The effect of dietary protein on reproduction in the mare. VI. Serum progestagen concentrations during pregnancy

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ABSTRACT

Sixty-four Thoroughbred and Anglo-Arab mares aged 6–12 years were used, of which 40 were non-lactating and 24 lactating. Foals from these 24 mares were weaned at the age of 6 months. Non-lactating and lactating mares were divided into 4 dietary groups each. The total daily protein intake and the protein quality (essential amino-acid content) differed in the 4 groups of non-lactating and 4 groups of lactating mares. The mares were covered and the effect of the quantity and quality of dietary protein on serum progestagen concentrations during pregnancy was studied. A sharp decline in serum progestagen concentrations was recorded in all dietary groups from Days 18 to 40 of pregnancy, with some individual mares reaching values of less than 4 ng/mℓ. Serum progestagen concentrations recorded in some of the non-lactating mares on the low-quality protein diet increased to higher values ($p < 0.05$) than those of mares in the other 3 dietary groups at 35–140 days of pregnancy. A similar trend was observed for the lactating mares on a low-quality protein diet at 30–84 days of pregnancy. No such trends were observed in any of the other dietary groups. High-quality protein supplementation increased serum progestagen concentrations during the 1st 30 days of pregnancy. Lactation depressed serum progestagen concentrations until after the foals were weaned.

Key words: equine, pregnancy, protein nutrition, serum progestagen.

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INTRODUCTION

Conception and maintenance of pregnancy in mammals involves complex interactions of biochemical, endocrine and immunological factors between the embryo, placenta and mother. After fertilisation the embryo is transported through the fallopian tube to the uterine environment where a wide range of proteins and other nutrients are secreted by the endometrial glands to meet the nutritional demands of the embryo before implantation. Simultaneously the cyclical endocrine changes associated with reproduction resume a secretory pattern necessary to maintain pregnancy.

Several hormones, particularly progesterone, play an important role in the maintenance of pregnancy. During early pregnancy, LH concentrations are very similar to concentrations that are normally found during dioestrus[10]. LH concentrations do not increase in response to rising oestrogen concentrations as in the non-pregnant mare[19] and also remain low from 150 days of pregnancy until parturition[19].

It would appear that the cyclical 10–11 d FSH peaks continue during the 1st 78 days of pregnancy [10,14]. Other studies indicate that season might have a major influence on FSH secretion during early pregnancy[11]. At Day 35 of pregnancy, specialised trophoblast cells develop in the allanto-chorionic band around the embryonic sac[5,27] and penetrate the endometrium at Days 35–38 to form endometrial cups[5] that produce PMSG and are of foetal origin[6,22]. Should foetal loss occur before Day 36 of pregnancy, PMSG will not be produced[2]. PMSG appears in the blood from Day 40 and reaches maximum values from Days 55 to 65. Thereafter levels decline to reach low concentrations from 100 to 150 days of pregnancy[1]. Although the precise function of PMSG is unknown, follicular development is not stimulated during early pregnancy[11]. Secondary follicles that develop as early as 15–20 days of pregnancy usually undergo atresia. Large follicles that develop at 35–45 days of pregnancy mature and ovulate with the formation of secondary corpora lutea[3,27]. It is accepted that PMSG might play a role in the ovulatory process and subsequent luteinisation at this stage of pregnancy[3].

After conception, the primary *corpus luteum* (CL) remains active because the presence of the embryo prevents release of luteolytic $PGF2_a$ by the endometrium[18]. The first secondary CL usually develops by Day 35–40 of pregnancy but this could be delayed until Day 60[11]. During the 1st 14 days after ovulation the plasma progestagen concentration of the pregnant mare is very similar to that of the non-pregnant mare[26]. After Day 14 a decline in plasma progestagen occurs due to partial cellular degeneration of luteal tissue of the primary CL[26]. After each secondary ovulation the plasma progestagen concentrations increase and as many as 10–15 additional luteal structures may be present by Day 120–150 of pregnancy before degeneration occurs[24]. At this stage the placenta is well developed and maintains the function of progestagen production. However, in most mares a slight to marked decrease in serum progestagen concentrations may occur by Day 90 due to degeneration of the secondary CL before the placenta has completely taken over the production of progesterone[4].

