# ORNAMENTAL, PHYSIOLOGICAL AND ENZYMATIC EVALUATION OF SOME GLADIOLUS SPECIES

## Petronica AMIŞCULESEI, Maria APOSTOL, Elena Liliana CHELARIU, Liliana ROTARU, Lucia DRAGHIA

"Ion Ionescu de la Brad" University of Life Sciences from Iasi, 3 Mihail Sadoveanu Alley, Iasi, Romania

Corresponding author email: mariabrinza2007@yahoo.com

#### Abstract

The genus Gladiolus (Iridaceae family), with about 270 species, is native to different regions of South Africa and the Mediterranean area, in the first region having a much larger distribution with a larger number of species (more than 114 species). In the Mediterranean area, the ten to twelve species of gladiolus offered taxonomic confusion due to their similarities and morphological variations due to localized evolution. The study effectuated during the period 2018-2020 at two Gladiolus species (G. byzantinus and G. imbricatus), existing in the collection of the lasi University of Life Sciences, had as purpose the analysis of some ornamental characters and also the content of photosynthetic pigments and the enzymatic activity of the leaves. G. byzantinus formed denser shrubs (higher number of shoots per plant), but in G. imbricatus there were more leaves/ shoot, longer flower stalks, denser inflorescences (shorter distance between flowers). G. byzantinus bloomed about two weeks earlier, but flowering time was seven days longer in G. imbricatus. The content of assimilative pigments was higher in G. imbricatus and was correlated with a lower enzymatic activity.

Key words: assimilative pigments, enzymatic activity, gladiolus, ornamental characters.

## INTRODUCTION

The genus Gladiolus belongs to the family Iridaceae and includes about 270 species. This genus is native to different regions of South Africa, tropical Africa and the Mediterranean area, in the first region having a much larger distribution with a larger number of species (more than 114 species). In the Mediterranean area, the ten to twelve species of gladiolus offered taxonomic confusion due to their similarities and morphological variations due to localized evolution. This led to several descriptions of Mediterranean species, which were later treated by many authors as synonymous or placed in lower taxonomic ranks (Hamilton, 1980, quote de Mifsud et al., 2013).

The European species were cultivated at least 500 years ago and were first identified as a species in New Forest, Great Britain, in 1855. Trade in these species evolved in a very short time and occupied the top places. in the international trade in flowers (Cantor & Tolety, 2011).

As an underground organ, the *Gladiolus* species have a corm, that is renewed with each cycle of vegetation and flowering. Under conditions of humidity and appropriate temperature one can form up to 4-5 new corms, at the base of which are formed small cormlets (3-8 mm diameter), capable of flowering after 2-3 years (Şelaru, 2007). The stems are generally unbranched and the flower spikes are one-sided, with bisexual flowers, each subtended by two green bracts. Tepals are united at their base into a tube-shaped structure.

In 2010 a study was conducted in the Floriculture experimental field ("Ion Ionescu de la Brad" University of Agriculture Sciences and Veterinary Medicine of Iași) shows that the species *G. imbricatus* has a good adaptability to environmental conditions in this area, noting insignificant differences from the specimens in the natural habitat (decrease in plant height, a low delay in flowering by about 3-7 days), the species covering almost the same decorative period as in the natural habitat, decorating by flowers from May to June (Chelariu & Draghia, 2011).

When the temperatures are high and rainfall is reduced quantitatively, the number of days required for corms formation is about 45 to 50 (Tomiozzo et al., 2019). The crop success (the vegetative growth, development and, finally, the height of the flower stem, the number of flowers and the production of tubers) is strongly influenced by the size of the corms (Bose et al., 2003 and Uddin et al., 2002, quoted by Sarkar et al., 2014).

The size of the corms and the planting distance directly influence the vegetation start of the plants, their growth, as well as the formation of the stems and the number of flowers on the stem (Toporaş, 2008). Plant height, number of leaves/ plant, time required for flowering, flower stem length, number and size of new corms and cormlets were significantly improved by the use of larger parent corms (Sharma & Gupta, 2003).

Determining the assimilative pigments content in the process of plant growth and development provides useful information on the photosynthetic state during the phenophases of vegetation. Ecological conditions of culture and especially water stress reduce the photosynthetic rate per unit area of leaves (Graca et al., 2010). In addition, Abo El-Kheir (2007) indicated that the reduction of the available moisture content in the soil leads to both a significant decrease in the concentration of Chl. a, Chl. b; the total content of chlorophyll pigments (a + b), as well as the content of carotenoid pigments. At gladiolus, the increase in photosynthetic pigments content is highlighted as a response of plants to foliar application of antioxidants and is due to their role in strengthening the photosynthetic activity and the chlorophyll biosynthesis, or protecting the chloroplast from oxidative damage resulted from oxidative stress (Munne-Bosch et al., 2001).

