

Australian Government

Department of Health and Aged Care Office of the Gene Technology Regulator

The Biology of *Lolium multiflorum* Lam. (Italian ryegrass), *Lolium perenne* L. (perennial ryegrass) and *Lolium arundinaceum* (Schreb.) Darbysh (tall fescue)



Lolium arundinaceum Schreb. (tall fescue). (Figure from Burnett (2006) Grasses for dryland dairying. Tall fescue: Species and Cultivars. Department of Primary Industries, Victoria #AG1241). © State of Victoria, Department of Primary Industries 2006

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This document provides an overview of baseline biological information relevant to risk analysis of genetically modified forms of the species that may be released into the Australian environment.

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PREAMBLE

This document describes the biology of *Lolium multiflorum* Lam. (Italian ryegrass), *Lolium perenne* L. (perennial ryegrass) and *Lolium arundinaceum* Schreb. (tall fescue), with particular reference to the Australian environment, cultivation and use. Information included relates to the taxonomy and origins of cultivated *L. multiflorum*, *L. perenne* and *L. arundinaceum*, general descriptions of their morphology, reproductive biology, biochemistry, and biotic and abiotic interactions. This document also addresses the potential for gene transfer to occur to closely related species. The purpose of this document is to provide baseline information about the parent organism in risk analysis of genetically modified *L. multiflorum*, *L. perenne* and *L. arundinaceum* that may be released into the Australian environment.

Italian ryegrass, perennial ryegrass and tall fescue are all tufted grasses used for both pasture and turf in Australia. Italian ryegrass is a vigorously growing annual or biennial which is native to Europe. Perennial ryegrass is a long-lived, densely tillered perennial, which is native to Europe and neighbouring countries with a temperate climate. Tall fescue is a coarse-leaved perennial native to Europe and North Africa. These three grasses have been grouped together as they have many characteristics in common, are found together in pastures and turf, and interbreed as part of the *Festuca-Lolium* complex. The biology of these grasses will be presented together, with the individual species described only where they differ significantly.

Lolium arundinaceum is the current name for *Festuca arundinacea* following a recent reclassification (as discussed in Section 1). However, much of the older literature concerning tall fescue in the genus *Festuca* and its taxonomy is still of relevance to tall fescue and so will be included in this document.

SECTION 1 TAXONOMY

The *Lolium* and *Festuca* genera are classified within the family Poaceae (Wheeler et al., 2002). Poaceae was previously known as Gramineae (e.g. Mallett and Orchard, 2002a). Within the subfamily Pooideae, also called Festucoideae, the *Lolium* and *Festuca* genera belong to the tribe Poeae (also called Festuceae) (Wheeler et al., 2002). Currently, there are 39 genera listed within the Poeae tribe (Wheeler et al., 2000).

The *Lolium* and *Festuca* genera are closely related. The overall taxonomy of *Lolium* and *Festuca* is ill-defined and there is no comprehensive or definitive world-wide taxonomic treatment. These grasses are commonly referred to as the *Festuca-Lolium* complex (Jauhar, 1993). *Lolium* is thought to be of more recent origin than *Festuca* (Charmet et al., 1997; Pašakinskiene et al., 1998), and may have derived from *Festuca* via an inflorescence transformation from panicle to spike (Essad 1962 cited in Bulínska-Radomska and Lester, 1988).

The number of species in *Lolium* is thought to be around seven to ten, all of which are diploid (Charmet and Balfourier, 1994). There are no native *Lolium* species in Australia (Wheeler et al., 2000; Wheeler et al., 2002; Wipff, 2002; Jessop et al., 2006a, b). However, about seven introduced species of *Lolium* (Table 1) have become naturalised in Australia (Wheeler et al., 2000; Wheeler et al., 2002; Wipff, 2002; Jessop et al., 2006a).

There are 400 or more species within the genus *Festuca* of varying ploidy levels (Charmet and Balfourier, 1994; Jessop et al., 2006a), although species estimates do vary (Wheeler et al., 2000; Wheeler et al., 2002; Jessop et al., 2006a). The grasses in the genus *Festuca* were originally classified into six sections: Bovinae, Ovinae, Montanae, Scariosa, Sub-bulbosae and Variae (Gaut et al., 2000), and includes broad-leaved (Bovinae-subgenus *Schedonorus*) and fine-leaved species (Ovinae – subgenus *Festuca*). About twelve *Festuca* species (six native and six introduced) are present in Australia (Table 2). Originally more *Festuca* species were classified as being present in Australia, but three Australian species have been transferred to the genus *Austrofestuca* (Wheeler et al., 2002), and others have been transferred into various genera including Australopyrum, *Diplachne*, *Dryopoa*, *Festucella*, *Glyceria*, *Panicularia*, *Poa*, *Triodia*, *Tripogon*, *Uniola* and *Vulpia* (APNI, 2006).

Festuca and *Lolium* have been distinguished on the basis of inflorescence morphology. The former species have a spicate inflorescence with two sterile glumes and the latter species are paniculate with one sterile glume in all but the terminal spikelet (Stebbins, 1956), so this has been used to support their status as separate genera (Clayton and Renvoize, 1986). However, there is much debate as to whether this separation is phylogenetically accurate. A spontaneous mutation in *L. multiflorum* occurred which converted the spicate inflorescence into a paniculate form, thus erasing the taxonomic distinction between the two genera, and illustrating how closely related the genera are (Jauhar, 1993). Many studies have found that molecular, cytological, morphological, and fertility data do not support separate genera, instead highlighting the especially close relationship between *Lolium* and broad-leaved

Festuca (subgen. *Schenodorus*) (Bovinae) species (Peto, 1933; Crowder, 1953; Stebbins, 1956; Terrell, 1966; Lehväslaiho et al., 1987; Bulínska-Radomska and Lester, 1988; Aiken et al., 1992; Darbyshire and Warwick, 1992; Loos, 1993; Charmet and Balfourier, 1994; Xu and Sleper, 1994; Stammers et al., 1995; Charmet et al., 1997; Gaut et al., 2000).

The closest relationships are among L. pratense, L. arundinaceum, L. perenne and L. multiflorum. Lolium-Festuca crosses (×Festulolium Aschers. & Graebner.) can be fertile and have an ability to backcross with either of the parents (Wipff, 2002). In 1956, Stebbins proposed that Lolium was merely a section of the large and diverse Festuca genus (Stebbins, 1956). Taking more recent evidence into account, Jauhar (1993) suggested that Lolium should become a subgroup of *Festuca*, with the outbreeding species (*L. perenne*, L. arundinaceum and L. multiflorum) including in the Section Bovinae, and the in-breeders remaining in the Section Lolium. Other taxonomists have recognised the close relationship between some species in the Festuca and Lolium genera by placing the species of Festuca subgenus Schedonorus in the genus Schedonorus P. Beauv (Soreng and Terrell, 1997). However, only the inflorescence would distinguish the genus Schedonorus from Lolium (Bulínska-Radomska and Lester, 1988; Darbyshire, 1993). Hence, Darbyshire has proposed that the broadleaved species in *Festuca* subg. *Schedonorus* be reclassified to *Lolium* subg. Schedonorus (Darbyshire, 1993). This subgenus includes tall fescue (Lolium arundinaceum (Schreb.) Darbysh.), meadow fescue (Lolium pratense (Huds.) Darbysh.) and giant fescue Lolium giganteum (L.) Darbysh. Although there is still debate regarding this nomenclature (Aiken et al., 1997), these are the scientific names that will be used in this Biology document.

The taxonomy is further complicated by the fact that some taxonomists give species status to some hybrids of *Lolium* and/or *Festuca* sp. Many of the *Lolium* species hybridise freely, e.g. *L. perenne*, *L. multiflorum* and *L. rigidum* (Wheeler et al., 2002) resulting in fertile progeny (Wipff, 2002), with the hybrid populations showing a continuum of variation (Wheeler et al., 2002). Hybrids also occur between the *Lolium* species *L. arundinaceum*, *L. pratense* and *L. giganteum* (Wipff, 2002). In fact, *L. pratense* and *L. arundinaceum* were previously classified as a single species, *F. elatior* in older literature (Gibson and Newman, 2001).

Table 1. Lolium species in Australia

Scientific name	Synonyms/Varieties/ Subspecies	Common name	Areas present in Australia	Flowering time	Natural hybrids (and their respective synonyms if available)
L1 ^{b,h,o} Lolium arundinaceum (Shreb.) Darbysh.,	*a,b,t)d,e,f <i>Festuca arundinacea</i> Shreb. a,b,hSchedonorus phoenix (Scop.) Holub., Poa phoenix Scop., ^c Festuca elatior var. arundinacea, ^{f,h} Festuca elatior L., ^h Schedonorus arundinaceus (Shreb.) Dumort., Festuca arundinacea Schreb., ⁿ Festuca arundinacea var. glaucescens	^{a,b,c,h,t} Tall fescue, ^{b,t} Tall meadow fescue, Alta fescue, Reed fescue	^{a,b,d,i,t,y} NSW, Vic, ^x Tas, SA (^{c,g} naturalised), WA, ^{b,e,i,t,y} Qld, ^{d,e,y} ACT, ^y NT	^t October - April	^{k,o} L1 × L7 = ^k F. ×aschersoniana Doerfler ^{k,o} L1 × L2 (S) = F. ×fleischeri Rohlena ^r L1 × F9 ^{k,o} L4 × L1 ^{k,o} L5 × L1
L2 ^w Lolium giganteum (L.) S.J.Darbysh	^{(*w)f} Festuca gigantea (L.) Vill.	^w Giant fescue, Tall brome, Giant green fescue	^{f,y} ACT		^{k,o} L1 × L2 (S) = <i>F. ×fleischeri</i> Rohlena °L2 × L7
L3 ^{(*a,b,t)e,f} Lolium loliaceum (Bory & Chaub.) HandMazz	^g Lolium subulatum Vis., Rottboellia Ioliacea Bory & Chaub., Lolium rigidum Gaudin var. rottboellioides Heldr. Ex. Boiss., Lolium rigidum Gaudin ssp. lepturoides (Bois.)Sennen & Mauricio, Lolium Iepturoides Boiss.	^{a,b,t} Stiff ryegrass, ^{b,t} Rigid ryegrass, Annual ryegrass, Wimmera ryegrass	^{d,e,i,t,y} NSW, Vic, Tas, SA (^g naturalised), WA, ^{d,e,I,y} Qld, ^{d,e,y} NT, ^{e,y} ACT	^t September - February	^s L3 × L5 (S) ^{j,s} L3 × L8 (S) ^s L3 × L9 (S) + (F) ^{j,s} L3 × L10 ^s (S)
L4 ^{(*a,b,t)d,e,f,h} Lolium multiflorum Lam.	^g Lolium italicum A.Braun, Lolium perenne L. var. multiflorum (Lam.)Parn., ^h Lolium perenne var. multiflorum (Lam.) Husn., ^o Lolium perenne var. aristatum Willdenow.	^{a,b,h,o,t} Italian ryegrass, ^{b,c,t} Westerwolds ryegrass, ^o Annual ryegrass	^{d,i,t,y} Tas, SA ^{d,e,i,t,y} (^g naturalised), NSW, Vic, WA, Qld, ^{d,e,I,y} ACT, ^{d,e} NT (^c naturalised in Australia and sown pasture species)	^t September - December	^{k,l,o,s} L4 × L5 ^{o,s} (F) = * ^{b,e,f} Lolium ×hybridium Hausskn., ^b Lolium multiflorum Lam. × L. perenne L., ^e Lolium multiflorum × L. perenne , ^k Festulolium holmbergii (Doerfler)P.Fourn. ^o L4 × L9 (F) = * ^b Lolium ×hubbardii Jansen & Wachter ex B.K.Simon, ^b Lolium ×hubbardii Jansen & Wachter, Lolium multiflorum Lam. × L. rigidum L., ^{e,f} Lolium hubbardii B.K.Simon ^s L4 × L10 (F) ^{k,o} L4 × L1 ^{k,o} L4 × L7 °(S) + (F)

Scientific name	Synonyms/Varieties/ Subspecies	Common name	Areas present in Australia	Flowering time	Natural hybrids (and their respective synonyms if available)
L5 ^{(*a,b,t)d,e,f} Lolium perenne L.	^e Lolium perenne var. italicum (A.Braun) Rodway, Lolium perenne L. var. perenne, ^{e.t} Lolium perenne var. cristatum Pers.	^{a,b,c,h} Perennial ryegrass, ^b English ryegrass	^{d,e,i,t,y} NSW, Vic, ^x Tas, SA (^g naturalised), Qld, NT, ^{d,e,I,y} ACT, WA (^c naturalised in Australia and sown pasture species)	^t August - February	^s L3 × L5 (S) ^{k,l,o,s} L4 × L5 ^{o,s} (F) = * ^{b,e,f} Lolium ×hybridium Hausskn., ^b Lolium multiflorum Lam. × L. perenne L., ^e Lolium multiflorum × L. perenne , ^k Festulolium holmbergii (Doerfler)P.Fourn. ^s L4 × L8 (S) ^{o,s} L5 × L9 (F) ^s L5 × L10 (S) ^{k,o} L5 × L1 ^{k,o} L5 × L7 °(S) + (F) = "Festulolium loliaceum ^r L5 × F9
L6 ^{(*w)f} Lolium persicum Boiss. & Hohen.		^w Persian ryegrass, Persian darnel	^{f,y} ACT, ^y SA		^m L6 × L10 (F)
L7 <i>Lolium pratense</i> (Huds.) Darbysh.,	^e Festuca elatior var. pratensis Hack, ^a Festuca elatior L., ^{a,b} Schedonorus pratensis (Hudson) Beauv., ^b Festuca elatior sensu J.M.Black, (*a,g,t)b,e,fFestuca pratensis Huds., ^{b,g} Festuca elatior L. subsp. pratensis (Huds.) Hack, ^g Festuca elatior auct.non L: J.M.Black(1943)	^{a,t} Meadow fescue	^{a,b,d,e,i,t,y} NSW, ^{d,y} Vic, ^{b,d,t,y} SA (^g naturalised), ^y WA, ^{d,e,l,y} ACT, ^y Qld, ^y Tas (^b naturalised in Australia)	^t Summer- Spring	^{k,o} L1 × L7 = ^k F. ×aschersoniana Doerfler ^o L2 × L7 ^{k,o} L4 × L7 ^o (S) + (F) ^{k,o} L5 × L7 ^o (S) + (F) = ^u Festulolium loliaceum (^{f,v} Festuca loliacea Huds.)
L8 ^{(*w)f} Lolium remotum Schrank	^w Lolium linicola	^w Hardy ryegrass	^{d,y} Vic, ^{e,y} ACT, ^y WA		^{j,s} L3 × L8 (S) ^s L5 × L8 (S) ^{j,s} L8 × L10 ^s (S)
L9 ^{(*a,b,t)d,e,f} Lolium rigidum Gaudin	^g Lolium subulatum auct.non Vis: J.M.Black(1943), Lolium perenne L. ssp. rigidium (Gaudin)A.Love & D.Love, Lolium multiflorum Lam. var. rigidum (Gaudin)Trab., ^t Lolium subulatum Vis, Lolium rigidum Gaudin subsp. lepturoides (Boiss.)	^{a,b, c,t} Wimmera ryegrass, ^{b,c,t} Annual ryegrass, ^{b,t} Rigid ryegrass, ^o Stiff ryegrass	^{d,e,i,t,y} NSW, Vic., ^x Tas, SA (^g naturalised), Qld, NT, ^{d,e,I,y} ACT, WA (^c naturalised in Australia and sown pasture species)	^t August - December	^s L3 × L9 (S) + (F) ^o L4 × L9 (F) = * ^b Lolium ×hubbardii Jansen & Wachter ex B.K.Simon, ^b Lolium ×hubbardii Jansen & Wachter, Lolium multiflorum Lam. × L. rigidum L., ^{e,f} Lolium hubbardii B.K.Simon ^{o,s} L5 × L9 (F) ^s L9 × L10 (S) + (F)

Scientific name	Synonyms/Varieties/ Subspecies	Common name	Areas present in Australia	Flowering time	Natural hybrids (and their respective synonyms if available)
	Sennen & Mauricio				
L10 ^{(*a,b,t)e,f} Lolium temulentum L.	^{g.t} Lolium arvense With., ^e Lolium temulentum var. arvense (With.) Lilj., Lolium temulentum L. var. temulentum, ^f Lolium temulentum var. linicola Benth., * ^{b,g,t} Lolium temulentum L. forma temulentum, * ^{b,g} Lolium temulentum L. forma arvense (With.) Junge, ^g Lolium temulentum L. var. leptochaeton A.Braun, * ^t Lolium temulentum var. arvense Lilj.	^{a,t} Darnel, ^b Bearded darnel, ^{b,t} Drake	^{a,d,l,y} Vic, ^{a,d,i,t,y} NSW, SA (^g naturalised), WA, Qld, ^{d,e,f,y} ACT, ^{a,b,i,t,x,y} Tas.	^t June - January	$^{j,s}L3 \times L10 \ {}^{s}(S)$ $^{s}L4 \times L10 \ (F)$ $^{s}L5 \times L10 \ (S)$ $^{m}L6 \times L10 \ (F)$ $^{j,s}L8 \times L10 \ {}^{s}(S)$ $^{s}L9 \times L10 \ (S) + (F)$
* ^b Lolium ×hubbardii Jansen & Wachter ex B.K.Simon	^b Lolium ×hubbardii Jansen & Wachter, Lolium multiflorum Lam. × Lolium rigidum L., ^{e,f} Lolium hubbardii B.K.Simon	[▶] Ryegrass	^{d,y} Vic, Tas, SA, ^y WA (^g naturalised), ^{e,y} Qld		
^{*b,e,f} Lolium ×hybridium Hausskn.	^b Lolium multiflorum Lam. × Lolium perenne L., ^e Lolium multiflorum × L. perenne	^b Ryegrass	^{d,y} Vic, Tas, SA (^g naturalised), WA, NT, ^y NSW		

(Note: ACT was only differentiated from NSW in ^dAVH (Atlas of Living Australia, 2022b), ^eAPC (CHAH, 2006), ^fAPNI (APNI, 2006) and ⁱMallett and Orchard (2002a)).

Lolium ×hubbardii and Lolium ×hybridium are recorded in the table twice as both species and hybrids. Although they are hybrids they are also given species status in some literature (APNI, 2006; Jessop et al., 2006a).

Key:* = introduced into Australia, • = native to Australia, (F) = Fertile (if known), (S) = Sterile (if known), (S) + (F) = some progeny may be fertile (if known).

Literature sources: ^a Wheeler et al. (2002), ^b Jessop et al. (2006b), ^c Lamp et al. (2001), ^d AVH (2022), ^e CHAH (2006), ^f APNI (2006), ^g Barker et al. (2005), ^h Wheeler et al. (2000), ⁱ Mallett and Orchard (2002b), ^j Jenkin (1954), ^k Gibson and Newman (2001), ^l Giddings et al. (1997b), ^m Senda et al. (2005), ⁿ Kopecký et al. (2006), ^o Wipff (2002), ^p Thorogood (2003), ^q Meyer and Watkins (2003), ^r Ruemmele et al. (2003), ^s Jenkin and Thomas (1938) (*L. multiflorum* is referred to as *L. italicum*), ^t Sharp and Simon (2002), ^u Giddings et al. (1997a), ^v GRIN (2005), ^w Randall (2002), ^x de Salas and Baker (2017), ^y Atlas of Living Australia (2022b).

Table 2. Festuca species in Australia

Scientific name	Synonyms/Varieties/ Subspecies	Common name	Areas present in Australia	Flowering time	Natural hybrids (and their respective synonyms if available)
F1 (•x)e,f <i>Festuca archeri</i> E.B.Alexeev			^{e,x, y} Tas		
F2 • ^{a,e,f,t} Festuca asperula Vickery		^{a,t} Graceful fescue	^{a,d,e,i,t, y} NSW, Vic, ^{a,d,e, y} Tas, ^{I,y} ACT	^t December - February	
F3 ^{(•b,g,t)e,f} Festuca benthamiana Vickery	^{e,g} Festuca duriuscula var. aristata Benth.	^b Fescue	^{e,y} Vic, ^{b,d,i,t,y} SA, ^y NSW	^t October - November	
F4 •ª <i>Festuca glauca</i> Vill.			^{a, y} NSW, ^y ACT, ^y SA,		
F5 ^{(*w)e,f} Festuca longifolia Thuill.		^w Hard fescue, Blue fescue	°Qld?		
F6 • ^{a,e,f,t} Festuca muelleri Vickery			^{a,d,e,i,t, y} NSW, Vic, ^{d,e,l, y} ACT	^t December - March	
F7 * ^{a,e,f,t} Festuca nigrescens Lamk.	^a Festuca rubra L. subsp. commutate Gaud., ^t Festuca rubra L. var. commutate Lam.	^a Chewings fescue	^{a,d,e,i,t,y} NSW, Vic, ^y Tas, ^a SA, WA, ^{d,e,I, y} ACT	^t November - January	
^{f,y} Festuca ovina L.	^e Festuca ovina subvar. durissima Hack., Festuca ovina subsp. eu-ovina Hack.		^y ACT, ^y NSW, ^y Vic		
F8 (• ^t) ^{e,f} Festuca plebeia R.Br.	· · · · · · · · · · · · · · · · · · ·		d,e,i,t, YTas	^t December - February	
F9 (* ^t) ^{b,d,e,f} Festuca rubra L.	 *aFestuca rubra L. subsp. rubra, ^eFestuca rubra subsp. commutata Gaudin, Festuca rubra L. var. rubra, ^bFestuca duriuscula sensu J.M.Black, Festuca asperula sensu H.Eichler. ^gFestuca duriuscula L., Festuca asperula auct.non Vickery: H.Eichler(1965) 	^{a,c,t} Red fescue, Creeping fescue	^{a,b,c,d,e,i,t, Y} NSW, Vic, ^Y Tas, ^{b,d,e,t,} ^Y SA (^g naturalised), ^{d,e,I, Y} ACT, ^{i,t,} ^Y WA (^b naturalised in Australia)	^t October - January	^r L1 × F9 ^r F9 × F10 = H1 ^r F9 × H1 (backcross) ^r L5 × F9

(Note: ACT was only differentiated from NSW in ^dAVH (Atlas of Living Australia, 2022b), ^eAPC (CHAH, 2006), ^fAPNI (APNI, 2006) and ⁱMallett and Orchard (2002a)).

Key:* = introduced into Australia, • = native to Australia, (F) = Fertile (if known), (S) = Sterile (if known), (S) + (F) = some progeny may be fertile (if known).

Literature sources: ^a Wheeler et al. (2002), ^b Jessop et al. (2006b), ^c Lamp et al. (2001), ^d AVH (2022), ^e CHAH (2006), ^f APNI (2006), ^g Barker et al. (2005), ^h Wheeler et al. (2000), ⁱ Mallett and Orchard (2002b), ^j Jenkin (1954), ^k Gibson and Newman (2001), ^l Giddings et al. (1997b), ^m Senda et al. (2005), ⁿ Kopecký et al. (2006), ^o Wipff (2002), ^p Thorogood (2003), ^q Meyer and Watkins (2003), ^r Ruemmele et al. (2003), ^s Jenkin and Thomas (1938) (*L. multiflorum* is referred to as *L. italicum*), ^t Sharp and Simon (2002), ^u Giddings et al. (1997a), ^v GRIN (2005), ^w Randall (2002), ^x de Salas and Baker (2017), ^v Atlas of Living Australia (2022b).

(The following species were listed as being present in Australia but no specific distribution or further taxonomic information was found. ^f*Festuca billardierei* Steud. (synonyms ^e*Festuca scabra* Labill., *Triticum scabrum* R.Br, *Agropyron scabrum* (R.Br) P.Beauv.), ^f*Festuca duriuscula* L. (synonym ^e*Festuca ovina* var. *duriuscula* (L.) Hack.), F10 = , hybrids ^rF9 × F10 = H1, ^rF10 × H1(backcross)), and ^f*Festuca stuartiana* Steud.).

SECTION 2 ORIGIN AND CULTIVATION

2.1 Centre of diversity and domestication

Western Europe is the main centre of origin of Poeae (Festuceae) (Wipff, 2002; Meyer and Watkins, 2003). Italian ryegrass, perennial ryegrass and tall fescue are native to Europe, temperate Asia and Northern Africa (Lamp et al., 2001). They are defined as 'cool season grasses' because of their preferential adaptation to cool and moist environments (Romani et al., 2002). Tall fescue is a widely adapted Eurasian grass, with natural populations found in sites varying from arid to very wet. The limits of its natural range are determined by extreme cold and by rainfall below 450 mm per year (Easton et al., 1994).

Perennial forage grasses are not domesticated in the strict sense, as 'wild' collections are generally phenotypically indistinct from cultivated forms. The exception to this rule is Italian ryegrass which was selected from a continuum of *Lolium* sp. and now has species status as *L. multiflorum* (Casler and Duncan, 2003). In contrast, cultivars of turfgrasses are considered as domesticated from their wild forms (Casler and Duncan, 2003).