It is accepted that progesterone plays an important role in the maintenance of pregnancy throughout the gestation period, and should serum progestagen concentrations decrease to levels below critical concentrations at any stage of pregnancy, embryonic or foetal loss will occur[28]. At 25–40 and 60–90 days of pregnancy serum progestagen concentrations normally decline and embryonic or foetal loss can easily occur if serum progestagen concentrations are further decreased by any form of stress, including nutritional stress[16,28]. Owing to the importance of progesterone production in maintenance of pregnancy, the purpose of this study was to investigate the effect of dietary protein quality on serum progestagen concentrations in both lactating and non-lactating mares during pregnancy.

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Table 1: **Comparison of periods after ovulation when mean serum progestagen concentrations** (ng/mℓ) **increased, decreased and when maximum values were obtained in lactating and non-lactating mares in 4 dietary groups during the 1st 50 days of pregnancy.**

Nutritional group[a]	Days after ovulation			
	Increase to 1st maximum	1st maximum values	Decreasing values	Increasing values
Non-lactating				
NL 1	1–12	12–15	27–35	39–42
NL 2	1–15	15–18	30–37	43–47
NL 3	1–12	12–15	34–40	43–47
NL 4	1–12	12–18	27–37	42–45
Lactating				
L 1	1–18	12–15	33–38	41–49
L 2	1–12	12–18	30–39	40–43
L 3	1–15	15–20	30–37	41–45
L 4	1–12	12–18	36–39	41–45
Range	1–18	12–20	27–40	39–49

[a]NL = non-lactating; L = lactating.

MATERIALS AND METHODS

Sixty-four Thoroughbred and Anglo-Arab mares aged 6–12 years were used, 40 of which were non-lactating (NL) and 24 lactating. Foals were weaned at the age of 6 months.

Initially, mares were kept as 8 different groups, of which 4 groups were non-lactating (NL) and 4 lactating (L), in paddocks of 50 × 30 m. Both non-lactating and lactating mares were teased and covered as described previously[30]. After weaning, the pregnant mares in each nutritional group were placed together and kept as 4 dietary groups in paddocks of 80 × 30 m. The daily diets of the mares were composed as follows: Group 1 (NL): 6 kg tef hay, 2 kg cubes; Group 2 (NL): 6 kg lucerne hay, 2 kg cubes; Group 3 (NL): 6 kg tef hay, 2 kg cubes, 0.2 kg fishmeal; Group 4 (NL): 6 kg lucerne hay, 2 kg cubes, 0.2 kg fishmeal. The 4 groups of pregnant mares that had foaled (L), received similar diets but their cubes were increased to 4 kg/d during the last 6 weeks of pregnancy. After foaling, their diets were increased to 6 kg cubes and 7 kg hay respectively while the fishmeal remained at 0.2 kg/d. Fishmeal was added to the morning feed when the foals were prevented from eating with the mares. When foals were 3 months old, 1 kg cubes and 2 kg hay were added to meet their dietary requirements. The crude protein and amino-acid content of the diets were determined according to previously described methods[29]. Pregnancy tests were performed rectally by ultrasonography from Days 14–16, 18–22, 28–30 and 33–36 post-ovulation and thereafter monthly by rectal palpation until 8 months of pregnancy.

Blood samples were collected from 06:30 to 07:30 each day and the serum stored as described previously[31]. Blood samples were collected from each mare as follows:

1. From the day of ovulation until Day 33 of pregnancy: 1 sample daily for the 1st 3 days and then 1 sample every 3rd day until Day 33.
2. One sample daily from Days 35–50 of pregnancy.
3. One sample weekly from Day 50 of pregnancy until foaling.

Statistical analysis

The fact that the same mares were sampled repeatedly presented a problem regarding analysis of variance, on account of possible covariances between samples obtained from the same individual. It was thus decided to investigate the covariance between repeated samples on the same individual, using the following mixed linear model of Harvey[12]:

$$Y_{ijkl} = \mu + a_i + b_{ij} + F_k\, b(x - x)^n + e_{ijkl}$$

with Y_{ijkl} = a specific observation of progestagen concentration; μ = the overall mean; a_i = the fixed effect of the i-th diet-pregnancy status group (i = Group 1: not-lactating ... Group 4: lactating); b_{ij} = the random effect of the j-th mare in the i diet/pregnancy status group; F_k = the fixed effect of the k-th sampling week or sampling day (k = 0 ... 50); x = specific sampling date; e_{ijkl} = random error term, used to test the other effects for significance; b = regression coefficients for regressions to the power of n (n = 1,2,3); $\bar{x}$ = overall mean for the sampling date in days or weeks.

