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SERUM BIOCHEMICAL PROFILE OF RABBIT DOES OF ALGERIAN LOCAL POPULATION (AURES REGION) AT DIFFERENT PHYSIOLOGICAL STAGE

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ABSTRACT

One hundred and twenty local Algerian female rabbits in first parity were used in this present study at the experiment farm of the University of Batna 1. The animals were divided into four equal groups with 30 replicates (empty female, pregnant female, pregnant-lactating female and lactating female only). Natural mating was used, the kits were weaned at 28 days. Four blood samples were collected on females at different physiological stages. The physiological status affected significantly ($p < 0.05$) cumulative BCS, milk yield, litter size and litter weight of does at birthing. Total protein, albumin, globulin, uric acid and creatinine concentrations as well as T3 and progesterone levels were higher ($p < 0.05$) in does of P-L group (5.70 ± 1 g/dl, 2.98 ± 0.07 g/dl, 1.88 ± 0.06 g/dl, 34.9 ± 3.4 mg/dl, 1.3 ± 0.4 mg/dl, 44.3 ± 0.93 ng/dl, 18.98 ± 1.5 ng/ml) respectively. The physiological status increased significantly ($p < 0.05$) total lipids, triglycerides and total cholesterol in does of P, P-L and L groups compared with their concentrations in E group. The physiological status affected alanine aminotransferase (ALT), aspartate aminotransferase (AST) and Alkaline phosphatase activities in does during pregnancy and suckling period (5.65 ± 0.1 UI/L, 22 ± 1.3 UI/L vs 5.45 ± 0.22 , 20.35 ± 4.3 UI/L). Does of P, P-L, and L groups had higher ($p < 0.05$) leptin levels. Insulin and glucose concentrations were increased in P and P-L groups compared to E and L groups.

Key words: Algerian population, Aures area, Rabbit, blood component, physiological status, reproductive performance.

INTRODUCTION

In recent years, several works have examined the reproductive traits of Algerian local population does (Moumen 2017) but no standardization study has been done on the plasmatic parameters of the Algerian population of rabbits reared in the Aures region during their different physiological stages. The body condition and energy balance of female rabbits appears to be correlated to short and long-term reproductive efficiency. Metabolites concentrations and fat mobilization also affect the fertility rate (Brecchia et al., 2008). The nutritional status can also be apprehended with the help of blood markers. The digestive and metabolic adaptations in response to under-nutrition has been described in many species including ruminants (Belkacem L et al., 2018). Several researches, studying essentially laboratory rodents, have shown that certain energetic nutrients (fatty acids, glucose) are capable by themselves to modify the key genetic expression of the metabolism. The objective of this article is to examine some blood parameters at different physiological stages as specific markers of the body status.

MATERIALS AND METHODS

Animals and experimental design

The trial was carried out at the experimental rabbit farm of the Veterinary Department of the University Batna 1. 120 nulliparous does of local Algerian population (Aures area) were submitted to natural mating. Does were first mated at the age of 16.5 weeks with an average body weight of 3107.1

± 53.5g and with body condition scoring (BCS) of loin and rump ranging from 2 to 3 on a scale of 1 – 5 following a 42-day rhythm. All does were kept in single cage made of galvanized wire. Sexual receptivity was evaluated before mating and does with red and turgescient vulva considered receptive. A pregnancy diagnosis by abdominal palpation was performed 10 days after the mating. A commercial pellet and water from nipple drinkers were provided *ad libitum*, the diet formulation and analytical data are presented in table 1. The environment temperature and relative humidity were collected (ranges: 15-20°C and 60-75%, respectively) and light schedule of 16L/8D was used. The building was artificially ventilated (0.3m³/s).

Table 1: Chemical composition of diet

Nutrients (%)	
Fat (%)	4.2
Starch (%)	14.0
Crude protein (%)	17.2
Crude fiber (%)	16.5
DE (kcal/kg)	2440

Blood samples and estimation of biochemical components

Four blood samples were collected from all females at: 08:00h, at different physiological stages (empty female, pregnant female, pregnant-lactating female and lactating female only) from the marginal ear vein, drawn into vacutainer tubes containing 0.85 U.I of Heparin, immediately centrifuged at 2000 x g for 10 mn (SIGMA-2K15 centrifuge) and plasma stored frozen until assayed for hormones and metabolites. Serum total proteins, albumin, total lipids, triglyceride and total cholesterol concentrations as well as alanine aminotransferase (ALT), aspartate aminotransferase (AST) and Alkaline phosphatase activities were determined *colorimetrically* using commercial kits produced by Stanbio Company, USA by computerized spectrophotometer model Milton Roy 1201. Serum Globulin values were calculated by subtracting albumin values from their corresponding total proteins values of the same sample. Insulin, leptin, total triiodothyronine (T3) and progesterone (P4) hormones were determined using radioimmunoassay (RIA), at the research laboratory of Pharmaceutical Mineral Chemistry.

