RESULTS

This work was carried out during the period from **December 1991** to **November 1992** on stool samples which were obtained from two groups of patients

Group I : 350 cases of diarrhea attending the Out- Patient Clinic and Rehydration center of Benha and Zagazig University Hospitals. Their ages ranged from one to 24 months. They were 220 moles and 130 females.

Group II : 54 normal apparently healthy infants Their ages were comparable to those of group I. They were 36 males and 18 females. Potavirus antigen was searched for the stool samples of both groups by the following methods :

1. Latex agglutination method.
2. Enzyme- Linked Immunosorbent Assay (ELISA).
3. Tissue culture and Indirect Immunofluorescent Technique.

Table (1) showed the number of positive cases among stools of cases and control subjects using latex agglutination test using specific monoclonal antibodies against rotavirus. It was obvious that 77 out of 350 diarrheal cases (22%) and 4 out of 54 control subjects (7.4%) had positively rotavirus antigen in their stools which were tested by specific monoclonal antibodies against rotavirus. Z test for cases and control group was 3.48 and the P value was highly significant ($P < 0.001$).

Table (2) showed bacterial association in positive cases for rotavirus antigen using latex agglutination test. The table showed that 56 out of 77 positive

cases (72.7%) were only positive for rotavirus. E. coli associated with ratavirus were obtained in 14 (18.2%), shigella associated with rotavirus in 4(5.2%), and E. coli and shigella associated with rotavirus in 3(3.9%).

Table (3) showed the number of positive cases among studied stools of cases and control subjects using ELISA technique. It was shown that 85 out of 350 diarrheal cases (24.3%), and 5 out of 54 control subjects (9.3%) were positive for rotavirus antigen in their stools. Z test for positive cases and control group was 3.28 and P value was very highly significant ($P < 0.001$).

Table (4) showed the number of positive cases to rotavirus among studied stools of cases and control group using indirect immunofluorescent technique after passage on MA-104 tissue culture. It was shown that 89 out of 350 cases (25.4%) and 7 out 54 control group (12.9%) were positive to rotavirus antigen. Z test for positive cases and control groups was 2.44 and P value was highly significant ($P < 0.01$).

Table (5) showed the results of indirect immunofluorescent versus ELISA and latex agglutination tests for detection of rotavirus antigen in stools of cases and control groups. The high percentage of positive cases was among the results of indirect immunofluorescent (25.4 %) followed by ELISA (24.3 %) and latex agglutination test (22 %). Z test between indirect immunofluorescent and ELISA methods was 0.34 and P value was not significant ($P > 0.05$). Z test between indirect immunofluorescent and latex agglutination methods was 1.06 and P value was not significant ($P>0.05$). Z test beteen ELISA and latex agglutination methods was 0.72 and P value was not significant ($P > 0.05$).

Table (6) showed the sensitivity, specificity and predictive values of ELISA and latex agglutination methods versus indirect immunofluorescent technique. The sensitivity and specificity of latex agglutination method (88.31 % and 96.55% respectively) and ELISA method (87.64 % and97.32% respectively) were nearly the same. The predictive value of positive of ELISA was very high (91.77 %) as compared with that of latex agglutination test (76.41 %). The predictive value of negative of ELISA (95.85 %) and latex agglutination test (96.55%) were nearly the same.

Table (7) showed the subgroup determination of rotavirus using indirect immunofluorescent technique. Subgroup I was detected in 37 out of 89 positive cases (41.6 %), while subgroup II was detecteds in 52 out of 89 positive cases (58.4 %). Subgroup I was detected in 2 out of 7 positives in control group (28.6 %) while subgroup II was detected in 5 out of 7 positive control group (71.4 %). Z test for subgroup I between cases and control groups was 0.39 and P value was not significant ($P > 0.05$) and Z test for subgroup II between cases and control group was 0.73 and P value was not significant ($P > 0.05$). Z test between subgroup II and subgroup I was 2.27 and P value was significant ($P < 0.05$).

Table (8) showed the relation between rotavirus infection and the type of feeding. The highest incidence of rotavirus positive antigen in cases was detected among artificially fed group (35 %) which was more than that of breast fed group (16.8 %). In control group, the highest incidence was detected also in artificially fed group (23 %) more than that of breast fed group (5.9 %). The weaned groups showed a moderate incidence between breast fed and artificially fed infants in both cases (24.4 %) and control group (12.5 %). Z test between breast and artificially fed groups was 4.07 and P value was highly significant ($P < 0.001$). Z test between

breast fed and weaned infants was 1.45 and P value was not significant (P > 0.05). Z test between artificially fed and weaned infants was 1.79 and P value was not significant (P > 0.05). Also in table (8) Z test between cases and control groups of breast fed infants which was 1.62 and P value was not significant (P >0.05), Z test between cases and control groups of artificially fed infants was 0.96 and P value was not significant (P > 0.05), and Z test between cases and control groups of weaned infants was 1.53 and P value was not significant (P > 0.05).

