

# Effect of Biotin Supplementation to the Diet of Pregnant Goats on Productive and Reproductive Traits and Performance of their Kids during Suckling Period

Alsaied Alnaimy Habeeb\*  
and Ahmed Elsayed Gad

Department of Biological Applications,  
Radioisotopes Applications Division, Nuclear  
Research Center, Atomic Energy Authority,  
Egypt

## Abstract

Evaluation of the biotin supplementation to the diet of doe pregnant goats on milk yield, blood components, and hormonal levels and weight changes of their kids during the suckling period was the objective of this study. Thirty pregnant Zaraibi goats in the first parity were randomly divided into three similar groups. The experiment was started at two weeks before mating and lasted until the end of the suckling period and weaning their kids. The 1<sup>st</sup> group fed diet without biotin while the 2<sup>nd</sup> and 3<sup>rd</sup> groups, each doe was fed diets with biotin daily at the rate of 5 mg and 10 mg, respectively. Biotin additives to the diet of goats increased significantly doe kidding twins, litter weight of kids, milk yield and dry matter intake and decreased significantly the time of return to estrous postpartum compared to control group. Biotin additives also increased significantly concentrations of blood biochemical components, thyroid hormones, female sex hormones and decreased significantly cortisol level compared to the control group. Live Body Weight (LBW) and Daily Body Gain (DBG) of their suckling kids increased significantly with increasing biotin level in the diets of their mothers. These results suggest that dietary biotin is required at the rate of 10 mg daily for optimal pregnant goats.

**Keywords:** Goats; Biotin; Milk yield; Blood components; Hormones

## Corresponding author:

Alsaied Alnaimy Habeeb

✉ dr\_alnaimy@yahoo.com

Department of Biological Applications,  
Radioisotopes Applications Division, Nuclear  
Research Center, Atomic Energy Authority,  
Egypt.

**Tel:** 002-01014456768

**Fax:** (+202) 0552325966

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## Introduction

Biotin (vitamin B<sub>7</sub>, C<sub>10</sub>H<sub>16</sub>N<sub>2</sub>O<sub>3</sub>S) is a water-soluble B-vitamin and formerly known as vitamin H or coenzyme R [1]. Biotin is present in feedstuffs in both bound and free forms and much of the bound biotin is apparently unavailable to animal species. The primary source of biotin for ruminants is that synthesized by microorganisms in the rumen [2]. Rations high in concentrate reduces the net synthesis of biotin by over 50% due to the acid environment and shift in rumen microbes [3]. Most diets fed to animals contain between 0.1 mg and 0.2 mg of biotin/lb of DMI which equals 4 mg to 10 mg of biotin daily, but net ruminal biotin synthesis was negligible i.e. 1 mg to 5 mg per day and 1 mg to 3 mg per day is absorbed [4]. Biotin is an essential coenzyme in carbohydrate, fat and protein metabolism and is needed for the body to process amino acids, to make glucose from the propionate produced in the rumen (Gluconeogenesis) and to make fat from the acetate produced in the rumen (Lipogenesis) [5]. Biotin is also

required for some of the rumen bacteria, especially, the fiber-digesting bacteria and also needed for the rumen microbes to produce propionate because biotin is a cofactor in the microbial enzyme methylmalonyl-CoA-carboxytransferase, which catalyzes a step in the synthesis of propionic acid [6]. Biotin is required for the normal function of the thyroid and adrenal glands, reproductive and nervous systems. Most researchers reported that milk yield and milk components increase due to biotin supplementation to the diet of dairy cattle [7,8]. Studies of biotin supplementation to the diet of goats are rare. Evaluate biotin supplementation to the diet of doe pregnant goats on milk yield, blood components, and hormonal levels and weight changes of their kids during the suckling period was the objective of this study.

## Materials and Methods

### Location and ethics

The experimental work was carried out in Goat Farm of Biological

Application Department, Radioisotopes Applications Division, Nuclear Research Centre, Atomic Energy Authority, at Inshas, Egypt (latitude 31° 12' N to 22° 2' N, longitude 25° 53' E to 35° 53' E). The ethics contain relevant information on the Endeavour to reduce animal suffering and adherence to best practices in veterinary care according to the International Council for Laboratory Animal Science guidelines. Experimental animals were also cared using husbandry guidelines derived from the Egyptian Atomic Energy Authority standard operating procedures.

