Emergence of pasture grasses from different sowing depths: importance of coleoptile and mesocotyl width

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Abstract

Inter-specific relationships between mean seed weight, coleoptile and mesocotyl length and width and emergence from 10 and 30 mm sowing depths were examined for prairie grass (*Bromus willdenowii* Kunth., mean seed weight 12.2 mg), annual ryegrass (*Lolium multiflorum* Lam., 5.10 mg), tall fescue (*Festuca arundinacea* Schreb., 2.60 mg), perennial ryegrass (*Lolium perenne* L., 1.71 mg), cocksfoot (*Dactylis glomerata* L., 0.65 mg) and timothy (*Phleum pratense* L., 0.48 mg) under controlled environment conditions. At 10 mm sowing depth, emergence percentage was 100% for prairie grass and annual ryegrass and 98%, 89%, 66% and 49% for tall fescue, perennial ryegrass, cocksfoot and timothy, respectively. At 30 mm sowing depth, emergence percentage was > 90% for the two largest seeded grasses: values for the other species decreased with seed weight to a low of 21%for timothy. For all species, at 10 and 30 mm sowing depths, coleoptile + mesocotyl length was greater for emerged plants than for non-emerged plants but across species, emergence % was not significantly correlated with coleoptile + mesocotyl length. Across species, there were significant positive correlations between emergence percentage and coleoptile and mesocotyl width.

For three lots of timothy with mean seed weights of 0.81 mg, 0.43 mg and 0.26 mg, emergence percentage decreased with decreased seed weight from 69% to 23% at 10 mm sowing depth and from 41% to 13% at 30 mm sowing depth. Coleoptile + mesocotyl length and coleoptile and mesocotyl width decreased with decreased seed weight at 10 and 30 mm sowing depth. For emerged and non-emerged plants of cocksfoot and timothy regardless of seed size, mean coleoptile + mesocotyl length was > 10 mm at 10 mm sowing depth. It is concluded that at 10 and 30 mm sowing depth, coleoptile + mesocotyl length is a factor determining emergence percentage within grass species, but that decreased emergence percentage with decreased seed weight across species is not due to decreased coleoptile + mesocotyl length. It is proposed that decreased emergence percentage with decreased seed weight across species at 10 and 30 mm sowing depth and for seed lots of timothy at 10 mm sowing depth is primarily due to decreased coleoptile and mesocotyl width resulting in decreased shoot strength and hence reduced ability to penetrate the substrate.

Additional key words: Bromus willdenowii, Dactylis glomerata, Festuca arundinacea, Lolium multiflorum, Lolium perenne, Phleum pratense

Introduction

Perennial ryegrass (*Lolium perenne* L.) is the main pasture grass sown in perennial pastures in New Zealand (Langer, 1990). Alternative species which are commercially available and, which under certain conditions can have an advantage over perennial ryegrass with respect to productivity and/or palatibility, include cocksfoot (*Dactylis glomerata* L.) and timothy (*Phleum pratense* L.) (Charlton and Thom, 1984; Rumball, 1984; Langer, 1990). However, both of these grasses have disadvantages with respect to perennial ryegrass in that they often show lower emergence and establishment percentages and are slower to establish (Langer, 1990; Porter *et al.*, 1993).

Emergence percentage, establishment percentage and rate of establishment of a particular grass are dependent on environmental factors and genotypic and phenotypic characteristics of the seed and seedling (Hill *et al.*, 1985; Askin, 1990). Mean seed weight is substantially less for cocksfoot and timothy than for perennial ryegrass and there is some evidence that this is a factor causing low emergence and establishment percentages of these grasses. For example, in a study of seven pasture grass species which included perennial ryegrass, cocksfoot and timothy, emergence and establishment percentages were found to be positively correlated with mean seed weight (Porter *et al.*, 1993). Also, although emergence percentage for all species decreased with increased sowing depth, the magnitude of the decrease was greater with smaller seeded grasses.