Tall fescue was first documented in Victoria in 1901, with the first commercial cultivar, "Demeter" released in the 1930's (Grassland Society of Southern Australia Inc, 2008c). The first commercial cultivar of perennial ryegrass, named "Victorian" was also released at a similar time (Grassland Society of Southern Australia Inc, 2008b).

2.2 Commercial uses

Italian ryegrass (annual or short-lived perennial), perennial ryegrass and tall fescue (both perennial species) are used for forage and turf purposes throughout the temperate regions of the world including North and South America, South Africa, Australia and New Zealand (Lamp et al., 2001).

All three grass species are often present together. Due to its ability to establish quickly and grow rapidly in its first year, Italian ryegrass is often included in permanent pasture mixtures to provide feed while the slower growing perennials become established. It also provides good winter growth, whereas perennial ryegrass and tall fescue have little or no growth in winter (Lamp et al., 2001).

2.3 Cultivation in Australia

Italian ryegrass, perennial ryegrass and tall fescue are used in both pasture and turf in Australia.

2.3.1 Pasture

All three species are used for dairying and sheep grazing predominantly in the temperate areas of Australia (New South Wales, Victoria, Western Australia, and Tasmania) (Blair, 1997; Lazenby, 1997; Callow et al., 2003). Pastures are usually a mix of species that can include Kentucky bluegrass (*Poa pratensis*), white clover (*Trifolium repens*), fescues (*Festuca* and *Lolium spp.*), ryegrasses (*Lolium spp.*), chicory (*Chicorium intybus*), red clover (*Trifolium pratense*) and others.

Annually oversown Italian ryegrass is the main forage for dairy cows for the cool season in the subtropics of Australia (Lowe et al., 1999b). However, there are drawbacks in the use of annual pasture including annual establishment cost, grazing problems in excessively wet weather and short growing season. Studies assessing the merits of sowing perennial pasture types in the subtropics have shown that modern cultivars of both perennial ryegrass and tall fescue can also support good milk production as well as overcoming the drawbacks of using annually sown pasture (Lowe et al., 1999a).

Most introduced pasture species in Australia were northern European in origin. However, it has been recognised that the Mediterranean climate (hot, dry summers, rainfall predominantly in autumn, winter and spring and mild winters which allow some growth) resembles that of southern Australia more closely than that of northern Europe. Cultivars from the Mediterranean are thus being used to develop cultivars for use in Australia (Lamp et al., 2001) (see Section 2.4.1).

Tall fescue

Tall fescue is widely adapted to a range of growing conditions and two types of tall fescue are currently grown in Australia – those that originate from temperate Europe or America (i.e. spring/summer active varieties) and those that originate from the Mediterranean (i.e. winter active/summer dormant).

Temperate (also known as Continental) varieties are suited to areas with 650 – 700 mm or more rainfall, or on water-logged soils in Victoria (Vic) with 900 mm rainfall. They are therefore recommended for Queensland (Qld), north coast of New South Wales (NSW), high rainfall areas of northern and central NSW tablelands and slopes, Vic, irrigated areas of South Australia (SA) and south coast of Western Australia (WA), and high rainfall areas of Tasmania (Tas) (Milne, 2009). Slow growth will occur during winter if there is sufficient soil moisture (Easton et al., 1994; Harris and Lowien, 2003). The temperate varieties can be further divided into soft- and tough-leaved types. The tough-leaved varieties flower earlier and are hardier with better tolerance to low summer rainfall, mismanagement and lower soil fertility (Grassland Society of Southern Australia Inc, 2008c).

Mediterranean varieties are suited to areas with dry summers and 450 – 550 mm rainfall and are therefore better adapted to summer drought than the temperate varieties (Easton et al., 1994; Harris and Lowien, 2003). The level of summer dormancy varies between cultivars, and can range between totally dormant to some summer production. These varieties are more suited to western areas of NSW tablelands and slopes, southern NSW, south-east SA, central and east coast of Tas and south-west WA (Milne, 2009).

Use of tall fescue is limited by its slow establishment, particularly when compared with ryegrass (Easton et al., 1994). Mature tall fescue may be cut in early Spring for silage (Burnett, 2006a).

Ryegrass

Perennial ryegrass is used for dryland pastures or irrigated for grazing, hay or silage. It has excellent seedling vigour which makes it easy to establish and it has a rapid recovery after grazing (Lowien et al., 2004). It is widely sown in the high-rainfall (600 – 800 mm) zones of south-eastern Australia despite the fact it shows poor persistence that makes it susceptible to invasion by less desirable species and causes loss of quality and production (Waller and Sale, 2001). This poor persistence is due in part to the Mediterranean-like hot, dry summer climate to which the traditional perennial ryegrass cultivars are not suited. Breeding programmes using Mediterranean germplasm together with grazing management strategies have been proposed as a means of increasing persistence and productivity (Waller and Sale, 2001). Despite this poor relative productivity, perennial ryegrass was shown to be one of the best performing fodder grasses amongst 22 tested in diverse low-rainfall Australian sites (Reed et al., 2008), highlighting its present and future importance.

The Pasture Species Database provides the following information about Italian ryegrass (Grassland Society of Southern Australia Inc, 2008a):

It is an annual/short rotation grass used for pasture and high-quality hay and silage production in areas of high rainfall (> 650 mm), a temperature range of 0-30° C and under conditions of high fertility. It is not tolerant of dry conditions. Its major use is as a special purpose winter/spring fodder crop for beef and dairy; use in sheep production requires careful management. It establishes quickly and can withstand heavy grazing.

Italian ryegrass is widely grown both under irrigated and dryland conditions in the temperate regions of Australia, and, for example, is the major grass species used in the high rainfall areas of south-western Australia (Bolland et al., 2001). It is also oversown into summer-growing subtropical and tropical perennial pastures in eastern Australia (Qld and NSW) to sustain forage production during lower winter temperatures (Lowe et al., 1999a).

2.3.2 Turf

All three species are also used for turf primarily in temperate regions (Lamp et al., 2001; Gardenet, 2006; Canturf, 2017; Yates, 2017). Growing mixes of species in turf is thought to be advantageous to monocultures for various reasons, including better use of soil moisture during times of low rainfall (Skinner et al., 2004). Most turfs are made up of a mixture of grasses, such as ryegrasses (*Lolium spp.*), fescues (*Festuca* and *Lolium spp.*), buffalo grasses (*Stenophrum* and *Buchloe spp.*), couch or Bermuda grasses (*Cynodon spp.*), bentgrasses (*Agrostis spp.*) and field and bluegrasses (*Poa spp.*) (Canturf, 2017; Yates, 2017).

2.3.3 Commercial propagation

Seed production

The Australian pasture seed industry has a research and development program that is administered by AgriFutures Australia (AgriFutures Australia, 2018). Seed for cultivars such

as those listed in Appendix 1 can be produced under a seed certification scheme (Seed Services Australia, 2020) that ensures a minimum standard for purity, seed germination and seed-borne disease. Seed certification, which is overseen by a national authority in Australia (Australian Seeds Authority, 2022), is voluntary and documents seed for its genetic purity and physical quality.

Certified seed is classed according to its generation along the pedigree. Breeders seed is used to produce Pre-basic, which is then used to produce Basic, which in turn is used to produce First Generation or C1 certified seed. Most certified seed is C1 class grown from Basic seed.

The information in Figure 1, which is an extract from the South Australian Seed Certification Scheme (Seed Services Australia, 2020), provides an example of the sorts of conditions applying to the production of certified seed of a grass species in Australia. Such production may require practices that are not commonly applied to pastures, such as wide row spacing, irrigation, residue burning, isolation from neighbouring species that may cross pollinate and herbicide application to control weeds.

Figure 1. C	Certified Seed Crop	Standard for Tall Fescue	(Seed Services Austr	alia, 2020)
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Sowing Seed Basic seed.	
Paddock History Land must not have grown or been sown to tall fescue in the previous two (2) years, unless it was the same cultivar and certification class where a minimum one (1) year break between crops is recommended to meet varietal purity standards. New crops at the seedling inspection containing mature or volunteer tall fescue plants will be rejected from certification.	Isolation For areas larger than 2 hectares: <u>Basic</u> : 100 metres from other cultivars <u>Certified</u> : 50 metres from other cultivars For areas of 2 hectares or less, double the isolation distances.
Inspections Seedling inspection Pre-harvest inspection Registration inspection	Stand Life Basic: two (2) years (maximum) Certified: five (5) years (maximum) Where Basic stands are down-graded certified seed may be produced for a further three (3) years. Crops that have thinned out significantly from the previous year will be rejected. Classes C1: from areas sown with Basic seed.

	Seed Quality Standards
	Basic Class
Contaminant Basic Certified Other off-types or varieties of tall fescue 1 per 30 m² 1 per 10 m² Seed produced from regenerated seedlings in 3	Minimum Pure Seed (% by mass) 99.0% Minimum Germination (% by count) 75.0% (including fresh ungerminated seed) Maximum Other Seeds (% by mass) 0.3% Nominated species (see Table 1 & Note 3)
Plants of other species, the seeds of which are difficult to distinguish in a laboratory test or which will readily cross- pollinate with the crop being grown for seed 1 per 30 m ² 1 per 10 m ²	<u>Certified class</u> Minimum Pure Seed (% by mass) 9.0% Minimum Germination (% by count) 75.0% (including fresh ungerminated seeds) Maximum Other Seeds (% by mass) 3.0% of which no more than 1.0% shall be seeds other than <u>Lolium sp</u> .

Seed production, whether for certified or non-certified seed, is closely associated with seed processing, which primarily involves seed cleaning and packaging and, if required, dressing the seed with fungicides and/or insecticides (NSW DPI, 2017).

Turf growing

While turfgrass grown in commercial turf farms can be established either through vegetative means (sprigs or plugs¹) or by direct seeding, it is common for the cool-season grasses such as ryegrass and tall fescue to be established from seed (Perez et al., 1995). After sowing and germination, the turf is managed using a variety of approaches that may include frequent irrigation, regular mowing, vacuum removal of clippings, fertiliser application and weed control. The turf is maintained for up to 24 months before being harvested and supplied to end users such as landscapers, sports field managers and home gardeners (Perez et al., 1995). Specialised turf harvesters (or sod cutters) are designed to cut rectangular sods of turf that are then transported as rolls or pads before being laid directly on a prepared surface to provide an 'instant' lawn. Cost effective and less complete turf coverage for areas not requiring immediate use may involve the planting of sprigs or plugs by methods developed by individual turf suppliers.

2.3.2 Scale of cultivation

Appendix 1 lists examples of commercial cultivars of perennial ryegrass, Italian ryegrass, and tall fescue grown as pasture or turfgrasses in Australia.

A breakdown of the cultivation statistics for each of the three grasses is not possible to obtain because, apart from seed production, none of the species is harvested for individual sale and therefore production statistics for the three species are subsumed within the statistics for more general categories such as pasture or turf. However, estimates of the use of perennial ryegrass and tall fescue in Australian pasture have been made. Pasture

¹ Sprigs are small pieces of stem with leaves and some root development; plugs are usually squares of sod measuring approximately 50 mm wide x 50 mm deep; sods are rolls or pads of mature grass (typically approximately 2 m long for coverage of small areas and up to 25 m long for coverage of large areas) with a layer of roots and growing medium at the base.

containing perennial ryegrass was estimated to cover an area of 35,420 km² and pasture containing tall fescue an area of 10,992 km² across the Australian states, compared to 78,151 km² and 35,420 km² for white clover and lucerne respectively (Hill and Donald, 1998). The relative importance of the three species can be further gauged from the figures for seed production. Perennial ryegrass is the most important sown pasture grass species in temperate Australia and other temperate regions of the world (Cunliffe et al., 2004). Within Australia, 2003 sales of ryegrass seed for pasture were estimated at 6,200 tonnes, with approximately 60% of this being perennial ryegrass. Annual sales of tall fescue seed were estimated at between 400-500 tonnes (RIRDC, 2003). A breakdown of certified seed production is given in Table 3.

In 2003, ryegrass (approximately 4,600 tonnes) and tall fescue (approximately 700 tonnes) were the only pasture seeds imported into Australia in significant amounts (RIRDC, 2003, 2014).

Species	Cultivar	2015/16	2016/17	2017/18	2018/19	2019/20
Lolium perenne	Victorian	395	532	423	334	324
	Proprietary Varieties	645	3289	1001	1039	1098
Lolium multiflorum	Proprietary Varieties	4826	5026	4144	4218	6042
Festuca arundinaceum	Demeter	N/A	27	40	N/A	N/A
	Proprietary Varieties	141	75	63	42	4

Table 3. Production (tonnes) of certified seed of three grass species in Australia between 2015 and 2020*

* data obtained from Australian Seeds Authority (Australian Seeds Authority Ltd., 2020). N/A, not available.

2.3.3 Cultivation practices

Autumn and early winter (March – June) are the best times to sow tall fescue. Sowing in September in high altitude areas can also be successful if there is high rainfall. Tall fescue has poor seedling vigour, with the roots and crown developing slowly. As a result, tall fescue is sensitive to competition from more vigorous pasture and weed species (Harris and Lowien, 2003). However, some of the new cultivars have improved seedling vigour.

Like ryegrass, tall fescue has a high requirement for nitrogen and its persistence is likely to be poor if high soil fertility is not maintained (Grassland Society of Southern Australia Inc, 2008c). Generally tall fescue is sown in combination with legumes and as long as these are fertilised regularly with phosphorous and sulphur they will provide sufficient nitrogen for tall fescue growth (Harris and Lowien, 2003). Seeding rates for tall fescue in pastures in Australia vary from 6-10 kg/ha for dryland pastures (Harris and Lowien, 2003). Up to 25 kg/ha is recommended by some authors for irrigated fields or areas of high rainfall (Grassland Society of Southern Australia Inc, 2008c). Perennial ryegrass can withstand close continuous grazing and is ideally suited to intensive sheep and cattle grazing. It is vigorous and competitive above and below ground – leading it to preferentially establish in competition with other plants or crops (Donald, 1958). Italian ryegrass with its more upright and open growth habit is suited to grazing systems with lengthy intervals between grazing, or for silage production (Jung et al., 1996). Seeding rates for perennial ryegrass are 8-20 kg/ha (Fulkerson, 2007; Grassland Society of Southern Australia Inc, 2008b; Griffiths et al., 2011). Rates for Italian ryegrass are slightly higher at 10-20 kg/ha for the diploids or 15-25 kg/ha for tetraploids (Grassland Society of Southern Australia Inc, 2008a).

2.4 Crop Improvement

Pasture

Factors which are considered when a new pasture plant introduction is made include; adaptation to the environment, e.g. climatic conditions and soil factors; higher yields than the resident species and increased winter/autumn production; higher nutritive value/herbage quality; seedling vigour; even spread of growth; persistence and tolerance to grazing; ability to combine with other grasses and legumes; adequate seed production; pest and disease resistance; no adverse effects on animals; ease of harvest; herbicide tolerance and lower endophyte toxicity; and increased seed yield (Cunningham et al., 1994; Blair, 1997; Oram and Lodge, 2003).

Turf

The major selection criteria for turf fall into three main groups:

1) turf growth characteristics and appearance (which includes visual quality, shoot/turf density, percentage ground cover/turf density, leaf texture, turf colour, spring 'greenup', seedling vigour and establishment, seed yield, and maturity),

2) disease resistance, and

3) resistance or tolerance to environmental factors (including wear, acid, salt and drought stress) (Stewart, 2002; Meyer and Watkins, 2003; Thorogood, 2003).

2.4.1 Breeding

Breeding in Italian ryegrass, perennial ryegrass and tall fescue for pasture and turfgrass has been extensively reviewed (Cunningham et al., 1994; Easton et al., 1994; Reed, 1996; Meyer and Belanger, 1997; Meyer and Watkins, 2003; Oram and Lodge, 2003; Thorogood, 2003; Bonos et al., 2006; Sampoux et al., 2011; Sampoux et al., 2013). Breeding objectives depend on where and how the grass is to be grown (Thorogood, 2003). Grasses are phenotypically variable and adapt readily to their environment (Casler and Duncan, 2003). Breeding for turf selects plants that can be mown close to the ground and maintain dense, high-quality turf. This requires selection for finer, shorter leaf blades, finer stems and shorter internodes (Casler and Duncan, 2003). Classical breeding approaches such as phenotypic and genotypic recurrent selection have been widely used in grasses that are cross-pollinated and are predominantly selfincompatible. Identification of superior genotypes and their subsequent interbreeding to produce new combinations of genotypes with improved expression of specific characters is the basis of this process. Selection of germplasm from new cultivars, old pasture, turf types, the wild, or combinations of these, can be used (Stewart, 2002; Bonos et al., 2006). Quantitative trait loci (QTL) have been identified in perennial ryegrass for some complex traits such as water-soluble carbohydrates (Turner et al., 2006), crown rust (Puccinia coronata Corda) resistance (Thorogood et al., 2001), delayed leaf senescence (Thorogood et al., 1999), biomass yield (Anhalt et al., 2009), and freezing tolerance (Hulke et al., 2012). QTLs have also been identified for grey leaf spot resistance in Italian ryegrass (Takahashi et al., 2014). Crown rust resistance was also identified in a perennial x Italian ryegrass population (Sim et al., 2007). Experiments have also been conducted to produce tetraploid perennial ryegrass cultivars using colchicine treatment, due to the improved performance of European tetraploids (Nair, 2004). Molecular and genomics approaches have also been employed in grass breeding including production of restriction fragment length polymorphism (RFLP) markers, simple sequence repeats and expressed sequence tag-simple sequence repeats (EST-SSR) markers (Zhang et al., 2006).

Many improved cultivars for use in pasture or turf are available in Australia (see Appendix 1). Information in Table 4 and Table 5 give some indication of the types of characteristics which have been selected for. Owing to the poor persistence of perennial ryegrass in pastures (see Section 2.3.1) particular attention has been paid to the improvement of this species and in the 1980s a National Perennial Ryegrass Improvement Program (NRIP) was initiated (Waller and Sale, 2001).

Cross-breeding between *Festuca* and *Lolium* species is used to introduce traits (e.g. improved drought and frost tolerance and rust resistance) not present in the individual species (Humphreys and Thomas, 1993; Oertel and Matzk, 1999; Skibinska et al., 2002; Humphreys et al., 2005; Kosmala et al., 2006).

Festulolium (*Festuca* × *Lolium* crosses) have been released as short-term perennials, but further work to improve hardiness, palatability and digestibility is necessary (Oram and Lodge, 2003). Natural and artificial hybrids are also known to occur between some *Festuca* and *Vulpia* species (Ainscough et al., 1986). Some genes that enable *Vulpia* to grow successfully on infertile, acid soils could therefore be transferred to tall fescue by standard backcrossing procedures (Oram and Lodge, 2003).

Species	Key features (turf cultivars)	Key features (pasture cultivars)
Perennial ryegrass	 Dark green Heat and stress tolerance High endophyte Dense turf 	 High rust-tolerance Heat tolerance Versatile grazing management Persistence

Table 4. Examples of key features in Italian ryegrass, perennial ryegrass and tall fescue cultivars bred for either turf or pasture use in Australia*.

Species	Key features (turf cultivars)	Key features (pasture cultivars)
Tall fescue	 Wear tolerance Disease resistance Shade tolerance Rapid establishment Active in cool season Absence of browning Very dark green Fine leaf texture Strong, vigorous, dense turf Dwarf type Heat and drought tolerant Vigorous establishment Resists weed invasion Endophyte mediated pest resistance Shade and sun-tolerance Brown patch and leaf spot tolerance Hard wearing 	 Surplus convertible to silage Selective herbicides available Rapid establishment Excellent winter and summer growth Insect tolerance Palatability Versatile grazing management Good summer growth Palatability Tolerance to disease, pests and drought Persistence Lower water requirement than ryegrass Cool season growth Rust resistant Improved survival under hot conditions
Italian Ryegrass		 Quick growth Persistence Rotational grazing Australian cultivar Seedling vigour Disease resistance Palatability

*(Barenbrug Australia, 2021; AGF Seeds, 2022; Barenbrug Australia, 2022; PGG Wrightson, 2022; Upper Murray Seeds, 2022).

2.4.2 Genetic modification

Genes have been introduced into Italian ryegrass, perennial ryegrass and tall fescue using various methods, including biolistics, protoplasts, whiskers and *Agrobacterium*. These are reviewed by a number of authors (Lee, 1996; Wang and Ge, 2006; Wang and Brummer, 2012) and examples are listed as follows:

Italian ryegrass

Biolistics (Ye et al., 1997; Dalton et al., 1999); protoplasts (Wang et al., 1997); whiskers (Dalton et al., 1998); and *Agrobacterium* (Bettany et al., 2003).

Perennial ryegrass

Biolistics (Spangenberg et al., 1995b); protoplasts (Wang et al., 1997); whiskers (Dalton et al., 1998); and *Agrobacterium* (Wang et al., 1997; Wu et al., 2005; Beechey-Gradwell et al., 2022b).

Tall fescue

Biolistics (Spangenberg et al., 1995c; Cho et al., 2000; Wang et al., 2001), protoplasts (Wang et al., 1992); whiskers (Dalton et al., 1998); and Agrobacterium (Bettany et al., 2003).

All three grass species have been genetically modified (Wang and Ge, 2006; Wang and Brummer, 2012). Some of the known traits as of November 2022 are listed in Table 5. None

of these GM grasses have been approved for commercial release in Australia or internationally.

A number of these GM plants have been trialled in Australia and overseas including those with altered lignin content, reduced pollen allergens, improved forage quality, and herbicide tolerances.

Trait	Plant species	Reference	
Phosphinothricin tolerance	Tall fescue	(Bettany et al., 2003; Wang et al., 2003; Wang and Ge, 2005)	
	Perennial Ryegrass	(Wu et al., 2012)	
	Tall fescue	(Bettany et al., 2003)	
Colour marker	Italian ryegrass	(Bettany et al., 2003; Takahashi et al., 2005; Takahashi et al., 2006)	
	Tall fescue	(Wang et al., 2003; Wang and Ge, 2005);	
Hygromycin tolerance	Perennial ryegrass	(van der Maas et al., 1994);	
	Italian ryegrass	(Bettany et al., 2003; Takahashi et al., 2005; Takahashi et al., 2006)	
Crown rust resistance	Italian ryegrass	(Takahashi et al., 2010)	
Herbicide resistance	Tall fescue	(Lee et al., 2008; Sato et al., 2013)	
Cold tolerance	Tall fescue	(Hu et al., 2005)	
Altered nutrition	Tall fescue	(Wang et al., 2001)	
	Perennial ryegrass	(Ye et al., 2001)	
	Italian ryegrass	(Hisano et al., 2004)	
Decreased lignin levels	Tall fescue	(Chen et al., 2003; Chen et al., 2004)	
Brown patch resistant/grey leaf spot resistant	Tall fescue	(Dong et al., 2008b)	
Bispyribac-sodium tolerance	Tall fescue	(Sato et al., 2009)	
Abiotic stress tolerance	Tall fescue	(Lee et al., 2012)	
Heat stress tolerance	Tall fescue	(Kim et al., 2012; Wang et al., 2017)	
Heat tolerance/methyl viologen tolerance	Tall fescue	(Kim et al., 2010)	
	Tall fescue	(Zhao et al., 2007b; Ma et al., 2014)	
Salt tolerance	Perennial ryegrass	(Cen et al., 2016)	
	Italian ryegrass	(Takahashi et al., 2010)	
Drought tolorance	Tall fescue	(Zhao et al., 2007a)	
Drought tolerance	Perennial ryegrass	(Patel et al., 2015)	
Drought tolerance/increased salt tolerance	Perennial ryegrass	(Wu et al., 2005)	
	Perennial ryegrass	(Bhalla et al., 2001; Petrovska et al., 2005)	
Reduced pollen allergen	Italian ryegrass	(Bhalla et al., 2001; Petrovska et al., 2005)	

Trait	Plant species	Reference	
Disease resistance	Tall fescue	(Dong et al., 2007; Dong et al., 2008a; Zhou et al., 2016)	
	Perennial ryegrass	(Xu et al., 2001)	
Increased metabolisable energy content	Perennial ryegrass (Beechey-Gradwell et al., 2022a)		
Altered senescence	Perennial ryegrass	(Li et al., 2004)	
	Tall fescue	(Buanafina et al., 2010; Sato et al., 2018)	
Enhanced digestibility Perennial F	Perennial Ryegrass	(Faville et al., 2010; Tu et al., 2010; Badenhorst et al., 2018)	
Pest resistance	Perennial Ryegrass	(Wu et al., 2012)	
Atrazine degradation	Perennial Ryegrass	(Vail et al., 2015)	

Another breeding goal includes reducing toxicity of endophyte infected material through genetic manipulation of the host or endophyte or both (Oram and Lodge, 2003).

