

MANAGEMENT PRACTICES EFFECT ON SEED FEATURES OF ITALIAN RYEGRASS FOLLOWING STORAGE PERIOD

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Abstract: Italian ryegrass seed crop was established in 2007 with two sowing densities ($D_1 = 60$ cm row spacing and 5 kg ha^{-1} seeding rate; $D_2 = 20$ cm row spacing and 20 kg ha^{-1} seeding rate) and using two spring nitrogen rates (0 and 150 kg N ha^{-1}). Seed germination and thousand seed weight (TSW) of Italian ryegrass was observed in first production year. After harvest in June, seeds were stored under standard storage conditions and sampled 90 days after harvest (DAH), and then 2000 DAH. At 90 DAH, seeds were tested for TSW, as well as germination energy and total germination percentage at incubation temperatures of 10, 15, 20 and 25°C . Ryegrass seeds had the best germination energy 90 DAH at 20°C and maximum total germination at 15°C , which implies that early autumn (September-October) is proper sowing period for freshly harvested seeds of Italian ryegrass. Italian ryegrass seeds could maintain satisfactory germination energy (59.3%) and total germination (77.3%) up to 2000 DAH. High seed quality was obtained and applied treatments did not change seed quality significantly unlike storage period which had considerable influence on seed quality. The data can serve for the determination of a proper storage duration management between harvest and sowing of the tested species under ambient conditions of Serbia.

Key words: Italian ryegrass, nitrogen application, seed features, sowing density, storage period

Introduction

Historically, grass seed quality has been synonymous with germination, i.e. the measurement of the percentage of seeds growing normally under standardised, controlled, optimum laboratory conditions, set so that seed is given every chance to

germinate to its full potential (ISTA, 2010). During post-harvest maturation, different species vary in the length of dormancy breaking or germination increases. Seed dormancy and slow seedling development often limit establishment of forage grass stands (Stanisavljević et al., 2011). However, seed dormancy and delayed germination of forage grasses under natural conditions can be beneficial because they postpone germination and the initial growth of seedlings until environmental conditions improve (Stanisavljević et al., 2012). Maximum germination is achieved after the period of seed maturation and dormancy loss. Further seed storage leads to certain physiological and biochemical processes that result in seed ageing and germination decreases (Bewley and Black, 1994). Seed germination conservation in the course of ageing depends largely on storage conditions (Walters et al., 2004). In Serbian conditions, approximately two and a half months elapses between the time of seed collection and the sowing of forage grasses during the autumn. Sowing in this period provides sufficient time for germination, seedling development, and survival during the winter. Compared with sowing seed during the spring of the following year, autumn sowing provides for substantially better turf formation, more forage yield, and increased seed yield (Salehi and Khosh-Khui, 2005). Harvested seeds can be used to establish new crop by sowing in autumn (August-September) of the same year or in spring (March-April) of the succeeding year. Laws and regulations governing the seed trade mainly determine minimum germination standards of approximately 70–75%. Seed size, usually measured as thousand seed weight (TSW), may be an indicator of quality, as increasing TSW can result in improved seedling growth (Hill et al., 1998). Bean (1973) pointed out that TSW was significantly and positively correlated with subsequent seedling dry weight for Italian ryegrass. Hampton (1986) reported that increasing seed weight from 3.2 g to 5 g, seedling performance was increased in field sowings in Italian ryegrass from 68% to 85%. Efficient germination of Italian ryegrass seeds is essential for successful establishment of meadows and pastures. Italian ryegrass is one of the best forage grasses in Serbia, producing high quality forage. *Lolium* species account for about 23% of the 52 million ha of grassland in Europe with Italian ryegrass being the most prevalent species (Humphreys et al., 2010). According to Simić et al. (2009) excellent ryegrass seed yield was achieved in Serbia in the first year, but local production covers only 50% of forage production needs for this seed. The application of different nitrogen rates and seeding techniques affected Italian ryegrass seed yield in Serbia (Vučković et al., 2003; Simić et al., 2009), but it is not clear whether management practice influenced ryegrass seed quality (Simić et al., 2010). Vučković et al. (1998) achieved higher germination energy of Italian ryegrass seed grown at row distance of 50 cm and applying seeding rate of 4 kg ha⁻¹ than at the distance of 20 cm and applying higher seeding rates. Seeds reach maximum germination during storage, but if storage is prolonged germination is decreased and lost, which signifies the process of seed ageing. Stanisavljević et al. (2011) noticed that a reduction in seed

