Effects of pine oil, sugar and covers on germination of serrated tussock (*Nassella trichotoma*) and kangaroo grass (*Themeda triandra*) in a Pot Trial

David A. McLaren[1,2], Masha Fridman[1] and Julio Bonilla[1]
[1] Department of Primary Industries, Victorian AgriBiosciences Centre, Bundoora, VIC 3083
[2] La Trobe University, Bundoora, VIC 3086
(david.mclaren@dpi.vic.gov.au)

Summary Weeds cost Australia more than \$4.5 billion in control and lost production costs based on present day figures (Sinden *et al.* 2004). Many of these weed issues could have been prevented had proactive eradication of new weed infestations had taken place. The evolution of weed control practices now places an extremely high emphasis on eradication of new weed incursions before their populations become an uncontrollable scourge on agriculture and the environment. A key component of a weed eradication programs is destroying or controlling the weed seed bank (Panetta 2004). The use of conventional herbicides for weed seed bank control is confined to pre-emergent herbicides or soil fumigants. Increasingly, the reliance on use of synthetic herbicides has resulted in environmental (Freemark and Boutin 1995) and human health (Cox and Surgan 2006, Weisenberger 1993) issues and is also leading to increasing incidences of herbicidal resistance among many weed species (Heap 2001). Therefore, efforts to develop alternative means of weed control, which are not only eco-friendly, but also cost effective and biologically robust are required (Duke *et al.* 2002).

Essential oils comprise a complex mix of volatile low-molecular weight isoprenoids, monoterpenes and sequisterpenes and some exhibit herbicidal activity (Angelini *et al.* 2003, Koli *et al.* 1998, Mathews *et al.* 2006, Batish *et al.* 2004, Tworkoski 2002). Pine-oil has been used with some success to control branched broomrape (*Orobanche ramaosa)* seed banks in a South Australia weed eradication program. (Mathews *et al.* 2006).

Carbon addition (sugar, wood chips, sawdust) to soil has been shown to increase microbial populations and CO_2 production as a consequence while also reducing soil nitrogen (Eschen *et al.* 2007, Jonasson *et al.* 1996). Reducing soil nitrogen has been shown to generally be more beneficial to indigenous grassland species compared to exotic species (Eschen *et al.* 2007). Sokoloff (1951) showed that the addition of molasses (carbon) to compacted soil, reduced seed germination of a range of grass and weed species. He postulated that this was due to the anaerobic growth of bacteria that deprived the seeds in the seed bank of oxygen and that the bacteria would also have some direct pathogenic impacts to seeds. Addition of sugar has been shown to reduce germination of chilean needle grass in a grassland seed inundation trial (Faithfull 2012).

This experiment was set up to investigate the effects of the essential oil (pine-oil) and carbon (sugar) and whether the possible fumigant effects of the essential oil could be improved through addition of covers and/or carbon. Mechanical damage to seeds, especially in the seed coat, makes seeds more susceptible to invasion by microbes (Kremer and Spencer 1989). This experiment was designed to use the essential oil (pine-oil) to inflict some damage to the seed coat and the consequent application of carbon (sugar) to provide a pulse (priming) of bacteria to rot down the weed seeds. We selected *Nassella trichotoma* (Nees) Hack. ex Arechav and *Themeda trianda* (R.Br.) Stapf as biassay plants with a view to testing whether pine-oil could be developed as a weed suppressant or as a novel organic bioherbicide. We also wanted to compare a relatively small seeded weedy C_3 exotic grass (*N. trichotoma*) to an indigenous large seeded C_4 grass (*T. trianadra*) and whether there were any differential impacts. This experiment seeks to identify organic techniques for managing weed seed banks. If this technique is effective on *N. trichotoma* it could then be utilised for eradication of other priority weed species seed banks. This would be especially applicable to *Nassella tenuissima,* where the majority of infestations requiring eradication of the seed bank occur in urban situations where organic techniques would be preferable to conventional synthetic herbicides.

This experiment compared 5 rates of pine-oil (0, 0.1, 0.25, 1.0, 2.5 and 5.0% v/v) to 4 rates of carbon applied as sugar or molasses (sugar: 0, 0.1, 0.2, 0.31 kg C m^{-2}; molasses: 0, 0.1, 0.2, 0.31 kg C m^{-2}) and two types of cover (nil and plastic) and two grass species, *N. trichotoma* (a small seeded C_3 exotic weed) and *T. triandra* (a large seeded C_4 indigenous grass). A herbicide treatment (flupropanate, 2 L ha^{-1}) was included to enable comparison to a traditional control technique. Experiments were undertaken as a glass house pot trial with all treatments replicated 3 times in a fully randomised block design. For each treatment pot (175 mm diameter), 500 *N. trichotoma* and 50 *T. trianda* seeds were counted and spread evenly onto the top of a soil commercial potting mix and covered with 1 cm of the same soil potting mix soil and pots were watered a day before treatment to imbibe seeds. Pine-oil was applied using a watering can at a rate of 20 000 L ha^{-1} which was the equivalent rate applied in the South Australian branched broomrape program (Mathews *et al*. 2006). Carbon treatments were applied directly after pine-oil application and were spread evenly across the pot surface. Plastic (100 µm) covers were applied last to the pot surface with the cover edge being pushed into the soil around the pot circumference ensuring an airtight seal. Covers were removed from treatments 1 week after application.