SECTION 3 MORPHOLOGY

3.1 Plant morphology

Lamp et al (2001) describe the vegetative morphology of Italian ryegrass, perennial ryegrass and tall fescue as follows:

"Italian ryegrass is annual to biennial usually, but cultivars that may persist for more than two years have been developed. **Leaf blades** green to dark green, hairless, flat, upper surface evenly ribbed, lower surface smooth and shiny. Length up to 40 cm, width 5-12 mm. Young leaves are rolled in the bud. **Auricles** small and narrow. **Ligule** white, translucent, shorter than wide. **Leaf sheath** hairless with fine longitudinal ribs as in leaf blades, rounder at back.

Perennial ryegrass is a tussock-forming perennial with a fibrous root system, up to 60 cm high. **Leaf blades** dark green, hairless, flat, upper surface evenly ribbed, lower surface smooth and shiny. Length up to 30 cm, width up to 7 mm. Young leaves usually folded in the bud (V-shaped cross-section) but occasionally rolled (spiral cross-section), particularly in young plants. **Auricles** small and narrow. **Ligule** white, translucent, shorter than wide. **Leaf** purple.

Tall fescue is a perennial tussock-forming grass with a fibrous root system, grows up to 2 m high. **Leaf blade** 10-60 cm long, usually 3-10 mm but can be up 15 mm, green, hairless except for a few hairs on and near the auricle, pronounced longitudinal grooves on upper surface, lower surface smooth and glossy, margins rough to touch when fingers are moved down the margins. Young leaves rolled in the bud. **Auricles** with a few hairs on them. **Ligule** membranous and very short. **Leaf sheath** hairless, rounded at back, may be smooth or rough. Mainly green but can be red to brownish purple at base."

3.2 Reproductive morphology

Lamp et al (2001) describe the reproductive morphology of Italian ryegrass, perennial ryegrass and tall fescue as follows:



Italian ryegrass (Lolium multiflorum Lam.)

From: USDA-NRCS PLANTS Database / Hitchcock, A.S. (rev. A. Chase). 1950. *Manual of the grasses of the United States*. USDA Miscellaneous Publication No. 200. Washington, DC. (USDA, 2006a).

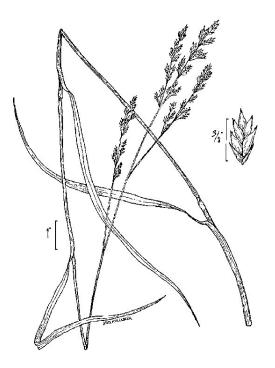
"Italian ryegrass: **Inflorescence** a spike up to 30 cm in length. The spikelets edge-on to the rachis (cf. *Agropyron*), in which the spikelets are side-on to the rachis. **Rachis** is recessed opposite each spikelet, which more or less fits into the recess. **Spikelets** consist of 10-20 florets laterally flattened, green, 15-25 mm long. **Glume** – 1 per spikelet, which is in the axil of the glume, lanceolate, about 10 mm long, outer surface ribbed like the upper surface of the blade, 5 nerved, covers less than the lower half of the spikelet. **Lemma** lanceolate, 5-8 mm long, 5 nerved. **Awn** nearly terminal, fine, straight, about 10 mm long. **Palea** similar to lemma in shape and size, 2 nerves with tiny hairs along them. **Anthers** 3, yellow or purple" (Lamp et al., 2001).



Perennial ryegrass (Lolium perenne L.)

From :USDA-NRCS PLANTS Database / Britton, N.L., and A. Brown. 1913. An *illustrated flora of the northern United States, Canada and the British Possessions*. Vol. 1: 281 (USDA, 2006a).

"Perennial ryegrass: **Inflorescence** a spike up to 20 cm in length; the spikelets are edge-on to the rachis. Rachis is recessed opposite each spikelet, which more or less fits into the recess. **Spikelet** usually 7-9 florets per spikelet, laterally flattened, green, 10-15 mm long. **Glume** 1 per spikelet, which is in the axil of the glume, terminal spikelet has 2, lanceolate, 7-10 mm long, outer surface ribbed like the upper surface of the leaf blade, 5 nerved, covers approximately the lower half of the spikelet. **Lemma** lanceolate, about 5 mm long, 5 nerved. **Palea** similar to lemma in shape and size, 2 nerves with tiny hairs along them. **Anthers** 3, yellow" (Lamp et al., 2001).



Tall fescue (Lolium arundinaceum)

From: USDA-NRCS PLANTS Database / USDA NRCS. *Wetland flora: Field office illustrated guide to plant species*. USDA Natural Resources Conservation Service (USDA, 2006a).

"Tall fescue: **Inflorescence** an open panicle, 10-20 cm long, erect or nodding, green or purplish. **Spikelets** contain 4-8 florets. Shape elliptic to oblong, 8-18 mm long, flattened laterally. **Glumes** usually unequal, pointed, keeled. Lower glume wider, lanceolate to lanceolate oblong, 4.5-7 mm long. Lower glume 1 nerved, upper 37 nerved. Upper glume about one-third as long as spikelet. No awns. **Lemma** 6-9 mm long, rounded on back, lanceolate to lanceolate oblong, 5 nerves. May or may not have a short terminal awn. **Palea** about as long as lemma, rough keels, 3 nerves. **Anthers** 3" (Lamp et al., 2001).

SECTION 4 DEVELOPMENT

4.1 Reproduction

Grasses can reproduce both sexually and vegetatively. Italian ryegrass, perennial ryegrass and tall fescue are all wind pollinated out-crossers. The main mode of reproduction is by seed, but tall fescue and perennial ryegrass can be maintained vegetatively in the sward for many years as some cultivars have short rhizomes (USDA, 2006a). This occurs rarely in turf and is more typical on spaced plants in sandy soils (Stewart, 2002; Meyer and Watkins, 2003). Grasses can also spread by tillering (Najda, 2004; USDA, 2006a) – see Section 4.1.1 below for further discussion. In intensely managed turf or pasture, where the grass is mowed short or grazed regularly, flower heads are removed preventing sexual reproduction, so reproduction may sometimes be entirely vegetative (Grime, 1979). Italian ryegrass regenerates entirely by seed (Alaska Natural Heritage Program, 2005).

4.1.1 Asexual reproduction

Grasses classified as 'annuals' complete their growth cycle in a single growing season and reproduce only by seed whereas those classified as 'perennials' reproduce vegetatively as well as by seed. Most of the commonly grown forage grasses, even if they complete their growth cycle in a single season (e.g. some cultivars of Italian ryegrass– see Appendix 1), function as perennials because they can reproduce vegetatively (Alaska Natural Heritage Program, 2005).

There are three types of growth habit that allow perennials to spread vegetatively and persist: bunch (also tufted or tussock-forming), stoloniferous, and rhizomatous (Langer and Hill, 1991). A stolon is a prostrate, above-ground stem that arises from a parent plant and bears nodes from which self-sustaining plants with roots can develop even if the physical connection with the parent is broken (Crampton, 1974). A rhizome is very similar except that it is defined as an underground stem. All three types of growth are dependent on the development of tillers, the basic unit of grass structure. A tiller is a shoot that arises from an axillary bud within a leaf sheath and can develop its own root system to effectively become a separate plant (Oregon State University, 2017). In bunchgrasses, the tillers grow vertically (intravaginal branching) either from the crown or from above-ground nodes (aerial tillers) whereas in the formation of stolons or rhizomes, the tillers grow out horizontally (extravaginal branching) (Oregon State University, 2017). Note, however that the vertical stems produced at the nodes of stolons and rhizomes can, themselves produce vertical tillers. While many of the species with stolons or rhizomes are classed as sod-forming, the extension growth of structures produced by extravaginal branching in some species can be very limited so that the species is classed as being a bunchgrass (see discussion of perennial ryegrass, below).

Bunchgrasses have minimal lateral spreading compared with the sod-forming grasses and ultimately it is seed that allows significant spread of the species. However, they may form dense clumps if they tiller extensively and the true perennials may live as long as 100 years with the centre dying out leaving an outer ring of active growth (Crampton, 1974). Crown derived tillers that become partially separated from the rest of the clump (for example, as a result of hoof damage) are able to strike more roots and survive independently (Langer and Hill, 1991).

Sod-forming grasses could potentially live indefinitely by continual vegetative reproduction and, in many cases have a reduced seed-forming ability (Crampton, 1974). They can usually recover quickly from excessive grazing, trampling or mowing and form a uniform cover of foliage and stems. For example, trampling can promote the spread of perennial ryegrass by pushing tillers apart and burying them so that they spread underground and then produce new tillers where they surface (Matthew et al., 1989).

Perennial ryegrass is classed as a bunchgrass (Thorogood, 2003; Oregon State University, 2017). The survival from one season to the next (perennation) and lateral spread, albeit limited, in pastures depends mainly on the production of upright tillers. However, the

species can produce stolons and this may account for the tendency of perennial ryegrass to dominate in heavily grazed pastures (Waller and Sale, 2001). The propensity for stoloniferous development is linked to cultivar genotype as well as to environmental factors such as soil type, degree of shading and grazing pressure (Donaghy, 2001). The species has also been described as producing short rhizomes from which plants can resprout quickly following fire (Sullivan, 1992).

Italian ryegrass is a bunchgrass that is only spread by seed (Carey, 1995). However, it tillers profusely and can therefore be a persistent pasture species in those climates which support a biennial growth cycle.

Tall fescue is classed by some as a sod-forming grass with short rhizomes (Oregon State University, 2017) and by others as a tufted bunchgrass that may or may not have rhizomes (Meyer and Watkins, 2003). In pasture cultivars, most rhizome growth occurs after the second Autumn post-sowing (Milne, 2009). While the rhizomes of pasture tall fescue do not spread as far as those of other species, their activity can be high in flood irrigated environments and is one reason why tall fescue persists better in this environment relative to perennial ryegrass (Milne, 2009). Recently there has been the development of the cultivar RTF ™ (Rhizomatous Tall Fescue) with an extensive rhizomatous habit that makes the cultivar more appealing to the turfgrass industry than conventional fescues because of its ability to fill in bare areas and to self-repair. With regard to tiller formation and persistence, the rate of new tiller formation in tall fescue is about one-third the rate of perennial ryegrass, but tall fescue tillers survive three times longer and are much larger (Burnett, 2006a; Milne, 2009).

4.1.2 Sexual reproduction

All three grass species reproduce sexually by producing seed, although this may not be the main form of reproduction in mown turf or heavily grazed pastures (Section 4.1.1). They produce an inflorescence in the form of a spike or panicle (see Section 3.2). In the Kangaroo Valley cv of perennial ryegrass, it takes approximately 132 days from seedling emergence to spike emergence, and then a further 16 days for anthesis to occur when planted in NSW (Shah et al., 1990). In Melbourne, grass pollen was detected from August to May, with the peak from November to January when most grass plants were flowering (Smart and Knox, 1979). The pollen is spread by wind.

In perennial ryegrass when the florets are mature the lodicules at the base of the floret swell with cell sap and force open the palea and lemma. The anthers of the stamens are extended on long filaments. The anthers split lengthwise from the tip to release clouds of pollen. At the same time the feathery stigmas project on either side of the floret ready to receive pollen. The basal older florets of the midspike flower first and then progresses toward the outermost floret and basal and apical spikelets (Thorogood, 2003).

Genetics of reproduction

All three grass species are self-incompatible. There has been debate about the number of loci controlling the self-incompatibility in perennial ryegrass (Spoor, 1976; McCraw and

Spoor, 1983). However, the presence of a two-locus (SZ) multiallelic gametophytic incompatibility system, which prevents self-seed setting and inbreeding depression is now generally accepted (Cornish et al., 1979; Fearon et al., 1983). Despite the presence of this self-incompatibility system, perennial ryegrass will set seed when selfed (Spoor, 1976; Thorogood, 2003). In general, the number of seeds set on selfing plants is considerably lower than in crosses, self-seed setting has been reported to vary from 2.2% to 32.3% and 100% in an inbred line (Jenkin, 1931; Jenkin and Thomas, 1938; Beddows et al., 1962; Foster and Wright, 1970; Thorogood and Hayward, 1991; Meyer and Watkins, 2003). Italian ryegrass possesses a similar 2 locus (SZ) incompatibility system which may be overcome to produce self-pollinated progeny (Fearon et al. 1983). As in perennial ryegrass, self-fertilisation is prevented when both the S and Z alleles present in the pollen are matched in the style.

Photoperiod and/or vernalisation requirements

Flowering in most temperate perennial grasses requires dual induction. The primary induction is brought about by low temperature or short days (acting independently or in combination), while the change to long days and higher temperatures are usually needed for secondary induction (Cooper and Calder, 1963; Heide, 1994; Meyer and Watkins, 2003; Thorogood, 2003). The primary induction enables initiation of inflorescence primordial and the secondary induction causes culm elongation, inflorescence development and anthesis (Heide, 1994). In some species such as perennial ryegrass it is only after the switch to secondary induction that floral initiation begins.

Perennial ryegrass has the most extreme vernalisation requirement of the three species. Little seeding in perennial ryegrass (cv. Yatsyn) in the subtropics is seen because of the lack of vernalisation (F. Wilson pers comm. in Lowe et al., 1999a), whereas Italian ryegrass and tall fescue seeded profusely in late spring and early summer.

Perennial ryegrass has an obligate vernalisation requirement of at least two weeks at less than 4°C before the inflorescence development will initiate (Cooper, 1957; Fejer, 1960; Cooper and Calder, 1963), although under certain conditions short day treatment may eliminate the requirement for a period at low temperatures (Evans, 1960). The requirement for primary induction of perennial ryegrass increases with the latitude of origin of the germplasm (Aamlid et al., 2000). In some perennial ryegrass varieties high temperatures can substitute for long days for secondary induction (Aamlid et al., 2000). Vernalisation is possible in the embryo, seedling or mature plant (Cooper and Calder, 1963).

In *Lolium*, the differences in inductive requirements are clearly related to past climatic and agronomic selections (Cooper, 1960). Separate plants of the same genotypes adjust to their surroundings and as such may flower at different times. The flowering period of the species also varies with location, although the anthesis period (the time of day during which pollen shedding begins and ends) is indicative of the species. Westerwolds ryegrass (*Lolium multiflorum*) which has been selected as a summer-annual catchcrop shows no inductive requirements, while in the perennial Italian varieties an obligatory vernalisation requirement

prevents tillers formed during mid-late summer from flowering and ensures an overlapping succession of vegetative tillers from year to year (Cooper, 1960).

Extreme primary induction requirements are found in the genus *Festuca*, including in tall fescue (Heide, 1994). In the UK, few tillers flower in the calendar year that they are produced (Gibson and Newman, 2001). Tall fescue is predominately self-sterile. The flowers are hermaphrodite, homogamous and wind-pollinated (Gibson and Newman, 2001). Tall fescue needs vernalisation to flower, then day lengths greater than 12 hours (Hannaway et al., 2004).

Grass species allocate different resources to flowering. Reproductive allocation (RA), defined as weight of reproductive structures as a proportion of total above-ground biomass was measured as 7.6% for tall fescue and 18.7% for perennial ryegrass. It was shown for 40 grass species to be negatively correlated with potential maximum height (Wilson and Thompson, 1989).

4.2 Pollination and pollen dispersal

Pollen

Plants (within and among species) can vary substantially in floral fertility, number of panicles, amount of pollen and quantity of seed produced. Tall fescue plants shed pollen in the early to mid-afternoon hours (Meyer and Watkins, 2003) and perennial ryegrass anthesis occurs once daily around midday and is more profuse on warm, bright days (Thorogood et al., 2002). In the UK, perennial ryegrass pollen was released at 0500-0600 and 1100-1300 except on dull days when anthesis was suppressed. In Melbourne, a bimodal release was also seen with a major peak between 1400-1800h and a minor peak between 0600-1000h (Smart and Knox, 1979).

The pollen production of perennial ryegrass in Victorian pasture has been estimated as 5.4×10^3 pollen grains per anther, 23.0×10^5 pollen grains per spike and 2.11×10^{13} pollen grains per hectare (Smart et al., 1979). This gives an estimate that 1 hectare of perennial ryegrass pasture could produce 464 kg of pollen per season. The estimate for the amount of pollen from perennial ryegrass on roadside verges is 10 fold lower at 48 kg per hectare per season due to competition from other plants (Smart et al., 1979).

Pacini et al. (1997) stated that tall fescue pollen could survive in open air for 48 hours but was completely non-viable 72 hours after opening of anthers. However, Wang et al. (2004a) found that pollen could survive up to 22 hours under controlled conditions in a growth chamber, whereas under sunny atmospheric conditions, viability was reduced to 5% in 30 minutes with complete loss of viability in 1½ hours. Under cloudy conditions pollen remained viable for up to 4 hours, with about 5% viability after 2½ hours. When pollen viability was lower than 5% no seed set could be obtained. Relatively high temperatures (36°C and 40°C) and high doses of radiation reduced pollen viability, while humidity did not. Pollen viability data is sparse for perennial ryegrass, with a report showing pollen to be viable for one day (Pfundt, 1910), and another showing 33% of perennial ryegrass flowers

could set seed when pollinated with 24 hour old pollen, using pollen stored in vials in the cold and dark (Gregor, 1928).

Temperature (in the range 14-26°C) also affects the growth of pollen tubes in perennial ryegrass with higher temperatures giving better pollination (Elgersma et al., 1989).

Pollination

As grasses are mainly wind pollinated, some of the following factors are expected to influence pollination levels, including:

- plant factors such as timing of flowering of pollen donors and receptor plants, level of pollen production (higher in cultivars with high levels of sexual reproduction), plant height, form, size, number of panicles, position of flowers and pollen weight and density
- 2) climatic conditions such as wind speed, direction and humidity
- ecological factors such as distance between the donor and acceptor plants (isolated plants are more likely to hybridise with pollen from a distant source than individual plants growing in groups), density of the donor plants, and geographical and/or vegetative barriers
- 4) genetic factors such as ploidy level and genetic compatibility

Source: (Rognli et al., 2000; Johnson et al., 2006).

As Italian ryegrass, perennial ryegrass and tall fescue are highly outcrossing, wind-pollinated species, extensive gene flow can occur. Both pollen shedding profiles and pollen viability data is lacking, yet necessary to estimate the possible gene flow.

In trials with perennial ryegrass, although pollen deposition declined with distance up to 80 m, there was considerable variation in dispersal of pollen over time (early to late anthesis) as well as in traps of various orientations (forward, backward and upward) (Giddings et al., 1997b, a). Further trials with perennial ryegrass showed that the amount of pollen deposited does not always decrease smoothly with increasing distance from the source. It is suggested that pollen clouds are taken high up into the atmosphere, move with weather and are deposited in times of calm weather, so it is therefore conceivable that pollen could move significant distances from the source. Both wind speed and turbulence are expected to be factors (among others) in this process (Giddings et al., 1997b). However, it is unknown whether the pollen collected in pollen traps is viable (Wang et al., 2004b).

A study in Australia (Cunliffe et al., 2004) showed that gene flow² in perennial ryegrass had a leptokurtic distribution with high gene flow close to the source which declines to a horizontal asymptote at 36 m. Beyond this, levels decreased from <5% at 36 m to <2% at 144 m (the limit of testing distance), depending on wind direction. In this study, the recipient

² Gene flow was assessed by measuring seed production as relative fertility – the fertility at each recipient position expressed as a percentage of fertility of centrally located recipient plants.

plants were isolated from any sexually compatible pollen that could compete with the pollen from the source plants. These results are supported by a more recent study of *L. perenne* in Argentina, which examined pollen-mediated gene flow from glyphosate resistant plants to receptor plants that were glyphosate susceptible. Effective gene flow was detected in receptor plants that were less than 35 m from the source plants, while no gene flow was detected in receptor plants that were greater than 35 m from the source plants (Yanniccari et al. 2018).

Studies on pollen flow from fields of Italian ryegrass to adjacent fields of perennial ryegrass in the United States have shown that little outcrossing occurred beyond 6 m from the field border in perennial ryegrass (Copeland and Hardin, 1970). Pollen-mediated gene flow between Italian ryegrass (source plants) and perennial ryegrass (receptor plants) was shown to decrease exponentially with increased distance from the pollen source at distances of up to 32 m in field studies in Ireland. This study also found hybridisation at stochastic low levels at distances between 64 m and 192 m (Mullins et al 2009).

In GM tall fescue gene flow experiments transgenes were detected at 50 m and 100 m with frequencies of 0.29-0.88%. The highest frequencies (0.88% at 50 m and 0.58% at 100 m) were in the direction of the prevailing wind (Wang et al., 2004b). In a further study, tall fescue transgenes were detected in recipient plants at up to 150 m from the central plot. The highest frequencies (5% at 50 m, 4.12% at 100 m and 0.96% at 150 m) were recorded in the prevailing wind direction. No transgenes were found at 200 m in any direction (Wang et al., 2004c).

In the USA, the isolation standard required by USDA for foundation seed of cross-pollinated grasses is 274 m (Code of Federal Regulations (USA), 2016), and the allowed level of seed contamination of other species is 0.1% (Montana Seed Growers Association, 2008). In South Australia the isolation distance for basic seed is 200 m from other grasses if the area is less than 2 ha, or 100 m if greater than 2 ha (see Figure 1), and the basic seed must be 99% pure at minimum (Seed Services Australia, 2020).

4.3 Fruit/seed development and seed dispersal

Development of seed

The seed of grasses is more correctly called the caryopsis and it is technically a fruit (Langer and Hill, 1991). It consists of the endosperm which is surrounded by the aleurone layer then the fused testa (seed coat) and pericarp (fruit wall). The scutellum separates the embryo, which comprises the radical, enclosed in the coleorhiza and the plumule surrounded by the coleoptile. The dry grass seed normally contains 14% water. Tall fescue seed are mature 29-30 days after anthesis at which time the endosperm is hard and flinty (Gibson and Newman, 2001). Seeds of the three species range from 2.6-4 mm long and 0.7-1.7 mm wide (Weiller et al., 1995).

Due to factors such as plant breeding, seed production between different cultivars of Italian ryegrass, perennial ryegrass and tall fescue can vary greatly (Table 6). Generally, seed yield

of perennial ryegrass is low and unpredictable (Elgersma et al., 1989), and inversely correlated with biomass yield (Studer et al., 2008, and references therein). However, estimated seed production in a NSW study was 14,040 seeds m⁻² for perennial ryegrass and 9,740 seeds m⁻² for tall fescue (Lodge, 2004), and perennial ryegrass yielded 35,000-160,000 seeds m⁻² (Hampton and Hebblethwaite, 1983). New Zealand seed production guidance suggests a seed yield of 150 g m⁻² (Brown et al., 1990). In the United States, Italian ryegrass cultivars gave numbers of pure live seed (PLS) of between 675-1,289 m⁻² (Venuto et al., 2002).

Perennial ryegrass and Italian ryegrass differ in several traits related to seed yield. Often, Italian ryegrass spikes are larger than perennial ryegrass, and have more spikelets and florets per spikelet and larger seeds and awns. The greater number of spikelets and florets, and larger seeds result in higher seed yield in Italian ryegrass (Brown et al., 2010). Italian ryegrass plants are very adaptable to different weed densities and exhibit high seed production potential (4,598 – 7,196 seeds plant⁻¹) from low density populations (Alarcon-Reverte and Moss, 2007). Perennial ryegrass has an average seed yield 350 – 1,000 kg ha⁻¹ (FAO, 2006).

	Tall fescue	Perennial ryegrass	Italian ryegrass
Seed production	100 (Cole and Johnston, 2006)	150-180 (Cole and Johnston,	
	100.2 ± 5.51 (cv Safari) (Fairey and Lefkovitch, 2001)	2006) 76-183 (Elgersma, 1990)	
	117.1 ± 6.39 (cv Tomahawk)	150 (Brown et al., 1990)	
	(Fairey and Lefkovitch, 2001)	35-100 (FAO, 2006)	
Mean	172.7 ± 11.26 (cv Safari) (Fairey	80 (cv Bulgarian) (Stoeva, 2005)	
seed/panicle	and Lefkovitch, 2001)	81.1-97.4 (Elgersma, 1990)	
(mg)	135.3 ± 8.86 (cv Tomahawk) (Fairey and Lefkovitch, 2001)		
	160 (cv Albena) (Stoeva, 2005)		
	182 (cv Elena) (Stoeva, 2005)		
1000 seed weight (g)	2.07 ± 0.077 (cv Safari) 1.87 ±	1.739 (cv Taya) (Larsen and Andreasen, 2004)	Multiflorum
	0.070 (cv Tomahawk)		diploid varieties: 2.0 - 2.5 tetraploid varieties: 3.0 - 4.6 (FAO, 2008)
	1.8 - 2.5 (FAO, 2008)	diploids: 1.3 - 2.7 tetraploids: 2.0 - 4.0 (FAO, 2008)	
	1.2 (cv Albena) (Stoeva, 2005)		
	2.3 (cv Elena) (Stoeva, 2005)	1.8 (cv Bulgarian) (Stoeva, 2005)	Westerwold
		1.780 ± 0.272 (Thompson et al., 1993)	diploid varieties: 2.5 - 3.0 tetraploid varieties: 3.7 - 5.1 (FAO, 2008)
		2.5-4.3 (Hill et al., 1985)	
		1.32-1.46 (Elgersma, 1990)	

Table 6. Seed produc	tion in three g	rass species
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Seed dispersal

Grass seeds are capable of germination after passing through the digestive systems of grazing animals such as cattle and sheep (Yamada and Kawaguchi, 1972; Yamada et al.,

1972; Johns and Greenup, 1976; Janzen, 1984; Chambers and MacMahon, 1994; Hulme, 1994; Fischer et al., 1996) or horses (Campbell and Gibson, 2001). The potential of cattle to disperse the seeds of perennial ryegrass, Italian ryegrass and tall fescue has been assessed in two feeding studies (Yamada & Kawaguchi 1972; Yamada et al. 1972). Seeds of the three species could be recovered from faeces 12-24 hours after feeding. Viable seeds were recovered for all species and seedlings started to emerge after one week (Yamada & Kawaguchi 1972; Yamada et al. 1972). In a study of seed dispersal by sheep, seeds of Italian and perennial ryegrass were transported in the wool of grazing sheep, and in the case of perennial ryegrass the seeds remained in the wool for 1-2 months (Fischer et al., 1996). A further study of seed dispersal after ingestion by goats reported that 1.6% of perennial ryegrass seeds remained viable after digestion, and 0.4% were able to form seedlings (Harrington et al., 2011); the seeds were completely excreted by 48 h post ingestion.

