

CLINICAL REVIEW AND CASE REPORT

Sulfur-associated polioencephalomalacia in cattle grazing plants in the Family Brassicaceae

RA McKenzie,^{a*} AM Carmichael,^b ML Schibrowski,^b SA Duigan,^b JA Gibson^c and JD Taylor^c

Polioencephalomalacia was diagnosed histologically in cattle from two herds on the Darling Downs, Queensland, during July–August 2007. In the first incident, 8 of 20 18-month-old Aberdeen Angus steers died while grazing pastures comprising 60% *Sisymbrium irio* (London rocket) and 40% *Capsella bursapastoris* (shepherd's purse). In the second incident, 2 of 150 mixed-breed adult cattle died, and another was successfully treated with thiamine, while grazing a pasture comprising almost 100% *Raphanus raphanistrum* (wild radish). Affected cattle were either found dead or comatose or were seen apparently blind and head-pressing in some cases. For both incidents, plant and water assays were used to calculate the total dietary sulfur content in dry matter as 0.62% and 1.01% respectively, both exceeding the recommended 0.5% for cattle eating more than 40% forage. Blood and tissue assays for lead were negative in both cases. No access to thiaminase, concentrated sodium ion or extrinsic hydrogen sulfide sources were identified in either incident. Below-median late summer and autumn rainfall followed by above-median unseasonal winter rainfall promoted weed growth at the expense of wholesome pasture species before these incidents.

Keywords Brassicaceae, cattle, pasture weeds, polioencephalomalacia, clinical review, sulfur

Abbreviations AST, aspartate aminotransferase; CK, creatine phosphokinase; DM, dry matter; PEM, polioencephalomalacia

Aust Vet J 2009; 87:27–32

doi: 10.1111/j.1751-0813.2008.00387.x

Polioencephalomalacia (cerebrocortical necrosis, PEM) is an uncommon outcome when ruminants graze crops such as rape or canola (*Brassica napus*), kale (*Brassica oleracea*) or weeds also belonging to the Family Brassicaceae.^{1–3} One Australian study of cattle grazing Brassica crops in Victoria and Tasmania identified 66 disease incidents in 5376 herds during summer and autumn 1995, none of which were identified as PEM,⁴ but some of the four cases of sudden death recorded may have involved this disease. PEM is much more common in grain-fed cattle,⁵ in which high-sulfur diets are currently thought to be a major cause.^{6,7}

^aDepartment of Primary Industries & Fisheries' Biosecurity Sciences Laboratory, Animal Research Institute, Locked Mail Bag No.4, Moorooka, Queensland 4105, Australia; ross.mckenzie@dpi.qld.gov.au

^bBell Veterinary Services, Bell, QLD, Australia

^cDepartment of Primary Industries & Fisheries' Animal Disease Surveillance Laboratory, Toowoomba, QLD, Australia

*Correspondence author.

Thiaminase-producing ruminal bacteria have been suggested as the cause in feedlot PEM incidents,^{8,9} but this is now doubted.¹⁰

Reports that link the sulfur content of green plants in pasture or fodder crops to PEM in the cattle grazing them are rare. In New Zealand, 26 of 99 heifers were affected when grazing *Brassica oleracea* var. *acephala* (chou moellier) that contained 8500 mg sulfur per/kg dry matter (0.85% sulfur in DM).¹¹ In North America, approximately 35 of 3500 cattle died of PEM after 7 to 10 days grazing grass (species not identified), turnips (*Brassica rapa*) and Essex rape (*Brassica napus*) with sulfur contents in DM of 0.19%, 0.63% and 0.91%, respectively, and drinking water containing 57 mg sulfate/L.⁶ In Turkey, 256 of 5050 cattle in two herds died of PEM after being fed a diet containing 0.45% sulfur based on barley sprouts (*Hordeum vulgare*).¹² In New South Wales, Australia, 12 of 200 steers became blind when grazing a failed *Brassica napus* (canola) oilseed crop that had 5-cm regrowth after two rainfall events, 2 and 1 week previously, and that contained 0.67% sulfur in DM (JH McNally, personal communication 31 July 2006; RA McKenzie, unpublished data 2006), but this case may have been so-called 'rape blindness', thought to be an entity separate from PEM.

