Modeling the growth dynamics of water lettuce, *Pistia stratiotes* L. in wastewater

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Abstract: This study was aimed at assessing water lettuce (*Pistia stratiotes* L.) biomass growth, which was tested at the Faculty of Civil Engineering and Architecture of Niš under partially controlled conditions during a 70-day-long test, with a mixture of communal wastewater and water from the shaft at the hydraulic engineering demonstration facility as a source of nutrient matter. The biomass measured after the 70-day experiment ranged from 4.31 to 4.71 kg WW/m^2 (average 4.48 kg WW/m^2). The daily absolute growth rate (AGR) was 58.81 g/m^2 day, the daily increase rate (DIR) was 16.16 %/day, the average daily relative growth rate (RGR) was 0.0359 g/g day, and the biomass doubling time (DT) was 32.94 days. The following models were used to model the dynamics of water lettuce biomass growth: the exponential model (average MSE 0.0485, average coefficient of determination (R^2) to 0.9757); the logistic model (mean squared error (MSE) 0.0049, R^2 0.9976), and the sigmoidal model (average MSE 0.0003, average R^2 0.9999). All models have a high accuracy; however, the exponential models give a continuous increase in biomass over time, practically to infinity, without taking into account that under conditions of increased plant density and reduced availability of resources, biomass growth slows down and, therefore, they are not suitable for application in real conditions. The logistic model (environmental capacity 6.1680 kg/m^2 after about 150 days, t_i 53.8587 days, t_a 32.8295 days, t_b 74.8879), and sigmoidal model (environmental capacity 5.2903 kg/m^2 after about 150 days, t_i 50.2972 days, t_a 34.3072 days, t_b 66.2872 days) adequately describe the biomass growth of the growth phase of water lettuce with high precision, which is essential for planning appropriate preventive and active measures to control the spread of water lettuce as an invasive plant and minimize negative impacts on waterbodies in Serbia.

Keywords: water lettuce; biomass production; biomass growth dynamics model.

Abbreviations: daily absolute growth rate (AGR), daily increase rate (DIR); daily relative growth rate (RGR); biomass doubling time (DT); dry weight (DW), wet weight (WW); probable biomass after time t on wet biomass basis (W_t); initial biomass on wet biomass basis (W_o); growth limit value of the population or load capacity (K); the rate constant or growth (r); integration constant (a); inflection point (t_i); mean squared error (MSE); root mean square error (RMSE); mean absolute error (MAE); coefficient of determination (R^2).

INTRODUCTION

Introduced invasive plants are an important factor in global changes, and pose a serious threat to biodiversity [1,2]. Previous research has documented the strong negative impact of invasive aquatic plants on aquatic flora and fauna [3,4]. Many scientists have focused on understanding the population dynamics of invasive species to discover the optimal method for managing and preventing the spread of these species within or outside their natural habitats [3-9].

Water lettuce (*Pistia stratiotes* L.) is considered one of the most widespread aquatic plants in the world and one of the most notorious invasive plants. It is distributed primarily in areas of tropical and subtropical climates [5], but it has also spread to other climatic zones, such as temperate climates, except for Antarctica [10]. It is widespread throughout Africa, where it was first recorded in the middle of the 19th century; Central and South America and South Asia, where it was classified as invasive [10]; throughout Oceania except for the North Island of New Zealand, where it was eradicated; in North America in the southern and southeastern USA [11] where it is most often classified as an invasive species, but also in more northern states up to the Great Lakes [10], and in Canada in the Ontario region

[11]. It is known as one of the aquatic macrophytes that strongly affects the environment and human activities in slow and stagnant water systems [12]. That is why numerous studies on the growth dynamics of water lettuce in tropical and subtropical countries have been conducted during the last decades [3,10,13-15].

In the last decades, there has been an increase in problems associated with the spread of invasive aquatic plants [6] in zones with a temperate continental climate, despite the weather conditions in these zones, which are generally unsuitable for establishing a permanent population of tropical invasive species. However, in these areas, research into the growth dynamics of invasive species is relatively rare, especially when it comes to water lettuce. In Europe, the presence of water lettuce was first mentioned in Great Britain in the 19th century [16]. During the 20th century, water lettuce was introduced to several European countries, and data on its appearance in Europe have shown a continuous expansion. In 2012, water lettuce was registered in 11 European countries [17], only to have water lettuce expand to 15 European countries in 2017 [10]. To this day, the presence of water lettuce has been occasionally or permanently registered in several other European countries.

In the European and Mediterranean Plant Protection Organization (EPPO) region, the presence of water lettuce in the natural environment was for the first time recorded in the Netherlands in 1973, but a permanent population of the plant was not established [18]. Occasional occurrences of water lettuce have been recorded in Great Britain in Somerset and the Bridgwater and Taunton Canal [10], in the continental part of Spain [10], in Austria [10], in several regions in northern and central Italy [19], in Croatia in the Zagreb district [20], in Russia around Moscow and in several areas south of Moscow towards the Caspian Sea [21]; in Ukraine, water lettuce appears relatively often in natural and artificial water bodies in the cities of Kyiv and Zaporizhzhia, in the Udi river on the outskirts of the city of Kharkiv [22], in the Seversky Donets river in the Kharkiv region, where in 2013 water lettuce completely covered the surface of the water on a section of about 40 km, causing an ecological disaster [23].

