THE PATHOLOGY OF EXPERIMENTAL *LASIOSPERMUM BIPINNATUM* (THUNB.) DRUCE (ASTERACEAE) POISONING IN SHEEP. II. PULMONARY AND MISCELLANEOUS LESIONS

M. C. WILLIAMS, Department of Pathology, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort 0110, Republic of South Africa

ABSTRACT

WILLIAMS, M. C., 1990. The pathology of experimental *Lasiospermum bipinnatum* (Thunb.) Druce (Asteraceae) poisoning in sheep. II. Pulmonary and miscellaneous lesions. *Onderstepoort Journal of Veterinary Research*, 57, 263–275 (1990).

Key words: plant poisoning; pneumotoxicosis; Clara cell

Poisoning with the plant *Lasiospermum bipinnatum* was studied in 9 lambs at various dose levels. Dyspnoea and tachypnoea, which were dose-related, were observed in 4 of the lambs. Gross and microscopic pulmonary lesions were found in all the lambs receiving plant material originating from one source but not in those given plant from another locality. The severity of the lesions appeared to be dose-dependent. Macroscopic lesions included pulmonary and mediastinal emphysema, congestion and oedema. Microscopically Clara cell hypertrophy and hyperplasia, and interstitial pneumonia were the most outstanding findings. It is speculated that the pulmonary lesions were induced by a furanosesquiterpene or tryptophan or a combination of both of these toxins in the dosed plant material. Miscellaneous and inconsistent lesions observed in the experimental animals included widespread haemorrhage (1 lamb), transudations into the body cavities and adrenocortical hyperplasia.

INTRODUCTION

Lasiospermum bipinnatum, commonly known as ganskweek, has been reported as a cause of poisoning in ruminants in the field in South Africa on a number of occasions (Walsh, 1909; Adelaar, Terblanche, Smit, Naudé & Codd, 1964; Fair, Tustin & Adelaar, 1970; Thornton, 1977). Adelaar *et al.* (1964) were the first to show, experimentally, that the plant is poisonous to sheep. The lesions are not reported in any detail, but the most striking changes were found in the liver. Hepatic lesions described by others as well as those produced during the present experiments have been reported in detail elsewhere (Williams, 1990). This paper describes hitherto unrecorded, significant pulmonary lesions with experimental *L. bipinnatum* poisoning, as well as other minor or inconsistent pathological changes.

MATERIALS AND METHODS

These have been documented in a previous publication (Williams, 1990).

RESULTS

Clinical signs

General clinical signs and those related to hepatic dysfunction in the 9 poisoned sheep have been reported (Williams, 1990). Clinical signs relating to pulmonary dysfunction were observed in 4 of the 9 lambs poisoned (C, D, E & F). All these animals were given toxic material collected from near Cradock. The 2 sheep dosed with plant collected from near Graaff-Reinet (B & G), the 3 sheep receiving very high or very low doses of Cradock material (A, H & I), as well as the controls (J & K), did not show abnormalities of respiration.

Lambs given 6 or 8 g/kg/day of the Cradock batch of ganskweek (C, D & E) showed severe dyspnoea on the 5th or 6th day after commencement of dosing. The onset of respiratory distress was fairly sudden, the affected animals exhibiting deep and rapid inspiratory and expiratory movements up until the time they died or were killed. Lamb F, which received 4 g/kg/day of the Cradock material, was moderately depressed and anorexic on Day 6. It was not dosed on Day 7, and dosing was resumed the following day when the animal seemed to be in good health. Mild

depression and a slight increase in the rate and depth of respiration were noted on Day 10, but the lamb appeared to be quite well on the subsequent days until slaughtered on Day 14.

Gross pulmonary and miscellaneous pathology

Gross pulmonary lesions were found in all the sheep that received plant material collected at Cradock (Lambs A, C, D, E, F, H and I). The severity of the changes was variable but was clearly dependent on the dose level of plant material. In general, the lungs did not collapse when the thoracic cavity was opened and they had a more or less doughy or meaty consistency, particularly in Lambs C, D and E.

In the animal given the highest dose of ganskweek (Lamb A), the lungs had a deep purple-red colour, the lobules were more distinct and the lungs felt heavy. Patches of lung tissue, where the alveoli appeared to be overdistended with air, were observed. Copious amounts of bloody fluid exuded from the cut surface of the lung, attesting to the presence of severe congestion and oedema of the parenchyma. The bronchi and bronchioles were filled with turgid foam, and the laryngeal and tracheal mucosae showed severe, multiple, linear, petechial and ecchymotic haemorrhages which extended into the proximal portions of the bronchial tree.

