

Antelope and poisonous plants: 1. Gifblaar *Dichapetalum cymosum* (Hooker) Engler & Prantl containing monofluoroacetate

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ABSTRACT

Ten tame eland (*Taurotragus oryx*), four kudu (*Tragelaphus strepsiceros*), two gemsbok (*Oryx gazella*), six springbok (*Antidorcas marsupialis*) and 33 domestic goats were drenched by stomach tube with extracts of the poisonous plant gifblaar (*Dichapetalum cymosum*) (Hooker) Engler and Prantl (= *D. venenatum* Engler and Gilg). Semi-quantitative analyses of the monofluoroacetate (MFA) content of the leaves were conducted at regular intervals and the proven lethal (ca. LD₁₀₀) caprine dose was found to be equivalent to 1.01–1.60 mg/kg MFA. Eland and kudu only succumbed at high dosage levels of 6–8 mg/kg MFA and proved to be much less susceptible to gifblaar poisoning than goats. Springbok and gemsbok were as susceptible as goats, although confirmation is needed in gemsbok to eliminate possible interaction between the immobilising drugs used and MFA. During separate voluntary intake trials, eland ate limited offerings of both low and highly toxic stages of gifblaar. At these dosages they showed no ill effects. Kudu on the other hand were more cautious and intake was minimal.

The lesions caused by gifblaar are described. A few macroscopic lesions observed in some of the animals have not been recorded previously. These include: oedema and haemorrhages of the gall bladder with occasional blood-stained bile; oedema of the abomasum, periportal area of the liver, pancreas and pulmonary valves; adrenal haemorrhages and petechiae in the urinary bladder. Histopathological changes were described in various organs such as the myocardium where replacement fibrosis became evident in animals which survived for two or more days.

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1 INTRODUCTION

One of the most potent known poisons, monofluoroacetate (=sodium fluoroacetate) (MFA), which causes blockage of the Krebs cycle and cardiac arrest in ruminants, is found in the plant gifblaar (*Dichapetalum cymosum*) (Hooker) Engler and Prantl (= *D. venenatum* Engler and Gilg) (Steyn, 1928; Marais, 1944; Steyn, 1949). Gifblaar occurs in large areas of southern Africa including the north-eastern part of South West Africa/Namibia (Map 1). Because it causes heavy annual mortality in domestic stock, gifblaar was selected for the first series of experiments, to determine the susceptibility of antelope to indigenous toxic plants.

The investigation was conducted in the Grootfontein district of South West Africa/Namibia and consisted of two separate studies. The first experiment was to determine the voluntary intake of gifblaar leaves while the second experiment comprised toxicity trials where the animals were dosed with an extract of the plant material.

2 EXPERIMENT 1:
VOLUNTARY INTAKE

2.1 Procedure

For more than a year, sublethal doses of specific quantities of counted leaves of gifblaar were periodically offered to a varying number of eland (*Taurotragus oryx*), kudu (*Tragelaphus strepsiceros*), springbok (*Antidorcas marsupialis*) and cattle. All these animals were starved and water was withheld for 24 hours before exposure. The number of animals that ingested gifblaar as well as the number of leaves ingested within 1–3 hours was determined. The intake was correlated with the MFA content at the time (Table 1).

During the determination of voluntary intake, a sample of each batch of leaves was dried and submitted to Onderstepoort for MFA assay. The moisture content lost by the process of drying was determined during May

(44%) and December (60–70% depending on the age of the leaves).

The dried leaves were thoroughly ground in a laboratory hammermill at Onderstepoort (0,5 mm mesh) and suspended in water. Guinea pigs (*ca.* six weeks of age) which were starved and withheld from water for 24 hours were dosed with this suspension by means of a dosing catheter. Food and water were given *ad lib* afterwards. The 24 hours and 48 hours LD₅₀ of gifblaar for each batch was determined using the method of Litchfield and Wilcoxon (1948). The LD₅₀ of the sodium salt of MFA (sodium fluoroacetate 98%, Purum Fluka AG) for guinea pigs was determined in the same way and found to be 0,5 (0,404–0,62) mg/kg.

The % MFA in the dried leaves was calculated from these findings (Table 1). Consequently, by considering the loss of moisture determined previously, the approximate MFA content for each batch of green leaves could be calculated.

