Phosphorus and the grazing ruminant. 1. The effect of supplementary P on sheep at Armoedsvlakte

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The performance in terms of bodymass and reproduction of two groups of free-grazing Dorper ewes, with (+P) and without (-P) phosphorus (P) supplementation, was investigated over 4,5 years at Armoedsvlakte (notorious for its P-deficient soils) to establish whether sheep are as susceptible to a P deficiency as cattle. The P status was monitored in blood and rib bone samples, taken at different stages of the reproductive cycle. Effects of feed intake on bodymass of both groups are discussed; the mass of - P ewes was 9,6% less than the + P group. The only effect on reproductive performance was that the +P group tended to wean heavier lambs. Levels of P in rib bone samples clearly indicated cyclic changes in the P reserves, associated with changes in physiological status. Bone P levels successfully identified the - P ewes as being deficient in P. S. Afr. J. Anim. Sci. 1986, 16: 1-6

Die prestasie in terme van liggaamsmassa en reproduksie van twee groepe Dorperooie met (+P) of sonder (-P) fosfor(P)-aanvulling, is oor 4,5 jaar in 'n gebied met 'n kenmerkende P-tekort (Armoedsvlakte) ondersoek om te bepaal of skape net so vatbaar is vir 'n P-tekort as beeste. Die P-status is gedurende verskillende stadia van die reproduktiewe siklus met behulp van bloed- en ribbebeenmonsters gemonitor. Die effek van voerinname op liggaamsmassa van beide groepe word bespreek. Die massa van die - P-groep was gemiddeld 9,6% laer oor die proefperiode as dié van die + P-groep. Die enigste effek van P-aanvulling op reproduksie was die neiging van die + Pgroep om swaarder lammers te speen. P-vlakke in beenmonsters het sikliese veranderinge in die P-reserwes met gepaardgaande veranderinge in fisiologiese status aangedui. 'n P-tekort is met behulp van beenontledings in die - P-ooie geïdentifiseer.

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Introduction

Sheep in South Africa can apparently remain normal and healthy in areas where cattle would suffer an extreme P (phosphorus) deficiency (Du Toit, Malan & Rossouw, 1930). Underwood, Shier & Beck (1940, cited by McDonald, 1968) gave P supplements to sheep grazing in an area where soils were severely deficient in P, but found no favourable response to supplementation in growth, wool production, reproductive performance, or growth of lambs. Similar results were obtained by Steenkamp (1967). Anoestrus in sheep, resulting from a P deficiency, has not been recorded according to Hemingway (1967). McDonald (1968) concluded that '... despite a great many tests, there has been no clear demonstration of a primary deficiency in grazing sheep . . . and it remains to be seen whether there are any conditions in which the pasture plants are so tolerant of a low phosphorus supply that sheep can obtain sufficient protein and energy without consuming enough phosphorus. No such situation has yet been recorded.'

Despite these reports, there seems to be some evidence of a P deficiency in sheep, e.g. the bone chewing and wasting disease referred to by Russell & Duncan (1956, cited by Butler & Jones, 1973) which arises from a simple P deficiency in the herbage and which occurs 'world wide in cattle and sheep'; the adverse effects of a P-deficient diet on bodymass gain and feed intake of young lambs (Preston & Pfander, 1964); the improved mass gains and decreased mortality rates following supplementation of sheep during periods of long droughts (Kotze, 1950; Van der Vyver & Van Niekerk, 1965; Cloete, 1971); and the observation of Little, Siemon & Moodie (1978) that despite the absence of any difference in bodymass of ewes and lambs or in the mean interval from weaning to oestrus, between groups with and without supplementary P, supplementation did maintain the level of skeletal P reserves.

Uncertainty therefore exists as to whether sheep, even reproducing ewes (Louw, 1979) benefit from a P supplement, and the aim of this study is therefore to investigate this controversial question: Are sheep susceptible to a P deficiency?

Experimental procedures

This trial was conducted in the north-western Cape, at the Armoedsvlakte Research Station, notorious for its P deficient soil and pastures since the pioneering work of Sir Arnold Theiler at the turn of this century and, therefore, ideally suited for this study. P levels as low as 0,029% P have been recorded at this site (Bisschop, 1964).

