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BACTERIAL AND PARASITIC CAUSES ASSOCIATED WITH MORTALITIES IN YOUNG RABBITS AND THEIR SUBSEQUENT BIOCHEMICAL CHANGES

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ABSTRACT

The present study was carried out in order to investigate some of the bacterial and parasitic causes associated with mortalities in young rabbits till weaning and the subsequent biochemical changes. A total of 180 morbid and apparently healthy rabbits were selected from privately owned rabitraries of variable sizes at El Bohaira Province, Egypt, suffering from increase mortalities in young rabbits till weaning. It was found that 93.3% (168) of the examined rabbits were bacteriologically positive. More than one isolate could be detected in 78.3% and only 21.7% revealed uni-isolate. E coli, Enterobacter Spp., Citrobacter, Proteus, Klebsiella Spp., Pseudomonas aregenosa, Staph aureus, Staph. epedermidis, Staph. intermedium and Streptococci were isolated from the examined morbid cases. Parasitological investigations revealed that 55% (99) of the examined rabbits were positive for Eimeria and Cryptosporidium spp. Five species of Eimeria were recognized, they are: E. perforans, E. media, E. magna, E. irresidua and E. intestinalis. Hepatic coccidiosis could not be detected in our work. Cryptosporidium Spp. could be detected in 12 (6.7%) of cases. The biochemical changes accompanied with each bacterial and parasitic affections were estimated. The economic and public health importance of the problem has been discussed and the suggested measures for solving the problem were done.

INTRODUCTION

Rabbit production is a growing industry in Egypt, and proved an economical profit. The increased mortalities in newly born and young rabbits is a result of many different causes **(Okerman, 1987)**. During the recent years interest has been focused on diarrhea in rabbits since it is responsible for high economic losses. Broiler rabbits are extremely sensitive to digestive tract diseases which occur specially in newly born and newly weaned rabbits **(Blanco et al., 1996)**.

Weaning is stressful period in which Juvenile rabbits are susceptible to diseases So early weaning and separation from the dam increase susceptibility to bacterial infections **(Harcourt-Brown, 2002)**. Pathogenic strains of E. Coli , Clostridia Spp., Coccidia or Rotavirus are likely to be present in the environment of newly weaned rabbits. The intensively reared domestic rabbits are more susceptible to harbour infections **(Lelkes and Change, 1987)**.

The common causative agent of enteritis in domestic rabbits described by **Percy et al. (1993)** were Eimeria Spp., E. Coli, Colistredium Spp. Salmonella, Bacillus Piliforms and Rota virus. Eimeria spp. were often found in conjunction with other pathogenic agents as E. coli specially in young rabbits around weaning **(Harkness, 1997; Rodriguez-Delara et al., 2008)**.

Coccidiosis is among the most serious rabbit diseases as it causes high mortalities of young rabbits **(Soulsby, 1982)**. Rabbit coccidiosis occurs in two forms, intestinal form caused by different Eimeria species as E. exigua and E. perforance, the other form is the hepatic one and caused by E. stiedae **(Caudert et al., 2000; Al-Mathal, 2008)**. Identification of Eimeria species based on size, shape, structure and sporulation time of the sporulated oocysts. Itestinal Eimeriasis were seldom exist singly in one rabbit, most infection were being a mixture of two or more species **(Catchpole and Norton, 1979)**.

Cryptosporidium is another coccidian parasite affecting rabbits. The name Cryptosporidium Cuniculus was offered by **Rehg et al. (1979)** to the protozoan parasite found attached to the brush border of intestinal epithelial cells in apparently healthy rabbits. The pathogenic significance of Cryptosporidia in old rabbits is unknown but in young rabbits leads to serious growth retardation and diarrhea **(Okerman, 1988)**.

Diseased conditions of rabbits such as enteritis are associated with variable biochemical changes. Diarrhea resulted in hypokalemia and changes in the acid-base status **(Licois et al., 1978)**.

The present study was aimed to investigate the bacterial and the parasitic causes of mortalities in young and growing rabbits till weaning and the subsequent biochemical changes.

MATERIALS AND METHODS

1- Sampling :

A total of 60 privately owned broiler rabbitaries of variable sizes at El Bohira Povince were included in the present study. Their main complain was diarrhea and increased mortalities in young rabbits till around weaning . Samples from 180 morbid and apparently healthy rabbits of different breeds and sexes between 2-6 weeks old were obtained .

2- Bacteriological investigations :

Specimens were collected from liver, spleen, lungs, kidneys, intestine, heart and blood

Media :

a- Fluid media :Nutrient broth and Selenlet-F broth.

b- Semisolid media: Nutrient agar for isolates preserving.

c- Solid Media: Blood agar, Nutrient agar, Mac-Conky agar, S. S agar and Bird Barker agar. All were according to **Oxoied, 1998.**

Isolation and identifications : The collected samples were prepared according to **Cruickshank et al. (1975)**. After preparation, samples were cultured on Nutrient broth, Selenlet-F broth tubes and incubated over night at 37°C followed by culturing on the solid media and incubated aerobically at 37°C over night. The growing colonies in variable media were examined morphologically. Culturally, Gram stained and biochmically tested according to **Edwards and Ewing (1972), Cruickshank et al. (1975), Fingold and Baron (1986), and Carter and Cole (1991).**

3- Parasitological investigation :

- Samples were recovered from liver, intestine and fecal matter .
- Fecal concentration flotation method according to **Soulsby (1982)**.
- Intestinal or fecal smears were done, air dried, fixed with methanol and stained with modified Ziehl-Neelsen stain using technique performed by **Henriksen and Pohlenz (1981)**.
- Sporulation of oocysts in fecal culture by using 2.5 % potassium dichromate solution, according to **Soulsby (1982)**, and the sporulation time for each species was estimated.
- direct smear from gall bladder and liver were done and examined microscopically.
- Identification of different Eimeria species was realized according to criteria admitted by **Pellerdy (1965) and Coudert et al. (1988)**.
- Identification of Cryptosporidium oocysts was confirmed according to **Levine (1984)** and previous authors who had dealt with Cryptosporidia of rabbits **Arafa (1992) and Shtibashi et al. (2008)**. Different dimensions of oocysts were measured using the calibrated ocular micrometer.