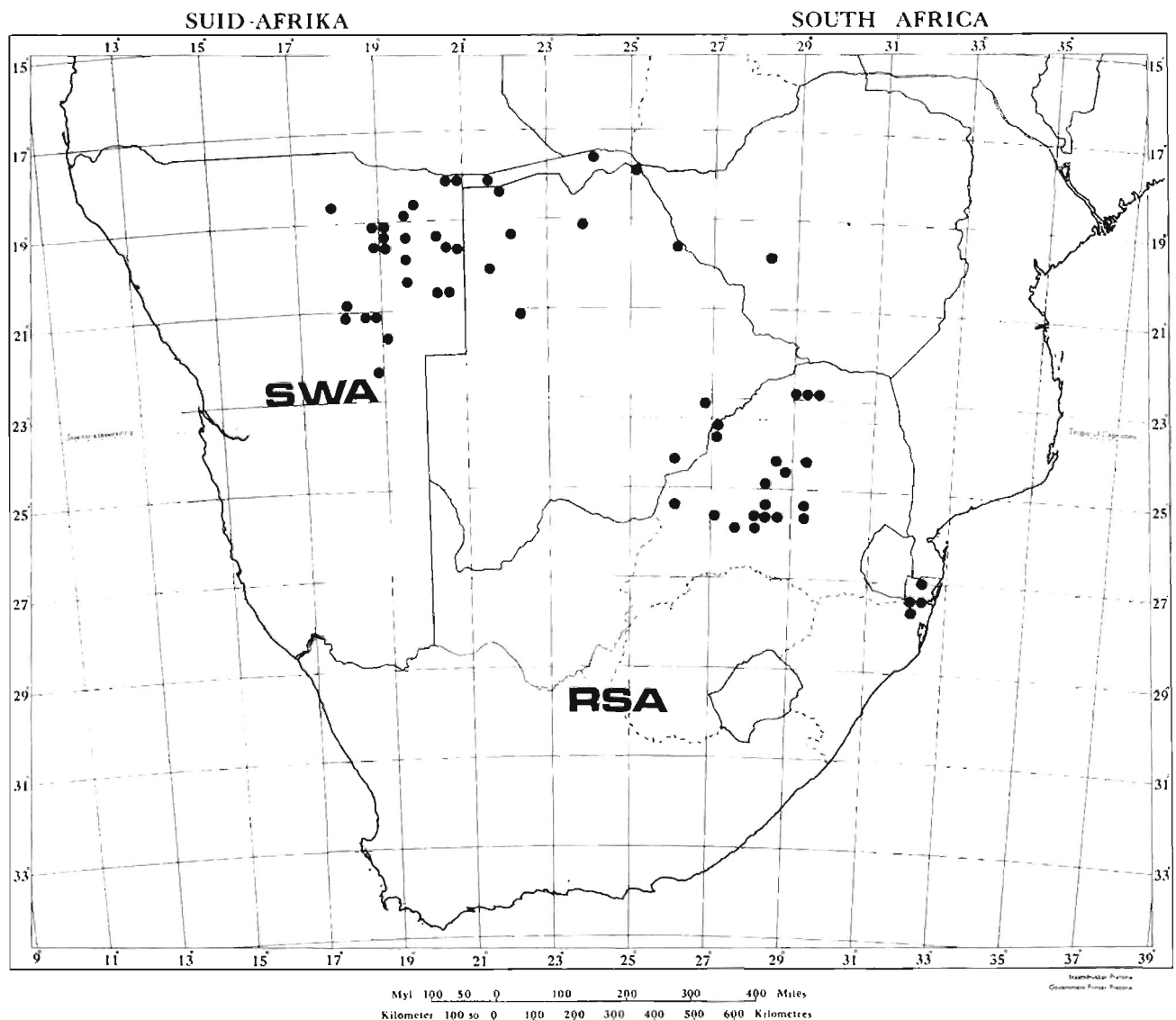


FIGURE 1: The distribution of gifblaar (*Dichapetalum cymosum* (Hooker) Engler & Prantl (National Herbarium, Pretoria, and SWA/Namibia Herbarium, Windhoek, Nov. 1980). Each dot represents a confirmed locality.

2.2 Results

Contrary to previous findings (Norval and Basson, 1974) tame eland frequently ingested varying quantities of gifblaar leaves (0–80) of both low and high toxic content. Some individuals seemed to be more cautious and either avoided gifblaar completely or only ingested a small number of leaves. The findings were the same for cattle and sheep but kudu were much more wary and their intake of gifblaar was both exceptional and limited. There was no correlation between intake and MFA content of the plant.

3 EXPERIMENT 2: TOXICITY TRIALS

3.1 General procedure

Extracts from the leaves of *D. cymosum* were prepared as follows. Plants were collected during September–October, which according to Steyn (1928) is the most toxic period of the plant. After defoliation, the mid-veins of the older and coarser leaves were usually removed and discarded. The mass of the leaves was determined on a triple beam balance. A suspension was either prepared within 2–3 hours or the leaves were stored at 4°C and a suspension prepared a few days later.

As the toxicity varied considerably from week to week, the extracts are dealt with separately (Table 2, A–E). Consequently the results of the dosing trials with the various extracts should be compared separately rather than jointly (Table 6). A similar suspension was prepared from old leaves collected from less toxic plants during May (Table 2, extracts F and G). Samples of these leaves were dried in the shade at room temperature for 24 hours (extract G2) and eight days (extract G3) before the extracts were prepared. Moisture loss was 33% and 44,16% respectively.

Since the active principle, MFA, is water soluble (Marais, 1944), cold tap water was used as a medium

TABLE 1: MFA assays of dried gifblaar

Date	24 and 48 h LD ₅₀ for guinea pigs g/kg (confidence limits in parentheses)	% MFA
May 1975	1,0 (0,48–2,1)	*0,05 (0,019–0,125) Extract G ₁₋₃
28.10.1975	0,65 (0,47–0,9)	0,077 (0,044–0,127)
23.12.1975	2,5 (1,94–3,22)	0,02 (0,0125–0,031)
5.2.1976	3,5 (2,8–5,25)	0,0143 (0,007–0,0215)
16.3.1976	0,7 (0,36–1,36)	0,071 (0,029–0,16)
13.5.1976	0,79 (0,53–1,17)	0,063 (0,034–0,113)
23.6.1976	1,48 (1,11–1,97)	0,034 (0,02–0,054)
9.9.1976	0,1 (0,064–0,155)	*0,5 (0,25–0,93) Extract D ₁
13.9.1976	0,17 (0,108–0,267)	*0,29 (0,15–0,55) Extract D ₂
15.12.1976	1,6 (1,06–2,4)	0,031 (0,016–0,056)
6.9.1977	0,25 (0,178–0,350)	*0,20 (0,114–0,377) Extract E

*Some of the extracts were prepared from these batches (as indicated)

for suspension and extraction. Blending was done in a Waring Blender and the suspension was left for ca. 12 hours overnight in closed containers at room temperature before being used for dosing. The suspension was strained through gauze or hessian to facilitate dosing by stomach tube with an automatic Phillips “Vis-a-matic” drencher or a vacuum pump. Details about the processing are contained in Table 2. The MFA content was assayed in extracts D₁, D₂, E and G₁₋₃ (Tables 1 and 2). Ten eland, four kudu, two gemsbok (*Oryx gazella*), six springbok and 33 domestic goats were used as experimental animals. All of them, except the eland, came from areas where gifblaar does not occur. The individual animals were dosed with the watery extract (Table 2) to determine their susceptibility to gifblaar. As the assays at Onderstepoort could only be conducted after completion of the dosing trials, the degree of toxicity of gifblaar and ca. LD₁₀₀ for goats had to be determined by dosing them with the various extracts.

