

Pimelea Update -October 2022

Summary of research progress to combat Pimelea toxicity

- Laboratory trials showed sodium bentonite and fully activated biochar (heated to 1000°C) were effective adsorbents, binding 95 per cent of the Pimelea (simplexin) toxin in rumen fluid in test tubes.
- Cattle feeding trials showed administering bentonite to animals fed daily on Pimelea appeared to reduce toxicity impacts. However, dung analysis suggested neither bentonite or biochar (at the doses used) effectively bound the toxin through the entire digestive tract.
- Producer experiences indicate cattle feed more readily on bentonite than biochar. The feeding trial provided 80 grams of bentonite / head / day or 0.3g/kg bodyweight. Producers recommend intake of 100 to 200 grams of bentonite / day.
- Unfortunately, there is no successful rumen drench. None of the 100 plus rumen fluid samples of bacteria broke down the Pimelea (simplexin) toxin. This was confirmed through tests in an artificial rumen in the laboratory, followed by cattle feeding trials with the most promising rumen fluid samples.
- Biodegradable biopolymer materials were tested to develop a rumen bolus that could trickle feed small amounts of simplexin into the rumen. This would help sustain a small population of bacteria that could degrade the simplexin toxin, even when cattle were not grazing on Pimelea plants. Research confirmed the most promising bolus is polyhydroxyalkanoate (PHA) which lasts for up to four months in the rumen. Rumen boluses have a role for slow-release of drugs and other products such as methane-reducing compounds.

Background

South-west Queensland producer pledges of cash and in-kind during 2017 to 2018 with co-matched funding from Meat and Livestock Australia's MLA Donor Company highlighted the severe impact of Pimelea to the cattle industry across inland Australia. The initial \$150K project enabled collection of 110 rumen fluid samples from affected and non-affected cattle and other ruminants (goats, sheep and kangaroos). These timely rumen samples were the basis of research into a rumen drench.

In 2019, MLA invested \$1.5million of producer levy funds into toxic plant research. Combating Pimelea toxicity as a first priority. No further call-in of producer pledges was required with this significant injection of research funding from MLA for a three-year period. The Pimelea research was led by Professor Mary Fletcher QAAFI and DAF microbiologist Diane Ouwerkerk.

In search of a rumen inoculum (drench) with bacterial isolates that can break down the Pimelea toxin called 'simplexin'.

Unfortunately, there was no success in developing a rumen drench. Testing the combinations from 110 field collected rumen bacterial samples resulted in none that could breakdown simplexin.. Testing required running fermentations for 63 day cycles, through an artificial, anaerobic rumen at the Ecosciences laboratory in Brisbane (Figure 1). The artificial rumen was fed daily with Pimelea plant material or a 50:50 mix of Pimelea and buffel grass hay. Daily rumen fluid samples were chemically analysed for presence and degradation of Pimelea material and the simplexin toxin. The most promising rumen fluid samples were used in a cattle feeding trial in 2021. **None of the rumen bacterial samples prevented the onset of Pimelea toxicity symptoms.**



Figure 1: Diane Ouwerkerk DAF with the artificial rumen at Ecosciences laboratory in Brisbane where field-collected rumen fluid samples are tested for ability to breakdown Pimelea plant material and degrade the simplexin toxin.

Rumen adsorbent studies

All three tested adsorbents had some effect in binding the simplexin toxin in laboratory experiments using rumen fluid. The most effective was sodium bentonite (Truefeed®) at 12mg/ml, which bound 95 per cent of the simplexin toxin (Figure 2). Other adsorbents Nutrilick® biochar and Elitox® bound 60 to 30 per cent of simplexin, depending on the concentration of adsorbent present. Both sodium and calcium bentonite remained bound to simplexin for the longest period. **Cattle feeding trials and blood samples confirmed sodium bentonite and fully activated biochar bind to the simplexin toxin in the rumen which prevented toxicity symptoms.**

The cattle feeding trial provided 80g (= 0.3g/kg bodyweight) of bentonite per day, whereas producers from Roma and Begonia prevented Pimelea toxicity symptoms by feeding 100 to 200g per day to cattle (Ross Turner, Nutrien Roma). The mixing ratio of one bag of bentonite to one bag of supplementary lick resulted in sleek fresh coats and no losses due to Pimelea.

If lick containing bentonite is fed all year, cattle are less likely to go off lick when green pasture growth appears after September. Dry Pimelea stalks with seed tufts in amongst pasture tussocks during spring and summer are high risk for onset of toxicity symptoms in cattle.