4 - Biochemical Investigation :

- Calorimetrically analyzed using test kits for the blood serum.
- Blood glucose level was determined according to **Siet and Schiele (1981)**.
- Serum total proteins were determined according to **Doumas et al. (1981)**.
- Serum albumin was carried according to **Drupt (1974)**.
- Globulin was calculated as the difference between total protein and albumin .
- Inorganic phosphate was determined according to **Daly (1972)**.
- Serum calcium level was estimated according to **Gindler and king (1972)**.
- Sodium and Potassium were determined according to **Oser (1979)** .
- Serum Aspartate aminotranferase (AST) and Serum Alanine aminotransferase (ALT) were estimated according to **Reitman and Frankel (1957)** .
- Statistical analysis were carried according to **Snaedecor and Cochran (1982)**.

Apparently healthy rabbits which were negative for bacteriological and parasitological examination, were used as a control group.

Table (1): Incidence of Bacterial isolates singly or mixed from investigated rabbits among isolates (N=360) & among investigated cases (N=180)

Bacterial isolate	Single				Mixed				Total			
	Among Isolate		Among cases		Among isolate		Among cases		Among Isolate		Among cases	
	N	%	N	%	N	%	N	%	N	%	N	%
E. coli	30	8.3	30	16.6	54	15	54	30	84	23.3	84	46.6
Enterobacter Spp	6	1.6	6	3.3	33	9.2	33	18.3	39	10.8	39	21.6
Ent. Cloaca	6		6		21		21				27	
Ent. agglomer					12		12				12	
Citrobacter Spp					24	6.6	24	13.3	24	6.7	24	13.3
Cit. frundii					12		12				12	
Cit. diversus					12		12				12	
Proteus Spp.					24	6.6	24	13.3	24	6.7	24	13.3
Prot. vulgaris					12		12				12	
Prot. mirablis					12		12				12	
Klebsiella Spp	6	1.6	6	3.3	15	4.1	15	8.3	21	5.8	21	11.6
Kleb. Pneum	6		6		9		9				15	
Kleb. Oxylocia					6		6				6	
Pseudomonas erugenosa					6	1.1	6	3.3	6	1.1	6	3.3
Staph . cocci	30	8.3	30	16.6	108	30	108	60	138	38.3	138	76.6
Staph .aures	18	5.0	18	10	54	15	54	30	72	20	72	40
Staph. epedermids	6		6		36		36		42		42	
Staph. Intermedia	6		6		18		18		24		24	
Strept. & Enterococci	6	1.6	6	3.3	18	5	18	10	24	6.6	24	13.3
Enteroc. fecales												
Strept. ini												
Total	78	21.7	78		282	78.3	282		360	100%		

Table (2): Incidence of parasitic infestation among investigated rabbits (N=180).

Total parasitic incidence		Total Eimeria incidence		Total Cryptospordia incidence		Single Eimeria incidence		Single Cryptospordia incidence		Mixed Eimeria & Cryptospordia	
N	%	N	%	N	%	N	%	N	%	N	%
99	55	96	53.3	12	6.7	87	48.3	3	1.6	9	5.1

Table (3): Frequency of Eimeria species revealed from investigated rabbits (N=180).

Species	*E.preforance*			*E.media*			*E.magna*			*E.irresidea*			*E.intestinales*			Total		
Freq	T	S	M	T	S	M	T	S	M	T	S	M	T	S	M	T	S	M
No	32	5	27	22	3	19	17	3	14	14	-	14	11	-	11	96	11	85
%	17.8	2.8	15	12.2	1.6	10.6	9.4	1.6	7.8	7.8		7.8	6.1		6.1	53.3	6.1	47.3

N = number S = single incidence M = mixed incidence T = total incidence

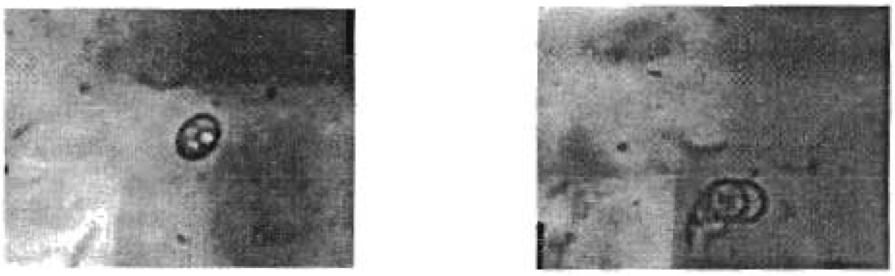

Fig. (1): Non sporulated and sporulated oocysts of Eimeria perforans.

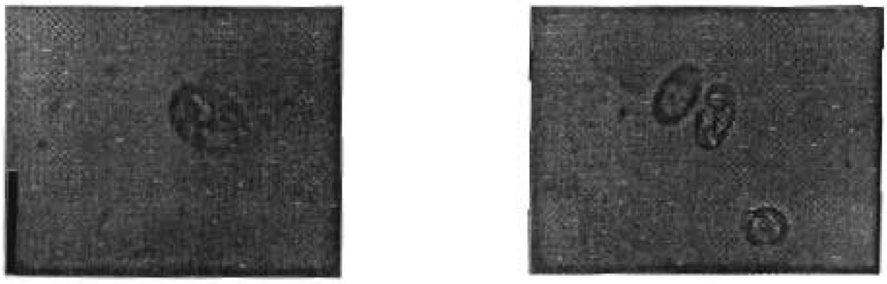

Fig. (2): Non sporulated and sporulated oocysts of Eimeria media.

Fig. (3): Non sporulated and sporulated oocysts of Eimeria magna.

Fig. (4): Non sporulated and sporulated oocysts of Eimeria irresidua.

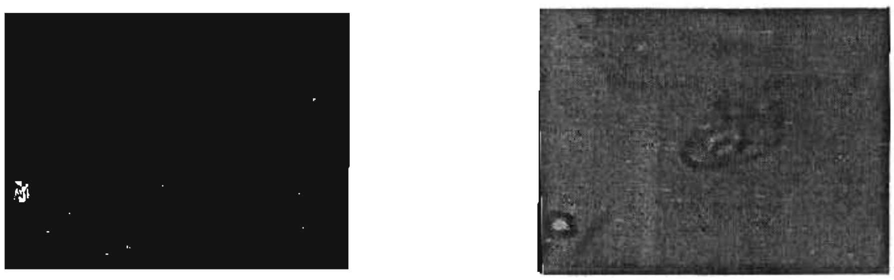

Fig. (5): Non sporulated and sporulated oocysts of Eimeria intestinalis.

