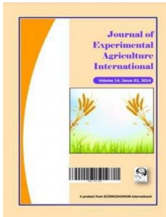

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Influence of Germination Aids on Germination of Different *Capsicum sp.*

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Authors' contributions

This work was carried out in collaboration between both authors who designed the study. Author NJ managed the literature searches and performed the experimental parts of the study. Author MW performed the statistical evaluation of the data and wrote the manuscript. Both authors read and approved the final manuscript.

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ABSTRACT

Aims: To provide data on the influence of common germination aids for additional pepper species than *Capsicum annuum*, additional species and wild types have been tested with priming media applicable to both hobby and commercial growers.
Place and Duration of Study: Germany, January to September 2017.
Methodology: 3x 50 seeds of four different *Capsicum* cultivars "CAP357" (*C. pubescens* L.), "Ancho Poblano" (*C. annuum* L.), "Ulupica from La Paz" (*C. eximium* Hunz.) and "Inca Surprise" (*C. baccatum* var. *baccatum* L.) were primed with either deionized water, 1%(w/v) KNO_3, 1%(w/v) gibberellic acid or 1%(w/v) acetylsalicylic acid. 3x 50 untreated seeds of each cultivar were as control. The seeds were then put to germination on moistened filter paper, maintaining constant temperature (29 ± 1°C) and air-humidity (85 ± 5%). Seeds were evaluated daily regarding radicle formation for 30 days.
Results: Highest germination rate and germination speed were observed using gibberellic acid as priming agent with overall germination >90%, followed by potassium nitrate, which improves germination especially for seeds that take longer to germination. Acetylsalicylic acid caused

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germination rates and speed to decrease. Hydropriming with water showed no significant effect compared to unprimed seeds.

Conclusion: Priming with appropriate media can effectively speed up germination on varieties that take longer to germination such as wild types. On already fast germinating seeds the effect is smaller. However, a more uniform germination can be achieved.

Keywords: Capsicum; germination; priming; seeds; potassium nitrate; gibberellic acid.

ABBREVIATIONS

ANN : Capsicum annuum L. cv. "Ancho Poblano"
BAC : Capsicum baccatum var. baccatum L. cv. "Inca Surprise"
EXI : Capsicum eximium Hunz. cv. "Ulupica from La Paz"
PUB : Capsicum pubescens L. cv. "CAP357"
MGT : Mean Germination Time
G50 : Time to 50% Germination
GA-3 : Gibberellic acid
ASA : Acetylsalicylic acid

1. INTRODUCTION

Hot (chili) pepper or paprika (*Capsicum spp.*) is one of the most consumed vegetable worldwide. It is not only a large market in various regions of the world, especially India and Pakistan, but the diversity of the *Capsicum* family recently has attracted many hobby growers all over the world. A high germination rate is favorable for every grower but can become substantially relevant for commercial growers, where every plant will eventually generate income.

In order to ensure successful germination, many germination aids or seeds priming agents have been investigated in the past [1]. Potassium nitrate KNO_3 has already been identified as good priming agents by Rivas et al. [2] and O'Sullivan et al. [3] in 1984. More recently various other priming agents such as acetylsalicylic acid [4], 1-hexadecene [5], polyethylene glycol [4,6], NaCl [4,7,8] and again KNO_3 [4,6,9], all reporting beneficial influence on germination speed, seedling uniformity and/or salt tolerance of *Capsicum*. Gibberellic acid (gibberellin) is also likely to speed up the germination process as previous studies on pepper and tomato show [10,11].

The germination percentage is one of the first and important parameters in order to evaluate seed health and can also be used to predict future seedling and plant development. High germination percentage and uniform germination time are closely linked to a positive development of the plant. Priming seems to be most advantageous for seed lots with low initial (unprimed) germination percentage [12,13].

