ວາລະສານວິທະຍາສາດມະຫາວິທະຍາໄລສຸພານຸວົງ, ຄົ້ນຄວ້າວິໄຈສະຫະສາຂາວິຊາ, ວາລະສານເປີດກວ້າງ
ສະບັບທີ 6, ເຫຼັ້ມທີ 2, ກໍລະກົດ - ທັນວາ 2020, ເລກທະບຽນ ISSN 2521-0653

ຜົນຂອງການໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ຕໍ່ການແຕກງອກຂອງເມັດພັນບັກວີດ ຊະນິດຫວານ ແລະ ຊະນິດຂົມ[1]

ຊໍຊົ່ງ ເບຣ່ຍເຕຍ[2], ຊາດຊາຍ ໂຂນົງນຸດ[*] ແລະ ໂຈນໂຣ ພາກ[**]

ພາກວິຊາ ວິທະຍາສາດ ແລະ ເຕັກໂນໂລຊີອາຫານ, ຄະນະກະເສດສາດ ແລະ ຊັບພະຍາກອນປ່າໄມ້, ມະຫາວິທະຍາໄລສຸພານຸວົງ, ລາວ.

[*] ສາຂາເຕັກໂນໂລຊີຊີວະພາບ, ຄະນະອຸດສາຫະກຳກະເສດ, ມະຫາວິທະຍາໄລຊຽງໃໝ່, ປະເທດໄທ.

[**] ພາກວິຊາ ເຕັກໂນໂລຊີຊີວະສຸຂະພາບ, ຄະນະວິທະຍາສາດຊີວະການແພດ, ມະຫາວິທະຍາໄລແຫ່ງຊາດກາງອອນ, ສ. ເກົາຫຼີ.

ບົດຄັດຫຍໍ້

ບົດຄວາມສະບັບນີ້ໄດ້ອະທິບາຍສັກກາຍຍະພາບການໃຊ້ເຊື້ອເບັກທີເຣຍເອນໂດໄຟຕ໌ສາຍພັນ *Herbaspirillum rubrisubalbicans* ເພື່ອກະຕຸ້ນການງອກຂອງເມັດບັກວີດຊະນິດຫວານ ແລະ ຊະນິດຂົມ (common and tartary buckwheat). ງານທົດລອງໄດ້ຈັດຕັ້ງປະຕິບັດ ໂດຍເພາະເມັດໃນຕູ້ບ່ຽມເພາະທີ່ບໍ່ມີແສງ ແລະ ຄວບຄຸມອຸນຫະພູມທີ່ 20, 25 ແລະ 30ºC ເປັນເວລາ 7 ວັນ. ໃຊ້ແຜນການທົດລອງແບບ CRD, ມີ 3 ຊ້ຳ ແລະ 3 ສິ່ງທົດລອງ, ນຳໃຊ້ເມັດພັນບັກວີດຈຳນວນ 30 ເມັດ/ສິ່ງທົດລອງ/ຊ້ຳ. ຜົນການທົດລອງສະແດງໃຫ້ເຫັນວ່າ ການໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ກັບເມັດບັກວີດມີຜົນຊ່ວຍເລັ່ງອັດຕາການແຕກງອກຂອງເມັດເພີ່ມຂຶ້ນ ໂດຍເພີ່ມຄວາມແຂງແຮງໃນການງອກສູງເຖິງ 90-95% ແລະ ເພີ່ມອັດຕາການງອກໂດຍລວມສູງເຖິງ 95-100% ໂດຍສະເພາະເມັດບັກວີດທີ່ໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ຄວາມເຂັ້ມຂຸ້ນ 10 ແລະ 20% (v/v) ເປັນເວລາ 4 – 8 ຊົ່ວໂມງກ່ອນເພາະເມັດ ເຊິ່ງເຫັນວ່າມີຜົນສູງກວ່າໃນສະພາວະອື່ນໆ. ຢ່າງໃດກໍ່ຕາມ, ເມື່ອເພີ່ມຄວາມເຂັ້ມຂຸ້ນຂອງເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ເປັນ 40% (v/v) ແລະ ໄລຍະເວລາໃນການໃສ່ເປັນ 12 ຊົ່ວໂມງ ມີຜົນເຮັດໃຫ້ອັດຕາການແຕກງອກລຸດລົງຢ່າງວ່ອງໄວທຽບກັບສະພາວະອື່ນໆ. ໃນການສຶກສາຄັ້ງນີ້ເຫັນໄດ້ວ່າ ສະພາວະອຸນຫະພູມທີ່ເໝາະສົມຕໍ່ການແຕກງອກຂອງເມັດບັກວີດທັງສອງຊະນິດແມ່ນຢູ່ລະຫວ່າງ 20 - 30ºC ແລະ ລະຫວ່າງຊະນິດພັນພົບວ່າ ການໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ມີຜົນຕໍ່ການແຕກງອກຂອງເມັດບັກວີດຊະນິດຫວານສູງກວ່າບັກວີດຊະນິດຂົມ ເມື່ອທົດສອບພາຍໃຕ້ສະພາວະເງື່ອນໄຂຕ່າງໆ. ຜົນການທົດລອງສະຫຼຸບໄດ້ວ່າ ເມັດພັນບັກວີດເມື່ອໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ສາຍພັນ *Herbaspirilum rubrisubalbicans* ໃນອັດຕາຄວາມເຂັ້ມຂຸ້ນ ແລະ ໄລຍະເວລາທີ່ເໝາະສົມ ເຊັ່ນ ຄວາມເຂັ້ມຂຸ້ນປະມານ 10-20% (v/v) ເປັນເວລາ 4-8 ຊົ່ວໂມງ ມີປະສິດທິພາບສົ່ງເສີມການແຕກງອກ, ຄວາມແຂງແຮງຕົ້ນອ່ອນ ແລະ ຕົ້ນກ້າມີການພັດທະນາການ ແລະ ຈະເລີນເຕີບໂຕໄດ້ດີໃນບັກວີດທັງສອງຊະນິດ.

ຄຳສຳຄັນ: ເບັກທີເຣຍເອນໂດໄຟ, ການແຕກງອກ, ຊະນິດບັກວີດ.