Statistical analysis

The statistical analysis was performed with MedCalc version 15.2.1 according to the following model: $Y_{ij} = \mu \pm T_i \pm e_{ij}$ where μ = the overall mean, T_i = the fixed effect of treatments, (1 = empty female, 2 = pregnant female, 3 = pregnant-lactating female, 4 = lactating female), e_{ij} = residual error. Least significant difference was used to compare between means.

RESULTS AND DISCUSSION

During pregnancy and suckling period, females had similar body weight but milk yield differs significantly ($p < 0.05$) between the two experimental groups (P-L and L). Litter size and litter weight of does at birthing was higher ($p < 0.01$) in L group than in P and P-L groups. The improvement in does weight during pregnancy and suckling period may be due to the increased appetite of rabbits and consequently increased feed intake and good absorption in the intestinal tract. Kids born of rabbits does of P and L groups weighed more and were of greater length than from does of P-L group. The difference between doe milk yields between experimental groups may be due to differences in litter size.

The increase in litter size and consequently litter weight of does at birthing may be due to the high number of ovulation and fertility in female rabbits of L group (Rebollar et al., 2014) (Table 2). Blood glucose values are again very low in pregnant-lactating female rabbits since the mammary gland is also a sensor for the synthesis of milk lipids. According to the obtained results, we see that the females started with an average glycemia (0.22 g/l). The latter increased during the second and third week of pregnancy (0.65 g/l) then diminished until parturition. Our results remain below the values of the European rabbits and the rabbits reared in tropical areas (0.79 g/l). The lowest level of protein was recorded in the second week of pregnancy ($p < 0.05$), a slight increase was then noticed however, it ended by decreasing at the end of pregnancy. The increase in total protein, albumin and globulin

concentrations in does of L group may be due to increase in feed intake and good absorption in the intestinal tract.

Table 2: Reproductive performances of rabbit does at different physiological stages (n=30/group)

Parameters	Physiological status				P
	E	P	P-L	L	
Initial BW (g)	3000±37	3053±47	3188.4±63	3187±67	ns
Cumulative BCS	3.00	2.2	1.96	2.56	*
BW at kindling (g)	-	2839.3±70	2703.3±68	3050±57	*
Average MY during suckling period	-	-	2200.7±36	2913.4±105	*
LS at birth	-	6.9±2.3	7.4±2.37	9±1	**
LW at birth (g)	-	390.5±13.6	402±11	501±9	*
LS at weaning	-	5.2±2.3	5±1.2	8±2	**
LW at weaning (g)	-	2040±67	2200±25	3720±39	*