We report here sulfur-associated PEM in cattle that grazed either large amounts of *Sisymbrium irio* (London rocket) combined with *Capsella bursapastoris* (shepherd's purse), or large amounts of *Raphanus raphanistrum* (wild radish), all in the Brassicaceae (synonym Cruciferae), at Bell and Jandowae, respectively, on the Darling Downs in Queensland during the late winter of 2007. These plant species have not been linked with this syndrome previously, nor have they been investigated for their sulfur content.

Laboratory Methods

Tissues, aqueous humour and blood were submitted to the Animal Disease Surveillance Laboratory, Toowoomba for histopathological examination and toxicological and clinical pathology tests. Formalin-fixed tissues were processed to 5- μ m haematoxylin–eosin-stained sections using standard methods and examined by light microscopy. Haematological profiles were obtained from EDTA blood samples by microhaematocrit and automated electronic examinations for haemoglobin, packed cell volume, erythrocyte and leucocyte counts, and erythrocyte indices. Plasma fibrinogen concentration was measured in EDTA blood samples centrifuged in capillary haematocrit tubes using a refractometer to determine the difference in plasma protein concentrations between one unheated tube and another after heating in a water bath for 3 min at 58 to 60°C to precipitate fibrinogen. Clinical chemistry serum profiles were obtained using an automated analytical system (Olympus AU400[®]). Calcium, magnesium, total protein, total bilirubin, creatinine, urea, γ -glutamyl transferase, CK and AST were assayed using commercial kits (ThermoTrace, South

Oakleigh, Vic, Australia) and albumin and glutamate dehydrogenase with other kits (Randox Laboratories Ltd, Antrim, UK). Reference ranges used to interpret haematological and clinical chemistry results were those established at the DPI&F laboratories. Kidney and EDTA blood samples were assayed for lead by colorimetric determination after triple-acid digestion.¹³

Plant investigations

Collected fresh plants were divided into three parts (flowering and fruiting tops, leaves and stems, and roots), oven dried and ground before assay. The DM content of plants as received at the Biosecurity Sciences Laboratory was determined for each plant part. Sulfur, molybdenum and copper concentrations in the plant samples were assayed by inductively-coupled plasma atomic emission spectrometry after digestion with nitric acid/perchloric acid (R Lascelles, personal communication 2007).¹⁴ Plant and aqueous humour nitrate contents were assayed colorimetrically using nitrate and nitrite test strips (Merckoquant®; Merck, Darmstadt, Germany) as previously described.¹⁵ Fertile pressed and dried botanical specimens were made from each batch of plant species collected and submitted to Queensland Herbarium as vouchers for the chemical assays.¹⁶

Total dietary percentage of sulfur in DM was calculated by the method of Gould.⁶ The feed sulfur contribution was calculated by multiplying the percentage of sulfur in DM for each feed component by the proportion of the diet it represented, then adding these data for all components. The water sulfur contribution was calculated by first calculating the daily sulfur intake in grams from the water sulfur content multiplied by the daily water intake derived from body weight, lactation status and ambient temperature data, then by dividing this result by one tenth of the daily dry matter intake, assumed to be 2% of body weight. Daily water intake is 8%, 10% and 18% of body weight (kg) for beef cattle (10%, 15% and 15% when lactating) for ambient temperatures of 5°C, 21°C and 32°C respectively.⁶ Then the total sulfur content in DM is the sum of the feed and water contributions.

Case reports

Case 1 (Bell)

A herd of 20 18-month-old Aberdeen Angus steers averaging 350 kg body weight grazed a 5-acre (2 ha) lane and an adjacent 40-acre (16 ha) paddock on a gentle slope for 2 weeks before one died on approximately 11 July 2007, followed by another three at intervals during the next week. The herd was then moved to a 70- to 80-acre (28–32 ha) paddock on flats beside a creek on 18 July. At the two sites, drinking water was provided from different artesian bores. Three steers were then found dead in the creek paddock at intervals during the next 3 weeks, but none of the seven carcasses was necropsied because decomposition was too advanced when they were found. On 8 August 2007, a recumbent steer was discovered by the owner in the creek paddock and examined on the same day by a veterinarian (AMC). Its rectal temperature was 41°C. It had tachycardia (heart rate 160–180 beats/min) and tachypnoea. It was comatose with mild clonic seizures. It had no blink reflex, its corneas were dry and pupils dilated. The steer died immediately after jugular blood samples were collected into EDTA and plain tubes.

