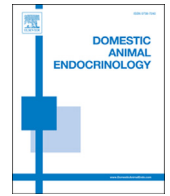




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## Short Communication

# Relationship between serum adiponectin concentration, body condition score, and peripheral tissue insulin response of dairy cows during the dry period



J. De Koster<sup>a,\*</sup>, C. Urh<sup>b</sup>, M. Hostens<sup>a</sup>, W. Van den Broeck<sup>c</sup>, H. Sauerwein<sup>b</sup>, G. Opsomer<sup>a</sup>

<sup>a</sup> Department of Reproduction, Obstetrics and Herd Health, Faculty of Veterinary Medicine, Ghent University, Salisburylaan 133, 9820 Merelbeke, Belgium

<sup>b</sup> Institute for Animal Science, Physiology and Hygiene Unit, University of Bonn, 53115 Bonn, Germany

<sup>c</sup> Department of Morphology, Faculty of Veterinary Medicine, Ghent University, Salisburylaan 133, 9820 Merelbeke, Belgium

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## ABSTRACT

The aim of the present study was to describe the relationship between serum adiponectin concentration and peripheral tissue insulin response in dairy cows with a variable body condition score (BCS) during the dry period. Cows were selected at the beginning of the dry period based on BCS (BCS <3.75, n = 4; BCS >3.75, n = 5). Animals were followed from the beginning of the dry period by weekly blood sampling and assessment of BCS and backfat thickness. Weekly blood samples were analyzed for adiponectin concentration using a bovine specific ELISA. Hyperinsulinemic euglycemic clamp tests were performed at the end of the dry period to measure peripheral tissue insulin response. Insulin dose response curves were established for both glucose and fatty acid metabolism. Regression analysis revealed that the serum concentrations of adiponectin dropped at the end of the dry period ( $P < 0.05$ ) and were negatively associated with BCS ( $P < 0.05$ ). At the level of the glucose metabolism, serum concentrations of adiponectin were positively correlated with insulin responsiveness (reflecting the maximal effect of insulin;  $r = 0.76$ ,  $P < 0.05$ ), but not with insulin sensitivity (reflecting the insulin concentration needed to achieve halfmaximal effect;  $r = -0.54$ ,  $P = 0.13$ ). At the level of the fatty acid metabolism, greater adiponectin concentrations were negatively correlated with lower NEFA levels during the HEC test reflecting the insulin responsiveness of the NEFA metabolism ( $r = -0.61$ ,  $P = 0.08$ ), whereas there was no association with the insulin sensitivity of the NEFA metabolism ( $r = -0.16$ ,  $P = 0.67$ ). In conclusion, serum concentrations of adiponectin were negatively associated with the BCS of dairy cows during the dry period and positively associated with insulin responsiveness of the glucose and fatty acid metabolism.

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## 1. Introduction

Adiponectin, a protein exclusively produced by adipocytes, exerts insulin sensitizing and anti-inflammatory properties in different tissues and is recognized to play an

important role in the pathogenesis of the human metabolic syndrome [1,2]. Obesity, in humans and rodents, is associated with decreased circulating concentrations of adiponectin and a downregulated expression of AdipoR1 and AdipoR2. Both factors contribute to a disruption of normal metabolic function of different tissues which eventually will lead to the development of insulin resistance, type 2 diabetes mellitus, and cardiovascular disease [1,3,4].

\* Corresponding author. Tel.: +32 9 2647528; fax: +32 9 2647797.

E-mail address: [jenne.dekoster@ugent.be](mailto:jenne.dekoster@ugent.be) (J. De Koster).

Overconditioned dairy cows are known to be more susceptible for metabolic and infectious disorders, which is known as the fat cow syndrome [5,6]. Similarly to the human metabolic syndrome, the adipose tissue is an important contributor in the development of the fat cow syndrome due to the excessive release of fatty acids [6,7]. The adipose tissue of dairy cows is also capable of producing different adipokines, including adiponectin [8–10]. However, the metabolic role of adiponectin in dairy cows is not yet fully explored. Recent *in vitro* research demonstrated that adiponectin decreases tumor necrosis factor  $\alpha$  production by bovine monocytes [11], whereas it stimulates lipid oxidation in bovine hepatocytes [12].

In a previous article, we described the relationship between the accumulation of body fat and glucose and fatty acid metabolism of peripheral tissues in response to insulin. Overconditioned cows were shown to have a decreased insulin sensitivity (increased EC50glucose) and a decreased insulin responsiveness (decreased maxglucose) of the glucose metabolism, whereas insulin action at the level of the fatty acid metabolism was not influenced by BCS [13]. The aim of the present study was to describe the relationship of the serum adiponectin concentration during the dry period with: (a) BCS, (b) adipose depot weight, and (c) variables derived from the insulin dose response curves of the glucose and fatty acid metabolism for cows of variable BCS (BCS 3–5) during the dry period.

