.7	1.8	105.9	0.6	0.3	50.0	30
6	4.1	3.4	82.9	2.3	2.7	117.4	1.3	0.0	00.0	25
7	5.7	3.3	57.9	2.4	2.2	91.7	1.3	0.0	0.0	12
8	5.2	3.3	63.5	2.7	2.0	74.10	0.6	0.2	33.3	30
Mean	4.2	2.9	70.7	2.2	2.1	94.9	0.75	0.19	29.5	
S.D.	1.012	0.717	9.354	0.611	0.687	14.86	0.363	0.204	29.84	
S.E.	0.358	0.254	3.307	0.216	0.243	5.254	0.128	0.072	10.55	___
T				8.7		5.33				
P<				.00001*		.0005*				

Table (36) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (1.0mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.8	2.3	82.1	1.4	1.9	135.7	0.4	0.0	0.0	25
2	5.7	4.0	70.2	3.0	3.3	110.0	0.5	0.1	20.0	30
3	3.9	3.0	76.9	2.6	1.5	57.7	1.3	0.4	30.8	30
4	2.9	2.1	72.4	1.8	1.4	77.8	1.1	0.4	36.4	30
5	3.4	2.5	73.5	2.0	1.9	95.0	0.7	0.1	14.3	30
Mean	3.7	2.8	75.0	2.2	2.0	95.2	0.80	0.2	20.3	
S.D.	1.181	0.76	4.639	0.64	0.762	29.87	0.387	0.187	14.30	
S.E.	0.528	0.34	2.074	0.286	0.341	13.36	0.173	0.084	6.39	___
T				5.17		2.39				
P<				0.001*		0.05*				

* Significant difference copmared with the corresponding values at the onset.

Table (37): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in 4 times normal calcium in solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		4.1±0.78	3.1±0.52	75.6±11.92	2.5±0.35	2.1±0.67	84.0±17.38	1 ±0.12	0.5 ±0.16	50 ± 18.8	30
Morphine (0.2 mg%)		4.4±1.67	3.1±1.24	69.3±11.77	2.5±1.14	2.5±0.85	106.3±31.4	0.62±0.32	0.18±0.23	23.5±28.07	27.5
	T	0.4	0.0	0.92	0.0	0.91	1.52	2.87	2.6	1.9	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	<0.02*	<0.05*	N.S.	
Morphine (0.5 mg%)		4.2±1.01	2.9±0.72	70.7±9.35	2.2±0.61	2.1±0.69	94.9±14.86	0.75±0.36	0.19±0.2	29.5±29.84	26.3
	T	0.2	0.58	0.86	1.07	0.0	1.26	1.29	2.97	1.46	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	<0.02*	N.S.	
Morphine (1.0 mg%)		3.7±1.18	2.8±0.76	75 ±4.64	2.2±0.64	2±0.76	95.2±29.87	0.8±0.39	0.2 ±0.19	20.3±14.3	29
	T	0.68	0.77	0.11	0.99	0.23	0.78	1.21	2.86	2.89	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	<0.02*	<0.02*	

* *Significant decrease compared with the control.*

Table (37): *Comparison between morphine subgroups carried out in 4 times normal calcium in krebs solution.*

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Morphine (0.2 mg%)		4.4±1.67	3.1±1.24	69.3±11.77	2.5±1.14	2.5±0.85	106.3±31.4	0.62±0.32	0.18±0.23	23.5±28.07	27.5
Morphine (0.5 mg%)		4.2±1.01	2.9±0.72	70.7±9.35	2.2±0.61	2.1±0.69	94.9±14.86	0.75±0.36	0.19±0.2	29.5±29.84	26.3
	T	0.28	0.38	0.25	0.64	0.98	0.91	1.07	0.0	0.38	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S .	N.S.	N.S.	
Morphine (0.2 mg%)		4.4±1.67	3.1±1.24	69.3±11.77	2.5±1.14	2.5±0.85	106.3±31.4	0.62±0.32	0.18±0.23	23.5±28.07	27.5
Morphine (1.0 mg%)		3.7±1.18	2.8±0.76	75±4.64	2.2±0.64	2±0.76	95.2±29.87	0.8±0.39	0.2±0.19	20.3±14.3	29
	T	0.78	0.47	1.01	0.87	1.02	0.6	0.94	0.0	0.23	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S .	N.S.	N.S.	
Morphine (0.5 mg%)		4.2±1.01	2.9±0.72	70.7±9.35	2.2±0.61	2.1±0.69	94.9±14.86	0.75±0.36	0.19±0.29	29.5±29.84	26.3
Morphine (1.0 mg%)		3.7±1.18	2.8±0.76	75±4.64	2.2±0.64	2±0.76	95.2±29.87	0.8±0.39	0.2±0.19	20.3±14.37	29
	T	0.74	0.22	0.86	0.0	0.25	0.02	0.0	0.0	0.64	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	

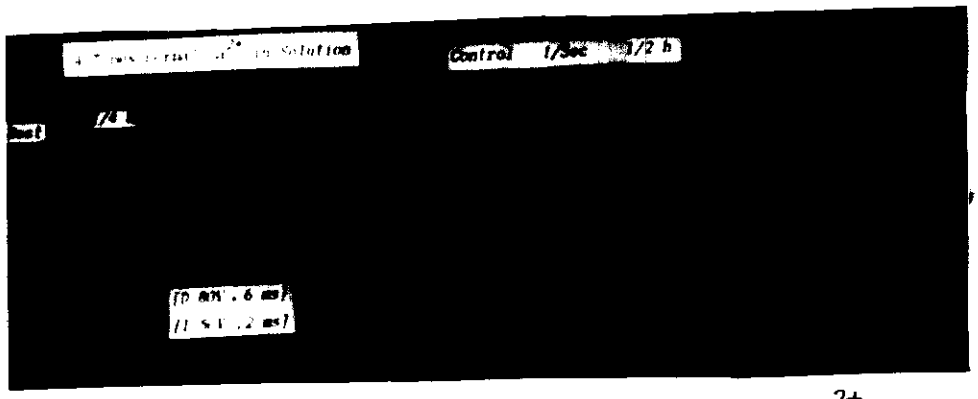

Fig. (25) : Control Experiment Carried out in 4 Times Normal Ca^{2+} in Solution.

Fig.(26) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.2mg%).

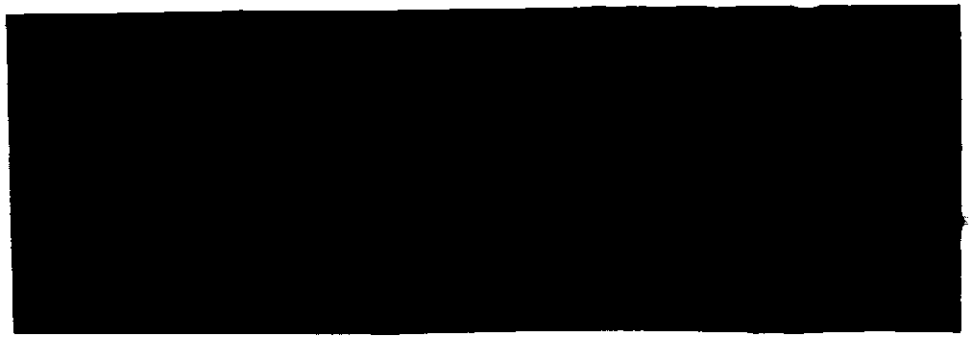

Fig.(27) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.5mg%).

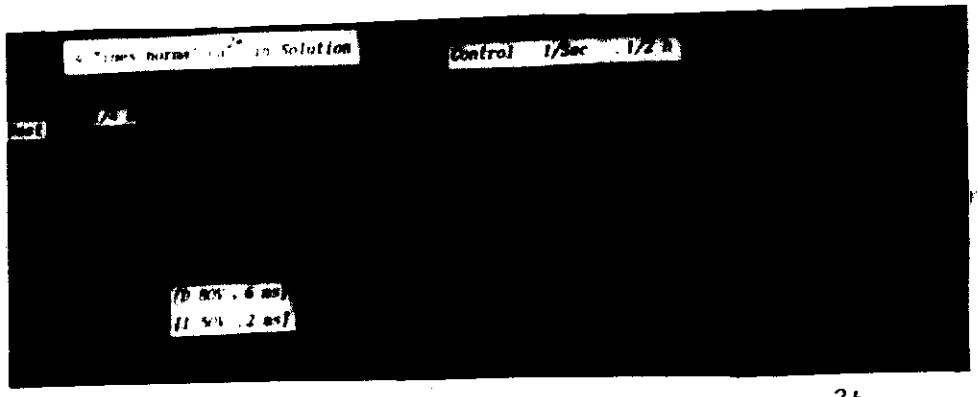

Fig. (25) : Control Experiment Carried out in 4 Times Normal Ca^{2+} in Solution.

Fig.(26) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal Ca^{2+} in Krebs Solution in Presence of Morphine (0.2mg%).

Effect of Naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing 4 times the normal calcium of krebs solution.

The doses of naloxone tried were 0.01mg% and 0.02mg%

Table (38) and figure (29) represent the effect of naloxone at a dose 0.01mg% (five experiments). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 4.5±1.73cm. The mean height of the indirect contraction at the onset of the experiment was 3.1±1.35cm. The I/D% was 69.1±7.75%. After quarter of an hour rest in a solution containing 4 times the normal concentration of calcium and naloxone (0.01mg%), the mean values of the direct and indirect responses became 3±1.34cm and 2.3±1.29cm respectively. The I/D% was 75.8±21.09%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 2.1±1.27cm, 1±0.42cm and 51.9±26.98% respectively.

