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Effect of Climatic Conditions on Flowering of Walnut Genotypes in Romania

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Abstract

The timing and lowering type of walnut genotypes (*Juglans regia* L.) is one of the important factors that lead to high and constant production. Therefore, the selection of homogame and spring frost tolerant genotypes has been among the most important priorities in breeding programs of walnut. The present paper analyzed the degree of dichogamy and the flowering time of 19 promising genotypes in walnuts from the sandy area, the S-V Oltenia region, Romania. Observations made over two calendar years have shown that the flowering period is influenced by environmental factors, especially temperature. The flowering time has changed from one year to the other, the high temperatures in spring accelerate the evolution of the male flowers,but the female flowers are not greatly influenced by these temperatures. Thus the type of flowering was not constantly, changing according to the climatic conditions of the two years. Knowing the type and timing of flowering of walnut genotypes is very important for pollination, because homogamy types are quite rare in the analyzed population.

Keywords: Dichogamy, Flowering, Temperature, Walnut.

Introduction

At present, for sustainable crops, particular attention is paid to the placement of genotype in areas where ecological factors are part of the biological requirements of the species. Although they are grown in vast areas of Romania, there are few areas where walnuts have high productivity, which shows that, in addition to their genetic characteristics, ecological factors play a decisive role in obtaining great and high quality productions. The values of ecological factors during the flowering and pollination period influence the production of nuts. Manescu *et al.* (1975) advocated the need for consistency between the biological requirements of the species and the environmental factors to obtain high quality and constant product. Cosmulescu *et al.* (2002, 2010) and Pintea (2004) showed that the flowering period is lower if the temperatures are higher, and the higher flowering period is the one which leads to a higher percentage of pollination that increases the productivity. The production of nuts is largely dependent on weather conditions, especially during pollination and fruit growth, and the amount of pollen available in the air plays an important role (Prentovic *et al*., 2014). Moderate to high heritability (0.39-0.88) was estimated by between-family variance, suggesting that the majority of phenotypic variations were under additive genetic control, according to Rezaee *et al.* (2009). Walnuts are

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monoecious species. Typically the period of pollen shedding does not completely overlap the period of female receptivity. This is known as dichogamy. Dichogamy is the major factor in decreasing productivity, the problem of dichogamy is being aggravated by the short period of pollen release by male flowers and by the receptivity of female stigma. Of the 20 walnut selections studied in India, 14 were protanders, the proton or protogin character is not constant, the degree of dichogamies ranging from 0 to 100% (Kumar and Sharma, 2013). Luedeling and Gassner (2012) showed that high temperatures have influenced the flowering of walnuts and the appearance of leaves in California. A study by Lee *et al*. (2016) on the impact of climate change on crops in Yolo County (USA) showed that walnut is one of the most vulnerable crops, and projections indicated that some varieties may be particularly affected in years of warm winters, seriously reflected on productivity. Floral characters such as bearing habit, abundance of male and female flowers coupled with dichogamy are also important traits used in characterization and variation studies in walnut (Sharma and Das, 2003). Ecological condition may affect dichogamy in walnut (Sen, 1998). A large variation of adaptation characters such as the early and dwelling periods and the dichogamy offer the possibility of adapting the crop to various agro-ecological environments (Khadivi-Khub *et al*., 2015). The timing and type of flowering of walnut genotypes is one of the important factors that lead to high and constant production. Therefore, the selection of homogamy and spring frost tolerant has been among the most important priorities in the breeding programs of walnut. Starting from the above, the paper analyzed the degree of dichogamy and the flowering time of some perspective walnut genotypes in the sandy area on the left of the Jiu river, S-V Oltenia region, Romania.

Materials and Methods

Twenty genotypes of the Bechet population have been studied, located on the sands on the left side of the Jiu river, the S-V Oltenia region (43°47′ N; 23°57′ E), a temperate climate area. All studied trees are seed originated andgrowing naturally.

The phenological observations and climatic factors were recorded in 2 calendar years (2016, 2017), during the walnut flowering period. The degree of dichogamy was calculated using the formula used by Solar (1995) and Cosmulescu *et al*. (2001a): DD% = (1 - number of days when female and male flowers coincide in blooming / number of days when female flowers are flowering) x 100.