The lungs of Lambs C and E had a similar appearance (Fig. 1). They were enlarged, and the whole surface was broken up into a mosaic pattern of deep purple-red areas comprising a number of atelectatic adjacent lobules interspersed with groups of light pink, raised, emphysematous lobules. The dark red areas of atelectasis were mainly located in the dorsal portions of the lung. The interlobular septa were widened, sometimes markedly so, by the accumulation of minute air bubbles. A large number of gas bubbles up to 2 mm in diameter were noted under the visceral pleura, particularly on the diaphragmatic lung surfaces. In both cases, the lungs were severely congested and moderately oedematous. In Lamb E, emphysema had extended from the lungs to involve the mediastinum and pericardium (Fig. 1). From there it stretched caudally to the diaphragm and cranially, via the thoracic inlet, to halfway up the neck, in a peri-oesophageal location.

In Case D, the lungs were enlarged, pinkish-red and had a fleshy appearance and feel (Fig. 2). They
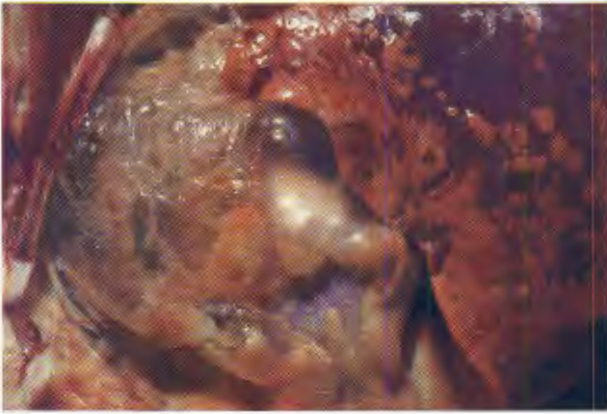
FIG. 1 Lamb E: Thoracic cavity. Severe emphysema of mediastinum and pericardium. Lungs show patchy atelectasis (dark red areas).
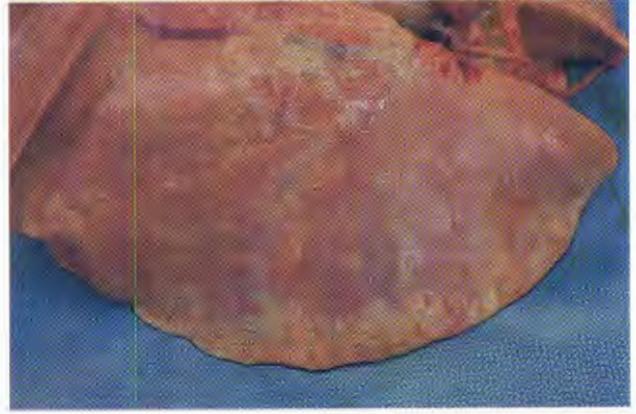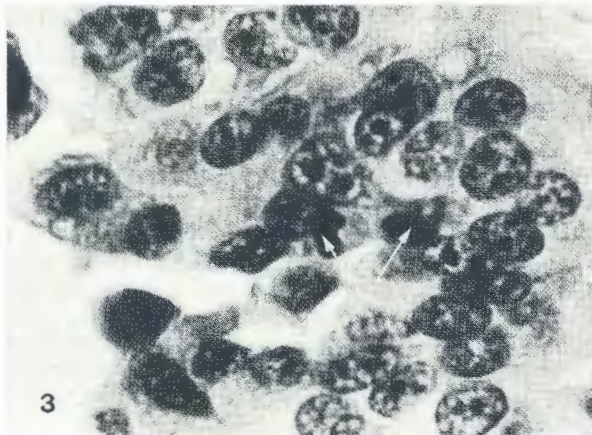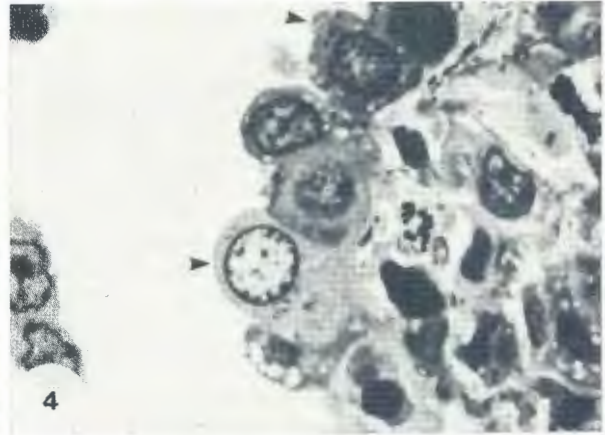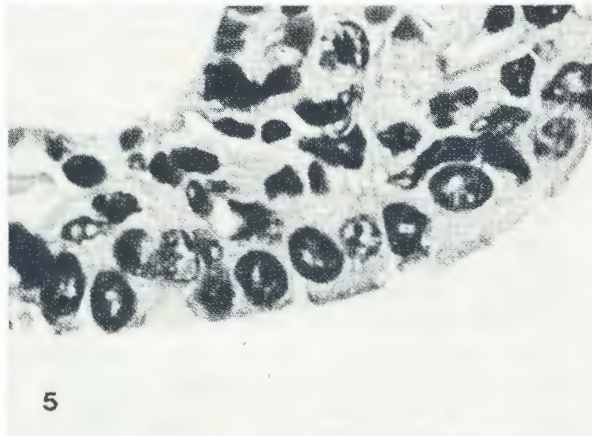

