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Polymorphism of the somatotropic axis genes in cattle – physiology and productivity

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Aim and introduction

This paper is a review of the possible use of indicator traits in dairy cattle for increasing the rate of genetic improvement. The indicator traits considered are related to hormones involved in physiological regulation of metabolism. The genes coding for such hormones have various types of polymorphisms, and are considered as candidate genes influencing both the hormones and ultimately the production traits.

Hormones of the somatotropic axis receive special attention for several reasons. First of all, it was revealed as early as 1937 that injections of pituitary extracts to lactating cows stimulated milk production [Azimov and Krouze 1937]. The increase was

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approximately 20 percent and lasted only few days. Purified synthetic bovine somatotropin (BST) derived from genetically modified cell using recombinant techniques have confirmed the early results in several studies [e.g. Bauman and Vernon 1993], also giving approximately 20 percent more milk during treatment. Not only milk production, but also growth rate in several species including pigs, cattle, humans, and chicken can be boosted by therapeutic administration of GH. Furthermore, it has been observed that mammary gland development in cattle is influenced by GH treatment [Sejrsen et al. 1986]. Consequently, GH is the obvious candidate for a physiological indicator trait for several production traits in cattle and in other species. However, GH does not work alone but in company with several other hormones, receptors and binding proteins. These may together form a portfolio of indicators that could be combined to best aim at a specific trait.

**Synthesis and release of growth hormone**

Growth hormone is synthesised in the pituitary gland, and coded for by a single gene consisting of five introns (Fig. 1). Several base substitution polymorphisms have been detected both in the promoter and the coding and non-coding regions. One of the base substitutions gives rise to an amino acid change at position 127 in the peptide, where a leucine is exchanged for valine. As this mutation is functional, it has been the subject of a number of studies, and among them, its possible value as genetic marker for both GH itself, and for production traits.

Growth hormone is released from the anterior pituitary to the circulation in secretory bursts in response to signal peptides from the hypothalamus (Fig. 2). The signal peptides are growth hormone releasing hormone (GHRH), and somatostatin (SS) which inhibits secretion. In cattle and chicken, GH is also released by thyrotropin releasing hormone (TRH). A synergistic interaction between GHRH and TRH has been described, giving rise to a multiplicative amplification of the release in presence of light, and only an additive effect during darkness [Lapierre et al. 1987]. The peptide hormone ghrelin originating from stomach cells also have GH-releasing activity in several species [Kojima et al. 1999]. This peptide responds to meal feeding and is as such related to appetite.

The GH released to circulation has a direct effect on the liver where it is a signal
to synthesis and release of Insulin like Growth Factor-1 (IGF-1). IGF-1 acts directly on mammary gland tissue during lactation. IGF-1 affects other tissues and supports accretion of protein into muscles. The actions of IGF-1 are modulated by its binding proteins, so that only a small IGF-1 fraction circulates as the biologically active “free” form. Important to the effects of GH is the population of receptors sitting in the liver cell membranes and transferring the signal by binding to free IGF-1 molecules. Direct effects of GH include the support of bone length growth. Furthermore, GH has a direct effect on adipose tissue where it inhibits accretion of fat by increasing lipolysis.

The pituitary also synthesises other hormones influencing metabolism, (Thyroid Stimulating Hormone – TSH), reproduction hormones (Follicle Stimulating Hormone and Luteinizing Hormone) and hormones controlling stress response (ACTH). Each of these peptide hormones is the key regulator for its own axis, similar to the somatotropic axis. Moreover, there are obviously interactions between these axes, as indicated by GH being released in some species not only by GHRH but also by TRH, which is the hypothalamic peptide, primarily aimed at TSH. Therefore, studies of hormones in circulation will most often require measurements of more than the target hormone in order to obtain a more complete picture. Among the important secondary responses is the glucose-insulin system.

**Growth hormone secretion in juvenile cattle selected for high or low milk yield**

In a number of experiments it was established that circulating GH in lactating cows was higher in those having high milk yield [e.g. Klemetsdal et al. 1992]. It was also established that secretion of GH in response to TRH was higher in cows from lines selected for high yield than in control line cows. These observations were in good agreement with the treatment effects of exogenous GH. However, in order to be useful
as predictor of dairy merit, measurements of GH need to be performed before the age of selection, which is at about one year.

The other obstacle to GH measurements is the nature of the random secretory spikes causing plasma concentrations to be very noisy as indicated by the two calves in Figure 3, where blood was sampled at 20-minute intervals over 24 hours (data from L. Hanninen, personal communication).

