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PLASMA LEVELS OF TRIIDOTHYRONINE, THYROXINE AND CORTISOL IN NEWBORN CALVES

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The aim of this investigation was to determine the concentrations of triiodothyronine (T3), thyroxine (T4) and cortisol in blood plasma from calves during the first 32 h of postnatal life, as well as at 7 days old. The experiment involved two groups of calves. The first group was offered the standard amount of colostrum while the second group received half the recommended amount. The results obtained showed that the calves were born with high plasma concentrations of T3, T4 and cortisol up to four times greater than values found at 7 days old. In the calves given the full ration of colostrum there was a gradual increase of mean T3 concentration to levels which plateaued from 8 to 28 h after birth, while the level of T4 increased slightly but significantly up to 18 h. Sharp highly significant increases in T3 and T4 concentrations were observed during the first 4 h of postnatal life in the group of calves given half the usual amount of colostrum to levels which persisted up to 20 h. Except at 4 and 20 h there were no significant differences between the groups. At 7 days old the levels of plasma T3 and T4 in both groups of calves were within the limits found for adult animals.

The plasma concentrations of cortisol decreased during the first 18 h after birth in both groups of calves and then increased somewhat. At 32 h the mean concentration was higher in group two than in group one. At 7 days old cortisol concentrations were up to four times lower than the level immediately after birth.

It was concluded that a decrease in the amount of colostrum consumed had only minor effects on the plasma concentrations of the examined hormones in neonatal calves.

Key words: calves, colostrum, cortisol, thyroxine, triodothyronine,

INTRODUCTION

Examination of the functional status of the thyroid gland in the perinatal period has been the subject of many investigations (Fisher *et al.*, 1966; Nathanielsz *et al.*, 1969; Hernandez *et al.*, 1972; Davicco *et al.*, 1982; Jovanović *et al.*, 1982). All authors agreed that newborn domestic animals are born with high peripheral levels of thyroid hormones. The influence of factors such as breed (Davicco *et al.*, 1982), nutrition including fasting, an immunoglobulin diet and colostrum (Grongnet *et al.*, 1985; Hammon and Blum, 1998) on thyroid hormone levels during the first 28 hours of postnatal life has been studied. However, from the results presented for thyroid hormone levels during the first 36 hours after birth by these and many other authors, it is apparent that there are inconsistencies, even though animals of the same age and species were concerned (Grongnet *et al.*, 1985; Stanko *et al.*, 1991; Chen *et al.*, 1996).

Prepartum increases in fetal plasma cortisol concentrations are also well documented for many mammalian species, including ruminants, and are related to adaptation of the neonate to extrauterine life (Hunter et al., 1977). Thus, cortisol has been shown to be a physiological regulator of most of the fetal gluconeogenic enzymes, as well as monodeiodination of T4 to T3 in peripheral tissues of lambs towards the end of gestation (Fowden *et al.*, 1993; Sensky *et al.*, 1994). A statistically significant correlation between T3 and cortisol levels on the day of birth was found in Holstein calves (Nikolić *et al.*, 1996). Both hormones may play a role in the regulation of gut permeability and growth (Johnston and Oxender, 1979; Slebodzinski *et al.*, 1995).

The aim of our investigation was to determine the concentrations of T3, T4 and cortisol in blood plasma from dairy calves kept under the same conditions but offered different amounts of colostrum.

MATERIAL AND METHODS

Animals

The experiment involved two groups of six calves of the Holstein-Friesian breed from Jabučki Rit farm (Belgrade Agricultural Corporation). The mean body mass of the calves at birth was 38.4±2.2 kg for group one and 36.7±2.8 kg for group two. All calves were born within 2 days and immediately placed in individual boxes in a byre where the temperature ranged from 18 to 22°C.

Colostral intake

The calves were suckled on pooled colostrum 2 h, 12 h and 24 h after birth through artificial teats. Calves in the first group were offered 1.5 L for the first two meals followed by 2 L colostrum for the third. Calves in the second group received 0.75 L for the first two meals and 1 L for the third.

