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KALE ANAEMIA IN RUMINANTS

I. SURVEY OF THE LITERATURE AND EXPERIMENTAL INDUCTION OF KALE ANAEMIA IN LACTATING COWS*)

By

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The use of Brassicae species — kale and rape — as autumn forage crops is a relatively new practice in this country, but one that is becoming increasingly economically attractive. Introduction of these crops poses the question of their feeding value in relation to their safety for ruminants when grown and fed under local climatic and husbandry conditions.

A feeding trial was run at Ultuna Agricultural College outside Uppsala to assess the effect on the yield, composition and properties of milk after feeding cows different amounts of locally-grown kale; these aspects will be described separately. Blood samples were taken at intervals to follow haemoglobin, packed cell volume, serum Ca, inorganic P and Mg, serum protein, S-GOT and S-OCT activities and in vitro erythrocyte uptake of I^{131}-triiodothyronine as a measure of thyroid activity. The results of these blood studies are reported here; a subsequent trial on sheep will be reported separately.

Much of what we know about kale anaemia in cattle was outlined in the first descriptions of *Rosenberger* (1939, 1943, 1950) in Germany. In his 1943 paper, a summary of his clinical observations and experimental results, he outlined the principal characteristics of kale anaemia

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- all types of marrow-stem kale regardless of variety, soil and fertilization are capable of inducing anaemia, and the severity of clinical manifestations is principally proportional to the amount of kale fed and the duration of the feeding period,
- the basic (and herd) manifestation is anaemia — probably haemolytic, and that frank haemolysis with haemoglobinuria need not necessarily occur in all cattle exposed,
- recovery — clinical and haematological — is fairly rapid following removal of kale from the diet, but recovered animals are still susceptible to kale anaemia.

Following Rosenberger's description other reports of kale anaemia have been published, mainly from the United Kingdom where kale is cultivated extensively as an autumn and winter forage crop (*Evans* 1951, *Penny et al.* 1961, 1964, *Clegg & Evans* 1962, *Dunbar & Chambers* 1963, *Clegg* 1967). *Penny et al.* in particular have contributed some haematological details. Even in this country where kale and rape feeding is not widely practised there are reports of lambs on kale (*Olsson* 1963) and cattle on rape (*Löfstedt* pers. comm.) developing severe clinical manifestations.

Although *Rosenberger*'s (1939) first report was mainly concerned with the haemoglobinuria of heavy kale feeding, he later (1943) proposed the term "kale anaemia" to centre attention upon the general reaction of cattle to large amounts of kale in the ration. Clinically evident haemolysis is apparently an individual variable in this context. The clinical reports have naturally been concerned with these dramatic manifestations — a circumstance which has undoubtedly contributed to the impression that kale feeding is only sporadically dangerous. Rosenberger's observation that heavy kale feeding regularly induces a drop in haemoglobin level and erythrocyte counts has been confirmed by *Penny et al.* (1964), *Steger et al.* (1964), and by the results to be presented here. The same point was made by *Schofield* (1947) for rape feeding.

Other species are apparently resistant to kale anaemia — rabbits, guinea-pigs and mice (*Rosenberger* 1943) and rabbits (*Penny et al.* 1964) with an equivocal response in 2 rabbits (*Penny et al.* 1961). Not even Rosenberger's goat developed anaemia.

Haematological studies. Only *Rosenberger* (1939, 1943), *Penny et al.* (1961, 1964) and *Steger et al.* have reported serial haematological studies of cattle during a period of kale feeding.

Clegg & Evans described some haematological findings for a clinically affected herd. Erythrocyte inclusions — Heinz bodies also present in some other types of haemolytic anaemia — were first described in kale anaemia by *Penny et al.* (1961), although *Rosenberger* (1943) mentioned punctate basophilia.

Actually, *Rosenberger* gave details for only 2 experiments. His experiment covered serial erythrocyte counts and relative haemoglobin levels for 2 heifers fed mainly on kale in unspecified amounts and for 2 fed kale silage. In the 1943 paper, the corresponding values are given for a cow fed 25 kg kale per day and for a young bull fed a large but unspecified amount of kale. The general pattern of the results was fairly uniform — a distinct drop in RBC counts and Hb levels after 10 to 14 days to reach a low after 14 to 21 days, a levelling off at this low level and, after kale was withdrawn from the ration, a distinct rise in RBC counts and haemoglobin levels within 7 to 10 days. Using the colour index as a guide, Rosenberger characterized the anaemia as hyperchromic, a conclusion which has been challenged (*Clegg & Evans*), but is born out by *Steger et al.*; our sheep results reported in a separate paper also indicate a macrocytic hyperchromic response.