FIG. 2 Lamb D: Lung. Diaphragmatic surface. Solid looking, pinkish-red colour with patchy interstitial emphysema.
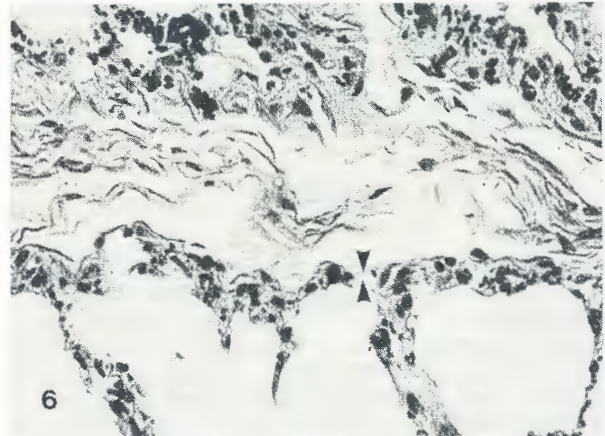

FIG. 3 Lamb D: Lung. Thickened terminal bronchiolar epithelium composed mainly of proliferating Clara cells. A dividing cell in telophase is present (arrows). HE × 1 000.

FIG. 4 Lamb D: Lung. Terminal bronchiole showing hyperactive Clara cells (arrows). Toluidine blue, Epon-embedded. × 1 000.

FIG. 5 Lamb J: Lung. Terminal bronchiole with normal Clara cells in a single layer (control animal). HE × 1 000.

FIG. 6 Lamb D: Lung. Widened, interlobular septa due to emphysema. Note ruptured alveolar wall (arrows). HE × 200.

were not congested or oedematous, but showed clear evidence of interstitial emphysema, characterized by widened interlobular septa and the formation of small bullae, especially on the ventral edges of the caudal lobes. Emphysema extended to the lung hilus and thence to the mediastinum and perioesophageal tissues and, finally, via the thoracic inlet, to the cervical subcutis. Very large bullae were present in the mediastinum and peri-aortic tissues,

and from the latter location air had spread to the perirenal connective tissue.

The lungs of Lambs F, H and I showed mild lesions. They were slightly heavier than normal, moderately congested, and had a more solid appearance on cut surface. A few lobules in the cranioventral portions of the lungs showed red hepatization. The pleural capillaries and lymphatics and the
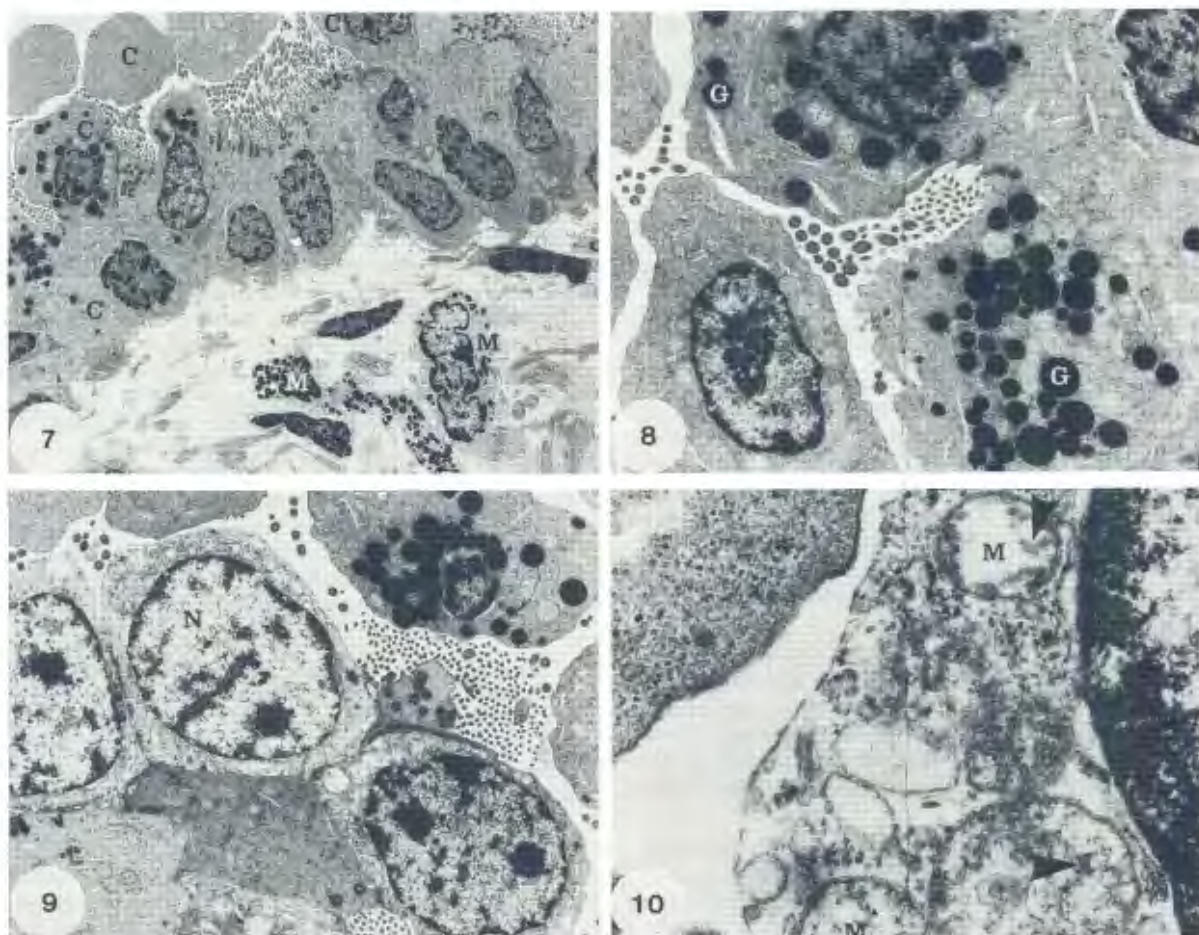
FIG. 7 Lamb E: Lung. Terminal bronchiole showing proliferation and desquamation of Clara cells (C). Mast cells (M) are in the oedematous peribronchiolar connective tissue. × 1 800.