The content of assimilative pigments in the leaves of *G. grandiflorus* is significantly influenced not only by ecological conditions, vegetation phenophase but also by organic fertilizer (Hassan et. al., 2020; Abdou et al., 2013; Khalil, 2015; Abdou et al., 2018, 2019). Also, ascorbate peroxidase (APX) and catalase (CAT) are enzymes that metabolize  $H_2O_2$  caused by stress and control the potential impact to maintain the cellular concentration of  $H_2O_2$  at a level necessary for the normal growth and development of plants (Gill & Tuteja, 2010; Ray et al., 2012; Anjum et al., 2014b).

This paper presents an analysis of the ornamental characters and also the content of photosynthetic pigments and the enzymatic activity of two *Gladiolus* species (*G. byzantinus* and *G. imbricatus*), growing in conditions of the North East region of Romania (Iasi city).

# MATERIALS AND METHODS

The observations and measurements were performed during 2018-2020. in the experimental field of the Floriculture discipline, "Ion Ionescu de la Brad" University of Life Sciences from Iasi. The laboratory performed within analyzes were the Horticultural Research Center of the Faculty of Horticulture from Iasi.

The experimental plot is located in temperatecontinental climat with excessive nuances. During the analyzed period, the average annual temperatures registered a slight increase, starting from an average of 10.5°C in 2018 and reaching the value of 12.0°C in 2020. The rainfall regime varied a lot during the experiment. The highest amount of precipitation was recorded in 2020 and the driest year was 2019.

Soil is a chernozem cambic with sandy-loam texture, with pH 7.8

The establishment of the crops was made on October-November, with corms having a mass between 5.06 and 3.5 g from the species *G. byzantinus* and *G. imbricatus* from the own collection of the Floriculture discipline. The corms were planted in rows, at a depth of 10 cm in a very well prepared substrate. The planting distances are 25-30 cm between rows and 10-15 cm distance between plants per row (Cantor & Pop, 2008).

The *G. byzantinus* (Figure 1a) presents as an underground organ an ovoid corm, covered with fibrous tunics. The leaves, 4-5 number, are ensiform and can reach a height of 70 cm, 1-2.5 cm wide. The floral stem is erect and reaches 0.5-1 m in height; presents 5-15 flowers, brightly colored in red-purple, very beautiful, arranged unilaterally, less bilaterally, often unbranched, blooming in May-June (Kerguélen & Lonchamp 1999).

The *G. imbricatus* (Figure 1b) is a rare, legally protected perennial species protected at European level by Directive 92/43 / EC (Annex I) of 21 May 1992 (on the conservation of natural habitats and of wild fauna and flora) (https: / /ro.wikipedia.org/wiki/F%C3%A2nea% C8%9Ba Izvoarelor Cri%C8%99ul Pietros).

As an underground organ it has globular corms. The stem is erect, unbranched and cylindrical. It is 30-60 cm tall and usually has three lanceolate leaves.

The inflorescence is a unilateral racemose crown and has 4 to 12 zygomorphic flowers, 2.5 cm long, with purple-violet tepals (Frantsuzenok & Nikonovich, 2005). The fruit is a short, obovate, obtuse, trimmed capsule (Brickell & Cathez, 2004). It grows on wet meadows and swamps and blooms from May to June. In Romania it is found in the meadows of the sub-Carpathian regions (Păun et al., 1980) and in the spontaneous flora of Maramureş, Transylvania, Banat, Maramureş, Oltenia, Muntenia, Moldova. In the Iasi area it was reported in the Bârnova forest (Oprea, 2005).



a. b. Figure 1. *G. byzantinus* (a) și *G. imbricatus* (b) (original photo)

The experience included two variants (each species representing a variant) distributed in randomized blocks with three repetitions (20 plants/repetition).

The examination of the morphological characters and phenology was made on the plants from the collection.

The results were compared to the average of the variants (considered control), and the interpretation was made using the analysis of the variance, with the LSD test (Saulescu and Saulescu, 1967).

The symbols used to indicate the significance of the differences from the control are: ns = nonsignificant; o/x = negative/positive significantdifference; oo/xx = negative/positive distinct significant difference; ooo/xxx = negative/positive very significant difference. The assimilative pigments were tested by the spectrophotometric method according to Lichtenthaler (2001). The extraction of photosynthetic pigments was performed from the leaves of the studied species according to the Current Protocols in Food Analytical Chemistry (Hartmut et al., 2001). To extract the photosynthetic pigments, fresh material was 0.03-0.05 weighed (between g). After weighing, the samples were placed in a grinding mortar and then quartz sand and Ca CO<sub>3</sub> (in powder form) were added to prevent the conversion of chlorophylls to porphyrins. To grind the tissue, 2-3 ml of pure acetone is added to the mortar in several stages until the vegetable material has been well ground, after which the liquid has been passed in a graduated cylinder. The process was repeated until the acetone was no longer colored, the volume of the filtrate was finally brought to 10 ml and then centrifuged for 10 minutes at 10,000 rpm. After extraction, the extracted samples were read using the UV-VIS spectrophotometer at E661.6 for a chlorophyll, E644.8 for b chlorophyll and E470 for carotenoid pigments. Sampling was performed using the T70 UV/VIS Spectrophotometer PG.

