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PROCEEDINGS OF THE 12TH INTERNATIONAL HERBAGE SEED GROUP CONFERENCE

Launceston, Tasmania, Australia
16-19 November 2025

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We are pleased to present the proceedings of the 12th International Herbage Seed Conference. The proceedings consist of volunteered oral papers, and peer-reviewed poster abstracts presented at The Tramsheds Function Centre in Launceston, Tasmania. The proceedings papers have been edited for style and clarity, but they have not been peer reviewed.

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ORAL PRESENTATIONS

Emerging and alternative seed crops

Breeding improved cultivars of *Chloris gayana* (Rhodesgrass) 1988-2025: a case history

Donald Loch^{123*}

Abstract

Rhodesgrass (*Chloris gayana* Kunth) is one of the major C₄ forage grasses, native to Africa but now widely sown and naturalised throughout the tropical-subtropical world. Prior to 1990, there were four released cultivars in Australia, all developed from imported ecotypes: 'Pioneer' and 'Katambora' (diploids); and 'Callide' (tetraploid) and 'Samford' (predominantly tetraploid). Rhodesgrass is an outbreeding species amenable to the development of improved cultivars through selection of superior individual plants which are then combined to make a single synthetic cultivar. This paper traces the development of methodology since breeding was initiated in 1988 at the request of, and with financial support from, a commercial seed company. A simplified system of Recurrent Mass Selection (RMS) was developed to speed up generational advancement by spraying out all culled plants and cutting all existing inflorescences from the selections to restrict cross-pollination to the selected group. This approach was extended to the tetraploids in the early 2000s, as well as developing more advanced diploid varieties. Future breeding and marketing strategies are discussed.

Keywords: rhodesgrass, *Chloris gayana*, Recurrent Mass Selection, breeding methodology

Introduction

Rhodesgrass (*Chloris gayana* Kunth), first cultivated in its native Africa around 130-years ago, has since become widely sown throughout the tropical and subtropical world and is now one of the major C₄ forage grasses (Loch et al. 2004). Apart from its primary roles as a pasture, hay, or ley crop, it has been extensively used to stabilise disturbed sites (e.g. mined land), and is widely naturalised in regions and areas where it is well adapted. It is also one of the more salt tolerant C₄ forage grasses, increasingly sown on moderately saline soils or where irrigation waters are marginal.

Rhodesgrass was introduced to Australia in about 1902 by soldiers returning from the Boer War and has since become widely sown and naturalised in the 600-1200 mm rainfall belt from northern New South Wales (NSW) through to central Queensland and is also grown on the Atherton Tableland in north Queensland (Oram 1990; Loch & Harvey 1999). This original introduction - formally registered as the cultivar 'Pioneer' in 1966 - was eventually followed in the late 1950s by imported commercial seed of 'Katambora' and the release of 'Callide' and 'Samford' (both derived from experimental accessions) in the early 1960s. In 1988, breeding to improve the original cultivars for specific uses was commenced at the request of, and with financial support from, Selected Seeds Pty Ltd to develop improved hay varieties. This paper traces the progressive development of breeding methodology and documents the commercial success (or lack thereof) for each of the new cultivars developed and released over the next 27 years through to the present day.

Plant Characteristics Relevant to the Breeding of Improved Rhodesgrass Cultivars

Collectively, the following genetic and physiological attributes of the species *C. gayana* determine what is possible through breeding and the methodology used to develop and maintain new cultivars. Unless specifically noted, this information has been drawn from Loch (1983) and Loch et al. (2004).

Unlike the majority of commercial tropical pasture grasses (around 60%) which reproduce asexually through apomixis, Rhodesgrass is an outbreeding species amenable to the development of improved cultivars through the selection of superior genotypes which are then combined to make a single new synthetic cultivar. Genetically, potential breeding material comes from Rhodesgrass ecotypes (held as gene bank accessions) and the four older Australian cultivars that occur in two different ploidy levels,

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diploid ('Katambora', 'Pioneer') and tetraploid ('Callide', 'Samford') (Oram 1990). Both ploidy levels are sexually outbreeding, but do not cross successfully: only one triploid seedling was recorded in chromosome counts by CSIRO during the 1960s, but it proved to be a very weak plant and soon died (R.J. Jones, pers. comm.). These two basic genetic groups are also physiologically different, most notably in their flowering behaviour. The diploids are day-neutral (DN) flowering throughout the growing season in contrast to the quantitative short-day (SD) tetraploids with restricted periods of strong flowering from late in the growing season through until early in the next growing season. Each tetraploid cultivar or ecotype also comprises a range of flowering behaviour, from a weak SD (earlier flowering) response through to strong, almost obligate, SD response (very late flowering).

Progressive Development and Application of Breeding Methodology

Rhodesgrass breeding in Australia was initiated by the Queensland Department of Primary Industries (QDPI) in 1988 in response to a request and financial support from a commercial seed company. This section of the present paper traces the development of methodology in four stages over the 37 years since that date with the aim of maximising progress in a shorter period of time.

Stage 1 (1988-99). Rhodesgrass breeding started in a project funded by Selected Seeds Pty Ltd develop an improved hay variety for emerging markets in the Middle East. As project leader, I had a good understanding of the species – an important advantage in any breeding programme - courtesy of my Ph.D. work to solve seed production problems with the tetraploids (Loch 1983), but I was not a specialist breeder. To enable repeated cutting cycles for hay production throughout the growing season, the project focused on the diploid group with their unrestricted flowering behaviour. Breeding procedures followed advice from specialist plant breeders. Starting with 1200 'Katambora' and 400 'Pioneer' plants from a range of different locations (plus 17 diploid accessions), the project resulted in the release in 1993 of two new synthetic cultivars: 'Finecut' (based on 10 single-plant clones from 'Katambora') and 'Topcut' (based on 7 single-plant clones from 'Pioneer'). In both cases, selection advanced by only one generation but was spread across four years: firstly, to select a shortlist of 104 superior single plants based on visual assessment of plant habit (erect), leafiness, and early flowering; secondly to intercross these in a randomised array; thirdly, establishing a dry matter production trial to select the final clones that constituted the two new synthetic varieties; and, fourthly, establishing plots of the new synthetics in isolation to produce breeder's seed. While advice was to base new synthetic varieties on at least 30 selected genotypes to minimise the risk of inbreeding depression, no such problems have been encountered then or since (e.g. an alternative 15-clone synthetic designated Capital proved less productive than 'Finecut', so was not released). Rather than develop a time-consuming method based on separating leaf from stem, leafiness was appraised visually consistent with end user practice, but taking care not to mark this down in finer-leaved plants relative to broader-leaved types. We also soon realised that visual assessments of leafiness were positively related to leaf length, further emphasising its importance and ensuring that this rating was an integral selection criterion in all subsequent breeding programmes.

A simplified system of Recurrent Mass Selection (RMS) similar to Glenn Burton's (1994) Recurrent Restricted Phenotypic Selection was developed to speed up generational advancement. All culled plants were killed and all existing inflorescences removed from the remaining plants to restrict cross-pollination to the selected group. An equivalent number of ripe inflorescences were later harvested from each selected plant, bulked, and used to establish the next generation for selection. This approach was first applied to develop a late flowering, low growing, creeping, tight-matted variety based on eight prostrate plants identified in the initial screening experiment. Following four cycles of RMS and seed increase at Biloela (QLD), the finished variety was subsequently registered as 'KP4' (Loch & Roche, 2008) and later marketed under the brand name Tolgar® by another seed company.

Stage 2 (2000-09). The commercial success of 'Finecut' led to further breeding requests, from this point onwards handled through GeneGro Pty Ltd and Stage 2 trial activities managed by Margaret Zorin. RMS was extended to a three-step selection process to incorporate salinity tolerance into agronomic attributes: (1) germination under saline conditions, (2) growth and survival under saline

conditions, and (3) improved agronomic characteristics under non-saline conditions. Tetraploid spaced plant trials were conducted from late summer through to early winter to select early and late flowering phenotypes separately. Three new diploid cultivars developed following four generations of RMS were released through Selected Seeds: 'Reclaimer' (dual purpose hay-grazing) and 'Gulfcut' (finer-textured hay) selected from 'Finecut'; 'Salcut' (hay) from 'Topcut' (Zorin & Loch, 2009a, 2009b, 2009c). Two diploid cultivars – 'KP8' (prostrate for re-vegetation) and 'KG2' (semi-erect grazing; marketed as Endura®) from 'KP4' (Zorin & Loch 2010d, 2010e) – and three tetraploids for grazing – 'Sabre' (early flowering) and 'Toro' (late flowering) from 'Callide' and 'Mariner' (late flowering) selected from 'Samford' (Zorin & Loch 2010a, 2010b, 2010c) – were released through a second seed group with licence agreements later novated to Barenbrug Australia Pty Ltd; with the exception of 'Sabre' (five generations), these synthetics were developed through four generations of RMS.

Stage 3 (2010-16). RMS methodology developed for rhodesgrass was applied to CPI 41192, a predominantly tufted accession of *Digitaria eriantha* Steud., across three generations of selection. A stoloniferous spreading growth habit was increased from 13% to 90% of the outbreeding population together with increased overall leafiness in the final synthetic cultivar 'DMJ-012' (Loch 2025a).

Stage 4 (2017-25). An improved Finecut-type 'FC 5' following five generations of RMS was released to Selected Seeds, increasing the selected plant type from 22.5% to 69% in the F₅ generation (Loch 2024). Selection for leaf to show higher up among the inflorescences - a desirable hay attribute - reduced peduncle length significantly. Six further generations of RMS have been applied to six other breeding families - Reclaimer, Gulfcut, KG, Sabre, Toro and Mariner – and releases are being made from these. 'KG8' is less stoloniferous and spreading and >50 days earlier flowering than 'KG2' (Loch 2025b). Similarly, 'SBR 6' flowered 21 days earlier than 'Sabre' in a PBR growing trial (Loch 2025c).

Seed Production and Marketing

Three criteria underpin a successful outbreeding cultivar: (1) breed a commercially attractive cultivar; (2) maintain the cultivar as close as possible to the original; and (3) promote the cultivar. Because there is no such thing as a stable outbreeding cultivar other than the way it is multiplied, seed harvested from blocks at QDPI's Walkamin Research Facility re-established from breeder's seed every four years (K.G. Cox, pers. comm.) is used to establish commercial seed production paddocks on a pedigree system. From Selected Seeds' records 2007-25 (B. Richards, pers. comm.), the leading varieties 'Finecut' internationally (3,210.1 tonnes of bare [uncoated] seed – an estimated 4,000-4500 t over cultivar life) and 'Reclaimer' domestically (838.6 t) were better promoted than 'Gulfcut' (76.0 t). Production of 'Topcut' ceased in 2011 (47.8 t) as did the market for its parent 'Pioneer' somewhat earlier. Collectively and valued at AU\$12.00 per kg, the farm-gate value of seed marketed from these four cultivars is in excess of AU\$50 million. Based on royalty report data, 2012-25 production now under Barenbrug Australia totalled 183.2 t ('KG2') and 37.7 t ('Mariner') with 'Toro' (19.7 t) and 'Sabre' (1.8 t) no longer in production, for an estimated overall farm-gate value of AU\$2.9 million.

Future Strategies

All Stage 3 and 4 releases will be marketed under trademark brands to reduce the risk of varietal substitution (Loch 2025d) already noted with commercial Finecut post-PBR. With 10 or more RMS generations in some breeding families, further selection will be restricted to smaller numbers of elite plants from each generation to increase selection pressure for faster genetic advancement. After 23 generations of RMS on 'Pensacola' *Paspalum notatum* Flügge, Glenn Burton (pers. comm.) observed plant types from rare genes in the population that had not been evident in his early generations.

References

- Burton GW (1992) Recurrent restricted phenotypic selection. *Plant Breeding Reviews* 9, 101-113.
DOI: <https://doi.org/10.1002/9780470650363.ch5>.
- Loch DS (1983) *Constraints on seed production of Chloris gayana cultivars*. Ph.D. Thesis, University of Queensland, 330 pp.

- Loch DS (2024)[†] Rhodes grass (*Chloris gayana*) 'FC 5'. *Plant Varieties Journal* **37**(2), 83-89.
- Loch DS (2025a) Finger grass (*Digitaria eriantha*) 'DMJ-012'. *Plant Varieties Journal* **38** (in press).
- Loch DS (2025b) Rhodes grass (*Chloris gayana*) 'KG8'. *Plant Varieties Journal* **38** (in press).
- Loch DS (2025c) Rhodes grass (*Chloris gayana*) 'SBR 6'. *Plant Varieties Journal* **38** (in press).
- Loch DS (2025d) Branding with marketing trademarks adds another layer of complexity to traditional botanical taxonomy and nomenclature. *International Turfgrass Society Research Journal* **15** (in press).
- Loch DS and Harvey GL (1997) Rhodes grass (*Chloris gayana*) 'Finecut', 'Nemkat', 'Topcut'. *Plant Varieties Journal* **10**(4), 47-49.
- Loch DS and Harvey GL (1999) *Chloris gayana* in Australia. *In*: D.S Loch & J.E. Ferguson (Eds.) *Forage seed production. 2. Tropical and subtropical species* (pp. 341-349). CAB International, UK.
- Loch DS, Rethman NFG and van Niekerk WA (2004) Rhodesgrass. *In*: L.E. Moser, B.L. Burson & L.E. Sollenberger (Eds.) *Agronomy Monograph 45, Warm-Season (C₄) Grasses* (pp. 833-872). ASA-CSSA-SSA, Madison WI, USA. DOI: <https://doi.org/10.2134/agronmonogr45.c25>
- Loch DS and Roche MB (2008)[†] Rhodes Grass (*Chloris gayana*), 'KP4'. *Plant Varieties Journal* **21**(1), 65, 202-206.
- Oram RN (1990) *Register of Australian Herbage Plant Cultivars (Third Edition)*. CSIRO, Australia.
- Zorin M and Loch DS (2009a)[†] Rhodes Grass (*Chloris gayana*), 'Gulfcut'. *Plant Varieties Journal* **22**(3), 98, 262-267.
- Zorin M and Loch DS (2009b)[†]. Rhodes Grass (*Chloris gayana*), 'Salcut'. *Plant Varieties Journal* **22**(3), 99, 268-273.
- Zorin M and Loch DS (2009c)[†] Rhodes Grass (*Chloris gayana*), 'Reclaimer'. *Plant Varieties Journal* **22**(3), 100, 274-279.
- Loch DS and Zorin M (2010a)[†] Rhodes Grass (*Chloris gayana*), 'Sabre'. *Plant Varieties Journal* **23**(1), 57, 169-176.
- Loch DS and Zorin M (2010b)[†] Rhodes Grass (*Chloris gayana*), 'Mariner'. *Plant Varieties Journal* **23**(1), 58, 177-184.
- Loch DS and Zorin M (2010c)[†] Rhodes Grass (*Chloris gayana*), 'Toro'. *Plant Varieties Journal* **23**(1), 59, 185-192.
- Loch DS and Zorin M (2010d)[†] Rhodes Grass (*Chloris gayana*), 'KP8'. *Plant Varieties Journal* **23**(2), 88, 273-279.
- Loch DS and Zorin M (2010e)[†] Rhodes Grass (*Chloris gayana*), 'KG2'. *Plant Varieties Journal* **23**(2), 89, 280-286.

[†] Online issues of the *Plant Varieties Journal* can be downloaded at <https://www.ipaustralia.gov.au/tools-and-research/professional-resources/ip-rights-journals/plant-varieties-journals>

Cocksfoot seed production: closing date and spring nitrogen response

Phil Rolston^{1*}, Richard Chynoweth², Murray Kelly³ and Bede McCloy⁴

Abstract

Cocksfoot (orchard grass, *Dactylis glomerata*) is a multi-year seed crop that in New Zealand is grazed following seed harvest, through the autumn and winter. The final day of grazing when animals are removed so the crop can undergo reproductive development is referred to as the closing date. Two grazing trials were undertaken, one with cv 'Savvy' and the other with cv 'Kara' each with three different closing dates. Defoliation was at 10 cm height, using a tractor mounted mower. The combined data showed seed yields declined when closing was delayed after early to mid-winter (June or July). Later closing (August) reduced seed head number. The percent seed yield decline was described with the polynomial curve $y = -3.009x^2 + 35.562x - 3.519$ (when x = month).

In five spring nitrogen (N) trials carried out in Canterbury, the optimum N requirements were between 148 and 167 kg N ha⁻¹, including soil mineral N measured to 60 cm soil depth. The trial areas had received 50 kg N ha⁻¹ in autumn. In one trial investigating either 0, 40 or 80 kg N ha⁻¹ (autumn applied), no difference in either head numbers or seed yield was recorded when followed by a standard spring N application of 120 kg N ha⁻¹. Data from two trials was influenced by lodging at higher N application rates. When lodging was controlled by plant growth regulator (PGR) application, the optimum N rates were approximately 14 kg N ha⁻¹ higher. The optimum spring N rates can be estimated as:

$$\text{Spring N application rate (kg ha}^{-1}\text{)} = 165 - \text{soil mineral N (0-60 cm)}$$

The average soil mineral N (NH₄ + NO₂) value was 42 kg N ha⁻¹, thus the optimum applied spring N averaged 123 kg N ha⁻¹.

Keywords: *Dactylis*, orchard grass, grazing, defoliation, seed yield

Introduction

Cocksfoot (orchard grass, *Dactylis glomerata* L.) is a multi-year seed crop that in New Zealand is grazed following seed harvest, through the autumn and early winter. The time when grazing animals are removed so the crop can undergo reproductive development is referred to as the closing date. Grazing removes excess and older leaf material which harbours disease that can infect the new leaf canopy during spring and summer. Grazing is undertaken with either sheep or young cattle, often dairy heifers that are less than one year old and can add additional income earned from the seed crop. Initiation of floral components takes place during spring in response to long or lengthening days, following primary induction that occurred due to short days during the winter. Generally, cocksfoot cultivars require short days (less than about 11 hours) to fulfil primary induction (Heide, 1987). Since daylength is perceived in the leaves, cocksfoot requires leaf area during winter and spring to complete primary and secondary induction. The closing date trials were undertaken to define the reductions in seed yield penalty if the defoliation period was extended beyond early winter.

Spring nitrogen (N) rates are influenced by the available soil mineral N (NO₃ and NH₄) and the rate of applied "bag" N. Nitrogen is an expensive input and excess N causes increased vegetative growth and increases the risk of lodging. Previous trials with straw shortening plant growth regulators on cocksfoot have shown that early lodging reduces seed yield (Rolston et al. 2014). The trials reported were undertaken in seed growers' commercial fields to define optimum spring application N rates.

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Material and Methods

Defoliation-Closing date. Two trials, representing two cultivars ‘Kara’ 2012-13 and ‘Savvy’ 2016-17, evaluated three closing dates, the earliest closing, late June (‘Kara’) and early July (‘Savvy’) was grazed and the subsequent closing dates in August and September were cut at 10 cm height, using a tractor mounted mower. Both trials were undertaken near Methven, New Zealand.

Nitrogen. There were two trials in the Methven area in 2019, both utilising ‘Savvy’ with five spring N rates. In 2020 there were three trials with 9 rates of spring N. In one trial 3 autumn N rates (0, 40 and 80 kg N ha⁻¹) were followed by 120 kg N ha⁻¹ in spring. The cultivar and sites were: ‘Savvy’ (Methven); ‘Safin’ (Wakanui) and ‘DG50’ (Darfield). Soil mineral N (NO₃ + NH₄) for 0-60 cm was assessed at each trial site in late winter. Spring applied N rates ranged from 0 to 250 kg N ha⁻¹.

General. In all trials plots were 2.0 or 2.5 m wide x 10 m in length with four replicates in a randomised block design and inputs other than the nitrogen were managed by the seed grower. Pre-harvest, seed head density was assessed by cutting 2 rows x 1 m (0.6 m²) weighing and counting a subsample containing at least 50 seed heads. At harvest the plots were cut with a plot windrower at ~40% seed moisture and one week later combine harvested when seed moisture was <14%. The field dressed seed was cleaned to a First Generation Seed Certification standard (MPI 2014) and used to calculate the machine dressed seed yield. To compare seed yields across years all the data was adjusted relative to the highest yielding treatment in each trial = 100.

Results and Discussion

Defoliation-Closing date. The best treatment yield for ‘Kara’ was 990 kg ha⁻¹ and for ‘Savvy’ was 730 kg ha⁻¹. Cocksfoot seed crops should be closed from grazing during in June and or July, with early cultivars likely requiring earlier closing compared with later cultivars so they can regrow green leaf area to perceive daylength. Delays in closing (August) reduced the number of seed heads and therefore reduce seed yield (Figure 1.). The percent seed yield decline was described with the polynomial curve $y = -3.009x^2 + 35.562x - 3.519$ (when $x = \text{month}$). Unlike ryegrass and cereals, monitoring the growing point and continuing to graze until it’s vulnerable for removal by grazing animals is detrimental. In cocksfoot the removal of leaf area in late winter and spring can reduce the number of tillers that become reproductive. Thus, you never find reproductive growing points and can keep grazing too long into spring.

Nitrogen. The highest yielding treatments in each trial in 2019 were ‘Savvy’ 540 and 560 kg ha⁻¹ for the two sites and in 2020, ‘Savvy’ 990 kg ha⁻¹, Safin 1330 kg ha⁻¹, ‘DG50’ 830 kg ha⁻¹. The average soil mineral N (0-60 cm) was 42 kg ha⁻¹. In the trial investigating either 0, 40 or 80 kg N ha⁻¹ (autumn applied), no difference in either head numbers or seed yield was recorded when followed by a standard spring N application of 120 kg ha⁻¹.

Seed head density increased as N supply (soil mineral N + applied N) increased up to ~95 kg N ha⁻¹ or when ~50 kg N ha⁻¹ was applied, following which seed head number was stable (in the range of 750-850 heads m⁻²) when additional N was applied. Thus, small amounts of N are required to encourage seed head production with the remainder of the applied N influencing canopy expansion and duration. This is similar to results presented from Oregon where seed head density was increase up to 40 kg N ha⁻¹ (100 lb N ac⁻¹) (Anderson et al. 2017; Anderson et al. 2018).

From five spring N trials grown in Canterbury, the optimum N requirements are between 148 and 167 kg N ha⁻¹ (Figure 1.), including soil mineral N measured to 60 cm. Data from two trials was influenced by lodging at higher N application rates (Figure 1B). When lodging was controlled via PGR application, the optimum N rates were approx. 20 kg N ha⁻¹ higher (Figure 1A and Figure1B.). Thus, the optimum spring N rates can be estimated as:

$$\text{N application rate (kg/ha)} = 165 - \text{soil mineral N (0 - 60 cm)}$$

Assuming a soil mineral N of zero, the economically optimum application rates range from 136-150 kg of applied N. However, in these trials the soil mineral N averaged 42 kg N ha⁻¹, thus, the optimum applied N ranged from 106 – 125 kg ha⁻¹ (Figure 2).

Excess nitrogen was associated with crop lodging in two of the five nitrogen trials where 80-100 kg of applied N ha⁻¹ maximised seed yield. Lodging was associated with yield depressions. If lodging at higher N rates was prevented with plant growth regulator (PGR) the economic optimum N response was increased from 136 to 150 kg N ha⁻¹ (Figure 2B). Applied nitrogen applications should be split throughout the spring at appropriate application rate so not to induce lodging. The PGR program should be considered in conjunction with nitrogen application rates when developing a cocksfoot management plan.

These results are similar to those presented from Oregon where in two of three trials, 112 kg N ha⁻¹ maximised seed yield while in the third season no response to spring applied N was recorded (Anderson et al. 2017; Anderson et al. 2018; Anderson et al. 2019). While data on soil mineral nitrogen testing was not reported, previous work has shown that with the wet Oregon winters the soil mineral N values are near zero (J. Hart, OSU pers comm).

Nitrogen should be applied to match crop demand, particularly in early spring, thus applications should begin when crop growth resumes following winter in August followed by one or two additional applications so not to induce rapid growth and promote lodging.

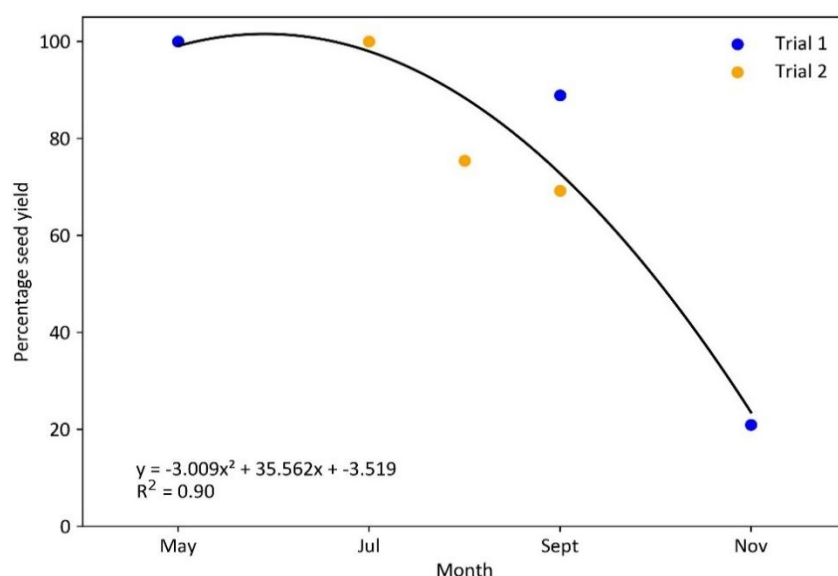


Figure 1. Cocksfoot seed yield from two trials expressed as a percentage of the earliest closing date. Trial 1 was in cultivar ‘Kara’ in 2012/13 and Trial 2 in cultivar ‘Savvy’ in 2016/17 season. Both trials were near Methven.

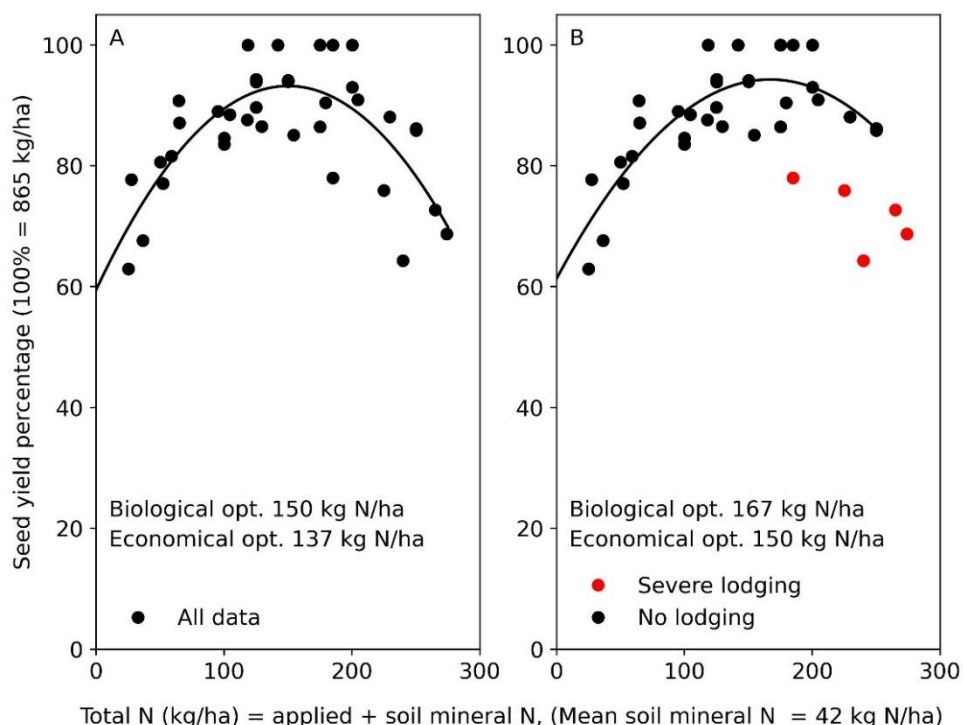


Figure 2. Seed yield of cocksfoot from five trials grown in Canterbury, expressed as a percentage of the maximum yielding treatment (i.e. nitrogen was treated as the limiting factor) for a range of nitrogen treatments. Figure A presents a relationship for all data including, where lodging influenced seed yield, while figure B shows the relationship developed by excluding heavily lodged data (that in red). Both the biological optimum and economical optimum are expressed as ‘total N’ (the sum of applied and soil mineral measured to 60 cm). Biological optimum is the curve maximum, economical optimum calculated using a seed price of \$NZ 5.25/kg and a nitrogen price of \$NZ 1.75/kg N⁻¹.

Acknowledgements

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References

- Anderson NP, Chastain TG, Moore AD and Garbacik CJ (2017) Spring-applied nitrogen and plant growth regulator effects on orchardgrass seed yield. In N. Anderson, A. Hulting, and D. Walenta (Eds.), *Seed Production Research Report*: Oregon State University, Ext/CrS 154.
- Anderson NP, Chastain TG, Moore AD and Garbacik CJ (2018) Spring-applied nitrogen and plant growth regulator effects on seed yield of second-year orchardgrass. In N. Anderson, A. Hulting, and D. Walenta (Eds.), *Seed Production Research Report*: Oregon State University, Ext/CrS 160.
- Anderson NP, Chastain TG, Moore AD, Garbacik CJ and Donovan BC (2019) Spring-applied nitrogen and plant growth regulator effects on seed yield of third-year orchardgrass. In N. Anderson, A. Hulting, D. Walenta, and C. Mallory-Smith (Eds.), *Seed Production Research Report*. (pp. 10-12): Oregon State University, Ext/CrS 162.
- Heide OM (1987) Photoperiodic control of flowering in *Dactylis glomerata*, a true short-long-day plant. *Physiologia Plantarum* **70**, 523-529. <https://doi.org/10.1111/j.1399-3054.1987.tb02853.x>
- MPI (2014) Ministry for Primary Industries Seed Varietal & Phytosanitary Certification Programme. Appendix 1. Seed Field Production Standards 2014. accessed 6 January 2023 from <https://www.mpi.govt.nz/dmsdocument/115-Export-Certification-Standard-Appendix-1-Seed-Field-Production-Standards>.

Rolston P, Chynoweth R, Kelly M, McCloy B and Trethewey J (2014) Seed yield response of cocksfoot (*Dactylis glomerata* L.) to stem shortening plant growth regulators. *New Zealand Journal of Agriculture Research*. **57**, 332-341.

Potential of using grain harvesters to harvest annual medic seeds

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Abstract

Annual medics (*Medicago* spp.) are widely grown on neutral and alkaline soils in southern Australia. When medic pods ripen, they fall to the ground and the seed industry use Horwood Bagshaw Clover Harvesters (HB) to suction harvest the pods and thresh the seeds. HB are also used to harvest subterranean clover, and recent research has highlighted issues with HB including high amounts of soil in the seed sample, slow operational speed (0.3 ha hour⁻¹), soil degradation, pre-treatments required to prepare for harvest machines, as well as their age (at least 30-years old) and reliability. The number of medic seed growers has reduced by 60% in the last quarter a century and seed companies state they are unable to get enough growers to meet demand. Without a new efficient method of producing medic seed the medic seed industry is at risk of collapsing and key pasture legumes for alkaline soils will be lost.

We conducted a literature search on harvesting annual medics seeds or pods, and then met with medic seed growers and seed companies to discuss potential new ways to harvest medic seeds. Harvesting medic pods with a grain harvester followed by threshing with a stationary thresher has potential to overcome the issues from using HB, greatly reduce labour inputs, and introduce modern machinery. Three medic cultivars have been bred using mutation breeding to hold pods better. These are reported as being able to be harvest pods from naturally senesced medic plants. Plants of non-pod holding medic cultivars can be harvested with a grain harvester if they are desiccated early. Harvested pods can be threshed with a stationary HB, but seed growers would like a better method developed to thresh pods. Pods of the burr medic cultivar Scimitar can be stored for two summers and sown in February and March. Seed growers report that Cavalier burr medic is the easiest cultivar to harvest with a HB while the strand medic cultivar Seraph is the most difficult due to its small pod and seed size. Potential exists for medic breeders to develop new cultivars with traits of pod holding, large seed size, and seed softening like Scimitar. The medic seed industry has a low number of growers and further research into harvest and threshing of medic pods is required. This together with medic breeding work has the potential to make harvesting medic seed easier and meet the demand for medic seeds.

Keywords: annual medics, seed production, Horwood Bagshaw Clover Harvester, desiccation

Introduction

Annual medics are key pasture legume species for alkaline soils in southern Australia and have been sown over 25 Mha (Nichols et al. 2012). Annual medics provide high quality and high protein feed to livestock and fix nitrogen which benefits following grain crops. Annual medics and subterranean clover seeds are harvested with a Horwood Bagshaw Clover Harvester (HB) which suction harvests pods, threshes pods and sieves out seed (Didar 2003; Moss et al. 2021). Moss et al. (2021) report that the seed industry faces a serious risk from the current practice of using HB which causes soil degradation, is expensive to operate, slow (0.3 ha hour⁻¹) and a labour-intensive process, with poor reliability and maintainability of harvesters that are now at least 30 years old. Seed companies report difficulty in finding medic seed growers and are unable to meet the demand for medic seed. Development of new ways to harvest medic seeds with modern equipment has the potential of saving the medic seed industry from collapse. We reviewed the literature for harvesting medic seeds/pods, and then held a meeting with medic seed growers and seed companies and discussed potential ways to improve the harvestability of medic seeds.

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Materials and Methods

We reviewed the literature on harvesting medic seeds and pods and invited medic seed growers from South Australia and Victoria, and seed companies to a workshop at Turretfield (SA) on harvesting medic seed. A PowerPoint on harvesting medic seeds and pods was presented and allowed for discussion during and after the PowerPoint presentation.

Results and Discussion

Medic plants set seeds inside pods which become hard as they mature and readily fall to the ground when they ripen. Seed growers rake the trash off, loosen pods with light tillage and suction harvest and thresh pods with a HB (Didar 2003). The number of medic growers has reduced by 60% in the last 25 years. The harvesting operation is slow at 0.3 ha hour⁻¹ which is much slower than combine harvesters at 8.3 ha hour⁻¹ (Moss et al. 2021). Seed growers readily agreed that harvesting medic seeds is slow, monotonous, and labour intensive. One grower stated that two people are allocated to harvesting lentils (one operating the harvester and one collecting and moving the seed) whereas when harvesting medic seed, five people are allocated due to the preparation treatments required. In high yielding years (e.g. 1500 kg ha⁻¹) ground speed is 0.8 km hour⁻¹.

Harvesting medic pods. Attempts have been made at harvesting medic pods before they fall. Mutation breeding using gamma radiation was used to develop the strand medic (*M. littoralis*) 'Jaguar' and the trait was transferred into barrel medic (*M. truncatula*) cultivars to develop cultivars 'Lynx' and 'Cheetah' (Lake and Drewry, 2005). A patent was obtained for these three cultivars and the method used to develop them, which has now expired. Pristine Forage Technologies (2009) show photographs of pods holding on senesced plants and harvesting with a grain harvester, but they do not present any data on pod yields or report on what to do with the harvested pods. Ballard (2010) set up eight five-hectare paddocks of different medic species, but was unable to harvest any with a grain harvester, but did harvest 10,000 kg pods of 'Santiago' burr medic (*M. polymorpha*) from a commercial farm (60-70% of total pods). One seed grower reported that he harvests medic pods of the barrel medic cultivars 'Lynx' (pod holding) and 'Penfield' (spineless).

Medic seeds are desiccant tolerant and have high vigour 400 growing degree days (GDD) after flowering and pods fall 900 GDD (Gallardo et al. 2003). Peck et al. (2022, 2023) noted when peak flowering started and finished and tracked GDD to target desiccation when pods from many flowers are 400-900 GDD old. Desiccation day was confirmed by observing many grey pods on medic plants, some pod fall, and a forecast of at least four fine days with light winds. At Kingsford (close to the main medic seed growing area) 1000-2500 kg pods ha⁻¹ of barrel and strand medics and 3200 kg pods ha⁻¹ of burr medic were harvested in a wet year, and 790-1080 kg pods ha⁻¹ of barrel and strand medic in a dry year. Seed growers were interested in this approach and in future research into windrowing medic plants to harvest pods. Some growers report they would be prepared to dry pods with a grain drier if necessary, and that early harvest would avoid seed loss that can occur with summer rainfall.

Threshing pods. The seed growers report that medic pods are too hard for a combine harvester to thresh and would need to be threshed with a stationary thresher. Ballard (2010) report on a series of modifications to a HB that allowed pods harvested with a grain harvester to thresh pods at a rate of 1000 kg pods hour⁻¹ with a clean out rate of 83% compared to 60% with a HB. The seed grower who harvested pods with a grain harvester threshed pods with a stationary HB but had problems with build-up of thrash. He now contracts someone who uses a thresher that was made to thresh Sulla (*Hedysarum coronarium*) pods. Pods thresh easier when they are warm (Ballard 2010), and seed growers think it would be worthwhile to investigate if warming pods up with a grain drier will assist threshing of pods. Peck et al. (2024) conducted seed softening studies to determine if pods stored for two or more summers could be sown in February or March and have enough softening for sowing rates to be <50 kg pods ha⁻¹. They found that only the burr medic 'Scimitar' achieved this (30 kg pods ha⁻¹). This suggests that 'Scimitar' pods could be harvested, cleaned and sold in a similar way as occurs with French serradella (*Ornithopus sativus*) pod segments.

Preferred cultivars. Seed growers were asked which cultivar they like to harvest and which they do not like to harvest. It was quickly agreed that the burr medic cultivar 'Cavalier' was the easiest to harvest and that the strand medic cv. 'Seraph' (only current strand medic cultivar) was the worst. Growers report that 'Cavalier' has large pods which are easy to suck up, large seeds (~4 mg) which makes it easy to thresh and sieve. 'Cavalier' also has high demand from seed companies. Peck and Howie (2012) report that burr medics have ~1.4x the yield of barrel and strand medics. The small pod size of 'Seraph' makes it difficult for pods to be sucked up, which results in more soil being sucked up, and its small seed size makes it difficult to sieve. 'Seraph' seed size is 1.25 x cv. 'Angel' (IP Australia), but the seed industry would benefit from its seed size being increased further.

Breeding solutions. Several breeding solutions are possible that can assist the seed industry. The pod holding patent has now expired (IP Australia) which means that there is no restriction on developing more pod holding cultivars. Strand medic is an important species for sandy loams in low rainfall areas, and it is in danger of not being grown by seed growers. The small seed size and pod size are the main concern. While 'Seraph' has 1.25x larger seed than prior strand medic cultivars, it still has smaller seed than current cultivars of other medic species. The Australian Pastures Genebank (APG) has over 250 accessions with seed size >3.5 mg. This indicates that a larger seeded strand medic cultivar could be developed which is expected to aid harvesting. Peck and Hill (2024) report that 'Scimitar' pods can be stored for two summers and sown in February and March. This suggests that APG accession of barrel and strand medics could be screened for seed softening patterns like 'Scimitar'. Accessions could be also screened to determine if any soften much more readily in the dark as occurs for several serradella cultivars, which forms the basis of early autumn sowing.

Conclusion

The medic seed industry harvest seeds with HB and is unable to meet the demand for medic seed. Cultivars with improved ability to hold pods along with early desiccation or swathing provide potential for harvest of medic pods with grain harvesters. This needs to be tested at scale. Research is required into the best way to thresh the pods with a stationary thresher. Potential exists for medic breeders to breed cultivars that assist seed harvest. Potential traits to breed for include, pod holding, larger seed size, and seed softening characteristics that allow February- March sowing of pods that have been stored for 1-2 summers. Further research into pod harvesting, pod threshing, and plant breeding has the potential to expand the size of the medic seed industry and hence the size of medic pastures that can be sown.

Acknowledgements

We thank the medic seed growers and seed company people who attended the meeting on new ways to harvest medics seeds. We thank Nick Koch from Seed Services Australia (SARDI) for providing contact details of medic growers who produce certified seed. Work on desiccation to assist pod harvesting was funded by the South Australian Grains Industry trust (SAGIT).

References

- Ballard L (2010) Improvements to Harvesting and Thrashing Medic and Biserrula Pods RIRDC Publication No. 10/001
- Didar AR (2003) Theoretical analysis of harvest collection of medic pods by pneumatic means. PhD thesis, The University of Adelaide, SA
- Lake AWH and Drewry RE (2005) 2005200437 Improved pasture, forage and seed production technology through pod and leaf retention on annuals of the *Medicago* genus (annual medics). <https://ipsearch.ipaustralia.gov.au/patents/2005200437>
- Gallardo K, Le Signour C, Vandekerckhove J, Thompson RD and Burstin J (2003) Proteomics of *Medicago truncatula* seed development establishes the time frame of diverse metabolic processes related to reserve accumulation. *Plant Physiology* **133**, 664-682

- IP Australia (2025) Plant breeders rights database. IP Australia, Australian Government, Canberra, ACT, Australia. Available at www.ipaustralia.gov.au/get-the-right-ip/plant-breeders-rights
- Pristine Forage Technologies (2009) Pod Holding (ph) Medics <https://pristineforage.com.au/pod.htm>
- Nichols PGH, Revell CK, Humphries AW, Howie JH, Hall EJ, Sandral GA, Ghamkhar K and Harris CA (2012) Temperate pasture legumes in Australia – their history, current use, and prospects. *Crop and Pasture Science* **63**, 691-725.
- Moss WM, Guzzomi AL, Foster KJ, Ryan MH and Nichols PGH (2021) Harvesting subterranean clover seed in southern Australia – current technology, practices and issues. *Crop and Pasture Science* **72**, 223-235.
- Peck DM and Howie JH (2012) Development of an early season barrel medic (*Medicago truncatula* Gaertn.) with tolerance to sulfonylurea herbicide residues. *Crop and Pasture Science* **B**, 866-874.
- Peck D, Webb J, Hill J, Rowe T and Watzke E (2022) Harvesting annual medic pods Eyre Peninsula Farming Systems Summary 2022, 148-151
https://pir.sa.gov.au/__data/assets/pdf_file/0007/432619/epfs-summary-2022.pdf
- Peck D and Hill J (2023) Harvesting and use of annual medic pod on farm. Eyre Peninsula Farming Systems Summary 2023, 151-154 https://pir.sa.gov.au/__data/assets/pdf_file/0009/465939/epfs-summary-2023.pdf#:~:text=Department%20of%20Primary%20Industries%20and%20Regions%20%28PIRSA%29%2C%20I,carried%20out%20at%20the%20Minnipa%20Agricultural%20Centre%20%28MAC%29.
- Peck D and Hill J (2024) Sowing of annual medic pods harvested with a grain harvester. Eyre Peninsula Farming Systems Summary 2024, 141-144
https://pir.sa.gov.au/__data/assets/pdf_file/0011/477227/2024_EPFS_BOOK_FINAL.pdf

ORAL PRESENTATIONS

**Seed production in a challenging
and changing environment**

A new low coumarin, multi-stem, annual *Melilotus albus* cultivar for Australia

Stan Paynter^{1*}, Gerald Smith² and Donald Loch³

Abstract

Multiple plant attributes (low coumarin, multi-stem growth habit, annual life cycle) were combined in experimental lines of *Melilotus albus* Medik (sweet clover) following three cycles of hybridisation, progeny testing and selection in a seven-year breeding programme by Texas A&M AgriLife Research at Overton, TX, USA. The breeding line TX-SC-909 was selected for evaluation in Australia by Selected Seeds Pty Ltd. Its breeding history (2001-07), introduction to and evaluation in Australia (2018-25), and impending commercialisation under Plant Breeder's Rights as 'TX-909' (2024-25) are described.