Fifty maiden Dorper ewes were randomly assigned to two groups (-P and +P) of 25 each. The -P group had free

access to a salt lick only and the + P group to a lick consisting of 44% salt (NaCl), 44% dicalcium phosphate (diCaP) and 12% molasses powder (Kalori 3000, Kynoch Feeds, Reg No V2809; Act 36 1947). A mean stocking rate of 7,8 ha/LSU, i.e. well within the 7 ha/LSU recommended by the Department of Agriculture, was maintained throughout the trial. The ewes were weighed monthly, feed and water not being withheld prior to weighing. The mating season lasted from 1 December to 31 January and all lambs were weaned at 100 days of age.

Feed intake was determined using the following equation, viz

$$OMI = \frac{100}{100 - \% OMD} \times \frac{OM \text{ excreted } (g/day)}{1}$$

where OMI = organic matter intake; OMD = organic matter digestibility; and OM = organic matter.

The pasture OMD was estimated from samples collected by oesophageally fistulated (OF) ewes and analysed using the modified (Engels & Van der Merwe, 1967), two-stage *in vitro* technique of Tilley & Terry (1963). Digestibility was then predicted using the regression equation of Engels, De Waal, Biel & Malan (1981). Faecal output was determined indirectly, using Cr_2O_3 as the external indicator, viz

$$\frac{OM \text{ excreted}}{(g/day)} = \frac{\text{mass } Cr_2O_3 \text{ dosed/day}}{\frac{1}{\sqrt{0}} Cr_2O_3 \text{ in faecal } DM} \times \frac{\frac{1}{\sqrt{0}} OM \text{ in faeces}}{1}$$

One gelatin capsule containing 1 g Cr_2O_3 was administered twice daily by means of a stomach tube.

Crude protein content (percentage N \times 6,25) of pasture samples collected by OF sheep was determined according to AOAC (1980).

Blood and rib bone samples were taken at different stages of the reproductive cycle (early pregnancy, late pregnancy, lactation and dry) for the duration of the trial. At each of these 'experimental periods', it was usual practice to sample six ewes of each group. Rib bone samples were taken according to the biopsy technique of Little (1972) and analysed for P, Ca, Mg, ash, and specific gravity (SG) as described by De Waal (1979). Although the concentrations of minerals were expressed both as percentage DM (dry matter) and per unit volume (mg/cm³) of fresh bone, the latter form of expression will be adopted, because it appears to be a better indicator of changes in bone mineral content, as was also suggested by Little (1972) and Little & McMeniman (1973).

Blood samples were taken by jugular puncture, gently mixed with three drops of heparin, spun down within 3 h of sampling and then analysed for P, Ca and Mg, according to De Waal (1979).

The trial commenced during January 1979 and continued until June 1983. Data were analysed as a split plot design with repeated measurements over years, this procedure being an approximation of more appropriate multivariate analyses (Winer, 1962).

Results and Discussion

Bodymass and feed intake

The two groups of ewes had the same bodymass (39 kg) when the trial commenced, but during the first pregnancy and lactation the -P ewes already tended to be lighter. This tendency proved consistent throughout the remainder of the experimental period (Figure 1) with the bodymass of the -Pewes being 9,6% lower. The differences between the groups followed a recurring pattern which suggested that each year, the -P group mobilized more of their body reserves during



Figure 1 The bodymass of the Dorper ewes with (+P) and without (-P) phosphorus supplementation $(+P: \circ - \circ)$ and $-P: \bullet - \circ)$.

late pregnancy and early lactation (May – August), resulting in greater differences at the start of the dry, i.e. non-reproducing, non-lactating period (September), although these differences were not significant (P < 0.05) in 1981. Because this dry period coincided with the pasture's spring/summer growing season, it enabled the – P ewes to restore much of their body reserves. The two groups therefore differed only slightly at the following mating season.