Table (39) represents the effect of naloxone at a dose 0.02mg% (five experiments). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 4±1.02cm. The mean height of the indirect contraction at the onset of the experiment was 2.8±0.97cm. The I/D% was 68.3±10.53%. After quarter of an hour rest, the mean values of the direct and indirect responses became 2.8±1.11cm and 2.1±0.79cm respectively. The I/D% was 76.4±6.52%.

After repeated stimulation for 1/2h, the mean values of the direct, indirect and I/D% were 1.4±0.51cm, 0.8±0.51cm and 50.2±19.54% respectively.

Figure (30) represents one of the experiment done in this group.

Table (40) shows a comparison between the mean values of the three studied parameters (direct, indirect & I/D%) in all the groups carried out in 4 times normal calcium in krebs solution, in absence and in presence of naloxone.

As observed, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control at the onset of the experiment.

After 15 min. rest, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control i.e, naloxone at the two used doses (0.01mg% & 0.02mg%) had no effect on the response of the rat phrenic diaphragm preparation after 15 min. rest bathed in calcium 4 times krebs solution.

At the end of the experiment (table 40), the mean value of the indirect response in naloxone subgroup at the smaller dose (0.01mg%) was 1.0±0.42cm, the corresponding value in the control was 0.5±0.16cm. Naloxone at this dose (0.01mg%) increased the indirect response (P<0.02) at the end of experiment. However, the I/D% not increased, this means that naloxone at this dose (0.01mg%) also increased the direct response. The large dose of naloxone (0.02mg%) had no apparent effect.

Table (38) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.5	3.0	66.7	3.0	2.0	66.7	2.2	0.9	40.9	$\frac{1}{2}$ h
2	2.6	1.7	65.4	1.9	1.4	73.7	1.1	0.3	27.3	
3	5.1	4.2	82.4	2.7	2.9	107.4	1.5	1.2	80.0	
4	7.0	4.8	68.6	5.3	4.3	81.1	4.3	1.3	30.2	
5	3.2	2.0	62.5	2.2	1.1	50.0	1.6	1.3	81.3	
Mean	4.5	3.1	69.1	3.0	2.3	75.8	2.1	1.0	51.9	
S.D.	1.7255	1.3491	7.748	1.3444	1.2942	21.09	1.271	0.424	26.98	
S.E.	0.7717	0.6033	3.465	0.6012	0.5788	9.433	0.568	0.19	12.06	

Table (39) : The response of isolated rat phrenic diaphragm preparation bathed in 4 times normal calcium in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	2.9	1.6	55.2	1.6	1.3	81.3	0.7	0.3	42.9	$\frac{1}{2}$ h
2	4.8	4.0	83.3	2.7	2.3	85.2	2.1	1.4	66.7	
3	4.3	2.7	62.8	3.3	2.4	72.7	1.4	0.5	35.7	
4	2.9	2.1	72.4	2.0	1.4	70.0	1.3	0.4	30.8	
5	5.0	3.4	68.0	4.4	3.2	72.7	1.6	1.2	75.0	
Mean	4.0	2.8	68.3	2.8	2.1	76.4	1.4	0.8	50.2	
S.D.	1.019	0.967	10.53	1.1068	0.786	6.515	0.507	0.505	19.54	
S.E.	0.456	0.432	4.71	0.495	0.351	2.914	0.227	0.226	8.738	

Table (40) : Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in 4 times normal calcium in krebs solution and the corresponding values in naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		4.1±0.78	3.1±0.52	75.6±11.92	2.5±0.35	2.1±0.67	84.0±17.38	1.0±0.12	0.5 ±0.16	50.0±18.8
Naloxone (0.01mg%)		4.5±1.73	3.1±1.35	69.1±7.75	3.0±1.34	2.3±1.29	75.8±21.09	2.1±1.27	1.0 ±0.42	51.9±26.98
	T	0.51	0.0	1.04	0.88	0.33	0.71	2.13(↑)	2.89	0.14
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	<0.02*	N.S.
Naloxone (0.02mg%)		4.0±1.02	2.8±0.97	68.3±10.53	2.8±1.11	2.1±0.79	76.4±6.52	1.4±0.51	0.8 ±0.51	50.2±19.54
	T	0.18	0.66	1.06	0.63	0.0	0.92	1.89	1.62	0.02
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

* Significant increase compared with the control.

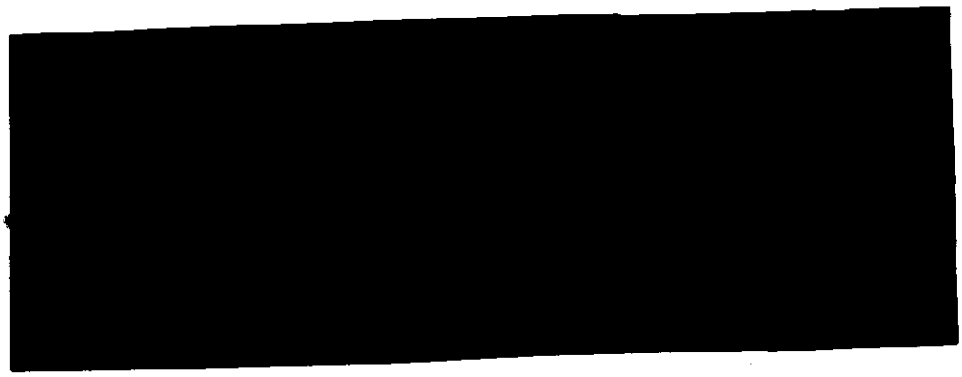

Fig.(29) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.01mg%).

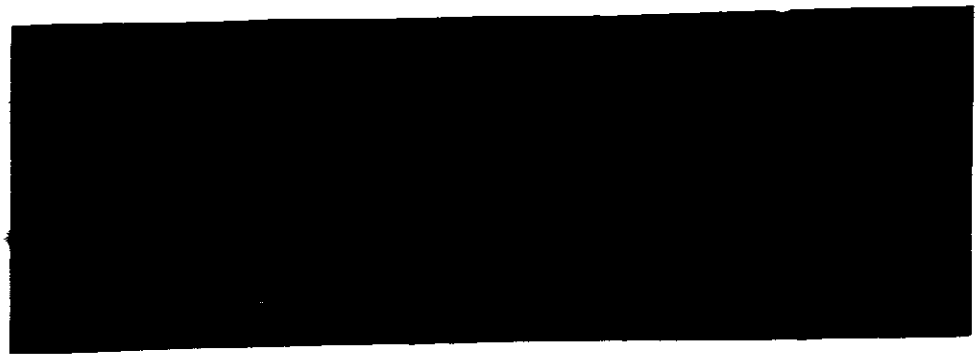

Fig.(30) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal Ca^{2+} in Krebs Solution in Presence of Naloxone (0.02mg%).

Group II (B): the effects of stimulation of opioid receptors by morphine and their blockage by naloxone in presence of various concentrations of K^+ in solution.

According to K^+ concentrations in the krebs, this group included

Group II(B)-1 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution.

Group II(B)-2 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in half the normal K^+ concentration in krebs solution.

Group II(B)-3 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in double the normal K^+ concentration in krebs solution.

Group II(B)-4 : This group demonstrates the effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in 4 times the normal K^+ concentration in krebs solution.

Each of the above groups included a control group, morphine subgroups and naloxone subgroups.

Group II(B)-1 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution.

Control experiments (n = 5).

The results are shown in table (41). It can be seen that, the mean height of the direct contraction at the onset of the experiment was 3.9±1.11 cm. The mean height of the indirect contraction at the onset of the experiment was 2.8±0.7cm. The percentage ratio between the indirect and direct response (I/D%) was 72.1±6.0%. After quarter of an hour rest in potassium free krebs solution the mean values of the direct and indirect responses became 3.8±0.59 cm and 2.4±0.39cm respectively. The I/D% was 65.6±12.56%.

At the end of experiment, after repeated stimulation at a rate of 1/sec for 1/2h, the mean values of the direct, indirect and I/D% were 1.4±0.95cm, 0.1±0.09cm and 9.8±9.9% respectively. As observed from this table there was CNMB in two preparations after 17 and 30 min.

Figure (31) represents one of the experiment included in this group.

The effect of morphine.

The doses of morphine tried were 0.2mg% and 0.5mg%

Table (42) and figure (32) show the effects of morphine at a dose 0.2mg% (five experiments). It can be observed that, the mean height of the direct

contraction at the onset of the experiment was 3.9±1.19cm. The mean height of the indirect response at the onset of the experiment was 2.4±0.56cm. The I/D% was 61.7±10.64%.

After quarter of an hour rest in potassium free krebs solution and morphine (0.2mg%), the mean values of the direct and indirect responses became 4.3±0.89cm and 2.1±0.22cm respectively. The I/D% was 51.3±11.43%.

After repeated stimulation (at the end of experiment) the mean values of the direct, indirect and I/D% were 2.3±0.98cm, 0.2 ±0.19cm and 10.5±6.82% respectively. As observed from this table there was CNMB in one preparation after 12 min. repeated stimulation.

Table (43) and figure (33) show the effects of morphine at a dose 0.5mg% (five experiments). It can be seen that, the mean height of the direct contraction at the onset of the experiment was 4.3±1.21cm. The mean height of the indirect contraction at the onset of the experiment was 3.0±0.85cm. The I/D% was 70.0±6.01%. After quarter of an hour rest in potassium free krebs solution and morphine (0.5mg%), the mean values of the direct and indirect responses became 4.4±0.84cm and 2.9±0.78cm respectively. The I/D% was 65.2 ± 10.49%.

