Metabolic profile of zebu cows (*Bos indicus*), cystic or with anovulatory anestrus

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Abstract

This study was carried out in the Adamawa region (Cameroon) with the main objective to evaluate the metabolic profile of zebu cows, cystic or with anovulatory anestrus and subsequently to propose a tool for diagnosing the risks of these pathologies. Thus, 53 zebu cows (20 cystic and 33 with anovulatory anestrus) were chosen and the blood obtained was centrifuged (3000 g, 10 min, 4° C) to obtain the serum. Serum concentrations of glucose, total cholesterol, albumin, total proteins, urea, calcium and phosphorus were quantified by spectrophotometry at a wavelength ranging from 340 to 630 nm based on the parameter tested. The metabolic profile of cows with anovulatory anestrus was characterized by low serum levels of glucose (< 2.6 mmol / L), total cholesterol (< 2.3 mmol / L), urea (< 3.8 mmol / L), calcium (< 2.2 mmol / L) and phosphorus (< 1.05 mmol / L); high serum levels of total proteins (> 80 g / L) and total globulins (> 45.5 g / L); and normal albumin (27.7-40.4 g / L) with an average Body Condition Score (BCS) of 3. The metabolic profile of cystic cows differed from that of anovular females only by normal total cholesterol level between 2.3 and 6 mmol/L and high urea (> 6.5 mmol /L). These findings could be used for the diagnosis and treatment of these pathologies.

Keywords: Zebu, metabolic profile, ovarian cysts, anovulatory anestrus, Ngaoundere, Cameroon.

Profil métabolique des vaches zébus (Bos indicus) kystiques ou en anoestrus anovulatoires

Résumé

Cette étude a été réalisée dans la région de l'Adamaoua (Cameroun) avec pour principal objectif d'évaluer le profil métabolique des vaches zébus présentant des kystes ovariens ou des anœstrus anovulatoires afin de proposer un outil de diagnostic des risques de ces pathologies. Ainsi, 53 vaches zébus (20 kystiques et 33 en anæstrus anovulatoires) ont été prélevées et le sang obtenu a été centrifugé (3000 g, 10 min, 4°C) pour l'obtention du sérum. Les concentrations sériques de glucose, cholestérol total, albumine, protéines totales, urée, calcium et phosphore ont été quantifiées par spectrophotométrie à une longueur d'onde variant de 340 à 630 nm selon le paramètre testé. Il en ressort que le profil métabolique des vaches présentant un anestrus anovulatoires était caractérisé par des concentrations sériques faibles en glucose (< 2,6 mmol/l), cholestérol (< 2,3 mmol/l), urée (< 3,8 mmol/l), calcium (< 2,2 mmol/l) et phosphore (< 1,05 mmol/l); élevées en protéines totales (> 80 g/l) et globulines totales (> 45,5 g/l); normale (27,7- 40,4 g/l) en albumine avec une Note d'État Corporel (NEC) moyenne (3). Le profil métabolique des vaches kystiques ne diffère de celui des anæstrus anovulatoires que par une cholestérolémie normale (2,3-6 mmol/l) et une urémie élevée (> 6,5 mmol/l). Ces résultats pourraient constituer un outil de diagnostic ou de traitement de ces pathologies.

Mots-clés: Zébu, profil métabolique, kystes ovariens, anoestrus anovulatoires, Ngaoundéré, Cameroun.

INTRODUCTION

Livestock is an indispensable means to reach the point of food self-sufficiency in Cameroon. Adamawa, one of the 10 regions of Cameroon, is an area where the breeding of cattle of various breed (local and exotic) predominates with 38 % of the national herd (MINEPIA, 2003). Nevertheless, productivity remains low due mainly to zootechnical, sanitary, nutritional and reproductive problems (Kouamo et al., 2016). The problem of infertility or low production, coupled with the desire of the population to control the reproduction process, have led to the development of new assisted reproductive technologies (ARTs) (Rahman et al., 2008). The applications of these ARTs such as superovulation, artificial insemination (AI), embryo transfer (ET) and in vitro fertilization (IVF) in cows are useful for increasing productivity, preserving genetic potential of sub-fertile or dead animals and for reducing reproductive pathologies (Deuleuze *et al.*, 2009).

