In vitro germination of pollen grains of wild fig (*Ficus carica* L. *var caprificus*)

Klijanje zrnaca peludi divlje smokve (Ficus carica L.var.caprificus)

Dijana Vego, I. Miljković

ABSTRACT

The aim of this study is to determine pollen germination and germination energy of selected types of wild fig in the region of Herzegovina. Pollen from wild fig was collected during anthers cracking from the end of June to the beginning of July. The pollen germination and germination energy for 12 types of wild fig were found. The pollen germination was researched by Lindler's method of hanging cap. The germination energy was established on substrate with 1% agar-agar and 10% solution of sucrose in Petri dishes on the temperature of 21°C. The pollen fixation in Petri dishes was carried out with formaldehyde. The results in percents were obtained after counting germinated pollen grains under a microscope binocular and after analysing the situations on 5 observations for places germination and germination energy. It was established that the best conditions in researching pollen germination of wild fig were in 3% solution of sucrose and 0.01% solution of boric acid on temperature of 30 °C and the best conditions in researching on germination energy were in 3% solution of sucrose + 1% agar-agar + 0.01% of boric acid on temperature of 30 °C.

The best pollen germination and germination energy was established for types 4 and 2.

Key words: types, caprification, pollen germination, germination energy

SAŽETAK

Cilj ovog rada bio je odrediti klijanje peludi i snagu klijanja izabranih vrsta divlje smokve na području Hercegovine. Pelud divlje smokve sakupljena je za vrijeme pucanja prašnika od kraja lipnja do početka srpnja. Klijanje peludi i snaga klijanja određeni su za 12 vrsta divlje smokve. Klijanje peludi određivano je Lindlerovom metodom viseće kapice. Snaga klijanja utvrđena je na supstratu od 1% agar-agara i 10% otopine saharoze u Petrijevim zdjelicama na temperaturi od 21 °C. Učvršćivanje peludi u petrijevim zdjelicama učinjeno je s formaldehidom. Rezultati u postocima dobiveni su nakon brojanja izniklih zrnaca peludi pod povećalom i nakon analiziranja stanja klijanja peludi divlje smokve bili u 3% otopini saharoze i 0,01% otopini + 1% agar-agara + 0,01%

borne kiseline na temperaturi od 30 °C. Najbolje klijanje peludi i snaga klijanja utvrđeni su za vrste 4 i 2.

Ključne riječi: vrste, kaprifikacija, klijanje peludi, snaga klijanja

INTRODUCTION

A male fig plant, called caprifig, gives fruit in three crop cycles of each growing season, such as in summer (profichi), fall (mammoni) and winter (mamme). The main caprifig crop coincides with the main summer crop cycle of female trees. Although most male fig trees do not produce edible fruit, they contribute a pollen source for caprification, meaning the transfer of pollen grains from male trees to female trees by a vector. Blastophaga psenes L. Within a male syconium, female flowers reside close to the peduncle, while male flowers are placed around the ostiolum (opening at the flower end of the receptacle). The media used for pollen germination vary according to the plant species (Vasil, 1960; Baker and Baker, 1979). Pollens of some species need more complicated media (Cetin et al., 2000.). Pollen grains are morphologically simple and the process of tube formation is a relatively uncomplicated example of growth and development. For these reasons, and because of the rapid rate of tube formation in vitro exhibited by some species, pollen tube formation has become a model system for studying growth and development in plants. The reserved nutrients in pollen are important for the regulation of sucrose concentration used for in vitro pollen germination. From the literature cited, the need of sucrose concentration varies with the nutriment reserve of pollens. Many organic and inorganic substances such as sucrose, boric acid, calcium nitrate, potassium nitrate and magnesium sulfate exert an effect on in vitro pollen germination (Parton et al., 2002., Kopp et al., 2002., Galletta, 1983.). Boric acid is the stimulating agent for pollen germination and pollen tube elongation, since it is involved in the translocation and metabolism of sucrose. Bore, derived from the stigma and style, plays a role in pectin production in the pollen tube (Gibernau et al., 2003.). Effects of chemical substances on pollen germination and tube length different depending on the cultivars and doses. Researches studied the effects of different factors, including chemicals on pollen germination and tube growth (Eaton and Chen, 1969, Vasilakakis and Porlingis, 1984, Filiti et al., 1986, Egea et al., 1992.). Polen germination and tube growth of fruit trees are the most important characteristics related to pollen quality. For an economical fruit set a higher rate of pollen germination is needed in fruit trees. Excessively low rates may cause fruit set to fail because of ovule degradation before the pollen tube reaches the ovary (Egea et al., 1992).