The trial run by *Penny et al.* (1964) covered RBC and reticulocyte counts, osmotic fragility, determinations of mean corpuscular volume and Heinz body counts for 5 test heifers and 5 control heifers during kale feeding (approx. 30 kg per day) for 17 weeks and a further 10 weeks after withdrawal of kale. Their results were presented graphically and represented the mean values for the 2 groups. With this reservation, the haematological pattern seems to fit in with that obtained by Rosenberger — a drop in RBC counts within a few weeks, then levelling off at this lower level throughout the period of kale feeding followed by a rise, ultimately to more or less normal levels, beginning within a few weeks after withdrawal of kale from the ration. The curve for MCV suggests a macrocytic response from the time of the RBC fall, but there was no change in the mean corpuscular haemoglobin concentration. A transient but slight rise in reticulocyte counts (less than 1 %) clearly followed the drop in RBC counts. There was no definite shift in osmotic fragility.

Heinz bodies first appeared in the erythrocytes and reached their peak values (25—30 % of erythrocytes) more or less concomitantly with the drop in RBC counts, the increase in MCV, and the reticulocytosis. The Heinz body counts declined again rapidly from the peak value, but the bodies were present in erythrocytes throughout the period of kale feeding. The count rapidly dropped, and only a few bodies were present in the erythrocytes a few weeks after withdrawal of kale.

Heinz bodies in the erythrocytes of cattle fed kale were first reported by *Penny et al.* (1961) in a heifer with clinical haemolysis (55 % of the erythrocytes) and in 4 heifers fed about 20 kg kale per day (reaching a peak of 27 % of erythrocytes and disappearing soon after kale was withdrawn).

Haematological observations were reported by *Clegg & Evans* for a number of cows in a kale-fed herd in which clinical haemolysis had occurred. From their RBC counts, haematocrit (PCV) and Hb determinations they determined MCV and MCHC, and concluded that whether or not clinical haemolysis had occurred in a particular animal the anaemia could be characterized as macrocytic and normochromic with hypochromasia during recovery (i.e. after withdrawal of kale). *Clegg & Evans* were also able to confirm the presence of Heinz bodies in the erythrocyte.

Anaemia induced by rape and other Brassicae

Feeding cattle large amounts of white cabbage (*Schubert* 1954, *Clegg & Evans*) and Brussel sprouts (*Clegg & Evans*) has resulted in clinically manifest haemolysis.

Grazing cattle on rape can have serious clinical consequences (cf. *Coté* 1944). Severe haemolysis can also occur in sheep grazed on rape (*Stamp & Stewart* 1953). *Michael* (1953) has reported somewhat different signs in sheep on rape — photosensitization, corneal opacity, tympanism and unthriftiness.

Williams et al. 1965 observed a significant goitrogenic effect of feeding kale (2—3 kg per day for 9 weeks) to ewes.

As is the case for kale anaemia, haemolysis with evident haemoglobinuria occurs only sporadically and apparently capriciously among animals feeding on rape with great differences between herds and between different years (cf. *Schofield*). No doubt for this reason there has been speculation about the part played by accessory factors such as soil and particularly phosphorus deficiency, occurrence of frosts, degree of pigmentation of the plants (*Coté, Evans*). *Schofield*, however, has reported some haematological studies which indicate — as is the case for kale — that rape regularly induces an anaemia, but haemoglobinuria occurs in only a few of the animals exposed. In Schofield's experiments, there was an obvious drop in RBC counts within 3 weeks, then a levelling off at this lower level followed by a slight rise. Again the anaemia was characterized as macrocytic.

Other reactions induced by Brassicae

The goitrogenic properties of kale, cabbage and rape seed because of their content of 1-5-vinyl-2-thioxazolidone and 3-butenyl isothiocyanate are well known (cf. *Garner* 1957). There is, for example, a regulation in this country limiting rape seed to 25 % of oil cake mixtures.

