AENSI Journals

Advances in Environmental Biology

ISSN-1995-0756 EISSN-1998-1066

Journal home page: http://www.aensiweb.com/AEB/

Generative meristem, anther development and microsporogenesis in *Lepidium sativum* L.

Ahmad Majd, Elham Bagheri Abyaneh, Sayeh Jafari, Golnaz Tajaddod, Fahimeh Salimpour

Department of Biology, Faculty of Biological Sciences, North-Tehran Branch, Islamic Azad University, Tehran, Iran.

ARTICLE INFO

Article history:
Received 2 April 2014
Received in revised form
13 May 2014
Accepted 28 June 2014
Available online 23 July 2014

Keywords:
Lepidium sativum L., development, generative meristem, microsporogenesis, pollen grain

ABSTRACT

Lepidium sativum L. (Garden cress) is a cool season annual herb, belonging to Brassicaceae family. In this research, generative meristem, anther development and microsporogenesis in *Lepidium sativum* L. were considered. The flowers, in different developmental stages, were collected, fixed in Formalin -glacial acetic acid- alcohol (FAA), stored in 70% ethanol, embedded in paraffin, sliced at 8-10 µm by rotary microtome and Stained by periodic Acid Schiff (PAS) and contrasted with hematoxylin. The results demonstrated that generative meristem was very active and protruded. Anther walls development followed the dicotyledonous type and were tetrasporangiate and composed of epidermal layer, endothecium layer, middle layers and tapetum layer. Microspore tetrads were tetrahedral and pollen grains had generative and vegetative nucleus.

To Cite This Article: Ahmad Majd, Elham Bagheri Abyaneh, Sayeh Jafari, Golnaz Tajaddod, Fahimeh Salimpour., Generative meristem, anther development and microsporogenesis in *Lepidium sativum* L. ***Adv. Environ. Biol., 8(12),*** *247-250, 2014*

INTRODUCTION

The genus Lepidum belongs to tribe Lepidieae and section Monoploca [20] of Brassicaceae family [11,1,2] and consists of approximately 175 species [15] is the largest genus in the Brassicaceae [11]. It is distributed world wide, primarily in temperate and subtropical regions; the genus is poorly represented in arctic climates and in tropical areas; and it grows in the mountains [1,2].

Lepidium sativum L. (Garden cress) is a cool season annual plant, It is a fast-growing, edible plant botanically associated with watercress and mustard and sharing their peppery, tangy flavor and aroma. Seeds, leaves and roots are economically valuable. This important green vegetable consumed by human beings, most typically as a garnish or as a leaf vegetable [22,3]. Persian used to eat the leaves of this plant even before bread was known [19]. This annual herb can reach a height of 60 cm with many branches on the upper part. The white to pinkish flowers are only 2 mm (1/12 of an inch) across, clustered in branched racemes. It has 6 stamens with oblong anther. The leaves are variously lobed and entire, Fruits are oblong-ovate or elliptic, about 5 mm long, with two seeds per pods, seeds are reddish brown, oblong and wingless, [23]. This valuable plant contains significant amounts of calcium, iron, folic acid, vitamin C and A. Additionally, it contains high amount of protein. The major fatty acid in Garden cress is linolinic acid [9]. Garden cress (*Lepidium sativum* L.) is one the valuable food stuff that abounds not only in nutrients but also in health enhancing [23]. People in South Asia, used this plant in traditional medicine [4,8]. In Europe and America, the leaves are used in salad. In many countries of Africa, *Lepidium sativum* L. seeds are thought to be a useful medicinal remedy to cure respiratory disorders [13,17]. Notably, there is not enough information about the generative meristem, basic anther structure and male gametophyte developmental stages of *Lepidium sativum* L.. Development of male gametophyte involves a series of occurrences to produce and release mature pollen grains from anther [14].

The aim of this research was to investigate a detailed study on generative meristem and microsporogenesis of *Lepidium sativum* L., not only for improving the knowledge of microsporogenesis developmental events but also for systematic evaluation.