Permanent populations of water lettuce have been also recorded in Belgium, mainly in East Flanders [24]; in France, in the canal along the Rhône, where in 2016 it was recorded along 17 km of the canal, including several sections with 100% coverage [10]; in Spain in the Canary Islands, where due to favorable climatic conditions it creates significant problems and is considered invasive [10]; in Slovakia in the rivers Čierna Voda and Malý Dunaj, where water lettuce appears every year, often with a mass appearance, but it is indicated that the plant is highly likely to appear in other locations as well [25]; in Germany in warm parts of the Erft River [26]; in eastern Slovenia, near the Terme Čatež, where a permanent population of water lettuce has been established on the natural river Topla, into which the excess water from the thermal spring and the pool in the spa is poured, which has spread over about 25 hectares in a length of 3 km, all the way to the place where the Topla flows into the Sava river [27].

In Serbia, the appearance of water lettuce was first documented in the thermal spring Banjica in the Sićevačka Gorge near Niš in 1994 [12], and later in the thermal spring in Rgošte near Knjaževac [28]. In the flowing waters of Serbia, the appearance of water lettuce was first recorded in Vojvodina, a northern Serbian province, in the River Begej near Srpski Itebej, near the Romanian border, about 1.2 km downstream from it [12]. Considering the closeness of the Romanian border and that water lettuce has been present in Romania for several years [10], the emergence of water lettuce in the Begej River may be a consequence of spontaneous spread from Romania. Although this possibility is present, discarded plants from aquariums cannot be completely excluded as a potential source of water lettuce in the Begej River [12]. Since the end of the 1970s, water lettuce has been used together with water hyacinth for wastewater treatment under controlled conditions at the wastewater treatment plant in Sokobanja [29,30], managed by staff from the Faculty of Civil Engineering in Niš. Research in this area at the Faculty of Civil Engineering and Architecture of Niš has continued to this day [29-31].

Water lettuce grows in slow-moving rivers and reservoirs, irrigation canals, ponds, and lakes, where it often forms thick floating mats, spreading from the peripheral areas of the waterbody towards open water [32]. The unrelenting growth of water lettuce often causes a drastic reduction in the diversity of autochthonous communities of water flora and fauna [32].

It has a negative effect on the functions of freshwater systems. Water lettuce and water hyacinth (considered one of the world's most productive plants) coexist in the same water, with water lettuce being the dominant and harder to control. It is listed as an invasive species in the Global Invasive Species Database (GISD) 2023. Since 2012 it has been on the EPPO List of invasive alien plants, and since 2017 it has been on the EPPO A2 List of Pests Recommended for Regulation.

It is assumed that the increase in temperature due to climate change anticipated in the 21st century will benefit the further expansion of most of the introduced invasive aquatic plants [26], including water lettuce. The frequency and intensity of hot daily temperature extremes are predicted to increase globally. It is very likely that the duration, frequency, and/or intensity of heat waves will increase. It is also expected that by the end of the 21st century, in most regions except the high latitudes of the Northern Hemisphere, the warmest day that has so far occurred once in twenty years is likely to become an event that occurs every other year, and that the extreme maximum daily temperature that occurs once in twenty years is expected to increase by about 1°C to 3°C by the middle of the 21st century and by about 2°C to 5°C by the end of the 21st century, depending on the region [33].

An increase in temperature will cause an increase in the duration of the growth period. The amount of biomass, as well as the expansion of the area affected by aquatic macrophytes [34,35] can lead to the establishment of their permanent populations in the waterbodies and further expansion in the zones with a temperate continental climate; therefore, a quantitative assessment of the impact of these species on ecosystems is necessary to prioritize highly invasive species for management [36,37]. Future studies must ensure that relations between the ecosystem stocks, ecosystem flows and services and species traits are clearly defined [36]; therefore, it is necessary to understand invasive species biomass growth dynamics and to define a reliable model and stages of plant biomass growth.

In light of new information about the emergence of water lettuce in waterbodies in Serbia and the real danger of the further propagation of this invasive plant, this study aimed to (i) measure and analyze water lettuce biomass growth dynamics in experimental conditions of a temperate continental climate; (ii) to test different mathematical models for modeling its growth dynamics, and (iii) to select a reliable model describing the biomass growth dynamics and stages of water lettuce growth, which would provide valuable insights into biomass increment over time and the different growth stages of water lettuce as key elements for planning preventive and adequate active measures to control its spread as an invasive plant and minimize its negative impacts on waterbodies in Serbia. Additionally, this study aimed to select an appropriate model for modeling the growth dynamics of water lettuce. This study provides relevant information such as absolute growth rate, relative growth rate, biomass doubling time, and similar parameters to better understand the dynamics of water lettuce biomass growth in a moderately continental climate.

MATERIALS AND METHODS

Species description

Water lettuce (*Pistia stratiotes* L.) (Supplementary Fig. S1) is an aquatic plant belonging to the family *Araceae*, Genus: *Pistia,* which has only one species *P. stratiotes* [38]. Originating from the Pantanal region in South America (Bolivia, Brazil, Paraguay) [10], water lettuce is a free-floating plant with a rosette of ovate-to-spatulate leaves covered with short hairs (Supplementary Fig. S1). The upper sides of the leaves are light green, while the lower sides are almost white. The plants have a large feathery root system that hangs freely in the water. The inflorescence is axillary and inconspicuous, with short stalks in the center of the rosette [2]. The spadix, with one female and several male flowers enclosed in a whitish spathe, is pale green, hairy outside, and smooth inside [13]. The morphological structure of water lettuce makes it one of the most notorious weeds in the world. Flowering begins early in the plant's life, at the fourth or fifth leaf stage. After flowering and fertilization, the stem bends and pushes the berries under water, where up to 4-6 seeds per berry can be released. [13]. Flowering plants produce numerous viable seeds [26], with enormous densities of up to several thousand per m^2 [13,39]. Water lettuce seeds are more resistant to frost than the plants themselves and can survive at temperatures as low as -5°C. They germinate only at 20°C under intense light [13]. In areas where, during certain parts of the year, the conditions for the growth of water lettuce are unfavorable (increased salinity, drought), sexual reproduction is crucial for plant survival. In periods of unfavorable conditions, the seeds sink to the bottom of the sediment. After conditions for growth have improved, the seeds germinate, enabling the survival of water lettuce in many areas [14].