FIG. 8 Lamb E: Lung. Clara cells containing characteristic, highly osmiophilic secretory granules (G). × 5 900.

FIG. 9 Lamb E: Lung. Clara cells with large, vesicular nuclei (N) and scant cytoplasm. × 4 300.

FIG. 10 Lamb E: Lung. Portion of the cytoplasm of a Clara cell shown in Fig. 9. Note numerous, swollen mitochondria (M) and disintegration of cristae (arrows). × 36 000.

interlobular septa were more prominent than normal, the latter resulting in pulmonary lobulation being more distinct. In 2 of the cases (F and I), single, isolated, emphysematous bullae about 15 mm in diameter were present under the visceral pleura.

No lesions were encountered in the respiratory system of the 2 control sheep.

Widespread, severe haemorrhage was a striking finding in the lamb that received the highest dose of ganskweek (Lamb A). In this animal, petechial and ecchymotic haemorrhages were observed in the liver, left dorso-lateral subcutis, mediastinum, thymus, peri-aortic tissue, superficial cervical lymph nodes, pericardium, coronary grooves, diaphragm and laryngeal, tracheal and bronchial mucosae.

Accumulation of small to moderate amounts of transudate, sometimes containing fibrin clots, in the body cavities occurred inconsistently in most of the poisoned animals. Mild to moderate ascites (20–100 mℓ) was observed in Lambs A, B, C, G and H. Mild hydrothorax (10–50 mℓ) was found in Lambs A, C and E, and moderate hydropericardium (10–20 mℓ respectively) in B and C. Excessive transudate was not found in the body cavities of control sheep.

In the 3 lambs (B, C and D), dosed 8 g/kg/day of toxic plant, the cortices of the adrenal glands were regarded as being moderately widened.

Light microscopical pathology

Pulmonary lesions

Microscopic pulmonary lesions were observed in lambs dosed with the Cradock batch of ganskweek but not in those given plant material originating from Graaff-Reinet or in control animals. The most striking finding, present in most of the animals, was proliferation of non-ciliated epithelial cells lining the lumen of terminal portions of the bronchioles (Fig. 3 & 4). These cells, collectively known as Clara cells, also showed degenerative changes and in some cases there appeared to be an increase in the number of desquamating cells.

Excepting Lamb A, in which Clara cell hyperplasia was not present, proliferation of these cells was most marked in the lungs of sheep receiving 8 g/kg/day and became progressively less obvious with decreasing dose levels. In control lambs, the Clara cells were seen to be cuboidal and formed a single, more or less continuous layer at the junction of the bronchioles and respiratory atria (Fig. 5). In the most severely affected animals (C and D), they had formed a layer 2 to 3 cells thick at this location (Fig. 3). The proliferating cells also showed cytological changes, which seemed to be progressive with the increasing dose rate. Normal Clara cells (control sheep) had oval, moderately vesicular nuclei, containing 1 or 2 small nucleoli and pale eosinophilic

cytoplasms. In proliferated cells, the nucleus was moderately enlarged and hyperchromic, and contained 1 very large nucleolus or 2 fairly large ones. The cytoplasm was variably more basophilic. Mitotic figures and conjoined daughter cells were frequently observed, especially in Lamb D.

