SOME BIOCHEMICAL ALTERATIONS ASSOCIATED WITH CARBOHYDRATE DEFICIENCY IN PREGNANT GOATS, WITH PARTICULAR REFERENCE TO PREGNANCY TOXAEMIA

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SUMMARY

The goal of the present study was to investigate some biochemical alterations in pregnant goats suffering from carbohydrate deficiency. The main clinical signs were anorexia depression, locomotion disturbances from staggering gait to sternum recumbancy which might have resulted in reduced energy intake and precipitated pregnancy toxaemia. Blood and urine samples were collected from 12 diseased animals before and after treatment trials. Samples were also collected from 8 late pregnant goats that were proved healthy. Laboratory findings showed ketonuria, lower serum values of glucose, cholesterol, triglycerides and insulin. Higher serum liver enzymes, urea, creatinine, total lipids, free fatty acids, β-OHB and cortisol, were observed. The response of the affected goats to conventional therapeutic treatment was variable. It could be concluded that, the disturbance in ration constituents especially carbohydrates leads to pregnancy disease which can be prevented by the subsequent correction of carbohydrate and lipid parameters in affected goats with balancing the nutritional intake of the goats with the increased late pregnancy needs of the dam and her fetus to avoid and prevent the occurrence of this syndrome.

INTRODUCTION

Carbohydrate deficiency results from a drop in the plane of nutrition during last pregnancy and/or management changes that create a brief period of fasting leading to pregnancy disease **(Kimberling, 1988)**. Undernourishment or starvation of multiple pregnant ewes resulted in Pregnancy ketosis **(Reid, 1968 and Bergman, 1984)**. A failure of dietary energy and energy from gluconeogenesis to meet the increasing foetal glucose demands in the last 6 weeks of a multiple pregnancy leads to pregnancy toxaemia **(Sargison, 1994)**. The determining cause of toxaemia is an alteration in the energy metabolism, as a consequence of an imbalance between glucose offer and demand, thereby giving rise to negative energy balance. This imbalance is caused by a reduction in energy supply due to poor or inadequate nutrition, in addition to the increasing re-

quirements of the fetus in its last prenatal growth phase and the pressure of the gravid uterus upon the digestive organs within the abdominal cavity **(Hay and Baird, 1991 and Van Saun, 2000)**. The consumption of low energy levels or poor utilization of the available energy supply gives rise to a gradual reduction in blood glucose levels, with depletion of the liver glycogen reserves and mobilization of the fatty depots for use as an unusual energy source with the subsequent formation of ketone bodies and fatty liver infiltration **(Radostits et al., 2000)**. The condition can be diagnosed by clinical examination of animals and confirmed biochemically by a marked increase in the concentration of β-hydroxybutyrate and a corresponding decease in the plasma glucose concentration **(Scott and Woodman, 1993)**. The present study aimed to investigate some biochemical alterations associated with carbohydrate deficiency in pregnant goats, with particular reference to pregnancy toxaemia.

MATERIALS AND METHODS

A total number of 40 late pregnant balady goats of 3-5 years old were obtained from a private farm in Abo-Hammad, Sharkia Governorate. The history of the flock indicated that these animals were inadequately fed (rice straw, grass and sometimes concentrates). The clinical signs were anorexia, depression, locomotion disturbances from staggering gait to sternum recumbancy of some cases which might have resulted in reduced energy intake and precipitated pregnancy toxaemia.

Blood and urine samples were collected from 12 clinically affected goats before and 7 days after treatment. Samples were also collectd from 8 late pregnant animals that were proved healthy after precise clinical and laboratory examinations. Blood samples were collected from the jugular vein in clean and dry centrifuge tubes for separation of serum to estimation of serum glucose **(Caraway, 1976)**, enzyme activities of aspartate aminotransferase (AST), alanine aminotransferase (ALT) **(Reitman and Frankel, 1957)**, alkaline phosphatase (ALP) **(Tietz and Shuey, 1986)**, blood urea nitrogen (BUN) **(Patton and Crouch, 1977)**, creatinine **(Henry, 1974)**, total lipids **(Chabrol and Charonat, 1937)**, triglycerides **(Trinder, 1969)**, total cholesterol **(Watson, 1960)**, free fatty acids **(Schuster and Pilz, 1979)** and β-hydroxybutyrate (β-OHB) **(Mercer et al., 1986)** by using diagnostic reagent kits supplied by BioMerieux, Marey-L Etoile, France. The hormonal assay of cortisol and insulin were done by using direct radioimmunoassay technique with coat A eount (DPC) kits according to **Burtis et al. (1994)**. Urine samples were immediately used for detection of ketone bodies using eoumbour-9 test-strips (Boehringer Monnheim, Germany).