Water lettuce easily reproduces vegetatively. It is a clonal plant that forms small colonies with daughter plants attached to the parent plant via stolons up to 20 cm long. Separating the daughter plants that form new colonies intensifies the spread of water lettuce. The growth rate is incredible: one rosette can spread over an area of hundreds of square meters [39] and cover an entire pond with a thick carpet of interconnected rosettes in a short time [40].

The optimal growth of water lettuce is at 22-30°C [13] in conditions of high concentration of nutrients and plenty of light. However, it can even develop and grow at temperatures from 10°C [26]. Plants are sensitive to low temperatures and frost and decay when ice-bound and at temperatures slightly above 0°C [10]. Exceptionally, if there is no ice cover, smaller water lettuce plants can survive the winter with flat leaves in contact with the water if the water temperature is >10°C, which prevents the plants from suffering frost damage. [26]. Optimal growth of water lettuce is at pH 4 [13]. It is resistant to high salt concentrations and can withstand 200 mM NaCl in water [41].

Experimental setup and sampling

The experiment was performed in outdoor experimental conditions on an intermittent model system (Supplementary Fig. S2) at the Faculty of Civil Engineering and Architecture in Nis (43.33°N, 21.89°E). The experiment was carried out with a series of 5 polyethylene tubs of the same shape and size to ensure plants' growth under outdoor air temperature and sunlight conditions. Each tub had a total depth of 0.5 m, a bottom surface area 0.30 m^2, and top surface area of 0.40 m^2. The initial amount of water in each tub was 50 L, and it was obtained by mixing 50% of water from the shaft at the hydraulic engineering demonstration facility and 50% of sewage water collected at the outlet of the main collector of the general sewage system of the city of Nis near the village of Medoševac. The initial water level was marked in all tubs. To ensure a constant level of water lost by evapotranspiration and to replenish nutrients, wastewater was added to the tubs every 7 days to the initial levels.

Water lettuce plants were taken for the experiment from the open shaft at the hydraulic engineering demonstration facility of the Faculty of Civil Engineering and Architecture in Nis, which is filled with atmospheric runoff from the surrounding grassy areas. In the middle of May, when the daily temperatures stabilize above 15°C, water lettuce was sown in the shaft from the plant material that is stored during the winter in the laboratory at the wastewater treatment plant in Sokobanja as parent material for the following season [42]. Immediately after sowing three individual plants, their initial wet biomass was measured. The following parameters were measured: dry weight (DW), wet weight (WW), and the daily absolute growth rate (AGR), daily increase rate (DIR); daily relative growth rate (RGR); biomass doubling time (DT); dry weight (DW), wet weight (WW), probable biomass after time t on wet biomass basis (W_t), and initial biomass on wet biomass basis (W_o), were calculated.

During the experiment, the wet biomass of the individuals in each tub was measured every seven days with a digital analytical balance with an accuracy of 0.1 g. Before measuring, the plants were carefully removed from the tubs and left on a paper towel for 5-7 min to drain the water from the root system. The measured biomass of water lettuce is converted into kg/m^2 for easier comparison.

The experiment started on June 29, 2004 after stabilization of the minimum air temperature above 15°C. It was completed on September 7, 2004 because of the plants' decay due to low air temperatures (below 5°C) at the beginning of September during the night and morning hours. During the experimental period, complete daily meteorological data (air temperature, precipitation, relative humidity, sunshine) were obtained from the Republic Hydrometeorological Service of Serbia (RHMZ) (Supplementary Table S1). Mean values of meteorological data by weekend during the research period are presented in Supplementary Table S2. The data showed that the air temperature ranged between 9.7°C and 38.6°C. The average mean daily air temperature was 21.8°C, the average minimum daily air temperature 15°C, and the average maximum daily air temperature 29.3°C. Daily sunshine ranged from 0 to 12.8 h/day, with an average of 8.5 h/day, with short one-day intervals with less sunshine. The meteorological data for the entire test period of ten weeks were generally within the range favorable to the plant growth [10,29,38]. However, at the beginning of the 11^{th} week, 71 days from the start of the experiment, there was a sudden drop in the morning air temperature to 7.4°C and the minimum night temperature to 5.7°C. Low morning and minimum daily temperatures continued during the eleventh week. They reached the minimum on September 11 2004 when the morning air temperature was 5.4°C. The minimum night temperature was 2.5°C (Supplementary Table S1), which negatively impacted the growth and development of the plants [10,26,29,31]. Almost immediately after the drop in air temperature, the color of water lettuce leaves in all tubs changed to a pale-yellow green, and rapid drying of the leaves and decay of the plants was observed: because of this, the experiment was terminated, and the biomass at the end of the 11^{th} week was not measured.

Statistical analysis

To analyze the biomass growth rate of water lettuce and compare the results obtained in different natural slow-flowing and stagnant waterbodies in locations with different climatic conditions, the following parameters are calculated [9,43,44]:

$$AGR = (W_t - W_o)/(t - t_o)$$

$$DIR = [(W_t - W_o)/(t - t_o)] \times 100/W_o$$

$$RGR = (\ln W_t - \ln W_o)/(t - to)$$

$$DT = \ln 2/RGR$$

In previous research, the most extensively used to model the growth dynamics of water lettuce were the exponential (Malthusian growth model) and the logistical models (Verhulst growth model) of growth. In this paper, a sigmoidal model (based on Boltzmann's sigmoid equation) was also applied, which was not previously used to model the biomass growth dynamics of water lettuce.