An immediate necropsy revealed no significant abnormalities. Aqueous humour was collected into a sterile vial. The brain and slices of heart,

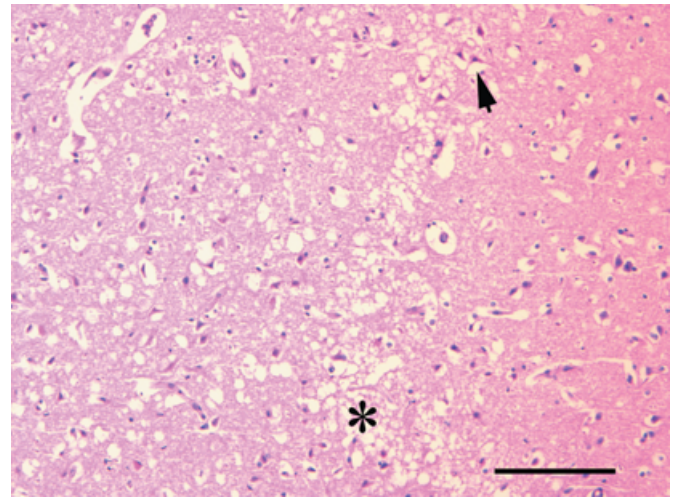


Figure 1. Polioencephalomalacia in the cerebral cortex with neuronal necrosis (arrow) and laminar spongiosis (*) of grey matter. Haematoxylin and Eosin stain. Case 1. Scale bar = 250 µm.

liver, kidney, lung, spleen, urinary bladder, rumen, reticulum, abomasum and small intestine were fixed in 10% formalin.

Haematological values were within reference ranges. Serum total bilirubin concentration was increased to 30 µmol/L (reference range 1–10) and serum creatine phosphokinase (CK) activity was increased to 1640 IU/L (reference range 10–200). Other serum values were within reference ranges. Lead was not detected in whole blood (lower limit of detection 0.10 mg/L). Nitrate was not detected in the aqueous humour (lower limit of detection 10 mg/L).

Histological examination of brain sections revealed severe PEM in the cerebral cortex with laminar spongiosis and neuronal necrosis (Figure 1). There was mild alveolar and interstitial emphysema in the lung. No significant abnormalities were seen in other tissues.

The pasture at both sites consisted of only forbs and no grass. At both sites there was an 80 to 90% ground cover consisting of approximately 60% *Sisymbrium irio* L. (London rocket) (Queensland Herbarium vouchers AQ751757, AQ751758) and 40% *Capsella bursapastoris* (L.) Medik. (shepherd's purse) (Queensland Herbarium vouchers AQ751759, AQ751760) (Figures 2–4). Both of these plant species were flowering and fruiting. Batches of both plants (≈ 5 kg fresh weight each) were collected by the owner as 'grab' samples from one part of the paddock on 23 August and sent to Biosecurity Sciences Laboratory, Yeerongpilly, Brisbane, for formal identification and chemical assay, arriving next day in fresh condition.

Bore water samples from both sites were collected for chemical assay (see Table 1). The total sulfur content of DM from combined feed and water was calculated to be 0.62%, assuming cattle body weights of 350 kg, 20°C ambient temperature and no discrimination by cattle between pasture components, and using the overall mean sulfur content of flowering and fruiting tops and leaf and stem components of the plants. Water contributed only 0.0020% sulfur (Lane bore) or 0.0056% sulfur (Creek bore) to the total.



Figure 2. Case 1 at Bell. Paddock with a dense cover of *Sisymbrium irio* (London rocket) and *Capsella bursapastoris* (shepherd's purse). Image created on 30 August 2007.



Figure 4. *Capsella bursapastoris* (shepherd's purse), flowering and fruiting plant. Bell, 30 August 2007.



Figure 3. *Sisymbrium irio* (London rocket), flowering and fruiting plant. Bell, 30 August 2007.



Figure 5. Case 2 at Jandowae, Queensland. Paddock with dense cover of *Raphanus raphanistrum* (wild radish). Image created on 30 August 2007.