---

[1] ***ການອ້າງອີງພາສາລາວ:***

ຊໍຊົ່ງ ເບຣ່ຍເຕຍ ແລະ ຄະນະ. (2020). ***ຜົນຂອງການໃສ່ເຊື້ອເບັກທີເຣຍເອນໂດໄຟດ໌ຕໍ່ການແຕກງອກຂອງເມັດພັນບັກວີດຊະນິດຫວານ ແລະ ຊະນິດຂົມ***, *ວາລະສານວິທະຍາສາດມະຫາວິທະຍາໄລສຸພານຸວົງ, ສະບັບທີ: 6, ເຫຼັ້ມ 2, ໜ້າທີ: 37-47.*

[2] ***ຜູ້ຮັບຜິດຊອບຂຽນ:***

ຊໍຊົ່ງ ເບຣ່ຍເຕຍ, ພາກວິຊາ ວິທະຍາສາດ ແລະ ເຕັກໂນໂລຊີອາຫານ, ຄະນະກະເສດສາດ ແລະ ຊັບພະຍາກອນປ່າໄມ້, ມະຫາວິທະຍາໄລສຸພານຸວົງ, ໂທ: 020-23512104; ອີເມລ: s_briatia@hotmail.com.

Effect of Endophytic Bacterium Inoculation on Sprouts and Microgreens Growth of Common Buckwheat (*Fagopyrum essculentum*) Species

Xoxiong Briatia[3], Chartchai Khanongnuch[*] and Cheol Ho Park[**]

Department of Food Science and Technology, Faculty of Agriculture and Forest Resource, Souphanouvong University, Laos

[*] *Division of Biotechnology, School of Agro-Industry, Faculty of Agro-Industry, Chiang Mai University, Thailand*

[**] *Department of Bio-Health technology, College of Biomedical Science, Kangwon National University, Chuncheon, Korea*

ABSTRACTS

This paper describes the potential use of endophytic bacterium strains (*Herbaspirillum rubrisubalbicans*) to stimulate seed germination in both common and tartary buckwheat. The experiment was conducted in a growth chamber under dark conditions and temperature maintained at 20, 25 and 30ºC, for 7 days. Thirty seeds for each treatment with three replications in completely randomized design (CRD) were used. The results showed that seeds inoculated with endophytic bacterium induced increases in seed germination parameters, in some cases achieving increases seed germination vigor rate up to 90-95% and total seed germination rate up to 95-100%, particular when the seeds inoculated with 10 and 20% (v/v) concentrations and treatment durations for 4 to 8 h, which showed higher than other germinated conditions. However, increased inoculants concentration and treatment durations up to 40% (v/v) and 12 h may be decreased seed germination parameters rapidly compared to others. In present study, the optimum temperature conditions for both common and tartary buckwheat seed germination test would be ranged from 20 to 30ºC, and between the species found that seed inoculation with endophytic bacterium showed higher seed germination parameter in common buckwheat than tartary buckwheat under each tested conditions. Finally, our conclusion that seed application with *Herbaspirilum rubrisubalbicans* strain in a proper concentrations and durations (10 to 20% (v/v), for 4 to 8 hours) may be recommended as a most effective treatment to promote seed germinations, seedling vigor and seedling establishment in both common and tartary buckwheat species.

***Key words**: Endophytic bacterium, seed germination, buckwheat species.*

[3] ***Corresponding Author**:*

Xoxiong Briatia, Department of Food Science and Technology, Faculty of Agriculture and Forest Resource, Souphanouvong University, Laos

Tel: 020-23512104; E-mail: s_briatia@hotmail.com)

1. Introduction

Buckwheat (*Fagopyrum esculentum* and *Fagopyrum tataricum*) is an alternative crop that belongs to the Polygonaceae family, and has a strong ecological adaptability so that it can grow well in almost all kinds of disadvantageous living environments (Born and Corns, 1958). Buckwheat is grown in many Europe and Asia countries (Popović *et al*., 2014), as an important functional food crop, the most popular one is called buckwheat noodles, which very popular in Japan, China, Korea and Italy (Akaya and Sun. 1992; Lin and Zhang, 2001). Recently, many researchers have focused on the development of buckwheat as a potential functional food material (Lin and Zhang, 2001), which lead the production areas and average yield have a rising tendency. Popović *et al*. (2014) reported that during the year of 2010-2011 about 2.113 million hectares of buckwheat was sown annually worldwide and the average yield was 913 kg ha^{-1}. However, the yield is still low compared to other grain crop. Several researches on buckwheat production, breeding, and cultivation have been conducted, and reported many achievements were related to agricultural techniques (Akaya and Sun, 1992; Wang and Campbell, 2000). Seed is one of the most important inputs for higher grain production and quality seeds required for rapid germination and synchronous seedling emergence, and development. Born and Corns (1958) found that a high degree of dormancy is normally exhibited by seeds of *F. esculentum* and *F. tartaricum* species at harvest and after storage in a period of time. Seed dormancy is regarded as the failure of an intact viable seed to complete germination under favorable conditions and provides an escape from suboptimal germination conditions in seasonal or spatially heterogeneous environments (Born and Corns, 1958; Wang and Campbell, 2000). Low seed vigor and seed dormancy is undesirable characteristics of farmers and researchers that results in a low productivity. Several studies on seed germination and seed emergence revealed the beneficial effects of seed priming by several ways such as light, heat, smoke, soaking, leaching, temperature, chilling, scarification, gamma rays and salinity (Koger *et al*., 2004; Mathur, 1989; Tzortzakis, 2009), and cutting or removing the seed coat and pericarp (Wang and Campbell, 2000), can promote the germination of dormant seeds. Moreover, seed treatment with plant growth hormones such as gibberellic acid (GA_3), indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), naphthalene acetic acid (NAA), and benzyladenine (BA), can improve seed germination percentages (Dhoran and Gudadhe, 2012; Zare *et al*., 2011). However, poor seed germination is a greatest concerns, thus, seed germination test is very important processed to determine the seed vigor, emergence and seedling establishment (ISTA, 2006). There are many factors influencing seed germination in general, plant growth promoting rhizobacteria, microbial inoculants have been used for treatment seed germination and seedling growth in various crops (Mia *et al.*, 2012; Peng *et al.*, 2009). Significant increases in seed germination and seedling vigor of agronomical important crops in response to inoculation with plant growth promoting rhizobacteria has been reported (Oliveira *et al*., 2002; Gholami *et al.*, 2009). Similarly, reported that *Azospirillum, Pseudomonas, Azotobacter, enterobacter, Acienetobacter* (Shaukat *et al*., 2006) and effective microorganisms (EM1) (Ertekin, 2011), could improve seed germination.

