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# POLLEN GRAIN TRAITS OF OIL SPECIES FROM THE NOVI SAD COLLECTION

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The collection of oil species in Novi Sad contains 12 species represented with 1-4 cultivars or landraces. In the continuous work on this collection in the sense of breeding of some of those species and their usage as a source of «desirable genes» we analyzed pollen grain morphology (shape and size), as well as pollen viability. To determine mentioned pollen traits we used Axiovert 40C microscope together with a software package (AxioVision LE; Rel.4.3.) for measurement of pollen length and width. Pollen viability was determined using a staining method (ALEXANDER, 1969). The results showed that species differ by pollen grain shape (round, egg-shaped, triangular and rod) as well as by shape of exine (thick and spiky, thick to thin). In some species there was a specific number of

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apertures present (1-11). The size of viable pollen grains ranged from 29,10/12,58 $\mu$  (coriander) to 176,63/169,94 $\mu$  (oil gourd), while non-viable pollen grains were always smaller (27,27/10,97 $\mu$  to 119,62/100,86 $\mu$ ) at the same plant species. Pollen viability of most species was around 80%. Lowest pollen viability was found in white flax (56,98%), and the highest in oil pumpkin (91,43%).

*Key words*: oil species, pollen grain shape and size, pollen viability.

### INTRODUCTION

Breeding of oil species has a long tradition in Novi Sad Institute regarding sunflower, rapeseed and castor oil plant. Most of other oil species like flax, sesame, safflower, false flax, caper spurge, mary thistle, chufa sedge, coriander, dill, okra and lens represent a possible source for obtaining oil of different quality (SCHUSTER, 1992). Many of these species have very high seed oil and protein content. High protein content in the residuum after pressing, enables it's usage in processing industry for human and domestic animal nutrition.

New market demands and new trends in healthy food production assert the need for breeding of those other oil species. The collection of oil species in Novi Sad contains 12 species represented with 1-4 cultivars or landraces. Evaluation of this collection implied above all to test the production value of individual species and the variability of oil and protein content (MARJANOVIĆ-JEROMELA *et al.*, 2007).

For further work on the usage of this collection in the sense of breeding individual species or their usage as a source of «desirable genes» we also analyzed pollen grain morphology (shape and size), as well as pollen viability.

## MATERIALS AND METHODS

The oil species collection is grown on an experimental field of the Institute of Field and Vegetable Crops in Rimski Sancevi. Following species were included for evaluation: flax (*Linum usitatissimum* L.), safflower (*Carthamus tinctorius* L.), sesame (*Sesamum indicum* L.), mary thistle (*Silybum marianum* syn. *Carduus Marianus*), caper spurge (*Euphorbia lathyris* L.), coriander (*Coriandrum sativum* L.), dill (*Anethum graveolens* L.), chickpea (*Cicer arietinum*), oil pumpkin (*Cucurbita pepo var.oleifera*) and oil gourd (*Cucurbita pepo convarietas citrullinina* – varietas styriaca). Species were represented with 1-4 cultivars, or landraces.

Sowing was performed by hand in mid April of 2008. with 25cm of spacing between rows. The in row spacing was obtained by rarefying and it depended on the plant species and it's architecture. The size of trial plot was 3,6m<sup>2</sup> with three replications. Mineral fertilizers were used before sowing and plants were regularly irrigated. Samples for the analysis of pollen morphology and viability were taken at flowering time.

Axiovert 40C microscope was used to analyze pollen traits, and the AxioVision LE; Rel.4.3 software for determination of pollen grain length and width. Pollen viability was determined using a staining method (ALEXANDER, 1969).