From this analysis, it was possible to obtain variance components to estimate the repeatability (intraclass correlation) of prostagen concentration in mares. This effect was barely significant and the obtained repeatability estimates were below 0.10. On the basis of these results, it was decided that observations on the same mare were sufficiently uncorrelated to meet the prerequisites for analysis of variance. The data were subsequently subjected to least squares analysis of variance to account for uneven subclasses according to a 4 × 2 factorial analysis with diet and reproductive status as factors. Day or week of sampling was included as a linear, quadratic or cubic continuous independent variable, to test for significant differences in the regression coefficients between diets and lactation status. If significant differences were obtained, subsequent analyses were carried out within diet-lactation status, where the degrees of freedom for day or week of sampling were partitioned in orthogonal polynomials depicting linear to quintic trends in progestagen concentrations. R^2 values for these regression coefficients were obtained, to indicate goodness of fit of these equations.

RESULTS

The total daily intake of protein (g) and certain amino-acids (g) of both NL and L mares in the 4 dietary groups has been described[30].

The mean serum total unconjugated progestagen concentrations for both the NL and L mares during the 1st 50 days of pregnancy are presented in Fig. 1A,B. The results of mares that were no longer pregnant at 90 d are not included. In Table 1 a comparison is made at the stages after ovulation that progestagen concentrations increased, decreased and when maximum and minimum values were observed during the 1st 50 days of pregnancy.

According to these results, the pattern of serum progestagen concentrations followed a similar trend irrespective of

Table 2: **Percentage variation in total progestagen concentration during the 1st 50 days of pregnancy that could be explained by the different regression coefficients of the orthogonal polynomials in lactating and non-lactating mares on 4 diets.**

Feeding[a]:	TC		LC		TCF		LCF	
Groups:	1		2		3		4	
Physiological status[b]:	NL	L	NL	L	NL	L	NL	L
Number of mares:	6	4	8	4	8	4	9	5
Linear	84.2**	91.4**	70.9**	72.3**	65.7**	56.4**	61.2**	58.8**
Quadratic	2.4	0.2	1.7	3.5	0.96	0.32	1.7	3.5
Cubical	7.0**	4.0	22.0**	10.7**	28.4**	33.0**	20.5**	24.1**
Quartic	4.7*	0.23	3.5**	4.5	0.5	2.2	12.9**	4.4*
Quintic	0.6	0.1	0.7	0.7	0.1	3.9*	0.7	0.2

[a]T = tef hay; L = lucerne hay; C = cubes F = fishmeal.
[b]NL = non-lactating; L = lactating.
*, **values in the same row with different superscripts differ significantly at 5 % (*) and 1 % (**) levels.

lactation in all dietary groups. Within 24 h post-ovulation, progestagen concentrations increased to reach maximum values of 8–20 ng/mℓ from Days 12–20, followed by a decrease to reach the lowest values from Days 27–40. From 39–49 d post-ovulation, progestagen concentrations increased. Although these are the mean values for each group, considerable individual variation existed between mares and in some mares, plasma progestagen concentrations decreased to values of less than 4 ng/mℓ 30–40 d post-ovulation.

Statistical analysis of these results revealed a significant interaction ($p < 0.05$) between physiological status (lactating and non-lactating) and diet. The regression of progestagen concentration on time (stage of pregnancy) revealed a significant cubical relationship during the 1st 50 days of pregnancy. In NL mares on the 4 different diets, the individual class regressions demonstrated a significant difference for linear regression (Table 2). Similar results were obtained in L mares but significant differences occurred between dietary groups regarding the linear, quadratic and cubical relationships. In both L and NL mares on the low-quality protein diet (Group 1 NL and L) a strong linear tendency was observed that was not present in the other 3 dietary groups. Although absolute differences in total serum progestagen concentrations were not significant throughout this period, it is more important to look at the pattern of progestagen secretion. L mares in Group 1 (tef hay and cubes) did not show a rapid increase in progestagen concentrations during the 1st 12 days after ovulation but a gradual increase in progestagen during this 1st 50-day period was found instead. The significant ($p < 0.01$) cubical tendencies in all the other groups indicated a high rate of progesterone secretion during this early period of pregnancy. Supplementation with fishmeal irrespective of the source of roughage decreased the variation explained by the linear function (Table 2). At the same time the variation accounted for by the cubical function was increased in the fishmeal-supplemented groups compared to the groups that did not receive fishmeal (Table 2).