To determine the activity of catalase (CAT) and ascorbate peroxidase (APX), 0.5 g of plant material was ground on ice in K phosphate buffer with a pH of 7.0. To obtain the supernatant from which the enzymatic activity was analyzed, the extract obtained from milling was centrifuged at 12,000 rpm for 20 minutes, at 4°C. The APX activity was determined according to the method of Chen & Asada (1989) by monitoring the decrease in absorbance at 290 nm. The reaction mixture (3 ml) consisted of 1.5 ml of phosphate buffer (pH 7.0), 300 µl of ascorbic acid, 600 µl of H2O2 and 600 µl of enzyme extract. One unit of enzyme activity was calculated as the amount of enzyme required for the oxidation of 1.0 mM ascorbate/min/g fresh substance. The total activity of CAT was tested by measuring the initial rate of H<sub>2</sub>O<sub>2</sub> disappearance according to the method of Aebi (1984). The reaction mixture (1 ml) contained 1.5 ml of phosphate buffer (pH 7), 1.2 ml of H<sub>2</sub>O<sub>2</sub> and 300 µl of enzyme extract. The decrease in absorbance was monitored over time, at 240 nm. One unit activity (units/min/g fresh of enzymatic

substance) is calculated as the amount of enzyme needed to release half of the oxygen peroxide from  $H_2O_2$ . Experiments were performed in triplicate and data for APX and CAT were presented as mean values with standard deviations.

### **RESULTS AND DISCUSSIONS**

In the Figure 2 are presents data on the share of corms started in vegetation. In the species *G. byzantinus* the percentage of corms started in vegetation is 89.0%, with 2.5% lower than in *G. imbricatus*, in which the share of starting corms in vegetation is 91.5%. Regarding the number of plants that formed floral stems, the observations showed that the plants of *G. imbricatus* formed floral stems in proportion of 97,2% and in *G. byzantinus* only 59.6% of the plants started in vegetation formed stems.



Figure 2. The share of corms in the vegetation and of the plants that formed floral stems

According to the calendar data that marked the main phenophases: the start of vegetation, the appearance of flower stalks, the onset of flowering and the end of flowering, the time required for each of them was calculated. From Figure 3 it can be seen that the number of days from planting to the start of vegetation is 132 for *G. byzantinus* and 151 for *G. imbricatus* (Figure 3).

It was found that the number of days from the onset of vegetation to the appearance of the flower stem is equal between the two species, however the number of days calculated from the appearance of the stem to the opening of the first flower in the inflorescence varied from 14 days at *G. byzantinus* and seven days at *G. imbricatus* (Figure 3).

The duration of flowering, expressed by the number of days from the opening of the first flower to the complete passage of flowers is longer for the species *G. imbricatus*, totaling a number of 16 days compared to 13 days for the species *G. byzantinus* (Figure 3).



Figure 3. Duration of the main phenophases (no. of days)

The aspects related to the phenology of the two species are also shown in the diagram, in Figure 4, from which it can be seen that *G. byzantinus* starts in vegetation and blooms two weeks earlier. but has a shorter flowering time by approx. seven days compared to *G. imbricatus*.



Figure 4. Phenological diagram

In the Tables 2, 3 and 4 were present data on some morphological properties of plants from the two species of *Gladiolus*. The length of the

leaves varied from 35.5 cm (G. byzantinus) to 56.3 cm (G. imbricatus). Very significant positive differences were recorded in the species

*G. imbricatus*, in which the leaf length exceeded by 22.3% the average of the experiment and by 6.8% *G. byzantinus* (Table 2). The width of the leaves had similar values for the two species (approx. 2 cm) and the differences were non significant. Regarding the number of shoots per plant (Table 1), higher values were at *G. byzantinus* (1.6 shoots per plant, compared to *G. imbricatus* which formed only one shoot per plant).

