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Full Length Article

Influence of Different Pre-Sowing Treatments on Seed Dormancy Breakdown, Germination and Vigour of Red Clover and Italian Ryegrass

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Abstract

A two-year study was conducted using seed of six cultivars of *Trifolium pratense* (red clover) and three cultivars of *Lolium multiflorum* (Italian ryegrass), to test the effect of different treatments including exposure to 50% H_2SO_4 for10, 30, 60, 90 min; to 80^{o}C temperature for 10, 30, 60, 90 min; gibberellic acid (GA_3) at 250, 500, 1000 and 1500 mg L^{-1}; and potassium nitrate (KNO_3) at 0.1, 0.2, 0.35 and 0.5%. Germination, dormancy and vigour of both red clover and ryegrass were assessed. In case of *T. pratense* seed, there was a significant ($p \leq 0.05$ or $p \leq 0.01$) interaction between all the cultivars and treatments. A significant ($p \leq 0.05$) interaction was also observed between the *L. multiflorum* cultivars and H_2SO_4 including temperature treatments. However, no significant interactions were noted between the GA_3 or KNO_3 treatment, germination and vigour. The results showed that by selecting the optimal treatment (H_2SO_4 30' or H_2SO_4 60'*T. pretense* and GA_3 1000 mg L^{-1} *L. multiflorum*) for a particular cultivar it is possible to significantly improve germination and vigour, and thus ensure technologically more effective and economical establishment of a grass-legume mixture of *L. multiflorum* and *T. pratense*.

Keywords: *T. pratense*; *L. multiflorum*; Germination; Seed; Vigour

Introduction

In central and south-eastern Europe, *T. pratense* (red clover) and *L. multiflorum* (Italian ryegrass) are important high-quality forage crops (Jurmescu *et al.*, 2012). These are grown both as main crops and in mixtures with other species. The latter approach is prevalent and offers many advantages (Simić *et al.*, 2011). Numerous studies in this region on both of these forage species report highly variable morphological properties, consequent forage quality and yield fluctuations (Petrović *et al.*, 2014; Tomić *et al.*, 2015). However, very few studies provide results pertaining to the variability of seed quality and viability. According to Zimmermann *et al.* (1998), seeds of perennial forage legumes (*Medicago sativa*, *Lotus corniculatus*, *Trifolium repens*, *T. pratense* and *Melilotus albus*) are hard and dormant. In the case of red clover, the specific morphology, anatomy of the seeds and the chemical composition of the seed coat (polyphenols, lignin, tannin, cellulose pectin and hemicellulose) prevent the entry of water and gasses and thus thwart germination, especially of dark seeds (Galussi and María, 2017; Velijević *et al.*, 2017).

The dormancy of perennial grass seeds is controlled by mechanisms within the structure that covers the embryo and/or mechanisms within the embryo, or a combination of the two (Adkins *et al.*, 2002). Seed dormancy generally depends on the forage grass species (Stanisavljević *et al.*, 2015), but also varies in the same species (Stanisavljević *et al.*, 2012). Under field conditions, dormant seeds germinate later and cannot compete with already developed plants, such that don't have considerable effect on crop establishment (Van Assche *et al.*, 2003). Germination and vigour play a decisive role in crop establishment and the achievement of projected species ratio (s) of crop mixtures. Consequently, the objective of the present study was to determine the variability of *T. pratense* and *L. multiflorum* cultivars with regard to germination, dormancy and vigour, by applying different seed treatments and selecting the optimal treatment that will improve germination and vigour, and thus facilitate the achievement of the projected ratio of the grass-legume mixture of *L. multiflorum* and *T. pratense*.

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Materials and Methods

Experimental Details and Treatments

Experimental material: The seed for the study were obtained from conventional seed crops and the same parcels of land in Serbia and Bosnia & Herzegovina (42^o 34'-45^o 51' N, 17^o 35'-21^o 55' E, 65-598 m.a.s.l.), after standard (combine) harvesting. The seed of the red clover diploid cultivars (cv. Sana, Petnica, Una, Nike, K-17, and K-39) were collected in the first half of August (second cut), and of the Italian ryegrass diploid cultivars (cv. Aubade, Draga, and K-13) in the latter half of May (first cut). After harvesting, the seed were dried to 12% moisture contents, processed, placed in paper bags, and stored under normal conditions in a seed warehouse: *T. pratense* for about two months and *L. multiflorum* for approximately four months.

Treatments: At the end of September, which is the sowing time of the studied cultivars, the seed were treated with H_2SO_4 (50%) for 10 (C1), 30 (C2), 60 (C3) and 90 (C4) min; exposed to a temperature of 80^{o}C for 10 (D1), 30 (D2), 60 (D3) and 90 (D4) min ('); treated with gibberellic acid (GA_3) in concentrations of 250 mg L^{-1} (E1), 500 mg L^{-1} (E2), 1000 mg L^{-1} (E3) and 1500 mg L^{-1} (E4), or treated with potassium nitrate (KNO_3) in concentrations of 0.1% (F1), 0.2% (F2), 0.35% (F3) and 0.5% (F4).Seed germination, dormancy and vigour were determined after the above treatments in four replications, including control (C0, D0, E0, F0). The seeds were tested for seven days in dark germination cabinets, on filter paper, at a temperature of 20^{o}C after pre-chilling at 5°C. The final count was taken on the 14th day for *L. multiflorum* and the 10th day for *T. pratense* (ISTA, 2016). A tetrazolium test was applied to hard seeds in order to separate dead seeds (ISTA, 2008). Seedling vigour was determined in terms of embryonic shoot length (cm) and root length (cm).

Statistical Analysis

The results were analysed for variance (ANOVA, F test). Tukey's multiple range test was applied to establish differences between the treatments. The relationship between traits was established by Pearson's correlation test. Germination data and dormancy percentages were arcsine transformed (√x/100) (Snedecor and Cochran, 1980) before the variance analysis. Minitab 16.1.0 software was used for data processing.

Results

The results showed that the year effect had no significant influence on germination, dormancy and vigour. The cultivars and other treatments (H_2SO_4 C, Temperature D, KNO_3 E, Gibberellins F) had a significant ($p \leq 0.05$ – $p \leq 0.001$) effect. In the case of *T. pratense*, significant ($p \leq 0.05$ or $p \leq 0.01$) interactions were noted for cultivar B x H_2SO_4 C, B x Temperature D, B x KNO_3 E and B x Gibberellins F (Table 1). In case of *L. multiflorum*, there were significant ($p \leq 0.05$) interactions of B x C and B x D, with respect to germination, dormancy and vigour, and of B x F for vigour, while the other interactions were not significant (Table 1).