Immediately before dosing the mass of each animal was determined, either with a portable cattle scale or a large spring balance. Except for the first eland and some springbok, the animals were starved for 24 hours before

TABLE 2: Watery extracts of *D. cymosum*: procedure

Extract	A ₁	A ₂	B ₁	B ₂	C	D ₁	D ₂	E	F	G ₁	G ₂	G ₃
Date collected	1974 14/10	1974 14/10	1974 25/9	1974 25, 27/9	1974 25, 27/9	1976 8/9	1976 13/9	1977 6/9	1975 5/5	1975 12/5	1975 12/5	1975 12/5
Date blended	14/10	15/10	25/9	30/9	2/10	8/9	13/9	6/9	5/5	12/5	13/5	26/5
Extraction time (h)	12–14	12	13	15	13	12	12	12	12	12	12	12
Mass of leaves (g)	505	1334	314	2027	480	4244	1360	3815,5	558	1555,7	247*	804*
Mass of mid-veins removed (g)	20	—	61,5	27	—	—	—	—	—	—	—	—
Balance of leaves (g)	485	1334	304	1965,5	453	4244	1360	3815,5	558	1355,7	247	804
Water added (ml)	5200	13150	2000	12666	2500	25429	8160	22893	5250	10840	2100	7000
Residue (after straining) (g)	388,5	1173	294,5	1508	360	3600	1208	3575	530	1401	419,5	1683
Balance of extract (ml)	4680	12950	1890	11325	2066	23435	8080	20600	ND	ND	ND	ND
Concentration of extract (ml extract/g of leaves)	9,65	9,7	6,2	5,76	4,56	5,5	5,94	5,4	±9,0	±8,0	±6,5	±8,7
% MFA in dried leaves	ND	ND	ND	ND	ND	0,5	0,29	0,2	ND	0,05	0,05	0,05

ND = Not determined
* = Dried material

they were weighed and drenched. The majority of antelope was caught in a crush without the aid of chemical agents, but where indicated, some animals were immobilised with a combination of etorphine HC1 (M99, Reckitt) and a neuroleptic, using a pneumatic Palmer Cap-Chur gun (Tables 7 and 8). The antidote, cyprenorphine (M285, Reckitt) was given soon after dosing to prevent possible interaction between etorphine and MFA.

After drenching, animals had access to food and water, and were kept under close observation for 24–48 hours. Those animals which survived were observed for a further 12–14 days to determine the degree of recovery or the onset of latent effects.

Autopsies were carried out wherever possible and organ specimens were collected in buffered 10% formalin from all the animals that had died. The tissues were prepared for histopathological studies in a routine manner. The sections of 3 to 5 μ in thickness were stained with haematoxylin and eosin. Frozen sections of some livers, kidneys and myocardium were stained with oil red O.

3.2 Goats

3.2.1 Dosing

A total of 27 goats was drenched with various extracts (Table 3, extracts A–E) prepared during September–October (spring) of 1974, 1976 and 1977. Dosages which were calculated on the basis of fresh leaves, ranged from 0,5–4,0 g/kg. During May 1975 (early winter) another six goats were used (Table 5, extracts F–G). Dosage rates of fresh leaves (extracts F & G₁) and dried leaves (extracts G₂ & G₃) in these animals ranged from 3–8 g/kg, calculated on a wet basis.

3.2.2 Results

The onset of symptoms in many of the goats dosed with the spring extracts of gifblaar was usually within 2 to 3 hours. Initially they showed indolence and inappetence and preferred a recumbent position. When aroused they would walk a short distance only to adopt the same position once more. Shady areas were preferred. The animals urinated frequently and appeared uneasy and depressed.

Their gait was often slow and ataxia was usually a very ominous sign. Tachycardia and dyspnoea or polypnoea with gasping were noted in the advanced stages. Bleating of a moaning nature occurred frequently. Some ataxic animals showed circling movements, excitement and terminal spasms. Lateral recumbency with spasms or galloping movements similar to heartwater were seen occasionally. Spells of excitement characterised by sudden bewildered running, often into fences and bushes, and with fatal spasmodic collapse were noticed with the highly toxic material of 1976. Where an extract

TABLE 3: Gifblaar experiment: September–October: goats

Goat No.	Extract	Sex	Equivalent leaf dosage (g/kg)	Onset symptoms (h)	Death (h)
1	A ₁ (1974)	F	0,5	—	—
2	A ₁	F	1	2	—
3	A ₁	F	2	2	—
4	B ₁ (1974)	F	2	2–3	18
5	B ₁	F	3	2–3	6
6	A ₁	F	4	2	5
7	B ₁	F	4	2–3	17
8	B ₂ (1974)	F	4	2	17
9	D ₁ (1976)	F	4	±3	5
10	D ₁	F	3	±3	6
11	D ₁	F	2	±3	±60
12	D ₁	F	2	±3	4
13	D ₂ (1976)	F	2	±3	12
14	D ₂	F	2	±3	±20
15	D ₂	F	2	±3	±44
16	D ₂	F	3	3	±20
17	D ₂	F	3	3	8
18	D ₂	F	3	3	±8
19	D ₂	F	3	3	±12
20	D ₂	F	1	3	±44
21	D ₂	F	1	3	21
22	E (1977)	F	4	2–3	±16
23	E	M	3	2–3	±16
*24	E	F	2	2–3	33×24
25	E	F	1	2–3	—
26	E	F	0,5	2–3	—
27	E	M	0,5	—	—

h = Post-drenching period in hours

* = Died after 33 days

equivalent to 3–4 g/kg of fresh leaves proved to be an *ca.* LD₁₀₀ for goats during spring 1974, the toxicity of leaves during the same period in 1976 increased so markedly that 1 g/kg proved to be lethal to goats (Table 6). It could have been even less, as lower dosages were not administered.