Figure 6. *Raphanus raphanistrum* (wild radish), flowering plant. Jandowae, 30 August 2007.

Table 1. Sulfur, copper, molybdenum and nitrate content of pasture weeds and sulfur, copper and molybdenum content of water supplies associated with two bovine polioencephalomalacia incidents on the Darling Downs, Queensland, in 2007

Case	Material analysed (Queensland Herbarium voucher numbers for plants)	Plant part	Plant dry matter % as received by the laboratory	Sulfur % (plant); mg/L (water)	Copper mg/kg (plant); mg/L (water)	Molybdenum mg/kg (plant); mg/L (water)	Potassium nitrate %
1 (Bell)	<i>Sisymbrium irio</i> (AQ 751757, AQ751758)	Flowering & fruiting tops	14.5	0.96	10.0	0.54	0.5
		Leaves & stems	12.1	0.58	5.5	0.25	2.5
		Roots	11.4	0.57	6.2	0.35	0.7
	<i>Capsella bursapastoris</i> (AQ751759, AQ751760)	Flowering & fruiting tops	22.7	0.54	6.7	0.60	0.1
		Leaves & stems	17.4	0.27	7.8	0.54	–
		Roots	43.9	0.18	12.7	0.52	< 0.05
	Water, lane	–	–	4.0	0.015	0.010	–
Water, creek paddock	–	–	11.0	0.010	0.010	–	
2 (Jandowae)	<i>Raphanus raphanistrum</i> (AQ751761, AQ751762)	Flowering & fruiting tops	12.8	1.26	15.5	0.73	2.8
		Leaves & stems	9.0	0.72	16.0	0.80	2.9
		Roots	15.8	0.48	14.5	1.04	1.8
	Water	–	–	15.0	0.010	< 0.05	–

All plant chemical assay data are on a dry matter basis.

Case 2 (Jandowae)

A herd of 150 cattle averaging 300 kg body weight grazed a 140-acre (57 ha) paddock of heavy clay soil that had previously supported a failed millet crop, but that now carried a 60 to 70% ground coverage of forbs, almost all of which were *Raphanus raphanistrum* L. (wild radish, jointed charlock) (Queensland Herbarium vouchers AQ751761, AQ751762) (Figures 5, 6). The locality had received 190 mm of rain in June 2007 and 63 mm some days before the herd was placed in the paddock (Figure 7). Drinking water was provided from an artesian bore. On 7 August 2007, six days after first grazing these plants, a Friesian heifer appeared to be blind and died soon after being noticed. No necropsy was done. On 17 August, an 18-month-old Brahman × Lowline heifer became ill and was examined by a veterinarian (MLS) on that day. It was seen circling, unaware of its surroundings, apparently blind with no menace reflex and head-pressing. Jugular blood samples were collected into EDTA and plain tubes and tested as above.

Erythrocyte count was increased at $9.94 \times 10^{12}/L$ (reference range 5.8–8.9) and there was marginal hypocalcaemia with serum calcium at 2.01 mmol/L (reference range 2.1–2.8). Other haematological and clinical chemistry values were within reference ranges. Lead was not detected in whole blood (lower limit of detection 0.10 mg/L).

The heifer was immediately treated by slow IV injection with 2.5 g thiamine hydrochloride (20 mL of 125 mg/mL solution, Vitamin B₁ Nature Vet), 0.5 g flunixin meglumine (10 mL of 50 mg/mL solution, Flunixin® Norbrook Laboratories Australia) and 1.5 g oxytetracycline hydrochloride (15 mL of 100 mg/mL solution, Engemycin 100®, Intervet Australia) and similar doses were given daily for 4 days for the flunixin and oxytetracycline and for 5 days for the thiamine. The owners reported clinical improvement after 2 days and full recovery in 4 weeks.

On 19 August, a 3 year-old Lowline cow became ill and was examined by a veterinarian (MLS) on that day. The cow was laterally recumbent

with a heart rate of 144 beats/min and a rectal temperature of 37.6°C. There were skin abrasions on the face and head consistent with the cow having been head-pressing. The menace reflex was absent. She was immediately treated by slow IV injection with 2.5 g thiamine hydrochloride (20 mL of 125 mg/mL solution), 30 mg dexamethasone sodium phosphate (6 mL of 5 mg/mL solution, Dexapent® Illium Veterinary) and 2.0 g oxytetracycline hydrochloride (20 mL Engemycin 100® Intervet Australia). Jugular blood samples were collected into EDTA and plain tubes, as above.