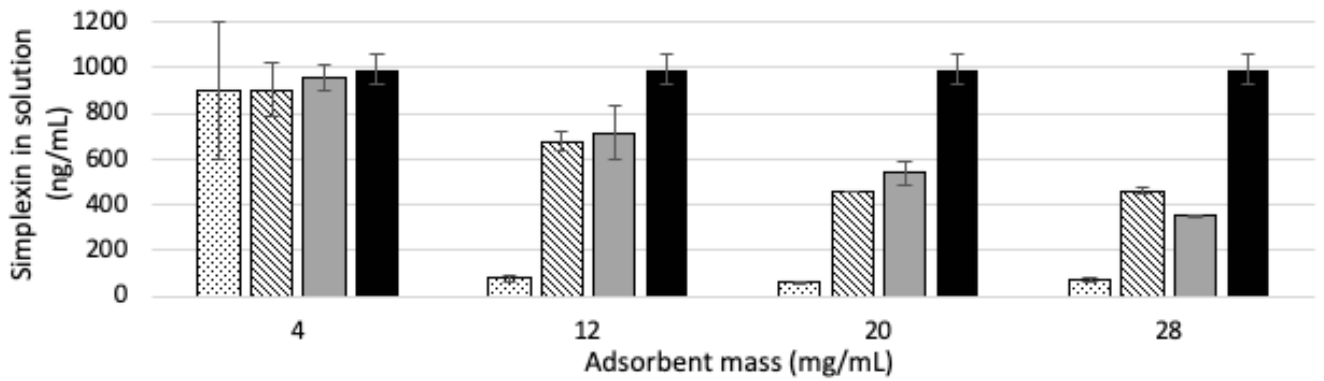


Figure 2. Mean results (\pm SD) for adsorbents: Bentonite, Biochar, Elitox, Blank (no adsorbent).

Reference: Gordon R, Hungerford N, Laycock B, Ouwerkerk D, Fletcher M. 2019. *Adsorbents for the sequestration of the Pimelea toxin, simplexin.* Poster 352 at the Northern Beef Research Update Conference, Brisbane, 2019 <http://www.nabrc.com.au/wp-content/uploads/2019/09/NBRUC-Conference-Proceedings.pdf>.

Biodegradable biopolymers for a slow-release, rumen bolus

The MLA research project also tested biodegradable rumen bolus compounds that could trickle-feed small amounts of Pimelea plant material into the rumen, to sustain effective bacteria. The most promising bolus combination for sustained release was polyhydroxyalkanoate (PHA). The bolus lasted for four months in the fistulated cattle feeding trial. Although no effective rumen bacteria were found, the biopolymer bolus has potential future use for slow-release delivery of drugs and other products such as methane-reducing compounds into the rumen.



Key contacts for rumen research:

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Symptoms of Pimelea toxicity include ruffled coat, diarrhoea, swelling in the brisket and lower jaw. Simplexin constricts blood vessels between the heart and lungs.

Other research and activities - Pimelea ecology research

Dr Rashid Saleem from University of Queensland UQ Gatton campus studied *Pimelea* ecology to ascertain optimal trigger points for forecasting field germination of the toxic native plant.

- Optimal soil temperature range for *Pimelea* germination is between 10°C and 25°C (Figure 4a,b).
- *Pimelea* seeds have a dormancy period. The embryo has a maturation process and the hard seed coat of fresh seed (Figure 4c) requires at least three months weathering to become permeable enough to allow seed germination.
- When seeds were treated with the stimulant, gibberellic acid at 1.15milliMolar concentration, approximately 80 per cent of seed germinated after 50 days. Gibberellic acid breaks seed dormancy.