Keywords: *Melilotus albus*, low coumarin, multi-stem habit, annual life cycle, legume

Introduction

Melilotus albus Medik. (sweet clover) is a drought-tolerant legume species of Eurasian origin. It is now grown worldwide, mainly as a forage plant as well as a food source for honeybees under subtropical to cold temperate conditions, especially in North America where it is well adapted to the alkaline and neutral soils of the Great Plains (Turkington et al. 1978; Popay 2021). *M. albus* includes both biennial (second year flowering) and annual genotypes, with biennial cultivars (e.g. 'Denta', 'Polara') preferred in colder regions (northern USA and Canada) and annuals in southern USA.

M. albus also varies in terms of plant coumarin content. This phytochemical is a precursor to dicoumarol, which can cause a toxic bleeding disease in animals, hence low coumarin genotypes are preferred. Prior to the initiation of a breeding programme at Overton, TX, in the early 2000s, the available low coumarin cultivars were all biennial, northern types. This breeding programme led to the release by Smith et al. (2017) of 'Silver River', a rust-resistant, annual sweet clover adapted to southern and central Texas. Separately, the first author identified for further evaluation in Australia a short, multi-branching, low coumarin genotype designated TX-SC-909 among other experimental lines developed through the Overton breeding programme. The present paper describes the multiple breeding steps to develop TX-SC-909, its import to Australia and early testing under subtropical conditions at Gatton (Queensland, QLD) by Selected Seeds Pty Ltd, and its description for Australian Plant Breeder's Rights (PBR) as a new cultivar 'TX-909'.

Breeding of 'TX-909'

The objective of the breeding programme at Overton (TX, USA) was to create a low coumarin and multi-stem annual sweet clover through a three-way cross of 'Emerald', 'Denta', and TX-4798.

Background. The inheritance of low coumarin in sweet clover is determined by the homozygous recessive form (*cu cu*) of a single gene (Goplen et al. 1957), but current low coumarin cultivars are all biennial northern types. Biennial growth habit in sweet clover is also controlled at a single locus (*a a* = biennial and *A_* = annual; Smith, 1927). The ability of annual sweet clover to develop a multi-stemmed crown, similar to alfalfa (*Medicago sativa*), is also under genetic control (Hartwig 1942). This multi-stem type is often referred to as a dwarf plant where *D_* = single/few stems and tall growth and *d d* = shorter, multi-stem growth.

Initial Hybridisations. Hand pollinations and bee cage crosses were made in 2001 between 'Denta' (low coumarin biennial with the status of each plant confirmed by testing in advance) and 'Emerald' (high coumarin, multi-stemmed annual) sweet clover. The high coumarin allele (*Cu*) carried by 'Emerald' was

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used as a genetic marker to identify true hybrids. Seed was harvested only from the 'Denta' plants, germinated, and grown in the greenhouse for 60 days before testing for coumarin content to determine actual hybrids by the presence of high coumarin using a rapid assay (Gorz & Haskins 1958). From all the hybrid combinations, nine 'Denta' parents and 14 'Emerald' parents contributed to this hybrid population. From 338 hand crosses, 36 hybrids were identified, together with 47 hybrids from bee cage crosses.

F₂ Production and Evaluation. All hybrids were self-pollinated (using a hand rolling technique) in the greenhouse to produce about 240,000 F₂ seeds during spring-summer in 2002. In September 2002, 1,500 F₂ seeds from each of the seven maturity groups in the 'Emerald' parent were germinated and transplanted to the greenhouse, giving a total of 10,500 plants for evaluation and simultaneous screening for low coumarin (*cu cu*), fine stem or multiple stem trait (*ff*), and annual growth habit (A A). 'Emerald' and 'Denta' seedlings were also planted for use as checks.

F₃ Family Evaluations. Progeny testing of 143 F₃ families plus three checks (two entries of *M. albus* 'Denta' and the cultivar 'Hubam') to identify those that bred true for the annual trait was conducted at Thrall, TX (Oct 2003 - Mar 2004). Entries were arranged in a randomised complete block design with two replications and planted in 1.83 m rows (1.22 m between rows). Hand scarified seeds were tested for germination and planted at 40 seeds per row. Evaluation of the low coumarin F₃ families at Thrall started in early March 2004. Notes were made of plant height, forage potential and stand.

Additional Crosses. Seed of three elite sweet clover F₃ lines identified at Thrall (2022-10, 2024-21 and 2022-32) were started in the greenhouse in Oct 2004. In May 2005, a low coumarin elite F₃ line (2024-21) was used as the female parent in a cross with a high coumarin, single stemmed breeding line TX-4798 which was resistant to sweet clover rust (*Uromyces striatus* Schroet.). Seedlings from the progeny of the hybridised 2024-21 plants were tested for coumarin in 2006, with high coumarin identifying a true hybrid. The 16 hybrids identified were self-pollinated in the greenhouse using the hand-rolling technique. A bulk of the 19 F₂ populations was grown in the greenhouse in 2007 and screened for low coumarin and for the multi-stem trait. Forty-eight plants with both low coumarin (*cu cu*) and the multi-stem growth trait (*ff*) were identified and were intermated using native/naturalised bee populations. Seeds from all 48 plants were bulked and designated TX-SC-909 (pedigree: Emerald/Denta//TX-4798). Seed of TX-SC-909 was increased at Overton from 2008.

Evaluation in Australia

TX-SC-909 was introduced to Australia in 2019. The 35 plants grown in post-entry quarantine provided a small quantity of seeds to initiate seed build-up and trialling under subtropical conditions in the Lockyer district (QLD) by Selected Seeds Pty Ltd. The first seed bulk-up was planted in May 2020 and successfully harvested in January 2021. Planting of a second seed increase was delayed until June 2021 by wet autumn conditions, but grew successfully until destroyed by a hailstorm in October 2021. It was noted that early growth from the autumn plantings was uneven and not competitive with difficult-to-control weeds. As sweet clover in the US has typically been an autumn-planted crop that germinates and remains dormant until spring, it was decided to try planting in September to coincide with the spring growth spurt and have a shorter cropping cycle with a slightly later harvest. In 2022, crops were planted in both autumn and spring for comparative purposes, but both were badly damaged by high summer rainfall during flowering and seed maturation. Thereafter, only spring plantings have been made. The 2023 spring planting was destroyed by flooding, but the 2024 crop was successfully harvested and the 2025 spring planting is developing well.

Description of 'TX-909' for PBR (Loch, 2025)

Materials and Methods. A comparative growing trial to describe 'TX-909' was conducted on a red volcanic ferrosol soil at Cleveland, QLD (Latitude 27°31' S, Longitude 153°15' E) with supplementary irrigation as required over the experimental period. The comparator 'Jota' (a low coumarin annual described by Trigg 2004) was selected as the closest variety of common knowledge.

Mini-sward rows of three treatments - 'TX-909' (from 2 generations) and 'Jota' - were arranged in six randomised blocks with ten plants per 1.5 m mini-sward plot planted at 15 cm spacing. Seed was sown in crackpot tubes (7 September 2024) and transplanted into the field on 22 October 2024. A proprietary blended fertiliser CK55(S) was applied to give 40 kg N, 44 kg P, 37 kg K, and 20 kg S per hectare; and S-metolachlor (Dual Gold®) at 2 L ha⁻¹ was applied for weed control. Group AL inoculant (RRI128) was applied as a slurry to emerging seedlings in crackpots on 13 September 2024.

Days to first flowering were recorded for each plot. Leaf and inflorescence measurements (five per plot) were made on 18 December 2024, respectively from ± 10 th and ± 5 th visible nodes back from the main stem tip. Main stem length (\equiv plant height) and node numbers (five plants sampled per plot) were determined (16 January 2025). Samples of ripe pods from each plot were, sun-dried, threshed using sanding sheets, screened through a 1.18 mm sieve and aspirated before weighing 200-seed subsamples to determine seed size. Analyses of variance (ANOVAs) were conducted with GenStat.

Results. As shown pictorially in Figure 1 and documented in Table 1, 'TX-909' is a shorter and denser plant than 'Jota' with fewer main stem nodes and shorter internodes. Both cultivars are comparable in terms of flowering date, inflorescence length, and leaf shape, but 'TX-909' has significantly smaller leaves with crenulate, rather than serrate, margins. 'TX-909' also produces smaller seeds that are yellowish-green colour compared with 'Jota' (yellow seeds).

Table 1. Comparative developmental and morphological data for *Melilotus albus* from PBR growing trial.

Attribute	'TX-909'	'Jota'	LSD ($P = 0.05$)
Sowing to first flowering (days)	72.7	72.3	2.3
Height of main stem (cm)	97.9	167.4	11.4
No. of main stem nodes	30.1	35.3	2.1
Mean internode length on main stem (cm)	3.3	4.8	0.3
Terminal leaflet length (mm)	31.8	35.5	1.7
Terminal leaflet width (mm)	12.5	13.9	1.3
Terminal leaflet length:width ratio	2.57	2.57	0.23
Petiolule length (mm)	6.9	8.9	1.1
Lateral leaflet length (mm)	27.2	30.8	1.5
Lateral leaflet width (mm)	10.2	11.7	1.1
Lateral leaflet length:width ratio	2.68	2.66	0.22
Leaflet margins	Crenulate	Serrate	
Inflorescence length (mm)	157.1	163.5	13.5
Number of seeds per pod	1	1	
Ripe pod colour	Black	Dark brown	
Seed colour (RHS Colour Chart)	Yellowish-green (146B-C)	Yellow (162A)	
1000-seed weight (g)	2.15	2.41	0.09

Looking to the future

TX-SC-909 – the first low coumarin, multi-stem, annual sweet clover - has been registered in Australia under the PBR cultivar name 'TX-909' and will be marketed under the trademark Jade SC®.



Figure 1. 'TX-909' with comparator 'Jota' showing differences in plant height, density and branching.

References

- Goplen BP, Greenshields JER and Baenziger H (1957) The inheritance of coumarin in sweet clover. *Canadian Journal of Botany* **35**, 583-593.
- Gorz HJ and Haskins FA (1958) Rapid tests for free and bound coumarin in sweetclover. *Agronomy Journal* **50**, 211-214.
- Hartwig EE (1942) Inheritance of growth habit, cotyledon color, and cup-leaf in *Melilotus alba*. *Journal of the American Society of Agronomy* **34**, 160-166.
- Loch DS (2025) Sweet clover (*Melilotus albus*) 'TX-909'. *Plant Varieties Journal (Online)* **38** (in press).
- Popay I (2021). *Melilotus albus* (honey clover). *CABI Compendium Online* 33693. DOI: <https://doi.org/10.1079/cabicompendium.33693>.
- Smith GR, Evers GW, Ocumpaugh WR, Forbes TDA, Ong K and Foster-Malone J (2017) Registration of 'Silver River' sweetclover. *Journal of Plant Registrations* **11**, 112-115. DOI: <https://doi.org/10.3198/jpr2016.11.0066crc>.
- Smith HB (1927) Annual versus biennial growth habit and its inheritance in *Melilotus alba*. *American Journal of Botany* **14**, 129-146.
- Trigg P (2004) *Melilotus albus* (sweet clover) 'Jota'. *Plant Varieties Journal* **17**(3), 128–129.
- Turkington RA, Cavers PB and Rempel E (1978) The biology of Canadian weeds. 29. *Melilotus alba* Desr. and *M. officinalis* (L.) Lam. *Canadian Journal of Plant Science* **58**, 523-537.

Insecticide resistance management strategies for the clover seed weevil (*Tychius picirostris*) in Oregon white clover seed production

Navneet Kaur^{1*}, Seth J Dorman², Christy Tanner³ and Nicole Anderson⁴

Abstract

Tychius picirostris Fabricius (Coleoptera: Curculionidae), the clover seed weevil (CSW), is a major pest of white clover (*Trifolium repens* L.) seed crops in Oregon, USA, where larval feeding during seed development reduces yield and quality, causing substantial economic losses. For decades, CSW management has relied on bifenthrin (IRAC Group 3A), but since 2017 growers have reported reduced efficacy, prompting concern of resistance. Laboratory bioassays in 2022-2023 confirmed very high resistance to bifenthrin (RR50 = 178-726) and moderate resistance to malathion (Group 1B; RR50 = 7.8-32.8), underscoring the need for alternative chemistries and insecticide resistance management (IRM) guidelines. From 2022 to 2024, on-farm insecticide efficacy trials were conducted in commercial white clover seed fields in western Oregon. Early-season (pre-bloom or PB) application using contact insecticides (malathion, isocycloseram [Group 30], indoxacarb [Group 22]) targeted adults, either alone or in sequence with mid-season (full bloom or FB) application of systemic insecticides (chlorantraniliprole and cyantraniliprole [Group 28]) targeting larvae. Adult abundance was monitored with 20-sweep net samples, and larval densities were estimated from 30 inflorescences per plot extracted with Berlese funnels. Across sites and years, isocycloseram consistently suppressed adult populations, cyantraniliprole reduced larval densities, while indoxacarb showed variable performance. Although seed yields did not differ significantly among treatments, yet efficacy data supported product registration in Oregon and highlighted the value of chemical rotation plans for resistance management. Based on these findings, we recommend discontinuing bifenthrin and adopting an integrated resistance management (IRM) program that applies contact insecticides during spring adult migration when ≥ 2 weevils per sweep are detected and systemic insecticides during full bloom when ≥ 3 per 30 inflorescences are observed.

Keywords: *Tychius picirostris*, white clover seed, insecticide resistance management, rotational strategies, resistance mechanisms

Introduction

The clover seed weevil (CSW), *Tychius picirostris* Fabricius (Coleoptera: Curculionidae), is a persistent pest of white clover (*Trifolium repens* L.) seed crops in Oregon, which produces more than 90% of U.S. white clover seed. Adult feeding occurs on developing buds and flowers, but the greatest injury is caused by larvae that consume developing seeds within florets. Larval feeding directly reduces seed yield and quality, resulting in significant economic losses for growers.

Historically, CSW management has relied almost exclusively on foliar applications of bifenthrin, a pyrethroid insecticide (IRAC Group 3A). While initially effective, repeated reliance on bifenthrin created strong selection pressure for resistance. Laboratory bioassays conducted by Tiwari et al. (2023) documented very high resistance to bifenthrin (178- to 725-fold) and moderate resistance to malathion (9.2- to 32.8-fold) in Oregon field populations compared to a susceptible strain. Although chlorantraniliprole showed no evidence of resistance, its efficacy was limited under laboratory conditions (Tiwari et al. 2023). These results highlight the risks of single-chemistry reliance to avoid selection pressure and the urgent need for resistance management.

Since 2022, our project team has actively evaluated newer insecticide chemistries and alternative modes of action to identify viable rotational products (Tiwari et al. 2024). Incorporating these products

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into chemical rotation is a core strategy for reducing reliance on the broad-spectrum insecticides and lowering selection pressure by incorporating diverse modes of actions (MoAs) (Kaur et al. 2024). This integrated approach provides the foundation for developing practical IRM guidelines for growers.

Materials and Methods

Two commercial white clover (*Trifolium repens* L.) seed fields, Site 1 (44.3385, -123.1935) and Site 2 (44.4595, -123.1792), were selected in 2024 based on clover seed weevil (CSW), *Tychius picrostris*, abundance. Trials were arranged in a randomised complete block design with seven insecticide treatments (Table 1) and one untreated control, replicated three times at each location. Two application timings were evaluated: pre-bloom (PB) sprays applied on 29 May at Site 1 and 5 June at Site 2 (<10% flowering), and full-bloom (FB) sprays applied on 20 June at both sites (~100% bloom with 10% brown-down) (Table 1). Plots measured 8.8 m × 91.4 m to accommodate grower swather width. Applications were made using an ATV-mounted CO₂-pressurised boom sprayer with TeeJet XR11002VS nozzles, calibrated to deliver 140 L ha⁻¹ at 138 kPa. A methylated seed oil surfactant (Wilbur-Ellis Super Spread® MSO, 1% v/v) was included in all treatments.

Table 1. Details of insecticides including trade name, active ingredient (AI), label rate, application timing and crop growth stage of white clover seed crop.

Trade Name	AI (IRAC Group)	Kg (ai) ha ⁻¹	Date of Application	Crop Growth Stage
Control	—	—	—	—
Steward	Indoxacarb (22)	0.83	29 May 2024	Pre-bloom
Vantacor	Chlorantraniliprole (28)	0.18	20 June 2024	Full-bloom
A21377X	Isocycloseram (30)	0.15	29 May 2024	Pre-bloom
Vantacor	Chlorantraniliprole (28)	0.18	20 June 2024	Full-bloom
Malathion	Malathion (1B)	1.46	29 May 2024	Pre-bloom
A21377X	Isocycloseram (30)	0.15	29 May 2024	Pre-bloom
Exirel	Cyantraniliprole (28)	1.50	20 June 2024	Full-bloom

Adult CSW abundance was monitored 7 and 14 days after treatment (DAT) for PB applications, and 7, 14, and 21 DAT for FB applications. At each sampling date, 20 sweeps were taken per plot using a 38 cm heavy canvas sweep net, rotating 90° between sweeps. For larval abundance, four 30 cm PVC quadrats were tossed into each plot, and 30 inflorescences were randomly collected in plastic bags. Inflorescences were placed in Berlese funnels under 40 W lamps, and larvae were collected in vials of 70% ethanol.

All analyses were conducted in R (v4.3.1; R Core Team) using the *lme4* package (Bates et al. 2015), *emmeans* (Lenth 2023), and *ggplot2* (Wickham 2016). Adult and larval abundance data were fitted to a negative binomial distribution and analysed using generalised linear mixed models with treatment and DAT as fixed effects. Plot replicate and spray timing were included as random intercepts, and DAT was modelled as a random slope.

Results and Discussion

No phytotoxicity was observed in any treatment plots at either site. Significant adult suppression ($P = 0.0006$) (Table 2) was also observed in the plots treated with indoxacarb, malathion, and isocycloseram compared to untreated control at Site 1. On the other hand, significant adult suppression was only observed in isocycloseram-treated plots compared to untreated control plots at Site 2. No effect of indoxacarb on adult suppression was observed at Site 2. A 56% reduction of adult weevil abundance ($P = 0.044$) was present in isocycloseram-treated plots than the untreated control at Site 2.

Table 2. Adult CSW abundance after pre-bloom application of a single product (7 and 14 DAT evaluation) at Sites 1 and 2

Treatment AI (trade name)	Site 1	Site 2
	Overall mean adult weevils per 20 sweep samples	
Control	44.17 a	65.33 a
Indoxacarb (Steward)	33.25 b	55.17 a
Malathion (Aquamul 8)	25.33 bc	41.00 ab
Isocycloseram(A21377X)	21.42 c	28.75 b
P-value	0.0006	0.044

Pooled data of larval abundance per 30 inflorescence samples collected from various insecticide-treated plots at Site 1 and 2 are presented in Figure 1. Only three insecticide treatments (isocycloseram followed by chlorantraniliprole, malathion followed by chlorantraniliprole, and cyantraniliprole alone; Figure 1) provide significant suppression ($P < 0.001$) of larval abundance compared to the untreated control. Yield data were only collected at Site 1 (data not shown). Treatment means for seed yield at Site 1 were not statistically different among insecticide treatments. Overall poor yield at Site 2 resulted in no data collection.

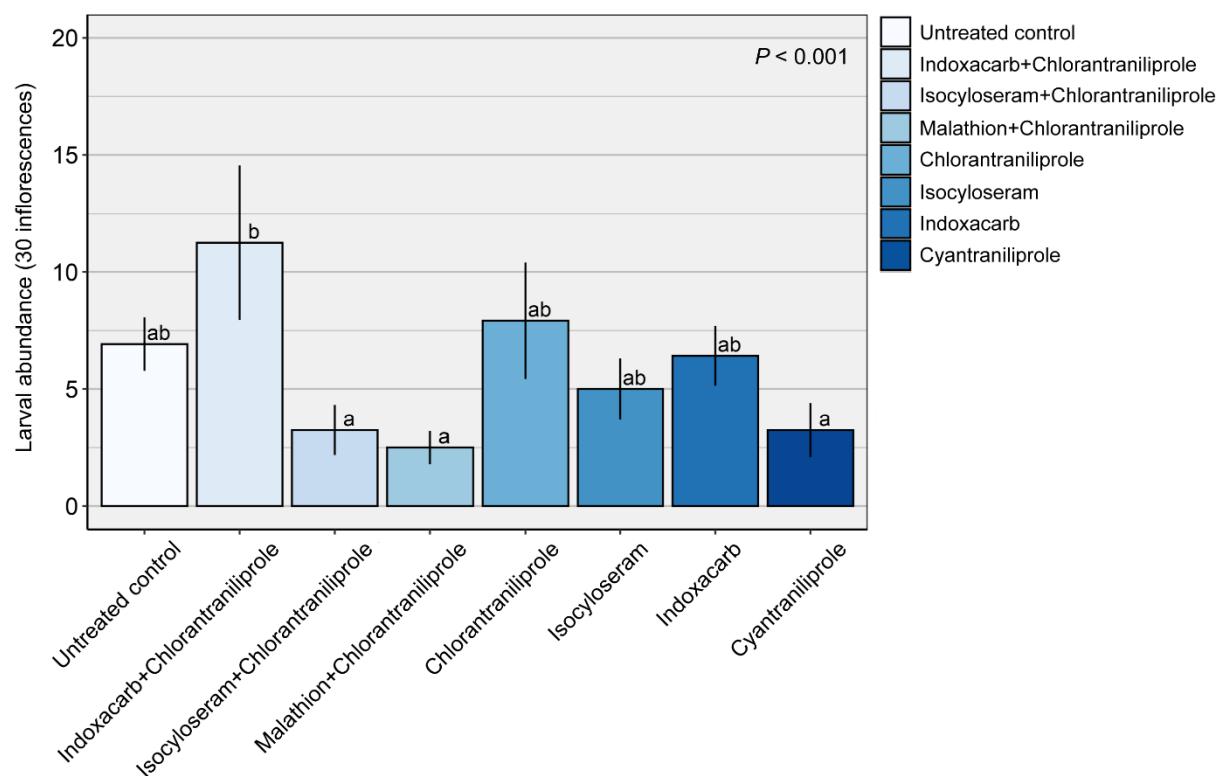


Figure 1. Overall larval abundance per 30 inflorescence samples collected during the post-bloom evaluation period from Sites 1 and 2

References

- Kaur N, Tiwari G, Anderson NP, Lightle D, Tanner C and Dorman SJ (2024) Management guidelines for the clover seed weevil in Oregon white clover seed production. OSU Extension publication EM 9429.
- Tiwari G, Kaur N, Anderson NP, Tanner KC, Lightle DM, Willette A, Donovan B and Dorman SJ (2024) Evaluating foliar insecticides and economic thresholds for *Tychius picirostris* management in Oregon white clover seed production. *Journal of Economic Entomology* **117**, 1892-1900.
- Tiwari G, Kaur N, Anderson NP, Tanner KC, Lightle D, Otani J, Jorgensen A, Willette A, Yoder C and Dorman SJ (2023) Field-evolved pyrethroid resistance in *Tychius picirostris* populations in Oregon white clover seed crops. *Journal of Economic Entomology* **117**, 609-617.

Development of devices for mass capture of insect pests in forage legume seed crops

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Abstract

Alfalfa (*Medicago sativa* L.) and red clover (*Trifolium pratense* L.) seed crops are regularly attacked by insect pests (weevils and bugs), resulting in grain yield losses that can be very substantial. Current solutions for managing these insects, even those based on synthetic chemistry, are not effective enough to ensure the production levels achieved in the past, twenty or thirty years ago. Today, it is therefore important to find new solutions in the field that can be combined with each other, enabling the development of cropping systems that are more respectful of the environment and biodiversity, but also capable of producing good yield results.

To this end, one of the approaches being tested by the FNAMS since 2022 is the use of mechanical devices to mass capture insect pests in the field. Two pieces of equipment are currently being tested: a mass vacuuming device, currently in the prototype stage, and a vegetation beater, already on the market for use against the Colorado potato beetle. The pests targeted by these tools are different, depending on their mobility and their host crop, which makes this equipment complementary on a farm.

The trials carried out to date (2024 and 2025) on micro-plots at the FNAMS experimental station in Brain-sur-l'Authion (France) show very encouraging results in terms of capturing the target insect pests, particularly weevils (*Tychius aureolus* and *Protopion trifolii* in particular). The aim of these trials was to specify the capture efficiency according to: 1) insect flights, 2) the stage and development of the crop, 3) the speed at which the tools are used, and 4) the number of consecutive passes the tools are used. However, a great deal of work still needs to be done to characterise the optimum conditions for using the tools. Development is also needed to limit the collateral effects of these devices on communities of auxiliary insects (predators and pollinators) and on the grain productivity of the crops on which they are used.

Experimental work on this equipment will be carried out as part of a multi-year project funded by the French Ministry of Agriculture and Food Sovereignty. The aim is to be able to deploy these devices operationally on French farms by 2030.

Keywords: insect, pest, device, capture, legume

Introduction

The alfalfa weevil, *Tychius aureolus* Kiesenwetter, is a seed-eating coleoptera that damages seed-bearing alfalfa. It is one of the main pests affecting alfalfa pods in France. The adult lays its eggs in the pod and the larva consumes the seeds during its development. Early studies (D'Aguilar and Perrier 1973) report average yield losses of 14 to 21%. A more recent analysis of multi-year monitoring data indicates that a population of 50 tychius captured in a 25 sweep-net sampling could result in a loss of 30 to 60 kg ha⁻¹ of alfalfa seeds (with an average seed yield of approximately 440 kg ha⁻¹; Coussy 2023).

The red clover weevil (*Protopion trifolii* L.) is the main insect pest in red clover seed production in France. The female lays her eggs in the flower buds and the larva consumes the ovaries or seeds in the inflorescences. Several years of experimentation have shown that the loss of seed yield caused by this insect averages 38% (with a maximum observed loss of 70%; Coussy and Joffre 2018).

Equipment produced by FIELDWORKERS© is already on the market for mass capture of Colorado potato beetle larvae in potato crops. This tool, the 'Colorado beetle catcher', beats the vegetation in the crop

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row and collects the insects in bins placed on the ground between rows. The very first tests of this tool were carried out in 2024 by the FNAMS on alfalfa, and an experimental trial was conducted in 2025.

A prototype mass vacuuming device developed by POLLINOVA© is also being tested by the FNAMS. Initial evaluations began in 2022 on seed-bearing red clover crops. In 2024 and 2025, experimental trials were conducted to evaluate the effectiveness of clover weevil capture based on various factors.

The experiments conducted were funded by the French Interprofessional Organisation for Seeds (SEMAE) and the French Ministry of Agriculture and Food Sovereignty.

Materials and Methods

Evaluation of the Colorado beetle catcher on alfalfa and red clover in 2025. A trial on seed-bearing alfalfa in its first year of production and a trial on seed-bearing red clover, also in its first year of production, were conducted at the FNAMS experimental station in Brain-sur-l'Authion (France) in 2025. The aim was to evaluate the effectiveness of the tool in capturing *T. aureolus* in alfalfa and *P. trifolii* in red clover. Both crops were planted with a row spacing of 50 cm. In each trial, two elementary plots corresponding to a beetle-catcher width over 10 m in length were marked out. Each elementary plot corresponds to a tractor speed (2 km hour⁻¹ and 4.5 km hour⁻¹) in order to assess whether speed affects capture efficiency. On each elementary plot, the effect of two additional factors was also assessed: 1) the number of consecutive passes over the same area (1, 2, 3 passes), and 2) the position of the capture bins on the tool (the outermost pairs of bins on the tool, numbered 1+2 and 7+8). The number of consecutive passes aims to assess whether the number of insects captured decreases as the Colorado beetle catcher is passed over a short period of time, but also to assess whether insects are still being captured on the third consecutive pass over the same area. Sampling is carried out by collecting insects from the two pairs of bins (1+2 and 7+8) after each pass. In alfalfa, four samples were taken on 4 June, 19 June, 24 June and 8 July. In red clover, only two samples were taken (24 June and 8 July), but only the 24 June sample can be analysed because the tool was incorrectly positioned on 8 July, skewing the results.

Evaluation of the mass vacuuming device on red clover in 2024 and 2025. A trial on seed-bearing red clover in its second year of production and a trial on seed-bearing red clover in its first year of production were conducted at the FNAMS experimental station in Brain-sur-l'Authion (France) in 2024 and 2025, respectively. In both years, the clover was planted with a row spacing of 35 cm. As with the alfalfa trials, two elementary plots corresponding to the mass vacuuming device width (1.5 m) and 10 m long were marked out, with each elementary plot corresponding to a tractor forward speed (2 km hour⁻¹ and 5.7 km hour⁻¹). In 2024, the factor of the number of consecutive passes over the same area (1, 2, 3 passes) was also evaluated. In 2025, the factor of the position of the suction openings on the tool (3 openings: left, centre, right) was evaluated. The sampling dates in 2024 were 26 June, 3 July and 10 July; and in 2025, 3 June, 18 June and 2 July. During each sampling, the insects sucked up were collected in insect-proof bags and then counted in the laboratory.

Results and Discussion

Evaluation of the Colorado beetle catcher on alfalfa and red clover in 2025. In 2025 on alfalfa, *T. aureolus* density on the trial was very low, with a maximum of 11 specimens captured in 25 sweep-net sampling. Only a few weevils were collected in the Colorado beetle catcher bins (an average of 3.3 *T. aureolus* per double bin (1+2 or 7+8, per sampling date). Nevertheless, when comparing the capture results of the sweep net and Colorado beetle catcher, the capture efficiency appears to be very similar between the two (a maximum of 0.42 weevils m⁻² captured with the sweep net compared to 0.68 weevils m⁻² captured with the Colorado beetle catcher), which confirms the value of the tool (no low efficiency compared to a proven method such as the sweep net).

Statistical analyses show that the forward speed and position of the bin on the Colorado beetle catcher did not significantly affect the efficiency of *T. aureolus* capture ($P = 0.20$ and $P = 0.22$, respectively). The opposite is true for the number of consecutive passes ($P = 2 \times 10^{-7}$): on average, across all dates, 0.63 weevils m⁻² were captured on the first pass, 0.28 on the second pass and 0.15 on the third pass. For all

dates on which weevils were captured with the tool, this represents an average of 63% of captures on the first pass, 29% on the second and only 8% on the third. Sampling has shown that other insects such as the alfalfa weevil *Hypera postica* Gyllenhal, the mirid bugs *Lygus rugulipennis* Poppius and *Adelphocoris lineolatus* Goeze, and even aphids can be captured relatively effectively with this tool.

One of the problems with using the Colorado beetle catcher is that it can cause collateral damage to crops by collecting flowers and pods during the period when they are present. In our trial, during the flowering phase, a number of flowers were collected, but it is impossible to say whether these were senescent or aborted flowers, the collection of which would not result in a subsequent loss of pods. During the ripening phase, pods were collected and therefore could not produce seeds, resulting in a predictable decrease in yield. For use in fields, it would therefore be necessary to try as much as possible to use the tool before the pods appear, which unfortunately does not necessarily coincide with the peak presence of *T. aureolus* and mirid bugs. Statistical analyses showed no significant difference in flower and pod collection depending on the forward speed or number of passes (for flowers, $P = 0.07$ and $P = 0.99$ respectively; for pods, $P = 0.64$ and $P = 0.62$ respectively). To reduce damage to the crop, if the tool is to be used from the flowering and/or pod stage onwards, it would probably be best to make only one pass (which still allows the majority of insects to be captured).

The Colorado beetle catcher used on 24 June on red clover performed very well, with the bins passing easily between the rows 50 cm apart, as the crop was not too developed and was not lodging. The density of *P. trifolii* on the plot was high and the tool enabled effective capture. As with alfalfa, statistical analyses show that the forward speed and the position of the bin on the tool did not significantly affect the efficiency of weevil capture ($P = 0.17$ and $P = 0.43$ respectively), unlike the number of passes ($P = 0.001$): on average on 24 June, 57.1 weevils m^{-2} were captured on the first pass, 21.9 on the second pass and 13.1 on the third pass. This represents an average of 62% of captures on the first pass, 24% on the second and 14% on the third. This means that under the test conditions (high *P. trifolii* density), there were still a number of captures on the third pass, even though the first two passes accounted for an average of 86% of total captures from a bin at a given speed and date.

Evaluation of the mass vacuuming device on red clover in 2024 and 2025. In 2024 and 2025, the density of *P. trifolii* on red clover was very high, with a maximum of 532 and 1,090 specimens captured in 25 sweep-net samplings, respectively. The results obtained with the sweep net and the mass vacuuming device show that the capture efficiency appears to be very similar between the two techniques (Figure 1) confirming, as for the Colorado beetle catcher, the usefulness of the device (no low efficiency compared to a proven method such as the sweep net). These results seem logical, as a behavioural study in the 2024 trial has shown that the vast majority of *P. trifolii* are found in the upper parts of plants, which allows for relatively effective capture with either the sweep net or the mass vacuuming device.

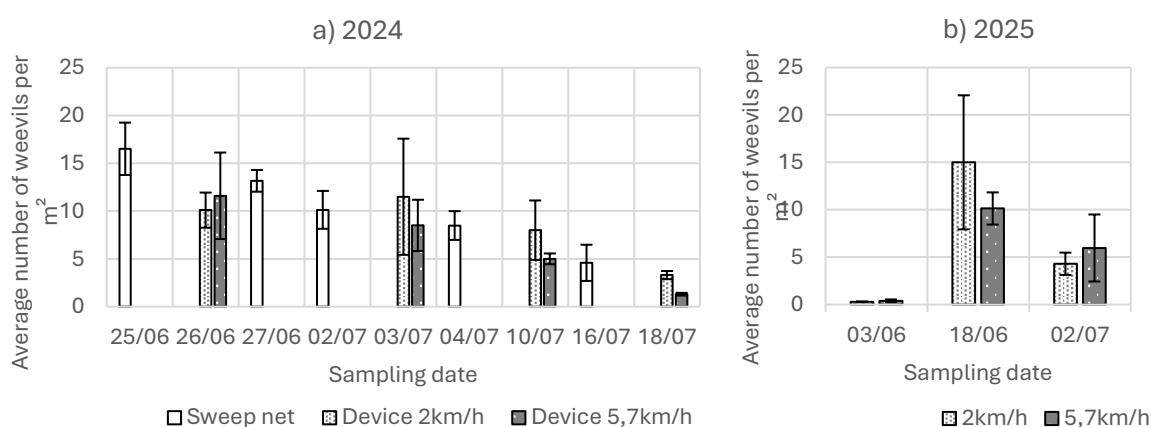


Figure 1. Average number of *P. trifolii* captured per square metre depending on the capture method and date

In 2024, it was not possible to conclude on the single effect of forward speed on the capture efficiency of *P. trifolii*, as an interaction between speed and the position of the suction opening was observed

($P = 2 \cdot 10^{-5}$), potentially due to a heterogeneous distribution of weevils in the plot. However, the 2025 trial showed no interaction and, under these conditions, forward speed had no significant effect on capture efficiency ($P = 0.17$). In 2024, the number of consecutive passes was evaluated as an experimental factor and its effect on capture efficiency proved to be significant ($P < 2 \cdot 10^{-16}$): on average, across all dates, 7.4 *P. trifolii* m⁻² were captured on the first pass; 3.0 on the second pass and 2.0 on the third pass. Across all dates on which weevils were captured with the mass vacuuming device, this represents an average of 60% of captures on the first pass, 24% on the second and 16% on the third, which is fairly comparable with the results obtained in 2025 with the Colorado beetle catcher. In 2025, the factor of the number of consecutive passes was not evaluated, unlike that of the position of the suction opening on the tool. A significant effect of this factor was observed ($P = 0.002$), with the right suction opening capturing more *P. trifolii* on average than the centre and left openings (63.1, 29.1 and 40.5 weevils, respectively), but no precise explanation could be given for this, apart from the uneven distribution of insects in the vegetation on the two sampling dates.

As with the Colorado beetle catcher, the mass vacuuming device can cause some damage to the plants being sucked up. However, this damage is less significant with this tool because the plants are not beaten. On the other hand, it is less selective with insects because they cannot escape from the collection bags once they have been sucked up. To remedy this problem and limit the capture of beneficial insects (predators or pollinators), exclusion filters have been placed on the suction openings of the prototype. The prototype still needs to be improved in order to release the filtered insects as they are collected.

Work on evaluating and developing the tools will continue over the next years in order to precisely identify the potential yield gains resulting from their use and the most optimal strategies for their use in terms of labour time and cost.

References

- d'Aguilar J and Perrier D (1973) Morphologie et biologie comparées de deux espèces de *Tychius* (Col. Curculionidae) dans les luzernières du Lauragais. *Ann. Zool. Ecol. Anim.* **5**(4), 483-489.
- Coussy B (2023) Ravageurs de la luzerne porte-graine - Quelles nuisibilités en parcelles de multiplication? Bulletin Semences 289, juillet-août-septembre 2023: 30-32.
- Coussy B and Joffre A (2018) Apion du trèfle violet - Et maintenant, comment damer l'apion? Bulletin Semences 262, juillet-août 2018: 24-27.

Green on green spot spraying Italian ryegrass in tall fescue grown for seed

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Abstract

Precision spot-spraying technologies offer new opportunities to selectively manage weeds in perennial cropping systems while reducing non-target herbicide exposure. This study evaluated the performance of the Weed-IT Generation 4 “green-on-green” optical spot-spray system for the selective control of Italian ryegrass (*Lolium multiflorum*) within first year fall-planted tall fescue (*Festuca arundinacea*, syn., *Schedonorus arundinaceus* (Shreb.) Dumort.) grown for seed. The system detects chlorophyll fluorescence and reflectance to identify photosynthetically active vegetation, triggering nozzles when green reflectance exceeds background canopy levels. Two sensor sensitivity settings (Level 5: high sensitivity; Level 6: low sensitivity) and three ground speeds (4.8, 9.6, and 14.5 km hour⁻¹) were tested and compared with a hand-crew treatment and an untreated control. Glyphosate (680 g ae ha⁻¹) mixed with spray dye was used to evaluate system accuracy and crop response. Visible dye deposition on Italian ryegrass plants averaged 79-91% accuracy among sensor treatments and 97% for the hand-crew application. Increasing travel speed reduced false-positive detections within the tall fescue canopy, resulting in lower crop phytotoxicity (15-48%) compared with lower speeds. Aerial imagery (red-green-blue-RGB) was collected at 0, 8, 14, 30, 51, and LiDAR imagery at 104 days after application (DAA) to quantify crop phytotoxicity and regrowth. Higher sensitivity and lower speeds (Level 5: 4.8 km hour⁻¹) produced the greatest crop phytotoxicity (up to 50% LAI reduction relative to the check), whereas lower sensitivity (Level 6) and higher speeds (9.6-14.5 km hour⁻¹) had greater selectivity. LiDAR biomass trends mirrored LAI responses, with less reduction under lower speeds.

Introduction:

The Willamette Valley of Oregon is the primary grass seed-producing region in the United States, encompassing over 150,000 ha and generating more than 270 million kg of seed annually, with a farm-gate value exceeding USD 630 million (USDA-NASS 2023). The region’s mild, wet winters and dry, warm summers favour cool-season grass seed crops such as tall fescue (*Festuca arundinacea*, syn., *Schedonorus arundinaceus* (Shreb.) Dumort.) and perennial ryegrass (*Lolium perenne*), but these same conditions also promote the establishment of weedy winter annual grasses, particularly Italian ryegrass (*Lolium multiflorum*) and annual bluegrass (*Poa annua*) (Daugovish et al. 2020).

Italian ryegrass is among the most problematic weeds in tall fescue production due to its similar morphology and seed characteristics, which complicate mechanical separation and reduce marketable seed purity (Gould et al. 2018). Its extended germination period, vigorous growth, and prolific tillering enable dense infestations that compete for light, nutrients, and moisture, ultimately reducing crop vigour and yield (Davis et al. 2017). Persistent seedbanks and widespread herbicide resistance further exacerbate management challenges (Busi et al. 2020; Nandula et al. 2019).

Due to selective postemergence herbicide limitations, producers often rely on manual “rogueing” or spot-spray crews using nonselective herbicides. These approaches are effective but costly and labour-intensive at field scale. Optical spot-spray systems such as WEED-IT employ active sensors to detect chlorophyll fluorescence and trigger precise, real-time applications when green biomass exceeds a defined threshold (Ahmad et al. 2021; López-Granados 2018). This study evaluated the potential of a sensor-guided WEED-IT system for “green-on-green” spot spraying of Italian ryegrass within tall fescue seed fields. Two sensor sensitivity levels and three travel speeds were tested to quantify weed detection accuracy, crop phytotoxicity, and canopy regrowth using aerial leaf area index (LAI) and LiDAR-derived biomass data.

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Methods

Field establishment and pre-emergence management. Tall fescue ('Titanium') was established in fall 2024 at the Oregon State University research farm following standard carbon seeding practices commonly used in Oregon's grass seed industry. These practices involve the surface application of 2.5 cm wide activated carbon over the seed row during planting to adsorb herbicides near the seed zone, thereby enabling the safe use of pre-emergent herbicides during crop establishment. Immediately following seeding, diuron (0.56 kg ai ha⁻¹) was broadcast applied. The field received approximately 4.25 cm of rainfall within 7 days of application.