However, in the case of bovine breeding, in spite of all these ARTs, several individual factors (fattening condition, postpartum weight loss, calving interval, calving difficulties, non-delivery, uterine involution) and breeding condition (feed, season, mode of reproduction) constitute limits to fertility (Mialot, 1996).

The reproductive performance of domestic animals is severely disrupted if the body's energy and protein requirements are not met, either in the case of undernutrition or bad nutrition in extensive livestock system or sharp increase in needs (lactation, repeated pregnancy) in intensive livestock system (Monget *et al.*, 2004). These reproductive performance disorders are also the result of the action of several infectious agents (Manas *et al.*, 2012). Diet is one of the most important environmental factors in breeding control. Thus, animals in poor body condition have poor reproductive performance (Brisson, 2003).

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Pathological conditions such as ovarian cysts (OC) and anovulatory anestrus are the major problems in cattle, which affect fertility leading to great economic loss to farmers. These negative effects on reproductive performance and livestock economy have been quantified (Fourichon *et al.*, 2000; Kouamo *et al.*, 2016). Would there be a causal relationship between the variation in serum metabolite levels and the appearance of OC and anovulatory anestrus? Thus, this study was carried out with the general objective to evaluate the metabolic profile of zebu cows, cystic or with anovulatory anestrus. Specific objectives were to characterize slaughtered cows presenting the two pathologies, to study the changes in serum metabolite levels and propose a tool to diagnose the risks of the occurrence of OC and anovulatory anestrus from metabolites.

MATERIALS AND METHODS

Study area and animal selection

This study was carried out in Ngaoundere, Adamawa region, Cameroon (Latitude19°7'39N and Longitude13° 35'4 E), characterized by a sudano-sahelian climate. The samples were collected at the Ngaoundere Municipal Slaughterhouse (NMSH) and analyzed in the Veterinary Research Laboratory of IRAD Wakwa. The Adamawa region is characterized by a dry season from November to March and a rainy season from April to October. Each year an average rainfall of 1496.7 mm is recorded. The minimum and maximum average temperatures recorded were 15.2 ° C and 29 ° C, respectively.

The study was conducted on 53 zebu cows, cystic (20) or with anovulatory anestrus (33). Eighty-four (67.7 %) and forty (32.3 %) cows were originated from Mayo Rey and Vina division, respectively. The animals were raised in an extensive system. Breed, weight, BCS and age of cows were evaluated as described by Lhoste (1969), Njoya et al. (1997), Natumyana et al. (2008) and Moussa Garba et al. (2013), respectively. Ovarian and genital tract diagnosis were performed in all ante-mortem cows by transrectal palpation and confirmation of OC and anovulatory anestrus was performed post-mortem after slaughter. OC was considered as an anovulatory follicle with a diameter greater than 24 mm in the absence of a functional corpus luteum (CL) (Hanzen et al., 2008). The classification of types of anovulatory anestrus was based on the presence of follicles, CL (1 and 2 for hemorrhagic and diestrus CL, respectively) and / or cysts on the ovaries (Peter et al., 2009; Nguyen and Hanzen, 2013). Thus, five types of anovular anestrus were identified: type 0 (absence on both ovaries, follicle with diameter ≥ 2 mm, CL and cysts); type I (presence on one ovary, follicles of diameter between 2 and 7 mm with the absence of CL and cysts on both ovaries); type II (presence on one ovary, at least one follicle of diameter > 7 mm and follicles between 2 and 7 mm in the absence of CL and cysts on both ovaries); type III (presence on one ovary, ovarian cyst in the presence or not of follicles diameter > 7mm but in the absence of CL1 and CL2 on both ovaries); type IV (presence on one ovary, a CL2 in the presence or not of follicles ≥ 8 mm diameter and the presence of a pyometra).

Blood collection and biochemical analysis

Blood from the jugular vein was collected aseptically from each Gudali (Venoject®). After collecting blood, the serum was separated out by centrifugation (3000 g, 10 min, 4°C), and aliquots were stored at -20°C until assayed. Serum sample was evaluated for the concentration of glucose, total cholesterol, urea, total proteins, albumin, calcium and phosphorus. Total globulins were calculated by subtracting albumin from total proteins. All biochemical parameters were determined by a spectrophotometric method using commercial kits supplied by Cromatest®.

Statistical analysis

Data obtained were recorded and statistically analyzed using Statistical Package for Social Science software version 20. Analysis of Variance and Turkey HSD tests were used to compare different groups. The level of significance was recorded at the 5% level of confidence.