The H_2SO_4 treatment for 90 min. (C4) for *T. pratense* resulted in a complete break of dormancy, but also increased the number of dead seeds (data not shown). Consequently, the germination rate in this case was similar to control treatment (C0). Following treatment with H_2SO_4 for 10 min. (C1),30 min. (C2) and 60 min. (C3), 2% to 7% of seed remained dormant but germination improved by 35% (cv. Sana) to 15% (cv. K-39). On average, the optimal acid treatment (H_2SO_4 for 30 min. – C2) of the six studied cultivars ($\overline{X}$ B) of red clover improved germination by 21%. Depending on the cultivar, the optimal H_2SO_4 (50%) treatment was C1, C2 or C3 improved shoot and root growth of *T. pratense*. The same treatments applied to *L. multiflorum* cv. Aubade seed had no significant effect on shoot growth, and when applied to the other two cultivars had a significant effect on both root and shoot growth (Table 3). In general, *L. multiflorum* seed were more sensitive to the acid treatments, corroborated by C4 and C3 with no dormant seeds. The optimal acid treatment was C2, which improved germination by 18% compared to C0 ($\overline{X}$ B). Germination of the cultivars improved by 24% (cv. K-13) to 10% (cv. Draga) (Table 3). Exposing red clover seed to a temperature of 80^{0}C for 90 min (temperature of 80^{0}C, 90 min. – D4), did not fully break seed dormancy, but increased the number of dead seeds, which ultimately resulted in a smaller improvement of germination compared to acid treatment (Table 2). The cultivars responded differently, such that the optimal treatment for cv. Petnica was 80˚C for 10 min. – D1 (G, 91%), whereas for cv. K-17 was D4 (G, 93%). By selecting the optimal temperature treatment for a particular cultivar it was possible to improve germination by 24% (cv. Sana and Petnica) to 10% (cv. Una). Contrary to *T. pratense*, the temperature treatments improved germination and vigour of *L. multiflorum*, compared to the H_2SO_4 treatment (Table 2). The optimal treatment (80˚C for 30 min. – D2) resulted in a high germination rate (97%) ($\overline{X}$ B). Selection of the optimal treatment per cultivar improved germination by 31% (G, 98%, cv. K-13) to 19% (G, 97% cv. Draga).

Following the optimal temperature treatment (80^{0}C for 30 min. – D2) of *T. pratense* seed, the shoot was 1.0 cm longer and the root 0.2 cm longer, compared to D0 ($\overline{X}$ B). Optimal treatment per cultivar resulted in longer shoots, compared to the control treatment, by 1.4 cm (cv. K-17) to 0.6 cm (cv. Una) (Table 4). The optimal temperature treatment (D1) of L. *multiflorum* seed increased the length of the shoot by 3.5 cm and of the root by 1.7 cm, compared to the control treatment ($\overline{X}$ B). Depending on the cultivar, the shoot length increased by 5.1 (cv. K-13) to 1.6 cm (cv. Draga) (Table 5).

Table 1: ANOVA by year, cultivar and treatment (C, D, E and F) with regard to germination (G), dormancy (D) and vigour of red clover and Italian ryegrass seeds

Parameter	*T. pratense*					*L. multiflorum*				
	d.f	G,%	D,%	Vigour		d.f	G,%	D,%	Vigour	
				Shoot cm	Root cm				Shoot cm	Root cm
Year (A)	1	NS	NS	NS	NS	1	NS	NS	NS	NS
Cultivar (B)	5	**	**	*	*	2	**	***	*	*
H_2SO_4 (C)	3	***	***	***	***	3	***	***	**	**
Temperature (D)	3	***	***	**	**	3	**	***	*	*
KNO_3 (E)	3	*	**	*	*	3	**	**	*	*
Gibberellins (F)	3	*	**	*	*	3	*	**	*	*
Interactions										
AxB	5	NS	NS	NS	NS	3	NS	NS	NS	NS
AxC	3	NS	NS	NS	NS	3	NS	NS	NS	NS
AxD	3	NS	NS	NS	NS	3	NS	NS	NS	NS
AxE	3	NS	NS	NS	NS	3	NS	NS	NS	NS
AxF	3	NS	NS	NS	NS	3	NS	NS	NS	NS
BxC	15	*	**	*	*	6	*	*	*	*
BxD	15	*	*	*	*	6	*	*	*	*
BxE	15	*	*	*	*	6	NS	NS	NS	NS
BxF	15	*	*	*	*	6	NS	NS	*	*

F test, statistical significance levels: *$p \leq 0.05$, **$p \leq 0.01$, ***$p \leq 0.001$, NS – not significant ($p \geq 0.05$)

Table 2: Seed germination [(a)] and dormancy [(b)] after treatments: H_2SO_4 (C1-C4), temperature (D1-D4), GA_3 (E1-E4), and KNO_3 (F1-F4)