The exponential model is presented with the equation [31,45]:

$$W_t = W_o \cdot e^{rt}$$

The logistical model is presented with the equation [9]:

$$W_t = K/(1+e^{a-rt})$$

The sigmoidal model is presented with the equation [46]:

$$W_t = W_o + (K-W_o)/(1+e^{-r\cdot(t-ti)}),$$

where AGR is the daily absolute growth rate (g/m²·day), DIR is the daily increase rate (%/day), RGR is the daily relative growth rate (g/g·day), DT is the biomass doubling time (day), W_t is probable biomass after time t on wet biomass basis (kg/m²), W_o is initial biomass on wet biomass basis (kg/m²) (initial state), K is growth limit value of the population or load capacity (kg/m²), r is the rate constant or growth (1/day), a is integration constant, t – time (day), and t_i – inflection point (day).

The integration constant a in the logistical model is defined through the expression:

$$a = \ln((K-W_o)/W_o)$$

The logistical function has an inflection point

$$t_i = \ln((K - W_o)/W_o)/r = a/r$$

The point of inflection divides the logistic and sigmoidal function into convex and concave branches, on which characteristic points can be defined, which are obtained if the third derivative of the function is equal to zero [46]:

$$t_A = t_i + \ln(2-\sqrt{3})/r = t_i - 1.317/r$$

$$t_B = t_i + \ln(2+\sqrt{3})/r = t_i + 1.317/r$$

The agreement of the model with the experimental data is assessed through the mean squared error (MSE), root mean square error (RMSE), mean absolute error (MAE), and coefficient of determination (R^2). The best model is defined as the model with the least MSE, RMSE, or MAE, or the model with the highest R^2.

For data processing and analysis, Microsoft Excel 2019 and free component-based software suite for machine learning and data mining Orange v3.32 developed at Bioinformatics Laboratory, Faculty of Computer and Information Science, University of Ljubljana, Slovenia,

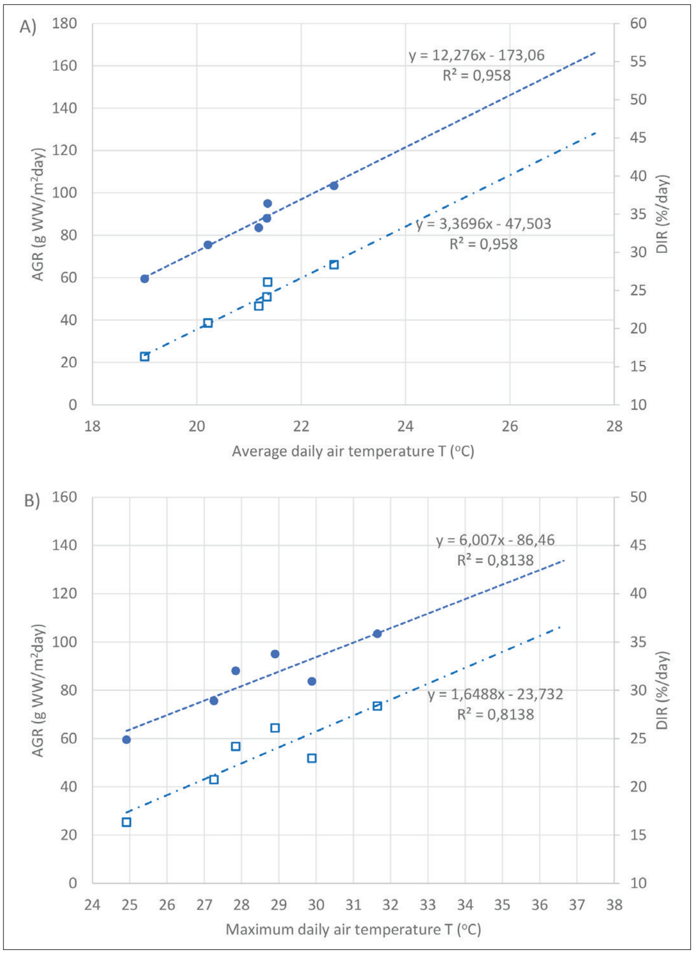

Fig 1. Variation of AGR and DIR as a function of average daily air temperature (**A**), and variation of AGR and DIR as a function of the maximum daily air temperature (**B**).

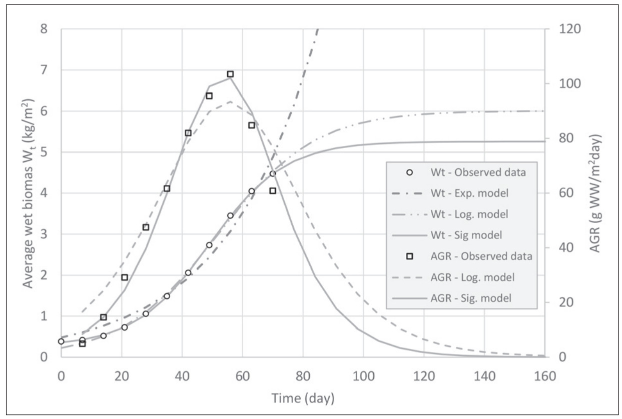

Fig 2. Exponetial, logistic, and sigmoidal models. Measured and modeled amounts of water lettuce W_t and daily absolute growth rate ADR over time.

Table 1. Average values of AGR, DIR, RGR, and DT for the research period in each tub.

Tub	AGR (g WW/m²·day)	DIR (%/day)	RGR (g/g·day)	DT (day)
Tub 1	58.43	15.24	0.0351	25.20
Tub 2	58.98	16.08	0.0358	25.18
Tub 3	58.01	16.47	0.0361	24.49
Tub 4	62.19	17.30	0.0368	47.21
Tub 5	56.45	15.69	0.0355	42.64
Min	56.45	15.24	0.0351	24.49
Max	62.19	17.3	0.0368	47.21
Average	58.81	16.16	0.0359	32.94

Daily absolute growth rate (AGR), daily increase rate (DIR); daily relative growth rate (RGR); biomass doubling time (DT).

were used. The linear regression dependence of the variables shown in Figs. 1 and 2 was established using the command Excel Trendline.