RESULTS

Characterization of cows

The animals were between 4 to 14 years old $(6.43 \pm 0.42 \text{ years})$, with a BCS of 2.64 ± 0.09 . The average weight of the cows (Min-Max) was $341.02 \pm 12.07 \text{ Kg}$ (187.02-518.56 Kg). The origin and weight of the cows have no significant influence (P> 0.05) on the occurrence of ovarian pathologies. The breed and age of the cows have a significant influence (P <0.05) on the occurrence of anovulatory anestrus while they have no significant influence on the occurrence of OC (Table 1).

Global nutritional status of zebu cows

The mean values of blood glucose, calcium and phosphorus of the animals examined were lower than the physiological values; those of total cholesterol, albumin and urea are included in the reference range. Only the total protein and total globulins levels were above the reference values (Table 2). The majority of cows had low glucose (62.1 %), calcium (67.2 %), phosphorus (63.7 %), total cholesterol (56.4 %), urea (44.3 %); normal in albumin (69.3 %) and high in total proteins (70.2 %) and total globulins (66.9 %) (Table 2).

Metabolic changes in females, cystic or with anovulatory anestrus

The mean BCS did not vary significantly (P> 0.05) between OC and anovulatory anestrus. The majority of cows presenting these two ovarian pathologies have an average BCS (3/5). Overall, most metabolites excepted albumin have low serum concentrations in the present study. However, total proteins and total globulins levels were significantly higher (P < 0.05) in all two ovarian pathologies. Serum urea concentrations remained high in cystic cows but low in females with anovulatory anestrus (Table 3).

Proposition of a diagnostic tool for OC and anovular anestrus based on the variation of metabolites

The metabolic profile of cystic cows was similar to females with anovulatory anestrus but differs by normal total cholesterol and high urea (Table 4).

Discussion

The proportions of ovarian abnormalities (anovulatory anestrus and OC) and characteristics of slaughtered animals have been reported by Kouamo *et al.*, (2016) in a previous study in the same environment. The zebu cows

with anovulatory anestrus have the same metabolic profile and thus the same diagnostic tool as reported by Manas *et al.*, (2012) and Kumar *et al.*, (2015). In this study, the low glucose concentration observed in the cows presenting the two pathologies is similar to the study of Reddy *et al.*, (2012), Naafia *et al.*, (2013) and Mimoune *et al.*, (2017).

Table 1: Proportions of OC and anovulatory anestrus according to origin, breed, age and weight

Parameters	Factors	OC	anovulatory anestrus	Type 0	Type 1	Type 2	Type 4
Origin	Mayo Rey	80 (16) ^a	63.6 (21) ^a	100 (2) ^a	64.3 (9) ^a	53.3 (8) ^a	100 (2) ^a
	Vina	20 (4) ^a	36.4 (12) ^a	0 (0)a	35.7 (5) ^a	46.7 (7) ^a	0 (0)a
	P-value	0.55	0.53	0.33	0.76	0.19	0.33
Breed	Akou	40 (8)a	24.2 (8) ^a	100 (2)a	14.3 (2) ^a	20 (3) ^a	50 (1)a
	Bokolo	0 (0)a	15.1 (5) ^a	0 (0)a	28.6 (4) ^b	6.67 (1) ^a	0 (0) ^a
	Djafoun	40 (8) ^a	24.2 (8) ^a	0 (0)a	21.4 (3) ^b	26.7 (4) ^a	50 (1)a
	Gudali	20 (4) ^a	36.4 (12) ^b	0 (0)a	35.7 (5) ^b	46.7 (7) ^a	0 (0)a
	P-value	0.67	0.00	0.43	0.00	0.31	0.64
Age (Years)	[4-8]	80 (16) ^a	93.9 (31) ^a	100 (2) ^a	85.7 (12) a	100 (15) ^a	100 (2) ^a
	[9-14]	20 (4)a	6.06 (2) b	0 (0)a	14.3 (2) a	0 (0)b	0(0)a
	P-value	0.99	0.02	0.47	0.56	0.04	0.47
Weight (Kg)	<250	0 (0) ^a	9.09 (3) ^a	0 (0)a	14.3 (2)	6.67 (1) ^a	0 (0)a
	[250-350]	40 (8) ^a	60.6 (20) ^a	50 (1) ^a	11.3 (8)	66.7 (10) ^a	50 (1) ^a
	>350	60 (12) ^a	30.3 (10) ^a	50 (1) ^a	28.6 (4)	26.7 (4) ^a	50 (1) ^a
	P-value	0.68	0.16	0.93	0.21	0.38	0.93

 $^{^{(}a,b,c)}$ percentages in a column with different superscripts are significant at p < 0.05.