Usually, on shallow sowing (around 10 mm), the primary leaf of a developing grass seedling extends within a protective sheath-like structure, the coleoptile, to a point close to the soil surface. The primary leaf then emerges from a pore near the tip of the coleoptile and, with little impedence to growth, emerges from the soil (Langer, 1990). If the primary leaf emerges underground, it is less likely to reach the surface as it is less rigid than the coleoptile and folds more easily (Andrews *et al.*, 1991; Hines *et al.*, 1991). The coleoptile is limited in growth and for some grass species at deeper sowings, the young shoot within the coleoptile can be carried to the soil surface by elongation of the mesocotyl, a subcoleoptile internode (Newman and Moser, 1988; Robson *et al.*, 1988). Potential mesocotyl elongation has been reported to increase with seed size across grass species (Robson *et al.*, 1988). Thus, lower emergence percentage of small seeded grasses in comparison with larger seeded grasses may be due to them having a shorter coleoptile + mesocotyl. The primary objective of the present study was to test this possibility. Also, the importance of within lot seed weight with regard to emergence from different sowing depths was assessed for timothy, the species in the study with the least mean seed weight.

Materials and Methods

Seed of prairie grass (*Bromus willdenowii* Kunth. cv. Grasslands Matua, mean seed weight 12.2 mg), annual ryegrass (*Lolium multiflorum* Lam. cv. Grasslands Tama, 5.10 mg), tall fescue (*Festuca arundinacea* Schreb. cv. Grasslands Roa, 2.60 mg), perennial ryegrass (cv. Grasslands Nui, 1.71 mg), cocksfoot (cv. Grasslands Wana, 0.65 mg) and timothy (cv. Grasslands Kahu, 0.48 g) was obtained from Cudden and Stewart Ltd, Christchurch, N.Z.

In experiment 1, seed of all six species was sown at 10 and 30 mm depth in 0.8 litre pots (10 seeds per pot) containing a vermiculite/perlite/sand (1:1:1 by volume) mix, soaked in a basal nutrient solution containing 5 mol/m^3 potassium nitrate (Andrews *et al.*, 1989). There were five replicate pots per treatment. The pots were maintained at 12 ± 1°C in the dark and flushed with nutrient solution every 3 days. Emerged plants were counted each week after planting. Emergence from each pot was taken as complete either when all plants had emerged or when no further plants emerged during a three week period. When counting had finished, coleoptile and mesocotyl length and width were determined for all emerged plants and all non-emerged plants which were recovered.

In experiment 2, timothy seed was sieved to produce seed lots with a mean seed weight of 0.81 mg, 0.43 mg and 0.26 mg. Seed of each lot was sown in pots (10 seeds per pot) at 10 and 30 mm as in experiment 1. There were six replicate pots per treatment. Growth conditions and measurements taken were as in experiment 1.

Both experiments were of completely randomised design and were repeated once as described. Germination percentage was determined for seed of all species including the different lots of timothy at 15°C in the dark. Six replicates of fifty seeds of each seed type were placed in petri dishes containing 10 Whatman no 1 filter papers maintained saturated with nutrient solution. Counts of germinated seed were taken weekly until germination percentage did not change. Germination percentage was > 80% for all species except tall fescue (61%). All values determined for emergence percentage were standardised with respect to germination percentage (i.e., emergence percentage presented = actual emergence % ÷ the proportion of seeds which germinated) prior to data analysis.

An analysis of variance was carried out on all data with species and sowing depth as factors in Expt 1 and seed lot and sowing depth as factors in Expt 2. An arcsine transformation was carried out on emergence % data prior to analysis. Ranking of species at each sowing depth in Expt 1 is on the basis of a LSD ($p < 0.05$) value derived from an analysis of variance carried out on data from that particular sowing depth. A general linear model analysis (Minitab Version 12, Minitab Inc., Pennsylvania USA) was carried out on data from Expt 1 using emergence % (arcsine transformed data) as the response variable, depth as a factor and coleoptile + mesocotyl length, coleoptile width and mesocotyl width as covariates (Table 2). All effects described in both experiments have a probability $p < 0.05$ and were obtained in the repeat experiments.