Increased haemoglobin concentration (16.4 g/dL; reference range 9.7–14.0), packed cell volume (51%; 28–42) and erythrocyte count ($10.63 \times 10^{12}/L$; 5.0–7.7) were interpreted as evidence of dehydration. There was a slight leucocytosis ($16.4 \times 10^9/L$; 5.2–15.0) and increased plasma fibrinogen concentration (1.4 g/dL; 0.3–0.7) suggesting an inflammatory response. Clinical chemistry results indicated muscle damage (CK 164,380 IU/L; 10–200; AST 2989 IU/L; 30–170), azotaemia (creatinine 559 µmol/L; 40–220; urea 23 mmol/L; 2.0–8.5), slight increase in total bilirubin concentration (20 µmol/L; 1–10), moderate hypermagnesaemia (magnesium 1.48 mmol/L; 0.65–1.30) and marked hypocalcaemia (calcium 1.29 mmol/L; 2.1–2.8) with marginal hypoalbuminaemia (albumin 27.4 g/L; 30–45). Other clinical pathology values were within reference ranges.

The cow died on 20 August and was necropsied by a second veterinarian (SAD) 2 h later. There was emphysema of lungs, pericardium and perirenal fat. No abnormalities were noted in other organs and tissues. The brain and slices of lung, heart, spleen, liver, kidney, small and large intestines were fixed in 10% formalin for histopathological examination. Unpreserved kidney was collected for toxicological testing.

Histological examination of tissues revealed severe acute PEM in the cerebral cortex with necrotic neurones, swollen capillary endothelial cells and cortical spongiosis. There were scattered small foci of hepatocyte necrosis with neutrophil infiltration, alveolar and interstitial emphysema in the lung, and a few small foci of myocardial necrosis. These were all interpreted as incidental findings. No lesions

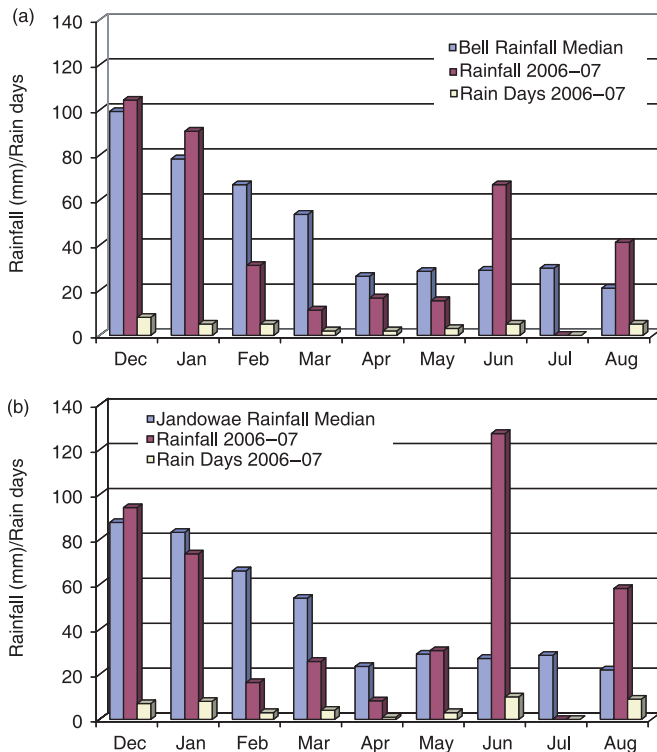


Figure 7. Rainfall December 2006 to August 2007, at a) Bell store (Meteorological Station 041005) and b) Jandowae Post Office (Meteorological Station 041050). Data from the Climate and Consultancy Section, Brisbane Regional Office, Australian Bureau of Meteorology.

were seen in other tissues. Lead was not detected in kidney tissue (lower limit of detection 1.0 mg/kg).