Table (9) showed the distribution of rotavirus infection of diarrheal stools of cases and control subjects according to age. There were divided into 3 age groups : group (1) (< 6 months), group (2) (7-12 months) and group (3) (13- 24 months). Z test between cases and control subjects in group (1) was 1.97, and P value was significant (P < 0.05). Z test between cases and control subjects of group (2) was 2.49, and P value was highly significant (P <0.01). Z test between cases and control subjects of group (3) was 0.54 and P value was not significant (P > 0.05). On the other hand, Z test between cases of group (1) and group (2) was 1.01, and P value was not significant (P > 0.05). Z test between cases of group (1) and group (3) was 3.36, and P value was very high significant (P < 0.001). Z test between cases of group (2) and group (3) was 3.52 and P value was very high significant (P < 0.001).

Table (10) showed the distribution of rotavirus infection in stools of cases and control subjects according to sex. Z test between male cases and control group was 2.24 and P < 0.05 (significant). Z test between female cases and control group was 1.06 and P > 0.05 (not significant). Z test between male and female cases was 0.49 and P > 0.05 (not significant). Z test between male and female controls was 0.55 and P > 0.05 (not significant).

Table (11) showed the seasonal changes in the prevalence of rotavirus infection associated with diarrheal gastroenteritis. The table showed that the highest level of rotavirus infection was detected in winter (December, January and February) 45 cases with an incidence of (50.6 %), while the level in summer was 17 cases with an incidence of (19.1 %). Z test between winter and summer seasons was 4.9, and P value was very high significant ($P < 0.001$).

Table (12) showed the distribution of rotavirus infection according to residence. The prevalence of rotavirus infection was more among cases in rural areas 57 with an incidence of (27.8 %), while in urban areas 32 with an incidence of (22 %). Z test between rural and urban areas was 0.26, and P value was not significant ($P > 0.05$). Also, Z test between cases and control subjects in urban areas was 1.31, and P value was not significant ($P > 0.05$), and Z test between cases and control subjects in rural areas was 3.19, and P value was very high significant ($P < 0.001$).

Table (13) showed the relation of rotavirus infection and the degree of dehydration. The majority of cases with rotavirus infection 72 (80.9 %) had no dehydration, 6 (6.7 %) of cases suffered from mild dehydration, 8 (9 %) of cases suffered from moderate dehydration and 3 (3.4) of cases suffered from severe dehydration.

Table (1) : Latex agglutination method for detection of rotavirus antigen in stools of cases and control subuects

Cases			Control			Z Test	P value	Signifi-cance
Total No.	No. of +ve	%	Total No.	No. of +ve	%			
350	77	22	54	4	7.4	3, 48	<0.001	S.

No. of + ve = positive number

S. = significant

N.S. = non signifivcant

Table (2) : Bacterial association in positive cases of rotavirus antigen by latex agglutination test.

	Total + ve cases	
	number	%
Rotavirus infection alone	56	72.7
Rotavirus infection with E.coli	14	18.2
Rotavirus infection with Shigella	4	5.2
Rotavirus infection with E.coli and Shigella	3	3.99
total	77	100%

** Out of 350 diarrheal cases,the incidence of rotavirus infection alone was(16%),rotavirus with E.coli was(4%),rotavirus with shigella was(1.1%),and mixed infection between rotavirus,E.coli and shigella was(0.85%).

Table (3) : ELISA technique for detection of rotavirus antigen in stools of cases and control subjects.

Cases			Control			Z Test	P value	Signifi-cance
Total No.	No. of +ve	%	Total No.	No. of +ve	%			
350	85	24.3	54	5	9.3	3.28	< 0.001	S.

Table (4) : Indirect immunofluorescent technique after passage on MA- 104 tissue culture for detection of rotavirus antigen in stools of cases and control subjects.

Cases			Control			Z Test	P value	Signifi-cance
Total No.	No. of +ve	%	Total No.	No. of +ve	%			
350	89	25.4	54	7	12.9	2.44	< 0.01	S.

Table (5): Indirect immunofluorescent method versus ELISA and latex agglutination test for detection of rotavirus antigen in stools of diarrheal cases and control subjects.

	Cases			Control			Z test	P value	Signif-icance
	Total No.	No. of+ve	%	Total No.	No. of+ve	%			
Latex agglutination test	350	77	22	54	1	1.9	3.48	<0.001	S.
ELISA methed	350	85	24.3	54	5	9.3	3.28	<0.001	S.
Indirect immunofluorescent technique	350	89	25.4	54	7	12.9	2.44	<0.01	S.