TABLE 4: Gifblaar experiment: September–October: antelope

Antelope	Extract	Sex	Dosage (g/kg)	Onset symptoms (h)	Death (h)
Gemsbok					
OG 1	B ₂	F	3	5	10
OG 2	B ₂	M	4	4	10
Springbok					
AM 1	B ₂ (1974)	M	4	?	20
AM 2	C (1974)	F	4	5	6
AM 3	C	M	3	5	6
AM 4	E (1977)	F	4	?	6
Kudu					
TS 1	B ₂	M	3	?	—
TS 2	B ₂	M	4	—	—
TS 3	D ₂	F	4	±4	±24
TS 4	D ₁	F	3	±4	—
Eland					
TO 1	A ₂	M	4	?	—
TO 2	D ₁	M	4	?3	26
TO 3	D ₁	M	3	?3	—
TO 4	D ₁	M	4	?3	—
TO 5	E	F	4	—	—
TO 6	E	F	3	—	—
TO 7	E	F	3	—	—
TO 8	E	F	2	—	—

h = Post-drenching period in hours

In those goats which received extracts of the May collection, the onset of clinical signs was delayed, rather uncertain and not as distinct as in the previous group. For instance, two goats which received extract F (Table 5) were mostly recumbent and unwilling to move. One goat (dosage 8 g/kg) died 16 hours and the other (4 g/kg) 48 hours after dosing.

Secondly, three goats which were given extract G₁ at dosages ranging from 3–5 g/kg, showed inappetence for a 3–4 hour period which commenced approximately 2 hours after dosing. Nothing more dramatic was noticed until 30 hours when one animal (dosage rate 5 g/kg) became recumbent and refused to eat. This goat bleated in a moaning way and remained recumbent for almost 40 hours. It eventually recovered. The other two goats also showed inappetence at 30 hours and developed peculiar spells of excitement at 48 hours. Shaking movements of the head, kicking at the flanks, sudden running with sternal recumbency were repeatedly seen. However, both animals recovered.

Thirdly, a goat which was dosed with extract G₂ at 8 g/kg showed anorexia and an inclination to sternal recumbency at 10 hours. Almost continuous bleating followed at 21 hours. The animal then adopted a lateral recumbent position in which it died at 32 hours. Galloping movements were seen at times before death.

TABLE 5: Gifblaar experiment: May

	Extract	Sex	Equivalent leaf dosage (g/kg)	Onset symptoms (h)	Death (h)
Goat					
28	F	F	4	3–4	48
29	F	F	8	3–4	16
30	G ₁	F	3	3–4, 48	—
31	G ₁	F	4	3–4, 48	—
32	G ₁	F	5	3–4, 30	—
33	G ₂	F	8	10	32
Springbok					
AM 5	G ₁	M	2,7	?	17
AM 6	G ₁	M	3,9	?	27
Eland					
TO 9	G ₁	M	4	—	—
TO 10	G ₃	F	8	—	—

h = Post-drenching period in hours

Extracts G₂ and G₃ were prepared from dried leaves and the dosage calculated on a wet basis.

Apart from differences in the clinical picture, the *ca.* LD₁₀₀ for goats was considerably higher (*ca.* 8 g/kg) when the May extracts were used. It was important to consider these factors and establish the toxic levels of the different extracts in goats in order to calculate dosage rates in the antelope and determine their susceptibility to gifblaar poisoning.

Finally, the MFA assays indicated that the *ca.* LD₁₀₀ of MFA for goats was found to be equivalent to 1,01–1,60 mg/kg (Table 6).

3.3 Eland

These animals originated from a known gifblaar area in the southern Kavango. The eland which were yearlings and sub-adults, were in captivity for at least six months before commencement of the trials. They had access to natural browse and were fed lucerne hay and Rider cubes (Epol) twice daily. Rock salt and fresh water were available *ad lib.*

3.3.1 Dosing

One two-year-old bull (TO 1) which was not starved previously, was immobilised (Table 7) and dosed by stomach tube with the October extract (A₂). Nine other eland were dosed during various seasons without the aid of any immobilising agents. They were herded into a crush after a starvation period of *ca.* 24 hours. Seven eland (TO 2–8) received the highly toxic material (Table 4, extracts D and E) collected during spring (1976 and 1977), whereas two eland (TO 9 and TO 10) were drenched with the May extracts (Table 5, G₁ and G₃). The details about the procedures are given in Tables 4, 5 and 7.

3.3.2 Results

Because eland TO 1 was not starved beforehand, there was an increase in pressure on the diaphragm during drenching which was caused by the unusually large volume of the extract on the full rumen of the recumbent and immobilised animal. This produced such an adverse spasmodic reaction that cardiac arrest was feared. The reaction was too soon for the toxic agent to have had any effect, as a minimum interval after dosing of approximately 2 hours is required for the appearance of symptoms and 5 hours for death to occur. This is in accordance with the known latent period required for MFA (Jensen, Tobiska and Ward, 1948; Robinson, 1970; Vickery and Vickery, 1973).

Following the administration of the antidote the animal rose and did not reveal any sign of intoxication until 5 hours later when the following symptoms were noticed: sternal recumbency, uneasiness, inappetence, salivation with mouth slightly opened and occasional protrusion of the tongue. The symptoms disappeared within an hour and an uneventful recovery was made.

The three eland drenched with the most toxic extract (D₁) did not show any serious symptoms. They were found to be recumbent at times but rose whenever approached. Recumbency was always interrupted by periods of normal browsing. All of them appeared normal, quite at ease and were found feeding the following day. However, one animal (TO 2) (dosage rate 4 g/kg) died suddenly a few hours after this observation.

Of four eland dosed with the E extract (Sept. 1977), none showed any symptoms of gifblaar poisoning. The

two eland (TO 9 and TO 10) which received the May extract (4 and 8 g/kg respectively) did not develop any untoward effects.

Based on the MFA assays, at least 6–8 mg/kg MFA was required to kill one of two eland (Table 6).

3.4 Kudu

The kudu were caught in the south-western Otjo-vasandu area of the Etosha National Park. They were accommodated for 38 days before the studies commenced. Their natural diet was substituted twice daily with lucerne hay and Rider cubes and they had free access to water.