Fig. (6): Cryptosporidium oocysts stained with modified Ziehl-Neelsen stain.

DISCUSSION

The rapid expansion of rabbit production in recent years in Egypt is accompanied by several problems. Mortalities in baby rabbits became a problem of almost concern for economic losses in rabbit production **(Okerman, 1987)**.

In the present study, the included 60 broiler rabbitaries were had complains of increased mortalities in young rabbits till around weaning. Bacteriological and parasitic investigations as well as the subsequent biochemical alterations were carried out.

The bacteriological studies revealed that 93.3 % (168 out of 180) of cases harboured a bacterial infection from which 21.7% yielded a single pure isolate and 78.3% yielded mixed culture. These results varied from those reported by **Hatab and mostafa (2003)** whom reported 63% bacteriological positive cases with 47.6 % and 52.4 % harbored single and mixed infection, respectively. This difference may turned to their study on apparently healthy baby kids.

Bacteriological examination of samples revealed that E. Coli was predominant isolate where it could be detected as sole cause in (30) 16.6% and mixed with other pathogens in (54) 30% of the investigated rabbits. The over all incidence of E. coli (84) 23.3% among all isolates . Nearly similar results were recorded by **Fahmy et al. (1985); Okerman (1987); Percy et al. (1993) and Boucher (2005)**, they concluded that, E. Coli infection is the primary causative agent in most outbreaks of diarrhea in rabbits. Our results disagreed with those reported by **Ghoniem et al. (1971); Ibrahim (1985); Abd EL-Gawad (1988); Alaha & Yousef (1999) and Abd EL-Rhman et al. (2005)**, that could be attributed to their study on diarrheal rabbits only.

Staph. auseus was the second predominant isolate where it could be detected in (18) 10% as sole cause, (54) 30% as mixed infection with total incidence of (72) 20 %among isolates. Similar results were recorded by **Fahmy et al. (1985), and El-Sayed and Moustafa (2007)** whom reported an incidence of 16.67%.

Members of Enterobacteriacae were frequently isolated in variable incidences. Enterobacter species were isolated in incidence of 10.8%. Enterobacter cloaca were detected as sole bacterial cause in (6) 3.3% and (12) 11.7% as mixed infection among cases. Enterobacter cloaca is a part of normal flora of gastrointestinal tract and widely distributed in environment. It is recognized as a major cause of infection and may cause a serious cause of nosecomal **(Dudley et al., 1980 and Gaston, 1988)**. The mechanism of infection by Ent. cloaca still a matter of disputation. Many authors turned it to endogenous translocation from the digestive tract. This idea was proven to be true when **Lambert-Zechovsky et al. (1992)** reported results of molecular analysis strongly supported that endogenous nature of systemic bacteria and meningitis due to Ent. cloaca .

Citrobacter, Proteus Spp. were detected as mixed infection with incidence of 6.6 %. Citrobacter frundii and Citrobacter diversus; Prot. vulgaris and Prot. mirabilis were biochemically identified. Our results agreed with those reported by **Dana Kremples (2006)** who recorded that Citrobacter frundii is opportunistic pathogen can cause a disease in compromised animals but is not usually a primary pathogen. It was recorded as respiratory invader.

Klebsiella pneumoniae is a typical member of Enterobactereacea that produce Entero toxins following penetration through intestine or respiratory mucosa, **EL-Sayed and Mostafa (2007)**. Often infection are not detected until respiratory signs occur lately but systemic infection are also common **(Jense, 1992)**. Klebsiella pneumoniae could be detected in (6) 3.3% as sole pathogen. It could be detected in mix with others in (9) 5 % with overall incidence of 8.3%. On the other hand, Klebsiella oxytocia was detected in (6) 3.3%. Our result agreed with those recorded by **Abd EL-Rahman et al. (2005)**, higher incidences were recorded by **Fahmy et al. (1985); Okerman (1987) and EL-Sayed and Mostafa (2007)** whom isolated Klebsiella pneumonia from 14.06 % of the examined rabbits.

Psudomonus aerogentosa were detected from 6 (3.3%) of examined cases, this is lower than that reported by **EL-Sayed and Mostafa (2007)** whom reported an incidence of 5, 10 and 10% in apparently healthy, diseased and dead kits, respectively .

Streptococci and Enterococci were detected as sole cause in (6) 3.3% and as mixed isolate in (18)10% with over all incidence of 13.3%. Similar results ware recorded by **Fahmy et al. (1985); Okerman (1987) and El-Sayed and Mostafa (2007)** where the later reported an infection rate of 13.02%.

Two major Coccidian parasites were found to infect rabbits, Eimeria and Cryptosporidia. Parasitic investigation of examined rabbits in our study recorded that (99) rabbits 55% had infestation with Eimeria and Cryptosporidia species as single or mixed forms (Table, 2). Concerning Eimeria species, the total incidence was (96 out of 180) 53.3% (Table, 2), this result agreed with **El-Masry (1983)** who recorded an incidence of 53.58 % in Egypt and **Jithendran and Bhat (1995)** 57.3% in India. Higher incidence in Egypt was found by **AbdAlla, 1988** (72.2%); **Ibrahim, 1990** (65.8%) and El-Tayar, 1996 (80.2%). An incidence rate of 96% was recorded in England by Catchpole and **Norton (1979)** and` 70% by **Percy et al. (1993)** in Canada. **Nosal et al. (2009)** found the level of infection was high in young rabbits with prevalence of (94.9 to 100%). While our result was higher than that recorded by **Peeters et al. (1984)** in Belgium who recorded 18.5% and **Aish (1994)** in Egypt 20% in foreign breeds and 42.8% in native breeds. The age, breed and health condition of sampled animals as well as management practices and environmental condition could explain the differences in the results.