Spot-spray technology and experimental design. In March 2025, a Generation 4 WEED-IT optical spot-spray system (Rometron B.V., The Netherlands) was evaluated for 'identifying' Italian ryegrass emerged within tall fescue research plots. all fescue was between 7-10 cm and Italian ryegrass was between 20-30 cm in height. The system was mounted on a John Deere Gator (Moline, IL, USA) with nozzles spaced 13 cm apart. The sensors detect chlorophyll reflectance to trigger individual nozzles for localised herbicide delivery once the background level threshold is passed. Treatments included two sensor sensitivity thresholds (Level 5: high sensitivity; Level 6: low sensitivity) and three ground speeds (4.8, 9.6, and 14.5 km hour⁻¹) arranged in a randomised complete block design with four replications. Backpack spot spraying was used as the grower standard. Glyphosate (680 g ae ha⁻¹) was applied as the non-selective herbicide, and a dye was added to the spray solution to enable visual detection of targeted plants. Immediately after application, dye deposition on Italian ryegrass tillers was recorded to quantify system detection accuracy.

Crop injury and remote sensing assessment. Crop injury and regrowth were quantified using a combination of visual ratings and aerial remote sensing. Visual assessments of Italian ryegrass control and tall fescue phytotoxicity were performed at 7 and 14 DAA, while canopy recovery was evaluated using imagery collected at 30 and 51 DAA. A DJI Matrice 300 RTK equipped with a DJI Zenmuse P1 RGB sensor and an L2 LiDAR unit acquired sub-centimetre imagery and 3-D canopy structure data. RGB imagery was used to compute the Triangular Greenness Index (TGI) (Equation 1) to assess spatial chlorophyll variation, while LiDAR-derived point clouds were used to estimate canopy height and biomass regrowth over time.

$$\text{Equation 1: TGI} = -0.5 \times [190(R_{\text{red}} - R_{\text{green}}) - 120(R_{\text{red}} - R_{\text{blue}})]$$

Results and Discussion

Visual assessment of Italian ryegrass control and tall fescue phytotoxicity. Visible dye deposition at application indicated spot-spraying accuracy across all treatments, with each Italian ryegrass plant visually assessed for dye coverage (mean of 18 plants per plot). Accuracy ranged from 79-91% for applications and 97% for the hand-crew treatment. Increasing speed reduced false-positive activations, lowering crop injury, which ranged from 15-48% at 14 DAA based on visual assessment.

Table 1. Visible spray accuracy and crop phytotoxicity following Italian ryegrass spot spraying in tall fescue using a Weed-IT green-on-green system.

Visual Evaluation of Italian Ryegrass Spot Sprayed in Tall Fescue with a Weed-IT Spot Sprayer at Two Sensitivity Levels and Three Speeds			
Sensitivity	Speed (km hour ⁻¹)	% Accuracy	% Phytotoxicity
5	4.8	87 ab	48 e
5	9.6	79 b	38 d
5	14.5	91 ab	26 cd
6	4.8	80 b	25 cd
6	9.6	80 b	18 bc
6	14.5	81 b	15 b
Hand-crew	Walking	97 a	9 ab

Percent accuracy was determined visually at the time of application by assessing dye deposition on Italian ryegrass foliage (average of 18 plants per plot). Percent phytotoxicity was visually rated 14 days after application

(DAA) to quantify crop injury from tall fescue herbicide application. Means followed by the same letter within a column are not significantly different according to Fisher's protected least significant difference (LSD) test at $\alpha = 0.05$.

Aerial imagery assessment of tall fescue phytotoxicity and regrowth. Aerial imagery confirmed uniform canopy conditions prior to spraying (0 DAA) (Table 2). By 14 DAA, reductions in leaf area index (LAI) relative to the untreated check (UTC) were observed at higher sensitivity (Level 5) and lower speeds (4.8 km hour⁻¹), where canopy loss exceeded 25% relative to the check. Higher speeds and lower sensitivity (Level 6) resulted in less canopy phytotoxicity and greater LAI compared with higher sensitivity and lower speed.

At 30 and 51 DAA, high-sensitivity, low-speed treatments continued to exhibit the greatest canopy reduction (40–50% below UTC), whereas lower sensitivity and higher speeds had less reduction in biomass due to phytotoxicity (15–25%). The hand-crew treatment showed moderate initial phytotoxicity but maintained higher LAI relative to the check at later dates (15–20%).

At harvest (104 DAA), LiDAR-derived biomass trends closely mirrored LAI responses: lower sensitivity and moderate-to-high travel speeds resulted in the least biomass reduction relative to the UTC, whereas high-sensitivity, low-speed treatments exhibited the greatest loss. LiDAR data indicated that sensor sensitivity as well as speed influenced biomass accumulation as compared with in-season LAI measurements, where travel speed alone had the greater effect on canopy recovery.

Table 2. Leaf area index (LAI) was derived from aerial imagery to assess canopy phytotoxicity and regrowth following an herbicide application.

<i>Using Leaf Area Index (LAI) and LiDAR measurements obtained from aerial imagery after spot spraying to quantify crop injury and regrowth</i>							
Sensitivity	Speed	LAI 0 DAA	LAI 8 DAA	LAI 14 DAA	LAI 30 DAA	LAI 51 DAA	LiDAR 104 DAA
5	4.8	0.29 a	-12.92 a	-25.48 b	-49.42 e	-49.50 f	-42.2 b
5	9.6	3.09 a	-1.05 a	-12.74 ab	-36.64 d	-36.17 e	-58.71 b
5	14.5	4.17 a	3.26 a	-12.42 ab	-30.31 cd	-29.19 de	-11.82 ab
6	4.8	4.17 a	5.88 a	-7.32 ab	-25.79 bc	-21.52 cd	-6.96 ab
6	9.6	2.54 a	-0.94 a	1.35 a	-17.89 b	-17.07 c	26.57 a
6	14.5	8.49 a	-11.97 a	-0.78 a	-21.11 bc	-15.63 bc	15.17 ab
Hand-crew	Walking	-7.81 a	-4.15 a	-9.00 a	-15.92 b	-20.34 cd	13.68 ab

Values represent the percentage change in LAI relative to the untreated check (UTC) at each timepoint (0, 8, 14, 30, and 51 DAA). Data for each timepoint were analysed separately using analysis of variance (ANOVA) with treatment as a fixed effect and replicate as a blocking factor. When significant effects occurred ($P < 0.05$), means were separated using Fisher's protected least significant difference (LSD) test. Means sharing the same letter within a column are not significantly different. Harvest biomass at 104 DAA, obtained from LiDAR data, is also expressed relative to the UTC.

References

- Ahmad F, Rehman SA, Naz MM and Iqbal MT (2021) Development of an intelligent precision sprayer using real-time machine vision and active sensing. *Computers and Electronics in Agriculture* **190**, 106444.
- Busi R, Vila-Aiub MM, Yu Q and Powles SB (2020) Evolution of herbicide resistance in Italian ryegrass (*Lolium rigidum*) and implications for management. *Weed Research* **60**, 303–316.
- Daugovish O, Hanson BD and Orloff S (2020) Integrated management of annual grass weeds in cool-season crops. *Weed Technology* **34**, 657–667.
- Davis AS, Gibson CM and Hulting AG (2017) Emergence patterns and competitive ability of Italian ryegrass (*Lolium multiflorum*) in temperate cropping systems. *Weed Science* **65**, 626–638.
- Gould SA, Young WC III and Hulting AG (2018) Characterization of Italian ryegrass seed contamination in perennial ryegrass and tall fescue seed lots. *Crop Science* **58**, 2015–2024.

- López-Granados F (2018) Weed detection for site-specific weed management: mapping and real-time approaches. *Advances in Agronomy* **151**, 65–119.
- Nandula VK, Reddy KN and Shankle MW (2019) Seed dormancy and after-ripening behavior of Italian ryegrass (*Lolium multiflorum*) biotypes. *Weed Biology and Management* **19**, 66-75.
- USDA-NASS (2023) *Oregon Agricultural Statistics Annual Bulletin*. Washington, DC: U.S. Department of Agriculture National Agricultural Statistics Service.

Sustainable management strategies for the seed gall nematode (*Anguina funesta*) parasitizing annual ryegrass (*Lolium multiflorum*) seed in Oregon

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Abstract

Oregon's grass seed industry specialises in producing forage grasses including annual ryegrass (ARG, *Lolium multiflorum*), a host for the seed gall nematode (SGN, *Anguina funesta*). SGN causes yield-limiting seed galls and are strictly regulated in international trade. From 2019 to 2020, over 500 metric tons of Oregon ARG seed were rejected from international ports due to SGN detection. A 2022 field survey of 22 ARG fields in the Willamette Valley of Oregon resulted in SGN detection in 50% of the fields throughout the growing season. Several approaches managing SGN are under evaluation. Previous reports indicate that there may be genetic resistance to SGN in other *Lolium* species. Therefore, a breeding population of 240 public accessions of *L. multiflorum* have been seeded with two seed galls and planted in the field. Seed were harvested to evaluate for galls in July 2025 and to identify potential resistant families for future study. To date, no nematicides are labelled for the control of SGN. Varied fluopyram timings and rates, as well as an untreated control, are being evaluated in the field with and without growth regulation for SGN control. Seed yield and galled seed data was collected showing limited differences between treatments. Cultural control methods are also being considered, including seed cleaning and utilizing high energy pulses on seed galls. Preliminary data suggests that these could be viable treatments to reduce SGN inoculum. Successful control options for the SGN in ARG seed production are important to reduce the spread of this nematode globally and maintain healthy forage production.

Keywords: integrated pest management, export, annual ryegrass, seed gall nematode, resistance breeding

Introduction

Oregon produces 70% of the world's cool-season grass seed, including annual ryegrass (ARG; *Lolium multiflorum*) for forage production. This grass species can be parasitised by the seed gall nematode, *Anguina funesta* (hereafter SGN), which can transmit toxic *Rathayibacter* species to grazing animals (Murray et al. 2017; Murray et al. 2025). Recent reports have indicated the presence of SGN in 50% of surveyed ARG seed fields (Rivedal et al. 2024). The presence of SGN in Oregon ARG seed has led to an increase in seed lot rejections in some markets due to zero tolerance limits, particularly in Asia (Rivedal et al. 2024). In 2024, the U.S. produced >104 million kg of ARG with ~66 million kg sold into export markets at an estimated value of US\$49.5 million (National Agricultural Statistics Service 2024; Peace River Forage Association 2014). From 2019 to 2021, at least 500,000 kg were rejected at Asian ports due to the presence of SGN (Rivedal et al. 2024) resulting in an economic loss of approximately US\$730,000. Currently, multiple Asian markets are inaccessible to U.S. ARG producers due to SGN infested seed. Methods to control SGN are needed to improve export success and reduce initial inoculum in the field.

Control of SGN in Oregon grass seed systems is challenging for multiple reasons. Currently, there are no labelled nematicides for use on grass seed crops. The lack of labelled products is partially due to the challenge of timing and cost of a nematicide application during seed production. Other cultural controls, like crop rotation, have limited utility in Oregon ARG production since many fields are perennially cropped (Rivedal et al. 2024), and the seed galls can survive for many years in the soil

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(Murray et al. 2017). Genetic resistance is not currently available to Oregon seed producers, and all previous resistant varieties have been identified for environments other than seed production (i.e., Australian pasture grass; Riley and Barbetti 2008; Murray et al. 2025). Local breeding efforts are necessary to find the appropriate, nematode-resistant germplasm, but are not yet available for Oregon ARG producers. Seed cleaning is an area of cultural intervention that could provide improved SGN control, but has yet to be evaluated due to the similar size of seed galls to healthy seed. Utilising different seed cleaning tools (gravity table, air screen cleaner) along with non-chemical interventions like high energy pulses may reduce overall SGN inoculum in seed lots. In this study we evaluate three management strategies for SGN in ARG seed production in Oregon: 1) genetic resistance through traditional breeding methods; 2) chemical control with fluopyram in the field; and 3) cultural control through seed cleaning and high energy pulses. Together, these methods will provide a framework for best management practices to control SGN in ARG seed production.

Materials and Methods

Breeding trial. In November 2024, five seeds of each of 240 ARG accessions were grown in plug trays. Each seed was sown with two SGN galls. After eight weeks, plugs were planted at the Oregon State University Botany Field Lab in an augmented design with diploid variety ‘Gulf’ and tetraploid variety ‘HW-51’ checks. On June 24, 2025, all five plants were hand harvested, placed collectively into large bags and allowed to air dry for four weeks. After drying, each accession was hand threshed, and cleaned with a Clipper cleaner with limited airflow. Cleaned seed was collected, weighed, and three 25 cc subsamples were collected for seed counting and visual gall evaluation. A repeated trial was planted in October of 2025.

Nematicide trial. In October 2024, two randomised complete split-plot trials were established at the Oregon State University Botany Field Lab. Each trial consisted of either diploid variety ‘Gulf’ or tetraploid variety ‘HW-51’. In each trial, three whole plots (8.5 x 6.7 m) were established and split into eight split plots (4.3 x 1.2 m). Plots were seeded at a rate of 28 kg ha⁻¹ with 10% of the seeding weight replaced with galls. Whole plots were divided in half, and four received a growth regulator application (trinexapac-ethyl, TNE, 420 g ai ha⁻¹) in spring, and the other half did not. Four nematicide treatments, untreated control, autumn application, spring application, and autumn + spring application, were applied to two split plots (one TNE+, one TNE-) per whole plot. Applications were made in November 2024 and April 2025 using 247 g flupyrpyr/ha for all applications. Plots were swathed, and combined in July 2025. Seed was cleaned on a clipper cleaner, and three 25 cc subsamples were collected for seed counting and visual gall evaluation. A repeated trial was planted in October 2025.

Seed cleaning. Two commercial ARG seed lots with natural infestation of SGN from 2024 were used to compare gall removal on an air screen and gravity table cleaner. Four different air speeds were evaluated for each cleaner and two 3000 cc seed samples per seed lot were tested per speed. Seed was collected into clean or dirty cuts as would be done for a commercial cleaning line. Preliminary comparisons of gall status across cuts and air speed were conducted for each cleaner.

Lisi Global seed treatments. Utilising Lisi Global’s high energy pulse method, we evaluated five energy profiles: untreated control; 200 V at 20 J cc⁻¹ and 5 m sec⁻¹; 280 V at 20 J cc⁻¹ and 5 m s⁻¹; 200 V at 20 J cc⁻¹ and 2 m s⁻¹; and 200 V at 40 J cc⁻¹ and 5 m s⁻¹. For each energy profile six replicates of 90 seeds plus 10 galls were exposed to the treatment. The number of seeds that germinated as well as the number of live and dead nematodes per gall were recorded.

Results and Discussion

Breeding trial. In total, there were 16 plots each of the diploid and tetraploid checks. Visual evaluation of clean seed indicated that all diploid plots had gall detections (100% prevalence), whereas 15 of 16 plots had gall detections for tetraploid plots (93% prevalence). While prevalence of SGN was high for checks, incidence was still quite low, with an average of 0.304% infection for diploid ‘Gulf’ plants, and 0.384% infection for tetraploid ‘HW-51’ plants. This supports the hypothesis that both diploid and

tetraploid ARG is susceptible to disease in Oregon. It also highlights some of the challenges of working with this pathosystem – infection rates are very low, meaning there is no guarantee that each plant is infected despite high inoculation volume (two galls is equivalent to ~2000 individual nematodes). This highlights the need for more basic biological studies to understand the SGN-ARG life cycle. The other accessions are still being evaluated for galls; however, variability of infection has been noted from initial counts. The goal is to develop resistant germplasm that can be incorporated into industry breeding programs that ensure lower susceptibility to SGN.

Nematicide trial. In the nematicide trial, galls were detected across nematicide treatments, growth regulator treatments, and replicates, regardless of ARG variety. The average incidence for ‘Gulf’ plots ranged from 0.08% to 1.4% per 25 cc seed, while the average incidence for ‘HW-51’ plots ranged from 0% to 1.7% per 25 cc seed. Since infection was still found across the field trial, regardless of treatment, it was determined that higher rates, and sprays closer to germination may be required to limit infection caused by SGN. This will be evaluated in the recently established 2025 nematicide trial.

Seed Cleaning. For both cleaners, four air speeds were evaluated on two replicate samples from two commercial seed lots. The air screen cleaner removed galls from healthy seed at a range of airspeeds, but mid-range air speeds led to the highest reduction in galls with the lowest loss of clean seed (120 CFM). The gravity table was highly effective at separating galls from healthy seed, leading to significant gall reductions across clean cuts for three of four tested air speeds (40-172 CFM). However, gravity tables are expensive and infrequently available to ARG seed producers in Oregon. Additionally, since this work was conducted on naturally infested seed lots, additional study using samples with known rates of infection would be beneficial to further refine clean out abilities of both methods.

Lisi Global seed treatments. For the five different treatments tested, germination rates of the 90 treated seeds averaged between 82 and 88%. Untreated control galls had live nematodes present in 95% of tested galls, with 83% germination of seed. For all other treatments, the incidence of live nematodes was reduced to less than 80% of treated galls. Treatment 280 V at 20 J cc⁻¹ and 5 m s⁻¹ had the lowest live nematode incidence, with 51% of galls having living nematodes after treatment. Germination for this treatment was 84%, indicating a potential starting treatment to increase gall kill efficacy and maintain germination rates at or above untreated seed levels. This method is non-destructive to both seeds and galls but could lead to overall reductions in viable nematode populations. Further evaluation of this method as part of a seed cleaning line are currently under evaluation.

These studies provide new information on the potential success of multiple treatment options for SGN in ARG seed production in Oregon. When final best practices are developed, they will be shared widely with growers in Oregon and across the globe to help reduce infection by this nematode.

References

- Murray TD, Schroeder BK, Schneider WL, Luster DG, Sechler A, Rogers EE and Subbotin SA (2017) *Rathayibacter toxicus*, other *Rathayibacter* species inducing bacterial head blight of grasses, and the potential for livestock poisonings. *Phytopathology* **107**, 804-815. <https://doi.org/10.1094/PHYTO-02-17-0047-RVW>
- Murray TD, Duarte A, Luster DG, McKirdy SJ, Rogers EE, Schroeder BK and Subbotin SA (2025) Seed Gall Nematodes and their association with toxigenic bacteria. *Ann. Rev. Phytopath.* <https://doi.org/10.1146/annurev-phyto-121823-033153>.
- Riley IT and Barbeti MJ (2008) Australian anguinids: their agricultural impact and control. *Aust. Plant Pathol.* **37**, 289–297. <https://doi.org/10.1071/AP08022>.
- Rivedal HM, Temple TN, Starchvick RJ, Braithwaite E, Lowder SR, Dorman SJ, Núñez Rodríguez LA, Peetz AB and Zasada IA (2024) Comparison of molecular and morphological identification

methods for *Anguina* seed gall nematodes in Oregon grasses grown for seed. *PhytoFrontiers*
<https://doi.org/10.1094/PHTOFR-01-24-0001-R>

ORAL PRESENTATIONS

Sustainable seed production

Abstract

The perennial forage seed producers and production area are in declining trend in favour of simplified annual crop rotations in the Peace River region of Canada. High cost of perennial termination for transitioning from perennial to annual crop phases in the rotations is a deterring factor. A three-year cropping systems experiments was started in 2024 to examine the prospect of integrating cover crops in the transitional phase between perennial creeping red fescue seed crop and succeeding annual crops. Following the herbicidal spray for the termination of creeping red fescue stand, five full-season cover crop treatments were replicated four times in randomised complete block design. Biomass production, leaf area index and nitrous oxide (N₂O) emission were evaluated during the cover crop phase. The cut biomass was left for decomposition on the field for soil improvement. The legacy effects of cover crops are being evaluated on succeeding annual crops of canola followed by wheat in terms of yield, weed suppression and soil health indicators. Cover crop treatments containing hairy vetch as a sole crop (7.5 t ha⁻¹) and its mixture with sweet clover (7.0 t ha⁻¹) or fall rye (6.6 t ha⁻¹) produced higher double-cut biomass yield compared to the mixture of sweet clover and tillage radish (3.1 t ha⁻¹) and the single-cut biomass of annual pea (4.3 t ha⁻¹). The leaf area index as the surrogate measure of weed suppression potential also followed the similar trends as biomass yield. The cover crops of perennial legumes exhibited higher fluxes of N₂O emission towards the end of the cropping season. However, the hairy vetch sole crop treatment had the least intensity of N₂O emission with respect to dry biomass productivity. The legacy effects of cover crop treatments on economic yield, expressed as canola equivalent yield (CEY) of succeeding crops in 2025 highlighted the profitability of sweet clover-based systems.

Keywords: biomass, creeping red fescue, hairy vetch, leaf area index, nitrous oxide

Introduction

Perennial forage seed production in the Peace River region of Canada has undergone a marked decline as simplified annual crop rotations have replaced diversified systems. Between 2001 and 2023, the area of perennial seed crops decreased by 65% (from 138,000 to 49,000 ha), while the number of producers fell by 72% (from 1,086 to 303) (Yoder 2025, personal communication). This decline has reduced the agroecological benefits of perennials, which include improved soil organic matter, erosion control, and enhanced productivity of luvisolic soils. Simplified rotations have also increased vulnerability to pests, herbicide-resistant weeds, and environmental stresses (Khanal 2022; Khanal et al. 2021).

A major barrier to sustaining perennial-based diversification is the high cost of rejuvenation and termination forage seed stands, particularly creeping red fescue (*Festuca rubra* L.), which is resilient but difficult to rejuvenate or remove (Deleuran et al. 2013; Fairey & Lefkovitch 2001). Although perennial growth habits allow multiple harvests, indefinite tillering and root-boundness reduce seed yield after the first year and complicate stand termination. Transitioning to annual crops often requires herbicide- or tillage-intensive interventions (Holmes 2018). Alternatives that reduce termination costs while maintaining soil health are urgently needed.

Cover crops offer a promising solution. They provide soil cover, scavenge nutrients, and suppress weeds through rapid canopy development measured by leaf area index (McKenzie-Gopsill et al. 2022). Legume-based cover crops also supply nitrogen by biological fixation, though they may increase nitrous oxide (N₂O) emissions following termination (Bressler & Blesh 2023). Integrating cover crops between perennial and annual phases could mitigate termination costs, sustain soil health, and enhance system resilience, but empirical data under Peace River conditions remain scarce.

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This study was initiated in 2024 to evaluate cover crop-mediated termination of creeping red fescue and its legacy effects on succeeding annual crops. Specifically, the objectives were to: (1) identify cost-effective options for perennial stand termination and transition to annual cropping; (2) assess improvements in soil health, greenhouse-gas mitigation, and reduced reliance on synthetic inputs through legume-based cover crops; The underlying hypotheses were that (i) cover crops suppress fescue regrowth and weeds through competitive and allelopathic effects, (ii) cover crop mixtures exert synergistic suppression, and (iii) cover crops green manuring enhances soil microbial activity, nitrogen availability, and yield of succeeding crops. By addressing these aims, the project seeks to generate agroecological and economic solutions for sustainable crop diversification in the Peace River region.

Materials and Methods

The experiment was conducted during 2024-2025 at Beaverlodge Research Farm, Alberta, Canada (55°12' N, 119°24' W). The soils were classified as Gray Luvisols and are characteristic of boreal continental climate of the Peace River region. The study evaluated five cover crop treatments: (1) hairy vetch (*Vicia villosa* Roth) (seed rate at 45 kg ha⁻¹), (2) hairy vetch (22.5 kg ha⁻¹) + sweet clover (*Melilotus officinalis* L.) (22.5 kg ha⁻¹), (3) hairy vetch (22.5 kg ha⁻¹) + fall rye (*Secale cereale* L.) (50 kg ha⁻¹), (4) sweet clover (22.5 kg ha⁻¹) + tillage radish (*Raphanus sativus* L.) (10 kg ha⁻¹), and (5) field pea (*Pisum sativum* L.) (200 kg ha⁻¹, control). Plots were established on terminated creeping red fescue stands. Fescue termination involved two applications of glyphosate (Roundup WeatherMAX, 3.2 L ha⁻¹). Treatments were arranged in a randomised complete block design (RCBD) with four replications. Seeding occurred on 13 May 2024. No fertiliser was applied to allow assessment of cover crop legacy effects. Weed control relied solely on cover crop competition, with mowing together with cover crops twice to prevent weed seed set.

The cover crop biomass was sampled by hand clipping within 1 m² quadrats on 1 August and 19 September in 2024. Fresh and dry weights were recorded, and canopy development was assessed using a LAI-2200C plant canopy analyser (LI-COR Environmental, USA). Drone-based digital and NDVI imagery was collected periodically to monitor canopy cover and crop vigour. Nitrous oxide (N₂O) fluxes were measured weekly using a LI-7820 N₂O/H₂O trace gas analyser (LI-COR Environmental, USA). Post-harvest soil samples were collected for chemical and biological analyses.

In 2025, all plots were seeded to canola. Each plot was split into two strips: one with conventional herbicide weed control and one without. No fertiliser was applied. Measurements included soil sampling, N₂O fluxes, drone imagery, crop vigour ratings, weed surveys (species composition, density, biomass), plant population counts, and crop height. Canola biomass and seed yield were determined from quadrat harvests. Regrowth and seed production of biennial sweet clover and fall rye were also recorded.

All data were analysed using PROC GLIMMIX in SAS v.9.4 (SAS data and AI solutions), with block as a random effect and treatment as fixed. Means were separated using Tukey's HSD at $P \leq 0.05$. Economic analysis included seed yield expression into canola equivalent yield (CEY), calculated as the price ratio of non-canola to canola multiplied by non-canola seed yield.

Results and Discussion

This exploratory study investigated the potential of cover crops to mediate termination of creeping red fescue seed stands after three production seasons. The central hypothesis was that rapidly growing, nitrogen-fixing and nitrogen-scavenging cover crops suppress regrowth of terminating crops through shading and allelopathic effects, with biomass accumulation and leaf area index (LAI) serving as the primary suppressive mechanisms.

Cover crop biomass yields, and LAI differed significantly among treatments ($P < 0.05$) (Figure 1A). Hairy vetch (HV) monoculture and its mixture with sweet clover (HV + SC) produced the highest double-cut biomass yields of 7.5 and 7.0 t ha⁻¹, respectively. The HV + fall rye (HV + FR) mixture yielded 6.6 t ha⁻¹, while sweet clover + tillage radish (SC + TR) and field pea (FP) produced 3.1 and 4.3 t ha⁻¹, respectively. Peak LAI values mirrored biomass trends, reaching 4.55 for HV, 4.58 for HV + SC, 3.95 for HV + FR, 1.46

for SC + TR, and 2.62 for FP (Figure 1B). High LAI corresponded with visual assessments of weed suppression, consistent with earlier findings that rapid canopy closure is a key mechanism of weed control by cover crops (Bressler & Blesh, 2023; Groß et al, 2024).

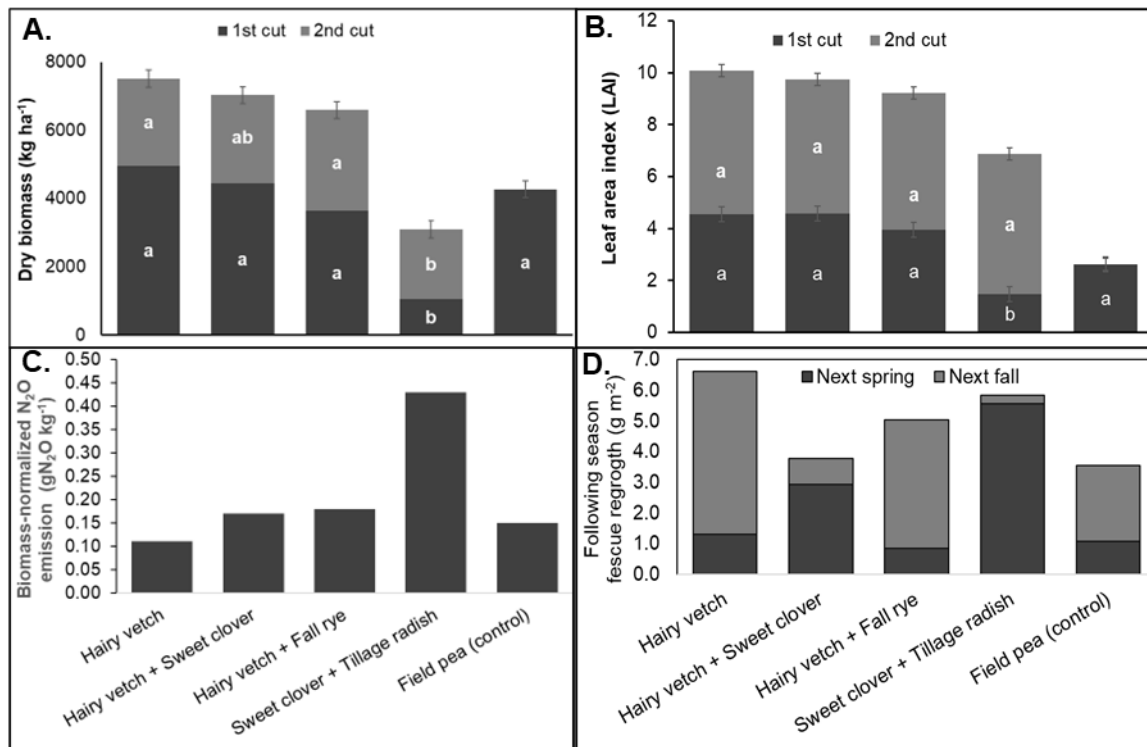


Figure 1. Dry biomass (panel A), leaf area index (LAI) (panel B), nitrous oxide (N₂O) emission intensity per unit cover crop biomass (panel C) and follow season regrowth of terminating crop of creeping red fescue of different cover crops evaluated at Beaverlodge Research Farm, Alberta, Canada in 2024. Treatment columns followed by the same letter are not significantly different ($P > 0.05$). Error bars represent the standard error (SE).

Nitrous oxide (N₂O) fluxes were monitored weekly. Hairy vetch monoculture and HV + SC mixtures exhibited higher emission peaks late in the season, likely due to rapid mineralization and subsequent denitrification of first-cut biomass. However, when normalised to biomass, HV recorded the lowest emission intensity (0.11 g N₂O kg⁻¹ biomass), followed by FP (0.15 g N₂O kg⁻¹ biomass). SC + TR produced the highest intensity (0.43 g N₂O kg⁻¹ biomass), while HV + FR and HV + SC were intermediate (0.18 and 0.17 g N₂O kg⁻¹ biomass, respectively) (Figure 1C). These findings corroborate earlier reports that biomass removal and timing of incorporation strongly influence N₂O release (Böldt et al. 2024; Bressler & Blesh 2023). Thus, while high-biomass treatments enhanced weed suppression, they also carried a trade-off in terms of elevated N₂O emissions. The relatively low emission intensity of HV suggests that careful species selection and residue management can mitigate greenhouse gas risks (Abdalla et al. 2019; Sievers & Cook 2018).

The legacy effects of cover crops were assessed on the 2025 canola crop. That season was marked by severe drought, with regional agricultural emergencies declared (Agricultural Service Boards 2025) and provincial surveys reporting that only 20% of crops were in desirable condition (Government of Alberta 2025). Consequently, canola establishment was poor across treatments. Experimental deviation also arose because annual white sweet clover was inadvertently substituted with biennial yellow sweet clover, and fall rye behaved as a biennial. As a result, harvests included both canola and cover crop seeds. Yields were converted into canola equivalent yield (CEY) for comparison.

Fescue regrowth suppression varied across the cover crops and succeeding crops sequences. The hairy vetch and FP plots showed reduced regrowth in spring, but suppression later shifted in favour of biennial crops (SC and FR) (Figure 1D). However, regrowth was patchy, and differences were not statistically significant. Weed pressure was consistently higher in FP legacy plots, while sweet clover-

containing treatments reduced weed biomass and coverage (Table 1). This supports earlier findings that sweet clover cultivars can suppress weeds and sustain subsequent wheat yields (Moyer et al. 2007).

Table 1. Weed dynamics during succeeding canola production in 2025 at site A of Beaverlodge Research Farm, Alberta, Canada.

Treatments	May 27, 2025			August 28, 2025			Canola equivalent yield (kg ha ⁻¹)
	No. of Weed Species	Weed Cover (%)	Weed dry biomass (gm m ⁻²)	No. of Weed Species	Weed Cover (%)	Weed dry biomass (gm m ⁻²)	
1. HV	3.50 ab	25.13 ab	17.44	4.25 ab	40.0 ab	168.8 ab	847 c
2. HV + SC	2.25 bc	6.50 b	7.11	3.00 ab	20.3 b	42.4 ab	10043 b
3. HV + FR	2.75 abc	14.38 ab	5.76	3.50 ab	53.8 ab	160.2 ab	959 c
4. SC + TR	1.75 c	2.38 b	4.57	1.50 b	13.5 b	17.3 b	15432 a
5. FP (control)	3.75 a	43.75 a	82.29	4.75 a	92.8 a	210.5 a	215 c
p-values	0.003	0.013	<.0001	0.0097	0.0034	0.0136	<0.0001
SEM	0.316	9.94	6.047	0.540	12.517	43.445	1023

HV = hairy vetch; SC = sweet clover; FR = fall rye; TR = tillage radish; FP = field pea. SEM = Standard error of mean (n=4). Numbers followed by different letters are significantly different at $P < 0.05$ according to Tukey's test. Note: Major weed species observed during 2025 (variable proportion in relation to cover crops treatment and succeeding crop growth) were clover, Shephard's purse, dandelion, field violet, lamb quarters, foxtail, and corn spurry

Economic yield, expressed as CEY, highlighted the profitability of sweet clover-based systems. Sweet clover and tillage radish mixture (SC + TR) produced the highest CEY, followed by HV + SC. The high seed yield and market value of sweet clover contributed substantially to these outcomes, confirming its potential as a strategic intercrop during perennial-annual transitions. Sweet clover not only enhances profitability but also reduces reliance on synthetic nitrogen inputs (Moyer et al. 2007). In contrast, FP legacy plots produced the lowest CEY, while HV legacy plots yielded sub-optimal returns due to poor canola establishment under drought stress.

The ongoing evaluation of legacy effects on the 2026 wheat crop will provide further insights into soil health and system resilience. Studies in semi-arid systems indicate that functional group richness does not always outperform monocultures, but legumes and mixtures can improve soil organic carbon and nitrogen cycling (Jones et al. 2025; Opoku et al. 2024). Global syntheses also show that cover crops can increase soil organic carbon and yields, though trade-offs with N₂O emissions remain (He et al. 2025).

Overall, the study demonstrates that cover crops, particularly hairy vetch and sweet clover mixtures, can play a significant role in terminating perennial fescue stands while providing weed suppression, soil fertility benefits, and economic returns. However, trade-offs exist between biomass productivity and greenhouse gas, nitrous oxide (N₂O) emissions. Biennial legumes such as sweet clover appear especially promising, offering both profitability and ecological services under challenging climatic conditions. Continued monitoring of soil health and crop performance will be essential to confirm the long-term sustainability of these strategies in the Peace River region.

References

- Abdalla M, Hastings A, Cheng K, Yue Q, Chadwick D, Espenberg M, Truu J, Rees RM and Smith P (2019) A critical review of the impacts of cover crops on nitrogen leaching, net greenhouse gas balance and crop productivity. *Global Change Biology* **25**(8), 2530-2543.
- Agricultural Service Boards (2025) *Municipalities Declaring Disasters in 2025: A Growing Concern – Updated September 16, 2025*, 2025. Available online:

<https://agriculturalserviceboards.com/2025/07/29/municipalities-declaring-disasters-in-2025-a-growing-concern/>.

- Böldt MJ, Smit HPJ, Loges R, Taube F, Kluß C and Reinsch T (2024) Evaluating nitrous oxide emissions in low input systems using different cover crop strategies over the winter period. *Agriculture, Ecosystems & Environment* **364**, 108895.
- Bressler A and Blesh J (2023) A grass–legume cover crop maintains nitrogen inputs and nitrous oxide fluxes from an organic agroecosystem. *Ecosphere* **14**(2), e4428.
- Deleuran LC, Kristensen K, Gislum R and Boelt B (2013) Optimizing the number of consecutive seed harvests in red fescue (*Festuca rubra* L.) and perennial ryegrass (*Lolium perenne* L.) for yield, yield components and economic return. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science* **63**(1), 1-10.
- Fairey NA and Lefkovitch LP (2001) Effect of post-harvest management on seed production of creeping red fescue, tall fescue, and Kentucky bluegrass in the Peace River region of north-western Canada. *Canadian Journal of Plant Science* **81**(4), 693-701.
- Government of Alberta (2025) *Alberta Crop Report: Crop conditions as of June 30, 2025*, 2025. Available online: <https://open.alberta.ca/dataset/2e0c96ee-50bf-4891-8f16-224233f372ce/resource/dcf5c8c7-86bf-4895-881f-15b5859c6599/download/agi-tedab-alberta-crop-report-2025-06-30.pdf> [Accessed].
- Groß J, Kümmerer R, Heuermann D, Gentsch N, Schweneker D, Feuerstein U, Guggenberger G, von Wirén N and Bauer B (2024) Improving dual cover crop mixtures to increase shoot biomass production and weed suppression potential. *Frontiers in Agronomy* **6**.
- He Q, Lu C, Cowie A, Zhao S, Liu DL, Yi B, Shi L, Zhang S, Qiu T, Shi Y, Huete A, Siddique KHM, Yu Q and Li L (2025) Optimizing cover cropping application for sustainable crop production. *npj Sustainable Agriculture* **3**(1), 10.
- Holmes M (2018) *Effect of Forage Stand Termination Method and Fertilization History on Greenhouse Gas Emissions, Nutrient Supply Rates, and Soil Carbon Dynamics*. Master of Science University of Saskatchewan. Available online: <https://harvest.usask.ca/server/api/core/bitstreams/98b87c11-338f-498a-a88c-1ee0bb4cfb3a/content>.
- Jones CA, Zabinski CA, Miller PR, D'Agati K, Tallman SM and Housman ML (2025) Soil health responses to cover crop functional group and richness in semiarid Montana. *Soil Science Society of America Journal* **89**(2), e70045.
- Khanal N (2022) Integration of perennial forage seed crops for cropping systems resiliency in the Peace River region of western Canada. *Canadian Journal of Plant Science* **103**(1), 1-12.
- Khanal N, Azooz R, Rahman N, Klein-Gebbinck H, Otani JK, Yoder CL and Gauthier TM (2021) Value of integrating perennial forage seed crops in annual cropping sequences. *Agronomy Journal* **113**(5), 4064-4084.
- McKenzie-Gopsill A, Mills A, MacDonald AN and Wyand S (2022) The importance of species selection in cover crop mixture design. *Weed Science* **70**(4), 436-447.
- Moyer JR, Blackshaw RE and Huang HC (2007) Effect of sweetclover cultivars and management practices on following weed infestations and wheat yield. *Canadian Journal of Plant Science* **87**(4), 973-983.
- Opoku A, Ogunleye AM, Solomon JKQ and Payne WA (2024) Cover crop systems impact on biomass production, carbon-to-nitrogen ratio, forage quality, and soil health in a semi-arid environment. *Heliyon* **10**(20).
- Sievers T and Cook RL (2018) Aboveground and Root Decomposition of Cereal Rye and Hairy Vetch Cover Crops. *Soil Science Society of America Journal* **82**(1), 147-155.

Alternative harvesting practices in western Oregon grass seed production systems

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Abstract

Annual bluegrass (*Poa annua* L.) (ABG) is a common weed in Oregon grass seed production systems and is sometimes unwantedly harvested with grass seed crops, which leads to an increase in stakeholder seed cleaning costs. While the morphology of ABG is diverse with plant heights reported to be as high as 40.6 cm, the majority of ABG panicles reside below tall fescue (*Festuca arundinacea*) (TF) panicles at the time of harvest. This morphological difference could be used to the benefit of stakeholders by swathing or direct harvesting above ABG. Internationally, some grass seed growers direct harvest or use a combine instead of a swather to make the first cutting to collect mature seeds, although this technique is not common in Oregon, likely because drying of grass seed is not a routine practice. It is hypothesised that if a combine were used to make the first cutting in an ABG contaminated stand of TF, the equipment could be set to cut above ABG and below TF panicles, resulting in a TF crop that has a reduced amount or perhaps no ABG present in the crop. Because drying facilities are not common in Oregon, it may be beneficial to explore the impacts of direct harvesting TF at lower than recommended swathing seed moisture (<35%). In this case, avoiding the swather altogether may capture more total seed than the traditional method of swathing and later combining.

To elucidate these questions, a preliminary trial took place in 2024 in Corvallis, Oregon on a second-year stand of '4th Millenium' TF sown on 30 cm row spacing. The experiment was arranged as a randomised complete block design that explored two swathing heights (5 cm and 28 cm), two initial cutting methods (swather or combine), two seed moistures (27% and 43%), as well as two direct harvest treatments at 10% moisture at 5 cm or 28 cm in height. The 2024 data analysis observed no differences in clean crop yield between harvest heights or across swathing seed moistures, suggesting promise for this technique. While not significant in 2024, when the combine was used instead of a swather for the initial cutting, seed yields trended higher compared to the traditional harvesting method. Interestingly, the lack of differences in seed yield between swathing at 27% and 43% suggests there is a potential benefit to stakeholders to further research swathing at moistures below the current minimum local university recommended swathing moisture of 35%. Using a combine to swath at a lower seed moisture than 35% would likely reduce drying costs, increasing the likelihood of stakeholder acceptance of this method. In 2025, an additional harvesting experiment took place on a first-year stand of TF. Factors included three swathing moistures (18%, 35%, & 45%), two swathing heights (5 cm and 28 cm) and two swathing methods (swather and combine). Findings suggest that ABG contamination was reduced when harvesting at 28 cm compared to 5 cm.

Keywords: swathing, seed moisture, annual bluegrass, tall fescue, harvest

Introduction

Suppressing grassy weeds in grass seed production systems relies on best practices at the time of establishment, timely herbicide applications, and labour-intensive hand-rogueing activities. In some instances, the incidence of weeds like annual bluegrass (*Poa annua* L.) (ABG) can still dominate the understory of grass seed fields, creating challenges for seed cleaning activities post-harvest. For some species of grasses like tall fescue (*Festuca arundinacea*) (TF), lodging is less pronounced, resulting in standing panicles at the time of harvest with the majority of the seed crop higher in the canopy than the shorter ABG weed seeds, although some ABG plants are reported to grow as high as 40.6 cm (Warwick 1979). This physical separation of crop and weed seed at the time of harvest presents an opportunity to test the impacts of alternative TF harvesting practices with the objective to avoid harvesting ABG alongside the crop seed. Potential alternative harvesting practices include swathing

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above the ABG panicles compared to swathing closer to the base of the plants, or direct harvesting the grass seed crop at a higher height to collect only the crop seed panicles. An underlying uncertainty in Oregon grass seed production systems are the logistics to directly harvest TF prior to excessive shattering of the crop but with the challenge of processing seed that is still too humid for direct storage. This is especially concerning in the Pacific Northwest United States because drying of grass seed crops is not standard practice with a very limited number of seed drying facilities available. Drying is also likely to be considered cost-prohibitive to growers compared to traditional harvesting practices where swathing rows are permitted to dry in the field until seed is harvested at moisture levels suitable for storage. With the goal of exploring alternative harvesting practice in western Oregon, two field experiments took place that compared traditional swathing versus direct combining at two different heights and at different seed moisture contents.