Blood sampling

Blood samples were obtained by puncture of v. jugularis with a sterilised needle into heparinised tubes. The tubes were immediately centrifuged at 3000 rpm in order to separate out the plasma. Blood samples were taken at birth (Oh) and 4h, 6h, 8h, 16h, 18h, 20h, 28h, 32h and 7 days later.

Analyses

Commercial ^{125}I-RIA kits validated for bovine plasma were used to determine T3 and T4 in accordance with the instructions (Isotope Institute Budapest Hungary). Cortisol was determined by a direct ^{3}H-RIA method (Csernus, 1982). Intra- and interassay CVs ranged from 3.2-8.7 and 4.1-10.3 % respectively.

Statistical analysis

The results are expressed as mean ± standard deviation (SD), standard error (SE) and CV for each group of calves. Probability and the statistical significance of differences between mean values were calculated using Student's t-test and split-plot analysis of variance followed by the least significant difference (LSD) test.

RESULTS

Mean values for T3 and T4 concentrations in blood plasma from the calves in group one are given in Table 1. and 2., respectively. It can be seen that the calves were born with high concentrations of T3 (8.26 nmol/L) and T4 (215.1 nmol/L).

At 4 h after birth and 2 h after the first intake of colostrum mean T4 concentration had increased slightly to 250.4 nmol/L. In comparison with the initial level the difference became statistically significant 6 h after birth ($P < 0.05$). The highest plasma T4 concentration (279.0 nmol/L) occurred at 18 h after birth. The mean level of T4 then decreased, falling below the initial concentration at 28 h after birth to reach levels similar to those in adult cattle at 7 days old (Stojić *et al.* 2001).

In the same group of calves mean T3 concentration increased from 8.26 nmol/L at birth to levels significantly higher during the interval of 8 to 28 hours postnatally. By 7 days after birth plasma T3 had decreased four fold to achieve values within the range found for adult cattle (Fig. 1).

Plasma levels of the examined thyroid gland hormones found in the second group of calves are shown in Table 3. and 4. Mean values differed from those for group one at 4h and 20h after birth ($P < 0.05$).

Thus there were immediate sharp increases in mean T3 and T4 concentrations during the first 4 h of postnatal life from 7.33 to 12.02 nmol/L and 176.2 to 303.8 nmol/L respectively. The second plasma samples were obtained 2h after suckling the calves with half the recommended amount of colostrum (0.75 L). The mean concentrations of T3 and T4 remained markedly higher than the initial values up to 28 h and 20 h of postnatal life respectively. At 7 days of age plasma concentrations of both thyroid hormones in this group of calves were also within the range found for adult animals (Fig. 2). Plasma T3 and T4 concentrations were closely correlated ($r = 0.827$; $P < 0.0001$).

The results for plasma cortisol concentrations for I and II group of calves are shown in Table 5. and 6., respectively. High mean values were observed at birth followed by decreases to a minimum around 18 h of postnatal life. Mean values then increased in both groups, more markedly in group two, which received less colostrum, than for group one. Namely, at 32 h after birth the mean value for group one was half the initial value, while that for group two was only 17% lower (Fig. 3). The difference between the groups at this time was statistically significant ($P < 0.01$). Plasma cortisol concentrations were not correlated with thyroid hormone concentrations in either group.

Table 1 . The mean plasma levels of T_3 (nmol/L) in the I experimental group of calves / colostral intake time ↓

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	8.26	9.76	10.15	11.83	10.24	10.82	10.28	10.50	9.63	9.36	2.22
SD	1.93	1.22	1.17	2.31	0.82	0.83	0.71	0.82	1.27	2.31	1.10
SE	0.79	0.50	0.48	0.94	0.33	0.34	0.29	0.33	0.52	0.94	0.45
CV(%)	23.4	12.5	11.5	19.5	8.0	7.7	6.9	7.8	13.2	2.5	49.5