A trial of treatment aimed to correct energy and stimulating appetite, where the diseased goats were received daily I/v injection of 250 ml of 25% dextrose (once a day) in addition to 10

gm sodium bicarbonate (once a day) and 250 ml Ringer solution orally/head (twice a day) until goats eat normally.

The obtained data were statistically analyzed by using the analysis of variance according to **Snedecor and Cochran (1982).**

RESULTS

Upon clinical examination, the affected goats were with variable degrees of affection ranged from dullness, depression, inappetance and star gazing to sternum recumbancy of some cases. The smell of acetone was detected in the breath. Results of the trial treatment showed that 6/12 of the affected goats showed clinical improvement and 4/12 were partially recovered, while 2/12 of the affected goats were died inspite of treatment.

Laboratory findings as shown in Table (1) have showed a significant increase (P<0.001) in liver enzymes activities; AST, ALT and ALP and (P<0.01) in kidney parameters (urea and creatinine) in affected goats compared to healthy ones.

Serum concentrations of glucose, cholesterol and triglycerides showed a siguificant decrease (P<0.01) while total lipids were significantly increased (P<0.01) in the diseased group as compared to the control ones. Serum free fatty acids and _-OHB values showed a significant elevation (P<0.001) in affected goats than healthy ones.

Cortisol level revealed a siguificant increase (P<0.001), while insulin level showed significant decrease (P<0.01) in the diseased goats as compared to the healthy ones.

Ketone bodies were detected in the urine of affected goats indicating ketonuria.

DISCUSSION

The affected goats had a history of insufficient feed supply especially carbohydrate diet prior to the onset of clinical disease. A qualitatively insufficient feeding program together with the significant increased glucose consumption by the rapidly growing fetuses induce progredient hypoglycemia and ketosis. Other factors like stress and decreased ruminal volume may help triggering the metabolic breakdown of fat **(Brus, 1989)**. Anorexia, depression, dullness, locomotion disturbances from staggering gait to sternum recumbency of some cases were signs observed in affected goats. These observations are in agreement with **El-Sebale et al. (1992), Nasser et al. (1998)** and **Mohamed et al. (2005)**. The clinical signs were confirmed by the presence of ketone bodies in urine of the affected goats indicating ketonuria. Once the energy imbalance, in affected

goats, has become established, the host systems attempt to maintain sufficient blood glucose levels to satisfy the needs of the most vital tissues of the fetus. This done by liver gluconeogenesis from propionate derived from carbohydrate digestion, amino acids and glycerol. When the availability of propionate proves insufficient, gluconeogenesis makes use of body reserves in the form of fat and proteins via the secretion of glucocorticoides, thereby generating large amount of acetyl coenzyme A **(Brus, 1989)**.

Laboratory findings revealed that the increase in the activities of serum enzymes (AST, ALT & ALP) in diseased goats is an evidence for the degree of hepatic damage. These results were coincided with that reported by **El-Sebaie et al. (1992), Nasser et al. (1998) and Mohamed et al. (2005)**. This is in accordance with the fact that in the event of energy deficiency, the body uses its fatty tissue reserves as a source of energy. Thereby leading to important lipolysis, which in turn increases the presence of circulating free fatty acids that reach the liver and induce fatty infiltration with subsequent liver degeneration **(Brus, 1989 and Radostits et al., 2000)**.

The elevated values of serum urea and creatinine in diseased goats can be explained by the observation of **Parry** and **Tylor (1956)** who found fatty infiltration in the tubular epithelium of the kidneys of ketotic ewes. **West (1996)** and **Makuyana et al. (2002)** attributed that increases to reduced glomerular filtration as result of extensive degenerative changes of the kidneys.