3.4.1 Dosing

Two young kudu bulls (TS 1 and TS 2) were immobilised 40 hours after starvation and drenched by stomach tube with gifblaar extract B₂ (dosage rate 3 and 4 g/kg respectively) (Tables 4 and 8). For close observation they were retained in a wooden pen for 24 hours and provided with water and their usual ration. They were subsequently released in a small camp for further observation for another 12 days. Two adult cows (TS 3 and TS 4) were caught in a crush without the aid of any immobilisation and dosed with the highly toxic extract

D₁ (Sept. 1976) at 4 and 3 g/kg respectively (Table 4). They were released in a small camp for observations.

3.4.2 Results

The youngest kudu bull (TS 2) which received 4 g/kg did not show any signs of poisoning at any stage after dosing. His appetite was unaffected and both food and water were taken within a few minutes after the administration of the antidote against the immobilising agent. However, kudu TS 1 (dosage rate 3 g/kg) was inclined to recumbency, but rose whenever disturbed or approached and appeared normal otherwise. It showed no interest in food or water. The next morning both kudu were released into a small camp where food and water were taken normally. No other symptoms developed.

The two kudu cows (TS 3 and TS 4) were apparently affected 3 hours after dosing. Both animals were inclined to sternal recumbency and, although intervals of browsing did occur in the late afternoon, they were more frequently seen in a recumbent position than the four eland which were dosed on the same day. The following day (21 hours post-drenching) both cows were recumbent, more listless and oppressed. Shivering and stretching of the neck occurred in both animals. TS 3 (4 g/kg leaves or 6–8 mg/kg MFA) (Table 6) died approximately 24 hours after dosing. TS 4 (3 g/kg) remained recumbent the whole day and appeared to be oppressed. However, she recovered uneventfully on the third day.

TABLE 6: Seasonal comparison of dosing trials

Date collected	% MFA in dried leaves	Dosage		Goats	Eland	Kudu	Springbok	Gemsbok
		Leaves g/kg	MFA (approx.) mg/kg					
Sept. 1974		0,5		0/1				
		1,0		0/1				
		2,0		0/1				
		3,0		0/1				
		4,0		2/2				
Sept. 1976	0,29	1,0	1,01 (0,87–1,16)	2/2				
	0,29	2,0	2,01 (1,74–2,32)	3/3				
	0,5	2,0	3,5 (3–4)	2/2				
	0,5*	3,0	5,25 (4,5–6)	5/5				
	0,5*	4,0	7 (6–8)	1/1				
Sept. 1977	0,2	0,5	0,4	0/2				
		1,0	0,8	0/1				
		2,0	1,6	1/1				
		3,0	2,4	1/1				
		4,0	3,2	1/1				
Oct. 1974		2,0		1/1				
		3,0		1/1				
		4,0		1/1				
May 1975	0,05	3,0	0,84	0/1				
		4,0	1,12	1/2				
		5,0	1,4	0/1				
		8,0	2,24	2/2				

The ratios (fifth column) e.g. 1/2 represent the mortalities (left) out of total number dosed (right).

* Some goats received the D₂ Extract (0,29% MFA), but the antelope D₁ Extract (0,5% MFA).

TABLE 7: Details about the dosing programme of cland

No.	TO 1	TO 2	TO 3	TO 4	TO 5	TO 6	TO 7	TO 8	TO 9	TO 10
Date	Oct. 74	Sept. 76	Sept. 76	Sept. 76	Sept. 77	Sept. 77	Sept. 77	Sept. 77	May 75	May 75
Sex	M	M	M	M	F	F	M	F	M	F
Approx. age (years)	2	±1	±1	±1	2	2	2	2	1,5	1,5
Mass of animal (kg)	170	136	116	129	180	220	175	167	145	180
Dosage: Leaves (g/kg)	4	4	3	4	4	3	3	2	4	8
Total dose (ml)	6596	3003	1921	2848	3887	3563	2835	1803	4000	6100
Extract dosed	A ₂	D ₁	D ₁	D ₁	E	E	E	E	G ₁	G ₃
Immobilising agents (mg):										
M99 ¹ (etorphine hydrochloride)	4	—	—	—	—	—	—	—	—	—
Azaperone ²	100	—	—	—	—	—	—	—	—	—
Immobilisation time (min./s.)	12/0	—	—	—	—	—	—	—	—	—
*Interval (min.)	38	—	—	—	—	—	—	—	—	—
Antidote: M285¹ (cyprenorphine) (mg)	12	—	—	—	—	—	—	—	—	—
Recovery time (min./s.)	ND	—	—	—	—	—	—	—	—	—
Starvation (h)	—	24	24	24	24	24	24	24	24	24
Water withheld (h)	—	24	24	24	24	24	24	24	24	24

*Interval = time between dosing and administration of antidote
 ND = not determined

¹Reckitt
²Janssen Pharm.