RESULTS

Water lettuce biomass growth

The measured values of water lettuce wet biomass for the entire research period for each tub are presented in Supplementary Table S3. The results of the calculation of AGR, DIR, RGR, and DT for each tub are presented in Supplementary Tables S4-S8. The results of the calculation of AGR, DIR, RGR, and DT for average wet biomass for all tubs are presented in Supplementary Table S9. The average values of AGR, DIR, RGR, and DT for the entire research period for each tub are presented in Table 1. The mean value for AGR was 58.81 g/m² day, for DIR 16.16%/day, for RGR 0.0359 g/g day, and for DT 32.94 days.

The effect of air temperature on the growth of water lettuce biomass

There is a correlation between the absolute (AGR) and relative (DIR) variation of the daily growth of biomass of examined species and the maximum daily air temperatures averaged weekly, so a regression correlation can be established between these parameters.

It should be noted that the analysis of the dependence of AGR and DIR on the mean daily air temperatures averaged weekly, i.e., the maximum daily air temperatures averaged weekly, did not take into account the data in the initial phase of the test because in this phase the plants had adapted to the new environmental conditions and biomass growth was not representative for the analysis. The analysis considered data from 35 to 70 days in the phase of stable exponential growth between the inflection points. Initially, individuals of water lettuce in all tubs have a uniform biomass/weight, and also there is no large dispersion of the measured values of biomass growth in individual tubs over time. That is why the analysis of the influence of air temperature on the growth of water lettuce biomass, on AGR and DIR, was not performed with the measured values of biomass growth for each tub individually, but with the average values of biomass growth, which were obtained for each time interval as the mean value of the measured values of biomass growth in all five tubs. (Supplementary Table S9).

The variation of AGR and DIR in the function of maximum daily air temperatures averaged every week, observed and modeled with linear regression for all tubs jointly, is shown in Fig. 1. The variation of AGR and DIR in the function of mean daily air temperatures averaged weekly, observed and modeled with linear regression for all tubs jointly, is shown in Fig. 2.

Figs. 1 and 2 show a strong regression relationship between AGR and DIR and the mean weekly temperature, i.e., the maximum daily air temperatures averaged weekly. The correlation coefficient between AGR and DIR and the mean daily air temperatures averaged weekly ranged from 0.7741 to 0.9013 (average for all tubs 0.8381), and between AGR and DIR and the maximum daily air temperatures averaged weekly from 0.5796 to 0.9080 (average for all tubs 0.7206). For the regression dependencies obtained for the average amount of biomass in all tubs, the correlation coefficient between AGR and DIR and the mean daily air temperatures averaged weekly was 0.9580, and between AGR and DIR and the maximum daily air temperatures averaged weekly, 0.8138.

To analyze the potential impact of temperature changes due to climate change, average annual air temperatures for the region of Niš and the wider region of Belgrade for the last four decades were analyzed. The results for Belgrade are given to compare the effects of increasing temperature in the same climate zone. In Supplementary Fig. S3 are presented the variations and trend change of mean yearly air temperatures, and in Supplementary Fig. S4, the yearly air temperature anomalies from 1979 to 2021 for Niš and the wider region of Belgrade. Fig. S3 shows how climate change has already affected the Niš and Belgrade regions [47] over the last 40 years. Based on the trend line, the mean annual air temperature for 40 years has increased for the region of Niš by about 2.2^{o}C and for the region of Belgrade by about 1.9^{o}C.

The temperature anomalies are presented for each year from 1979 to today for the regions of Niš and Belgrade. Anomalies show how much warmer or colder it was than the 30-year, 1980-2010 climate average. Thus, the red years were warmer, and the blue years colder than usual. For the region of Belgrade, the most significant thermal anomaly was 1.8°C, and the average was 0.82°C, while for the region of Niš, it was 1.93°C, and the average was 0.90°C. It is also noted that in the last 20 years, from 2000 to today, only two years were colder than usual, and all the others were warmer.

Considering that the temperature changes and temperature anomalies for the regions of Niš and Belgrade have been very similar in the past 40 years, it is likely that the majority of other locations will also experience a similar temperature increase and warmer months throughout the year.

Water lettuce growth dynamic modeling

The parameters of the water lettuce biomass growth dynamics model in each tub, and average parameters are shown in Tables 2, 3, and 4.

Table 2. Parameters of exponential water lettuce growth dynamics model for biomass values in each tub.

Tub	W_o (kg/m^2)	r (g/g·day)	MSE	RMSE	MAE	R^2
1	0.4822	0.0331	0.0580	0.2408	0.2154	0.9723
2	0.4447	0.0341	0.0519	0.2278	0.2034	0.9749
3	0.3972	0.0355	0.0455	0.2132	0.1882	0.9775
4	0.4357	0.0352	0.0638	0.2526	0.2286	0.9728
5	0.4321	0.0339	0.0426	0.2063	0.1878	0.9773
Min	0.3972	0.0331	0.0426	0.2063	0.1878	0.9723
Max	0.4822	0.0355	0.0638	0.2526	0.2286	0.9775
Average	0.4384	0.0343	0.0523	0.2281	0.2047	0.9750

Initial biomass on wet biomass basis (W_o); the rate constant or growth (r); Mean squared error (MSE); Root Mean Square Error (RMSE); Mean Absolute Error (MAE); Coefficient of determination (R^2).

Table 3. Parameters of logistical water lettuce growth dynamics model for biomass values in each tub.