*BW: body weight, BCS: body condition score, MY: milk yield, **P<0.01; *P<0.05.*

Immunoglobulin is the main component of antibodies, and increase in the immunoglobulin level indicates a good immune status of the animal (Ballou et al., 2009). Concerning total lipids, there was a strong mobilization of body fat in pregnant females (97 mg/dl ± 0.10). This significantly high lipemia (P<0.05) can be explained by the energy needs that were more significant during pregnancy and lactation. Regarding cholesterolemia, a highly significant increase (p<0.05) was observed in -pregnant females. However, a marked decrease (p<0.05) was recorded in cholesterolemia in lactating females, same changes were observed in other species and in rabbits. Regarding triglyceridemia, we have recorded a significant raise (37.7 ± 35.6; p<0.001) in pregnant females. Triglycerides were the constituents of lipoproteins. Hydrolysis by lipoprotein-lipase of triglycerides circulating in free fatty acids captured by underlying tissues, particularly the muscle, was considered as a limiting step for the use of triglycerides for energy purposes. The increase in total lipids, triglycerides and total cholesterol concentrations in does of P group may be due to the mobilization of body reserves to the needs of fetal growth and hormone synthesis (El Moghazi et al., 2014). The decrease in ALT activity in does of all experimental groups may be due to a decrease in gluconeogenesis process in the liver. The increase in alkaline phosphatase in animals during suckling period may be due to an increase in milk biosynthesis (Salman, 2017). Creatinine and urea are two small molecules eliminated by the kidney of mammals. If creatinine is relatively constant in an individual but varies across breeds according to the muscular mass. Uremia, however, can vary in function of extra-renal factors (protein intake and liver functioning). The creatinine value recorded was close to the physiological norms 1.3-2.7 mg/dl vs 1.2-0.72 mg/dl (Emma K et al., 2016). The T3 plasma levels were lower (p<0.05) in L group. Brecchia et al., 2008 found that T3 blood concentrations decreased during fasting. Thus, the thyroid hormone concentration clearly reflects the energetic balance of the doe, and food deficiency reduced thyroid function, so that the animals could spare energy by decreasing adaptive thermogenesis. Thyroid hormones, which are mainly implicated in the regulation of tissue growth and metabolism, are influenced by many factors, including nutrition this is supported by the finding of Vanderpas (2006) who concluded that the increase in basal metabolic rate is accompanied by increased appetite and subsequently increased body weight. Insulin rate, increases greatly at the end of pregnancy. Since there is evidence of active transfer of both insulin and leptin into the brain, these hormones could have a role in signaling the metabolism state of the animal (Brecchia et al., 2008). Pregnant and lactating does showed an increase (P<0.05) of leptin level. Leptin receptors were detected in different structure of rabbits, including follicles at different stage of development, and oviducts, suggesting that leptin may regulate steroidogenesis of pre- and post-ovulatory follicles as well as fertilization and early embryonic development by providing a favorable local environment to gamete (sperm and oocyte) transport, sperm capacitation and oocyte maturation. The higher (p<0.05) progesterone levels in P and P-L groups than its concentrations in E and L groups may be attributed to maintaining pregnancy (Table 3).

Table 3: Effect of the physiological stage on metabolic profile and hormones in doe rabbits

Parameters	Physiological status			
	E(n=5)	P(n=5)	P-L(n=5)	L(n=5)
Total proteins (g/dl)	3.60 ^c ±0.5	4 ^b ±0.90	5.70 ^a ±1	6.10 ^a ±0.1
Albumin (g/dl)	2.01 ^c ±0.1	2.81 ^b ±0.05	2.98 ^a ±0.07	3.50 ^a ±0.10
Globulin (g/dl)	1.22 ^c ±0.02	1.69 ^b ±0.01	1.88 ^a ±0.06	2.80 ^a ±0.04
Total lipids (mg/dl)	31 ^d ±1	97 ^c ±0.10	69 ^b ±0.2	78.01 ^a ±0.6
Triglycerides (mg/dl)	15.4 ^d ±5.8	65.80 ^c ±23	44.7 ^b ±17.6	37.7 ^a ±25.6
Cholesterol (mg/dl)	28.7 ^d ±16.7	70 ^c ±18.2	58.3 ^b ±24.1	38.8 ^a ±23.9
ALT (IU/L)	4.98 ^b ±0.2	5.65 ^a ±0.1	5.21 ^a ±0.3	5.45 ^a ±0.22
AST (IU/L)	18.34 ^a ±0.4	22 ^a ±1.3	21.4 ^a ±1.14	20.35 ^a ±4.3
Alk-phosphatase (IU/L)	12.8 ^b ±0.85	40.50 ^a ±8.9	39.8 ^a ±0.53	38.2 ^a ±0.66
Uric acid (mg/dl)	7.9 ^a ±1.4	21.2 ^c ±3.8	34.8 ^b ±3.4	24 ^a ±3.3
Creatinine (mg/dl)	0.72 ^b ±1.1	1.4 ^a ±0.4	1.3 ^a ±0.4	1.2 ^a ±0.8
T3 (ng/dl)	22.4 ^c ±0.53	31.6 ^a ±0.6	44.3 ^b ±0.93	29.2 ^a ±0.6
Progesterone (ng/ml)	0.35 ^c ±4.2	20 ^b ±0.2	18.98 ^b ±1.5	10.7 ^a ±0.15
Leptin (ng/ml)	1.5 ^a ±0.1	2.50 ^b ±0.2	1.7 ^a ±.3	2.02 ^b ±0.9
Insulin (µg/ml)	22.2 ^a ±1.2	45.5 ^b ±2	41.08 ^b ±3.2	28 ^c ±1.02
Glucose (g/l)	0.22 ^c ±0.04	0.65 ^b ±0.1	0.35 ^a ±0.2	0.4 ^a ±0.1

a, b, c, d, Means in the same row within each physiological status having different.

