

Boron Determination and Pollen Morphology Studies on Endangered Endemic *Scabiosa hololeuca* Bornm. (Caprifoliaceae)

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Abstract

Deficiency and toxicity levels of boron is too close in plants which need to be trace amounts of boron for vital activities. B requirements show great differences between the plants. Leaves and generative organs deposit the highest amount of B. In contrast to this, root, fruit and seed deposit minimal amounts of B. Also pollen production level is directly related with the B content of soil. Within this scope, boron determination and pollen morphology studies were performed on endemic *Scabiosa hololeuca* in this study. B content of root, stem and flower were measured by curcumin method for *S. hololeuca*. Pollen grain microphotographs of examined taxon have been taken from preparates which were made by Wodehouse ve Erdtman techniques. Examinations and measurements have been performed on these and morphometric results given. According to the curcumin technique, B element as been measured 7,17 mg kg⁻¹ dry weight (ppm.) in root, 8,48 mg kg⁻¹ dry weight (ppm.) in stem and 8,13 mg kg⁻¹ dry weight (ppm.) in flower of studied taxon. Type of the pollen grain of studied taxon has determined as tricolpatae according to the Wodehouse and Erdtman methods. We believe that comparisons and evaluations of obtained data from like these studies will make important contributions to plant taxonomy in future.

Keywords: *Scabiosa hololeuca* Bornm. (Caprifoliaceae), Boron Amount, Pollen Morphology.

INTRODUCTION

Deficiency and toxicity levels of boron is too close in plants which need to be trace amounts of boron for vital activities [4]. B requirements show great differences between the plants. Plants can be classified in three groups as low, medium and high to their B requirements. B requirements of dicotyledons are 3-4 times higher than monocotyledons [2; 12]. B can be absorbed by actively or passively in plants. But generally, nonionised B(OH)₃ form B is passively taken by plants. Leastwise, B(OH)₄⁻ ions can be observed by plants too [14; 19]. In contrast to other microelements, only transportation of B shows differences species from species. B is mostly deposited in leaves and generative organs of plants. After this, it is deposited in respectively roots, fruits and seeds. In this scope, our study aims to determinate the B contents and palynological features of endangered endemic *Scabiosa hololeuca* Bornm (Caprifoliaceae).

MATERIALS and METHODS

Boron analysis

Boron Analysis in Plants Boron analysis has been performed on stem, leaf and root of studied taxa by curcumin technique. 0,5 g of dried and crushed plant materials was put into ceramic cinerarium and they were burned until white ash was obtained. When was the ceramic cinerarium getting cold, 5 ml 1.0 N HCl added into it. Cinerarium was kept on water bath for a while and then contents of ceramic burning capsules were quantitatively transferred into volumetric flask (50 ml) by distilled water. Contents of volumetric flask were fulfilled to 50 ml by distilled water and they were shook. We waited for a while till silicium pile up at the bottom of the volumetric flask. After this, 1 ml of clear plant solutions was transferred into ceramic capsules. 4 ml curcumin-oxalic acid solution added on to each clear plant solution

and they were waited on the 55 ± 3 °C water bath till solution evaporated. When ceramic capsules got cold to room temperature, 10 ml ethyl alcohol was added in to capsules and they were mixed by glass stick for dissolving the residue. Then each of these solutions were transferred into volumetric flasks (25 ml) and they were completed their degree by ethyl alcohol. Each solutions absorbance value were measured at 540 nm by Jasco V-530 UV/VIS branded spectrophotometer. Boron content calculations from absorbance values were performed according to Kacar (1972).

Pollen Morphology

Pollen samples were taken from dried herbarium specimens of Eskişehir Osmangazi University, Faculty of Science and Letters, Department of Biology Herbarium (OUFE). In palynological studies, flowers of the 10-15 different plant specimens which were collected from different locations were used for acquiring the pollen grains. In plant collection stage, specimens of each taxa were put into different paper envelopes and so melding the different pollen grains were prevented. This study was carried out to investigate whether different factors in different regions have an effect on pollens in the same structure. Pollen morphology of investigated taxa was examined by light microscope. Faegri and Iversen's terminology was used for naming the exine layers. In light microscope investigations, preparation of pollen grains was performed with regard to Wodehouse (1935) and Erdtman (1969) methods [24; 25]. Exine and intine thickness of pollen grains belonging to studied taxa were measured min 20 max 50 times. Arithmetic means of these measurements were taken. Identification (100x) and counting (10, 40 and 100x) of pollens were made by Prior brand binocular microscope. Each space of ocular micrometer is 0.98 µm. Microphotographs of pollens were taken by Spot In-SIGHT Color Digital camera that attached to Olympus brand light microscope. In identification of pollens, various basic palynological books and literatures were used [26; 16; 10;



Figure 1. *Scabiosa hololeuca* in nature.