However, no documentations reported about use of endophytes in improvement of seed germination and seedling growth of *F. esculentum* and *F. tataricum* species. Thus, this study was conducted to elucidate the isolation and identification of endophytic bacterium and investigated the effect of selected endophytic bacterium inoculation on seed germination of common and tartary buckwheat species.

2. Materials and Methods

The experiment was conducted at the Microbiology Laboratory, Department of Food Science and Technology, Faculty of Agriculture and Forest Resource, Souphanouvong University, Laos and the Microbiology Laboratory, Division of

Biotechnology, Faculty of Agro-Industry, Chiang Mai University, Thailand. The cultivar of buckwheat (*Fagopylum*) used in this study were *F. esculentum* "Common buckwheat" and *F. tataricum* "Tartary buckwheat". The seeds were obtained from a farmer of Longlanh Village, Luang Prabang, Laos. Endophytic bacterium strains (*Herbaspirillum rubrisubalbicans*) was isolated from common buckwheat seedling stems and use throughout the experiment.

Inoculums preparation

The endophytic bacterium (*H. rubrisubalbicans*) was grown in modified nutrient agar (NA) and nutrient broth (NB) (Atlas, 1999). A single colony was selected and inoculated into NB medium, then incubated at 37°C for 8-10 h under shaking conditions. Exponentially growing cells were harvested by centrifugation at 4,000 g for 20 min, and resuspended in either sterilized normal saline (0.85% NaCl) solutions to obtain the final cell densities of 10^8cfu.ml^{-1} and used as inoculums.

Seed preparation and seed treatment

The inoculums were prepared with the proper diluents for the treatments by using sterile distilled water to obtain a final concentration of 10, 20 and 40% (v/v), respectively. Both of buckwheat seeds were surface-sterilized with 2.5% (v/v) sodium hypochlorite for 3 min, and rinsed thoroughly in sterile distilled water for 4times with 1 min duration. Seeds were soaked in liquid suspension of endophytic bacterium (10^8cfu.ml^{-1}) by separately each treatment for 0, 4, 8 and 12 h, respectively, at 30±1°C.

Seed germination test

Buckwheat seed germination tests were carried out by a top of paper (TP) method. Thirty seeds for each treatment with three replications in completely randomized design (CRD) were used. The seeds were placed directly onto a sterilized moist filter paper in petridishes and covered with lid, thereafter transferred to germinate in a growth chamberunder the dark condition, and temperatures maintained at 20±1°C, 25±1°C and 30±1°C, for 7 days, and sterile distilled water was also supplied when necessary. Germination parameters:number of seed germination per day (GD), seed germination vigor rate (GV) in the first of four days after seeding and total of seed germination rates (GR) in seven days after seeding were investigated according to the International Rules for Seed Testing Association for the *Fagopyrum* species (ISTA, 2006), and seed germination index (GI) was calculated according to the formula described by Perry (1984): ***GI = n/d***, where, n = number of seedlings emerging on day 'd' and d = day after planting.

Statistical analysis

Statisticalanalysis of all tests was carried out using Statistix software version 8.0.FL. Data was analyzed with ANOVA and Least significant difference (LSD) test at $P \leq 0.05$ levelwas used to separate the means when the ANOVA F-test indicated a significant effect of the treatments.

3. Results

Seed germination test at 20±1°C

Common buckwheat seed germination parameters: seed germination vigor rate (%GV), total seed germination rate (%GR), seed germination index (GI) and number of seed germination per day (GD) significantly affected by all treatment concentrations ($P \leq 0.005$) (Fig. 1). The best germination parameters were showed in seeds inoculated with 10 and 20% (v/v) concentrations for 4 to 8 h. Hence, seed application with 10 to 20% (v/v) concentrations was the most effective treatment in the present study compared to others. However, the germination parameters differed significantly with the treatment durations, while, increased treatment duration up to 12 h, found seed germination parameters tended to decrease.

Seed inoculation significantly enhanced seed germination parameter of tartary buckwheat species (Fig. 2). However, the rate of enhancement varied with inoculants concentration. All inoculants concentration increased seed germination parameter greater than control. The highest enhancement of seed germination parameters significantly difference, whereas seed treatment for 4 to 8 h, respectively, showed better seed germination parameters

than others. In contrast, when the treatment duration increased up to 12 h, found the germination parameters decreased comparisons with other durations.