After repeated stimulation for 1/2h (at the end of experiment), the mean values of the direct, indirect and I/D% were 2.2±0.56cm, 0.7 ±0.18cm and 36.1±18.19% respectively.

Table (44) compares the mean values of the direct, indirect and I/D% in the control group carried out in potassium free krebs solution and the corresponding values in morphine subgroups. As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment.

As also observed from table (44), there was no significant difference between morphine subgroups and control after 15 min. rest i.e, morphine

(0.2mg% & 0.5mg%) had no effect on the rat phrenic diaphragm preparation bathed in potassium free krebs solution after 15 min. rest.

At the end of experiment (Table 44), the mean value in the indirect response in morphine subgroup at a dose 0.5mg% was 0.7±0.18cm., the corresponding value in the control was 0.1±0.09cm. As observed morphine at this dose (0.5mg%) stimulated the indirect contraction (P<0.0005), the I/D% was increased (P<0.02). The smaller dose of morphine (0.2mg%) had no such effect. The direct contraction was not affected by the two used doses of morphine.

Table (41) : The response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.3	2.5	75.8	3.7	2.3	62.2	1.0	0.1	10.0	30
2	2.5	1.7	68.0	4.5	2.4	53.3	0.6	0.1	16.6	30
3	4.0	3.1	77.5	2.9	2.1	72.4	0.9	0.2	22.2	30
4	4.1	3.1	75.6	4.1	2.3	56.1	1.3	0.0	0.0	17
5	5.5	3.5	63.6	3.7	3.1	83.8	3.0	0.0	0.0	30
Mean	3.9	2.8	72.1	3.8	2.4	65.6	1.4	0.1	9.8	
S.D.	1.110	0.702	6.003	0.594	0.387	12.56	0.951	0.087	9.90	
S.E.	0.497	0.314	2.685	0.266	0.173	5.615	0.425	0.039	4.428	

Table (42) : The response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	5.0	3.0	60.0	5.0	1.9	38.0	2.7	0.5	18.5	30
2	3.0	1.6	53.3	3.9	2.3	59.0	1.8	0.2	11.1	30
3	4.9	2.5	51.0	5.2	2.4	46.2	3.7	0.0	0.0	12
4	2.6	2.0	76.9	3.0	2.0	66.7	1.1	0.1	9.1	30
5	4.0	2.7	67.5	4.3	2.0	46.5	2.2	0.3	13.6	30
Mean	3.9	2.4	61.7	4.3	2.1	51.3	2.3	0.2	10.5	
S.D.	1.1948	0.561	10.64	0.887	0.218	11.43	0.977	0.194	6.82	
S.E.	0.534	0.251	4.757	0.397	0.098	5.111	0.437	0.087	3.051	

Table (43) : The response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.7	2.9	61.7	4.7	2.9	61.7	2.8	0.6	21.4	1/2h
2	4.2	3.3	78.6	5.4	3.3	61.1	2.3	0.7	30.4	
3	2.3	1.6	69.6	3.1	1.7	54.8	1.5	0.9	60.0	
4	4.8	3.4	70.8	4.6	3.8	82.6	1.8	0.9	50.0	
5	5.5	3.8	69.1	4.4	2.2	65.9	2.7	0.5	18.5	
Mean	4.3	3.0	70.0	4.4	2.9	65.2	2.2	0.7	36.1	
S.D.	1.210	0.845	6.01	0.840	0.776	10.49	0.564	0.180	18.19	
S.E.	0.541	0.378	2.688	0.376	0.347	4.693	0.252	0.081	8.133	

Table (44) :Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in potassium free krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		3.9±1.11	2.8±0.7	72.1±6.0	3.8±0.59	2.4±0.39	65.6±12.56	1.4±0.95	0.1 ±0.09	9.8 ±9.9	27.4
Morphine (0.2 mg%)		3.9±1.19	2.4±0.56	61.7±10.64	4.3±0.89	2.1±0.22	51.3±11.43	2.3±0.98	0.2 ±0.19	10.5±6.82	26.4
	T	0.0	1.0	1.9	1.05	1.51	1.88	1.48	1.05	0.13	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Morphine (0.5 mg%)		4.3±1.21	3.0±0.85	70.0±6.01	4.4±0.84	2.9±0.78	65.2±10.49	2.2±0.56	0.7 ±0.18	36.1±18.19	30
	T	0.54	0.41	0.55	0.15	1.29	0.05	1.62	6.71	2.84	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	<0.0005*	<0.02*	

** Significant increase compared with the control.*

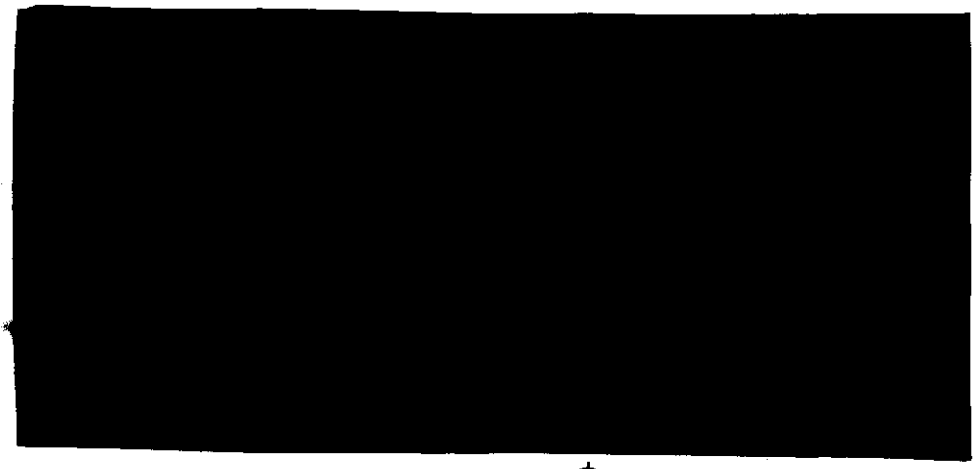

Fig.(31) : Control Experiment Carried out in K^+ Free Krebs Solution.

Fig.(32) : The Response of Rat Phrenic Diaphragm Preparation Bathed in K^+ Free Krebs Solution in Presence of Morphine (0.2mg%).

Fig.(33) : The Response of Rat Phrenic Diaphragm Preparation Bathed in K^+ Free Krebs Solution in Presence of Morphine (0.5mg%).

Effect of Naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution.

The doses of naloxone used were 0.01mg% and 0.02mg%

Table (45) shows the response of the rat phrenic diaphragm bathed in K^+ free krebs solution in presence of naloxone at a dose 0.01mg%. It included five experiments. It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.9±1.57cm. The mean height of the indirect contraction at the onset of the experiment was 2.3±0.74cm. The I/D% was 65.6±24.15%. After quarter of an hour rest in potassium free krebs solution and naloxone (0.01mg%), the mean values of the direct and indirect responses became 4.4±1.8cm and 3.1±1.48cm respectively. The I/D% was 74.3±26.41%.

After repeated stimulation for 1/2h. (at the end of experiment) the mean values of the direct, indirect and I/D% were 1.6±1.06cm, 0.26±0.42cm and 23.3±19.27% respectively. As observed from this table there was CNMB in one preparation after 30 min. repeated stimulation.

Figure (34) represents one of the experiment done in this group.

Table (46) shows the response of the rat phrenic diaphragm bathed in K^+ free krebs solution in presence of naloxone at a dose 0.02mg%. It included five experiments. It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.2±1.31cm. The mean height of the indirect contraction at the onset of the experiment was 2.1±0.36cm. The I/D% was 70.1±13.69%.

After quarter of an hour rest in potassium free krebs solution and naloxone (0.02mg%), the mean values of the direct and indirect responses became 3.0±1.24cm and 1.9±0.19cm respectively. The I/D% was 70±17.92%.

After repeated stimulation for 1/2h (at the end of experiment) the mean

values of the direct, indirect and I/D% were 1.4±0.85cm, 0.14±0.17cm and 13.8±15.26% respectively. As observed from this table there was CNMB in two preparations after 17 and 25 min. repeated stimulation.

Figure (36) represents one of the experiment included in this group.

Table (47) compares the mean values of the direct, indirect and I/D% parameters in the control group carried out in potassium free krebs solution and the corresponding values in presence of naloxone at its two doses. As observed, there was no significant difference in the three studied parameters between naloxone subgroup at a dose 0.01mg% and control at the onset of the experiment. Also, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroup at a dose 0.02mg% and control at the onset.

After 15 min. rest,naloxone at a dose 0.01mg% had no effect on the response of the rat phrenic diaphragm preparation bathed in potassium free krebs solution. The mean value of the indirect contraction in naloxone subgroup at a dose 0.02mg% was 1.9±0.19cm., the corresponding value in the control was 2.4±0.39cm. As observed naloxone at this dose (0.02mg%) decreased the indirect contraction ($P<0.05$), however, the I/D% was not decreased and the direct contraction was also not affected by this dose.

At the end of experiment, naloxone at its two doses had no effect on the response of the preparation bathed in potassium free krebs solution.