Results and Discussion

In a study of seven pasture grass species which included the six species used here, emergence percentage from 10, 30 and 60 mm sowing depth was positively correlated with mean seed weight (Porter *et al.*, 1993). Also, although emergence percentage for all species decreased with increased sowing depth, the magnitude of the decrease was greater for smaller seeded grasses. Similar results were obtained in experiment 1 (Table 1).

Table 1. Emergence percentage and coleoptile and mesocotyl length and width of six pasture grass species sown at different depths. Variability shown is SEM, n = 5. Value in brackets below grass species is mean seed weight.

Species	Sowing depth (mm)	Emergence (%)	Coleoptile + mesocotyl length (mm)		Coleoptile length (mm)		Coleoptile width (mm)		Mesocotyl width (mm)	
			emerged	non-emerged	emerged	non-emerged	emerged	non-emerged	emerged	non-emerged
Prairie grass	10	100 ±2.8	22.0 ±0.91	-	22.0 ±0.91	-	0.98 ±0.012	-	-	-
(12.2 mg)	30	93 ±5.2	24.1 ±0.45	12.2 ±1.08	24.1 ±0.45	12.2 ±1.08	1.03 ±0.030	1.02 ±0.058	-	-
Annual ryegrass	10	101 ±2.5	44.1 ±2.61	-	36.2 ±2.08	-	0.74 ±0.013	-	0.63 ±0.016	-
(5.10 mg)	30	96 ±4.6	47.9 ±1.59	15.0 ±1.00	34.5 ±1.17	15.0 ±1.00	0.74 ±0.032	0.75 ±0.005	0.68 ±0.022	-
Tall fescue (2.60	10	98 ±5.3	19.1 ±0.61	-	18.1 ±0.54	-	0.63 ±0.022	-	0.66 ±0.014	-
mg)	30	83 ±2.9	33.7 ±1.66	18.0 ±0.79	24.7 ±1.10	14.7 ±0.67	0.72 ±0.021	0.47 ±0.068	0.62 ±0.017	0.50 ±0.089
Perennial ryegrass	10	89 ±2.9	20.9 ±0.94	11.3 ±1.13	18.1 ±0.79	11.3 ±1.13	0.47 ±0.029	0.37 ±0.028	0.51 ±0.016	-
(1.71 mg)	30	78 ±3.1	29.0 ±2.56	16.0 ±3.03	22.2 ±1.33	15.4 ±2.84	0.58 ±0.020	0.60 ±0.044	0.52 ±0.020	0.54 ±0.024
Cocksfoot	10	66 ±2.4	16.5 ±0.91	14.7 ±0.33	13.6 ±0.93	12.7 ±0.33	0.40 ±0.025	0.33 ±0.026	0.35 ±0.031	0.30 ±0.012
(0.65 mg)	30	28 ±3.7	26.7 ±1.73	23.1 ±0.82	19.3 ±1.38	20.1 ±0.82	0.53 ±0.023	0.41 ±0.038	0.43 ±0.018	0.38 ±0.022
Timothy	10	49 ±3.1	19.7 ±2.57	16.4 ±1.71	15.9 ±1.86	15.1 ±0.71	0.38 ±0.034	0.34 ±0.031	0.37 ±0.019	0.34 ±0.019
(0.48 mg)	30	21 ±3.8	27.3 ±1.80	21.4 ±2.06	18.3 ±0.92	13.6 ±1.36	0.43 ±0.036	0.37 ±0.020	0.41 ±0.021	0.36 ±0.025

At 10 mm sowing depth, emergence percentage was around 100% for the three largest seeded species (prairie grass, annual ryegrass and tall fescue) but values for the other species decreased with decreased seed weight thereafter to a low of 49% for timothy. Similarly, at 30 mm sowing depth, emergence percentage was > 90% for the two largest seeded grasses but values decreased consistently with decreased seed weight thereafter to a low, in this case, of 21% for timothy. The findings here for timothy and cocksfoot that a substantial proportion of seedlings fails to emerge from 10 mm sowing depth and that the major proportion of seedlings does not emerge from 30 mm sowing depth are consistent with field results. For example, in six hand sown field experiments, mean emergence percentage from 30 mm sowing depth was 31% and 20% for cocksfoot and timothy respectively (Porter *et al.*, 1993; Andrews unpub.)

