RESULTS

This study was carried out from November 2005 to May 2006 on 50 non pregnant women of child bearing age (15 to 49) years, suffering from vaginal discharge, irritation or soreness, cervicitis and cervical erosion. Cases were selected from the out patient clinics of Gynaecology and Obstetrics Department, Benha University Hospital.

Endocervical swabs were collected from each women for detection of the presence of *C.trachomatis* by Giemsa and DFA staining methods.

Results of Giemsa stained smears :(Table3 &Figure2):

All specimens taken were examined for the presence of characteristic intracytoplasmic masses of *Chlamydial* infection which either appear blue- mauve to dark purple, depending on the stage of development.

Eight women (16%) were positive for the presence of *Chlamydia* in their specimens, while 42 women (84%) were negative.

Examination of Giemsa stained smears revealed the presence of inflammatory reaction in many positive cases manifested by the presence of polymorph nuclear cells.
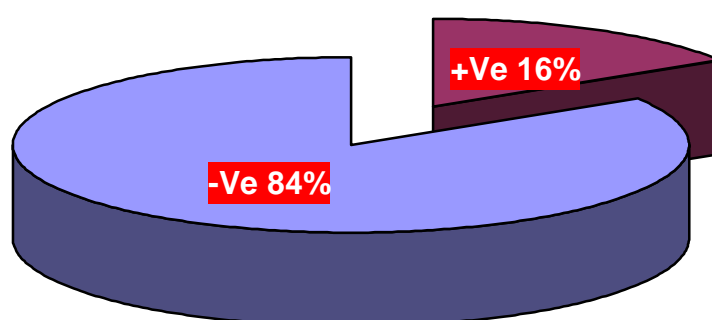
Table (3): Distribution of *C. trachomatis* infection in the studied samples according to Giemsa staining results.

Total No. of sample	+Ve samples for Giemsa		-Ve sample for Giemsa	
	No.	%	No.	%
50	8	16	42	84

Z =6.56   P< 0.001

Figure (2): Distribution of *C. trachomatis* infection in the studied samples according to Giemsa staining results.

Results of direct fluorescent antibody (DFA) staining technique (Table 4 &Figure 3):

All specimens taken were examined for the presence of apple green fluorescence contrasted by reddish brown background of the counter stain.

There were 15 women (30%) positive for the presence of *Chlamydia* in their specimens, while 35 women (70%) were negative.

Table (4): Distribution of *C. trachomatis* infection in the studied samples according to DFA test results.

Total No. of sample	+Ve sample for DFA		-Ve sample for DFA	
	No.	%	No.	%
50	15	30	35	70

Z =3.09   **P< 0.01**

Figure (3): Distribution of *C. trachomatis* infection in the studied samples according to DFA test results.
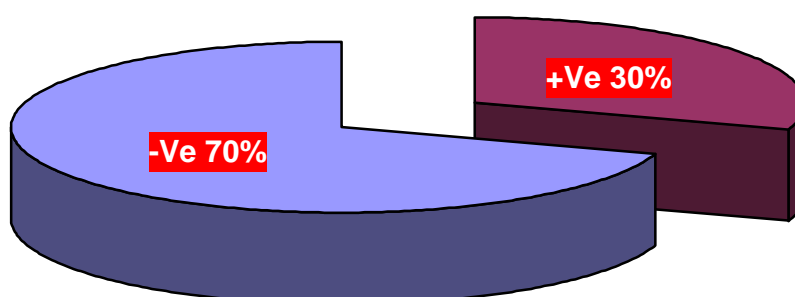
Table (5): Comparison between DFA test and Giemsa staining results according to diagnosis of *chlamydial* infection:

Test		DFA test		Total No. of tested samples
		+Ve	-Ve	
Giemsa stain	+Ve	8	0	8
	-Ve	7	35	42
Total		15	35	50

Sensitivity= 53.3%

Specificity= 100%

C.trachomatis infection according to the age :(Table6& Figure4).

Out of 50 examined cases there were 21 women(42%) aged from 15 to 25 years, 15 (30%) women were aged from 26 to35 years, and 14 (28%) women were aged from 36 to 49 years.