Tub	K (kg/m²)	a	r (g/g·day)	W_o (kg/m²)	t_i (day)	t_a (day)	t_b (day)	MSE	RMSE	MAE	R^2
1	6.0097	3.2477	0.0625	0.2248	51.9862	30.9049	73.0676	0.0046	0.0677	0.0547	0.9978
2	6.1483	3.3454	0.0624	0.2093	53.6526	32.5310	74.7742	0.0063	0.0796	0.0676	0.9969
3	6.2689	3.4872	0.0627	0.1860	55.5749	34.5863	76.5635	0.0060	0.0776	0.0670	0.9970
4	6.3063	3.5028	0.0660	0.1844	53.0693	33.1158	73.0228	0.0070	0.0835	0.0679	0.9970
5	6.1066	3.2930	0.0599	0.2187	55.0103	33.0095	77.0112	0.0055	0.0741	0.0579	0.9971
Min	6.0097	3.2477	0.0599	0.1844	51.9862	30.9049	73.0228	0.0046	0.0677	0.0547	0.9969
Max	6.3063	3.5028	0.0660	0.2248	55.5749	34.5863	77.0112	0.0070	0.0835	0.0679	0.9978
Average	6.1680	3.3752	0.0627	0.2046	53.8587	32.8295	74.8879	0.0059	0.0765	0.0630	0.9972

Initial biomass on wet biomass basis (W_o); growth limit value of the population or load capacity (K); the rate constant or growth (r), integration constant (a); Mean squared error (MSE); Root Mean Square Error (RMSE); Mean Absolute Error (MAE); Coefficient of determination (R^2).

Table 4. Parameters of sigmoidal water lettuce growth dynamics model for biomass values in each tub.

Tub	K (kg/m²)	r (g/g·day)	W_o (kg/m²)	t_i (day)	t_a (day)	t_b (day)	MSE	RMSE	MAE	R^2
1	5.2554	0.0810	0.2693	49.0861	32.8352	65.3370	0.0007	0.0274	0.0247	0.9996
2	5.2647	0.0824	0.2705	50.0490	34.0629	66.0351	0.0015	0.0390	0.0290	0.9993
3	5.2696	0.0840	0.2676	51.4014	35.7258	67.0770	0.0007	0.0256	0.0194	0.9997
4	5.4594	0.0869	0.2743	49.9544	34.7939	65.1149	0.0070	0.0835	0.0679	0.9970
5	5.2023	0.0780	0.2480	50.9952	34.1182	67.8722	0.0021	0.0459	0.0386	0.9989
Min	5.2023	0.0780	0.2480	49.0861	32.8352	65.1149	0.0007	0.0256	0.0194	0.9970
Max	5.4594	0.0869	0.2743	51.4014	35.7258	67.8722	0.0070	0.0835	0.0679	0.9997
Average	5.2903	0.0825	0.2659	50.2972	34.3072	66.2872	0.0024	0.0443	0.0359	0.9989

Initial biomass on wet biomass basis (W_o); growth limit value of the population or load capacity (K); the rate constant or growth (r), integration constant (a); Mean squared error (MSE); Root Mean Square Error (RMSE); Mean Absolute Error (MAE); Coefficient of determination (R^2).

For the exponential model of growth dynamics of water lettuce for different tubs, the coefficients of determination are 0.9723 to 0.9775 (on average 0.9750), while for the exponential model of growth dynamics of water lettuce obtained with the average amount of biomass in all tubs, tubs coefficient of determination is 0.9757. High values of the coefficient of determination close to unity and low values of the MSE, RMSE, and MAE indicators show a high agreement of the exponential model with the measured amounts of water lettuce biomass. The growth rate of the exponential models for different tubs is from 0.0331 to 0.0355 g/g day (average 0.0343 g/g day).

For the logistic model of water lettuce growth dynamics for different tubs, the determination coefficients are from 0.9969 to 0.9978 g/g day (on average 0.9972 g/g day). High values of the coefficient of determination close to unity and low values of the indicators MSE, RMSE, and MAE show a high agreement of the logistic model with the measured amounts of water lettuce biomass in all tubs. The environmental capacity in partially controlled growth conditions for water lettuce obtained by the logistic model for different tubs is from 6.0097 kg/m² to 6.3063 kg/m² (average 6.1680 kg/m²) for a period of about 150 days, which coincides with the period of favorable climatic conditions for a temperate-continental climate, which is 5-6 months, from May to October.

For the sigmoidal model of water lettuce growth dynamics for different tubs, the coefficients of determination are from 0.9970 to 0.9997 g/g day (on average 0.9989 g/g day). High values of the coefficient of determination close to unity and low values of the MSE, RMSE, and MAE indicators show a high agreement of the sigmoidal model with the measured amounts of water lettuce biomass. The capacity of the environment in partially controlled growth conditions for water lettuce obtained by the sigmoidal model is from 5.2023 kg/m² to 5.4594 kg/m² (average 5.2903 kg/m²) for a period of about 150 days, which coincides with the period of favorable climatic conditions for a temperate-continental climate, which is 5-6 months, from May to October.

Considering the obtained values of model parameters in different tubs and averaged model parameters, water lettuce biomass growth dynamics models created with averaged model parameters are used for further analysis.

Average measured water lettuce wet biomass and exponential, logistic, and sigmoidal models of biomass growth dynamics created with averaged model parameters are presented in Fig. 2. Average measured water lettuce wet biomass and logistic model of biomass growth dynamics created with averaged model parameters, and daily absolute growth rate ADR over time, observed and modeled by the logistic model, are presented in Fig. 3. Average measured water lettuce wet biomass and sigmoidal model of biomass growth dynamics created with averaged model parameters, and daily absolute growth rate ADR over time observed and modeled by the sigmoidal model are presented in Fig. 4. Figs. 3 and 4 also show characteristic points and stages of water lettuce biomass growth for the logistic and sigmoidal models.

