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Thyme, Celery and Salinomycin Implication on Antioxidant Capacity and Neurotransmitters Related to Milk Production in Pregnant Barki Ewes

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Abstract: The experiment goal was the investigation of thyme (T), celery (C) and salinomycin effects on immune response, neurotransmitters related to milk production in Barki ewes. Total 72 mature ewes (2-3 years & 40±1.5 Kg BW) randomly pined equally into five groups. Group-1 was control; groups 2 & 3 received 20g/head/day T and C, respectively. Group-4 received 10g T+ 10g C/head/day, group-5 treated with salinomycin 1g/head/day. Samples collected during 2nd, 3rd trimester of pregnancy and on delivery day (DD); milk yield assessed on 15, 30 and 45-day postpartum. T and/or C and salinomycin increased ($P<0.05$) superoxide dismutase (SOD), reduced glutathione (GSH) and glutathione disulfide (GSSG) during mid-, late-pregnancy and DD compared to control, celery and thyme increase malondialdehyde (MDA) ($p<0.05$) during mid-and late-pregnancy, respectively compared to other groups. Nitric oxide (NO) levels increased in thyme X celery (TxC) group during mid-pregnancy and DD with insignificantly compared with other groups. During mid-pregnancy TxC treatment increased ($p<0.05$) serotonin (5-HT) levels compared with other groups, the same was dopamine (DA), norepinephrine (NE) and tryptophan (Trp) levels ($P>0.05$). During late pregnancy 5-HT, DA, NE & Trp increased ($P<0.05$) in the thyme and/or celery group. While on DD salinomycin increased neurotransmitters ($P<0.05$) with an insignificant increase in other groups. Milk yield increased ($P<0.05$) during 15, 30 and 45 days postpartum in T and/or C groups than control and salinomycin. In conclusion, the applied treatments had a significant effect on reproductive performance, immune response in ewes throughout pregnancy and DD periods, and milk production during the postpartum period.

Keywords: Thyme, Celery, Salinomycin, Immune System, Neurotransmitters, Milk Yield, Ewes

1. Introduction

Pregnancy sustainability and increase milk yield after parturition are vital goals for animal breeders that guarantee the normal and healthy growth of newborn offspring. Milk production is a unique physiologic completion of the reproductive cycle that is regulated by an array of interacting endo- and neurocrine process. prolactin (PRL) is the most hormone required for lactogenesis, which provides a comprehensive signal that fosters synthesis and secretion of milk components [1, 2]. Medicinal plants and ionophores had a wide consideration as feed additives that improve farm animals' antioxidant status and ameliorate physiological stress generated from changes in the reproductive (i.e., pregnancy and/or parturition) or productive (lactation) stage because of their chemical composition and functional groups; such as phenols, thymol, carvacrol, terpenes [3] for thyme; and mono, bi, and sesquiterpenes hydrocarbons [4] for Celery, that gives them the antioxidant, and antimicrobial potentiality [5-7]. Celery supplementation reduced glutathione, catalase,

lipid peroxidation [8], and showed a suppressive effect against malonaldehyde generation [9]. Salinomycin is a fermentation product of various Streptomyces *spp* used on large scale as a growth stimulus as well as animal health and welfare enhancer via the shedding of pathogens [10]. Rare investigations have examined the effect of T, C and salinomycin on reproductive performance and sexual-immune response throughout pregnancy and parturition in sheep, rather than milk production during the postpartum period. The objective here was to examine T, C, and Salinomycin effects on neurotransmitters related to milk production, antioxidant capacity and sexual hormone profile in ewes throughout pregnancy and delivery day, and milk production during the postpartum period.

2. Materials and Methods

The current study was conducted in sheep experimental farm, Nuclear Research Center, Egyptian Atomic Energy Authority. The study was approved by the EAEA Committee, Cairo, Egypt.

2.1. Herbal Plants

Thyme (dried leaves) and seeds of celery were brought from HARAZ herbal store, Cairo, Egypt. Then ground separately and used in the study.