germination of Italian ryegrass was not recorded before 750 DAH. Furthermore, acceptable germination was recorded up to 810 DAH (81%) and without a reduction of the seedling vigour. In practice, there may also be also interest in storing the seeds of Italian ryegrass over the next two autumn sowings.

Successfully growing Italian ryegrass requires a high and consistent germination energy, coupled with fast and vigorous seedling growth. Italian ryegrass seed crop is usually harvests or disperses under natural conditions mainly in June. This experiment was conducted to determine the differences among Italian ryegrass seed parameters obtained by management practices of the seed crop in the first production year, using different sowing density and spring nitrogen application. Also, the aim of the experiment was to determine TSW, the level of seed dormancy at the different incubation temperatures, germination energy and total germination of Italian ryegrass seeds, immediately upon harvesting (90 DAH) and 5.5 years after harvesting.

Material and methods

The experiment was set up in autumn 2007 with seeds of the tetraploid Italian ryegrass K-29t in the vicinity of the city Šabac, western Serbia (44°47' N, 19°35' E, 80 m asl). The plot for harvest was 10 m², and it was replicated four times in a randomized complete block design. Soil in the experimental area was humofluvisol (2.54% humus), with rinsed limestone, pH in KCl: 5.25; K₂O: 15 mg kg⁻¹; P₂O₅: 3 mg kg⁻¹. Italian ryegrass seed crop was established with two sowing densities (D₁ = 60 cm row spacing and 5 kg ha⁻¹ seeding rate; D₂ = 20 cm row spacing and 20 kg ha⁻¹ seeding rate) and using two spring nitrogen rates (0 and 150 kg N ha⁻¹). Seed from the primary growth in 2008 was harvested at the peak of seed ripeness, in the first production year after the establishment. Harvested seeds were cleaned manually, placed into paper bags and stored dry under ambient storage conditions. Seed samples were drawn 90 days after harvest (DAH) and then 2000 DAH.

Three characters were measured 90 DAH to provide an estimate of seed quality: (1) 1000-seed weight (TSW), (2) germination energy and (3) total germination and two characters were measured 2000 DAH: (1) germination energy and (2) total germination. Four replicates of 100 seeds were germinated on filter paper according to the ISTA Rules (*ISTA, 2010*). The seeds were incubated 90 DAH at different temperatures of 10, 15, 20 and 25°C and 2000 DAH at temperature 20°C. Germination energy count was made after 5 days and the total seedling after 14 days.

The data were analysed by two-way ANOVA using Statistica 10.0 (StatSoft, Inc. Tulsa, OK, USA) software. LSD multiple range test was used to detect significant differences among means at the 5% level of probability.

Results and Discussion

Average TSW varied among different seed sowing densities and nitrogen rates from 3.89 g to 4.32 g (Table 1), and that variation could be explained by environmental conditions during seed development and ripening. Sparse plants at D₁ density enabled to take more nutrients and had full light treatment. On the other hand, dense sward affected assimilates supply and may have limited seeds from achieving their potential final weight, consequently reducing TSW. Nitrogen application at seed crop had diminishing effect on sowing density treatments, either decreasing TSW at D₁ density or increasing TSW at D₂ density. This is in accordance with former results of *Akpan and Bean (1980)*, who reported that the seed from spaced plants had a higher TSW and seedling dry weight than the seed from narrow drills. Otherwise, TSW as seed quality indicator was conditioned by the factors of vegetation area in crop establishment, whereas nitrogen had an influence only in extremely different densities (sparse and dense crop). It is in agreement with findings of *Choi et al. (2002)* who noted the TSW increase when the inter-row space was raised from 15 cm to 45 cm.