For wheat (*Triticum aestivum* L.) which appears not to show mesocotyl elongation, emergence percentage from deeper sowings is positively correlated with coleoptile length within and between cultivars (Hines *et al.*, 1991). Within cultivars, coleoptile length is positively correlated with seed weight in wheat (Cornish and Hindmarsh, 1988). For pasture grasses, potential mesocotyl elongation has been reported to increase with seed size across species (Robson *et al.*, 1988). The primary objective of the present study was to determine if the lower emergence percentage of small seeded grasses in comparison with large seeded grasses is due to them having a shorter coleoptile + mesocotyl. In the initial and repeat experiment 1, and in two field experiments which included 10, 30 and 50 mm sowing depth treatments, prairie grass did not show mesocotyl extension (Table 1; Andrews unpub.). With the exception of prairie grass, at least some plants of all species showed mesocotyl extension at all sowing depths in experiment 1 (Table 1). Also, for all species except prairie grass, the proportion of coleoptile + mesocotyl length as mesocotyl increased with increased sowing depth. The results obtained in experiment 1 indicate that coleoptile + mesocotyl length is an important factor determining emergence percentage within grass species for two reasons (Table 1). Firstly, mean coleoptile + mesocotyl length for tall fescue, perennial ryegrass, cocksfoot and timothy increased with increased sowing depth. Secondly, for all species at all sowing depths, coleoptile + mesocotyl length was greater for emerged than non emerged plants. Nevertheless, across species, emergence percentage was not significantly correlated with mean coleoptile + mesocotyl length and low emergence of cocksfoot and timothy relative to the other species cannot be explained by differences in coleoptile + mesocotyl length (Table 2). For example, at 10 mm sowing depth, coleoptile + mesocotyl length was similar for tall fescue and timothy but emergence % was substantially lower for timothy. Also, at 10 mm sowing depth, mean coleoptile + mesocotyl length for emerged and non-emerged cocksfoot and timothy seedlings was > 10 mm. At 30 mm sowing depth, mean coleoptile + mesocotyl length was similar for prairie grass (the largest seeded grass) and cocksfoot and timothy but emergence percentage was 93% for prairie grass and only 26% and 21% for cocksfoot and timothy respectively. However, across species, there was a significant positive correlation between emergence % and coleoptile width (Tables 1,2). There was also a significant positive correlation between emergence % and mesocotyl width (N.B. prairie grass was not included in the analysis). It seems likely that shoot strength will increase with increased coleoptile and mesocotyl width. It is proposed that low shoot strength resulting in reduced ability to penetrate the substrate is the primary factor causing low emergence percentage of cocksfoot and timothy relative to the other grasses at 10

Table 2. Results of a General Linear Model analysis of data from Expt 1. Emergence % (arcsine transformed data) was used as the response variable, depth was a factor and coleoptile + mesocotyl length (C+M length), coleoptile width (C width) and mesocotyl width (M width) were covariates:

Source	DF	Seq SS	Adj SS	Adj MS	F	P
C+M length	1	1.9650	0.0039	0.0039	0.13	0.715
C width	1	3.0604	0.1319	0.1319	4.59	0.038
M width	1	0.9901	0.3532	0.3532	12.29	0.001
Sowing depth	1	1.7937	1.7937	1.7937	62.41	0.000
Error	42	1.2071	1.2071	0.0287		
Total	46	9.0163				

Table 3. Emergence percentage and coleoptile and mesocotyl length and width of three timothy lots of different mean seed weight sown at different depths. Variability shown is SEM, n=6.