TABLE 8: Details about dosing programme of antelope

No.	Kudu				Gemsbok		Springbok					
	TS 1	TS 2	TS 3	TS 4	OG 1	OG 2	AM 1	AM 2	AM 3	AM 4	AM 5	AM 6
Date	Sept. 74	Sept. 74	Sept. 76	Sept. 76	Sept. 74	Sept. 74	Sept. 74	Sept. 74	Sept. 74	Sept. 77	May 75	May 75
Sex	M	M	F	F	F	M	M	M	M	F	M	M
Approx. age (years)	1,5	0,6	Adult	Adult	1	1	2	1	1	±2	±3	±3
Mass of animal (kg)	153	78	146	144	68	88	33	25	25	25	39,5	36
Dosage (g/kg)	3	4	4	3	4	3	4	4	3	4	3	4
Total dose (ml)	2639	1794	3224	2385	1564	1518	759	342	456	540	740	1000
Extract dosed	B ₂	B ₂	D ₁	D ₁	B ₂	B ₂	B ₂	C	C	E	G ₁	G ₁
Immobilising agents (mg):												
M99 ¹ (etorphine hydrochloride)	4	2	—	—	2	2	1,5	1,5	1,5	—	2	2
Azaperone ²	—	—	—	—	—	—	—	—	—	—	100	100
Acetylpromazine ³	10	5	—	—	10	10	—	—	—	—	—	—
Hyoscine hydrobromide ⁴	—	—	—	—	10	10	—	—	—	—	—	—
Sernylan ⁵ (phen-cyclidine hydrochloride)	60	30	—	—	—	—	—	50	50	—	—	—
Serenace ⁶ (haloperidol)	—	—	—	—	—	—	—	2,5 ⁺	2,5 ⁺	—	—	—
Rompum ⁷ (xylazine)	—	—	—	—	—	—	15	—	—	—	—	—
Immobilisation time (min./s.)	7/5	13/40	—	—	5/36	4/22	5/40	10/47	2/45	—	ND	ND
*Interval (min.)	—	—	—	—	15	—	—	—	—	—	—	—
Antidote: M285 (cyprenorphine) (mg)	12	6	—	—	6	6	5	5	5	—	4	4
Recovery time (min./s.)	0/58	2/0	—	—	1/25	1/40	4/45	0/45	2/45	—	ND	ND
Starvation (h)	40	40	24	24	41	41	—	—	—	24	24	24
Water withheld (h)	12	12	24	24	13	13	—	—	—	24	24	24

*Interval = time between dosing and administration of antidote
 + = drug given following morning
 ND = not determined

¹Reckitt
²Janssen Pharm.
³Boots Pure Drug Co.
⁴Burroughs & Wellcome Co.
⁵Parke-Davis
⁶Searle
⁷Bayer

3.5 Gemsbok

These animals were captured at the same time and in the same area as the kudu. They were fed Rider cubes and veld hay treated with Morea 30 (30% urea in an ethyl alcohol molasses complex) and Moranol (van Dorsen) which is an alcohol-molasses compound. Nine litres of the former and 18 litres of the latter, diluted in 180 litres of water were sprayed over the hay.

3.5.1 Dosing

Two gemsbok, one male (OG 2) and one female (OG 1), were immobilised with a combination of etorphine, acetylpromazine (Boots), and hyoscine (Burroughs Wellcome) (Table 8), drenched with 3 and 4 g/kg respectively of extract B₂ and then revived.

3.5.2 Results

Both animals were inclined to lie down; OG 2 within 4 hours and OG 1 within 5 hours. They were drowsy and OG 1 (dosage 4 g/kg) showed muscular tremors. Both became progressively worse and at 9 hours, OG 1 showed severe ataxia when aroused. It died 15 hours later. At this stage severe imbalance was also evident in OG 2, but at 10 hours it was standing calmly in the pen when it died within minutes after a sudden fatal seizure (Table 6).

3.6 Springbok

3.6.1 Dosing

Three springbok (AM 1, AM 2, and AM 3) were captured by immobilisation. AM 1 which was immobilised with M99 and xylazine (Rompun, Bayer), was drenched with 4 g/kg of extract B₂ within 1 hour after capture. Sixteen hours after capture AM 2 and AM 3 were tranquillised with 2,5 mg haloperidol (Serenace, GD. Searle & Co.) (Hofmeyr, Luchtenstein and Mostert, 1977) and then dosed with 4 and 3 g/kg respectively of extract C (Table 8). In order to combat stress and other untoward effects due to capture, all three springbok were treated prophylactically with a vitamin E-selenium preparation (BO-Se Cyanamid) and a long-acting antibiotic.

Springbok AM 4 which had been in captivity for 10 months was dosed with 4 g/kg of extract E, without being chemically immobilised. Two other springbok (AM 5 and AM 6) which had been captive for three years were immobilised with etorphine and azaperone (Janssen Pharm.) (Table 8) and then dosed with 3 and 4 g/kg of extract G₁, respectively.

3.6.2 Results

AM 1 showed inco-ordination for at least 15 minutes after revival, but this was evidently due to the effect of the xylazine. No further ill effects were noticeable until the animal was found dead after 20 hours. Both AM 2 and AM 3 developed sudden severe intermittent convulsions with bleating 5 hours after dosing. The former died within 15 minutes and the latter within half an hour after the onset of symptoms (Table 6).

AM 4 was found dead 6 hours after dosing. No symptoms were noticed beforehand. The remaining two springbok (AM 5 and AM 6) recovered within a minute or two after the administration of the antidote, but both revealed drowsiness and ataxia until death intervened 17 hours and 27 hours respectively after dosing (Table 6). It was evident that the animals became affected by the gifblaar extract while still under the influence of azaperone. Peak effects of azaperone are known to last for 2 hours with a rapid decline between the second and eighth hours (Marsboom, 1969). However, the azaperone dose recommended by Pienaar (1973) for the immobilisation of springbok is considered excessively high, hence the prolonged drowsiness and ataxia which set in.

3.7 Pathology

Most of the cases were acute, some sub-acute and only one goat died 33 days after dosing.

3.7.1 Antelope

No macroscopic observations could be made in the one kudu and one eland that had died. Specimens were, however, collected by a stock inspector. All the other animals were autopsied.

Cyanosis and congestion were always present. The *livers* were usually congested, sometimes with petechiae or haemorrhages beneath Glisson's capsule (OG 2). Degeneration was suspected. The *lungs* were mildly congested with a few petechiae. Pulmonary oedema was present in springboks AM 4, AM 5 and AM 6. The *heart* was usually dilated. Myocardial degeneration, mainly in the papillary muscles, was seen in gemsbok OG 1, and springboks AM 2, AM 3 and AM 4. Mild subepi- and subendocardial petechiae were usually noticeable but were never very prominent, except in AM 6. Congestion of the *small intestines*, occasionally with mild enterorrhagia, was fairly common, but absent in OG 2 and AM 4. The *kidneys* were also congested and mild nephrosis was suspected in the two gemsbok and AM 4. Some animals (AM 2 and AM 3) revealed a few petechiae in the *urinary bladder* and congestion and/or haemorrhages of the *adrenals* (OG 2 and AM 2). Mild *cerebral* oedema was suspected in AM 3. AM 2, AM 3, OG 1 and OG 2 revealed oedema of the *gall bladder*

with some haemorrhages whilst AM 3 had a considerable amount of blood in the bile contents. *Abomasal* oedema was seen in both gemsbok and springbok AM 3 and also oedema of the *pulmonary valves, pancreas* and *periportal* area in gemsbok OG 2.

