

# Influence of spring and autumn frosts on *Impatiens glandulifera* populations in the Sofia region (Bulgaria)

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## Abstract

The present study aims to analyze and determine to what extent spring and autumn frosts affect the population size and life cycle of this invasive alien species (IAS). The study area is located in the immediate vicinity of the capital Sofia and covers the Iskar River stream and its tributaries in the gorge between Plana and Lozenska Mountains (Southwestern Bulgaria). The study was carried out in the period 2020-2022. Six permanent sample plots (PSP), each measuring 10 m<sup>2</sup>, were established in characteristic localities of *I. glandulifera* populations along the Iskar River and its tributaries (Table 1). Each year, during the spring and autumn frosts, the percentage of dead individuals of *I. glandulifera* relative to the total number of individuals of this species in the PSP was recorded. During the autumn frosts, the number of alive individuals in the following phenological stages was also recorded: flowering; fruiting; dispersal of fruits and seeds; end of growing season. The results show that frosts in the study area did not significantly affect the population size and life cycle of *I. glandulifera*. The death of the whole population occurs only in permanent periods of negative daytime temperatures, however a significant part of the individuals at that time are in a generative stage of development. The temperature conditions in the area favor the invasiveness of *I. glandulifera*, extending the vegetation period of this species and the possibilities for its reproduction and spread.

## Keywords

Invasive alien plant species, frost, ecological factors, riparian habitats

## Introduction

*Impatiens glandulifera* Royle, or Himalayan balsam, is widely considered to be a highly problematic invasive alien species (IAS), having spread across more than thirty countries during the past century (Coakley, S., C. Petti 2021). Within its native range, Himalaya, this species is frost tolerant, and it was found at elevations of 1,800 to 4,000 m a.s.l. (Beerling and Perrins 1993). At the same time, studies of the distribution of this IAS in Europe show that *Impatiens glandulifera* is susceptible to frost, both in the early seedling stage and as a mature plant, potentially limiting its non-native distribution (GLANSIS, 2023).

In recent years, a number of studies related to the distribution, impact and ecological features of Himalayan balsam have been carried out in Bulgaria (Petrova et al., 2013, Kachova et al. 2020, Glogov et al, 2021, Glogov, Georgieva, 2021), but a specific study about the influence of frosts on the development of this species in the country has not been done so far. Only Georgieva (2021) touches on this problem in her study of the morphological traits and growth dynamics of *Impatiens glandulifera* in Bulgaria. The author confirms the data from foreign studies regarding the influence of temperature as a leading factor on the growth and spread of *I. glandulifera*.

The present study is focused on one of the country regions with the largest population of *Impatiens glandulifera* (Glogov, 2021). It aims to analyze and determine to what extent spring and autumn frosts affect the population size and life cycle of this IAS.

## Materials and methods

The study area is located in the immediate vicinity of the capital Sofia and covers the Iskar River stream and its tributaries in the gorge between Plana and Lozenska Mountains (Southwestern Bulgaria). The average altitude is 650 m. a.s.l. The maximum average monthly temperature is in July (21.5°C), and the minimum is in January (-1.5°C). The wind is strongest during the winter months (December to March) and its predominant direction is northwest. The maximum amount of precipitation is in the month of May, and the minimum - in December. The average annual amount of precipitation is 590.7 mm. The snow cover usually lasts from the end of November to the end of March.

The study was carried out in the period 2020-2022. Six permanent sample plots (PSP), each measuring 10 m<sup>2</sup>, were established in characteristic localities of *I. glandulifera* populations along the Iskar River and its tributaries (Table 1). Each year, during the spring and autumn frosts, the percentage of dead individuals of *I. glandulifera* relative to the total number of individuals of this species in the PSP was recorded. During the autumn frosts, the number of live individuals in the following phenological stages was also recorded: flowering; fruiting; dispersal of fruits and seeds; end of growing season (Pavlov, Dimitrov, 2012).