RESULTS

According to the curcumin technique, B element has been measured 7,17 mg kg⁻¹ dry weight (ppm.) in root, 8,48 mg kg⁻¹ dry weight (ppm.) in stem and 8,13 mg kg⁻¹ dry weight (ppm.) in flower of studied taxa. Type of the pollen grain of studied taxon has determined as tricolpate according to the Wodehouse and Erdtman methodes (Figure 2 and 3).

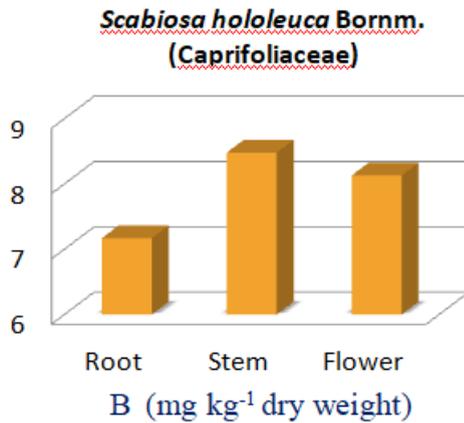


Figure 2. Boron contents in *Scabiosa hololeuca*

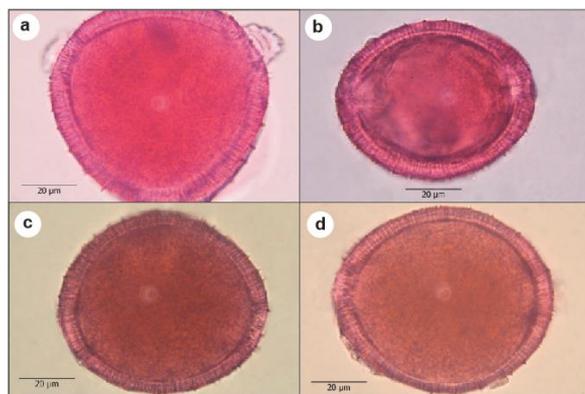


Figure 3. Pollen grains of *Scabiosa hololeuca* (a: Polar view (W), b: Evcatorial view (W), c: Polar view (E), d: Evcatorial view (E))

DISCUSSIONS

In some plants, difficulties in Boron transportation are

be taken B externally for healthy development of generative organs [4]. B directly affects the anthers in reproductive development. Pollen amount in anthers are connected by B. Also B is necessary for development of pollen tube and pollen germination [7]. In spite of the B concentration is high stylus, stigma and ovary; it is less in pollen grains than others [3]. B deficiency affects the fertility of anther in barley, wheat and rice. Also it affects the fertility of pistil in avocado and corn. Because of the inadequate development of pollen tube in B famine plants, some abnormalities observed from researchers [21]. For example, development of egg cell stopped and it didn't become an embryo in peanut; fruits of avocado didn't matured; fruits misshaped and they fallen off after maturation in mango [7].

Pollen grains of endangered endemic *Scabiosa hololeuca* are identified as tricolpate and their exine structure is tectate-echinate. According to the literatures, aperture features and exine structure are essential criteria for determination the phylogenetic relationships between taxa [22; 16; 6; 24; 25; 23]. Except variational ones, All morphological differences are directly related with genetic differences as be in the pollen grains of related taxa [6].

In addition to the systematic features of taxa, morphological features of pollen grains should be important distinctive characters. We believe that, this study will bring light into phylogenetic relationship between investigated taxa.