### Experimental animals, feeding and management

Thirty Zaraibi pregnant goats aged 18-20 months and weighed  $30.0 \pm 2.5$  kg in the 1<sup>st</sup> parity were randomly divided into three similar groups; ten does in each, were used in this research. The experimental goats were healthy and clinically free of external and internal parasites. Animals were provided the feedstuffs consisted of Concentrating Feed Mixture (CFM) and Clover Hay (CH) according to its physiological status; pregnant and lactation to be adequate in protein and energy to cover their nutrient requirements [9]. The CFM composed of 37.4% wheat bran, 27.0% yellow corn, 12.5% soybean meal (44% crude protein), 10.0% undecorticated cottonseed cake, 5% rice bran, 4% sugarcane molasses, 3% limestone, 1% sodium chloride and 0.1% vitamin and minerals premix. Each kg of CFM contains 0.8% calcium, 0.6% phosphorus, 0.07% magnesium and 0.65% potassium. Each kg of vitamin and minerals premix contains 40 g Mn, 3 g Cu, 0.3 g I, 0.1 g Si, 200 mg Co, 45 g Zn, 30 g Fe, 20 g biotin and 200 mg choline chloride as well as 13500, 4500 and 36 IU vitamins A, D<sub>3</sub>, and E, respectively. Feed CFM was offered once daily at 10 hours at a rate of 3.5% of the body weight of the experimental animals and CH was offered at midday ad libitum and fresh drinking water was available all time. Proximate analysis of the CFM and CH were carried out according to AOAC [10].

Chemical composition values (on DM basis %) of CFM used in feeding the does are 18.00% crude protein, 13.50% crude fiber, 4.80% ether extract, 53.70% nitrogen-free extract and 10.00% ash. The corresponding values for CH were 13.24%, 30.19%, 1.67%, 42.90% and 12.00%. Calculated nutritive values of the CFM and CH according to NRC [9] were 4.00 and 2.64 net energy (mg/kg DM) and 60.82 and 48.00 total digestible nutrients (%) respectively. Each experimental group was housed in the separate yard with a concrete pen, partially asbestos roofed in semi-open sheds during the experimental period. Each yard was divided to 10 partitions using pieces of hardwood, each doe in one partition provided with here feeds and water.

### Experimental procedure

The experiment was conducted on Zaraibi pregnant goats starting from two weeks before mating date (day 23 of August 2016) and lasted during pregnancy period (5 months) and suckling period till weaning their kids (3 months) at end of April 2017. Thirty Zaraibi pregnant goats were divided randomly into three equal groups. The 1<sup>st</sup> group was kept without treatment as control while in the 2<sup>nd</sup> and 3<sup>rd</sup> groups; each doe was supplemented daily with 5 and 10 mg synthesized commercial biotin, respectively. Synthesized commercial biotin was produced as a powder for

Animal Pharmaceuticals CAS No.:58-85-5, Purity: 98.5-99% from Hangzhou Fanda Chemical Co., Ltd., Zhejiang, China.

Biotin was added to the diet of goats of 2<sup>nd</sup> and 3<sup>rd</sup> groups by dilution the powder of biotin in warm water and spraying CFM to be more uniform biotin activity and enhance the mixing and distribution of biotin in CFM. Diets of the 2<sup>nd</sup> and 3<sup>rd</sup> groups were prepared by spraying biotin to the CFM to be satisfied one month. Biotin in CFM fed the three experimental animals contains a basal level of 20 g biotin /kg premix.

### Productive and reproductive traits measurements in does

Productive traits of goats including the type of birth (the number of doe kidding single and twins), litter size and litter weight per experimental group and per doe at birth and monthly were estimated. Litter weight of kids born per group was calculated by multiple litter size with average litter weight at birth. The number of mortality kids per group from birth to weaning was also estimated. Gestation length was determined by the difference between the date of jumping of male on doe and date of kidding. Days from kidding to the day of the first estrous noticed in doe after kidding were estimated.

### Dry matter intake and milk yield determination

Feed intake (FI) from CFM and BH was measured individually for each doe two times, once during the mid-pregnant period and once during the mid-lactation period and averaged to obtain FI for each experimental group at each time. The average DM of CFM and BH were determined from the weight difference before and after oven drying overnight at 105°C. DMI was calculated by multiplying daily fresh feed intake from CFM and BH of each animal with the DM percentage which being 90.0% and 88.0% for CFM and BH, respectively, The total DMI (CFM+CH) of each group was also calculated. Daily milk yield per each doe was recorded at four times; at two days after birth and then monthly during suckling period (3 months) using isolation of all kids out of their mothers at midday until completely hand milking until stripping the udder on midday of the second day.

### Blood samples and estimation of blood components and hormonal levels

Single blood sample with anticoagulant (heparin) was collected on day 10 prior to the day of expected parturition and also on days 2, 30, 60 and 90 postpartum. Blood samples had taken from the jugular vein of each doe before morning meal and placed on ice water immediately following collection. Blood samples were centrifuged at 3000 rpm for 25 minutes to obtain plasma and stored at -20°C until plasma analyses for total protein, albumin,  $\gamma$ -globulin and glucose concentrations using reagent kites manufactured by Diamond Diagnostic Company (Egypt). Globulin value was calculated by subtraction of albumin value from their corresponding total protein value. Plasma levels of total thyroxin (T<sub>4</sub>), Triiodothyronine (T<sub>3</sub>), cortisol, progesterone (P<sub>4</sub>) and estradiol-17 $\beta$  hormones (E<sub>2</sub>) were estimated by the radioimmunoassay (RIA) technique using the coated tubes kits purchased from Diagnostic Products Corporation, Los Angeles,

CA, USA and counting in the Laboratory of Biological Applications Department, Atomic Energy Authority, using computerized Gamma Counter. The tracer in the two hormones was labeled with iodine-125 (<sup>125</sup>I).