Life capability of pollen is capability of surviving or continuation of development and it is quite different from term fertility, i.e. sterility. Some positive correlations between in vitro germination and pollen staining have been reported (Widrlechner et al., 1983.). A great variability of wild fig pollen germination in vitro has been observed (Ölçer, 1968). One of the conditions for successful germination of pollen in vitro is relatively low sucrose concentration. Temperature is also important for pollen germination in several fruit-trees. King (1963.) had shown that pollen of pecan and fig germinated the best at temperature, which approximated field temperature during blooming period of the species. Complex nutrient media has enhanced pollen germination too (Koncalova and Jicinska, 1980.).

MATERIALS AND METHODS

Fresh pollen was collected from 3 selected trees for each caprifig type among the natural population, which had matured pollen during anthers cracking from the end of June to the beginning of July (2001 – 2003) early in the morning. The pollen germination and germination energy for 12 types of wild fig were found. The pollen germination was researched by Lindler's method of hanging cap (Duraković et al., 2002). The cultures were stored in room temperature and in diffuse laboratory light. Pollen was germinated at the temperature of 21°C (pH 7) in different solutions of sucrose: I.) 5% sucrose, II.) 10% sucrose, III.) 15% sucrose and IV.) 20% sucrose. The pollen grains were removed using a brush for a homogenous distribution of the material. Counting germinated pollen grains i.e. tubes was done after 12 hours. The measurements are based on 15 readings.

After establishment of bad and unsatisfactory pollen germination in 2001 and on the basis of own observations and acquired data form literature, the research was expanded next year on pollen germination in 3% solution of sucrose and 0.01% boric acid at the higher temperature of 30°C. So, in 2002 and 2003 the research included two more temperature levels with the same procedure and corresponding solutions: V.) 3% sucrose + H₃BO₃ (21°C), VI.) 5% sucrose (30°C), VII.) 20% sucrose (30°C), VIII.) 3% sucrose + H₃BO₃ (30°C).

The germination energy was established on substrate with 1% agar-agar and 10% solution of sucrose in Petri dishes at the temperature of 21°C. The control of germination energy was monitored in the following intervals: A) 10 hours

later, B) 15 hours later, C) 20 hours later, D) 24 hours later. The pollen fixation in Petri dishes was carried out with formaldehyde.

The research was expanded during 2002 and 2003 by introduction of the new media for pollen germination (3% solution of sucrose in agar-agar and boric acid (0.01%) with temperature increasing as follows: E) After 10 hours, 3% sucrose+agar-agar+ H₃BO₃ to 21°C, G) After 10 hours, 10% sucrose+agar-agar+ H₃BO₃ on 30°C, K) After 10 hours, 3% sucrose+agar-agar+ H₃BO₃ on 30°C, F) After 24 hours, 3% sucrose+agar-agar+ H₃BO₃ on 30°C, H) After 24 hours, 10% sucrose+agar-agar+ H₃BO₃ on 30°C, H) After 24 hours, 10% sucrose+agar-agar+ H₃BO₃ on 30°C, H) After 24 hours, 10% sucrose+agar-agar+ H₃BO₃ on 30°C).

The results in percentages were obtained after counting germinated pollen grains under microscope (x100) and after analysing situations on 15 observation places for pollen germination and germination energy.

RESULTS

Type 2 (6.88%) in 2001 had better pollen germination in 5% solution of sucrose than type 4 (4.30%) in 2002 and type 9 (4.04%) in 2003. In 10% solution of sucrose types 2 (9.09%) and 4 (8.01%) in 2001 and 2002, and type 4 (6.04%) in 2003 had the best pollen germination. In 15% solution of sucrose pollen germination was the best in type 4 in all three years (10.99%, 14.17%, 9.93%) and it was also the best type in 20% solution of sucrose (17.33%, 26.03%, 19.78%). In 2003 better pollen germination was in type 9 (4.04%, 5.96%, 9.51%, 11.38%). Generally, all of the types had similar and better pollen germination in 2003 than in previous years when variations between types were expressed. All of the types had germinated pollen in 2003 in 15% and 20% solution of sucrose.