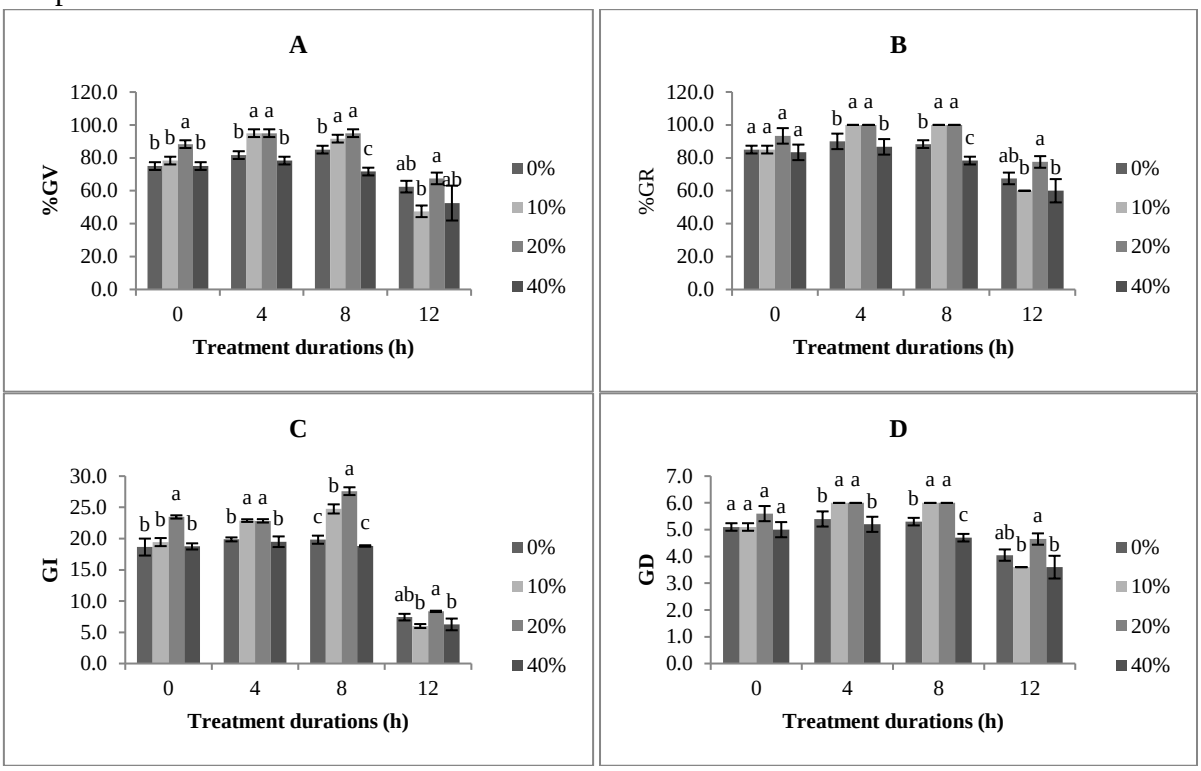

Figure 1. Effect of endophytic bacterium inoculation on seed germination of common buckwheat at 20±1°C, Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

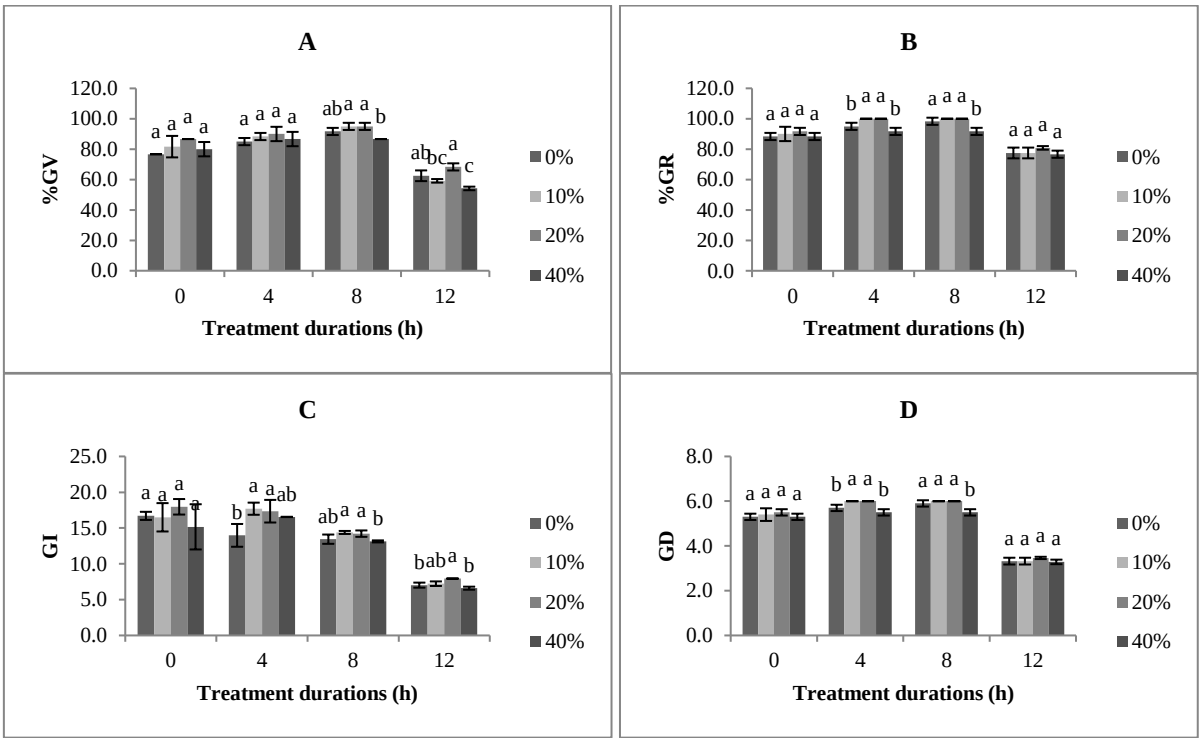

Figure 2. Effect of endophytic bacterium inoculation on seed germination of tartary buckwheat at 20±1°C.

Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

Seed germination test at 25±1°C

Seed inoculation significantly enhanced seed germination parameters: seed germination vigor rate (%GV), total seed germination rate (%GR), seed germination index (GI) and number of seed germination per day (GD) of common buckwheat species. However, the rate of enhancement varied depending on the treatment concentration and treatment durations (Fig. 3). All treatment concentration (10, 20 and 40%, v/v), increased seed germination parameter higher compared to control. The highest germination parameters were observed in seed treated with 10 and 20% (v/v) for 4 and 8 h. Whereas, treatment durations increased up to 12 h, showed seed germination parameter decreases compared to other treatment durations.

Tartary buckwheat seed germination parameters: seed germination vigor rate (%GV), total seed germination rate (%GR), seed germination index (GI) and number of seed germination per day (GD) were significant differences among the treatment concentration and durations (Fig. 4). All seed treated with 10, 20 and 40% (v/v) concentrations, respectively, showed greater germination parameters than control. The highest germination parameters were observed for the seeds treated with 10 and 20% (v/v), for 0, 4 and 8h, respectively. However, the germination parameter was differed significantly with treatment durations. Nevertheless, increased treatment duration might decrease seed germination parameters particularly when treatment duration up to 12 hours.