		Leaf leng	gth	Number of shoots / plant			
Species	Absolute values (cm)	Relative values (%)	Difference/ Significance	Absolute values (no.)	Relative values (%)	Difference/ Significance	
G. byzantinus	35.5	77.34	-10.4000	1.6	123.08	0.3 <sup>x</sup>	
G. imbricatus	56.3	122.66	10.4 <sup>xxx</sup>	1	76.92	-0.30	
$\bar{x}$	45.9	100.0	100.0 0		100.0	0	
		4 $LSD_{5\%} = 0.3$					
			$LSD_{1\%} = 4.1$	1	$LSD_{1\%} = 0.5$		
			$LSD_{0,1\%} = 7$	LSD <sub>0.1%</sub> = $1.0$			

The height of the floral stem is a very appreciated character, especially in the case of capitalizing the plants as cut flowers. From the statistical analysis of the data on the height of the floral stems, the longer stems were highlighted in *G. imbricatus*, which exceeded the average of the experience by 11,65%, respectively with significant positive differences (Table 2).

Table 2. The biometric features of floral stems and inflorescences

	Floral stem height			Height to the first flower			Inflorescence length		
Species	Absolute values (cm)	Relative values (%)	Diff./ signif.	Absolute values (cm)	Relative values (%)	Diff./ Signif.	Absolute values (cm)	Relative values (%)	Diff./ signif.
G. byzantinus	61.4	88.35	-8.100	38.8	83.44	-7.800	22.7	98.69	-0.3 <sup>ns</sup>
G. imbricatus	77.6	111.65	8.1 <sup>xx</sup>	54.3	116.77	7.8 <sup>xx</sup>	23.3	101.75	0.3 <sup>ns</sup>
$\bar{x}$	69.5	100.0	0	46.55	100.0	0	23.0	100.0	0
	$LSD_{5\%} = 4.4$ $LSD_{1\%} = 7.2$			$LSD_{5\%} = 2.7$ $LSD_{1\%} = 4.4$			$LSD_{5\%} = 1.4$ $LSD_{1\%} = 2.3$		
	$LSD_{1\%} = 7.2$ $LSD_{0.1\%} = 13.5$			$LSD_{1\%} = 8.3$			$LSD_{1\%} = 4.3$		

The first basal flower in the inflorescence was located at a height of approx. 39 cm above ground level in *G. byzantinus* and over 54 cm in *G. imbricatus* (Table 2).

Although the values indicating the length of the inflorescences are not statistically assured (Table 2), the differences between the two species being very small, it should be noted the share of the inflorescence in the total floral stem (37% in *G. byzantinus* and almost 30% in *G. imbricatus*) (Figure 5).

An important element for the both use of these species in landscaping and for their use as cut flowers is the number of flowers in the inflorescence.



Figure 5. The length of the inflorescence in the floral stem (%)

Also in this character, G. imbricatus had better results, respectively 12 flowers/inflorescence (with 21.21% above the average of the experience), compared to 7.8 flowers/ inflorescence in G. *byzantinus*. The differences from the mean were significant (positive in G. *imbricatus* and negative in G. *byzantinus*).

From the data presented above (Table 3) it was found that the length of the inflorescences was relatively close to the species analyzed. However, the difference was in the density of the inflorescences, determined by the distance between the flowers (Table 3). Thus, *G. byzantinus* was characterized by looser inflorescences, with a distance between flowers of 3.7 cm, unlike *G. imbricatus*, with denser inflorescences, the distance between flowers being 1.7 cm, with over 50% lower (Table 3).

	1	No. flowers/infl	orescence	The distance between the flowers in the inflorescence			
Species	Absolute values (no.)	Relative values (%)	Difference/ Significance	Absolute values (cm)	Relative values (%)	Difference/ Significance	
G. byzantinus	7.8	78.79	-2.100	3.7	137.04	1 <sup>xx</sup>	
G. imbricatus	12	121.21	2.1 <sup>xx</sup>	1.7	62.96	-100	
x	9.9	100.0	00.0 0		100.0	0	
	$LSD_{5\%} = 1.3$ $LSD_{5\%} = 0.5$						
		$\begin{array}{llllllllllllllllllllllllllllllllllll$					

Table 3. Flowers and inflorescences characteristics

It is known that photosynthetic activity is related to the content of photosynthetic pigments (Macintyre et al., 2002). Chlorophyll, which captures light and transfers energy in order to conduct photochemical reactions, is one of the most active photochemical compounds in photosynthesis. Photosynthetic efficiency and cell growth are associated with the quantifycation of chlorophyll and have been highlighted on algae cultures's systems (Masojidek et al., 2000; Tremblin et al., 2000).

Conventionally, chlorophyll content is measured using spectroscopic and chromatographic methods (Lichtenthaler & Wellburn, 1983; Gilmore & Yamamoto, 1991).

Photosynthetic pigments represented bv chlorophylls and carotenoids are those that provide valuable information on the installation of physiological stress in plants in different culture conditions. The values of chlorophyll content in leaves are closely correlated with the ecological conditions, the stress caused by high temperatures and drought, inducing a decrease in the total content of photosynthetic pigments. In addition to decreasing of the total content of photosynthetic pigments, this stress also induces an increase in the content of carotenoid pigments in the leaves.