Intakes of supplemental P varied between 1,6 and 0,35 g/ewe/day (Read, 1984). The relatively poor response of the + P ewes to this supplement may possibly have resulted from a multiple nutrient deficiency involving not only P but also crude protein (e.g. McDonald, 1968; Myburgh & Du Toit, 1970; Little, 1975; Tuen, Hodge, Smith, Day & Murray, 1982) and/or digestible energy (Leibholz, 1974; McMeniman & Little, 1974) and/or trace minerals.

According to NRC (1975) recommendations, the CPI (crude protein intake) seemed adequate for the dry ewes only and not even for these during 1980 (Table 1). As the CP content of the diet selected by OF sheep varied between 19 and 5% and the OMD between 65 and 48% (Read, 1984), the inadequate CPI's were clearly not due to protein-deficient pastures but rather to poor intakes of DOM. The latter resulted in an energy balance more negative than that of CP, because not even the maintenance requirements were always satisfied (Table 1). Despite this, P-supplemented cattle at Armoedsvlakte satisfied their requirements for both ME and CP (Read, Engels & Smith, 1984). This suggests that the sheep

Table 1 A comparison of the intakes $(g/W_{kg}^{0.75}/day)$ of digestible organic matter (DOM) and crude protein (CP) of the + P ewes with their requirements according to NRC (1975) recommendations^a

		Ď	ОМ	СР		
Year	Physiological status ^b	Intake	Require- ments	Intake	Require- ments	
1979	L	37,6	49,8	3,91	8,40	
	D	38,2	27,5	6,23	4,73	
1980	Р	28,1	47,6	4,89	8,06	
	L	21,9	68,2	2,96	11,60	
	D	26,3	27,5	3,01	4,73	
1 9 81	L	28,9	68,2	2,43	11,60	
	D	33,5	27,1	8,48	4,64	
1982	Р	25,3	31,5	6,66	5,43	
	L	27,8	65,6	6,31	11,09	
	D	20,5	27,1	6,14	4,64	

^aAssuming that 1 g DOM = 16 kJ ME (Engels & Malan, 1975) ^bL = lactating; D = dry; P = pregnant

were unable to consume adequate quantities of the pasture components which they preferred. This supports Freer's (1981) suggestion that a balance is maintained between a lowered level of preference and a decrease in feed intake if the more acceptable components are too sparse to satisfy the animal's appetite readily, a situation not unlike that of 'starving in the midst of plenty' (Stobbs, 1973).

Besides this apparent inability to satisfy their requirements, the -P ewes were further subjected to the depressing effects of a P deficiency on intake, as illustrated in Table 2 where (except for lactation in 1980 which cannot be explained) the +P group had consistently higher intakes of DOM. This depression of intake of the -P sheep may represent the 'initial and basic' effect of a P deficiency as demonstrated with cattle (Little, 1968).

In such studies, where growth and therefore body size is affected by the treatments, feed intake expressed per metabolic size ($W_{kg}^{0,75}$) may not be the most suitable as this denominator tends to cancel the differences in body size. Rather, the severity of the depression in feed intake caused, e.g. by a P deficiency, may be best illustrated when intakes are expressed as total consumed (g/day) as in Table 2.

It was only during 1982 that the CPI of the -P ewes was considerably lower than that of the +P ewes (Table 2) probably because the inadequate DOMI (of -P) was coupled with a tendency for selecting a diet lower in CP content during both pregnancy (12,33 vs 13,54% CP) and the dry period (11,46 vs 15,79% CP) for -P and +P, respectively according to Read (1984).

Reproductive performance

Over the first three lambing seasons (1979 - 1981), P supplementation had little effect on any aspect of reproduction (Table 3). This agrees with other reports (McMeniman & Little, 1974; Little, *et al.*, 1978) and particularly with the conclusion of the latter authors that a short-term deficiency is apparently of no consequence for the subsequent reproductive performance of ewes. A prolonged deficiency may, however, be manifested in a slightly lower reproductive efficiency, as observed during 1982 and 1983 (Table 3), despite the suggestion (Hemingway, 1967) that anoestrus in sheep due to a P deficiency, has '... not yet been recorded' probably

Table 2Digestible organic matter (DOM) intake (g/day)and crude protein (CP) intake (g/day) of the + P and - Pewes