Although a definite link between feeding cattle on kale and impaired fertility has not been established, the reports by *Reed* (1961), *Melrose & Brown* (1962) and *Williams et al.* suggest the possibility of an association. *Reed* considered conception rates in relation to kale acreage as a measure of the amount of kale fed. On farms not growing kale, the "corrected" conception rate was 66.0 % and on farms with more than 12 % kale acreage, the "corrected" conception rate was 50.9 %. In the herd followed by *Melrose & Brown,* the first insemination conception rates for animals fed kale were from 41 to 48 % and for animals not fed kale, from 53 to 68.5 %. The ultimate conception rates were practically identical, but the interval until conception was much longer for the kale-fed animals.

Williams et al. studied the effect of feeding kale to ewes on e.g. the duration of oestrus, the returning rate, the number of corpora lutea, and normal and abnormal embryos. Kale-feeding reduced the duration of oestrus, increased returning rate and the number of abnormal embryos. The reproductive disturbances in kale fed cattle has been suggested to be due to the presence of oestrogenic material (*Chury* 1960). However, *Pickard & Crighton* 1967 could not demonstrate by biological assay any oestrogenic activity in kale. The herd described by *Clegg & Evans* went through a period of anoestrus after an episode of kale anaemia, but the plane of nutrition in their herd was low and the quality of the animals and management apparently poor.

The present investigation was carried out in connection with experiments going on at the Agricultural College with the primary purpose to study the effect of feeding different amounts of kale on the yield, composition, and properties of milk. A study was made of changes in the levels of total protein and individual protein fractions, S-GOT and S-OCT levels, serum calcium, inorganic phosphorus and magnesium values, haemoglobin levels and packed cell volume, and the in vitro erythrocyte uptake of triiodothyronine.

MATERIAL AND METHODS

The study was conducted on 30 cows of the Swedish Red and White (SRB) breed with ages ranging from 3 to 11 years. The animals were divided into 3 groups of 10 animals each with the groups as similar as possible as to age, milk production and last calving date.

Details of these points will be published separately from the Agricultural College.

The trial started September 20th, when all animals were taken from pasture into the barn, and continued until October 30th. The control group received no kale in the ration, and another group (ad lib.) was given gradually increasing amounts of kale and then free access. The animals of the third group received 10 kg of kale daily. All animals were also fed 6 kg of hay per day and sufficient concentrates to maintain their previous level of milk production.

Blood samples were taken 3 days prior to the start of the experimental period, twice during the trial — after 25 and 39 days — and 3 weeks after the end of the experiment.

Packed cell volume and haemoglobin determinations were made on heparinized blood (*Paulson & Åberg* 1965) and determination of in vitro erythrocyte uptake of I^{131}-triiodothyronine according to *Thorell* (1965). Serum calcium and magnesium were measured by an absorption flame photometer in a Perkin-Elmer model 202 atomic absorption spectro-photometer. The level of inorganic phosphorus in the blood serum was determined according to *Taussky et al.* (1953), glutamic-oxaloacetic transaminase (GOT) by the method of *Reitman & Frankel* (1957), ornithine-carbamyl transferase (OCT) by the method of *Reichard* (1957), and total protein by the method described by *Weichselbaum* (1946).

The serum protein fractions were separated by paper electrophoresis using the apparatus described by *Valmet & Svensson* (1954) in the TRIS-borate-buffer method of *Aronsson & Grönwall* (1957). Amido black was used for staining of the proteins and a Beckman-Spinco Analytrol for the determination of the protein fractions.

The material was statistically analyzed; conventional statistical symbols are used.

RESULTS

The animals with free access to the kale increased their consumption up to a daily maximum of 60—70 kg. No clinical signs could be observed. An account of milk production, butterfat content, taste and quality of the milk is to be published separately from the Agricultural College.

Table 1. Haemoglobin, g/100 ml blood and t-values.

Sample	Group: control A (n=10) $\bar{x} \pm s$	kale ad lib. B (n=10) $\bar{x} \pm s$	kale 10 kg C (n=10) $\bar{x} \pm s$	Comparison A–B
1. 3 days before kale period	11.03 ± 0.91	11.06 ± 0.88	10.66 ± 1.19	
2. Kale feeding, 25 days	12.58 ± 1.29	10.89 ± 1.20	11.53 ± 1.00	3.03**
3. Kale feeding, 39 days	11.44 ± 0.78	9.21 ± 1.10	10.64 ± 0.60	5.24***
4. 19 days after kale feeding	11.26 ± 0.56	10.30 ± 1.43	10.65 ± 0.87	1.93
Comparison				
1—2	3.11**		1.77	
1—3		4.17***		
2—3	2.37*	3.29**	2.41*	
2—4	3.00**		2.09	
3—4		1.91		

The haemoglobin values (Table 1) for the control group and the 10 kg group increased between the 1st and 2nd blood sampling and then dropped to the original level. In the ad lib. group the level became progressively lower during the trial, but 3 weeks after the end of the experimental period it had increased to slightly below the original value. The haemoglobin values in this group were almost identical with those of the control group before the trial started, but at the 2nd and 3rd samplings they were significantly lower.

