

Investigation Of The Protective Role Of Albendazole On Male Fertility, Hormonal And Some Biochemical Parameter Induced By Gastrointestinal Nematode On Rams

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ABSTRACT

The present study was designed to evaluate the effect of gastrointestinal nematodes on male fertility and some biochemical parameters in rams and modulating this effect by using albendazole. Twenty rams aged 1.5 - 2 years old weighing 40 – 45 kg belonged to a private farm at Sharkia Province were used in this trial. Rams were divided into four equal groups (5 each). 1st group clinically healthy rams free from parasite (control group), 2nd group clinically healthy rams free from parasite received 5mg albendazole/kgm bwt one dose, 3rd group rams gastrointestinal nematodes infested non treated and 4th group gastrointestinal nematodes infested and treated with 5mg albendazole/kgm bwt. one dose. Blood, fecal and semen samples were collected from all rams at 7th, 14th and 28th day post treatment for biochemical analysis evaluate semen character and evaluate efficacy of albendazole against gastrointestinal nematodes.

Fecal examination revealed presence of gastrointestinal nematodes eggs as haemonchus spp, trichuris spp, trichostrongylus spp., Ostertagia circumcieta and mixed infection Trichuris + Haemonchus and Ostertagia +Trichostrongylus. Reduction of egg count of gastrointestinal nematodes 100 % at 14th day post albendazole treatment.

Healthy rams received albendazole show significant decrease in testosterone hormone, sperm cell concentration, individual motility, alive sperm, total protein, albumin and globulin. Beside significant increase in sperm abnormality and insignificant decrease in ejaculate volume, coupled with insignificant increase in serum and seminal plasma trace elements, ALT, AST, Alp, urea and creatinine.

Rams infested with gastrointestinal nematodes show significant decrease in testosterone hormone, sperm cell concentration, individual motility, live sperm, serum and seminal plasma trace element, total protein, albumin and globulin and significant increase in total abnormality, liver enzymes (AST, ALT & ALP) urea and creatinine. Beside insignificant decrease in ejaculate volume.

Infested rams treated with albendazole show return the previous parameter to nearly normal levels at 28th day post treatment

It could be concluded that the albendazole effective against gastrointestinal nematodes and ameliorating its adverse effect in rams fertility and biochemical parameters but its have transit adverse effect in male fertility so; it is not used during mating period.

Key words: Ram- Gastrointestinal nematodes -Fertility - Semen- Albendazole – Biochemical parameters

INTRODUCTION

Sheep and goats, representing an important source of animal protein in third world countries (1). Parasitic infections especially gastrointestinal nematode poses a serious health threat and limits the productivity of livestock (2) unthrifty (3). reduced fertility (4). Gastrointestinal nematodes sensitize animals to shortage of trace elements and vitamins lead to destruction of gastrointestinal mucosa (5).

There are many drugs used against any parasites routinely administered in farm animals (6). Many studies proved that some anthelmintic drugs cause a transitory depression of fertility, suggest avoiding the treatment of animals during the mating season (7). Albendazole, a benzimidazole derivative, is authorized for used in treatment and control of gastrointestinal nematodes as well as liver flukes in ruminant species (8). They inhibit the energetic metabolic of parasites and bind with tubulin, inhibiting its polymerisation and thus interfering with microtubule-dependent glucose uptake (9). It is permitted for use in both pregnant and non-pregnant animals (10). Its antiparasitic drugs used in treatment domestic animals and used against a variety of nematodes (11).

The present study focused on evaluation of the efficacy of albendazole in rams naturally infested with gastrointestinal nematode, studying the effect of infection and treatment in semen picture and some biochemical parameters in rams.

MATERIAL AND METHODS

Drugs

Albendazole (parabenzole tablet)^R each tablet contain 250mg albendazole produce by pharmasweed company Egypt. It is recommended dose as 5mg/kgm body weight (1 tablet/50 kgm body weight) (12)

Animals

Twenty rams of native breed, 1.5 -2 years old weighting 40–45kg Bwt. at aprived farm in Sharkia Province Egypt were used in this study.

Experimental design

rams were divided into four groups five in each, 1st group healthy rams free from internal and external parasite (control group), 2nd group healthy rams free from internal and external parasites (continuous faecal examination was carried out for 3 weeks befor experiment to be sure that these rams free from helminthes , also external parasites) treated with 5mg albendazole/kgm. Bwt orally. 3rd group infected with gastrointestinal nematodes non treated and 4th group naturally infected only with gastrointestinal nematodes were treated with 5mg albendazole/kgm. bwt orally.