Table (45) : The response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	5.9	1.7	28.8	6.3	1.9	30.2	0.8	0.0	0.0	1/2h
2	2.6	2.4	92.3	3.2	2.6	81.3	1.1	0.3	27.3	
3	2.6	1.6	61.5	2.7	2.0	74.1	1.0	0.1	10.0	
4	5.3	3.4	64.2	6.4	5.5	85.9	3.4	1.0	29.4	
5	3.2	2.6	81.3	3.4	3.4	100.0	1.6	0.8	50.0	
Mean	3.9	2.3	65.6	4.4	3.1	74.3	1.6	0.26	23.3	
S.D.	1.568	0.735	24.15	1.799	1.479	26.41	1.059	0.419	19.27	
S.E.	0.701	0.3286	10.8	0.8044	0.6614	11.81	0.474	0.1870	8.619	

Table (46) : The response of the isolated rat phrenic diaphragm preparation bathed in K^+ free krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.9	1.9	65.5	2.4	1.8	75.0	1.2	0.0	0.0	17
2	3.0	2.3	76.7	3.2	2.1	65.6	1.1	0.4	36.4	30
3	2.1	1.7	81.0	2.1	1.7	81.0	0.8	0.1	12.5	30
4	2.4	1.9	79.2	2.3	2.0	87.0	1.0	0.2	20.0	30
5	5.4	2.6	48.1	5.1	2.1	41.2	2.9	0.0	0.0	25
Mean	3.2	2.1	70.1	3.0	1.9	70.0	1.4	0.14	13.8	24.6
S.D.	1.306	0.364	13.69	1.236	0.187	17.92	0.852	0.167	15.26	
S.E.	0.584	0.163	6.125	0.553	0.084	8.012	0.381	0.075	6.826	

Table (47): Comparison between the mean values = S.D. of the direct, indirect and I/D% parameters in the control group carried out in potassium free krebs solution and the corresponding values in naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		3.9=1.11	2.8=0.7	72.1=6.0	3.8=0.59	2.4=0.39	65.6=12.56	1.4=0.95	0.1 =0.09	9.8 =9.9	27.4
Naloxone (0.01mg%)		3.9=1.57	2.3=0.74	65.6=24.15	4.4=1.8	3.1=1.48	74.3=26.41	1.6=1.06	0.26=0.42	23.3=19.27	30
	T	0.0	1.1	0.58	0.71	1.02	0.67	0.31	0.84	1.39	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Naloxone (0.02mg%)		3.2=1.31	2.1=0.36	70.1=13.69	3.0=1.24	1.9=0.19	70.0=17.92	1.4=0.85	0.14=0.17	13.8=15.26	24.6
	T	0.91	1.98(↑)	0.3	1.3	2.6	0.45	0.0	0.47	0.49	
	P	N.S.	N.S.	N.S.	N.S.	<0.05*	N.S.	N.S.	N.S.	N.S.	

* *Significant decrease compared with the control.*

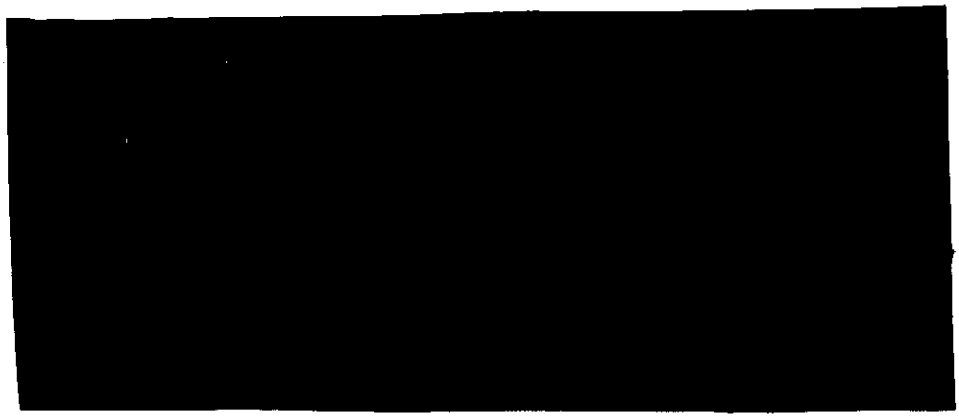

Fig.(34) : The Response of Rat Phrenic Diaphragm Preparation Bathed in K^+ Free Krebs Solution in Presence of Naloxone (0.01mg%).

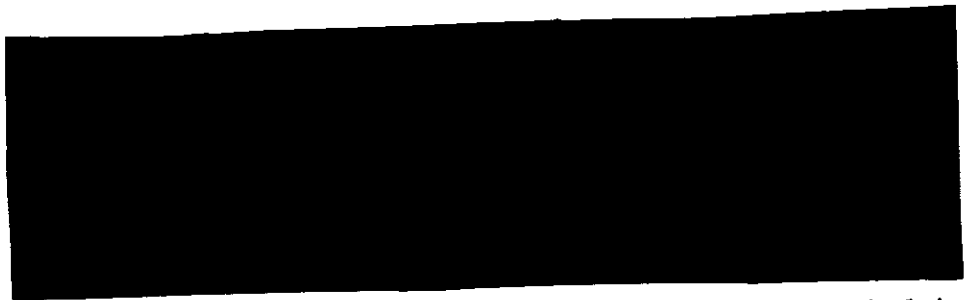

Fig.(35) : The Response of Rat Phrenic Diaphragm Preparation Bathed in K^+ Free Krebs Solution in Presence of Naloxone (0.02mg%).

Group II(B)-2 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing half the normal K^+ concentration in krebs.

Control experiments (n = 5)

The results are shown in table (48). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 5.9±0.98 cm. The mean height of the indirect contraction at the onset of the experiment was 3.4±0.73cm. The percentage ratio between the indirect and direct response (I/D%) was 58.5±15.99%. After quarter of an hour rest in potassium half krebs solution the mean values of the direct and indirect responses became 5.2±1.56 cm and 3.1±1.36cm respectively. The I/D% was 59.9±19.92%. At the end of experiment, after repeated stimulation for 1/2h. the mean values of the direct, indirect and I/D% were 2.7±1.68cm, 0.7±0.46cm and 29.8±20.74% respectively.

Figure (36) represents one of the experiment done in this group.

The effect of morphine.

The doses of morphine used were 0.2mg% and 0.5mg%

Table (49) shows the effects of morphine at a dose 0.2mg% . It included five experiments. It can be seen that the mean height of the direct contraction at the onset of the experiment was 5.5±1.96cm. The mean height of the indirect contraction at the onset of the experiment was 3.2±0.88cm. The I/D% was 60.5±11.77%.

After quarter of an hour rest in presence of morphine (0.2mg%), the mean values of the direct and indirect responses became 5.2±2.01cm and 2.9±0.74cm

respectively. The I/D% was 60.5±11.69%.

After repeated stimulation for 1/2h. (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 3±1.31cm, 0.45±0.47cm and 13.2±12.96% respectively.

Figure (37) represents one of the experiment done in this group.

Table (50) represents the effects of morphine at a dose 0.5mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 4.4±1.47cm. The mean height of the indirect contraction at the onset of the experiment was 2.6±0.44cm. The I/D% was 62.3±13.36%.

After quarter of an hour rest in a solution containing half the normal potassium concentration of krebs solution and morphine (0.5mg%), the mean values of the direct, indirect and I/D% parameters became 4±1.57cm and 0.9±1.15cm respectively. The I/D% was 49.4±14.6%.

After repeated stimulation for 1/2h. (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 1.9±1.61cm, 0.12±0.08cm and 6.9±4.55% respectively.

Figure (38) represents one of the experiment done in this group.

Table (51) compares the mean values of the direct, indirect responses and I/D% parameters in the control group and the corresponding values in morphine subgroups. As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment.

As also observed from table (51), there was no significant difference in the three studied parameters (direct, indirect & I/D%) between morphine subgroup at a dose 0.2mg% and control after 15 min. rest and at the end of the experiment i.e, morphine (0.2mg%) had no effect on the rat phrenic diaphragm preparation bathed in potassium half krebs solution.

As observed from table (51), the mean value of the indirect contraction in morphine subgroup at a dose 0.5mg% was 0.9±1.15cm. after 15 min. rest, the corresponding value in the control was 3.1±1.36cm. As observed morphine at this dose (0.5mg%) decreased the indirect contraction ($P<0.05$) after 15 min. rest, I/D% was non significantly decreased. At the end of experiment, the mean value of the indirect response in morphine subgroup at a dose (0.5mg%) was 0.12±0.08cm , the corresponding value in the control was 0.7±0.46cm. As observed morphine at this dose decreased the indirect response ($P<0.05$) at the end, the I/D% was decreased ($P<0.05$).

