Germination and development differences between summer and winter annual ryegrass (*Lolium rigidum*) populations

Michael Thompson[1] and **Bhagirath S. Chauhan[2]**

[1] Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, Gatton, 4343, QLD, Email: b.chauhan@uq.edu.au

[2] School of Agriculture and Food Sciences (SAFS) and Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, Gatton, 4343, QLD

Abstract
Annual ryegrass (*Lolium rigidum*) is a highly competitive and costly winter annual weed species that in recent years has been observed growing in fields during summer, threatening greater costs if this occurrence becomes more widespread. In this research, seeds of *L. rigidum* were collected from populations that grew during winter and summer, which were then germinated to identify temperature requirements for germination and duration of dormancy, as well as pot experiments to analyse differences in growth, development and production. Winter and summer populations were capable of germinating over a range of temperatures with each population differing in the level of dormancy. All populations took longer to develop and reach maturity when germinated in summer compared with late autumn. These results demonstrate the capability of *L. rigidum* to germinate and grow throughout summer, which suggests there is potential for this weed to become a greater issue throughout the year.

Keywords
Temperature, dormancy, weed development, weeds.

Introduction
Annual ryegrass (*Lolium rigidum* Gaud.) is one of the most economically damaging weeds in southern Australian winter cropping systems, resulting in substantial yield losses up to $93 million per annum (Llewellyn et al. 2016). Various methods are used for its control ;however, herbicide application is the primary control option. As such, annual ryegrass is the most costly weed in Australia in terms of herbicides. It typically grows during winter and matures in spring; however, in recent years this weed has been observed growing throughout the summer months. This poses a serious threat to the Australian agricultural industry if annual ryegrass becomes more common in summer.

Germination is an important step required for the establishment of annual ryegrass, with various factors determining the successful germination and subsequent survival of the plant. Germination begins with the imbibition of water and can be influenced by the temperature of the environment (Woodstock, 1988). Water availability will therefore be a determining factor in the successful establishment of annual ryegrass in summer cropping systems. Previous studies have demonstrated the capabilities of annual ryegrass to germinate over a range of temperatures (Gramshaw 1976, Chauhan et al. 2006; Rahman and Asaduzzaman 2019). An ability to germinate at higher temperatures may allow non-dormant seeds to germinate in high summer temperatures. Whether differences exist between the optimum temperature for germination of summer and winter annual ryegrass is unknown.

Annual ryegrass maintains a period of dormancy after maturation that is lost over the summer months (Gramshaw 1976). This allows the seeds to survive the summer months when rainfall is sparse and temperatures are high, and germinate in autumn. However, the behaviour of the weed is now changing and it is adapting to a wider range of seasons. There is a current lack of knowledge on the level of dormancy of summer growing annual ryegrass populations.

Due to the damaging economic effect of this weed, it is important to understand why populations are now growing during both summer and winter seasons. This study aimed to identify and compare the optimum germination temperatures of winter and summer annual ryegrass, and to determine whether any difference in dormancy duration exists between these two types of annual ryegrass. Additionally, this study investigated the growth and development of winter and summer collected populations when grown during both winter and summer.

Methods

Plant material

Populations of annual ryegrass were collected from various locations throughout New South Wales (NSW), Australia. For all germination temperature and dormancy experiments, 20 winter populations (8 collected in 2018 and 10 collected in 2019, with two additional populations sourced from earlier years and grown for seed production in 2018) and eight summer populations were used (collected in 2019 and grown for seed production in 2020). A common garden experiment was conducted in the winter season of 2019 and the summer season of 2020 using 10 winter populations and either 7 or 8 summer populations for the 2019 and 2020 experiments, respectively.