## 2. Materials and methods

All experimental procedures were approved by the ethical committee of the Faculty of Veterinary Medicine (EC2010/149—University Ghent, Belgium). The study design is described in detail by De Koster et al [13]. Ten clinically healthy, pregnant Holstein Friesian dairy cows were selected at the beginning of the dry period based on BCS according to the scale of Edmonson et al [14]. The upcoming parity number of the animals was 2 ( $n = 4$ ); 3 ( $n = 4$ ); 4 ( $n = 1$ ), and 5 ( $n = 1$ ). Five animals were considered to have a normal BCS (BCS <3.75) and 5 animals were considered to be overconditioned (BCS >3.75). Cows were followed starting 2 mo before the expected parturition date by weekly assessment of BCS, backfat thickness, and weekly blood sampling. In the third week (–21 to –17 d) before the expected parturition date, cows were weighed and catheters (Cavafix Certo 338–14G, B. Braun, Instrulife, Oostkamp, Belgium) were placed in both jugular veins. The next day, the animals underwent a hyperinsulinemic euglycemic clamp (HEC) test. Water and hay were always available but corn silage was withheld from 12 h before until the end of the test. The HEC tests were performed as described in detail by De Koster et al [13]. Briefly, after assessment of basal blood glucose concentration, 4 consecutive insulin infusions were administered at increasing doses of insulin: 0.1, 0.5, 2, and 5 mIU/kg per min (Actrapid 100 IU/mL, human recombinant insulin, Novo Nordisk, Bagsvaerd, Denmark). At regular time points, blood glucose concentration was determined using a hand-held glucometer (Precision Xceed, Abbott Diabetes Care, Verdifarm, Beringen-Paal, Belgium) and compared with the basal blood glucose concentration measured before the start of

the HEC test. When the blood glucose concentration dropped, the speed of the concomitant glucose infusion (30% glucose, Eurovet, Verdifarm, Beringen-Paal, Belgium) was increased to keep the blood glucose concentration near basal levels. A steady state was reached when no or minor changes of the glucose infusion (CV <10%) were necessary to keep the blood glucose concentration constant and near basal levels for at least 30 min. During each steady state, the steady state glucose infusion rate (SSGIR), the steady state insulin concentration (SSIC), and the steady state NEFA concentration were calculated as the average GIR, the average insulin concentration and the average NEFA concentration during the steady state period. Within 2 h after collection, all blood samples were centrifuged for 20 min ( $2,460 \times g$ ,  $7^\circ\text{C}$ ) and stored at  $-80^\circ\text{C}$  until analysis. Samples for NEFA and insulin determination were taken in gel-coated blood tubes (Vacutest, Novolab, Geraardsbergen, Belgium). Serum insulin concentrations were determined using a human specific insulin electrochemiluminiscent immunoassay (ECLIA, Roche, Basel, Switzerland), intra-assay and interassay CV were 1.1% and 6.0%, respectively. Serum NEFA concentrations were determined in a commercial laboratory (Mediclab, Aalst, Belgium) using an enzymatic endpoint method, intra-assay and interassay CV were 1.0% and 1.1%, respectively. Serum adiponectin concentrations were determined by an indirect, competitive bovine specific ELISA [9], intra-assay and interassay CV were 4.5% and 5.6%, respectively.

One week after the HEC test (–13 to –10 d before expected parturition date), cows were euthanized at the Department of Morphology (Faculty of Veterinary Medicine, Ghent University, Belgium). The day before euthanasia, live BW of the cows was determined using a large animal floor scale (Bascules Robbe NV, Torhout, Belgium). Immediately after euthanasia, adipose tissue was dissected from the 4 major adipose depots (subcutaneous, abdominal, intrapelvic, and thoracic), weighed and subdivided as described in detail by De Koster et al [13]. The subcutaneous adipose depot contains all the adipose tissue that is located subcutaneously at the back, the sternum, and the tailbase (including the adipose tissue in the fossa ischiorectalis). The abdominal adipose depot contains all the adipose tissue that is located in the omentum majus, omentum minus, mesenterium, and, the perirenal, and retroperitoneal adipose tissue. The intrapelvic adipose depot contains all the adipose tissue that is located in the pelvic cavity. The thoracic adipose depot contains all the adipose tissue that is located at the inside of the ribs, in the mediastinum, and, around the heart (including the adipose tissue in the coronary grooves).

All statistical analyses were performed using SAS version 9.4 (SAS Institute Inc, Cary, North Carolina, USA). One cow (BCS = 2.82) was excluded from final statistical analysis because steady state conditions during the HEC test were not reached in 2 of the 4 insulin infusion periods of the HEC test. Body condition score and BFT during the dry period were modeled using the PROC MIXED function with cow as random factor and time points during the dry period as repeated measurements within cow. Significant differences between time points were checked using the Bonferroni adjustment for multiple comparisons. For each

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