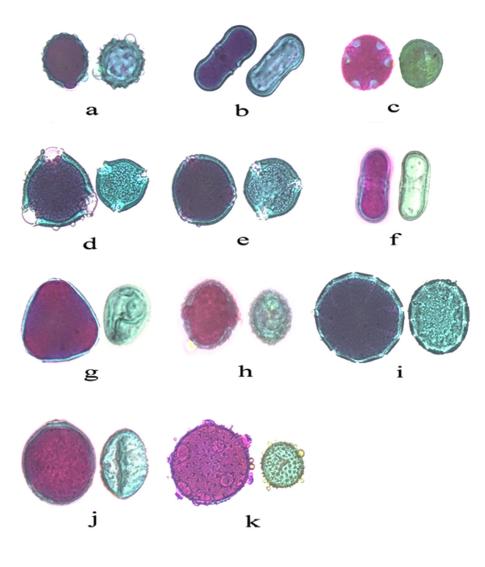
## **RESULTS AND DISCUSSION**

Results of pollen grain morphology analysis of species from oil species collection showed that above all there were differences between species, but also between viable and non-viable pollen grains of the same species. Non-viable pollen grains were mostly smaller and often of irregular shape in comparison to the viable ones.Species differed by shape and size of viable pollen grains, as well as by exine thickness. Most species had round (ball shaped) pollen grains (mary thistle, caper spurge, sesame, oil gourd) (Fig.1.). Egg shaped were the pollen grains of safflower and oil pumpkin. Stick shaped were the pollen grains of coriander and dill, while white flax, blue flax and chickpea had triangular form of pollen grains (Fig.1).Some species had pollen with thick and spiky exine (mary thistle), some had thick and spikeless exine (coriander), while most species had pollen grains with thin exine (caper spurge, safflower, oil gourd) (Fig.1).Number of pores or septs on the pollen grain varied in the analyzed species between 1 (oil pumpkin) to 11 (sesame). Most of the species had 2-3 septs (coriander, dill, safflower, white and blue flax, chickpea) (Fig.1).

Size of the viable pollen grains was from  $29,10/12,58\mu$  (coriander) to 176,63/169,94 $\mu$  (oil gourd), while non-viable pollen grains were always smaller (27,27/10,97 $\mu$  to 119,62/100,86 $\mu$ ) in the same oil species. Pollen viability in most species was around 80%. Lowest pollen viability was found in white flax (56,98%), and highest in oil gourd (91,43%) (Tab.1).

	Pollen grain size (µ)				
Species	Viable		Non-viable		Pollen viability (%)
	Length	Width	Length	Width	
Mary thistle	51,43	48,79	43,00	41,87	85,51
Coriander	29,10	12,58	27,27	10,97	75,34
Caper spurge	54,39	49,16	46,95	41,59	84,21
White flax	68,31	63,01	52,69	51,08	56,98
Blue flax	66,64	62,51	50,66	49,71	84,13
Dill	28,52	13,17	25,38	11,72	79,36
Chickpea	27,41	25,66	21,70	17,09	86,76
Safflower	54,65	51,07	49,75	41,26	60,87
Sesame	86,27	75,05	55,95	52,10	67,65
Oil pumpkin	70,79	65,24	54,72	49,86	81,97
Oil gourd	176,63	169,94	119,62	100,86	91,43

Table 1. Pollen grain size and viability of oil species from the Novi Sad collection



- Figure 1. Pollen grain morphology of oil species from the Novi Sad collection (viable – red, non-viable – green)
  - a) Mary thistle; b.) Coriander; c.) Caper spurge; d.) White flax;
  - e.) Blue flax; f.) Dill; g.) Chickpea; h.) Safflower; i.) Sesame;
  - j.) Oil pumpkin; k.) Oil gourd

The difference in size and shape between viable and non-viable pollen grains found in this work were also determined in sunflower (ATLAGIĆ, 1990), rape seed (ATLAGIĆ *et al.*, 2007) and sugar beet (MEZEI *et al.*, 2005). The results of this study also showed that the staining method by Alexander (ALEXANDER, 1969), which was previously intensively used on sunflower, is also possible to use on other analyzed oil species.

Pollen grain morphology (mature, viable) is a taxonomical trait. Sometimes species from the same genus have similar morphology, while we found that morphology was specific for most of the analyzed species (coriander and dill have similar shape and size). ČANAK and PARABUĆSKI (1966) stated that pollen grain size in most plant species varies in the range of 15 to 50  $\mu$ , and that the largest pollen grain is (150-200  $\mu$ ) in gourd, which was confirmed by the measuring taken in this trial.

The pollen grain appearance and the number of pores (septs) are species specific. MÄGDEFRAU AND EHRENDORFER (1978) have described in detail the pollen grain morphology. The part of the solid exine through which the pollen germination takes place is called "aperture". Apertures are openings through one part or whole exine. There are two types of apertures: pori or pores which are mostly isodiametric and colpi or furrows which are boat shaped. The appearance of pores depends on the viewing angle on pollen grain (polar or equatorial). Considering it is a important taxonomical trait, the authors made a pollen grain distribution, according to the pori and colpi number and appearance with a very detailed review - schematic. Pollen grains with different number of colpi are called: monocolpate, tricolpate, stephonocolpate, heterocolpate. Pollen grains with different number of pores are called: monoporate, triporate, stephonoporate, pantoporate. If the pollen grain has visible both colpi and pori than they are named according to their number: monocolporate, tricolporate, stephonocolpate, heterocolpate, stephonocolporate, the appearance and port than they are named according to their number: monocolporate, tricolporate, stephonocolporate, heterocolporate, tricolporate, stephonocolporate, heterocolporate, tricolporate, stephonocolporate, heterocolporate, the pollen grain has visible both colpi and pori than they are named according to their number: monocolporate, tricolporate, stephonocolporate, heterocolporate (MÄGDEFRAU and EHRENDORFER, 1978).

