CONTROL OF *HYPERICUM PERFORATUM* L. (ST JOHN'S WORT) BY A GRAZING MANAGEMENT SYSTEM THAT USES MERINO SHEEP

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Abstract A grazing management system for the control of St John's wort (*Hypericum perforatum* L.) growing on steep, inaccessible, rangeland hill country is currently being investigated at the weeds CRC Orange Agricultural Institute. St John's wort infested pastures can be poisonous for grazing animals. The system is being designed around grazing the plant during periods when the risk of poisoning is low but the negative effect of grazing on plant growth recovery is high. White wool producing Merino sheep are to be used as the grazing animal. This paper presents a synopsis of the findings of this investigation so far. The following concepts are addressed. Differences in hypericin content of St John's wort biotypes. Seasonal variations in the development of St John's wort plants. Seasonal variations in hypericin production by wort plants. Influence of sunlight exposure on sheep wort tolerance. Influence of wool cover on sheep wort tolerance. Wort and hypericin tolerance levels in the Australian Merino. Variation in wort tolerance between different Merino bloodlines, and finally, the determination of when wort pastures might be safe for white Merino sheep to graze.

INTRODUCTION

There is anecdotal evidence to suggest that repeated grazing by livestock of St John's wort infested pastures will eventually suppress wort growth and reduce its prevalence. Grazing could offer a cheap and practical solution to wort control on steep, inaccessible, rangeland hill country. However, the ingestion of wort by grazing animals can be detrimental to their health, in particular, the presence of large amounts of hypericin in the weed can result in a severe, sunlight induced, skin reaction called photosensitiseation (Bourke 1997).

Animals with pigmented skin are more resistant to wort poisoning. Consequently coloured breeds of goats, black faced breeds of sheep and coloured breeds of cattle have been traditionally recommended for grazing wort pastures. For a variety of reasons none of these recommendations have been adopted by livestock producers with large wort infestations. White wool producing Merino sheep are the most common livestock enterprise encountered on the wort infested hills of the Great Dividing Range in south-eastern Australia. This livestock enterprise has both community acceptance and commercial viability in these regions. Consequently a grazing management system for wort control using white wool producing Merino sheep is currently being investigated at the weeds CRC Orange Agricultural Institute. This paper presents a synopsis of the findings of that investigation so far.

MATERIALS AND METHODS

Seasonal hypericin determinations The seasonal hypericin determinations reported for wort plant material were established using a modified version of the method of Southwell and Campbell (1991). 2g of dried ground plant was extracted by soxhlet, firstly with t-butyl methyl ether (200ml for 4-8h) to remove the green chlorophyll, and then with absolute ethanol (100ml for 8h) to remove the red hypericin. The ethanolic solution was adjusted to 500ml and the ultra-violet spectra measured between 500-700nm. Hypericin absorption occurred at 591nm and any small amounts of chlorophyll present at 665nm. Where chlorophyll absorption was present it was corrected for, the resultant hypericin concentration was then determined and expressed in ppm.

Wort plant samples were collected at two field sites on the Central Tablelands of NSW every 3 weeks over a two year period. Two biotypes were studied, a broad leafed wort community growing at Orange and a narrow leafed community growing at Tuena. The sample for hypericin analysis was taken from a bulked collection of all soft growth material present on 50 plants selected at random on each sampling occasion. Soft growth being leaves, small fine stems and when present, flowers and immature fruit capsules. These plant parts have previously been shown to have higher levels of hypericin than the more lignified supporting stalks (Southwell and Campbell 1991), in addition, we have observed sheep selectively graze these soft growth portions and reject the coarser stalks. The plant material was air dried at room temperature indoors. Results reported are for the period late April 1997 to early December 1998. The growth stage of the wort plants was observed and recorded at each 3 weekly sample collection. The field sites sampled were not grazed, mown or defoliated by other mechanical or chemical means during the two year collection period.

Sheep dosing trials The hypericin content of wort plant material used in the sheep dosing trials was also determined by the above method, this material consisted of field grown wort, air dried at room temperature inside a shed. This wort was harvested at the flowering growth stage and once dried, the coarse stalks were discarded and the remaining plant material milled to a fine powder. The wort dose was put up in a water based slurry and administered via a stomach tube. The sheep used in the dosing trials were adult Australian Merino ewes drawn from 13 different blood lines. Superfine, fine and medium wool types were used. So far a total of 96 sheep have been studied. Wort intolerance was determined by monitoring daily rectal temperature elevations, together with observing animals for early signs of a clinical response to hypericin poisoning, these are, agitation, rubbing of the head and face against fixed objects, mild diarrhoea and mild depression. Once an animal was determined to be showing intolerance it was immediately moved indoors and allowed to recover. The protocol used was approved by an animal care and ethics committee that ensured it fell within the current regulatory guidelines for use of animals in research.

