

# EFFECT OF AGE ON THE INTERRELATIONS BETWEEN CERTAIN BLOOD COMPONENTS AND MILK YIELD DURING EARLY LACTATION PERIOD IN AYRSHIRE COWS INJECTED WITH VITAMIN D<sub>3</sub> PRIOR TO CALVING

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Interrelations between the number of trichloroacetic acid (TCA) soluble whole blood minerals and the milk yield have been examined by SAARINEN (1950), SAARINEN (1953), LANE et al. (1968), and LANE & CAMPBELL (1969).

SAARINEN's (1950) data consisted of 274 arterial blood samples taken from Finnish Ayrshire cows and was characterized by the following mean and range values: stage of lactation 68.6 days (20-252), daily milk yield 20.34 kg (10.9-33.6), the Ca/P of the diet 2.07 (1.07-3.91), daily calcium intake in g/cow 160.44 (67-555), daily phosphorus intake in g/cow (33-157), blood calcium (Ca) 6.52 mg% (4.01-10.62), and blood inorganic phosphorus (P) 4.30 mg% (2.66-7.38). Partial correlations in his study revealed that the blood Ca level was positively influenced by the Ca/P of the diet and milk yield, while the effects of the daily intakes of calcium and phosphorus were nonsignificant. The blood P level, in turn, was significantly affected by the daily intake of phosphorus from the diet (positively) and by the milk yield (negatively). The stage of lactation had a nonsignificant effect on the blood Ca and P levels (SAARINEN 1950). A positive simple correlation, 0.12, was found between blood Ca and P levels (SAARINEN 1953).

In the data of LANE et al. (1968) that was obtained from lactating as well as dry Guernsey cows during all seasons of the year, the blood magnesium (Mg) level correlated positively with the blood Ca level (0.15\*). In lactating cows the blood Mg was correlated with the level of milk production (0.11\*). LANE & CAMPBELL (1969) obtained also significant correlations for hematocrit (Hc) with blood P (0.18\*), Ca (-0.35\*), and Mg (-0.12\*) levels. Similar but nonsignificant trends were noted in nonlactating cows. Sire, season, pregnancy, age, and stage of lactation were found to affect the blood components by LANE and coworkers; however, the effect of age on the interrelations between the various blood components was not reported in their study.

The purpose of this study was to investigate the possible effect of age on the interrelations between the serum Ca, P, Mg, and blood hemoglobin (Hb) levels and the daily milk yield, during heavy lactation in cows treated with massive doses of vitamin D shortly before calving.

#### Material and methods

In 1963, blood samples were drawn from V. jugularis of 38 apparently healthy lactating Ayrshire cows at the Viik Experimental dairy herd during the first 75 days following calving. Approximately one half of the samples were taken during the late indoor feeding period and the other half during the pasture season. Feeding and management of the herd has been recently described by KOSSILA (1967, p. 32—36). Each cow was injected intramuscularly twice with 5 million I. U. vitamin D<sub>3</sub> shortly before calving. The serum Ca, P, and Mg levels and blood Hc and Hb values were determined as previously described (see KOSSILA et al. 1970). The milk yield of each cow was weighed on the day when the blood sample was taken. The live weights and the degree of fatness of the cows were estimated 5 days *post partum*, and corrections to the live weight in regard to the degree of fatness were made in order to obtain the so-called corrected body weight (ref. KOSSILA 1967, p. 38). The writer has earlier noted that the milk yield is more closely correlated to the corrected body weight than to the live weight of the cows. Both the absolute (in kg) and the relative (in kg/100 kg corrected body weight) daily milk yields have been considered in this study. Corrections for the fat content of milk were not made, because it was assumed that variations in the milk fat content are probably of minor importance in determining the loss of minerals with milk from the cow's body during lactation.

Statistical calculations were made according to CROXTON & COWDEN (1955).

#### Results

The data was divided into two groups, group I comprising 19 cows (67 blood samples) with 1 or 2 calvings, and group II, 19 cows (65 blood samples) with more than 2 calvings (mostly 3 to 6 calvings). The average blood Hc and Hb values, serum Ca, Mg, and P con-

Table 1. Mean and range values of the investigated blood characteristics, level of milk yield, and birth weight of calf in young (I), old (II), and all (I+II) cows.