In the UK, a study in grassland showed no significant predation of grass seed by molluscs or arthropods, but did show removal by rodents (Hulme, 1994), although this was reduced by seed burial. Perennial ryegrass seed may be harvested and transported by ants (Campbell, 1966; McGowan, 1969). Viable Italian ryegrass seeds have been found in the faeces of the European hare with the authors concluding that hares could be a means of seed dispersal (Vignolio and Fernández, 2006). In the UK, geese and buntings have been shown to graze on the seeds of Lolium spp. (Patton and Frame, 1981; Buckingham et al., 2011). In a small, contained feeding study of bird species that have been identified as horticultural pests in Australia, 0.03% of perennial ryegrass seeds were excreted by corellas, 0% by galahs, and up to 0.4% by house sparrows (Woodgate et al., 2011). However, as only small amounts of perennial ryegrass seeds were consumed by the studied bird species, the germination potential of the excreted seeds could not be determined due to the insufficient number of seeds excreted. No specific literature was available on seed dispersal by birds through exozoochory (adhesion to the exterior of the animal), however perennial ryegrass seeds lack structures to enhance dispersal by this method (e.g. hooks). Seed dispersal in irrigation water has been observed for Lolium spp. in Chile, with germinable seeds recovered from the irrigation water (Tosso et al., 1986). Human activity is also a likely source of seed dispersal with perennial ryegrass seed transported on cars (Hodkinson and Thompson, 1997), or spilled during seed transport in the agricultural supply chain.

The external factors described above are responsible for any long-distance seed dispersal from ryegrasses. For perennial ryegrass, most seeds fall adjacent to the parent plant (DiTomaso and Healy, 2007) and lack structures to enhance their dispersal by wind or water. Shattering seed heads may aid in short distance dispersal, although the number of seeds spread by shattering depends on the genotype and abiotic conditions (Elgersma et al., 1988; Fu et al., 2019).

4.4 Seed dormancy and germination

Both seed size and shape can affect the presence of seeds in the soil seed bank. This is probably related to the ease of burial of the seed. The common mechanisms of burial (such

as penetrating cracks in soil, being washed into soil by rain, ingestion by earthworms) will operate more effectively with small, compact seeds. Earthworms can also ingest seeds in the seed bank and bring them to the surface where they can germinate (Thompson et al., 1987). Earthworms have been shown to predate perennial ryegrass seeds and seedlings (Eisenhauer et al., 2010). A study in northern NSW (Armidale and Tamworth) indicated that ant predation of tall fescue was significant, but did not examine if this predation affected germination (Johns and Greenup, 1976). Germination requirements, dormancy mechanisms and resistance to pathogens also contribute to persistence in soil (Thompson et al., 1993).

Perennial and Italian ryegrass both form transient type I seed banks where seed banks are present in summer; a transient type I seed bank enables a species to take advantage of seasonal gaps in vegetation cover (Thompson and Grime, 1979). This is related to their large seed, lack of pronounced dormancy mechanisms, ability to germinate in a range of temperatures or in light and dark (Thompson and Grime, 1979). However, Italian ryegrass has been shown to have an ability to germinate at higher temperatures in the light/dark condition than in constant darkness (Ichihara et al., 2009).

It is likely that the length of dormancy could vary widely among and within cultivars and even among and between individual plants of the same cultivar due to genetic and environmental factors. The environment in which seeds develop on the parent plant often plays a role in determining dormancy status. Temperature, water supply, shading, day length and nutrient supply are the main factors attributed to modifying the proportion of seeds exhibiting dormancy in a number of plant species. For example, Italian ryegrass seeds become larger and more dormant as temperature is reduced from 27°C to 15°C, and tall fescue seeds respond similarly (Steadman et al., 2004).

A study in NSW of tall fescue and perennial ryegrass indicated that 14 months after seed production the seed bank contained 14% of the perennial ryegrass and 10% of the tall fescue seed that was naturally dispersed in the field. Some seed was produced by volunteers in this study, therefore some of the seed recorded at 14 months may have been second generation seed. Viability of the seed bank was not tested. After 26 months no seed bank remained of either species (Lodge, 2004). Perennial ryegrass seeds were not found six years after burial in a seed bank study in the UK (Akinola et al., 1998). Another UK study found that in a permanent pasture perennial ryegrass comprised 7.5% vegetative cover and 0.6% of the transient seedbank during April prior to ryegrass seed set, but when seed set was prevented over the summer, no perennial ryegrass was found in the seed bank by October (Williams, 1984). Perennial ryegrass seed has been seen to persist in the soil for less than 5 years (Thompson et al., 1993). Field studies of Italian ryegrass seed persistence in Brazil found that 55% of *L. multiflorum* seeds germinated by 3 months after burial. Less than 6% of seeds were recovered from the seed bank 18 months after burial (Cechin et al., 2020).

Burial depth may also influence the persistence of viable seeds in the soil seed bank. A series of small plot studies were conducted in temperate conditions in Denmark that simulated the impact of a range of soil tillage practices on the germination of *L. perenne* seeds. Data from

several seasons showed that seeds left on the soil surface or buried up to 5 cm depth had survival rates of less than 1% after 12 months. When seeds were immediately buried at a depth of 25 cm, seed germination ranged from 0 to 16%, but if they remained at the surface for a period of time before burial at 25 cm, germination rates were typically less than 1% (Jensen 2010). Germination experiments with *L. perenne* seed in the glasshouse indicated that 90% of seed emerged at 1 cm seed burial depth, while no seed emerged at the soil surface or at depths of 6 - 7 cm (Javaid et al. 2022).

Italian ryegrass, perennial ryegrass and tall fescue germinate rapidly without pre-treatment as they all lack a physiological dormancy (Hill and Pearson, 1985; Lodge, 2004). One month after harvest mean germination rates of perennial ryegrass (cv. Kangaroo Valley) and tall fescue (cv. Demeter) seeds were 70.5% and 62.5%, respectively (Lodge, 2004). In two Italian ryegrass cultivars (Tribune and Lemtal) 50% of the seed germinated within three weeks of harvest (Hides et al., 1993). In comparison to other grasses, perennial ryegrass (cv. Derby) is relatively quick to germinate. In an Australian study in it took 2.8 days (in spring) to 6 days (in winter) for 50% of perennial ryegrass seeds to germinate in the field (Lush and Birkenhead, 1987).

Perennial ryegrass was superior in its ability to germinate under conditions of moisture stress compared to six other grass and legume species (McWilliam et al., 1970). In constant temperature experiments, temperatures within the range 5-30°C did not limit the germination of seeds of perennial ryegrass (cv. Victorian) (McWilliam et al., 1970). In addition, it has been reported that maximum germination occurred at 30/10°C (12 h/12 h) and high germination levels may also occur above 30°C for cv. Kangaroo Valley (Lodge, 2004).

At constant temperatures, germination of Italian ryegrass (cv. Grasslands Tama, Ucivex) and tall fescue seeds (cv. Demeter, AF5, AF6, Kenhy) was lowest at 15°C and 10°C compared to other temperatures tested (Hill et al., 1985). Germination of tall fescue and Italian ryegrass was also reduced at 35°C (or 30/25°C for cv. Grasslands Tama) (Hill et al., 1985). Maximum germination occurred at 30/20, 30/15 and 30/25°C (12 h/12 h) for tall fescue (Lodge, 2004). Optimum germination of perennial ryegrass seeds occurred over a wider range of temperatures (15/25, 20/25, 20/30 and 25/30°C), but varied between two cultivars (Shen et al., 2008). Soil temperatures of 15°C reduced seed germination but did not influence seed survival (Shen et al 2008). Recent glasshouse studies of perennial ryegrass found that 25°C was the optimal temperature for seed germination and seedling growth (Javaid et al 2022).

Germination rates for both Italian ryegrass and perennial ryegrass are inhibited by increasing salinity or alkalinity (Lin et al 2018). Javaid and co-authors (2022) also found that high salinity reduced seed germination rates of perennial ryegrass in glasshouse trials, which they suggested may be the result of reduced water uptake under high salt conditions.

In the United States, a study showed no germination of naturally dispersed tall fescue seed in undisturbed soil (Smith, 1989). In NSW, seedling emergence of naturally dispersed tall

fescue and perennial ryegrass occurred in May/June, with 31% of tall fescue plants and 73.8% of perennial ryegrass plants surviving until November and none until the following March (Lodge, 2004). However, under controlled conditions, seeds of tall fescue and Italian ryegrass maintained germinability for at least 12 months (10°C and 95% relative humidity) (Kulik and Justice, 1967) although after 5 years the percent germination of Italian ryegrass seed dropped off rapidly (Rutledge and McLendon, 1996).

4.5 Vegetative growth and dispersal

After imbibition the caryopsis swells slightly, the root emerges through the coleorhiza from the proximal end of the grain after 3-5 days, just before the coleoptile. The coleoptile is purplish in colour and grows up between the palea and lemma, splitting at the top when it has grown 5-10 mm to allow emergence of the first leaf (Gibson and Newman, 2001).

Early growth rates are dependent on the seed reserves, with growth rates during the first 20 days being related to the seed or caryopsis weight. This was also influenced by the perenniality, which gave slower than expected growth, and polyploidy, which increased growth rates (Hill et al., 1985). The number of live leaves per tiller is regarded as relatively constant. For perennial ryegrass a mean of 3 leaves per tiller was observed, with a slightly higher mean of 3.7 for tall fescue (Yang et al., 1998). Both ryegrass and tall fescue produce their first root below their last live leaf and have approximately twice the number of active roots on tiller axis than leaves as roots turnover more slowly than leaves (Yang et al., 1998).

Perennial ryegrass plants can be very long lasting (30+ years), depending on management and environmental factors. However, the effective life of a pasture is 5-10 years (Grassland Society of Southern Australia Inc, 2008b). The persistence of perennial ryegrass also varies between cultivars. For example, for cv. Yatsyn, only about 40% of the plants survived under grazing from one year to the next (Lowe et al., 1999a), similar to results seen from a sheepgrazed perennial ryegrass pasture in SW Vic (Waller et al., 1999). Tall fescue has been described as a long-lived perennial (10+ years) (Hannaway et al., 2004). Italian ryegrass cultivars are either annuals (Westerwolds types) or biennials (Grassland Society of Southern Australia Inc, 2008a). In the subtropics of Qld, tall fescue was the most persistent grass in irrigated pasture under grazing and Italian ryegrass the least persistent (Lowe et al., 1999a).

Perennial ryegrass tillers produced after reproductive growth in the spring form the basis of the plant population for the following year. Few tillers (<10%) survive for more than twelve months so the plant is dependent on tiller replacement for survival (Grassland Society of Southern Australia Inc, 2008b). Perennial ryegrass is known to form compact tussocks with large numbers of long leaf blades (Fustec et al., 2005).

Vegetative dispersal

Perennial ryegrass and tall fescue can form clones, with adventitious roots, from cut stem pieces kept in water. It was therefore assumed that weed control by cutting the stems of these plants could contribute to their dispersal (Uchida and Arasea, 2005). No literature is available on the likelihood of vegetative dispersal occurring in this manner under field

conditions. Tiller growth as a means of perennial ryegrass vegetative dispersal is also possible over short distances in pasture. The average distances of aerial tiller dispersal over the two year study were 4.5-4.8 cm and 3.6-4.2 cm. The maximum length of aerial tiller dispersal found in this study was 15 cm in the first year and 15.5 cm in the second year. While aerial tillering may not lead to long distance dispersal of perennial ryegrass it is important for maintaining the population (Sawada, 1991).

SECTION 5 BIOCHEMISTRY

Italian ryegrass, perennial ryegrass and tall fescue are not pathogens and not capable of causing disease in humans, animals or plants. However, they do contain endophytes which produce alkaloids that act as deterrents to insect herbivory but also affect utilisation as animal feed. Grass pollen are one of the most important airborne allergen sources worldwide and cause hay fever in many susceptible individuals.

5.1 Toxins

Throughout the world Italian ryegrass, perennial ryegrass and tall fescue are widely grown for turf and forage. In contrast to many plants that possess chemical defences to predation, grasses are not well defended. Most grasses have co-evolved with grazing animals and survive defoliation by their growth habit rather than intrinsic toxicity (Cheeke 1994). An examination of a number of toxic plant databases listed tall fescue (University of Purdue, 2006; Cornell University, 2015); and perennial ryegrass (University of Purdue, 2006; Food and Drug Administration, 2008) as toxic to animals. However, the Canadian Poisonous Plants Information System (Canadian Biodiversity Information Facility, 2017) did not list any of the three species as toxic. This inconsistency arises because the grasses alone are not ordinarily toxic (even though they produce their own alkaloids), but become so when in symbiosis with endophytic fungi, or when experiencing mineral deficiencies. However, in a review of plant toxicants which may be present in milk, the genus *Festuca* (which includes tall fescue) was listed as containing pyrrolizidine alkaloids which are known to be hepatotoxic and/or pneumotoxic in horses, cattle, sheep, goats, pigs (Panter and James, 1990), and alpacas (Sampaio et al., 2008).

Endophyte related diseases

Italian ryegrass, perennial ryegrass and tall fescue can all contain toxin-producing fungal endophytes (see details below and Section 5.4 for more details). The endophytes produce alkaloid toxins which can be harmful to livestock. In tall fescue and perennial ryegrass this is predominantly caused by the toxins ergovaline and lolitrem B. Ergovaline is a vasoconstrictor that can cause or exacerbate heat stress by constricting blood vessels as well as other symptoms (Strickland et al., 2009). For sheep and cattle an ergovaline concentration of between 0.75-1.25 mg/kg dry plant tissue can pose a risk if these grasses are the sole food (Harris and Lowien, 2003). For perennial ryegrass a lolitrem B concentration of between 1.5-2.5 mg/kg dry plant material can pose a risk. However, as a general rule if the hay or silage makes up <50% of total ration there is unlikely to be a problem (Kemp et al., 2007).

Ryegrass staggers (fescue toxicosis, summer slump)

Ryegrass staggers occurs in animals grazing pastures containing perennial ryegrass and tall fescue infected with endophytes (see Section 6.4). The main cause is Lolitrem B from perennial ryegrass. It is most concentrated in older tissues, the base of the plant and seed. The concentration increases under drought or with high soil nitrogen when the plant is water stressed (Reed, 1999b; Kemp et al., 2007). The highest incidence is in summer and autumn. Symptoms in affected animals can vary from tremors in mildly affected stock, to lack of coordination and collapse in more severe cases (Reed, 1999b). It can also cause slower weight gains, decreased milk production, poor appetite, retention of winter coat, reproductive problems, and elevated temperature (University of Purdue, 2006). Ryegrass staggers is not usually fatal, and animals usually recover unaided (Reed, 1999a), although mortality can occur due to misadventure such as affected animals falling over cliffs or into water (Cheeke, 1995).

Fescue foot and fat necrosis (dry gangrene)

Fescue foot and fat necrosis affect animals grazing pasture infected with endophytes. It is caused by ergovaline (Kemp et al., 2007) and is a problem generally associated with tall fescue, although it is theoretically possible that perennial ryegrass could produce this disorder (Kemp et al., 2007). 'Fescue foot' is a painful swelling of the fetlocks, which causes lameness. It tends to develop in the late autumn and winter, and the extremities (typically tail, ears, and rear feet) undergo necrosis (University of Purdue, 2006). Animals need to be removed from the fescue pasture otherwise gangrene can set in which may cause death of the animal (Lamp et al., 2001). Fat necrosis develops when lesions develop in fat inside the abdomen causing the animal to die (Lamp et al., 2001; University of Purdue, 2006). Both these conditions contribute to poor animal performance (resulting in low meat and milk production) (Lamp et al., 2001).

Ergot

Ergot is another fungal disease associated with pasture grasses, including perennial ryegrass and tall fescue. The disease is caused by a number of species of *Claviceps* depending on grass type (e.g. *Claviceps purpurea* is associated with perennial ryegrass and tall fescue), and is toxic to grazing animals and humans on consumption (Clarke, 1999a). Summer ill-thrift or winter lameness in livestock is associated with ingestion of ergot alkaloids (Harris and Lowien, 2003). Clinical signs include behavioural changes, swelling, lameness, abortions, convulsions, gangrene, and death. In sub-lethal cases, once the source of ergot is removed, recovery from neurologic signs is likely, but recovery from the vascular effects and gangrene is unlikely (University of Purdue, 2006).

Grass tetany

Cattle grazing on pastures which are low in magnesium, calcium and sodium, and high in potassium and nitrogen, are at risk of developing grass tetany. Most veterinary texts define 'grass tetany' as a deficiency of magnesium (Mg) and many different circumstances can

cause this condition to arise. Low levels of blood magnesium in cattle (hypomagnesaemia) are usually associated with low levels of blood calcium, particularly in pregnant cows (late in gestation) and those with calves. At these low levels muscle movement is restricted and breathing fails, often resulting in animal death. The disorder is prevalent on the northern, central and southern tablelands and slopes, but has also occurred elsewhere in Australia (Elliot, 1999).

5.2 Allergens

Grass pollen is one of the most important airborne allergen sources worldwide. Allergic sensitisation may affect as many as 20% of the general population and as much as 40% of atopic individuals. Symptoms of grass pollen allergy are most often distinctly seasonal and predominantly consist of rhinitis and conjunctivitis (Andersson and Lidholm, 2003). As many as 21 genera of grasses have been implicated in grass allergy, but the main contributor is perennial ryegrass due to its large pollen output. Italian ryegrass and tall fescue also produce prolific amounts of pollen and in certain environments and seasons can cause allergic rhinitis, or 'hay fever' (Mallett and Orchard, 2002a; Kmenta et al., 2017). In Australia, Australian Institute of Health and Welfare (2011) estimated that 15% of the population are affected by allergic rhinitis. Indeed, as many as 37% of individuals are immunoreactive to perennial ryegrass pollen in some populations (Scala et al., 2010). Further estimates suggest that in Melbourne a person may be exposed to 0.5-1.0 mg grass pollen during the 4 months of the pollen season (Smart et al., 1979; Scala et al., 2010), which, when combined with the observations that human respiratory intake of 1µg of grass pollen may be sufficient to cause symptoms of hay fever in susceptible individuals (Smart et al., 1979), indicates that grasspollen allergy is a significant health problem. In fact, these allergies are responsible for considerable public health and economic burdens in Australasia (Davies et al., 2015). Experience overseas estimates that 84% of USA residents are exposed to perennial ryegrass pollen (Lankow et al., 2015).

The allergens identified have been classified into a number of groups, with Groups 1 and 5 being the most prevalent and immunoreactive (Klein-Tebbe and Davies, 2014). About 90% of allergic individuals display IgE antibody reactivity to Group 1 allergens, and 55-85% to Group 5 antigens. In several species, these antigens have been shown to account for a considerable part of the specific IgE binding to pollen extract (Andersson and Lidholm, 2003).

Type 1 grass allergens are typically glycoproteins of approximately 30 kDa, which are released quickly from pollen upon hydration (Cosgrove et al., 1997). The group 5 pollen allergen, Lol plb, is located mainly on starch granules in the pollen grain so when the ryegrass pollen encounters water, the grains burst releasing approximately 1,000 starch granules from each pollen grain (Singh et al., 1991). The identification and characterisation of allergens is difficult due to the high degree of cross reactivity between different grass species (van Ree et al., 1998; Wissenbach et al., 1998), and the existence of isoallergens within a single group of allergens from one species (Wissenbach et al., 1998).

Many pollen allergens have been cloned and characterised, including several *Lolium* pollen allergens. The Structural Database of Allergenic Proteins (SDAP) lists a number of allergenic proteins from *L. perenne* including Lol p 1 (Perez et al., 1990; Griffith et al., 1991) (and isoallergens Lol p 1.0101, 1.102 and 1.0103), Lol p 2 (Ansari et al., 1989a; Sidoli et al., 1993), Lol p 3 (Ansari et al., 1989b), Lol p 5 (Singh et al., 1991; Klysner et al., 1992) (and isoallergens Lol p 5 1.0101 and 1.0102), and Lol p 11 (van Ree et al., 1995). Additional allergenic proteins, Lol p 4, Lol p 10 and another isoallergen for Lol p 5 (Lol p 5c), are listed in a review by Andersson and Lidholm (2003). Lol p 1 proteins are classed as Group 1 allergens. No allergenic proteins are listed for Italian ryegrass or tall fescue in comprehensive reviews (Andersson and Lidholm, 2003) or on SDAP (Ivanciuc et al., 2002; Ivanciuc et al., 2003). However, taking into account high sequence-level similarity of proteins from Italian and perennial ryegrasses, and reports of widespread antigenicity of Italian ryegrass pollen (Schäppi et al., 1999; Taketomi et al., 2006) it is reasonable to consider that Italian ryegrass pollen, along with the pollen of other related Poaceae, are also antigenic.

Besides the protein component of pollen, carbohydrate structures such as N-glycans are also thought to be an important part of allergenicity, particularly in relation to cross reactivity between different plant allergens. The most abundant N-glycans in perennial ryegrass pollen are those carrying both xylose and core α -1,3-linked fucose residues (such as MOXF3 and MMXF3) (Wilson and Altmann, 1998). (1 \rightarrow 3)- β -D-glucan has also been measured from perennial ryegrass pollen at approximately 600 ng/106 pollen grains, which was estimated to be a level sufficient to cause allergic reactions (Rylander et al., 1999).

5.3 Other undesirable phytochemicals

Some grass species have been thought to exhibit allelopathy. Allelopathy is the direct or indirect effect of one plant on another through the production of chemical compounds that escape into the environment (Rice, 1979). This is most pronounced in tall fescue.

The allelopathic potential of tall fescue has been documented and is thought to be the reason that it is difficult to establish legumes in tall fescue pastures (Smith and Martin, 1994). Aqueous and ethanol extracts of tall fescue leaf and stem tissue have been shown to exhibit allelopathic effects upon alfalfa, Italian ryegrass, rape (*Brassica rapa*), red clover (*Trifolium pratense L.*), ball clover (*T. nigrescens*), crimson clover (*T. incarnatum*) and birdsfoot trefoil (*Lotus corniculatus*) (Peters, 1968; Stephenson and Posler, 1988; Luu et al., 1989; Smith and Martin, 1994; Chung and Miller, 1995b; Springer, 1996; Applebee et al., 1999).

A wide range of organic acids in the tall fescue extracts were involved in the inhibition of birdsfoot trefoil, with seedling growth rates being more affected than germination (Luu et al., 1989). The inhibition has been shown to be dose-dependent and occurred in soil as well in the glasshouse (Stephenson and Posler, 1988). It varies between tall fescue genotypes (Peters and Mohammed Zam, 1981), and between seasons, with the greatest inhibition of birdsfoot trefoil growth occurring for extracts prepared in the autumn, when the tall fescue

plants were growing strongly (Stephenson and Posler, 1988). Increased carbon dioxide levels were seen to enhance the phytotoxicity of tall fescue to alfalfa (Applebee et al., 1999).

The allelopathy observed for tall fescue and perennial ryegrass is associated with the presence of an endophyte , but the effect size is smaller than the allelopathic effect of different grass species (Cripps et al., 2013). The presence of tall fescue inhibits seed germination and reduces seedling shoot and root lengths in clover, whether the tall fescue contains an endophyte or not. However, the presence of the endophyte in tall fescue did reduce the length and density of clover root hairs which might impact on its ability to take up nutrients and water (Springer, 1996). Summer-active cultivars of tall fescue are highly competitive with alfalfa, whereas summer-dormant were not, and the effect could be magnified by endophyte infection in some cultivars (Malinowski et al., 2011).