Figs. 1 to 3 show that logistic and sigmoidal water lettuce biomass growth models describe well the water lettuce biomass growth defined in this study. Figs. 1 to 3 show a better agreement between the measured and modeled values of AGR, as well as the measured and modeled values of water lettuce biomass with the sigmoidal model than with the logistic model.

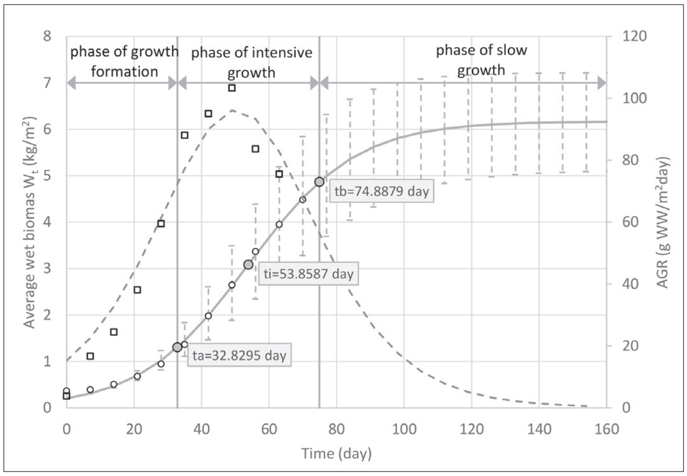

Fig 3. Logistic model. Measured and modeled amount of water lettuce biomass W_t and daily absolute growth rate ADR over time, with error bars indicating a 95% confidence interval for the forecasted values.

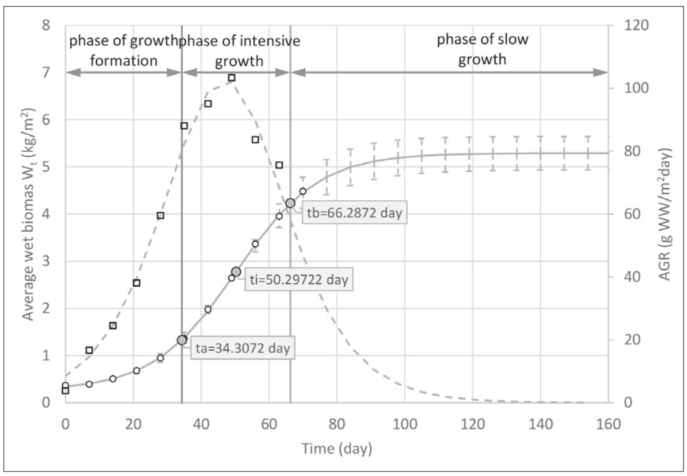

Fig 4. Sigmoidal model. Measured and modeled amount of water lettuce biomass W_t and daily absolute growth rate ADR over time, with error bars indicating a 95% confidence interval for the forecasted values.

DISCUSSION

The effect of air temperature on the growth of water lettuce biomass

Based on linear regression dependences between changes in AGR and DIR depending on the average temperature, i.e., the maximum daily air temperature obtained for the average amounts of biomass in all tubs, it can be concluded that there is no biomass production, i.e., water lettuce stops growing when the average air temperature is approximately 14°C; the daily absolute growth rate (AGR) increases by 12.276 g/m²·day for each °C with the increase of in mean daily air temperature of 1°C; the daily increase rate (DIR) increases by 3.369%/day for each °C with the increase in mean daily air temperature of 1°C.

With the relationships established in this way, extrapolation for different climatic conditions can be easily performed, enabling the formation of prognostic models of water lettuce biomass growth dynamics. The increase in temperature due to global warming and climate change will favor further expansion of invasive aquatic plants. Results in this study showing an increase in the average air temperature for the region Niš of about 2°C over the last 40 years means an increase in the daily AGR by about 25 g/m^2 day and DIR by about 6.7%/day, which for a season of 150 days means an increase in biomass of several tens of tons per ha, but also an increase in the duration of the growth period, as well as an expansion of the area affected by water lettuce. Air and water temperature are of fundamental importance to the growth stages of aquatic organisms and affect metabolism and growth rate. Increasing temperature can affect overwinter survival and seed germination date with far-reaching effects on size at germination and food availability. Such changes in ecosystem structure due to global warming will make the system more vulnerable to invasions by exotic species [48].

Water lettuce productivity

A wide range of values of water lettuce productivity in tropical and subtropical regions, where the conditions for water lettuce growth are favorable, has been recorded in the literature. The total amount of biomass in tropical and subtropical regions ranges between several tons to several hundred t DM/ha, in Nigeria 46.8-108.8 t DM/ha, in Ghana 52.1-61.4 t DM/ha [7], in Uruguay 70.9 t DM/ha [15], in Brazil 100.6 t DM /ha [8], in the region of the South Nile Delta, Giza Province in Egypt 34.2 t DM/ha. The high amount of biomass in tropical and subtropical climates is a consequence of favorable conditions for the growth of water lettuce in these countries. Variations in water lettuce biomass can be attributed to nutrient availability and environmental factors, such as habitat conditions, which vary with latitude, such as temperature, day length, solar radiation, and growing season length. However, variations in the amount of water lettuce biomass may be the result of other biotic as well as abiotic factors [7].

Studies that deal with water lettuce in locations with a moderate continental climate are few, and most of them deal only with the appearance of water lettuce in a specific location and do not provide data on the growth dynamics of water lettuce, so the data on the productivity of water lettuce in conditions of a moderate continental climate that can be compared with the results obtained in this study are limited.