Treatments C, D, E, F	Cultivar of *T. pratense*						$\overline{X}$
	Sana	Petnica	Una	Nike	K-39	K-17	
[(a)]C0	56±1.05^{c}	67±1.10^{c}	73±1.11^{c}	72±1.12^{b}	75±0.90^{b}	72±1.12^{c}	69
$H_2SO_4$10’	87±1.02^{a}	86±1.12^{a}	86±0.95^{b}	87±1.12^{a}	88±1.12^{ab}	88±1.10^{a}	87
$H_2SO_4$30’	91±1.09^{a}	89±1.01^{a}	90±1.10^{a}	86±1.11^{a}	90±1.10^{a}	91±1.09^{a}	90
$H_2SO_4$60’	89±0.95^{a}	87±0.98^{a}	87±1.05^{ab}	88±1.00^{a}	90±1.05^{a}	91±0.95^{a}	89
$H_2SO_4$90’	78±1.12^{b}	78±1.03^{b}	74±1.12^{c}	71±0.90^{b}	49±1.07^{c}	67±1.12^{c}	70
$\overline{X}$	80	81	82	81	78	83	-
[(b)]C0	38±1.12^{a}	25±0.92^{a}	22±1.12^{a}	19±1.12^{a}	19±1.03^{a}	21±0.95^{a}	24
$H_2SO_4$10’	6±1.08^{b}	6±1.04^{b}	7±1.10^{b}	6±1.06^{b}	5±1.07^{b}	5±1.11^{b}	6
$H_2SO_4$30’	4±0.95^{bc}	5±1.09^{bc}	4±1.03^{bc}	4±1.05^{bc}	5±1.10^{b}	4±1.05^{b}	4
$H_2SO_4$60’	3±1.11^{cd}	2±1.12^{bc}	2±1.00^{cd}	2±0.95^{cd}	2±1.12^{bc}	3±0.98^{b}	2
$H_2SO_4$90’	0±0.00^{d}	0±0.00^{c}	0±0.00^{d}	0±0.00^{d}	0±0.00^{c}	0±0.00^{c}	0
$\overline{X}$	10	8	7	6	6	7	-
[(a)]D0	56±1.05^{d}	67±1.10^{b}	73±1.11^{bc}	72±1.12^{c}	75±0.90^{c}	72±1.12^{c}	69
Temp. 80^{o}C 10’	77±1.08^{ab}	91±1.12^{a}	77±1.12^{b}	79±1.08^{b}	81±1.12^{bc}	86±1.10^{b}	83
Temp. 80^{o}C 30’	80±1.11^{a}	90±0.95^{a}	83±0.98^{a}	84±1.12^{a}	85±1.12^{b}	91±1.12^{a}	85
Temp. 80^{o}C 60’	71±1.09^{b}	89±0.98^{a}	77±0.95^{b}	76±1.10^{b}	86±1.12^{b}	94±1.06^{a}	81
Temp. 80^{o}C 90’	65±1.00^{c}	89±1.03^{a}	71±1.07^{c}	70±1.12^{c}	92±1.12^{a}	93±1.05^{a}	80
$\overline{X}$	65	85	76	76	84	87	-
[(b)]D0	38±1.12^{a}	25±0.92^{a}	22±1.12^{a}	19±1.12^{a}	19±1.03^{a}	21±0.95^{a}	24
Temp. 80^{o}C 10’	18±1.10^{b}	10±1.10^{b}	15±1.11^{b}	13±0.95^{b}	12±1.12^{b}	21±1.11^{a}	13
Temp. 80^{o}C 30’	8±1.07^{c}	8±0.95^{b}	9±1.03^{c}	7±0.98^{c}	10±1.00^{b}	9±0.94^{c}	9
Temp. 80^{o}C 60’	8±1.03^{c}	8±1.12^{b}	8±1.02^{c}	6±1.01^{c}	6±0.99^{c}	4±1.00^{d}	7
Temp. 80^{o}C 90’	6±1.00^{c}	7±1.10^{b}	7±1.00^{c}	6±1.12^{c}	6±0.96^{c}	4±1.06^{d}	6
$\overline{X}$	16	12	12	10	10	10	-
[(a)]E0	56±1.05^{c}	67±1.10^{c}	73±1.11^{c}	72±1.12^{b}	75±0.90^{b}	72±1.12^{c}	69
$GA_3$250 mg l^{-1}	71±1.12^{b}	68±1.00^{b}	72±1.12^{a}	71±1.03^{b}	75±1.10^{b}	75±0.90^{ab}	72
GA_3 500 mg l^{-1}	78±1.03^{a}	77±1.04^{a}	77±1.07^{a}	78±0.93^{a}	82±1.12^{a}	76±1.00^{a}	78
GA_3 1000 mg l^{-1}	74±1.00^{ab}	79±1.10^{a}	75±0.97^{ab}	76±0.98^{a}	79±1.09^{a}	75±1.10^{ab}	76
GA_3 1500 mg l^{-1}	60±0.90^{c}	63±1.01^{b}	63±1.10^{d}	63±1.12^{c}	62±1.05^{c}	62±1.05^{c}	62
$\overline{X}$	68	71	72	72	75	72	-
[(b)] E0	38±1.12^{a}	25±0.92^{a}	22±1.12^{a}	19±1.12^{a}	19±1.03^{a}	21±0.95^{a}	24
$GA_3$250 mg l^{-1}	15±1.07^{b}	15±0.90^{b}	15±1.01^{b}	15±1.08^{b}	14±0.99^{b}	15±1.12^{b}	15
GA_3 500 mg l^{-1}	14±1.10^{d}	12±0.96^{b}	13±0.97^{b}	12±1.10^{b}	12±0.94^{b}	13±0.95^{b}	13
GA_3 1000 mg l^{-1}	15±0.93^{b}	12±1.04^{b}	15±1.00^{b}	13±0.97^{b}	12±1.03^{b}	13±1.00^{b}	13
GA_3 1500 mg l^{-1}	13±1.01^{d}	13±1.12^{b}	13±1.10^{b}	15±1.03^{b}	131.12^{b}	12±1.08^{b}	13
$\overline{X}$	19	15	16	15	14	19	-
[(a)] F0	56±1.05^{c}	67±1.10^{c}	73±1.11^{c}	72±1.12^{b}	75±0.90^{b}	72±1.12^{c}	69
KNO_3 0.1%	63±1.12^{a}	66±1.12^{a}	78±1.12^{ab}	73±1.10^{ab}	77±1.12^{ab}	81±1.01^{a}	73
KNO_3 0.2%	60±1.10^{ab}	70±1.03^{a}	85±1.09^{a}	73±0.93^{ab}	80±1.05^{a}	80±0.91^{a}	75
KNO_3 0.35%	59±0.92^{ab}	68±1.08^{a}	75±0.96^{ab}	77±1.06^{a}	74±0.92^{b}	81±1.05^{a}	72
KNO_3 0.5%	56±1.03^{b}	67±1.11^{a}	75±1.00^{ab}	71±1.12^{b}	74±1.02^{b}	73±1.12^{b}	69
$\overline{X}$	59	68	77	73	76	77	-
[(b)] F0	38±1.12^{a}	25±0.92^{a}	22±1.12^{a}	19±1.12^{a}	19±1.03^{a}	21±0.95^{a}	24
KNO_3 0.1%	32±1.03^{a}	25±1.12^{a}	18±0.96^{a}	20±1.03^{a}	17±1.12^{ab}	19±1.06^{ab}	22
KNO_3 0.2%	38±1.12^{a}	25±1.03^{a}	19±1.08^{a}	16±0.99^{ab}	16±0.92^{ab}	18±1.12^{ab}	22
KNO_3 0.35%	37±0.97^{a}	24±1.00^{a}	20±1.10^{a}	16±0.92^{ab}	15±1.01^{ab}	16±0.97^{b}	21
KNO_3 0.5%	36±0.90^{a}	23±1.10^{a}	20±0.97^{a}	14±1.12^{b}	14±1.04^{b}	17±1.00^{ab}	21
$\overline{X}$	36	24	20	17	16	18	-