Table 2. The mean plasma levels of T_4 (nmol/L) in the I experimental group of calves. colostral intake time ↓

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	215.14	250.41	265.02	270.63	268.22	279.03	240.01	200.92	201.64	195.00	74.85
SD	44.97	39.67	45.73	49.94	61.62	58.03	23.44	17.99	20.27	38.33	33.69
SE	18.36	16.19	88.67	20.39	25.16	23.69	9.57	7.34	8.27	15.65	13.75
CV(%)	20.09	15.8	17.3	18.4	23.0	20.8	9.8	8.9	10.1	19.7	45

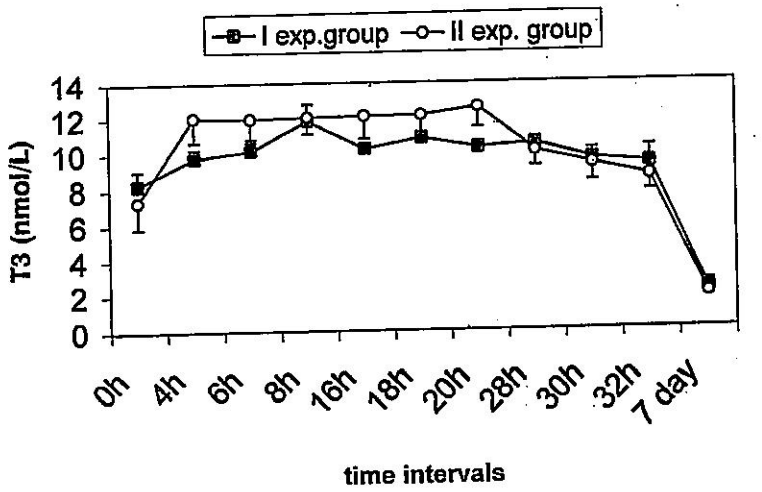

Figure 1. Mean plasma T3 concentrations ($\bar{X}\pm SE$ nmol/L) in I and II experimental group of calves.

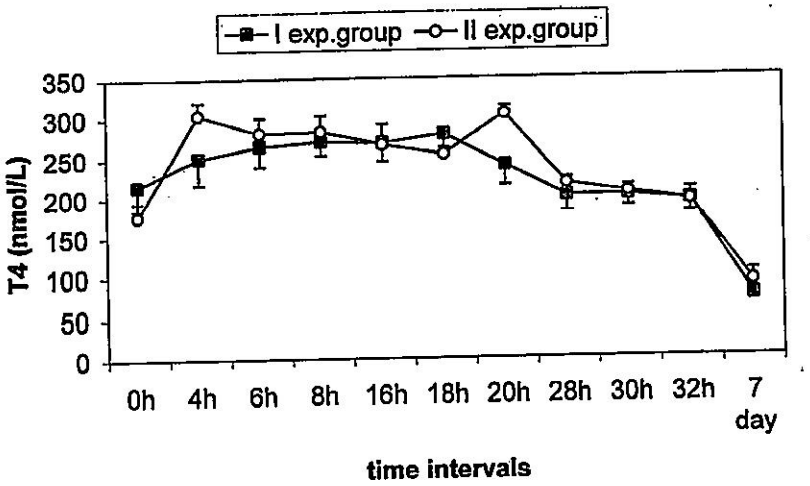

Figure 2. Mean plasma T4 concentrations ($\bar{X}\pm SE$ nmol/L) in I and II experimental group of calves.