### Changes in weight gain of kids

Live Body Weight (LBW) of born kids (kg) for each experimental group either male or female were recorded and weighed at birth and monthly intervals until weaning at 90 days old. Monthly daily body weight gains (DBG) of kids per group from birth to at weaning were also calculated.

### Statistical analysis

Data were statistically analyzed by analysis of variance using the general linear model procedure by the computer program of the SAS software [11]. Duncan's multiple ranges of t-tests were used for testing the significant differences between means [12].

## Results

### Effect of biotin on productive and reproductive efficiency of pregnant goats

Does kidding twins and litter size per doe increase significantly with increasing the level of biotin in the diet of goats? Litter weight of kids at weaning was differing significantly due to biotin, being improved significantly over control by 30.3 kg ( $p < 0.05$ ) and 73.48 kg ( $p < 0.01$ ) in groups received daily 5 mg and 10 mg biotin, respectively. Gestation length was not affected significantly due to adding biotin to the diet of pregnant goats. It is interesting to observe that time of return to estrous in dairy goats postpartum was before weaning by 14 and 25 days in does receive 5 mg and 10 mg daily from biotin, respectively, while the resumption of the estrous cycle it does not receive biotin was after weaning by 7.0 days **Table 1**.

### Effect of biotin on DMI

DMI of CFM and BH was significantly affected by the biotin levels.

Total DMI before and after kidding periods were progressively increased significantly over control (received 0 g biotin) by 166 g/day (11.5%,  $p < 0.05$ ) and 321 g/day (22.3%,  $p < 0.01$ ) it does receive 5 mg and 10 mg daily from biotin respectively **Table 2**.

### Effect of biotin on milk yield

Levels of biotin in the diet of goats affect significantly on overall mean of daily milk yield (g/h). Milk yield in two experimental groups which received daily 5 and 10 mg biotin were higher significantly by 37.21g milk/day (19.7%;  $p < 0.05$ ) and 75.07 g milk/day (39.75%;  $p < 0.01$ ) than milk yield of goats received daily 0 mg biotin **Table 3**.

Days after birth affects significantly on daily milk yield. The highest milk yields in the three experimental groups were at day 30 after birth while the lowest milk yields were at day 90 after birth (at weaning their kids) **Table 3**.

### Effect of biotin on blood proteins and glucose concentrations

Levels of biotin in the diet of goats affect significantly on overall mean of total protein, globulin, and  $\gamma$ -globulin. The improvements were progressively increased significantly with increasing the biotin level in the diet of goats from 5 mg to 10 mg. Biotin at the level of 10 mg/head/day increased significantly total protein, globulin, and  $\gamma$ -globulin by 14.1%, 20.5%, and 33.3%, respectively. Albumin value was not significantly affected by biotin supplementation **Table 4**.

Blood total protein, globulin and  $\gamma$ -globulin also affected significantly due to the timing of blood samples. The highest values were at 30 days after birth while the lower values were at 2 days after birth (**Table 4**). Glucose value increased significantly from 40.9 mg/dl to 47.2 mg/dl (15.4%;  $p < 0.05$ ) in goats received daily 5 mg biotin and to 54.9 mg/dl (34.2%;  $p < 0.01$ ) in goats received daily 10 mg biotin (**Table 4**). These results indicate that biotin addition tended to improve blood metabolic parameters and feeding 10 mg/d of biotin may be the optimal **Table 4**.

**Table 1** Effects of biotin on productive and reproductive traits of pregnant goats.

Productive and reproductive traits of does	Control, 0 mg biotin/daily	5 mg biotin/daily	10 mg biotin/daily
Number of does	10	10	10
Type of birth of kids	7single+3 twins	6 single+4 twins	5 single+5 twins
Doe kidding single, No (%)	7 (70) <sup>a</sup>	6 (60) <sup>b</sup>	5 (50) <sup>c</sup>
Doe kidding twins, No (%)	3 (30) <sup>c</sup>	4 (40) <sup>b</sup>	5 (50) <sup>a</sup>
No. of kids born per group (litter size)	13	14	15
Sex of kids (male=M and female=F)	7F+6M	7 F+7M	8F+7M
No. of kids born per doe	1.3	1.4	1.5
Litter size at weaning	11	12	14
*Litter weight/ group at weaning, kg	122.1	152.4	195.58
Improvement in litter weight/ group		+30.3 kg	+73.48 kg
Significance		$p < 0.05$ .	$p < 0.01$ .
Gestation length, day	151.2 <sup>a</sup> ± 0.75	153.2 <sup>a</sup> ± 0.55	150.1 <sup>a</sup> ± 0.88
Days from kidding to first oestrous	97.0 <sup>a</sup> ± 3.7	76.0 <sup>b</sup> ± 4.2	65 <sup>c</sup> ± 4.3
First estrous in relation to weaning	After 7 days	Before 14 days	Before 25 days