Table 2: Distribution of cows according to global nutritional status

Nutritio	onal		Blood concentrations (%)			Reference values*	
Parameters	M±SD	Min-Max	Low	Normal	High	Keierence	e values"
Glucose	2.45±0.34	0.18-9.8	62.1	30.6	7.26	2.6-4.9	mmol/L
Total cholesterol	2.47±0.34	0.05-8.44	56.4	37.9	5.65	2.3-6.0	mmol/L
Total Proteins	154.3±25.27	25.2-700	21.0	8.87	70.2	59.5-80	g/L
Albumin	30.3±0.91	16.6-44.16	30.6	69.3	0	27.7-40.4	g/L
Total Globulins	124±25.32	0.05-673.08	19.3	13.7	66.9	26.2-45.5	g/L
Urea	4.35±0.46	0.06-11.91	44.3	37.9	17.7	3.8-6.5	mmol/L
Calcium	1.86±0.20	0.25-3.29	67.2	17.2	15.6	2.22-2.7	mmol/L
Phosphorus	0.95±0.14	0.02-3.28	63.7	33.9	2.4	1.05-2.83	mmol/L

^{*}Kouamo et al., 2011. M±SD: Mean ± Standard Deviation. Min= minimum Value; Max= Maximum value.

Table 4: Occurrence risk of OC and anovulatory anestrus according to metabolites variation

Serum metabolites	Criteria a	ssociation
Glucose	Low (< 2.6 mmol/ L)	Low (< 2.6 mmol/ L)
Total Cholesterol	Normal (2.3- 6.0 mmol/ L)	Low (<2.3 mmol/ L)
Albumin	Normal (27.7- 40.4 g/ L)	Normal (27.7- 40.4 g/l)
Total Proteins	High (> 80g/ L)	High (> 80g/ L)
Total Globulins	High (> 45.5g/ L)	High (> 45.5g/ L)
Urea	High (> 6.5mmol/ L)	Low (< 3.8 mmol/ L)
Calcium	Low (< 2.22 mmol/ L)	Low (< 2.22 mmol/ L)
Phosphorus	Low (<1.05 mmol/ L)	Low (<1.05 mmol/ L)
BCS/5	Average (3)	Average (3)
Risks	OC	Anovular anestrus

Table 3:Distribution of cows according to nutritional status and ovarian pathologies: OC and anovulatory anestrus