Degenerative lesions in the Clara cells, as interpreted from mildly increased cytoplasmic vacuolation, seemed to occur only in the high-dose group. Increased desquamation of apical blebs of cytoplasm or of whole epithelial cells, on the other hand, appeared not to be related to dose levels and was clearest in Lambs A, F and I.

Circulatory disturbances of the pulmonary vasculature were manifested as congestion and oedema. Congestion of the lung parenchyma was apparent in all the lambs, including the controls. It was very severe in Lamb A, marked in C and E, moderate in B and D and mild in the remaining animals. Fibrin-rich fluid, indicative of oedema, was seen only in the alveoli of the 3 sheep that had died naturally (A, C and E). It was most marked in Lamb A and least noticeable in E.

Neutrophils were observed in the alveolar capillaries in all the sheep and in the interstitium and alveolar lumens of some. Moderate to marked increases, with cells being present in the alveolar spaces, were noted in Lambs A, B, C and E. Mild neutrophilic leukostasis was seen in the capillaries in the other lambs.

Small, scattered aggregations of eosinophils, neutrophils and mast cells were found in the lungs of Lambs A, C, D and E. Sections cut from different blocks of lung tissue from the same animal did not invariably show these aggregations. This was true even for Lamb C, which had the most marked infiltration of these cells.

The interstitial and alveolar emphysema noted under the description of gross pathology in Lambs A, C, D and E was confirmed histologically (Fig. 6).

Other organs

The majority of cells of all 3 zones of the adrenal cortex of Lambs B, C, D, E and F were swollen, with excessively vacuolated cytoplasms and vesicular nuclei. No changes were observed in the adrenal cortices of Lambs H, I, J and K. The adrenal gland of Lamb A exhibited post-mortem autolysis.

Significant histological lesions were not found in sections of kidney, myocardium, skeletal muscle, lymph node, spleen, brain, abomasum, small and large intestine and pancreas.

Electron microscopical pathology

Sections of the lungs of Lamb E showed the most severe lesions of those examined with the EM (Lambs E, F, G, H and I). Hyperplasia of the non-ciliated bronchiolar epithelial cells (Clara cells) was the most outstanding change in the lungs and occurred in the distal portions of the bronchioles (Fig. 7). In Lamb E, there appeared to be a marked increase in desquamating Clara cells, many of which contained characteristic, highly osmiophilic, secretory granules (Fig. 8). Some of the Clara cells had very large vesicular nuclei and prominent nucleoli, with a narrow rim of lucent cytoplasm (Fig. 9). The cytoplasm of these cells contained numerous, invariably more electron-lucent, swollen mitochondria (Fig. 10). Disintegrating, fuzzy cristae were observed in some mitochondria. These highly active cells probably represented newly formed Clara cells

and only occasional, small secretory granules were found in them. Numerous mast cells were also observed in the oedematous peribronchiolar stroma (Fig. 7).

DISCUSSION

Significant pulmonary lesions or the clinical sign of dyspnoea related to lung pathology have not been reported in either natural or experimental gans-kweek poisoning in ruminants. Varying degrees of pulmonary congestion and oedema (Adelaar *et al.,* 1964; Kellerman, Basson, Naudé, Van Rensburg & Welman, 1973) and, in some cases, ecchymotic haemorrhages (Adelaar *et al.,* 1964; Fair *et al.,* 1970) are the only changes recorded. In the present study, variable degrees of congestion were found in the lungs of all the lambs, including those of the control animals, but pulmonary oedema was observed only in sheep which had died naturally (Lambs A, C and E). A degree of lung congestion was probably induced by barbiturate administration at the time of euthenasia, while pulmonary oedema most likely developed agonally in the 3 sheep that died naturally.

Specific microscopic and macroscopic, dose-dependent pulmonary lesions were found in all the lambs that had received plant material originating from Cradock (A, C, D, E, F, H and I), but were absent from the lungs of lambs given the Graaff-Reinet material (B and G). The primary injury appeared to be to the non-ciliated epithelial cells (= Clara cells) (Smith, Greenburg & Spjut, 1979; Widdicombe & Pack, 1982) occurring in the distal portions of the bronchioles. Under the light microscope, these cells appeared to be hypertrophied and hyperplastic in some lambs. In other animals, the Clara cells showed degenerative changes and, in a few cases, there seemed to be an increased rate of desquamation of whole cells and/or their cytoplasmic apices. The light microscopic observations were confirmed by electron microscopic examination of terminal bronchioles in Lamb E (Fig. 7–10).