The locality types of *I. glandulifera* populations were determined according to the classification of Kachova et al. (2020). The names of the soils are according to the World Referent Base of Soil Resources (2014). The count of individuals in the juvenile phase in each of the PSPs was carried out in 5 sub-plots with dimensions of 1 m<sup>2</sup> each, after which the data were averaged for the whole PSP.

**Table 1.** Geographic data for each of the PSPs

PSP №	Coordinates	Altitude (m a.s.l.)	Locality type	Soils	Specificity
1	42°34'58.674"N 23°25'39.212"E	600	Constantly flooded riparian type	Fluvisols	along the Iskar River bank
2	42°34'44.555"N 23°25'34.075"E	610	Swamp on stagnant water	Histosols	swamp at 20m from the main stream of the Iskar River
3	42°34'2.076"N 23°25'48.594"E	650	Screes without direct contact with river running waters	Leptosols	forest scree 50m from the main stream of the Iskar River
4	42°33'27.642"N 23°25'53.594"E	610	Periodically flooded riparian type	Fluvisols	at the mouth of the Veleka River - the tributary of the Iskar River
5	42°30'27.873"N 23°30'29.057"E	650	Periodically flooded riparian type	Fluvisols	along the banks of the Okolska River - tributary of the main Iskar River
6	42°34'5.006"N 23°25'42.95"E	750	Constantly flooded riparian type	Fluvisols	along the Iskar River bank

## Results

The first seedlings of *I. glandulifera* in the study area appeared in mid-April, except for the populations in PSP3, where the conditions for the species are less favorable due to the distance from water and the primitive type of soils; and in PSP5, which is located at a higher altitude. For the period 2020-2022, late frosts were found from April 16 to 18 and first frosts from November 15 to 18 (Table 2) with an average duration of 2.4 days. The relative daily temperatures in the periods with frost in April 2020 have varied from -5°C to 17°C., and in April 2021 - from -4°C to 15°C. In 2022, no frosts were observed in the study area after April 6, when the specific species had not yet sprouted. The relative diurnal temperatures in the first autumn frost periods in the three years were similar and ranged from -5°C to 11°C. Quantitative data on changes in population numbers of *I. glandulifera* are presented in Table 2.

A statistically significant difference in population mortality from spring frost was found between the two years, being lower in 2021 (Table 3, Fig. 1 ). No statistically significant difference in autumn frost mortality was observed between the three years (Table 3, Fig. 2)

A comparison between spring 2020 and fall 2020 mortality (Fig. 3) showed no statistically significant difference ( $p=0.0503$ ), while a comparison between spring 2021 and fall 2021 showed a statistically significant difference ( $p=0.0054$ )

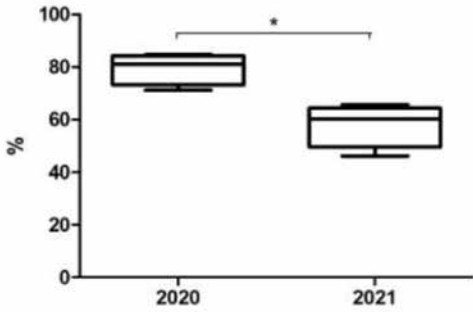
**Table 2.** Data on changes in the number of *I. glandulifera* individuals in PSPs as a result of the influence of spring and autumn frosts in the period 2020-2022.