In the present investigation, five species of Eimeria were identified among the investigated rabbits. They were E. preforans, E.media, E. magna, E. irresidua, and E.intestinalis (Table, 3). In Egypt, 4 Eimeria species were detected by **Ibrahim (1990)**, and 5 species by **Ahmed (1952); Abd El-Rahman (1972) and Abd Alla (1988), while Zahran (1979) and El Tayar (1996)** detected 8 species. This variation may be due to differences in age and breed of examined animals as well as differences in management practices. In other countries. 13 species were noted by **Rodriguez (1973)** in Spain, 8 species by **Catchpole and Norton (1979)** in Britain, 9 and species by **Peeters et al. (1981, 1984)** in Belgium, respectively, and 10 and 4 species by **Nosal et al. (2008 and 2009)**, respectively. In the present work infection with more than one species of Eimeria was common in 47.3% of examined rabbits. Eimeria prefo-ranes has the highest frequency (17.8%) either a sole species (2.8%) or mixed with other species (15%) followed by Eimeria media (Table, 3). Nearly similar results were reported by **Stodart (1971); Meshkow (1973); Catchpol and Norton (1979); Peeters et al. (1981); Ajayi et al. (1987); AbdAlla (1988) and Ibrahim (1990)**. In this study, Eimeria stiedae was not recorded. This was in accordance with that by **Catchpole and Norton (1979) and Peeters et al.(1981)**. From the descriptive point of view and sporulation time recorded in present work there were an agreement in the morphological features of the obtained oocysts with those recorded by **Pellerdy (1965); Levine(1985); Ibrahim (1990)**. Fig. (1,2,3,4,5).

Concerning Cryptosporidium species, this investigation is considered one of the rare studies about rabbit cryptosporidiosis reported in Egypt. Protozoan parasite Cryptosporidium has been identified as the cause of numerous water born, food born and day care outbreaks of diarrheal disease world wide **(Fayer et al., 2000)**. The result that we reported in the present work have shown Cryptosporidium infection occrued in young rabbits with prevalence of (12 out of 180) 6.7%, and occrued as a single parasite in 1.6% and mixed with Eimeria species in 5.1% (Table 2). In Egypt, **Arafa (1992)** recorded an incidence of 11.19% in naturally infected rabbits and added that one month rabbits appeared more susceptible (11.1%) than 2 and 3 months of age (9.4 and 7.3%, respectively). **Aish (1994) and El- Tayar (1996)** reported that all examined rabbits were free from Cryptosporidium spp. These variations in results may be attributed to health condition of examined rabbits, age, management and environmental condition. In Tunisia, **Soltane et al. (2007)** concluded that Cryptosporidium spp. not found in examined rabbits of 1-2 months of age. Prevalence of Cryptosporidium spp. in rabbits was 19.7% and 3.3% in diarrheic and normal feces respectively **(Shiibashi et al., 2006)**.

Concerning shape and size of the Cryptosporidium oocysts, faecal and intestinal smears stained with the modified Ziehl-Neelsen technique revealed pink, relatively spherical oocysts with smooth wall and greenish background as shown in figure (6), the revealed oocysts were of variable sizes measured 4.5-5.9 x 4.3-5.6 m, these morphological features were nearly agreed with those stated by **Inman and Takeuchi (1979); Arafa (1992) and Shiibashi et al. (2006)**. Cryptosporidium species rabbit genotype was identified by **Robinson and Chalmers (2009)** as the etiological agent in an out break of water born human cryptosporidiosis, the source was a wild rabbit that had entered a treated water tank. they add that both pet and wild rabbits are a potential source of human cryptosporidiosis and as such, good hygiene practices are recommended during and after handling rabbits or exposure to their feces, or potentially contaminated surfaces and water supplies should be protected. Further studies are required to analyzed the genotype of the Cryptosporidium oocysts to determine its zoonotic impact.

Table (4) revealed the biochemical alterations in variable bacterial infections. For the serious septicemic effects of E. coli, Staph aureus as single invader in contrast with both of them in the same case were studied in addition to other bacterial infections all were compared to check the significant effects on the studied biochemical parameters.

Glucose It is well known that, rabbits depend on bacterial fermentation in the cecum which yield volatile fatty acids. These fatty acids continuously absorbed as an energy source and sustained glucose blood level. Blood samples from 96 hours food deprivated rabbit showed no alteration in blood glucose levels **(Kozma et al., 1974)**.

The obtained results in table (4) revealed significant decrease in serum glucose level in rabbit invaded by other bacteria rather than E Coli, Staph. aureus together, or rabbits affected with Staph aureus only and in rabbits affected with bacterial infection mixed with Cryptosporidia, which may attributed these changes to the lake of absorption of glucose from the damaged intestine due to the harmful and destructive effects of bacterial and Cryptosporedia on the intestinal villa. These results agreed with those reported by. **El-Desouky and Nabila (2005); Hernandes et al. (2007) and Shahira and Sahar (2009).**

Total serum protein and it's fractions have a great importance in clarifying the healthy state of animals. In the present study as illustrated in table (4,5), a decrease in the total protein in bacterial and parasitic infection, was significant in cases of bacterial and cryptosporedial mixed infection, also in case of mixed infection with E Coli and Staph. aureus, in addtion to Staph. auras infection alone. That decrease in total protein and its fractions could be attributed to inability of animal gut to absorb and assimilate the dietary protein, furthermore, in state of anorexia , chronic enteropathy induce the inability of protein synthesis **(Ahmed, 2002; Radostits et al., 2000 and Shahira and Sahar, 2009)**.

Caecotrophs is a source of amino acids in rabbit. Parasitism prevent rabbits ingestion of Caecotrophs leading to reduce the amino acid which is necessary for protein syntheses **(Harcourt-Brown and Baker, 2001)**.

In the current study, the obtained data as illustrated in table (4,5) revealed a non-significant increase in the activities of liver enzymes, transaminase (ALT, AST) in diarrheic rabbits due to bacterial, and bacterial and parasitic infestations. AST significantly increase in mixed infection of E Coli and Staph. aureus. That could be attributed to tissues damage induced by bacterial and /or parasitic affection and its toxins **(Russel, 2003)**.

It is well known that, rabbit liver enzymes activities is lower than that in other species and there is less organ specificity **(Rosenthal, 1997)**. An increase in ALT signifies cell damage although the degree of the increase dose not correlate with the severity of hepatic disease and is not a prognostic indicator

(Willard et al., 1999). Moreover **Benson and Paul -Murphy (1999)** recorded that rabbit AST is found in liver, heart and skeletal muscles. The damage of these organs due to infection may be the visible cause of increase this level of activities.

Electrolyte pattern including Sodium and Potassium (table 4) revealed a significant decrease in sodium level (P< 0-05). That could be attributed to the loss of large amount of sodium ions with intestinal secretion and diarrhea **(Fadl- alla, 1989; EL-Sengary et al., 2004, and El-Desouky and Nabila, 2005)**.