Tukey's test, $p \leq 0.05$, ± standard error of mean, was applied to assess the significance by column, separately for C, D, E and F

The GA_3 treatment of T. *pratense* seed significantly ($p \leq 0.05$) reduced dormancy relative to the control treatment, but there was no significant ($p \leq 0.05$) difference among treatments GA3 1000 mg L^{-1} (E1) through GA3 1500 mg L^{-1} (E4) (Table 2). The optimal treatment (GA3 500 mg L^{-1} – E2) improved germination by 9% compared to the control treatment ($\bar{X}$ B). Selection of the optimal treatment per cultivar improved germination by 22% (cv. Sana) to 4% (cv. Una and K-17). However, the GA_3 treatment did not improve germination as much as the H_2SO_4 and temperature treatments (Table 2). Conversely, the GA_3 treatment of *L. multiflorum* seed resulted in a germination rate of 97% (treatment E3 $\bar{X}$ B). All the cultivars achieved the same germination rate (97%). The cultivars responded similarly to the other treatments. Following the E2 treatment of *T. pratense*, the shoot was 0.8 cm longer and the root 0.3 cm longer than after the control treatment. The cultivars measured a longer shoot than the control, from 1.3 cm (treatment E1 of cv. Sana and treatment E2 of cv. K-17) to 0.2 cm (treatments GA3 250 mg L^{-1} E1 and E2 of cv. Nike), while the roots were longer by 0.5 cm (E1, cv. Sana) to 0.3 cm (E2, cv. K-17). *L. multiflorum* seed generally had the highest vigour. The optimal treatment (E4) increased the length of the shoot by 7.8 cm and of the root by 1.9 cm (treatment E1 $\bar{X}$ B), compared to the control treatment ($\bar{X}$ B) (Table 5). Considering all the cultivars, the shoot length increased from 7.8 cm (KNO_3 0.35% – E3, cv. Aubade) to 6.4 cm (KNO_3 0.5% – E4, cv. Draga), and the root length from 2.4 cm (KNO_3 0.2% – E2, cv. Aubade and E4, cv. K-13) to 1.5 cm (E2, cv. Draga).

The KNO_3 treatment of *T. pratense* seed had a significant ($p \leq 0.05$) effect on cv. Nike, K-17 and K-39 and reduced dormancy by 4–5%. The optimal treatment (F2) improved germination by 6% ($\bar{X}$ B) and, depending on the cultivar, by 9% (treatment F3 of cv. K-17) to 1% (F3, cv. Petnica). The optimal treatment (F4) of *L. multiflorum* seed improved germination by 22% ($\bar{X}$ B); the cultivars responded in a similar manner to this treatment, which was corroborated by no significant interaction of cultivars Bx KNO_3 F (Tables 1 and 5). The optimal treatment (KNO_3 0.2%) of the red clover seed increased the lengths of the shoot and root by 0.7 and 0.3 cm, respectively compared to the control treatment ($\bar{X}$ B). Selection of the optimal treatment per cultivar increased the length of the shoot by 1.8 cm (treatment KNO_3 0.1% – F1 of cv. K-17) to 0.2 cm (F2, cv. Petnica), and increased the length of the root by 0.8 cm (F4, cv. Sana) to 0.4 cm (F3, cv. Una). A much larger increase in the shoot length was noted in the case of Italian ryegrass (5.6 cm after treatment F3, compared to the control treatment), and in the root length (0.6 cm by F3 and F1, relative to the control treatment $\bar{X}$ B). A positive correlation was established between germination and vigour of all the cultivars of both species. In the case of the red clover, it ranged from the strongest ($p \leq 0.001$) for cv. Sana (shoot r=0.933 and root r=0.748) to no significant correlation with cv. Nike in respect of the root (r=0404). The strongest correlation ($p \leq 0.001$) between germination and shooting was noted in the case of Italian ryegrass cv. K-13 (r=0.806), while there was no significant correlation (r=0.179) between germination and rooting of cv. Draga (Table 6).

Table 3: Seed germination [(a)] and dormancy [(b)] after treatments: H_2SO_4 (C1-C4), temperature (D1-D4), GA_3 (E1-E4), and KNO_3 (F1-F4)