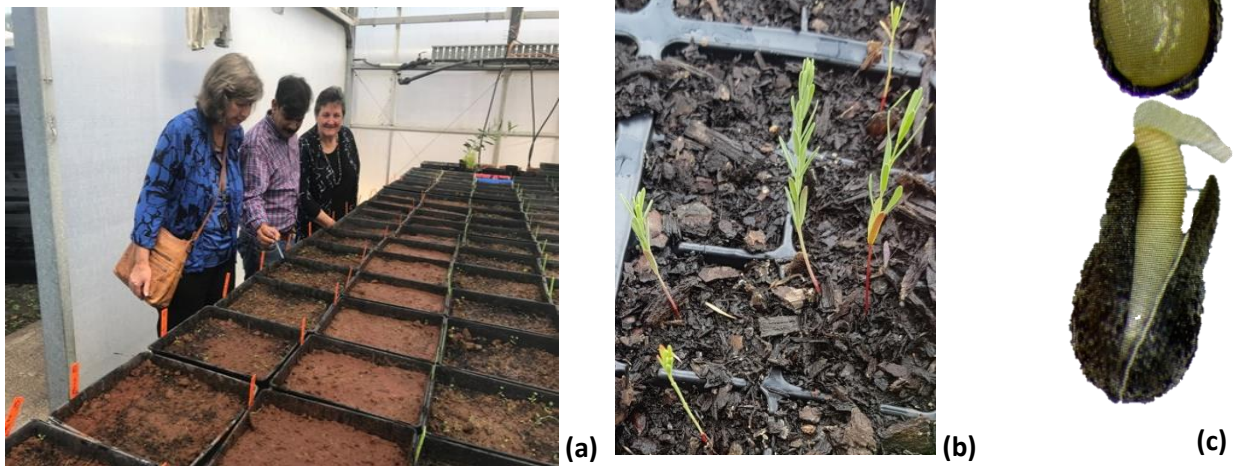


Figure 4. (a) Mary Fletcher, Rashid Saleem, Marie Vitelli with germinable seed bank trays from soil collected from two field sites. (b) *Pimelea* seedlings often have a characteristic lower red stem. (c) Hard seed coat, internal embryo and germinating *Pimelea trichostachya* seed. Photos by R. Saleem.

Residual herbicide trial to suppress *Pimelea* germination

During 2020, Rashid conducted a pot trial at UQ Gatton to assess residual herbicides to suppress *Pimelea trichostachya* seedling emergence. Treating a small paddock or graded tracks to suppress *Pimelea* during high risk seasons may be a useful option to free an area of this toxic plant.

In the pot trial, tebuthiuron pellets (Graslan) at a product rate of one gram per square metre prevented *Pimelea* seedling emergence. At lower rates (0.25g to 0.75g/m²), any emerging seedlings died within seven days.

The commercial formulation of a 20 per cent (coarse) pellet and 10 per cent (finer) experimental formulation of tebuthiuron were used in the trial (Figure 5). The finer formulation enabled a more even distribution on the soil surface than the tebuthiuron pellet.

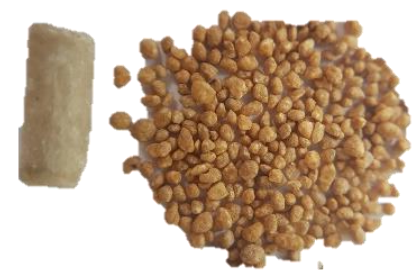


Figure 5. One 20% pellet and equivalent 10% formulation of tebuthiuron.

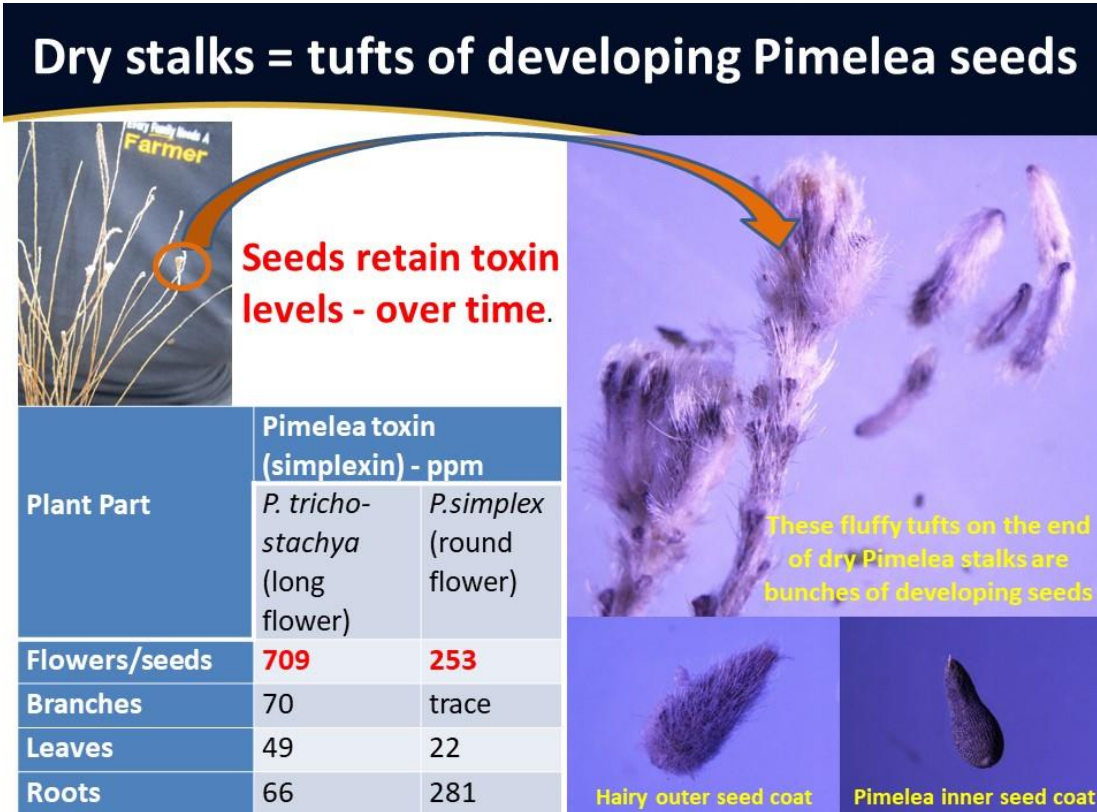
A small-scale field trial needs to be conducted (up to 1ha in total area, under APVMA permit 7250 <https://permits.apvma.gov.au/PER7250.PDF>). Other residual herbicides for consideration in an experimental trial include metsulfuron-methyl at 30, 40 and 50 grams per hectare (plus wetter) and picloram granules (Tordon granules) at 50, 75 and 100 grams per square metre.