Out of 21 women aged from 15 to 25 years, 4 (19.04) were positive for Chlamydia by Giemsa stain, 9 (42.85%) were positive by DFA test.

Out of 15 women aged from 26 to 35 years, 4 (26.66%) were positive for Chlamydia by Giemsa stain, 5 (33.33%) were positive by DFA test.

Out of 14 women aged from 36 to 49 years, no women were positive forChlamydia by Giemsa stain, 1 (7.4%) was positive by DFA.

Table (6): *C.trachomatis* infection in different age groups of the examined women

Age	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%		
15-25(A)	21	42	4	19.04	9	42.85	0.996	>0.05
26-35(B)	15	30	4	26.66	5	33.33		
36-49(C)	14	28	0	0	1	7.14		
Total	50	100	8		15			

Z_1 = A vs. B= 0.54 P_1 >0.05	Z_1=A vs. B =0.58 P_1>0.05
Z_2 = A vs. C =1.74 P_2 <0.05	Z_2=A vs. C =2.29 P_2<0.05
Z_3 = B vs. C=2.1 P_3 <0.05	Z_3=B vs. C =1.74 P_3<0.05

There is no significant difference between age groups as regard results of Giemsa stain and DFA test, but the table showed that, young women (15 to 35) had higher frequency of occurrence of *C.trachomatis* than older women.

Figure (4): *C.trachomatis* infection in different age groups of the examined women:
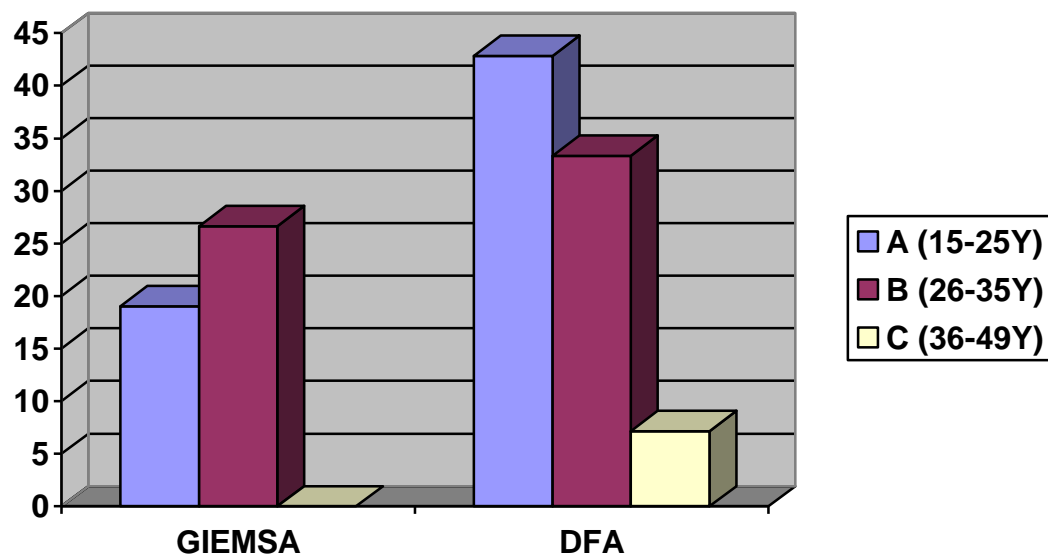
C.trachomatis infection according to the residence :(Table 7 &Figure5):

Out of 50 examined women there were 30 (60%) women from rural. And 20 (40%) women were from urban area.

Out of 30 rural women, 7(23.33%) were Chlamydia positive by Giemsa stain, 12 (40%) were positive by DFA test.