Treatments C, D, E, F	Cultivar of *L. multiflorum*			$\bar{X}$
	Aubade	Draga	K-13	72
[(a)]C0	70 ± 1.12^{d}	78 ± 1.10^{d}	67 ± 1.03^{c}	85
$H_2SO_4$10'	87 ± 1.11^{b}	88 ± 1.12^{a}	81 ± 1.00^{b}	90
$H_2SO_4$30'	92 ± 1.08^{a}	88 ± 0.90^{a}	91 ± 0.99^{a}	82
$H_2SO_4$60'	81 ± 1.06^{c}	84 ± 1.11^{b}	81 ± 1.07^{b}	80
$H_2SO_4$90'	79 ± 1.12^{c}	83 ± 1.05^{b}	79 ± 1.12^{b}	-
$\bar{X}$	82	84	80	13
[(b)]C0	13 ± 1.12^{a}	13 ± 1.12^{a}	14 ± 1.10^{a}	6
$H_2SO_4$10'	5 ± 1.11^{b}	7 ± 1.07^{b}	7 ± 1.05^{b}	2
$H_2SO_4$30'	1 ± 1.00^{c}	2 ± 1.03^{c}	2 ± 0.97^{c}	0
$H_2SO_4$60'	0 ± 0.00^{c}	0 ± 0.00^{c}	1 ± 1.01^{c}	0
$H_2SO_4$90'	0 ± 0.00^{c}	0 ± 0.00^{c}	0 ± 0.00^{c}	-
$\bar{X}$	4	4	5	72
[(a)]D0	72 ± 1.12^{d}	78 ± 1.10^{d}	67 ± 1.03^{d}	96
Temp. 80°C 10'	97 ± 1.00^{a}	96 ± 0.99^{a}	96 ± 1.12^{a}	97
Temp. 80°C 30'	98 ± 1.01^{a}	97 ± 0.95^{a}	97 ± 0.95^{a}	93
Temp. 80°C 60'	93 ± 1.03^{b}	93 ± 1.07^{b}	93 ± 1.07^{b}	92
Temp. 80°C 90'	90 ± 1.10^{c}	89 ± 1.04^{c}	98 ± 1.03^{a}	-
$\bar{X}$	90	91	88	13
[(b)]D0	13 ± 1.12^{a}	13 ± 1.12^{a}	14 ± 1.10^{a}	3
Temp. 80°C 10'	2 ± 1.10^{b}	3 ± 1.03^{b}	4 ± 1.12^{b}	2
Temp. 80°C 30'	1 ± 1.01^{c}	2 ± 1.11^{b}	3 ± 0.94^{b}	0
Temp. 80°C 60'	0 ± 0.00^{d}	0 ± 0.00^{d}	0 ± 0.00^{d}	0
Temp. 80°C 90'	0 ± 0.00^{d}	0 ± 0.00^{d}	0 ± 0.00^{d}	-
$\bar{X}$	3	4	4	72
[(a)]E0	70 ± 1.12^{d}	78 ± 1.10^{d}	67 ± 1.03^{c}	91
$GA_3$250 mg l^{-1}	91 ± 1.10^{b}	92 ± 1.12^{b}	90 ± 1.08^{c}	93
GA_3 500 mg l^{-1}	93 ± 1.03^{b}	92 ± 1.07^{b}	94 ± 1.12^{b}	97
GA_3 1000 mg l^{-1}	97 ± 1.05^{a}	97 ± 1.10^{a}	97 ± 1.00^{a}	92
GA_3 1500 mg l^{-1}	91 ± 0.98^{b}	93 ± 1.01^{b}	91 ± 0.97^{c}	-
$\bar{X}$	89	90	88	13
[(b)] E0	13 ± 1.12^{a}	13 ± 1.12^{a}	14 ± 1.10^{a}	3
$GA_3$250 mg l^{-1}	3 ± 1.04^{b}	2 ± 1.09^{b}	4 ± 1.12^{b}	1
GA_3 500 mg l^{-1}	1 ± 0.96^{c}	1 ± 1.11^{c}	1 ± 1.02^{c}	0
GA_3 1000 mg l^{-1}	0 ± 0.00^{d}	0 ± 0.00^{d}	0 ± 0.00^{d}	0
GA_3 1500 mg l^{-1}	0 ± 0.00^{d}	0 ± 0.00^{d}	0 ± 0.00^{d}	-
$\bar{X}$	3	3	4	72
[(a)] F0	70 ± 1.12^{d}	78 ± 1.10^{d}	67 ± 1.03^{c}	93
KNO_3 0.1%	94 ± 1.10^{a}	93 ± 1.12^{a}	92 ± 1.12^{a}	93
KNO_3 0.2%	94 ± 1.00^{a}	93 ± 1.08^{a}	92 ± 1.00^{a}	92
KNO_3 0.35%	91 ± 1.01^{b}	92 ± 0.93^{a}	93 ± 1.05^{a}	94
KNO_3 0.5%	96 ± 0.99^{a}	92 ± 1.07^{a}	94 ± 1.09^{a}	-
$\bar{X}$	89	90	88	13
[(b)] F0	13 ± 1.12^{a}	13 ± 1.12^{a}	14 ± 1.10^{a}	2
KNO_3 0.1%	3 ± 1.07^{b}	2 ± 1.03^{b}	2 ± 1.12^{b}	2
KNO_3 0.2%	2 ± 0.96^{bc}	2 ± 0.92^{b}	2 ± 1.07^{b}	1
KNO_3 0.35%	1 ± 1.05^{cd}	1 ± 1.05^{c}	2 ± 0.91^{b}	1
KNO_3 0.5%	0 ± 0.00^{d}	0 ± 0.00^{d}	2 ± 1.04^{b}	-
$\bar{X}$	4	4	4	

Tukey's test, $p \leq 0.05$, ± standard error of mean, was applied to assess the significance by column, separately for C, D, E and F

Table 4: Vigour (shoot cm $^{(a)}$, root cm $^{(b)}$) after treatments: H_2SO_4 (C1-C4), temperature (D1-D4), GA_3 (E1-E4), and KNO_3 (F1-F4)

