Effects of chemical and biological treatments on germination of onion (*Allium cepa* L.) seeds*

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[Abstract] The effects of osmopriming (OP) in PEG 8000 and hydropriming (HP) on onion "Wolska" seed germination were studied. Moreover, fungicides (Penncozeb and Apron) and biological (Biozym, Promot) treatments on seed germination were investigated. After osmopriming at 10℃ all the *T* values and MGT were reduced significantly compared with control. Hydropiming at 10℃ gave the better results than osmopriming. On the other hand, *T* values and MGT after hydropriming at 20 ℃ did not differ significantly from those in control an OP. Fungicide treatment at 10 ℃ delayed speed of germination. All the *T* values and MGT in fungicide treatment at 20 ℃ did not differ significantly from control. Biological treatments did not affect their speed of germination both at 10 ℃ and 20 ℃. OP and HP at 10 ℃ significantly improved germination capacity (GC) compared with control. Biozym and Promot treatments, osmopriming and hydropriming at 20 ℃ did not affect significantly germination capacity (GC) compared with control.

[Key words] onion; seed germination; priming; fungicide; biological agent

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Onion (*Allium cepa* L.) is one of the most important vegetable crops. Because of its unique flavour and odour, it is an excellent food source in the world. It is also an important vegetable crop in Poland, about 700 thousands tons of onion bulbs are produced in Poland yearly. However, onion seeds usually have a poor quality. Various seed treatments are used widely to improve seed quality. Out of them, seed priming is very common and effective, which has been devised to improve the rate and uniformity of seed germination as well as seed viability[1]. Preplant priming improvements were greater in poor seeds than in more vigorous seeds[2,3]. The effects of priming include increasing germination rate; more uniform emergence; germination under a broader range of environments; improving seedling vigour and growth[4].

The treatment of seeds with microorganisms which are beneficial to plant growth has been the subject of considerable investigation for many years, often with mixed results[5]. Many fungi and bacteria have been tested as seed treatments to provide short-term protection against seed rots and damping-off fungi (*Pythium* spp.) in the soil[6]. In this function, biological seed treatments largely have been effective because the pathogens are limited in time and space and the area of host tissue available for infection is relatively small and can be effectively covered with antagonists[7]. Such microorganisms have been collectively called bio-protectants. Fungi (*Trichoderma* spp.) and bacteria (*Enterobacter* and *Pseudomonas* spp.) have been tested for this purpose.

Penncozeb 80 WP (a. i. 80% mancozeb) was applied for treating seeds in this experiment.

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Maneb is one of the organic sulfur fungicides which is sometimes mixed with zinc ion and results in the formulations known as zinc ion maneb, called mancozeb. The addition of zinc reduces the phytotoxicity of maneb and improves its fungicidal properties[8].

The main goal of this research was to investigate the effects of priming and fungicide and biological treatments on germination of onion seeds.

1 Materials and methods

1.1 Seeds

Onion 'Wolska' seeds supplied by CNOS Seed Company in Pozna were used in the experiment.

1.2 Hydropriming

Seeds were placed in 100 mL flasks and 500 μL of distilled water per 1 g of seeds was added. Then flasks were sealed with Parafilm and incubated in darkness at 20 ℃ for 2 days. Afterwards, the seeds were surface dried with blotting paper, placed in semi-open Petri dishes and dried back at 20 ℃ and 45% relative humidity for 48 h to an equilibrium moisture content.

1.3 Osmopriming

Seeds were primed for 7 days in darkness at 15 ℃ by placing 50 seeds in 9 cm diameter Petri dishes on 4 blotters moistened with 5 mL of Polyethylene glycol (PEG 8000, Sigma Chemical Co.) solution of the osmotic potential of -1.5 MPa. The Petri dishes were sealed with Parafilm. After priming the seeds from each replicate were washed separately under the running tap water for 5 min. and next rinsed three times in sterile distilled water to remove PEG. Then, they were surface dried with blotting paper. Afterwards, the seeds were placed in semi-open Petri dishes and dried back at 20 ℃ and 45% R. H. for 48 h to an equilibrium moisture content.