In 2001 types 3, 5, 6 and 11 did not germinate, and partially even types 8, 10 and 12. During next years the mentioned types have showed very low pollen germination or partial germination but it was always with higher sucrose concentrations. As a rule pollen germination became better with increasing sucrose percentage. An temperature of 21°C the best pollen germination was in 20% solution of sucrose.

In 2001 pollen germination in 20% solution of sucrose ranged from 0 (types 3, 5, 6, 8, 10 and 11) to 17.33% (type 4), in 2002 from 0 (types 8 and 10) to 26.03% (type 4) and in 2003 germination varied from 2.59% (type 5) to 19.78% (type 4).

Table 1: Pollen germination (%) of researched types of wild fig (*Ficus carica* var.caprificus) (combinations I – IV)

Tablica 1: Klijanje istraživanih vrsta divlje smokve (*Ficus carica* var. *caprificus*) (komb. I-IV)

-		I			II			Ш			IV			
		5% sucrose (21°C)			10% sucrose (21°C)			15% sucrose (21°C)			20% sucrose (21°C)			
		x			x			x			x			
Type of wild fig	2001	2002	2003	2001	2002	2003	2001	2002	2003	2001	2002	2003		
1	5,90	0	0	5,91	0	3,12	6,86	1,95	5,29	9,01	3,19	5,62		
2	6,88	2,63	2,98	9,09	4,58	4,93	10,75	5,01	6,05	13,80	9,24	7,85		
3	0	0	2,78	0	1,78	3,64	0	4,76	5,74	0	4,42	9,11		
4	6,83	4,30	3,37	8,15	8,01	6,04	10,99	14,17	9,93	17,33	26,03	19,78		
5	0	0	0	0	0	0	0	1,74	1,86	0	2,36	2,59		
6	0	0	0	0	0	1,46	0	2,30	3,24	0	4,77	3,92		
7	3,09	0	2,96	5,40	2,41	3,73	6,24	5,91	5,19	7,34	6,83	5,34		
8	1,59	0	0	2,82	1,48	1,69	0	0	3,67	0	0	6,55		
9	4,15	2,37	4,04	4,18	4,86	5,96	5,52	4,62	9,51	6,42	5,28	11,38		
10	2,59	0	0	4,16	2,00	2,43	0	0	4,43	0	0	5,54		
11	0	0	0	0	0	0	0	0	2,13	0	3,21	2,90		
12	0	1,37	0	0	3,17	0	4,23	3,03	3,26	3,10	3,44	5,87		

General conclusion is that pollen germination in previously described conditions of temperature and sucrose concentration (21°C; 5%; 10%; 15% and 20% solution of sucrose) was low. Types 2 and 4 had better germination than other researched types.

Temperature increasing from 21°C to 30°C, combinations VI, VII and VIII (table 2), leaded to better conditions for pollen germination of wild fig and it is obvious from comparison earlier results of research where pollen germination in researched types was pretty good. On the other hand, increase of sucrose solution concentration had no influence on increase of pollen germination unlike previously described combinations I, II, III and IV. Good pollen germination was obtained even with use of low concentrations of sucrose solution, but at temperature of 30%. Better pollen germination was in 5% solution of sucrose than in 20% solution.

Here is selected type 2 with good pollen germination (from 20.96% to 50.96%) and type 4 (from 18.59% to 41.03%). In 2003 good pollen germination was established in type 9 (from 16.18% to 31.60%). On the contrary, type 5 had low germination (from 2.09% to 4.92%) and type 6, too (from 3.17 to 5.56%). Partial pollen germination in 2002 had types 7 (0-8.05%) and 11 (0-7.07%) whose pollen germinated only in 20% solution of sucrose and it did not germinate in 5% solution of sucrose.

Table 2: Pollen germination (%) of researched types of wild fig (*Ficus carica* var. caprificus) (combinations V – VIII)

Tabica 2: Klijanje peludi (%) istraživanih vrsta divlje smokve (*Ficus carica* var. *caprificus*) (komb. V-VIII)