** Z test between indirect immunofluorescet and ELISA methods=0.34 and P > 0.05(not significant).

** Z test between indiect immunofluorescent and latex agglutination =1.06 and P > 0.05(not significant).

** Z test between ELISA and latex agglutination tests=0.72 and P > 0.05(not significant).

Table (6) : Senstivity, specificity and predictive values of ELISA and latex agglutination tests versus indirect immunofluorescent technique.

	Sensitivity	Specificity	Predictive value	
			+ ve	-ve
ELISA method	87.64%	97.32%	91.77 %	95.85%
Latex agglutination method	88.31%	92.30%	76.41 %	96.55%

Table (7) : Subgroup determination of rotavirus infection by using indirect immunofluraecent technique.

	Cases		Control		Z test	P value	Signif-icance
	+ve No.	%	+ve No	%			
Subgrop I	37	41.6	2	28.6	0.39	> 0.05	N.S.
Subgrop II	52	58.4	5	71.4	0.73	> 0.05	N.S.
Total	89	100	7	100			

** Z test between subgroup(2) and subgroup(1) of cases=2.27
and P < 0.05(significant).

Table (8) : Relation between rotavirus infection and the type of feeding using indirect immunofluorescent technique.

Type of feeding	Cases			Control			Z test	P value	Signif-icance
	Total No	+ve No	%	Total No	+ve No	%			
Breast fed (1)	113	19	16.8	17	1	5.9	1.62	> 0.05	N.S.
Artificial fed (2)	114	40	35	13	3	23.0	0.96	> 0.05	N.S.
weaned (3)	123	30	24.4	24	3	12.5	1.53	> 0.05	N.S.
Total	350	89	25.4	54	7	12.9			

** Z test between artificial feeding and breat feeding =4.07
and P < 0.001(significant).

** Z test between weaned and breast feeding = 1.45
and P > 0.05(not significant).

** Z test between artificial feeding and weaned infants=1.79
and P > 0.05(not significant).

Table (9) : Distribution of rotavirus infection in stools of cases and control subjects according to age using indirect immunofluorescent technique.

Age	Cases			Control			Z test	P	Signif-icance
	Total No	+ve No	%	Total No	+ve No	%			
< 6 months (group 1)	55	21	38.2	18	3	16.6	1.97	< 0.05	S.
7-12 months (group2)	160	49	30.6	25	3	12.0	2.49	< 0.01	S.
13-24 months (group 3)	135	19	14.0	11	1	9.0	0.54	> 0.05	N. S.
Total	350	89	25.4	54	7	12.9	2.44	< 0.01	S.

** Z test between cases of group(1) and group (2)=1.01
and P > 0.05 (not significant).

** Z test between cases of group(1) and group(3)=3.36
and P < 0.001(significant).

** Z test between cases of group(2) and group(3)=3.52
and P < 0.001(significant).

Table (10) : Distribution of rotavirus infection in stools of cases and control sujbects according to sex using immunofluorescent technique.

Type of Sex	Cases			Control			Z test	P value	Signifi-cance
	Total No	+ve No	%	Total No	+ve No	%			
males	220	54	24.5	36	4	11.1	2.24	< 0.05	S.
females	130	35	26.9	18	3	16.7	1.06	> 0.05	N.S.
Total	350	89	25.4	54	7	12.9	2.44	< 0.01	S.

** Z test between cases of males and females = 0.47
and P > 0.05(not significant).

** Z test between conrol groups of males and females = 0.55
and P > 0.05(not significant).

Table (11) : Seasonal changes in the prevalence of rotavirus infection during December 1991 through November 1992

Season	No. of Cases	No. of +ve	+ve R.V %
Winter (Dec. Feb)	95	45	50.6%
Spring (Mar. May)	61	12	13.5%
Summer (Jun-Aug)	105	17	19.1%
Autumn (Sept-Nov)	89	15	16.8%
Total No.	350	89	100%

** Z test between winter and summer seasons = 4.9

and $P < 0.001$ (significant).

Table (12) : Urban versus rural distribution of rotavirus infection in stools of cases and control subjects

	Cases			Control			Z test	P	Signif-icance
	Total No	+ve No	%	Total No	+ve No	%			
Urban	145	32	22	20	3	15	1.31	>0.05	N.S.
Rural	205	57	27.8	34	4	11.8	3.19	<0.001	S.
Total	350	89	25.4	54	7	12.9			

** Z test between cases of rural and urban areas = 0.26 and P > 0.05(not significant).

Table (13) : Rotavirus infection versus the degree of dehydration using indirect immunofluorescent technique

Degree of dehydration	Case	
	Number	%
No dehydration*	72	80.9
Mild dehydration	6	6.7
Moderate dehydration	8	9.00
Severe dehydration	3	3.4
Total No. of + ve	89	100%

* Corrected dehydration

Discussion