Corresponding Author: Elham Bagheri Abyaneh, Department of Biology, Faculty of Biological Sciences, North-Tehran Branch, Islamic Azad University, Tehran, Iran.
E-mail: e.bagherii@yahoo.com

MATERIAL AND METHODS

Inflorescences at all different developmental stages (tiny buds to mature flowers) were collected and fixed in FAA solution (20% formalin, 10% acetic acid, 70% ethanol, v/v), dehydrated during alcohol series, embedded in paraffin and sectioned with a thickness of 8-10µm using a rotary microtome. Staining was carried out with PAS (Periodic Acid Schiff) technique according to protocol suggested by Yeung [24]and contrasted with Meyer's Hematoxylin. Several sections for each generative meristems and anther developmental stages were investigated with a Zeiss Axiostar plus light microscope. Many samples were studied before each stage and photomicrographs were made from the most effective ones.

Results:

Results obtained from this study demonstrated that generative meristem cells were very active and generative meristem was massive and protruded. The superficial layers of generative meristem, were sporangiare meristem (Sp.m) and the downward regions, which were less stained were receptacle meristem (Re.m) (Fig1 b).Our findings demonstrated that sepal and petal primordia were formed sooner compare to formation of the stamen and ovule primordial (Fig1 c) and sepal and petal primordia passed the subsequent developmental stages faster than the stamen and ovule primordia did (Fig1 G). Therefore, from outside to inside the sepals (Se), Petals, stamen primordia (st.p) and ovary primordia (ov.p) were detectable and ultimately stamen and ovary primordia evolved completely. However, development of stamen primordia (st.p) was faster than ovary primordia (ov.p) in early stages. Developed stamens were consisted of filament and anther. Each young anther was consisted of 4 pollen sacs (tetrasporangiate) with connective tissue in the center (Fig k). Each pollen sac was concluded of peripheral cells forming an undifferentiated walls and a mass of uniformed cells (archeosporial cells) (Fig1 I). Then the wall differentiated to epidermis, endothecium, middle layer and tapetum layer (Fig1 L). Taptum layer in *Lepidium sativum* L. was secretory type in this development stage. At this stage, microspore mother cells (Microsporocytes), were also detectable (Fig1 J). Microsporocytes are recognizable by their large volume, dense cytoplasm, and conspicuous nuclei. Each microspore mother cell undergoes meiosis; during which M.M.C undergo successive type. In this type of cytokinesis, each nuclear division was followed by cell wall formation. The first nuclear division of meiosis (meiosis I) is accompanied with the formation of two haploid cells in a dyad form (Fig1 M& N). Then the second meiotic division occur in dyadic cells (meiosis II), which was followed by wall formation and ultimately tetragonal tetrads which were enclosed with special callosic walls, appeared (Fig 0&P). At the stage of tetrad, the anther wall was composed of epidermis, endothecium and tapetum. This means that the middle layer was degenerated. At next stage, haploid cells (meiospores) were segmented after the decomposition of the special wall, and each of the meiospores is turned into young meiospores with big and central vacuoles and small and peripheral nucleus. Exine (Ex) evolved in this stage. At the stage of anther dehiscence, there was one-layer anther wall of endothecium and the tapetal layer was digested completely (Fig1 Q) to nourish the pollen grains. Additionally we found the mitotic division of microspores unequal. Therefore, a darker generative nucleus and a lighter vegetative nucleus were detectable (Fig1 R). Fig.1 shows detailed occurrences.

