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Copper and selenium supplementation of ewes grazing on pastures low in copper and selenium: Effect on reproduction and concentration of plasma copper and blood selenium during pregnancy

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Influence of copper and selenium supplementation in SA Mutton Merino ewes, kept on pastures with low copper and selenium concentrations, was investigated. Ewes which came into oestrus and conceived during March to April, were found to have concentrations of plasma copper of 60 to 80 μg/dl. Inadequate copper intake by ewes during late pregnancy resulted in decreased concentrations of plasma copper, thereby leading to low concentrations of copper in the liver of young lambs. Selenium supplementation of ewes grazing pastures with a selenium content of 0,03—0,09 μg Se/g DM, has been found to raise concentrations of blood selenium but had no effect on the concentrations of selenium in the liver and kidneys of young lambs.

Die invloed van koper- en seleniumaanvulling by SA Vleismerino-ooie, wat op weiding met 'n lae koper- en seleniumkonsentrasie aangehou is, is ondersoek. Ooie wat tydens Maart tot April in estrus gekom en suksesvol gepaar is, het plasmakoperkonsentrasies van 60 tot 80 μg/dl gehad. Onvoldoende koperinname tydens laatdragtigheid het tot 'n daling in die plasmakoperkonsentrasie van die ooi gelei, terwyl die lewerkoperkonsentrasie van hierdie lammers kort na geboorte abnormaal laag was. Seleniumaanvulling van ooie op weiding met 'n seleniumkonsentrasie van 0,03–0,09 μg Se/g DM, het bloedseleniumkonsentrasies van ooie verhoog, maar het min invloed op die lewer- en nierseleniumkonsentrasies van lammers kort na geboorte gehad.

Keywords: Blood selenium, copper supplementation, fertility, plasma copper, pregnancy, selenium supplementation,

Low copper concentrations are frequently found in the soil and plants of the South Western coastal belt of the Republic of South Africa (Van der Merwe & Perold, 1967). Whether this deficiency is reflected in the concentration of plasma copper and in the reproductive performance of sheep of different breeds, is unknown. An induced copper deficiency has been found to delay the commencement of the breeding season in SA Mutton Merino ewes, but once the ewes exhibited normal oestrous cycles in the breeding season, the occurrence of oestrus and conception was normal (Van Niekerk & Van Niekerk, 1989c). Although these ewes developed low concentrations of plasma copper, especially in late pregnancy, and lower than normal plasma progesterone concentrations, pregnancy otherwise proceeded normally. However, lambs born from these copper-deficient ewes were very weak at birth, their mortality rate was high and growth rate poor (Van Niekerk & Van Niekerk, 1989c). Howell

(1968) found that ewes aborted when suffering from a copper deficiency, whereas Allcroft & Parker (1949) found a low conception rate in copper-deficient cows. Munro (1957), Mahadevan & Zubairy (1969) and Van Rensburg (1961) found a high incidence of anoestrous conditions in cows which suffered from a copper deficiency. The fertility of rams suffering from an induced copper deficiency was severely affected, as spermatozoa and testosterone production were suppressed (Van Niekerk & Van Niekerk, 1989a).

Selenium deficiencies were diagnosed biochemically in several flocks grazing in the coastal area of South Africa (F. van Niekerk, 1988, unpublished results). The effect of the low concentrations of blood selenium on the fertility of these ewes is unknown. Hartley & Grant (1961), Hartley (1963) and Schales (1974) found improved reproduction in ewes after selenium supplementation, contrasted with the findings of Gardiner, Armstrong, Fels & Glenross (1962) and Davies (1966). According to Hartley (1963), ewes suffering from a selenium deficiency come into oestrus and conceive but resorb the embryo at the time of implantation. In cows suffering from a selenium deficiency, retained placentas are quite common (Trinder, Woodhouse & Renton, 1969; Jullen, Conrad, Jones & Moxon, 1976).

The influence of copper and selenium supplementation on the trace element status and the incidence of oestrus, conception and lambing percentage of ewes, grazing on pastures low in copper and selenium, was studied at six weeks before mating, at mating and at midpregnancy.