2.2. Experiental Animal Management and Treatments

Total of 72 mature ewes (2-3 years & 40±1.5 Kg mean body weight) were randomly pined equally into 5 groups as in Table 1. The concentrate feed mixture (CFM) used in the feeding trial of ewes consisted of (as a percentage): 24.0 corn, 22.5 sugar beet pulp, 20.0 wheat bran, 30.0 undecorticated cotton seed cake, 1.0 sodium chloride, 0.3 mineral mix, 2.0 dicalcium phosphate, 0.1 AD_3E, and 0.1 sodium bicarbonate. Ewes were fed twice daily at 07:00 and 21:00 hrs. Fresh drinking water was available at all times. The animals received treatments 6weeks before breeding and continued until the end of the experiment.

Table 1. *The experimental design used in feeding the study.*

Feedstuffs program	Control	Treated groups			
	G1	G2	G3	G4	G5
CFM, %	60	60	60	60	60
Berseem %	40	40	40	40	40
Thyme (T)	---	20g/h/d	---	---	---
Celery (C)	---	---	20g/h/d	---	---
Thyme × Celery (TxC)	---	---	---	10×10g/h/d	---
Salinomycin (S)	---	---	---	---	1g/h/d

G1=control, G2=Thyme, G3=Celery, G4=TxC, G5=Salinomycin and CFM=Concentrate feed mixture.

2.3. Blood Sampling, Milk Yield, and Hormones Analysis

Evacuated non-heparinized glass tubes were used to collect blood samples from the jugular vein, then left at room temperature from 30 to 60 min for clotting. Later the tubes centrifuged at 3000 rpm for 15 min to harvest serum. The latest was stocked up on -70°C until analysis. Samples were taken during the 2nd and 3rd trimester of pregnancy as well as on delivery day (DD). Milk yield was recorded for individuals once every two weeks started from the second week until 8 weeks of lactation. Twenty-four hours before hand milking, the lambs were kept away from their dams. Ewes were completely hand milked. Blood E2-17β (pg/ml) & P4 (ng/ml) levels were determined using ELISA kits (Cat. No. MBS734529; MBS269207, respectively).

2.4. Antioxidant's Capacity Estimations

The activities of GSH, GSSG were determined according to the procedure of chromatographic method with standard Sigma-Aldrich Cat. No. 70-18-8, 27025-41-8, respectively [11]. MDA and SOD serum contents were assessed following kits' instruction of Bio-diagnostic® firm (Cairo, Egypt) (Cat. No: SD 2521 & MD 2529, respectively). Nitric oxide was calculated as the sum of nitrite and nitrate by using anion exchange column (Hamilton PRP-X100) using the parameters (150*4.1mm, 10 _m, mobile phase 45:55 of 0.1 M NaCl-methanol); wavelength was adjusted to 230 nm, according to HPLC procedure [12].

2.5. Determination of the Neurotransmitter Concentrations by HPLC Method

The HPLC system consisted of a quaternary pump; a column oven, Rheodine injector and 20µl loop, UV variable wavelength detector. The report and chromatogram were taken from the data acquisition program purchased from Chemstation. The sample was immediately extracted from the trace elements and lipids by the use of solid-phase extraction CHROMABOND column NH2 phase cat. No. 730031. The sample was then injected directly into an AQUA column 150 mm 5µ C18, purchased from Phenomenex, USA under the following conditions: mobile phase 20mM potassium phosphate, pH 2.5, flow rate 1.5ml/min, UV 190 nm. Noradrenalin, dopamine, and serotonin were separated after 12 minutes .The resulting chromatogram identified each monoamine position and concentration from the sample as compared to that of the standard, and finally, the determination of the content of each monoamine as µg per mL [13].

2.6. Statistical Analysis

Data were expressed as mean±SE. Statistical analysis of

the obtained data was performed using the general linear model (GLM) using SAS software [14]. Significant differences among means were evaluated using Duncan's Multiple Range Test.