	Group I (N = 67)		Group II (N = 65)		Groups I+II (N=132)
	Mean $\pm$ S	Range	Mean $\pm$ S	Range	Mean $\pm$ S
Hematocrit value %	33.63 $\pm$ 0.12	27.8—40.4	32.87 $\pm$ 0.19	26.2—38.3	33.26 $\pm$ 0.08
Hemoglobin, g/100 ml blood	10.20 $\pm$ 0.01	8.2—12.1	9.95 $\pm$ 0.02	7.6—12.3	10.08 $\pm$ 0.01
Serum calcium, mg %	9.69 $\pm$ 0.01	8.5—11.4	9.48 $\pm$ 0.01	7.8—10.9	9.59 $\pm$ 0.00
» inorg. phosphorus, mg%	5.65 $\pm$ 0.02	3.6— 8.5	5.31 $\pm$ 0.03	2.5— 9.1	5.48 $\pm$ 0.01
» magnesium, mg%	1.78 $\pm$ 0.00	1.0— 2.6	1.76 $\pm$ 0.00	1.1— 2.6	1.77 $\pm$ 0.00
Daily milk yield in kg	17.51 $\pm$ 0.25	8.1—28.2	24.74 $\pm$ 0.31	11.3—33.6	21.07 $\pm$ 0.24
Daily milk yield in kg/100					
kg corrected body weight	4.05 $\pm$ 0.01	2.0— 6.4	5.07 $\pm$ 0.01	2.4— 6.7	4.55 $\pm$ 0.01
Birth wight of the calf, kg	33.26 $\pm$ 1.23	25.0—41.0	34.53 $\pm$ 1.97	27.0—48.0	33.89 $\pm$ 0.79

N = number of cases,

S = standard error of mean,

centrations, absolute and relative daily milk yields, and also the mean birth weights of the calves in group I, group II, and in all cows (I + II) are given in Table 1.

Table 1 indicates that on an average the blood Hc and Hb and serum mineral levels of younger cows (group I) were somewhat higher, while the absolute and relative milk yields and the birth weight of the calf were lower compared to corresponding values in older cows (group II). The most pronounced difference between the two age groups was in the milk yield.

Coefficients of simple and partial correlations among blood Hb ( $X_1$ ), serum Ca ( $X_2$ ), Mg ( $X_3$ ), P ( $X_4$ ), and absolute daily milk yield ( $X_5$ ) or relative daily milk yield ( $X_{5a}$ ) were calculated separately for group I, group II, as well as for all cows (I + II). These results have been summarized in Table 2.

Since the preliminary calculations had revealed a very close correlation between the Hc and Hb values in group I (0.83) as well as in group II (0.89), of these two blood characteristics, only the Hb value was included as a variable in the further statistical calculations.

The results presented in Table 2 indicate that the significance of the simple and corresponding partial correlations turned out to be very much alike.

In younger cows, significant partial correlations were obtained for Hb with Mg (0.29\*) and the milk yield ( $-0.28^*$ ), and for Ca with the milk yield ( $0.54^{***}$ ). Positive trends were found for Hb with Ca (0.18), and negative trends for Ca with Mg ( $-0.13$ ), and for P with the milk yield ( $-0.15$ ).

In older cows, significant partial correlations were obtained for Hb with the milk yield ( $-0.42^{***}$ ), and for Mg with P ( $0.25^*$ ). A positive trend was found for Hb with P (0.14), and negative trends for Mg with Hb ( $-0.12$ ) and the milk yield ( $-0.24$ ), and for Ca with the milk yield ( $-0.11$ ).

In all cows, a significant partial correlation was found for Hb with the milk yield ( $-0.31^{***}$ ). Positive trends were found for Mg with Hb (0.11) and P (0.14). Negative trends were found for Mg with Ca ( $-0.14$ ) and the milk yield ( $-0.10$ ), and for P with the milk yield ( $-0.12$ ).

### Discussion

In this study age had a significant effect on certain correlations between blood (serum) components and milk yield. Furthermore, the level of significance of the correlations remained nearly the same irrespective of whether the absolute or relative milk yield was used as one of the variables in the statistical calculations, and the significance of the partial correlations was of the same order as that of the corresponding simple correlations (Table 2).

The correlation trends found in this study differ in several respects from those presented by SAARINEN (1950), SAARINEN (1953), LANE et al. (1968), and LANE & CAMPBELL (1969) apparently as a result of the differences in the methods and materials. The cows of this study received massive doses of vitamin D prior to calving, which treatment aids in the attaining of normal serum Ca and P levels more rapidly after calving (ref. KOSSILA et al. 1970). Apparently vitamin D was not used for the said purpose in the studies of Lane & coworkers and Saarinen. On the other hand, it is not known exactly how long the effect of vitamin D, when administered shortly before calving, persists during the ascending phase of lactation.