A batch of *Raphanus raphanistrum* of approximately 5 kg fresh weight was collected by a veterinarian (MLS) as a 'grab' sample from one part of the paddock on 21 August and sent to the Biosecurity Sciences Laboratory for formal identification and chemical assay, arriving next day in fresh condition. A bore water sample was also collected for chemical assay (Table 1). The total sulfur content of DM from combined feed and water was calculated from these data to be 1.01%, assuming cattle body weights of 300 kg and 20°C ambient temperature, and using the overall mean sulfur content of flowering and fruiting tops and leaf and stem components of the plants. Water contributed only 0.0075% sulfur to the total.

Discussion

Known causes of PEM in ruminants are thiaminase,¹⁷ lead,¹⁸ hydrogen sulfide (manure gas),¹⁹ sodium ion (salt)²⁰ and sulfur^{6,7} poisonings, and cobalt deficiency.²¹ Known thiaminase sources available to grazing ruminants on the Darling Downs in descending order of potency²² are *Marsilea* spp. (nardoos), *Cheilanthes sieberi* (mulga or rock fern) and *Pteridium esculentum* (bracken fern). Neither affected cattle herd had known access to any of these plants. Lead poisoning was excluded in both cases by assays of blood and kidney tissue. Cobalt deficiency has not been diagnosed on the Darling Downs and was not investigated in these cases. Calculating

the contributions of feed and water to the total sulfur intake of the affected cattle by the method of Gould⁶ (with conservative assumptions) using data shown in Table 1, yielded a total sulfur content of the DM from combined feed and water of 0.62% for case 1 and 1.01% for case 2. If the cattle had discriminated in favour of flowering and fruiting tops in either case, or in favour of *Sisymbrium irio* in case 1, their sulfur intakes would have been greater. In neither case did water contribute significantly to sulfur intake. Thus, the total sulfur intake in DM in both cases exceeded the guideline value for maximum tolerable sulfur concentration in DM of 0.4%,⁶ or of 0.5% when the high proportion of forage in the diet was considered.⁵ Ruminant tolerance of dietary sulfur varies inversely with the proportion of concentrates in the diet.⁵ Cattle and other ruminants on diets containing less than 15% forage are at risk of PEM when dietary sulfur is as low as 0.35%.⁵ On the other hand, when diets contain more than 40% forage, ruminants are at risk of PEM when dietary sulfur exceeds 0.5%.⁵ Concentrations of copper and molybdenum in the plants and water (Table 1) were insufficient to moderate the effects of sulfur.^{5,6} Consequently, these cases were diagnosed as sulfur-associated PEM, a conclusion that was strengthened further when no more cattle were affected after free access to the plant taxa involved was prevented. Sulfur-associated PEM has been reported mostly in hand-fed cattle or cattle drinking water with a large sulfur content.^{6,23} PEM in grazing cattle that drink water containing small concentrations of sulfur is less commonly reported and our cases add to these and to the number of plants known to be capable of inducing it.

Members of the Family Brassicaceae are well known for containing large quantities of sulfur.^{14,24,25} Sulfur-containing plant defence compounds, including elemental sulfur, hydrogen sulfide, glutathione, phytochelatins, glucosinolates and sulfur-rich proteins, are crucial for the survival of plants under biotic and abiotic stress, mediating resistance to fungal and insect attack.²⁶ In line with this role, sulfur (as glucosinolates) is at its greatest concentration in the inflorescences and seed capsules²⁷ (Table 1).

The three plant species associated with these PEM incidents are naturalised weeds originating in Europe, but widespread in Australia.²⁸ The Brassicaceae in Australia currently contains 89 known species and interspecific hybrids in 59 genera,^{28,29} including garden plants, vegetable, fodder and oil seed crops, weeds of pasture and cultivation, and native forbs. Those most likely to pose a risk of causing PEM in grazing cattle are the species used as fodder, vegetable and oil seed crops, particularly if grazed when carrying seed capsules. Sheep grazing seeding *Brassica napus* (rape) in central New South Wales have suffered a syndrome consistent with PEM, but unconfirmed histologically.³⁰ Weedy species such as *Brassica tournfortii* (wild turnip), *Capsella bursapastoris* (shepherd's purse), *Diplotaxis tenuifolia* (sand rocket), *Hirschfeldia incana* (Buchan weed), *Lepidium draba* (hoary cress), *Raphanus raphanistrum* (wild radish), *Rapistrum rugosum* (turnip weed), and *Sisymbrium irio* (London rocket) pose a lesser threat because they are less likely to be deliberately grazed in large amounts. Nevertheless, PEM has been diagnosed histologically in cattle grazing dense populations of *Rapistrum rugosum* in the Narrabri district of New South Wales during 1987 (GI Carter, BA Vanselow, unpublished data 1987) and again during 2003 (SM Slattery, JG Boulton, unpublished data 2003), and in the Coonamble district of New South Wales during 2006 (EC Bunker, unpublished data 2006). The sulfur content of these plants was not investigated, but excessive sulfur intake may have been the cause of those incidents.