Table 1. Effect of Italian ryegrass sowing density and nitrogen rate on thousand seed weight

Treatment	Nitrogen rate		
	N ₁	N ₂	Average
D ₁	4.316	4.071	4.193
D ₂	3.887	4.066	3.977
Average	4.102	4.068	
LSD _{0.05(D, N)} = 0.115			
LSD _{0.05(DxN)} = 0.163			

D1 = 60 cm row spacing and 5 kg ha⁻¹ seeding rate; D2 = 20 cm row spacing and 20 kg ha⁻¹ seeding rate; N1 = 0 kg N ha⁻¹; N2 = 150 kg N ha⁻¹

Unlike results for TSW, seed germination energy and total germination was not affected by sowing density and nitrogen rate in any investigated temperature traits (Table 2). This is in agreement with results of *Choi et al. (2002)*. At all incubation temperatures 90 DAH (from 10 to 25°C), as well as 2000 DAH at 20°C there was not a statistically significant differences among seeds produced in dense or sparse sward, with or without N application. Physiological immaturity of seed was a possible cause of the low germination energy at temperature treatment of 10°C (<5% germination energy for 5 days), but total germination after two weeks reached 90% (tab. 2). *Akpan and Bean (1980)* reported that an increase in temperature from a 15°/10°C regime to a constant 25°C environment increased germination energy. An increase in germination energy occurred between temperatures 10 and 15°C, indicating the start of dormancy loss 90 DAH. Starting

from 15°C, germination energy was increased (> 80%) and such was at 20 and 25°C. After 2000 DAH germination energy was decreased to 59.3%. Total seed germination of K-29t was high and uniform, except for 2000 DAH.

Table 2. Effect of sowing density and nitrogen rate on germination energy and total germination (%)

Treatment	Days after harvest (DAH)										
	90					2000	90				2000
	Germination energy (%)					Total germination (%)					
	<i>Temperature</i>										
	10°C	15°C	20°C	25°C	20°C	10°C	15°C	20°C	25°C	20°C	
Sowing density	T1	T2	T3	T4	T5	T1	T2	T3	T4	T5	
D1	4.9	92.7	89.6	85.3	61.5	91.6	96.0	94.7	92.0	78.5	
D2	3.6	80.4	86.4	82.0	57.2	90.9	93.6	91.8	92.0	76.2	
LSD _{0.05}	8.45	13.81	9.53	8.52	8.45	5.43	3.88	7.19	4.22	7.67	
Nitrogen rate											
N1	5.3	89.6	87.8	84.0	57.7	91.1	94.7	92.9	92.9	77.8	
N2	3.1	83.6	88.2	83.3	61.0	91.3	94.9	93.6	91.1	76.8	
LSD _{0.05}	8.45	13.81	9.53	8.52	8.45	5.43	3.88	7.19	4.22	7.67	
Mean	4.2	86.6	88.0	83.7	59.3	91.2	94.8	93.2	92.0	77.3	
SD	4.9	1.2	6.4	5.6	5.9	4.0	2.7	4.6	2.9	4.8	

D1 = 60 cm row spacing and 5 kg ha⁻¹ seeding rate; D2 = 20 cm row spacing and 20 kg ha⁻¹ seeding rate; N1 = 0 kg ha⁻¹ nitrogen rate; N2 = 150 kg ha⁻¹ nitrogen rate; SD-standard deviation

There is no complete concordance between our results and data of *Stanisavljevic et al. (2011)*. They reported the best germination and vigour of Italian ryegrass seedlings between 270 and 330 DAH, which equates to spring sowing time (March-April) in the succeeding year. Italian ryegrass seeds maintained satisfactory germination levels up to 630 DAH (81%) and 810 DAH (81%), respectively. The results indicated that early spring is the best sowing period for Italian ryegrass, but our results indicated that temperate autumn conditions are also suitable sowing period for Italian ryegrass.

