SUMMARY OF BISERRULA PROJECT 2013

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A summary prepared for the AWET

Throughout 2013, all planned data and sample collection has been collected in preparation for data analysis and thesis construction.

Due to unforeseen circumstances, Georgia Ladmore's thesis is due in April 2014, by which time all analysis related to this study will be completed. A copy of the thesis will be submitted at that time.

SURVEY OF PRODUCERS

Producers known to be using Biserrula for feeding livestock were contacted and asked to complete a survey investigating the risk factors for photosensitisation following Biserrula consumption. Thirty two producers responded, and the contribution of these farmers was integral to the success of this component of the study. Producers were distributed across Western Australia and New South Wales.

Wool (n=27) and meat (n=20) sheep producers were represented, as well as one beef producer. All survey respondents used 'Casbah' Biserrula, and the majority of pastures were part of crop rotation (93.3%). Clay was the dominant parent soil material (33.3%) and the soil texture was typically either sandy loam (46.7%) or light clay (26.7%). Only 33.3% of producers had a current soil test. The majority of paddocks had fertiliser applied (92.6%) The majority of paddocks (70%) were planted prior to 2008.

Of the producers that grazed Biserrula, 55.2% had experiences of photosensitised stock. Sheep of all ages and sexes were affected, although ewes were most commonly reported. No farmers reported photosensitisation to be caused by other plants grazed concurrently.

Of the animals affected, 46.7% were described as mildly affected, 40% were moderately affected and 13.3% were severely affected. The framework for categorisation was as follows:

MILD = Skin is red, weepy and swollen but not blackened. Eyes, muzzle and ears are affected. Swelling of the ears in young stock. Mild dermatitis, affecting exposed skin areas only. Some eye and nasal discharge may be present. Irritation and pain will be evident by scratching of head and face. Udders will be affected in dairy cattle. Animals may show reduction in appetite.

MODERATE = Skin is very red, blackened and flaking. There will be severe swelling of ears and soft tissues of the face. Focal or generalised hair or wool loss. Dermatitis affecting whole body. Animals may show significant nasal and eye discharge. Eyes may be swollen shut. Animals will show difficult in eating and / or reduced appetite. Irritation and pain will be evident by continuous scratching of affected regions. Animals will preferentially seek shade or stand in dams to reduce irritation.

SEVERE = The skin will be sloughing and necrotic in patches with weeping lesions. Severe dermatitis including secondary lesions. Significant loss of wool or fur. Animals will be depressed and have poor appetite. They will seek shade, show skin scratching and pain related behaviours or may, in extreme cases, show a complete lack of movement or response to normal stimuli.

No clear risk factors have been identified yet, but thorough data analysis is pending. There may be an association between physiological state and risk of photosensitisation.

Only 18.8% of producers that experienced photosensitisation used a veterinarian to confirm the diagnosis. This is important for the dissemination of information back to producers- in most cases, this information needs to be fed directly back to the farmers through agricultural officers, farmers groups and agronomists, rather than via veterinary channels. Of the surveyed producers, 90.3% want to be contacted with the results of the study.

ESTABLISHMENT OF HAEMATOLOGICAL & BIOCHEMICAL REFERENCE INTERVALS

To ensure meaningfulness of subsequent results, current laboratory techniques were used to establish haematology and biochemistry reference intervals for mixed breed weaned lambs. Blood was collected from 74 weaned, 6- to9-month-old mixed breed lambs. Data was ultimately used for 71 lambs. Haematology and biochemistry panels were performed using routine automated methods. The reference intervals were determined as within 2 x the standard deviation of subjects in the cohort. Intervals derived from this study were different to the intervals used by this laboratory previously. Formulation of haematological and biochemical reference intervals for this signalment of animals is necessary for continuing research using lambs as subjects.

CASE SERIES: CLINICAL EVALUATION & NECROPSY OF AFFECTED ANIMALS

Following the survey distribution, an offer was received from a Biserrula producer with affected lambs. These lambs were unweaned and had been in paddocks of Biserrula since birth. Ewes were largely unaffected. Ten subclinical animals and five clinically photosensitised animals were sampled for haematological and biochemical testing.

Significant ($p<0.05$) differences were noted between the subclinical and photosensitised animals. Haematocrit was significantly lower in photosensitised animals. Creatinine was significantly higher in photosensitised animals. Bilirubin was significantly lower in photosensitised animals. Chloride was significantly lower in photosensitised animals.

Five of the worst affected lambs were sacrificed for necropsy. Fleece samples were taken for fleece testing. On necropsy and histopathological inspection, the only consistent, clinically relevant lesions were skin lesions.

Despite the small sample size, the lack of enzymatic, histologic nor gross pathological lesions are encouraging suggestions that the toxin(s) involved in Biserrula photosensitisation is/are a primary photosensitation agent.

EXPERIMENTAL TRIAL: GRAZING NAÏVE LAMBS ON CONTROL, MIXED & BISERRULA PASTURES

Seventy four Merino and White Suffolk x Merino lambs were used for this experiment. Dorpers were also grazed simultaneously as part of a separate experiment investigating growth rates and pasture preferences on pasture legumes. Experimental lambs were bled prior to, and at the exit of the trial. The three treatment groups were control, mixed, and Biserrula grazing. The Biserrula used for this experiment was 'Mauro' variety. The control group grazed subterranean clover. The mixed group grazed a split pasture which contained Biserrula and subterranean clover. The Biserrula group grazed Biserrula.

Sheep were introduced to the plots on the 17th October, 2013. Clinical signs consistent with photosensitisation were first observed on the 21st October, 2013. The trial was complete by the 29th October 2013. Between the 22nd-29th October 2013, 14 animals, including photosensitised animals from Biserrula plots, were removed for

necropsy. Fleece samples were taken for fleece testing. Due to the unexpected rapidity of onset of photosensitisation, it is unlikely that fleece testing will yield useful results, and as such, these samples were not sent for processing, although these samples have been stored for future use. Again, on necropsy and histopathological inspection, the only consistent, clinically relevant lesions were skin lesions.

The lack of enzymatic, histologic nor gross pathological lesions add further weight that the toxin(s) involved in Biserrula photosensitisation is/are a primary photosensitation agent.

Anecdotal reports of photosensitisation of sheep whilst grazing 'Mauro' Biserrula can now be supported by this clinical example. Dorpers grazing the same plots as experimental animals were observed to exhibit signs of photosensitisation. The relative incidence and severity of photosensitisation between different sheep breeds would be an interesting avenue for further studies.

Further work is needed to establish the mechanism of action and toxin(s) involved Biserrula photosensitisation, however, the experimental work completed throughout 2013 has provided interesting insights into the disease progression. In particular, the onset of clinical signs within 96 hours was unexpected, and this is useful information to disseminate to farmers using Biserrula, as well as for the planning of future experiments.