Maximum daily wort intake by sheep To determine the maximum daily amount of plant dry matter these Merino sheep might reasonably be expected to ingest, 33 were fed lucerne chaff ad lib for 14 days as a sole ration. The average daily intake was determined to be 17g/kg live weight, for a 45kg sheep this would be approximately 770g. Lucerne chaff has greater palatability than St John's wort consequently the maximum daily wort intake should be equal to or less than this amount.

RESULTS AND DISCUSSION

Hypericin content of wort biotypes During the period of this study a 2 to 3 fold greater level of hypericin production was frequently recorded for the narrow leafed biotype compared with the broad leafed. The maximum production occurred in both biotypes in early summer, with broad leafed measuring about 2500ppm and narrow leafed about 5000ppm, the minimum production occurred in late winter, with broad leafed measuring about 50ppm and narrow leafed about 100ppm.

Seasonal variations in wort development The following 5 growth stages and time periods were observed for both biotypes:

1. Growth of upright flower spikes from mid Sept to early Nov.
2. Full flowering from mid Nov to late Dec.
3. Development of fruiting capsules from early Jan to mid Mar.
4. Growth of prostrate winter stems from late Mar to mid Jun.
5. Winter stem static growth phase from late Jun to early Sept.

Variations in hypericin production Hypericin production in wort plants appeared to be strongly associated with the development of the upright growing flower stems. That is, hypericin levels started to rise rapidly in the Spring once the new season flower spike shoots exceeded a height of 5 to 10cm, continued that rise as the flower heads developed further and reached a maximum when the plant was in full flowering.

The hypericin production level changes recorded for each of the 5 growth stages of the narrow leafed biotype were:

1. From mid Sept to early Nov an initial level of about 300ppm rising to 600ppm.
2. From mid Nov to late Dec an initial level of about 1000ppm rising to 5000ppm.
3. From early Jan to Mid Mar an initial level of about 3000ppm falling to 1500ppm.
4. From late Mar to mid Jun an initial level of about 1500ppm falling to 300ppm.
5. From late Jun to early Sept a seasonally variable amount between 100 and 300ppm.

The hypericin production level changes in the broad leafed biotype were:

1. From mid Sept to early Nov an initial level of about 100ppm rising to 500ppm.
2. From mid Nov to late Dec an initial level of about 500ppm rising to 2500ppm.

3. From early Jan to Mid Mar an initial level of about 1000ppm falling to 400ppm.

4. From late Mar to mid Jun an initial level of about 400ppm falling to 100ppm.

5. From late Jun to early Sept a seasonally variable amount between 50 and 100ppm

Influence of sunlight exposure on sheep None of 11 sheep dosed with wort plant material at doses of up to 5.7g of wort per kg live weight but kept indoors (out of direct sunlight), reacted to this amount of wort ingestion. None of the treated sheep kept outdoors reacted to wort doses of up to 5.7g/kg on days when an extensive cloud cover existed (no bright sunlight exposure). All of the potentially adverse clinical effects of wort ingestion in sheep seemed to be dependant upon bright light activation of the circulating wort poison, generally regarded as hypericin and related chemical compounds. Wort ingestion did not affect sheep in any way during these experiments in the absence of bright sunlight exposure. The complete recovery of sheep affected by early signs of wort poisoning typically occurred within 12 hours of their movement indoors out of bright sunlight. Hypericin is a pigment compound and like any colouring agent or dye its chemical structure can be altered when exposed to particular wavelengths of light, it would seem that it is this photoactivated chemical state that makes hypericin pharmacologically active, hence potentially poisonous

Influence of wool cover on sheep For sheep dosed with wort plant material at from 2.5 to 5.7 g/kg live weight, there were significantly more clinical reactors amongst recently shorn sheep then amongst sheep carrying 4 months or more of wool growth. In the former group 45 out of 47 sheep reacted and in the latter group only 13 out of 49. Recently shorn sheep have a much greater area of skin blood vessels exposed to direct sunlight than do woolly sheep, this probably means they have a much greater ability to activate any circulating hypericin and make it poisonous.