Bechet is situated in the silvosteppe area of the Romanian Plain, the average temperature is 11 C and the annual average rainfall is 500 mm. The warmest month of the last 50 years was August 2003 with an average temperature of 26.4°C, and the coldest month was January 2006 with an average temperature of -5.5°C. The warmest year of the last 50 years was 2002 with an average of 12.8°C, and the coldest was 2006 with an average of 11.5°C.

Statistical analysis

The data obtained from the measurements were statistically processed using the descriptive statistical program.

Results

The temperature conditions the processes of assimilation, breathing and sweating, the various phases of growth and fructification, the latent life of the trees during the winter rest, etc. The most important climatic factor influencing the flowering process at walnut is the average air temperature during the flowering period and the amount of accumulated active temperature up to the date when the phenophase starts.

Observations on the 19 genotypes of the Bechet population, located on the sands on the left side of Jiu

river, showed that walnut blossoming in this area takes place in April. Using the classification made by Cosmulescu (2010), depending on the time of flowering, the studied genotypes were grouped into 4 classes:early- April 3-9; mid-early - 10-12 April; semi-late - April 13-15; late - after 15 April. The flowering time differed from one year to the other (Table 1). Most genotypes (13 of 19 had a mid-early blossoming in 2016 and in 2017, 9 genotypes had a nearly flowering (Table 1). The time and duration of flowering have changed with environmental factors (Table 2).

Table 1. Timing of flowering and dichogamy degree for the walnut genotypes analyzed

Genotype	Dichogamy degree (%)		Timing of blossoming for male/female flowers*	
	2016	2017	2016	2017
B1	79.00	55.25	SE/SE	E/SE
B5	69.00	51.94	SE/SL	E/SE
B6	57.33	74.00	SE/SL	E/E
B7	52.33	69.59	SL/SE	E/SL
B8	99.00	99.00	SE/SE	SL/SE
B9	52.33	46.06	E/SL	E/SE
B10	14.38	45.15	SE/E	E/SL
B14	49.00	45.67	SE/L	SE/SL
B15	62.64	52.33	SE/SE	SE/SL
B16	65.67	30.25	SE/E	E/E
B19	71.73	91.31	SE/E	SL/SL
B22	84.71	56.14	SL/SE	E/SL
B24	80.82	52.85	SE/E	SL/SL
B28	59.00	52.85	SE/E	SL/SL
B29	59.00	45.67	SE/E	SE/SE
B32	59.00	59.00	E/E	SE/SE
B33	77.57	65.67	E/E	E/SL
B36	56.14	32.33	SE/E	E/E
B39	60.54	37.46	E/E	SL/E
Mean	63.64±17.61	55.92±17.40		

*E=early; SE=semi-early; SL=semi-late; L=late

Table 2. The period of flowering, the duration and the medium temperatures during the flowering for the walnut genotypes in Bechet area

Genotype	Type of flowers	Time of flowering		Duration of flowering (days)		Medium temperature (°C)	
		2016	2017	2016	2017	2016	2017
B1	female	12.04–21.04	5.04–18.04	10	14	15.3±2.94	10.86±2.47
	male	12.04-19.04	10.04-18.04	8	9	16.38±2.06	11.22±2.86
B5	female	12.04-21.04	9.04-25.04	10	17	15.3±2.94	9.59±3.16
	male	13.04-19.04	10.04-18.04	7	9	16.57±2.14	11.22±2.86
B6	female	11.04-22.04	8.04-19.04	12	12	14.75±2.95	10.58±3.08
	male	13.04-19.04	8.04-17.04	7	9	16.57±2.14	11.5±2.41

Table 2. Continued.