Knowledge about the Clara cell has recently been reviewed (Greenburg, Lewis & Levy, 1975; Widdicombe & Pack, 1982) and a number of reports dealing with comparative ultrastructural features of these cells in mammals have been published (Plopper, Mariassy & Hill, 1980 a, b & c; Smith *et al.,* 1979). Clara cells in different species have no common definitive ultrastructural features, and within the same section in the same animal the cells may show very different morphologies (Widdicombe & Pack, 1982). Despite this variability, it is possible to state that the Clara cell of sheep is characterized by abundant ovoid secretory granules and SER (Plopper *et al.,* 1980b). Aside from its presumed role as a secretory cell in the terminal airways, it has recently been shown that the Clara cell is the chief site in the lung for cytochrome P-450-dependent, mixed function oxidase (MFO) activity (Boyd, 1977). The oxidases are bound to microsomes on the SER, where they serve to polarize lipophilic molecules, including drugs and toxins, in order to facilitate their excretion (Bradley & Carlson, 1980; Widdicombe & Pack, 1982).

Toxins, such as 4-ipomeanol (Boyd, 1977; Durham, Boyd & Castleman, 1985), carbon tetrachloride (Boyd, Statham & Pongo, 1980), 3-methyl-furan (Boyd, Statham, Franklin & Mitchell, 1978) and 3-methylindole (Bradley & Carlson, 1980; Bray & Carlson, 1979; Huang, Carlson, Bray & Bradley, 1977) have a selectively injurious effect on Clara

cells. Since they are all highly lipophilic compounds, they readily cross cell membranes and enter lung cells, including non-ciliated bronchiolar cells. Because of their lipid nature, they are preferentially metabolized by the MFO system in the Clara cell. The resulting reactive metabolite(s) are cytotoxic and cause injury to the cell. It must be noted, however, that other pulmonary cells may also be damaged by these compounds. For example, Type I and Type II pneumocytes are affected by 3-methylindole (Bradley & Carlson, 1980; Breeze, Pirie, Selman & Wiseman, 1975; Pirie, Breeze, Selman & Wiseman, 1976) and endothelial cells of pulmonary capillaries by 4-Ipomeanol (Bray & Carlson, 1979).

A furanosesquiterpene has recently been isolated from *L. bipinnatum* (L. A. P. Anderson, personal communication, 1989). These are lipophilic compounds which are known to be metabolized by the MFO enzyme system (Jubb, Kennedy & Palmer, 1985) and may have been responsible for the pulmonary lesions. This leaves unanswered the question as to why a specific batch of ganskweek appears to cause only hepatic lesions at 2,5 and 8 g/kg/day (Graaff-Reinet plant), while another induces lesions in both the liver and lung at 1, 2, 4, 8 and 12 g/kg/day (Cradock batch).

Prozesky, Kellerman, Jordaan, Welman & Joubert (1985) produced pulmonary lesions with *Hertia pallens* (Asteraceae), which included oedema, mild to moderate diffuse interstitial pneumonia and epithelial hyperplasia of especially the smaller bronchi and bronchioli. These lesions are similar to those described above for *L. bipinnatum*.

The toxin 3-methylindole is a bacterial metabolite of tryptophan produced naturally in the rumen. High levels of tryptophan occurring in lush regrowth (foggit) pasture can induce a pulmonary syndrome in cattle known *inter alia* as "fog fever" (Pirie, Breeze, Selman & Wiseman, 1974). The dark green, lush foliage of the ganskweek plants collected at Cradock contrasted sharply with the pale green, sparse leaves of the decumbent plants collected near Graaff-Reinet. Perhaps the Cradock ganskweek had high levels of tryptophan. This could account for the pulmonary lesions found in the current experiments.

Moderate to marked infiltration of certain cells into the interstitium and alveolar spaces of the lungs was noted only in the lambs given 6 g/kg/day, or more, of ganskweek (A, B, C, D and E). In those animals receiving Cradock plant material, scattered aggregates of mast cells, eosinophils and neutrophils, involving several contiguous alveoli, were seen. In Lamb B (Graaff-Reinet material) only diffuse neutrophil infiltration was observed. The significance, if any, of the cell infiltrations in the pathogenesis of pulmonary emphysema in ganskweek poisoning is not clear but resembles that seen in fog fever (Pirie *et al.*, 1974).

The outstanding macroscopic features of the lungs of the sheep dosed with the Cradock batch of ganskweek were failure to collapse when the thoracic cavity was opened, a peculiar doughy or meaty feel, heaviness in the absence of oedema and the development of interstitial emphysema, which, in some cases, extended to the mediastinum and elsewhere. It is contended that these findings are directly related to the degree of changes occurring in the Clara cells. The severity was dose-dependent as they were most obvious in sheep given the highest doses and almost undetectable in the lamb that received the lowest dose rate.