Table (48) : The response of isolated rat phrenic diaphragm preparation bathed in half normal K^+ concentration in krebs solution to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	7.5	3.5	46.7	7.5	3.8	50.7	5.0	1.4	28.0	1/2h
2	5.4	2.7	50.0	4.4	2.6	59.1	1.1	0.7	63.6	
3	5.0	4.2	84.0	5.3	4.8	90.6	1.3	0.1	7.7	
4	6.2	4.0	64.5	5.6	3.5	62.5	3.9	0.8	20.5	
5	5.5	2.6	47.3	3.3	1.2	36.4	2.4	0.7	29.2	
Mean	5.9	3.4	58.5	5.2	3.1	59.9	2.7	0.7	29.8	
S.D.	0.984	0.731	15.99	1.558	1.36	19.92	1.684	0.464	20.74	
S.E.	0.44	0.327	7.153	0.697	0.608	8.907	0.753	0.2074	9.276	

Table (49) : The response of isolated rat phrenic diaphragm preparation bathed in half normal K^+ concentration in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	7.0	3.5	50.0	7.0	3.3	47.1	4.7	0.8	17.0	1/2h
2	7.2	3.5	48.6	7.0	3.6	51.4	2.6	0.15	5.8	
3	5.6	4.0	71.4	5.4	3.2	59.3	3.2	1.3	34.4	
4	5.3	3.1	58.5	4.1	2.9	70.7	1.1	0.05	4.5	
5	2.3	1.7	73.9	2.3	1.7	73.9	3.4	0.15	4.4	
Mean	5.5	3.2	60.5	5.2	2.9	60.5	3.0	0.45	13.2	
S.D.	1.964	0.878	11.77	2.009	0.738	11.69	1.31	0.47	12.96	
S.E.	0.878	0.392	5.264	0.898	0.33	5.232	0.586	0.210	5.796	

Table (50) : The response of isolated rat phrenic diaphragm preparation bathed in half normal K^+ concentration in krebs solution to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.4	2.4	70.6	2.8	1.8	64.3	1.4	0.2	14.3	1/2h
2	4.6	2.5	54.3	3.6	2.1	58.3	1.1	0.05	4.5	
3	6.7	3.4	50.7	6.7	2.4	35.8	3.7	0.2	5.4	
4	4.5	2.4	53.3	4.1	1.3	31.7	2.0	0.05	2.5	
5	2.9	2.4	82.8	3.0	1.7	56.7	1.3	0.1	7.7	
Mean	4.4	2.6	62.3	4.0	0.9	49.4	1.9	0.12	6.9	
S.D.	1.465	0.439	13.86	1.573	1.151	14.60	1.61	0.076	4.549	
S.E.	0.655	0.196	6.12	0.704	0.515	6.53	0.474	0.034	2.034	

Table (51): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in half normal potassium concentration in krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		5.9±0.98	3.4±0.73	58.5±15.99	5.2±1.56	3.1±1.36	59.9±19.92	2.7±1.68	0.7 ±0.46	29.8±20.74
Morphine (0.2 mg%)		5.5±1.96	3.2±0.88	60.5±11.77	5.2±2.01	2.9±0.74	60.5±11.69	3.0±1.31	0.45±0.47	13.2±12.96
	T	0.41	0.39	0.23	0.0	0.29	0.06	0.31	0.58	1.52
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Morphine (0.5 mg%)		4.4±1.47	2.6±0.44	62.3±13.86	4.0±1.57	0.9±1.15	49.4±14.6	1.9±1.61	0.12±0.08	6.9 ±4.55
	T	1.9	2.1	0.4	1.21	2.76	0.95	0.9	2.76	2.41
	P<	N.S.	N.S.	N.S.	N.S.	0.05*	N.S.	N.S.	0.05*	0.05*

* *Significant decrease compared with the control.*

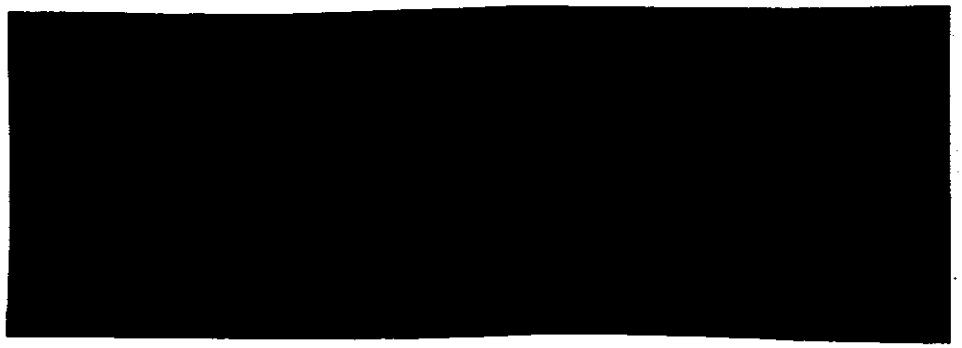

Fig.(36) :Control Experiment Carried out in Half Normal K^+ in Krebs Solution.

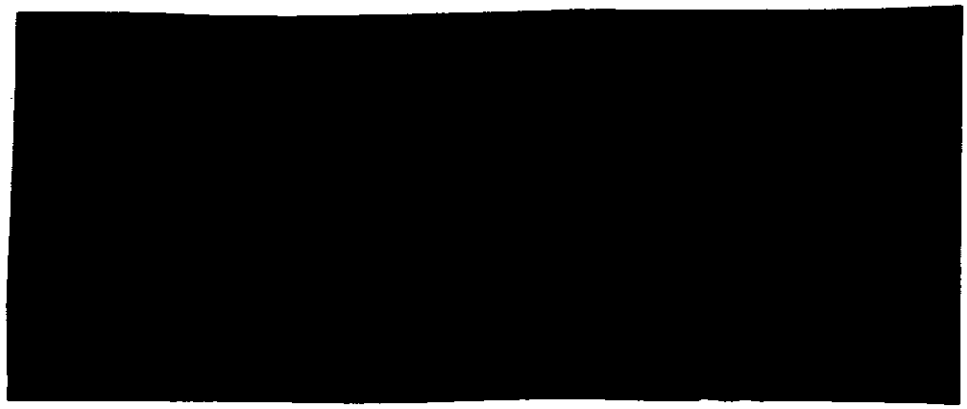

Fig.(37) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal K in Krebs Solution in Presence of Morphine (0.2mg%).

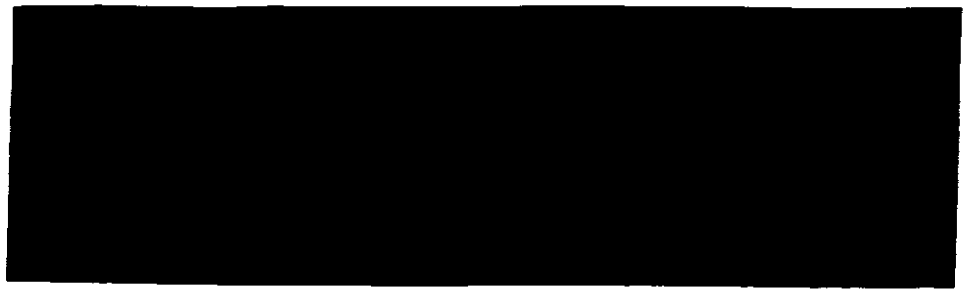

Fig.(38) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal K^+ in Krebs Solution in Presence of Morphine (0.5mg%).

The effect of Naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing half the normal K^+ concentration in krebs.

The doses of naloxone investigated were 0.01mg% and 0.02mg%

Table (52) represents the response of rat diaphragm bathed in half normal K^+ in the krebs solution in presence of naloxone at a dose 0.01mg% . It included five experiments. It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.7±1.36cm. The mean height of the indirect contraction at the onset of the experiment was 2.6±0.72cm. The I/D% was 72±6.85%. After quarter of an hour rest, the mean values of the direct and indirect responses became 3.3±1.16cm and 2.3±0.66cm respectively. The I/D% was 76.3±28.7%.

After repeated stimulation for 1/2h. (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 1.8±0.95cm, 0.4±0.19cm and 21±8.39% respectively.

Figure (39) represents one of the experiment done in this group.

Table (53) and figure (40) represent the response of rat diaphragm bathed in half normal K^+ in the krebs solution in presence of naloxone at a dose 0.02mg% . It included five experiments. It can be observed that the mean height of the direct response at the onset of the experiment was 4.9±1.26cm. The mean height of the indirect contraction at the onset of the experiment was 3.4±0.37cm. The I/D% was 72.2±10.71%. After quarter of an hour rest, the mean values of the direct and indirect responses became 5.0±1.13cm and 3.2±0.38cm respectively. The I/D% was 68.1±17.39%.

At the end of experiment, the mean values of the direct, indirect and I/D% parameters were 3.2±0.7cm, 0.9±0.27cm and 28.6±14.99% respectively.

Table (54) compares the mean values of the studied parameters in the control group carried out in half the normal K^+ concentration in the krebs

solution and the corresponding values in presence of naloxone at its two doses. As seen, there was no significant difference between naloxone at a dose 0.01mg% and control in the studied parameter (indirect response) after 15 min. rest and at the end of experiment i.e, naloxone (0.01mg%) had no observed effect on the indirect contraction of the preparation bathed in half potassium concentration in krebs solution. It can be also seen that, there was no observed effect of naloxone (0.02mg%) on direct, indirect and I/D% after 15 min. and at the end of the experiment when compared with the control values.