Italian ryegrass has also shown allelopathy (McKell et al 1969), particularly affecting clovers and medics (Chung and Miller, 1995b). Aqueous extracts of Italian ryegrass foliage inhibited the germination and seeding growth of alfalfa (Smith and Martin, 1994), although this effect is less severe than that observed with tall fescue extracts. No allelopathic response of wheat to Italian ryegrass was seen, but below-ground competition reduced wheat height, leaf number, tillering, area and dry weight (Stone et al., 1998).

There have been multiple reports of allelopathy from perennial ryegrass affecting other grasses and forage legumes such as zoysiagrass (Zuk and Fry, 2006), bermudagrass (McCarty et al., 2010), alfalfa (Chung and Miller, 1995a), nodding thistle (Nicholson et al., 1990), and white clover (Mattner and Parbery, 2001). Auto-allelopathy has also been observed between two populations of perennial ryegrass, where the growth of one population reduced the growth of the other in mixed cultures when the growth medium was not replaced (Kraus et al., 2002). Furthermore, aqueous extracts of perennial ryegrass were shown to inhibit growth of the same populations they were derived from (Newman and Rovira, 1975). In other species such as US prairie plants, no allelopathic effects of tall fescue extracts were observed (Renne et al., 2004).

5.4 Beneficial phytochemicals

Pasture Nutrition

Tall fescue has a high nutritive value compared to perennial ryegrass, although trials on dairy cows in southeast Qld showed similar average daily milk production and milk quality from perennial ryegrass and tall fescue pastures (Harris and Lowien, 2003).

The herbage yield and composition of pasture grasses are influenced by many environmental (e.g. climatic and weather conditions, soil type and nutrients in the soil, and fertilisation and cutting management) and genetic factors (e.g. forage type, species and cultivar) (Tas, 2006).

Generally, pasture is most palatable, most easily digested and most nutritive earlier in the season when leaves are young (Sullivan, 1992; Carey, 1995; Walsh, 1995). Although the nutritive value of pasture declines with age, total yields increase, which results in a conflict

between quality and yield. Taking this into consideration, early cutting of pasture for hay and silage (at heading emergence) is probably the most cost-effective way of producing feed for both production and maintenance of stock, despite the associated sacrifices in total yield (Andrews, 1997).

There is a tendency for leaves to become coarse, tough and unpalatable with age, making digestion more difficult for grazing animals. For example, in a perennial ryegrass study, *in vitro* digestibility was between 80-88% during the vegetative, early bloom and heading stages of growth but dropped to 71% when the grass was mature. Protein levels are also higher early in the season, or at immature stages of development. For example, in a tall fescue study conducted in the US, protein was 16.2% in May compared to 9.1% in July, and in perennial ryegrass crude protein levels dropped from 13.2% to 9.1% between the heading and mature stages of growth. In the same study, protein levels increased with increased applications of nitrogen fertiliser, by 8% and 5.8% in the vegetative and early stages of growth, respectively (Sullivan, 1992; Carey, 1995; Walsh, 1995; USDA, 2006b).

SECTION 6 ABIOTIC INTERACTIONS

Populations of Italian ryegrass, perennial ryegrass and tall fescue are variable in their response to the environment. Grasses in general are highly plastic and are capable of responding to fluctuating environments and stresses by morphological and physiological changes as to phenotype (Bradshaw, 1965). Therefore, it has been possible to breed many cultivars with improved adaptation to a wide range of environments and conditions (Lodge, 2004; Sampoux et al., 2011; Sampoux et al., 2013).

6.1 Nutrient requirements

The major nutrient deficiency that limits grass growth in soils in temperate regions of Australia is nitrogen. However, sowing pastures which contain a legume component reduces the need for applications of nitrogen fertilisers (Andrews, 1997). Depending on the use of the pasture, nutrient requirements vary. For example, the greatest losses via nutrient removal in animal products, from high productivity dairy pastures, is nitrogen and potassium, wool and dairy enterprises are similar in terms of sulfur losses, and beef and dairy enterprises are similar in their loss of phosphorus (Sale and Blair, 1997).

Nitrogen is important in turfgrass for healthy growth and colour, although excessive nitrogen can lead to shallow-rooted grass that is more susceptible to stresses such as drought and heat (Fagerness et al., 1998). Phosphorus is also important, although the fibrous root system of grasses is efficient at extracting phosphorus from the soil. The addition of potassium to turfgrass is important for improving stress tolerance. The micronutrient that is most often deficient in turf, especially in alkaline soils, is iron (Fagerness et al., 1998). Iron deficiency results in chlorosis or yellowing of foliage due to a lack of chlorophyll synthesis.

Italian ryegrass requires a highly fertile soil to perform well and responds well to applications of nitrogenous fertilisers (Lamp et al., 2001). It has a wide range of soil adaptability, being tolerant of acidic to alkaline soils (pH 5.0 to 7.8). Below pH 5.0, aluminium toxicity may be a

problem and higher pH can cause chlorosis due to iron and manganese deficiencies. The best growth occurs when soil pH is maintained between 5.5 and 7.5 (Hannaway et al., 1999).

Perennial ryegrass grows best on fertile, well-drained soils but has a wide range of soil adaptability, and tolerates both acidic and alkaline soils (pH range of 5.1 to 8.4; Thorogood, 2003). It responds well to applications of nitrogen (both as absolute N and N fixed by legumes) and phosphorus, and is moderately tolerant of acid soils although there is a sensitivity to Al concentration when soil pH is low (pH_{Ca} < 4.4) (see discussion in review by Waller and Sale, 2001). Nitrogen application has been proposed as a measure to improve persistence of perennial ryegrass in intensively grazed dairy pastures (Harris et al., 1996).

Tall fescue is adapted to a wide range of soils, including those that are acid or alkaline, saline, of low fertility, medium to heavy textured, or subject to waterlogging (Lamp et al., 2001; Meyer and Watkins, 2003; Lodge, 2004), but is more productive on highly fertile soils (eg basalt to fine granite; Harris and Lowien, 2003). It responds well to applications of phosphorous, nitrogen and sulphur. For example, tall fescue will grow on less fertile soils that are regularly fertilised to maintain phosphate and sulphur (Easton et al., 1994; Harris and Lowien, 2003). It grows better on alkaline, saline, wet and compacted soils than other cool season grasses. Optimum growth occurs at a pH of 5.5 to 6.5, but tall fescue can tolerate a pH range of 4.7 to 8.5 (Meyer and Watkins, 2003). Experiments have shown increased tillering (emergence, number of proportion of buds developing into tillers) following application of nitrogen fertiliser (Wilman and Pearse, 1984) and increased seed yield (Simpson and Bull, 1970).

6.2 Temperature requirements and tolerances

Italian ryegrass is characterised by rapid establishment (even in very wet conditions). Its growing season is autumn, winter and spring, and also summer if sufficient moisture is available. It is particularly valued for its winter growth which is higher than perennial ryegrass (Lamp et al., 2001). It was found to have the fastest primary (uncut) growth during establishment, and a broader tolerance of temperature than perennial ryegrass and prairie grass (*Bromus catharticus*) (Hill et al., 1985). In subtropical southern Qld where the hot, humid summers accelerate the decline in perennial ryegrass density, farmers plant Italian ryegrass (Callow et al., 2003). It can grow in a temperature range of 0-30°C with optimum growth occurring between 15-18°C. It is tolerant of cold winters (Grassland Society of Southern Australia Inc, 2008a).

Perennial ryegrass is less tolerant of heat than tall fescue, although there is variation between cultivars (Razmjoo et al., 1993). It is best adapted to cool, moist climates where winterkill is not a problem. During hot summers, perennial ryegrass becomes dormant and it will not tolerate climatic extremes of cold, heat, or drought. Optimum growth occurs between 20 and 25°C. Forage production suffers when temperatures exceed 31°C (daytime) and 25°C (night-time) regardless of moisture availability (Thorogood, 2003). It is tolerant to cold winters and can maintain growth down to 4°C (Grassland Society of Southern Australia Inc, 2008b). Perennial ryegrass accessions can have freezing tolerances (LT₅₀) as low as - 10.3°C to -13.95°C for most cultivars grown in the US (Hulke et al., 2007) or -3°C to -12.6°C for northern European cultivars (Humphreys and Eagles, 1988), although this difference may be related to experimental protocols. Generally cultivars with poor winter hardiness were very early heading, whereas those with better winter hardiness were later heading (Humphreys, 1989). A positive correlation exists between LT₅₀ estimates and lowest temperature of coldest month in the place of origin of the accession (Humphreys and Eagles, 1988), illustrating the plasticity of this species. Perennial ryegrass is less shade tolerant than other perennial pastures (Dodd et al., 2010).

Tall fescue is a shade tolerant species and is more cold tolerant than perennial ryegrass, but in a turf environment needs a higher soil temperature for germination than other grasses such as Kentucky bluegrass or bentgrass (Meyer and Watkins, 2003) as germination is limited by temperatures less than 12°C (Grassland Society of Southern Australia Inc, 2008c). Cultivars with a Mediterranean origin have a growing season from autumn to late spring, and are dormant over winter. Other tall fescue cultivars have a growing season from spring to autumn, with little or no growth over winter (Lamp et al., 2001). It is tolerant of hot conditions and can grow from 0-35°C (Grassland Society of Southern Australia Inc, 2008c).

6.3 Water stress

There is variation between the three species of grass in their ability to tolerate dry conditions. A study in the UK indicated that tall fescue could survive four years without rain, whereas perennial ryegrass died after two years and Italian ryegrass only survived for 12-15 months (Wilman et al., 1998).

Tall fescue

Tall fescue develops a relatively deep root system compared to perennial ryegrass and other cool-season turfgrasses. This trait, along with high evapotranspiration and leaf water potential, provides tall fescue with higher drought tolerance when compared to others in the Lolium-Festuca complex (Aronson et al., 1987; Sheffer et al., 1987; Wilman et al., 1998). Water stress in tall fescue leads to leaf rolling, stomatal closure and a reduction in transpiration rate (Renard and Francois, 1985). The dense fibrous root stock of tall fescue forms an extensive root system that has the ability to draw water from >1 m in the soil profile (Garwood and Sinclair, 1979). In a comparison with seven other grasses in the Lolium-Festuca complex, tall fescue had the greatest number and weight of roots at 50-100 cm depth, although frequent cutting of shoots reduces the root system and hence the plants' drought tolerance (Gibson and Newman, 2001). This has also been seen with a combination of severe moisture stress and grazing pressure (Lazenby, 1997). Generally tall fescue recovers rapidly from drought, especially in endophyte infected plants, where enhanced accumulation of cell solutes and a decreased osmotic potential following drought has been observed (Lodge, 2004). However, there is variability in the degree of drought tolerance between cultivars and regions depending on environmental factors (Aronson et al., 1987; Sheffer et al., 1987; Ervin and Koski, 1998; Lodge, 2004). Mediterranean cultivars are able to 'avoid' drought due to morphological adaptations such as small plant size and

higher root to shoot ratio. The temperate cultivars are more able to make physiological adjustments such as osmotic adjustment in leaf blade tissue or decrease in the rate of leaf senescence and can grow through dry conditions (Assuero et al., 2002).

Tall fescue is also tolerant of flooding and has been recorded as occurring in pastures that are inundated with water for up to 4 weeks (Gibson and Newman, 2001) or up to 8 months of flooding during winter (Razmjoo et al., 1993).

Perennial ryegrass

Perennial ryegrass is sensitive to drought (Garwood and Sinclair, 1979), which leads to a reduction in herbage production under mild moisture deficit and dormancy or death under severe drought. The minimum annual rainfall requirement is 457 to 635 mm (Thorogood, 2003). However, the inability of perennial ryegrass to survive dry summers in areas where annual rainfall is below 650-700 mm limits its use in Australia (Waller and Sale, 2001). There are differences in the level of drought tolerance between perennial ryegrass cultivars given the wide geographic and climatic range of this grass species. For example, seed of accessions from Algeria, a consistently dry habitat, had greater drought tolerance compared to commercial cultivars from Australia and New Zealand (Reed et al., 1987). Some perennial ryegrass cultivars can survive drought for 2 years with cutting, but do not produce as much yield as tall fescue under the same conditions. This difference in drought tolerance and yield is determined by the different root systems of the two grass species. Perennial ryegrass roots are able to draw water from approximately 80 cm deep in the soil, while tall fescue was able to draw water from greater than 100 cm in the soil (Garwood and Sinclair, 1979).

Numerous drought tolerance or avoidance mechanisms have evolved including variation in size or number of stomata, depth of epidermal ridging, leaf water conductance and leaf osmotic potential (reviewed in Casler et al., 1996). Studies in both temperate (Waller et al., 1999) and subtropical (Lowe et al., 1999a) Australia indicated that perennial ryegrass survived the hot dry conditions by dying back and then, when conditions improved, producing new tillers in the centre of the crown.

Perennial ryegrass is moderately tolerant to waterlogging or flooding, less than tall fescue (Razmjoo et al., 1993). It will tolerate extended periods of flooding (up to 25 days) when temperatures are below 27°C.

Italian ryegrass

Italian ryegrass is not tolerant of dry conditions and did not survive 2 years of drought without irrigation (Garwood and Sinclair, 1979). However, Italian ryegrass is moderately tolerant to waterlogging (Grassland Society of Southern Australia Inc, 2008a).

Variation between cultivars is seen. For example, the Italian ryegrass cultivar Aristocrat is useful in areas prone to severe rainstorms as it has been bred for greater resistance to seed shedding and lodging during adverse weather (Oram and Lodge, 2003).

6.4 Herbicides

All three grasses are used in turf and pasture, and control of turf weeds such as *Poa annua* and pasture weeds such as *L. rigidum* and *Avena spp.* is desirable. For this reason, many cultivars are tested for their susceptibility to various herbicides.

P. annua in turf can be controlled by applying split applications of ethofumesate e.g. 2.2 + 1.1 or 2.2 + 2.2 kg/ha (Dernoeden and Turner, 1988). At these concentrations, perennial ryegrass cultivars could be safely treated with the herbicide. *P. annua* has shown some susceptibility to sulfosulfuron, an acetolactate synthase-inhibiting herbicide. Perennial ryegrass cv. Paragon was found to be tolerant to the herbicide if applied at rates up to 6-22 g/ha whereas tall fescue cv. Coronado was not tolerant to application rates necessary for adequate *P. annua* control (Lycan and Hart, 2004).

In a study which tested the tolerance of perennial pasture grass seedlings to pre- and postemergent herbicides, simazine, atrazine, flamprop-*m*-methyl, imazethapyr, fenoxapropethyl, and triallate caused less severe toxicity to perennial ryegrass cv. Kangaroo Valley and tall fescue cv. Demeter, than other herbicides tested such as fluazifop and tralkoxydim. The less toxic herbicides caused yield reductions between 0-45%, 30 days after spraying. Simazine caused yield losses of 20-50% in both perennial ryegrass and tall fescue, which may be deemed acceptable in swards with high weed burdens (Dear et al., 2006). Atrazine and fenoxaprop-p-ethyl also showed some selectivity for weeds versus perennial grass species.

Herbicide resistance

Herbicide resistance in plants is a common phenomenon that occurs as a consequence of the widespread use of herbicides for weed control. It was first recognised in Australia in 1981 when annual ryegrass (*Lolium rigidum*) developed resistance to diclofop-methyl (Heap, 1988). The Herbicide Resistance Action Committee (HRAC), whose aim is to create a uniform classification of herbicide modes of action in as many countries as possible, has proposed a number of herbicide groups, which were updated in 2020 (HRAC, 2020). An increasing problem is cross resistance across the groups and Integrated Weed Management techniques are becoming more important for achieving weed control (Vic DPI, 2005).

Italian ryegrass biotypes show resistance (or multiple resistances) to a range of herbicides (Heap, 2022). Of particular impact is resistance to glyphosate and diclofop-methyl. Glyphosate is a Group 9 herbicide (inhibition of EPSP synthase, Legacy HRAC Group G) and resistance has been reported in two Italian ryegrass populations from Chile and Oregon (Perez-Jones et al., 2007) and, with the worldwide usage of the herbicide, may be expected to occur more widely. For instance, a population of multiple-herbicide-resistant Italian ryegrass was identified in Oregon, USA (Liu et al., 2014; Liu et al., 2017). The mechanism for this resistance is thought to involve lower spray retention, lower foliage uptake from the abaxial leaf surface and altered translocation patterns (Michitte et al., 2005; Michitte et al., 2007). Diclofop-methyl is a Group 1 herbicide (inhibition of acetyl CoA carboxylase, Legacy HRAC Group A). The presence of Italian ryegrass in wheat crops is a significant problem (see

Section 8) and, while post-emergent spraying with diclofop-methyl has been successful, diclofop-methyl resistant *L. multiflorum* has developed (Hoskins et al., 2005). In countries other than Australia, Italian ryegrass biotypes have also been found showing single or multiple-resistances to herbicides from multiple functional groups (Liu et al., 2017; Heap, 2022).

Perennial ryegrass has recently begun to show extensive herbicide resistances in Argentina, New Zealand and Portugal, but not in Australia (Ghanizadeh et al., 2015; Heap, 2022). Glyphosate resistance in perennial ryegrass has been shown to be a heritable trait able to be transmitted to *L. perenne* × *L. multiflorum* hybrids (Yanniccari et al., 2015). An analysis of the glyphosate resistance trait has revealed that it confers a burden on the organism (Yanniccari et al., 2017) suggesting that the trait might be self-limiting in the absence of herbicide selection. The International Herbicide-Resistant Weed Database does not record any entries for tall fescue (Heap, 2022), nor are any apparent in the wider scientific literature.

6.5 Other tolerances

The plasticity of grasses has meant that in natural populations there is an ability to adapt to a variety of abiotic stresses. These include tolerance to heavy metals, sulphur dioxide (SO₂) and salinity (Wilson and Bell, 1985; Casler and Duncan, 2003).

Salt

Salt tolerance has evolved naturally in many grasses, especially those in coastal marshes and rocky alpine habitats (Casler and Duncan, 2003). The three grass species differ in their tolerance to saline conditions, with perennial ryegrass being the most salt tolerant and Italian ryegrass the least. Experiments selecting for salt tolerance in perennial ryegrass indicated that it could grow in solutions containing 200 mM NaCl (Ashraf et al., 1986, 1989), but it is not generally seen in saline habitats (Venables and Wilkins, 1978). However, this may be due to its inability to withstand waterlogging as many saline habitats such as salt marshes have waterlogged soils (Ashraf et al., 1986). It has been classified as moderately salt tolerant (Rogers, 2007).

Tall fescue shows higher salt tolerance than either species of ryegrass (Grassland Society of Southern Australia Inc, 2008c) with a small reduction in turf quality at 4.7dSm⁻¹ salinity level (deciSiemens per metre) (Alshammary et al., 2004). However, nitrogen status affects the response of tall fescue to salinity. Turf grown in non-limiting conditions for nitrogen is less salt tolerant than turf grown in moderately nitrogen deficient turf (Bowman et al., 2006). Italian ryegrass is not tolerant of salinity (Grassland Society of Southern Australia Inc, 2008a). Despite these limitations, perennial ryegrass and tall fescue feature amongst the most successful perennial grasses in saline and waterlogged conditions (Dear et al., 2008).

Sulphur dioxide

Tolerance to sulphur dioxide (SO₂) pollution is present in populations of perennial ryegrass that had been grown in polluted areas of the UK. This tolerance evolved within 3.5 years for

tolerance to acute levels (3,000-6,000 μ g m⁻³ SO₂ for 6h) and a year longer for tolerance to chronic pollution (317 μ g m⁻³ SO₂ for 123 days) (Wilson and Bell, 1985). Further research confirms that perennial ryegrass is tolerant of SO₂, even where the grass was not pre-adapted to SO₂ as it was in the UK study (Li et al., 2015).

Metals

Tall fescue has been shown to display some tolerance to elevated levels of aluminium (Grassland Society of Southern Australia Inc, 2008c), whereas perennial ryegrass is described as not particularly tolerant (Grassland Society of Southern Australia Inc, 2008b), and Italian ryegrass is not tolerant (Grassland Society of Southern Australia Inc, 2008a). Perennial ryegrass has been shown to tolerate lead, cadmium, and zinc contamination to the extent required for use as a phytoremediant of contaminated land (Bidar et al., 2009), and Italian ryegrass is highly tolerant of cadmium (Sabreen and Sugiyama, 2008).

Grazing

One of the other major abiotic stresses faced by a pasture or turfgrass is that of defoliation, either by a grazing animal or through mowing. In experiments, perennial ryegrass was less tolerant to defoliation than tall fescue (Cullen et al., 2006). Defoliation restricted new tiller development and caused tiller and plant death in perennial ryegrass. However, in nature perennial ryegrass is thought to employ a defoliation avoidance strategy by changing tiller size and density to prevent defoliation by grazing animals (Chapman and Clark 1984).

Fire

Most well-established perennial grasses can survive a cool-moderate burn³ (Ward, 1995). Green tall fescue pasture has some resistance to fire; perennial ryegrass is damaged more by fire than other temperate improved species because the surface crowns are easily burnt (McGowen, 1997). Survival of perennial ryegrass is inversely proportional to the fire intensity (Table 7).

Fire intensity ³	Survival of plants
Unburnt	100%
Cool burn	98%
Moderate burn	79%
Hot burn	42%
Very hot burn	0%

Table 7. Survival of perennial ryegrass plants following fires at Hamilton (Victoria)*

* data taken from (McGowen, 1997)

³ Cool-moderate burn (50 – 150°C soil surface temperature) – most dead plant material burnt, some seed and perennial grasses survive unhurt. Usually, a small residue of unburnt pasture remains; Hot burn (150 – 250°C soil surface temperature) – all dead plant material, many seeds, young and weaker perennial grasses destroyed. Topsoil charred and bare; Very hot burn – soil virtually sterilised; all plant material and seed is destroyed in the top organic matter layer (Ward, 1995; McGowen, 1997).

SECTION 7 BIOTIC INTERACTIONS

7.1 Weeds

Some of the main weeds found in the turfs and pastures in Australia are listed in Appendices 2 and 3, respectively. The main pasture weeds identified in a study in the subtropics were paspalum (*Paspalum dilatatum*), barnyard grass (*Echinochloa spp.*) and both red and white clover (*Trifolium repens* and *T. pratense*) (Lowe et al., 1999a). In Victoria, both Bathurst burr and Amsinckia are considered to be significant pasture weeds (Department of Primary Industries, 2007b, a).

7.2 Pests and pathogens

Some of the main nematode pests in Australian turfs and pastures are listed in Appendix 4, insect pests in Appendices 5 and 6, and diseases in Appendices 7 and 8, respectively.

The main insect pests of perennial ryegrass in Australia are black field cricket, black headed pasture cockchafer, red headed pasture cockchafer, common army worm, common cutworm, pasture tunnel moth, red legged earth mite, lucerne flea, and cereal rust mite (Cunningham et al., 1994; Project 30:30, 2012). Pasture scarabs and Corbie grubs attack roots just below the ground. This attack is tolerated better by tall fescue than perennial ryegrass (Harris and Lowien, 2003). However tall fescue is affected by red legged earth mites, blue oat mites, field crickets, slugs and snails (Lowien and Harris, 2004).

The main fungal pathogens of perennial ryegrass in Australia are crown rust, stem rust, net blotch and blind seed disease (Cunningham et al., 1994). Crown rust can seriously damage perennial ryegrass turf in the Autumn, especially under conditions of low fertility (Meyer and Belanger, 1997). Stem rust and blind seed disease can be serious problems for seed production in southern Australia. Blind seed disease reduced seed quality and yield and has cost the Victorian seed industry up to \$2.5 million in some years, especially when it is humid during seed harvest (Cunningham et al., 1994). Barley yellow dwarf virus (BYDV) and ryegrass mosaic potyvirus (RMV) have been reported in perennial ryegrass in Australia (Eagling et al., 1989; Eagling et al., 1992).

One of the major problems of tall fescue as turf is susceptibility to brown patch, caused by *Rhizoctonia solani* (Sleper and West, 1996). However, the two most important fungal diseases selected against in breeding programs for tall fescue are crown rust and stem rust (Sleper and West, 1996).

Damping off can cause severe seedling loss, especially if seed is sown into a cold, damp seedbed (Harris and Lowien, 2003). Ergot can also infect tall fescue, causing black-purplish elongated ergots in the seed head, containing alkaloids that are toxic to livestock (Harris and Lowien, 2003) (see Section 5.1).