Materials and Methods

In the summer of 2024, an alternative harvesting experiment exploring traditional versus alternative harvesting treatments took place on a second-year stand of TF in Corvallis, Oregon at the Hyslop Field Research Laboratory. The research field was sown in autumn 2022 with the cultivar '4th Millenium' on 30.5 cm row spacing at 13.5 kg ha⁻¹. Annual nitrogen inputs totalled 179 kg ha⁻¹. Herbicides were used to suppress dichotomous weeds and the herbicide metribuzin was used to suppress volunteer grasses on October 6th, 2023. The experiment was laid out as a randomised complete block design with ten treatments, and the experiment was replicated three times. A summary of the treatments is shown in Figure 1.

The traditional harvesting method consisted of a swather (John Deere 2270 swather, Moline, IL, USA) used to cut the crop that created a swathing row that was permitted to dry in the field and was later collected with a combine (Zürn model 150, Ravenstein-Merchingen, Germany) equipped with a pick-up header. The alternative harvesting treatment consisted of using a research combine (Zürn model 150, Ravenstein-Merchingen, Germany) equipped with a cutter bar and reel to cut the crop, while permitting the combine to collect any seed as the crop material flowed through the combine. The material that passed through the combine was left in the field and was recleaned with the same combine using a pick-up header at a later date. The initial pass of the combine had settings that consisted of a threshing drum speed of 500 rpm, blower at 400 rpm, concave setting set to maximum spacing, middle screen size of PS8, and bottom screen size of RD8. Harvesting occurred at two different seed moistures (27% or 43%) and two different harvesting heights (5 or 28 cm). In addition, direct harvesting at 10% seed moisture at different harvesting heights (5 or 28 cm) took place. Direct harvesting and final harvesting with the combine pick up header occurred with a Zürn 150 with a threshing drum speed of 1450 rpm, concave setting set to the minimum spacing, middle screen size of PS8, and bottom screen size of RD8. The blower setting for plots originally cut at 43% moisture was 850 rpm whereas the blower setting was 450 rpm for plots originally cut at 27% moisture. The dependent variable collected in 2024 consisted of TF seed yield. There was no ABG present in 2024; therefore, ABG seed yield was not collected.

In the summer of 2025, a second iteration of the harvesting experiment occurred on a first-year stand of TF in Corvallis, Oregon at the Hyslop Field Research Laboratory. This research field was sown in autumn 2024 with the cultivar '4th Millenium' sown on 45.7 cm row spacing at 13.5 kg ha⁻¹. The experiment was laid out as a randomised complete block design with nine treatments, and the experiment was replicated three times. A summary of the 2025 treatments is shown in Figure 2. Annual nitrogen inputs totalled 179 kg ha⁻¹. Herbicides were used to suppress dichotomous weeds. In 2025, the alternative harvesting method where the combine was used to cut the crop occurred only at the 28 cm height because it became apparent in 2024 that the 5 cm cutting height was not practical for the combine. In 2025, seed harvest occurred at three seed moistures (18%, 35%, and 45%) and two heights (5 and 28 cm). All harvesting settings were identical to the 2024 harvest with the exception that the blower speed was set to 450 rpm for all treatments when the combine pick-up header was used. Dependent variables in 2025 consisted of TF seed yield, ABG seed yield, and 1000 count seed weight of the TF seed crop.

Results and Discussion

In 2024, there was no evidence that TF seed yield was reduced when swathing occurred at either 43% or 27% seed moisture across the same swathing treatments or heights (Figure 1). These results were unexpected because the current recommended swathing seed moisture percentages in western Oregon for TF turfgrass type seed crops range from 35% to 45% (Silberstein et al., 2010). Swathing with the combine at 27% seed moisture at a 28 cm cutting height resulted in a higher seed yield compared to swathing with a traditional swather at 43% moisture at a 5 cm cutting height. These results suggest that there may be merit in exploring lower than recommended seed moisture percentages for swathing TF seed crops in western Oregon. This is also promising regarding adoption of direct harvesting by stakeholders, because a lower seed moisture content will require less fuel to dry the crop.

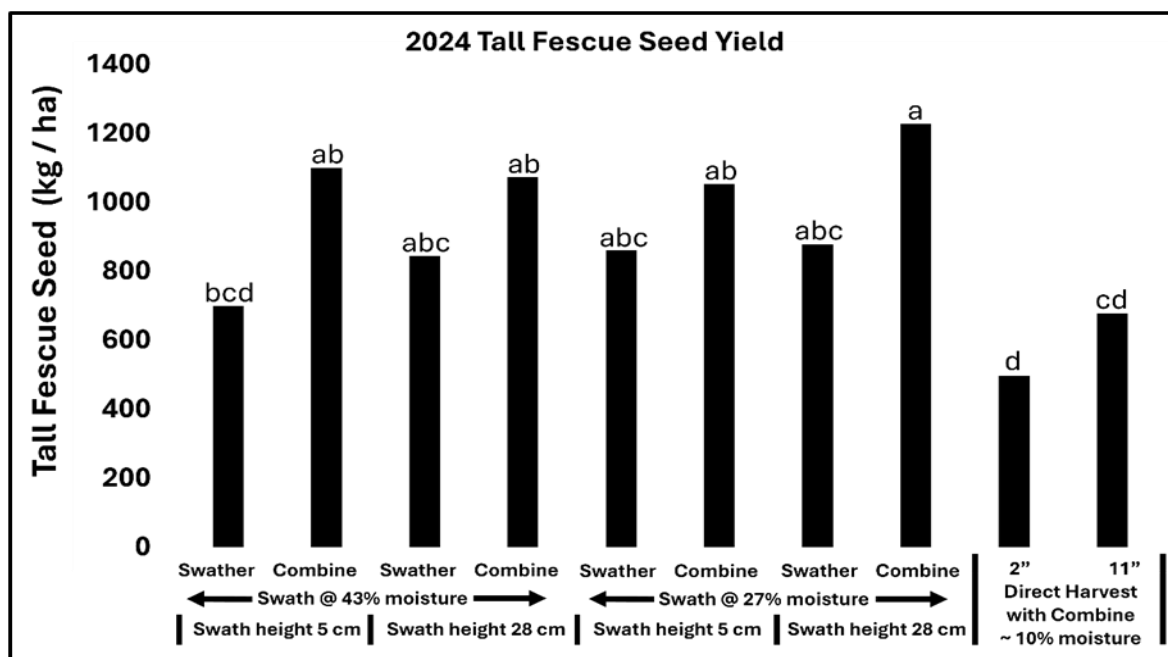


Figure 1. Average 2024 tall fescue ‘4th Millenium’ seed yield from a field established in autumn 2022 in Corvallis, Oregon. Harvest parameters included different seed moisture percentages, different swathing heights, and alternative harvesting techniques. Means not sharing the same letter are significantly different at an $\alpha \leq 0.05$ according to Tukey’s HSD pairwise comparison analysis.

In 2025, the first-year stand TF seed crop yield was meagre compared to more established stands; therefore, yield results remain preliminary and the influence of these same treatments on a more mature stand may result in different observations. Similar to 2024, TF seed yield was the same across seed harvest moistures when using a traditional swather at 5 cm or cutting with a combine at 28 cm (Figure 2). Treatments that used the swather at 28 cm resulted in a reduction in yield as the seed moisture percentage decreased. During the 2025 harvest, it was noted that the swather reel rotation speed increased at the 28 cm compared to the 5 cm height, caused by a machine setting on the swather that adjusted the reel speed based on mechanical resistance. It is speculated that this change in reel speed resulted in an increase in seed shatter at the lower seed moisture percentages. It is unclear why this did not occur in the 2024 harvest; however, it is possible that there was more biomass in 2024 with a more mature stand that was sown on 30.5 cm row spacing compared to a first-year stand in 2025 that was sown on 45.7 cm row spacing. Future research that explores a reduced reel speed at higher swathing heights is needed to elucidate these findings.

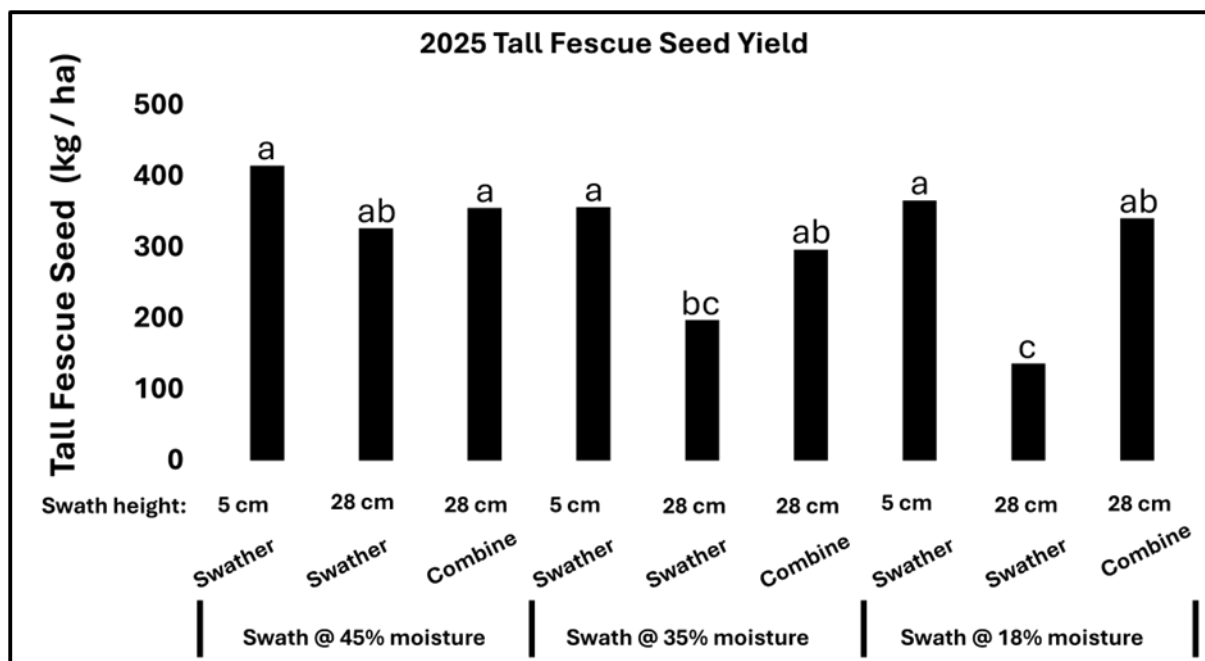


Figure 2. Average 2025 tall fescue ‘4th Millenium’ seed yield from a field established in fall 2024 in Corvallis, Oregon. Harvest parameters included different seed moisture percentages, different swathing heights, and alternative harvesting techniques. Means not sharing the same letter are significantly different at an $\alpha \leq 0.05$ according to Tukey’s HSD pairwise comparison analysis.

References

- Silberstein TB, Mellbye ME, Chastain TG and Young WC III. (2010) Using seed moisture as a harvest management tool. Oregon State University Extension Service. EM9030.
- Warwick SI (1979) The biology of Canadian weeds. 37 *Poa annua* L. *Canadian Journal of Plant Science* 59(4), 1053-1066. <https://doi.org/10.4141/cjps79-165>.

Can a fungal volatile organic compound mitigate abiotic and biotic stress impacts in perennial ryegrass grown for seed production?

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Abstract

Trichoderma fungi produce the volatile organic compound 6-pentyl-alpha-pyrone (6-PP) which can prime plant defences against abiotic and biotic stress. Coating seeds with 6-PP is one delivery option, and this method was used to assess the ability of 6-PP to mitigate abiotic (heat) and biotic (*Rhizoctonia solani* Kuhn) stress in perennial ryegrass (*Lolium perenne* L.). For seeds of cv. 'Base' coated with 6-PP at rates of 75 and 114 mM germination was 90% which did not differ from the untreated control, but germination for the 228 and 350 mM rates was 81% and 85%. 6-PP seed coating did not affect the seed to seedling transmission of six *Epichloë* endophytes but for two of the six seed to seed transmission was reduced by around 20%. 6-PP seed coating (75 and 114 mM) was then assessed as a primary strategy against the soil-borne pathogen *R. solani* in a glasshouse experiment. 6-PP significantly increased seedling emergence and reduced the disease impact caused by the pathogen, with the higher rate increasing root length by 77% and root volume by 131% over that of the pathogen control. 6-PP seed coating (75 mM) was finally used to assess the ability of the seed crop of cv. 'Base' produced to withstand heat stress (180°C h) at flowering or during seed development (70% seed moisture content). Plants grown from 6-PP coated seed had a 25% greater seed yield and 15% higher thousand seed weight than the control when the heat stress was applied at flowering, and the seeds produced had a germination of 82% cf. 45% for the control. When heat stress was applied during seed development the germination of seeds produced from the plants grown from 6-PP coated seed was 78% and that of the control was 48%. 6-PP applied as a seed coating enhanced stress resilience in perennial ryegrass and may offer a strategy for mitigating the negative impacts of climate change on seed yield and quality.

Keywords: disease suppression, *Epichloë* endophytes, germination, heat stress, seed coating, 6-PP

Introduction

Herbage seed growers will face increasing problems from the impacts of climate change induced abiotic and biotic stress on seed production (Hampton et al. 2016) and there is a need for solutions to support seed growers in facing these environmental challenges. One possible mechanism for mitigating these impacts is the interaction between plants and fungal volatile organic compounds (FVOC) produced by strains of the rhizosphere-inhabiting *Trichoderma* fungi (Moran-Diez et al. 2021). FVOCs are able to activate plant defence mechanisms (Poveda 2021). This defence priming (Conrath et al. 2015) can provide the plant with long-term resistance to abiotic and biotic stress (Moran-Diez et al. 2021).

The FVOC 6-pentyl-alpha-pyrone (6-PP), a ketone produced by several *Trichoderma* spp. can enhance plant stress resilience (Mendoza-Mendoza et al. 2024) and can be applied within a seed coating. This method was used to determine: i) whether 6-PP affected the germination of perennial ryegrass (*Lolium perenne* L.) or transmission of *Epichloë* endophytes, and ii) whether 6-PP could provide perennial ryegrass plants with resistance to biotic stress (from the soil-borne pathogen *Rhizoctonia solani* Kuhn) and abiotic stress (heat stress during flowering and during seed development).

Materials and Methods

Seeds of perennial ryegrass cultivar 'Base' were supplied by PGG Wrightson Seeds Ltd (Lincoln, New Zealand (NZ)) and used for the germination test and both stress experiments. For the *Epichloë* transmission assessments the seed lots listed in Table 1 were supplied by Barenbrug NZ (Courtenay, NZ).

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6-PP was purchased from Sigma-Aldrich (St. Louis, Missouri, USA). To ensure 6-PP would adhere to the seed coat, it was mixed with a polymer (Seedworx Stix 1, Centor Oceania, Victoria, Australia) and ethanol as a solvent (Echaide Aquino 2024) before the seeds were added. This was followed by gentle agitation by hand to coat the seeds which were then air-dried in a laminar flow cabinet prior to storage at 4°C until use.

Table 1. Effect of 6-PP seed treatment on *Epichloë* endophyte seed to seedling and seed to seed transmission in perennial ryegrass seed lots.

Perennial ryegrass cultivar ¹	<i>Epichloë</i> endophyte ²	Transmission (%)			
		Seed to seedling		Seed to seed	
		Control ³	6-PP ⁴	Control ³	6-PP ⁴
Trojan	NEA	56	52	71	53*
Array	NEA2	85	90	81	63*
Maxsyn	NEA4	76	86	71	63
Maxsyn	NEA12	88	76	81	85
Governor	AR1	78	65	68	74
Governor	AR37	65	84	50	73

¹Seeds kindly supplied by Barenbrug NZ; ²proprietary endophyte strain; ³untreated seed; ⁴75 mM 6-PP; *significantly ($P < 0.05$) lower than the control.

1. Germination test. 6-PP was applied to seeds at rates of 75, 114, 228 and 350 mM. Untreated seeds and polymer only coated seeds served as the controls. For each treatment four replicates of 50 seeds were sown into trays containing seed raising mix. The trays were then placed in a glasshouse at a temperature which averaged 25°C and germination was assessed using an internationally agreed method at 12-days after sowing (DAS) (ISTA 2024).

2. Endophyte transmission.

2.1 Seed to seedling. 6-PP was applied to seeds at 75 mM with untreated seeds as the control. Endophyte transmission to seedlings was tested at the PGG Wrightson Seed Laboratory, Lincoln using the grow-out test (immunoblotting) (Hillis 2019; Rinklake et al. 2020).

2.2. Seed to seed. For each of the six cultivars (Table 1), 6-PP was applied to seeds at 75 mM with untreated seeds as the control. In February 2024, eight replicates for each cultivar x treatment were sown into 4 L pots containing potting mix (10 seeds per pot, later thinned to six plants pot⁻¹). The pots were placed in a shade house, watered as required, and left to produce seeds. At maturity seeds were hand harvested and tested for the presence of viable endophyte using the grow-out method.

3. Biotic stress. A colony of *Rhizoctonia solani* (strain Rs73-13b) was used to prepare a wheat-bran based inoculum (Kandula et al. 2015). Sterilised pots (2 L) were filled with 1.75 L potting mix, and for all except the no pathogen control, *R. solani* inoculated wheat bran (0.5 g 100 g⁻¹ potting mix) was added. For each treatment (Table 2) there were eight replicates with 10 seeds sown per pot. At 40 DAS, plant number per pot was recorded. Plants were removed from the pots, the roots washed and scanned using a Win RHIZO Scanner (Regent Instruments Inc, California, USA); root length, surface area and volume were measured before root dry weight was recorded.

Table 2. Effect of 6-PP seed treatment on *Rhizoctonia solani*¹ impacts on perennial ryegrass plant performance 40 days after sowing.

Treatment	Pathogen ²	Plant number per pot ³	Root ⁴		
			Length (cm)	Surface area (cm ³)	Volume (cm ³)
Untreated seed	-	8.8 a	626.7 a ⁵	117.5 a	1.8 a
Untreated seed	+	1.9 c	271.8 c	45.7 b	0.6 b
75 mM 6-PP	+	4.0 b	472.9 ab	80.5 ab	1.1 ab
114 mM 6-PP	+	5.4 b	479.9 ab	90.7 a	1.5 a

¹Strain Rs 73-13b; ²pathogen absent (-) or present (+) in growing medium; ³out of 10 seeds sown per pot; ⁴all root data are means per pot; ⁵data with different letters indicate significantly different means at $P < 0.05$.

4. Abiotic stress. Using the same method as described in 2.2, 16 pots containing plants grown from 6-PP treated seed (75 mM) and 16 pots containing plants grown from untreated seed were prepared. At mid flowering eight pots from each treatment were transferred to the NZ Biotron and subjected to 180°C hours of heat stress (30°C day/25°C night) (Rashid et al. 2017) before being returned to the shade house. The second set of plants were moved to the Biotron during seed development (ca. 70% seed moisture content) and subjected to the same period of heat stress before being returned to the shade house. Before, during and after the heat stress leaf relative water content and chlorophyll content were recorded (Echaide Aquino 2024). At maturity seeds were hand harvested, hand cleaned and weighed to determine seed yield and thousand seed weight. Germination was then tested using the between paper method (ISTA 2024).

Results and Discussion

1. Germination. For the 75 and 114 mM application rates, germination was 90% and this did not differ from that of the two controls (data not presented). However, at the 228 and 350 mM application rates, germination was reduced (to 81% and 85% respectively), because of the production of abnormal seedlings (stunted roots). A reduction in maize seed germination when seeds were placed on filter paper impregnated with 6-PP has been reported (El-Hasan et al. 2009). However, these authors used radicle emergence at 4-days to assess germination, a time insufficient to accurately assess any effects of 6-PP on normal seedling development (ISTA 2024). Chemical seed treatment induced phytotoxicity can result in abnormal root development, with the extent depending on the active ingredient, application rate and seed lot quality (Taylor & Salanenke 2012). 6-PP at 75 and 114 mM did not affect the germination of wheat, prairie grass, forage rape or radiata pine (Hampton unpub.).

2. Endophyte transmission. 6-PP seed treatment had no effect on the seed to seedling transmission of the *Epichloë* endophytes assessed (Table 1). For seed-to-seed transmission 6-PP reduced transmission of the NEA and NEA2 endophytes by 18% but transmission for the other four endophytes did not differ from that of the controls (Table 1). With its known anti-fungal activities there was a possibility that 6-PP would have a negative impact on *Epichloë*, but currently there is no evidence of any negative effect of 6-PP on beneficial fungi (Mendoza-Mendoza et al. 2024). Zhu et al. (2023) reported that 6-PP stimulated beneficial rhizosphere microbes which promote plant growth, and 6-PP had a synergistic interaction with *Rhizobia*, which resulted in increased nodule numbers in white clover (Hampton, unpub.).

3. Biotic stress. At 40 DAS the number of plants present per pot (Table 2) were the same as the number of seedlings which had emerged, as no post-emergence seedling death occurred. For the untreated seeds the pathogen reduced plant number by 78%; the two 6-PP treatments significantly increased plant number over that of the pathogen control (Table 2). The 114 mM 6-PP treatment significantly increased root length, root surface area and root volume over that of the pathogen control (Table 2).

6-PP is known to have potent anti-pathogen activities through disruption of pathogen cell wall membranes, inhibition of enzymes critical for plant pathogen metabolic processes, induction of oxidative stress and alteration of gene expression (Mendoza-Mendoza et al. 2024). 6-PP directly

inhibits *R. solani* mycelium growth and reduces sclerotia formation (van Zijll de Jong et al. 2023). 6-PP is also known to prime plant defence systems conferring resistance to pathogen attack through the upregulation of defence related genes (Mendoza-Mendoza et al., 2024). The significant increases in root growth for plants grown from 6-PP treated seeds is similar to previous reports and this has been related to 6-PP's auxin-like activity (Garnica- Vergara et al. 2016).

4. Abiotic stress. For plants which received the heat stress during flowering leaf relative water content was significantly greater for the 6-PP plants before but not during or after the stress, while leaf chlorophyll content (SPAD value) was greater during stress but not before or after the stress (Table 3). For the heat stress during seed development 6-PP increased leaf relative water content before the stress only, but SPAD values were increased during the stress (Table 3). 6-PP positively affects photosynthesis effectiveness and plant water use efficiency (Mendoza-Mendoza et al. 2024). Echaide-Aquino (2024) reported that dwarf bean leaf chlorophyll content and water content were increased during drought stress in plants grown from 6-PP treated seeds.

Table 3. Effect of 6-PP seed treatment on two perennial ryegrass plant stress parameters following heat stress¹

Heat stress							
Heat stress at flowering				Heat stress during seed development			
1. Leaf relative water content (%)							
Treatment	Before stress	During stress	After stress	Before stress	During stress	After stress	
Control	70.9 a ²	76.6 a	83.4 a	59.5 a	62.3 a	72.0 a	
6-PP	82.2 b	75.4 a	78.2 a	76.3 b	61.8 a	74.1 a	
2. Photosynthetic capacity (SPAD value)							
Treatment	Before stress	During stress	After stress	Before stress	During stress	After stress	
Control	41.6 a	31.1 a	37.5 a	31.3 a	31.8 a	23.0 a	
6-PP	44.0 a	41.0 b	36.8 a	26.6 a	39.5 b	20.0 a	

¹heat stress (180°C/h) applied either during peak flowering or at around 70% seed moisture content; ²data with different letters indicate significantly different means at $P < 0.05$.

For plants which received the heat stress during flowering, 6-PP increased both FD and MD seed yields by 25% as dressing losses did not differ between the treatments. Thousand seed weight was also significantly greater for the 6-PP treatment. However, seed yield and thousand seed weight did not differ when the heat stress was applied during seed development (Table 4). For both stress timings, the germination of seeds produced from plants grown from 6-PP treated seeds was significantly greater than for seeds of the control plants; stress during flowering = 82% (6-PP) and 45% (control); stress during seed development = 78% (6-PP) and 48% (control). The reason for the increased seed yield for 6-PP plants following heat stress during flowering is not clear. Heat stress has a negative effect on pollen viability, germination and tube growth (Hampton et al. 2016), but whether 6-PP –induced stress priming was able to reduce the impact of heat stress on pollen quality remains to be determined. Whatever the mechanism, 6-PP plants were better able to cope with the heat stress than the control plants. 6-PP application increases the activity of antioxidant enzymes including superoxide dismutase and peroxidase and also non-enzymatic antioxidants including proline which are crucial for neutralising the damaging reactive oxygen species (ROS) that accumulate in plants during heat stress. Rashid et al. (2017) reported that loss of seed quality following heat stress was correlated with a decrease in antioxidant enzymes and increase in ROS in the seeds. 6-PP also stimulates the expression of defence related plant genes (Moran-Diaz et al. 2021), although the molecular mechanisms through which 6-PP exerts this bioactivity is largely unknown (Mendoza-Mendoza et al. 2025).

These results indicate that 6-PP can be applied as a seed coating to enhance stress resilience in perennial ryegrass, and therefore may offer a strategy for mitigating the negative effects of climate change on seed yield and quality.

Table 4. Effect of 6-PP seed treatment on perennial ryegrass seed yield and quality following heat stress¹

Heat stress at flowering			Heat stress during seed development			
1. Seed yield (g/pot)						
Treatment	FD ² (g)	MD ³ (g)	Dressing loss (%)	FD ² (g)	MD ³ (g)	Dressing loss (%)
Control	11.21 a ⁴	9.43 a	15 a	13.48 a	11.26 a	16 a
/6-PP	14.14 b	12.03 b	15 a	14.43 a	12.44 a	13 a
2. Seed quality						
Treatment	TSW ⁵ (g)	Germination (%)		TSW ⁵ (g)	Germination (%)	
Control	2.69 a	45 a		2.82 a	48 a	
6-PP	3.10 b	82 b		2.74 a	78 b	

¹heat stress (180°C/h) applied either during peak flowering or at around 70% seed moisture content; ²FD = field dressed; ³MD = machine dressed; ⁴data with different letters indicate significantly different means at $P < 0.05$.

⁵TSW = thousand seed weight.

References

- Conrath U, Beckers GJM, Langenbach CJG and Jaskiewicz MR (2015) Priming for enhanced defence. *Annual Review of Phytopathology* **53**, 97-119.
- Echaide Aquino JF (2024) Can the presence of a fungal volatile organic compound improve plant resilience to abiotic stress? Unpublished PhD thesis, Lincoln University, Canterbury, New Zealand.
- El-Hasan A and Buchenauer H (2009) Actions of 6-pentyl- α -pyrone in controlling seedling blight incited by *Fusarium moniliforme* and inducing defence responses in maize. *Journal of Phytopathology* **156**, 79-87.
- Garnica-Vergara A, Barrera-Ortiz S et al. (2016) The volatile 6-pentyl-2H-pyran-2-one from *Trichoderma atroviride* regulates *Aribidopsis thaliana* root morphogenesis via auxin signalling and ETHYLENE INSENSITIVE 2 functioning. *New Phytologist* **209**, 1496-1512.
- Hampton JG, Conner AJ, Boelt B, Chastain TG and Rolston MP (2016) Climate change: seed production and options for adaptation. *Agriculture* **6**, 33-48.
- Hillis S (2019) Transmission and survival of perennial ryegrass endophyte during field based seed production. Unpublished MAgSC thesis, Lincoln University, Canterbury, New Zealand.
- ISTA.(2024) International Rules for Seed Testing. International Seed Testing Association, Wallisellen, Switzerland.
- Kandula DRW, Jones EE, Stewart A, McLean KL and Hampton JG (2015) *Trichoderma* species for biocontrol of soil-borne plant pathogens of pasture species. *Biocontrol Science and Technology* **25**, 1052-1069.
- Mendoza-Mendoza A, Esquivel-Naranjo EU et al. (2024) Uncovering the multifaceted properties of 6-pentyl- α -pyrone for control of plant pathogens. *Frontiers in Plant Science* **15**, 1420068.
- Moran-Diez ME, Martinez de Alba AE, Rubio MB and Hermosa R (2021) *Trichoderma* and the plant heritable priming response. *Journal of Fungi* **7**, 318.
- Poveda J (2021) Beneficial effects of microbial volatile organic compounds (MVOCS) in plants. *Applied Soil Ecology* **168**, 104118.
- Rashid M, Hampton JG, Rolston MP, Khan KM and Saville DJ (2017) Heat stress during seed development affects forage brassica (*Brassica napus* L.) seed quality. *Journal of Agronomy and Crop Science*, 1-8.

- Rinklake I, James A, Brownfield L et al. (2020) Relative *Epichloë* endophyte fungal biomass in ryegrass tillers grown pre and post vernalization. *Agronomy New Zealand* **50**, 95-107.
- Taylor AG and Salanenke YA (2012) Seed treatments ameliorated tracer uptake. *Seed Science Research Suppl.* **81**, S86-S90.
- Van Zijl de Jong E, Kandula J, Rostas M et al. (2023) Fungistatic activity mediated by volatile organic compounds is isolate dependant in *Trichoderma* sp. "*atroviride* B". *Journal of Fungi* **91**, 20.
- Zhu Z, Wu G, Deng R et al. (2023) Spatiotemporal biocontrol and rhizosphere microbiome analysis of *Fusarium* wilt of banana. *Communications in Biology* **6**, 27.

Assessing environmental drivers of insect pests in grass seed crops to develop areawide decision support tools

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Abstract

Successful mitigation of agricultural insect pests depends on integrated pest management practices incorporating multiple techniques for effective population management below economically damaging levels. Pest surveillance remains the cornerstone of IPM programs, enabling appropriately timed management action. Phenological models using weather data are commonly used as decision support tools to predict the timing of ‘when’ economically important life stages are expected to occur. Furthermore, geostatistical models that consider landscape-level variation in environmental drivers of pest densities may inform ‘where’ outbreaks are expected to occur along spatial gradients of abiotic and biotic risk factors. Thus, predictors of pest populations can be leveraged to generate spatiotemporal risk assessments. Migratory and overwintering moth species in the family Noctuidae (order Lepidoptera) comprise a complex of serious pests that threaten the profitability of grasses grown for seed in the Willamette Valley, Oregon, USA, as well as seed production globally. Noctuid pests, including black cutworm, true armyworm, and winter cutworm, inflict crop damage by direct feeding on plant crowns and roots in the larval developmental stage. Management action with foliar insecticides is most effective when larvae are immature (early instar stage) due to insecticide susceptibility and pest behaviour. For noctuid pests (and other priority pests) in grass seed systems, real-time phenological models can provide field practitioners with information to better allocate pest monitoring and management resources to reduce input costs.

Keywords: insect phenology, predictive modelling, decision support, population dynamics, areawide pest management

Introduction

Migratory and overwintering moth pests within the Noctuidae family pose a substantial threat to agricultural production globally due, in part, to their strong flight capability, polyphagous feeding behaviour, broad host range, and multivoltine life history (Showers 1997). Synthetic pesticides are widely utilised for pest control worldwide and are a primary strategy for managing noctuid pests. An array of factors contributes to insecticide resistance evolution, including pesticide-based programs that rely on excessive applications of broad-spectrum insecticides and limited rotations with other modes of action. Pest management strategies employed within an areawide framework may offer compounded benefits regarding pest suppression and insecticide resistance management by reducing regional population densities of highly mobile pests, lowering reliance on chemical inputs for crop protection (Brewer & Dorman 2025). A key tenet of effective areawide pest suppression and insecticide resistance management involves pest surveillance to determine when populations are present at susceptible life stage(s) and above economically damaging levels, enabling appropriately timed management action. Regionally validated phenology models can inform decision support platforms that predict the timing of susceptible pest life stages across broad spatial scales (Dorman et al. 2024). Furthermore, spatialised phenological predictions using real-time environmental data can increase the resolution, efficiency, and usability of pest forecasting tools for crop management implementers (Barker et al. 2020).

Insects exhibit a positive correlation between their rate of development and increasing temperature. Insect phenology and temperature-dependent development vary regionally based on local weather patterns and crop hosts (Valtonen et al. 2011). The relationship between temperature and insect

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phenology is often used to develop degree-day models that predict insect life stages utilizing daily heat summations and lower and upper developmental thresholds. Understanding the developmental timeline of key noctuid pests (black cutworm, true armyworm, and winter cutworm) in grasses grown for seed production, a primary host with the greatest land area across the Willamette Valley compared to other crop hosts, is critical for developing regionally specific phenology models utilizing heat summation calculations.

As such, our objectives were to: 1) develop crop host and species-specific phenology models to predict the timing of susceptible noctuid life stages across various armyworm and cutworm species, and 2) incorporate models into a decision support tool to share real-time forecasts and recommendations with growers and crop consultants.

Materials and Methods

Temperature-dependent development. To evaluate the temperature-dependent development of three critical noctuid pests (black cutworm, true armyworm, and winter cutworm), laboratory colonies were established from field populations and maintained at $22 \pm 1^\circ\text{C}$, $60 \pm 5\%$ RH, and 16L:8D photoperiod. Neonate larvae, within 24-hours from egg hatch, were placed in individual incubators (Percival® model DR-36VL) set to one of five constant temperatures, ranging from 10 to 32°C ($60 \pm 5\%$ RH and 16L:8D photoperiod) based on preliminary experiments. Two diet treatments were evaluated at each constant temperature, including an artificial diet (soy wheat-germ diet) and a perennial ryegrass diet (cultivar 'Fastball 3GL') consisting of fresh grass tissue clippings. For both diet types, enough artificial diet (approximately 0.7 g) or grass clippings (approximately 1.2 g) were placed in 1 oz plastic cups to cover the bottom of the cup. Single neonate larvae were confined in individual cups (one specimen per cup) with aerated lids wrapped with thrips screen ($150 \times 150 \mu\text{m}$) to prevent escape. Temperature and RH were recorded in 30-minute intervals with two iButton® data loggers (Thermochron Temperature Loggers; Sydney, Australia) placed in additional lidded plastic cups with both artificial diet and grass seed diet types to replicate the test environment. Diet was replaced at least three times per week, and data was recorded daily. Data collection for the larval life stage included the date, current larval instar (evaluated based on exuviae and head capsule width), and mortality if present. Larvae were allowed to pupate in 1 oz plastic cups; the date of pupation was recorded, and pupae were sexed before adult emergence.

Following adult emergence, individual moths were placed in nylon mesh enclosures ($23 \times 23 \times 28 \text{ cm}$) and fed a 10% sucrose solution dispensed on a cotton ball. The temperature, RH, and photoperiod regimen remained the same as previously described for larval and pupal development. When female and male adults had emerged, single female-male pairs were placed in one enclosure. Strips of nylon mesh and paper were affixed to the top of the enclosure to serve as an oviposition substrate. The date of oviposition, adult mortality, total number of eggs, and subsequent egg hatch (number of viable eggs hatched per matriline) were recorded daily.

Adult flight phenology and biofix prediction. To predict flight phenology of the adult life stage for each species and determine the biofix date for degree day accumulation, an attract-based trap network was deployed in commercial grass seed fields (perennial ryegrass, tall fescue, Kentucky bluegrass) across the Willamette Valley and eastern Oregon. Over 100 commercial grass seed farms across four years (2021 to 2024) were monitored weekly for a minimum of ten consecutive weeks from May through October. Adult abundance was quantified using one green funnel trap (Unitrap™) baited with a female sex pheromone lure or black light trap per field, positioned $>15 \text{ m}$ from field borders. Insecticidal pest strips (10% dichlorvos active ingredient) placed in traps to prevent escape were replaced every three weeks. Collected specimens were transferred to the laboratory to confirm identification using Leica S9i stereomicroscopes (Leica Microsystems, Wetzlar, Germany).

Data analysis. All data analyses were performed in R (v4.3.1 R Core Team). The temperature-dependent development rate for each life stage by diet type was determined using simple linear regression; development rates were also evaluated for combined data across diet type.

Seasonal adult flight phenology of adults was analysed with nonlinear logistic regression models using generalised least squares in the *nlme* package. All site years included in the study had >20 total seasonal trap captures across respective sampling windows. Phenological models were developed using daily minimum and maximum temperatures and three common degree day calculation methods, including simple average, single triangle, and single sine. Daily land surface temperature data was extracted from PRISM gridded rasters in 800 × 800 m resolution using the *prism* package. Adult count data was converted to the seasonal cumulative proportion of trap catch at each site-year location.

A range of degree day accumulation start dates were evaluated from 1 January to 1 May in five-day increments, totalling >70 candidate models across the three degree day calculation methods evaluated for each species. The accuracy and performance of candidate models were compared using mean absolute error (MAE), root mean squared error (RMSE), and pseudo coefficient of determination (R^2) metric that estimates the fit of nonlinear regression models. Optimal models for each degree day calculation method (simple average, single triangle, single sine) were cross validated by randomly splitting the data by year into ‘test’ and ‘train’ datasets. Model predictions based on the ‘train’ data were evaluated against ‘test’ observations using the *car* package. Simple linear regression was used to evaluate the accuracy and model fit of predictions compared to observed values across degree day calculation methods (with equal minimum temperature threshold and degree day start date parameters) using RMSE and R^2 metrics, respectively, in the *plm* package.

Results and Discussion

For detailed statistical methodology and data interpretation, refer to Slone and Dorman et al. (2025). Refereed papers on true armyworm and winter cutworm phenology, as well as landscape composition and configuration predictors of noctuid population dynamics in Oregon using Bayesian inference and gut-content analyses, are forthcoming.

Temperature-dependent development experiments provide information on optimal survival and development rates (e.g., 27 and 32°C for black cutworm). Additionally, mean lower and upper developmental thresholds evaluated with controlled laboratory experiments are critical for determining degree-day requirements for life stage-specific phenological events. For black cutworm, we estimated thermal requirements of 76, 372, 187, 648 degree days (with a lower threshold of 9.8°C) for egg, larvae, pupae, and oviposition to adult eclosion life stages, respectively. Phenology model parameters can be used in conjunction with real-time weather data to generate spatiotemporal pest forecasts used by field implementers (growers and crop consultants) to improve management timing of noctuid (and other) pests. Additionally, we found development times varied significantly with different food sources, highlighting the importance of regionally specific and crop-specific models to ensure phenological forecasts for noctuid pests are maximally accurate.

Findings from temperature-dependent development experiments and modelling of noctuid adult seasonal flight can be incorporated into general and crop-specific phenology models using the predicted median flight as the biofix for adult arrival (black cutworm and true armyworm) or emergence from overwintering sites (winter cutworm) and oviposition (egg-laying) to initiate degree day accumulation for subsequent life stages. Furthermore, model parameters can be incorporated into a decision support platform that shares real-time areawide interpolation risk maps, species-specific life stages across time and space, using high-resolution gridded weather data. Space-time forecasts of priority pests using an areawide pest management framework are critical for coordinating pest surveillance and management resources to promote sustainable pest management practices and achieve optimal pest suppression (population densities below economic concern) while reducing the environmental footprint and economic input costs associated with pesticide overuse.

Phenology models for noctuid pests and other economic pests in grass seed, vegetable, and orchard crop systems in Oregon have been incorporated into a practical decision support tool for crop consultants and growers to readily access in the field to serve as a support tool to assist with pest management decisions on their farms (web link: <https://oregonpests-usdaars.hub.arcgis.com/pages/pest-forecasts>). The pest phenology application is featured on a

publicly available web platform (Oregon Pest Monitoring Network) that shares pest monitoring resources with the agricultural community statewide. These predictions combine real-time weather data with pest biology information (e.g., seasonal adult flight phenology, life-stage specific developmental thresholds, and developmental rates on affected crops) to visualise the location and timing of pest activity (Figures 1A and 1B). Predictions on the timing of pest life stages based on cumulative growing degree day thresholds are spatialised using land surface temperature raster data (800 m-resolution) on interactive maps, allowing end-users to zoom and scroll on pest predictions in various regions across Oregon (Figure 1B).

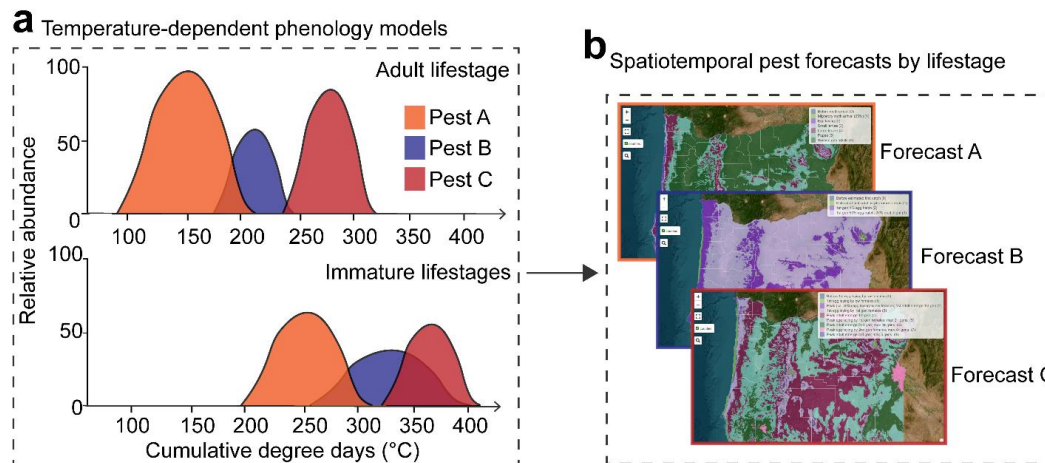


Figure 1. Pest phenology model parameters for key pests (A); decision support tool translating models into spatialised pest forecasts with a colour legend to indicate predicted life stages (B)

Temperature-dependent phenology forecasts strictly based on land surface temperature data are excellent tools for spatiotemporal predictions of phenological events. Additional environmental variables (precipitation, wind speed, soil temperature, air pressure), as well as the landscape scale differences in cropland composition and configuration of plant hosts, can affect the severity and interannual variation in pest outbreaks. Statistical approaches for developing pest-specific risk (severity) forecasts (for example, Bayesian inference, machine learning approaches), in addition to extracting real-time spatial data on multiple environmental predictors, are computationally demanding and difficult to incorporate into a practical, high-speed application for agricultural decision makers. More advanced models are particularly helpful, however, considering range expansion and interannual migration patterns, respectively, are driven by multiple environmental cues. Research to develop spatiotemporal forecasts and decision aid pipelines with additional abiotic and biotic risk factors is ongoing to predict the severity of pest outbreaks.