Immediately after harvest and breaking seed dormancy (90 DAH) was highest correlation between germination energy and total germination (Table 3), but 2000 DAH that correlation was decreased to twice lower (0.69 and 0.36%, respectively). Correlation between total germinations and germination energies (DAH90/DAH2000) was similar (0.49% and 0.44, respectively).

Table 3. Correlation coefficients between germination energy and total germination following different storage period of seed

	TG/90	TG/2000	GE/2000
GE90	0.69		0.44
GE/2000		0.36	
TG/90		0.49	

GE/90 = germination energy 90 DAH; TG/90 = total germination 90 DAH; GE/2000- germination energy 2000 DAH; TG/2000- total germination 2000 DAH.

Conclusion

The high seed quality of Italian ryegrass was confirmed with this research and it is slightly influenced by different variants of establishment or by N spring application, but much more by storage period. The data can serve for the determination of a proper storage duration management between harvest and sowing of Italian ryegrass. Although there was a reduction in the total germination during seed ageing of Italian ryegrass up to 2000 DAH (77.3%), this reduction gave almost satisfactory germination in a sense of the market requirement (77%). Average TSW varied from 3.89 to 4.32 g. TSW was influenced by factors forming crop density, while nitrogen had a lower influence on that parameter. Germination increased with the increase temperatures from 10 to 25°C. The results of germination in this study provide aspects for recommendation the best sowing date and, in particular, about the minimum and maximum storage period between harvests and sowing of Italian ryegrass.

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Uticaj agrotehnike na osobine semena italijanskog ljulja pri različitim dužinama skladištenja

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Rezime

Setva semenskog useva italijanskog ljulja obavljena je u 2007 godini u dve gustine setve ($D_1=60$ cm međuredno i 5 kg ha^{-1} setvena norma; $D_2=20$ cm međuredno i 20 kg ha^{-1} setvena norma) i primenom dve količine azota u prihrani (0 and 150 kg ha^{-1}). Posmatran je klijavost i masa 1000 semena italijanskog ljulja u prvoj proizvodnoj godini. Posle junske žetve, seme je skladišteno u standardnim skladišnim uslovima i uzorkovano 90 dana posle žetve (DPŽ), a potom 2000 DPŽ. Posle 90 DPŽ seme je ispitivano na masu 1000 semena, energiju klijanja i ukupnu klijavost na temperaturama klijanja od 10, 15, 20 i 25°C. Seme ljulja je imalo

najbolju životnu sposobnost 90 DPŽ na 20°C i maksimalnu ukupnu klijavost na 15°C, što sugerije da je rana jesen (septembar-oktobar) odgovarajući period za setvu sveže poźnjevenog semena italijanskog lJulja. Seme italijanskog lJulja moēe zadržati zadovoljavajuću energiju klijanja (59,3%) i klijavost (77,3%) i 2000 DPŽ. Dobijeno je kvalitetno seme i primenjeni tretmani pri gajenju semenskog useva nisu menjali značajno kvalitet semena, za razliku od duēine perioda skladištenja, koje je imalo značajan uticaj na kvalitet semena.

Podaci mogu poslužiti za određivanje duēine perioda skladištenja semena između ųetve i setve italijanskog lJulja u uslovima Srbije.