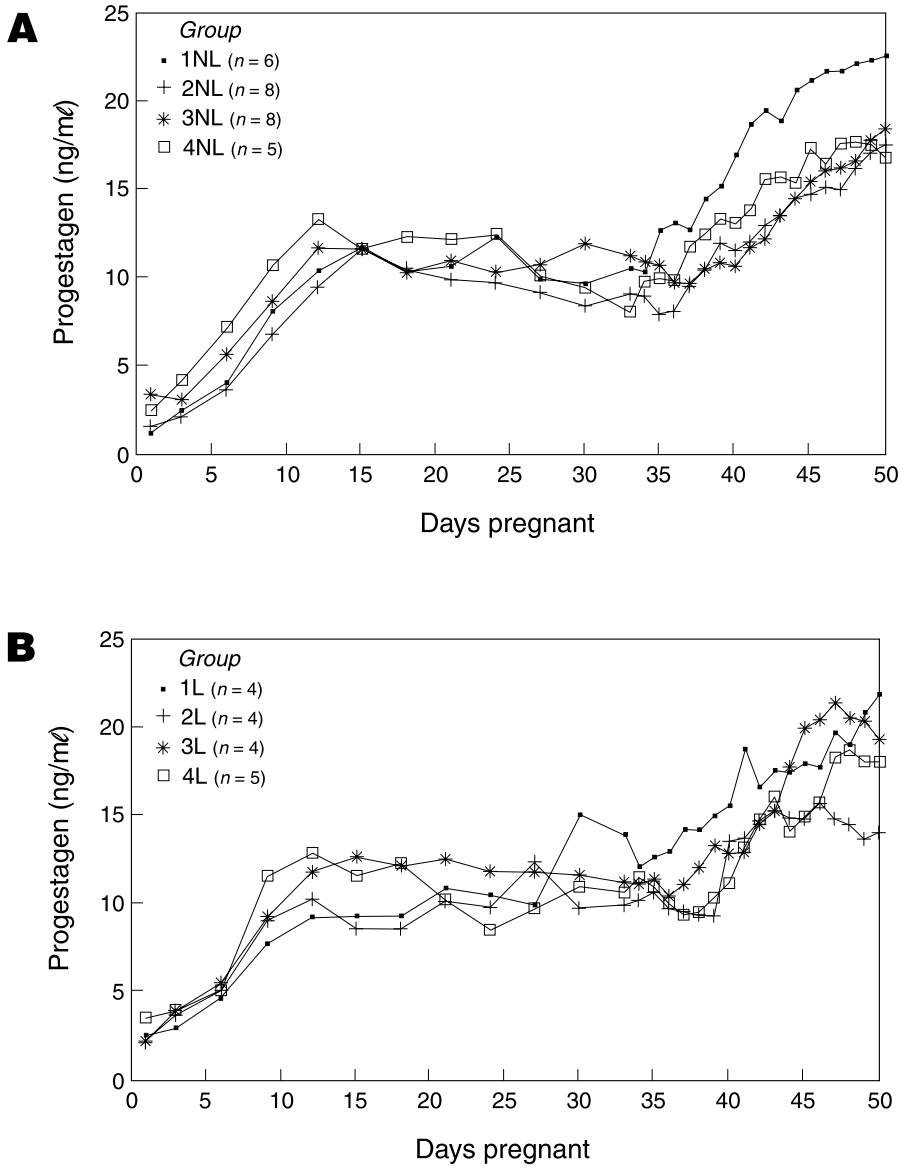

Fig. 1: **Mean serum progestagen concentrations** (ng/mℓ) **of (A) non-lactating** (NL) **and (B) lactating** (L) **mares in 4 dietary groups during the 1st 50 days of pregnancy.**