Similar to these authors, but in more detail about pollen grain morphology in the sense that pollen morphology is closely related to its function was written by RODRIGUEZ (2000). Pollen grains are generally classified according to their physical appearance. There are three criteria of classification: 1) the number and position of the apertures; 2) the shape of the pollen grain as a whole; and 3) the fine elaborate structure on the sexine. Apertures are any missing parts of the exine, which are independent of the exine pattern. Apertures are big and they cut across the fine structure pattern on the surface of the pollen grain. There are two types of apertures: pori or pores are mostly isodiametric apertures, although the can be slightly elongated with rounded ends; colpi or furrows are long and boat shaped with pointed ends. Colpi are thought to be more primitive. In living pollen grains these apertures are not actually open. Instead, a very thin layer of exine covers them. Grains with pori are called *porate*; those with colpi are called *colpate*; and those with both pori and colpi are called *colporate*. If their apertures are arranged equidistantly around the equator of the pollen grains they are assigned the prefix zono-; if they are scattered all over the surface of the pollen grain they are assigned the prefix panto-.

The number of apertures is also indicated by prefixes: *mono-* for one aperture; *di-* for two apertures; *tri-* for three apertures; and so on.

The shape of a pollen grain refers to the shape of their outline in polar and equatorial views. The shape of a grain can sometimes be useful in identifying of pollen species, but not usually. It may vary considerably within one grain type, and sometimes within one species.

The conclusion of this detailed research of correlation between pollen grain structure and function in four Angiosperm species is that many forms of pollen grains, including variations in their shape, size, number and arrangement of apertures, and the fine sculpturing of their sexine, are adaptations to help the pollen grain better perform its function of fertilizing the female gametophytes and forming seeds that will give rise to new generations of plants.

Pollen grain morphology has been studied by many authors and not only for taxonomical porpoises. PFAHLER and PFAHLER (1991) studied the influence of genotype on pollen grain morphology in sesame. The results obtained by these authors are very significant because they show that pollen grains from dehiscent anthers (free pollen) and non dehiscent anthers did not differ in shape or exine wall pattern but did differ by the dimensions of pollen. They also indicated the influence of environment on pollen dimensions.

The findings considering pollen grain traits for the species from oil species collection, especially the values of pollen viability, indicate good fertilization potential in most of the studied species. That information in combination with cross compatibility represents valuable and usable data for breeding programs. Studying the cross compatibility of different genotypes (cultivars, populations) from the same specie, as well as the cross compatibility of different species is necessary in further research of the oil species collection.

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## OSOBINE POLENA ULJANIH VRSTA IZ NOVOSADSKE KOLEKCIJE

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## Izvod

Kolekcija uljanih vrsta u Novom Sadu raspolaže sa 12 vrsta zastupljenih sa 1-4 sorte, odnosno lokalne populacije. Za dalji rad na korišćenju ove kolekcije u smislu oplemenjivanja pojedinih vrsta ili korišćenje istih kao izvor «poželjnih gena» ispitivana je morfologija polenovih zrna (oblik i veličina), kao i vitalnost polena. Za odredjivanje osobina polena korišćen je mikroskop Axiovert 40C, a za merenje dužine i širine polenovih zrna softver AxioVision LE; Rel.4.3. Vitalnost polena je odredjena bojenom metodom (ALEXANDER, 1969). Rezultati ispitivanja su pokazali da su se vrste razlikovale po obliku polenovih zrna (okrugla, jajasta, trouglasta i štapićasta) kao i po izgledu egzine (debela i bodljikava, debela do tanka). Za neke vrste je karakteristično postojanje odredjenog broja septi (1-11). Veličina vitalnih polenovih zrna se kretala od 29,10/12,58 $\mu$  (korijander) do 176,63/169,94 $\mu$  (uljana bundeva), dok su sterilna zrna bila uvek sitnija (27,27/10,97 $\mu$  do 119,62/100/86 $\mu$ ) kod istih biljnih vrsta. Vitalnost polena kod najvećeg broja vrsta je bila oko 80%. Najnižu vitalnost polena je imao lan beli (56,98%), a najvišu uljana tikva (91,43%).

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