\*Litter size at weaning × kids body weight at weaning. Statistical differences between treatments in the probability of the type of birth was conducted by Chi-square test and significant results were subsequently evaluated using the multiple Z-tests to compare corresponding proportions

**Table 2** Effect of biotin on Dry Matter Intake (DMI) of does during the experimental period.

Dry matter intake (DMI), g/day	Control, 0 mg biotin/daily	5 mg biotin/daily	10 mg biotin/daily
DMI from CFM during pregnancy	1020 <sup>c</sup> ± 10.9	1150 <sup>b</sup> ± 20.4	1270 <sup>a</sup> ± 15.8
DMI from CFM during lactation	1230 <sup>c</sup> ± 14.7	1310 <sup>b</sup> ± 12.8	1390 <sup>a</sup> ± 20.1
Average DMI from CFM	1125 <sup>c</sup> ± 34.2	1230 <sup>b</sup> ± 45.9	1330 <sup>a</sup> ± 30.9
DMI from BH during pregnancy	277 <sup>c</sup> ± 12.8	325 <sup>b</sup> ± 11.8	375 <sup>a</sup> ± 10.7
DMI from BH during lactation	351 <sup>c</sup> ± 20.4	425 <sup>b</sup> ± 18.9	485 <sup>a</sup> ± 18.8
Average DMI from BH	314 <sup>c</sup> ± 22.0	375 <sup>b</sup> ± 24.1	430 <sup>a</sup> ± 30.0
Total DMI	1439 <sup>c</sup> ± 32	1605 <sup>b</sup> ± 34	1760 <sup>a</sup> ± 35
Increase in DMI, g/day		166	321
Increase %		11.5	22.3
Significantly		p<0.05	p<0.01

Different letters in the same raw of each item indicate significant differences (a<b<c<d, p<0.05).

**Table 3** Effect of biotin supplementation on daily milk yield (g/head) of does from birth to weaning their kids.

Time of daily milk yield estimation	Daily milk yield of doe (g/day/head)		
	Control 0 mg biotin/daily	5 mg biotin/daily	10 mg biotin/daily
Number of does	10.0	10.0	10.0
Day 2 after birth	138.50 <sup>b</sup> ± 4.29	213.50 <sup>a</sup> ± 14.19	216.00 <sup>a</sup> ± 13.69
Day 30 after birth	244.00 <sup>a</sup> ± 8.60	265.00 <sup>b</sup> ± 15.45	358.50 <sup>a</sup> ± 5.97
Day 60 after birth	227.50 <sup>c</sup> ± 11.42	256.00 <sup>b</sup> ± 20.27	291.00 <sup>a</sup> ± 12.59
Day 90 after birth	82.50 <sup>c</sup> ± 4.03	93.50 <sup>b</sup> ± 3.50	111.00 <sup>a</sup> ± 9.37
Overall daily milk yield	188.86 <sup>c</sup> ± 29.64	226.07 <sup>b</sup> ± 32.00	263.93 <sup>a</sup> ± 41.32
Increase over control (g/day)		37.21	75.07
Change, %		+19.70	+39.75
Significance		p<0.05	p<0.01

Different letters in the same raw of each item indicate significant differences (a<b<c<d, p<0.05).

**Table 4** Effect of biotin on blood immunity parameters and glucose concentrations in dosage before and after birth their kids.