However, all major research mainly focuses on *Capsicum annuum* L. cultivars as they are commercially most relevant, while other domesticated species such as *Capsicum pubescens* (Rocoto), *Capsicum baccatum* and wild types are rarely subject of germination studies. There are emergence data of various chili pepper cultivars [14], however, the study investigated cold temperature germination without prior priming. To our knowledge, there are no dedicated germination studies on *Capsicum* species other than *Capsicum annuum* L.

The present research now provides data for simple and inexpensive seed preparation comparing the germination rates and speeds of a range of *Capsicum sp.* seeds treated with different media prior to germination. The results may not only be relevant for commercial pepper growers but also to hobby growers who want to improve or accelerate their seed germination. Media for seed treatment except for gibberellic acid are widely available and fairly easy to handle for both commercial and hobby growers.

2. METHODOLOGY

2.1 Germination Procedure

Seeds from "CAP357" (*C. pubescens* L., PUB), "Ancho Poblano" (*C. annuum* L., ANN), "Ulupica from La Paz" (*C. eximium* Hunz., EXI) and "Inca Surprise" (*C. baccatum* var. *baccatum* L. wild type, BAC), have been harvested from self-grown plants in autumn 2016 and stored in the dark at 2-6°C after dehydration over silica gel. The plants of these cultivars have been the most healthy and productive in the season 2016 when seeds have been harvested.

50 seeds of each cultivar were subjected to each of the following priming agents for 24 h: deionized water, KNO_3 (1%(w/v)), acetylsalicylic acid (1%(w/v)), and gibberellic acid (1%(w/v)). 50 untreated seeds of each lot were used as control group. Due to a limited number of seeds, a separate viability test has not been performed, however, the influence of the priming agents can be evaluated by comparing data to those of the control group.

Seeds were germinated in petri dishes on moistened filter paper in an incubator under controlled conditions (29 ± 1°C, 85 ± 5%rh) for 30 days. Status of germination was checked every 24 hours with radicle formation and subsequent seedling emergence indicating successful germination [9,15]. Abnormal radicles or seedlings have not been detected. Weighing of the seeds has been performed before and after priming in lots of 50 seeds/lot.

Every experiment described above has been performed in triplicates (3x 50 seeds for each cultivar and priming agent).

2.2 Calculations

Overall Germination (G) describes the percentage of successfully germinated seeds (no abnormal seedlings) in each lot after 30 days. Mean Germination Time (MGT) is a key parameter often used [15,16] to compare the speed of seed germination. It is calculated by Equation 1:

$$MGT = \frac{\sum_{i=1}^{n}(n_i \cdot d_i)}{\Sigma n_i} \quad (1)$$

with n_i being the number of germinated seeds on each day d. A lower MGT represents a faster germination. Time to 50% germination (G_{50}) as previously described [17] and modified [18] is another index to describe the speed of seed germination.

$$G_{50} = t_i + \frac{\left(\frac{N}{2} - n_i\right) \cdot (t_j - t_i)}{n_j - n_i} \quad (2)$$

N denotes the total number of germinated seeds, n_i and n_j represent the cumulative number of germinated seeds adjacent to N/2, respectively ($n_i < N/2 < n_j$). t_i and t_j account for the respective days on which n_i and n_j have been counted.

For ANOVA with post-hoc Tukey test on significance ($P < .05$) within tested groups statistical evaluation has been performed using Origin Pro 2016.

3. RESULTS AND DISCUSSION

Weighing of dry and primed seeds (Table 1) revealed that water uptake is independent of the priming medium but varies for the different *Capsicum* species. Water uptake ranges from 62 % for BAC to 150 % for PUB. A similar water uptake regardless of the priming agent is not unexpected considering all priming media are aqueous solutions (1%). Seeds of smaller size (BAC and EXI) have less surface available for water intrusion, while larger seeds (ANN and PUB, which additionally have a rougher, more uneven surface) are likely able to take up more water.