Wort and hypericin tolerance in Merinos Wort intolerance was observable clinically within 24 to 30 hours of wort ingestion under favourable sunlight conditions. A single day dose of wort was observed to exert an adverse response in some sheep on each of the four days that followed, provided sunlight remained bright during that particular period. Affected sheep recovered overnight only to re-react following another 3 to 5 hours of sunlight exposure on the following day. The daily wort intake tolerance level for Merino sheep, carrying 4 months or more of wool, was determined to be approximately 2.5g/kg live weight. This was for broad leafed biotype dried wort plant material in the full flowering stage of growth and containing approximately 1100ppm hypericin. This amount of plant would equate to about 2.75mg of hypericin per kg live weight per day. For a 45kg sheep eating wort plant material only and ingesting about 770g of dry matter a day, this would equate to a wort pasture hypericin tolerance level of 160ppm during the most intense sunlight period of the year, from early November to early February. A greater level of tolerance would be anticipated during periods of low sunlight intensity, for example from early May to early August.

Differences between Merino bloodlines There appeared to be small differences in wort tolerance between different bloodlines of sheep as well as between individual sheep within a bloodline. Fine and superfine bloodline sheep appeared to be somewhat more tolerant of wort ingestion than those from medium wool bloodlines, however this trend needs to be tested further using a much larger sample size before it can be validated. It may be an association with a greater wool staple density in finer wool sheep as opposed to a more open staple in the fleeces of medium wool types. This would influence the penetration of sunlight down onto the skin surface, hence it would affect the overall ability of the blood vessels in the skin to activate the hypericin ingested in the wort. Individual sheep variation in wort tolerance would suggest that it is worth considering selection from within a chosen wort grazing flock, against individual sheep that seem to develop a clinical reaction to wort grazing, even when grazing is restricted to relatively safe times of the year. By culling these individuals over successive years a mob of wethers of superior wort tolerance could be quickly accumulated

When do wort pastures become safe? Hypericin levels of 160ppm or less occurred in the broad leafed biotype pasture between early May and mid October in 1997 (a very dry season at Orange) and between mid June and mid October in 1998 (a very wet season at Orange). In the narrow leafed biotype pasture the relevant periods were between late June and early October in 1997 (very dry at Tuena) and between early August and early September in 1998 (very wet at Tuena). The reduced sunlight intensity during these periods of the year would mean that sheep could tolerate wort pastures with more than 160ppm hypericin. Likewise it would be highly unlikely that the daily pasture ingestion would only consist of wort plant material, therefore the overall hypericin level of the wort component in the pasture could exceed 160ppm by a factor of 25 to 50% and still not cause any adverse effects in the flock, that is up to 200-240ppm hypericin. This may afford much greater scope for the safe grazing of the broad leafed biotype in all winters, but it would still only afford limited latitude for the safe grazing of the narrow leafed biotype in very wet winters (such as 1998).

The reduction of the wort density in the pasture, following successive years of grazing, will afford an ever increasing length in the annual safe grazing period because it will effectively decrease the potential total daily hypericin intake of the sheep. Likewise, it is envisaged that heavy winter grazing of a wort pasture may significantly delay the time of onset of the rapid spring rise in hypericin production by the plant. This would mean that a longer relatively safe spring grazing period may be possible than the present study would initially suggest.

CONCLUSIONS

Based on the findings of this study so far, the following approach to grazing management for the control of St John's wort is suggested. Firstly, use a flock of white wool producing adult Merino sheep from a fine (<20 microns) or superfine (<17 microns) wool producing bloodline. If using ewes, rather than wethers, make sure they are neither pregnant nor lactating during the wort grazing period. Only use adult sheep (not weaners) and make sure they are carrying a fleece of at least 4 months wool growth. Initially graze broad leafed wort biotypes from May 1 to Oct 14 (19 weeks) and narrow leafed biotypes from July 1 to Sept 14 (6 weeks). In subsequent years, as the amount of wort present in the pasture decreases, gradually increase the period of grazing by both starting at an earlier date and finishing at a later date. However, in Spring always make sure the sheep are moved off the wort pastures before the new season flower spike shoot growth exceeds 5 to 10cm in height because poisonous levels of hypericin will rapidly develop. Reducing the size of paddocks, particularly if this involves the specific fencing off of areas with high wort infestations, will make grazing management more effective, as will the use of very high stocking rates during the limited wort grazing period proposed.

This system should exert a significant impact on wort survivability, by repeatedly attacking the plant during its relatively sensitive over-wintering growth phase. The system should exert a strong annual suppressive pressure on the spring growth vigour of the plant, but at the same time considerably reduce the risk of health and production problems in the animals being used to graze it. Landholders would be well advised to take steps to ensure that the diminishing wort growth is replaced by other more appropriate pasture species, so that these can then compete with the declining wort and prevent its re-establishment.

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