Experimental groups	Sampling time	Blood biochemical components in does				
		T. protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	γ globulin (g/dl)	Glucose (mg/dl)
Control	Day 10 bb	6.8 <sup>b</sup> ± 0.1	3.2 <sup>b</sup> ± 0.1	3.6 <sup>b</sup> ± 0.1	0.90 <sup>d</sup> ± 0.01	40.5 <sup>ab</sup> ± 1.5
	Day 2 ab	6.4 <sup>c</sup> ± 0.2	3.5 <sup>b</sup> ± 0.1	2.9 <sup>c</sup> ± 0.2	0.89 <sup>d</sup> ± 0.01	38.9 <sup>b</sup> ± 1.8
	Day 30 ab	7.8 <sup>a</sup> ± 0.3	3.9 <sup>a</sup> ± 0.2	3.9 <sup>a</sup> ± 0.2	1.09 <sup>a</sup> ± 0.01	43.5 <sup>a</sup> ± 1.6
	Day 60 ab	6.8 <sup>b</sup> ± 0.3	3.3 <sup>b</sup> ± 0.2	3.5 <sup>b</sup> ± 0.2	0.99 <sup>b</sup> ± 0.01	42.4 <sup>a</sup> ± 2.1
	Day 90 ab	7.0 <sup>b</sup> ± 0.3	3.3 <sup>b</sup> ± 0.2	3.7 <sup>ab</sup> ± 0.2	0.94 <sup>c</sup> ± 0.01	39.6 <sup>ab</sup> ± 1.4
	<b>Overall</b>	<b>6.96<sup>c</sup> ± 0.2</b>	<b>3.44 ± 0.1</b>	<b>3.52<sup>c</sup> ± 0.2</b>	<b>0.96<sup>c</sup> ± 0.04</b>	<b>40.9<sup>c</sup> ± 0.86</b>
	5 mg biotin/daily	Day 10 bb	7.1 <sup>b</sup> ± 0.1	3.3 <sup>b</sup> ± 0.1	3.8 <sup>b</sup> ± 0.1	1.02 <sup>b</sup> ± 0.01
Day 2 ab		6.9 <sup>b</sup> ± 0.1	3.5 <sup>ba</sup> ± 0.1	3.4 <sup>c</sup> ± 0.1	1.02 <sup>b</sup> ± 0.01	44.8 <sup>b</sup> ± 1.9
Day 30 ab		7.9 <sup>a</sup> ± 0.3	3.8 <sup>a</sup> ± 0.1	4.3 <sup>a</sup> ± 0.2	1.25 <sup>a</sup> ± 0.01	51.9 <sup>a</sup> ± 2.1
Day 60 ab		7.8 <sup>a</sup> ± 0.3	3.6 <sup>a</sup> ± 0.1	4.2 <sup>a</sup> ± 0.2	1.15 <sup>ab</sup> ± 0.01	46.8 <sup>ab</sup> ± 2.3
Day 90 ab		7.8 <sup>a</sup> ± 0.3	3.6 <sup>a</sup> ± 0.1	4.2 <sup>a</sup> ± 0.2	1.13 <sup>ab</sup> ± 0.01	44.7 <sup>b</sup> ± 1.4
<b>Overall</b>		<b>7.50<sup>b</sup> ± 0.2</b>	<b>3.56 ± 0.07</b>	<b>3.92<sup>b</sup> ± 0.2</b>	<b>1.11<sup>b</sup> ± 0.02</b>	<b>47.2<sup>a</sup> ± 0.70</b>
10 mg biotin/daily		Day 10 bb	7.9 <sup>b</sup> ± 0.3	3.6 <sup>b</sup> ± 0.1	4.1 <sup>b</sup> ± 0.2	1.20 <sup>c</sup> ± 0.01
	Day 2 ab	7.8 <sup>b</sup> ± 0.1	3.6 <sup>b</sup> ± 0.1	4.2 <sup>ab</sup> ± 0.1	1.28 <sup>b</sup> ± 0.01	50.6 <sup>b</sup> ± 1.8
	Day 30 ab	8.2 <sup>a</sup> ± 0.1	4.0 <sup>a</sup> ± 0.1	4.2 <sup>ab</sup> ± 0.1	1.33 <sup>a</sup> ± 0.01	63.8 <sup>a</sup> ± 2.1
	Day 60 ab	7.9 <sup>b</sup> ± 0.2	3.5 <sup>b</sup> ± 0.1	4.3 <sup>a</sup> ± 0.1	1.31 <sup>a</sup> ± 0.01	57.9 <sup>ab</sup> ± 2.1
	Day 90 ab	7.9 <sup>b</sup> ± 0.3	3.5 <sup>b</sup> ± 0.1	4.4 <sup>a</sup> ± 0.2	1.28 <sup>b</sup> ± 0.01	51.2 <sup>b</sup> ± 1.5
	<b>Overall</b>	<b>7.9<sup>a</sup> ± 0.07</b>	<b>3.64 ± 0.09</b>	<b>4.24<sup>a</sup> ± 0.05</b>	<b>1.28<sup>a</sup> ± 0.02</b>	<b>54.9<sup>a</sup> ± 1.66</b>

Different letters in the same column of each group indicate significant differences (a<b<c<d, p<0.05). Different letters in the same column of overall mean each item indicate significant differences. (A<B<C, p<0.05). bb: before birth; ab: after birth

### Effect of biotin on the hormonal level in female goats

**Metabolic hormones (Thyroid hormones):** Overall means of T<sub>4</sub> and T<sub>3</sub> levels increased significantly from 35.36 mg/ml and 102.04

mg/dl in goats received 0 mg biotin daily to 40.06 mg/ml (13.3%; p<0.05) and 116.98 mg/dl (14.6%; p<0.05) in goats received 5 mg biotin daily and increased again to 46.04 mg/ml (30.2%; p<0.01) and 125.62 mg/dl (23.1%; p<0.01) in goats received 10 mg biotin