In order to highlight the growth and development of the G. *imbricatus* and G. *byzantinus* species under the culture

conditions of Iasi during research period 2018-2020, the content of photosynthetic pigments was analyzed by the spectrophotometric method. In order to obtain conclusive results, the harvesting of the vegetal material was carried out during the flowering period of the plants and after the end of the flowering. The samples were collected at the same time of the day these being represented by leaves that have reached maximum maturity. After determinations were made, in both phenophases of vegetation, was observed a higher content of assimilating pigments in G. imbricatus species compared to G. byzantinus species (Table 4). The results regarding the total content of photosynthetic pigments showed higher values during the flowering period of the plants at both species. The total content of photosynthetic pigments increased in the flowering period of the plants compared with the post-flowering period by 0.29 mg/g s.p. at *G. imbricatus* and by 0.71 mg/g s.p. at the *G. byzantinus*. Studies showed that under normal ecophysiological conditions the ratio of chlorophyll *a*/chlorophyll b is around 3:1 (Lichtenthaler et al., 1981). The results of chlorophyll pigments content as well as the values of the Chl. a/ Chl. b ratio can provide useful information on the interaction between plants and the environment (Richardson et al., 2002). The values of this ratio were within the theoretical limits, ranging between 2.93 at *G. imbricatus* and 2.58 at *G. byzantinus*. Was observed, in case of both species *G. imbricatus* and *G. byzantinus*, an increase in the total content of carotenoid pigments after the flowering of the plants. Within this phenophase, by correlating the decrease of the values of the photosynthetic pigments content with the increase of the carotenoid pigments content, it is highlighted that the development of physiological processes in plants is manifested normally, the data confirming the results according to which the

content of chlorophyll pigments generally decreases during the installation of senescence (Fang et al., 1998). The ratio of chlorophyll pigments/ carotenoids was higher for the both species during the flowering period of the plants (3.75)in G. imbricatus and 4.11 in G. byzantinus). The increase in the content of carotenoid pigments in the period after the flowering of the plants determined a decrease of the chlorophyll / carotenoid pigments ratio, the values obtained by the two species being of 3.66 for G. imbricatus and 3.88 for G. byzantinus.

Table 4. Average content of photosynthetic pigments (mg g<sup>-1</sup> F.W)

Species	Phenophase	Chl. <i>a</i> mg/g F.W.	Chl. <i>b</i> mg/g F.W.	x+c mg/g F.W.	Σ	Chl.a/ Chl.b	Chl./Car.
G. imbricatus	flowering	2.94±0.03	$1.06\pm0.02$	$1.07 \pm 0.05$	5.06	2.77	3.75
	after flowering	2.79±0.02	0.95±0.04	$1.02\pm0.02$	4.77	2.93	3.66
G. byzantinus	flowering	2.77±0.04	0.94±0.02	0.90±0.03	4.62	2.94	4.11
	after flowering	2.24±0.03	$0.87 \pm 0.05$	$0.80{\pm}0.04$	3.91	2.58	3.88

Studies have shown that there was an increase in ascorbate peroxidase (APX) activity along with the activity of other enzymes in the case of plants where have been subjected to stress conditions (Shigeoka et al., 2002). APX can also regulate redox signaling pathways involved in plant development (Caverzan et al., 2012), and transcriptional expression of APX is dependent on the stage of plant or tissue development (Agrawal et al., 2003; Teixeira et al., 2006). Changes in the activity of antioxidant enzymes such as APX, superoxide dismutase (SOD) and catalase (CAT) have been described during senescence period of the plant and are related to cleansing processes (Alaey et al. 2011, Gerailool & Ghasemnezhad, 2011). There are several reports of antioxidant defense systems at ornamental plants, such as chrysanthemum (Bartoli et al., 1997), daylilies (Panavas & Rubinstein, 1998) and gladiolus (Ezhilmathi et al., 2007; Sairam et al., 2011).

In order to study the species *G. imbricatus* and *G. byzantinus* in the ecological conditions of Iaşi from 2018-2020, biochemical determinations were performed regarding the enzymatic activity of the leaves. During the flowering and after flowering, determinations were made regarding the enzymatic activity of ascorbate peroxidase (APX) and catalase (CAT).

The results obtained regarding APX at the two species for the two developmental phenophases were compared both with each other and with the average. The difference regarding the increase of ascorbate peroxidase's activity can be considered a practical measure for assessing the level of stress caused by ecological conditions or by the development of physiological processes which are specific to the phenophase of vegetation.

By comparing the results, the highest increases of APX activity was observed at *G. byzantinus* species (the average content being 567.59 UP/g/min FW during flowering and 629.86 UP/g/min FW after flowering). Lower values of APX were registred on *G. imbricatus* species (average content being 525.93 UP/g/min FW during flowering and 567.28 UP/g/min FW after flowering) (Figure 6).