Potassium showed increase (hyperkalemia) even non significantly increase as tabulated in table (4). that could be attributed to the subsequent acidosis associating with long standing enteritis in which false positive increase in Potassium value in attempt to compensate the occurring metabolic acidosis which known as K/H exchange **(Radostits et al., 2000)**.

Blood calcium levels are fixed in growing rabbits approximately 12 mg/dl in normal healthy one **(Harcourt-Brown and Baker, 2001)**. In the present study a decrease in serum calcium level, that turned to diarrhea which lead to decrease albumin levels. A proportional relation between level of each as calcium perform portion of albumin content **(Assane et al., 1993 and Duncan et al., 1994)**. Table (4) showed a significant decrease in serum calcium levels in bacterial infection and in mixed bacterial and parasitic affections. That could attributed to that rabbits calcium is not only absorbed from the gastrointestinal tract, it is also secreted into the gut across the intestinal mucosa, **(Barr et al., 1991)**. Moreover the decrease in serum calcium level could be attributed to the damage of intestinal mucosa due to both bacterial and parasitic and their toxins which impaired calcium absorption and secretion. Nearly similar results were reported by **Shigehiro et al., (2003); Assad and Nizar (2004), and Shahira and Sahar (2009)**.

Inorganic phosphorus in table (4,5) revealed non significant increase in its level These results agreed with those reported by **Cheeke (1987)** who claimed that phytates (Inositol Hexaphosphoric acid) that found in many planets specially grains contain phosphorus that is released into the digestive tract of rabbits due the action of caecal fermentation, investigation have shown that phytate phosphorus is available to rabbits .

In conclusion it is obvious that broiler small rabbits are more susceptible to serous problems specially around weaning. Diarrhea and pneumonia are predominant problems accompanied by Enterobacteriacae and Staph aureus and other bacteria in addition to parasitic infestations with Eimeria and Cryptosporidia. These affections causing biochemical alterations, So attention to the management factors and the nutrition of the pregnant dam to ensure adequate colostrums production with its valuable protective antibodies for protection of Kids. More over proper planning and adequate knowledge of the back ground of disease and application of preventive programs at right time. Intensive care and high biosafty measures for newly born.

Table (4). Effect of variable bacterial causes on biochemical parameters in diseased young rabbits

Biochemical parameters	*E. coli*		*Staph. aureus*		*E. coli* + *Staph. aureus*		Other bacteria		Control	
Glucose mg / dL	124.09±11.68	a	102.72±4.95	b	118.8±14.8	a	81.38±9.46	b	126.10±5.50	a
Asparate aminotransferase,U/l	32.32±2.23	b	28.39±2.25	b	42.80±6.25	a	27.00±3.21	b	28.80±2.92	b
Alanine aminotransferase,U/l	34.68±1.45	a	40.00±3.48	a	42.00±3.79	a	36.62±3.92	a	35.90 ± 2.77	a
Total protein g/dl	6.34±0.15	ab	5.84±0.12	b	5.88±0.54	b	6.61±0.23	a	6.53 ± 0.20	a
Albumin,g/dl	3.98±0.09	ab	3.79±0.09	b	3.64±0.39	b	4.02 ±0.13	ab	3.85±0.20	b
Globulin g/dl	2.36±0.14	ab	2.05±0.10	b	2.24±0.24	b	2.59 ± 0.21	a	2.68±0.11	a
Sodium	119.73±1.38	bc	121.11±3.23	bc	124.20±2.58	b	114.62±3.50	c	145.00±1.45	a
Potassium	5.35±0.25	a	6.22±0.40	a	5.54 ±0.38	b	5.48±0.48	a	5.75 ±0.49	a
Calcium	10.40±0.19	b	10.58±0.48	b	10.40±0.70	b	10.15±0.29	b	12.70 ±0.37	a
Phosphorus	6.52±0.40	abc	6.56±0.26	abc	6.54±55	abc	5.38±0.24	bc	5.98±0.24	b

Values are means and their standard errors.
Means within a row without a common letter differ significantly (p<0.05)

Table (5) Effect of bacterial infection alone and concurrent with Eimeria or Cryptosporidium on biochemical parameters in diseased young rabbits

Variable	Bacteria		Bacteria + Eimeria		Bacteria + Cryptosporidium		Control	
Glucose , mg / dl	107.72±6.60	a	115.30±8.84	a	50.50±7.54	b	126.10±5.50	a
Aspartate aminotransferase U/l	33.32±2.40	a	29.10±1.88	a	27.50 ± 7.59	a	28.50 ± 2.92	a
Alanine aminotransferase,U/l	38.32±2.27	a	37.00 ± 2.37	a	36.00±2.83	a	35.90 ± 2.77	a
Total protein , g/dl	6.39±0.13	a	6.31 ± 0.16	a	5.55±0.49	b	6.53 ± 0.20	a
Albumin , g/dl	4.09±0.11	a	3.89 ± 0.07	a	3.65 ± 0.32	a	2.85 ± 0.20	a
Globulin , g/dl	2.30±0.10	ab	2.42 ± 0.14	ab	1.90 ± 0.20	b	2.68 ± 0.11	a
Sodium	120.76±2.46	b	119.69±1.70	b	108.75±2069	c	145.00±1.45	a
Potassium	5.91±0.29	a	5.51 ± 0.29	a	5.33 ± 0.73	a	5.75±0.49	a
Calcium	10.22 ±0.27	b	10.42±0.26	b	11.35±0.81	b	12.70±0.37	a
Phosphorus	6.52 ± 0.27	ab	6.56±0.32	ab	6.25±0.45	ab	5.98±0.24	b

Values are means and their standard errors.
Means within a row without a common letter differ significantly (P<0.05).

REFERENCES

AbdAlla, A. R. Salma (1988) : Studies on endoparasities of rabbits in Assiut. M.V.Sc. Thesis. Fac. Med., Assiut Univ.

Abd EL-Gawad, A. M. (1988) : Some studies on Enterobacteriacae in rabbits. M. V. Sc., Fac. Vet . Med., Cairo Univ.

Abdel-Rhman, A. M. (1972) : Studies on coccidiosis in Assiut. M.V.Sc., Thesis, Fac. Vet. Med., Assiut Univ.