1.4 Fungicide treatment

Seeds were treated with Penncozeb 80WP (a. i. 80% mancozeb) 5 g/kg seed and Apron 35 SD (a. i. 35% metalaxyl) 1 g/kg seed. Both of the fungicides were recommended by the Institute of Plant Protection in Poland.

1.5 Biological treatment

Two microbial preparations were used for treating seeds: Biozym and Promot. The former contains different bacteria, fungi were mainly *Trichoderma* spp. and enzyme complexes. The latter contains only *Trichoderma koningii* and *T. harzianum*. Both microbiological products were produced in JH Biotech, INC USA. Onion seeds were treated with Biozym and Promot 30 g/kg seed respectively.

1.6 Seed germination

Germination tests were performed according to the ISTA Rules[9]. They were conducted at 20 ℃ and 10 ℃ in darkness using 6 replicates of 50 seeds at each treatment. Seeds were placed in 9 cm diameter Petri dishes containing 6 layers of blotting paper wetted with distilled water. Seeds were considered as germinating when there was a visible protrusion through the seed coat. Germinating seeds were counted daily until no new germs occurred and were removed daily from the dishes. The following parameters of germination were calculated: G_{max} (the percentage of maximum germinating seeds), germination rate (T_1, T_{10}, T_{25}, T_{50} and T_{90}, time to reach germination of 1%, 10%, 25%, 50% and 90% of the total number of germinating seeds), germination uniformity (U_{75-25}, time between 25% and 75% of G_{max}; U_{90-10}, time between 10% and 90% of total germination) and MGT (mean germination time).

Moreover, six replicates of 50 seeds from each treatment were incubated under the same conditions for determination of: 1) the percentage of normal seedlings, i. e. germination capacity (GC); 2) deformed and diseased seedlings; 3) dead seeds; and 4) fresh non-germinated seeds. All evaluations were made according to the ISTA Rules (1996) after 12 days.

1.7 Data analysis

Seed Calculator version 2.1 software[10] was applied to analyse germination data. All statistical results were evaluated by means of variance analysis (ANOVA) followed by Duncan's multiple range test.

2 Results

Fungicide treatment delayed speed of germination, all the T values and MGT were higher than that of control (Table 1). Treating seeds with Biozym and Promot did not affect their speed of germination.

After osmopriming (OP), all the T values and MGT were reduced significantly compared with control.

The values of T_{10}, T_{25}, T_{50} and MGT for hydroprimed seeds (HP) were even significantly lower than that for osmoprimed ones (OP). Hydropriming gave the better results than osmopriming.

After osmopriming (OP) and hydropriming (HP) at 10 ℃ the lower U values than in control were noticed. However, the differences were not statistically significant. Fungicides and biological preparations (Biozym and Promot) did not affect the uniformity of germination at this temperature compared with Control.

Table 1 Effects of seed treatment on seed germination at 10 ℃ d

Treatment	T_1	T_{10}	T_{25}	T_{50}	T_{90}	MGT	$U_{75\text{-}25}$	$U_{90\text{-}10}$
Untreated	4.79 b	6.04 b	6.84 b	7.80 b	10.00 b	7.95 b	2.03 a	3.96 a
Fungicide	5.41 a	6.80 a	7.65 a	8.65 a	10.74 a	8.73 a	2.05 a	3.95 a
Biozym	4.63 b	6.05 b	6.93 b	7.93 b	9.99 b	7.99 b	2.05 a	3.94 a
Promot	4.74 b	6.00 b	6.78 b	7.69 b	9.66 b	7.79 b	1.88 a	3.66 a
OP	4.00 c	5.05 c	5.73 c	6.55 c	8.65 c	6.77 c	1.78 a	3.60 a
HP	3.46 c	4.46 d	5.12 d	5.95 d	8.16 c	6.20 d	1.79 a	3.71 a

Note: MGT. Mean germination time; OP. Osmotic priming; HP. Hydropriming. In columns followed by the same letters are not significantly different at $\alpha = 0.05$ level according to Duncan's multiple range test. The following tables are just the same.

Treating seeds with fungicides did not affect their speed of germination compared with control. All the T values and MGT in fungicide treatment did not differ significantly from control (Table 2).