PSP №	Average number of seedlings in the PSP	Period of spring frost	Period of autumn frost					
		16-18 April	15-18 November					
		Number of dead individuals	Average number of adults	Number of dead individuals	Number of live individuals in flowering stage	Number of live individuals in fruiting stage	Number of live individuals in stage of dispersal of fruits and seeds	Number of live individuals in the end of the growing season
2020r.								
1	131	111	44	31	2	2	4	5
2	112	93	28	19	1	1	5	2
3	97	0	31	24	0	0	2	5
4	118	84	41	26	0	3	6	6
5	105	0	37	22	0	2	4	9
6	101	80	33	15	1	2	7	8
2021r.								
1	90	59	33	25	1	1	3	3
2	78	36	16	10	0	0	2	4
3	90	0	29	17	0	3	3	6
4	92	55	36	29	0	1	3	3
5	126	0	44	14	1	9	9	11
6	66	40	19	15	0	0	3	1
2022r.								
1	109	0	30	16	1	1	6	6
2	99	0	19	16	0	0	0	3
3	104	0	32	22	0	0	4	6
4	131	0	38	18	0	0	7	13
5	114	0	49	35	1	1	3	9
6	80	0	27	12	0	4	4	7

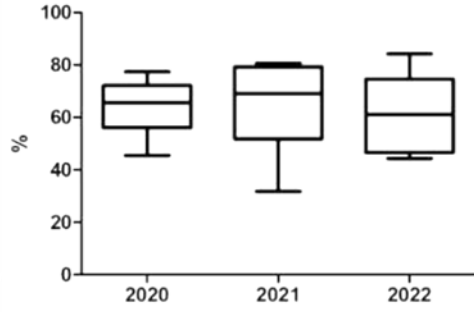
**Table 3.** Frost-induced mortality (mean  $\pm$  SD), %

Year	Spring	Autumn
2020	79,52 $\pm$ 6,01	64,03 $\pm$ 10,95
2021	58,05 $\pm$ 8,31	64,7 $\pm$ 18,45
2022	not applicable	61,58 $\pm$ 15,65

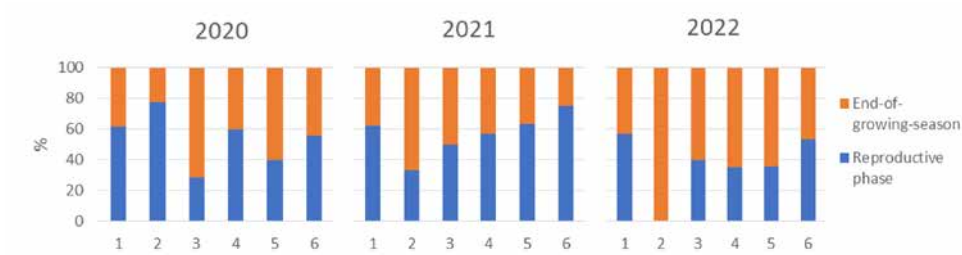
There was no statistically significant difference between the percentage of surviving individuals in the reproductive stage versus those in the end-of-growing-season stage in 2020 and 2021 (Figs. 4 and 5). In 2022, among the surviving individuals, those in the end-of-growing-season stage predominate, with a significantly lower number of surviving individuals in the reproductive stage being observed. (Fig. 4 and 6)



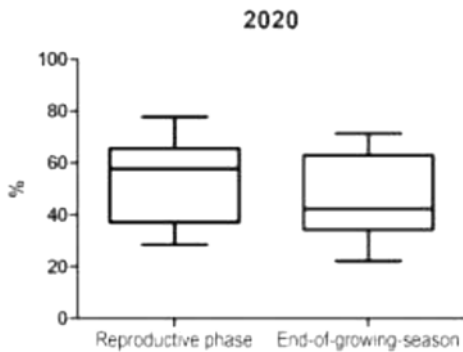
**Figure 1.** Comparison between frost-induced mortality rates in the spring of 2020 and 2021 in PSP No 1,2,4 and 6; Mann-Whitney U test,  $p=0,0286$ )



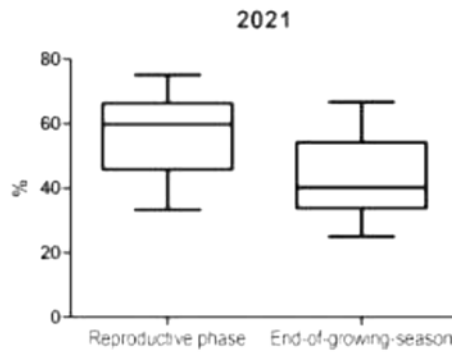
**Figure 2.** Comparison between frost-induced mortality rates in the autumn of 2020, 2021 and 2022 in PSP No 1,2,3,4,5 and 6) (paired t-test,  $p=0,9455$ )



