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TOTAL THYROXINE AND FREE THYROXINE INDEX IN PLASMA OF DAIRY COWS IN RELATION TO STRENGTH OF HEAT*

By

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ANDRESEN, Ø., J. AMRUD, L. E. GRØHOLT, G. HELLAND, K.-A. SCHIE & G. A. SYLLIAS: *Total thyroxine and free thyroxine index in plasma of dairy cows in relation to strength of heat.* Acta vet. scand. 1980, *21*, 108—112. — Total thyroxine (TT_4) and free thyroxine index (FT_4I) were measured in peripheral plasma of cows. The samples were collected at the time of insemination from 66 cows showing pronounced signs of the heat and from 56 cows showing weak or silent heat. Neither TT_4 or FT_4I in plasma differed significantly between the two categories of oestrous cows.

cow; oestrus; thyroxine; free thyroxine index.

"Silent heat" in dairy cattle is a problem in many herds and leads to delays of inseminations. Although the main reason for the occurrence of "silent heat" may be inadequate heat observation (*De Kruif* 1978), several other factors may also be involved, and among these there are indications that thyroxine could be of importance. Thus it has been reported that thyroidectomized cows do not display the usual signs of oestrus (*Spielman et al.* 1945). Nevertheless it was found by rectal palpation of the ovaries of these cows that ovulation occurred at regular intervals, and insemination at the time of ovulation was followed by conception. This could indicate that the thyroxine levels differ in cows showing strong and silent heat.

As more than 99.9 % of the thyroxine in plasma is bound to protein (*Refetoff et al.* 1970), the total level of thyroxine may

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be altered without any appreciable change in the level of free, physiologically active hormone. Besides measuring total thyroxine, a thyroid hormone uptake test was therefore also performed and a free thyroxine index calculated.

MATERIAL AND METHODS

Sampling

Samples were collected in five districts in southern Norway during the period from 15th of December 1978 to 15th of March 1979. The cows were at least 2nd calvers. The sampling was performed by veterinarians who also carried out the artificial insemination.

At the time of insemination the strength of heat was judged by the veterinarian and by the herdsman in collaboration, and samples of blood were collected from cows showing pronounced signs of heat and from cows showing very weak or silent heat. In general, samples were collected in pairs so that in each herd at least one cow showing pronounced and one with weak/silent heat were sampled. Sampling was performed by the use of heparinized Vacutainers®. On arrival at the laboratory, usually within one to two days, the samples were centrifuged and plasma stored at —20°C until required for analysis.

Analyses

Total thyroxine (TT_4) was measured by a modification of the radioimmunoassay described by *Larsen et al.* (1973). The buffer used was 0.04 M barbitalbuffer, pH 8.0, with 0.1 % merthiolate added. Ten µl aliquots of plasma were used in the assay, and all samples were analysed in duplicate. The properties of the antiserum have been described by *Kruse* (1976).

The intra-assay coefficients of variation for 7 samples analysed with 10 parallels were 4.4, 4.6, 6.5, 6.5, 6.7, 8.5 and 8.6 %. The inter-assay variation for the same samples analysed in duplicate on 7 different days were 5.3, 5.7, 5.9, 6.0, 7.4, 8.9 and 9.5 %.

The thyroid hormone uptake test was performed by the use of T_3 Uptake Diagnostic Kit (Nuclear Medical Laboratories Inc., Dallas, Texas), and the free thyroxine index (FT_4I) was calculated as the product of the T_3 uptake and the TT_4 value.

RESULTS AND DISCUSSION

The total thyroxine level in plasma was found to vary between cows, and levels from 24 nM/l to 76 nM/l were recorded. These levels are a little lower than those cited by *Slebodzinski* (1975), but the mean level (Table 1) is about the same as the one reported by *Strbák et al.* (1976) during late pregnancy in 23 cows.

Table 1. Total thyroxine (TT_4) and free thyroxine index (FT_4I) in plasma of cows showing strong and weak/silent heat, respectively (mean ± standard deviation).

District	Strong heat			Weak/silent heat		
	n	TT_4 (nM/l)	FT_4I	n	TT_4 (nM/l)	FT_4I
1	10	51.8	53.0	5	42.4	44.1
2	9	41.4	39.0	8	38.0	34.7
3	12	46.8	47.2	6	49.0	48.6
4	16	47.9	49.9	17	50.1	50.1
5	19	48.5	46.5	20	46.3	46.1
Mean	66	47.6	47.4	56	45.6	46.1
s		± 11.3	± 11.6		± 10.2	± 10.4

No significant difference either in total thyroxine level or in free thyroxine index could be revealed between cows showing strong and weak/silent heat (Table 1). The free thyroxine index was in general close to the thyroxine level, indicating that primary changes in the level of thyroid binding proteins were not the major reason for the variations in total thyroxine.

The time elapsing from calving to insemination varied from 25 to more than 150 days. The total thyroxine levels in plasma of cows in relation to days after calving did not differ between the two categories of cows (Fig. 1).

From the present study it appears that there is no simple correlation between strength of heat and plasma level of thyroxine. An explanation for this lack of relationship could be a varying sensitivity for thyroid hormones among cows. However, when the usual signs of heat are missing in thyroidectomized cows, it is more likely that a minimum level of thyroxine is necessary

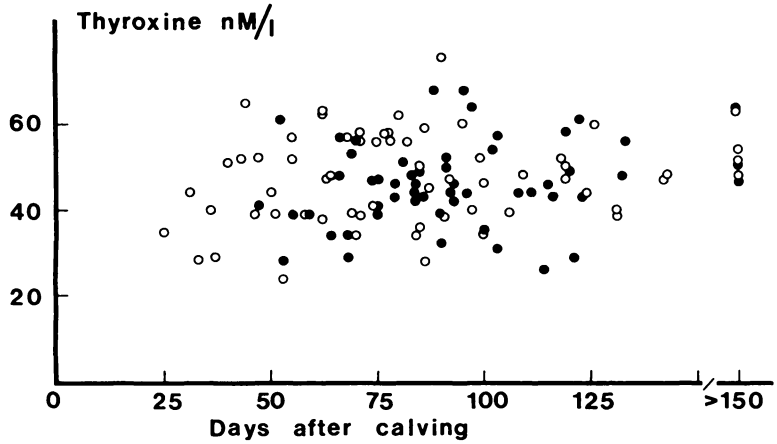

Figure 1. Total thyroxine in plasma of cows at the time of insemination in relation to time after calving.

o : cows showing strong heat,

• : cows showing weak or silent heat.

for the animal to come into heat. According to the present study this level must be rather low, as thyroxine levels of 20—30 nM/l plasma were occasionally observed in cows showing strong heat.

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SAMMENDRAG

Tyroksin og fri tyroksin indeks i kuplasma i relasjon til brunststyrke.

Total tyroksin (TT_4) og fri tyroksin indeks (FT_4I) ble målt i perifert plasma fra kuer. Blodprøver ble tatt fra 66 kuer som viste utpregede psykiske brunsttegn og fra 56 kuer som hadde meget svak eller stille brunst. Blodprøvene ble tatt samtidig med at kuene ble inseminert. Det ble ikke funnet noen forskjell hverken i TT_4 eller i FT_4I mellom de to grupper av dyr.

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