Demonstrate	Donout:		200042000000000000000000000000000000000	True	1		T. T.	D Value
rarameters	Repartition	3	anovulatory anestrus	Type 0	Type I	1ype 2	Type 4	r-value
	Mean±SD	3 ± 0.21^{a}	2.61 ± 0.08^{a}	2.5 ± 0.34^{a}	$2.5{\pm}0.13^{a}$	$2.73\pm0.12^{\text{a}}$	2.5±0.34 a	0.23
BCS	Thin (1-2)	5 (25 %) ^a	13 (39.4 %) ^a	$1(50\%)^{a}$	7 (50 %) ^a	4 (26.7 %) ^a	$1 (50 \%)^a$	
	Normal (3)	15 (75 %) ^b	20 (60.6 %)	$1(50\%)^{a}$	7 (50 %) ^a	11 (73.3 %)	$1(50\%)^a$	
	P-Value	0.00	0.00			0.00		
	Mean±SD	2.28±0.81a	2.21±0.32ª	6.165±46.2ª	1.72 ± 0.58^{a}	2.17 ± 0.87^{a}	2.02±16.3a	0.43
	Low	$1.18\pm0.30~(60~\%)^{a}$	1.33 ± 0.18 $(69.7\%)^a$	$1 (50 \%)^a$	1.31 ± 0.18 (78.6 %) ^a	$1.30\pm0.25~(66.7~\%)^{\text{a}}$	$1 (50\%)^a$	
Glucose	Normal	3.94 ± 0.38 (40 %) ^b	$3.34\pm0.30~(24.2~\%)^{b}$	0(0%)	$3.21\pm0.36~(21.4~\%)^{b}$	$3.44\pm0.40~(26.7~\%)^{b}$	$1(50\%)^a$	
	High	0 (0%)	7.8±0.60 (6.06%)	$1 (50\%)^a$	0 (0%)	5.8±0.78°(6.67 %)	(%0) 0	
	P-Value	0.01	0.00		0.01	0.00		
	Mean±SD	2.23±0.81ª	2.14 ± 0.31^{a}	3.3±20.84ª	1.44±0.68ª	2.78±1.26 ^a	1.07±10.1 a	0.11
	MO.	$141\pm0.23(40\%)^a$	1 22±0 14 (72 73%) a	$1(50\%)^a$	1 19+0 20 (92 9%) a	1 27+0 22a (53 3 %)	2 (100%)	
Total cholesterol	Normal	2 77+0 19 (60%) ^b	3 71+0 28 (71.73%) b	$1(50\%)^a$	4 75+0 74 (7 14 %) b	3.25+0.27 ⁶ (33.3%)	(%0)0	
	High	0.00) (1.0=/./2	(%)(4) (%)(%)(%)(%)(%)(%)(%)(%)(%)(%)(%)(%)(%)((%)()()	(%) 1 (%) 0	7 60+0 43° (13 3 %)	(%0)0	
	P-Value	0.02	0.00	(6/0) 0	0.00	0.00	(6/0)0	
	Mean±SD	31±2.18ª	30.59 ± 0.85^{a}	28.5±26.75 ^a	31.4 ± 3.54^{a}	30.3±2.99 a	29.7 ±83.8ª	0.95
	Low	(%0) 0	25.2 ± 1.11 (36.4 %) ^a	$1 (50\%)^a$	$24.6\pm1.49(35.7)^{a}$	25.9±2.01 (33.3 %) a	1 (50%)	
Albumin	Normal	$31\pm 2.53 (100 \%)$	33.7±0.84 (63.6 %) ^b	$(50\%)^a$	35.1±1.11 (64.3 %) ^b	32.5±1.42 (66.7 %) b	1 (50%)	
	High	(%0) 0	(%0) 0	(%0) 0	0 (0%)	0 (0%)	(%0) 0	
	P-Value		0.00		0.00	0.01		
	Mean±SD	81.71±58.14 ^b	113.5±22.6°	109.2 ± 45.9^{a}	81.9±15.8b	149.5±93ª	69.9±346.2 ^b	0.00
	Low	$56.2\pm11.5\ (20\ \%)^{a}$	40.3±38.9 (24.2 %) ^a	0(0%)	$46.6\pm10.8(21.4\%)^{\mathrm{a}}$	34.9±77.05 (26.7 %) ^a	$1 (50\%)^a$	
Total Proteins	Normal	$67.4\pm11.5 (20\%)^{a}$		(%0)0	69.8±13.21 (14.3 %) ab	$2.49\pm109 (13.3 \%)^{a}$	(%0) 0	