In this study, special attention is paid to the correlation found between the serum Ca level and the milk yield, which was highly significant in younger cows ( $0.54^{***}$ ) but non-

Table 2. Simple and partial correlations among blood hemoglobin ( $X_1$ ), serum calcium ( $X_2$ ), magnesium ( $X_3$ ), inorganic phosphorus ( $X_4$ ), and daily milk yield in kg ( $X_5$ ) or daily milk yield in kg per 100 kg body weight corrected for fatness ( $X_{5a}$ ) in young (I), old (II), and all (I+II) cows.

		I (N = 67)	II (N = 65)	I + II (N = 132)
Hb.Ca	$r_{12}$	-0.01	-0.02	-0.02
	$r_{12 \cdot 245}$	0.18	-0.05	0.01
	$r_{12 \cdot 245a}$	0.15	-0.06	0.02
Hb.Mg	$r_{13}$	0.31**	0.02	0.17
	$r_{13 \cdot 245}$	0.29*	-0.12	0.11
	$r_{13 \cdot 245a}$	0.28*	-0.10	0.10
Hb.P	$r_{14}$	0.09	0.13	0.12
	$r_{14 \cdot 235}$	0.02	0.14	0.06
	$r_{14 \cdot 235a}$	0.02	0.12	0.05
Hb.Milk	$r_{15}$	-0.27*	-0.41***	-0.34***
	$r_{15a}$	-0.29*	-0.42***	-0.37***
	$r_{15 \cdot 234}$	-0.28*	-0.42***	-0.31***
	$r_{15a \cdot 234}$	-0.27*	-0.43***	-0.34***
Ca.Mg	$r_{23}$	-0.16	-0.15	-0.14
	$r_{23 \cdot 145}$	-0.13	-0.15	-0.14
	$r_{23 \cdot 145a}$	-0.12	-0.14	-0.14
Ca.P	$r_{24}$	0.00	-0.11	-0.04
	$r_{24 \cdot 135}$	0.09	-0.07	-0.01
	$r_{24 \cdot 135a}$	0.07	-0.08	-0.01
Ca.Milk	$r_{25}$	0.53***	-0.06	0.03
	$r_{25a}$	0.42***	-0.08	0.04
	$r_{25 \cdot 134}$	0.54***	-0.11	0.01
	$r_{25a \cdot 134}$	0.42***	-0.12	0.02
Mg.P	$r_{34}$	0.07	0.26*	0.17
	$r_{34 \cdot 125}$	0.05	0.25*	0.14
	$r_{34 \cdot 125a}$	0.04	0.25*	0.14
Mg.Milk	$r_{35}$	-0.19	-0.21	-0.17
	$r_{35a}$	-0.21	-0.16	-0.18*
	$r_{35 \cdot 124}$	-0.02	-0.24	-0.10
	$r_{35a \cdot 124}$	-0.07	-0.18	-0.11
P.Milk	$r_{45}$	-0.14	-0.07	-0.17
	$r_{45a}$	-0.16	-0.09	-0.17
	$r_{45 \cdot 123}$	-0.15	0.03	-0.12
	$r_{45a \cdot 123}$	-0.15	-0.01	-0.11

\*\*\* =  $P < 0.001$

\*\* =  $P < 0.01$

\* =  $P < 0.05$

significant in older cows (-0.11). It seems improbable that increasing the serum Ca level stimulates the secretion of milk in young cows, even though a low serum Ca level may be an inhibitory factor for milk secretion (ref. RAMBERG et al. 1967). Other explanations are possible.

The Ca/P of milk is usually about 1.2:1, while calcium and phosphorus are mobilized from bone in a higher ratio, i.e. in a ratio of 2:1. It has been noted that calcium mobilization

is faster in young and lactating than in old or nonlactating cows (HANSARD et al. 1954, PAPPENHAGEN 1959). Hence it is possible that in young cows, as a result of active bone mineral mobilization, a rising surplus of calcium in respect of phosphorus, has remained in the circulation at the same time when the milk yield, and in consequence, also the lactational requirements for Ca and P have increased, these phenomena explaining the significant positive correlation found for serum Ca with the milk yield and the negative correlation found for serum P with the milk yield respectively.