	V			VI	V	II			
	3% sucrose (21°			oucrose C)		ucrose °C)			
	Ī			$\bar{\mathbf{x}}$	ž	x			
Type of wild fig	2002	2003	2002	2003	2002	2003	2002	2003	
1	0	5,32	5,96	8,20	6,30	7,07	8,71	13,25	
2	6,10	5,19	50,96	30,74	20,96	22,74	39,95	36,85	
3	0	4,26	6,76	16,74	4,43	13,81	12,53	20,76	
4	6,13	4,71	41,03	27,94	18,59	18,86	48,27	35,38	
5	0	0	2,09	4,92	2,83	3,32	3,93	5,41	
6	0	0	4,63	5,56	5,21	3,17	5,74	7,84	
7	0	4,02	0	8,30	8,05	9,70	0	17,64	
8	0	0	5,25	12,88	5,32	7,89	7,31	14,88	
9	4,15	6,36	7,39	31,60	5,08	16,18	10,12	42,76	
10	3,19	2,71	8,34	7,87	4,68	4,85	13,06	13,60	
11	0	0	0	8,28	7,07	5,36	0	8,47	
12	0	0	5,48	7,58	4,59	4,70	8,3	10,87	

Table 3: Pollen germination energy (%) of researched types of wild fig (*Ficus carica* var. *caprificus*) (combinations A – D)

Tablica 3: Snaga klijanja peludi divlje smokve (komb. A-D)

	A After 10 hours 10% sucrose + agar (21°C)			B After 15 hours 10% sucrose + agar (21°C)				С		D After 24 hours 10% sucrose + agar (21°C)		
								er 20 hour aucrose + a				
								(21°C)				
	x			x			x			x		
Type of wild fig	2001	2002	2003	2001	2002	2003	2001	2002	2003	2001	2002	2003
1	5,72	0	0	7,63	3,27	3,85	9,86	3,80	4,50	10,42	6,28	6,40
2	4,54	2,22	2,81	8,73	6,53	6,67	15,60	8,61	7,20	16,69	13,04	16,86
3	0	0	4,48	2,88	4,68	5,77	5,46	8,62	8,16	6,75	15,01	15,14
4	4,75	3,68	4,02	7,06	5,02	8,73	17,16	15,87	16,20	20,98	22,14	20,70
5	0	0	0	0	3,73	3,00	2,10	2,53	3,00	4,13	2,25	3,86
6	0	0	0	0	4,04	0	2,57	5,19	2,30	3,57	5,50	3,73
7	3,13	0	2,86	5-79	4,50	5,94	6,70	7,00	6,58	7,44	10,87	8,40
8	2,20	0	0	4,23	3,27	6,05	6,30	4,74	9,73	6,66	3,90	11,96
9	4,71	2,12	3,87	6,01	5,62	8,65	8,84	10,04	10,81	9,88	12,58	11,87
10	3,62	0	0	3,31	2,50	5,53	4,43	2,48	6,28	5.67	2,91	7,00
11	1,29	0	0	2,24	1,50	2,85	5,75	3,57	6,26	5,34	5,17	8,71
	0	2,92	1,99	2,87	4,65	6,44	4,49	5,50	6,88	6,05	6,90	7,16

Better pollen germination was obtained by adding boric acid (0.01%) in 3% solution of sucrose with temperature of 30°C in most of researched types. Adding boric acid to sucrose at temperature of 21°C had no influence on pollen germination.

Results of researching pollen germination energy are shown in tables 3 and 4. In 2001 after 10 hour's type 1 had better germination (5.72%) and types 2, 4 and 7 (4.54%, 4.75% and 4.71%). Low pollen germination was recorded in 2002 after 10 hours in type 4 (3.68%) while in 2003 types 3 and 4 had the best germination (4.48% and 4.02%).

Table 4: Pollen germination energy (%) of researched types of wild fig (*Ficus carica* var. *caprificus*) (combinations E – H)

		E		E G			K		F		L		Н	
	After 10 hours 3% sucrose + agar + H ₃ BO ₃ (21°C)		After 10 hours 10% sucrose +agar (30°C)		After 10 hours 3% sucrose + agar+ H_3BO_3 ($30^{\circ}C$)		After 24 hours 3% sucrose + agar+ H ₃ BO ₃ (21°C)		After 24 hours 3% sucrose + agar+ H ₃ BO ₃ (30°C)		After 24 hours 10% sucrose + agar (30°C)			
	x		x		x		x		x		x			
Type of wild fig	2002	2003	2002	2003	2002	2003	2002	2003	2002	2003	2002	2003		
1	0	1,88	2,28	3,65	4,58	4,49	7,98	9,40	38,49	33,38	13,68	11,42		
2	2,79	3,21	4,10	5,58	7,42	8,74	15,98	7,55	61,65	62,17	9,99	13,40		
3	3,85	5,54	5,98	7,74	15,92	10,71	64,19	60,38	92,11	90,06	75,51	56,58		
4	4,22	5,71	5,44	8,29	12,42	14,83	61,65	62,91	96,55	95,37	82,04	77,87		
5	4,07	0	2,88	2,30	5,84	3,24	6,92	6,02	28,32	23,04	4,17	4,09		
6	2,80	2,25	3,25	3,64	6,76	5,49	7,29	6,55	32,76	27,00	4,34	4,27		
7	0	3,28	0	3,87	4,25	4,60	38,53	31,37	68,31	61,54	32,87	27,11		
8	0	0	0	4,27	0	6,51	27,48	20,00	64,79	58,80	18,70	16,47		
9	4,19	4,75	3,96	5,18	7,86	7,60	40,04	44,39	78,28	70,17	19,09	23,43		
10	0	0	2,74	2,18	9,15	3,35	17,16	9,39	37,25	24,89	15,64	9,66		
11	0	0	0	2,74	0	5,45	3,95	4,46	26,10	33,14	4,02	5,22		
12	2,98	3,09	3,48	3,02	5,95	3,96	7,55	7,90	32,23	31,82	5,23	6,05		