Pulses can be characterized using various criteria and detection algorithms. Mainly, these give the number and the size of the pulses, and the baseline or nadir level between pulses. Woolliams et al. [1993] found that British Friesian calves of lines selected for high milk yield had slightly higher baseline GH than those from a low-yield line. Through a similar pulse analysis approach Klindt [1988] found a negative association between GH pulse amplitude and frequency and breeding value for milk yield in 26 mature Holstein bulls. Although results were promising, the testing procedure required many samples and was labour-consuming.

Testing of GH response to a standardized stimulation had been applied in humans for diagnosing GH deficiency as malfunction of the pituitary, using arginine infusions. Loevendahl et al. [1991] took such approach and used arginine, TRH, GHRH, and a combined dose of GHRH and TRH as acute stimuli for GH secretion in British Friesian calves of lines divergently selected for high or low milk yield. The GH response to both GHRH and TRH stimulation given separately was clearly larger in calves from “high” than from “low” line. Discrepancy between lines was smaller using arginine and not...
significant when GHRH was given combined with TRH. The results were confirmed in another cohort of British Friesians from the same selection lines [Woolliams et al. 1993], and in Red Dane calves using arginine or TRH [Loevendahl and Sejrsen 1993]. However, the US-Holstein high- and low-milk yield-selected lines used first by Massri et al. [1985] and later by Baumgard et al. [2002] did not respond differently to GHRH stimulation. Similarly, the results of Loevendahl and Klemetsdal (Fig. 4) based on Norwegian cattle selection lines stimulated to release GH following a range of GHRH doses and TRH showed no difference between lines. Based on the published evidence from selection line experiments, there is either a weak positive or no association between induced GH release and breeding value for milk yield.

Effects of L/V polymorphism in population studies of GH release in juvenile dairy cattle

The leucine/valine (L/V) polymorphism has been studied as a genetic marker using a candidate gene approach, for milk yield [e.g. van der Werf et al. 1996]. However, the direct effect on GH release has been studied in Polish Friesians by Grochowska et al. [2001], and in Danish Holsteins, Red Danes and Jerseys by Soerensen et al. [2002]. In Polish Frisians the VV genotype was associated with the largest release of GH in response to TRH [Grochowska et al. 2001].

The allele frequency differs between breeds, with Holsteins having low frequency of V compared with Jerseys that have almost equal frequency of the L and V alleles (Tab. 1). In Jerseys genotype LL-calves of both sexes released more GH in response
to GHRH than VV, with an intermediate response in LV heterozygotes [Soerensen et al. 2002]. However, in a larger cohort of Jersey calves this relationship was not confirmed (Loevendahl and Holm, unpublished). The recent findings question the results of the candidate gene approach, at least when many modifying factors in the studied axis remain unknown.

Polymorphisms in other genes of the growth hormone axis, namely the GH receptor (GH-R) and the IGF-1 gene, have been studied as candidate genes. Recently, Grochowska et al. [2002], demonstrated phenotypic inter-breed differences expressed as efficiencies of the GH-R in its binding constants. These findings in general support that GH axis is involved in regulation of metabolism during lactation, and that its components may be valuable as indicator traits. However, it is also a demonstration that a broad range of systems needs to be studied to gain sufficient insights.

**Combined use of more indicator traits and single gene polymorphisms**

More recently a project aiming at developing physiological predictors has been conducted at DIAS, using future AI-bulls and heifers from the Future Genetics (DK) nucleus herd. Juvenile animals were tested at 9 months of age using a panel of physiological challenges, including a GHRH stimulation test, an adrenaline stimulation test, and a glucose tolerance test. Preliminary results indicate that response variables have intermediate to high heritability [Loevendahl and Soerensen 2001]. From a similarly designed but smaller experiment we also have similar results. By combining information from into a “physiological index”, an accuracy of $r_{IA} = 0.16$ was obtained in heifers, and $r_{IA} = 0.07$ in 142 AI-bulls [Soerensen et al. 2000]. This accuracy of prediction is comparable to the use of MAS suggested by others [Boichard et al. 2002], and could be applied immediately. By combining MAS and physiological predictors a further improvement in accuracy should be achieved.

**Conclusions**
Although polymorphisms in the GH gene itself and in GH-R gene have some physiological effects, these are not consistent across age or time and sub-populations of dairy cattle. Physiological indicators based on GH alone carry insufficient information for reliable genetic selection in juvenile cattle. However, by combining QTL-based and a number of physiological indicators into an index, this will provide sufficient information to facilitate marker-assisted selection.

REFERENCES