7.3 Endophytes

Tall fescue, Italian ryegrass and perennial ryegrass can all contain endophytes. Endophytes are fungi that live between the plant cells of many forage grasses (Kemp et al., 2007). The

fungi and grasses have a mutualistic symbiotic relationship where the fungus derives its nourishment and means of reproduction from the grass and the grass host benefits from enhanced tolerance to biotic and abiotic environmental stresses (Joost, 1995). These endophytes complete their lifecycle within host tissues, are maternally transmitted in seed, are completely non-pathogenic and are asexual (Tsai et al., 1994). Growth of the fungi is systemic through the aerial parts of the plant (Rasmussen et al., 2007). Sparsely branched hyphae grow parallel to the long axis of plant cells in intercellular spaces. During host flowering the fungus grows into ovules and seeds, with the rate of vertical transmission approaching 100% (Clay and Schardl, 2002). In perennial ryegrass leaf blades, the ratio of fungal compartments (one genome copy = one compartment) to plant cells may be as high as 1:2 to 1:1 (Rasmussen et al., 2007).

The most studied symbioses are tall fescue with *Neotyphodium coenophialum* (previously known as *Acremonium coenophialum*) and perennial ryegrass with *N. lolii*; also known as *Epichloë festucae* var. *lolii* (previously known as *Acremonium lolii*) (Schardl et al., 1997). They are evolutionarily derived from the sexual (teleomorphic) fungi of genus *Epichloë* (*Ascomycotina, Clavicipitaceae*) which causes grass choke disease. Phylogenetic evidence suggests that *N. coenophialum* is an interspecific hybrid with three *Epichloë* ancestors (Tsai et al., 1994). In most old pastures in Australia about 90% of perennial ryegrass plants are infected with *N. lolii* (Reed, 1999b). The endophyte in Italian ryegrass is found only in the nodal region rather than in the upper portion of the leaf sheath (Latch et al., 1987) and is thought to be *N. occultans* C.D. Moon, B. Scott & M.J. Chr. (Dombrowski et al., 2006; Sugawara et al., 2006). The infection rate varies as, although half of the wild Italian ryegrass populations collected contained endophyte, eight cultivars from a Welsh plant breeding station were not infected with endophyte (Latch et al., 1987).

There is a rapid decline in endophyte (*N. lolii*) when perennial ryegrass seed is stored at room temperature for more than 24 months (Reed et al., 1987), although viable endophyte has been found in some plants grown from 14 year old seed (Latch et al., 1987). This effect has been found to vary amongst tall fescue cultivars and endophyte accessions (Hill and Roach, 2009). Endophyte infection remaining greater than 80% after 18 months in the 'Flecha' cultivar infected with the AR542 endophyte, and 0% in the 'Advance' cultivar infected with the same endophyte (Hill and Roach, 2009). In general, however, the endophyte could be eliminated from most cases by storage for 22 months.

These fungi do not cause any disease in the grasses, and under most circumstances they are beneficial to the growth and survival of infected plants (Clay, 1990; Joost, 1995; Schardl et al., 1997; Schardl and Phillips, 1997; Clay and Schardl, 2002; Grewal and Richmond, 2003; Schardl et al., 2004; Lowe et al., 2008). For example, in tall fescue, *N. coenophialum* increases tillering and root growth, improves drought tolerance, and protects against certain nematodes, fungal pathogens, insect and mammalian herbivores. Endophyte infection of tall fescue and perennial ryegrass has been shown to be a highly desirable trait for crop performance in Australia (Hume and Sewell, 2014).

Perennial ryegrass and tall fescue containing endophytes have been more persistent and productive in areas of NSW where African Back Beetle (*Heteronychus arator*), Prunose and Dusky Pasture Scarabs (*Sericesthis spp.*) are found on the tablelands and coastal areas (Kemp et al., 2007), and are subject to less grazing by herbivores (Pennell et al., 2016). However, growth comparisons indicate that endophyte infection in perennial ryegrass may only be advantageous under particular environmental conditions (Marks et al., 1991). Endophyte infection may also provide protection against fungal pathogens (Lowe et al., 2008; Wiewióra et al., 2015). Further, presence of the endophyte is associated with improved water stress tolerance in perennial ryegrass (Hahn et al., 2008; Kane, 2011). Endophyte-containing Italian ryegrass plants produce more vegetative tillers, root biomass and seed than uninfected plants (Elmi and West, 1995).

Endophytes in tall fescue are able to affect plant osmotic relations to improve drought tolerance (Malinowski et al., 1998), and recovery after drought (Elmi and West, 1995). Endophyte-infected tall fescue maintained significantly higher photosynthetic rates at temperatures above 25°C than uninfected plants in the glasshouse (Clay, 1990). Tall fescue infected with an endophyte accumulates more phosphorus than non-infected plants when grown in phosphorus-deficient soils. In response to phosphorus deficiency, endophyte-infected plants produce a Fe³⁺ reducing activity on the root surface and have increased phenolic concentration in roots and shoots (Malinowski et al., 1998). These soil-level chemical changes may underlie the observation of significantly altered soil ecology and soil gas exchanges in endophyte-infected tall fescue plots (Iqbal et al., 2012). Endophyte-infected tall fescue has numerous agronomic advantages over endophyte free cultivars (Hume and Sewell, 2014).

Seeds from infected tall fescue and perennial ryegrass germinated at a higher frequency than uninfected seeds and had more tillers and a greater biomass (Clay, 1987). Infected tall fescue produced twice the number of filled seed than uninfected, although for perennial ryegrass there was no difference between infected and uninfected (Clay, 1987).

Endophytes in Italian ryegrass might delay the appearance of herbicide resistance (Vila Aiub and Ghersa, 2001). Uninfected plants showed faster germination at suboptimal temperatures (Gundel et al., 2006b) and under water stress (Gundel et al., 2006a); although this may be due to altered seed dormancy levels (Gundel et al., 2006a). The effects of endophyte infection in Italian ryegrass seem most pronounced under drought stress, where reduced aphid densities and increased tillering were observed compared to uninfected controls (Miranda et al., 2011). Field experiments in a range of ecological settings showed that Italian ryegrass enhanced invasive establishment, as well as biomass accumulation (Uchitel et al., 2011).

The concentration of endophytes and associated alkaloid varies between plant cultivars (Rasmussen et al., 2007). The plant's nutritional status may also affect the concentration of both endophyte and alkaloid in the plant (Rasmussen et al., 2007). Alkaloid concentration in tall fescue and perennial ryegrass is also altered by management practices, such as mowing.

A decreased mowing frequency resulted in higher alkaloid production in the two species, while the levels of some alkaloids were also increased when the mowing height was increased (Salminen and Grewal, 2002; Salminen et al., 2003). The concentration of multiple alkaloid toxins is dependent on nutrient status, and also varies with the local environment (Helander et al., 2016). The alkaloids produced by endophytes in tall fescue may decrease predation on the plants by birds (Madej and Clay, 1991), rabbits (Pennell et al., 2016), and mice (Finch et al., 2016). Furthermore, an effect of endophyte infection is some measure of protection against insect predation in perennial ryegrass and tall fescue (Saari et al., 2010; Hume and Sewell, 2014).

Despite being potentially toxic to grazing livestock (see Section 5.1), the good forage and agronomic characteristics of tall fescue infected with N. coenophialum and perennial ryegrass infected with N. lolii (alternatively known as Epichloë festucae var. lolii), in conjunction with their exceptional fitness and low management requirements places them amongst the most important forage grasses (Schardl and Phillips, 1997; Hume and Sewell, 2014). Strains of endophytes which do not produce toxic alkaloids, but which still produce compounds that are beneficial to plant persistence have been developed (Harris and Lowien, 2003) and are included with the majority of the ryegrass sold in New Zealand (Easton, 2007), where the agronomic advantage of endophyte mutualism is most apparent. In Australia, comparatively little research has been undertaken on specific advantageous endophytes (Hume and Sewell, 2014), but a few studies have shown selected variant endophytes to perform at least as well as the wildtype (toxic) strain (Moate et al., 2012), or less well than wildtype (Lowe et al., 2008). In the USA, studies with tall fescue infected with non-ergot alkaloid producing endophyte strains provided cattle growth performance that did not differ from that of endophyte-free tall fescue (Parish et al., 2003). In Australia, many tall fescue varieties are commercially available containing a low toxicity endophyte which still helps protect the plant from insect attack and environmental stresses (Burnett, 2006b). It is also possible to remove the endophyte from perennial ryegrass and tall fescue seed using a fungicide such as prochloraz (Leyronas et al., 2005), or by ageing seeds beyond about 24 months. Renovated pastures in which endophyte infected tall fescue was reduced to <10%, was sown with endophyte free seed and after 3 years rotational cattle grazing endophyte infected plants did not invade and increase (Tracy and Renne, 2005).

SECTION 8 WEEDINESS

Weeds are plants that spread and persist outside their natural geographic range or intended growing areas such as farms or gardens. Weediness in Australia is often correlated with weediness of the plant, or a close relative, elsewhere in the world (Panetta, 1993; Pheloung et al., 1999; Groves et al., 2005). The likelihood of weediness is increased by repeated intentional introductions of plants outside their natural geographic range that increase the opportunity for plants to establish and spread into new environments (e.g. escapes of commonly used garden plants) (Groves et al., 2005).

As noted earlier in this document, *Lolium* species have many weedy characteristics. They are wind pollinated, are primarily an outcrossing species with vegetative abilities, can adapt rapidly to their environment, produce large amounts of seed, and are easily dispersed (see discussion in Section 4.3).

8.1 Weediness status on a global scale

The most comprehensive compilation of the world's weed flora is produced by Randall (Randall, 2017). Most of the information contained in this book has been sourced from Australia and North American countries but also includes numerous naturalised floras from many other countries. Randall (2017) lists 15 species of *Lolium* which have been identified as having a documented weedy history, including all the *Lolium* species introduced into Australia - including the hybrids *Lolium* × *hubbardii* and *Lolium* × *hybridum* which were previously listed as naturalised, and the hybrid *Lolium arundinaceum* × *Lolium perenne* (refer to Table 1). Forty-six species of *Festuca* have also been identified with a history of weediness; including all *Festuca* species introduced to Australia (refer to Table 2). Risk assessments undertaken overseas identified perennial ryegrass as having a high risk of invasiveness in Hawai'i and other Pacific islands (Hawai'i-Pacific Weed Risk Assessment, 2016).

8.2 Weediness status in Australia

Groves et al. (2003) lists Italian ryegrass, perennial ryegrass and tall fescue as weeds in native and agricultural ecosystems in Australia, all three species are given a rating of 4 and 3 in these respective areas (refer to Table 8 for details). Italian and perennial ryegrass are primarily agricultural weeds, commonly found in fields, waste places and roadsides whilst tall fescue is most often found in open and damp sites along roadsides, creeks, swampy verges and in open paddocks (Sharp and Simon, 2002). Perennial ryegrass and tall fescue are considered as weeds Australia wide, and Italian ryegrass is considered a weed in all areas of Australia except the Northern Territory (NT) (Lazarides et al., 1997). In SA, tall fescue is described as "becoming a weed in wetter areas" (Wheeler et al., 2002; Wipff, 2002; Jessop et al., 2006a). Perennial ryegrass is naturalised throughout the higher rainfall temperate areas, extending into Southern Qld with casual occurrences near Cairns and Alice Springs (Kloot, 1983; Atlas of Living Australia, 2022a). Randall (2007) accords invasive weed status to tall fescue, perennial ryegrass, and Italian ryegrass, with perennial ryegrass and Italian ryegrass being recorded as weeds of the Australian natural environment, of Australian agriculture, and as escaping cultivation. In Vic, perennial ryegrass and Italian ryegrass are listed as environmental weeds with a medium risk rating, while tall fescue has a high risk rating (White et al., 2022). Furthermore, perennial ryegrass, Italian ryegrass, and tall fescue were all listed as possessing extreme risk of weediness according to the Western Australian weediness risk assessment system (Randall, 2016, 2017).

Other *Festuca* and *Lolium* species present in Australia are also considered to be weedy. *L. rigidum* and *F. elatior* are weeds Australia wide, while *L. loliaceum*, *L. temulentum* and *F. rubra* are considered as weeds in all areas of Australia except the NT. *F. nigrescens* and *L. pratense* are weeds in NSW and Tas, and WA, SA, NSW and Tas, respectively (Lazarides et al., 1997). *L. loliaceum* is commonly found on roadsides, in waste places, sandy areas, and often in maritime habitats. *L. rigidum* is primarily an agricultural weed found in crops, fields, roadsides, waste places, rocky hillsides, and sandy areas. It is regarded as one of the most significant weeds of cereal-based cropping systems. It has a transient soil seed bank (Maia et al., 2009) and persistent use of herbicides has resulted in resistance to a number of Group 1 (Legacy HRAC Group A), 2 (Legacy HRAC Group B), 12 (Legacy HRAC F), 14 (Legacy HRAC Group G), 15 and 0 (Legacy HRAC Group K), and 10 (Legacy HRAC Group N) herbicides (Vic DPI, 2005; Heap, 2022)⁴, and herbicide-resistant populations are becoming more widespread and resistant in the Australian wheat belt (Owen et al., 2014). *F. nigrescens* is common in drier open habitats on roadsides and in pasture; and *F. rubra* occurs in damp open habitats (Sharp and Simon, 2002).

No *Festuca* or *Lolium* species are registered as weeds of national significance to Australia (Weeds Australia, 2022).

Species	Australian rating natural ecosystems	Australian rating agricultural ecosystems	QLD	NSW	VIC	TAS	SA	WA	NT
Lolium arundinaceum	4	3	1	+	1	+	2	3	+
Lolium Ioliaceum	4	2		1	+	+	2	+	1
Lolium multiflorum	4	3	1	3	3	+	2	3	
Lolium perenne var. cristatum	2	+			+				
Lolium perenne var. perenne	4	3	1	3	3	+	3	3c	1
Lolium perenne × Lolium rigidium	2	+	+						
Lolium pratense	3	+		+			+	+	
Lolium remotum	3	+						+	
Lolium rigidum	5	5	2	5	5	5 (crops)	4	5	1
Lolium temulentum var. arvense	3	1	1	+	+	+	1		
Lolium temulentum var. temulentum	3	1	+	+	+		0	1	
Lolium ×hubbardii	3	1	+	+			1		
Lolium ×hybridum	3	1	+	+			1		
Festuca nigrescens	4	+		+		+			
Festuca rubra ssp. rubra	4	1		+	+	+	1	+	

Table 8. Assessment of weediness of Lolium and Festuca species in natural and agricultural ecosystems
in Australia (Groves et al., 2003)

⁴ Note that the Classification system used by VDPI is slightly different from the HRAC system described in Section 6.1.4. The Groups listed relate to the HRAC system.

Key:

0 – Reported as naturalised but only known naturalised population now removed or thought to be removed 1 – Naturalised and may be a minor problem but not considered important enough to warrant control at any location

2 – Naturalised and known to be a minor problem warranting control at 3 or fewer locations within a State or Territory

3 – Naturalised and known to be a minor problem warranting control at 4 or more locations within a State or Territory

4 - Naturalised and known to be a major problem at 3 or fewer locations within a State or Territory

5 – Naturalised and known to be a major problem at 4 or more locations within a State or Territory

+ - Present in a State or Territory but not given a rating as an agricultural weed, either because it was not considered a problem or because it was not known to occur in agricultural areas at present
 c - Under active control in part of a State

8.3 Weediness in agricultural ecosystems

In terms of ability to spread in the environment, *L. perenne* and *L. multiflorum*, both being classed as bunchgrasses (see Section 4.1.1) rely mainly on spread by seed. In turf environments and in pasture that is being grazed or mown, the likelihood of seed developing is low. However, in ungrazed pasture and unmanaged habitats seed has an opportunity to form. Seeds of both species germinate rapidly and with high rates, leading them to be present in the seedbank for only a relatively short time (Sullivan, 1992; Carey, 1995) (see also discussion in Section 4.4).

The ryegrasses in general are significant weeds of wheat worldwide and their control has been investigated extensively (Kuk and Burgos, 2007). Control of unwanted ryegrasses is complicated by the parallel development of multiple herbicide tolerances in multiple locations worldwide (Heap, 2022). A sample of perennial ryegrass taken from an Adelaide Hills vineyard in 2019 was found to be highly resistant to the herbicide, paraquat (Group 22), following the use of this herbicide at sublethal rates over a number of years (Boutsalis et al., 2021). Italian ryegrass can be a difficult-to-control contaminant in turfgrass farms and causes decreased marketability of cool-season sod (Beam et al., 2005). Perennial ryegrass is highly vigorous and competitive, leading to significant reductions in growth of other grasses (*Phalaris* in this case) in competition experiments (Donald, 1958). The underlying basis was perennial ryegrass' competitiveness for light and nutrients.

While tall fescue can develop short rhizomes, it is not regarded as a spreading grass and there is little evidence in the literature that tall fescue *per se* is a significant weed of either pasture or cropping systems. It is slow to establish and is sensitive to competition from more vigorous pasture and weed species (Harris and Lowien, 2003). However, the association of tall fescue with a fungal endophyte (see Section 7.3.1) often means that grazing stock may lose productivity in an infected pasture and that removal of the infected sod is desirable (Smith, 1989; Defelice and Henning, 1990).

Tall fescue is a wide-bladed grass, texturally different from a turf such as Kentucky bluegrass and its appearance in fine-leaf turf may therefore be undesirable and warrant removal.

Volunteer tall fescue growing near certified seed production enterprises also requires control measures to prevent contamination of the seed (Mueller-Warrant and Rosato, 2005).

8.4 Weediness in natural ecosystems

The ryegrasses and tall fescue occur as typical weed species in riparian zones in rural and urban areas of Australia. For example:

- Perennial ryegrass is listed as one of the perennial grasses that is a weed along waterways and wetlands of cool, high rainfall areas of WA (Pen, 2000) and is both invasive and has a high impact in a floodplain riparian woodland in the Northern Inland Slopes Bioregion of Vic (Department of Sustainability and Environment, 2004a).
- Italian ryegrass is listed as one of the introduced species that have replaced native samphire and rush communities along the Swan/Canning and Leschenault estuaries (WA) flushed by stormwater (Pen, 2000) and is invasive in a floodplain riparian woodland in the Northern Inland Slopes Bioregion of Victoria (Department of Sustainability and Environment, 2004a).
- Tall fescue is invasive and has a high impact in a eucalypt swampy woodland in the Warrnambool Plain bioregion of Vic (Department of Sustainability and Environment, 2004b).

8.5 Control measures

Herbicide is the main control method for the cool-season perennial grasses as described below (see Section 6.4 for a discussion of the herbicide Groups and of herbicide resistance).

Of the non-selective herbicides, glyphosate (Group 9 – inhibitors of EPSP synthase) is a commonly used systemic herbicide for grass control and glufosinate ammonium (Group 10 – inhibitors of glutamine synthase) is effective against most perennial grasses that do not produce rhizomes (Dernoeden, 1999).

Use of selective herbicides for grass control is more limited. Diclofop-methyl (Group 1 – inhibitors of acetyl CoA carboxylase) has traditionally been used for controlling ryegrass weeds in wheat but resistance has developed. The advent of diclofop-methyl resistant ryegrasses (see Section 6.4) has meant that the use of herbicides from other groups may be necessary to achieve control. In general, the Group 2 herbicides (inhibitors of acetolactate synthase), especially the sulfonylureas, have proved to be successful for control of the cool-season grasses. For example:

- mesosulfuron-methyl, a relatively new herbicide may be suitable for controlling Italian ryegrass in wheat even though mesosulfuron-methyl resistant Italian ryegrass can develop because of the existence of cross resistance to chlorsulfuron in some diclofopmethyl resistant accessions (Kuk and Burgos, 2007; Rauch et al., 2010).
- nicosulfuron has been suggested as an appropriate herbicide to use for Italian ryegrass control in tall fescue or tall fescue/Kentucky bluegrass turf (Beam et al., 2005).
- chlorsulfuron can be used for spot spraying of tall fescue and perennial ryegrass clumps in more tolerant turf species such as Kentucky bluegrass (*Poa pratensis*) (Maloy and

Christians, 1986; Dernoeden, 1999). Similarly, tall fescue is less tolerant to sulfosulfuron than Kentucky bluegrass or perennial ryegrass (Lycan and Hart, 2004).

• triasulfuron used as a pre-emergent herbicide is effective against tall fescue and perennial ryegrass (Dear et al., 2006).

Examples of other herbicides used for control of the ryegrasses and tall fescue are given in Table 9; the references given describe the optimal conditions (e.g. herbicide rate, time of application, stage of growth)

Herbicide	Group	Mode of Action	Grass controlled	Reference
Mesosulfuron- Methyl	2 (Legacy HRAC Group B)	Inhibition of branched chain amino acid synthesis	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015) (Vazan et al., 2011) (Gunnarsson et al., 2017)
lodosulfuron- methyl	2 (Legacy HRAC Group B)	Inhibition of branched chain amino acid synthesis	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015) (Gunnarsson et al., 2017)
Pyroxsulam	2 (Legacy HRAC Group B)	Inhibition of branched chain amino acid synthesis	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015) (Gunnarsson et al., 2017)
Florasulam	2 (Legacy HRAC Group B)	Inhibition of branched chain amino acid synthesis	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015)
Cyanizine (pre- emergent)	5 (Legacy HRAC Group C1)	Inhibition of photosynthesis at photosystem II	Perennial ryegrass, tall fescue	(Dear et al., 2006)
Metribuzin (pre- emergent)	5 (Legacy HRAC Group C1)	Inhibition of photosynthesis at photosystem II	Perennial ryegrass, tall fescue	(Dear et al., 2006)
Isoproturon	5 (Legacy HRAC Group C2)	Inhibition of photosynthesis at photosystem II	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015)
Paraquat	22 (Legacy HRAC Group D)	Photosystem I electron diversion	Tall fescue	(Smith, 1989)
Oxyfluorfen	14 (Legacy HRAC Group E)	Inhibition of protoporphyrinogen oxidase	Tall fescue	(Mueller-Warrant and Rosato, 2005)
Flumioxazin	14 (Legacy HRAC Group E)	Inhibition of protoporphyrinogen oxidase	Perennial ryegrass	(Teagasc, 2015)
Diflufenican	12 (Legacy HRAC Group F1)	Inhibition of PDS	Perennial ryegrass, Italian ryegrass	(Teagasc, 2015)
Glyphosate	9 (Legacy HRAC Group G)	Inhibitor of EPSP synthase	Tall fescue	(Smith, 1989)
Pronamide	3 (Legacy HRAC Group K1)	Inhibitor of microtubule assembly	Perennial ryegrass	(Horgan and Yelverton, 2001)
Pendimethalin	3 (Legacy HRAC Group K1)	Inhibitor of microtubule assembly	Tall fescue	(Mueller-Warrant and Rosato, 2005)
Metolachlor	15 (Legacy HRAC Group K1)	Lipid synthesis inhibition (not ACCase)	Tall fescue	(Mueller-Warrant and Rosato, 2005)
Prosulfocarb	15 (Legacy HRAC Group K1)	Lipid synthesis inhibition (not ACCase)	Italian ryegrass	(Teagasc, 2015)

Table 9. Examples of herbicides used in control of the ryegrasses and tall fescue

Management of grass weeds other than through herbicide application is a possibility. For example:

- Both Italian ryegrass and perennial ryegrass are good soil stabilizers and therefore are sometimes deliberately seeded with other, more aesthetically pleasing grasses to aid establishment. However, because both show vigorous seedling growth and thus are very competitive, the ryegrass component in the final stand often needs to be reduced. Close, early mowing following establishment can often inhibit the ryegrasses (Brede and Duich, 1984; Brede and Brede, 1988). The fact that Italian ryegrass can outcompete regenerating native vegetation is one reason why there has been a move away from reseeding burned chaparral sites in the USA with the species (Carey, 1995)
- If it becomes necessary in a sports field or other grassed amenity area to remove or replace a dominant species, the general principles are that the management strategy is shifted to favour the 'desired' grass and that persistent overseeding of the 'desired' grass eventually crowds out the 'unwanted' grass. As an example, in the conversion of perennial ryegrass golf fairways to Kentucky bluegrass fairways, high seeding of the bluegrass into the mature ryegrass fairway turf followed by low mowing (0.6 cm twice weekly for 4 weeks) allows for successful establishment of the bluegrass (Kraft et al., 2004).
- A biological control involving the planting of diclofop-susceptible Italian ryegrass has been proposed as a method of reducing the evolution of diclofop-methyl resistance in Italian ryegrass (Ghersa et al., 1994).
- On a small scale, ryegrasses and tall fescue can be successfully removed by physically digging up the clumps (Dernoeden, 1999).
- In pastures, tall fescue is susceptible to overgrazing which may therefore be used to reduce the persistence of the grass; perennial ryegrass is less susceptible (Kemp et al., 2001).

A basic reality of modern farming systems is the emergence of herbicide tolerance amongst crop species and weeds. Management of unwanted ryegrasses and fescues, such that herbicide tolerance does not develop or is slow to develop, takes an integrated approach combining selective herbicide use with many of the non-herbicidal methods listed above. Commonly, farmers will manage volunteer ryegrass populations by crop rotation, and herbicide rotation.