Optimum germination temperature and dormancy of winter and summer annual ryegrass

To determine the optimum germination temperature for annual ryegrass seeds at seed collection and to compare the dormancy duration of winter and summer annual ryegrass populations, germination experiments were established in Petri dishes inside incubators. Seeds were placed on filter paper in Petri dishes and moistened with 5 ml water. Twenty five seeds were placed in each Petri dish, with three Petri dishes per population. All Petri dishes were placed in sealed zip-lock bags and then incubated under a range of day/night temperatures. All winter populations were incubated at five temperature regimes (15/5 °C, 20/10 °C, 25/15 °C, 30/20 °C and 35/25 °C), while all summer populations were incubated at three temperature regimes (15/5 °C, 25/15 °C and 35/25 °C) due to insufficient seeds. Germination was counted every 7 days for 28 days. To test dormancy, the same experimental setup was repeated for a single temperature (the temperature with the highest average germination across populations) every 45 days until dormancy was lost. Dormancy was considered lost once ≥ 85% germination was achieved.

Development and production of annual ryegrass in a common garden experiment

A common garden experiment was established in May 2019 (winter season) and January 2020 (summer season) to analyse the development and production of winter compared with summer populations. Seeds of 10 winter populations and 7 summer populations were sown in pots filled with potting mix in winter 2019, with the same populations used for the summer 2020 experiment plus one additional summer population. Days until emergence, fourth leaf stage, anthesis and maturity were recorded. At maturity, height and tiller number were recorded and seeds were separated from panicles. Total shoot and seed biomass were recorded alongside total seed number. Dormancy was then tested using the same protocol as mentioned previously.

Results and Discussion

Optimum germination temperature and dormancy of winter and summer annual ryegrass

Germination of winter and summer annual ryegrass was tested shortly after seed collection to identify the optimum germination temperature and duration of dormancy. For both sets of winter collected populations (collected in 2018 and 2019) and summer collected populations, the 25/15 °C temperature regime allowed the highest germination percentages when averaged across populations regardless of the season or year of collection (Table 1). Winter and summer populations were both able to germinate across the other temperature regimes tested, although the amount of germination was lower than at 25/15 °C. A previous study by Gramshaw (1976) observed that optimal germination of *L. rigidum* occurred at day temperatures between 24 and 29 °C, and night temperatures between 9 and 14 °C.

Duration of dormancy varied greatly within the winter populations. The average dormancy of winter populations was 120 and 100 days for the 2018 and 2019 collected populations (Table 2), respectively, although most populations lost dormancy before these averages. One winter population lost dormancy after 28 days, while two populations took as long as 270 days. Experiments for the summer populations are still underway, with some populations taking 90 days to break dormancy and others taking longer than 90 days. The seeds in this experiment were after-ripened in dry conditions at a steady temperature of approximately 25 °C. The levels of dormancy may change when stored in other conditions, as factors such as soil type, temperature and moisture can affect dormancy status (Narwal et al. 2008; Steadman et al. 2003). This experiment demonstrates the variation in dormancy across populations and that even some winter populations may lose dormancy in time to germinate during summer.

Table 1. Germination percent under five alternating day/night temperature regimes and the average duration of dormancy (days) of winter and summer collected annual ryegrass populations

Season	Germination (%)					Duration of dormancy (days)
	15°C/5°C	20°C/10°C	25°C/15°C	30°C/20°C	35°C/25°C	
Winter (2018)	34±4	42±4	59±4	52±4	18±4	120±30
Winter (2019)	17±2	21±2	25±2	16±2	4±1	100±11
Summer	14±1	-	20±2	-	19±4	> 90

Development and production of annual ryegrass in a common garden experiment

When grown throughout the winter season, winter and summer populations of annual ryegrass emerged at similar rates, while winter populations were quicker to reach the fourth leaf stage, anthesis and maturity (Table 2). When grown throughout the summer season, winter populations were quicker to reach all stages. Annual ryegrass emerged and reached the fourth leaf stage quicker during summer, but took around 100 days longer to reach anthesis and maturity.