3.7.2 Goats

As some specimens were collected by stock inspectors, the incidence of the lesions in the various animals cannot be given accurately. However, most of the lesions were similar to those found in the antelope. Congestion, degeneration and enlargement of the *liver*, with congestion and suspected degeneration of the *kidneys* as well as *pulmonary* and *intestinal* congestion were most frequently encountered. The *myocardium*, either entirely or zonally (such as the endocardial region and papillary muscles), often had a parboiled appearance. Mild sub-endocardial and subepicardial haemorrhages were seen occasionally. Larger haemorrhages were noticeable in the wall of the right auricle of a few animals. Approximately 50% of the goats revealed variable degrees of congestion, oedema and even mild haemorrhages of the *gall bladder*. Lesions that were less frequently encountered included *pulmonary* oedema, enterorrhagia, *abomasal* oedema, very mild haemorrhages and suspected oedema of the *urinary bladder*.

The spleen was usually either small or of normal size but sometimes mildly enlarged.

3.8 Histopathology

Two gemsbok, one eland, one kudu, six springbok and 23 goats were examined. The most important lesions, although not constant, were present in both the antelope and goats. For this reason the histopathological findings of both groups are dealt with simultaneously. Furthermore, as there appeared to be no correlation between the dosage rate, or survival time and severity of the lesions, except in the myocardium, the animals are not dealt with individually.

In the *heart* the papillary muscles and the subendocardial zone of the left ventricle were most consistently affected. Other areas including the inner and outer walls of both ventricles and the apex were also frequently involved. Although not always very obvious, degeneration of the myocardium was a constant feature. The most common findings were swelling of the fibres, hydropic rarefaction and mild fatty changes, frequently accompanied by foci or patches of more advanced lesions, especially in the papillary muscles. The latter changes were characterised by zonal rarefaction and irregular narrow intermittent eosinophilic bands. The most advanced lesions were overt Zenker's necrosis. Mineralisation was present in two animals that died at 21–24 hours. In those animals that survived for approximately two days or more, macrophage mobilisation, mild in-

filtrative and proliferative (fibroblastic) changes were noticeable in the necrotic foci. Congestion, some haemorrhages, especially in the auricles, and occasional oedema were sometimes present. In the *skeletal muscles*, Zenker's changes were seen in scattered individual fibres of a few animals.

The *livers* were usually moderately to severely congested and oedematous with varying degrees of fatty degeneration. Whenever abundant proteinaceous material or even globules were present in dilated spaces of Disse, this was regarded as oedema. The distribution of the fatty changes varied, being either zonally (peripherally or centrilobularly) or diffusely spread in the lobuli. Centrilobular necrosis accompanied by neutrophil infiltration and peripheral fatty changes occurred in a few animals. A restricted zone of necrosis and haemorrhage was occasionally seen around the gall bladder. The latter was frequently congested and oedematous, sometimes with concomitant haemorrhages and necrotic foci. A small to moderate number of large eosinophilic globules was found in the hepatocytes in a third of the cases.

Congestion of the *kidneys* was present in most of the animals. With the exclusion of a small number of animals, nephrosis was a common feature. It varied from cloudy swelling, vacuolar or fatty degeneration to frank necrosis. Hyalin globules were occasionally found in the cytoplasm of the cortical cells. The capsular spaces of Bowman and many of the cortical tubuli, especially the proximal convoluted tubuli, were frequently dilated and contained much proteinaceous material. Eosinophilic globules were often seen in the capsular spaces. Reflux of tubular epithelium in these spaces was present in one goat. Necrosis was usually seen in the distal convoluted tubuli, ascending loop of Henle, spiral tubuli and outer medullary area; but occasionally other tubuli and the glomeruli were also affected.

The *brains* of some animals were either congested, mildly swollen and/or oedematous. A very mild or mild status spongiosus of the white substance occurred in a few of the goats and antelope. The spongiosity was unrelated to any specific dosage level or any length of survival period. Mild glial swelling was present in a few animals.

The *spleen* was either congested or somewhat contracted and about 30% showed variable degrees of karyorrhaxis of the splenic corpuscles. This was sometimes also noticeable in the *lymph nodes*, which were unfortunately not regularly studied. Oedematous lymph nodes were encountered occasionally.

Apart from occasional *abomasal* oedema, congestion and haemorrhages were more frequent in the *small intestine*. These changes were at times accompanied by necrosis of the tips of the villi.

Some of the *urinary bladders* showed very mild congestion, haemorrhages and occasionally mild oedema.

The *lungs* were congested and about 30% revealed oedema. Pneumonitis and acute thrombosis of some vessels were present in one goat with a survival period of 32 hours (dosage rate 8 g/kg of extract G₂) (Table 5).

The *adrenals* were sometimes congested and a few contained intracytoplasmic eosinophilic globules in various zones such as the *zona fasciculata*, *zona glomerulosa*, *zona reticularis* and in exceptional cases even in the medulla. Specimens of the *pancreas* were collected from a few animals, and in two of these, congestion, haemorrhages and focal necrosis were observed. The latter was macroscopically noticeable as greenish areas. Microscopically they represented necrotic foci around affected pancreatic ducts some of which contained numerous bacilli. The greenish colour is thought to be due to bile seepage or regurgitation of bile.

Springbok AM 4 revealed no significant degenerative lesions in either myocardium or parenchymatous organs such as the liver and kidneys. Similarly, in a few goats degeneration was so mild that it could easily have been overlooked.