The packed cell volume (PVC) followed the same pattern as the haemoglobin values (Table 2).

The in vitro erythrocyte uptake of triiodothyronine (Table 3) was greater in all groups at the 2nd sampling. At the 3rd sampling the 10 kg group had an almost significantly lower uptake than the control group, just as the ad lib. group did at the 4th sampling. The rise in the beginning apparently depended upon the lower serum protein level.

Table 2. Packed cell volume and t-values.

Sample	Group: control A (n=10) x̄ ± s	kale ad lib. B (n=10) x̄ ± s	kale 10 kg C (n=10) x̄ ± s	Comparison A-B	Comparison B-C
1. 3 days before kale period	34.8 ± 2.9	33.9 ± 3.5	33.7 ± 3.8		
2. Kale feeding, 25 days	34.4 ± 2.5	32.6 ± 4.0	32.5 ± 3.2		
3. Kale feeding, 39 days	33.5 ± 2.1	28.5 ± 3.8	31.6 ± 1.3	3.65**	2.42*
4. 19 days after kale period	33.7 ± 1.5	31.8 ± 4.7	32.1 ± 2.8		
Comparison					
1—3		3.29**	1.67		
2—3		2.36*			
3—4		1.72			

Table 3. In vitro erythrocyte uptake of I^{131}-triiodothyronine and t-values.

Sample	control A n	control A x̄ ± s	kale ad lib. B n	kale ad lib. B x̄ ± s	kale 10 kg C n	kale 10 kg C x̄ ± s	Comparison A-B	Comparison A-C
1. 3 days before kale period	9	6.30 ± 0.84	9	6.08 ± 0.82	10	5.77 ± 0.66		
2. Kale feeding, 25 days	10	7.56 ± 0.64	10	7.32 ± 0.63	10	7.16 ± 1.19		
3. Kale feeding, 39 days	10	5.64 ± 0.54	10	5.10 ± 0.66	10	5.10 ± 0.48	2.00	2.38*
4. 19 days after kale period	10	5.75 ± 0.69	10	5.17 ± 0.51	9	5.52 ± 0.57	2.11*	
Comparison								
1—2		3.65**		3.72**		3.22**		
2—3		7.26***		7.66***		5.31***		

The total serum protein level (Table 4) was significantly lower in all groups 3 weeks after the beginning of the experiment. The drop was the greatest in the ad lib. group (3.26 ± 0.52), but the degree of decrease was not significantly different from the other 2 groups (2.31 ± 1.69 and 2.54 ± 1.75). After a further 2 weeks of kale feeding the total protein level returned to normal. The albumin-globulin ratio showed no change at any time (values not presented).

Table 4. Total serum protein, g/100 ml and t-values.

Sample	Group: control A n	$\bar{x} \pm s$	kale ad lib. B n	$\bar{x} \pm s$	kale 10 kg C n	$\bar{x} \pm s$
1. 3 days before kale period	10	7.74 ± 0.56	8	8.29 ± 0.39	9	8.17 ± 0.81
2. Kale feeding, 25 days	10	5.42 ± 1.57	8	5.03 ± 0.30	10	5.47 ± 2.47
3. Kale feeding, 39 days	10	7.59 ± 0.58	10	7.72 ± 0.02	10	7.56 ± 0.65
4. 19 days after kale period	10	7.90 ± 0.31	10	8.30 ± 0.70	10	8.06 ± 0.66
Comparison						
1—2		4.38***		19.2***		2.95***
2—3		4.09***		12.2***		2.25*
3—4				1.97		1.71

The S-GOT activity (Table 5) was significantly higher at pasture before the kale period than after 3 weeks confinement in the barn. The S-OCT activity showed a slight increase during the first period of confinement in the barn. The S-OCT value of the control group was almost significantly higher on day 39 than the pre-trial level.