REFERENCES

- [1] Aytuğ B., Aykut, S., Merev, N., Edis, G., 1971, İstanbul Çevresi Bitkilerinin Polen Atlası. İ. Ü. Yayın No:1650, O. F. Yayın no:174.
- [2] Bellaloui, N. and Brown, P.H., 1998, Cultivar differences in boron uptake and distribution in celery (*Apium graveolens*), tomato (*Lycopersicon esculentum*) and wheat (*Triticum aestivum*). Plant and Soil, 198, 153-158 p.
- [3] Blevins D.G., Lukaszewski K.,M., 1998, Boron in Sturcture and Function. Annual Rev. Plant Physilogy Plant Molecular Biology, 49, 481-500 p.
- [4] Brown P. H., Bellaloui N., Wimmer M. A., Bassil E. S., Ruiz J., Hu H., Pfeffer H., Dannel F., Römheld V., 2002, Boron in Plant Biology. Plant Biology 4, 205-223 p.
- [5] Charpin, J., Surinyach, R., Frankland AW., 1974. Atlas of European allergenic pollens. Sandoz Editions, Paris, pp. 20-23.
- [6] Cronquist, A., 1968. The evolution and classification of the flowering plants. Thomas Nelson Ltd. Edinburgh, London.
- [7] Dell, B., Huang, L., 1997. Physiological response of plants to low boron. Plant and Soil, 193, 103-120 p.
- [8] Erdtman, G., 1966. Pollen Morphology and Plant Taxonomy, Angiospermae. Hafner, New York.
- [9] Erdtman, G., 1969. Handbook of Palynology Morphology, Taxonomy, Ecology. An Introduction to the Study of Pollen Grains and Spores. Hafner Pub. New York.
- [10] Erdtman, G., Roger, P., Wodehouse, R.P., 1954. An Introduction to Pollen Analysis. Waltham, Mass., USA, Published by the Chronica Botanica Company. Stockholm, Almquist and Wiksell, 239 s.
- [11] Faegri, K., Iversen, J., 1975. Textbook of pollen analysis. 3rd edition. Munksgaard, Copenhagen.
- [12] Hakkı, E.E., Atalay, E., Babaoğlu M.B., Soylu, S., Duralı, D., Gezgin, S., 2005. Bitkilerde düşük ve yüksek bora toleransta tür içi ve türler arası farklılık. IVX. Ulusal Biyoteknoloji Kongresi, 31 Ağustos-02 Eylül 2005, Eskişehir, Bildiri Kitabı, 74-77 p.
- [13] Hu, H., Brown, P.H., 1997. Absorption of Boron by plant roots. Plant and Soil, 193, 49-58 p.
- [14] Kacar, B., 1972. Bitki ve Toprağın Kimyasal Analizleri: II, Bitki Analizleri. Ankara Üniversitesi Ziraat Fakültesi

Yayımları, No:453, Ankara.

- [15] Kapp, R.O. 1968. How to know Pollen and Spores. WM. C. Brown Company.
- [16] Kuprianova, A., 1967. Apertures of pollen grains and their evolution in Angiosperms. *Paleobot. Playnology*, 3: 73-80.
- [17] Kuprianova, L.A., 1965. The Palynology of The Amentiferae. The Academy of Sciences of the USSR, Moscow, 214 s.
- [18] Moore, P.D., Webb, J.A., Collinson, M.E., 1991. Pollen analysis. Oxford Blackwell Scientific Publications, London, pages: 110-112.
- [19] Nable, R.O., Banuelos, G.S., Paull, J.G., 1997. Boron toxicity. *Plant and Soil*, 193, 181-198.
- [20] Pehlivan, S., 1995. Pollen morphology of some Turkish endemic *Centaurea*. *Grana* 34: 29-38.
- [21] Shelp, B.J., Marentes, E., Kitheka, A.M., Vivekanandan, P., 1995. Boron mobility in plants. *Physiologia Plantarum*, 94, 356-361.
- [22] Skvarla, J.J., 1966. Techniques of pollen and spore electron microscopy. I. staining, dehydration and embedding. *Oklah Geol Notes* 26: 179-186.
- [23] Takhtajan, A.L., 1980. Outline of the classification of flowering plants (Magnoliophyta). *Bot Rev* 46.
- [24] Walker, J.W., 1974a. Evolution of exine structure in the pollen of primitive Angiosperms. *Am J Bot* 61: 891-902.
- [25] Walker, J.W., 1974b. Aperture evolution in the pollen of primitive Angiosperms. *Am J Bot* 61: 1112-1137.
- [26] Wodehouse, R.P., 1959. Pollen grains. Their structure, identification and significance in science and medicine. Hafner, New York.