**Table 5** Effect of biotin on hormonal levels in experimental does before and after birth their kids.

Experimental groups	Sampling time	Metabolic hormones		Stress hormone	Female sex hormones	
		T <sub>4</sub> (mg/ml)	T <sub>3</sub> (mg/dl)	Cortisol (mg/ml)	P <sub>4</sub> (mg/ml)	Estradiol <sub>17β</sub> (mg/ml)
Control	Day 10 bb	35.5 <sup>ab</sup> ± 0.9	101.0 <sup>ab</sup> ± 1.2	13.5 <sup>a</sup> ± 0.32	9.0 <sup>a</sup> ± 0.2	136.8 <sup>a</sup> ± 6.7
	Day 2 ab	33.8 <sup>b</sup> ± 0.8	98.4 <sup>b</sup> ± 1.1	10.9 <sup>b</sup> ± 0.3	1.8 <sup>e</sup> ± 0.1	55.6 <sup>e</sup> ± 5.7
	Day 30 ab	36.8 <sup>a</sup> ± 1.1	105.6 <sup>a</sup> ± 1.6	10.8 <sup>b</sup> ± 0.4	3.5 <sup>d</sup> ± 0.2	79.9 <sup>d</sup> ± 6.8
	Day 60 ab	36.6 <sup>a</sup> ± 1.2	106.2 <sup>a</sup> ± 2.1	10.1 <sup>b</sup> ± 0.4	4.9 <sup>c</sup> ± 0.2	99.9 <sup>c</sup> ± 8.9
	Day 90 ab	34.1 <sup>ab</sup> ± 1.0	99.0 <sup>b</sup> ± 2.0	10.0 <sup>b</sup> ± 0.3	7.8 <sup>b</sup> ± 0.3	120.8 <sup>b</sup> ± 9.3
	<b>Overall</b>	<b>35.36<sup>a</sup> ± 0.6</b>	<b>102.04<sup>c</sup> ± 1.6</b>	<b>11.1<sup>A</sup> ± 0.6</b>	<b>5.40<sup>c</sup> ± 1</b>	<b>98.6<sup>C</sup> ± 14.4</b>
5 mg biotin/daily	Day 10 bb	39.2 <sup>ab</sup> ± 1.1	112.0 <sup>b</sup> ± 2.0	10.7 <sup>a</sup> ± 0.32	10.9 <sup>a</sup> ± 0.3	155.5 <sup>a</sup> ± 6.6
	Day 2 ab	36.5 <sup>b</sup> ± 1.2	110.8 <sup>b</sup> ± 2.5	7.9 <sup>b</sup> ± 0.31	2.9 <sup>e</sup> ± 0.3	60.7 <sup>e</sup> ± 7.5
	Day 30 ab	42.9 <sup>a</sup> ± 1.0	123.7 <sup>a</sup> ± 2.3	7.8 <sup>b</sup> ± 0.28	4.8 <sup>d</sup> ± 0.2	88.9 <sup>d</sup> ± 8.8
	Day 60 ab	43.9 <sup>a</sup> ± 1.2	124.8 <sup>a</sup> ± 2.6	8.0 <sup>b</sup> ± 0.12	6.8 <sup>c</sup> ± 0.1	110.7 <sup>c</sup> ± 9.3
	Day 90 ab	37.8 <sup>b</sup> ± 1.1	113.6 <sup>b</sup> ± 2.1	7.7 <sup>b</sup> ± 0.26	9.8 <sup>b</sup> ± 0.2	134.8 <sup>b</sup> ± 9.2
	<b>Overall</b>	<b>40.06<sup>a</sup> ± 1.4</b>	<b>116.98<sup>b</sup> ± 1.8</b>	<b>8.42<sup>B</sup> ± 0.6</b>	<b>7.04<sup>B</sup> ± 1</b>	<b>110.1<sup>B</sup> ± 16.7</b>
10 mg biotin/daily	Day 10 bb	45.9 <sup>ab</sup> ± 1.3	123.0 <sup>b</sup> ± 2.3	7.2 <sup>a</sup> ± 0.21	12.8 <sup>a</sup> ± 0.4	172.4 <sup>a</sup> ± 6.3
	Day 2 ab	42.2 <sup>b</sup> ± 1.2	120.8 <sup>b</sup> ± 2.6	5.6 <sup>b</sup> ± 0.32	3.9 <sup>e</sup> ± 0.3	70.6 <sup>e</sup> ± 5.1
	Day 30 ab	48.6 <sup>a</sup> ± 1.3	132.5 <sup>a</sup> ± 3.0	5.5 <sup>b</sup> ± 0.22	5.6 <sup>d</sup> ± 0.2	90.6 <sup>d</sup> ± 5.2
	Day 60 ab	49.9 <sup>a</sup> ± 1.4	130.4 <sup>a</sup> ± 3.2	5.9 <sup>b</sup> ± 0.25	7.9 <sup>c</sup> ± 0.2	120.8 <sup>c</sup> ± 6.2
	Day 90 ab	43.6 <sup>b</sup> ± 1.0	121.4 <sup>b</sup> ± 2.7	5.8 <sup>b</sup> ± 0.23	11.9 <sup>b</sup> ± 0.3	150.6 <sup>b</sup> ± 9.5
	<b>Overall</b>	<b>46.0<sup>A</sup> ± 1.4</b>	<b>125.62<sup>A</sup> ± 1.5</b>	<b>6.00<sup>C</sup> ± 0.4</b>	<b>8.42<sup>A</sup> ± 1</b>	<b>121.0<sup>A</sup> ± 18.7</b>

Different letters in the same column of each group indicate significant differences (a<b<c<d, p<0.05). Different letters in the same column of overall mean each item indicate significant differences (A<B<C, p<0.05). bb: before birth; ab: after birth