Fecal samples

Rectal faecal samples were obtained from each rams before and at 5, 7, 14 and 28 days post treatment per rectum with new gloves for each animal. Collected samples were labelled and transported to the laboratory where they immediately examined. The concentration and floatation technique was used to detect the presence of eggs of nematodes in the samples (13). Worm identification through culturing faecal samples (14) and the identification of gastrointestinal nematodes larvae was done (15). Degree of infestation was determined by counting the egg/gram faeces through MC Master Technique (15), before and 28 days after treatment for each rams.

Blood samples

Blood samples were collected from all rams via Jugular vein puncture at 7th, 14th and 28th days post treatment, then centrifuged for obtain clear serum and used for determination of trace elements (Selenium, copper, iron, upper and zinc) by using atomic absorption spectrophotometer (16). Serum testosterone hormone (17), total proteins (18) albumin (19) and serum globulin were calculated as difference between total proteins and albumin. AST, ALT (20) ALP (21), urea (22) and creatinine (23).

Examination of semen sample

Semen samples were collected from each rams before and at 7th, 14th and 28th days post treatment to estimate ejaculate volume, sperm cell concentration, motility%, alive sperm% and total abnormalities% (24) post spermatological examination semen samples were centrifuged at 3000 rpm for 10min for obtain seminal plasma for estimation trace elements.

Statistical analysis

The obtained data were analyzed (25).

RESULTS AND DISCUSSION

In the present study, parasitological examination of fecal samples revealed presence of gastrointestinal nematodes eggs as trichuris spp, trichostrongylus spp., haemonchus spp, Ostertagia circumcieta as single infection and mixed infection Trichuris+ Haemonchus and Ostertagia + Trichostrongylus (table, 1). This result was in agreement with (26) stated that haemonchus spp is a common abomasal nematode, Trichuris spp. are common sheep parasites and trichostrongylus spp., have been reported from intestine and abomasum of goats. Same prevalence of gastrointestinal nematodes was recorded by (27)

Oral administration of albendazole to infected rams with gastrointestinal nematodes revealed that such eggs gastrointestinal nematodes were completely disappeared at 14th day post treatment (table, 1). Similar results were recorded previously (28- 29) they found that efficacy of albendazole was 99.6% at days 14th post treatment. Meanwhile reduction in faecal gastrointestinal nematodes gastrointestinal nematodes egg counts post using albendazole in sheep was 100% at 14th day post treatment (30). Additionally, albendazole was highly efficacious in removal gastrointestinal nematodes in sheep and goats (31).

The present study (table, 2 and 3) demonstrated that there was insignificant increase in serum and seminal plasma trace elements (copper, iron, zinc and selenium) in healthy rams received albendazol but infested rams with gastrointestinal nematodes show significant decrease in serum and seminal plasma trace elements, these changes regained nearly normal level at 28 days post treatment. The classical feature of parasitic infestation in ruminants is decrease in serum trace elements (32). Our findings are in agreement with that (33) they found significantly lower in serum trace elements levels in sheep infected with gastrointestinal nematodes. Reduction in serum trace elements elements in infested rams may be due to gastrointestinal nematodes induced intestinal damage and decrease absorption in infested animal (34). Gastrointestinal nematodes nematode infection induced significant reduction in seminal plasma trace elements (35).

In the present study, it has been noticed that, healthy rams received albendazol and infested rams with gastrointestinal nematodes resulted significant decrease in testosterone hormone, sperm cell concentration, individual motility, live sperm and significant increase in total abnormality. Beside insignificant decrease in ejaculate volume. Decrease in testosterone hormone may be due to gastrointestinal nematodes infection affect central nervous system and depress the action of anterior pituitary lobes on the gonads (36). Ram suffering from lack of selenium show decrease in testosterone hormone (37). Decrease in zinc caused impairs responsiveness of leydig cell to gonadotro-phins lead to reduction in testosterone hormone secretion and inferior semen picture in ram (38). Same results were recorded by (39) they stated that metabolic regulation of sperm appears to be mediated by zinc, as a regulator of enzyme activity in the sperm. In addition, (40) mentioned that reduction in sperm motility due to decrease in androgen production. Decrease in trace element induces significant decrease in semen volume, sperm number, sperm motility, alive sperm (41). Our findings were reinforced with those recorded by (42) the author found that,

deficiency of zinc and/or selenium resulting in a decrease in semen quality and production due to the importance of zinc and selenium in stabilizing nuclear chromatin of spermatozoa. Albendazole treatment had harmful effects on semen quality in healthy rats (43). Also, albendazole induced reduction in physical semen characteristics in bulls (44). Moreover, albendazole treatment deleteriously affected sperm motility (45). Our results were supported (46) stated that decreased physical semen characteristics may be due to degenerations in testicular tissues post albendazole dosing. Ricobendazole induced reduction sperm count, motility and raise in sperm abnormalities (47).