Treatments C, D, E, F	Cultivar of *T. pratense*						$\overline{X}$
	Sana	Petnica	Una	Nike	K-39	K-17	
$^{(a)}$C0	2.8±0.07^{c}	3.5±1.23^{c}	3.8±0.56^{b}	3.7±1.12^{b}	4.0±0.69^{b}	3.6±0.75^{b}	3.5
$H_2SO_4$10’	5.2±1.20^{a}	4.7±1.16^{ab}	5.6±1.11^{a}	4.3±0.87^{a}	5.4±1.01^{a}	3.7±0.98^{b}	4.8
$H_2SO_4$30’	4.6±1.17^{b}	5.1±1.20^{a}	4.6±1.12^{b}	4.4±0.99^{a}	5.0±0.89^{a}	4.6±0.57^{a}	4.7
$H_2SO_4$60’	4.7±0.23^{b}	4.6±0.92^{b}	4.3±0.67^{b}	4.6±1.00^{a}	4.9±1.10^{a}	4.1±1.04^{ab}	4.5
$H_2SO_4$90’	4.50.16.^{b}	4.7±0.35^{ab}	4.1±1.18^{b}	3.3±0.42^{b}	2.5±0.09^{c}	3.8±1.19^{b}	3.8
$\overline{X}$	4.4	4.5	4.5	4.1	4.4	4	-
$^{(b)}$C0	0.9±1.23^{b}	1.1±1.12^{c}	2.0±1.18^{b}	1.4±0.07^{c}	1.6±1.20^{c}	1.4±1.19^{c}	1.4
$H_2SO_4$10’	2.7±1.09^{a}	2.7±0.92^{a}	3.1±1.00^{a}	2.7±1.10^{a}	3.3±0.98^{a}	3.0±0.84^{a}	2.9
$H_2SO_4$30’	2.8±0.74^{a}	1.8±0.09^{bc}	2.7±0.63^{a}	1.7±0.09^{bc}	2.8±1.01^{ab}	2.5±1.15^{ab}	2.4
$H_2SO_4$60’	2.6±0.17^{a}	2.5±0.99^{ab}	2.7±1.02^{a}	2.0±1.03^{b}	2.3±1.09^{b}	2.2±1.09^{b}	2.4
$H_2SO_4$90’	1.5±1.19^{b}	1.5±1.20^{c}	2.0±0.92^{b}	1.7±0.90^{bc}	1.0±1.16^{d}	1.4±1.12^{c}	1.5
$\overline{X}$	2.1	1.9	2.5	1.9	2.2	2.1	-
$^{(a)}$D0	2.8±0.07^{c}	3.5±1.23^{c}	3.8±0.56^{b}	3.7±1.12^{b}	4.0±0.69^{b}	3.6±0.75^{b}	3.5
Temp. 80°C 10’	4.1±1.12^{a}	4.4±1.10^{a}	4.0±0.98^{ab}	3.6±1.02^{b}	4.7±1.12^{a}	4.2±1.12^{bc}	4.2
Temp. 80°C 30’	3.8±0.63^{b}	4.5±1.19^{a}	4.4±1.00^{a}	4.5±1.19^{a}	4.7±1.21^{a}	5.0±1.19^{a}	4.5
Temp. 80°C 60’	3.7±1.10^{b}	3.9±1.09^{ab}	3.7±0.83^{b}	3.7±1.23^{b}	4.5±0.90^{ab}	4.5±0.82^{b}	4.0
Temp. 80°C 90’	3.2±1.19^{c}	4.0±1.12^{ab}	3.6±1.12^{b}	3.7±0.07^{b}	5.0±1.01^{a}	4.8±1.18^{ab}	4.1
$\overline{X}$	3.5	4.1	3.9	3.8	4.6	4.4	
$^{(b)}$D0	0.9±1.23^{b}	1.1±0.09^{c}	2.0±1.18^{b}	1.4±0.07^{c}	1.6±1.20^{c}	1.4±1.19^{c}	1.4
Temp. 80°C 10’	1.4±1.12^{ab}	1.5±1.10^{ab}	1.8±1.19^{ab}	1.4±1.12^{a}	1.2±1.08^{b}	1.2±1.12^{c}	1.4
Temp. 80°C 30’	1.5±1.19^{a}	1.8±1.09^{a}	1.8±0.65^{ab}	1.4±1.23^{a}	1.5±1.12^{ab}	1.7±1.21^{b}	1.6
Temp. 80°C 60’	1.2±1.13^{ab}	1.5±1.15^{ab}	2.0±1.09^{a}	1.4±1.09^{a}	1.6±1.10^{ab}	1.4±0.87^{bc}	1.5
Temp. 80°C 90’	1.4±0.99^{ab}	1.4±0.85^{ab}	1.4±1.00^{c}	1.2±1.12^{a}	1.7±1.03^{a}	1.9±1.64^{a}	1.5
$\overline{X}$	1.3	1.5	1.7	1.3	1.4	1.6	
$^{(a)}$E0	2.8±0.07^{c}	3.5±1.23^{c}	3.8±0.56^{b}	3.7±1.12^{b}	4.0±0.69^{b}	3.6±0.75^{b}	3.5
$GA_3$250 mg l^{-1}	4.1±1.12^{a}	3.4±1.17^{ab}	3.4±1.12^{c}	3.9±1.21^{a}	4.2±1.12^{ab}	3.7±1.12^{b}	4.0
GA_3 500 mg l^{-1}	3.7±1.20^{ab}	3.9±1.12^{b}	4.5±1.17^{a}	3.9±1.19^{a}	4.8±1.21^{a}	4.9±1.21^{a}	4.3
GA_3 1000 mg l^{-1}	3.9±1.16^{ab}	4.1±1.00^{a}	4.0±0.98^{b}	3.8±1.13^{a}	4.4±1.19^{ab}	3.7±1.10^{b}	4.0
GA_3 1500 mg l^{-1}	3.3±1.12^{b}	3.0±1.07^{c}	3.3±1.12^{c}	2.8±1.12^{b}	3.0±0.87^{c}	2.9±1.12^{c}	3.1
$\overline{X}$	3.6	3.6	3.8	3.6	4.1	3.8	
$^{(b)}$ E0	0.9±1.23^{b}	1.1±0.09^{c}	1.3±1.18^{b}	1.4±0.07^{c}	1.6±1.20^{c}	1.4±1.19^{c}	1.3
$GA_3$250 mg l^{-1}	1.4±1.12^{b}	1.1±1.21^{b}	0.8±1.12^{c}	1.3±1.12^{b}	1.6±1.18^{a}	1.3±1.23^{b}	1.3
GA_3 500 mg l^{-1}	1.1±1.00^{b}	1.8±1.02^{a}	1.6±1.20^{ab}	1.9±1.23^{a}	1.5±10.89^{ab}	1.7±0.07^{a}	1.6
GA_3 1000 mg l^{-1}	1.3±1.21^{b}	1.4±0.95^{ab}	2.0±0.89^{a}	1.2±1.19^{b}	1.4±0.95^{b}	1.4±0.35^{b}	1.5
GA_3 1500 mg l^{-1}	1.1±1.03^{b}	1.2±1.12^{b}	1.3±1.13^{b}	0.9±0.09^{c}	1.0±1.12^{c}	0.7±0.84^{c}	1.2
$\overline{X}$	1.2	1.3	1.4	1.3	1.4	1.3	
$^{(a)}$ F0	2.8±0.07^{c}	3.5±1.23^{c}	3.8±0.56^{b}	3.7±1.12^{b}	4.0±0.69^{b}	3.6±0.75^{b}	3.5
KNO_3 0.1%	3.6±1.12^{a}	3.6±1.08^{a}	4.0±1.12^{bc}	3.7±1.23^{ab}	4.2±1.12^{b}	5.4±1.12^{a}	4.1
KNO_3 0.2%	3.0±1.22^{b}	3.7±1.00^{a}	4.3±1.22^{a}	3.9±1.09^{ab}	5.2±1.20^{a}	5.0±1.17^{a}	4.2
KNO_3 0.35%	2.9±1.15^{b}	3.3±0.86^{ab}	4.1±1.11^{b}	4.1±1.01^{a}	4.0±1.18^{b}	3.1±0.73^{b}	3.6
KNO_3 0.5%	2.9±0.96^{b}	3.5±0.55^{a}	2.9±1.00^{d}	3.5±0.99^{b}	3.0±1.13^{c}	3.0±1.19^{b}	3.1
$\overline{X}$	3.0	3.4	3.8	3.8	4.1	4	
$^{(b)}$ F0	0.9±1.23^{b}	1.1±0.09^{c}	2.0±1.18^{b}	1.4±0.07^{c}	1.6±1.20^{c}	1.4±1.19^{c}	1.4
KNO_3 0.1%	1.6±1.12^{a}	1.5±1.21^{a}	2.0±1.05^{ab}	1.8±1.12^{c}	1.6±1.17^{c}	1.9±1.12^{a}	1.7
KNO_3 0.2%	1.5±1.19^{a}	1.5±1.12^{a}	1.8±1.12^{ab}	1.5±1.17^{d}	1.8±1.12^{b}	1.9±1.23^{a}	1.7
KNO_3 0.35%	1.5±0.98^{a}	1.3±0.92^{a}	2.4±1.21^{a}	2.0±0.89^{b}	2.0±1.03^{a}	1.5±0.67^{b}	1.8
KNO_3 0.5%	1.7±1.01^{a}	1.4±0.85^{a}	2.0±1.00^{ab}	3.0±0.91^{a}	1.3±1.00^{d}	1.0±0.09^{c}	1.7
$\overline{X}$	1.4	1.4	2.3	1.9	1.5	1.5	