Out of 20 urban women, 1(5%) were Chlamydia positive by Giemsa stain, 3 (15%) were positive by DFA test.
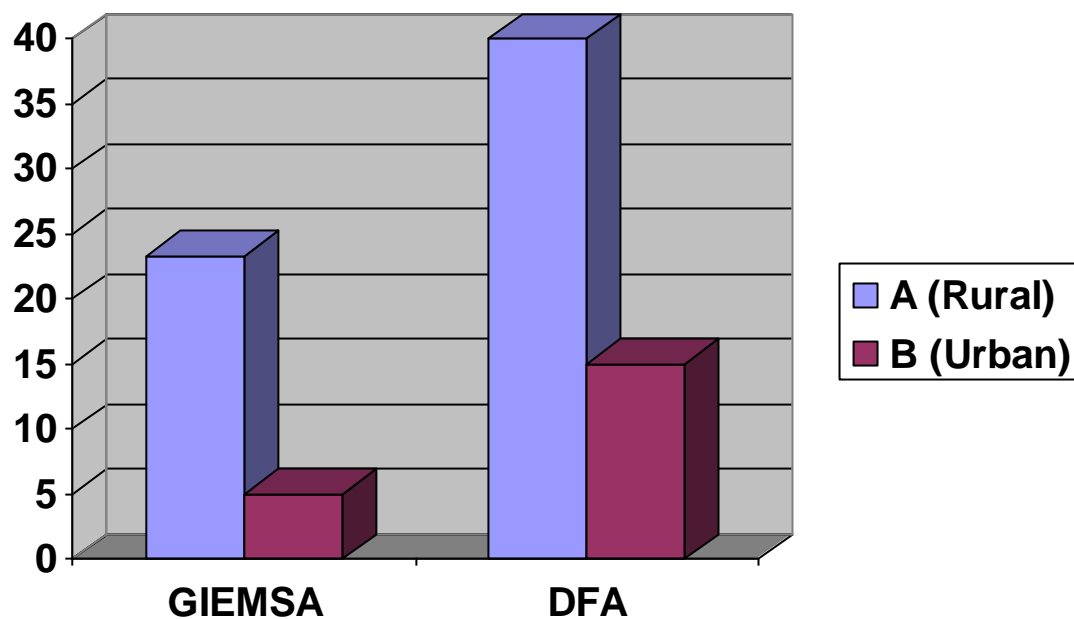
Table (7): *C.*trachomatis infection according to the residence of the examined women:

Residence	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%		
Rural (A)	30	60	7	23.33	12	40	0.16	>0.05
Urban(B)	20	40	1	5	3	15		
Total	50	100	8		15			

Z_1=A vs. B =1.73 P_1<0.05	Z_2=A vs. B=1.89 P_2<0.05

There is no significant difference between rural and urban residence according to results of Giemsa stain and DFA, but the table and figure showed that, rural women had higher frequency of occurrence of *C.trachomatis* than urban women.

Figure (5):): *C.*trachomatis infection according to the residence of the examined women :

C.trachomatis **infection according to previous abortion(s):**

(Table8 &Figure6).

Out of 50 examined women there were 35women (70%) have no history of previous abortion, 10 (20%) were aborted once, and 5 (10%) were aborted more than one time.

Out of 35 women. who had no previous history of abortion, 3 (8.57%) were Chlamydia positive by Giemsa stain, 7 (20%) were positive by DFA test.

Out of 10 women who aborted once, 3 (30.7%) were *Chlamydia* positive by Giemsa stain, 5 (50%) were positive by DFA test.