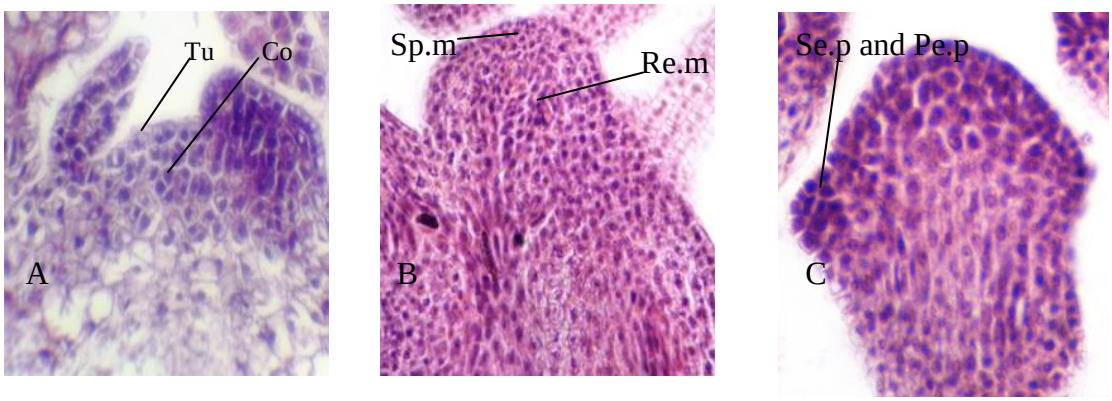

Fig. 1: A: Longitudinal section of *Lepidium sativum* L., vegetative meristem and tunica (Tu) and corpus (Co) layers B: Protruded generative meristem contains of receptacle meristem (Re.m) and sporangiare meristem (Sp.m) C: Formation of sepal and petal primordia

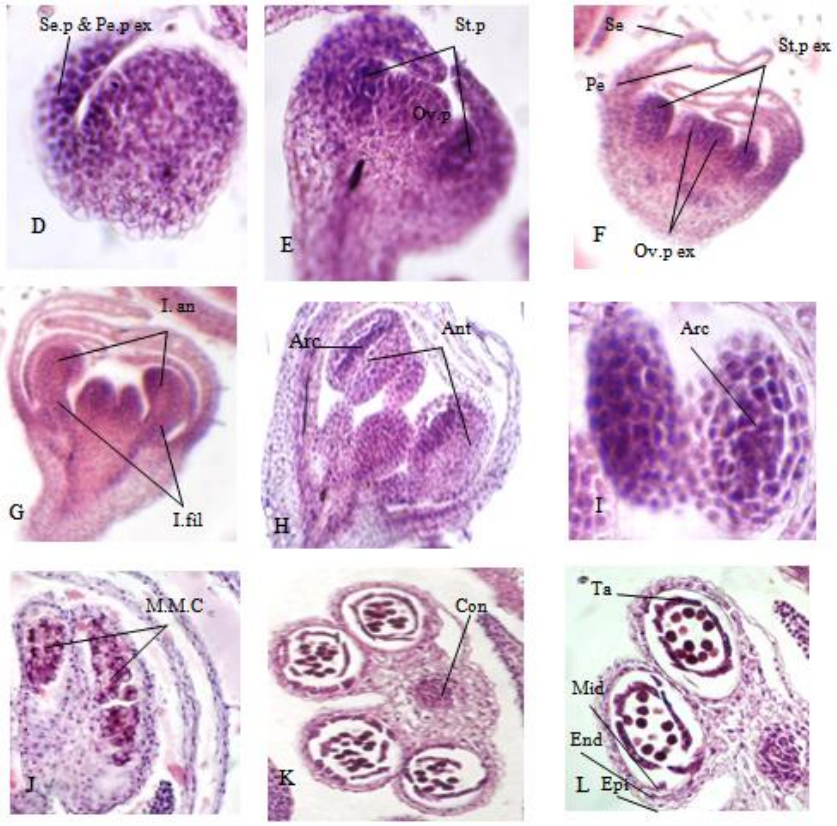

Fig. 1 D: Extension of sepal and petal primordial(Se.p & Pe.p ex) E: Formation of stamen primordia (St.p) and ovary primordia (Ov.p) F: sepal(Se) and peta (Pe) formation, stamen (St.p ex)and ovary primordial extension (Ov.p ex) G: Stamen primordia develop to initial filament(I. fil) and initial anther (I. an) H: Anther and archeospore formation. I: Archeospore cells J: Microspore mother cells (M.M.C) K: Tetrasporangiate anther with connective tissue (Con) L: Each anther contains four layers: epidermis (Epi), endothecium (End), middle layer (Mid) and tapetum (Ta).