Table 3. Table 3. The mean plasma levels of T_3 (nmol/L) in the II experimental group of calves / colostral intake time ↓

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	7.33	12.02	11.96	12.02	12.09	12.11	12.57	10.08	9.33	8.63	1.94
SD	3.64	3.33	2.87	2.27	3.16	2.90	2.87	2.14	2.42	2.06	0.46
SE	1.49	1.36	1.17	0.93	1.29	1.18	1.17	0.87	0.99	0.84	0.19
CV(%)	49.6	27.7	24.0	18.9	26.1	23.9	22.8	21.2	25.9	23.9	23.7

Table 4. The mean plasma levels of T_4 (nmol/L) in the II experimental group of calves/ / colostral intake time ↓

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	176.19	303.84	280.97	282.47	266.39	254.06	304.04	216.35	205.15	193.98	90.96
SD	75.07	81.81	62.36	42.51	55.09	40.47	61.85	47.87	36.29	37.90	18.84
SE	30.63	33.40	25.46	17.35	22.47	16.52	25.25	13.54	14.81	15.47	7.69
CV(%)	42.6	26.9	22.2	15.0	20.7	15.9	20.3	22.1	17.7	19.5	20.7

Table 5. The mean plasma levels of cortisol (nmol/L) in the I experimental group of calves / colostral intake time

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	92.80	67.48	50.65	53.03	38.54	31.14	35.54	56.22	57.9	45.83	20.59
SD	13.62	27.37	15.02	19.56	12.95	14.99	11.71	35.67	21.28	16.23	12.24
SE	6.09	12.24	6.72	8.75	5.79	6.70	5.24	15.95	9.50	7.25	5.46
CV(%)	14.7	40.6	29.6	36.9	33.6	48.1	23.9	63.4	36.7	35.4	59.4

Table 6. The mean plasma levels of cortisol (nmol/L) in the II experimental group of calves./ colostral intake time

	age of calves (hours/days)										
n=6	0h	4h	6h	8h	16h	18h	20h	28h	30h	32h	7. day
$\bar{X}$	84.67	56.04	54.28	52.33	30.97	29.42	46.37	49.76	64.97	70.89	22.96
SD	20.64	17.66	19.41	9.22	5.37	9.24	12.76	16.05	30.61	19.15	5.95
SE	8.43	7.21	7.94	3.76	2.19	3.77	5.21	6.55	12.50	7.82	2.43
CV(%)	24.4	31.5	35.7	17.6	17.3	31.4	27.5	32.2	47.1	27.0	25.9

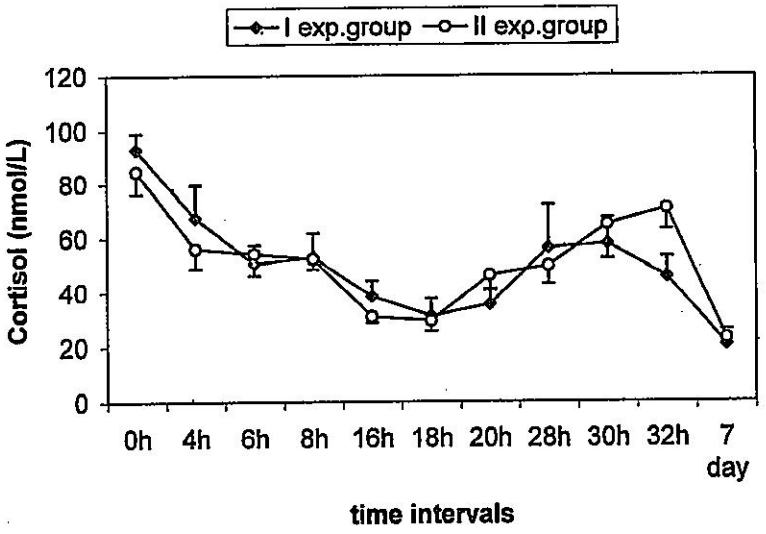

Figure 3. Mean plasma cortisol concentrations ($\bar{X}\pm SE$ nmol/L) in I and II experimental group of calves.

Thus our results indicate that the calves which consumed insufficient amounts of colostrum soon after birth showed rapid increases in thyroid hormone concentrations which were maintained for the first 20 to 28 h of postnatal life. There was also an increase in plasma thyroid hormone concentration in the group of calves which received normal amounts of colostrum but it was more gradual and of lower intensity (Fig. 4 and 5). Nevertheless, there was no indication of a general effect of reduced colostrum intake on T3, T4 and cortisol concentrations ($F = 0.48$, 0.27 and 0.37 respectively) while differences associated with sampling time were highly significant ($F = 28.8$, 24.3 and 7.4: $P < 0.0001$) with no interaction between the variables.