Out of 5 women, who aborted more than one time, 2 (40%) were positive by Giemsa stain, 3 (60%) were positive by DFA test.
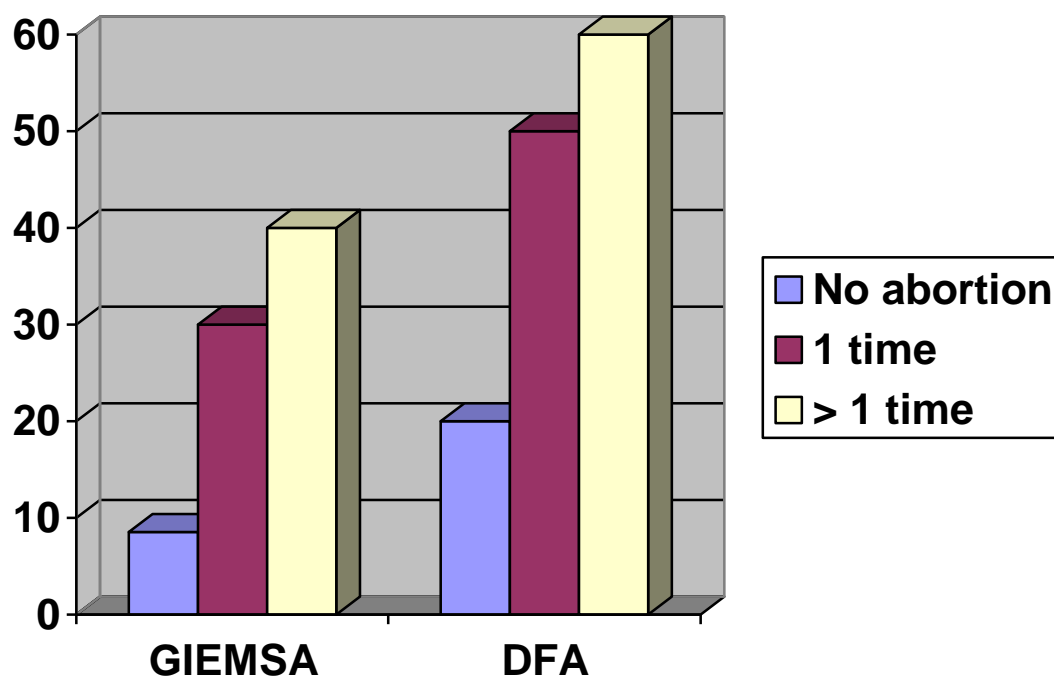
Table (8):*C.trachomatis* infection according to previous abortion (s):

Previous abortion	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%		
No abortion(A)	35	70	3	8.57	7	20	0.187	>0.05
One time(B)	10	20	3	30	5	50		
More than one time(C)	5	10	2	40	3	60		
Total	50	100	8		15			

Z_1=A vs. B =1.75	P_1<0.05	Z_1=A vs. B = 1.89	P_1<0.05
Z_2=A vs. C = 1.98	P_2<0.05	Z_2=A vs. C = 1.93	P_2<0.05
Z_3=B vs. C =0.38	P_3>0.05	Z_3=B vs. C = 0.36	P_3>0.05

No significant difference between aborted and non aborted women according to results of Giemsa stain and DFA, but aborted women (B and C) had higher frequency of occurrence of *C.trachomatis* infection than non aborted women (A)

Figure (6): *C.trachomatis* **infection according to previous abortion (s):**

C.trachomatis **infection according to the used method of contraception:** (Table 9).

Out of 50 examined there were 9 women (18%) not used any contraceptive method, 11 (22%) were using hormonal contraceptive, 30 (60%) were using IUD.

Out of 9 women, who not used any contraceptive method, no women were *Chlamydia* positive by Giemsa stain, 1 (11.11%) were positive by DFA test.

Out of 11 women, using hormonal contraceptive, 4 (36.36%) were *Chlamydia* positive by Giemsa stain, 6 (54.54%) were positive by DFA.

Out of 30 women, who used IUD, 4 (13.33%) were *Chlamydia* positive by Giemsa stain, 8 (26.66%) were positive by DFA test.

Table (9) *C.trachomatis* **infection according to the used method of contraception :**

Method of contraception	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%		
No contraception(A)	9	18	0	0	1	11.11	0.664	>0.05
Hormonal(B)	11	22	4	36.36	6	54.54		
IUD(C)	30	60	4	13.33	8	26.66		
Total	50	100	8		15			

Z_1=A vs. B=2.02 P_1<0.05	Z_1=A vs. B =2.02 P_1<0.05
Z_2=A vs. C=1.2 P_2>0.05	Z_2=A vs. C = 0.97 P_2>0.05
Z_3=B vs. C=1.7 P_3<0.05	Z_3=B vs. C = 1.7 P_3<0.05

There is no significant difference between using or non using contraceptive method according to results of Giemsa stain and DFA, but, women who used hormonal contraceptive method had higher frequency of occurrence of *C.trachomatis* than other women.