Tablica 4: Snaga klijanja peludi divlje smokve (komb. F-H)

After 15 hours the best pollen germination was observed in type 2 in 2001 (8.73%) and in 2002 (6.53%), but in 2003 the best was type 4 (8.73%). After 20 hours type 4 had adequate germination from 15.87% to 17.16% and after 24 hours with pollen germination from 20.70% to 22.14%.

Application of new media for pollen germination and temperature increase (2001 and 2003) had on influence on pollen germination.

Type 4 showed the best pollen germination energy after 10 hours in combinations E, G and K. Satisfying pollen germination was noticed even in other types by introduction of 3% solution of sucrose + agar-agar + H_3BO_3 on temperature of 30°C.

Accordingly, germination energy was better with addition of boron and with lower concentration of sucrose (3%) but at higher temperature.

After 24 hours on the media F (3% solution of sucrose + agar-agar + H_3BO_3 at temperature of 21°C) types 4, 3 and 9 had the best pollen germination (61.65%, 64.19%, 40.04%, 62.91%, 60.38%, 44.39%) and on the media H (10% solution of sucrose + agar-agar, on temperature of 30°C) types 4 and 3 (82.04%, 75.51%) (77.87%, 56.58%). Low pollen germination was observed in types 5, 6 and 11 (4.17%, 4.34%, 4.02%, 4.09%, 4.27%, 5.22%).

After 24 hours in 3% solution of sucrose + agar-agar + H_3BO_3 (combination L) and at temperature of 30°C the best germination energy was in types 4 (96.55%) and 11 (26.10%) in 2002 and types 4 (95.37%) and 5 (23.04%) in 2003. Besides type 4 with best germination in both years, types 3 (92.11%, 90.06%) and 9 (78.28%, 70.17%) showed satisfying germination energy.

Monitoring the beginning pollen germination it can start after 10 hours, but it is complete after 24 hours when most pollen grains germinate, especially in some types of wild fig. Types 3, 5, 6 and 11 did not germinate after 12 hours, but they showed better germination energy after 15 hours and even more after 20 and 24 hours.

DISCUSSION

One of the important characteristics of successful pollination is good pollen germination. According to Dakuzoğuz (1953.) optimal and satisfying germination is higger than 30%. In researching fruit (apple, pear, cherry and other) pollen it is usual to use the temperature of 21°C. However, in our researching in 2001 at the temperature of 21°C very low pollen germination was established in fig, but at the same time jigger in solutions with higher sucrose percentage. Even in such low germination better results had types 2 and especially 4. Type 4 showed the best pollen germination energy.

Kuden (1998.) introduced 39 varieties and clones of wild fig with their percentage of germination and amount of pollen. High percentages of germination were recorded (77%-100%) and most frequently medium amount of pollen, particular by nine Turkish cultivars of wild fig (*Bostanci, Siyah ilek, Çiçekli II, Ak erkek II, Cankurt, Armut ilek, Yanako II, Kara erkek I, Kızılburun*).