		DOM	l intake	CP intake			
Year	Physio- logical status ^a	$-\mathbf{P} + \mathbf{P}$	Level of signifi- cance ^b	- P	+ P	Level of signifi- cance ^b	
1979	L	423 606	**	52,6	62,9	NS	
	D	575 750	**	102,6	122,4	NS	
1980	Р	572 648	NS	102,8	112,8	NS	
	L	489 416	NS	53,3	56,1	NS	
	D	437 458	NS	50,3	52,4	NS	
1981	L	377 530	**	45,7	44,6	NS	
	D	550 612	NS	164,4	156,8	NS	
1982	Р	402 575	*	91,1	151,6	**	
	L	285 580	**	75,1	131,8	**	
	D	352 415	NS	80,6	123,3	**	

^aL = lactating; D = dry; P = pregnant

^bNS = non significant; *P < 0.05; **P < 0.01

Table	3	Reproductive	performance	and	growth	of
lambs	of	-P and $+P$	ewes		-	

	Treat	Lambing season						
Measurement	ment	1979	1980	1981	1982	1983		
Conception rate (%)	- P	80,0	95,5	85,7	80,0	87,5		
•	+ P	73,1	87,0	81,0	100,0	78,6		
Lambing percentage ^a	- P	80,0	100,0	90,5	95,0	87,5		
	+ P	73,1	91,3	90,5	106,3	92,9		
Weaning percentage ^b	- P	80,0	95,5	66, 7	90,0	81,3		
	+ P	73,1	87,0	85,7	88,2	92,9		
Birth mass (kg)	- P	4,43	4,88	4,19	4,50	4,79		
	+ P	4,68	4,88	4,58	4,97	4,46		
Weaning mass ^c (kg)	- P	20,5	21,3	15,3	20,9	26,1		
	+ P	21,8	24,0	18,0	26,1	25,5		
ADG ^d (g)	- P	161	164	110	163	213		
·	+ P	171	191	131	211	210		

^aDefined as (lambs born/ewes mated) \times 100

^bDefined as (lambs weaned/ewes mated) \times 100

°Corrected for 100 days of age

^dAverage daily gain from birth to weaning

because of the long recovery period from the end of lactation to the start of the following pregnancy (Hill, 1962).

As in the trial of Little, *et al.* (1978), there appeared to be no consistent differences between the birth mass of lambs born to -P and +P ewes, although in contrast to these authors, the +P ewes seemed to wean heavier lambs (Table 3). This may be due to an enhanced milk production of the +P ewes, resulting from the stimulating effect of the P supplement on the intake of these ewes.

Phosphorus status

Changes in bone parameters

After only 5 months deprivation of P, the -P ewes tended to have lower levels of bone mineralization; their percentage rib bone ash was significantly (P < 0,05) lower than that of the +P group during the pregnancy, and all bone parameters investigated differed significantly during the following lactation period (1979). The overall pattern that emerges from Table