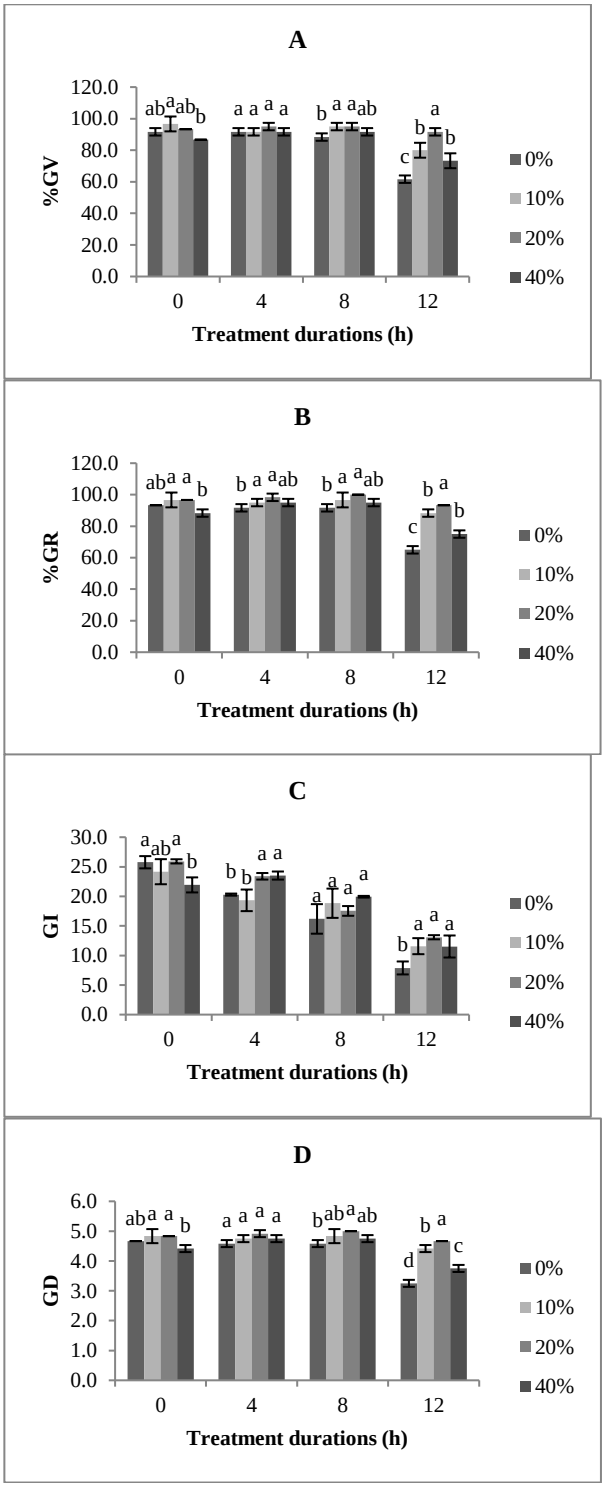

Figure 3. Effect of endophytic bacterium inoculation on seed germination of common buckwheat at 25±1°C.

Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

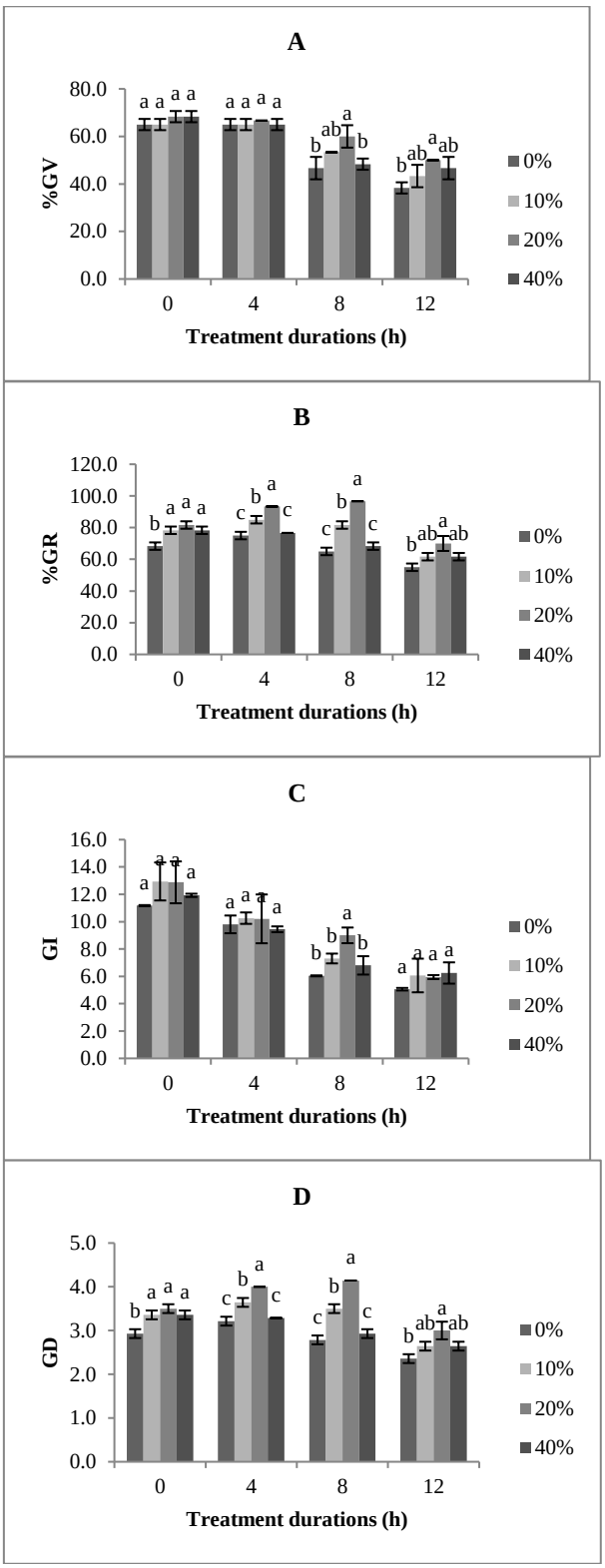

Figure 4. Effect of endophytic bacterium inoculation on seed germination of tartary buckwheat at 25±1°C, Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

Seed germination test at 30±1°C

The seed germination parameters: seed germination vigor rate (%GV), total seed germination rate (%GR), seed germination index (GI) and number of seed germination per day (GD) of common buckwheat was significant differences among the inoculants concentration and treatment durations ($P \leq 0.005$). Inoculation seed with 10, 20 and 40% (v/v) concentrations showed relatively greater germination parameters than control (Fig. 5). The highest germination parameters were observed in seeds inoculated with 10 and 20% (v/v) for 0, 4, 8 and 12 h, followed by 40% (v/v) for 0, 8 and 12 h, respectively compared to control. However, seed treatment for 8 and 12 h, decreased seed germination parameters compared to others, while, the best inoculants concentration and treatment durations were obtained from seeds inoculated with 10 to 20% (v/v) concentrations for 4 hours.