Both biological preparation treatments (Biozym and Promot) did not affect their speed of germination, considering the values T_{25}, T_{50}, T_{90} and MGT in Biozym treatment as well as T_{50}, T_{90} and MGT in Promot treatment.

Osmopriming (OP) did not improve speed of germination in spite of lower T values, i.e. T_1, T_{10} and T_{25} were lower than control at 20 ℃.

Generally, T values and MGT after hydropriming (HP) did not differ significantly from those in control and OP. Only T_1, T_{10} and T_{25} were considerably lower.

After fungicide, Biozym and Promot treatments, seeds did not show any significant differences in both U values at 20 ℃ compared with control (Table 2).

The uniformity values in OP treatment did not differ significantly from those in fungicide treatment, whereas, the values were significantly higher than that in control. On the other hand, after hydropriming, the uniformity values were significantly high than in control.

Table 2 Effects of seed treatment on seed germination at 20 ℃ d

Treatment	T_1	T_{10}	T_{25}	T_{50}	T_{90}	MGT	$U_{75\text{-}25}$	$U_{90\text{-}10}$
Untreated	2.12 a	2.58 a	2.91 ab	3.36 ab	5.03 cd	3.67 ab	1.10 c	2.45 c
Fungicide	1.92 ab	2.56 a	3.01 a	3.62 a	5.57 bc	3.94 a	1.43 bc	3.01 bc
Biozym	1.77 b	2.32 b	2.70 bc	3.20 b	4.70 d	3.42 b	1.15 c	2.38 c
Promot	1.81 b	2.31 b	2.66 c	3.14 b	4.89 cd	3.45 b	1.15 c	2.58 c
OP	1.70 b	2.20 b	2.58 c	3.15 b	5.93 ab	3.72 ab	1.55 b	3.73 b
HP	0.39 c	1.17 c	1.95 d	3.12 b	6.37 a	3.52 b	2.69 a	5.19 a

The highest percentage of germinating seeds (G_{max}) at 10 ℃ equal 91.3 was observed in control (Table 3). Fungicides and Promot did not affect significantly Gmax compared with control. On the other hand, Biozym, osmopriming (OP) and hydropriming (HP) considerably reduced this parameter.

Germination capacity (GC) for control at 10 ℃ was very low (18.0%). Osmopriming (OP) and hydropriming (HP) significantly improved this parameter compared with control. The lowest percentage of deformed seedlings was obtained in OP treatment (32.7%). The diseased seedlings and dead seeds were not observed in fungicide treatment. Considering fresh non-germinated seeds, there were no significant differences in this parameter between Biozym, Promot treatments, osmopriming and control. After hydropriming, the increase in the percentage of fresh non-germinated seeds was observed compared with control and OP.

Table 3 Effects of seed treatment on germination parameters at 10 ℃ %

Treatment	G_{max}	GC	Deformed seedlings	Diseased seedlings	Dead seeds	Fresh non-germinated seeds
Untreated	91.3 a	18.0 c	69.7 b	3.0 a	5.3 b	4.0 b
Fungicide	87.7 abc	15.0 d	72.0 b	0.0 b	0.0 c	13.0 a
Biozym	83.3 cd	8.0 e	80.7 a	0.7 b	7.7 ab	3.0 b
Promot	89.0 ab	17.7 cd	71.0 b	0.7 b	6.7 ab	4.0 b
OP	86.3 bc	51.7 a	32.7 d	1.7 ab	10.7 a	3.3 b
HP	79.7 d	40.7 b	40.7 c	0.3 b	6.0 b	12.3 a

The highest percentage of germinating seeds (G_{max}) at 20 ℃ equal 90.0 was observed in Biozym treatment (Table 4). Biozym treatment gave better results than fungicide, Promot treatments and OP treatment, however, the differences were not statistically significant. Fungicide, Promot and hydropriming (HP) treatments did not affect significantly G_{max} compared with control.

Biozym and Promot treatments, osmopriming (OP) and hydropriming (HP) did not affect significantly germination capacity (GC) compared with control.

There were no significant differences in the percentage of deformed seedlings at 20 ℃ between control and all the treatments.