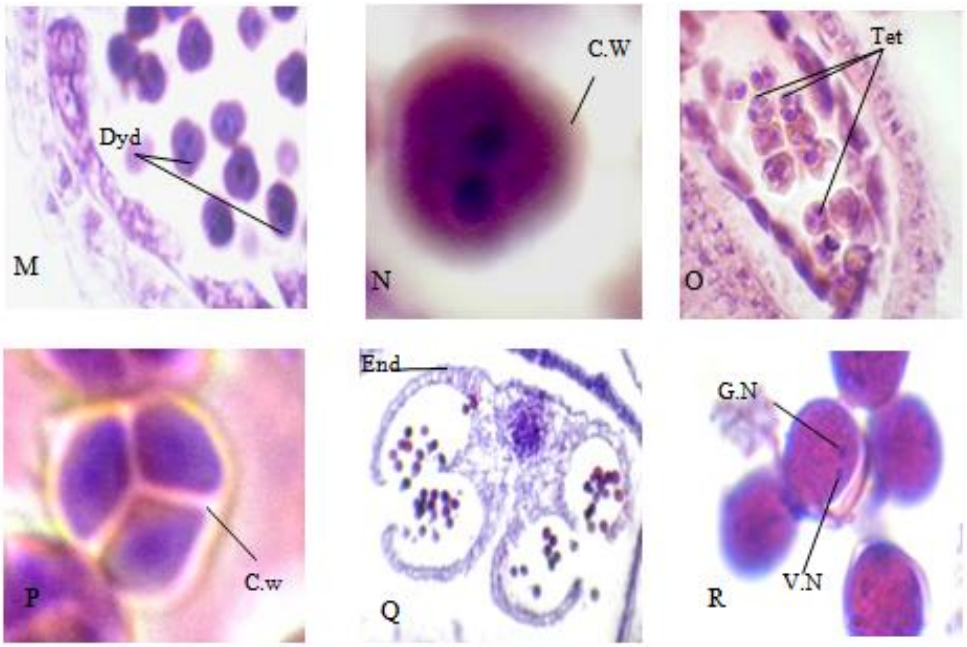

Fig. 1. M: Pollen grains in dyad stage N: A dyad cell with callosic wall (C.W) O: Tetrahedral tetrads (Tet) in the anther loculus P: Formation of callosic wall in a tetrad cell Q: Cross section of adult anther with endothecium layer(End) R: pollen grain with generative (G.N) and vegetative nucleus(V.N)

Discussion:

Results obtained from this study revealed that generative meristem was very active and their cells were divided continuously, and meristem became massive and protruded which was consistent with the findings of jafari and Niknam [12], in *Ziziphus jujuba* L. Developmental stages of *lepidium sativum* L. anther wall followed the dicotyledonous type, which was composed of an epidermal layer, an endothelial layer, one middle layer and tapetum which is consistent with Davis [7]. The tapetum was secretory that was similar with the findings of Chehregani et al., 2009 for *Lepidium vesicarium* L.. There was a significant correlation between microspore mother cell divisions and anther`s tapetum development that is coincides with other reports for dicotyledonous plants [7,10,6]. The wall-bearing tapetum phase was ended at the tetrad developmental stage and Tapetum cells started breaking down prior to anther dehiscence, which was according to findings of Murgia et al., [16]. The anther tapetum is the main supplier of nutrients and cell wall precursors for developing pollen grains [18]. Prior to anthesis, the tapetum cells were consumed completely and ultimately bi-nucleated pollen grain released which is in accordance with findings of Chehregani et al., [6].