Seed inoculation on seed germination parameters of tartary buckwheat species was significantly affected by all inoculants concentration and treatment durations (Fig. 6). The seed inoculated with 10 and 20% (v/v) concentrations were the most effective treatment in the present study. However, the germination parameters differed significantly with treatment durations, while the suitable duration for the treatment was 4 to 8 h with 10 to 20% (v/v) concentrations. In contrast, increased seed treatment durations might decrease the germination parameters of tartary buckwheat species.

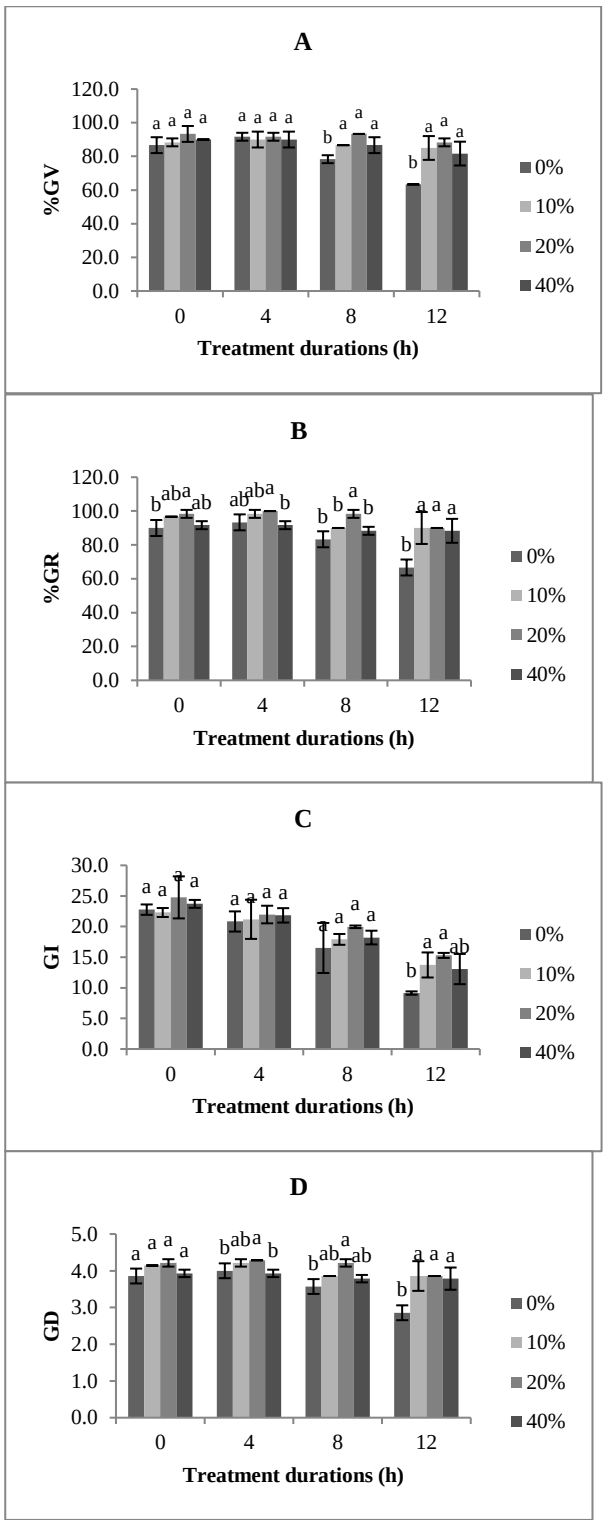

Figure 5. Effect of endophytic bacterium inoculation on seed germination of common buckwheat at 30±1°C,
Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