Seasonal conditions at Bell and Jandowae before and during the winter of 2007, principally the below-median rainfall from February to April 2007 followed by above-median rainfall in June and August 2007 (Figure 7), resulted in poor to absent residual summer pasture cover and thus an unusual dominance of pastures by Family Brassicaceae plants that was a necessary precondition for the reported PEM incidents. The usual late-winter growth of nutritious medic ('clovers'), such as *Medicago polymorpha* (burr medic), was preceded and possibly retarded by the brassicaceous weed growth. *Sisymbrium irio* was particularly abundant throughout the region at the time of the reported incidents. Anecdotal evidence suggests that other PEM incidents occurred in cattle in the region at that time, none of which were confirmed by laboratory examination.

Managing incidents of sulfur-associated PEM in grazing cattle involves treating affected cattle as early as possible with thiamine and glucocorticoids^{6,7} and immediately finding alternative feed and water of tolerable sulfur content⁵ for the herd. Recommended therapy is 10 to 20 mg thiamine/kg IV two or three times on the first day followed by the same dose IM for 2 to 3 more days, supplemented with 1 to 2 mg dexamethasone/kg IV to aid reduction of cerebral oedema.⁷ Affected animals that are ambulatory and eating have a good prognosis.⁷ The successful treatment of one of our cases is consistent with the view that the outlook for animals treated early can be positive. Preventing this toxicosis is difficult in circumstances such as those in our cases where available pastures are dominated by plants high in sulfur. Gradual adaptation of the ruminal microbe population to utilising sulfur without excessive hydrogen sulfide production occurs over the first 1 to 3 weeks of access to a high-sulfur diet,⁶ which gradually reduces the risk of PEM developing.

Acknowledgments

We thank the owners of the affected cattle for their willing cooperation. Nigel Fechner, Queensland Herbarium, Environmental Protection Agency Queensland, identified the plants (NF:1022/07). Grant Madill produced histological sections. Brian Burren assayed tissues for lead and ran the clinical chemistry tests. Lana Bradshaw ran the haematological tests. Peter Martin advised on plant processing and Adam Pytko and Madeleine Modina helped process the plants. Plant and water sulfur, copper and molybdenum concentrations were assayed by SGS Agritech, Toowoomba QLD. Neville Wedding, Brisbane Regional Office, Bureau of Meteorology, provided rainfall data.