References

- Barker BS, Coop L, Wepprich T, Grevstad F and Cook G (2020) DDRP: Real-time phenology and climatic suitability modeling of invasive insects. *PLoS One*. 15(12):e0244005. <https://doi.org/10.1371/journal.pone.0244005>.
- Brewer MJ and Dorman SJ (2025) Editorial: Areawide pest management and agroecosystem resilience to suppress invasive insects. *Front. Insect Sci.* 5, 1605737. <https://doi.org/10.3389/finsc.2025.1605737>.
- Dorman SJ, Kaur N, Anderson NP, Sim RE, Tanner KC, Walenta DL and Cooper WR (2024) Flight phenology and landscape predictors of invasive *Coleophora deauratella* populations in Oregon and New Zealand red clover. *J. Pest Sci.* 97, 631–643. <https://doi.org/10.1007/s10340-023-01684-8>.
- Showers WB (1976) Migratory ecology of the black cutworm. *Annu. Rev. Entomol.* 42, 393–425. <https://doi.org/10.1146/annurev.ento.42.1.393>.

- Slone E, Green J, Kaur N, Walenta DL, Anderson NP, Cruse C and Dorman SJ (2025) Forecasting the seasonal phenology of *Agrotis ipsilon* in Oregon grass seed and vegetable agroecosystems. *Front. Insect Sci.* **4**, 1505524. <https://doi.org/10.3389/finsc.2024.1505524>.
- Valtonen A, Ayres MP, Roininen H, Pöyry J, Leinonen R (2011) Environmental controls on the phenology of moths: predicting plasticity and constraint under climate change. *Oecologia* **165**, 237–248. <https://doi.org/10.1007/s00442-010-1789-8>.

Occurrence and management of aphid-vectored yellow dwarf viruses in perennial grass seed crops

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Abstract

Epidemiology and management of aphid-transmitted yellow dwarf viruses (YDVs) have received international attention in small grains, but research regarding YDVs in grass seed production is limited. An integrated pest management program is needed to reduce the impact of YDVs in grass seed crops that are grown for more than one year. The objectives of this work were to: 1) survey commercial grass seed production fields to determine spatiotemporal virus composition, 2) evaluate the effects of nitrogen (N) fertiliser rate, and the timing and frequency of foliar insecticide applications on aphid abundance, YDV disease incidence, and seed yield in two perennial ryegrass cultivars, and 3) develop high-throughput phenotyping methods to screen cultivars for host plant resistance. To determine the incidence and diversity of YDVs, perennial ryegrass (n=20) and tall fescue (n=30) seed fields in Oregon were surveyed in 2021-2022. In 82% of fields, a *Luteovirus*-type YDV was detected, and 65% had detection of a *Polerovirus*-type YDV. In small-plot field trials conducted from 2021 to 2024, high N rates increased YDV incidence in perennial ryegrass. Seed yield was greatest for the less susceptible cultivar when protected with one insecticide treatment per season. A higher-than-recommended N rate did not increase seed yield across treatment combinations in first-year stands but did increase seed yields in second and third-year stands when YDV infection was >50%. Phenotyping methods were evaluated to assess potential host-plant resistance to YDVs using perennial ryegrass cultivars (n=27) with high-throughput automated video tracking for aphid behaviours that may confer resistance, and compared to traditional phenotyping methods. Several cultivars showed potential tolerance to YDVs. This research provides new knowledge of the spatial composition of aphid-transmitted YDVs, integrated pest management guidelines, and high-throughput methods for breeding programs to develop cultivars that are resistant to YDVs.

Keywords: aphids, yellow dwarf virus, perennial ryegrass, tall fescue, high-throughput phenotyping

Introduction

Yellow dwarf viruses (YDVs) are considered one of the most damaging viruses affecting cereal crops worldwide (Walls et al. 2019), but little attention has been given to understanding the epidemiology and identifying best management practices for YDVs in cool-season grass seed crops. Yellow dwarf viruses comprise two virus genera, *Luteoviruses* (commonly known as barley yellow dwarf viruses) and *Poleroviruses* (commonly known as cereal yellow dwarf viruses), that are composed of multiple species (Miller & Lozier 2022). These viruses are vectored by multiple species of aphids, which feed on infected phloem to acquire the virus before transmitting it to non-infected plants through salivary gland excretions. Visual symptoms of YDV infection include leaf discoloration, plant dwarfing, a reduction in root biomass, and delayed flowering or sterile seed heads (Aradottir & Crespo-Herrera 2021). Symptomology in noncereal grasses is often not as visually pronounced, and therefore, it has been difficult to know exactly how widespread YDVs are in commercial grass seed fields and how often they are responsible for reductions in seed yield.

In Oregon (USA), the world's largest producer of cool-season grass seed, it is common to produce perennial ryegrass (*Lolium perenne* L.) and tall fescue [*Schedonorus phoenix* (Scop.) Holub] seed crops for two to five years. Thus, grass seed crops in Oregon are believed to be more susceptible to yield reductions from YDVs, as there are multiple seasons for virus transmission to occur, as compared to other regions of the world where grass seed crops are produced for only one year. In Oregon, reductions in stand longevity, particularly in perennial ryegrass, have been attributed, in part, to YDV.

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However, quantifying the precise impact of YDV in perennial grass seed crops has been difficult due to the absence of focused research on YDV and the presence of asymptomatic infections.

In most temperate grass seed production regions, there are two prominent aphid alate flights, one in spring and one in autumn. Flight is generally dependent on a minimum temperature threshold of 15°C in the spring, and virus transmission is significantly reduced when temperatures drop below 12°C in the autumn (Lowles et al. 1996). Delayed planting in the autumn and use of insecticidal seed treatments have been successful in reducing the occurrence of YDVs and associated grain yield losses. However, the perennial nature of grass seed crops in Oregon, combined with the inability to delay planting and lack of registered insecticidal seed treatments, poses several challenges to managing aphid-transmitted YDVs in this system. Industry observations have concluded that a significant percentage of commercial grass seed fields are routinely treated with foliar insecticide applications to reduce aphid populations, which alone are not considered an economically damaging pest, to reduce the occurrence of YDV infections. The seasonal phenology of aphid species that transmit YDVs in grass seed crops complicates this unvalidated strategy, as prolonged exposure to aphids, namely *Rhopalosiphum padi* (L.), from early spring through late autumn, limits the utility of chemical control with foliar insecticides. Thus, host plant resistance and selecting phenotypes resistant to the aphid-YDV pathosystem is likely the most effective management approach to manage YDVs in grass seed cropping systems. The objectives of this work were to: 1) conduct surveys on commercial grass seed farms to determine spatiotemporal virus composition, 2) evaluate the effects of nitrogen fertiliser rate, and the timing and frequency of foliar insecticide applications on aphid abundance, YDV disease incidence, and seed yield in two perennial ryegrass cultivars, and 3) develop high-throughput phenotyping methods to screen cultivars for host plant resistance.

Materials and Methods

Spatiotemporal sampling. Commercial perennial ryegrass (n=20) and tall fescue (n=30) seed fields in Oregon were surveyed in 2021-2022 for YDV incidence and diversity, alate aphid population densities, and aphid virus status to generate spatiotemporal data on the virus-vector system. To sample the abundance of aphid alates immigrating into the fields, yellow sticky traps (15.6 × 20.7 cm) were mounted approximately one meter above the ground, on both the north and south-facing field edges. Traps were checked weekly for eight weeks, and samples were stored at -20°C until counts were performed with a stereomicroscope. Five aphids per card were selected for YDV testing.

To assess YDV incidence in perennial ryegrass and tall fescue plants, each field was divided into four roughly equal-sized quadrants, and five random leaves from five representative plants every 10 m, along a 100 m transect, were collected. Samples were stored at -80 °C until processing. To extract total nucleic acids, the CTAB method (Doyle & Doyle 1990) and the Dellaporta method (Dellaporta et al. 1983) were used for the aphid and plant leaf samples, respectively. Multiple sets of multiplex RT-PCR were conducted, and samples were sequenced to identify YDV genus and species (Rivedal et al. 2024).

Evaluating field management strategies. Two adjacent field trials were conducted over three crop seasons (2021-2024) at Oregon State University's Hyslop Experimental Farm. Turf-type perennial ryegrass cultivars 'Top Gun II' and 'Fastball 3GL' were planted in September 2021 and managed as an irrigated crop. The experimental design for both trials was a split-plot, randomised block design with four replications. Main plots (15.7 × 15.2 m) were spring-applied nitrogen (N), and subplots (3.4 × 15.2 m) were foliar insecticide timing treatments.

Spring N treatments were applied just prior to stem elongation (BBCH 29) at two rates: 135 and 225 kg N ha⁻¹. The labelled rate of flupyradifuron insecticide (Sivanto Prime®, Bayer Crop Science) was applied at five timings: untreated control (no insecticide), autumn only, spring only, autumn + spring, and autumn + spring + summer. Seed was harvested by swathage with a modified small-plot swather when seed moisture content reached 35-43%. Dried windrows were threshed with a plot combine, and seed was dressed and weighed to determine clean seed yield.

Movement of aphids into the trial area was monitored weekly for the entirety of the three-year period using outward-facing yellow sticky traps placed on the field edges. After two consecutive weeks of

aphid alate detection on the sticky card traps, aphid populations within plots were assessed using sweep net sampling (25 pendulum swings sweep⁻¹) for five to eight weeks in spring and autumn. Plant samples were collected to evaluate the presence of YDVs by collecting five leaf samples at five random points within each plot. Samples were collected one to five days before and 30 days following each insecticide application. Nucleic acid extraction and PCR assays for both the aphid and plant leaf samples were performed according to the procedures previously described.

Phenotyping methods to determine plant host resistance. Local populations of *R. padi* aphids were collected from commercial grass seed farms and allowed to asexually reproduce to form a greenhouse colony. A subset of the colony was transferred to new perennial ryegrass plants every two weeks to form a non-viruliferous (YDV-) colony. Select plants that were YDV+ were removed from the field, potted in the greenhouse, and the aphids were allowed to feed on these plants for three days to form a YDV+ colony.

A two-factor factorial experimental design with randomised blocks was used to phenotype plant resistance and tolerance to the aphid-YDV pathosystem across perennial ryegrass and tall fescue cultivars. Factors included cultivar (n=27) and aphid YDV status (n=3). All treatment combinations were replicated four times, and the experiment was replicated twice. Plants receiving an aphid treatment were inoculated by attaching clip cages containing either YDV+ or YDV- aphids and allowing them to feed for 10 days. After 12 weeks, five leaves per plant were collected for nucleic acid extraction and PCR analysis, performed using procedures previously described.

Video tracking experiments were conducted in the laboratory by recording aphid feeding behaviour with monochrome GigE cameras. A 3-D printed arena containing eight individual cells (9 × 6 mm) was placed under each camera, allowing for 16 individual aphids to be analysed with video tracking during a recording session. A leaf attached to a grass seedling was placed in slits under each arena cell. Individual aphids were then placed in the arena cells and filmed for eight hours. Grass seedlings from the different cultivars were evaluated for both YDV+ and YDV- aphids. After an additional six weeks, five leaves per plant were collected for nucleic acid extraction and YDV detection.

Aphid behaviour was also tracked using automated video tracking software with settings adjusted to 25 frames per second. Arena cells were divided into leaf area and non-leaf area zones. Aphid velocity was used to determine when an aphid started and ended a probing (feeding) event. The number of probing events, duration of feeding events, and distance travelled were recorded.

Results and Discussion

In 82% of surveyed perennial ryegrass and tall fescue seed fields, a *Luteovirus*-type YDV was detected, and 65% had detection of a *Polerovirus*-type YDV. Perennial ryegrass fields had *Luteovirus* incidence of 28% compared to 64% in tall fescue fields. *Polerovirus* incidence was estimated at 3.5% in perennial ryegrass and 9.9% in tall fescue ($P < 0.0001$). Plant YDV species diversity was similar in perennial ryegrass and tall fescue fields, with BYDV-PAV being the most prevalent in both species (data not shown). Similarly, diverse YDV communities were found associated with sampled aphids.

In the small plot seed yield trials, aphid abundance in sweep net samples varied among insecticide timing treatments across three field seasons (Figure 1). There were fewer aphids in sweep net samples in 'spring', 'autumn + spring', and 'autumn + spring + summer' insecticide treatments compared to the untreated control and 'autumn' treatments. Aphid abundance was not different across whole-plot cultivars and split-plot N rates. There were no interactions in aphid abundance between insecticide timing and cultivar, insecticide timing and N factors, or cultivar and N factors.

Across three field seasons, YDV incidence did not differ among insecticide timing treatments (Figure 2). However, differences in YDV incidence were detected across cultivars ($P < 0.0001$) and N rates ($P = 0.011$). The high N rate increased YDV incidence in both cultivars. There were no interactions for insecticide timing treatments for cultivar or N rates.

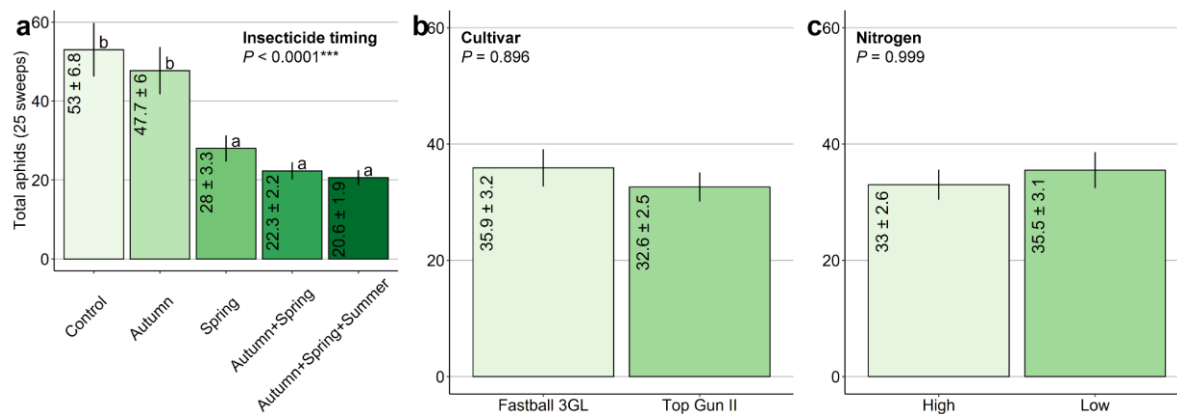


Figure 1. Comparisons of mean aphid abundance per sweep net sample across (a) foliar insecticide timing treatments, (b) cultivar, (c) spring nitrogen (N) fertiliser rate in three-year perennial ryegrass field trials.

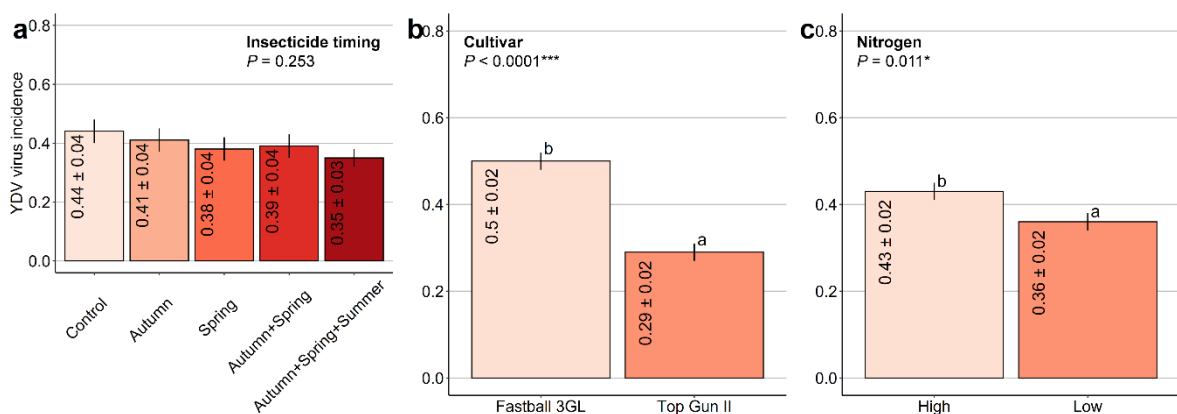


Figure 2. Comparisons of mean yellow dwarf virus (YDV) incidence per plot transect (n=5 leaves) across (a) foliar insecticide timing treatments, (b) cultivar, (c) spring nitrogen (N) fertiliser rate in three-year perennial ryegrass field trials.

Seed yield was different across insecticide timing treatments ($P = 0.001$), cultivars ($P = 0.004$), and N rates ($P = 0.0001$) (Figure 3). The 'autumn + spring + summer' insecticide treatment had 7.3% higher seed yield across cultivars and N rates compared to the untreated control. Seed yield was greatest for the less susceptible cultivar when fully protected with insecticide treatments throughout the season. A higher than recommended N fertiliser rate did not increase seed yield across treatment combinations in first-year stands but did increase seed yield in second and third-year stands when YDV infection >50%. Selecting resistant cultivars and reducing aphid populations during the autumn and spring flights can effectively maximise seed yield potential in perennial ryegrass. Furthermore, a lower N fertiliser rate can be used in first-year stands without reducing seed yield potential.

Phenotypic screening of grass cultivars in greenhouse experiments revealed mixed results for aphid fecundity and tolerance to YDV (data not shown). Differences in YDV infection across cultivars were not detected when exposed to YDV+ aphids. Aphid fecundity across aphid YDV+ and YDV- treatments varied among perennial ryegrass cultivars ($P < 0.001$), but differences were not detected among tall fescue cultivars. Differences in aphid treatment effects were observed for perennial ryegrass ($P < 0.001$) and tall fescue ($P < 0.001$), with the YDV+ aphid treatment having lower root biomass than the YDV- and control (no aphid) treatments.

A high-throughput method using automated video tracking addresses some of the challenges of traditional host plant resistance phenotyping. Aphid feeding behaviours, including the number of short probes and distance travelled using automated video tracking, were highly correlated across treatment groups. In both the greenhouse experiments and video tracking, YDV- aphids displayed greater fecundity, a reduction in short probing events, and shorter travel distances when exposed to YDV-

perennial ryegrass and tall fescue leaves. Greater phloem feeding events were observed in YDV- aphids in perennial ryegrass cultivars only. High-throughput phenotyping can help select YDV resistant germplasm for incorporation into traditional breeding programs.

References

- Aradottir GI and Crespo-Herrera L (2021) Host plant resistance in wheat to barley yellow dwarf viruses and their aphid vectors: A review. *Curr. Opin. Insect Sci.* **45**, 59-68.
- Dellaporta SL, Wood J and Hicks JB (1983) A plant DNA miniprep: Version II. *Plant Mol. Biol. Rep.* **1**, 19-21.
- Doyle JJ and Doyle JL (1990) Isolation of plant DNA from fresh tissue. *Focus* **12**, 13-15.
- Lowles AJ, Tacheel GM, Harrington R, Clark SJ (1996) The effect of temperature and inoculation access period on the transmission of barley yellow dwarf virus in *Rhopalosiphum padi* (L.) and *Sitobion avenae* (F.). *Ann. Appl. Biol.* **128**, 45-53.
- Miller WA and Lozier Z (2022) Yellow dwarf viruses of cereals: Taxonomy and molecular mechanisms. *Annu. Rev. Phytopathol.* **60**, 121-141.
- Rivedal HM, Dorman SJ, Temple TN, Lockwood TC, Walenta DL, Oshiro C, Cruse C, Starchvick RJ, Woodward MM and Anderson NP (2024) Prevalence and diversity of aphid-vectored yellow dwarf viruses in Oregon perennial grass seed crops. *PhytoFront.* **4**(4), 651-661.
- Walls J III, Rajotte E and Rosa C (2019). The past, present, and future of barley yellow dwarf management. *Agriculture* **9**(23), 1-16.

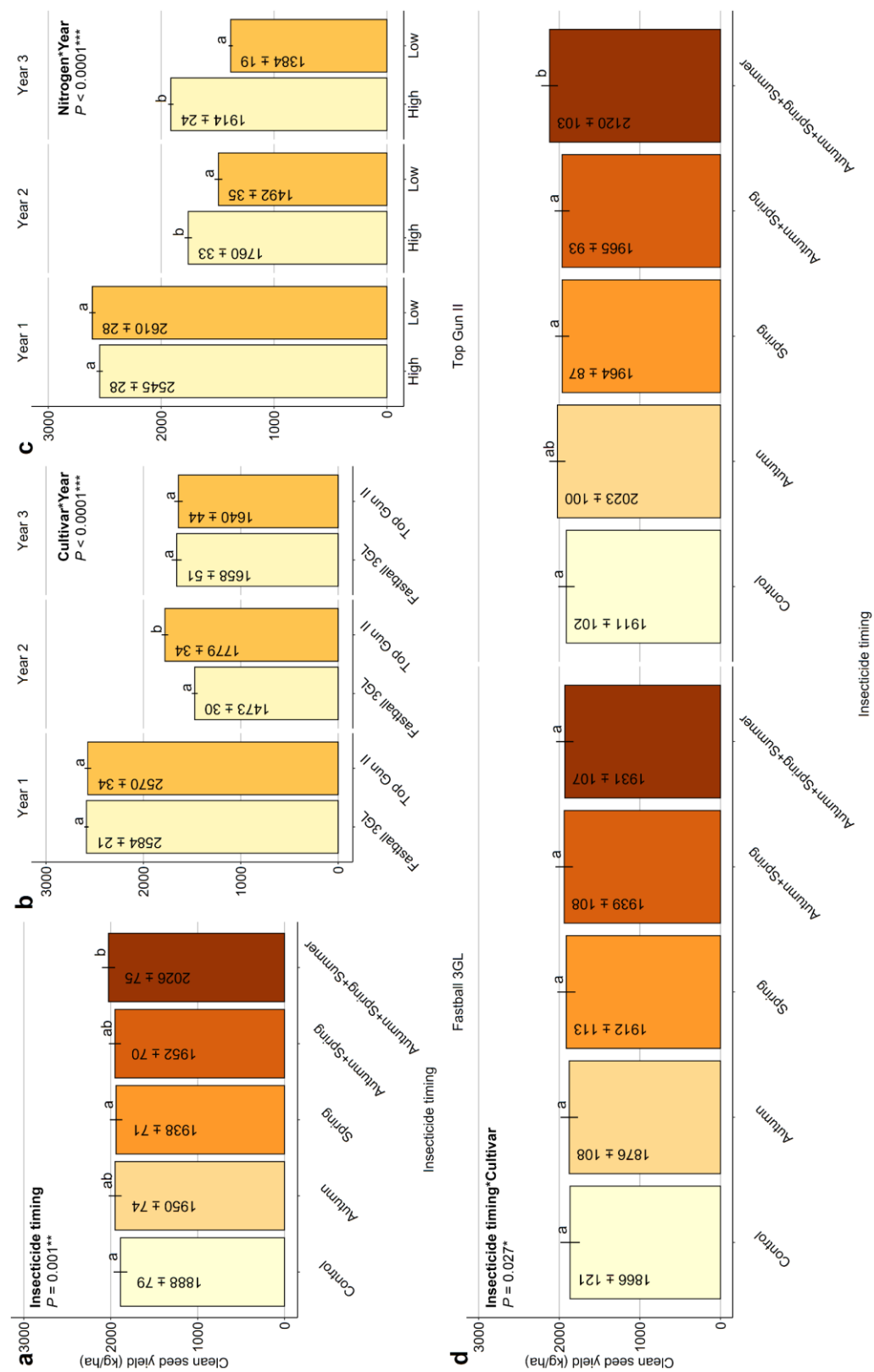


Figure 3. Comparisons of clean seed yield across (a) foliar insecticide timing treatments, (b) cultivar, (c) spring nitrogen (N) fertilizer rate, and (d) cultivar by foliar insecticide timing treatments in three-year perennial ryegrass field trials.

ORAL PRESENTATIONS

**Agronomic advances, innovation
& technology**

Seed packaging bag types - impact on endophyte viability and seed quality in perennial ryegrass during storage

Aung Myo Thant^{1*}, John Hampton¹, Phil Rolston² and Ivan Lawrie³

Abstract

Using woven polypropylene (WPP) plastic bags for seed packaging challenges the New Zealand seed industry's alignment with sustainability goals. The recycling scheme for farm plastic waste is still voluntary, especially for small seed bags (20-25 kg). The seed industry's concerns are that alternative seed packaging may not maintain *Epichloë* endophyte viability in stored perennial ryegrass (*Lolium perenne* L.) seed lots. A storage experiment was conducted using two perennial ryegrass seed lots with different endophyte strains stored in six types of bags (woven polypropylene (WPP), bi-axially oriented polypropylene (BOPP), low density polyethylene (LDPE), multi-wall paper (MWP), enviro-barrier paper (EPB) and moisture shield paper (MSP)) for a year. The seeds were stored in cool/dry (4°C, 30%RH), ambient (15-25°C, 65%RH), and warm/moist conditions (25°C, 80% RH). Seed moisture content (SMC), germination, seed vigour, and endophyte viability were assessed every 1.5-months (warm storage) and every 3-months (ambient and cool storage). At 12 months, SMC had increased by 2-3.5% in ambient and warm storage, while it decreased by 1-2% in cool storage. LDPE and BOPP bags maintained stable SMC across storage conditions. Germination remained stable in ambient and cool storage but dropped significantly in warm conditions, from 92% to between 49-67% after a year. BOPP and LDPE bags resulted in less germination loss (about 10%). Seed vigour declined in all storage environments, with the highest vigour loss in warm storage and the lowest in cool storage. Endophyte viability fell significantly in ambient and warm conditions, but remained stable in cool storage, regardless of bag type. Overall, the storage environment had a greater impact on the endophyte survival than the bag type. While all bag types were suitable, their physical robustness and environmental footprints require evaluation before recommendations for change.

Keywords: perennial ryegrass, endophyte, packaging, seed storage, seed quality

Introduction

The New Zealand seed industry produces around 85,000 tonnes of seeds annually. This generates about NZ\$281 million in exports and contributes approximately NZ\$329 million to the national GDP (NZGSTA 2024; FAR 2025). The industry supplies seeds for pastoral farming, arable crops and vegetable production both domestically and internationally. Despite its success, the seed industry faces several challenges, with the current use of plastic bags for seed packaging being a significant environmental concern.

The industry's use of woven polypropylene (WPP) plastic bags for seed packaging complicates its efforts to meet sustainability goals. The recycling scheme for farm plastic waste in New Zealand is still at an early stage and remains voluntary, particularly for small seed bags (20-25 kg). While alternative packaging options exist, the industry is concerned that these alternatives may not preserve the viability of *Epichloë* endophytes in perennial ryegrass seed lots from the first entry into storage until the sowing of the next crop. Perennial ryegrass is the backbone of pastoral agriculture in the country, and more than 90% of perennial ryegrass cultivars contain *Epichloë* endophytes (Caradus et al. 2021). Since endophytes are only transmitted through seeds (Easton et al. 2001; Gagic et al. 2018; Hume et al. 2020; Gundel et al. 2009), maintaining endophyte viability is crucial for commercial use (Caradus & Johnson 2019).

At the request of the seed industry, a storage experiment was conducted with two cultivars of perennial ryegrass, each containing a different endophyte strain, stored in six types of packaging bags available in New Zealand. Two bag types were not included due to either their high cost (aluminium

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foil bags) or their potential to contaminate seed lots (jute bags). Perennial ryegrass seeds can be safely stored in ambient conditions in New Zealand for up to a year, but in commercial settings, they are typically kept in cool/dry environments to preserve endophyte viability. This study included cool and ambient storage conditions, along with an additional storage condition that simulated a warm/moist environment. The latter scenario reflects the conditions seeds might encounter during shipping from New Zealand, especially when crossing the equator to the northern hemisphere.

Materials and Methods

For each of two perennial ryegrass cultivars (cv 'Base' containing AR37 endophyte and cv '4front' containing NEA2 endophyte), 250 g of seeds were stored in six different types of packaging bags over one year. The trial was replicated three times using a split-plot design, with cultivar as the main plot factor and packaging bag type as the sub-plot factor. To avoid the possibility of contamination resulting from frequent sampling from a 25 kg bag, a number of small trial bags (45 cm × 30 cm) were prepared and one of these trial bags was removed from storage at each sampling time. The six bag types were woven polypropylene (WPP), bi-axially oriented polypropylene (BOPP), low-density polyethylene (LDPE), multi-wall paper (MWP), enviro-barrier paper (EPB), and moisture shield paper (MSP). Each bag's mouth opening was single-stitched shut using a cotton thread.

The bags were stored in three different environments: cool/dry (4°C, 30% RH), ambient (15-25°C, 65% RH), and warm/moist (25°C, 80% RH). Initial seed quality was assessed before storage (Table 1). During the experiment, seed moisture content (SMC), germination, seed vigour and endophyte viability were assessed every 1.5 months (warm storage) and every 3 months (ambient and cool storage). However, only the quality data for the assessment at 12 months are reported here (Table 1).

Seed quality testing for SMC (high temperature oven method), germination (top of paper method), and seed vigour (accelerated ageing test) was conducted in accordance with the International Seed Testing Association rules (ISTA, 2023) except that both germination and seed vigour tests used four replicates of 50 seeds.

Endophyte viability was evaluated using a grow-out test. Seeds were sown in potting mix in a glasshouse and 5-6 weeks after emergence the basal part of one tiller per plant was cut above the point of root initiation from 200 randomly selected seedlings per treatment per replicate (200 × 3 = 600 seedlings total) and blotted on a Nitrocellulose membrane (NCM) sheet (Amersham Protran 0.45 µm NC, GE Healthcare Life Science). The NCM sheets were processed by the Barenbrug NZ seed laboratory. The sheets were agitated in a blocking solution containing a primary anti-endophyte antibody (monoclonal antibody supplied by Cropmark Seeds) and a secondary antibody (goat anti-mouse IgA alkaline phosphatase). Viable endophyte presence was indicated by a purple blot colour, while a colourless blot meant no endophyte had been transmitted. Further details regarding the immunoblotting procedure can be found in Hillis (2009) and Rinklake et al. (2020).

Results and Discussion

Seeds are hygroscopic, either gaining or losing moisture depending on the relative humidity (RH) of the surrounding air to finally achieve equilibrium seed moisture content (SMC) (Hay et al. 2022). Initially 'Base' had a higher SMC than '4front', and this difference was still evident after one year of storage (Table 1). For both cultivars, SMC increased as storage room RH% increased. There was a significant effect of bag type on SMC; seeds in the LDPE bag had the lowest SMC in both ambient and warm storage, but the highest SMC in cool storage. This suggests that LDPE bags regulate SMC based on storage conditions. Seeds in the BOPP bag also had a higher SMC than WPP in cool storage, but not in the other storage conditions (Table 1). There was a highly significant interaction for SMC between cultivars and bag type in both cool/dry and ambient storage conditions (Table 1). After one year of storage SMC between cultivars and among bag types differed, but in cool/dry storage LDPE bags had a mean SMC similar to that of 'Base' while in ambient storage LDPE bags had a mean SMC similar to that of '4front'.

Before storage both cultivars had similar high germination and vigour, and after one year of storage, they did not differ for either quality factor in cool or ambient storage, but '4front' had a greater germination in warm storage (Table 1). There was no significant difference in germination or seed

vigour among bag types in cool and ambient storage, although both had reduced over the year of storage in both storage environments to a level which would negatively impact the suitability of the seed for sowing (Finch-Savage & Bassel 2016). Seed germination loss (via the production of abnormal seedlings/dead seeds) is an indicator of seed physiological deterioration (as recorded by the vigour test). In the warm storage environment, seed germination had decreased from the initial 92% to 67% or less, and seed vigour from the initial >85% to 21% or less (poor seed vigour) over the course of a year. This is consistent with Hume et al. (2011) who reported that after 12 months of storage perennial ryegrass seed viability loss was about 23% in warm conditions (Queensland) compared with about 6% loss in cooler ambient conditions (New Zealand). In warm storage, both LDPE and BOPP bags were able to slow down the loss of both germination and seed vigour (approximately 10% greater than other bag types) but none of the bag types were able to maintain seed quality to a commercially acceptable level.

Endophyte viability was the quality factor most influenced by storage environment. In cool storage, the viability of both endophyte strains after one year of storage did not differ from that before storage and viability did not differ among bag types (Table 1). However, endophyte viability had declined rapidly in both ambient and warm storage conditions with higher temperature accelerating the loss. Only cool storage was able to maintain viability above 70%, the commercial threshold for endophyte-infected perennial ryegrass seeds (Hume & Baker 2005; Easton & Tapper 2005). Although after one year in ambient storage, both LDPE and BOPP bags preserved a slightly higher endophyte viability (65-67%) compared to other bag types, this still fell slightly short of the 70% threshold. Endophyte viability was maintained in ambient storage for 9 months in this trial (data not presented), with the loss mostly occurring within the following three months. Hume et al. (2011) and Rolston et al. (1986) had previously reported that endophyte viability could be maintained for at least 6 months under ambient New Zealand conditions. In warm storage, endophyte viability collapsed completely, having dropped to below 40% after just three months of storage (data not presented). Rolston et al. (1986) previously reported rapid loss of endophyte viability in warm storage, particularly when the SMC was above 11%.

Following one year of storage of seeds of two perennial ryegrass cultivars containing endophytes in six different bag types, it was evident that the storage environment had a greater impact on seed quality and endophyte viability than did bag type. Endophyte viability was maintained only under cool/dry storage, a storage environment that also best maintained seed germination and vigour. However, when compared with pre-storage germination and vigour test results, seed quality was reduced under all storage conditions, with the losses in germination and vigour increasing as storage temperature increased. This result challenges the contention that perennial ryegrass seed quality can be maintained during storage under ambient New Zealand conditions for up to a year. Storage in BOPP and LDPE bags slightly reduced the loss of endophyte viability during ambient storage and seed quality during warm storage, but the differences, while statistically significant, were biologically meaningless. Any of the five packaging types could be used as a replacement for WPP, but to meet sustainability goals, an assessment of their robustness and environmental footprint will first be required and is currently being undertaken.

Table 1. Seed quality and endophyte viability of perennial ryegrass seeds after 12 months of storage in three different environments

	Cool/Dry (4°C, 30%RH)				Ambient (15-25°C, 65%RH)				Warm/Moist (25°C, 80%RH)			
	SMC	Germ	Vigour	Endophyte	SMC	Germ	Vigour	Endophyte	SMC	Germ	Vigour	Endophyte
<u>Cultivars - Endophyte</u>												
Base - AR37	10.0 a	83	65	92 a	12.4 a	79	57	54 b	13.6 a	49 b	6	11
'4front' - NEA2	9.5 b	84	65	83 b	11.8 b	80	66	68 a	13.4 b	63 a	19	9
<u>Bag Type</u>												
WPP (Control)	9.6 c	81	66	90	12.1 c	82	64	58 b	13.5 a	51 b	11 b	10
BOPP	9.9 b	85	64	88	12.1 c	81	60	67 a	13.5 a	64 a	21 a	13
LDPE	10.7 a	84	70	87	11.8 d	78	65	65 a	13.3 b	67 a	20 a	12
MWP	9.5 c	84	66	89	12.3 a	82	63	59 b	13.5 a	53 b	9 b	10
EBP	9.4 c	84	60	88	12.2 b	79	60	59 b	13.5 a	51 b	6 b	7
MSP	9.5 c	83	65	88	12.2 b	79	60	58 b	13.5 a	49 b	9 b	7
<u>Pr > F</u>												
Cultivars	**	ns	ns	**	***	ns	ns	**	*	**	ns	ns
Bags	***	ns	ns	ns	***	ns	ns	***	***	***	***	ns
Cultivars x Bags	***	ns	ns	ns	***	ns	ns	ns	ns	ns	**	ns

Initial seed quality of perennial ryegrass seed lots before storage

'Base' AR37 → SMC (seed moisture content) =12.0%; Germination = 92%; Vigour (AA test) = 87%; Endophyte = 92%

'4front' NEA2 → SMC (seed moisture content) =10.4%; Germination = 92%; Vigour (AA test) = 89%; Endophyte = 84%

WPP = Woven polypropylene; BOPP = Bi-axially oriented polypropylene; LDPE = Low-density polyethylene; MWP = Multi-wall paper; EBP = Enviro-barrier paper; MSP = Moist shield paper

* = < 0.05, ** = < 0.01, *** = < 0.001, ns = not-significant

References

- Caradus JR, Chapman DF, Cookson T, Cotching B, Deighton MH, Donnell L, Ferguson J, Finch SC, Gard S, Hume DE, Johnson LJ, Kerr GA, Norriss MG, Peddie K and Popay AJ (2021) *Epichloë* endophytes – new perspectives on a key ingredient for resilient perennial grass pastures. In: Douglas, G. (ed.) *Resilient pastures—Grassland research and practice series*. New Zealand: New Zealand Grassland Association, 17, 347-360. DOI: 10.33584/rps.17.2021.3435
- Caradus JR and Johnson L (2019) Improved adaptation of temperate grasses through mutualism with fungal endophytes. In: Schouten, A. (ed.) *Endophyte biotechnology: potential for agriculture and pharmacology*. Oxford: CAB International, pp. 145-183. DOI: 10.1079/9781786399427.0085
- Easton HS, Christensen MJ, Eerens JPJ, Fletcher LR, Hume DE, Keogh RG, Lane GA, Latch GCM, Pennell CGL, Popay AJ, Rolston MP, Sutherland BL and Tapper BA (2001) Ryegrass endophyte: A New Zealand grassland success story. *Proceedings of New Zealand Grassland Association* **63**, 37-46.
- Easton HS and Tapper BA (2005) *Neotyphodium* research and application in New Zealand. In: Roberts, C.A., West, C.P. and Speirs, D.E. (eds.) *Neotyphodium in cool-season grasses*. Hoboken, NJ: Wiley-Blackwell. pp. 35-42.
- Finch-Savage WE and Bassel GW (2016) Seed vigour and crop establishment: extending performance beyond adaptation. *Journal of Experimental Botany* **67**, 567-591.
- FAR (Foundation for Arable Research) (2025) New Zealand's arable industry (Accessed: 1 May 2025).
- Gagic M, Faville MJ, Zhang W, Forester NT, Rolston MP, Johnson RD, Ganesh S, Koolaard JP, Easton HS, Hudson D, Johnson LJ, Moon CD and Voisey CR (2018) Seed transmission of *Epichloë* Endophytes in *Lolium perenne* is heavily influenced by host genetics. *Frontiers in Plant Science* **9**, 1-16.
- Gundel PE, Martínez-Ghersa MA, Garibaldi LA and Ghersa CM (2009) Viability of *Neotyphodium* endophytic fungus and endophyte-infected and noninfected *Lolium multiflorum* seeds. *Botany* **87**, 88-96.
- Hay FR, Rezaei S and Buitink J (2022) Seed moisture isotherms, sorption models, and longevity. *Frontiers in Plant Science* **13**, 1-14. DOI: 10.3389/fpls.2022.891913.
- Hillis S (2019) Transmission and survival of perennial ryegrass endophyte during field based seed production: Unpublished MAgSc thesis, Lincoln University, Canterbury, New Zealand.
- Hume DE and Baker J (2005) Growth and management of endophytic grasses in pastoral agriculture. In: Roberts, C.A., West, C.P. and Speirs, D.E. (eds.) *Neotyphodium in cool-season grasses*. Hoboken, NJ: Wiley-Blackwell. pp. 201-226.
- Hume DE, Stewart AV, Simpson WR and Johnson RD (2020) *Epichloë* fungal endophytes play a fundamental role in New Zealand grasslands. *Journal of the Royal Society of New Zealand* **50**(2), 279-298. DOI: 10.1080/03036758.2020.1726415.
- Hume DE, Schmid J, Rolston MP, Vijayan P and Hickey MJ (2011) Effect of climatic conditions on endophyte and seed viability in stored ryegrass seed. *Seed Science and Technology* **39**, 481-489.
- ISTA (International Seed Testing Association) (2023) ISTA International rules for seed testing, Wallisellen, Switzerland.
- NZGSTA (New Zealand Grain and Seed Trade Association) (2024) New Zealand seed exports are increasing in value. <https://www.nzgsta.co.nz/new-zealand-seed-exports-are-increasing-in-value> (Accessed: 1 March 2024).
- Rinklake I, James A, Brownfield L, MacKnight R and Eady C (2020) Relative *Epichloë* endophyte fungal biomass in ryegrass tillers grown pre and post vernalization. *Agronomy New Zealand* **50**, 95-107.
- Rolston MP, Hare MD, Moore KK and Christensen MJ (1986) Viability of *Lolium* endophyte fungus in seed stored at different moisture contents and temperatures. *New Zealand Journal of Experimental Agriculture* **14**, 297-300.

The Grady Sensor: determining seed moisture content in seconds

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Abstract

Seed moisture content (SMC) is the most reliable indicator of optimal harvest timing in many seed crops, including grass seed. Measuring SMC as grass seed crops approach maturity is recommended to determine optimal harvest timing. Currently, to measure SMC, seeds need to be stripped from heads by hand, weighed, dried until all the moisture has been lost, then re-weighed, and SMC manually calculated. Collecting SMC information in this manner is arduous, time consuming, and prone to error. Consequently, this procedure has resulted in inadequate SMC testing or failure to test in a timely fashion. In addition, SMC is an important factor in the storage of harvested seed, which typically needs to be stored under 12% SMC, to ensure high seed quality. The goal of this project is to develop a portable sensing device - The Grady Sensor - that allows for rapid and accurate SMC measurement of grass seed. The sensor employs near-infrared (NIR) spectroscopy principles that water molecules absorb specific NIR wavelengths. By analysing the light reflected from the seed surface, the sensor predicts SMC based on the intensity of the reflected light at moisture-sensitive wavelengths. Over two years, multiple sensor prototypes have been developed, and their performance has been validated through field tests in Oregon and New Zealand. Sensor readings were compared to laboratory oven gravimetric SMC values of samples collected from major grass seed species, including tall fescue, annual ryegrass, perennial ryegrass, orchardgrass, creeping red fescue, creeping bentgrass, Chewings fescue, and Kentucky bluegrass. The sensor readings demonstrated a significant linear relationship with the oven SMC. Mean absolute errors of sensor SMC predictions were within 1.2 to 4.6% across all grass species. The results indicate that the prototype is a reliable replacement for the traditional oven drying method.