Figure 6. Ascorbate peroxidase activity determined in leaves of *G. imbricatus* and *G. byzantinus* [(U/g/min) fresh substance]

The determinations performed during after flowering period showned the APX activity increased with 41.35 UP/g/min F.W. at *G. imbricatus*, and with 62.27 UP/g/min F.W. *G. byzantinus* species in comparation with the values obtained during flowerin period. The results obtained for the two species in the two vegetation periods were compared with the average value of the results. The results obtained showed againste the average an increase in APX only in the case of *G. byzantinus* species after flowering when the percentage was 9.98%.

The increase of APX activity in the postflowering period does not indicate an abiotic stress caused by the culture conditions but highlights the development of physiological processes depending on the vegetation phenophase.

The biochemical results obtained in the case of the two species were correlated with the biometric measurements and the phenological determinations, highlighting a good development of these species at the climatic conditions in Iasi from 2018-2020.

Regarding the results of catalase activity (CAT) by comparing the results, the same trend was highlighted as in the case of APX (Figure 7). The highest increases in CAT activity were highlighted in the species *G. byzantinus* in both vegetation phenophases. Within the two species, the lowest values of CAT activity were obtained in *G. imbricatus*, the average content being 79.14 UP/g/min F.W. at flowering and 89.03 UP/g/min F.W. (Figure 7).



Figure 7. Catalase activity determined in leaves of *G. imbricatus* and *G. byzantinus* [(U/g/min) fresh substance]

Compared to the average value of CAT obtained by the two species, during the flowering

period, the decrease was 10.26% in *G. imbricatus* and 2.41% *G. byzantinus*. Within the two species, significant increases of CAT compared to the average were registred only by the species *G. byzantinus* in the post-flowering period, the values presenting increases compared to the average by 11.72%.

# CONCLUSIONS

In the ecological conditions of Iasi, the two species of gladiolus studied have distinctive ornamental characters and can be used in various ways.

*G. byzantinus* formed denser shrubs (higher number of shoots per plant).

*G. imbricatus* there were more leaves/ shoot, longer flower stalks, denser inflorescences (shorter distance between flowers).

*G. byzantinus* bloomed about two weeks earlier, but flowering time was seven days longer in *G. imbricatus*.

The content of assimilating pigments was higher in *G. imbricatus* and lower in *G. byzantinus*.

In both species, the content of photosynthetic pigments was higher in the case of determinations made during the flowering period. The chlorophyll *a* / chlorophyll *b* ratio had values above 2.5, which suggests that the plants showed a normal growth and development from a physiological point of view. The increase of the content in carotenoid pigments from the period after flowering caused the decrease of the chlorophyll pigments/ carotenoids ratio, the values obtained by the two species were of 3.66 for G. imbricatus and 3.88 for G. byzantinus.

The enzymatic activity of both ascorbate peroxidase (APX) and catalase (CAT) recorded higher values especially in the samples colected in the post-flowering period. The results obtained in *G. byzantinus* showed increases compared to the average by 9.98% of APX and 11.72% of CAT.

The results regarding the enzymatic activity are correlated with those of the photosynthetic pigments content, the specie that showed the increase of the enzymatic activity (*G. byzantinus*) also registered the decrease of the chlorophyll pigments content.

*G. byzantinus* and *G. imbricatus* can be used both as cut flowers and in landscaping.