The variations in the serum mineral content (Ca, P, Mg) were very small (Fig. 1).

Both the Ca and the inorganic P levels declined for the con-

Table 5. Serum GOT- and OCT-activity and t-values.

Sample	S-GOT, Karmen-Wroblewski: control A n	$\bar{x} \pm s$	kale ad lib. B n	$\bar{x} \pm s$	S-OCT, Reichardt, units: control A n	$\bar{x} \pm s$	kale ad lib. B n	$\bar{x} \pm s$
1. 3 days before kale period	10	112.9 ± 45.8	8	142.6 ± 40.9	10	1.92 ± 1.54	8	2.41 ± 1.33
2. Kale feeding, 25 days	10	58.9 ± 11.4	10	59.6 ± 6.7	10	4.69 ± 3.90	8	3.51 ± 1.49
3. Kale feeding, 39 days	10	60.3 ± 11.4	10	64.3 ± 6.6	10	3.31 ± 1.18	10	2.81 ± 1.26
Comparison								
1—2		3.62**		5.65***		2.08		
1—3						2.27*		

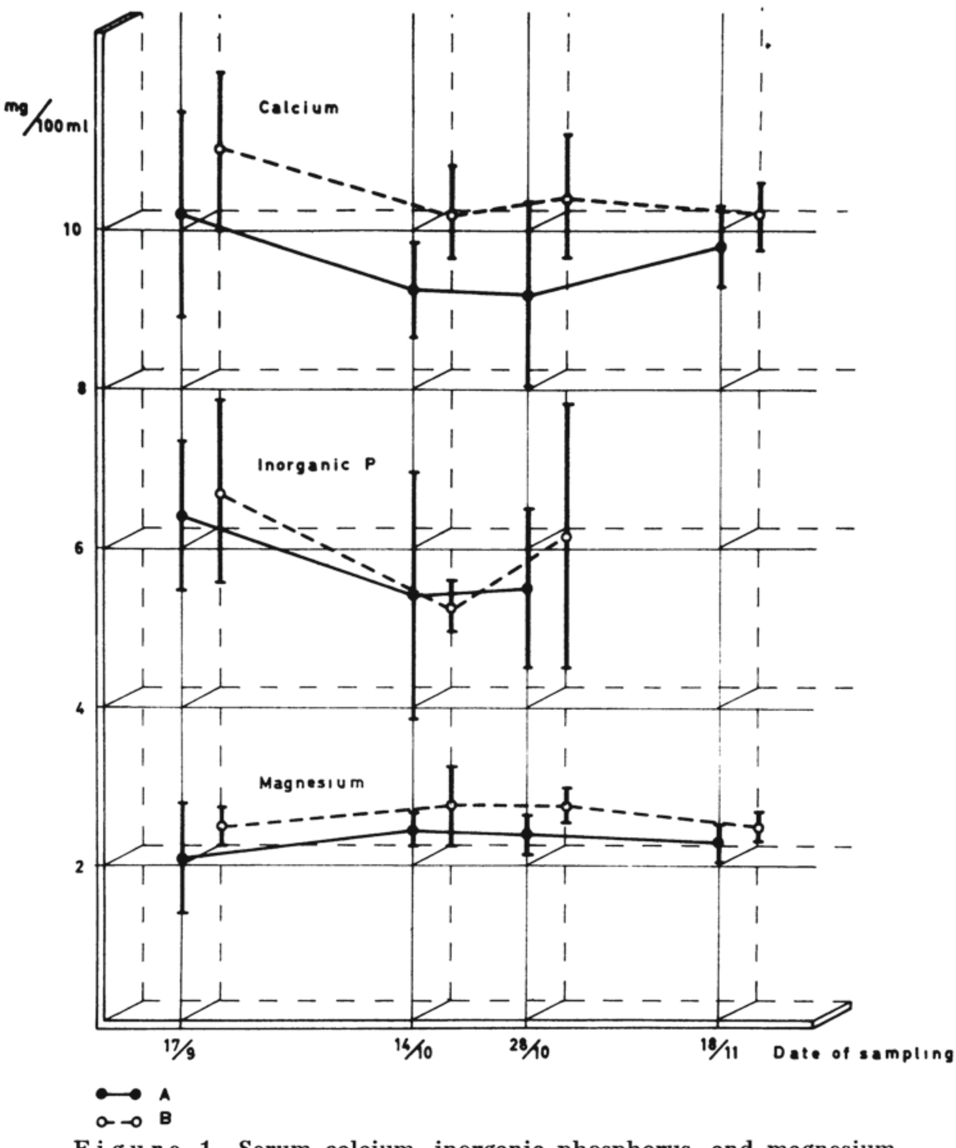

Figure 1. Serum calcium, inorganic phosphorus, and magnesium in the control group (A) and the highest kale-fed group (B).