The experimental farm is situated in the sandy coastal area of the Southern Cape. Primary copper and selenium deficiencies in pastures and soil have been identified on this farm (personal communication: N. Kritzinger, 1986). SA Mutton Merino ewes (n = 192) were kept on 10 ha of irrigated pastures, consisting of kikuyu and rye-grass, for a period of eight months without any trace element supplementation prior to the experiment. This area was divided into eight camps which were grazed on a rotational basis. Before ewes were put into a camp, hand-picked pasture samples were collected from 10 different spots in the camp, dried, pooled and stored for the determination of trace minerals.

Ewes were divided into nine groups (n = 21—22). One group received no supplementation while four groups (2—5) received oral copper (2 × 2,5 g CuO needles) and four groups (6—9) oral selenium supplementation (10 mg SE as sodium selenite), as shown in Table 1.

Blood samples were taken six and three weeks before mating, at mating and at monthly intervals thereafter for four months from the same ewes (n = 5) in each group. The ewes, kept as a single flock, were exposed to fertile rams in March and April. Mated ewes were identified on a daily basis by placing marking ink on the rams. Mating took place over a period of six weeks. Livers and kidneys of lambs which died at or within a week after birth were collected and stored in clean plastic bags at −20 °C until analysed for trace minerals. A strict program to control internal parasites was followed and the ewes were inoculated yearly against enzoötic abortion, blue tongue and pulpy kidney.

Blood (20 ml) was drawn from the jugular vein into heparinized vacuum tubes (Vac-U-Test) and was divided into two portions, one of 10 ml which was stored at −20 °C for selenium analysis and one of 10 ml which was centrifuged,

Table 1 Time of copper and selenium supplementation

Group	Time of supplementation		
	Six weeks before mating	At the beginning of mating	Mid-pregnancy
2	–	Cu	–
3	–	Cu	Cu
4	Cu	–	–
5	Cu	–	Cu
6	–	Se	–
7	–	Se	Se
8	Se	Se	–
9	Se	Se	Se

the plasma removed and stored at −20 °C for the determination of copper. Minerals were determined in biological material and pasture samples, using the techniques described by Van Niekerk & Van Niekerk (1989b).

Results were evaluated according to standard, one-way analysis-of-variance procedures, using the P7D program of the BMDP statistical packet (Engelman, 1981). Differences between treatment means were compared by the Bonferroni method (Van Ark, 1981). In some cases, only the standard deviation (± SD) of the mean is indicated.

The copper content of the pasture varied between 4,23 to 6,66 μg Cu/g DM during the experimental period and the selenium content between 0,03 to 0,09 μg Se/g DM. No significant differences in the concentration of either plasma Cu (groups 2—5) or blood Se (groups 6—9) were found within the supplemented groups, irrespective of the frequency of supplementation.

Therefore, the results of the supplemented groups were pooled for further analysis (Tables 2 & 3). Concentrations of plasma copper in healthy animals have been reported to be 60—150 μg Cu/dl, with a high proportion of the values lying between 80—120 μg Cu/dl (Underwood, 1977). Concentrations below 60 μg/dl are usually accepted to be indicative of a copper deficiency. Concentrations of plasma copper in the control and selenium-supplemented groups were approximately the same as those of the copper-supplemented groups up to the third month of pregnancy. After the third month of pregnancy, the concentrations of plasma copper in the control and selenium-supplemented groups decreased from concentrations in excess of 80 μg/dl to less than 60 μg/dl, whereas concentrations in the copper-supplemented groups remained in excess of 80 μg/dl. The decrease in the concentrations of plasma copper during late pregnancy, in the groups without copper supplementation, indicates that the copper intake or the reserve of these ewes was inadequate as concentrations of plasma copper did not decrease where ewes received adequate copper during late pregnancy (Butler, 1963; Van Niekerk & Van Niekerk, 1989c). The copper concentration in the total products of conception increased 27 times between trimesters one and three (Moss, Madsen, Hansard & Gamble, 1974).

Blood selenium concentrations of less that 50 ng/ml have been suggested to be indicative of a selenium deficiency (Caple, Andrewartha, Edwards & Halpin, 1980). None of the blood selenium concentrations (Table 3) suggested such a

Table 2 Mean plasma copper concentrations (μg/dl) of the ewes in the control (C), copper (Cu)- and selenium (Se)-supplemented groups

	Weeks before mating			Months pregnant			
Group	6	3	Mating	1	2	3	4
C	72 ± 18	74 ± 20	86 ± 25	86 ± 17	99 ± 28	80 ± 15	53[a] ± 22
Cu	62 ± 16	69 ± 16	86 ± 17	96 ± 13	107 ± 18	94 ± 19	89[b] ± 17
Se	65 ± 17	64 ± 14	74 ± 12	79 ± 15	86 ± 20	84 ± 16	57[a] ± 19

[a,b] Values in the same column with different superscripts differ significantly ($P \leq 0,05$).