Table (52) : The response of isolated rat phrenic diaphragm preparation bathed in half normal K^+ concentration in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	5.8	3.7	63.8	4.9	2.0	40.8	1.8	0.6	33.3	1/2h
2	3.4	2.5	73.5	2.4	2.9	120.8	1.3	0.3	23.1	
3	3.9	2.6	66.7	4.0	3.1	77.5	3.4	0.5	14.7	
4	2.1	1.7	81.0	2.1	1.5	71.4	0.9	0.2	22.2	
5	3.2	2.4	75.0	3.1	2.2	71.0	1.7	0.2	11.8	
Mean	3.7	2.6	72.0	3.3	2.3	76.3	1.8	0.40	21.0	
S.D.	1.356	0.719	6.848	1.155	0.6595	28.7	0.953	0.187	8.389	
S.E.	0.606	0.322	3.063	0.517	0.295	12.83	0.426	0.084	3.752	

Table (53) : The response of isolated rat phrenic diaphragm preparation bathed in half normal K^+ concentration in krebs solution to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.8	3.2	84.2	4.6	3.1	67.4	2.8	0.8	28.6	1/2h
2	4.6	3.6	78.3	4.5	3.6	80.0	2.4	1.3	54.2	
3	5.0	3.3	66.0	5.2	3.6	69.2	4.1	0.9	22.0	
4	7.0	4.0	57.1	6.8	2.7	39.7	3.8	0.6	15.8	
5	4.1	3.1	75.6	3.8	3.2	84.2	3.1	0.7	22.6	
Mean	4.9	3.4	72.2	5.0	3.2	68.1	3.2	0.9	28.6	
S.D.	1.261	0.367	10.71	1.1325	0.3808	17.39	0.704	0.2739	14.99	
S.E.	0.5639	0.164	4.792	0.507	0.170	7.775	0.315	0.123	6.704	

Table (54): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in half normal potassium concentration in krebs solution and the corresponding values in naloxone subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.		
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%
Control		5.9±0.98	3.4±0.73	58.5±15.99	5.2±1.56	3.1±1.36	59.9±19.92	2.7±1.68	0.7 ±0.46	29.8±20.74
Naloxone (0.01mg%)		3.7±1.36	2.6±0.72	72.0±6.85	3.3±1.16	2.3±0.66	76.3±28.7	1.8±0.95	0.40±0.19	21±8.39
	T	2.9	1.7	1.7	2.19	1.18	1.05	1.04	1.34	0.88
	P<	0.02*	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Naloxone (0.02mg%)		4.9±1.26	3.4±0.37	72.2±10.71	5.0±1.13	3.2±0.38	68.1±17.39	3.2±0.70	0.90±0.27	28.6±14.99
	T	1.4	0.0	1.59	0.23	1.58	0.69	0.61	0.83	0.1
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

* *Significant decrease compared with the control.*

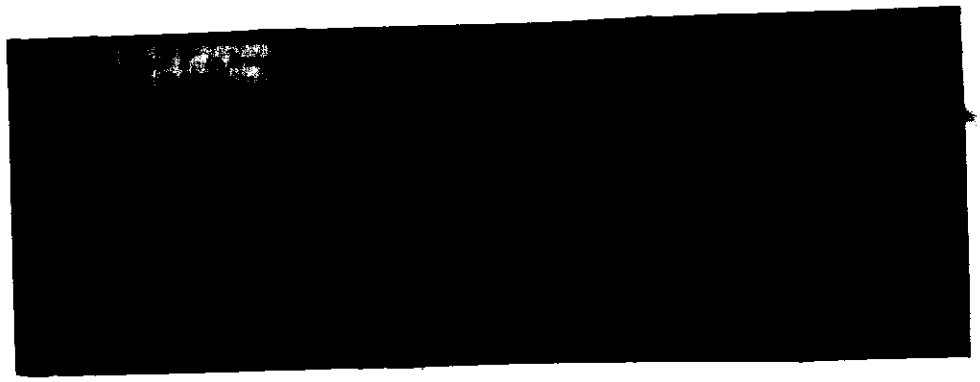

Fig.(39) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal K^+ in Krebs Solution in Presence of Naloxone (0.01mg%).

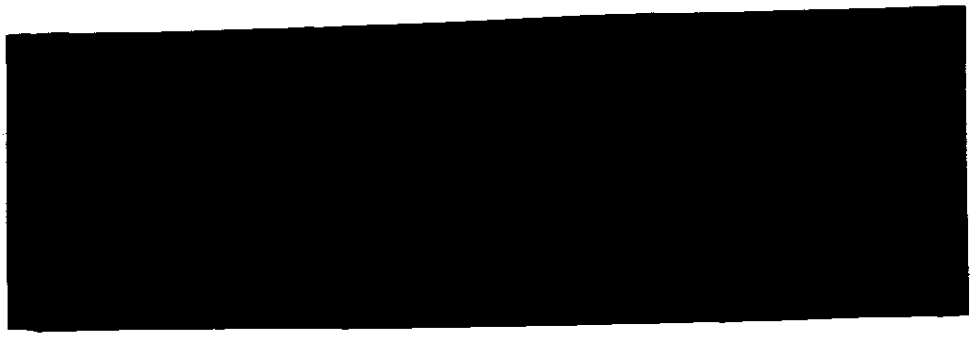

Fig.(40) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Half Normal K^+ in Krebs Solution in Presence of Naloxone (0.02mg%).

Group II(B)-3 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing double the K^+ concentration in krebs solution.

Control experiments (n=6)

The results are shown in table (55) and figure (41). It can be observed that, the mean height of the direct contraction at the onset of the experiment was 4.1±0.88 cm. The mean height of the indirect contraction at the onset of the experiment was 2.9±0.58cm. The percentage ratio between the indirect and direct response (I/D%) was 70.1±4.38%. After quarter of an hour rest, the mean values of the direct and indirect responses became 2.8±0.35 cm and 2.7±0.65cm respectively. The I/D% was 99±29.89%. As seen the direct response was significantly decreased (P<0.02) and the I/D% was increased (P<0.05) as compared with the corresponding values at the onset.

At the end of experiment, after repeated stimulation, the mean values of the direct, indirect and I/D% parameters were 1.5±0.35cm, 0.5±0.38cm and 31.1±24.87% respectively. As observed from table (55), there was CNMB in one preparation after 25 min. repeated stimulation.

The effect of morphine on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing double the K^+ concentration in krebs solution.

The doses of morphine tried were 0.2mg% and 0.5mg%

Table (56) shows the subgroup done to study the effects of morphine at a dose 0.2mg% . It included five experiments. It can be seen that, the mean height of the direct contraction at the onset of the experiment was 3.3±0.76cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±0.66cm. The I/D% was 74.4±5.99%. After quarter of an hour rest in presence of double the potassium concentration in krebs solution and morphine (0.2mg%), the mean values of the direct and indirect responses became 2.5±0.77cm and 2.3±0.85cm respectively. The I/D% was 88.4±19.86%.

After repeated stimulation (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 1.4±0.9cm, 0.5±0.46cm and 28.7±20.61% respectively. As observed from table (56) there was CNMB in one preparation after 27 min. repeated stimulation. Figure (42) represents one of the experiment done in this group.

Table (57) shows the subgroup done to study the effects of morphine at a dose 0.5mg% . It included five experiments. It can be observed that the mean height of the direct contraction at the onset of the experiment was 3.9±1.23cm. The mean height of the indirect contraction at the onset of the experiment was 2.5±0.73cm. The I/D% was 65.6±7.63%.

After quarter of an hour rest in presence of double the potassium concentration of normal krebs solution and morphine (0.5mg%), the mean values of the direct, indirect and I/D% parameters became 2.5±0.91cm and 2.2±0.78cm respectively. The I/D% was 85.7±18.22%.

After repeated stimulation for 1/2h, (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 1.5±0.76cm, 0.50±0.38cm and 28.7±10.65% respectively.

Figure (43) represents one of the experiment done in this group.

Table (58) represents a comparison between the mean values of the direct, indirect and I/D% parameters in the three groups done to demonstrate the effects of morphine in presence of double the K^+ concentration in krebs solution. As observed, there was no significant difference in the direct, indirect and I/D% parameters between morphine subgroups and control at the onset of the experiment. There was no significant difference in the three studied parameters (direct, indirect & I/D%) between morphine subgroups and control after 15 min. rest and at the end of the experiment. It is thus concluded that morphine (0.2mg% & 0.5mg%) had no observed effect on the response of the rat phrenic diaphragm preparation bathed in a solution containing double the K^+ concentration of krebs solution.

Table (55) : The response of rat phrenic diaphragm preparation bathed in double the normal K^+ in krebs to repeated stimulation at a rate 1/sec.

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.1	2.8	68.3	2.8	2.2	78.6	1.6	0.4	25.0	30
2	5.7	3.8	66.7	2.4	3.6	150.0	1.6	0.7	43.8	30
3	3.8	2.6	68.4	3.2	3.2	100.0	1.3	0.0	0.0	25
4	4.4	3.4	77.3	3.0	3.0	100.0	2.1	0.9	42.9	30
5	3.5	2.4	68.6	2.4	2.5	104.2	1.2	0.8	66.7	30
6	3.2	2.4	75.0	3.1	1.9	61.3	1.2	0.1	8.3	30
Mean	4.1	2.9	70.1	2.8	2.7	99.0	1.5	0.5	31.1	
S.D.	0.884	0.576	4.377	0.349	0.645	29.89	0.346	0.3768	24.87	
S.E.	0.361	0.235	1.787	0.143	0.263	12.20	0.141	0.1538	10.15	
T				2.89		2.66				
P<				0.02*		0.05*				

* Significant difference compared with the corresponding values at the onset.