The results obtained for serum glucose indicate that the diseased goats were hypoglycemic. These results could be confirmed with those obtained by **Storry** and **Rook (1962)** and **Jonsson** and **Pehrson (1972)** who found that the glucose levels are related to the animals energy status. values falling with a negative energy balance, while **Ford et al. (1990)** and **West (1996)** attributed that to the diminished glucose supply from increasing fetal demand as pregnancy progresses and decreased glucose production due to undernourishment. Schlumbohm and **Harmeyer (2004)** recorded that high ketone body concentrations suppress the endogenous production of glucose by approximately 30% and this facilitate the development of pregnancy toxaemia in pregnant sheep.

Concerning the lipogram picture, the results revealed significant ($P<0.01$) reduction of cholesterol and triglycerides levels and significant ($P<0.01$) increase of total lipids with significant ($P<0.001$) increase of free fatty acids in diseased goats as compared with healthy ones. The reduction of the cholesterol and triglycerides levels is may be attributed to the reduction of glucose level that participate in the formation of glycerol and triglycerides. In addition the liver infiltration with large amount of free fatty acids with the subsequent development of fatty liver have resulted in the inability of the liver to re-esterify fatty acids into triglycerides (**Hay** and **Baird, 1991**; **Radostits et al., 2000** and **Latimer et al., 2003**). On the other hand, the increase of to-

tal lipids level is due to the mobilization of free fatty acid from fat depots to be used as source of energy through increased lipolysis and glyconeogenesis in a trial to compensate the reduction of the glucose level (**Brus 1989** and **Henze et al., 1998**). The sharp increase in the concentrations of free fatty acids in affected goats could be attributed to the increased mobilization of fatty acids from the adipose tissues in response to an increased requirement for endogenous substrate for energy production during pregnancy (**Noble et al., 1971**). **Russel et al. (1967)** suggested that plasma free fatty acids would be the most useful index of the degree of undernourishment in pregnant ewes.

As shown in Table (1), the β-OHB revealed significant ($P<0.001$) increase in affected goats as compared to the healthy ones. This observation is in accordance with **Henze et al. (1998)** in sheep and with **Sackran et al. (2004)** in goats. Pregnancy disease is a condition which can be diagnosed by clinical examination of ewes with signs of hepatic encephalopathy which can be confirmed biochemically by marked increase in serum concentration of β-hydroxybutyrate and a corresponding decrease in plasma glucose concentration (**Scott et al., 1995**).

Concerning the serum cortisol level, it showed a marked increase in diseased animals when compared to healthy ones. **Kimberling (1988)** postulated that the stress and low caloric intake in pregnancy toxaemic animals have a profound effect on the kidney and adrenal gland with a significant reduction in renal blood flow and glomerular filtration rate which raise plasma renin activities and elevate plasma cortisol levels. Another explanation, the increase of serum cortisol level may be the consequence of increased glucocorticoids output in response to stress or reduced excretion by the liver (**Ford et al., 1990 and Henze et al., 1998**).

Regarding the level of insulin hormone, it showed a significant ($P<0.01$) decrease in the diseased goats. The drop of the insulin level may be a reflex of decreased glucose level for facilitating the hepatic synthesis of glucose (**Hart et al., 1978**). In late pregnancy, pancreatic release of insulin tend to decrease resulting in lower blood glucose level and stimulated synthesis of ketone bodies in the liver (**Vernon et al., 1981 and Fernandez & Lee, 1987**).

Kimberling (1988) described that the total feed requirements for the single bearing ewe during the last 6 weeks is 1.5 x maintenance and 2 x maintenance for a twin bearing ewe. The additional energy for this period is best supplied by concentrate feeds as the rumen capacity is limited by fetal expansion. Accordingly the animals included in this study receive low feed requirements (rice straw, grass and less concentrates) which led to the occurrence of this condition.

It could be concluded that, the disturbance in ration constituents especially carbohydrates leads to pregnancy disease which can be prevented by the subsequent correction of carbohy-

drate and lipid parameters in affected goats with balancing the nutritional intake of the goats with the increased late pregnancy needs of the dam and her fetus to avoid and prevent the occurrence of this syndrome.