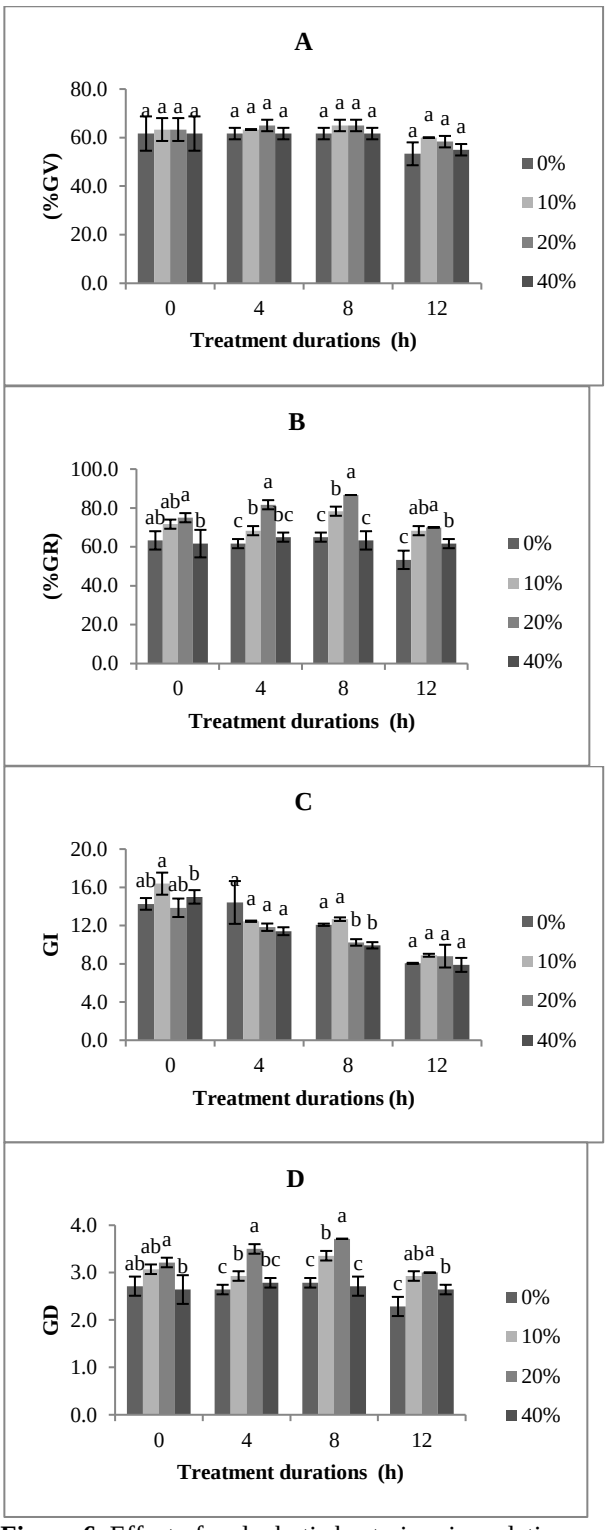

Figure 6. Effect of endophytic bacterium inoculation on seed germination of tartary buckwheat at 30±1°C, Means followed by a common letter are not significantly different at 5% level by LSD. ***A*** = number seed germination vigor rate in the first 4 days (%GV), ***B*** = total seed germination rate in the end of the experiment '7 days' (%GR), ***C*** = seed germination index (GI), and ***D*** = number seed germination per day (GD).

4. Discussions

Bacteria inoculations are able to increase seed germination rate, improve seedling emergence and plant growth, responses to external tress factors and protect plant from disease in different crops were clearly demonstrated (Gholami *et al*., 2009; Mia *et al*., 2012). The present study confirms that seed inoculation with endophytic bacterium particularly *H. rubrisubalbicans* strain significantly enhanced seed germination parameters in both common and tartary buckwheat species with higher vigor rate (%GV), total germination rate (%GR), greater value of germination index (GI) and more number of seed germination per day (GD) compared to un-inoculated seeds (control) and the germination parameters varied depending on the inoculants concentration, treatment durations and tested conditions (Fig. 1 to 6). These may be due to the production of phytohormone as phytohormone influences seed germination. Similarly improved of seed germination parameters by treated with PGPR strains and rhizobacteria has been reported in many cereal species (Gholami *et al.* (2009, Mia *et al.* 2012, Shaukat *et al.*, 2006). Common and tartary buckwheat seed inoculation with endophytic bacterium particularly *H. rubrisubalbicans* strain induced increases in seed germination parameter, in some cases achieving increases seed germination vigor rate up to 95% and total seed germination rate up to 100% greater than control. These might be due to the increased synthesis of hormones like gibberellins by endophytic bacterium (*H. rubrisubalbicans*) strain, which would have triggered the activity of specific enzymes that promote early germination such as α-amylase, which have brought an increase in availability of starch assimilation (Gholami *et al.*, 2009). During the seed germination, α-amylase in the aleurone layer plays an important role in hydrolyzing the endosperm starch into metabolizeable sugars, which provide the energy of emergence rate and growth in cereal crops (Mia *et al*., 2012).

Seed germination is known to be regulated by exogenous hormones. Many of bacteria strains can be produced and excrete in their cultures more than one hormone types, *Rhizobium* isolates synthesize gibberellins (GA) and auxin (Atzorn*et al*., 1988), *Azotobacter* spp. Synthesizes GA, auxin and cytokinins (Salmerson *et al*., 1990) and *Acetobacter* and *Herbaspirillum* spp. isolates synthesized indole-3-acetic acid (IAA) and GA (Bastia *et al*., 1998), which were improved seed germination rate, seedling vigor and growth. The above information might help to support to the present study, hence, seed application with endophytic bacterium (*H. rubrisubalbicans*) strain with suitable inoculants concentration such as 10 to 20% (v/v) may be recommended as a most effective treatment for common and tartary buckwheat species. Furthermore, Briatia *et al.* (2012) reported that common and tartary buckwheat seeds treated with 10% (v/v) deep sea water improved seed germination rate higher compared to control and other treatments. However, the present study found that increased seed inoculation concentrations up to 40% (v/v), the germination parameters tended to decrease rapidly compared to others. Similarly, Dhoran and Gudadhe (2012) found in *Asparagus sprengeri* Regel in seed germination rate and vigor indexes decreased rapidly when treated with growth regulator (GA) concentration above 60 ppm.

To improve seed germination parameters of common and tartary buckwheat species, the treatment durations may also needed to be considered. However, the present study showed highly effectiveness of the treatment durations was observed at 4 to 8 h, which better than the other treatment durations. Mia *et al.* (2012) reported rice seeds inoculated with rhizobium strains UMPR1006 and UMPR 1102, for 48 to 96 h improved percentage of seed emergence higher compared to others. In contrast, our present study found that increasing seed treatment duration up to 12 h decreased germination parameters rapidly compared to other durations. On the other hand, Ertekin (2011) reported that *Koelreuteriapaniculata* seeds soaked for 24 h and moist chilled was showed very low of seed germination rate, but increased if the

seeds were redried after soaking. The difference in germination of seeds may be related to the characteristics of seeds, varieties, species and environmental conditions (Ertekin, 2011). Enhancing seed germination and developing vigorous seedling are crucial for this study. The best germination temperature conditions for common and tartary buckwheat seed were ranged from 20 - 30ºC, when seeds inoculated with 10 and 20% (v/v) concentrations of endophytitc bacterium (*H. rubrisubalbicans*), which better than others. Born and Corns (1958) found that tartary buckwheat seeds after-ripened, seeds germinated over a wide range of temperatures under laboratory conditions. To ensure high germination, temperature, light, pre-soaking treatment and scarification of seed coat have been applied in various plant (Basbag *et al*., 2009). Optimum seed germination and seedling emergence occurred at relatively high temperature between 20 - 30ºC for several cereal and vegetable species including common and tartary buckwheat (Briatia *et al.*, 2012, Mai *et al*., 2012, Gholami *et al*., 2009 and Tzortzakis, 2009).

Seed germination varied significantly among the varieties, inoculants concentration and tested conditions ($P \leq 0.005$). Seeds inoculated with endophytic bacterium showed higher affected on seed germination parameters in common buckwheat than tartary buckwheat species under the different temperature conditions. It's possible because this endophytic bacterium (*H. rubrisubalbicans*) strain has been isolated from common wheat species, so it could be had more effect to common buckwheat than tartary buckwheat species.