	High	$95\pm6.67 (60\%)^a$	$150.1\pm24.0~(63.6~\%)^{\mathrm{b}}$	2 (100%)	96.3±6.22 (64.3 %) ^b	$218.8\pm51.4 (60\%)^{a}$	$1 (50\%)^a$	
	P-Value	0.20	0.01		0.01	0.14		
	Mean±SD	$50.7\pm58.24^{\rm b}$	82.9±22.7°	$80.7 \pm 72.7^{\rm a}$	$50.5{\pm}15^{\mathrm{b}}$	119.2 ± 92.6^{a}	$40.3\pm430^{\rm b}$	0.00
	Low	$22.6\pm10.9\ (20\ \%)^{a}$	$9.58\pm40.27(21.21\%)^{a}$	(%0)0	$13.2\pm12.4~(14.3\%)^{a}$	8.57±73.95 (26. 7 %) ^a	1 (50%)	
Total Globulins	Normal	$34.8\pm10.9~(20~\%)^{\text{a}}$	41.14±35.52 (27.27%) ^a	(%0) 0	43.3±7.15 (42.9 %) b	$36.8\pm85.39(20\%)^{3}$	0(00)	
	High	$65.4\pm6.27\ (60\ \%)^{a}$	135.28±25.84 (51.51%) ^b	2 (100%)	70.1±7.15 (42.9 %) ^b	$205.4\pm52~(53.3\%)^{a}$	1 (50%)	
	P-Value	0.20	0.00		0.00	0.09		
	Mean±SD	5.84 ± 1.07^{a}	3.84±0.42ª	2.17 ± 19.50^{a}	4.55±1.57a	$3.40{\pm}1.17^{\mathrm{a}}$	3.81 ± 12.01^{a}	0.28
	L_{0w}	$2.56\pm0.40\ (20\ \%)^{a}$	2.10±0.27 (48.5 %) ^a	2(100%)	$1.44\pm0.52\ (35.7\ \%)^{\mathrm{a}}$	$2.10\pm0.38~(53.3~\%)^{a}$	1 (50 %)	
Urea	Normal	$5.71\pm0.40(20\%)^{b}$	4.38±0.31(36.4 %) ^b	0 (0%)	4.38±0.42 (35.7 %) ^b	4.22 ± 0.44 (40 %) ^b	1 (50 %)	
	High	$6.98\pm0.23~(60~\%)^{6}$	$8.14\pm0.49(15.1\%)^{\circ}$	0 (0%)	7.93±0.52 (28.6 %)°	$8.96\pm1.08~(6.67~\%)^\circ$	(% 0) 0	
	P-Value	0.02	0.00		0.00	0.00		
	Mean±SD	1.65 ± 0.32^{a}	$1.94{\pm}0.16^{\mathrm{a}}$	$1.56\pm2.86^{\mathrm{a}}$	$1.73{\pm}0.96^{a}$	$2.27{\pm}0.46^{\mathrm{a}}$	1.27 ± 6.61^{a}	0.35
	\mathbf{Low}	$1.65\pm0.32~(100\%)$	$1.40\pm0.11~(60\%)^{a}$	2 (100%)	1.35 ± 0.07 (66.7 %) ^a	$1.63\pm0.16 \ (40\%)^{a}$	2 (100 %)	
Calcium	Normal	0 (0%)	$2.54\pm0.18(20\%)^{\rm b}$	0 (%)	$2.7\pm0.13~(16.7~\%)^{b}$	$2.49\pm0.19 \ (30\%)^{6}$	(% 0) 0	
	High	0 (0%)	$2.93\pm0.18(20\%)^{b}$	0 (%)	$2.99\pm0.13(16.7\%)^{b}$	$2.91\pm0.19\ (30\ \%)^{b}$	(% 0) 0	
	P-Value		0.00		0.01	0.01		
	Mean±SD	$0.71{\pm}0.33^{a}$	0.92 ± 0.13^{a}	1.99 ± 7.31^{a}	$0.76\pm0.24^{\mathrm{a}}$	$0.96\pm0.40^{\mathrm{a}}$	0.62 ± 0.25^{a}	0.51
Phosphorus	Low	$0.23\pm0.10~(60\%)^{a}$	0.57 ± 0.07 (69.7 %) ^a	(%0) 0	$0.64\pm0.08~(78.6~\%)^{a}$	$0.54\pm0.12(66.7\%)^{a}$	2 (100 %)	
	Normal High	$1.42\pm0.12~(40\%)^{b}$	$1.72\pm0.11(30.3\%)^{6}$	2 (100%)	$1.54\pm0.19(21.4\%)^{b}$	$1.81\pm0.17(33.3\%)^{b}$	(%0)0	
	ngm	0 (0/0)	0 (0/0)	0 (0/0)	0 (0/0)	0 (0 / 0)	0 (0/ 0) 0	