3. Results

C, T and TxC treatments increased (P<0.01) SOD activity more than control during the second and third trimester of pregnancy and DD with the highest values of 223.6, 215.0 & 210 U/mL, respectively through the 3rd trimester. On DD, all treatments revealed the lowest (P<0.01) SOD mean values compared with the pregnancy period. Throughout mid and late pregnancy applied treatments did not affect (P<0.01) MDA content than control, except celery (21.2 nmol/mL) at mid-pregnancy, Thyme (16.82 2 nmol/mL) and Salinomycin (16.732 nmol/mL) during late pregnancy. On DD, MDA was at the highest values either for treatments or control group in comparison with mid and late pregnancy period, the highest mean value was about 26.8 2 nmol/mL for Thyme (Table 2). TxC treatment showed the best enhancement in GSH activity (2.49 µmol/mL) followed by Thyme (2.17 µmol/mL) during mid-pregnancy, then Celery (2.34 µmol/mL), thyme and TxC during late pregnancy. The noticeable (P<0.01) increase in GSH at parturition was due to Thyme (Table 4). Salinomycin did not affect GSH other than control except an increase (1.948 µmol/mL; P<0.01) during mid-pregnancy. GSSG increased (P<0.01) due to C, TxC treatments during mid-pregnancy while at late pregnancy and DD salinomycin recorded the highest GSSG mean value 0.353 µmol/mL. Celery caused a marked (P<0.01) decrease in GSSG level at late pregnancy and parturition. There are no significant changes in NO levels due to treatments during mid-pregnancy, but through late pregnancy, all treatments lowered (P<0.01) NO less than control. Celery markedly decreased NO (0.29 µmol/mL; P<0.01) at parturition whereas TxC showed an opposite trend (Table 2).

In general, estimated neurotransmitters (5-HT, DA, NE & Trp) gradually increased (P<0.01) for more than 3 folds from mid-pregnancy to late-pregnancy trimester then significantly declined about 6 folds at parturition which recorded the lowest (P<0.01) mean values for all parameters in each group (Figure 1). During mid-pregnancy only 5-HT that significantly affected by treatments due to TxC supplementation with a mean value of about 1.27 *vs* 1.08 µg/mL for control. Celery and Thyme groups had the highest (P<0.01) levels of 5-HT, DA, NE & Trp throughout late pregnancy as compared to control. Salinomycin recorded an increase (P<0.01) in 5-HT, DA, NE & Trp concentrations at parturition, without any significant changes except these (Table 3).

It noticeable that E2 levels decreased (P<0.01) to 6.912 pg/ml at mid-pregnancy than late-pregnancy (9.486 pg/ml) then continued to rise to reach 11.565 Pg/ml on DD. whereas P4 concentrations showed an opposite trend through the previous stages, recoded the lowest overall mean value of 0.542 ng/mL; P<0.01 at parturition (Figure 2). T, C and TxC groups were higher (P<0.01) in E2 levels than control at late-pregnancy and DD, the highest E2 mean value was 12.38 Pg/mL for T treatment on DD. During late pregnancy, only TxC group recorded an increase (P<0.01) on serum P4 than control, while at mid-pregnancy P4 levels increased (P<0.01) than control due to treatments (except Salinomycin) with the highest mean value 4.11 ng/mL for Celery. No significant changes in P4 levels than control on DD (Table 4).

As shown in Table 5, ewes' supplementation with thyme and/or celery showed a significant increase (P<0.001) in total milk yield than control and salinomycin. The addition of 20g/h/d thyme to the basal diet increased (P<0.01) milk yield from 566 to 757g in comparison to control. The highest daily milk yield was on day-15 postpartum due to thyme and/or celery treatments, however, the salinomycin group recorded its own on day-30 PP with 712 versus 549 g/d; P<0.007 for control (Table 5).