Why dry Pimelea stalks are toxic to cattle?

Cattle tend to avoid grazing dense stands of odorous, green Pimelea plants. However, cattle cannot distinguish dry Pimelea stalks amongst buffel and other pasture tussocks. Producers have reported major cattle losses from ingesting dry Pimelea stalks. Dry stalks often contain a fluffy tuft of very toxic, immature seeds!

Flowers and seeds contain the highest levels of the simplexin toxin in Pimelea.

Dry stalks = tufts of developing Pimelea seeds



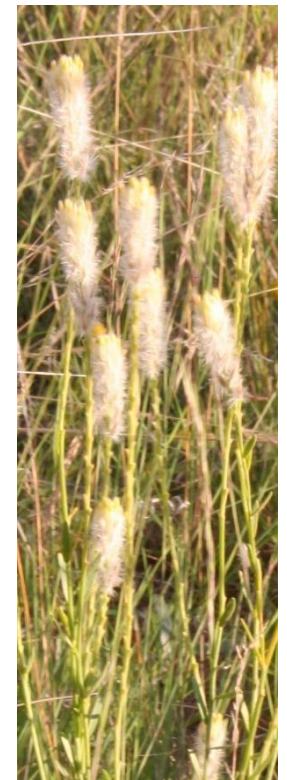
Seeds retain toxin levels - over time.

Plant Part	Pimelea toxin (simplexin) - ppm	
	<i>P. trichostachya</i> (long flower)	<i>P. simplex</i> (round flower)
Flowers/seeds	709	253
Branches	70	trace
Leaves	49	22
Roots	66	281

These fluffy tufts on the end of dry Pimelea stalks are bunches of developing seeds

Hairy outer seed coat Pimelea inner seed coat

Source: Understanding Pimelea poisoning of cattle (2009) https://futurebeef.com.au/wp-content/uploads/2011/09/Understanding_pimelea_poisoning_of_cattle.pdf



Green plants of flowering *Pimelea trichostachya* (above) and *P. simplex* (below). Photos J.Milson.

Risk of dry Pimelea stalks & seeds

Reduce exposure to dried *Pimelea trichostachya* amongst buffel.

- Seeds & flowers have highest level of toxin
- Consuming 2g dried plant (=1 plant) per day over 35 days = toxicity symptoms in 150kg weaner.
- Prevent seeds blowing into stock water.



What will Pimelea do this year?

Pimelea is often abundant after dry summers, followed by cool, wet winters. Rainfall events greater than 40mm during April to August may trigger Pimelea germination. Flowering can occur most months, in response to rainfall. Visible, standing dead plant material can occur anytime of the year and is a risk to grazing cattle. Livestock trampling, heavy rain and wind tend to flatten dead stems.

Cattle poisoning is more frequent between September to January when Pimelea is abundant.

Current herbicide management options for Pimelea

In small areas, where herbicide management is a feasible option, seven herbicides are registered for control of Pimelea as per APVMA Minor Use Permit 13549 in NSW and QLD <https://permits.apvma.gov.au/PER13549.PDF> . Spray young Pimelea plants to ensure no dead toxic stalks with seed tufts remain standing amongst pasture tussocks. These herbicides will also affect broadleaf legumes and forbs in treated areas.

Active constituent	Example of Product Trade Names	Rate plus add wetter to all products	Application method	Grazing WHP (days)
Fallow land				
Glyphosate	Roundup 360g/L	3L/ha 200ml/100L	Boom Spot spray	Nil
Pastures, rights of way, agricultural non-crop areas, commercial and industrial areas				
2,4-D	Crop Care Amine 625g/L	800ml/ha	Boom Spot spray	7
Metsulfuron-methyl	Metsulfuron 600g/kg	10g/ha 10g/100L	Boom Spot spray	Nil
2,4-D and picloram	Corteva Tordon 75-D	1.5L/ha 100ml/100L	Boom Spot spray	7
MCPA and Dicamba	Nufarm Kamba M	3L/ha 200ml/100L	Boom Spot spray	7
Diflufenican	Bayer Brodal Options	250ml/ha 17ml/100L	Boom Spot spray	14
Improved pastures containing medics				
2,4-D B	Nufarm Buttress	750ml/ha 50ml/100L	Boom Spot spray	7

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