In the current study, it was found that gastrointestinal nematodes infestation and healthy rams received albendazole revealed significant decrease in total protein, albumin and globulin in rams. This result was consistent with that reported (48,49) found that serum total protein, albumin and globulin levels decreased significantly in sheep and goats infested with gastrointestinal nematodes. Also, the obtained data coincide with (50) found that gastrointestinal nematodes infection induced significant decrease in albumin and globulin in cattle. Gastrointestinal nematodes infestation is associated with alteration in absorption or metabolism of protein ending in hypoproteinaemia (34,51). Further explanation of hypoalbuminemia comes from (52) stated gastrointestinal nematodes induces anorexia led to decrease in albumin. Albendazole induce significant decrease in albumin (53,54) in

healthy cattle and goats received albendazole respectively.

Analysis of the biochemical results indicated that, administration of albendazole in single dose in healthy rams induced insignificant increase in liver enzymes (ALT, AST and Alp) urea and creatinin meanwhile rams infected with gastrointestinal nematodes show significant increase in total abnormality, liver enzymes (AST, ALT and AP) urea and creatinine. Same results were recorded (47,55) in rats received ricobendazole and albendazole respectively. This result was in agreement with that (56) who reported that thiabendazole induced insignificant increase in concentrations of AST ALT and ALP in mice. Also, albendazole at dosage 30 mg/kg Bwt induce insignificant effect on hepatorenal function in goats (57). Elevation in liver enzymes in was observed in sheep infested gastrointestinal nematodes (58). The elevation in liver enzymes could be attributed to liver involvement by metabolic products of gastrointestinal nematodes (59).

Infested rams treated with albendazole ameliorates the adverse effects of gastrointestinal nematodes and returns the biochemical parameters and semen picture to nearly normal at 28th day post treatment due to the nematocde activity of abendazole.

Finally, it could be concluded that the albendazole, effective against gastrointestinal nematodes and ameliorating the adverse effect of gastrointestinal nematode in rams but its have adverse effect in male fertility so, it is not used during mating period.

Table 1. Efficacy of albendazole in rams naturally infested with gastrointestinal nematodes.

Nematode species	rams Number	Egg count / gram faeces				
		Pre Treated	5 Days Post Treatment	7	14	28
Trichuris.spp.	1	1560	610	43	0	0
Trichostrongylus.spp.	2	610	80	29	0	0
Ostertagia circumcieta	3	563	94	50	0	0
Haemonchus contorus	4	830	420	93	0	0
Mixed Inf. (Trichuris+ Haemonchus)	4	12540	270	61	0	0
Mixed Inf. Ostertagia + Trichostrongylus	5	983	363	93	0	0

Table 2. Effect of gastrointestinal nematodes and albendazole on serum trace elements in rams (n=5)

Parameter	Groups		Healthy rams			Rams infested with gastrointestinal nematodes		
	Non Treated	Treated	Days Post Treatment			Non Treated	Days Post Treatment	
			7	14	28		7	14
Copper (µmol/L)	25.20±	27.27±	25.15±	25.23±	12.46±	17.11±	21.79±	23.9±
	3.40	1.12	1.13	1.44	2.91*	1.60*	1.21	3.22
Selenium (Ug%)	108.14±	108.21±	108.20±	108.18±	96.19±	99.23±	103.38±	106.05±
	1.84	1.04	1.42	1.60	1.96*	2.88*	0.82*	0.98
Zinic (µmol/L)	22.08±	22.37±	22.18±	22.11±	11.28±	16.34±	20.39±	22.02±
	1.41	1.21	1.31	1.62	1.18**	1.63*	1.06	1.19
Iron (µmol/L)	47.20±	50.11±	48.17±	48.31±	34.82±	40.58±	44.29±	46.16±
	2.68	1.92	1.86	1.26	2.50**	1.18*	1.27	1.42

* P < 0.05

** P < 0.01

Table 3. Effect of gastrointestinal nematodes and albendazole on Seminal plasma trace elements in rams (n=5)