Table 3 Mean concentrations of blood selenium (ng/ml) of the ewes in the control (C), copper (Cu)- and selenium (Se)-supplemented groups

	Weeks before mating			Months pregnant			
Group	6	3	Mating	1	2	3	4
C	114 ± 47	148 ± 14	73 ± 20	102 ± 10	111 ± 12	101 ± 12	114 ± 21
Cu	120 ± 50	149 ± 34	97 ± 35	110 ± 31	116 ± 27	109 ± 25	110 ± 23
Se	91 ± 39	132 ± 43	102 ± 49	129 ± 23	142 ± 22	122 ± 22	141 ± 23

Table 4 Mean (± SD) liver and kidney trace element concentrations of the lambs in the control (C), copper (Cu)- and selenium (Se)-supplemented groups

			Trace element concentration (μg/g)					
Group	Organ	n[a]	Cu	Fe	Zn	Mn	Mo	Se
C	Liver	2	44 ± 19	632 ± 15	227 ± 38	19 ± 11	0,7 ± 0,3	0,33 ± 0,04
	Kidney		18 ± 2	497 ± 10	152 ± 18	3 ± 1	0,5 ± 0,1	2,59 ± 0,23
Cu	Liver	13	299 ± 119	526 ± 71	191 ± 47	13 ± 4	0,4 ± 0,3	0,54 ± 0,20
	Kidney		30 ± 24	331 ± 83	131 ± 36	3 ± 1	0,4 ± 0,2	2,26 ± 0,29
Se	Liver	18	41 ± 40	577 ± 84	200 ± 61	13 ± 5	0,5 ± 0,5	0,52 ± 0,21
	Kidney		18 ± 8	347 ± 149	134 ± 39	3 ± 2	0,4 ± 0,2	2,30 ± 0,45

[a] Number of lambs.

deficiency, which indicates that ewes, grazing pastures with a selenium concentration in excess of 0,03 μg/g DM, would not respond to selenium supplementation. This is in agreement with the results of Caple *et al.*, 1980.

The mean trace element concentrations in the liver and kidneys of the lambs born from ewes in the control, copper- and selenium-supplemented groups are shown in Table 4.

The decrease in the concentrations of plasma copper in the control and selenium-supplemented groups was reflected in the low concentrations of copper in the liver of the lambs. Approximately 40% of the total body copper of a new-born lamb accumulate in the liver, which makes this the key organ for estimating the copper reserves of the young lamb (Grace, 1986).

Present results indicated that when the concentration of plasma copper in the pregnant ewe decreased to 60 μg/dl or less during late pregnancy, it inevitably resulted in low concentrations of liver copper in lambs born from these ewes. The concentrations of liver copper in the lambs born from the ewes without copper supplementation ranged from 5 to 85 μg Cu/g DM. Selenium supplementation of ewes had no effect on the concentrations of selenium in the liver and kidneys of the lambs. Concentrations of iron, zinc, manganese and molybdenum in the liver and kidneys were within normal limits (Georgievski, Annenkov & Samokin, 1979).

Supplementation of copper and selenium had no effect on conception or fecundity of the ewes (Table 5). It must, however, be stated that mating took place during March and April – the period of highest sexual activity in sheep in the Southern Hemisphere (Dyrmondsson, 1973).

It is concluded that inadequate copper intake by, or

Table 5 The % ewes mated as well as the % ewes mated twice in the control (C), copper (Cu)- and selenium (Se)-supplemented groups

Group	n[a]	% Ewes mated	% Ewes mated twice	Conception rate (%)	Fecundity[b]
C	23	100	13	86	1,37
Cu	85	98	20	81	1,31
Se	81	98	17	84	1,36

[a] Number of ewes.

[b] Expressed as the number of lambs born per ewe lambed.

reserves of ewes during late pregnancy result in low (less than 60 µg/dl) concentrations of plasma copper in the ewes and low (less than 50 µg/g DM) concentrations of copper in the liver of their new-born lambs.

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