Table 2. Number of days taken to reach four developmental stages for winter and summer annual ryegrass populations grown in a common garden experiment during winter 2019 and summer 2020

Seasonality	Days			
	Emergence	Fourth Leaf	Anthesis	Maturity
2019 (Winter)				
Winter	8.2±0.1	22.1±0.3	144.4±1.5	180.9±1.1
Summer	8.0±0.2	26.0±0.9	148.6±1.9	187.5±1.0
2020 (Summer)				
Winter	3.5±0.1	15.1±0.2	229.4±8.4	285.6±6.2
Summer	4.4±0.2	18.3±0.7	247.8±5.8	293.8±2.6

Winter populations grew taller on average than summer populations in the winter season and produced more seeds and biomass (both seed and shoot biomass) (Table 3). When grown in summer, plants grew shorter than in winter and produced lower numbers of seeds. Tiller number was similar for all plants except the summer populations, which were similar in winter, but less in summer. The number of seeds produced per tiller was lower when plants were grown in summer. The longer duration of growth seen with plants emerging in summer means that management strategies will need to adapt. Timing of herbicide application should be considered as seedlings will emerge over a wider timespan. Summer emerging plants reached anthesis around the same time of the year as winter emerging plants, which may suggest late-season spraying as an effective option to reduce the number of seeds entering the seedbank (Steadman et al. 2006) as summer and winter emerging plants can be sprayed at the same time.

Table 3. Production parameters per plant of winter and summer collected annual ryegrass grown in a common garden experiment during winter 2019 and summer 2020

Seasonality	Height (mm)	Tiller number	Total shoot biomass (g)	Total seed biomass (g)	Total seed number
2019 (Winter)					
Winter	750.0±12.4	64.1±2.9	37.6±1.4	16.3±1.0	7033.4±486.5
Summer	649.0±16.2	63.3±3.7	29.4±1.8	10.2±0.8	6573.0±453.2
2020 (Summer)					
Winter	569.9±13.3	65.4±5.1	52.7±2.3	5.2±0.5	3017.1±260.2
Summer	571.7±11.6	46.2±3.3	31.3±1.6	3.1±0.2	1984.0±147.4

Conclusions

Annual ryegrass possesses the ability to germinate under a range of temperatures shortly after seed collection, however, dormancy remains high in many populations and the optimum germination temperature is 25/15 °C. Duration of dormancy varies greatly among winter populations, with some populations rapidly losing dormancy and others retaining long-term dormancy. Summer populations are still dormant shortly after seed collection, which suggests that decreased dormancy was not the primary ability that allowed these populations to establish in the summer season. However, dormancy testing of summer seeds occurred on seeds produced from a seed production experiment which was performed in different conditions to where the populations were collected. Therefore, the maternal environment may have played a role in the dormancy of the seeds. When both winter and summer populations are grown in summer, they take longer to reach anthesis and maturity than when grown in winter and produce shorter plants with fewer seeds. Therefore, when annual ryegrass germinates in summer, they may not produce seeds until the end of the winter cropping season.

References

Chauhan, BS, Gill, G, Preston, C (2006) Influence of environmental factors on seed germination and seedling emergence of rigid ryegrass (*Lolium rigidum*). Weed Science 54:1004-1012

Gramshaw, D. 1976. Temperature/light interactions and the effect of seed source on annual ryegrass (*Lolium rigidum* Gaud.) seeds. Australian Journal of Agricultural Research 27:779-786

Llewellyn RS, Ronning D, Ouzman J, Walker SR, Mayfield A, Clarke M (2016) Impact of weeds on Australian grain production: the cost of weeds to Australian grain growers and the adoption of weed management and tillage practices. Report for GRDC. GRDC/CSIRO, Canberra, ACT

Narwal S, Sindel BM, Jessop RS (2008) Dormancy and longevity of annual ryegrass (Lolium rigidum) as affected by soil type, depth, rainfall, and duration of burial. Plant and Soil 310:225

Rahman, A, Asaduzzaman, M (2019) Statistical modelling of seed germination and seedlings root response of annual ryegrass (*Lolium rigidum*) to different stress. Agricultural Research 8:262-269

Steadman KJ, Crawford AD, Gallagher RS (2003) Dormancy release in Lolium rigidum seeds is a function of thermal after-ripening time and seed water content. Functional Plant Biology 30:345-352

Steadman KJ, Eaton DM, Plummer JA, Ferris DG, Powles SB (2006) Late-season non-selective herbicide application reduces Lolium rigidum seed numbers, seed viability, and seedling fitness. Australian Journal of Agricultural Research 57:133-141

Woodstock, l. W. 1988. Seed imbibition: a critical period for successful germination. Journal of Seed Technology 12, 1-15