Table 3: **Percentage variation in total serum progestagen concentrations during pregnancy that could be explained by the different regression coefficients of the orthogonal polynomials in lactating and non-lactating mares on 4 diets.**

Groups[a]:	TC 1		LC 2		TCF 3		LCF 4	
Physiological status[b]:	**NL**	**L**	**NL**	**L**	**NL**	**L**	**NL**	**L**
Number of mares:	**6**	**2**	**6**	**4**	**8**	**4**	**8**	**5**
Linear	33.8**	45.2**	72.5**	65.7**	70.0**	86.2**	70.1**	69.9**
Quadratic	4.3**	1.0	1.4*	11.5**	0.4	0.1	2.9**	2.6**
Cubical	48.4**	5.0*	17.4**	1.1*	22.8**	1.7**	10.4**	1.1*
Quartic	0.9	0.02	0.7	10.1**	1.2*	0.6	2.1**	3.1**
Quintic	1.1	21.6**	1.9**	4.7**	0.01	4.6**	3.7**	12.9**

[a]T = tef hay; L = lucerne hay; C = cubes F = fishmeal.
[b]NL = non-lactating; L = lactating.
*, **values in the same row with different superscripts differ significantly at 5 % (*) and 1 % (**) levels.

The mean serum progestagen concentrations of NL and L mares in the 4 dietary groups during pregnancy are shown in Fig. 2. These results were statistically compared with the basic outlay as a 2 (L *versus* NL) × 4 (diets) factorial with a regression of serum progestagen on time of pregnancy (1–48 weeks). Degrees of freedom of time of sampling as indicated for the individual regression equations were divided into orthogonal polynomials to identify linear, quadratic, cubical, quartic and quintic tendencies. The variation accounted for by each regression is presented in Table 3. Comparison of the progestagen secretion pattern (Figs 1A,B and 2A,B) demonstrated that constant differences existed in the concentrations between groups and also that the pattern of the curves differed. Figs 1A and 2A indicate that the progestagen concentration in Group 1 (NL) increased from Week 7 to Week 20 above that of the other dietary groups. A similar tendency was observed for the lactating mares (Figs 1B and 2B) in Group 1 (L) on the low-quality protein diet.

Serum progestagen concentrations of the NL and L mares were very similar during the 1st 5 weeks of pregnancy. From Week 6 until Weeks 17–21, circulating progestagen concentrations in L mares were lower than those of NL mares. After weaning at 20–38 weeks of pregnancy, serum progestagen concentrations increased to higher values than those of NL mares. The pregnancy was divided into 3 time periods, namely (1) Weeks 1–16, (2) Weeks 17–21 and (3) Weeks 22–48. These 3 periods are referred to as (1) lactation, (2) period of weaning and (3) the late-pregnant, non-lactating period. These 3 periods also represent the different sources of progestagen production, *i.e.* the primary CL, the secondary CL and the foeto-placental unit. A 4 (diets) × 2 (physiological status) × 3 (periods) factorial analysis was carried out. Results of the