C.trachomatis infection according to the week of menstrual cycle at time of examination :(Table10).

Out of 50 examined women there were 24 (48%) women, who were at the 1st week of menstrual cycle, 10 (20%) were at 2nd week, 8 (16%) were at the 3rd week. And 8 (16%) were at the 4th week.

Out of 24 women, who were at the 1st week of menstrual cycle, 3 (12.5%) were Chlamydia positive by Giemsa stain, 7 (29.1%) were positive by DFA test.

Out of 10 women, who were at 2nd week of menstrual cycle,2 (20%) were Chlamydia positive by Giemsa stain, 3 (30%) were positive by DFA test.

Out of 8 women, who were at 3rd week of menstrual cycle,3 (37.5%) were Chlamydia positive by Giemsa stain ,4 (50%) were positive by DFA test.

Out of 8 women, who were at 4th week of menstrual cycle, no positive cases were Chlamydia positive by Giemsa stain, 1 (12.5%) were positive by DFA test.

Table (10): *C.trachomatis* **infection according to week of menstrual cycle:**

Week of menstrual cycle	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%	0.895	>0.05
1st week(A)	24	48	3	12.5	7	29.1		
2nd week(B)	10	20	2	20	3	30		
3rd week(C)	8	16	3	37.5	4	50		
4th week(D)	8	16	0	0	1	12.5		
Total	50	100	8		15			

Z_1=A vs. B =0.04 P_1>0.05

Z_2=A vs. C =1.07 P_2>0.05

Z_3=A vs. D =0.94 P_3>0.05

Z_4=B vs. C =0.86 P_4>0.05

Z_5=B vs. D =0.88 P_5>0.05

Z_6=C vs. D = 1.60 P_6>0.05

No significant difference between the weeks of the menstrual cycle according to results of Giemsa stain and DFA, but higher frequency of occurrence of *C.trachomatis* in 1st, 2nd, and 3rd weeks.

C.trachomatis infection as regards to the fertility state:

(Table11 &Figure7).

Out of 50 examined women there were 14 women (30%) were 1ry infertile, 4 (6%) were 2nd infertile, and 32 (64%) were fertile.

Out of 14 women, who were 1ry infertile, 3 (21.4%) were *Chlamydia* positive by Giemsa stain, 7 (50%) were positive by DFA.

Out of 4 women, who were 2nd infertile, 2 (50%) were *Chlamydia* positive by Giemsa stain, 3 (75%) were positive by DFA.

Out of 32 women, who were fertile, 3 (9.3%) were *Chlamydia* positive by Giemsa stain, 5 (15.62%) were positive by DFA.

C.trachomatis infection as regards to the fertility state:

Table (11): *C.trachomatis* **infection according to the fertility state of the examined women:**

Fertility state	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%		
1ry infertility(A)	14	30	3	21.42	7	50	0.538	>0.05
2ry infertility(B)	4	6	2	50	3	75		
fertile(C)	32	64	3	9.37	5	15.62		
Total	50	100	8		15			

Z_1=A vs. B=1.12 P_1>0.05

Z_2=A vs. C=1.11 P_2<0.05

Z_3=B vs. C=2.21 P_3>0.05

Z_1=A vs. B =0.88 P_1>0.05

Z_2=A vs. C = 2.44 P_2<0.05

Z_3=B vs. C = 2.69 P_3<0.05

No significant difference between fertile and infertile according to results of Giemsa stain and DFA test. But, infertile women (A and B) had higher frequency of occurrence of *C.trachomatis* than fertile women (C)