Keywords: grass seed crops, harvest timing, near infrared reflectance, seed moisture content, portable sensor

Introduction

Moisture content in grass seed plays an important role in achieving optimum seed quality, seed yield potential, and harvest efficiency. Seed moisture content (SMC) is a reliable indicator of physiological maturity in cool-season grass seed crops and also determines the conditions of storage and shelf life of seeds (Grabe et al. 1986; Silberstein et al. 2010; Szulc et al. 2020). Harvesting in the SMC correct range minimises seed shattering losses (Andrade et al. 1994). Seed shattering is lowest at high SMC and highest at low SMC. Thus, it is important that seed growers harvest at the recommended SMC range for each cool-season grass species (Silberstein et al. 2010).

The traditional method for determining SMC is to measure the change in weight of a sample after drying at a temperature that allows the water to be released from the seed (Hay et al. 2023). This method involves obtaining clean seed samples, weighing for the seed wet weight, drying in an oven at 130°C for two hours and re-weighing for the seed dry weight, after cooling down. Gravimetric SMC is calculated using:

$$\text{Gravimetric SMC} = \frac{\text{Weight}_{\text{wet}} - \text{Weight}_{\text{dry}}}{\text{Weight}_{\text{wet}}} \times 100\%$$

This process is time consuming, with increased risk of testing too late when the SMC has already fallen below recommended levels for harvest. There is a pressing need for a more efficient SMC measuring method that can provide more timely information for the decision making.

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Conflict of interest: All authors (except for Logan P. Snell) are stakeholders of Digital Seed Technology Inc. that manufactures and distributes The Grady Sensor.

Therefore, in this study, we developed The Grady Sensor that allows end-users to predict SMC in seconds. The sensor measures SMC non-destructively by employing near-infrared (NIR) spectroscopy principles where water molecules absorb specific NIR wavelengths. The sensor predicts the moisture content by using empirically established calibration equations and the intensity of the reflected light at moisture-sensitive wavelengths. Generally, higher values of reflected light intensity lead to lower predicted values of SMC in seeds and vice versa. The sensor is compatible with most economically important cool-season grass crop species, including tall fescue (*Schedonorus arundinaceus* (Shreb.) Dumort.), annual ryegrass (*Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot), perennial ryegrass (*Lolium perenne* L.), orchardgrass (*Dactylis glomerata* L.), creeping red fescue (*Festuca rubra* L. subsp. *rubra*), creeping bentgrass (*Agrostis stolonifera* L.), Chewings fescue (*Festuca rubra* L. subsp. *fallax* (Thuill.) Nyman), and Kentucky bluegrass (*Poa pratensis* L.).

Materials and Methods

1. Sensor design. The physical design of The Grady Sensor, including the main device body, a seed cup, and calibration standards, is shown in Figure 1A. The device consists of electrical components, including a microprocessor, battery power management circuit, NIR light-emitting diode (LED) circuit, and a photodiode circuit (Figure 1B). Circuits were embedded into a custom-designed printed circuit board (PCB) with all electronic components assembled onboard. A device enclosure was designed to house the PCB and other features that enable this device to be handheld and portable.

The LED circuit includes a constant current LED driver (TLC5916, Texas Instruments, city, country) and a LED array with LEDs at two specific wavelengths (WLs), namely WL-1 and WL-2, arranged in a circular array. During operation, the NIR LEDs are activated by the microprocessor, emitting light at the two wavelengths, one after another. This illumination intercepts the seed sample in the seed cup, where the incident light undergoes partial absorption and diffuse reflection from the seed surface. The reflected light intensity is captured by a centrally positioned InGaAs photodiode (G12181-210K, Hamamatsu Photonics, Iwata, Japan), which generates a photocurrent proportional to the photon flux received.

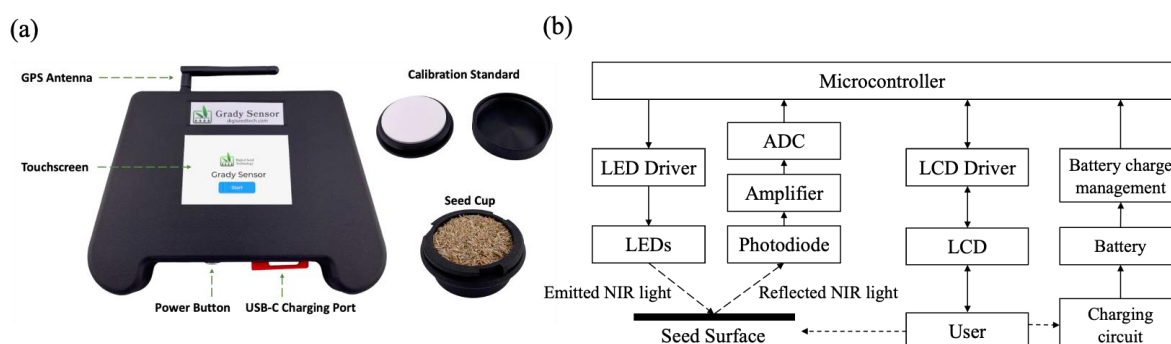


Figure 1. Design of the near-infrared (NIR) sensor including (a) physical device components and (b) the sensor printed circuit board (PCB) block diagram.

The photocurrent signal from the photodiode is processed to an analogous voltage output through a transimpedance amplifier. This voltage is subsequently amplified by a programmable gain value to ensure compatibility with the dynamic range of the subsequent analogue-to-digital converter (ADC). The amplified voltage is digitised by the ADC, which performs a comparison against a reference voltage and quantises the input into a binary representation for interfacing with the microcontroller.

The microcontroller interprets the binary ADC outputs and converts them into reflectance values, or the ratio of the reflected light intensity to the intensity of emitted lights, that is estimated using a calibration standard (FWS-99-02, Avian Technology, New London, New Hampshire, USA). These reflectance values are then applied to empirically established calibration equations stored in firmware, which map the optical response to SMC.

2. Data collection and sensor calibration. Calibration equations are established through regression analysis of wavelength readings provided by the sensor against reference gravimetric oven SMC across

the eight species and seed moisture levels. Seed samples have been collected from commercial seed production fields and experimental plots located in both New Zealand and Oregon. Nine sensor prototypes were developed for testing in New Zealand, and 13 prototypes were developed for testing in Oregon.

Seed moisture content was determined daily using the NIR sensor and gravimetric oven-drying methods (Silberstein et al. 2010). Data collection began at the onset of externally visible seed fill (presence of an endosperm) and continued through harvest. Each day, representative samples were taken from each field by stripping seed from a minimum of 30 seed heads and storing them in an airtight container in a cooler until ready for sensor measurement. Sensor readings were taken and recorded after stripping seed from the seed heads. Immediately after sensor measurement, the samples were removed from the cup, placed in a drying tin and weighed prior to drying in a laboratory circulated-air oven at 130°C for two hours. After drying, samples were cooled to room temperature and then weighed to allow for calculation of the SMC percentage.

White and dark measurements were recorded for each sensor prototype before measuring samples. White measurements were collected on the calibration standard with 99.8% nominal reflectance averaged across 250-2450 nm. The dark measurements were collected in a dark environment by mounting an empty seed cup to the sensor body with no light emitted from the LEDs. Raw readings of samples were then converted into reflectance values using Equations 1 and 2.

$$X_1 = \frac{R_1 - D_1}{C_1 - D_1} \quad (1)$$

$$X_2 = \frac{R_2 - D_2}{C_2 - D_2} \quad (2)$$

where, X_1 and X_2 are the calibrated reflectance values in LED group 1 and 2, respectively, R_1 and R_2 are raw sensor readings in WL-1 and WL-2, respectively, C_1 and C_2 are white measurements in WL-1 and WL-2, respectively, and D_1 and D_2 are dark measurements in WL-1 and WL-2, respectively.

The linear relationship between the sensing readings and oven SMC was assessed using the Coefficient of Determination (R^2) derived from linear regression analysis. To evaluate the effects of environment (i.e. fields) and sensor device variability on the linear relationship between sensor readings and oven SMC, two-way Analysis of Variance (ANOVA) tests were conducted at a significance level of $\alpha = 0.05$. Calibration equations were developed based on oven SMC and sensor readings from the latest NIR sensor prototypes. The dataset was randomly divided into training (80%) and testing (20%) sets. A partial least squares regression (PLSR) model was trained on the training set and validated on the testing set of each species. Model performance for estimating SMC was evaluated using the R^2 and Mean Absolute Error (MAE) between the oven SMC and model predictions.

Results and Discussion

1. Linear relationship between sensor readings and oven SMC. Linear regression results demonstrated a significant linear relationship ($P < 0.05$) between sensor readings at WL-1 and oven SMC values for all eight grass species (Figures 2A and 2C). Regression analyses of sensor readings at WL-2 revealed significant linear relationships ($P < 0.05$) for annual ryegrass, tall fescue, creeping bentgrass, Chewings fescue and Kentucky bluegrass, but not for orchardgrass, perennial ryegrass, and creeping red fescue (Figures 2B and 2D). This result is expected as WL-2 is designed as a reference wavelength to capture baseline reflectance less affected by SMC, thereby enabling normalisation of WL-1 readings to account for seed-specific factors such as seed surface texture and size.

Analysis of Variance tests were conducted to evaluate whether the linear regression parameters (slope and intercept) relating sensor readings to gravimetric SMC differed across multiple sensor prototypes and across fields where seed samples were collected. The ANOVA results showed no differences ($P > 0.05$) in the slopes of the linear relationships across different sensor prototypes for all species tested. This consistency in slopes indicates that the rate of change in sensor readings with respect to gravimetric SMC is uniform across devices. However, differences ($P < 0.05$) were observed in the intercepts of the linear relationships between devices for all species. This suggests that while the

sensitivity (as reflected by the slope) of the sensor to gravimetric SMC changes remains consistent, there are device-specific offsets in the baseline reflectance values. These offsets likely arise from minor variations in hardware components, such as LED intensity or photodiode sensitivity among prototypes. In practical applications, these offsets can be quantified by measuring sensor readings against calibration standards, which ensures consistent SMC predictions across all prototypes by including device-specific corrections to the calibration equations.

In contrast, ANOVA results indicated that the individual fields where seed samples were collected significantly affected the linear relationship between sensor readings and gravimetric SMC, for most species. Differences in the intercepts of the linear relationships were observed across fields for tall fescue, perennial ryegrass, orchardgrass, creeping bentgrass, red fescue, and Chewings fescue, but not for annual ryegrass where the two sampling fields had the same variety, and Kentucky bluegrass with three different varieties across three fields. In addition, differences in the slopes of the linear relationships were observed across fields for tall fescue, perennial ryegrass, annual ryegrass, orchardgrass, red fescue, and Chewings fescue, but not for creeping bentgrass or Kentucky bluegrass. The slope variations indicate that the sensitivity of the sensor's response to SMC changes differs across fields, for most species, potentially due to environmental factors such as soil type, irrigation practices, harvest conditions, or differences in variety.

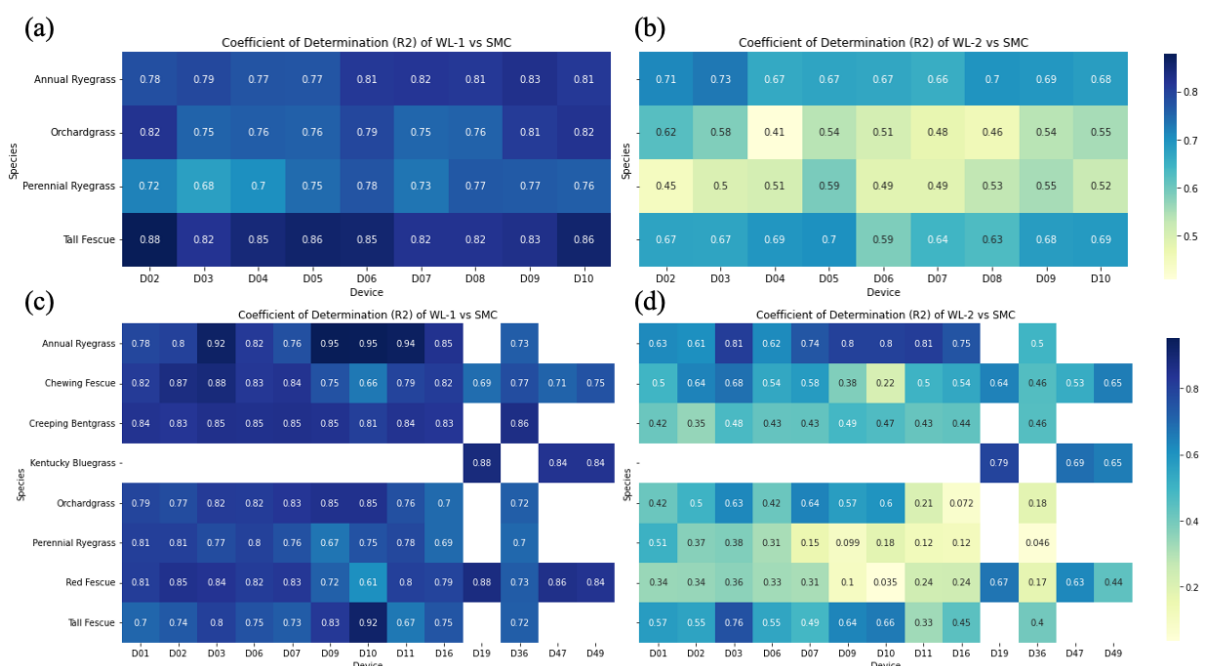


Figure 2. Heatmaps of Coefficient of Determination (R^2) between near-infrared (NIR) sensor readings and laboratory oven seed moisture content (SMC) across sensor devices for eight major cool-season grass species. (a) and (b) show data from nine sensor prototypes collected in New Zealand and (c) and (d) show from data collected in Oregon. Devices 19, 47, and 49 were tested in eastern Oregon while the other 10 devices were tested in western Oregon. (a) and (c) indicate the linear agreement between WL-1 and oven SMC while (b) and (d) indicate the linear agreement between WL-2 and gravimetric SMC.

2. Sensor performance in predicting SMC. Performance of PLSR models in predicting SMC using readings from the latest NIR sensor prototypes is shown in Figure 3. Across the eight grass species evaluated, the PLSR models demonstrated robust predictive accuracy for SMC, with average prediction errors ranging from 1.2 to 4.6% when validated against gravimetric SMC values. Among the species tested, Kentucky bluegrass showed the best predictive performance, achieving an R^2 of 0.94 and an MAE of 1.2%.

Despite significant field-specific differences in linear regression parameters for other species, the PLSR models resulted in robust prediction accuracy across varieties or fields for these species. It highlights

the sensor's ability to generalise across varieties and environmental conditions, and reproducibility in the sensor's response to moisture variations. Notably, a deviation in model agreement was observed for turf-type and forage-type tall fescue. The forage-type fields had higher prediction errors compared to the turf-type fields. The discrepancy suggests that varietal differences between forage-type and turf-type tall fescue may influence NIR reflectance properties. Consequently, the development of separate calibration equations for forage and turf-type varieties of tall fescue may be necessary to enhance model accuracy for this species.

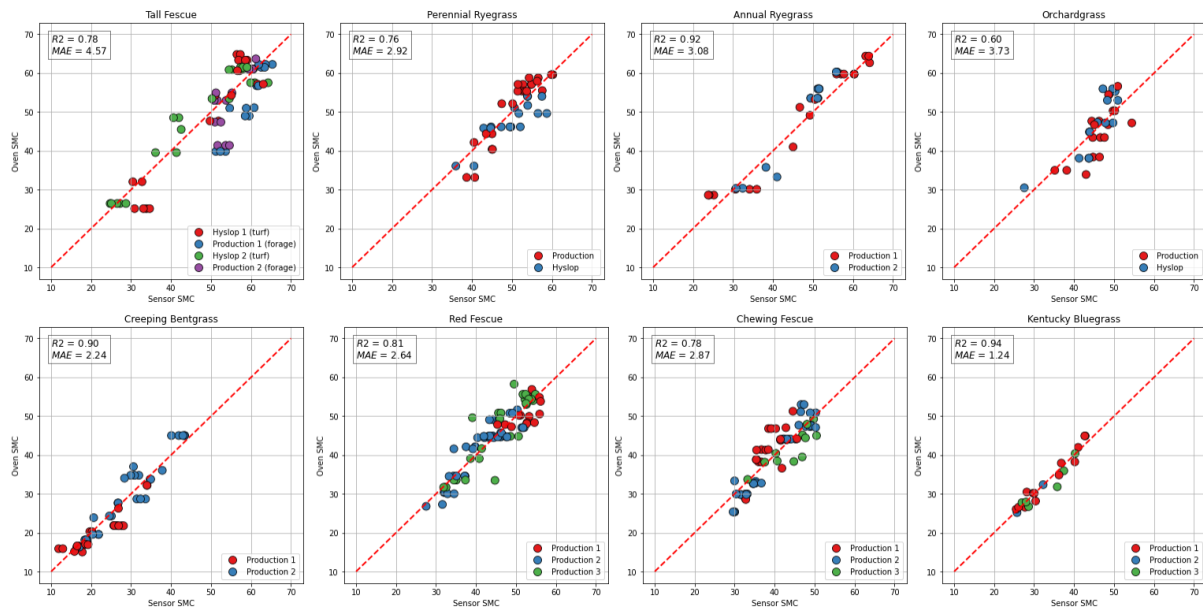


Figure 3. Scatterplots showing the relationship between near-infrared (NIR) sensor seed moisture content (SMC) and laboratory oven-derived SMC for tall fescue (*Schedonorus arundinaceus* (Shreb.) Dumort.), annual ryegrass (*Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot), perennial ryegrass (*Lolium perenne* L.), orchardgrass (*Dactylis glomerata* L.), creeping red fescue (*Festuca rubra* L. subsp. *rubra*), creeping bentgrass (*Agrostis stolonifera* L.), Chewing fescue (*Festuca rubra* L. subsp. *fallax* (Thuill.) Nyman), and Kentucky bluegrass (*Poa pratensis* L.). For each grass species, training data from all sampled varieties were combined and used to establish the estimation equation.

References

- Andrade R, Grabe D and Ehrensing D (1994) Seed maturation and harvest timing in turf type tall fescue. *Journal of Applied Seed Production* **12**, 34-46.
- Grabe DF, Benjamin E, Garbacik CJ and Chastain TG (1986). Development of moisture testing methods for seed crops. In H.W. Youngberg (ed.) *Seed Production Research*, Oregon State Univ., Crop Science Extension Report, **61**, 12-13.
- Hay FR, Rezaei S, Wolkis D and McGill C (2023) Determination and control of seed moisture. *Seed Science and Technology* **51**(2), 267-285.
- Silberstein TB, Mellbye ME, Chastain TG and Young WC (2010) Using seed moisture as a harvest management tool. Oregon State University, EM 9012.
- Szulc J, Gozdecka G and Poćwiardowski W. (2020) The application of NIR spectroscopy in moisture determining of vegetable seeds. *Czech Journal of Food Sciences* **38**(2), 131-136.

Irrigation responses of perennial ryegrass seed crops in New Zealand

Richard Chynoweth^{1*}, Phil Rolston² and Bede McCloy³

Abstract

The soils of the Canterbury Plains in New Zealand are characterised by variable topsoil depths, typically underlain by gravel. As a result, water holding capacity (i.e., the supply) is influenced by both the depth of topsoil and the rooting depth of the crop species. Crop water use, in turn, is primarily driven by atmospheric demand at any given time and the ability of the crop-soil system to meet that demand via evapotranspiration (ET), which includes evaporation from the leaf surface and soil. For instance, under north-westerly wind conditions common in Canterbury, water use can be substantial (up to 6 mm day⁻¹) due to high temperatures, strong winds, and low humidity, all of which accelerate water loss from plant stomata. During spring and summer, the accumulated potential water deficit (PET minus rainfall) for many grass seed crops often exceeds 300 mm. In contrast, many soils in the region can only supply between 60 and 120 mm of available water before moisture stress develops.

The response of perennial ryegrass (*Lolium perenne* L.) to water stress was evaluated over three seasons (2009–2011) near Chertsey, Canterbury. The trial site featured a shallow Templeton silt loam soil overlying gravel at approximately 60 cm depth, with a total water-holding capacity of around 120 mm and an estimated stress threshold of 60 mm. Soil moisture was monitored weekly in the 10–60 cm profile using a neutron probe, and irrigation was applied weekly via trickle tape to replace the PET, delivering approximately 12 mm hour⁻¹. The maximum potential soil moisture deficit (MPSMD) was calculated as the cumulative difference between daily ET and the sum of rainfall and irrigation inputs. Treatments were designed to simulate drought stress either before or after flowering, representing spring and early summer drought conditions, respectively. Plots were windrowed and machine harvested at ~40% seed moisture content.

Seed yield in the untreated (non-irrigated) control plots ranged from 410 to 1970 kg ha⁻¹ depending on seasonal rainfall, while seed yields under full irrigation ranged from 1800 to 2500 kg ha⁻¹. A reduction in seed yield occurred when MPSMD exceeded approximately 75 mm. When expressed relative to fully irrigated yields, seed production declined at a rate of ~0.22% for each additional millimetre of accumulated soil moisture deficit. Seed yield improvements under irrigation were attributed to increases in both seed number and seed weight (measured as thousand seed weight).

These findings highlight the importance of understanding the soil water supply and identifying the critical deficit threshold at which yield losses begin. Irrigation strategies should therefore aim to maintain soil moisture above this threshold to safeguard seed production.

Keywords: irrigation, soil moisture, deficit, rainfall, evapotranspiration

Introduction

Water stress is a recurrent constraint to seed production in the east coast regions of New Zealand, where evapotranspiration rates often exceed precipitation during the critical growth months of September, October, November, and December (NIWA, 2025). Under these conditions, soil moisture reserves are rapidly depleted. If the available soil water in the root zone cannot meet crop demand, wilting, accelerated leaf senescence, and tiller mortality may occur.

Perennial ryegrass (*Lolium perenne* L.) is an important seed crop in the rotation of arable farmers with approximately 12,000 ha grown annually (Chynoweth et al. 2015). The timing of water deficit exerts a decisive influence on yield formation (Chastain et al. 2015). Early-season drought, occurring during the tillering and stem elongation phases, can limit the initiation and survival of reproductive tillers by

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reducing leaf expansion rates and lowering photosynthetic capacity. This results in a reduced number of fertile seed heads per unit area, thereby constraining yield potential.

In contrast, late-season drought, occurring during anthesis, seed set, and grain filling, predominantly affects seed development (Chastain et al. 2015; Hebblethwaite 1977). Water stress during this period impairs photosynthetic assimilation and can force reliance on stored non-structural carbohydrates for seed filling (Rowarth et al. 1997). When carbohydrate reserves are insufficient, seeds fail to attain their potential size or weight, reducing thousand-seed weight and the proportion of seed meeting commercial standards. Additionally, increased canopy temperatures associated with late-season drought can exacerbate these effects by shortening the seed-filling duration which is determined by accumulated temperature (Chynoweth & Moot 2017).

This study was established to investigate the responses of perennial ryegrass seed crops to temporally distinct drought events, with the specific aim of quantifying yield penalties associated with early-versus late-season water deficits and identifying the physiological mechanisms underpinning these responses.

Methods

Three field experiments were conducted over consecutive seasons (2009-10, 2010-11, and 2011-12) at the FAR Arable Site, Chertsey, Canterbury, New Zealand. Each trial was established on a new perennial ryegrass (*Lolium perenne* L.) stand sown in April in 15 cm rows at 8-10 kg ha⁻¹, with suSCon® Green (15 kg ha⁻¹; ai 100 g kg⁻¹ chlorpyrifos) applied for grass grub (*Costelytra giveni*) control. 'Grasslands Samson' was grown in the first two seasons using a randomised complete block design, while season three included both 'Grasslands Samson' and 'One50' in a factorial design. Both cultivars are diploid ryegrasses widely used in New Zealand, with 'Grasslands Samson' having a mid-season heading date and 'One50' heading date 18 days later. All seed lines sown contained the AR1 endophyte.

The soil was a Templeton silt loam with ~60 cm topsoil over free-draining gravel and a water-holding capacity of ~120 mm (Lilburne et al. 2012). Irrigation was applied via trickle tape between rows at 11 mm hour⁻¹, scheduled using potential soil moisture deficit (PSMD) calculated from evapotranspiration and rainfall, and verified by weekly neutron probe readings (0-60 cm). The profile was generally restored to field capacity during winter rainfall (Table 1).

Table 1. Monthly rainfall and calculated potential evapotranspiration (PET) from sowing to harvest for three seasons, recorded at the FAR Arable Site, Chertsey, New Zealand.

Month	2009-2010			2010-2011			2011-2012		
	Rain	PET ¹	Deficit ²	Rainfall	PET	Deficit	Rainfall	PET	Deficit
April	54	57	-3	32	59	-27	90	56	34
May	129	33	96	224	26	198	76	33	43
June	14	22	-8	92	19	73	35	23	12
July	33	30	3	40	25	15	12	34	-22
August	41	49	-8	112	31	81	46	45	1
September	21	76	-55	33	65	-32	47	77	-30
October	76	101	-25	28	112	-84	107	95	12
November	18	118	-100	52	144	-92	59	132	-73
December	38	135	-97	38	144	-106	50	144	-94
January 1-11	19	55	-36	14	68	-54	38	154	-116
Seasonal total	443	676	-233	665	693	-28	560	793	-233
Sept - harvest	172	485	-313	165	533	-368	301	602	-301

Note: ¹ Monthly PET values calculated using the Priestly Taylor method. ² Positive monthly deficit equals soil moisture recharge while negative monthly deficit results in soil moisture draw down.

Harvest procedures were consistent across years where plots were windrowed at ~40% seed moisture and harvested 6–9 days later using a plot combine except for the untreated in season two that was

direct combined. Subsamples were machine-dressed to 1st generation seed certification standards and expressed as kg ha⁻¹. Thousand seed weight was determined from 200 seeds weighed to three decimal places.

PSMD was estimated using the Priestley–Taylor method (Jamieson 1982), and maximum seasonal deficits were recorded at MPSMD. Yield and component data were analysed by ANOVA, with significant differences ($P < 0.05$) separated using LSD (Seabold and Perktold 2010).

Results and Discussion

In season one, seed yield increased ($P < 0.05$) from 1,997 to about 2,500 kg ha⁻¹ when irrigation exceeded 205 mm (Table 2). Timing of drought, before or after anthesis, did not affect yield response with similar reductions (~500 kg ha⁻¹) occurring when water stress intensity was equal regardless of timing. The seed yield response was driven by increases in TSW. In season two, irrigation lifted seed yield from 410 kg ha⁻¹ in the control to 1,776 kg ha⁻¹ under full irrigation, a fourfold increase. A minimum of 220 mm of irrigation was required to maximise seed yield, when efficient use of rainfall occurred. Treatments replacing 66% of evapotranspiration (PET) achieved yields comparable to the full ET replacement in both seasons (Table 2) due to capacity to capture and hold rainfall. The mechanisms responsible for seed yield increases were changes in the number of seeds reaching maturity (Figure 1) and TSW.

Table 2. Seed yield of ‘Grasslands Samson’ perennial ryegrass for two seasons following nine irrigation treatments grown at FAR Arable Site, Chertsey Canterbury between 2009 and 2011.

Trt #	Treatment	2009-10		2010-11	
		Applied ¹ (mm)	Seed yield (kg ha ⁻¹)	Applied (mm)	Seed yield (kg ha ⁻¹)
1	No irrigation (Nil)	0	1997	5	410
2	Replace 33 % of ET	102	2070	116	1232
3	Replace 66 % of ET	205	2518	221	1651
4	Replace ET (R-ET)	310	2500	340	1776
5	Nil to anthesis f.b. R-ET	165	2054	161	1023
6	Nil to anthesis f.b. Replace 50 % ET	83	1984	83	1113
7	R-ET to anthesis f.b. Nil	145	1943	219	1452
8	R-ET to anthesis f.b. Replace 50% ET	227	2581	280	1605
9	Apply at 60 mm PSMD		-	316	1744
		<i>P</i> value	<0.001		<0.001
		LSD _{0.05}	290		204
		S.E.	56		75

Note. yellow highlighted treatments are in the top statistical group. f.b. = followed by, ¹ = applied irrigation.

In season three, cultivar and irrigation treatment interacted such that overall ‘Grasslands Samson’ outperformed ‘One50’, which matured later in hotter conditions and experienced increased drought intensity (Table 2). For example, both cultivars produced the same fully irrigated seed yield, but no irrigation reduced seed yield by 1,044 kg ha⁻¹ in ‘One50’ versus 498 kg ha⁻¹ in ‘Grasslands Samson’. Maintaining the seed yield of ‘One50’ required ~97 mm of additional irrigation, equivalent to 20 extra days of PET replacement.

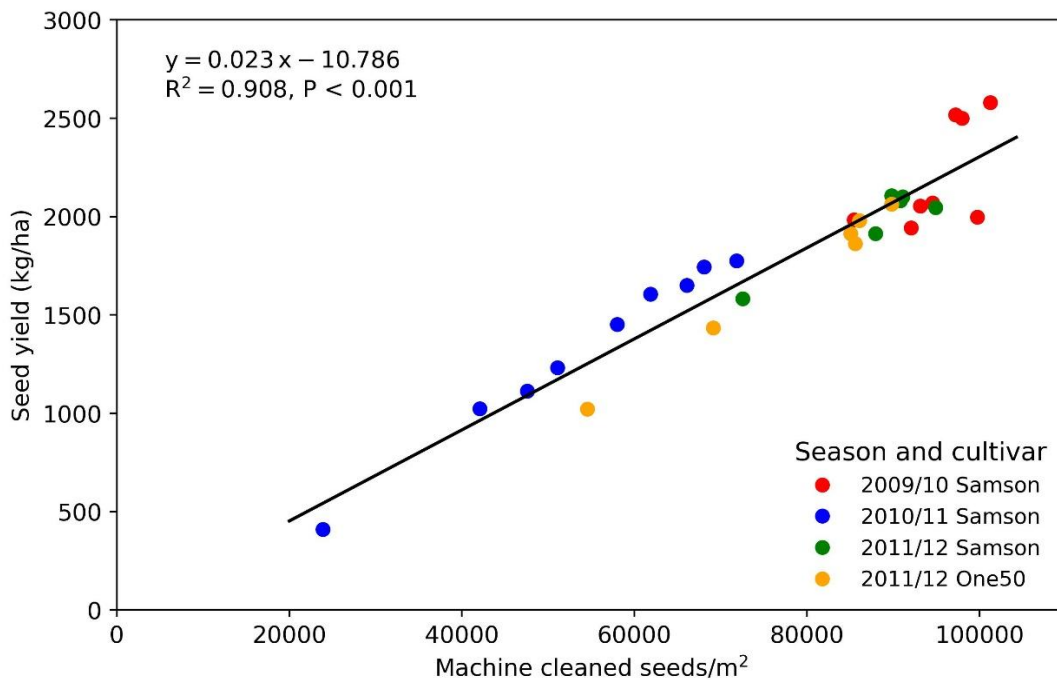


Figure 1. Seed yield response to the number of seed per square meter for two perennial ryegrass cultivars grown over three seasons and treated with different irrigation quantities and timings near Chertsey, Canterbury, New Zealand between 2009 and 2012

In season one there was no difference ($P < 0.05$) in the number of seeds produced per square meter with a weak positive Pearson correlation coefficient ($r = 0.63$) suggesting the seed yield response was driven by changes in TSW. In season two the number of seeds produced per square meter increased ($P < 0.05$) as seed yield increased ($r = 0.996$). In season three, there was no interaction between irrigation treatment and cultivar, but all irrigation treatments were different ($P < 0.05$) from the untreated. Overall, there was a strong correlation between the number of seeds per square meter and final seed yield ($r = 0.953$, $P < 0.001$) (Figure 2).

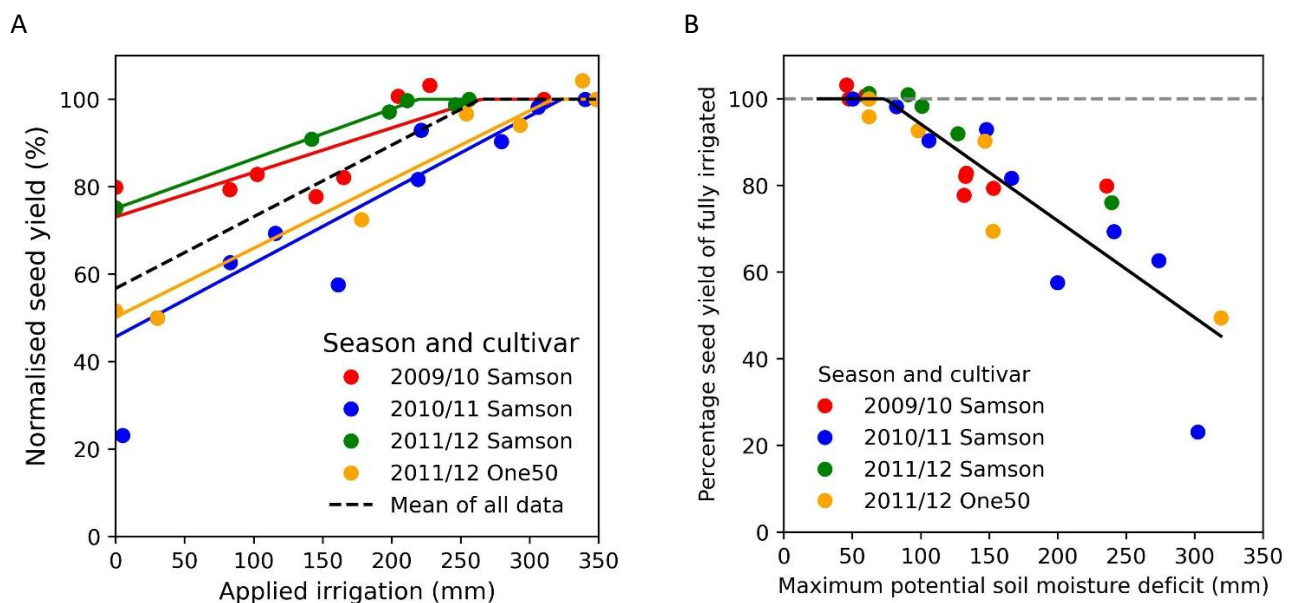


Figure 2. Normalised seed yield response of two perennial ryegrass cultivars following different irrigation treatments over three seasons and B normalised seed yield response of two perennial ryegrass cultivars following different drought intensities expressed as maximum potential soil moisture deficit when grown near Chertsey, Canterbury, New Zealand between 2009 and 2012. Breakpoint = 75

mm (± 14), Slope = $-0.223 (\pm 0.026) \% \text{ mm}^{-1}$ additional deficit, seed yield at 100% is 2105 kg ha^{-1} , $R^2 = 0.80$.

On average over all three seasons, seed yield increased with irrigation up to 268 mm. However, the appropriate application rates varied by season and cultivar e.g. for 'Grasslands Samson' appropriate irrigation quantities ranged from between 219 to 322 mm depending on seasonal rainfall and ET. Deficit irrigation, replacing 50-66% of PET, gave the same seed yield response as full replacement of PET due to the ability of the soil to capture and hold rainfall when it occurred. In fully irrigated treatments rainfall was commonly lost as drainage.

Table 2. Seed yield of two perennial ryegrass cultivars in response to six irrigation treatments when grown at FAR Arable Site, Chertsey Canterbury, New Zealand in the 2011-12 growing season.

Trt#	Treatment	'Grasslands Samson'		'One50'		Trt Mean (kg ha ⁻¹)
		Applied ¹ (mm)	Seed yield (kg ha ⁻¹)	Applied (mm)	Seed yield (kg ha ⁻¹)	
1	No irrigation (Nil)	0	1583	0	1020	1302
2	Replace ET (R-ET)	299	2081	391	2064	2072
3	Nil to anthesis f.b. R-ET	198	2245	293	1862	1954
4	R-ET to anthesis f.b. Nil	142	1914	178	1434	1674
5	Replace 50% ET	199	2100	254	1913	2007
6	R-ET each 3 or 4 days	297	2106	389	1979	2042
Cultivar mean			2014		1687	
Term			P value		LSD _{0.05}	
Cultivar			<0.001		*	
Treatment			<0.001		261	
Cultivar*Treatment			0.034		97	
S.E.M			52			

Note: f.b. = followed by, ¹ = applied irrigation, * = significantly different

The critical deficit (D_c) for this soil Templeton silt loam was identified as 75 mm above which seed yield declined at $0.223\% \text{ mm}^{-1}$ or 4.7 kg mm^{-1} . Therefore, on a warm summer day with a PET of 4.5 mm, each day above D_c costs $\$52 \text{ ha}^{-1}$ at a seed price of $\$2.50$. The seed yield decline was consistent among years and cultivars with increasing deficit (PSMD), regardless of when drought occurred. The consistency demonstrates that the timing of water stress was less critical than its severity (Figure 2B). Thus, the greater yield losses observed during post-anthesis drought (e.g. 'One50') were likely a consequence of higher daily PET during this phase of development, which caused D_c to be reached more rapidly, rather than greater sensitivity of the crop to water stress during later growth stages. Similar results have been shown for wheat, barley and tall fescue (Huetting et al. 2013; Jamieson et al. 1995). Commonly D_c can be estimated as 50% of the available water content (AWC) (Brown et al. 2010), for this Templeton silt loam with gravels below approximately 60 cm the AWC is approx. 120 mm (Lilburne et al. 2012) making D_c 63% of the AWC.

Summary

On average over all three seasons, seed yield increased with irrigation up to 268 mm. However, the appropriate application rates varied by season and cultivar e.g. for 'Grasslands Samson' appropriate irrigation quantities ranged from between 219 to 322 mm depending on seasonal rainfall and ET. Deficit irrigation, replacing 50 66% of PET, gave the same seed yield response as full replacement of PET due to the ability of the soil to capture and hold rainfall, thereby keeping the soil moisture status above D_c . In fully irrigated treatments, rainfall was commonly lost as drainage when the soil profile was near field full.

Acknowledgements

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References

- Brown H, Martin D and Craigie R (2010) Irrigation management for cropping - a grower guide., FAR Focus. Foundation for Arable Research, Lincoln New Zealand.
- Chastain TG, King CM, Garbacik CJ, Young WC III and Wysocki DJ (2015) Irrigation frequency and seasonal timing effects on perennial ryegrass(*Lolium perenne* L.) seed production. *Field Crops Research* **180**, 126-134.
- Chynoweth RJ and Moot DJ (2017) Seed growth of three perennial ryegrass cultivars sown on two dates and treated with trinexapac ethyl straw shortener. *Grass Forage Sci* **72**, 271–280. <https://doi.org/10.1111/gfs.12236>.
- Chynoweth RJ, Pyke N, Rolston MP and Kelly M (2015) Trends in New Zealand herbage seed production; 2004-2014. *Agronomy New Zealand* **45**, 47-56.
- Hebblethwaite PD (1977) Irrigation and Nitrogen Studies in s. 23 Ryegrass Grown for Seed: 1. Growth, Development, Seed Yield Components and Seed Yield. *The Journal of Agricultural Science* **88**, 605-614. <https://doi.org/10.1017/S002185960003728X>.
- Huetting KD, Chastain TG, Garbacik CJ, Young WC III and Wysocki DJ (2013) Spring irrigation of tall fescue for seed production. *Field crops research* **144**, 297-304. <https://doi.org/10.1016/j.fcr.2013.01.023>.
- Jamieson P (1982) Comparison of methods of estimating maximum evapotranspiration from a barley crop. *New Zealand Journal of Science* **25**, 175-181.
- Jamieson PD, Martin RJ and Francis GS (1995) Drought influences on grain yield of barley, wheat, and maize. *New Zealand Journal of Crop and Horticultural Science* **23**, 55–66. <https://doi.org/10.1080/01140671.1995.9513868>.
- Lilburne LR, Hewitt AE and Webb TW (2012) Soil and informatics science combine to develop S-map: A new generation soil information system for New Zealand. *Geoderma* **170**, 232–238. <https://doi.org/10.1016/j.geoderma.2011.11.012>.
- NIWA (2025) Drought indicator charts. National Institute of Water and Atmospheric Research. Retrieved from <https://niwa.co.nz/climate-and-weather/nz-drought-indicator-products-and-information/drought-indicator-charts>.
- Rowarth J, Chapman H, Novis P and Rolston M (1997). Water stress and seed yield in perennial ryegrass (*Lolium perenne* L.) growth in pots. *Journal of Applied Seed Production* **15**, 89-92.
- Seabold S and Perktold J (2010) Statsmodels: Econometric and statistical modeling with python, in: Proceedings of the 9th Python in Science Conference (SciPy 2010). Presented at the 9th Python in Science Conference, pp. 92-96.

Reducing grass seed cleaning loss by improving combine setups

Chris Smith^{1*} and Phil Rolston²

Abstract

Grass seed cleaning companies in New Zealand are reporting increasing difficulties with machine dressing of field-dressed seed, with cleaning losses now typically ranging from 21-24%, compared with less than 15% in the past.

Seed cleaning of six representative samples of ryegrass offal showed a large proportion of short (<30 mm) straw, suggesting over-threshing of seed heads at harvest. As a result, the Foundation for Arable Research (FAR) conducted a series of grower clinics in the 2024 and 2025 harvest seasons aimed at improving combine harvester performance and reducing harvest losses. These workshops provided a comprehensive review of combine harvester operations, focusing on productivity, measurement of losses, and practical strategies to mitigate against any losses. The 2025 sessions placed additional emphasis on improving sample quality in the grain tank, particularly for small-seeded crops such as grasses.

The primary adjustments made by growers through the workshops focused initially on ensuring the proper setup of their threshing systems. Subsequently, they fine-tuned the machine settings, by adjustments such as increasing the clearance of the concaves to reduce aggressive threshing and minimise straw breakage. Throughout this process, growers measured and monitored the impact of each adjustment to assess its effectiveness.

These modifications improved seed separation and resulted in a cleaner field-dressed sample with less broken straw. Participating growers reported not only significant reductions in harvesting losses but also increased confidence in machine setup, allowing for faster harvesting and enhanced overall productivity. Notably, dressing plants reported a drop in dressing losses in cocksfoot, averaging around 10%, compared with a previous three-year average of 15-18%.

Keywords: harvest, sample quality, measure, productivity, efficiency

Introduction

Grass seed production is one of the most technically demanding areas of arable farming. Unlike bulk grain crops, where losses can be easier to mitigate, small-seeded species such as ryegrass or cocksfoot demand precision at every stage of the process. Over the last few seasons, however, seed cleaning plants across New Zealand have reported increasing issues with the quality of field-dressed seed arriving from the combine. Losses during the cleaning process now typically sit between 21 and 24%, compared with historical levels of less than 15%.