REFERENCES

[1] Al-Shehbaz, I.A., 1986a. New Wool-alien Cruciferae (Brassicaceae) in Eastern North America. *Lepidium* and *Sisymbrium*. *Rhodora*, 88: 347-356.
[2] Al-Shehbaz, I.A., 1986b. The Genera of *Lepidieae* (Cruciferae; Brassicaceae) in the South Eastern United States. *J. Arnold Arboretum*, 67: 265-311.
[3] Bagheri Abyaneh, E., A. Majd, S. Jafari, G. Tajaddod and F. Salimpour, 2014. Influence of the Electromagnetic Fields on Some Biological Characteristics of *Lepidium sativum* L. Adv. Environ. Biol., 8(4): 980-984.
[4] Baquar, S.R., 1989. Medicinal and Poisonous Plants of Pakistan, Printas, Karachi, Pakistan.
[5] Chregani, A. and M., Seddaghat, 2009. Pollen Grain and Ovule Development in *Lepidium vesicarium* **(**Brassicaceae). International Journal of Agriculture & Biology, 11(5): 601-605.
[6] Chehregani, A., F. Mohsenzadeh and M. Ghanad, 2011. Male and Female Gametophyte Development in *Cichorium intybus*, Int. J.Agric& bio, 4: 603-606.
[7] Davis, O.L., 1966. Systematic Embryology of the Angiosperms. John Wiley Sons, New York, USA
[8] Duke, J.A., M.J. Bogenschutz-Godwin, J. Ducelliar and P.A.K. Duke, 2002. Handbook of Medicinal Herbs, CRC Press, Boca Raton, Fla, USA.
[9] Gocavi, S.S., N.G. Malleshi and M. Guo, 2004. Chemical Composition of Lepidium sativum L. Seeds and Its Fractions and Use of Bran as a Functional Ingredient, Plant Foods HumanNutr, 59: 105-111.
[10] Gustafsson, L., 1946. Apomixis in Higher Plants. Part I. the Mechanism of Apomixis. Actauniversity Lund, 42: 1-67.
[11] Hewson, H.J., 1982. The Genus Lepidium L. (Brassicaceae) in Australia. Brunonia, 4: 217-308.
[12] Jafari Marandi, S. and F. Niknam, 2012. Pollen and Anther Development in *Ziziphus jujuba* L. (Rhamnaceae). Advances in Environmental Biology, 6(8): 2339-2343.
[13] Kloos, H., 1976. Preliminary Studies of Medicinal Plants and Plant Products in Ethiopian Markets. J. Ethiop.Pharm. Assoc. 2: 18–28.
[14] McCormick, S., 2004. Control of Male Gametophyte Development. Plant cell, 16(S1): 42-53.
[15] Mummenhoff, K., H. Hurka and H.-J. Bandelt, 1992. Systematics of Autralian Lepidium Species (Brassicacea) and Implications for Their Origin: Evidence from IEF Analysis of Rubisco. Plant Syst. Evol, 183: 99-112.
[16] Murgia, M., M. Charzynska, M. Rougier and M. Cresti, 1991. Secretory Tapetum of *Brassica oleracea* L.: Polarity and Ultrastructural Features. Sexual Plant Reproduction, 4(1): 28-35.
[17] Nadkarni, KM., 1976. Indian Materia Medica, Popular Prakashan, Bombay, India. 1001.
[18] Piffanelli, P., JHE. Ross and D.J. Murphy, 1998. Biogenesis and Function of the Lipidic Structures of Pollen Grains. Sex Plant Reprod, 11: 65-80.
[19] Sharma, Sh. and N. Agrawal, 2011. Nourishing and Healing Prowess of Garden cress (Lepidium sativum Linn.). Indian Journal of Natural Products and Resources, 293: 292-297.
[20] Thellung, A., 1906. Die Gattung Lepidium (L.) R. Br. Eine Monographische Studie. Mitteilungen aus dem Botanischen Museum der Universität Zurich. XXVIII.
[21] The Wealth of India, Raw Materials, 1962. Publication and Information Directorate, CSIR, New Delhi, 6: 71-73.
[22] Tiwari, P.N. and G.S. Kulmi, 2004. Performance of Chandrasur (*Lepidium sativum*) Under Different Levels of Nitrogen and Phosphorus. J Med Arom Plant Sci., 26: 479-481.
[23] Watt, J.M. and M.G. Breyer Brandwjk, 1962. Medicinal and Poisonous Plants of Southern and Eastern Africa. 2nd Edn., Livingstone Ltd.,Edinburgh.
[24] Yeung, E.C., 1983. Histological and histochemical staining procedures. In: Cell culture and somatic cell genetics of plants, Orlando. Vasil, I. K. (Ed). Florida: Academics Press, pp: 689-697.