Figure (7): *C.trachomatis* **infection according to the fertility state of the examined women:**
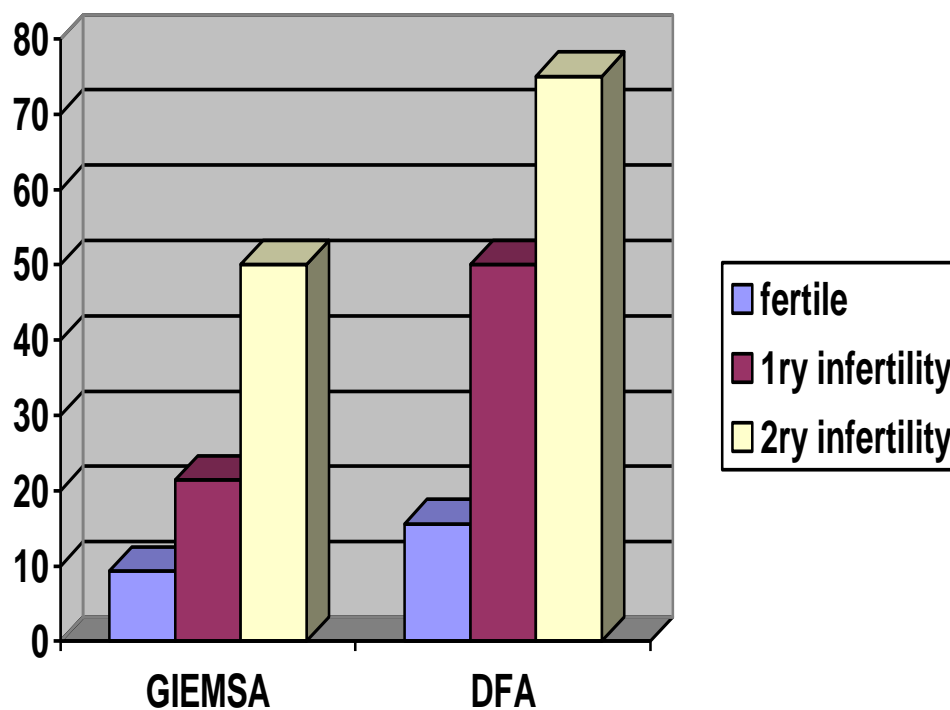
C.trachomatis infection according to symptoms and signs:

(Table12).

Out of 50 women, 19 (38%) were suffering from vaginal discharge only, 9 (18%) were suffering from irritation and soreness, 7 (14%) were had cervicitis and 15 (30%) were had cervical erosion.

Out of 19 women, who suffering from vaginal discharge only, 1(5.26%) was Chlamydia positive by Giemsa stain, 3 (15.78%) were positive by DFA test.

Out of 9 women, who suffering from irritation and soreness, no positive cases were Chlamydia positive by Giemsa stain,1 (11.11%) were positive by DFA.

Out of 7 women, who had cervicitis, 3 (42.85%) were Chlamydia positive by Giemsa stain, 4 (57.14%) were positive by DFA test.

Out of 15 women, who had cervical erosion.4 (26.66%) were Chlamydia positive by Giemsa stain, 7 (46.66%) were positive by DFA test.

Table (12): *C.trachomatis* **infection according to symptoms and signs of the examined women**

Symptoms & signs	No.	%	Giemsa		DFA		X^2	P
			No.	%	No.	%	0.915	>0.05
Discharge(A)	19	38	1	5.26	3	15.78		
Irritation & soreness(B)	9	18	0	0	1	11.11		
Cervicitis(C)	7	14	3	42.85	4	57.14		
Cervical erosion (D)	15	30	4	26.66	7	46.66		
Total	50	100	8		15			

Z_1=A vs. B = 0.33 P_1>0.05

Z_2=A vs. C = 2.11 P_2<0.05

Z_3=A vs. D =1.96 P_3<0.05

Z_4=B vs. C = 1.97 P_4<0.05

Z_5=B vs. D = 1.78 P_5<0.05

Z_6=C vs. D = 0.45 P_6>0.05

No significant difference between symptoms and signs according to results of Giemsa stain and DFA test, But higher frequency of occurrence of *C.trachomatis* in women had abnormal cervix (C and D).

Figure (8): Detection of inclusions in smears stained by Giemsa. Magnification (1000 x)

Figure (9): Detection of inclusion bodies in smears using fluorescein-labelled monoclonal antibodies specific to *C.trachomatis* (600x-1000x)