As regards the dyspnoea which developed in the lambs showing the most severe Clara cell hyperplasia (C, D and E), of considerable interest is a letter, written by the late T. F. Adelaar in 1971, in which the results of a toxicity test conducted at the Veterinary Research Institute, Onderstepoort, with ganskweek collected in the Adelaide district, Cape Province are given. Terminal polypnoea was noted in the single sheep that was dosed, and at necropsy this animal showed "general cyanosis" and "mediastinal emphysema", in addition to typical hepatic lesions. This corresponds to the dyspnoea, cyanosis and emphysema observed in Lambs C, D and E in the present trials and serves to indicate that pulmonary lesions may be more common in ganskweek poisoning than is generally realized.

It is considered that the pathogenesis of these significant lung lesions, and thereby the clinical sign of dyspnoea, is as follows: the basic lesion in the lung in ganskweek poisoning occurs in terminal bronchioles, and this may be important in the genesis of emphysema and dyspnoea. Complete obstruction or total collapse of the distal portions of the bronchioles, particularly as a physiological phenomenon during expiration due to lack of Clara cell secretions (Widdicombe & Pack, 1982), would probably result in atelectasis of the parenchyma served by the airway concerned. Numerous atelectatic areas were, in fact, found in Lambs C and E, the lungs having a mosaic appearance because of interspersed atelectatic and emphysematous lobules (Fig. 1). On the other hand, if terminal bronchioles should partially collapse or become partially obstructed by Clara cell hypertrophy, hyperplasia, degeneration or desquamation or a combination of these, inspired air may be able to pass through the narrowed bronchiolar lumen into the alveoli more readily than in the reverse direction. If this should happen, the carbon dioxide levels in the air accumulating in the alveoli would tend to rise and thereby aggravate hypoxia and promoting dyspnoea. Evidence to support this hypothesis was found by light microscopy. Several alveoli were seen to have ruptured into the interlobular septal connective tissue (Fig. 6). That no severe primary lesion of the alveolar wall could be found suggests that rupture had occurred as a result of increased intra-alveolar pressure rather than of a weakening of the alveolar wall. Possibly partial obstruction and partial collapse of teminal bronchioles both contributed to the development of pulmonary emphysema.

Transudate accumulation in the body cavities of the lambs in the current experiments most commonly occurred in the abdominal cavity, but hydropericardium and hydrothorax were also seen in some animals. The quantities of fluid were small but the occasional presence of fibrin clots in the transudate may be indicative of underlying endothelial damage. This however, was not demonstrated. Transudation into the body cavities in ganskweek poisoning has also been found in experimentally-induced disease in sheep by other workers (Adelaar *et al.*, 1964; Kellerman *et al.*, 1973). In addition, oedema of the abomasal mucosa (Adelaar *et al.*, 1964; Fair *et al.*, 1970), intestinal mucosa (Adelaar *et al.*, 1964) and the mediastinum, perirenal tissue and subcutis (Kellerman *et al.*, 1973) has been seen in odd animals.

Although an unequivocal haemorrhagic diathesis does not appear to be characteristic of ganskweek poisoning, fairly widespread petechiae and ecchymoses are a feature of poisoning in cattle (Fair *et al.*, 1970; Thornton, 1977). Adelaar *et al.* (1964) record mild localized haemorrhages in their experimental

cases. In the present study, severe, almost generalized haemorrhage was observed only in the lamb given the highest dose rate (Lamb A). This may be indicative of endothelial damage in various tissues, a contention which is supported by the EM finding of severe endothelial cell degeneration in the liver of Lamb B (Williams, 1990).

Lesions in ganskweek poisoning reported by others but not observed in the current study include nephrosis (Adelaar *et al.*, 1964; Fair *et al.*, 1970; Kellerman *et al.*, 1973), fatty change and necrosis in the myocardium (Kellerman *et al.*, 1973) and reactive hyperplasia of lymphoid tissue (Adelaar *et al.*, 1964).

REFERENCES

ADELAAR, T. F., TERBLANCHE, M., SMIT, J. D., NAUDÉ, T. W. & CODD, L. E., 1964. A hitherto unknown poisonous plant: *Lasiospermum bipinnatum* (Thunb.) Druce. Preliminary communication. *Journal of the South African Veterinary Medical Association*, 35, 11–16.

BOYD, M. R., 1977. Evidence for the Clara cell as a site of cytochrome P450-dependent mixed-function oxidase activity in lung. *Nature*, 269, 713–715.

BOYD, M. R. STATHAM, C. N., FRANKLIN, R. B. & MITCHELL, J. R., 1978. Pulmonary bronchiolar alkylation and necrosis by 3-methylfuran, a naturally occurring potential atmospheric contaminant. *Nature*, 272, 270–271.

BOYD, M. R., STATHAM, C. N. & PONGO, N. S., 1980. The pulmonary Clara cell as a target for toxic chemicals requiring metabolic activation; studies with carbon tetrachloride. *Journal of Pharmacology and Experimental Therapeutics*, 212, 109–114.