References

- AKPAN E.E.J., BEAN E.W. (1980): The effects of density, date of inflorescence emergence, date of harvesting and temperature upon seed quality in tetraploid hybrid ryegrasses. *Grass Forage Science*, 35, 1, 55-61.
- BEAN E.W. (1973): Seed quality: its variation, control and importance in breeding and varietal assessment. Report of the Welsh Plant Breeding Station, 194-208.
- CHOI G.J., JUNG E.S., RIM Y.W., LIM Y.C., KIM K.Y., SUNG B.R., PARK G.J. (2002): Effects of drill widths and nitrogen application levels in early spring on the growth characteristics and seed productivity of Italian ryegrass (*Lolium multiflorum* Lam.). *Journal of the Korean Society of Grassland Science*, 22, 3, 221-226.
- BEWLEY J.D., BLACK M. (1994): Seeds: physiology of development and germination. New York, 445p.
- HAMPTON J.G. (1986): Effect of seed and seed lot 1000 seed weight on vegetative and reproductive yields of "Grasslands Moata" tetraploid Italian ryegrass (*Lolium multiflorum*). *New Zealand Journal of Experimental Agriculture*, 14, 13-18.
- HILL M.J., HAMPTON J.G., HILL K.A. (1998): Seed quality of grasses and legumes. In: Fairey, D.T., Hampton, J.G. (Eds.), Forage seed production. Vol. 1: temperate species. CAB International, 219-242.
- HUMPHREYS M. ET AL. (2010): Ryegrasses. In: Boller, B.; Posselt, U.K.; Veronesi, F. *Fodder Crops and Amenity Grasses*. New York: Springer., 211-260.
- ISTA - INTERNATIONAL SEED TESTING ASSOCIATION (2010): International Rules for Seed Testing. Bassersdorf, Switzerland.
- SALEHI H, KHOSH-KHUI M (2005): Enhancing seed germination rate of four turfgrass genera by acid treatments. *Journal of Agronomy and Crop Science*, 191, 346-350.
- SIMIĆ A., VUČKOVIĆ S., MALETIĆ R., SOKOLOVIĆ D., ĐORĐEVIĆ N. (2009): Impact of seeding rate and interrow spacing on Italian ryegrass for seed in

the first harvest year. Turkish Journal of Agriculture and Forestry, 33, 5, 425-433.

SIMIĆ A., VUČKOVIĆ S., ĆUPINA B., KRSTIĆ Đ., STANISAVLJEVIĆ R., MILIVOJEVIĆ M. (2010): Impact of management practices on Italian ryegrass seed quality. Journal of Agricultural Sciences, 55, 2, 131-140.

STANISAVLJEVIĆ R., DRAGIĆEVIĆ V., MILENKOVIĆ J., DJUKANOVIĆ L. (2010): Effects of the duration of after-ripening period on seed germinations and seedling size in three fescue species. Spanish Journal of Agricultural Research, 8, 2, 454-459.

STANISAVLJEVIĆ R., ĐOKIĆ D., MILENKOVIĆ J., ĐUKANOVIĆ L., STEVOVIĆ V., SIMIĆ A., DODIG D. (2011): Seed germination and seedling vigour of Italian ryegrass, cocksfoot and timothy following harvest and storage. Revista Ciència e Agrotecnologia, 35, 6, 1141-1148.

STANISAVLJEVIĆ R., VUČKOVIĆ S., SIMIĆ A., MARKOVIĆ J., LAKIĆ Ž., TERZIĆ D., ĐOKIĆ D. (2012): Acid and Temperature Treatments Result in Increased Germination of Seeds of Three Fescue Species. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 40, 2, 220-226.

VUČKOVIĆ S., NEDIĆ M., ZARIĆ D., ZIVANOVIĆ LJ. (1998): Effect of row spacing and sowing rate on Italian ryegrass (*Lolium italicum*) seed yield and quality. Proceedings of 2nd Balkan Symposium on Field Crops, 16-20 June, Novi Sad, Yugoslavia, , 469-473.

VUČKOVIĆ S., SIMIĆ A., ĆUPINA B. STOJANOVIĆ I., STANISAVLJEVIĆ R. (2003): The effect of vegetation area size on grass seed yield. Journal of Agricultural Sciences, 1, 125-134.

WALTERS C., WHEELER L.M., STANWOOD P.C. (2004): Longevity of cryogenically stored seeds. Cryobiology, San Diego, 48, 2, 229-244.