Abd-El Rahman, A. A.; Veveen, A. H. and Fatma, A. M. (2005) : Isolation and pathogenicity of intestinal Pathogens associated with the enteritis complex rabbits with special reference to E.coli and Salmonella spp. Assiut Vet. Med. J., 51(106): 180-197.

Ahmad, A. S. Nagwa (2002) : Serum protein profile in cattle calves infested with Cryptosporidia Spp.Egyp. J. Comp. and Clin. Path. 5(1): 149-155.

Ahmed, A. T. (1952) : The problem of coccidiosis of rabbit in Egypt. M.V.Sc., Thesis. Fac. Vet. Med., Fuad 1 Univ.

Aish, H. M. S. (1994) : Parasitic infestation in foreign and native breeds in rabbits. M.V.Sc., Thesis, Fac.Vet. Med. Moshtohor, Zagazig Univ., Benha branch.

Aisha - Ragab, A. and Yousief. H. M. Z. (1999): Escherichia coli isolated from chickens and rabbits with special reference to their pathogenicity. J. Egypt. Vet. Med. Ass., 59 (1): 45 - 59.

Ajayi, S. A.; Ajayi, A. and Ogunbiyi, A. O. (1987) : Clinical laprine coccidiosis in Newzealand white rabbits - An outbreak in Vom, Nigeria. Bull. Anim. Health Prod. Afr., 35 : 314-316.

Al-Mathal, E. M. (2008) : Hepatic coccidiosis of the domestic rabbit Oryctolagus Cuniculus dmesticus L. in Saudi Arabia. World Journal Zoology, 3(1): 30-35.

Arafa, A. A., Entesar (1992) : Morphobiological studies on the Cryptosporididae of rabbits. Ph. D. Thesis. Fac. Vet. Med., Cairo Univ.

Assad, A. and Nezar, A. (2004) : Studies on the clinical, hematological and biochemical changes in the diarrheic calves. Alex. Vet. Med. J., 21 (2): 712-719.

Assane, M.; Gongnet, G. P.; Colibaly, A. and Sere, A. (1993) : Influence of dietary calcium phosphorus ratio on blood calcium, phosphorus and magnesium during the gestation in rabbit. Rep. Nut. Dev., 33: 223-228.

Barr, D. R.; Sadomaki, D. L. and Bourfeay, J. E. (1991) : Characterization of the renal and intestinal adaptation to dietary Calcium deprivation in growing female rates. mineral, electrolytes metab. 17: 32 - 40.

Benson, K. G. and Paul-murphhy, J. (1999) : Clinical pathology of the domestic rabbit.Acquisition and interpretation of samples. Vet. Clin. North. Am. Exot. Anim. Pract., 2(3):539-551.

Blanco, J. B.; Blaco, M.; Blanco, J.; Mora, A.; Balaguer, L.; Mourino, M.; Juarez, A. and Jansen, W. H. (1996) : O serogroups biotypes and E.A.E.genes in Escherichia coli strains isolated from diarrheic and healthy rabbit. J. Clin. Microbiol. 34(12): 3101-3107.

Boucher, S. (2005) : Salmonellosis in a batch of companion rabbits. Pratique medical and chirgical de La Animal de campaign. 40 (1): 43 - 46 .

Carter G. R. and Cole, J. (1991) : Diagnostic procedures in Veterinary Bacteriology and mycology. 5th Ed., 293 - 320.

Catchpol, J. and Norton, C. C. (1979) : The species of Eimeria in rabbits for meat production in Britain. Parasitology, 79 (2): 249-57.

Cheek, P. R.; Bromson, J.; Roobenson, K. L. and Batton, N. M. (1987) : Availability of calcium, phosphorus and magnesium in rabbit food and mineral supplement. Appl. Rat Res.8:72 - 74.

Coudert, P.; Licois, D. and Provot, F. (1988) : Differential diagnosis of Eimeria species from the rabbit (poster). Proceedings 4th congress of the world rabbit Science association, (3): 480.

Coudert, P.; Licois, D.; Drouet- Viard, F. and Provot, F. (2000) : Coccidiosis. In Rosell, Y.M.(ed), (En fermedades del Conejo),Vol. ll , chapter XVI, PP219-243, Mundi-Prensa Libros, Madrid, Spain.

Cruickshank, R.; Duguid, J. P.; Marmion, B. P and Swain, R. H. A. (1975) : The practice of medical microbiology, 12th Ed., Churchill, Livingstone, London, U. K.

Daly, J. A. (1972) : Colorimetric method for detection of serum inorganic phosphorus. Clin. Chemist., (18):263.

Dana - Kremples (2006) : Rabbits Citrobacter frundii. Ph. D. w.w.w bio Miami edu, hare, sneezing Html.

Doumas, B. T.; Cater, R. J.; Peters, T. Jr. and Schaffer, R. (1981) : A candidate reference method for determination of total protein in serum .I. Development and validation. Clinical chemistry,(27):1642-1650.

Drupt, F. (1974) : Colorimetric method for determination of Albumin. Phar. Bio., (9).

Dudley, D. J.; Guentzel, M. N.; Ibarra, M. J.; Moore, B. E. and Sagik B. P. (1980) : Enumeration of potential pathogenic bacteria from sewage sludges . App . Environ. Microbiol., 39 (1): 118 - 126.

Duncan, J. R.; Grasse, K. W. and Mohaffy, A. (1994) : Veterinary laboratory medicine, clinical Pathology, 3rd Ed., Ams. Lawa state Univ press.

Edwards, P. R. and Ewing, W. H. (1972) : Identification of Enterobacteriacae. Burgess publ., co, Minnecopolis Minnesota, 103 - 103.

El-Dessouky S. A. and Nabila, M. O. (2005) : Effect of C. parvum infection on the physiological and blood biochemical changes of buffalo calves with special reference to the prevalence infection among buffaloes. Assiut. Vet. Med. J., 51 (105): 108-123.

El-Masry, N. M. (1983) : Problems of coccidiosis in rabbits. M.V.SC. Thesis, Fac. Vet. Med., Zagazig Univ.

EL-Sayed, M. H. and Moustafa, A. H. (2007) : Some studies on the bacterial causes of morality in new Born rabbits. Assiut . Vet . Med . J., 53 (112): 1258 - 268 .