Year	Physiological status	Treat- ment	Treat-Mineral concentrationment(mg/cm³)			Ca: P ratio	Ash (%)	SG	
1979	Late pregnancy	- P	114,9 ^a	259,3ª	4,3 ^a	2,26	58,6 ^a	1,56 ^a	
		+ P	128,3 ^a	293,5ª	5,3ª	2,29	62,7 ^b	1,65 ^a	
	Lactation	$-\mathbf{P}$	94,4 ^a	188,0 ^a	3,9 ^a	1,99	52,0 ^a	1,28 ^a	
		+ P	138,1 ^b	261,9 ^b	5,4 ^b	1,90	61,4 ^b	1,52 ^b	
1980	Early pregnancy	– P	104,5ª	228,2 ^a	3,4 ^a	2,18	52,1 ^a	1,43 ^a	
		+ P	142,2 ^a	297,4 ^a	5,0 ^a	2,09	59,9 ^b	1,60 ^b	
	Lactation	$-\mathbf{P}$	115,5 ^a	255,9 ^a	7,8 ^a	2,22	59,5ª	1,51ª	
		+ P	125,4 ^a	268,7 ^a	7,2 ^a	2,14	58,8ª	1,54 ^a	
	Dry	$-\mathbf{P}$	85,5 ^a	151,5 ^a	3,5ª	1,77	46,8 ^a	1,27 ^a	
		+ P	133,1 ^b	263,9 ^b	6,2 ^b	1,98	60,4 ^b	1,58 ^b	
1981	Early pregnancy	$-\mathbf{P}$	122,1ª	246,4ª	4,3ª	2,02	52,4 ^a	1,42 ^a	
		+ P	167,2 ^b	337,8 ^b	6,4 ^a	2,02	63,0 ^b	1,63 ^b	
	Late pregnancy	$-\mathbf{P}$	118,8 ^a	252,5 ^a	2,3ª	2,13	57,3ª	1,52 ^a	
		+ P	146,0 ^a	318,9 ^a	4,2 ^a	2,18	63,2 ^a	1,67 ^a	
	Dry	$-\mathbf{P}$	108,0 ^a	246,4ª	1,4 ^a	2,28	59,9ª	1,50 ^a	
		+ P	135,9 ^b	295,0 ^b	3,2 ^b	2,17	60,7 ^a	1,62 ^b	
1982	Early pregnancy	$-\mathbf{P}$	93,3ª	191,9 ^a	3,6ª	2,06	55,1ª	1,53 ^a	
		+ P	135,3 ^b	267,6 ^b	5,7 ^b	1,98	61,3 ^b	1,68 ^b	
	Late pregnancy	$-\mathbf{P}$	124,9 ^a	262,8 ^a	5,9ª	2,10	57,1ª	1,61 ^a	
		+ P	137,8ª	293,7 ^a	7,7 ^b	2,13	62,3 ^b	1,64 ^a	
	Lactation	$-\mathbf{P}$	80,0 ^a	159,4 ^a	2,7 ^a	1,99	47,0 ^a	1,28 ^a	
		+ P	127,2 ^b	268,7 ^b	5,4 ^b	2,11	59,7 [⊳]	1,57 ^b	
	Dry	$-\mathbf{P}$	85,9 ^a	168,5ª	3,2ª	1,96	49,3ª	1,38 ^a	
		$+\mathbf{P}$	131,7 ^b	250,4 ^b	5,4 ^b	1,90	58,7 ^b	1,57 ^b	
1983	Early pregnancy	- P	105,1 ^a	206,4 ^a	7,4 ^a	1,96	54,3 ^a	1,48 ^a	
		+ P	127.2^{a}	243.2^{a}	6 5 ^a	1 91	58 2 ^a	1 57 ^a	

 Table 4
 Chemical composition of rib bone samples^a

^{a,b}Differences between treatments tested within individual periods and years; treatments with same superscripts do not differ significantly (P < 0.05)

4 is the rather drastic mobilization of bone minerals during lactation (as apparent at the start of the dry period). The -Pewes seemed unable to restore their mineral reserves by the start of the following pregnancy, although by late pregnancy there seemed to have been a slight increase in bone mineralization which decreased drastically during lactation. The +P ewes showed relatively constant levels of mineralization, except during lactation, when a slight tendency for resorption was possible. Like Little, et al. (1978), it may be concluded that supplementary P may have allowed the level of mineral reserves to be maintained. These observations clearly demonstrate the findings of earlier researchers as cited by Little & McMeniman (1973) . . . 'Early lactation is normally a time of resorption of skeletal calcium and phosphorus, which cannot be offset completely by mineral supplements, but the extent to which the skeletal reserves are depleted during the lactation is a function of dietary calcium and phosphorus concentrations'.

Because the CPI of the + P ewes was apparently inadequate for gestation and lactation (Table 1), and that of the – P ewes even lower (Table 2) at times, impaired bone mineralization might be expected in both groups; this is very dependent on protein intake, even overriding the effects of a mineral deficiency via the reduced formation of organic bone matrix (matrix osteoporosis) and thereby bone accretion rate (Sykes & Field, 1972). However, the + P ewes do not seem to have had impaired mineralization of the skeleton according to Table 4, because a P content of between 130 and 140 mg/cm³ fresh bone and a SG in excess of 1,60 are indicative of adequate levels of mineralization (Little & McMeniman, 1973).