Compared to untreated control treatments, pot trial seed bank experiments show strong interactions between pine-oil and sugar with *N. trichotoma* seed germinations reduced by 98–100%. This is a comparable result to the grass selective herbicide, flupropanate applied as a pre-emergent herbicide and suggests that organic seed bank manipulations with essential oils and carbon could be used as a management tool in appropriate circumstances. Covers increased the effectiveness of pine-oil applications in reducing seed germination by approximately 30% for *N. trichotoma* in all pine-oil treatments. This trend also occurred for *T. triandra* but was not as significant at the 0.5% pine-oil concentration. This highlights the importance of the volatile activity of essential oils and provides some evidence for future investigations to improve essential oil efficacy. The paper discusses how the combined actions of the essential oils and carbon may be depriving the soil seed bank of oxygen resulting in reduced germination as a consequence.

ACKNOWLEDGMENTS

The authors wish to thank the Victorian Government for supplying the necessary resources for undertaking this research and Certified Organics Pty Ltd. for the supply essential oil products tested in this project.

REFERENCES

Angelini, L.G., Carpanese, G., Cioni, P.L., Morelli, I., Macchia, M. and Flamini, G. (2003). Essential oils from Mediterranean Lamiaceae as weed germination inhibitors. *Journal of Agricultural Food Science* 51, 6158-6164.

Batish, D.R., Setia, N., Singh, H.P. and Koli, R.K. (2004). Phytotoxicity of lemon-scented gum oil and its potential use as a bioherbicide. *Crop Protection* 23, 1209-1214.

Cox, C. and Surgan, M. (2006). Unidentified inert ingredients in pesticides: Implications for human health and the environment. *Environmental Health Perspectives*. 114, 1803-1806.

Duke, S.O., Dayan, F.E., Rimando, A.M., Schrader, K.K., Aliotta, G., Oliva, A. and Romagni, J.G. (2002). Chemicals from nature for weed management. *Weed Science* 50, 138–151.

Echen, R., Mortimer, S.R., Lawson, C.S., Edwards, A.R., Brook, A.J., Igual, J.M., Hedlund, K. and Schaffner, U. (2007). Carbon addition alters vegetation composition on ex-arable fields. *Journal of Applied Ecology* 44, 95-104.

Freemark, K.E. and Boutin, C. (1995). Impacts of agricultural herbicide use on terrestrial wildlife in temperate landscapes: A review with special reference to North America. *Agricultural Ecosystems and Environments* 52, 67-91.

Heap, I. (2001). The International Survey of Herbicide Resistant Weeds. Available at www.weedscience.com.

Faithfull, I.G. (2012). 'Biodiversity impacts of Chilean needle grass, *Nassella neesiana* to Australia's indigenous grasslands'. PhD thesis, (Victoria University of Technology).

Jonasson, S., Michelson, A. and Schmidt, K. (1996). Microbial biomass C, N and P in two arctic soils and responses to addition of NPKfertiliser and sugar: implications for plant nutrient uptake. *Oecologia* 106, 507-515.

Kohli, R.K., Batish, D.R. and Singh, H.P. (1998). Eucalyptus oils for the control of *Parthenium* (*Parthenium hysterophorus* L.) *Crop Protection* 17 (2), 199-122.

Kremer, R.J. and Spencer, N.R. (1989). Impact of a seed-feeding insect and microorganisms on velvetleaf (*Abutilon theophrasti*) seed viability. *Weed Science* 37, 211-216.

Mathews, T., Miegel, D. and Hayton, D. (2006). Seed bank and seed bank reduction of *Orobanche ramosa* in South Australia. In, 15th Australian Weeds Conference, (eds) C. Preston, J.H.Watts and N.D. Crossman pp. 626-628 (Weed Management Society of South Australia, Adelaide).

Panetta, F.D. (2004). Seed banks: The bane of the weed eradicator. In, Proceedings of the 14th Australian Weeds Conference. (Wagga Wagga, Australia: Weed Society of New South Wales) pp. 523–526.

Sinden, J., Jones, R., Hester, S., Odom, D., Kalisch, C., James, R. and Cacho, O. (2004). 'The economic impact of weeds in Australia'. CRC for Australian Weed Management Technical Series No 8, 55 pp. (CRC for Australian Weed Management, Waite Campus, University of Adelaide).

Sokoloff, V.P. (1951). Biological control of weeds in compacted soil cultures. *Science* 14, 271-274.
Tworkoski, T. (2002). Herbicide effects of essential oils. *Weed Science* 50, 425-431.
Weisenberger, D.D. (1993). Human health effects of agrichemical use. *Human Pathology* 24(6), 571-576.