Percentage of pollen germination on 6 cultivars of wild fig (*Taşlik, Haci Mestan, Kıbrıslı Kűçűk Konkur, Çakin, Mordemirtaş*) were investigated by Zeybeoğlua et al. (1998.). Germination ranged from 29.04% to 15.97% by using

different substrates. The research confirmed the fact that pollen germination depends on the cultivation media. The best germination was obtained on medium containing 5% sucrose $+H_3BO_3+Ca(NO_3)_2+KNO_3+MgSO_4$. Good results were also obtained with 3%-5% sucrose solution with addition of salt and boric acid in 1% agar. Pollen germination was better at lower sucrose concentrations. This research also confirmed that temperature was very important factor for pollen germination. Pollen did not germinate at room temperature but it germinated at 30°C in the dark. Cultivars with germination over 30% were separated, in accordance with previous research (Dakuzoğuz, 1953.) where figs with pollen germination over 30% were considered as representative.

Also researched germination at temperature higher then 30° C for possible increase of pollen germination. Increase of temperature gave great results in improving pollen germination. The best pollen germination, even at higher temperature, had type 2 (31.35% - approximate value for 2 years) and type 4 (26.6%). In 2003 better pollen germination was recorded in type 9 (23.9%), while low germination was recorded in type 5 (3.39%) and type 6 (4.64%).

Assording to Ilgin et al. (2007.) the caprifig pollen did not germinate at all on a medium without sucrose.

Increasing sucrose concentrations up to 20% improved pollen germination percentages. However. germinations decreased with higher sucrose concentrations (25 or 30%). This finding showed the negative effect of higher sucrose concentrations on pollen germination as reported for some stone fruits (Bolat & Pirlak, 1999). In all caprifig types tested, the highest pollen germinations (between 60.08 and 68.48 %) were achieved on the media containing 20% sucrose. The germination percentages of some caprifig pollen were highest (over 70%) with the addition of 0.050% H₃BO₃, followed by 0.025% KNO₃. Germination percentages of the caprifig pollen were higher with the addition of these chemicals than the 20% sucrose alone. The pollen of Pistacia vera from Anacardiaceae need 50% sucrose concentration, Avocado cultivars germinate in 15% sucrose concentration (Sahar and Spiegel, 1984.).Vasil made some investigations on the pollen germination on 9 taxa of Cucurbitaceae and he used different concentrations of sucrose. His results show that pollen germination is optimum between 7.5% and 20% of sucrose concentrations. The best results for germination energy in almond were obtained at 24°C with 15% sucrose after 24 h (Slavgorodskii, 1980.).

From everything said before we can conclude that to research on pollen germination in a laboratory it is necessary to stimulate natural conditions. Namely, in years of research approximate daily temperatures during anthers pollination and caprification, i.e. in the period from June 25th to July 5th varied from 28.6 °C in 2001 and 2002 to 27.9 °C in 2003, which was similar to temperatures in laboratory conditions.

It is necessary to mention that increasing concentration solution of sucrose had no influence on germination with temperature increase while there was a positive influence on lower temperature (21 °C). So, good germination was established at low concentrations of sucrose solution and the confirmation for that was better pollen germination in 5% sucrose solution than in 20% solution.

Adding boric acid (0.01%) in 3% solution of sucrose and the temperature of 30 °C resulted in most types with better pollen germination. However, the same procedure at temperature of 21 °C was not efficient for pollen germination.

Media for germination are determined by temperature, percentage of sucrose and introduction of boron. So, establishing pollen germination in laboratory conditions is a very sensitive procedure and it is very important for the success of germination.

This research established the best conditions for pollen germination related to combination VIII, while combination L, was the best for pollen germination energy. Our results correspond with similar research in the world (Zeybekoğlu et al., 1998.).

Results from 3 years of research on pollen germination and germination energy selected type 4 as the best one. Better pollen germination than 30% (combination VIII) in 2002 and 2003 was recorded only for types 2 and 4 and in 2003 for type 9. Germination energy of more than 30% (combination L) was recorded in all types excepted in type 5 in 2002 and 2003, in types 6 and 10 in 2003 and in type 11 in 2002. However, previously separated types had significant germination because their percents of germination energy were a little bit under 30% (type 5 - 28.32%; 22.50%; type 6 - 25.83%).

Besides pollen germination important factors for evaluation of pollinators are: pollen germination energy, anthers development, pollen maturation, wasp's exit, duration of wasp's exit, amount of wasps, wasps attraction etc. This research may focus the selection of wild figs on, a detailed study of pollinators and their effectiveness "in the field "with the aim of providing satisfactory caprification.

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Author's address – Adresa autora

Dijana Vego Agronomski i prehrambeno-tehnološki fakultet Sveučilišta u Mostaru Biskupa Čule bb. 88 000 Mostar, BiH Ivo Miljković Čazmanska II/3, 1000 Zagreb