Seed weight (mg)	Sowing depth (mm)	Emergence (%)	Coleoptile + mesocotyl length (mm)		Coleoptile length (mm)		Coleoptile width (mm)		Mesocotyl width (mm)	
			emerged	non-emerged	emerged	non-emerged	emerged	non-emerged	emerged	non-emerged
0.81	10	69 ±5.2	18.8 ±0.60	16.0 ±0.73	15.7 ±0.39	14.0 ±0.63	0.41 ±0.008	0.33 ±0.023	0.43 ±0.016	0.42 ±0.034
0.43		51 ±6.1	16.7 ±0.52	15.0 ±0.44	13.8 ±0.48	13.0 ±1.11	0.35 ±0.016	0.30 ±0.021	0.38 ±0.023	0.30 ±0.021
0.26		23 ±3.4	15.4 ±1.11	11.5 ±0.50	12.4 ±0.45	10.1 ±0.35	0.33 ±0.028	0.30 ±0.001	0.37 ±0.035	0.28 ±0.014
0.81	30	41 ±4.0	29.5 ±1.98	20.4 ±2.04	21.0 ±0.93	17.2 ±1.47	0.43 ±0.016	0.43 ±0.023	0.43 ±0.017	0.43 ±0.023
0.43		21 ±5.1	21.8 ±1.16	16.1 ±0.43	16.8 ±0.25	13.1 ±0.68	0.38 ±0.014	0.35 ±0.017	0.40 ±0.014	0.35 ±0.025
0.26		13 ±4.7	16.5 ±0.50	12.0 ±0.58	13.3 ±0.72	10.5 ±1.44	0.34 ±0.025	0.34 ±0.005	0.35 ±0.038	0.33 ±0.005

and 30 mm sowing depths. Experiments will be carried out to test this proposal.

Emergence percentage within grass species has also been found to be dependent on seed weight. For example, the decrease in emergence percentage of perennial ryegrass with increased sowing depth from 12.5 to 70 mm was greater for small (0.9 - 2.1 mg) seeds than for large (2.5 - 3.6 mg) seeds (Arnott, 1969). Similarly for timothy in experiment 2, emergence percentage decreased with decreased seed weight at 10 and 30 mm sowing depth. Previously, it was reported that within grass species, potential mesocotyl extension is similar for seedlings from different seed weights but mesocotyl width decreases with decreased seed weight (Robson *et al.*, 1988). For timothy at 10 and 30 mm sowing depth in experiment 2, coleoptile + mesocotyl length and coleoptile and mesocotyl width decreased with decreased seed weight (Table 3). In view of the results obtained in Expt 1, it seems likely that decreased coleoptile + mesocotyl length and decreased coleoptile and mesocotyl width are factors resulting in decreased emergence percentage of timothy lots with decreased seed weight. However, at 10 mm sowing depth, mean coleoptile + mesocotyl length for emerged and non-emerged plants was greater than sowing depth, regardless of seed weight. It is proposed that the decrease in coleoptile and mesocotyl width with decreased seed weight results in a decrease in shoot strength and that this is the primary factor causing decreased emergence percentage of timothy with decreased seed weight at 10 mm sowing depth.

Conclusions

1. Across grass species and for different lots of timothy, there is a significant positive correlation between emergence percentage and mean seed weight at 10 and 30 mm sowing depths.
2. Coleoptile + mesocotyl length is a factor determining emergence percentage within grass species at 10 and 30 mm sowing depths.
3. Decreased emergence percentage with decreased seed weight across grass species at 10 and 30 mm sowing depth is not due to decreased coleoptile + mesocotyl length.
4. Decreased emergence percentage with decreased seed weight across species at 10 and 30 mm sowing depth and for seed lots of timothy at 10 mm sowing depth is likely to be due primarily to decreased coleoptile and mesocotyl width resulting in decreased shoot strength and hence reduced ability to penetrate the substrate.

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