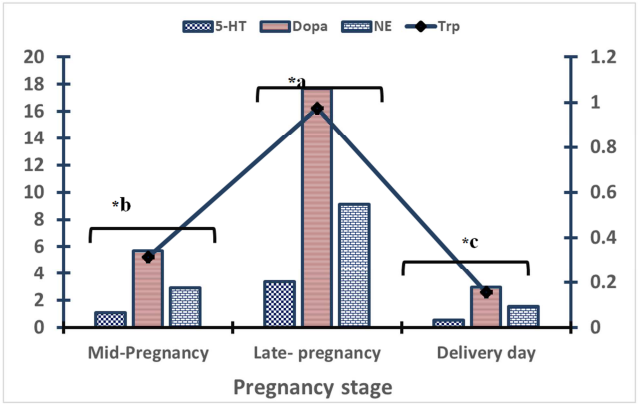

Figure 1. *Serotonin (5-HT), Dopamine (Dopa), Nor Epinephrine (NE) and Tryptophan (Trp) profile throughout mid-, Late pregnancy and Delivery day of Barki ewes in response to thyme, celery and salinomycin supplantation.*

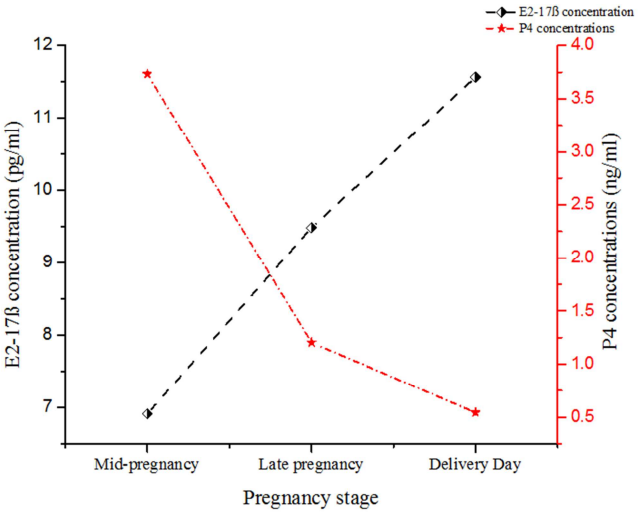

Figure 2. *The pattern of Estradiol-17β (pg/mL) and progesterone (ng/mL) of Barki ewes during mid-, late pregnancy and delivery day as affected by Thyme, Celery and Salynomicyn supplementations.*

Table 2. *Effect of thyme, celery and salinomycin on antioxidant capacity of pregnant Barki ewes.*

Parameters	Mid-pregnancy				
	Ctrl	**T**	**C**	**TxC**	**S**
SOD	161.3±1.6g	212.4±2.5bc	189.9±5.1d	202.7±1.4c	176.7±1.9ef
MDA	14.93±0.4f	14.83±0.2f	21.20±0.9bc	14.13±0.2f	14.60±0.4f
GSH	1.73±0.05f	2.17±0.04c	1.83±0.02def	2.49±0.03a	1.948±0.03d
GSSG	0.24±0.01fg	0.251±0.01fg	0.313±0.01bc	0.263±0.01ef	0.255±0.01efg
NO	0.503±0.03cd	0.447±0.03d	0.518±0.01bcd	0.573±0.01abc	0.498±0.02cd

Parameters	Late-pregnancy				
	Ctrl	**T**	**C**	**TxC**	**S**
SOD	173.1±0.2f	215.0±1.3ab	223.6±0.7a	210.8±4.7bc	183.3±1.1de
MDA	15.11±0.3f	16.83±0.2e	15.21±0.3f	14.95±0.4f	16.73±0.3e
GSH	1.91±0.1de	2.28±0.1cd	2.34±0.1b	2.21±0.1bc	1.78±0.02ef
GSSG	0.23±0.01g	0.235±0.01fg	0.200±0.01h	0.258±0.01ef	0.300±0.01bcd
NO	0.605±0.01a	0.445±0.02d	0.565±0.01abc	0.555±0.04abc	0.537±0.01abc