TABLE 9: Oral lethal dose of MFA in mg/kg body weight of various animals (Garner, 1957).

Horse	0,50–1,75
Sheep	0,25–0,50
Goat	0,30–0,70
Pig	0,30–0,40
Dog	0,06–0,20
Cat	0,30–0,50
Fowl	10,00–30,00

4 DISCUSSION AND CONCLUSIONS

The trials on voluntary intake have shown that certain antelope will eat gifblaar at least under some adverse circumstances such as starvation but that kudu are apparently much more wary than eland and springbok. It should, however, be taken into consideration that the animals had been in captivity for a prolonged period and that the regular feeding by man could have played a role in their acceptance of the plant.

MFA assays confirmed that the most toxic stage of gifblaar is during spring (September) and another high peak is reached during late summer – early autumn (March) (Steyn, 1928; Vickery and Vickery, 1973; Tanock, 1975).

The assays also indicate that the toxicity may vary from year to year and that a drastic variation may occur even within seven days of collecting the material. The lowest level of 0,0143% MFA compared with the highest level of 0,5% MFA represents a 35-fold increase. Due to this variation in toxicity and the fact that MFA assays were not done regularly, an evaluation and comparison between goats and antelope are only valid within each separate trial and not collectively or between those trials

where assays were made (Table 6). The *ca.* LD₁₀₀ of goats killed gemsbok and springbok but in no instances eland or kudu. During 1976 the lowest dosage rate of 1,01 mg/kg MFA used proved to be an approximate LD₁₀₀ for goats, but a five-fold increase (5,25 mg MFA/kg) failed to kill eland and kudu. This only happened at a dosage level of 6 to 8 mg MFA/kg which constitutes at least a seven-fold increase of the caprine LD₁₀₀. Considering all the evidence, it can be safely stated that eland and kudu need a six- to seven-fold increase of the *ca.* LD₁₀₀ of MFA for goats to produce some mortalities. It is evident therefore that both kudu and eland are much more resistant to gifblaar poisoning than goats. Garner (1957) gives the oral lethal dose of MFA for goats as 0,3–0,7 mg/kg (Table 9). According to Robinson (1970), the LD₅₀ for cattle is 0,247–0,625 mg/kg. These findings and our own investigation show that cattle are equally or even slightly more susceptible to gifblaar poisoning than goats.

Only one of the ten eland and two of the four kudu which were drenched were chemically immobilised. Consequently the advisability of such immobilisation in gemsbok and springbok comes into question. As the combination of immobilising agents varied, an adverse and possible synergistic effect between some of the drug combinations and MFA cannot be ignored entirely. Drugs in question include xylazine used in one springbok, the high dose of azaperone used in two springbok and in particular hyoscine which was used in both gemsbok. Hyoscine is a powerful cholinolytic agent and produces *inter alia* tachycardia which in the gemsbok may well have enhanced the toxic effect of MFA on the myocardium. It would therefore be advisable to repeat dosing trials on gemsbok preferably without using immobilising compounds or at least by omitting hyoscine.

Springbok AM 1 had fully recovered overnight from the effect of xylazine before it died the next afternoon, but the high dose of azaperone used in AM 5 and AM 6 obviously caused the animals to remain drowsy and ataxic after MFA had taken effect. However, the death of AM 4 a few hours after dosing it without the use of chemical immobilisation is substantial evidence that springbok are at least as susceptible as goats and that the immobilising agents played a negligible or no role at all in enhancing death in these antelope.

All the antelope except the first three springbok were given adequate time for adaptation in captivity, before commencement of the trials. The shortest period was five weeks in two kudu and two gemsbok and it exceeded six months in all the other animals. Apart from handling and dosing, any stress which could have precipitated capture myopathy and enhance death during the trials can therefore be regarded as negligible.

Some of the macroscopic lesions observed have not been recorded previously for gifblaar poisoning (Steyn, 1928; and Steyn, 1949). These lesions include oedema and haemorrhages of the gall bladder with occasional blood-stained bile, oedema elsewhere such as the

abomasum, periportal area, pancreas and pulmonary valves, and adrenal haemorrhages and petechiae in the urinary bladder. It is also noteworthy that the intestinal congestion described by Steyn (1928) and Steyn (1949) was frequently present, whereas enterorrhagia in the small intestines was found in a small percentage of animals.

Mild fatty changes proved to be a fairly common lesion in the myocardium and liver. It is sometimes preceded or replaced by hydropic changes or cloudy swelling. More advanced lesions include hyalin droplet degeneration (liver) and necrosis in both liver and myocardium. Replacement fibrosis in the latter is noteworthy in animals which survived for two or more days and is reminiscent of the lesions in Georgina River Poisoning caused by MFA in *Acacia georginae* (Whittem and Murray, 1963). Other changes that need emphasis are kariorrhaxis of the splenic corpuscles and lymph nodes, oedema of the liver and the mild status spongiosus of the brain in some cases.

It is accepted that some of the lesions encountered, notably tubular reflux and accumulation of eosinophilic globules in the renal capsular spaces, could have been caused by shock.

Gifblaar poisoning is of considerable economic importance. Although there are apparently no figures available on the annual losses of domestic stock caused by gifblaar in southern Africa, an estimate for the Grootfontein district alone would be at least 300 cattle a year. It is undoubtedly the most important cause of death amongst livestock in this area. Reports of mortalities in wild antelope are also lacking. However, the relatively high resistance of eland and kudu to gifblaar poisoning which became evident during the present studies, tempts one to emphasise the potential of selective game ranching in gifblaar areas and possibly also other areas where poisonous plants abound. It is also interesting to note that gifblaar areas generally fall within the natural habitat of eland and kudu, whereas the major natural habitat of springbok and gemsbok occurs elsewhere. Although small numbers of gemsbok are occasionally found in the gifblaar areas of South West Africa/Namibia, they are known grazers, and were consequently not exposed to MFA during their evolutionary development.

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