El-Sengary, F. H.; El-Kabany, M. F. and Nagwa, A. Helmy (2004) : Some immunological, hematological and biochemical studies on kids suffering from diarrhea. Alex. Vet. Med. J., 53 (112): 258-268.

El-Tayar, A. Amal (1996) : Studies on protozoal parasites of rabbits. M. V. SC. Thesis, Fac. Vet. Med. Alex. Univ.

Fadl-Alla, M. G. E. (1989) : Clinical, Hematological and Biochemical studies on calves suffering from diarrhea. M. V. Sc. Thesis, Fac. Vet. Med., Cairo Univ.

Fahmy, M. F.; Anisa, M. Mustafa and Abd EL-Ghany (1985) : Pathological studies on rabbit diseases. Zag. Vet . J., 12 (2): 285 - 304 .

Fayer, R.; Morgan, U. M. and Upton, S. J. (2000) : Epidemiology of Cryptosporidium: transmission, detection and identification. Int. J. Parasitol., 30: 1305-1322.

Finegold, S. M. and Baron, E. L (1986) :

Diagnostic Microbiology. 7th Ed., Mosby company st. Louis Toronto Princention.

Gaston, M. A. (1988) : Enterobacter and emerging nosecomal pathogen. J. Hosp. infect., (2): 197.

Ghoniem, N.; Zein El-Abdin, Y. and Abd El-Hady, H. (1971) : Studies on pathogenic micro organisms in domestic rabbits in (U A. R). Egypt. Vet. Med. Assoc., 31: 227 - 234.

Gindler, E. M. and King, J. D. (1972) : Rapid calorimetric determination of Calcium in biological fluid with methylene blue. Am. J. Clinc. Pathology. 58: 376-382.

Harcourt-Brown, F. M. and Baker, S. J. (2001) : Parathyroid hormone, hematological and biochemical parameters in relation to dental disease and husbandry in bet rabbits J. small Anim. Pract .42: 130-136.

Harcourt-Brown, F. (2002) : Text book of rabbit medicine. 1st Ed., Linacert house Jordan Hill Oxford OX2 8Dp

Harkness, J. E. (1997) : Gastro enteric conditions in rabbit. In proceeding of the house rabbit Society Veterinary Conference, 19 - 25 .

Hatab, M. E. M. and Mostafa, E. M. (2003) : Studies on some bacteria associated with abortion in rabbit. Assiut.Vet. Med. J.52 (109) 285 - 293 .

Henriksen, S. A. and Pohlenz, J. F. L. (1981) : Staining of Cryptosporidia by modified Ziehl-Neelsen technique. Acta. Vet. Scand., 22: 495-596.

Hernandez, J.; Andrew, L.; Pyon, A.; Kakali, M.; David, J.; Mari, M.; Griffith, D. and Robenson, P. (2007) : Substance P. is responsible for physiological alteration such as increase chloride ion secretion and glucose metabolism in crypyosporidiosis. American Society for Microbiology, 75 (3): 1137-1143.

Ibrahim, A. A. (1985) : Colibacellosis of rabbits. Assiut Vet. Med. J., 14 (27): 243 - 246.

Ibrahim, I. A. Omima (1990) : Some morphological studies on the internal parasites of rabbits in Giza governorate. M.V.SC. Thesis, Fac. Vet. Med., Cairo Univ .

Inman, L. R. and Takeuchi, A. (1979) : Spontaneous cryptosporidiosis in adult female rabbit.Vet. Pathol., 16: 89-95.

Jensen, J. M. (1992) : Infectious and parasitic diseases of rabbits. Intern. Zan. and wild Animal medicine. Fowler, Med. W.B Sunder Philadelphia, 200 - 203.

Jithendran, K. P. and Bhat, T. K. (1995): Subclinical coccidiosis in angora rabbits, a field survey in Himachal Pradesh, India. World Rabbit Science, 4(1): 29-32 .

Kozma, C.; Maklin, W.; Cummins, L. and Mauer, R. (1974) : The anatomy, physiology and biochemistry of the rabbit in the biology of the laboratory. Rabbits Academic press, 50-69.

Lambert- Zechovsky, N.; Bingen, E.; Denamur, E.; Brahimi, N.; Brun, P.; Mathieu, H. and Elion, J. (1992) : Molecular analysis provides evidence for the endogenous origin of bacteremia and meningitis due to Enterobacter cloacae in an infant. Cin. infect. Dis., 15 (1): 30 - 32.

Lelkes, L. and Change, C. L. (1987) : Microbial disbiosis in rabbit enteropathy. Lab. Anim. Sci., 36: 757 - 764 .

Levine, N. D. (1984) : Taxonomy and review of the coccidian genus Cryptosporidium (Protozoa, Apicomplexa). J. Protozool., 31: 94-98 .

Levine, N. D. (1985) : Veterinary protozoology. 1st Ed. Iowa state Univ. Press, Ames.

Licois, D.; Coudert, P. and Mongin, P. (1978) : Changes in hydro mineral metabolism in diarrhoeic rabbits 2. study of the modifications of electrolyte metabolism .Ann Rech. Vet. Scin. 9: 453 - 464.

Meshkow, S. (1973) : Etiological study of coccidiosis in rabbits. Veterinaria SbirkaBulgaria,70 (8): 14-18 .

Nosal, P.; Petryszak, A.; Nowosad, B. and Sobolewska, M. (2006) : Gastrointestinal parasites of rabbits in coproscopic investigations. Wiad parazytol., 52(4): 327-30 .

Nosal, P.; Kowal, Y.; Nowosad, B.; Bieniek, J. and Kowalska, D. (2009) : Dynamics of endoparasites infections in rabbits at different rearing regims. Wiad parazytol., 55 (2) : 173-177 .

Okerman, L. (1987) : Pre-weaning mortalities in rabbits. study of bacteriology and pathology. Land bouwting dschrift., 40 (5): 1295 - 1304.

Okerman, L. (1988) : Diseases of domestic rabbits. Book was published by black well scientific publications.

Oser, B. L. (1979) : Hawk's physiological chemistry. 14th Ed. MC. Grow Hill company, L. T. d London.

Oxoied, M. (1998) : The Oxoied Manual of cultured media ingredient and other laboratory Crevices 8th Ed. Oxoied limited.

Peeters, J. E.; Geeroms, R.; Froyman, R. and Halem, P. (1981) : Coccidiosis in rabbits: a field study. Res. Vet. Sci., 30 (3): 328 - 34.