On the other hand, in older cows, whose milk production has been significantly higher than that of younger cows (Table 1), the dietary minerals are apparently quantitatively more important than the bone minerals in maintaining the serum Ca and P levels adequate during the ascending phase of lactation. Moreover, LOMBA et al. (1968) have noted that the calcium excreted in faeces falls when the Ca requirements for milk are growing. The negative trend found for serum Ca with the milk yield (partial correlation  $-0.12$ ) in older cows (Table 2) may be taken to indicate that the rate of utilization of dietary and bone minerals for the maintenance of the serum Ca level with an increasing milk yield has been slightly below optimal.

Practically no correlation was found for serum P with the milk yield in older cows. LOMBA et al. (1969) noted that the utilization of phosphorus from the diet is highly variable, being, however, markedly more efficient in lactating than in dry cows. They also found a significant positive correlation between the amounts of phosphorus secreted in milk and urine. These observations may explain the phenomenon noted in this study that the milk yield, as a whole, had very little effect on the serum P level.

The serum Mg level was not found to be affected by the milk yield in young cows (Table 2). However, a nearly significant negative partial correlation was found for Mg with the milk yield ( $-0.24$ ) in older cows, in spite of the fact that the amount of Mg secreted in milk is not large. BAKKER (1960) reported lower serum Mg levels in high-yielders than in low-yielders. ROOK & STORRY (1963) and O'KELLEY & FONTENOT (1968) have demonstrated that the serum Mg level is positively influenced by the intake of Mg from the diet of the cows. Probably either the intake or the availability of dietary Mg in the case of older cows has not been quite high enough for the maintenance of the Mg level in serum with an increasing milk yield. According to LOMBA et al. (1968), even cows which produced 10–20 kg milk daily (compare Table 1 group II) had generally a negative Mg balance. They also found a significant positive correlation between the starch intake and Mg in urine ( $0.53^*$ ) in lactating cows, while the correlation between Mg in milk and Mg in urine was nonsignificant. It seems thus that the composition of the diet (contents of starch, Mg, and a number of other substances) is of greater importance than the level of milk production as far as the serum Mg level is concerned.

A significant positive partial correlation was found for Mg with P in older cows ( $0.25^*$ ) but practically no correlation respectively in younger cows ( $0.05$ ). This phenomenon seems to be hard to explain. However, in the previous study (KOSSILA et al. 1970) simultaneously rising Mg and P levels were found in a few ketotic cows, and it is known that older cows are more apt to develop this disturbance than younger ones.

In young, old, and all cows, significant negative correlations were found for Hb with the milk yield (Table 2). This correlation was more significant in older cows which produced more milk compared to younger cows (Table 1). This phenomenon is believed to be mainly

due to the greater increase of the plasma than the red cell volume during the ascending phase of lactation. This assumption is supported by the results of several studies. SMITH & KESLER (1969) found higher Hb values on Day 1 *post partum* than 5 weeks *post p.*; the trends in Hc were similar. TURNER & HERMAN (1931) found larger blood volumes and DALE *et al.* (1957) in addition also lower Hc values in lactating than in dry cows. High-yielders had larger blood volumes than low-yielders in the studies of BOGDANOV (1961) and ZABOEVA (1963). In Polish Red cows, the Hb was negatively correlated with the maximal daily milk yield (ZIEBA 1964). On the other hand, PATTERSON *et al.* (1960) failed to obtain significant correlations between Hb and the lactational ability in Holstein and Jersey cows; LANE & CAMPBELL (1969) found no correlation for Hc with the milk yield in Guernsey cows, while BOGDANOV (1961) reported higher Hb values for high-yielders in Black Pied cattle. FISHER (1962), in turn, found higher Hc values in lactating than in dry cows. However, in the dairy herd of the Viik Experimental farm, dry pregnant cows had higher Hc and Hb values than lactating nonpregnant ones as can be seen from the following compilation:

	dry pregnant			lactating nonpregnant (see Table 1)		
	N	Hb	Hc	N	Hb	Hc
group I	11	10.98	34.88	67	10.20	33.63
group II	9	11.12	35.33	65	9.95	32.87

The cows in the said herd have received mineral salt mixture containing, among others, iron, copper and cobalt (see KOSSILA 1967, p. 34) which elements are known to be essential for the formation of Hb. Hence, the negative correlation found for the Hb value with the level of the milk yield (Table 2) is not readily explainable on the basis of a possible shortage of these hemopoietic substances in the diet particularly since the quantity of these elements secreted with the milk is small.