**Table 6** Effects of biotin on body weight (kg) of kids and mortality rate in kids during the suckling period.

Change in weight gain of kids and kids mortality during the suckling period	0 mg biotin/daily	5 mg biotin/daily	10 mg biotin/daily
Kids body weight at birth, kg	2.35 <sup>c</sup> ± 0.12 (13)	2.89 <sup>b</sup> ± 0.06 (14)	3.16 <sup>a</sup> ± 0.05 (15)
Kids body weight after one month, kg	5.28 <sup>c</sup> ± 0.31 (12)	6.27 <sup>b</sup> ± 0.41 (14)	6.97 <sup>a</sup> ± 0.61 (14)
Kids body weight after 2 months, kg	8.27 <sup>a</sup> ± 0.40 (11)	9.77 <sup>a</sup> ± 0.42 (13)	10.11 <sup>a</sup> ± 0.53 (14)
Kids body weight after 3 months, kg	11.10 <sup>c</sup> ± 0.38 (11)	12.70 <sup>b</sup> ± 0.60 (12)	13.97 <sup>a</sup> ± 0.91 (14)
Total gain (at weaning-at birth), kg	8.75	9.81	10.81
Average daily gain, g	97.22 <sup>c</sup> ± 2.8	109.00 <sup>b</sup> ± 2.5	120.11 <sup>a</sup> ± 2.4
Increase in daily gain over control		+1.06 kg	+2.06 kg
Change, %		12.11	23.54
Significance		p<0.05	p<0.01
Mortality number of kids per group	2	2	1
Mortality rate, %	15.4 <sup>a</sup>	14.3 <sup>a</sup>	6.7 <sup>b</sup>
**Viability rate, %	84.6 <sup>b</sup>	85.7 <sup>b</sup>	93.3 <sup>a</sup>

Between parentheses are a number of live kids at birth and during the suckling period. Different letters in the same row indicate significant differences (a b<c<d, p<0.05)

No significant differences were found in LBW and DBWG of kids at birth and at weaning due to sex

\*\*Statistical differences between treatments in mortality rate and viability rate were conducted by Chi-square test and significant results were subsequently evaluated using the multiple Z-tests to compare corresponding proportions

daily, respectively. The thyroid hormonal profiles were studied on days -10 prior to kidding and on day +2, +30, +60, +90 days post kidding and found that the lowest thyroid hormonal levels were at day +2 after birth while the highest values were at day +30 and +60 after birth **Table 5**.

**Stress hormone (cortisol hormone):** Overall mean of cortisol level decreased significantly from 11.06 mg/ml in goats received 0 mg biotin daily to 8.42 ng/ml (23.9%; p<0.01) in goats received 5 mg biotin daily and decreased to 6.00 mg/ml (45.8%; p<0.001)

in goats received 10 mg biotin daily. The highest cortisol value was in pregnant goats i.e. at day 10 before birth while after birth, no significant differences in cortisol level due to timing after birth **Table 5**.

**Female sex hormones:** The levels of Progesterone (P<sub>4</sub>) and 17β-estradiol (E<sub>2</sub>) showed great variation. As expected the levels vary according to the stage of the reproductive cycle or presence of pregnancy or parturition. P<sub>4</sub> and E<sub>2</sub> levels recorded the highest values at day 10 before birth and the lowest values on day 2

and in the first month post-partum. The levels of two hormones increased at day 60 after birth and increased again at day 90 after birth **Table 5**. Levels of biotin in the diet of female goats significantly affected the overall means of female sex hormones.  $P_4$  and  $E_2$  levels in goats received 0 mg biotin daily were 5.4 mg/ml and 98.6 mg/ml and increased significantly to 7.04 mg/ml (30.4%;  $p < 0.01$ ) and 110.1 mg/ml (11.7%;  $p < 0.05$ ) in goats receive daily 5 g biotin and increased significantly to 8.42 mg/ml (55.9%;  $p < 0.001$ ) and 121.0 mg/ml (22.7%;  $p < 0.01$ ) in goats received 10 mg biotin daily, respectively **Table 5**.

The interaction between physiological status and biotin treatment effect showed that the highest female sex hormones levels in the three experimental groups were during pregnancy period and the lowest levels were after birth directly **Table 5**.

### Effect of biotin on LBW and DBG of suckling kids from birth to weaning

Adding biotin to the diets of female goats improved significantly positively LBW and DBG of their suckling kids at both birth and during suckling period until weaning **Table 6**.

LBW and DBG of suckling kids increased significantly with increasing biotin level in the diets of their mothers. LBW of suckling kids of mother's intake 5 mg/daily biotin was better than LBW of suckling kids of mother's intake 0 mg/daily biotin by 1.06 kg/90 days and LBW of suckling kids of mother's intake 10 mg/daily biotin was better than LBW of suckling kids of mother's intake 0 mg/daily biotin by 2.06 kg/90 days. No of mortality kids per group from birth to weaning decreased with adding 10 mg/daily biotin to the diet of pregnant goats. Viability rate improved during suckling period till at weaning due to biotin supplementation at the rate of 10 mg daily in the diets of goats **Table 6**.