#### REFERENCES

- Abdou, M.A.H., Aly M.K., & Ahmed, A.S.A. (2013). Effect of compost, biofertilization and some vitamins addition on *Gladiolus grandiflorus*. J. Plant Production, Mansoura Univ., 4(12): 1751-1761.
- Abdou, M.A.H., Badran, F.S., Ahmed, E.T., Taha, R.A., & Abdel-Mola, M.A.M. (2018). Effect of compost and some natural stimulant treatments on: II. Corms production and chemical constituents of (*Gladiolus* grandiflorus cv. Peter Pears) plants. Proc. of The 4th Conf. of SSFOP, Scientific J. Flowers & Ornamental Plants, 5(2):115-126.
- Abdou, M.A.H.; Aly, M.K., & Ahmed, A.S.A. (2019). Influence of organic manure, biofertilizer and/or some vitamin treatments on: A. Vegetative growth and flowering aspects of *Gladiolus grandiflorus* var. Gold field plants. *Scientific J. Flowers & Ornamental Plants*, 6(2):113-124.
- Abo, El-Kheir, M.S.A., & Mekki, B.B. (2007). Response of maize single cross-10 to water deficits during silking and grain filling stages. *World J. Agric. Sci.*, 3(3): 269-272.
- Aebi, H. (1984). Catalase in Vitro. Methods in Enzymology 105: 121-126. DOI: 10.1016/S0076-6879(84)05016-3.22. S. WANG, H. LI, C. LIN, 2013, Physiological, biochemical and growth responses of Italian ryegrass to butachlor exposure. *Pesticide Biochemistry and Physiology*, 106, 21-27.
- Agrawal, GK., Jwa, NS., Iwahashi, H., & Rakwal, R. (2003) Importance of ascorbate peroxidases OsAPX1 and OsAPX2 in the rice pathogen response pathways and growth and reproduction revealed by their transcriptional profiling. *Gene*, 322:93–103.
- Alaey, M., Babalar, M., Naderi, R., & Kafi, M. (2011). Effect of pre- and postharvest salicylic acid treatment on physio-chemical attributes in relation to vase-life of rose cut flowers. *Postharvest Biol. Technol.* 61: 91-94.
- Andréia Caverzan, Gisele Passaia, Silvia Barcellos Rosa, Carolina Werner Ribeiro, Fernanda Lazzarotto, & Márcia Margis-Pinheiro (2012). Plant responses to stresses: Role of ascorbate peroxidase in the antioxidant protection. Genet Mol Biol. 35 (4 Suppl): 1011–1019.doi: 10.1590/s1415-47572012000600016.
- Bartoli, C.G., Simontacchi, M., & Montaldi, E.R. (1997). Oxidants and antioxidants during aging of chrysanthemum. *Plant Sci*, 129: 157-165.
- Cantor Maria, & Ioana Pop (2008). Floricultură, Baza de date, Ed. Todesco, Cluj Napoca.
- Cantor Maria, & Tolety (2011). Wild crop relatives: Genomic și Breeding Resources. Planting and Ornamental Crop, Chapter Gladiolus. Ed. Springer, Berlin, Germania, page 133-159.
- Chelariu Elena-Liliana, & Lucia Draghia (2011). Studies Regarding the Behaviour in Crop Conditions of Some Species from Ornamental Flora, with Decorative Value, *Bulletin UASVM Horticulture*, 68(1).
- Chen, GX., & Asada, K. (1989). Ascorbate peroxidase in pea leaves: occurrence of two isozymes and the differences in their enzymatic and molecular

properties. *Plant and Cell Physiology 30*: 987-998. DOI: 10.1093/oxfordjournals, pcp. a077844.

- Ezhilmathi, K., Singh, V.P., Arora, A., & Sairam, R.K. (2007). Effect of 5-sulfosalicylic acid on antioxidant activity in relation to vase life of Gladiolus cut flowers. *Plant Growth Regul*, 51: 99-108.
- Fang, Z., Bouwkamp, J., & Solomos, T. (1998). Chlorophyllase activities and chlorophyll degradation during leaf senescence in non-yellowing mutant and wild type of *Phaseolus vulgaris* L, *Journal of Experimental Botany*, vol. 49, pg. 503-510.
- Gerailool, S., & Ghasemnezhad, M. (2011). Effect of salicylic acid on antioxidant enzyme activity and petal senescence in 'Yellow Island' cut rose flowers. *J. Fruit Ornamental Plant Res*, 19: 183-193.
- Gilmore, A.M., & Yamamoto, H.Y. (1991). Resolution of lutein and zeaxanthin using a non endcapped, lightly carbon-loaded C-18 high-performance liquid chromatographic col. J Chromatogr, 543:137–145.
- Graca, J.P., Rodrigues, F.A., Forias, J.R.B., Oliveira, M.C.N., Hoffmann-Campo, C.B., & Zingaretti, S.M. (2010). Physiological Parameters in sugarcane cultivars submitted to water stress. *Brazilian J. Plant Physiology*, 22 (3): 189-197.
- Hamilton, A.P. (1967). The discovery and present status of Gladiolus illyricus, Koch., in Britain. *Journ. Durham Univ. Biol. Soc.*, 13, 1-4.
- Hartmut, K., Lichtenthaler, H.K., & Buschmann, C. (2001). Extraction of Phtosynthetic Tissues: Chlorophylls and Carotenoids, *Current Protocols in Food Analytical Chemistry*, vol. 1, pg. F4.2.1-F4.2.6.
- Hassan, A.A., & Abd El-Azeim, M.M. (2020). Impacts of compost, biofertilizer and/or some antioxidant treatments on gladiolus (*Gladiolus grandifloras*), *Scientific J. Flowers & Ornamental Plants*, www.ssfop.com/journal ISSN: 2356-7864 doi: 10.21608/sjfop.2020.11456.
- Kerguélen, M., & Lonchamp, JP. (1999). Index synonymiquede la flore de France, http://www.inra.fr/flore-france.
- Khalil, A.R.M. (2015). Physiological Studies on Gladiolus Plant. M.Sc. *Thesis, Fac. Agric. Minia* Univ., 146 p.
- Lichtenthaler, H.K., & Wellburn, A.R. (1983). Determination of total carotenoids and chlorophyll a and b of leaf extract in different solvent. *Biochem Soc Trans* 603:591–592.
- Macintyre, H.L., Kana, T.M., Anning, T., & Geider, R.J., (2002). Photoacclimation of photosynthesis irradiance response curves and photosynthetic pigments in microalgae and cyanobacteria. J Phycol 38:17–38.
- Masojidek, J., Torzillo., G., Koblizek., M., Nidiaci., I., Komenda, J., Lukavska, A., & Sacchi, A. (2000). Changes in chlorophyll fluorescence quenching and pigment composition in the green alga *Chlorococcum* sp. grown under nitrogen deficiency and salinity stress. J Appl Phycol 12:417–426.
- Mifsud, S., & Hamilton, A.P. (2013). Preliminary observations from long-term studies of *Gladiolus L*. (Iridaceae) for the Maltese Islands, *Journal of Plant Taxonomy and Geography*, Volume 68, Issue 1.