References

- Cote FT. Rape poisoning in cattle. *Can J Comp Med* 1944;7:38–41.
- Perrett DR. Suspected rape poisoning in cattle. *Vet Rec* 1947;59:674.
- Wikse SE, Leathers CW, Parish SM. Diseases of cattle that graze turnips. *Compend Contin Educ Pract Vet Food Anim* 1987;9:F112–F121.
- Morton JM, Campbell PH. Disease signs reported in south-eastern Australian dairy cattle while grazing *Brassica* species. *Aust Vet J* 1997;75: 109–113.
- Klasing KC, Goff JP, Greger JL et al. *Mineral tolerance of animals*. 2nd rev. edn. The National Academies Press, Washington DC, 2005:372–385.
- Gould DH. Update on sulfur-related polioencephalomalacia. *Vet Clin N Am Food Anim Pract* 2000;16:481–496.
- Niles GA, Morgan SE, Edwards WC. The relationship between sulfur, thiamine and polioencephalomalacia: a review. *Bov Practit* 2002;36:93–99.
- Shreeve JE, Edwin EE. Thiaminase-producing strains of *Clostridium sporogenes* associated with outbreaks of cerebrocortical necrosis. *Vet Rec* 1974;94:330.
- Morgan KT, Lawson GHK. Thiaminase type 1-producing bacilli and ovine polioencephalomalacia. *Vet Rec* 1974;95:361–363.
- Haven TR, Caldwell DR, Jensen R. Role of predominant rumen bacteria in the cause of polioencephalomalacia (cerebrocortical necrosis) in cattle. *Am J Vet Res* 1983;44:1451–1455.
- Hill FI, Ebbert PC. Polioencephalomalacia in cattle in New Zealand fed choux moellier (*Brassica oleracea*). *NZ Vet J* 1997;45:37–39.
- Kul O, Karahan S, Basalan M, Kabakci N. Polioencephalomalacia in cattle: a consequence of prolonged feeding barley malt sprouts. *J Vet Med* 2006; 53A:123–128.
- Helrich K (editor). Lead in food 934.07. *Official methods of analysis of the Association of Official Analytical Chemists*. 15th edn. The Association, Washington DC, 1990;1:258–261.
- Benton Jones J Jr, Wolf B, Mills HA. *Plant analysis handbook: A practical sampling, preparation, analysis, and interpretation guide*. Micro-Macro Publishing Inc., Athens, Georgia, 1991:25–29, 34, 176–187, 196–197.
- McKenzie RA, Rayner AC, Thompson GK, Pidgeon GF, Burren BR. Nitrate–nitrite toxicity in cattle and sheep grazing *Dactyloctenium radulans* (button grass) in stockyards. *Aust Vet J* 2004;82:630–634.
- McKenzie RA. Plant poisoning? Which plant? [Leading Article]. *Aust Vet J* 1993;70:201–202.
- Pritchard D, Eggleston GW, Macadam JF. Nardoo fern and polioencephalomalacia. *Aust Vet J* 1978;54:204–205.
- Christian RG, Tryphonas L. Lead poisoning in cattle: brain lesions and hematologic changes. *Am J Vet Res* 1971;32:203–216.
- Hooser SB, van Alstine W, Kiupel M, Sojka J. Acute pit gas (hydrogen sulphide) poisoning in confinement cattle. *J Vet Diagn Invest* 2000;12: 272–275.
- Trueman KF, Clague DC. Sodium chloride poisoning in cattle. *Aust Vet J* 1978;54:89–91.
- MacPherson A, Moon FE, Voss RC. Biochemical aspects of cobalt deficiency in sheep with special reference to vitamin status and a possible involvement in the aetiology of cerebrocortical necrosis. *Br Vet J* 1976;132:294–308.
- McCleary BV, Chick BF. The purification and properties of a thiaminase I enzyme from nardoo (*Marsilea drummondii*). *Phytochemistry* 1977;16:207–213.
- Radostits OM, Gay CC, Hinchcliff KW, Constable PD. *Veterinary medicine: A textbook of the diseases of cattle, horses, sheep, pigs and goats*. 10th edn. Saunders Elsevier, Edinburgh, 2007:2006–2012.
- Whittle PJ, Smith RH, McIntosh A. Estimation of S-methyl sulphoxide (kale anaemia factor) and its distribution among brassica forage and root crops. *J Sci Food Agric* 1976;27:633–642.
- Willey N, Wilkins J. An analysis of intertaxa differences in sulfur concentrations in angiosperms. *J Plant Nutr Soil Sci* 2006;169:717–727.
- Rausch T, Wachter A. Sulfur metabolism: a versatile platform for launching defence operations. *Trends Plant Sci* 2005;10:503–509.
- Booth EJ, Walker KC, Griffiths DW. A time-course study of the effect of sulphur on glucosinolates in oilseed rape (*Brassica napus*) from the vegetative stage to maturity. *J Sci Food Agric* 1991;56:479–493.
- Hewson HJ. Brassicaceae (Cruciferae). *Flora Aust* 1982;8:231–357.
- Australian National Herbarium. *Australian plant name index*. <http://www.anbg.gov.au/cgi-bin/apni>. Accessed 12 December 2007.
- Seaman J, Carrigan M. Suspect rape poisoning (*Brassica napus*) in sheep. *Aust Soc Vet Pathol Rep* 1987;16:15–16.

(Accepted for publication 27 August 2008)