Tukey's test, $p \leq 0.05$, ± standard error of mean, was applied to assess the significance by column, separately for C, D, E and F

Discussion

Red clover and Italian ryegrass are very important high-quality forage crops in south-eastern and central Europe. Growing of forage legumes and grasses in a mixture offers many agronomic advantages compared to single crops (Simić *et al.*, 2011). When a crop mixture is considered, it is extremely important to achieve the projected legume-to-grass ratio, whereby seed quality and vigour are of overriding significance. However, germination and viability of forage legume and grass seeds are poorer during the period of post-harvest maturity (Stanisavljević *et al.*, 2011; Tiryaki and Topu, 2014). Still, regardless of these circumstances, seed suppliers operate on economic principles of making profit, ensuring a quick turnover of capital, avoiding storage of seed, and cutting costs, thus, it is in their interest to sell the seed as soon as possible.

A higher germination rate is in the interests of farmers because it leads to a lower seed rate and more economical crop establishment, while maintaining the projected number of plants. The cause of forage legume seed dormancy is a seed coat impervious to water and gasses,

Table 5: Vigour (shoot cm $^{(a)}$, root cm $^{(b)}$) after treatments: H_2SO_4 (C1-C4), temperature (D1-D4), GA_3 (E1-E4), and KNO_3 (F1-F4)

Treatments C, D, E, F	Cultivar of *L. multiflorum*			$\bar{X}$
	Aubade	Draga	K-13	72
$^{(a)}$C0	9.9±1.12^{a}	7.2±0.12^{bc}	6.7±1.23^{c}	7.9
$H_2SO_4$10’	9.8±0.10^{a}	9.0±0.26^{a}	9.8±1.09^{ab}	9.5
$H_2SO_4$30’	9.3±1.05^{a}	7.3±1.08^{bc}	10.4±0.95^{a}	9.0
$H_2SO_4$60’	9.3±0.63^{a}	8.6±1.23^{ab}	8.8±0.09^{b}	8.9
$H_2SO_4$90’	9.9±0.89^{a}	7.5±1.02^{b}	8.2±1.00^{bc}	8.5
$\bar{X}$	9.6	7.9	8.8	-
$^{(b)}$C0	6.5±1.21^{ab}	3.8±1.12^{c}	5.5±0.65^{c}	5.3
$H_2SO_4$10’	8.0±1.09^{a}	4.6±0.78^{bc}	7.5±1.18^{a}	6.7
$H_2SO_4$30’	5.6±0.87^{b}	9.2±1.20^{a}	6.1±0.87^{b}	7.0
$H_2SO_4$60’	7.7±1.22^{ab}	5.1±0.91^{b}	6.2±1.00^{b}	6.3
$H_2SO_4$90’	7.3±0.98^{ab}	4.4±1.09^{bc}	7.1±0.87^{ab}	6.3
$\bar{X}$	7.0	5.4	6.5	-
$^{(a)}$D0	9.9±1.12^{a}	7.2±0.12^{bc}	6.7±1.23^{c}	7.9
Temp. 80ºC 10’	13.6±1.10^{a}	8.8±0.99^{a}	11.8±1.12^{a}	11.4
Temp. 80ºC 30’	12.3±1.19^{ab}	8.4±1.00^{ab}	11.8±1.04^{a}	10.8
Temp. 80ºC 60’	12.5±1.21^{ab}	8.4±1.01^{ab}	11.7±0.95^{a}	10.9
Temp. 80ºC 90’	11.2±1.23^{b}	7.3±1.09^{b}	9.4±1.00^{b}	9.3
$\bar{X}$	11.9	8	10.3	
$^{(b)}$D0	6.5±1.21^{ab}	3.8±1.12^{c}	5.5±0.65^{c}	5.3
Temp. 80ºC 10’	6.7±1.13^{bc}	4.2±1.16^{ab}	6.9±1.12^{a}	5.9
Temp. 80ºC 30’	9.3±1.02^{a}	4.6±1.22^{ab}	7.2±1.20^{a}	7.0
Temp. 80ºC 60’	7.5±0.98^{b}	5.5±0.95^{a}	7.0±0.89^{a}	6.7
Temp. 80ºC 90’	7.1±1.03^{b}	4.3±1.00^{ab}	7.2±0.08^{a}	6.2
$\bar{X}$	7.4	4.5	6.8	
$^{(a)}$E0	9.9±1.12^{a}	7.2±0.12^{bc}	6.7±1.23^{c}	7.9
$GA_3$250 mg l^{-1}	16.7±1.10^{ab}	13.2±1.12^{a}	13.4±1.18^{a}	14.4
GA_3 500 mg l^{-1}	13.7±1.00^{b}	10.7±0.96^{c}	13.2±1.12^{a}	12.5
GA_3 1000 mg l^{-1}	17.7±1.21^{a}	11.4±0.87^{b}	16.3±0.85^{a}	15.1
GA_3 1500 mg l^{-1}	17.3±1.19^{a}	13.6±1.23^{a}	16.3±1.15^{a}	15.7
$\bar{X}$	15.1	11.2	13.2	
$^{(b)}$ E0	6.5±1.21^{ab}	3.8±1.12^{c}	5.5±0.65^{c}	5.3
$GA_3$250 mg l^{-1}	8.8±1.12^{a}	5.2±1.15^{a}	7.7±1.12^{ab}	7.2
GA_3 500 mg l^{-1}	8.9±1.18a	5.3±1.00^{a}	6.8±1.23^{b}	7.0
GA_3 1000 mg l^{-1}	7.3±1.03^{ab}	4.5±1.02^{ab}	6.3±1.18^{bc}	6.0
GA_3 1500 mg l^{-1}	7.6±1.10^{ab}	5.0±0.96^{a}	7.9±0.99^{a}	6.8
$\bar{X}$	7.8	4.8	6.8	
$^{(a)}$ F0	9.9±1.12^{a}	7.2±0.12^{bc}	6.7±1.23^{c}	7.9
KNO_3 0.1%	15.2±1.20^{ab}	10.3±1.12^{ab}	12.5±1.12^{a}	12.7
KNO_3 0.2%	16.0±1.04^{a}	10.0±1.23^{b}	13.6±1.10^{a}	13.2
KNO_3 0.35%	14.0±0.15^{ab}	12.7±0.45^{a}	13.9±1.09^{a}	13.5
KNO_3 0.5%	13.0±0.33^{b}	9.6±0.09^{bc}	12.3±0.93^{a}	11.6
$\bar{X}$	13.6	10	11.8	
$^{(b)}$ F0	6.5±1.21^{ab}	3.8±1.12^{c}	5.5±0.65^{c}	5.3
KNO_3 0.1%	8.2±1.12^{a}	4.1±1.21^{b}	5.5±1.12^{b}	5.9
KNO_3 0.2%	7.2±1.02^{ab}	4.2±1.18^{b}	5.5±1.01^{b}	5.6
KNO_3 0.35%	6.2±1.11^{b}	5.5±0.94^{a}	6.0±0.56^{a}	5.9
KNO_3 0.5%	6.5±0.98^{b}	3.7±0.33^{b}	5.5±1.18^{b}	5.2
$\bar{X}$	6.9	4.3	5.6	