## Discussion

The increase in litter size and litter weight of kids at weaning in goats due to biotin supplementation in their diet may be that biotin improved DMI from feeding stuffs and consequently improved protein and source of energy intake from feeding stuffs. Biotin also contributes to vital biological functions including synthesis of protein fractions as well as metabolic thyroid hormones. The decrease in time of return to estrous in dairy goats postpartum supplemented with biotin may due to that biotin increasing synthesis of metabolic substrates requirements for lactating goats. The nutritional level is quite important during early lactation because of animal use reserves for milk production which leads to a delay in the resumption of postpartum ovarian activity [13]. The evaluation of estrous behavior showed a negative linear regression between energy intake levels and time of return to estrous in dairy goats postpartum [14].

The higher DMI for biotin treated was attributable to a significantly higher voluntary intake from the feeding stuffs than for control. The increase of DMI may be improved nutrients and digestibility of feeding stuffs as well as fiber digestion [15]. The increase in blood protein parameters and metabolic hormones in goats received biotin may be also responsible for the increase

in milk yield of treated goats. Feeding supplemental biotin increased DMI and milk production significantly in dairy cows [16]. Biotin increased milk production by 1.29 kg per cow per day [17]. In addition, milk production linearly increased with biotin supplementation 36.9 kg, 37.8 kg and 39.7 kg per day for 0 mg, 10 mg and 20 mg per day of supplemental biotin respectively [18]. Biotin increased milk yield and milk components in dairy cows due to increasing the activity of liver pyruvate carboxylase and essential fatty acid metabolism [8]. Supplemental biotin helped cows to produce more glucose from the propionate produced in the rumen and this would increase the production of lactose and drive milk production [19].

Milk yield and milk components increases due to biotin supplementation to the diet of dairy cattle [20]. Cows supplemented with biotin demonstrated improved metabolic status specifically, blood glucose concentration was higher and blood NEFA concentration was lower post calving. The lower NEFA concentration in cows supplemented with biotin were mobilizing less body fat to support their milk production because a liver with less fat is better able to synthesize glucose for milk production and to detoxify ammonia from excess intake of protein [21]. In the same time, biotin is safe for the target animals with a margin of safety that is probably at least 10 times the requirements and use levels. The use of biotin in animal nutrition does not pose a risk to the environment and synthetic biotin is regarded as an effective source of the vitamin in animal nutrition when administered via feed or water for drinking [22].

The increase in total protein, globulin, and  $\gamma$ -globulin may be due to biotin supplementation increased DMI as well as nutrients feed digestibility and stimulate the protein synthesis. The increase in glucose value due to biotin supplementation may be due to biotin increase of the glucose transport to provide the energy required for peptide synthesis.

Biotin is involved in the maintenance of normal blood glucose levels via the gluconeogenic enzymes pyruvate carboxylase and Propionyl-CoA-Carboxylase [23]. Biotin helps the body metabolize proteins and process glucose; therefore it is, especially, important during pregnancy and lactating period [24]. Biotin may be has significant antioxidant properties that protect cells from oxidant damage and consequently increase the immunity function of goats. Similar results obtained in blood metabolites in Holstein cows due to biotin supplementation during peripartum period [25].

The increase in overall mean of  $T_4$  and  $T_3$  levels in goats received biotin may be that biotin stimulates the production of TSH which helps the thyroid gland to produce thyroid hormones. The lowest thyroid hormonal levels were at day +2 after birth while the highest values were at day +30 and +60 after birth. Lower  $T_3$  and  $T_4$  concentrations at day 2 after birth may be to reduce the rate of oxidation and the rate of continuous breakdown and formation of protein and fat in mammary gland tissue or may be to reduce the adverse effects of nutrient deficiency at the onset of lactation. The decrease in the concentrations of thyroid hormones on the +2 day after kidding may be an indication of enhanced utilization of hormones by mammary tissue as a result of the increased metabolism due to the stress of parturition [26]. The elevations in the concentration of thyroid hormones after parturition may

be associated with the metabolic changes in the whole body induced by lactation, which causes an increase in the demand for nutrients, as the kids remained with the dams, causing tactile stimulation of the mammary gland.

Lowering cortisol levels in treated goats as a response to biotin addition in their diet may be due to decrease ACTH hormone. The highest cortisol value was in pregnant goats i.e. at day 10 before birth while after birth, no significant differences in cortisol level due to timing after birth. During the last stage of pregnancy, there is an increased secretion of ACTH from the fetal pituitary

which stimulates rapid growth of the fetal adrenals, leading to a rise in the concentrations of cortisol. Thus an increased amount of plasma cortisol enters the maternal circulation and induces parturition by activating the production of  $\text{PGF}_{2\alpha}$  and coordinating the endocrine profile of the animal. The high level of cortisol during 10 days prepartum is indicative of stress of pregnancy. During parturition, there is an increased secretion of ACTH which stimulates adrenals to increase the level of cortisol.

The increase in overall mean of progesterone ( $\text{P}_4$ ) and estradiol- $17\beta$  ( $\text{E}_2$ ) levels in goats received biotin owing to increased metabolic activity due to high levels of  $\text{T}_3$  and  $\text{T}_4$  hormones.  $\text{P}_4$  and

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