- Munne-Bosch, S., Scharz, K., & Algere, L. (2001). Water deficit in combination with high solar radiation leads to midday depression of α-tocopherol in field grown lavender (*Lavandula stoechas*) plants. *Aust. J. Plant Physiol.*, 28:315-321.
- Oprea, A. (2005). Lista critică a plantelor vasculare din România. Editura Universității "Al. I. Cuza", Iași.
- Panavas, T., & Rubinstein, B. (1998). Oxidative events during programmed cell death of daylily (*Hemerocallis* hybrid) petals. *Plant Sci.* 133: 125-138.
- Regina Tomiozzo, Uhlmann, L.O., Camila Coelho Becker, Natalia Teixeira Schwab, Nereu Augusto Streck, N.A., & Balest, D.S. (2019). How to produce gladiolus corms. *Technical Article Ornam. Hortic*, 25 (3), https://doi.org/10.1590/2447-536X.v25i3.2048.
- Richardson, A.D, Duigan, SP., & Berlyn, G.P. (2002). An evaluation of non-invasive methods to estimate foliar chlorophyll content, *New Phytologist, vol. 153* (pg. 185-194).
- Sairam, R.K., Vasanthan, B., & Arora, A. (2011). Calcium regulates Gladiolus flower senescence by influencing antioxidative enzymes activity. *Acta Physiol. Plant.* 33: 1897-1904.
- Sarkar, M.A.H., Hossain, M.I., Uddin, A.F.M.J., Uddin, M.A.N., & Sarkar, M.D. (2014). Vegetative, floral and yield attributes of gladiolus in response to gibberellic acid and corm size, *Sci. Agri.* 7 (3), 142-146, DOI: 10.15192/PSCP.SA.2014.3.3.142146.
- Săulescu N.A., & Săulescu, N.N. (1967) Câmpul de experiență. Ed. Agro-Silvică, București.

- Șelaru Elena (2007). *Cultura florilor de grădin*ă. Editura Ceres, București.
- Sharma, J.R., & Gupta, R.B. (2003). Effect of corm size and spacing on growth, flowering and corm production in gladiolus, *Journal of Ornamnetal Horticulture*, Volume 6, issue 4, ISSN 2249-880X.
- Teixeira, F.K., Menezes-Benavente, L., Galvão, V.C., Margis, R., & Margis-Pinheiro M. (2006) Rice ascorbate peroxidase gene family encodes functionally diverse isoforms localized in different subcellular compartments. *Planta*, 224:300–314.
- Toporaș Maria (2008). Comportarea unor tipuri și soiuri noi de gladiolă în condiții ecologice specifice și optimizarea producerii materialului săditor, Universitatea de Științe Agronomice și Medicină Veterinară București, Facultatea de Horticultură.
- Tremblin, G., Cannuel, R., Mouget, J.L., Rech, M., & Robert, J.M. (2000). Change in light quality due to a blue-green pigment, marennine, released in oysterponds: effect on growth and photosynthesis in two diatoms, *Haslea ostrearia* and *Skeletonema costatum*. J Appl Phycol 12:557–566.
- Uhlmann, L.O., Camila Coelho Becker, Regina Tomiozzo, Streck, N.A., Schons, A., Balest, D.S., Mara dos Santos Braga, Natalia Teixeira Schwab, & Josana Andreia Langner (2019). Gladiolus as an alternative for diversification and profit in small rural property, Ornam. Hortic. 25 (2) https://doi.org/10.14295/oh.v25i2.1541.
- https://ro.wikipedia.org/wiki/F%C3%A2nea%C8%9Ba\_ Izvoarelor\_Cri%C8%99ul\_Pietros.