BRADLEY, B. J. & CARLSON, J. R., 1980. Ultrastructural pulmonary changes induced by intravenously administered 3-methylindole in goats. *American Journal of Pathology*, 99, 551–560.

BRAY, T. M. & CARLSON, J. R., 1979. The role of mixed-function oxidase in 3-methylindole-induced acute pulmonary

in goats. *American Journal of Veterinary Research*, 40, 1 268–1 272.

BREEZE, R. G., PIRIE, H. M., SELMAN, I. E. & WISEMAN, A., 1975. Fog fever in cattle: Cytology of the hyperplastic alveolar epithelium. *Journal of Comparative Pathology*, 85, 147–156.

DURHAM, S. K., BOYD, M. R. & CASTLEMAN, W. L., 1985. Pulmonary endothelial and bronchiolar epithelial lesions induced by 4-ipomeanol in mice. *American Journal of Pathology*, 118, 66–75.

FAIR, A. E., TUSTIN, R. C. & ADELAAR, T. F., 1970. Case report. Poisoning of cattle by ganskweek [*Lasiospermum bipinnatum* (Thunb.) Druce]. *Journal of the South African Veterinary Medical Association*, 41, 231–232.

GREENBURG, S. D., LEWIS, C. W. & LEVY, M. L., 1975. The Clara cell: a brief review with illustrative ultrastructure. *Patologia*, 13, 463–469.

HUANG, T. W., CARLSON, J. R., BRAY, T. M. & BADLEY, B. J., 1977. 3-methylindole-induced pulmonary injury in goats. *American Journal of Pathology*, 87, 647–666.

JUBB, K. V. F., KENNEDY, P. C. & PALMER, N., 1985. Pathology of domestic animals, 3rd edn., Vol. 2. New York: Academic Press.

KELLERMAN, T. S., BASSON, P. A., NAUDÉ, T. W., VAN RENSBURG, I. B. J. & WELMAN, WILHELMINA G., 1973. Photosensitivity in South Africa. I. A comparative study of *Asaemia axillaris* (Thunb.) Harv. ex Jackson and *Lasiospermum bipinnatum* (Thunb.) Druce poisoning in sheep. *Onderstepoort Journal of Veterinary Research*, 40, 115–126.

PIRIE, H. M., BREEZE, R. G., SELMAN, I. E. & WISEMAN, A. 1974. Fog fever in cattle: Pathology. *Veterinary Record*, 95, 479–483.

PIRIE, H. M., BREEZE, R. G., SELMAN, I. E. & WISEMAN, A., 1976. Indole-acetic acid, 3-methylindole and type 2 pneumocyte hyperplasia in a proliferative alveolitis of cattle. *Veterinary Record*, 98, 259–260.

PLOPPER, C. G., MARIASSY, A. T. & HILL, L. H., 1980a. Ultrastructure of the non-ciliated bronchiolar epithelial (Clara) cell of mammalian lung. I. A comparison of rabbit, guinea-pig, rat, hamster and mouse. *Experimental Lung Research*, 1, 139–154.

PLOPPER, C. G., MARIASSY, A. T. & HILL, L. H., 1980b. Ultrastructure of the non-ciliated bronchiolar epithelial (Clara) cell of mammalian lung. II. A comparison of horse, sheep, dog and cat. *Experimental Lung Research*, 1, 155–169.

PLOPPER, C. G., MARIASSY, A. T. & HILL, L. H., 1980c. Ultrastructure of the non-ciliated bronchiolar epithelial (Clara) cell of mammalian lung. III. A study of man with comparison with 15 mammalian species. *Experimental Lung Research*, 1, 171–180.

PROZESKY, L., KELLERMAN, T. S., JORDAAN, P., WELMAN, WILHELMINA G. & JOUBERT, J. P. J., 1985. An ovine hepatotoxicosis caused by the plant *Hertia pallens* (DC.) Kuntze (Asteraceae). *Onderstepoort Journal of Veterinary Research*, 52, 233–238.

SMITH, M. N., GREENBURG, S. D. & SPJUT, H. J., 1979. The Clara cell: A comparative ultrastructural study in mammals. *American Journal of Anatomy*, 155, 15–30.

THORNTON, D. J., 1977. Ganskweek (*Lasiospermum bipinnatum*) poisoning in cattle. *Journal of the South African Veterinary Association*, 48, 210–211.

WALSH, L. H., 1909. South African poisonous plants. Cape Town: Maskew Miller.

WIDDICOMBE, J. G. & PACK, R. J., 1982. The Clara Cell. *European Journal of Respiratory Diseases*, 63, 202–220.

WILLIAMS, M. C., 1990. The pathology of experimental *Lasiospermum bipinnatum* (Thunb.) Druce (Asteraceae) poisoning in sheep. I. Hepatic lesions. *Onderstepoort Journal of Veterinary Research*, 57, 249–261.