Overall Germination (Fig. 1) after 30 days is first of all strongly dependent on the species. While ANN and PUB display over 90% germination regardless of the priming method, germination of BAC and EXI is influenced by the choice of the priming agent. In general, no priming and simple deionized water yield almost the same results, and the use of ASA even decreases overall germination for all investigated species (in contrast to [4], where ASA improved germination). On the other hand, priming with KNO_3 or GA-3 markedly increases germination with GA-3 having the highest effect on seeds. MGT (Fig. 2a) and G_{50} (Fig. 2b) are both parameters describing the speed of germination. As with Overall Germination, speed is dependent firstly on the species and secondly on the used priming agent. Again, no priming and water priming yield comparable results, ASA prolongs germination of all species, while KNO_3 and GA-3 significantly shorten germination times, the latter being in accordance with [4]. In Fig. 3, both overall germination and germination speed are shown in a graph depicting cumulated number of germinated seeds over time.

Germination strongly depends on the individual species. Using no priming agent or only water, both *C. annuum* and *C. pubescens* already have high germination rates above 90% indicating very viable seeds, while *C. eximium* and the wild type of *C. baccatum* have decreased germination rates around 30 to 50%. Compared to no priming, simply priming with water (hydropriming) does not significantly enhance overall germination (except for EXI, see Table 2). Even the speed of germination expressed as MGT or G_{50} is not noticeably improved by hydropriming

Table 1. Seed weight dry and after 24 h of priming (50 seeds/measurement)

Priming agent	Weight (dry) [g]				Weight (primed) [g]			
	BAC	**PUB**	**EXI**	**ANN**	**BAC**	**PUB**	**EXI**	**ANN**
ASA[a]	0.139 ± 0.002	0.326 ± 0.011	0.276 ± 0.006	0.302 ± 0.002	0.220 ± 0.002	0.802 ± 0.009	0.493 ± 0.009	0.585 ± 0.006
Water	0.141 ± 0.005	0.323 ± 0.008	0.262 ± 0.010	0.306 ± 0.006	0.227 ± 0.005	0.810 ± 0.026	0.495 ± 0.004	0.598 ± 0.008
KNO_3	0.136 ± 0.002	0.319 ± 0.008	0.282 ± 0.01	0.300 ± 0.002	0.219 ± 0.002	0.800 ± 0.002	0.509 ± 0.009	0.584 ± 0.015
GA-3	0.135 ± 0.003	0.320 ± 0.005	0.271 ± 0.005	0.301 ± 0.002	0.226 ± 0.007	0.798 ± 0.006	0.487 ± 0.011	0.637 ± 0.045
None	0.137 ± 0.002	0.315 ± 0.011	0.270 ± 0.002	0.304 ± 0.002	n.a.	n.a.	n.a.	n.a.
Mean 50 seeds	0.137 ± 0.003	0.321 ± 0.008	0.272 ± 0.009	0.303 ± 0.003	0.223 ± 0.005	0.803 ± 0.013	0.496 ± 0.011	0.601 ± 0.031
					+62%	+150%	+82%	+99%

Means ± Standard Deviation in g of a 3-fold determination. [a]acetylsalicylic acid