Although not expressed on the same basis as those of Belonje & Van den Berg (1983), the composition of rib bone samples for the ewes, clearly reflect differences in the dietary intake of P, thereby completely refuting the title of these authors' article ('Failure of bone phosphorus levels to indicate dietary intake of phosphorus by sheep') and simultaneously supporting the statement (Little & McMeniman, 1973) that '... the method has substantial potential as an index of the phosphorus status of grazing ruminants'.

Mineral levels in blood plasma

There were no significant differences in plasma P_i levels between the -P and +P groups during early pregnancy. During late pregnancy, only the plasma P_i levels of the -Pewes declined sharply, whereas during lactation declines were observed in both groups (Table 5); again illustrating the mobilization of reserves in response to the requirements for milk production. Although more pronounced in the +Pgroup, plasma P_i levels of both groups increased during the dry period, probably because of their lower requirements during this period in addition to their having access to pasture with higher P concentrations (Hemingway, 1967).

Plasma Ca levels (Table 5) only differed significantly (P < 0.05) between the two groups during late pregnancy of 1979 and 1982. Mg levels (mean \pm SEM = 2.22 \pm 0.05 mg/ 100 ml) only differed significantly during late pregnancy of 1979.

Conclusion

P supplementation had no significant effect on the performance of the ewes. However, according to the levels of P in rib bone samples, the -P ewes were severely deficient in P. The supposition that sheep are not susceptible to a P deficiency may have arisen from the lack of any visible

Table 5	P _i and	Ca	levels	(mg/100	ml	plasma)	in	blood	samples ^a
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Year		Physiological status									
	Treat-	Ea	arly nancy	L preg	ate nancy	Lact	ation	D	bry		
	ment	Pi	Ca	Pi	Ca	Pi	Ca	Pi	Ca		
1979	– P + P	_	-	1,80 ^a 4,27 ^b	9,38 ^a 7,24 ^b	1,74 ^a 4,10 ^a	9,24 ^a 8,75 ^a	_	-		
1 9 80	- P + P	3,39 ^a 4,16 ^a	9,43 ^a 8,31 ^a	-	-	1,87ª 3,49ª	9,68ª 8,82ª	2,76 ^a 5,34 ^b	8,96 ^a 8,16 ^a		
1981	– P + P	6,15 ^a 4,80 ^a	10,53ª 9,85ª	3,09ª 4,77ª	8,43 ^a 7,67 ^a	_	-	5,01 ^a 7,70 ^b	8,24 ^a 7,34 ^a		
1982	- P + P	3,35 ^a 5,70 ^b	9,31ª 8,78ª	1,98 ^a 4,53 ^b	11,29 ^a 9,95 ^b	1,43 ^a 4,00 ^b	10,49 ^a 9,72 ^a	3,35 ^a 5,70 ^b	9,31 ^a 8,78 ^a		

^{a,b}Differences between treatments tested within individual periods and years; treatments with same superscripts do not differ significantly (P < 0.05)

symptoms of the condition and/or the absence of any adverse effects on their performance, together with various theories concerning species differences in susceptibility to P deficiency. Firstly, that sheep consume 1,5-2 times as much feed per unit of bodymass than cattle and are therefore more tolerant of low pasture P levels (McDonald, 1968); secondly, that with their smaller mouths and different mechanics of grazing, sheep are more selective grazers (e.g. Van Dyne & Heady, 1965; Dudzinski & Arnold, 1973; Engels, Ferreira, Swart & Niemann, 1978; Arnold, 1981) and therefore better able to select plant fragments (like young developing leaves) richer in P (Hemingway, 1967); thirdly, that cows are either pregnant or lactating or both every day of the year, whereas sheep have a non-productive period during which their condition may be restored (Louw, 1979), although McDonald (1968) contradicted this theory; and lastly, that sheep may retain P per unit of metabolic size more efficiently than cattle (Cohen, 1974). However, the implementation of a more sensitive and reliable indicator of the P status (bone P levels) in this trial, has enabled the identification of a P deficiency in sheep, thereby confirming that sheep are susceptible to this condition.