trol and ad lib. groups, but the Mg levels increased slightly. The inorganic phosphorus in the ad lib. cows was significantly (t-3.29**) lower at the 2nd (5.03 ± 0.30) than at the 1st sampling (6.43 ± 1.17), but there was no difference between the groups on either occasion.

DISCUSSION

This study demonstrated statistically that feeding lactating cows on marrow-stem kale gave a reduction in Hb values for the ad lib. group, but not for the group limited to 10 kg per day. Kale feeding was not consistently associated with changes in serum Ca, inorganic P and Mg, changes in thyroid function during the short feeding period as measured by in vitro erythrocyte uptake of I^{131}-triiodothyronine, changes in serum protein levels, or changes in S-GOT and S-OCT activities. Except for the Hb reduction in the ad lib. group, the other parameters were apparently affected to some extent by the change from pasture to stable feeding.

The results fit in with the pattern — outlined by *Rosenberger* (1939, 1943) — that the basic herd reaction to kale feeding is a drop in Hb values, that the degree of reduction is proportional to the amount of kale fed, that haematological recovery is fairly rapid after removal of kale from the ration and that local differences in the type of kale, cultivation etc. lack primary importance.

The heavy consumption of kale by our cattle, some 60 to 70 kg per day for 41 days, gave only a modest drop in Hb values and no clinical manifestations. Much less, 25 kg per day for 4 weeks, has been suggested as the upper safety limit (*Rosenberger* 1943). Furthermore, it does appear from published reports that there is a real difference from place to place and from year to year in the incidence and severity of clinical manifestations in ruminants fed kale or rape. That accessary factors contribute towards the anaemia cannot be completely discounted at present.

Hypophosphataemia has been suggested, notably by *Clegg & Evans* (1962) and *Clegg* (1966), but this apparently stems from observations on a particular episode and in analogy with post-parturient haemoglobinuria.

The key to the problem and the most obvious potential variable lies in the kale — and rape. The work that has been done towards a closer definition of the properties of Brassicae responsible for anaemia has not yielded results. *Steger et al.* (1964) suggested that the release of sulphur compounds in the rumen of kale-fed cattle binds copper and cobolt, but there is no experimental support for this.

Kale and other Brassicae are known to be goitrogenic. Thyroid function as measured by in vitro erythrocyte uptake of I^{131}-triiodothyronine was affected in both the control group and the kale groups (Table 3) quite apart from the transitory rise apparently related to the drop in serum protein levels after stabling. There was no evident effect on thyroid function attributable to kale during the relatively short kale feeding period. The effect of kale on haemoglobin levels and on thyroid activity appears to be separable.

Not only the aetiology but also the pathogenesis of kale anaemia offers possibilities for further work. Since a reduction in Hb levels regularly occurs in ruminants fed large amounts of kale, there is no evidence for a genetic defect in erythrocytes or haemoglobin which limits kale anaemia to susceptible individuals only. The occasional occurrence of haemoglobinuria among exposed animals makes it likely that kale anaemia is haemolytic. This is supported by indirect evidence — reticulocytosis, compensation by levelling off at lower RBC and Hb levels, and the rapid recovery after withdrawal of kale imply that haematopoiesis is stimulated rather than depressed. Two points brought forward by *Penny et al.* (1961, 1964) suggest a haemoglobin-erythrocyte defect — the occurrence of Heinz bodies in the erythrocytes and the demonstration that a pre-bled sheep, i.e. an animal with a younger erythrocyte population, was more resistant to kale anaemia.