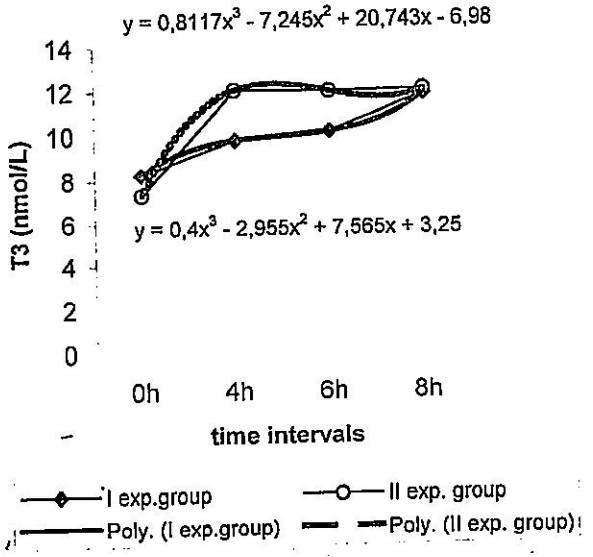

Figure 4. Plasma T3 increase during the first 8 hours of postnatal life with added trendlines.

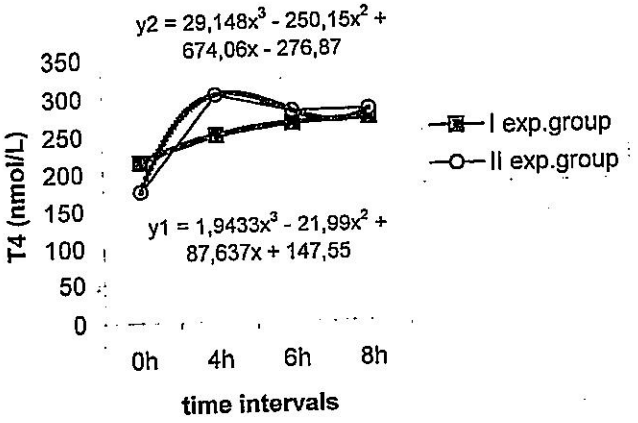

Figure 5. Plasma T4 increase during the first 8 hours of postnatal life with added trendlines.

DISCUSSION

Davicco and coworkers (1982) found lower levels of T4 at 24 h after birth than the initial values. In calves given 25g colostrum per kg body mass, which is a similar amount to that offered to calves in group two in our investigation, Grongnet

and coworkers (1985) found maximal T4 concentrations 24 h after birth, while calves which received less had maximal T4 levels at birth. These authors concluded that colostrum contains an unknown substance which stimulates thyroid function. Our results are not in accordance with this supposition. A brief increase in circulating T3 but not in T4 in calves after colostrum intake was recorded by Baumrucker and Blum (1994). Our study showed marked increases in thyroid hormone levels in both groups of calves during the same time interval but of different intensity. Thyroid hormone concentrations remained slightly elevated up to 32 h after birth. In 2-day-old calves offered limited amounts of milk replacer T3 levels were observed to be higher than in those fed on a full ration of colostrum (Kühne *et al.*, 2000).

At 7 days of age plasma thyroid hormone levels were much lower than at birth. Marked decreases in thyroid hormone concentrations in calves during the first week of postnatal life have been described by other authors (Jovanović *et al.*, 1982; Ronge and Blum 1988; Baumrucker and Blum, 1994; Hadorn *et al.*, 1997; Hammon and Blum, 1998). This phenomenon has been explained in connection with the role of thyroid hormones in energy metabolism during the first week of life (Kinsbergen *et al.*, 1994).