SECTION 9 POTENTIAL FOR VERTICAL GENE TRANSFER

Many factors may limit the formation of intra- and interspecific hybrids. These include ecological factors, such as distribution, and biological factors such as anthesis date, hours of pollen shed, genome and chromosome number (e.g. *Festuca* 2n = 14, 28, 35, 42, 56 and 70, and 2, 4, 5, 6, 8, and 10-ploid, *Lolium* 2n = 14 and 28, and 2 and 4-ploid (Sharp and Simon, 2002). The success of the hybrid is sometimes influenced by which of the two species in the cross acts as the male or female parent. The outcome of hybridisation can be variable, in some cases no seed is produced and in others, seed is produced but it is not viable so will not germinate. In other instances, the seed will germinate but result in weak seedlings which do not establish. Successful hybridisation results in established F1 hybrids, which could

sustain a hybrid population and/or backcross with parent plants, or if completely sterile maintain a vegetative population. Tables 1 and 2, and section 9.3 outline hybrids which may be relevant to Australia. However, due to confusion in the taxonomy, the list of hybrids presented may not be comprehensive.

9.1 Barriers to intraspecific crossing

Intraspecific gene transfer is the transfer of genetic material from parent to offspring by reproduction within the same species. This type of gene transfer can occur by sexual or asexual reproduction. Self-incompatibility and wind pollination are common in grass species; therefore, outcrossing is the normal mode of reproduction. Self-incompatibility in both Italian and perennial ryegrass is controlled by a two-locus incompatibility system. However, if self-pollination does occur both species will set seed; although the number of seeds may be low (see Section 4.1.2). This highly outcrossing mode of reproduction may result in hybridisation between species and sub-species, especially where spatial and temporal factors allow it.

9.2 Natural interspecific and intergeneric crossing

This section deals with gene transfer between plants of the same genera by sexual reproduction. Examples of this are presented in Table 1 and Table 2.

Successful gene transfer requires that three criteria are satisfied. The plant populations must:

- 1) overlap spatially
- 2) overlap temporally (including flowering duration within a year and flowering time within a day); and
- 3) be sufficiently close biologically that the resulting hybrids are fertile, facilitating introgression into a new population (den Nijs et al., 2004).

As discussed in Section 1, hybridisation between *Lolium* sp. and *Festuca* sp. is common, with offspring showing varying degrees of fertility.

Lolium and *Festuca* species can form hybrids (refer to Table 1 and Table 2). *Lolium* species can also form hybrids with species belonging to other genera for example, *Vulpia* (*×Festulpia* Melderis ex Stace & R. Cotton) and *Bromus* (*×Bromofestuca* Prodan.) (Watson and Dallwitz, 1992). As some of these species are present in Australia and some crosses form viable or vegetatively strong hybrids, gene transfer between *Lolium* and other genera can occur. Examples of some these hybrids are given in Table 10.

Hybrid	Name	Fertile?
L. multiflorum × L. perenne	L. × hybridium, Festulolium holmbergii (Doerfler) P.Fourn.	yes
L. multiflorum × L. rigidum	L. × hubbardii Jansen & Wachter ex B.K.Simon	yes
L. multiflorum × L. temulentum		yes
L. multiflorum × L. arundinaceum		
L. multiflorum × L. pratense		some

Table 10. Natural hybrids of tall fescue, Italian ryegrass and perennial ryegrass

Hybrid	Name	Fertile?
L. multiflorum × Dactylis glomerata	(Thorogood, 2003)	
L. perenne × L. Ioliaceum		no
L. perenne × L. multiflorum	L. × hybridium, Festulolium holmbergii (Doerfler) P.Fourn.	
L. perenne × L. remotum		no
L. perenne × L. rigidum		yes
L. perenne × L. arundinaceum		
L. perenne × L. pratense	Festulolium Ioliaceae	some
L. perenne × Festuca rubra		
L. perenne × Glyceria fluitans	(Thorogood, 2003)	
L. arundinaceum × L. pratense	F. × aschersoniana Doerfler	
L. arundinaceum × L. giganteum	F. × fleischeri Rohlena	no
L. arundinaceum × F. rubra		

9.3 Crossing under experimental conditions

Sexual hybrids between *L. multiflorum* and *L. arundinaceum* have been readily obtained only unidirectionally, while the reciprocal cross is very difficult (Eizenga and Buckner, 1986). In addition, chromosomal instability and poor female fertility can be a problem. This has been overcome by symmetric and asymmetric fusions performed between protoplasts of tall fescue and Italian ryegrass to make *Festulolium* hybrids (Takamizo et al., 1991; Spangenberg et al., 1995a) or from anther culture (Zwierzykowski et al., 1999). Anther culture has also been used to produce diploid hybrid progeny from tetraploid parents of *L. perenne*, *L. multiflorum* and *L. pratense* (Kopecky et al., 2005).

A viable *L. multiflorum* x *Dactylis glomerata* hybrid has been produced using auxin treatment and embryo rescue and this has been successfully backcrossed to *L. multiflorum* (Oertel et al., 1996). No hybrids were produced without auxin, or between *L. perenne* and *D. glomerata* (Oertel et al., 1996). D. glomerata has also been successfully crossed with *L. arundinaceum* to produce a *L. arundinaceum* x *D. glomerata* hybrid using embryo culture, however this hybrid was sterile (Matzk, 1981).

SECTION 10 SUMMARY

This document provides baseline information about Italian ryegrass, *L. multiflorum* Lam., perennial ryegrass, *L. perenne* L., and tall fescue *L. arundinaceum* (Schreb.) Darbysh. The Information included relates to the taxonomy and origins of these species, general descriptions of their morphology, reproductive biology, methods of control, biotic and abiotic interactions, and the potential for gene transfer to occur to closely related species. The purpose of this baseline information is to inform risk analysis of genetically modified forms of these species that may be released into the Australian environment.

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SECTION 12 APPENDICES

Appendix 1 –	Examples of cultivars of <i>L. perenne, L. multiflorum</i> and <i>L. arundinaceum</i> grown
commercially in	Australia

Species	Use	Plant type	Cultivar	Register of Australian Herbage Cultivars ⁵	PBR in Australia ⁶
L. perenne	Pasture	Very early maturing diploid	Boomer	No	Yes
			Drylander	No	No
			Everlast	No	No
			Fitzroy	No	Yes
			Kangaroo Valley	Yes	Yes
			Matilda	No	No
			Meridian	No	Yes
			Meridian Plus AR1	No	Yes
			Skippy	No	No
		Early maturing diploid	Ausvic	No	Yes
			Camel	No	Yes
			Kingston	No	Yes
			Roper	No	Yes
			Tomson	No	Yes
			Victorian	Yes	No
		Mid-season diploid	Aries HD	No	Yes
			Avalon	Yes	Yes
			Bolton	No	Yes
			Bronsyn	No	Yes
			Bronsyn Plus AR1	No	Yes
			Cannon	No	Yes
			Cannon AR1	No	Yes
			Cowmax CM 105HP	No	No
			Extreme	No	No
			Grasslands Commando	No	Yes
			Grasslands Nui	Yes	No
			Jackaroo	No	Yes
			Lincoln	No	Yes
			LM9928C	No	No
			Prolong	No	Yes
			Rich	No	No
			Samson	No	Yes
			Samson AR1	No	Yes
			Victoca		Yes
			Wintas	No	No
		Late season diploid	Abderdart	No	No
			Alto	No	Yes
			One-50	No	Yes

⁵ The Register of Australian Herbage Plant Cultivars is a voluntary registration system for recording the origins, distinctiveness and agronomic merit of cultivars. The system of registration was set up in the 1960s under the Standing Committee of Agriculture and Resource Management (Kelman, 2001). Like any non-statutory cultivar registration system, it does not confer any legal protection over the name of the plant.

⁶ Proprietary varieties in Australia are protected by Plant Breeder's Rights (PBR), which are exclusive commercial rights to a registered variety. The rights are a form of intellectual property and are administered under the Plant Breeder's Rights Act 1994 (for more information see the <u>IP Australia website</u>). All turfgrass cultivars listed have been developed overseas.

Species	Use	Plant type	Cultivar	Register of Australian Herbage Cultivars ⁵	PBR in Australia ⁶
		Late season tetraploid	Bealey	No	Yes
			Canasta	No	No
			Grasslands Sterling	No	No
			Grazmore	No	Yes
			Optima	No	No
			Sierra	No	No
			Quartet	No	Yes
	Turf		Premier II	N/A	No
			Barlennium	N/A	No
			Brightstar	N/A	No
			Stallion Supreme	N/A	No
L. multiflorum	Pasture	'Biennial' early season flowering diploid	Dargo	No	Yes
		'Biennial' mid-season flowering	Caversham	No	No
		diploid	Eclipse	No	Yes
		'Biennial' late season flowering	Concord	No	No
		diploid	Conquest	No	No
			Crusader	No	Yes
			Dargle	No	Yes
			Diplex	No	No
			Flanker	No	Yes
			Grasslands Status ^{Plus}	No	Yes
			Grasslands Warrior	No	Yes
			Hulk	No	Yes
			Mariner	No	Yes
			Marbella ^{Sud}	No	No
			Sonik	No	Yes
			Tabu	No	Yes
		'Biennial' late season flowering	Feast II	No	No
		tetraploid	Denver	No	Yes
		Annual early flowering diploid	Aristocrat	Yes	No
		Annual carry newening alpield	Noble	No	Yes
		Annual early flowering	Betta Tetila	No	No
		tetraploid	Drummer	No	No
		tettapiolo	Growmore Plus	No	No
			New Tetila	No	No
			Tetila (USA)	No	No
			Tetila Gold	No	No
		Annual mid accord flowering	Ceres Missile	No	No
		Annual mid-season flowering diploid	Ceres Pronto	No	No
				No	Yes
			Progrow		
		Annual mid account flowering	Surrey	No	No
		Annual mid-season flowering	Andy Crosslands Tama	No	No
		tetraploid	Grasslands Tama	Yes	No
			Robust	No	Yes
			Rocket	No	No
			Tetrone	No	No
			T Rex	No	No
			Winter Star	No	No
			Winter Star II	No	No
	Turf		Panterra	No	No
L. arundina-	Pasture	Temperate very early flowering	AU Triumph	No	No
ceum			Dovey	No	No
			Quantum	No	No

Species	Use	Plant type	Cultivar	Register of Australian Herbage Cultivars ⁵	PBR in Australia ⁶
			Quantum Max P	No	No
	Pasture	Temperate mid-late flowering	Advance	No	Yes
			Advance Max P	No	Yes
			Demeter	Yes	No
			Jesup	No	No
			Jesup Max P	No	Yes
			Lunibelle	No	No
			Torpedo	No	No
			Typhoon	No	No
	Pasture	Temperate late flowering	Carmine	No	No
			Vulcan II	No	No
	Pasture	Mediterranean mid-season	Flecha	No	Yes
		flowering	Flecha Max P	No	Yes
			Fraydo	Yes	Yes
			Origin	No	No
			Prosper	No	Yes
			Resolute	No	Yes
			Resolute Max P	No	Yes
	Turf		Barlexas	N/A	No
			Barrera	N/A	No
			Bingo	N/A	No
			RTF™	N/A	No

*Data derived from Register of Australian Herbage Plant Cultivars (CSIRO, 2022), last updated 18/11/2022;

(Harris and Lowien, 2003; Lattimore and McCormick, 2012; Pastures Australia, 2017; Barenbrug Australia, 2021; NSW DPI, 2022b, a, c, d)

Common name	Scientific Name	Common name	Scientific Name
Azolla	Azolla pinnata	Nutgrass	Cyperus rotundus
Bachelors Button	Cotula australis	Onion Grass	Romulea rosea
Bindii	Solvia pterosperma	Parramatta grass	Sporobolus africanus
Burr Medic	Medicago denticulata	Paspalum	Paspalum dilatatum
Cats ear	Hypochoeris radicata	Pennyweed	Hydrocotyle tripartite
Chickweed	Stellaria media	Pennywort	Hydrocotyle bonariensis
Chilean whitlow	Paronychia brasiliana	Petty Spurge	Euphorbia peplus
Creeping Mallow	Modiola caroliniana	Pig weed	Portulaca oleracea
Creeping Oxalis	Oxalis corniculata	Prairie Grass	Bromus catharticus
Crowsfoot	Eleusine indica	Salvinia	Salvinia molesta
Cudweed	Gnaphalium spicatum	Sheep's sorrel	Rumux acetosella
Dandelion	Taraxacum officinale	Summer Grass	Digitaria sanguinalis
Fleabane, Canadian fleabane, Horseweed	Conza canadensis	Water couch	Paspalum paspaloides
Goose grass	Eleusine indica	White Clover	Trifolium repens
Kidney Weed	Dichondra repens	White Root Lobelia	Pratia purpurascens
Lambs Tongue, Ribwort	Plantago lanceolata	Winter Grass	Poa annua
Mullumbimby Couch	Cyperus brevifolius	Wireweed	Polygonum aviculare

Appendix 2 – Common lawn and turf weeds in Australia (Cooper, 2006; Gardenet, 2006).

Common name	Scientific Name	Common name	Scientific Name
African lovegrass	Eragrostis curvula	Mimosa	Mimosa pigra
Annual grasses	Bromus, Hordeum, Vulpia, Lolium spp.	Noogoora burr	Xanthium spinosum
Bathurst burr	Xanthium spinosum	Parramatta grass	Sporobolus indicus
Blackberry	Rubus fruticosus	Parthenium weed	Parthenium hysterophorus
Bracken fern	Pteridium esculentum	Paterson's curse	Echium plantagineum
Burr grasses	Cenchrus spp.	Poa tussock	Poa labillardieri
Calomba daisy	Pentzia suffruticosa	Rubber tree	Calotropis procera
Caltrop	Tribulous spp.	Rubber vine	Cryptostegia grandiflora
Cape tulip	Homeria spp.	Sedges	Carex spp.
Capeweed	Arctotheca calendula	Serrated tussock	Nassella trichotoma
Common pear	Opuntia stricta	Sifton bush	Cassinia arcuata
Crofton weed, soursob	Eupatorium adenophorum	Sorrel	Rumex acetosella
Fireweed	Senecio madagascariensis	Speargrass	Heteropogon contortus
Galvanised burr	Sclerolaena birchii	St John's wort	Hypericum perforatum
Gorse	Ulex europaeus	Sweet briar	Rosa rubiginosa
Groundsel bush	Baccharis halimifolia	Thistles	Carduus, Carthamus, Cirsium, Onopordum, Silybum spp.
Harrisia cactus	Eriocereus martinii	Tiger pear	Opuntia aurantiaca
Horehound	Marrubium vulgare	Wiregrass	Aristida ramosa
Lantana	Lantana camara	Woody weeds	Dodonaea, Eremophila spp.

Appendix 3 – Common pasture weeds in Australia (Gardenet, 2006).

Appendix 4 – Common nematode pests of turf and pasture crops in Australia (Vargas, 2005)

Common name	Genus name	Common name	Genus name
Awl	Dolichodorus	Seed-gall	Anguina
Cyst (larvae)	Heterodera	Sheath	Hemicycliophora
Dagger	Xiphinema		Helicotylenchus
Lance	Hoplolaimus	Spiral	Rotylenchus
Needle	Longidorus		Tylenchus
Pin	Paratylenchus	Stubby root	Paratrichodorus
Ring	Criconemella	Stunt	Tylenchorhynchus
Root knot	Meloidogyne	Sting	Ipibora
Root lesion	Pratylenchus		

Appendix 5 – Common insect pests of turfgrasses in Australia (Gardenet, 2006)

Order/Family	Scientific name	Common name	Symptoms
Coleoptera Curculionidae	Listronotus bonariensis	Argentine stem weevil	Yellow mottling of grass, dispersed dead yellow patches of turf. The maggot may be seen in the crown of the plant. Usually starts on the edges of turf.
	Sphenophorus brunipennis	Billbug	Damage occurs primarily from November through to January. Infected turf will turn yellow initially and then brown as the larvae chew through stolons and rhizomes. The turf can easily be removed from the soil similar to scarab beetle damage. A second generation can occur in February if environmental conditions are favourable.
Scarabaeidae	Aphoditus tasmaniae and Adoryphorus couloni	Black headed cockchafer and Red headed cockchafer	Destruction of the turf sward may occur in autumn through the feeding activity of large larvae when these are present at densities of 200–500 per m ² . Heavy infestations can result in the roots of turf plants being severed to such an extent that the turf can be rolled back in sheets.
	Cyclocephala signaticollis	Argentinian scarab	Grubs are very damaging root feeders. This causes the plant to weaken and may die in times of slight heat or water stress. This can be identified easily as affected plants lose their stability and wilt.
	Heteronychus arator	African black beetle	Irregular dead areas of turf eaten below ground.
	Sericesthis geminate and S. pruinose	Lawn or Pruinose scarab	Caterpillars feed on roots, loosening the sod, this way reducing the turf's ability to respond to stress.
Lepidoptera Hepialidae	Oncopora spp.	Sod webworm, Underground grass grubs	The removal of leaves in irregular areas. The presence of a fine silk web. Excretion on leaves.
Noctuidae	Agrotis infusa A. ipsilon A. munda	Common cutworm or Bogong moth Greasy cutworm Brown or Pink cutworm	The cutworm caterpillar strips the grass of its foliage, limiting the grasses' ability to grow. A more devastating action of the cutworm caterpillar, and the reason for its name, is its tendency to cut off plants above, at or below the soil surface, thus the caterpillar actually destroys far more than it consumes.
Pyralidae	Pseudaletia convvecta Persectania ewingii Spodoptera mauritia	Common armyworm Southern armyworm Lawn armyworm	Stripped area of leaves to soil level.
	Herpetogramma licarsisalis	Webworm	The larvae are foliage feeders and often leave behind the 'frass' (faecal pellets). The damage shows up as small dead patches of grass among unaffected grass.
Orthoptera Gryllotalpidae	Scapteriscus didactylus	Changa mole Cricket	Tunnelling near surface loosening the sod and damaging roots.

Appendix 6 – Common insect pests of pasture grasses in Australia

Order/Family	Scientific name	Common name	Symptoms
Acarina Penthaleidae	Halotydeus destructor Penthaleus major	Redlegged earth mite Blue oat mite	"Silvering" or "whitening" of the attacked foliage. Mites lacerate the leaf tissue of the plants and suck up the discharged sap. Most damaging impact on newly establishing pastures, greatly reducing seedling survival and retarding development (Gregg, 1997). Emerging seedlings may be totally wiped out by high mite numbers. Feeding on established plants reduces productivity and plant function and in turn pasture palatability to livestock (Moritz, 1995).
Collembola Sminthuridae	Sminthurus viridis	Lucerne flea	Grasses are not preferred hosts but can sustain damage. Succulent green cells of leaves are eaten by a process of rasping up the leaf tissue. Damage appears as small holes in the leaves (McDonald, 1995).
Hemiptera		Aphids	Honeydew.
Coleoptera Elateridae		Wire worms	Larvae attack germinating seeds and the stems of seedlings. Established pastures can support substantial populations with little effect, but newly seeded pastures can be severely damaged (Gregg, 1997).
Lepidoptera Hepialidae	Oncopera rufobrunnea and O. intricata	Underground grass grub or corbie	Young larvae are gregarious and live under silken webbing spun among plant debris. Older larvae construct vertical silk-lined tunnels from which they emerge to feed at night on leaves and stems. May cause severe yield loss and increased weed invasion. Prefer grasses, especially ryegrass (Gregg, 1997).
Noctuidae	Agrotis infusa A. ipsilon A. munda	Common cutworm or Bogong moth Black cutworm Brown cutworm	Caterpillars feed at night cutting through stems at ground level (Gregg, 1997).
	Mythimna convvecta Persectania ewingii Spodoptera mauritia	Common armyworm Southern armyworm Lawn armyworm	Feed on grass component of pastures (Gregg, 1997).
Scarabaeida	Aphoditus tasmaniae	Black headed cockchafer	Larvae emerge at night to forage on stems and foliage close to ground level. Damage is restricted to pastures at least 3-4 years old because the early larval stages feed on dung and other organic matter on the soil surface, which is often lacking in new pastures (Gregg, 1997).
	Adoryphorus couloni	Red headed cockchafer	Larvae feed on the roots of grasses and may cut the roots just below ground level (Gregg, 1997).
	Antitrogus morbillosus	Tableland pasture scarab	The extent of damage by the grubs is dependent on the level of infestation, seasonal conditions and grazing management. Usually worsened in shallow soil and where plant growth is restricted by dryness and overgrazing, which reduces the plants capacity to replace severed roots (Goodyer and Nicholas, 2005).
	Anoplognathus spp.	Christmas beetles	

Order/Family	Scientific name	Common name	Symptoms
		(Goodyer and	
	Heteronychus arator	Nicholas, 2005) African black beetle (Goodyer and Nicholas, 2005)	Larvae feed on roots and may sever them (Gregg, 1997).
	Othnonius batesi	Black soil scarab (Goodyer and Nicholas, 2005)	
	Rhopaea spp.	Pasture white grub (Goodyer and Nicholas, 2005)	
	Sericesthis geminata Sericesthis nigrolineata	Pruinose scarab Dusky pasture scarab	Larvae damage the roots. In severe infestations the roots can be entirely removed so that the pasture is torn up by grazing animals or birds searching for larvae. Less severe damage can cause water stress as a result of root loss and an inability to recover from defoliation (Gregg, 1997).
Tenebrionidae		Pie dish beetles	Larvae attack germinating seeds and the stems of seedlings. Established pastures can support substantial populations with little effect, but newly seeded pastures can be severely damaged (Gregg, 1997).
Orthoptera Acrididae	Austroicetes cruciata	Small plague grasshopper	Defoliates (Gregg, 1997).
	Brachyexarna lobipennis	Striped winged meadow grasshopper	Defoliates (Gregg, 1997).
	Chortoicetes terminifera	Australian plague locust	Defoliates (Gregg, 1997).
	Locusta migratoria	Migratory locust	Defoliates (Gregg, 1997).
	Nomadacris guttulosa	Spur throated locust	Defoliates (Gregg, 1997).
	Phaulacridium vittatutum	Wingless grasshopper	Defoliates (Gregg, 1997).
Gryllidae	Teleogryllus commodus	Black field cricket	Feeds selectively on young foliage of valuable introduced species. Leads to change in pasture composition and weed invasion. Perennial pastures are less susceptible than annual. Crickets are also a major cause of seed removal (Gregg, 1997).
		Ants	Problem is most severe for surface sowings. Ants have been reported to remove up to half the sown seed prior to emergence (Scott, 1997).
		Slugs	May be a problem in protected drill rows in the slots from direct drilling (Scott, 1997; Burnett, 2006b).
		Weevils	

Common Name	Scientific Name	Environmental Factors	Symptoms
Algae	Many algal species	High surface moisture, inadequate drainage and insufficient light and air movement.	Very primitive plant that is bluish-black in colour and seen on the ground surface. It may be peeled off when dry.
Brown patch	Rhizoctani solani	A lush lawn with high humidity, excessive soil moisture, inadequate drainage, excessive nitrogen and thatch levels. High levels of nitrogen can increase severity.	An irregular roughly circular shaped "smoke ring" in appearance. Up to 50 cm in diameter, leaves darken turning brown. This disease attacks the leaf.
Curvularia	Curvularia spp.	Warm /hot weather and high humidity.	Yellowing areas, fading to brown. Up to 6 cm in diameter.
Dollar spot	Rustroemia floccosum, syn: Sclerotina homeocarpa	Temperature range between 20- 27°C with high humidity. Excessive thatch levels with nitrogen deficient soils.	Fine mycelium can be seen on the leaves early in the morning dew. Small circular straw coloured areas up to 5 cm in diameter. These areas may merge together. This is a leaf disease.
Fairy ring	Marasmius oread, Basidiomycetes	Unknown, though a change in nitrogen source or levels may stimulate growth.	Ring of darker coloured, fast growing grass stimulated by the fungi breaking down organic material in mat or thatch.
Fusarium	Fusarium nivale	Temperature ranges of 5-15°C. With excessive growth and a cool change in temperature. Poor soil drainage and turf possibly deficient in potassium (K) and/or phosphate (P).	There are small pale yellow to pink areas. These areas may enlarge and may merge together. This is a leaf disease.
Helminthosporium	Helminthosporium spp.	Warm moist conditions, wet soils and excessive nitrogen levels.	Small brown brown-red spots or lesions appear on leaves. They enlarge on the leaves and stem turning them blackish- purple in colour. The disease spreads in areas up to 5 cm in diameter. This is a leaf and stem disease.
Leaf spot	Bipolaris sorokiniana (syn. Helminthosporium sativum)	Hot, humid conditions.	Small dark purple to black coloured spots on the leaf blade. As spots enlarge, the centres often turn light tan. At temperatures exceeding 30°C, distinct spots are often absent and the entire leaf blade appears dry and straw coloured. A warm weather disease affecting leaf sheaths, crowns and roots. Severe thinning of turfgrass stand can occur in a short time.
Pythium blight, Grease spot, Cottony blight	Pythium spp.	Warm weather disease, most destructive between 30-35°C. Survives well in water and poorly drained soils.	Circular reddish brown spots, 2-15 cm in diameter. Infected leaves appear wet, dark and active mycelium can be seen in the morning. Leaves then

Common Name	Scientific Name	Environmental Factors	Symptoms
			shrivel and turn reddish brown.
Red thread	Corticium fuciforme	Temperatures greater than 20°C with dry nutrient deficient soils i.e. nitrogen. Occurs on water soaked turf.	Dark leaves in small areas and is more obvious in longer grass. Red-pink mycelium is seen on leaves. A leaf disease.
Rolf's Disease	Sclerotium rolfsii	Warm humid weather, dry soils with excessive thatch. Generally the turf is unhealthy.	Yellowish "donut" shaped area that is 6-8 cm in diameter. There is visible regrowth in the centre. The spores (sclerotia) may be present, they are 2 mm in diameter and orange-black or brown in colour.
Slime moulds	Physarium spp. Mycilago spp.	Moderate temperature with excessively wet soils with poor drainage.	Bluish-grey or yellowish-white. They are unsightly as they mass on the leaf surface. They spread up to 25 cm across. This is a saprophytic disease.
Smut, Leaf smut, Stripped or Flag smut.	Ustilago spp.	Moist, humid warm weather.	Blackish areas on leaves may be rubbed off with fingers.
Spring dead spot	Leptosphaeria narmari and L. korrae		Circular dead spots first appear in spring. The spots appear in many of the same places and expand for 3 to 4 years. After the second or third year, the disease often appears as rings of dead grass, and may disappear after 3 or 4 more years.
Take all patch	Gaeumannomyces graminis	Common to newly established turfs, diminishing with the build up of antagonistic microorganisms in soil. Favours high pH.	Patch of bronzed or bleached turf 10-15 cm diameter grows to 1 m over a period of years. Black runner hyphae can be seen under base of leaf sheath, on crowns and roots.