CONCLUSION

The lactating and pregnant females presented a strong mobilization of body fat, which can explain the energy needs that are more significant during pregnancy and lactation. It is also evident that excessive fatness of empty does should be controlled by reducing feed intake during the dry period. Both BCS evaluation, metabolites and hormonal analysis permit to manage properly the energy balance and to improve body status, reproductive performance and welfare of rabbit does. Naturally, this preliminary approach must be further studied, perhaps simplified and tested with a large livestock. Metabolites analysis represents a good tool for understanding the physiological mechanisms required to meet these objectives.

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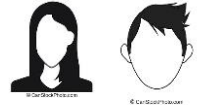
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Serum biochemical profile of rabbit does of Algerian local population (Aures region) at different physiological stage

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Context and objectives: The aim of this study was to determine the levels of certain plasmatic biochemical values during different physiological stages of the local rabbit doe.

Methods : One hundred and twenty (n=120) nulliparous does of local Algerian population were used for natural mating to study reproductive performance. The trial was carried out at the experimental rabbit farm of the Veterinary Department of the University Batna 1.

Four blood samples were collected from all females at: 08:00h, at different physiological stages from the marginal ear vein, drawn into vacutainer tubes, immediately centrifuged at 2000 x g for 10 mn (SIGMA-2K15 centrifuge) and plasma stored frozen until assayed for hormones and metabolites.

Results : Compared to the does of P and P-L group, the does of L group had higher ($p < 0.05$) sexual receptivity and fertility rate. Such an improvement was expected given that the overlapping between lactation.

Table 1: Reproductive performances of rabbit does at different physiological stages.

Parameters	Physiological status				P
	E	P	P-L	L	
Receptivity (%)	-	55.5	58	64.7	*
Fertility (%)	-	51.2	45.5	59.4	*
Initial BW (g)	3000±37	3053±47	3188.4±63	3187±67	ns
Cumulative BCS	3.00	2.2	1.96	2.56	*
BW at kindling (g)	-	2839.3±70	2703.3±68	3050±57	*
Milk yield (g)	-	-	-	-	-
-at 1 st week	-	-	435.1±96	631.4±136	*
-at 2 nd week	-	-	850.6±37	1316±156	**
-at 3 rd week	-	-	935±67	966±155	ns
Average MY during suckling period	-	-	2200.7±36	2913.4±105	*
LS at birth	-	6.9±2.3	7.4±2.37	9±1	**
LW at birth (g)	-	390.5±13.6	402±11	501±9	*
LS at weaning	-	5.2±2.3	5±1.2	8±2	**
LW at weaning (g)	-	2040±67	2200±25	3720±39	*

The higher total protein, albumin and globulin concentrations ($p < 0.05$) were in does of L group, Total lipids, triglycerides and total cholesterol concentrations in does of P group were ($p < 0.05$) higher .

Table 2: Effect of the physiological stage on serum protein fractions in doe rabbits.

Parameters	Physiological status			
	E	P	P-L	L
Total proteins (g/dl)	3.60±0.5	4±0.90	5.70±1	6.10±0.1
Albumin (g/dl)	2.01±0.1	2.81±0.05	2.98±0.07	3.50±0.10
Globulin (g/dl)	1.22±0.02	1.69±0.01	1.88±0.06	2.80±0.04

Table 3: Effect of the physiological stage on serum lipid profile in doe rabbits.

Parameters	Physiological status			
	E	P	P-L	L
Total lipids (mg/dl)	31±1	97±0.10	69±0.2	78.01±0.6
Triglycerides (mg/dl)	15.4±5.8	65.80±23	44.7±17.6	37.7±25.6
Cholesterol (mg/dl)	28.7±16.7	70±18.2	58.3±24.1	38.8±23.9

As an original experiment is still in progress, it would be premature to draw any conclusion. However, some correlations are noteworthy. The lactating and pregnant females presented a strong mobilization of body fat, which can explain the energy needs that are more significant during pregnancy and lactation