5. Conclusions

The results of this study suggested that common and tartary buckwheat seeds inoculated with endophytic bacterium (*H. rubrisubalbicans*) strain could enhance seed germination parameters (%GV, %GV, GI and GD) increased significantly differ to untreated seeds (control). The highest effect were observed in the 10 and 20% (v/v) concentrations with treatment duration for 4 - 8 h. However, increased inoculation concentrations and treatment durations up to 40% (v/v) and 12 hours decreased seed germination parameters of both common and tartary buckwheat species. Finally, conclusion that seed application with *H. rubrisubalbicans* strain in proper concentrations (10 - 20% (v/v) and durations for 4 - 8 h may be recommended as a most effective treatment to promote seed germinations, seedling vigor and seedling establishment in both common and tartary buckwheat species.

6. References

Akaya, M. and Sun, J., 1992. Tendency in buckwheat research around the world, *World Agriculture* 92(1): 25–26.

Atlas, R.M., 1993. Handbook of Microbiological Media.*CRC Press Inc, Florida*.

Atzorn, R., Crozier, A., Wheeler, C.T.andSandberg, G., 1988. Production of gibberellins and indole-3-acetic acid by *Rhizobium phaseoli* in relation to nodulation of *Phaseolus vulgaris* roots. *Planta.* 175(4): 532-8.

Basbag, M., Toncer, O. and Basbag, S., 2009. Effects of different temperatures and duration on germination of caper (Capparisovata) seeds.*Journal of Environmental Biology* 30(4): 621-624.

Bastia, F., Cohen, A., Piccoli, P., Luna, V., Baraldi, R. and Bottini, R., 1998. Production of indole-3-acetic acid and gibbereliins A1 and A3 by *Acetobacter diazotrophicus and Herbaspirilum seropedicae* in chemically-defined culture media.*Plant Growth Regul*. 25: 7-11.

Briatia, X., Hong, S.K., Sung, I.J., Chang, K.J., Park, B.J. and Park, C.H., 2012. Effect of deep sea water on seed germination, photoperiod and temperature on the growth and flowering of buckwheat species. *Korean J. Plant Res.* 25(3): 323-328.

Born, W.H.V. and Corns, W.G., 1958. Studies on seed dormancy, plant development, and chemical control

of tartary buckwheat (*F. tataricum* (L.) Gaertn.) I. Seed dormancy. *Canadian Journal of Plant Science*, 38: 357-365.

Dhoran, V.S. and Gudadhe, S.P., 2012. Effect of plant growth regulators on seed germination and seedling vigour in *Asparagus sprengeri* regelin.*I. Res. J. Biological Sci.*, 1(7): 6-10.

Ertekin, M., 2011. Effect of microorganisms, hormone treatment and stratification on seed germination of goldenrain tree (*Koelreuteriapaniculata*). *Int. J. Agric. Biol.*, 13: 38-42.

Gholami, A., Shahsavani, S. and Nezarat, S., 2009. The effect of plant growth promoting rhizobacteria (PGPR) on germination, seedling growth and yield of maize.*World Academy of Science, Engineering and Technology* 25: 19-24.

International Seed Testing Association (ISTA)., 2006. International rules for seed testing. *Seed Science and Technology*. Basserdorf, Switzerland.

Koger, C.H., Reddy, K.N. and Poston, D.H., 2004. Factors affecting seed germination, seedling emergence and survival of texas weed (*Caperoniapalustris*). *Weed Science* 52: 989-995.

Lin, S.Q and Zhang, Q.H., 2001. Advances in the Development of Functional Foods from Buckwheat. *Critical Reviews in Food Science and Nutrition* 41(6): 451–464.

Mathur, A., 1989. Mutation studies in buckwheat (*Fagopyrum*.) I. Effect of gamma rays on germination. *Fagopyrum* 9: 10-14.

Mia, M.A.B., Shamsuddin, Z.H. and Mahmood, M., 2012. Effect of rhizobia and plant growth promoting bacteria inoculation on germination and seedling vigor of lowland rice. *African Journal of Biotechnology* 11 (16): 3758-3765.

Oliveira, A.L.M., Urquiaga, S., Dobereiner, J. and Baldani, J.I., 2002. The effect of inoculating endophytic N_2-fixing bacteria on micropropagated sugar cane plants. *Plant and Soil* 242: 205-215.

Peng, C.C., Chen, K.C., Yang, Y.L., Lin, L.Y. and Peng, R.Y., 2009. Aqua-culture improved buckwheat sprouts with more abundant precious nutrients and hypolipidemic activity. *International of Food Science and Nutrition* 60(S1): 232-245.

Perry, A.D., 1984. Commentary on ISTA vigour test committee collaborative trials. *Seed Sci. and technol.* 12: 301-308.

Popović, V., Sikora, V., Berenji, J., Filipović, V., Dolijanović, Z., Ikanović, J. and DaliborDončić, D., 2014. Analysis of buckwheat production in the world and Serbia. *Economics of Agriculture, EP., (61)1: 53-62*.

Salmerson, V., Martinez-Toledo, M.V. and Gonzalez-Lopez, J., 1990. Nitrogen fixation and production of auxins, gibberellins, and cytokinins by an *Azotobacter chroococcum* strain isolated from the root of *zea mays* in the present of insoluble phosphate. *Chemosphere* 20: 417-422.

Shaukat, S., Affrasayab, S. and Hasnain, S., 2006. Growth responses of helianthus annus to plant growth promoting rhizobacteria used as a biofertilizer. *J. Agri. Res*., 1(6): 573-581.

Tzortzakis, N.G., 2009. Effect of pre-sowing treatment on seed germination and seedling vigour in endive and chicory. *Hort. Sci. (Prague)*, 36(3): 117-125.

Wang, Y.J. and Campbell, C.G., 2000. Breaking dormancy in buckwheat. *Fagopyrum* 17: 45-50.

Zare, A.R., Solouki, M., Omidi, M., Irvani, N., OladzadAbasabadi, A. and Mahdi Nezad, N., 2011. Effect of various treatments on seed germination and dormancy breaking in *Ferula assafoetida*L. (Asafetida), a threatened medicinal herb. *Trakia Journal of Sciences* 9(2): 57-61.