Appendix 8 – Common pathogens of pasture in Australia

Common Name	Scientific Name	Hosts	Environmental Factors	Symptoms/Damage
Barley yellow dwarf virus (BYDV)		Perennial ryegrass		Reductions in establishment, competitiveness, persistence, productivity and pasture quality (Clarke and Eagling, 1994).
Blind seed disease	Gloeotinia granigena	Perennial ryegrass	Extended periods of leaf wetness in late November/early December and air temperatures of 15-25°C favour the highest incidence.	Seed heads. Infection usually results in seed death. Seed from heavily infected stands has reduced commercial value because of poor germination rates (Vargas, 2005).
Brown blight	Drechslera siccans (syn. Helminthosporium siccans)	Perennial ryegrass, Italian ryegrass	The leaf-spot, crown and root rot stages occur during the cool weather of spring and fall, few signs of the disease are evident during warm summer months (Vargas, 2005)	Small brown spots which enlarge and may develop white centres and/or dark brown streaks appear on the leaf blade. Heavy infections can result in severe thinning (Vargas, 2005).
	Drechslera spp.	Perennial ryegrass	· · · · · ·	Foliage (Clarke and Eagling, 1994).
	Helminthosporium spp.	Perennial ryegrass, Tall fescue		Foliage (Clarke and Eagling, 1994).
Crown rust	Puccinia coronata	Perennial ryegrass, Tall fescue	Long, cool, wet winters or times when grass is growing most slowly (Vargas, 2005).	Initially yellow spots develop on the leaves. Later orange-brown pustules 0.5 to 1 mm long occur on both leaf surfaces. The black spore (teliospore) stage can be found as early as September, and through the summer and autumn. Infected leaves die prematurely and severely rusted plants become stunted (Clarke, 1999b).
Damping off	Fusarium, Pythium and Phytophora		Cool, moist conditions.	Damage is often done prior to emergence resulting in failure of seeds to emerge. If emergence occurs, seedlings collapse.
Ergot	Claviceps spp.	Perennial ryegrass, Italian ryegrass, Tall fescue		Dark purple sclerotia (resting body) develop in place of a healthy seed and protrude from the glume. There appearance is preceded by ay honey dew stage which appears 2-3 weeks after flowering (Clarke, 1999a).
Fairy ring	Marasmius oread, Basidiomycetess	Perennial ryegrass, Italian ryegrass and Tall fescue	Unknown, though a change in nitrogen source or levels may stimulate growth.	Ring of darker coloured, fast growing grass stimulated by the fungi breaking down organic material in mat or thatch.

Common Name	Scientific Name	Hosts	Environmental Factors	Symptoms/Damage
Leaf spot	Bipolaris sorokiniana (syn. Helminthosporium sativum)	Perennial ryegrass, Tall fescue	Hot, humid conditions.	Small dark purple to black coloured spots on the leaf blade. As spots enlarge, the centres often turn light tan. At temperatures exceeding 30°C, distinct spots are often absent and the entire leaf blade appears dry and straw coloured. A warm weather disease affecting leaf sheaths, crowns and roots. Severe thinning of turfgrass stand can occur in a short time (Vargas, 2005). Reduces yield and quality of tall fescue and its ability to recover after grazing. Longer the infection, greater the losses (Clarke and Eagling, 1994).
Net-blotch	Drechslera dictyoides Paul and Parbery (syn. Helminthosporium dictyoides Drechslera)	Tall fescue	In spring spores are produced and can be transferred to healthy plant tissue, but the leaf-spot phase is inconspicuous at this time. Crown and root infection is thought to take place in spring, but symptoms are not expressed until the warm, dry weather of summer.	Initially occurs as irregular dark purple to black transverse strands, resembling dark threads drawn across the leaf creating a net-like appearance (Clarke and Eagling, 1994).
Ryegrass mosaic virus (RMV)		Perennial ryegrass		Reductions in dry weight yields (Clarke and Eagling, 1994).
Stem rust	Puccinia graminis	Perennial ryegrass, Italian ryegrass, Tall fescue		Reddish-brown elongated, 1-10 × 0.5-2 mm pustules with ragged edges occur on the leaves, stems and seed head. Later in the season the pustules may turn black when the fungus produces a resting stage spore (teliospore). These spores, in Australia, have no further part in the life cycle of the fungus (Clarke, 1999b). Seed heads - problem for graziers and seed producers (Clarke and Eagling, 1994).
Slime moulds	Physarium spp. Mycilago spp.	Perennial ryegrass, Italian ryegrass and Tall fescue	Moderate temperature with excessively wet soils with poor drainage.	Bluish-grey or yellowish-white. They are unsightly as they mass on the leaf surface. They spread up to 250 mm across. This is a saprophytic disease.

Appendix 9 – Weed Risk Assessment of Perennial Ryegrass

Species: *Lolium perenne* L. (perennial ryegrass)

Relevant land uses⁷:

1.1 Nature conservation.

3.2 Grazing modified pastures & 4.2 Grazing irrigated modified pastures.

3.3 Cropping & 4.3 Irrigated cropping

3.4 Perennial horticulture & 4.4 Irrigated perennial horticulture

3.5 Seasonal horticulture & 4.5 Irrigated seasonal horticulture.

3.6 Land in transition & 4.6 Irrigated land in transition.

5.5 Services.

Background: In Australia, perennial ryegrass occurs in a wide range of environments, as deliberate plantings and volunteer populations. Perennial ryegrass is cultivated on irrigated turf production farms where improved cultivars are grown to produce ready-to-lay turf for home gardens, public gardens and public amenities. Furthermore, perennial ryegrass is an important forage crop and soil stabiliser in irrigated and dryland farming systems. Historically, perennial ryegrass was introduced to Australia for lawns, and pasture. Perennial ryegrass becomes a weed when its range of growth extends beyond the boundaries of areas of deliberate plantings, which is facilitated by its vigorous growth habit and propensity for seed dispersal – primarily by humans. This weed risk assessment is for **non-GM perennial ryegrass volunteers**.

Weeds are usually characterised by one or more traits, such as rapid growth to flowering, high seed output, and tolerance of a range of environmental conditions. Further, they may cause harm to human or animal health, safety and/or the environment. Many weedy characteristics are viewed as agronomic advantages, leading modern perennial ryegrass cultivars to possess some traits associated with weeds; perennial ryegrass is an invasive weed in Australia. The vigorous growth habit of perennial ryegrass means that the plant can easily escape areas of cultivation, and can become an aggressive competitor

⁷ According to the Australian Land Use and Management Classification system version 8, published October 2016 (ABARES, 2016)

producing high numbers of fertile seed. Groves et al. (2003) described perennial ryegrass as a naturalised non-native plant species⁸ and classified it as a category 4⁹ weed in natural ecosystems and category 3T¹⁰ weed in agricultural ecosystems, Australia-wide – noting that *L. perenne* is potentially toxic owing to its association with toxic endophytic fungi.

The Weed Risk Assessment (WRA) methodology is adapted from the Australian/New Zealand Standards HB 294:2006 National Post-Border Weed Risk Management Protocol. The questions and ratings used in this assessment are based on the South Australian Weed Risk Management Guide (Virtue, 2004). Questions 1–5 relate to the invasiveness of perennial ryegrass and questions 6-11 relate to the impact of perennial ryegrass on relevant land use area. Unless cited, information in this appendix is sourced from the main document, *The Biology of* Lolium multiflorum *Lam. (Italian ryegrass)*, Lolium perenne *L. (perennial ryegrass) and* Lolium arundinaceum (*Schreb.)* Darbysh (tall fescue).

This risk assessment is consistent with previous assessments of *L. perenne* in Australia and elsewhere, described in Section 8.2, and provides a baseline for the assessment of GM *L. perenne*.

⁸ A species that has been introduced, become established and that now reproduces naturally in the wild without human intervention (Groves et al., 2003)

⁹ Naturalised and known to be a major problem at 3 or fewer locations within a State or Territory.

¹⁰ Naturalised and known to be a minor problem warranting control at 4 or more locations within a State or Territory, and toxic.

Invasiveness questions	Perennial ryegrass (L. perenne)
1. What is the ability of perennial ryegrass to establish amongst existing plants?	Rating: High in all relevant land uses Perennial ryegrass is not a domesticated crop; it is a vigorous and competitive plant that spreads primarily by seeds. Rapid establishment and early vigour is a desirable agronomic trait in perennial ryegrass, yet it leads to invasiveness – especially in bare or disturbed sites. In situations of heavy grazing or mowing the plant is not able to set seed so the primary mode of growth is vegetative via tillers. Perennial ryegrass is likely to have a high ability to establish amongst existing plants, with a high likelihood of establishing in densely vegetated situations of agricultural and plantation land uses due to its vigour and shade tolerance.
2. What is the tolerance of perennial ryegrass to average weed management practices in the land use?	Rating: Low in all relevant land uses In relevant land uses, perennial ryegrass has low tolerance of primary cultural methods of weed control. It is tolerant of mowing, heavy grazing, and mild-moderate fire, but is susceptible to broad spectrum herbicides used for the control of a range of weeds in agricultural production areas, in residential gardens, and in open urban and recreational spaces. Herbicide resistance/tolerant populations of perennial ryegrass have been observed; however, these populations are not widespread in Australia. Perennial ryegrass seed does not persist in the seed bank long term, weed control over several seasons will effectively eliminate perennial ryegrass infestation. In recreational, residential or roadside land uses, physical methods of weed control such as mowing, slashing, or burning will not adequately control perennial ryegrass. There may be instances where perennial ryegrass is a 'welcome' volunteer in these land uses, for instance in stabilisation of disturbed land, or in patchy turf used for human recreation.

1. Invasiveness of perennial ryegrass

Invasiveness questions	Perennial ryegrass (L. perenne)
3a. What is the time to seeding in the land uses?	Rating: < 1 year in all relevant land uses Perennial ryegrass develops to maturity and sets seed in one growing season.
3b. What is the annual seed production in the land use per square metre?	Rating: High in all relevant land usesPerennial ryegrass develops seed heads within one growing season. Peer reviewed estimates of seed production indicate values greater than 14,000 seeds m ⁻² in an agricultural setting. Far higher estimates may be arrived at by combining published values for seed production (35-183 g m ⁻²) and seeds per gram (1,000 seed weight 1.7-1.8 g). Taking into account lower plant densities in volunteer populations these
3c. Can perennial ryegrass reproduce vegetatively?	Rating: Low in all relevant land usesPerennial ryegrass is a bunchgrass, whose primary means of reproduction is by the dispersal of seeds.Certain genotypes of perennial ryegrass may grow vegetatively via stolons and rhizomes under certain conditions; however, these are not the primary mode of growth. In heavily grazed, mown, or burnt crops vegetative growth and reproduction is the key means of plant reproduction, but perennial ryegrass is not known to spread rapidly by vegetative propagation.
4. Long distance dispersal (more than 100 m)	by natural means in land uses
4a. Are viable plant parts dispersed by flying animals (birds and bats)?	Rating: Unlikely in all relevant land usesThere is no evidence that flying animals play a major role in dispersal of perennial ryegrass seeds or vegetative propagules, even though perennial ryegrass seeds are commonly consumed by birds (e.g. Buckingham et al., 2011). Perennial ryegrass seed is probably digested beyond viability within the gizzard of birds, preventing widespread distribution of seed – barring death of birds before digestion is

Invasiveness questions	Perennial ryegrass (L. perenne)
	complete. It may be possible that seed is dispersed by regurgitative feeding of young birds. Perennial ryegrass seeds lack structural features to facilitate their dispersal amongst the fur or feathers of animals and birds.
	The dispersal of viable vegetative propagules of perennial ryegrass by flying animals has not been reported in Australia or overseas. Further, the success of dispersal would depend on vegetative
	segments being transferred to a conducive location to take root, which requires adequate moisture availability and soil contact.
4b. Are viable plant parts dispersed by wild	Rating: Occasional in all relevant land uses
land-based animals?	Transport of seeds or vegetative propagules in the hooves or other body parts of land-based animals is possible, yet not likely as these propagules have no particular features such as burrs, barbs or sticky surfaces that would attach to fur or feet of animals. Also, dispersal by vegetative propagules would depend on stolon segments being transferred to a conducive location to take root, which requires adequate moisture availability and soil contact. Perennial ryegrass is not well suited to this method of propagation.
	Dispersal via the digestive system of wild land-based animals is expected, given that perennial ryegrass seeds are known to survive digestion in livestock, however there is little available data regarding ingestion by wild animals and subsequent seed survival. In addition, seed-hoarding rodents or marsupials might transfer seeds over relatively short distances. However, there are no reported incidences of this in Australia. Short-distance transport of viable perennial ryegrass seed by ants and earthworms has been observed, but this is unlikely to exceed 100 metres in travel distance.
4c. Are viable plant parts dispersed by	Rating: Occasional in all relevant land uses
water?	Perennial ryegrass produces numerous lightweight seeds. The morphology of perennial ryegrass seeds may facilitate transport in water, however there is no information demonstrating that this is a dispersal

Invasiveness questions	Perennial ryegrass (L. perenne)
	mechanism. Perennial ryegrass populations are present in riparian zones, which may be a result of
	water dispersal or may reflect that there is sufficient moisture for establishment after transport by
	other means
4d. Are viable parts dispersed by wind?	Rating: Occasional in all relevant land uses
	Dispersal of perennial ryegrass seeds by wind is possible, however there is little information to confirm
	this. Perennial ryegrass seed heads develop close to the ground (up to 20 cm flower spike). Seeds are
	light (approximately 1.7 mg/seed). Perennial ryegrass seeds have no morphological characteristics aiding wind dispersal.
5. Long distance dispersal (more than 100	m) by human means in land uses
5a. How likely is deliberate spread via	Rating: Common in most relevant land uses
people?	Perennial ryegrass has been spread deliberately throughout the world by humans (as well as other
	natural means) as a species for lawns in gardens and public areas, ground cover, and pastures in
	temperate regions.
	The species is deliberately spread by people through the cultivation of perennial ryegrass turf for
	installation on properties potentially up to several hundreds of kilometres away from the turf farm. It is
	also deliberately spread throughout Australia, and the world, as certified seed for sowing in widely
	distributed pastures.
5b. How likely is accidental spread via	Rating: Common in all relevant land uses
people, machinery and vehicles?	Accidental dispersal of plant fragments and seed could occur during the deliberate transfer of perennial
	ryegrass from one pasture to another. The turf market involves transporting 'live' turf from the turf
	farm to the customer, where there could be an opportunity for fragments to break off turf rolls and

Invasiveness questions	Perennial ryegrass (L. perenne)
	establish where they land.
	Spillage during seed transport is a major source of volunteer populations along transport routes, and in
	agricultural supply centres. Further, plant fragments and seed produced by perennial ryegrass are
	reported to be widely distributed by agricultural vehicles and machinery.
5c. How likely is spread via contaminated produce?	Rating: Occasional in all relevant land uses
produce:	There is a strong perennial ryegrass seed production industry in Australia. Aside from crops specifically
	grown for perennial ryegrass seed, perennial ryegrass seed is an occasional contaminant of other types
	of seed crop – e.g. wheat or other ryegrasses. Perennial ryegrass volunteer populations are typically
	well controlled in relevant land uses.
	Perennial ryegrass is unlikely to spread vegetatively by contamination of other produce.
5d. How likely is spread via domestic/farm	Rating: Common in all relevant land uses
animals?	Fragments of plants could be spread in mud on animal hooves as animals are moved from one paddock
	to another, to a feedlot, or to other farms. This pathway is limited as perennial ryegrass seeds and
	vegetative propagules have no particular features such as burrs, barbs or sticky surfaces that would
	attach to fur or feet of animals. Nonetheless, perennial ryegrass seeds have been observed entangled in
	sheep wool and transported for 1-2 months. Vegetative dispersal would depend on stolon segments
	being transferred to an appropriate location to take root, which requires adequate moisture availability
	and soil contact.
	Perennial ryegrass biomass and seeds are consumed widely as fodder by ruminants. Whilst the
	optimum fodder quality occurs during early growth before seed has set, grazing mature seed-bearing
	pastures to stubble is a widespread land management practise. Dispersal of viable perennial ryegrass
	seed has been observed by endozoochory, and a small proportion of perennial ryegrass seed can be

Invasiveness questions	Perennial ryegrass (L. perenne)
	viable after digestion by ruminants so this likely forms a common pattern of dispersal.

2. Impact of perennial ryegrass

Impact questions	Perennial ryegrass (L. perenne)
6. Does perennial ryegrass reduce the establishment of desired plants?	Rating: 10-50% reduction in all relevant land uses
	Once established, perennial ryegrass forms a thick dense sward that can reduce the establishment of, and
	for some species displace, other plants. This is a desirable trait in turfgrass and pasture, however it inhibits
	the establishment of desirable species outside its intended range. Reports in Australia suggest that
	perennial ryegrass can displace native species such as spiny peppercress (Lepidium aschersonii) and the
	red darling pea (Swainsona plagiotropis) (Queensland Government, 2017).
	Perennial ryegrass is allelopathic, reducing the establishment of some plant species in the close vicinity.
7. Does perennial ryegrass reduce the yield	Rating: 10-25% reduction in all relevant land uses
or amount of desired vegetation?	Perennial ryegrass reduces the yield of desired vegetation when established. It is vigorous and can out- compete native or other desirable vegetation. Perennial ryegrass establishes rapidly and is especially competitive in disturbed land. Nonetheless, commonly used weed management practices would lead to low density of perennial ryegrass volunteers in relevant land uses, and hence would cause a moderate to low reduction in yield of desired vegetation.
8. Does perennial ryegrass reduce the quality	Rating: Low in all relevant land uses

Impact questions	Perennial ryegrass (L. perenne)
of products or services obtained from the land use?	 In general perennial ryegrass is a desirable grass species for turf and fodder due to its vigour and nutritional qualities. However, the establishment of volunteer perennial ryegrass populations could reduce the quality of products and services in some land uses. On land used for nature conservation, perennial ryegrass can reduce biodiversity – particularly of defined native species. In turf cultivation it may enter sod production areas of other turf species and reduce product quality. In pasture areas perennial ryegrass volunteer populations may displace more nutritious or otherwise preferred pasture species or lead to allelopathic reduction in yield of desired pasture species. Perennial ryegrass volunteers may contaminate seed harvests in other similar crops, however effective management of perennial ryegrass volunteer populations limits this possibility.
9. What is the potential of perennial ryegrass to restrict the physical movement of people, animals, vehicles, machinery and/or water?	Rating: none in all relevant land uses Perennial ryegrass is a low growing plant, and does not restrict the physical movement of people, animals, vehicles, machinery, and water even when densely planted in an agricultural setting. Volunteer populations, being less densely planted than agricultural crops would not restrict the movement of people, animals, vehicles or machinery. Dense swards that have had the opportunity to develop in seasonal channels or waterways may marginally slow the flow of water, but perennial ryegrass would not survive in permanently or frequently inundated waterways.
10. What is the potential of perennial ryegrass to negatively affect the health of animals and/or people?	Rating: Minor- intermediate in all relevant land usesThe pollen of perennial ryegrass is considered a major human allergen worldwide (Scala et al., 2010), and perennial ryegrass produces wind-carried pollen in vast quantities. The Australian population has high prevalence of perennial ryegrass pollen sensitivity, and the population comes into frequent contact with perennial ryegrass pollen due to its prevalence in agricultural land adjacent to population centres. Pollen- sensitivity may lead to serious complications such as asthma in susceptible individuals.Perennial ryegrass often contains potentially toxic endophytic fungi. These fungi produce a range of toxic

Impact questions	Perennial ryegrass (L. perenne)
	metabolites involved in biotic and abiotic stress tolerance. These toxins accumulate in the plant at various
	times of the year under certain conditions and can lead to ryegrass toxicoses in grazing livestock. Ryegrass
	toxicoses such as "ryegrass staggers" are rarely fatal to livestock. Grazing on a low density of perennial
	ryegrass volunteers is unlikely to lead to significant toxicity.
11. Major positive and negative effects of per	rennial ryegrass on environmental health in the land use
11a. Does perennial ryegrass provide food	Rating: Minor or no effect in all relevant land uses
and/or shelter for pests and pathogens in	A large range of pathogens and pests are reported to affect perennial ryegrass. Volunteer perennial
the land use?	ryegrass populations, being outside active pathogen management, might be expected to harbour
	pathogens and pests that could affect perennial ryegrass pastures, or other adventitious targets.
	Diseases harbored in perennial ryegrass volunteer populations may affect other grasses and some, e.g.
	powdery mildew, may affect multiple plant species; most of these pests and pathogens are specific to
	closely related crops. The vast majority of perennial ryegrass is grown in pastures, so it might be expected
	that the risk of pathogen spread from volunteer populations is outweighed by the risk of pathogen spread
	from minimally-managed pastures.
11b. Does perennial ryegrass change the fire	Rating: Minor or no effect in all relevant land uses
regime in the land use?	Volunteer perennial ryegrass may slightly increase fire risk in any of the land uses it may occur. Perennial
	ryegrass establishes rapidly, and rapidly reaches maturity before becoming senescent, leading to the
	build-up of tinder in areas with significant perennial ryegrass populations – increasing fire risk. Research
	suggests that areas where significant alien populations of perennial ryegrass have displaced native
	vegetation may become more fire-prone (D'Antonio and Vitousek, 1992).
11c. Does perennial ryegrass change the	Rating: Minor or no effect in all relevant land uses
nutrient levels in the land use?	As with all plants, perennial ryegrass will use soil nutrients for growth. Perennial ryegrass is an efficient

Impact questions	Perennial ryegrass (L. perenne)
	scavenger of nitrogen and may deplete this resource to low concentrations preventing access by other
	plants. While perennial ryegrass in cultivation responds well to fertilisation in an agricultural setting, its
	growth rate is much slower under nutrient limitation and will likely have minor effects on other species in
	the area or subsequent species planted in the same area.
11d. Does perennial ryegrass affect the	Rating: Minor or no effect in all relevant land uses
degree of soil salinity in the land use?	
	As a ruderal weed, volunteer perennial ryegrass may have a small positive effect on soil salinity, i.e.
	reducing the extent of percolation of water and salt transport through the soil profile by active
	transpiration in an otherwise non-vegetated area.
11e. Does perennial ryegrass affect the soil	Rating: Minor positive effect in some land uses
stability in the land use?	Perennial ryegrass has an extensive fibrous root system that has a positive effect in stabilizing light-
	textured or sandy soils, and in stabilizing recently disturbed soils.
11f. Does perennial ryegrass affect the soil	Rating: Minor or no effect in all relevant land uses
water table in the land use	As a ruderal weed, perennial ryegrass may have a small positive effect on the soil water table by reducing
	the extent of percolation of water through the soil profile by active transpiration in an otherwise non-
	vegetated area.
11g. Does perennial ryegrass alter the	Rating: Minor or no effect in all relevant land uses
structure of nature conservation by adding a	
new strata level?	In relevant land uses perennial ryegrass is a similar size to existing grass species and is not expected to add
	a new strata level.