Parameter	Groups		Healthy rams			Rams infested with gastrointestinal nematodes		
	Non Treated	Treated	Days Post Treatment			Non Treated	Days Post Treatment	
			7	14	28		7	14
Copper (Ug/dl)	3.06±	3.08±	3.18±	3.11±	1.37±	2.05±	2.95±	3.15±
	0.14	0.26	0.12	0.37	0.50*	0.11*	0.19	0.41
Selenium (Ug%)	74.05±	74.21±	74.18±	74.08±	70.31±	69.24±	71.06±	73.11±
	1.16	1.06	1.31	1.21	1.04*	1.15*	0.51*	0.37
Zinic (Ug/dl)	62.32±	62.48±	62.44±	62.33±	56.19±	59.22±	62.14±	62.54±
	1.18	1.17	1.28	1.32	0.86**	0.30*	0.52	0.42
Iron (Ug/dl)	36.12±	36.19±	36.14±	36.13±	30.02±	33.25±	34.39±	36.72±
	0.69	1.21	1.11	1.214	1.47**	0.85*	0.38*	0.48

* P < 0.05

** P < 0.01

Table 4. Effect of gastrointestinal nematodes and albendazole on sperm picture in rams (n=5)

Parameter	Groups		Healthy rams			Rams infested with gastrointestinal nematodes		
	Non Treated	Treated	Days Post Treatment			Non Treated	Days Post Treatment	
			7	14	28		7	14
Testosterone	2.87±	1.78±	1.98±	2.42±	1.32±	1.69±	2.10±	2.74±
	0.40	0.22*	0.34*	0.41	0.30**	0.48*	0.31	0.37
Semen volume ml	1.69±	1.41±	1.50±	1.54±	1.05±	1.24±	1.55±	1.64±
	0.42	0.33	0.21	0.19	0.25	0.29	0.27	0.35
Sperm cell conc. 109	3.02±	1.97±	2.31±	2.87±	1.95±	2.48±	2.69±	3.01±
	0.21	0.33**	0.11*	0.25	0.13**	0.10*	0.39*	0.14
Sperm motility%	84.69±	79.18±	80.06±	82.90±	75.59±	79.30±	80.19±	83.23±
	1.38	0.82**	0.24*	1.94	1.82**	1.28*	1.60*	1.29
Live Sperm%	82.51±	72.10±	76.18±	80.67±	69.47±	75.60±	80.27±	81.89±
	2.81	2.14*	1.60	1.97	1.75**	1.30*	1.07*	1.69
T.Abnormality %	10.09±	13.05±	12.48±	10.18±	14.73±	12.25±	11.53±	10.21±
	0.47	0.89*	0.60*	0.51	0.97*	0.34**	0.18*	0.2

* P < 0.05

** P < 0.01

Table 5. Effect of gastrointestinal nematodes and albendazole on Protein profile in rams (n=5)

Parameter	Groups	Healthy rams				Rams infested with gastrointestinal nematodes			
		Non Treated	Treated			Non Treated	Days Post Treatment		
			7	14	28		7	14	28
T.Protein		7.63±	5.89±	6.27±	7.41±	6.53±	6.78±	7.20±	7.46±
		0.30	0.60*	0.51*	0.68	0.27*	0.11*	0.17	0.14
Albumin		4.65±	3.85±	4.17±	4.39±	3.45±	3.75±	4.05±	4.08±
		0.26	0.22*	0.31	0.48	0.29*	0.18*	0.29	0.49
Protein Picture (mg/dl)	Globulin	3.98±	2.04±	2.10±	3.02±	3.08±	3.03±	3.15±	3.38±
		0.30	0.62*	0.56*	0.62	0.11*	0.21*	0.31	0.26
	A/G Ratio	1.20±	1.80±	1.44±	1.45±	1.12±	1.29±	1.32±	1.20±
		0.30	0.36	0.32	0.28	0.14	0.13	0.18	0.07
Liver enzymes (U/L)	AST	76.93±	77.75±	77.55±	77.13±	84.27±	83.18±	77.06±	76.62±
		1.53	1.43	1.82	1.21	1.05**	0.43*	0.97	0.83
	ALT	32.95±	33.07±	32.99±	32.85±	36.12±	34.76±	33.18±	32.98±
		0.78	0.95	0.49	0.95	0.99*	0.28*	0.38	0.78
	ALP	32.57±	33.77±	32.88±	32.63±	36.13±	35.05±	33.15±	32.60±
		0.12	0.63	0.41	0.53	0.86**	0.98*	0.42	0.94
Kidney function (mg/dL)	Urea	15.81±	16.90±	16.03±	15.80±	20.48±	18.97±	16.36±	15.89±
		0.70	0.21	0.48	0.52	0.84*	0.57*	0.88	0.67
	Creatinine	1.54±	1.59±	1.57±	1.55±	2.87±	2.02±	1.68±	1.58±
		0.19	0.17	0.37	0.15	0.41*	0.16*	0.42	0.20