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References

- AOAC, 1980. Official methods of analysis of the Association of Official Agricultural Chemists, (13th Ed.). Washington, D.C.
- ARNOLD, G.W., 1981. Grazing behaviour. In: Grazing Animals. Ed. Morley, F.H.W., Elsevier Scientific Publishing Company, Amsterdam, p. 79.
- BELONJE, P.C. & VAN DEN BERG, A., 1983. Failure of bone phosphorus to indicate dietary intake of phosphorus by sheep. Onderstepoort J. vet. Res. 50, 1.
- BISSCHOP, J.H.R., 1964. Feeding phosphates to cattle. Dept. Agric. Tech. Services. Science Bull. No. 365.
- BUTLER, G.W. & JONES, D.I.M., 1973. Mineral biochemistry of herbage. In: Chemistry and Biochemistry of Herbage. vol. 2. Eds. Butler, G.W. & Bailey, R.W., Academic Press, London, p. 127.
- CLOETE, J.G., 1971. Drought feeding of sheep. S. Afr. J. Anim. Sci. 1, 201.
- COHEN, R.D.H., 1974. Phosphorus nutrition of beef cattle. 4. The use of faecal and blood phosphorus for the estimation of phosphorus intake. *Aust. J. exp. Agric. Anim. Husb.* 14, 709.

- DE WAAL, H.O., 1979. Die voedingswaarde van veldweiding van die sentrale Oranje-Vrystaat vir skape met spesiale verwysing na die rol van proteïen- en fosforaanvullings. M.Sc.-tesis, Universiteit Stellenbosch.
- DUDZINSKI, M.L. & ARNOLD, G.W., 1973. Comparison of diets of sheep and cattle grazing together on sown pastures on the southern tablelands of New South Wales by principal components analysis. *Aust. J. Agric. Res.* 24, 899.
- DU TOIT, P.J., MALAN, A.I. & ROSSOUW, S.D., 1930. Studies in mineral metabolism. XII. Phosphorus in the sheep industry. Preliminary report. 16th Report Dir. Vet. Serve. Anim. Ind. Union of South Africa, 313.
- ENGELS, E.A.N., DE WAAL, H.O., BIEL, L.C. & MALAN, A., 1981. Practical implications of the effect of drying and treatment on nitrogen content and *in vitro* digestibility of samples collected by oesophageally fistulated animals. S. Afr. J. Anim. Sci. 11, 247.
- ENGELS, E.A.N., FERREIRA, B., SWART, J.A. & NIEMANN, P.J., 1978. Comparative feed intake and digestibility studies with sheep and cattle on roughages. S. Afr. J. Anim. Sci. 8, 149.
- ENGELS, E.A.N. & MALAN, A., 1975. The energy intake and excretion of sheep grazing oat pasture. *Agroanimalia* 7, 81.
- ENGELS, E.A.N. & VAN DER MERWE, F.J., 1967. Application of an *in vitro* technique to South African forages with special reference to the effect of certain factors on the results. S. Afr. J. Agric. Sci. 10, 983.
- FREER, M., 1981. The control of food intake by grazing animals. In: Grazing Animals. Ed. Morley, F.H.W., Elsevier Scientific Publishing Company, Amsterdam. p. 105.
- HEMINGWAY, R.G., 1967. Phosphorus and the ruminant. Outlook on Agriculture 5, 172.
- HILL, R., 1962. The provision and metabolism of calcium and phosphorus in ruminants. *Wld. Rev. Nutr. Diet.* 3, 129.
- KOTZE, J.J.J., 1950. Sheep farming in the sour-grassveld area. I. Influence of the lambing season. II. Mineral licks and dosing for internal parasites. Bulletin 294. Dept. Agric. Union of SA, The Govt. Printer, Pretoria.
- LEIBHOLZ, JANE, 1974. The flow of calcium and phosphorus in the digestive tract of the sheep. Aust. J. Agric. Res. 25, 147.
- LITTLE, D.A., 1968. Effect of dietary phosphate on the voluntary consumption of Townsville lucerne (*Stylosanthes humilis*) by cattle. *Proc. Aust. Soc. Anim. Prod.* 7, 376.
- LITTLE, D.A., 1972. Bone biopsy in cattle and sheep for studies of phosphorus status. Aust. vet. J. 48, 668.
- LITTLE, D.A., 1975. Effects of dry season supplements of protein and phosphorus to pregnant cows on the incidence of first post-partum oestrus. *Aust. J. exp. Agric. Anim. Husb.* 15, 25.
- LITTLE, D.A. & McMENIMAN, N.P., 1973. Variation in bone composition of grazing sheep in south-western Queensland, related to lactation and type of country. *Aust. J. exp. Agric. Anim. Husb.* 13, 229.