The diseased seedlings were not observed in fungicide treatment. The lowest percentage of dead seeds from all the treatments was obtained in *F* treatments. Considering fresh non-germinated seeds, there were no significant differences in this parameter in Biozym, Promot treatments and osmopriming compared with control. After hydropriming (HP), the increase in the percentage of fresh non-germinated seeds was observed compared with control and OP.

Table 4 Effects of seed treatment on germination parameters at 20 ℃ %

Treatment	G_{max}	GC	Deformed seedlings	Diseased seedlings	Dead seeds	Fresh non-germinated seeds
Untreated	82.0 bc	69.0 b	9.3 a	10.3 a	10.7 a	0.7 b
Fungicide	86.0 ab	84.0 a	7.3 a	0.0 c	1.3 c	7.3 a
Biozym	90.0 a	72.0 b	11.3 a	6.3 a	10.3 a	0.0 b
Promot	85.7 ab	71.3 b	8.7 a	8.0 a	11.3 a	0.7 b
OP	89.0 a	74.3 b	8.7 a	6.0 a	10.3 a	0.7 b
HP	75.0 c	74.0 b	8.0 a	2.7 b	5.3 b	10.0 a

3 Discussion

The values T_{10}, T_{25}, T_{50} and MGT at 10 ℃ for hydroprimed seeds (HP) were even lower than that of osmoprimed ones (OP). Hydropriming gave the better results than osmopriming. After hydropriming (HP) at 10 ℃ the lower U values than that of control were noticed. Recently, more attention is paid to hydropriming in the restricted volumes of water for the enhancement of seed performance. This method is more economical, ecological and more useful than osmopriming and matriconditioning for conditioning of large volumes of seeds. Hydropriming is a simple and inexpensive priming method used to invigorate seed, which involves hydration of seeds in distilled water followed by dehydration[11]. However, it is difficult to control the amount of water absorbed by seed[12]. A developed method, in which *Brassica oleracea* seeds were hydrated for short periods of up to 8h in columns of aerated water has been showed to result in clear and consistent improvements in germination rate, seedling root length and seed vigour[13]. Hydropriming would be beneficial to scale-up hydroprimed seed in the practice.

Osmopriming accelerated seed germination at 10 ℃. However there was no improvement in the speed of germination at 20 ℃. Brocklehurst and Dearman[14] found that after onion seeds were primed in PEG solution, the mean germination time was reduced by 3- 5 days. At the same time, their study on seedling emergence also revealed that priming generally reduced the spread of emergence times[15]. Ali et al.[16] also investigated the effect of priming in PEG solution on onion seed germination, and reported that most of the treatments led to a rapid germination response. Tylkowska and van den Bulk[17] showed that priming resulted in faster and more uniform germination for all three carrot seed lots, although the uniformity of germination as determined by the U_{75-25} values was not always significantly better after priming.

No reports were found on the effects of Promot and Biozym treatments on onion seed germination up to now. Hence, further research should be continued.

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化学和生物制剂处理对洋葱种子萌发的影响

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摘　要: 研究了洋葱种子“Wolska”在 PEG 8000 和水引发条件下的萌发情况, 以及杀菌剂(Penncozeb 硫锌制剂和 Apron 爱普荣)和生物制剂(Biozym 生物合剂, Promot 助长剂)对洋葱种子萌发的影响。结果表明, 10 ℃ PEG 引发处理中, 所有的达到最大萌发率的时间和平均萌发时间较对照极显著减小, 在该温度下, 水引发处理效果比 PEG 引发好; 20 ℃时, 水引发处理达到最大萌发率的时间和平均萌发时间与对照及 PEG 处理间没有显著差异。杀菌剂处理在 10 ℃时延缓了萌发速度, 而在 20 ℃时所有处理达到最大萌发率的时间和平均萌发时间与对照无显著差异。生物制剂处理在 10 ℃和 20 ℃下对萌发速度均没有改善; 与对照相比, 10 ℃的 PEG 引发与水引发处理极大地改善了萌发力; 20 ℃时的生物制剂处理、PEG 引发和水引发处理与对照相比, 均未显著改善萌发力。

关键词: 洋葱; 种子萌发; 引发剂; 杀菌剂; 生物制剂

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