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Table (1): Some laboratory findings in both apparently healthy and diseased goats before and after therapeutic treatment (mean±S.E).

Parameter		Healthy goats (n=8)	Diseased goats (n=12)	
			Before treatment	After treatment
AST	(U/L)	48.75±1.55	60.08±2.16***	55.83±2.54*
ALT	(U/L)	23.62±0.75	32.25±1.56***	26.58±0.89*
ALP	(U/L)	41.37±1.70	53.75±1.84***	47.91±2.20*
Urea	(mg/dl)	27.63±1.16	35.41±1.72**	29.75±1.23
Creatinine	(mg/dl)	1.41±0.12	1.95±0.12**	1.57±0.10
Glucose	(mg/dl)	53.87±2.54	42.75±2.29**	49.83±2.12
Cholesterol	(mg/dl)	67.50±2.06	53.91±3.20**	62.41±2.25
Triglycerides	(mg/dl)	53.62±3.02	38.33±2.94**	48.08±2.18
Total lipids	(mg/dl)	318.75±12.11	389.91±17.62**	346.25±14.01
Free fatty acids	(mg/dl)	21.62±1.66	36.50±2.24***	27.41±1.75*
-OHB	(mg/dl)	6.37±0.73	11.83±1.06***	8.91±0.95*
Cortisol	(ng/dl)	15.12±1.06	30.33±2.76***	20.16±1.59*
Insulin	(ng/dl)	7.62±0.65	5.01±0.24**	6.80±0.21

* P<0.05 **P<0.01 *** P<0.001

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الملخص العربى

بعض التغيرات البيوكيميائية المصاحبة لنقص الكربوهيدريت فى الماعز الحامل مع الإشارة المرجعية لتسمم الحمل

عادل على كامل

معهد بحوث صحة الحيوان

الهدف من هذه الدراسة هو استبيان بعض التغيرات البيوكيميائية والمصاحبة لنقص الكربوهيدريت فى الماعز الحامل ومدى الاستجابة للعلاج. اشتمل هذا البحث على فحص عدد ٤٠ من الماعز البلدية فى الأسابيع الأخيرة من الحمل باحدى المزارع الخاصة بمحافظة الشرقية، كانت تعانى من فقدان الشهية والخمول والرقاد. تم جمع عينات دم لفصل المصل من عدد ١٢ ماعز حامل (المجموعة المريضة) قبل وبعد العلاج بالإضافة إلى عدد ٨ ماعز سليمة بحالة صحية ظاهرية جيدة (المجموعة الضابطة).

أوضحت نتائج التحاليل البيوكيميائية لمصل الدم على انخفاض معنوى فى معدلات كل من الجلوكوز والكلوستيرول والدهون الثلاثية وهرمون الأنسولين فى الماعز المريضة بمقارنتها بالسليمة. بينما حدثت زيادة معنوية فى أنشطة إنزيمات الكبد)الاسبارتيت أمينوترانسفيريز، الألانين أمينوترنسفيريز، الألكالين فوسفاتيز) وأيضا فى مستوى البروتينا والكرياتينين ، بالإضافة إلى الدهون الكلية، الأحماض الدهنية والبيتاهيدروكسى بيوتيريت والكورتيزون فى الحيوانات المريضة بمقارنتها بالسليمة. أما نتائج تحليل البول فقد أوضح وجود ارتفاعا ملحوظا فى معدل الأجسام الكيتونية فى العنزات المريضة مقارنة بالسليمة. تم استخدام نظام علاجى فى الحالات المريضة وكانت الاستجابة متفاوتة حسب مرحلة الاصابة.

خلصت الدراسة إلى أن الاختلال فى مستوى بعض المكونات الغذائية وخاصة الكربوهيدرات والدهون يؤدى إلى مرض الحمل أو تسمم الحمل فى الماعز. وبالامكان تجنب تكرار هذا عن طريق توفير الغذاء المتوازن والمتكامل لجميع العناصر للماعز العشار للوفاء باحتياجاتها واحتياجات جنينها طوال فترة الحمل.