LITTLE, D.A., SIEMON, N.J. & MOODIE, E.W., 1978. Effects-

of varying phosphorus intake and requirements on measures of skeletal mineralization in the ewe. Aust. J. exp. Agric. Anim. Husb. 18, 514.

- LOUW, G.N., 1979. An evaluation of the application of stock licks in South Africa. S. Afr. J. Anim. Sci. 9, 133.
- McDONALD, I.W., 1968. The nutrition of grazing ruminants. Nutr. Abstr. Rev. 38, 381.
- McMENIMAN, N.P. & LITTLE, D.A., 1974. Studies on the supplementary feeding of sheep consuming mulga (Acacia aneura). I. The provision of phosphorus and molasses supplements under grazing conditions. Aust. J. exp. Agric. Anim. Husb. 14, 316.
- MYBURGH, S.J. & DU TOIT, J. DE V., 1970. A comparative study on the efficiency of water soluble phosphates and a dry phosphate lick for sheep fed on mature grass hay. *Onderstepoort J. vet. Res.* 37, 127.
- NATIONAL RESEARCH COUNCIL, 1975. Nutrient requirements of domestic animals. 5. Sheep. 5th rev. Ed. National Academy of Sciences, Washington.
- PRESTON, R.L. & PFANDER, W.H., 1964. Phosphorus metabolism in lambs fed varying phosphorus intakes. J. Nutr. 83, 369.
- READ, MARION V.P., 1984. Animal performance from natural pastures and the effects of phosphorus supplementation. M.Sc. (Agric) thesis, University of Stellenbosch.
- READ, MARION V.P., ENGELS, E.A.N. & SMITH, W.A., 1984. Phosphorus and the grazing ruminant. 2. The effect of

supplementary P on cattle at Glen and Armoedsvlakte. S. Afr. J. Anim. Sci. 16, 7.

- STEENKAMP, C.W.P., 1967. Die voedingswaarde van die weidings in die Petrusville-Hopetownse sandveldgebied. M.Sc.tesis, Universiteit Stellenbosch.
- STOBBS, T.H., 1973. The effect of plant structure on the intake of tropical pastures. I. Variation in the bite size of grazing cattle. *Aust. J. Agric. Res.* 24, 809.
- SYKES, A.R. & FIELD, A.C., 1972. Effects of dietary deficiencies of energy, protein and calcium on the pregnant ewe. I. Body composition and mineral content of the ewes. J. agric. Sci., Camb. 78, 109.
- TILLEY, J.M.A. & TERRY, R.A., 1963. A two-stage technique for the *in vitro* digestion of forage crops. J. Brit. Grassld. Soc. 18, 104.
- TUEN, A.A., HODGE, P.B., SMITH, P.C., DAY, P. & MURRAY, R.M., 1982. Mineral supplementation of Bos indicus cross cattle grazing native pastures in tropical Queensland. Proc. Aust. Soc. Anim. Prod. 14, 317.
- VAN DER VYVER, P.H.B. & VAN NIEKERK, B.D.H., 1965. Supplementary feeding of sheep on winterveld in the Karoo. *Farming in S.A.* 41, 15.
- VAN DYNE, G.M. & HEADY, H.F., 1965. Dietary chemical composition of cattle and sheep grazing in common on a dry annual range. J. Range Manage. 18, 78.
- WINER, B.J., 1962. Principles of experimental design. McGraw Hill: New York.

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