### Summary

Altogether 132 blood samples were drawn from V. jugularis of 38 apparently healthy lactating Ayrshire cows during the first 75 days following parturition. Each cow had been injected twice with 5 million I. U. of vitamin D<sub>3</sub> shortly before calving. The cows were divided into two groups, I comprising 19 cows with 1 or 2 calvings and II 19 cows with more than 2 calvings. The blood hemoglobin (Hb), and hematocrit (Hc) values, and the amounts of calcium (Ca), inorganic phosphorus (P), and magnesium (Mg) in serum were determined and coefficients of correlation between the blood components and the level of the daily milk yield were calculated. The following significant partial correlations were obtained: in group I, for Hb with Mg (0.29\*) and milk yield (−0.28\*), and for Ca with milk yield (0.54\*\*\*); in group II, for Hb with milk yield (−0.42\*\*\*), and for Mg with P (0.25\*); in groups I + II, for Hb with milk yield (−0.31\*\*\*). The significance of the simple and corresponding partial correlations was very similar, and it did not make much difference whether the absolute instead of the relative daily milk yield was used as a variable in the calculations. The results are discussed in detail.

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## SELOSTUS

IÄN VAIKUTUS VEREN ERÄIDEN KIVENNÄISKOMPONENTTIIEN JA PÄIVITTÄISEN  
MAITOTUOTOKSEN VÄLISIIN VUOROSUHITEISIIN LYPSSYLEHMILLÄ

VAPPU KOSSILA

*Helsingin yliopiston kotieläintieteen laitos*

Yhteensä 132 verinäytettä kerättiin 38 lehmästä laktaatiokauden 0. — 75. päivinä. Ennen poikimista lehmiiin oli injektoitu  $2 \times 5$  milj. ky. D<sub>3</sub>-vitamiinia. 19 lehmistä oli poikinut 1—2 kertaa (ryhmä I) ja 19 yli 2 kertaa (enimmäkseen 3—6 kertaa) (ryhmä II). Verinäytteistä määritettiin hematokriitti (Hc) ja hemoglobiini (Hb) arvot sekä seerumin kalsium (Ca), epäorgaaninen fosfori (P) ja magnesium (Mg). Päivittäin tuotettu maitomäärä punnittiin verenottopäivinä. Tutkimusajanjaksona saatiin seuraavat keskiarvot ryhmille I ja II: Hc 33.63 ja 32.87 %; Hb 10.20 ja 9.95 g/100 ml verta; Ca 9.69 ja 9.48 mg %; P 5.65 ja 5.31 mg %, Mg 1.78 ja 1.76 mg %; päivittäinen maitotuotos 17.51 ja 24.74 kg; suhteellinen maitotuotos päivässä (kg maitoa/100 kg lihavuuskunto korjattuna elopainoa) 4.05 ja 5.07 kg (Taulukko 1).

Veren Hc ja Hb tasojen väliset vuorosuhteet olivat kummallakin ryhmällä erittäin merkittävät (I 0.83 ja II 0.89) ja tästä syystä osittaisvuorosuhteita laskettaessa Hc arvoa ei sisällytetty muuttujien joukkoon.

Ikä vaikutti selvästi eräisiin vuorosuhteisiin (Taulukko 2). Ryhmässä I ilmeni seuraavien muuttujien välillä merkittävät osittaisvuorosuhteet: Hb.Mg 0.28\*, Hb.maitotuotos -0.28\*, Hb.suhteellinen maitotuotos -0.27\*, Ca. maitotuotos 0.54\*\*\* ja Ca.suhteellinen maitotuotos 0.42\*\*\*. Ryhmälle II saatiin seuraavat merkittävät osittaisvuorosuhteet: Hb.maitotuotos -0.42\*\*\*, Hb.suhteellinen maitotuotos -0.43\*\*\*, ja Mg.P 0.25\* (Taulukko 2). Yksinkertaisten ja vastaavien osittaisvuorosuhteiden merkittävyys oli yleensä samaa suuruusluokkaa ja tuloksiin ei maitotuotoksen laskentatapa (päivittäinen tuotos kiloina tai suhteellinen tuotos) näyttänyt sanottavasti vaikuttavan. Vuorosuhteisiin mahdollisesti vaikuttaneita tekijöitä on tarkastettu yksityiskohtaisesti.