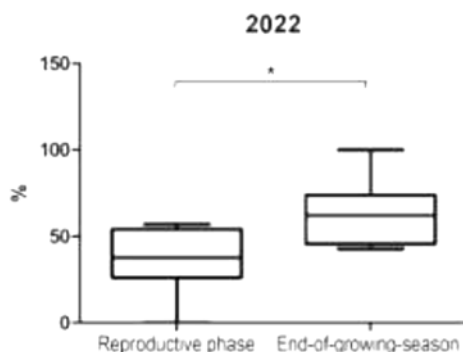
**Figure 3.** Comparison between mortality of individuals in autumn and spring populations



**Figure 4.** Ratio of the surviving individuals in the reproductive stage to those in the end-of-growing-season stage in the different PSPs during the three years.



**Figure 5.** Comparison of the surviving individuals in the reproductive stage versus those in the the end-of-growing-season stage for 2020 in all PSPs (unpaired t-test,  $p=0.4530$ )



**Figure 6.** Comparison of surviving individuals in the reproductive stage versus those in the the end-of-growing-season stage for 2021 in all PSPs (unpaired t-test,  $p=0.1233$ )

## Discussion

The results largely confirm the conclusions of previous studies that all life stages of this plant are frost intolerant (GLANSIS, 2023). In the frame of the studied periods during the day, temperatures rise and the frost does not persist, which is one of the reasons for the relatively high number of surviving individuals. Given the fact that it is theoretically sufficient for only one individual to reach the seed dispersal stage to restore and even increase the size of the entire population (Glogov, 2021), a conclusion can be drawn that these short-term frosts are not a decisive factor for the existence of *I. glandulifera* in the study area. The death of all the Himalayan balsam populations does not occur until after mid-November. In reality, the duration of the vegetation period of this annual species can reach 7 months, which is quite enough to ensure the smooth development and spread of its populations. The seeds have a long period of germination throughout April and the high mortality in late spring frosts is compensated by the emergence of new individuals.

The result that in the same population no significant difference was observed between mortality in spring and autumn may mean that the plant is not adaptive to the low temperature, because the same percentage of individuals that survived in spring to adulthood, died in the autumn. Field observations establish the beneficial influence of some additional factors on the survival of some individuals of *I. glandulifera* from the impact of frosts, such as proximity to water, which slows down the formation of frost. Water in plant cells acts as an insulator, cushioning the plant cell wall and protecting it from the damaging effects of a freezing night. Water also increases the soil's ability to retain heat from the sun, insulating the plant's roots and protecting them from harm (SF Gate, 2021). Another factor is the density of plant cover and the location of the individual and its degree of isolation from other plants in the PSP. Individuals found in denser communities survive frosts to a greater extent. The majority of plants killed by the frost had traces of generative organs, fruits and even flowers. The percentage of individuals in the end-of-growing-season stage is low. As a typical invasive species,

*I. glandulifera* uses every remaining day to develop its generative organs and produce new seeds.

It can be assumed that the end of the development of this species does not occur smoothly, but abruptly as a result of a sudden and permanent decrease in temperatures below 0°, which, according to our observations in the study area in the period 2020-2022, is registered after November 30, when the temperatures during the day vary from -7 to -2C°

## Conclusions

Frosts in the study area did not significantly affect the population size and life cycle of *I. glandulifera*.

The death of the whole population occurs only in permanent periods of negative daytime temperatures, however, a significant part of the individuals at that time are in a generative stage of development.

The temperature conditions in the area favor the invasiveness of *I. glandulifera*, extending the vegetation period of this species and the possibilities for its reproduction and spread.

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