The decline in plasma cortisol concentration during the neonatal period observed here also confirms the findings of other authors (Ronge and Blum 1988; Mao *et al.*, 1994; Hadorn *et al.*, 1997; Hristov *et al.*, 1994; Kühne *et al.*, 2000). The relatively high peripheral cortisol concentration found at 32 h after birth in the group of calves fed on half the recommended amount of colostrum in comparison with the group offered the full ration, is similar to the difference between groups observed at 31 h after birth by Hammon and Blum (1998). Namely, mean cortisol concentration in the group of calves given colostrum as their first meal, followed by milk replacer of lower energy content was higher at that time only than in the group of calves fed colostrum twice daily for 3 days. Brückmann and coworkers (2000) found that 3-day-old calves which later developed spontaneous *E. coli* linked diarrhea had significantly lower plasma cortisol concentrations than those which remained healthy. Nevertheless it should be remembered that total concentrations of cortisol may not reflect its biological activity. Namely, the active free fraction may increase relatively in hyperthermia and acidosis, while the binding activity of corticosteroid binding globulin can be altered differently according to the plasma free fatty acid profile (Obminski and Stupnicki 1996; Haourigui *et al.*, 1995).

Our results indicate the existence of neonatal reserves of thyroid hormones as well as a great potential for thyroid hormone synthesis in newborn calves. In our opinion these large readily available reserves of thyroid hormones are of great use in fulfillment of the considerable metabolic requirements during the first few critical days of neonatal life when the organism of the calf adapts to new environmental conditions. Reducing the amount of colostrum offered to the calves had only minimal effects on the plasma concentrations of the examined hormones.

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KONCENTRACIJA TRIJODTIRONINA, TIROKSINA I KORTIZOLA U KRVNOJ PLAZMI NOVOROĐENE TELADI

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SADRŽAJ

Cilj ovog rada bio je da se odrede koncentracije trijodtironina (T3), tiroksina (T4) i kortizola u krvnoj plazmi teladi tokom prvih 32 sata postnatalnog života kao i 7. dana života. Ogled je izveden na dve grupe teladi: prva grupa teladi je napajana sa normalnom količinom kolostruma, dok je druga grupa dobila duplo manju količinu kolostruma. Dobijeni rezultati ukazuju da se telad rađaju sa visokim vrednostima koncentracije T3, T4 i kortizola u krvnoj plazmi, koje su do četiri puta veće u odnosu na vrednosti ustanovljene 7. dana života. Kod teladi koja su dobijala normalnu količinu kolostruma koncentracija T3 postepeno raste i dostiže plato između 8-28 sata posle partusa, dok nivo T4 u plazmi statistički značajno raste do 18 sata posle partusa. Nagli i statistički visoko značajan porast koncentracije T3 i T4 u krvnoj plazmi tokom prvih 4 sata posle partusa su uočeni kod teladi koja su primila polovinu normalne količine kolostruma, i visoka koncentracija ovih hormona u plazmi se održava sve do 20. sata posle partusa. Nisu ustanovljene statistički značajne razlike u koncentraciji T3 i T4 između prve i druge ogledne grupe, osim 4. i 20. sata posle partusa. U starosti teladi od 7. dana koncentracija tireoidnih hormona u plazmi kod obe grupe teladi se nalazi u okvirima vrednosti nađenih kod odraslih jedinki.

Koncentracija kortizola u krvnoj plazmi opada kod obe ogledne grupe teladi u prvih 18. sati posle partusa, da bi kasnije došlo do blagog porasta koncentracije. U vremenu od 32. sata posle partusa nivo kortizola u plazmi je viši kod druge ogledne grupe u odnosu na prvu. U starosti teladi od 7. dana koncentracija kortizola je četiri puta niža u odnosu na vrednosti ustanovljene neposredno posle partusa.

Mišljenja smo da smanjenje unošenja kolostruma za 50% u odnosu na normalnu količinu ima minimalan uticaj na koncentraciju ispitivanih hormona u krvnoj plazmi novorđene teladi.