B7	female	9.04-23.04	9.04-25.04	15	17	14.67±2.63	9.59±3.16
	male	13.04-20.04	14.04-26.04	8	13	16±2.56	9±3.46
B8	female	12.04-21.04	13.04-24.04	10	12	15.3±2.4	8.75±3.38
	male	12.04-21.04	12.04-24.04	10	13	15.3±2.94	8.92±3.30
B9	female	9.04-23.04	9.04-25.04	15	17	14.67±2.63	9.59±3.16
	male	14.04-21.04	10.04-17.04	8	8	15.25±3.32	11.75±2.54
B10	female	11.04-23.04	13.04-25.04	13	13	14.69±2.83	8.85±3.26
	male	9.04-12.04	9.04-18.04	5	12	14±1.41	11.3±2.71
B11	female	11.04-21.04	9.04-24.04	11	16	15±2.96	9.56±3.26
	male	13.04-19.04	13.04-24.04	7	12	16.57±2.14	8.75±3.38
B14	female	11.04-22.04	11.04-25.04	12	15	14.75±2.95	9.27±3.23
	male	17.04-22.04	14.04-20.04	6	7	15±4.14	9.14±4.01
B15	female	10.04-20.04	11.04-25.04	11	15	15.45±2.46	9.5±3.23
	male	12.04-18.04	13.04-20.04	7	8	16.14±2.11	9.5±3.85
B16	female	10.04-21.04	9.04-24.04	12	16	15±2.82	9.56±3.26
	male	9.04-17.04	3.04-13.04	9	11	14.89±1.61	11.36±1.68
B19	female	12.04-22.04	13.04-25.04	11	13	15±2.96	8.85±3.26
	male	9.04-19.04	13.04-24.04	11	12	15.642.24	8.75±3.38
B22	female	16.04-24.04	5.04-25.04	7	21	14.78±3.30	9.62±2.88
	male	10.04-21.04	13.04-24.04	12	12	15±2.82	8.75±3.38
B24	female	12.04-22.04	13.04-25.04	11	13	15±2.96	8.85±3.26
	male	9.04-20.04	13.04-19.04	12	7	15.33±3.38	10±3.87
B28	female	11.04-20.04	13.04-25.04	10	13	15.5±3.59	8.85±3.26
	male	9.04-16.04	13.04-19.04	8	7	14.5±1.19	10±3.87
B29	female	12.04-21.04	11.04-25.04	10	15	15.3±2.94	9.27±3.23
	male	9.04-17.04	11.04-17.04	9	7	14.89±1.61	11.71±2.75
B32	female	8.04-22.04	10.04-24.04	15	15	14.8±2.65	9.4±3.31
	male	9.04-17.04	12.04-20.04	9	9	14.89±1.61	9.67±3.64
B33	female	9.04-22.04	7.04-24.0414	14	18	14.71±2.72	9.5±3.07
	male	9.04-19.04	13.04-24.04	11	12	15.64±2.24	8.75±3.38
B36	female	10.04-23.04	4.04-24.04	14	21	14.71±2.72	9.71±2.93
	male	9.04-17.04	7.04-13.04	9	7	14.89±1.61	11.14±1.57
B39	female	9.04-21.04	13.04-25.04	13	13	14.92±2.72	8.85±3.26
	male	8.04-6.04	9.04-17.04	9	9	14.67±1.22	11.78±3.38
Mean/range	female	8.04-24.04	4.04-25.04	11.81±2.11	8.6±1.90	14.98±0.28	9.39±0.55
	male	9.04-22.04	3.04-26.04	15.37±2.63	9.65±2.23	15.40±0.25	10.30±1.22

Discussion

The blossoming of female flowers in this area took place between April 9-16 in 2016 and April 5-13 in 2017 with a flowering period of 7-15 days in 2016 and 12-21 days in 2017. In Neiriz, Iran, Khadivi-Khub *et al*. (2015) reported a coefficient of variation for the blossoming of the female flowers of 104.20%. This genotypes of walnut in Iran's receptive period in female flowers and the pollen shedding period varied from the first decade of April to the first decade of May (Mahmoodi *et al*., 2016). The blossoming of the male flowers took place in 2016 between 9-14 April with a duration of 6-12 days and between April 3-14 in 2017, with a flowering period of 7-13 days. Similar results were obtained by Godeanu (1975), which reported a 4-8 day blossoming of male flowers, and female flowers between 6 and 15 days. Cosmulescu *et al*. (2001b) reported a 10-13 day period in blossoming of male flowers, full flowering being in 5 days and 15-day period in female flowers, while for walnut varieties grown in Ramnicu Valcea the full flowering is 9 days. This explains the influence of climatic conditions on the flowering time of walnuts. Studying 6 Romanian walnut varieties, Cosmulescu *et al*. (2010) shows that the flowering duration of the aments, or catkins, is between 8-11 days. Also, in Slovenia, the average flowering period for female flowers was 10.8 days for protandry varieties and 12.4 days for protogenetic varieties (Solar and Wine 1997). In Kashmir, India, the number of days in which the flowering of the two types of flowers on the same genotype coincides is at least 6 days, with a dichogamy level of less than 25% (Kumar, 2016). Pollen source is affecting nut and kernel diameter, kernel weight, length to diameter, shell-thickness and shell weight, according to Golzari *et al*. (2016).