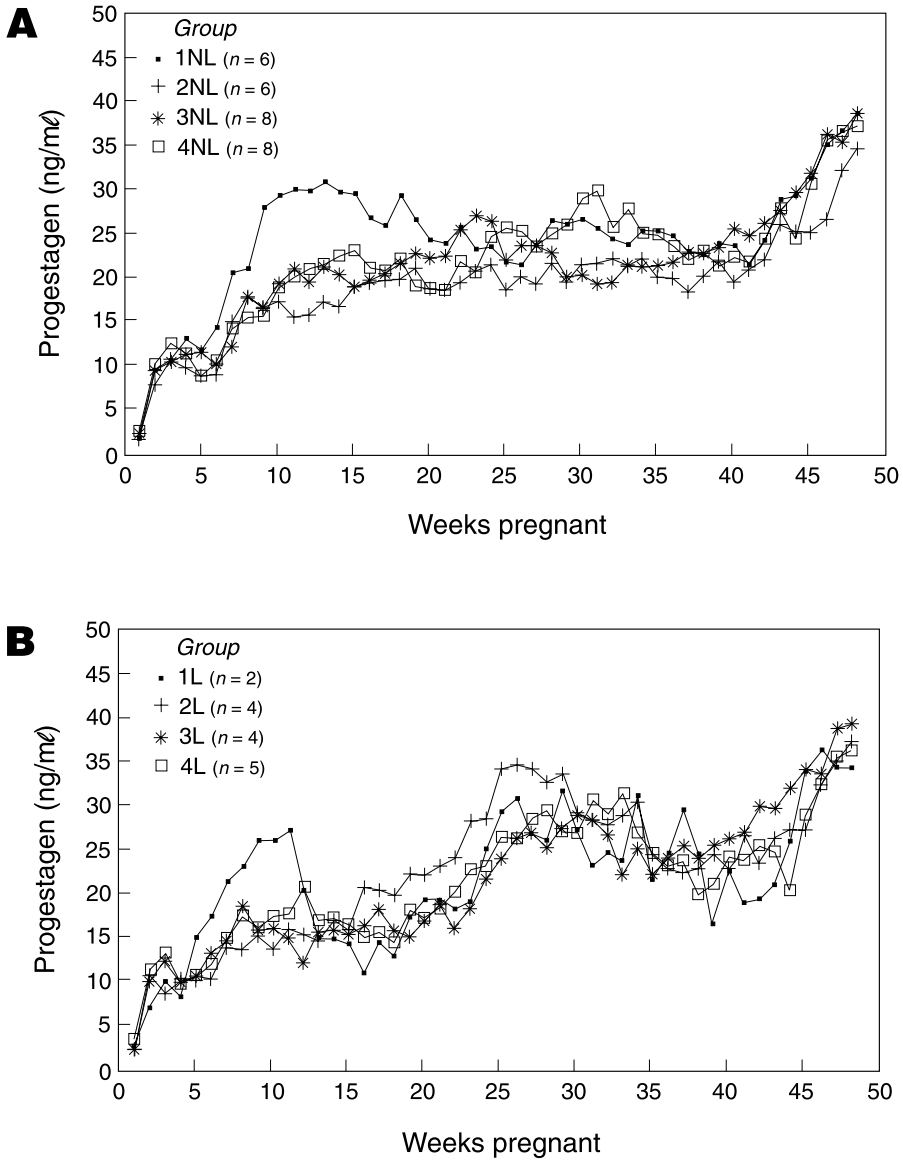

Fig. 2: **Mean serum progestagen concentrations** (ng/mℓ) **of (A) non-lactating** (NL) **and (B) lactating** (L) **mares in 4 dietary groups during pregnancy.**

NL mares were pooled and compared with those of the L mares. According to the results (Table 4), lactation reduced progestagen concentrations ($p \leq 0.05$) during the 1st 16 weeks of pregnancy (14.3 ng/mℓ *versus* 16.2 ng/mℓ). After weaning (Weeks 17–21) the serum progestagen increased in lactating mares from

Table 4: **Mean pooled serum progestagen concentration** (ng/mℓ) **of lactating** (n = 15) **and non-lactating** (n = 28) **mares for 3 periods during pregnancy.**

Period of pregnancy in weeks	Progestagen concentrations (ng/mℓ)	
	Lactating	Non-lactating
1–16	14.3[a] ± 0.05	16.2[b] ± 0.4
17–21	18.4[a] ± 0.9	21.9[b] ± 0.7
22–48	27.3[a] ± 0.4	25.3[b] ± 0.3

[a,b]Values in the same row with different superscripts differ significantly ($p < 0.05$).

18.4 ng/mℓ to 21.9 ng/mℓ, but this increase only reached significant ($p \leq 0.05$) levels of 27.3 ng/mℓ during late (22–48 weeks) pregnancy.

In Tables 5 and 6 the mean serum progestagen values for each period are shown for both NL and L mares on the low-quality protein diet (Group 1 NL). Serum progestagen concentrations increased ($p \leq 0.05$) to 20.9 ng/mℓ in the NL mares on the low-quality protein diet (Group 1 NL) compared to 13.5–15.4 ng/mℓ in the NL Groups 2, 3 and 4 and remained elevated during the period 17–21 weeks of pregnancy. A similar trend was found in L mares during the 1st 16 weeks. At 17–21 weeks of pregnancy the progestagen values declined to the lowest concentrations of 16.8 ng/mℓ in Group 1 (L) when compared to the 16.9–21.6 ng/mℓ in Groups 2 (L), 3 (L) and 4 (L) and remained so for the rest of the pregnancy period.