Parameters	Delivery Day					MSE	*P*-Value	
	Ctrl	**T**	**C**	**TxC**	**S**		**Stg**	**Trt.**
SOD	133.0±6.7i	161.0±4.2g	171.8±0.6f	148.8±5.7h	125.7±2.7i	10895.851	<0.0001	<0.0001
MDA	18.27±0.2d	26.80±1.0a	20.19±0.5c	21.63±0.6bc	21.97±0.7b	167.477	<0.0001	<0.0001
GSH	1.43±0.1g	2.14±0.04c	1.94±0.04d	1.91±0.1de	1.57±0.04g	1.028	<0.0001	<0.0001
GSSG	0.325±0.01b	0.28±0.01de	0.198±0.01h	0.29±0.01cd	0.353±0.01a	0.023	<0.0001	<0.0001
NO	0.523±0.03abcd	0.500±0.04cd	0.29±0.01e	0.595±0.05ab	0.545±0.04abc	0.072	0.0088	<0.0001

a,b,c… means within column with different superscript are significantly different at P<0.05.
Ctrl Control, T Thyme, C Celery, TxC thyme x celery, S salinomycin.
Stg pregnancy stage (mid- late pregnancy & delivery day), Trt treatment.
SOD superoxide dismutase, MDA malondialdehyde, GSH redused glutathione, GSSG glutathione disulfide, NO nitric oxide

Table 3. *Effect of thyme, celery and salinomycin on neurotransmitters related to milk production of pregnant Barki ewes.*

Parameters	Mid-pregnancy				
	Ctrl	**T**	**C**	**TxC**	**S**
5-HT	1.08±0.07e	1.115±0.08de	1.07±0.09e	1.27±0.02d	0.948±0.06ef
DA	5.74±0.35de	5.61±0.35de	5.675±0.52de	6.518±0.12d	4.885±0.30ef
NE	2.90±0.17ef	3.028±0.21e	2.883±0.25ef	3.345±0.09e	2.525±0.16fg
Trp	0.308±0.02ef	0.318±0.02ef	0.328±0.03e	0.353±0.01e	0.265±0.02fg

Parameters	Late-pregnancy				
	Ctrl	**T**	**C**	**TxC**	**S**
5-HT	3.188±0.04c	3.42±0.06 b	3.65±0.02a	3.568±0.06 ab	3.120±0.02c
DA	16.23±0.16c	17.90±0.23b	19.47±0.15a	19.06±0.31a	15.68±0.24c
NE	8.71±0.06 c	9.258±0.21b	10.105±0.09a	9.535±0.10b	8.043±0.05d
Trp	0.93±0.02 c	0.998±0.02b	1.07±0.01a	1.018±0.01b	0.860±0.01d

Parameters	Delivery Day					MSE	*P*-Value	
	Ctrl	**T**	**C**	**TxC**	**S**		**Stg**	**Trt.**
5-HT	0.455±0.02g	0.528±0.02g	0.465±0.02g	0.49±0.02g	0.81±0.15f	19.758	<0.0001	0.0007
DA	2.373±0.10g	2.763±0.10g	2.94±0.03 g	2.53±0.12 g	4.308±0.78f	537.070	<0.0001	<0.0001
NE	1.205±0.06h	1.443±0.06h	1.50±0.01h	1.33±0.06h	2.17±0.40 g	143.075	<0.0001	<0.0001
Trp	0.13±0.01hi	0.158±0.01h	0.095±0.01i	0.145±0.01hi	0.243±0.05g	1.660	<0.0001	0.0012

a,b,c… means within column with different superscript are significantly different at P<0.05.
Ctrl Control, T Thyme, C Celery, TxC thyme x celery, S salinomycin.
Stg pregnancy stage (mid- late pregnancy & delivery day), Trt treatment.
5-HT 5-hydroxytraptamine (serotonin), DA Dopamine, NE Norepinephrine, Trp Tryptophan.