The type of flowering was not constant therefore, it changed according to the environmental conditions of the two years. B22 and B36 genotypes that behaved like protandry varieties in 2016, in 2017 behaved like protogenetic varieties with a gap of 11 and 6 days, respectively. Genotypes B19, B24, B28, B29 that behaved like protanders in 2016, in 2017 were homogamy. Genotype B7, a protogin genotype in 2016, behaved like homogamous in 2017. The ability to move from protogin to homogamous can be explained embryologically: the male flower anteres are formed in summer-autumn, and the meiotic division takes place in the spring, as well pollen formation (Cosmulescu *et al*., 2010). In terms of flowering characteristics, Keles *et al*. (2014) found 11 prototype genotypes, 5 genotypes were protogyne and 4 genotypes were homogamy in their promising 20 genotypes in walnut seedling populations naturally grown in Gumushacikoy, Turkey. Akca and Ozongun (2004) selected 17 walnut genotypes، determined 11 genotypes of protogyny and 6 selections as protandry. Protandry is a genetically dominant character in *Juglans regia* (Akca and Sen, 1997).

Temperature is very important during the flowering period, especially during the opening of the male flowers (Cosmulescu *et al*., 2010). The analysis of the recorded data reveals that in most genotypes (80%), the female flowers blossomed at an average temperature of 11.77-12.86°C and the male flowers at an average temperature of 12.23-13.31°C (Table 3). Flowering time for female flowers was 7-15 days in 2016 at an average temperature of 14.67-15.50°C and 12-21 days in 2017 at an average temperature of 8.75-10.86°C, which shows that high temperatures reduce the flowering period, speeding up this process (Table 2). The flowering of male flowers in 2016 was 5-12 days at an average temperature of 14.00-16.57°C and 7-13 days in 2017 when the average temperature varied between 8.75-11.78°C (Table 2). Differences from year to year also appear among the flowers of the same genotype and show that elevated temperatures reduce the number of days of the flowering period, influencing more the blossoming of the male flowers. Genotype B10 in 2016 had a flowering period

of 5 days for male flowers, at an average temperature of 14°C and in 2017 when the temperature was lower (11.30°C), of 12 days and a blossoming period of female flowers for 13 days at different average temperatures of 14.69°C and 8.85°C respectively. Genotype B39 has maintained the flowering period for female (13-day) and male (9-day) flowers at medium temperatures to be different in 2016 compared to 2017.

Table 3. The distribution of medium temperatures during the flowering period of walnut genotypes analyzed in Bechet area

Female flowers		Male flowers	
Class – medium temperature (°C)	% Cumulative	Class – medium temperature (°C)	% Cumulative
11.77	5.00%	11.87	5.00%
11.98	25.00%	12.23	25.00%
12.2	60.00%	12.59	40.00%
12.42	80.00%	12.95	60.00%
12.64	90.00%	13.31	80.00%
12.86	95.00%	13.67	85.00%
More	100.00%	More	100.00%

As to the degree of dichogamy (Table 1), it differs from one year to the other influenced by the high temperatures that rush the evolution of male flowers, reducing the number of days in which the flowering of female and male flowers coincides. Genotype B8 and B32 retained their dichogamy level in the two years, with the lowest degree of dichogamy (30.25%) being recorded in B16 genotype in 2017. The problem of dichogamy is further aggravated by a short period of pollen shedding and stigma of receptivity. Dichogams in walnut prevents self-pollination and requires cross-pollination to set fruits (Kumar and Sharma, 2013).

Conclusions

The flowering period is influenced by environmental factors, especially temperature. The flowering time has changed from one year to the next. High spring temperatures accelerate the evolution of male flowers and female flowers are not greatly influenced by these temperatures. Thus the type of flowering was not constant, while changing according to the climatic conditions of the two years. Knowing the type and timing of flowering of walnut genotypes is very important for pollination because homogamy types are quite rare.

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