DISCUSSION

According to National Research Council (Washington DC)[17] recommendations and the feeding programme followed in this trial, none of the mares suffered from a protein deficiency[30].

The normal pattern of serum progestagen concentration of the mare during pregnancy originates from 3 sources: (1) primary CL (Days 1–35), (2) secondary CL (Days 30–120) and (3) foetal-placental unit from Days 60–90 until parturition[13]. The initial increase in progestagen after conception is produced by the primary CL. Maximum values are attained from Days 12–20 followed by a decline between Days 27 and 40 (Table 1), which is in agreement with previous studies[13,23]. This decline, which obviously coincides with degeneration of the primary CL, may vary considerably between individuals, reaching values of less than 4 ng/mℓ, at a stage that is considered to be a critical period in pregnancy, when up to 26 % of foetal losses are reported to occur[32]. When results from individual mares are pooled, these low levels are frequently masked because they usually do not occur on the same days and may easily be overlooked.

According to Van Niekerk[26] and Van Niekerk *et al.*[27], the primary CL degenerates when progesterone is secreted by the secondary CL. There is, however, evidence that the primary CL may persist until 160 days of pregnancy[23] and that pregnancy can be maintained without the formation of the secondary CL[25].

However, when the secondary CL begins to produce progesterone by Days 35–49 (Table 1), progestagen values immediately increased, which is in agreement with previous studies[26,27,31]. The increase in serum progesterone could also be partly due to the resurgence of the primary CL[7].

During the 1st 50 days of pregnancy the serum progestagen concentrations of NL (Group 1 NL) (Fig. 1A) and L (Group 1 L) (Fig. 1B) mares on the low-quality protein diet had a positive linear tendency ($p \leq 0.05$) (Table 2), indicating that progestagen concentrations were lower during the 1st 20 days than the concentrations in the other dietary groups irrespective of lactation. In the other groups (both L and NL) a strong cubical tendency was observed (Table 2). Mares in Group 1 (NL) did not have a definite decrease in serum progestagen concentrations and the increase began at about Day 25. In Group 1 (NL) a rapid increase in serum progestagen was observed from Day 34, resulting in much higher concentrations compared to the other 3 non-lactating groups. The significant ($p \leq 0.01$) cubical tendency observed in all groups except Group 1 (NL) indicates that progesterone production by the primary CL was impaired in this group. This suggests that supplementation of high-quality protein stimulates progesterone production by the primary CL during early pregnancy.

Table 5: **Mean serum progestagen concentration** (ng/mℓ) **of non-lactating mares for 3 periods during pregnancy.**

Period of pregnancy in weeks	Progestagen concentrations (ng/mℓ) in groups			
	1 (NL) (n = 6)	2 (NL) (n = 6)	3 (NL) (n = 8)	4 (NL) (n = 8)
1–16	20.9[a] ± 0.8	13.5[b] ± 0.8	15.0[b] ± 0.7	15.4[b] ± 0.7
17–21	26.1[a] ± 1.4	19.5[b] ± 1.4	21.9[b] ± 1.2	19.9[b] ± 1.2
22–48	26.4[a] ± 0.6	22.6[b] ± 0.6	25.4[b] ± 0.5	26.4[a] ± 0.5

[a,b]Values in the same row with different superscripts differ significantly ($p < 0.05$).

Table 6: **Mean serum progestagen concentration** (ng/mℓ) **of lactating mares for 3 periods during pregnancy.**

Period of pregnancy in weeks	Progestagen concentrations (ng/mℓ) in groups			
	1 (NL) (n = 2)	2 (NL) (n = 4)	3 (NL) (n = 4)	4 (NL) (n = 5)
1–16	16.1[a] ± 1.4	13.0[a] ± 1.0	13.3[a] ± 1.0	14.4[a] ± 0.9
17–21	18.6[a] ± 2.4	21.6[a] ± 1.7	17.1[a] ± 1.7	16.9[a] ± 1.5
22–48	26.3[a] ± 1.0	29.0[a] ± 0.7	27.4[ab] ± 0.7	26.8[a] ± 0.7

[a,b]Values in the same row with different superscripts differ significantly ($p < 0.05$).