Table (56) : The response of rat phrenic diaphragm preparation bathed in double the normal K^+ in krebs to repeated stimulation at a rate 1/sec in presence of morphine (0.2mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	3.7	2.9	78.4	2.6	2.9	111.5	1.7	0.6	35.3	30
2	3.1	2.2	71.0	1.9	1.8	94.7	1.3	0.2	15.4	30
3	4.1	3.3	80.5	2.9	2.8	96.6	1.0	0.5	50.0	30
4	3.5	2.3	65.7	3.6	2.9	80.6	2.8	1.2	42.9	30
5	2.1	1.6	76.2	1.7	1.0	58.8	0.4	0.0	00.0	27
Mean	3.3	2.5	74.4	2.5	2.3	88.4	1.4	0.5	28.7	
S.D.	0.762	0.6595	5.994	0.7714	0.8529	19.86	0.897	0.4583	20.61	
S.E.	0.341	0.295	2.681	0.345	0.3814	8.881	0.401	0.205	9.218	

Table (57) : The response of rat phrenic diaphragm preparation bathed in double the normal K^+ in krebs to repeated stimulation at a rate 1/sec in presence of morphine (0.5mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	4.9	3.4	69.4	3.8	2.9	76.3	2.5	1.0	40.0	
2	2.2	1.7	77.3	1.9	1.7	89.5	1.1	0.3	27.3	
3	5.0	3.0	60.0	3.0	2.2	73.3	2.2	0.8	36.4	1/2h
4	3.1	1.9	61.3	1.5	1.1	73.3	0.8	0.1	12.5	
5	4.5	2.7	60.0	2.5	2.9	116.0	1.1	0.3	27.3	
Mean	3.9	2.5	65.6	2.5	2.2	85.7	1.5	0.5	28.7	
S.D.	1.234	0.725	7.625	0.908	0.781	18.22	0.758	0.381	10.65	
S.E.	0.552	0.324	3.41	0.406	0.349	8.15	0.339	0.1703	4.761	

Table (58): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in double the normal potassium in krebs solution and the corresponding values in morphine subgroups.

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		4.1±0.88	2.9±0.58	70.1±4.38	2.8±0.35	2.7±0.65	99.0±29.89	1.5±0.35	0.5 ±0.38	31.1±24.87	29.2
Morphine (0.2 mg%)		3.3±0.76	2.5±0.66	74.4±5.99	2.5±0.77	2.3±0.85	88.4±19.86	1.4±0.90	0.5 ±0.46	28.7±20.61	29.4
	T	1.59	1.07	1.38	0.86	0.89	0.68	0.25	0.0	0.17	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Morphine (0.5 mg%)		3.9±1.23	2.5±0.73	65.6±7.63	2.5±0.91	2.2±0.78	85.7±18.22	1.5±0.76	0.5 ±0.38	28.7±10.65	30
	T	0.31	1.02	1.23	0.75	1.17	0.87	0.0	0.0	0.2	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	

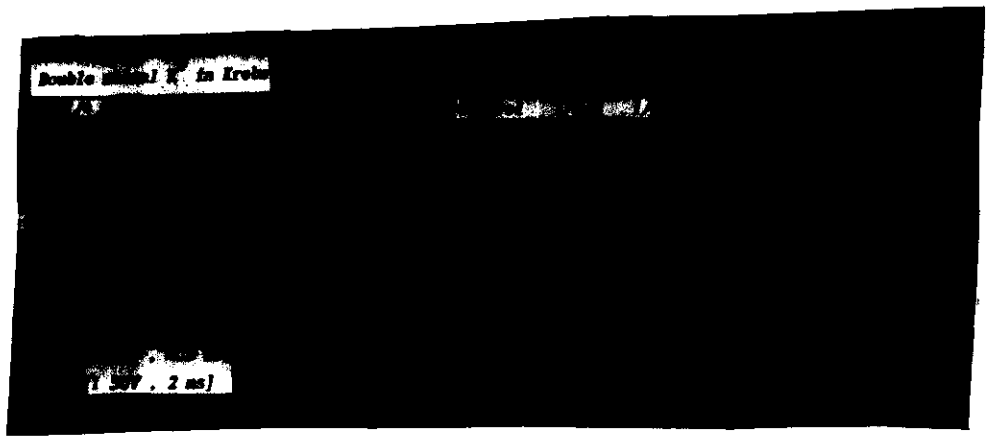

Fig.(41) :Control Experiment Carried out in Double Normal K^+ in Krebs Solution

Fig.(42) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal K^+ in Krebs Solution in Presence of Morphine (0.2mg%).

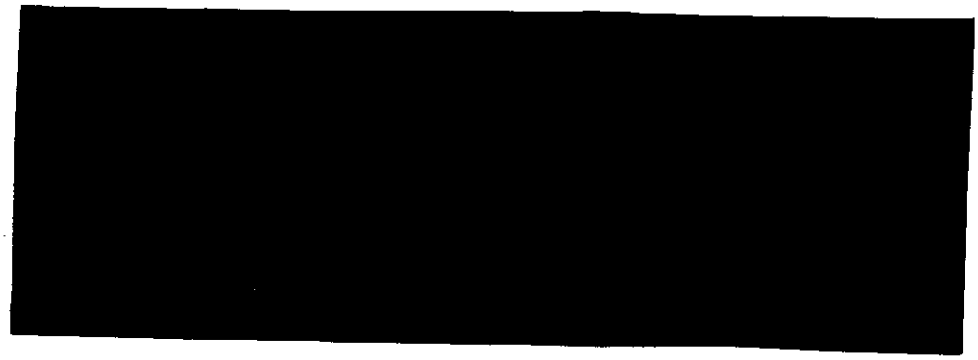

Fig.(43) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal K^+ in Krebs Solution in Presence of Morphine (0.5mg%).

The effect of naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing double the K^+ concentration in krebs solution.

The doses of naloxone investigated were 0.01mg% and 0.02mg%

Table (59) represents naloxone subgroup at a dose 0.01mg% . It included five experiments. It can be noted that the mean height of the direct contraction at the onset of the experiment was 3.3±0.54cm. The mean height of the indirect contraction at the onset of the experiment was 2.4±0.27cm. The I/D% was 73±6.78%. After quarter of an hour rest in a solution containing double the K^+ concentration in krebs solution and naloxone (0.01mg%), the mean values of the direct and indirect responses became 2.5±0.94cm and 2.3±0.88cm respectively. The I/D% was 91.0±13.13%. As seen, the I/D% was increased ($P<0.02$) compared with the corresponding value at the onset.

After repeated stimulation for 1/2h (at the end of experiment), the mean values of the direct, indirect and I/D% parameters were 1.5±0.41cm, 0.4±0.27cm and 24.3±16.31% respectively. As observed from this table there, was CNMB in one preparation after 10min of repeated stimulation.

Figure (44) represents one of the experiment done in this group.

Table (60) represents naloxone subgroup at a dose 0.02mg% . It included five experiments. It can be observed that, the mean height of the direct contraction at the onset of the experiment was 3.7±1.62cm. The mean height of

the indirect contraction at the onset of the experiment was 2.5±1.2 cm. The I/D% was 68.6±7.99%.

After quarter of an hour rest in presence of double the potassium concentration of krebs solution and naloxone (0.02mg%), the mean values of the direct and indirect responses became 2.6±0.98cm and 2.5±1.72cm respectively. The I/D% was 90.5±30.36%.

After repeated stimulation for 1/2h. (at the end of experiment) the mean values of the direct, indirect and I/D% parameters were 1.5±0.49cm, 0.6±0.45cm and 36.9±18.01% respectively.

Figure (45) represents one of the experiment done in this group.

Table (61) compares the mean values of the direct, indirect and I/D% parameters in the control group carried out in presence of double the potassium concentration of normal krebs solution and the corresponding values in naloxone subgroups.

As observed, there was no significant difference in the direct, indirect and I/D% parameters between naloxone subgroups and control at the onset of experiment, after 15 min. rest and at the end of the experiment i.e, naloxone (0.01mg% & 0.02mg%) had no observed effect on the response of the preparation bathed in a solution containing double the potassium concentration of normal krebs solution

Table (59) : The response of rat phrenic diaphragm preparation bathed in double the normal K^+ in krebs to repeated stimulation at a rate 1/sec in presence of naloxone (0.01 mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time min.
	direct	indirect		direct	indirect		direct	indirect		
1	2.6	2.0	76.9	1.4	1.0	71.4	1.4	0.0	0.0	10
2	2.9	2.4	82.8	2.2	2.2	100.0	1.5	0.5	33.3	30
3	4.0	2.7	67.5	2.6	2.7	103.8	1.1	0.2	18.2	30
4	3.5	2.5	71.4	4.0	3.4	85.0	2.2	0.6	27.3	30
5	3.3	2.2	66.7	2.5	2.4	96.00	1.4	0.6	42.9	30
Mean	3.3	2.4	73.0	2.5	2.3	91.0	1.5	0.4	24.3	
S.D.	0.543	0.274	6.778	0.943	0.878	13.13	0.409	0.269	16.31	
S.E.	0.243	0.123	3.031	0.422	0.392	5.874	0.183	0.120	7.293	
T						3.26				
P<						0.02*				

* Significant increase compared with the corresponding value at the onset.