 $^{(a,b,c)}$ percentages in a column with different superscripts are significant at p < 0.05.

A negative energy balance is an infertility factor responsible for several pathological disorders of the ovary. Mukherjee et al., (2011) reported three factors responsible for the low glucose concentration, namely the failure of neoglucogenesis or glycogenolysis, the increase in peripheral glucose absorption and endogenous hyperinsulinemia. Low glucose in blood causes depression of hypothalamic function, low secretion of GnRH and atrophy of the ovaries, which could lead to anovulatory anestrus. In addition, hypofunctional CL provokes a decrease of progesterone and affects negatively the fertility. CLs are small and poorly developed, with low progesterone production and LH peak asynchrony. Therefore, inadequate uterine environment is formed and this increases the abnormalities and the loss of embryos. Embryonic mortality or non-fertilization leads to the regression of the CL and the continuation of the subsequent follicular cycle whose dominant follicle will ovulate with the formation of a new CL. An energy deficiency alters the capacity of the follicles to produce enough estradiol to ensure ovulation, and it is associated to anovulatory anestrus or OC (Benoit et al., 1996). Funston et al., (1995) showed that a "glucoprivation" induced by a peripheral injection of 2-desoxy-glucose (2DG), results in a fall in LH and FSH secretion in sheep, despite an increase in peripheral insulin levels and causes the appearance of OC and anestrus. Average total cholesterol in anovulatory anestrus cows was lower than the physiological value. This result is similar with the findings of Rameez and Sukla (2012). On the contrary, Ahmed et al., (2017) reported normal serum total cholesterol levels in these cows. The higher the energy deficit, the lower the total cholesterol level, the greater the risk of anestrus. In addition, cholesterol is the precursor of the synthesis of steroid hormones that play a role in fertility, fecundity and whose imbalance could cause the occurrence of anovulatory anestrus.

Total proteins in cows, cystic or in anovular anestrus are greater than the physiological range and mean albumin levels remain normal (Ahmed et al., 2017). Total proteins consist of albumin and globulins; therefore, this high protein is only due to an increase in total globulins. A high blood total globulins level marks a possible infection against which the organism is fighting, using a large part of its energy reserves; which affects the functioning of the hypothalamic-pituitary axis at the origin of OC and anestrus. Indeed, the concentration of total proteins increases (to more than 80 g/L) in the case of blood concentration which takes place in the majority of cases when the animal shows a dehydration or when raising the total globulin level following an infectious, acute or chronic inflammatory process (Braun et al., 1992). Infectious agents present in the genital tract may hamper fertilization and early embryo development as well (Kouamo et al., 2016). A high incidence of OC and anovulatory anestrus is associated with a significant increase in total proteins, albumin and total globulins (Joe et al., 1998). Excess nitrogen seems to be the most harmful to reproduction in current feeding systems (Wallace, 1991). The average urea value of the majority of cows with OC (60 %) is greater than the physiological range. Jackson et al. (2011) and Mimoune et al. (2017) reported that excess urea affects the uterus and ovary. The elevated plasma urea nitrogen can change

the uterine fluid composition, lowering the uterine pH, and reducing the conception rates. This elevated plasma urea nitrogen level may be caused by the high protein content in the diet. Disturbances of ovulation in part because of faulty luteinizing hormone (LH) secretion, a prolonged duration of standing estrus or improper steroid genesis appear to be major causes of OC in dairy cattle (Purohit, 2001). The synthesis and clearance of urea requires enormous energy. A high concentration of urea greatly reduces the energy of the cow, and energy deficiencies disturb the hypothalamic-pituitary axis by reducing the secretion of gonadotropins, and favoring the appearance of OC.

Cows presenting one of the two pathologies have low calcemia and phosphoremia in their majority. These results are similar to those reported by several authors (Bindari et al., 2014; Yotov et al., 2014; Osman et al., 2017). Minerals such as calcium and phosphorus depend on feed intake in quantity and quality. The main source is plants ingested on the pasture. They are essential and are involved in many biological and reproductive processes (Kouamo et al., 2011). El-shahata et al., (2010) reported that calcium plays a key role in improving the number and size of pre-ovulatory follicles as well as the ovulation rate. Calcium plays a role in the regulation of "Gap junction" between cumulus cells; it is responsible for a disruption of cell cohesion, which contributes to the process of ovulation (Peracchia et al., 1978). The absence of calcium would negatively influence the function of the hypothalamic-pituitary axis. This will disrupt the functioning of the reproductive organs, favoring the appearance of OC and anestrus. Stankiewicz et al. (2015) reported low concentrations of calcium and phosphorus in the blood and follicular fluid of females with OC contrary to normal ones, hence the importance of these minerals in the occurrence of OC. Phosphorus is essential in all metabolic pathways, energy use and transfer, and is part of the structure of nucleic acids (Murray et al., 2003). Therefore, any imbalance would lead to metabolic disorders.

CONCLUSION

The present study indicated that the metabolic profile of cystic cows is similar to females with anovulatory anestrus but differs by normal total cholesterol and high urea. These findings could be used for the diagnosis and treatment of these pathologies.

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