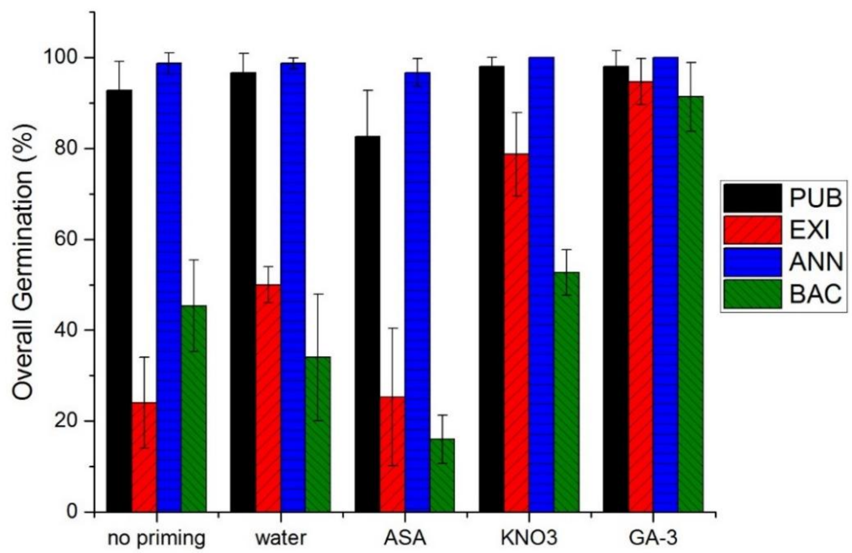

Fig. 1. Overall germination for different priming media

Means ± Standard Deviation of three experiments

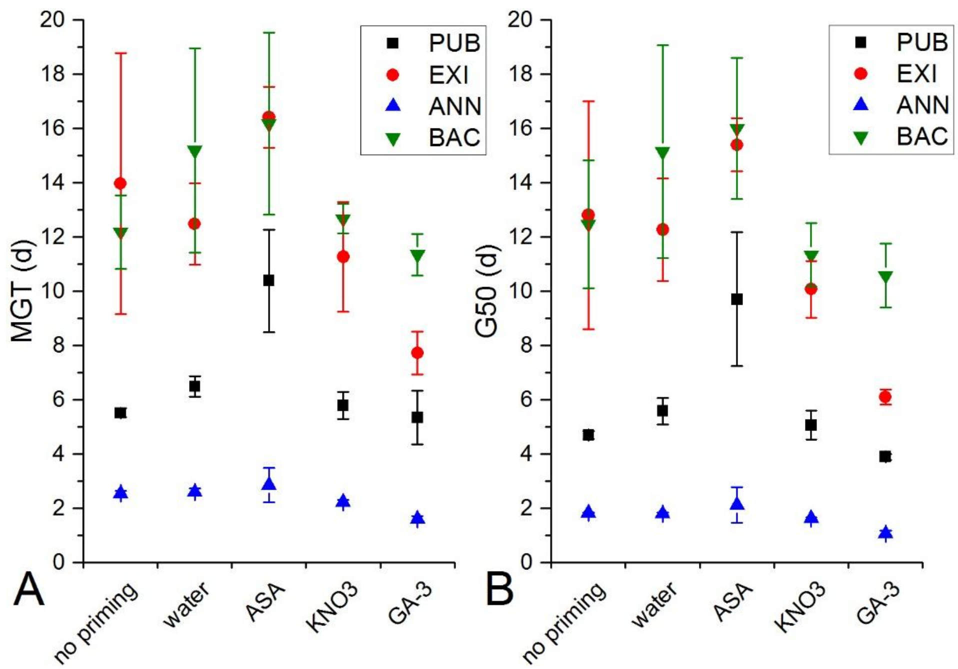

Fig. 2. a) Mean Germination Time (MGT) and b) Time to 50% germination (G50) for different priming media