Table (60) : The response of rat phrenic diaphragm preparation bathed in double the normal K^+ in krebs to repeated stimulation at a rate 1/sec in presence of naloxone (0.02mg%)

Exp. No.	Height of Contraction at onset		I/D%	Height of Contraction after 1/4h.		I/D%	Height of Contraction at end		I/D%	Time
	direct	indirect		direct	indirect		direct	indirect		
1	3.6	2.4	66.7	2.2	2.2	100.0	1.6	0.7	43.8	
2	2.8	1.6	57.1	3.3	2.0	60.6	1.2	0.5	41.7	1/2h
3	2.9	2.3	79.3	2.1	1.8	85.7	1.4	0.4	28.6	
4	6.5	4.6	70.8	4.0	5.5	137.5	2.2	1.3	59.1	
5	2.6	1.8	69.2	1.6	1.1	68.8	0.9	0.1	11.1	
Mean	3.7	2.5	68.6	2.6	2.5	90.5	1.5	0.6	36.9	
S.D.	1.621	1.20	7.993	0.982	1.717	30.36	0.489	0.447	18.01	
S.E.	0.724	0.537	3.575	0.439	0.768	13.58	0.219	0.20	8.056	

Table (61): Comparison between the mean values ± S.D. of the direct, indirect and I/D% parameters in the control group carried out in double normal potassium in krebs solution and the corresponding values in naloxone subgroups

Group		At Onset			After 15 Min. Rest			At End of Exp.			Time Min.
		Direct	Indirect	I/D%	Direct	Indirect	I/D%	Direct	Indirect	I/D%	
Control		4.1±0.88	2.9±0.58	70.1±4.38	2.8±0.35	2.7±0.65	99.0±29.89	1.5±0.35	0.5 ±0.38	31.1±24.87	29.2
Naloxone (0.01mg%)		3.3±0.54	2.4±0.27	73.0±6.78	2.5±0.94	2.3±0.88	91.0±13.13	1.5±0.41	0.4 ±0.27	24.3±16.31	26
	T	1.76	1.77	0.86	0.73	0.87	0.55	0.0	0.5	0.52	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	
Naloxone (0.02mg%)		3.7±1.62	2.5±1.2	68.6±7.99	2.6±0.98	2.5±1.72	90.5±30.36	1.5±0.49	0.6 ±0.45	36.9±18.01	30
	T	0.52	0.73	0.4	0.47	0.27	0.47	0.0	0.4	0.43	
	P	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	

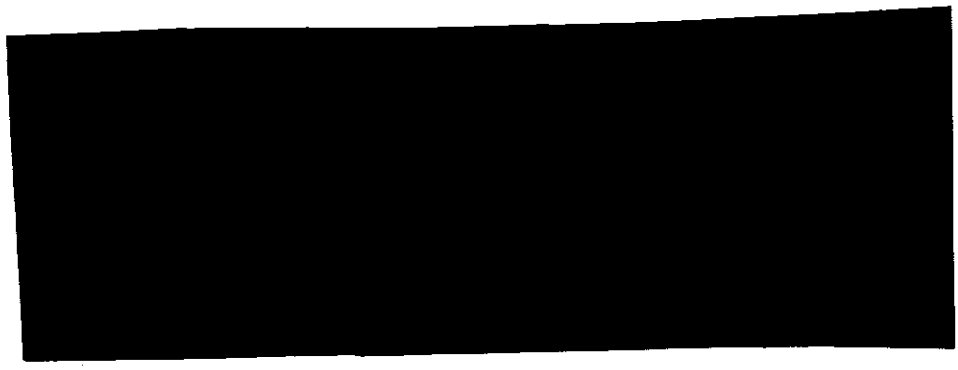

Fig.(44) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal K^+ in Krebs Solution in Presence of Naloxone (0.01mg%).

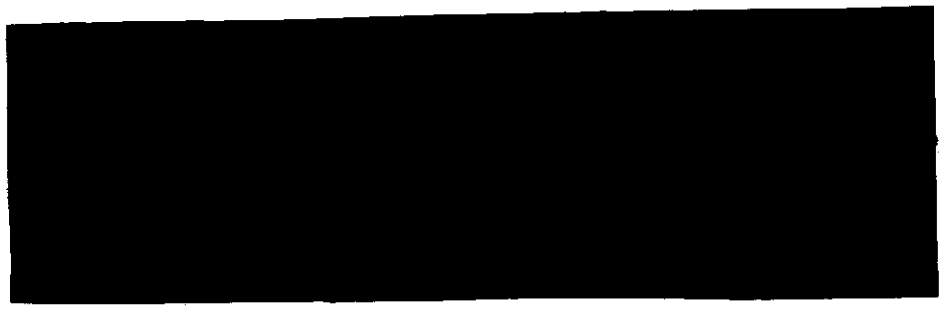

Fig.(45) : The Response of Rat Phrenic Diaphragm Preparation Bathed in Double Normal K^+ in Krebs Solution in Presence of Naloxone (0.02mg%).

GROUP II(B)-4 :

The effects of stimulation of opiate receptors by morphine and their blockage by naloxone on the response of the isolated rat phrenic diaphragm preparation bathed in a solution containing 4 times the normal K^+ concentration of krebs solution.

Control Experiment (n = 5) :

The mean height of the direct and indirect at the onset of experiment were 5.3 and 3.6cm respectivly. There was no indirect response at all after 15 min. rest bathing in 4 times normal K^+ in krebs and the direct contraction was very small (0.1cm) in three experiments. However, there was small indirect response (0.3cm) after 15 min. rest, in one preparation, continued for 1/2h. by repeated stimulation but with decline. The height if indirect contraction at the end of the experiment was 0.1cm. The height of direct contraction were 0.3cm and 0.1cm after 15 min. rest and at the end of this experiment respectively. In the last experiment the indirect response was 0.2cm and declined gradually till complete block after 20 min. repeated stimulation. The direct contraction was 0.3cm after 15 min. rest and at the end of this experiment.

Fig.(46) represents one of the experiment done in this group.

The effect of morphine.

The dose of morphine used was 0.5mg%. Five separate preparations were examined. The results were nearly similar as in the control group i.e., morphine had no effect on the preparations bathed with K^+ 4 times krebs solution.

Fig.(47) represents one of the experiment done in this group.

The effect of naloxone.

The dose of naloxone used was 0.02mg%. Five preparations were examined. There was slight recovery of the indirect response after 15 min rest compared with the control. The indirect contractions maintained with decline after 30 min. repeated stimulation in three experiments. One experiment only had no indirect response after 15 min. rest and the last one blocked after 3 min. repeated stimulation.

Fig (48) represents one of the experiment done in this group.

GROUP II(C) :

It was carried out on few preparations bathed with double normal Na^+ in krebs solution and 4 times normal Na^+ in krebs. In each instance two control experiments were done. The preparations bathed in 4 times normal Na^+ in krebs gave no response at all after 15 min. rest, while the preparation bathed in double normal Na^+ in krebs solution gave only very small direct contraction

(0.2cm) after 5 min. rest. Morphine was used at the dose 0.5mg% in one preparation bathed with double normal Na^+ in krebs, the response was the same as in control.. Naloxone was used at the dose 0.02mg% in one preparation bathed with double normal Na^+ in krebs solution and in another preparation bathed in 4 times normal Na^+ in krebs solution, the response was the same as in control.

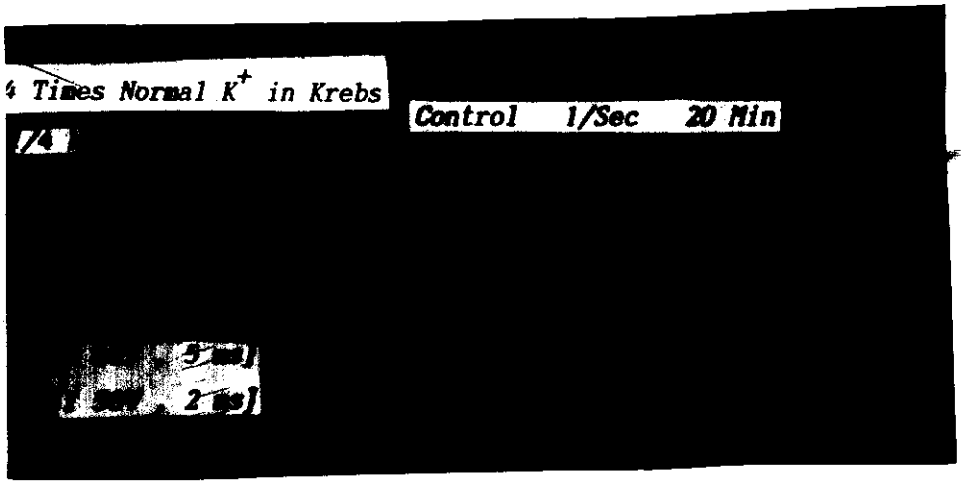

Fig.(46) :Control Experiment Carried out in 4 Times Normal K^+ in Solution.

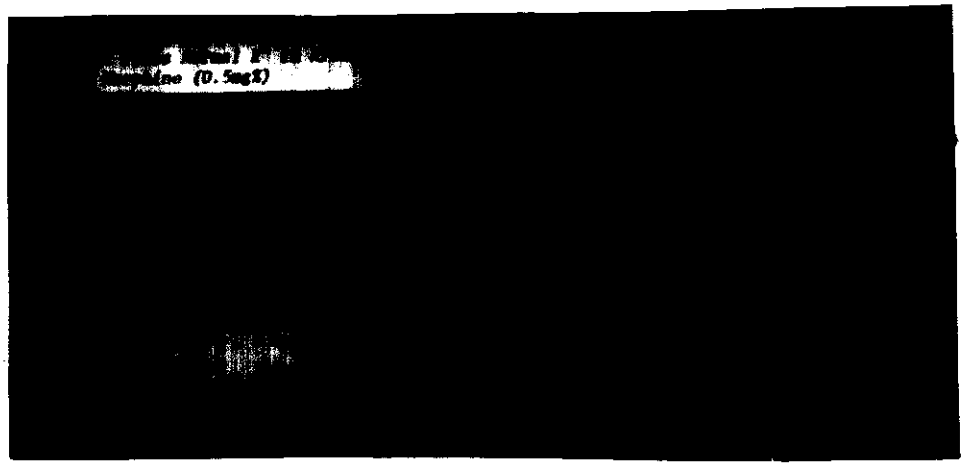

Fig.(47) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal K^+ in Krebs Solution in Presence of Morphine (0.5mg%).

Fig.(48) : The Response of Rat Phrenic Diaphragm Preparation Bathed in 4 Times Normal K^+ in Krebs Solution in Presence of Naloxone (0.02mg%).