Table 4. *Effect of thyme, celery and salinomycin on estradiol-17β and progesterone levels of pregnant Barki ewes.*

Parameters	Mid-pregnancy				
	Ctrl	**T**	**C**	**TxC**	**S**
E2	6.9±0.04i	7.483±0.06h	7.083±0.04hi	7.168±0.14hi	6.00±0.14j
PROG	3.518±0.04c	3.798±0.05b	4.115±0.02a	3.923±0.05b	3.310±0.03d

Parameters	Late-pregnancy				
	Ctrl	T	C	TxC	S
E2	8.768±0.11f	9.908±0.20e	10.893±0.27cd	9.525±0.28e	8.335±0.15g
PROG	1.183±0.07f	1.22±0.08ef	1.24±0.11ef	1.355±0.02e	1.025±0.07g

Parameters	Delivery Day					MSE	*P*-Value	
	Ctrl	T	C	TxC	S		Stg	Trt.
E2	11.19±0.07c	12.38±0.17a	11.89±0.01b	11.715±0.14b	10.648±0.14d	52.697	<.0001	<0.0001
PROG	0.500±0.02h	0.593±0.02±0.01h	0.587±0.01h	0.54±0.02h	0.49±0.01h	24.709	<0.0001	<0.0001

a,b,c… means within column with different superscript are significantly different at P<0.05.
Ctrl Control, T Thyme, C Celery, TxC thyme x celery, S salinomycin, Stg pregnancy stage (mid- late pregnancy & delivery day), Trt treatment, E2 estradiol-17β, PROG progesterone.

Table 5. *Effect of thyme, celery and salinomycin feeding on milk yield of Barki ewes.*

Milk yield	Ctrl	T	C	TxC	S	MSE	*P-Value*
15-day PP g^{-d}	$601±42^{c}$	$826±40^{a}$	$787±56^{ab}$	$822±65^{a}$	$670±45^{bc}$	149075	0.006
30-day PP g^{-d}	$550±41^{b}$	$769±41^{a}$	$713±45^{a}$	$706±51^{a}$	$712±31^{a}$	99993.3	0.007
45-day PP g^{-d}	$549±27^{bc}$	$677±32^{a}$	$612±30^{ab}$	$591±28^{ab}$	$501±36^{c}$	61873.4	0.002
Total Milk Yield g/d	$566±21^{b}$	$757±23^{a}$	$704±28^{a}$	$707±32^{a}$	$628±26^{b}$	2588	0.001

a,b,c… means within column with different superscript are significantly different at P<0.05.
Ctrl Control, T Thyme, C Celery, TxC thyme x celery, S salinomycin, PP postpartum.

4. Discussion

Many studies have shown that active herbal ingredients have a strong antioxidative effect due to scavenging of free radicals or by increasing the production of CAT, SOD and GPx as essential parts of the antioxidant defence system in cells [15]. In concomitance of the current results, administration of thyme and/or celery throughout ewes' estrous cycle enhanced immune response via increasing serum GSH, GSSG, SOD with a decrement in MDA and NO concentrations [16]. It stated that the phenolic OH group in thymol is responsible for its antioxidant properties, whereby it serves as a hydrogen donor and can neutralize the peroxyl radicals which are produced during the initial step of lipid peroxidation [17].

Prolactin is a peptide hormone primarily synthesized and secreted by adenohypophysis, then secreted into the bloodstream in response to an appropriate stimulus like suckling or milking process. The mechanisms that control PRL secretion range from the direct stimulation of the adenohypophysis to the suppression of the hypothalamic secretion of PRL inhibitory factor (PIF) and stimulation of the hypothalamus to secrete PRL releasing hormone neurons [18]. Prolactin secretion is normally tightly regulated by a short - loop negative - feedback mechanism, whereby prolactin stimulates the activity of tuberoinfundibular dopamine (TIDA) neurons to increase dopamine secretion into the pituitary portal blood. Dopamine (DA) is a neurotransmitter functions as a PRL-inhibiting factor (PIF) in the hypothalamic-hypophysial system [19]. During early and mid-pregnancy, there is a tight association between TIDA neuronal activity and prolactin secretion. However, late pregnancy is different because TIDA neuronal activity reduced during periods of low prolactin secretion before the antepartum prolactin surge, as well as during the antepartum prolactin surge [20]. During late pregnancy, plasma prolactin levels remain low until a large prolactin surge occurs during the dark period immediately preceding parturition [21] which declare and support the current results that showed a significant increase in DA during late pregnancy of Barki ewes, especially in thyme and/or celery treatment groups which were higher in DA than control, followed by a decline (P<0.01) on delivery day. In sheep, Eliot [22] recorded a significant increase in DA concentrations only during the last 30 min in spontaneous labor ewes. Highly increase in prolactin levels noticed during late pregnancy and early lactation period [23]. Dopamine converts into epinephrine and then the epinephrine converts into norepinephrine by the cortisol-dependent enzyme. Any form of stress that increases the cortisol level (such as near of delivery day) stimulates epinephrine production [24], which may be an explanation of DA and NE increases in the current study.