Means ± Standard Deviation of three experiments

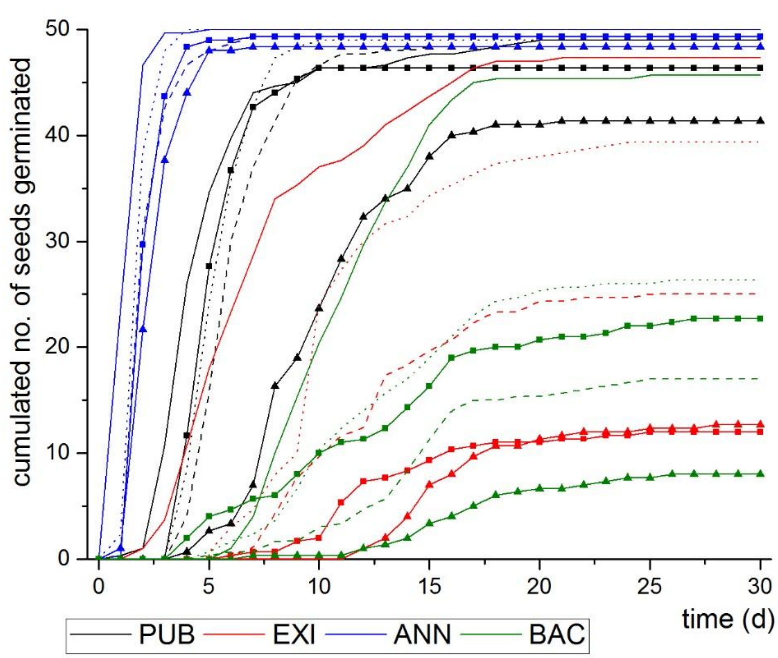

Fig. 3. The cumulated number of germinated seeds over time. No priming (-■-), water (dashed ---), ASA (-▲-), KNO3 (dotted ···), GA-3 (solid —)

Table 2. Results of ANOVA *post-hoc* Tukey test on significance. Comparison of overall germination and germination speed depending on priming medium *vs.* unprimed seeds

		***P* values compared to unprimed seeds**			
		Water	**ASA**	**KNO3**	**GA3**
G	PUB	.917	.305	.802	.802
	EXI	**.047**	.999	**<.001**	**<.001**
	ANN	.999	.658	.886	.886
	BAC	.564	***.017**	.852	**<.001**
MGT	PUB	.754	***.001**	.997	.999
	EXI	.945	.756	.688	.073
	ANN	.999	.723	.700	**.022**
	BAC	.554	.306	.999	.992
G50	PUB	.875	***.002**	.995	.905
	EXI	.998	.599	.559	**.023**
	ANN	.999	.752	.920	.063
	BAC	.680	.449	.977	.876

***Bold** P values (P < .05) indicate significant enhancement of germination,*
Bold** P values with **asterisk (P < .05) indicate significant negative influence on germination*

for 24 h. On the contrary, in cases of PUB and BAC, unprimed have slightly lower (although not significant) MGT and G_{50} values indicating faster germination.

Priming with ASA leads to significantly reduced overall germination for PUB, ANN and BAC and unchanged values for EXI. Additionally, germination speed is to some extent significantly decreased for all species. This leads to the conclusion that ASA is not suitable as priming agent for pepper varieties.

Potassium nitrate as a common priming agent leads to improved germination rates for all investigated species, however, a significant increase is only observed for EXI due to the already high germination rates of ANN and PUB. For BAC and EXI lower MGT and G_{50} are also observed, confirming a positive effect on germination on seeds that take longer to germinate. This positive effect has previously been described by several groups [3,4,9,2], however, the present study contributes additional data for other varieties than *C. annuum*. Again, results for germination speed do not significantly differ from unprimed or water-primed seeds, although germination is more uniform and has a smaller variance.

Using gibberellic acid GA-3 as priming agent yields the highest overall germination rates. Even BAC and EXI now exceed 90% overall germination. MGT and especially G_{50} are lower for all species indicating a faster and also more uniform germination, which supports previous findings [10,11]. In cases of ANN and EXI, G_{50} of GA-3-treated seeds is reached almost twice as fast as compared to unprimed seeds. When sowing older seeds or species that seem to be harder to germinate, priming with GA-3 is a very effective method to enhance germination.

4. CONCLUSION

Priming of pepper seeds can markedly enhance both overall germination and germination speed, especially for seeds that take longer time to germinate such as wild types or older seeds. Priming with water or no priming at all show similar effects, indicating that water priming is just lost time, at least when sowing on filter paper. KNO_3 and gibberellic acid as priming agents are both enhancing germination, the latter being more effective but also less easy to handle. Acetylsalicylic acid prolongs germination and reduces overall germination and is thus not recommended.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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