This increase in dressing losses represents not only an economic cost to growers and processors, but is also a warning sign: something is going wrong at harvest. In response, the Foundation for Arable Research (FAR) set out to work with growers to diagnose the problem, test solutions, and build confidence in combine harvester setup for small-seeded crops.

Investigations quickly pointed to the combine harvester. Six representative ryegrass offal samples from cleaning plants showed an average dressing loss of 23.5% (Table 1). A large proportion of this was short straw (<30 mm), accounting for 30% of material on average. This strongly indicated over-threshing during harvesting.

For small seeds, this over-threshing is particularly damaging. Excess broken straw makes separation less efficient, increases losses during cleaning, and ultimately costs growers yield and processors time and money. The question became: how can growers adjust their combines to strike the right balance - effective threshing without damaging the sample - and measure the results easily.

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Table 1. Composition of six representative offal samples from field-dressed ryegrass (Eg FDS31RYE) following seed cleaning.

	FDS31RYE	FDS37RYE	FDS22RYE	FDS48RYE	FDS33RYE	FDS29RYE	
	offal 1	offal 2	offal 3	offal 4	offal 5	offal 6	AVG
Dressing loss%	23	23	27	25	22	21	23.5
FRACTION	%	%	%	%	%	%	%
Straw (95% <30 mm)	28.6	20.7	30.8	72.1	13.1	14.5	30.0
Empty seed & leaf	23.9	28.1	23.2	9.6	12.8	29.1	21.1
Ryegrass seed **	0	22.5	35.8	6.9	40.7	0	17.7
Light ryegrass**	20.4	0.0	0	0	0	49.3	11.6
Barley/grain	15.1	5.2	0	0	0	0	3.4
Doubles + short rachis	11.6	9.7	3.6	3.8	10.3	2.8	7.0
Weed seed	0.4	12.4*	4.2**	7.6*	23.2*	4.2*	8.7
Soil	0	1.3	0	0	0	0	0.2
TOTAL	100.0	100.0	100.0	100.0	100.0	100.0	
**Ryegrass TSW (g)	1.27	2.5	2.17	2.00	2.39	1.48	
		*soft brome	**bromes & awn seed				

Materials and Methods

The Grower Clinics. To answer this, FAR launched a series of grower clinics during the 2024 and 2025 harvest seasons. These were practical, hands-on workshops designed to help growers step through their combine harvester from “front to back.” Looking at the four major brands in New Zealand; Claas, Case, New Holland and John Deere, with international experts on each brand. The clinics were then followed by ‘in-field’ visits at harvest focusing on specific machines, adjusting setups, measuring changes and recording the results, to optimise performance and productivity.

The focus of the clinics was on three pillars:

1. Productivity: ensuring combines could run efficiently, maximising output without sacrificing sample quality.
2. Measurement: equipping growers with tools to quantify losses and assess the effect of adjustments. Laying out protocols for a systematic approach to machine adjustments and methods of measuring losses as a consequence of those changes. Reinforcing the adage: “You cannot manage what you do not measure,” highlighting the need for systematic loss quantification.
3. Practical strategies: demonstrating how small, incremental changes to machine settings could deliver measurable improvements.

Importantly, the sessions were focused on knowledge exchange where growers were encouraged to share their experiences of what worked and what didn’t. Once harvest came around growers experimented, measured, and learned in real-time, gaining the confidence to adjust their machines beyond “default” factory settings.

Results

Measured harvest losses. Field measurements of harvest losses across 13 growers revealed a wide range, from less than 1% to more than 9% in ryegrass crops (Figure 1). During the clinics, many growers were able to adjust their machine settings, reducing losses and moving from the higher end of this range towards the lower end.

Grower adjustments. For most growers, the starting point was the threshing system. By slightly increasing the clearance of the concaves, machines became less aggressive, reducing straw breakage. Once this baseline was established, fine-tuning followed: airflow adjustments, sieve settings, and rotor speed tweaks.

The key was iteration. Growers measured losses behind the combine, checked the sample in the grain tank, weighing a known volume of tank sample to gauge the percentage of offal to good seed and

adjusted again. Data supplied by DLF demonstrated that increasing dressing loss was strongly associated with reduced sample weight in diploid ryegrass (Figure 2). This highlights the practical cost of poor combine setup - higher losses translate directly into lower cleaned seed yield. With each adjustment, they could see the immediate impact of their decisions - turning what had once been guesswork into an evidence-based process.

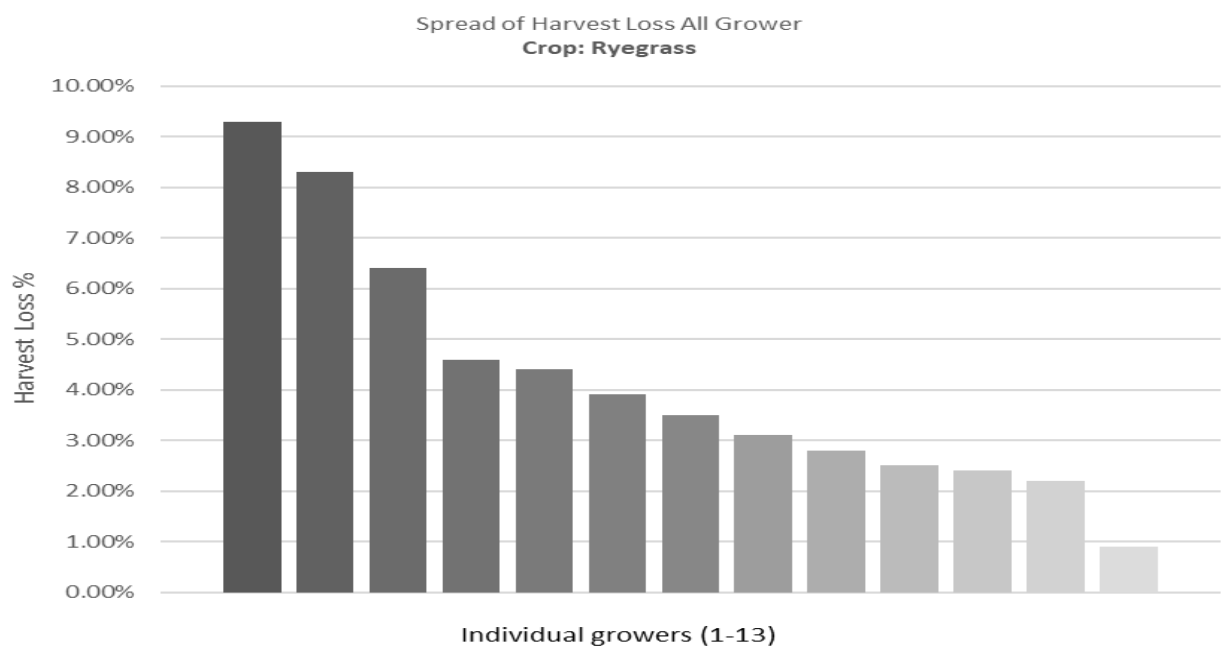


Figure 1. Harvest losses (%) measured off a combine for 13 ryegrass growers in the 2025 season.

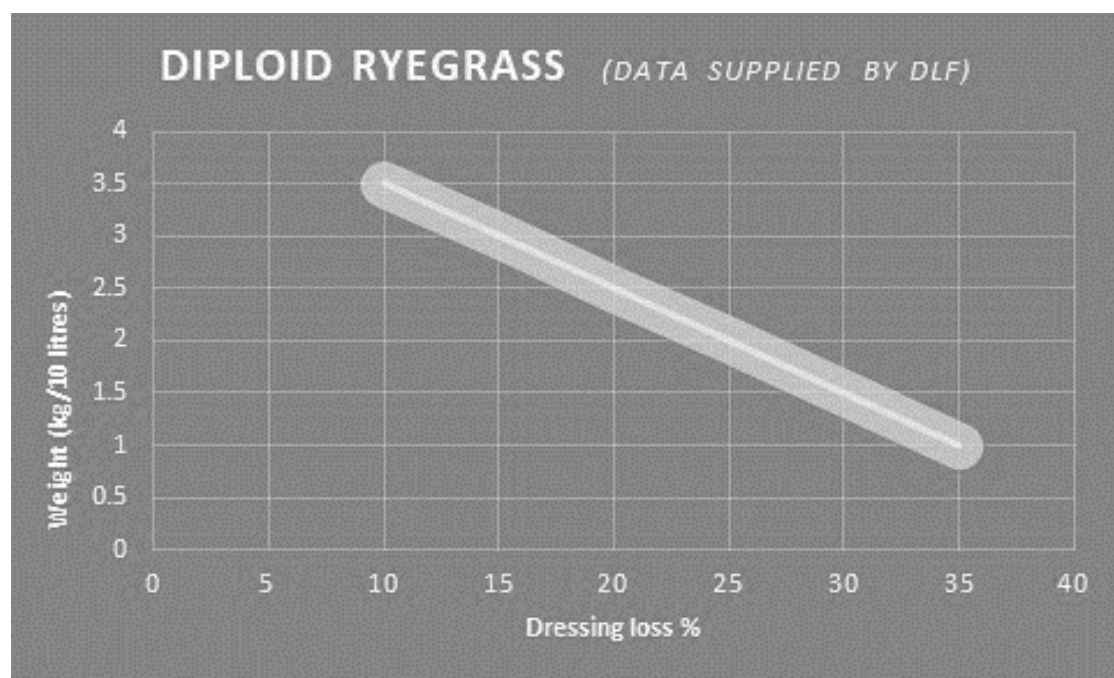


Figure 2. Relationship between dressing loss (%) and seed weight ($\text{kg } 10 \text{ L}^{-1}$) in diploid ryegrass.

Resulting benefits. Participating growers reported:

1. Lower harvest losses: Less seed left in the field, reducing both immediate yield loss and the burden of volunteer plants in subsequent seasons.
2. Cleaner samples: Less broken straw and offal in the tank, and related benefits that brings to cartage, the dressing plant storage and processing.
3. Greater efficiency: From higher productivity, enhanced harvesting capacity, potential lower engine load and therefore reduced fuel consumption per hectare.
4. Increased confidence: Growers felt empowered to take control of machine setup rather than relying on “default combine factory settings.”

Dressing plant data. Crucially, seed dressing plants noticed the difference too. For example, losses in cocksfoot for one seed cleaning company in Canterbury fell to an average of around 10% - a marked improvement on the previous three-year average of 15-18%.

Discussion

These findings highlight a simple but powerful truth: in small-seeded crops, small changes in machine setup can make a big difference. Over-threshing, while often unintentional, is costly. Yet with the right knowledge and measurement techniques, growers can prevent it, improving both profitability and efficiency.

The wider implications are significant. Less wastage means more saleable seed, less energy spent on cleaning, and a smoother relationship between growers and seed plants. At the same time, the process of learning and adapting builds capacity and resilience within the farming community.

Looking ahead, there is scope to go further. Technologies such as more precise on-combine sensors, digital advisors, and better understanding of automated settings could help optimise machine performance in real-time. But the foundation remains the same: understanding the principles of combine operation and taking the time to measure and adjust accordingly.

Four key learnings were:

1. Rising seed cleaning losses are linked to combine harvester setup including over-threshing at harvest.
2. Practical, hands-on clinics enabled growers to measure, identify and correct machine setup issues with confidence. With adjustments made one at a time, including increased concave clearance, reduced straw breakage and improved sample quality was possible.
3. Dressing plant data confirmed tangible benefits, with losses in cocksfoot falling significantly compared with recent averages.
4. Ultimately, investing time in setup and measurement pays off in higher quality seed, greater efficiency, and stronger grower confidence.

Acknowledgements

The Foundation for Arable Research thanks the participating growers, seed cleaning plants, and industry partners who contributed to these workshops and shared their experiences, especially Peter Broley (Primary Sales Australia), the independent experts Kassie van der Westhuizen, Brett Asphar, and Murray Skayman.

Trinexapac-ethyl and spring nitrogen effects on orchardgrass seed development

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Abstract

Orchardgrass (*Dactylis glomerata* L.) is an important forage seed crop, but unlike other cool-season grass seed crops such as perennial ryegrass and tall fescue, seed yields have not increased over time. Research from the literature suggests that plant growth regulators (PGRs), such as trinexapac-ethyl (TE), and spring nitrogen (N) application increase seed yield in orchardgrass by increasing seed number. However, no research has investigated the effects of PGRs and spring N on orchardgrass seed development. Field trials were conducted in 2018 and 2019 to investigate orchardgrass seed development and the effects of PGR and spring N treatments on this process. Treatments included an untreated control, TE (210 g ai ha⁻¹), spring N (112 kg ha⁻¹), and TE + N. Regression analyses were used to elucidate seed development in three spikelet positions: distal, central, and proximal. In 2018, seed weight increased over growing degree days (GDD) in a bi-phasic segmented pattern from distal and central spikelets, but increases were linear from proximal spikelets. In 2019, seed weight increased in proximal spikelets following a bi-phasic segmented function, and in central spikelets, the seed weight increase was also bi-phasic, except for the TE treatment. Seed growth rate varied among spikelet positions, ranging from 0.22 to 0.34 mg GDD⁻¹ per 100 seeds. The seed growth rate varied among TE and N treatments, ranging from 0.31 to 0.47 mg GDD⁻¹ per 100 seed. The TE + N treatment had the shortest seed filling duration and one of the smallest seed growth rate values, producing low seed weight. The TE + N treatment produced high seed number and seed yield, indicating a reduction in seed abortion or shattering. Seed carbon (C) and N content increased during seed development and peak deposition preceded physiological maturity. There was no effect of TE on deposition of C or N in orchardgrass seed.

Keywords: seed development, seed physiology, plant growth regulators, nitrogen, orchardgrass

Introduction

Orchardgrass (*Dactylis glomerata* L.) is a cool-season forage grass species that is native to western and central Europe. Orchardgrass seed yields are variable, and no studies have investigated seed development in this important grass species.

Seed development begins with double fertilisation in the floret and ends with mature seed. Seed development in cool-season grass has two phases which are seed growth and seed maturation (Grabe 1956). During seed growth, there is an initial and short period with cell division but no change in seed weight, followed by a linear increase in seed weight (Stoddart 1968). Increased seed weight in seed growth is a result of carbon (C) and nitrogen (N) accumulation in the endosperm and growth of the embryo (Weber et al. 1998). When maximum seed weight is reached, seed has attained physiological maturity. Seed maturation is marked by the start of seed moisture content decrease while the seed weight does change appreciably.

The seed development process affects seed yield in orchardgrass. Seed yield is primarily influenced by two components: seed number and seed weight (Chastain & Young 1998). Seed number is determined by establishment of seed yield potential at anthesis and realised during seed harvest (Chastain & Young 1998). Seed weight is determined by the seed growth period (seed filling duration = SFD) and by seed growth rate (SGR) (Egli 1998).

Plant growth regulators (PGRs), such as trinexapac-ethyl (TE) have been adopted in grass seed crop production around the globe and have increased seed yield in orchardgrass. Chastain et al. (2014) found that TE PGR influenced seed partitioning in perennial ryegrass (*Lolium perenne* L.) by increasing seed m⁻² in each spikelet. One of the studies that examines the effect of TE on orchardgrass reported

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seed yield increase by 37% in New Zealand with a TE application rate between 100 to 300 g ai ha⁻¹ (Rolston et al. 2014). Anderson et al. (2024) showed that orchardgrass seed yield increases were due to increased seeds m⁻². It has not been previously known whether there is an increase in carbon (C) and N available to fill the increased number of seeds. No information is available for the effects of TE and spring N on orchardgrass seed development, and no work has been conducted on effects of TE on C and N assimilation in grass seed. The objective of this study is to investigate seed development in orchardgrass and ascertain the effects of TE PGR and spring N on seed weight, seed growth (SGR and SFD), and progress towards physiological maturity in orchardgrass seed.

Materials and Methods

Orchardgrass ('Persist') field trials were conducted over two crop seasons (2018 and 2019) at Oregon State University's Hyslop Experimental Farm near Corvallis, Oregon (USA). Growing degree days (GDD) were used to mark progress of the crop during seed development and were calculated from 1 September each year. Air temperatures observed at Hyslop Farm were used for GDD calculation with a 5°C base temperature.

The experimental design was a split-plot, randomised block design with three replications. Main plots (11.6 x 13.4 m) were spring-applied N and subplots (3.4 m x 11.6 m) were PGR treatments. Treatments included: control (no N, no TE), TE (210 g ai ha⁻¹ TE), N (spring N at 112 kg N ha⁻¹), and TE + N (both TE and N). Spring N treatments consisted of dry granular urea (46-0-0), applied prior to stem elongation (BBCH 29; mean = 791 GDD). Applications of TE were made at early stem elongation (BBCH 32; mean = 858 GDD). The PGR was applied at walking speed by using a bicycle-type boom sprayer operated at 138 kPa with XR Teejet 8003VS nozzles.

Five panicles were collected periodically from random locations within each plot starting at BBCH 65 (peak anthesis) and continuing until BBCH 85 (seed harvest) in both years. Panicle samples were immediately transported to the laboratory for determination of floret number, seed number, floret weight, and seed weight. In 2018, 90 seed samples were collected from each of the three spikelet positions (distal, central, and proximal) and each of the three different panicle positions (apical, medial, and basal). One treatment was sampled on each sampling day. In 2019, 120 seed samples were collected on each sampling day from each of the three spikelet positions (distal, central, and proximal) from each treatment.

Seeds were differentiated from unfertilised florets or those ovules that aborted development shortly after fertilisation by microscopic examination (Chastain et al. 2014). The dry weight of florets and seeds were determined after oven drying. Seed and florets were placed in a metal sample container and weighed prior to oven-drying. Sample containers were placed in a laboratory air-oven and dried at 130°C for 2 hours. After drying was complete, a cover was placed on the sample container and was cooled to room temperature prior to weighing. The SFD was determined by subtracting GDD at pollination (start point of seed filling) from GDD at maximum seed weight.

Proximal and central seed (50 mg each) were collected to determine C and N deposition. Total N and C content of seed were determined by using an automated LECO CNS-2000 macro analyser (LECO Corporation, St. Joseph, MI). Seed C and N filling periods were determined by subtracting the start point of the filling period from the GDD where maximum C and N content was recorded.

Regression analysis was conducted to ascertain spring N and TE effects on seed development characteristics and C and N deposition using PROC GLM and PROC NLIN in SAS 9.4. Segmented or linear models were chosen if the data fits and the models were compared for goodness of fit. Growing degree days (base 5°C) were used in curve-fitting as the abscissa for seed development relationships.

Results and Discussion

The ANOVA revealed that there was a significant change in seed weight over GDD ($P < 0.05$) in both 2018 and 2019. In 2018, seed weight increase followed a bi-phasic pattern in both distal and central seed, while seed weight increase was linear in proximal seed (data not shown). In 2019, there were insufficient numbers of seed found in distal spikelet positions, thus only seed from central and proximal spikelet positions were examined. Seed weight from proximal positions increased ($P < 0.0001$) and

followed a segmented model for all four treatments tested ($R^2 > 0.90$) (Figure 1). Variation in the results is due to study being conducted in a field unlike the clonal seed sown in greenhouse trials in the Warringa et al. (1998) study. In contrast to the seed weight fitted to segmented model in this study, seed weight was fitted to Gompertz model in perennial ryegrass (Warringa et al. 1998), while Chynoweth and Moot (2017) fitted seed weight to a logistic model.

Seed growth rate (SGR) was 0.36 mg GDD^{-1} per 100 seed in proximal spikelet positions with the control. Seed filling ended at 1483 GDD in the control treatment with a maximum weight of 122 mg per 100 seed in proximal spikelet positions. The seed-filling period for the control started earlier at 1225 GDD compared to other treatments where seed-filling started at 1246 GDD. Maximum seed weight for seed in proximal spikelet positions was recorded at 1493 and 1498 GDD for the N and TE treatments, respectively (Figure 1). The SGR was higher in the TE treatment (0.36 mg GDD^{-1} per 100 seed) than with the N treatment (0.34 mg GDD^{-1} per 100 seed). With the combination of TE + N, SGR was 0.31 mg GDD^{-1} per 100 seed and maximum seed weight was reached at 1453 GDD.

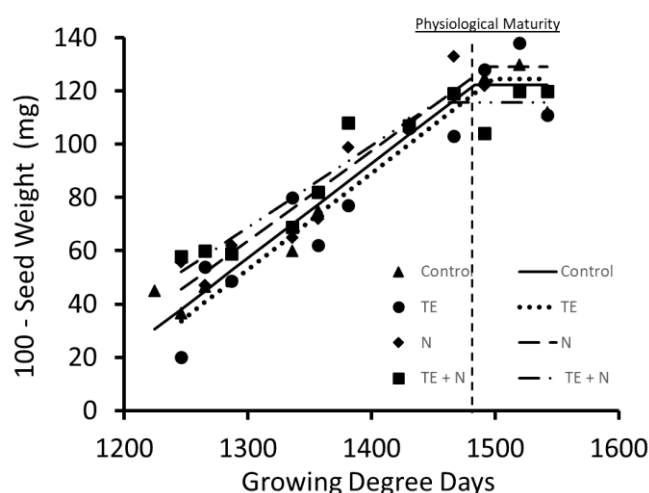


Figure 1. Relationship of growing degree days (GDD) and seed weight in orchardgrass over seed development in proximal spikelet positions in 2019. Physiological maturity is based on maximum seed weight in the control treatment.

The weight of seed developing in central spikelet positions increased ($P < 0.01$) for all treatments, following a segmented model with the exception for seed with the TE treatment where linear growth in seed weight was observed (Figure 2). The SGR for seed in the central spikelet positions was 0.47 mg GDD^{-1} per 100 seed with the control treatment. In the control treatment, maximum seed weight for central spikelet seed was 99 mg per 100 seed at 1420 GDD. Unlike other treatments, seed growth of the TE treatment followed a linear function without a plateau and a maximum seed weight in the central spikelet positions. The SGR for seed in the central spikelets for the TE treatment was 0.32 mg GDD^{-1} per 100 seed. The N treatment had an SGR of 0.33 mg GDD^{-1} per 100 seed in the central spikelet positions. The maximum seed weight for the N treatment was 108 mg per 100 seed and was observed at 1471 GDD in the central spikelet positions. The combination of TE + N produced an SGR of 0.31 mg GDD^{-1} per 100 seed with a maximum seed weight of 98 mg per 100 seed at 1460 GDD. The seed growth rate values for orchardgrass seed were similar to the seed growth rate values for perennial ryegrass seed after converting the values from Warringa et al. (1998) to GDD (Base 5°C). Both species appear to fill seed at the same seed growth rate, but SFD is longer in perennial ryegrass.

Deposition of C and N were different during seed growth with C accumulation exhibiting a quadratic function to a plateau in the segmented model (Figure 3), whereas N accumulation followed a linear to plateau segmented function (data not shown). Carbon content in seed increased over GDD ($P < 0.0005$) during seed development (Figure 3). Among treatments, deposition of C in seed was not different. Maximum seed C content for treatments ranged from 451 to 456 mg g^{-1} . The control had a C accumulation rate of $0.559 \text{ mg g}^{-1} \text{ GDD}^{-1}$ and the peak C content was reached at 1452 GDD. The C accumulation rate was $0.518 \text{ mg g}^{-1} \text{ GDD}^{-1}$ with the TE treatment and the accumulation of C peaked at

1462 GDD. The deposition of C peaked early at 1415 GDD with the N treatment. The rate of C accumulation for the N treatment was $0.887 \text{ mg g}^{-1} \text{ GDD}^{-1}$. The rate of C content increase was $0.515 \text{ mg g}^{-1} \text{ GDD}^{-1}$ with the TE + N treatment and deposition peaked at 1457 GDD. The highest rate of C accumulation was with the N treatment while other treatments had a lower but similar rate of C accumulation. The N treatment not only had the highest C accumulation rate but also reached peak C deposition earliest. The C content in orchardgrass seed increased over GDD corresponding to the increase in starch accumulated in the endosperm as observed in perennial ryegrass (Warringa et al. 1998). Such increase in starch was the most important contributor to seed development. While starch increase was not measured in this study, it is likely that starch increased due to the increase in carbon content in seed.

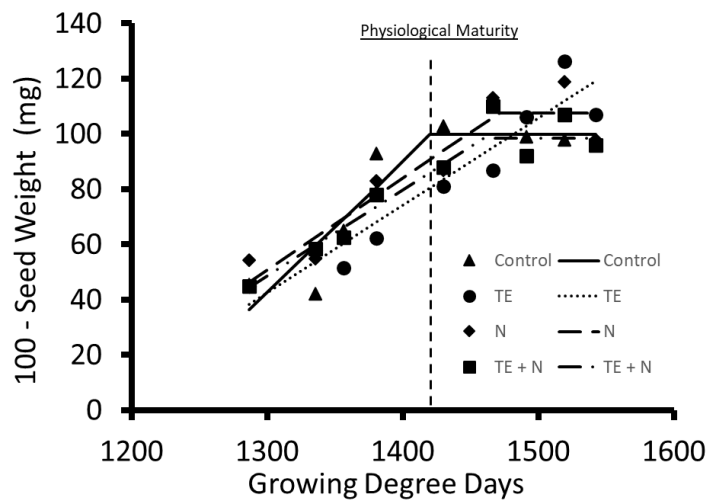


Figure 2. Relationship of growing degree days (GDD) and seed weight in orchardgrass over seed development in central spikelet positions in 2019. Physiological maturity is based on maximum seed weight in the control treatment.

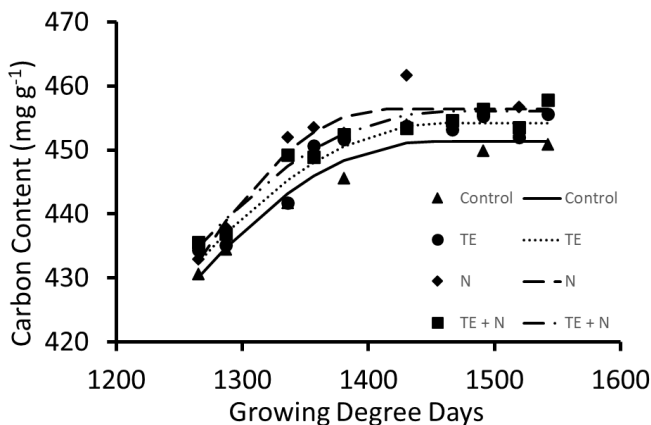


Figure 3. Relationship of growing degree days (GDD) and seed carbon (C) content over the course of seed development in orchardgrass in 2019. Seed (C) content was determined on seed collected from the proximal spikelet position. Lines represent regression function for each treatment.

Nitrogen content increased over GDD with all treatments ($P \leq 0.001$). The deposition of N over GDD with the TE + N treatment followed a linear function whereas N deposition in other treatments followed a segmented model (linear to plateau). With the control, the N deposition rate was $0.0637 \text{ mg g}^{-1} \text{ GDD}^{-1}$ with peak N accumulation occurring at 1457 GDD. The peak N content for the control was 29.7 mg N g^{-1} . The N deposition rate in the TE treatment was $0.0759 \text{ mg g}^{-1} \text{ GDD}^{-1}$ and the peak N content in seed was observed at 1438 GDD at a seed N content of 25.6 mg g^{-1} . With the N treatment, the rate of N deposition in seed was $0.0860 \text{ mg g}^{-1} \text{ GDD}^{-1}$ and the peak N content was 31.1 mg N g^{-1} at 1439 GDD. The TE + N treatment produced a rate of N deposition in seed of $0.0494 \text{ mg g}^{-1} \text{ GDD}^{-1}$. The rate of N deposition tended to be greatest in TE and N treatments and lower in control and TE + N

treatments. Results from this study concedes with N increase in perennial ryegrass seed which follows a segmented bi-phasic model (Warringa et al. 1998).

There was variation among treatments in the duration of N filling in seed (data not shown). For the control, duration of N filling was 192 GDD. The duration of N filling in TE and N treatments were 173 GDD and 174 GDD, respectively. Peak N filling in seed ranged from 1438 to 1457 GDD in proximal spikelets. These peaks in N accumulation took place before physiological maturity of the seed in proximal spikelets at 1453 GDD to 1498 GDD. This is consistent with similar results found in perennial ryegrass where starch accumulation preceded seed physiological maturity (Warringa et al. 1998).

The primary effects of TE PGR in increasing seed yield appear to be through increasing seed number but there does not seem to be any consistent influence of TE on the deposition of C or N in the seed. However, there were consistent effects of N application on the content of C and N deposited in the seed – both were increased with N application. This study increases our understanding of the development of orchardgrass seed and provides an underlying explanation for the seed yield increases observed in Anderson et al. (2024).

References

- Anderson NP, Morad MM and Chastain TG (2024) Spring nitrogen and plant growth regulator effects on seed yield of orchardgrass. *Crop Science* **64**(6), 3533-3540.
- Chastain TG and Young WC III (1998) Vegetative plant development and seed production in cool-season perennial grasses. *Seed Science Research* **8**, 295-301.
- Chastain TG, Young WC III, Silberstein TB and Garbacik CJ (2014) Performance of trinexapac-ethyl on *Lolium perenne* seed crops in diverse lodging environments. *Field Crops Research* **157**, 65-70.
- Chynoweth RJ and Moot DJ (2017) Seed growth of three perennial ryegrass cultivars sown on two dates and treated with trinexapac ethyl straw shortener. *Grass and Forage Science* **72**(2), 271-280.
- Stoddart JL (1968) Biochemical aspects of seed ripening in grasses. *Journal of the National Institute of Agricultural Botany* **11**, 370-377.
- Warringa JW, Struik PC, De Visser R and Kreuzer ADH (1998) The pattern of flowering, seed set, seed growth and ripening along the ear of *Lolium perenne*. *Functional Plant Biology* **25**, 213-223.

Understanding yield limitations in New Zealand red clover seed crops

Sean Weith^{1*}, Phil Rolston² and Richard Chynoweth³

Abstract

Red clover (*Trifolium pratense* L.) seed yields in New Zealand often fall short of their biological potential, typically producing 200-500 kg ha⁻¹, a range lower and more variable than those seen internationally. To investigate factors influencing realised seed yield, 16 commercial red clover crops were surveyed during the 2023-24 season across South, Mid and North Canterbury, South Island. At each site, pre-desiccation sampling measured aboveground biomass, stem density, flowerhead density, florets per inflorescence, and the proportion of florets containing seeds. These traits were used to estimate seed set potential seed yield, while realised seed yields obtained from growers allowed estimation of harvest losses. Across all sites, 60% of florets contained seed on average (range: 40-80%). While seed yields were higher than anticipated, most crops underperformed relative to their seed set potential, with realised yields ranging from 200 to 850 kg ha⁻¹ (mean: 473 kg ha⁻¹), compared with a mean potential of 790 kg ha⁻¹ (range: 166-2,012 kg ha⁻¹). Correlation analysis showed weak to moderate associations between yield components, with realised yield positively associated with flowers per stem ($r = 0.3$) and negatively with seeds per floret ($r = -0.3$). These results indicate that factors beyond key seed components, particularly canopy management, pollination or harvest losses may be limiting factors. This survey provides a benchmark dataset for identifying key drivers of red clover seed production. Further analysis will examine relationships between plant reproductive traits and final yields to improve understanding of the factors constraining seed production in New Zealand.

Keywords: red clover, survey, seed yield, Canterbury, New Zealand

Introduction

Red clover (*Trifolium pratense* L.) is a widely cultivated forage legume valued for both grazing and seed production. It provides high-quality feed for livestock due to its favourable nutritive value and digestibility, while also supporting farming system sustainability through biological nitrogen fixation. Reliable seed production is essential to ensure the continued supply of high-performing cultivars for livestock farmers and the seed industry. In New Zealand, red clover seed production is concentrated in the growing regions of the Wairarapa, Marlborough and Canterbury, with an estimated annual production area of about 1,000 ha (Chynoweth et al. 2018). Crops are generally managed as multi-year stands, with seed harvested for up to three consecutive years. However, seed yields are typically low to modest, ranging between 200-500 kg ha⁻¹, with the upper end of this range considered commercially desirable, but often not achieved (Karagić et al. 2010). These yields are well below those reported internationally, where diploid red clover cultivars produce between 400 to 500 kg ha⁻¹ in Western and Central Europe (Boller et al. 2010) and 500 to 1,200 kg ha⁻¹ in Oregon, USA (Anderson et al. 2018). The comparatively low and inconsistent yields in New Zealand therefore represent a major limitation to the reliability and profitability of seed production.

Seed yield in red clover is a complex trait influenced by both genetic and environmental factors, but examining the key yield components that underpin crop performance provides a useful approach for understanding yield variation. Among the main determinants are the number of flower heads per plant, the proportion of functional florets and seed number per head (Herrmann et al. 2006; Jing et al. 2021). Because red clover is allogamous with strong self-incompatibility, successful cross-pollination by insects, particularly bumblebees (*Bombus* spp.) and honeybees (*Apis mellifera*), is critical, and reduced pollinator density can markedly lower seed yields (Vleugels et al. 2019). Seed production is further constrained by abiotic stresses such as drought and late frosts, as well as biotic pressures including pests and diseases. In New Zealand, below-average yields have recently been linked to increasing pest pressure, particularly from red clover casebearer (*Coleophora deauratella*) and red

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clover thrips (*Haplothrips niger*) (Chynoweth et al. 2018). These challenges underscore the need to better understand the drivers of seed yield variability across production environments. To examine these factors under New Zealand conditions, a survey of 16 commercial red clover seed paddocks was conducted across South, Mid and North Canterbury during the 2023–24 growing season. The survey aimed to identify the main drivers of seed yield variation in commercial red clover seed production systems.

Materials & Methods

A cohort of 16 commercial red clover paddocks was sampled during the 2023-24 growing season. Sites were selected to represent variation in farm size, location, and management history and were distributed across South, Mid, and North Canterbury.

Three random samples were collected 2-14 days prior to desiccation from each site following the methods described by Oliva et al. (1994), with modifications. Plant material was cut to ground level from an area spanning two adjacent rows over a 50 cm length (approximately 0.3-0.4 m², depending on row spacing). Each sample was weighed fresh, and a bulk subsample of ~150 g was taken. From each subsample, the mean number of stems, flowers per stem, flowerheads and number of florets per inflorescence were recorded. Subsamples were oven-dried at 65°C for ~48 hours in labelled paper bags and dry weights were measured (Anderson et al. 2016).

Reproductive traits were assessed following a method described by Oliva et al. (1994), with modifications. At each site, 150-300 florets were randomly sampled, and the presence or absence of seed was recorded, assuming a maximum of one seed per floret. Thousand seed weight was estimated by weighing 200 seeds. Derived traits including stems m⁻², flower heads m⁻², florets m⁻² and seeds m⁻² were then calculated. These values were used to estimate seed set potential yield (florets producing harvestable seed).

All analyses of data were conducted using R (v 4.5.1) software programming language (R Core Team, 2025). All data was visualised using the 'ggplot2' package (Wickham 2011). Pairwise correlations among measured variables were calculated using the 'psych' package in R (Revelle 2017), with significance ($P \leq 0.05$) tested using the car package (Fox & Weisberg 2018).

Results and Discussion

Across all surveyed sites, the mean realised seed yield was 473 kg ha⁻¹ (range: 200-850 kg ha⁻¹) while seed set potential yield was 790 kg ha⁻¹ (range: 166-2012 kg ha⁻¹) (Figure 1). While yields were higher than anticipated, most crops underperformed relative to their seed set potential, reinforcing the substantial gap between yield achievable after pollination and early seed development and actual achieved yield. Correlation analysis indicated only weak to moderate associations between yield components, with realised seed yield showing a positive relationship with flowers per stem ($r = 0.3$) and a negative relationship with seeds per floret ($r = -0.3$) (data not presented). However, there was no strong relationship between realised seed yield with either florets m⁻² ($r = -0.08$) or seeds m⁻² ($r = -0.19$). These results suggest that factors beyond these key yield components are constraining yield, with canopy management, pollination efficiency or insect feeding likely acting as key bottlenecks. Previous research has highlighted both flowerhead number and seed number per head as important contributors to seed yield (Anderson et al. 2016; Jing et al. 2021). However, unlike the findings of Vleugels et al. (2019), which identified flowerhead number as a major determinant of seed yield, this study found only a weak association between flower heads m⁻² and realised seed yield ($r = -0.13$). This weak relationship may reflect variations in sampling, experimental conditions and unrecorded factors, such as pest damage, irrigation and forage removal, which can influence flowerhead production across sites.

On average, 60% of florets contained seed (Figure 1), suggesting that pollination is not the primary source of yield loss. However, red clover pollination is complex and influenced by factors not examined in this study, including pollinator species present and the availability of alternative pollen and nectar sources (Vleugels et al. 2019). Other unrecorded factors, such as paddock size and distance from pollinator nesting sites, may also affect pollination efficiency and merit further investigation. While

pollination did not appear to be the main driver of yield loss in this study, improvements could still provide modest gains, but are unlikely to close the gap between potential and realised yield on their own.

The gap between potential and realised seed yield suggests that late insect damage or harvest losses in New Zealand red clover are underestimated and may represent a major barrier to achieving maximum yields. Losses at or before harvest can substantially reduce returns, even when pollination and crop management are effective. Addressing this will require greater focus on optimising harvest timing, combine setup, and handling, alongside improvements in canopy and pollination management.

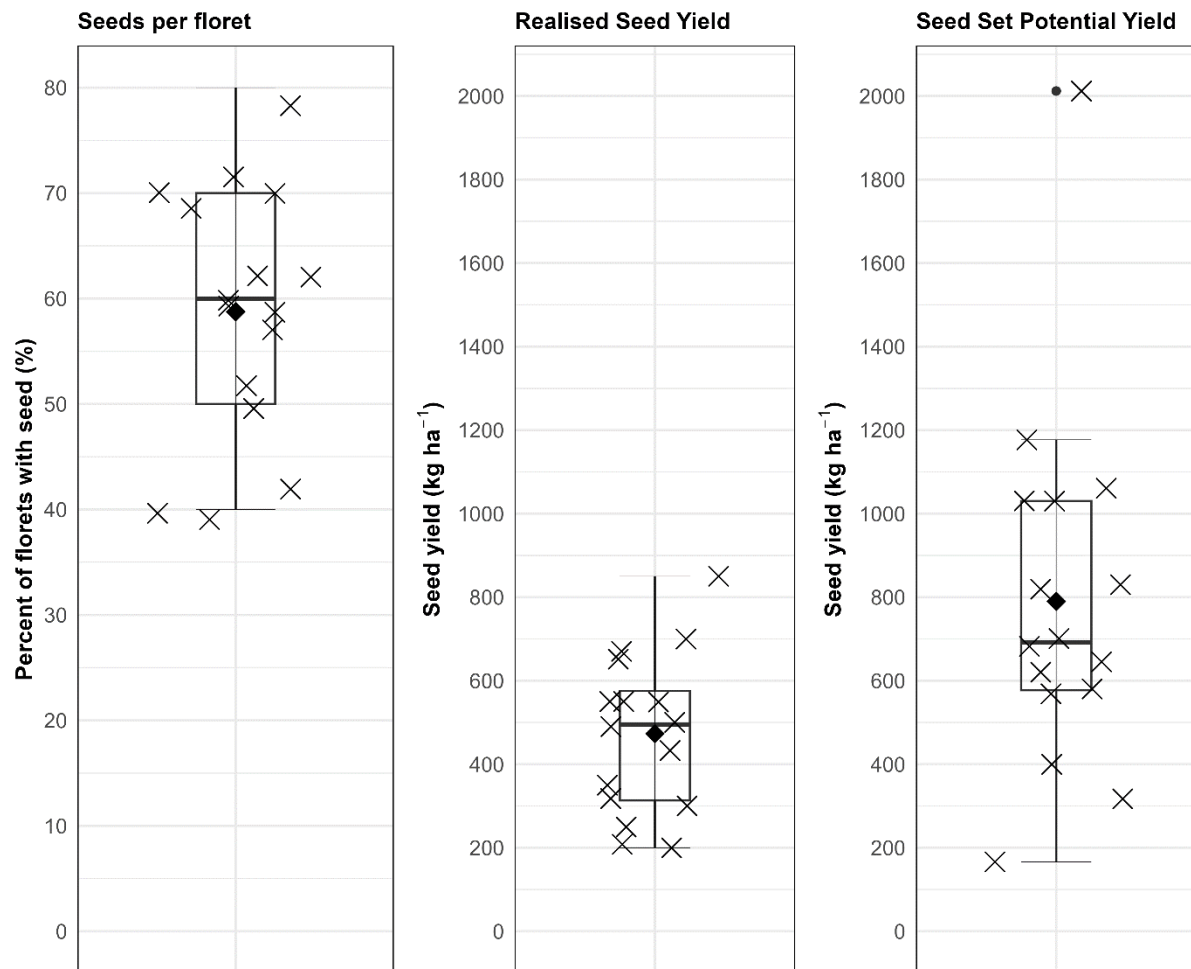


Figure 1. Variability in the percentage of florets containing seed and realised (actual) and potential seed yield (kg ha⁻¹) based on florets with developing seeds from 16 red clover (*Trifolium pratense* L.) crops surveyed across Canterbury, New Zealand during the 2023-24 growing season. In each box, the line represents the median, diamonds show the mean and circles indicate outliers.

Conclusion

There is a huge gap between achieved and achievable red clover seed yield, with most crops performing well below their potential. Weak correlations between yield components indicate that factors beyond flower and seed number may be contributing to seed loss. Harvest losses are likely underestimated and represent a significant barrier to maximum productivity. These findings highlight the need for integrated management of canopy structure, pollination and harvest practices to narrow the yield gap.

References

Anderson NP, Chastain TG and Garbacik CJ (2016) Irrigation and trinexapac-ethyl effects on seed yield in first-and second-year red clover stands. *Agronomy Journal* **108**, 1116-1123.