With the vast literature implicating dopamine in the control of prolactin secretion, some studies suggest that serotonin is a neurotransmitter involved in the stimulation of PRL release. Serotonin administration induced prolactin release by either injecting into the third ventricle [25], or systemic administration [26]. Furthermore, in rats, serotonin precursor, 5hydroxy-tryptophan (5-HTP), induced prolactin release [27]. On the other side, the current results show a 5-HT decline (P<0.01) from late pregnancy trimester to after parturition, in accordance to Moore [28] who found a 57.9% and 29.5% decline in circulating 5-HT occurred from peak prepartum (d -3&-1) to 5-HT nadir postpartum in Jersey and Holstein multiparous cows, respectively. From another point of view, throughout late pregnancy and the rapid growth of fetus that press on rumen results a decrease in feed intake and changes animal appetite which may lead to serotonin (mood and appetite regulator) increment.

Tryptophan (Trp) amino acid, is very important in pregnancy, because of the increased demand for maternal protein synthesis and fetal requirements for growth and

development, therefore, it is physiologically unfavorable if maternal Trp is depleted through pregnancy. Also, Trp is a precursor of many important metabolites such as serotonin (5-HT) [29]. Data of Tsuji [30] pointed to the increased requirement of the fetus for Trp. In addition to its essential role in protein synthesis and hence fetal growth, Trp requirements extend to the importance of its metabolites in pregnancy. On day 20 (Late-pregnancy), free Trp was hugely elevated (by 122%), presumably because of the continued decrease in albumin (21%) and, more importantly, the 76% increase in NEFA. The 21% decrease in total Trp on day 20 can be explained by increased tissue uptake of free Trp and the rapid equilibration between the free and albumin-bound fractions [31]. In general, thyme essential oil (carvacrol) is a brain-active molecule that influences neuronal activity through the modulation of neurotransmitters action [32].

Our results displayed an elevation ($P<0.05$) in E2 (during late-pregnancy and DD) and P4 (during mid-pregnancy) levels, also salinomycin treatment (during late pregnancy); which acts through inhibiting enzymes that metabolize steroid hormones and therefore increase their levels in serum. The earlier studies reported that Thyme and Celery are a source of phytoestrogen compounds, that show estrogenic activity via estrogen receptors and may affect estrogen synthesis and metabolism [33-35]. In addition, Apigenin is a flavonoid in C, T, and other herbs, which may mimic estrogen [36]. The essentiality of Progesterone lies in the success and maintenance of pregnancy. These sexual hormones' herbal mimics may bind to the respective receptors for the endogenous hormones in the target cells of the brain and reproductive tract and induce or inhibit biological responses similar to the sex hormones [33].

In the current study, thyme and/or celery increased ($P<0.05$) total and daily milk yield than control, thyme increased total milk yield by 33.8%, while celery and TxC were 25% higher than control. The high milk production is mediated by Trp, and 5-HT increases throughout the late gestation period since increasing Trp with 0.12%an increaserevealed ($P<0.05$) in milk yield by 16.5% [37].

5. Conlusion

Thyme and/or celery and salinomycin supplementations significantly affect antioxidant capacity, neurotransmitters, and steroid hormones profile related to milk production which in turn enhances reproductive performance in Barki ewes throughout pregnancy and parturition periods, which lead to increase milk yield during the postpartum period.

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