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SUMMARY

After a review of the literature the report of a trial with 30 dairy cows is presented. The cows were divided into 3 groups of 10; 1 group functioned as control (A), 1 had free access to marrow-stem kale (B), and the animals of the 3rd received 10 kg marrow-stem kale daily (C). The trial period lasted about 6 weeks. Blood samples from all animals were taken before the trial began, twice in the course of the experiment, and 3 weeks after its termination. Estimations were made for haemoglobin, haematocrit, the erythrocyte in vitro uptake of I^{131}-triiodothyronine, total protein and protein fractions, serum calcium, inorganic phosphorus and magnesium, as well as for the serum activity of glutamic-oxalacetic acid transaminase (GOT) and ornithine-carbamyl-transferase (OCT). During the trial period differences between groups A and B existed only in haemoglobin and haematocrit, the experimental animals having significantly lower values. The erythrocyte in vitro uptake of triiodothyronine demonstrated a tendency towards lower values in both experimental groups. Other blood values varied between different samples but not between groups. Special mention is made of the total protein value which was substantially lower for the 2nd sample than for the others. This may have been associated with the change from pasture to stall feeding which occurred at the beginning of the trial.

ZUSAMMENFASSUNG

Anämie beim Rind nach Markstammkohlfütterung.

Nach Durchgang der Literatur wird folgender Bericht über die Untersuchung von 30 Milchkühen, welche in 3 gleichgrosse Gruppen eingeteilt waren, abgegeben. Die Gruppen bestanden aus der Kontrollgruppe A und den Gruppen B und C. Gruppe B hatte freien Zugang zu Markstammkohl und Gruppe C bekam 10 kg Markstammkohl täglich. Die Versuchsperiode erstreckte sich über 6 Wochen. Von sämtlichen Tieren wurde vor Beginn des Versuchs eine Blutprobe entnommen, ausserdem zweimal während der Versuchsperiode und einmal 3 Wochen nach Abschluss derselben.

Die Proben wurden auf Hämoglobin, Hämatokrit, Erythrozyten in vitro-Aufnahme von J^{131}-Trijodothyronin, dem totalen Protein und den Proteinfraktionen, Serum-Kalzium, unorganischem Phosphor und Magnesium, sowie der Serumaktivität von Glutaminsäure-Oxalessigsäuretransaminase (GOT) und Ornitin-Carbamyl-Transferase (OCT)

analysiert. Die Unterschiede zwischen den Gruppen A und B lagen während der Versuchszeit nur in Bezug auf Hämoglobin und Hämatokrit vor, bei der die Versuchstiere signifikant niedrigere Werte hatten. Die Aufnahme der Erythrozyten in vitro von Trijodothyronin zeigte eine Neigung zu niedrigeren Werten für beide Versuchsgruppen. Die übrigen Blutwerte variierten zwischen den verschiedenen Entnahmen der Proben, aber nicht zwischen den Gruppen. Dies galt speziell dem Totalprotein, welches bei der zweiten Probeentnahme wesentlich niedriger war, als bei den übrigen. Dies dürfte mit dem Übergang vom Weidegang zur Stallfütterung zusammenhängen.

SAMMANFATTNING

Anemi vid utfodring med fodermärgkål åt idisslare.

Efter genomgång av litteraturen lämnas en redogörelse för en undersökning av 30 mjölkkor, som delades i 3 lika stora grupper, varav 1 utgjorde kontrollgrupp (A), 1 fick fri tillgång till fodermärgkål (B) och 1 fick 10 kg fodermärgkål dagligen (C). Försöksperioden var c:a 6 veckor. Blodprov från samtliga djur togs före försökets början, 2 gånger under försöksperioden och 1 gang 3 veckor efter dennas slut. Proverna analyserades på haemoglobin, haematokrit, erytrocyternas in vitro-upptag av J^{131}-trijodothyronin, total protein och proteinfraktioner, serum kalcium, oorganiskt fosfor och magnesium samt serumaktiviteten av glutaminsyra-oxalättiksyratransaminas (GOT) och av ornitin-carbamyl-transferas (OCT). Skillnader mellan grupperna A och B förelåg under försöksperioden endast beträffande haemoglobin och haematokrit, där försöksdjuren hade signifikant lägre värden. Erytrocyternas in vitro-upptag av trijodothyronin visade en tendens till lägre värden för båda försöksgrupperna. Övriga blodvärden varierade mellan olika provtagningar men ej mellan grupperna. Speciellt gällde detta totalproteinet, som var väsentligt lägre vid andra provtagningen än vid någon annan, vilket torde kunna sammanhänga med övergången från bete till stallfodring, som skedde vid försökets början.

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