Tukey's test, $p \leq 0.05$, ± standard error of mean, was applied to assess the significance by column, separately for C, D, E and F

and of grass seed dormancy the embryo or the substances that surround it (Adkins *et al.*, 2002; Kimura and Islam, 2012). As the proportion of dormant seed increases, the germination rate decreases; this is strongly affected by genetic predisposition and environmental conditions during the time of soakage and maturing of forage legume and grass seeds (Adkins *et al.*, 2002; Asci *et al.*, 2011; Stanisavljević *et al.*, 2015).

Table 6: Coefficient of correlation (r) between germination (G) and vigour (shoot cm S, root cm R)

Species	Cv.	Parameter	S, cm	R, cm
T. pratense	Sana	G, %	0.933***	0.748***
	Petnica	G, %	0.690***	0.627**
	Una	G, %	0.743***	0.671**
	Nike	G, %	0.887***	0.404NS
	K-39	G, %	0.893***	0.525*
	K-17	G, %	0.608**	0.698***
L. ultiflorum	Aubade	G, %	0.652**	0.361NS
	Draga	G, %	0.604**	0.179 NS
	K-13	G, %	0.806***	0.415 NS

Statistical significance levels: $*p \leq 0.05$, $**p \leq 0.01$, $***p \leq 0.001$, NS – not significant ($p \geq 0.05$)

Other researchers have reported conflicting findings after studying H_2SO_4, temperature, GA_3 and KNO_3 treatments of forage and/or ornamental grasses and legumes, and their effect on the enhancement of germination and vigour (Can *et al.*, 2009; Qadir *et al.*, 2011; Kimura and Islam, 2012; Stanisavljević *et al.*, 2012; Tiryaki and Topu, 2014). The results of the present study, with regard to the optimal acid treatment of Italian ryegrass, are consistent with to *Lolium perenne* (Salehi and Khosh-Khui, 2005). Bhattarai *et al.* (2008) reported a much higher H_2SO_4 concentration in the case of *Cenchrus ciliaris* (100%, 4 min). Treatments with H_2SO_4 of different species of the genus *Festuca* appear to have different effects on germination: for example, germination of *F. rubra* can be improved by 19%, of *F. ovina* by 13%, and of *F. pratensis* by 22% (Stanisavljević *et al.*, 2012). Seed treatment of the genus *Trifolium* with 95–97% H_2SO_4 resulted in germination rates from 90% *T. lappaceum* to 0% *T. badium* and *T. spumosum* (Can *et al.*, 2009).

One of the most popular methods is temperature scarification of seed because it is simple, easy to implement, and environmentally safe. According to Stanisavljević *et al.* (2013) drying of meadow fescue (*F. pratensis*) seeds at temperatures of 40°C and 50°C resulted in maximum germination of seed harvested with 45% moisture after three months. After eight months the best germination of all seeds was obtained at 22°C and 40°C. Consistent with the results of the present study, Kimura and Islam (2012) reported an optimal temperature of 80ºC for seed of the genus *Trifolium*. The treatment with GA_3 was less effective than mechanical scarification (Uzun and Aydin, 2004). Conversely, GA_3 significantly improved germination of the grass species *Leymus arenarius*, but the effect decreased with seed ageing (Greipson, 2001). The same treatment of *C. ciliaris* improved germination by 44.8%, as well as vigour (Nagar and Meena, 2015). Treatments with KNO_3 of grasses (*buffel grass, dhaman grass, blue panic grass*) improved germination by 22.1% (*buffel grass*), but had no such effect on *blue panic grass*. The germination improvement trend was consistent with seedling growth (Qadir *et al.*, 2011). The results of the present study concerning the positive correlation between germination and vigour agree with findings after testing of *F. pratensis, F. arundinacea, F. rubra, Lolium multiflorum, Dactylis glomerata, Phleum pratense, Brassica napus* (Stanisavljević *et al.*, 2011, 2014; Zhang *et al.*, 2013).

Conclusion

The results led to the conclusion that *L. multiflorum* and *T. pratense* respond differently to acid, temperature, GA_3 and KNO_3 treatments of seed. In general, the strongest response in terms of improved germination and vigour was noted after treatment of *T. pratense* with 50% H_2SO_4, and of *L. multiflorum* with gibberellic acid (1000 mg L^{-1}) and temperature(80^{o}C 30'). By selecting the optimal seed treatment of the considered cultivar, it is possible to improve germination, initial shoot and root growth.

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