* P < 0.05

** P < 0.01

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الملخص العربي

استبيان الدور الوقائي للبندازول على الخصوبة الذكورية، الهرمونات وبعض الوظائف البيوكيميائية المحدثة ببعض الديدان الداخلية (المعدية المعوية) في الكباش

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تم استخدام ٢٠ كبش فى سن ٥، ١-٢ سنة ووزنها يتراوح من ٤٠ الى ٤٥ كجم. بمحافظة الشرقية لدراسة كفاءة البندازول على الإصابة الطبيعية بالديدان الداخلية (المعدية المعوية). تم تقسيم هذه الكباش إلى اربع مجموعات متساوية كلا يحتوى على ٥ كبش. المجموعة الأولى كباش خالية من الديدان الداخلية (مجموعة محكمة). المجموعة الثانية كباش خالية من الديدان الداخلية وتم اعطائها البندازول بجرعه ٥ مجم/كجم من وزن الجسم، المجموعة الثالثة كباش مصابة اصابه طبيعيه بالديدان الداخلية (المعدية المعوية) ولم تعالج. المجموعة الرابعة كباش مصابة اصابه طبيعيه بالديدان الداخلية (المعدية المعوية) وعولجت بالبندازول بجرعه ٥ مجم/كجم من وزن الجسم. تم اخذ عينه دم، براز وسائل منوى من كل كبش فى كل المجموعات عند اليوم السابع والخامس عشر والثامن والعشرين من نهاية العلاج. لدراسة تاثير الديدان الداخلية والعلاج على بعض الوظائف البيوكيميائية وصورة الحيوانات المنوية.

بالفحص الباراسيتولوجى لعينات البراز وجد اصابات مختلفه من بويضات الديدان الداخليه مثل ترايكريس ،ترايكواسترنجليس،هيمونكس،اوسترتاجيا،وعدوى مشتركه من(ترايكريس مع هيمونكس- اوسترتاجيا مع ترايكواسترنجليس).وبفحص عينات البراز بعد العلاج وجدت ان البندازول قضى على تلك الديدان الداخليه فادى الى اختفاء بويضات تلك الديدان بنسبه ١٠٠% عند ١٥ يوم بعد العلاج.

وبدراسة التغيرات البيوكيميائية التى حدثت نتيجة اعطاء البندازول للكباش السليمه والخالیه من الديدان الداخليه وجد نقص معنوى فى هرمون التسترون، حجم القذفه، تركيز الحيوانات المنوية ومعدل الحركة وعدد الحيوانات المنوية الحية، البروتين الكلى الزلال والجلوبيولين بجانب زياده معنويه فى نسبة العيوب الشكلية فى الحيوانات المنوية الحية.كما ادى البندازول الى حدوث زياده غير معنويه فى العناصر النادرة (الزنك، النحاس الحديد والسيلنيوم) وانزيمات الكبد (ALT, AST, Alp) اليوريا والكرياتينين.

الاصابه بالديدان الداخليه فى الكباش ادت الى حدوث نقص معنوى فى ، العناصر النادرة (الزنك، النحاس الحديد والسيلنيوم) هرمون التسترون، تركيز الحيوانات المنوية ومعدل الحركة وعدد الحيوانات المنوية الحية، البروتين الكلى الزلال والجلوبيولين ونقص غير معنوى فى حجم القذفة بالاضافه الى زياده معنويه فى نسبة العيوب الشكلية فى الحيوانات المنوية الحية وانزيمات الكبد (ALT, AST, Alp) اليوريا والكرياتينين حتى اليوم الرابع عشر من نهايه العلاج وعادت الى وضعها الطبيعى عند اليوم ٢٨ بعد العلاج.

نستخلص من هذه الدراسة أن الإصابة بالديدان الداخليه فى الكباش أدى الى حدوث بعض التغيرات العكسية فى صورة المعادن النادرة وصورة البروتين وتغيرات فى تركيز بعض الهرمونات والخصوبة الذكرية ولكن استخدام البندازول بالجرعه العلاجيه ادى الى تخليص الكباش من تلك الديدان وأدى إلى تلافى تلك التأثيرات. ولكن البندازول وجد ان له اثار عكسيه على صورة الحيوانات المنويه. لذلك ينصح بعدم استخدام البندازول فى مزارع الكباش فى فتره التزاوج.