Peeters, J. E.; Pohl, P. and Charlier, G. (1984) : Infectious agents associated with diarrhea in mmercial rabbits:a field study. Ann. Resch. Vet.,15 (3):335 -340.

Pellerdy, L. P. (1965) : Coccidia and coccidiosis. Akademia Kiado, Budapest, Hungary,323-324.

Percy, D. H.; Muckle, C. A.; Hampson, R. J. and Brash, M. L. (1993) : The enteritis complex in domestic rabbits: a field study. Can. Vet. J., 34: 95 - 102 .

Radostitis, O. M.; Arundel, J. H. and Gay, C. C. (2000) : Veterinary medicine. 10th Ed. Bailliere Tidal and Philadelphia .

Rehg, J. E.; Lawton, G. W. and Pakes, S. P. (1979) : Cryptosporidium Cuniculus in the rabbits (Oryctolagus Cuniculus). Lab. Anim. Sci., 29: 656-660.

Reitman, S. and Franckel, S. (1957) : Calorimetric determination of S.AST, S.ALT enzymes activities. Ann. J. Clin. path. 28: 56.

Robinson, G. and Chalmers, R. M. (2009) : The European rabbit (Oryctolagus cuniculus) a source of zoonotic cryptosporidiosis. Zoonoses and Pulbic health, 23 .

Rodriguez, A. J. (1973) : Eimeria species parasitic in rabbit with reference to their occurrence in Spain. Supplemento scientifico del boletin informative, Consejo. Gen. Colog. Vet. Espana, 196: 37-43.

Rodriguez -Delara, R.; Cedillo-Pelaez, C.; Constantino-Casas, F.; Fallas -Lopez, M.; Cobos-Perolta, M. A.; Gutierrez-Olvera, C.; Juarez-Acevedo, M. and Miranda-Romero, L. A. (2008) : Studies on the evolution ,pathology and immunity of commercial fattening rabbits affected with epizootic outbreaks of diarrhea in Mexico: a case report. Res. Vet. Sci., 184 (2): 257-268.

Rosenthal, K. (1997) : Interpretations of selected clinical pathology values in ferrets and rabbits. Proceeding of Atlantic coast Veterinary Conference, 1-3.

Russel, S. (2003) : The effect of Cryptosporidiosis on certain biochemical constituents of calves. Vet. Parasit., 255-260.

Shahira, H. M. and Sahar, S. A. (2009) : Field trial for treatment of diarrhea in kids at Sharkia governorate. 6^{th} Int. Sci. Conf., 447 - 461.

Shigehiro, I. J.; Kazuyuki, S.; Yohoko, S.; Kouji, I.; Eiichiro, K. and Ryuji, A. (2003) : Effect of intravenous infusion of hypotonic lactated ringer solution on calves with diarrhea. J. Vet. Med. Sci., 65 (9): 1033-1036.

Shiibaashi, T.; Imai, T.; Sato, Y.; Abe, N.; Yukawa, M. and Nogai, S. (2006) : Cryptosporidium infection in Juvenile pet rabbits. J. Vet. Med .Sci., 68 (3): 281-282.

Siet, G.; Henny, J. and Schiele, F. (1981) : Interpretations des examines de laborator karga Ed. 206.

Snaedecor, G. W. and Cochran, W. G. (1982) : Statistical methods 8^{th} Ed Lowa State Uni. Vet. Press USA.

Soltane, R.; Guyot, K.; Del-Cas, E. and Ayadi, A. (2007) : Prevalence of Cryptosporidium spp. (Eucoccidiorida : Cryptosprtidae) in seven species of farm animals in Tunisia. Parasite, 14: 335 -338 .

Soulsby, E. J. L. (1982) : Helminthes. Arthropods and Protozoa of domesticated animals. 6^{th} Ed. London, Bailliere, Tindall and Cassel .

Stodart, E. (1971) : Coccidiosis in wild rabbits. Oryctolagus Cuniculus (L). At a site on the coastal plain in Eastern Australia. Aust. J. Zool., 19: 287-292.

Willard, M. D.; Tvedtem, H. and Turm Wold, G. H. (1999) : Small Animal clinical diagnosis by laboratory methods. 3^{rd} Ed . W. B. Saunders.

Zahran, O. T. (1979) : Studies on the methods of control of rabbit coccidiosis. M. V. Sc. Thesis, Fac. Vet. Med., Cairo Univ.

الملخص العربى

المسببات البكتيرية و الطفيلية المصاحبة لنفوق الأرانب الصغيرة والتغيرات الكيميائية المصاحبة

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أجريت هذه الدراسة لتقييم بعض المسببات البكترية والطفيلية المصاحبة لنفوق الأرانب الصغيرة وحتى الفطام ودراسة التغيرات البيوكيميائية المصاحبة. وخلال هذه الدراسة تم فحص 60 مزرعه بأحجام مختلفة بمحافظة البحيرة بها شكوى من زيادة معدلات النفوق في الأرانب صغيرة السن وحتى الفطام وتم تجميع عينات من أرانب مريضة اضافة لارانب سليمة ظاهريا كمجموعة ضابطة جميعها بذات الأعمار وقد أظهرت النتائج أن 168 من 180 الأرانب المختبرة بنسبة 93.3% ايجابية بكتريولوجيا منها 78.3% بها اصابة باكثر من معزول و 21% مصابة بمعزول واحد. وقد تم عزل العصيات القولونية ،الانتيروباكتر ،الستروباكتر ،البروتيس ،الكليبسيلا ،الزائفات الهوائية السودوموناس و المكورات العنقودية الذهبية. أوضحت الدراسة الطفيلية ان 99 حالة ممثلة 55% مصابة بطفيل الكوكسيديا والكريبتوسبوريديا، تم تصنيف خمسة انواع من الكوكسيديا المعوية. هذا ولم يتم التعرف على أو عزل الكوكسيديا الكبدية فى هذه الدراسه . وقد تم عزل وتصنيف طفيل الكريبتوسبوريديامن 12 حالة بنسبة 6.7%. تم دراسة التغيرات البيوكيميائية فى مصل الدم المصاحبة للإصابات البكتيرية والطفيلية. هذا وقد تم مناقشة النتائج واثرها الاقتصادى واهميتها على الصحة العامة وابداء التوصيات اللازمة لحل هذه المشكلة وطرق الوقاية لمنع حدوثها .