In considering serum progestagen concentrations for the total duration of pregnancy it is evident that the low-quality protein diet in both the L and NL mares in Group 1 affected the serum progestagen pattern. NL (Group 1 NL) mares (Fig. 2A) had higher serum progestagen concentrations from 35–140 d and L (Group 1 L) mares (Fig. 2B) had higher serum progestagen concentrations from 30–84 d than Groups 2 NL and 2 L, Groups 3 NL and 3 L and Groups 4 NL and 4 L. The reason for this increase is unknown. As seen in Tables 5 and 6, these increased values ($p < 0.05$) in Groups 1 NL and 1 L remained until Weeks 21 and 16 of pregnancy respectively. It must be remembered that results for the mares that lost their foetuses before Day 90 were not included.

The inverse relationship between level of nutrition and progesterone concentration has also been described in sheep[9], where increased plasma progesterone concentrations were found in ewes kept on 25 % of a maintenance diet[34]. The incidence of embryonic loss did not increase in ewes carrying singletons but increased in those with twins[33]. In a study where ewes were ovariectomised 4 days after ovulation, pregnancy was maintained by means of exogenous progesterone[20]. Half the ewes were kept on 25 % of a maintenance diet[20]. These ewes on the low dietary intake maintained higher plasma progesterone concentrations compared to the well-fed ewes[20]. In a study with boer goats on a restricted diet where weight loss of 15–19 % occurred during late pregnancy, progesterone values of 5–10 ng/mℓ above those of the adequately fed ewes were found[8]. It appears that a compensatory mechanism is activated by under-nutrition in both species where the CL is the main source of progesterone throughout pregnancy (goats) and also where the foeto-placental unit fulfils this function during the latter part of pregnancy (sheep). This effect was only observed in the present trial during the period when luteal structures were actively functional. Once the foeto-placental unit took over production of progesterone, no such compensatory difference was found. However, none of the mares in any of the groups were underfed, and the total protein intake of mares in Group 1 (NL) and Group 1 (L) complied with the minimum requirements outlined by the NRC[17]. The possibility thus exists that the mare is susceptible to deficiencies of certain essential amino-acids that could affect steroidogenesis from luteal structures but not from the foeto-placental unit as described in other species[8,15,21].

The depressive effect of lactation on circulating progestagen concentrations ($p < 0.05$), recorded during the 1st 21 weeks of pregnancy, when lactating mares are compared to non-lactating mares, followed by an increase in serum progestagen concentrations during 21–35 weeks of pregnancy in all the lactating groups (Fig. 2B), supports the contention that lactation is detrimental to the maintenance of pregnancy in the Thoroughbred[16] and that a higher incidence of foetal losses before weaning[16,28] is associated with a reduction in plasma progestagen concentrations.

It is thus clear that an increased risk of impaired progesterone production exists during certain stages of pregnancy, namely (1) Days 30–40 when the secondary CL develops and (2) Days 60–120 when the secondary CL undergoes regression and the foeto-placental unit takes over this function. In lactating mares, milk production peaks between 4 and 8 weeks *post partum,* which coincides with the time that mares should reconceive if successfully rebred at 30 d *post partum,* and when progesterone is still secreted by the primary CL. The suppressive effect of lactation on progesterone production is therefore important. By the time the foal is weaned at 6 months, the foeto-placental unit should be fully functional in the production of progesterone. This implies that special attention should be given to the diet of brood mares in terms of protein (quantity and quality) and energy requirements during lactation to optimise reproductive efficiency.

CONCLUSION

The maintenance of normal blood progesterone levels throughout pregnancy is important in preventing embryonic and foetal loss. It is known that the circulating progestagen concentrations in the pregnant mare originate from different sources. Between Days 35 and 45, secondary luteal structures become effective, and some mares could be at risk of foetal loss due to very low circulating progestagen concentrations, particularly lactating mares suffering from nutritional stress. Serum progestagen secretion from the primary and secondary luteal structures is stimulated when mares are fed a low-quality protein diet during pregnancy, which is probably a compensatory measure to avoid embryonic loss. Circulating progestagen concentrations are suppressed during lactation. Consequently the diet of pregnant mares, especially lactating mares, should contain adequate high-quality protein rich in the essential amino-acids throughout pregnancy to maintain normal progesterone levels and minimise foetal loss.

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