- Anderson NP, Garbacik C, Chastain T and Elias S (2018) Boron effects on red clover seed production and quality. *Seed Prod. Res* **9**, 9-11.
- Boller B, Schubiger FX and Kölliker R (2010) Red clover. *Fodder crops and amenity grasses*, 439-455.
- Chynoweth R, Rolston P, McNeill M, Hardwick S and Bell O (2018) Red clover casebearer moth (*Coleophora deauratella*) is widespread throughout New Zealand. *New Zealand Plant Protection* **71**, 232-239.
- Fox J and Weisberg S (2018) *An R companion to applied regression*, Los Angeles, CA, Sage publications.
- Herrmann D, Boller B, Studer B, Widmer F and Kölliker R (2006) QTL analysis of seed yield components in red clover (*Trifolium pratense* L.). *Theoretical and applied genetics* **112**, 536-545.
- Jing S, Kryger P and Boelt B (2021) Review of seed yield components and pollination conditions in red clover (*Trifolium pratense* L.) seed production. *Euphytica* **217**, 69.
- Karagić Đ, Jevtić G and Terzić D (2010) Forage legumes seed production in Serbia. *Biotechnology in animal husbandry* **26**, 133-149.
- Oliva RN, Steiner J and Young WC III (1994) Red clover seed production: II. Plant water status on yield and yield components. *Crop science* **34**, 184-192.
- R Core Team (2025) R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
- Revelle W (2017) *An overview of the psych package* [Online]. Available: <http://personality-project.org/r/overview.pdf>. [Accessed 27 April 2022].
- Vleugels T, Amdahl H, Roldán-Ruiz I and Cnops G (2019) Factors underlying seed yield in red clover: review of current knowledge and perspectives. *Agronomy* **9**, 829.
- Wickham H (2011) ggplot2. *Wiley Interdisciplinary Reviews: Computational Statistics* **3**, 180-185.

Forage chicory seed production – An update of techniques in New Zealand and Tasmania

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Abstract

Forage herbs like chicory (*Cichorium intybus* L.) have become an integral part of New Zealand and Australian pastoral systems. These species can be grown either as specialised monocultures or incorporated into pasture mixes, often in combination with clovers.

Forage chicory is a deep rooted, short-lived perennial belonging to the Compositae (Asteraceae) family. Its summer active leafiness is typically grazed in situ. Research and breeding by New Zealand's DSIR Grasslands in the 1970's resulted in the release of the first commercial cultivar 'Grasslands Puna' in 1985. Since then, several cultivars have been released; offering extended periods of forage productivity.

Seed production research was initiated in the early 1980's and set the basic methodology for commercial production. Initial yields ranged from 200-700 kg ha⁻¹, with occasional fields achieving 800-900 kg ha⁻¹. Seed production in Tasmania began in 1995. Chicory seed production in both countries was characterised by variable yields and germination results, and challenges from certain weed groups that restricted market access.

Herbicide research has had some but limited success, with only a small range of suitable herbicides. Greater understanding of crop rotations and cultural controls have enabled seed purity to improve. Asteraceae family weeds (thistle species) remain a challenge. Some pests and diseases can be significantly detrimental, mostly in establishment and early development of the crop. More recent trials have focused on managing crop architecture to reduce excessive vegetative growth and present pollinating insects with easier access to flowers. The field methods to mitigate these challenges are discussed.

Some chicory lines reputedly have some ability to self-pollinate, although most of the pollination is insect cross-pollination. A range of bee and blowfly species are essential and timing of flowering when these are at peak population, and in variable weather conditions is a challenge. As a highly indeterminate crop, chicory presents a significant challenge for pollination, with individual flowers open for only a few hours. Maximising pollination over a flowering period of approximately six weeks is complicated by declining temperatures, shortening daylength, and increasingly inclement weather conditions.

Decisions on cutting (swathing) time is a key decision, early flowers begin to shatter, yet the crop continues to flower, therefore several indicators are required to aid this decision. It takes 2-3 weeks for the crop to dry sufficiently for threshing. Weather conditions at harvest can be challenging and cause some seed losses. Considering the challenges, chicory seed yields and qualities have improved and become more reliable with an average of 750 kg/ha and some crops yielding over 1400 kg/ha.

Keywords: chicory, pollination, herbicides, seed production

Introduction

Since the introduction of 'Grasslands Puna' chicory in 1985 there has been considerable growth in its use in pasture systems in New Zealand (NZ), Australia, South America and recently in Europe and America. It can be used in pasture mixes or as a specialty pasture. The high animal growth rates have driven this growth (refs). Benefits researched include high in essential amino acid; broader mineral intake e.g. vitamins A, B, D, selenium, iron, zinc; and high levels of omega-3 polyunsaturated fats, leading to claimed superior cooking qualities.

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Development of Forage Chicory Seed Production

Initial research was conducted at DSIR Grasslands, Palmerston North, by Hare 1986, Hare, Rolston, et al. 1987, Hare & Rolston 1987, Hare, Rowarth et al. 1990, Rowarth et al. 1996. This established basic methods, many of which are still used 40 years later. Seed yields have improved as both research and seed growers have developed experience and overcome obstacles in establishment, weed, pest and disease control, crop bulk, pollination, cutting, harvest, seed drying and cleaning.

Early seed yields in the development of Puna achieved 200 kg ha⁻¹. Over the next few years, the yield increased to over 500 kg ha⁻¹ in 1987 (Hare et al 1987). Commercial crops showed similar increases from 200 kg ha⁻¹ to 470 kg ha⁻¹ with top field seed yield achieving 700 kg ha⁻¹ in 1990 and 800 kg ha⁻¹ in 1991 (Rowarth, Rolston et al. 1991). Current average seed yield for PGGWS crops is 760 kg ha⁻¹ with top crops achieving 1500 kg ha⁻¹. We have focused our production on growers who have high capabilities and available infrastructure to manage their crops well and this has reduced the lower end of averages. A network of capable field staff monitors crops and are supported by research agronomists to advise growers. Geographic spread around suitable seed production regions also reduces risk of loss to weather events. A small group of regular growers have developed the experience to manage the seasonal vagrancies that plague chicory seed production.

In Tasmania, similar seed yields are achieved. Despite less available infrastructure the advantages of higher temperatures and with less competitive crops achieves better pollination.

Recent developments in Seed Production Techniques

1. Establishment. No further development other than improved large scale cultivation equipment to produce a firm, fine seed bed. Trifluralin at low rates is still widely used. While autumn sowing is common, some growers in Tasmania prefer spring sowing, with grazing and lamb-finishing in the first summer and autumn.

2. Herbicides. The early techniques using flumetsulam from 2-leaf and diflufenican from 4-leaf are still widely used. A range of Group 1 grass herbicides can be used over a wide range of timings. The tri-mix of bromoxynil, ioxynil and mecoprop-P (Axall, now Image®) identified by Hare et al. 1993 is used over NZ crops. This has some plant growth regulator (PGR) effect on the crop which can be a useful tool. Australia has bromoxynil and ioxynil available, and the advantage of imazamox (Raptor®).

Recent herbicide trials have focused on early control of annual and prickly sow thistles (*Sonchus oleraceus* & *Sonchus asper*). The crop needs to grow to 6-leaf before application and with the weed at rosette stage up to 95% control can be achieved with later germinations controlled by the triple mix Image. Saflufenacil (Sharpen®) has also been in trials with greater efficacy but more crop damage than the bromoxynil. Later application at early stem extension (Image timing) of saflufenacil plus bromoxynil and ioxynil shows promise to be used as an alternative to Image. Bentazone and/or metribuzin can be useful for late control of some weeds.

3. Canopy/bulk management. Early productions did not use PGR and crops were very tall and prone to lodging. Currently several tools are used to manage the vegetative bulk during stem extension and flowering period:

- a. The herbicide PGR effect at early stem extension shortens early internodes so stems have a firm base.
- b. At about 2 week intervals a combination of DMI fungicide and chlormequat are applied to control sclerotinia and some other fungal diseases and shorten the next 1-3 internodes. We have tested several PGR options and found that chlormequat was the most economical product.
- c. We can alternate fungicide chemistry as when flowers are wet they can become infected with *Botrytis cinerea* which then causes seed rot. Several fungicides are effective including carbendazim, tebuconazole, prothioconazole, fluazinam, coppers and thiram.
- d. Monitor for aphids notably *Myzus persicae*. Use bee safe, IPM methods for control. If insecticides are needed make applications at night to avoid damage to pollinators.

e. Topping has also been used to manage reproductive growth and increase lateral branching.

4. Flowering. The early studies by Hare, Rolston et al (1987) were published in their landmark paper: Puna Chicory – A Perennial Herb for New Zealand Pastures. They determined that the flowering pattern, and peak seed yields of Puna occurred about 49-60 days after the start of flowering, 16-31 days after peak flowering. This is similar for modern cultivars and production techniques.

The indeterminate nature is a challenge as each individual flower is open for just 4-12 hours and must be pollinated within that time to produce a viable seed. Our cultivars are largely insect pollinated but some genetics may have a small degree of selfing and/or wind pollination. Our NZ productions have managed honeybee hives at 3 hives per hectare and increasing numbers have drone fly breeding nests as developed by BSI-Plant and Food Research (Brad Howlett's group). As a member of the Seed Industry Research Group (SIRC) we are also funding research into alternative native pollinators with University of Canterbury entomology groups.

During flowering the crops also face challenges from fungal diseases, notably *Sclerotinia sclerotiorum* and *Botrytis cinerea*. Standard practice is to mix chlormequat with DMI fungicide at about two- weekly applications for both canopy and disease control. Different DMI fungicides are used, such as carbendazim, tebuconazole, prothioconazole and thiram.

During this time the crop is frequently under pressure from aphids, especially the green peach aphid *Myzus persicae*. Crops must be monitored, and if insecticide is required, use IPM and bee safe products. Common insecticide products in NZ are Mavrik Aquaflo® (240 g L⁻¹ Tau-fluvalinate) and Coragen® (200 g L⁻¹ chlorantraniliprole).

5. Windrow/swathing timing. Early research, published in their landmark paper by Hare, Rolston, Rowarth et al. 1987, established that seed crop cutting time was best at 46 days after the start of flowering. This is still used as a base timing, but decisions are now made on a combination of factors:

- a. Observation of best pollination conditions, record the date(s).
- b. Monitor the crop colour as it changes from dark to mid green, then purple to brown.
- c. Similarly, the seeds can be stripped from pods to seed the colour change from cream to brown.
- d. Current practice is to use Tetrazolium Tests (TZ) during the ripening process to identify how the crop is maturing. See pics. Our experience is that when TZ reaches over 80% the crop can be cut. This is effectively about 3 days after the sample was taken.

Birds (finches and other passerines) are attracted to chicory seed so bird scare methods are often required. Laser lights and bird banger scarers are used where appropriate.

6. Harvesting. Chicory seed has a late summer/early autumn harvest so harvest hours per day are limiting.

The crop can be windrowed, mown or direct harvested depending on the crop architecture such as lodging, and vegetative bulk. Most crops are cut by some means.

Often the crop can be harvested 10-14 days after cutting or by direct harvest, but sometimes in cool or damp conditions it will be >21 days. Conditions on the day of harvest determine the success of the outcome.

Seed is ready dislodged from the pods with a slow drum speed and tight concave. In NZ both conventional and Axial Flow combines are used successfully. Tasmanian experience is that Axial Flow machines are best.

Seed is often dried either in batch dryers or on drying floors. Chicory seed is sensitive to heat damage, care must be taken not to exceed 30°C, especially at high levels of seed moisture content.

References

- Hare MD (1986) Development of Grasslands Puna chicory (*Cichorium intybus* L.) seed and determination of time of harvest for maximum seed yields. *Journal of Applied Seed Production* **4**, 30-33.
- Hare MD, Rolston MP, Crush JR and Fraser TJ (1987) Puna Chicory – A Perennial Herb for New Zealand Pastures. *Proceedings of the New Zealand Agronomy Society* **17**, 45-49.
- Hare MD and Rolston MP (1987) Effect of time of closing, and paclobutrazol (PP333) on seed yield of 'Grasslands Puna' chicory (*Cichorium intybus* L.). *New Zealand Journal of Experimental Agriculture* **15**, 405-410.
- Hare MD, Rowarth JS, Archie WJ, Rolston MP and Guy B (1990) Chicory seed production: Research and practice. *Proceedings of the New Zealand Grasslands Association* **52**, 91-94.
- Hare MD, Rolston MP, Foote AG, Archie WJ and Hagerty G (1993) Herbicide tolerance of Grasslands Puna chicory. *Proceedings of the New Zealand Plant Protection Society* **46**, 282-287.
- Rowarth JS, Hare MD, Rolston MP and Archie WJ (1996) Effects of nitrogen on seed yield of chicory (*Cichorium intybus* L.) cv 'Grasslands Puna'. *Journal of Applied Seed Production* **14**, 73-76.

POSTER PRESENTATIONS

Vegetative growth and seed production of endophyte-free and endophyte-infected tall fescue plants under defoliation conditions

Lucas Petigrosso^{12*}, María Echeverría¹, Jorge Castaño¹ and Osvaldo Vignolio¹

Abstract

Tall fescue, *Festuca arundinacea* (syn., *Schedonorus arundinaceus* (Shreb.) Dumort.), is a cool-season grass, exotic, invasive in the Flooding Pampa grasslands (Argentina). Tall fescue is a perennial C₃ forage and it is commonly infected with the endophytic fungus *Epichloë coenophiala*. The grass-fungus association is symbiotic-mutualistic, with vertical transmission.

The objective of this work was to evaluate the effect of defoliation by clipping, on vegetative and reproductive attributes endophyte-infected (E+) and endophyte-free (E-) of tall fescue plants. The experiment was carried out in a greenhouse. The materials used were a naturalised tall fescue population infected with the endophyte and the same population free of endophyte. On July 31, 2023, the seeds were sown in 3 L plastic pot filled with Argiudol soil, one seed per pot. Twenty-six pots, E+ and E-, were prepared.

On February 29, 2024, tall fescue plants, E+ and E-, were hand-mowed at 6 cm height from the ground level to standardise the height. The biomass was not included in the calculus.

Seven harvests were applied: March 20, April 9, April 25, May 10, July 5, August 20 and September 24. Shoot biomass was dried in ovens and weighed. As the seeds matured, they were harvested. Relative growth rate (RGR) was calculated as $RGR = (\ln w_2 - \ln w_1) / (t_2 - t_1)$, where w_1 and w_2 are the dry shoot biomass per plant at the first harvest (t_1 , March 20) and the accumulated biomass through seven successive harvests (t_2). Seed production was determined. Reproductive effort was calculated as seed weight / total shoot biomass. Seed germination was evaluated by the International Seed Testing Association (ISTA) rules. The endophyte transmission was analysed in seedling obtained from the seed produced by E+ plants.

The effects of defoliation on the variables were analysed using the Student's t-test ($P = 0.05$). E- plants flowered between October 7 and November 4, 2024. E+ plants began flowering between October 14 and November 4. Mature panicles were harvested between December 9 and 30, 2024, for E- plants, and between December 6, 2024, and January 2, 2025, for E+ plants. In both infection levels, the reproductive period was of 55 days.

The E+ and E- plants were not significantly affected by defoliation in germination power, germination energy, thousand seeds weight and reproductive effort. The number of panicles was not significantly different between E- and E+ plants. Endophyte transmission neither was significantly affected by defoliation. In other words, the vertical transmission, from maternal plants to seeds, was not affected. The RGR of E+ was higher than in E-, 0.0548 g/g/week and 0.0430 g/g/week, respectively. Seedling root length was significantly higher in E+ than in E-, 5.82 cm and 5.32 cm, respectively. Our results allow to hypothesise that the invasion of infected tall fescue plant is related to two crucial stages of its life cycle; the higher seedling root length which confers the ability to avoid stress during establishment and the higher RGR of the plant which confers more competitive ability.

Keywords: *Festuca arundinacea*, *Epichloë coenophiala*, seed production, defoliation, germination

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Impact of storage duration on fungal contamination and mycotoxin accumulation in forage oat seeds

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Abstract

Oat (*Avena sativa* L.) is an important annual forage crop in western China, especially on the Qinghai-Tibet Plateau. Oat hay and silage serve as main roughage for livestock during the winter and spring in alpine grassland regions. With the development of animal husbandry and increase of livestock head, the expanding cultivation area has led to a growing demand for oat seeds and increased production. However, oat seeds are rich in fat and protein, which makes them susceptible to fungal contamination and spoilage. As a result, they are not well-suited for long-term storage.

To investigate the effects of storage duration on fungal communities and mycotoxin levels in oat seeds, six varieties stored for 1, 2, 3 and 5 years were analysed, including naked oats ('Baiyan 2', 'Bayou 9' and 'Bayou 14') and hulled oats ('Longyan 2', 'Bayan 4' and 'Baiyan 7'). Fungi were isolated and identified through plate culture, morphological characterisation, and rDNA-ITS sequencing, while mycotoxins (TeA, AFB1, and CIT) produced by dominant fungal genera were quantified using HPLC-MS. The results showed that storage duration significantly influenced fungal load, diversity, and isolation rates. Total fungal counts generally declined over time, with spore concentrations ranging from 0.33 to 29.70 CFU grain⁻¹. Infection rates peaked (1.50-36.75%) after 1-2 years of storage before decreasing. Fungal species composition varied markedly across storage periods and varieties, with 'Baiyan 2' harbouring the lowest fungal load and 'Bayou 14' exhibiting the highest species richness. Forty-five fungal species (20 genera) were identified, dominated by *Alternaria*, *Aspergillus*, and *Penicillium*. Dominant species shifted with storage time and oat variety.

For mycotoxin profiles, varieties, storage duration, and their interaction exerted highly significant effects on TeA, AFB1, and CIT levels ($P < 0.01$), with the interaction showing the strongest impact. TeA was the predominant mycotoxin (16.88-348.67 $\mu\text{g kg}^{-1}$), followed by CIT (0.02-7.06 $\mu\text{g kg}^{-1}$) and trace AFB1 (0.01-1.04 $\mu\text{g kg}^{-1}$). All three toxins peaked at 2-3 years of storage before declining. Naked oats consistently exhibited lower fungal loads, fewer species, and reduced mycotoxin accumulation compared to hulled oats. Hulled oats averaged 3.69 CFU grain⁻¹ (surface spores) and an 8.83% internal infection rate, whereas naked oats averaged 2.69 CFU grain⁻¹ and 9.67% infection. Notably, 37 species (17 genera) were detected in hulled oats versus 33 species (17 genera) in naked oats, with 12 and 8 species being exclusive to each type, respectively. Mycotoxin averages were higher in hulled oats (TeA: 140.70 $\mu\text{g kg}^{-1}$; AFB1: 0.18 $\mu\text{g kg}^{-1}$; CIT: 2.74 $\mu\text{g kg}^{-1}$) than in naked oats (TeA: 123.85 $\mu\text{g kg}^{-1}$; AFB1: 0.23 $\mu\text{g kg}^{-1}$; CIT: 0.84 $\mu\text{g kg}^{-1}$).

The findings of this study provide a scientific basis for optimising oat seed storage strategies. Given that mycotoxin levels peak between 2-3 years of storage, it is recommended that under practical production conditions, oat seeds should not be stored beyond this critical window in order to minimise contamination risks. Enhanced drying, sealing, and regular monitoring of storage conditions are essential to control fungal growth and mycotoxin production.

Keywords: forage oat seeds; storage duration; seed borne fungi; mycotoxins

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Modelling the development stages of alfalfa seed production to anticipate the impacts of climate change

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Abstract

Climate change is a central concern for every agricultural sector, and forage seed production is no exception. In 2023, the FNAMS started a project to assess and anticipate its impacts on alfalfa (*Medicago sativa* L.) and its cultivation. The first step of this work was to model the developmental stages of alfalfa for seed production.

To determine the phenology of a crop, we must first characterise how temperature affects its development. For this, an empirical measure is used: the growing degree days (GDD). The accumulation of these degree-days allows for the estimation of the appearance dates of distinct stages: budding, flowering, green pods, brown pods, and ripe seed pods.

Based on an extensive database, this work allowed us first to identify alfalfa's threshold base temperature. The plant exhibits different temperature responses depending on its organs, particularly a developmental duality between germination organs (radicles and coleoptiles) and stems/internodes. The base temperature for germination organs is around 0°C, while for stems and internodes, it is around 9°C. The maximum growth temperature is estimated at around 30°C. Using a degree-day accumulation between 9°C and 30°C, we were able to determine key developmental stages (in growing degree days). Given that alfalfa is a perennial crop and that cutting in spring is a common practice in France for seed production, two heat accumulation scenarios were identified: one starting from January 1st of the production year (no cutting), the other starting after the cutting, during the same production year.

Knowing the stages is essential for simulating climate effects on crops. Within a pheno-climatic approach, modelling these stages allows for the calculation of stages' dates for each year and location, providing a more detailed analysis of the risks during each sensitive phase of the plant's development. Moreover, it becomes possible to observe how practices like cutting, which shift the stages to later, affect plant development, enabling evaluation of the duration of the shift, along with associated risks and opportunities. For instance, in the south-west, cutting on the 1st of May means the budding of alfalfa starts 35 days later (around the 4th of June), when a cutting the 20th of May induce the budding 26 days after.

Based on these findings, the FNAMS continues to study the effects of climate change and the influence of cutting dates and other technics on seed alfalfa production in several key regions in France.

Keywords: alfalfa, seed production, development stages, modelling, climate

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Increasing seed yield potential of Tasmanian white clover seed production crops

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Abstract

White clover (*Trifolium repens* L.) is an important forage crop in Tasmania, Australia, and is the second most dominant pasture species grown in the state. There has been an increasing demand from seed production companies to expand the area and volume of certified white clover seed grown in Tasmania. However, growers and seed production companies have noted that seed yields have not experienced any significant increase in the past few decades - compared to average ryegrass seed yield increases of over 50% in the same period - mainly through the evaluation and adoption of agronomic and crop production improvements.

While white clover has been grown in Tasmania as a commercial seed production crop for over sixty years, a majority of growers perceive low seed prices and high variability of seed yields between seasons as barriers for growing white clover specifically for certified seed production, resulting in white clover primarily being grown a forage crop for livestock production or as a dual-purpose forage/seed crop. To maximise dry matter production over winter, 'standard' row spacings of 10-15 cm are used for Tasmanian white clover seed crops, which is narrower than the row spacings conventionally used in other major seed production countries.

As part of *PRO-016025 Increasing yield potential of Tasmanian white clover seed production crops*, funded by the AgriFutures Australia Pasture Seeds Program, four small-plot replicated field trials were conducted across two seasons in the Tasmanian northern midlands to compare crop growth, dry matter (DM) production over winter and seed yield in the Tasmanian environment at the 'standard' 10-15 cm spacings, and at wider spacings of 30 cm, 40-45 cm and 60 cm. Three trials were established within commercial crops sown in autumn with 15 cm row spacings, and a fourth with 10 cm rows. Once emerged, rows were removed as needed to create the desired row spacings in a Latin square design. Other than row spacings, all plots were maintained by the trial cooperator as per the surrounding commercial crop. DM cuts were made just before grazing at one site each season. The methodology used to establish the row spacings halved the effective sowing rate and plant density as row spacings doubled.

As expected, preliminary results suggest that early crop growth was significantly ($P < 0.05$) reduced at wider row spacings, and had substantially less DM available over winter than the 'standard' row spacings. However, DM production contributed less than 10% to the overall profitability of the crop, and the reduction in DM production was partially offset by the reduced cost of sowing basic seed at wider row spacings. After paddocks were closed, row spacings did not affect the time it took the crop to reach full ground cover.

Under the trial conditions, row spacings 30 cm and 45 cm appeared to have higher pollination rates, higher seed yields and were more profitable than 'standard' 15 cm row spacings. The final analysis is currently being conducted.

Keywords: white clover, row spacing, seed yield, dry matter, Tasmania

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Advancing proprietary seed production through independent forage evaluation in Australia

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Abstract

The requirements for proprietary seed production are closely linked to the commercial landscape of forage seed sales, with Australia's diverse production environments creating both opportunity and complexity. This variability poses challenges for breeders and end-users, resulting in a wide range of forage solutions and making varietal selection difficult for farmers, consultants, and agronomists. Historically, the merits and performance of varieties in specific regions were obscured by inconsistent demonstration trials and marketing, leading to confusion and a lack of confidence in adopting improved genetics. To address this, Meat & Livestock Australia (MLA) implemented a nationally coordinated program to provide independent, standardised, and scientifically rigorous evaluation of forage varieties across temperate regions. The outcome is a collaborative seed industry effort that established the Pasture Trials Network (PTN), a national temperate program dedicated to standardised, independent evaluation and a scientifically rigorous, multi-site trial program following a MERIT (Monitoring, Evaluation, Report, Improvement Tool) process, assessing forage yield, quality, and persistence. These trials produce high-quality, robust agronomic and peer-reviewed outputs. The dual purpose of this initiative is to support cost-effective breeding and evaluation programs for seed companies and provide reliable, independent, and trusted data for informed sowing decisions by farmers, consultants, and agronomists. Unique components of the PTN program focus on four main elements that all trials must follow: Established protocol, with all trials complying with industry-accepted protocols; an annual audit program for compliance with the program's species protocol, agronomy and scientific processes; an independent review panel consisting of pasture and plant breeding specialists from across the industry, concentrating on agronomy and processes; independent analysis of data by a specialist MERIT statistician for generating outputs for communication.

Initially launching with perennial ryegrass (*Lolium perenne* L.), phalaris (*Phalaris aquatica*), lucerne (*Medicago sativa* L.), as well as subspecies of cocksfoot (*Dactylis glomerata*), tall fescue (*Festuca arundinacea*, syn., *Schedonorus arundinaceus* (Shreb.) Dumort.) and sub clover (*Trifolium subterraneum* L.), the program has increased over time to include short-term ryegrasses (*Lolium multiflorum* and *Lolium westerwoldicum*), white clover (*Trifolium repens* L.) and, more recently, forage oats (*Avena sativa* L.), and chicory (*Cichorium intybus*). Program outputs are communicated through the MLA PTN eTool or the Dairy Australia Forage Value Index (FVI), freely accessible online tools hosted by the industry. Data is presented across five seasonal points, with spring divided into early and late phases for greater detail. The two communication tools differ: the MLA PTN eTool is a multi-species platform focusing on individual trials, while the FVI provides ryegrass data along with a value for varieties within a macro-region. The MLA PTN eTool is updated regularly following trial completions and the commercial release of new varieties. This comprehensive and transparent data resource enables producers, advisors, and seed companies to make informed varietal choices for a specific region. Currently, trials are communicated through the MLA PTN eTool in a discrete manner, with ongoing efforts to build confidence in variety selection for seed purchase at sowing time. Using Genotype by Environment (GxE) analysis, combined with the PTN's near 15 years of data, potential relationships are being explored to help end users better understand how a variety performs under different environmental and management conditions, such as rainfall, soil type, and nutrient inputs, among others.

This ongoing project will continue with the PTN's aim of making quality data freely accessible to end-users. Beyond data delivery, the PTN contributes to industry capacity-building through training, collaboration with pasture-focused organisations across Australia, and other regional pasture

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initiatives. These outcomes collectively strengthen confidence, transparency, and sustainability in Australia's proprietary seed industry.

Keywords: MERIT testing, herbage performance, data communication, industry program

Development of a tool for automatically counting insect pests in alfalfa seed crops

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Abstract

Seed-bearing alfalfa (*Medicago sativa* L.) has to contend with infestations of insect pests such as leaf weevils, seed weevils and mirid bugs. Detecting these insects in the field is important for adapting and reasoning out cultural practices. At present, the monitoring of these insects is based on visual and manual counting with a sweep net, a laborious and time-consuming method that is prone to human error.

To overcome these constraints, the FNAMS and the University of Angers are working together since 2023 to develop a technique for automatically counting insect pests of seed-bearing alfalfa. This work is based on imaging coupled with artificial intelligence (Deep Learning). The approach involves several successive steps: 1) collecting data in the field, 2) annotating sample images, 3) developing the Deep Learning model, 4) assessing the predictive quality and 5) deploying the finished tool.

The process began with a phase of data collection in the field. Sampling was carried out in alfalfa fields using a sweep net. Each net sample was then transferred to a yellow basin, in which the insects caught were photographed and then counted. These images, taken under real field conditions, form the basis of a representative dataset (412 images from 2023 to 2025), which is then subjected to rigorous manual annotation to identify and locate the various pest species. These annotations were then used to train a deep learning model based on a convolutional neural network (CNN), designed to automatically detect, classify and count insects. Data augmentation techniques were applied to increase its robustness to variations in lighting, position and image quality. The model was then evaluated using an independent set of images to validate its performance. The best model showed an F1-score of 0.63 ± 0.09 , which shows interesting predictive potential, although the accuracy of the tool should be improved in the future. Finally, a mobile interface is to be developed to enable the solution to be deployed operationally, offering users the possibility of uploading images and obtaining an instant automated count, thus facilitating its integration into agricultural practices.

The results expected from this project highlight the potential of artificial intelligence to enhance pest surveillance and improve decision-making in agriculture. Automating the counting of pests not only saves time, but also limits the unwise use of pesticides through more targeted intervention. This initiative illustrates the value of collaboration between research institutes and agricultural operators in developing innovative tools suited to the current challenges facing agriculture. The FNAMS and the University of Angers plan to extend this approach to other crops and types of pests, in order to make widespread use of artificial intelligence in plant protection.

Keywords: insect, pest, Deep Learning, AI, alfalfa

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Foxtail barley resistance to quizalofop-p-ethyl in Peace Region creeping red fescue seed fields

Calvin Yoder^{1*}, Charles Geddes² and Doug Fehr³

Abstract

The active ingredient quizalofop-p-ethyl has been used for many years on seedling and established creeping red fescue (*Festuca rubra* L. spp. *Rubra*) seed fields to suppress or control grassy weeds such as wild oat, quack grass and foxtail barley (*Hordeum jubatum*). There are several products for quizalofop-p-ethyl including products like Assure[®] II, Contender[®], Elegant and Yuma[®] as examples. The continual use of quizalofop-p-ethyl or other Group 1 grassy weed herbicides has led to many fields across Western Canada having high populations of wild oats resistant to herbicides with this mode of action. Unfortunately, a number of fields with a history of long-term creeping red fescue seed crops have now been confirmed of having foxtail barley plants resistant to quizalofop-p-ethyl.

In 2022, foxtail barley seed heads were collected from three creeping red fescue seed fields with high populations of plants that appeared to be unaffected by an application of quizalofop-p-ethyl. Heads were thrashed and seeds processed to remove awns. Seed samples were sent to the AAFC Herbicide Resistant Lab in Lethbridge to determine if these populations of foxtail barley were indeed showing some resistance to quizalofop-p-ethyl. Seeds were planted and sprayed with varying rates of quizalofop-p-ethyl.

The dose-response experiment showed that two of the populations exhibited between 15 and 29-fold resistance to quizalofop (PR-1 and PR-2), while one of the populations was susceptible (PR-3). S-1 and S-2 are susceptible controls.

Collecting and processing foxtail barley seed is a terrible process. Fortunately, the AAFC Herbicide Resistance Lab in Lethbridge developed a molecular testing kit where foxtail barley leaves can be collected and submitted to the lab to test for resistance to quizalofop-p-ethyl.

Foxtail barley plants were collected from ten creeping red fescue fields in 2024 where plants were not controlled by an earlier application of quizalofop-p-ethyl. Two leaves from ten foxtail barley plants from each field were submitted for molecular testing to identify possible resistance to quizalofop-p-ethyl. Unfortunately, seven of the ten fields had foxtail barley plants showing some resistance to quizalofop-p-ethyl with five fields showing high levels of resistance.

The long-term use of quizalofop-p-ethyl in creeping red fescue seed fields is leading to grassy weed resistance in both wild oat and foxtail barley. To date there are no other herbicide options for managing foxtail barley in creeping red fescue. The active ingredient clethodim (e.g. trade name Centurion[®], Select[®], Arrow[®]) shows fair activity on foxtail barley plants but previous tolerance trials conducted in Alberta have shown clethodim to cause damage to both seedling and established creeping red fescue seed crops.

Creeping red fescue seed growers will require a longer period of time between fescue crops to clean up foxtail barley as much as possible prior to seeding fescue. Establishing strong competitive fescue stands will also be required to manage this weed.

Keywords: foxtail barley, creeping red fescue, quizalofop-p-ethyl, molecular test, Peace Region

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Can aerial imagery be used to quantify yield losses in vole infested tall fescue fields?

Christy Tanner^{1*}

Abstract

The gray-tailed vole (*Microtus canicaudus*), a key pest of grass seed production in the Willamette Valley of Oregon, USA, commonly causes yield losses of more than 200 kg ha⁻¹ when vole populations are high. Voles primarily feed in the areas immediately surrounding their burrows, resulting in a spatially variable pattern of crop damage. Previous research showed that aerial imagery data (Normalised Differential Vegetation Index (NDVI) and crop canopy height) could be used to distinguish areas of vole damage from undamaged areas in tall fescue (*Lolium arundinaceum*, syn., *Schedonorus arundinaceus* (Shreb.) Dumort.) seed production fields. This study investigated whether aerial imagery could be used to estimate seed yield loss in vole infested tall fescue seed production fields in 2023 (one field) and 2024 (three fields). Aerial imagery was collected at each field on three to five dates beginning in the spring (March or April), with the final flight occurring immediately before harvest. Seed yield was measured in small plots measuring one crop row in width by 50 cm in length. In 2023, 32 yield samples were collected from pairs of plots with one plot in an area with obvious vole damage, and the other collected in an area that appeared undamaged. In 2024, 50 yield plots were collected in each field. The location of 70% of plots was determined by walking along a transect and placing a plot every 10 paces, while the remaining plots were selected to ensure that the dataset included a range of vole damage levels.

The relationships between seed yield and aerial imagery-based measurements differed between fields and flight dates. In 2023, yield was positively correlated with NDVI measured in March through early May ($r = 0.85$ to 0.9 , $P < 0.05$), with correlation coefficients decreasing for flights in late spring ($r = 0.74$, $P < 0.05$) and at harvest ($r = 0.56$, $P < 0.05$). In 2024, there was a weaker relationship between NDVI and yield in March and April ($r = 0.56$ to 0.32 , $P < 0.05$). Yield was not significantly correlated with NDVI in mid-June for the 2024 fields, and two of the three fields showed negative correlations between seed yield and NDVI at harvest ($r = -0.34$ to -0.38 , $P < 0.05$). Crop canopy height had a more consistent relationship with yield, with statistically significant positive correlations for all fields and flight dates. The transect sampling method used in 2024 produced a dataset with a less uniform distribution of yield values, which may have led to decreased statistical power. However, differences in field characteristics such as stand age (the 2023 field was a first-year field, while the 2024 fields were second-year or older) may have impacted the findings. These results demonstrate that the relationships between aerial imagery measurements and seed yield can vary among fields and over the course of the growing season.

Keywords: drone, vole, tall fescue, seed yield, NDVI

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Classification of grass seed crop species using spaceborne imagery and Artificial Intelligence-driven models

Jing Zhou^{1*}, Qianyi Duan¹ and Nicole Anderson²

Abstract

Accurate classification of grass seed crop species is essential for estimating seasonal field acreages, informing market strategies, promoting crop diversification, and establishing long-term cropping histories. Unlike major commodity crops, grass seed crops lack reliable datasets and mapping products. This study investigates the use of spaceborne imagery and artificial intelligence (AI)-driven computer vision to remotely classify grass seed crops.

Our ground observation dataset comprises 15 grass seed species grown in Oregon, USA (2021-2023), covering over 4,000 data points. Satellite imagery was acquired from Sentinel-2 (S2) spanning January 1st to June 14th each study year. The imagery includes 12 bands across 400-2190 nm with a spatial resolution of 10 m pixel⁻¹, collected at five-day intervals, totalling 34 time stamps.

Statistical analyses identified the second and third weeks of May as the most critical temporal window for spectrally distinguishing among grass species using satellite imagery, coinciding with field inspection timing for crop purity. The near-infrared [835.1 nm (S2A) / 833 nm (S2B)], red edge [740.2 nm (S2A) / 739.1 nm (S2B)], and narrow near-infrared [864.8 nm (S2A) / 864 nm (S2B)] bands showed the highest spectral separability among major grass species.

A U-Net Temporal Attention Encoder (U-TAE) model was trained to classify grass seed crop species, integrating temporal and spectral data. The overall classification accuracy - defined as the ratio of correctly classified samples to total samples - was 0.89 across all 15 grass species with high accuracies for four major species, including tall fescue (0.93) (*Schedonorus arundinaceus* (Shreb.) Dumort.), perennial ryegrass (0.90) (*Lolium perenne* L.), annual ryegrass (0.87) (*Lolium perenne* L. ssp. *Multiflorum* (Lam.) Husnot), and Kentucky bluegrass (0.83) (*Poa pratensis* L.) (0.83).

Our findings provide actionable insights for industry stakeholders, enabling informed pricing, planting strategies, and reduced risk of cross-pollination. This work highlights the potential of AI and remote sensing in grass seed crop production, with future efforts focused at estimating field acreage and predicting production potential.

Keywords: grass seed crops, remote sensing, crop mapping, artificial intelligence

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Increasing ryegrass seed yields and returns for growers using biostimulants

Julie Finnigan^{1*} and Matthew Kleinig¹

Abstract

Increases in yield were achieved in ryegrass seed production over two consecutive seasons at properties in Tasmania's north. YieldOn, a biostimulant produced by Valagro®, was trialled to determine potential increases in seed yield following beneficial yield increases in other seed crops around Australia.

YieldOn is a natural biostimulant that optimises plant hormones and enhances the process of cytokinesis, ultimately improving the productivity of row crops. International research has shown improvements to nutrient uptake and efficiency, increases to phloem capacity and transport of sugars from leaves to the rest of the plant, increases in fatty acid biosynthesis and transport within seed crops, and importantly optimisation of cell division and expansion. Collectively this can improve seed vigour and development, leading to increases in seed weight and quality.

YieldOn was commercially applied to large demonstration areas of ryegrass seed crops at a rate of 2 L ha⁻¹, at growth stages 33 and 39 to the Italian diploid (*Lolium multiflorum* L.) 'Tempo' (5 ha), and annual tetraploid (*Lolium multiflorum* L.) 'Coaster' (2.9 ha). Being highly compatible with most products, YieldOn was combined with fungicides, trace elements, plant growth regulators (PGRs) and adjuvants where required, needing no additional passes in crop. While replicated plot sampling was taken, harvest yield results discussed were gained from both sites using commercial yield monitor data. No statistical analysis was conducted.

Data from these trials provided positive results in seed yield increases for ryegrass treated with YieldOn. YieldOn applied to Italian ryegrass 'Tempo' increased commercial seed yield by 100 kg ha⁻¹, while YieldOn applied to annual ryegrass 'Coaster' increased commercial seed yield by 200 kg ha⁻¹. These increases in commercial seed yields provided returns on investment of \$150 ha⁻¹ and \$290 ha⁻¹, with calculations based on pricing and costs at the time.

These trials using different seed cultivars, conducted by different growers, in different regions and across different seasons, have shown that positive increases in ryegrass seed yield can confidently be gained from the use of YieldOn. The use of biostimulants not only provides a sustainable and environmentally beneficial means of improving seed yield but is also very user-friendly requiring no extra passes to apply. These benefits combined with easily achievable returns on investment, makes YieldOn an excellent biostimulant for use in ryegrass seed production.

Keywords: yield increase, sustainable, ryegrass seed, biostimulants, positive ROI

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Overseeding a forage crop in established alfalfa: which impacts on weeds and seed production?

Serge Bouet^{1} and Mael Quemar¹*

Abstract

Alfalfa (*Medicago sativa* L.) is a sensitive crop to weed soiling during the winter. This study (research project SURSEME) investigates the impact to overseeding in an established alfalfa seed crop, a mix of cereals and protein crops in the autumn or a single forage rye crop:

- on weed development during the winter,
- on the alfalfa forage production and value, and
- on seed production.

In 2022, 2023 and 2024, three trials were carried out in west of France (Angers, FNAMS experimental station, on established alfalfa seed production with large inter-row (50 cm). These seed production trials were conducted with 1 forage harvest at the spring (cutting) followed by the seed harvest at the end of summer.

These trials compared in a 4 replicates experimental design:

- the effect of overseeding different forage mix of cereal and protein crops and a fertilised forage rye crop for methanization outlet, and
- two different dates of forage harvest for the mix: an early harvesting strategy based on alfalfa vegetative stage, and a classic harvesting strategy based on a late alfalfa bud stage (20 days later).

The results of this study make it possible to clarify the interest of overseeding in established alfalfa seed production:

- Forage mix allowed to produce between 2 tons/ha of dry matter per hectare more in early harvest strategy vs without overseeding; 2.8 of dry matter per hectare more in classic harvest and 4.5 more with fertilised rye crop for methanization.
- Forage value is improved with an early harvest of forage mix vs classic harvest.
- Overseeding allows faster soil coverage, which gives variable results on weed control:
 - o With an early forage mix harvest, weed is reduced in the following month.
 - o With a classic date of forage mix harvest, the impact to overseeding became minor, erased by the cleaning effect of the cutting compared to the control without overseeding. Similar result was obtained with the forage rye crop harvested at the same date.
- Overseeding has no significant impact on alfalfa seed yield. Even with a very competitive mix or forage rye crop.

To conclude, this practice seems to be promising to enhance forage production or production for methanizer outlet without impact on alfalfa seed yield. With an early harvest of forage mix, forage value is increased and a positive impact on weed was registered.

Keywords: alfalfa, overseeding, weeding, seed production

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Genome-wide association analysis of drought tolerance traits in alfalfa and functional validation of the candidate gene

Tianning Fang¹, Nan Song¹, Bao Ao¹ and Jiyu Zhang^{1*}

Abstract

As a globally important leguminous forage, alfalfa (*Medicago sativa* L.) plays a crucial role in supporting sustainable livestock production and enhancing agricultural resilience to climate change, making research on its drought resistance highly significant. In this study, drought-tolerant germplasm resources of alfalfa were used as materials, and the role of the candidate gene *MsASE1* in drought resistance was systematically investigated through Genome-Wide Association Studies (GWAS) combined with functional verification.

Drought tolerance evaluation was conducted on 199 *M. sativa* accessions. Based on cluster analysis of morphological and physiological indices, the materials were divided into 5 groups, and drought-tolerant plants were screened out. Through GWAS, the gene *MsASE1* significantly associated with the drought tolerance index was identified. Exon SNP haplotype analysis showed that the drought tolerance index of haplotype G (1.07) was significantly higher than that of the G:T type (0.66). Quantitative real-time PCR (QRT-PCR) results revealed that under mannitol-simulated drought stress, the expression level of *MsASE1* was significantly upregulated at 6 hours, peaked at 12 hours, and then decreased, presenting a time-dependent response. The expression level of *MsASE1* in drought-tolerant plants was significantly higher than that in drought-sensitive plants. Moreover, heterologous expression of *MsASE1* in yeast enhanced osmotic stress tolerance, suggesting a conserved role in stress tolerance.

In conclusion, this study is the first to identify and verify the role of the *MsASE1* gene in the drought resistance of *M. sativa*. It enriches the drought-tolerant gene pool of alfalfa and provides more options for drought-resistant breeding.

Keywords: alfalfa, drought stress, functional analysis

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