At the Faculty of Civil Engineering and Architecture in Niš (43°19' N, 21°54' E), the average was RGR 0.0359 g/g day. The average RGR obtained in this study is comparable to that of the region of the South Nile Delta in the range of 0.0336-0.0354 g DM/g·a day, despite it being an area with different climatic conditions.

In this study, the biomass measured after 70 days was 4.31 to 4.71 kg WW/m^2 (average 4.48 kg WW/m^2). The annual productivity after about 150 days was 6.01 to 6.31 kg WW/m^2 (average 6.17 kg WW/m^2) according to the logistic model, i.e., 5.21 to 5.46 kg WW/m^2 (average 5.29 kg WW/m^2) according to the sigmoidal model.

The quantities of biomass of water lettuce obtained in this study are very similar to the amounts of biomass of 6.3 kg/m2 after 60 days of research conducted at the pilot plant in Sokobanja in similar climatic conditions but with wastewater in a slow-flowing system [45]. They are also very similar to the values obtained in the Republic of Slovakia on the Čierna Voda River (48° 11' 38" N; 17° 26' 58" E), where the total quantity of water lettuce biomass was 5.6-9.4 kg/m^2 (average 7.3 kg/m^2) [25], and in Ukraine on the Seversky Donets River (49°27'-49°47' N, 36°20'-36°52' E), where the quantity reached 4.5-19.9 kg WW/m^2 (average 6.7 kg WW/m^2) on the parts of the river where the entire water surface was covered with water lettuce, i.e., 4.0-12.2 kg WW/m^2 (average 5.26 kg WW/m^2) on the parts where the plant coverage was up to 30% [49], proving similar biomass production of the examined species within a moderately continental climate.

Based on the obtained results, it can be concluded that under conditions of moderately continental climate and nutrient-rich water, the growth rate of water lettuce is very high when the air temperature exceeds 14°C and increases with rising air temperature. In these conditions, substantial biomass quantities can be obtained during a 150-day season, amounting to over 5 tons per hectare of water surface. Such large amounts of biomass, along with the high seed production of water lettuce, seed resilience to adverse environmental conditions, and high reproductive capacity, enable rapid spread and the establishment of permanent habitats for water lettuce. These habitats can occupy significant water areas and pose an ecological disaster, as observed in multiple locations with a moderately continental climate (canal along the Rhône River in France [10], rivers Čierna Voda and Malý Dunaj in Slovakia [25], Seversky Donets in Ukraine [23]).

Growth dynamics of water lettuce

The obtained values of the growth rate of water lettuce of the exponential models for 70 days (0.0331 to 0.0355 g/g day, average 0.0343 g/g day) are in agreement with previously conducted research where the growth rates were 0.0107 to 0.0237 g/g day for 21 days of research [31], i.e., 0.026 g/g day for an average air temperature of 20°C and 0.061 for an average air temperature of 30°C, for a period of 60 days [45].

The exponential growth model of floating macrophytes does not consider the instability of the water system in terms of resource availability and the reduction of plant biomass growth dynamics due to the increase in plant density, so that with increasing time, it gives a continuous increase in biomass, practically to infinity. For this reason, the exponential growth model of water lettuce, regardless of the high accuracy in the initial phases of plant biomass growth, cannot be applied for modeling plant biomass growth in a real environment in all growth phases.

Considering the above, the sigmoidal model is recommended as the most adequate for describing the dynamics of biomass growth and defining the growth phases of water lettuce. However, the logistic model can also adequately describe biomass growth dynamics and define water lettuce growth stages with negligibly less accuracy. Given that the sigmoidal model has not been used to model the biomass growth dynamics of water lettuce, further research should confirm its high accuracy and possible application for modeling in this area. The inflection point t_i bifurcates the logistic and sigmoidal functions to the convex and concave branches. For the logistic model of water lettuce growth dynamics for different tubs, the inflection points are obtained from 51.9862 days to 55.5749 days (average 53.8587 days). For the sigmoidal model of water lettuce growth dynamics for different tubs, inflection points were obtained from 49.0861 days to 51.4014 days (average 50.2972 days).

For $t \leq t_i$, logistic and sigmoidal functions are convex, and in this interval, the growth is progressive. For $t \geq t_I$, logistic and sigmoidal functions are concave, and in this interval, the growth is reduced. Such a regime of water lettuce growth is in agreement with the water lettuce growth studies in Egypt [7], Florida [5], and Slovenia [27], which established two phases of water lettuce biomass growth as follows: gradual biomass growth and rapid biomass decline phases, when the biomass quantity drastically decreases [7]. Two clearly defined intervals are also visible in the graph of AGR change over time. The AGR in the interval of progressive growth at low plant density increases to the point of inflection, when it reaches a maximum, and in the phase of degressive growth, with an increase in plant density, it starts to decrease. Water lettuce biomass growth ends when the maximum plant density is reached, i.e., the capacity of the environment K, when there is no more increase in biomass growth.

In the interval of progressive growth for the logistic model of water lettuce growth dynamics for different tubs, the characteristic point t_a from 30.9049 days to 34.5863 days (average 32.8295 days) was obtained. For the sigmoidal model, the characteristic point t_a from 32.8352 days to 35.7258 days (average 34.3072 days) was obtained.

In the degressive growth interval, for the logistic water lettuce growth dynamics model for different tubs, the characteristic point t_b from 73.0228 days to 77.0112 days (average 74.8879 days) was obtained. A characteristic point t_b for the sigmoidal model of water lettuce growth dynamics for different tubs from 65.1149 days to 67.8722 days (average 66.2872 days), was obtained. The interval $(-\infty, t_A]$ represents the growth formation phase, in the interval $[t_A, t_B]$, there is intensive growth, and interval $[t_B, -\infty)$ represents the slow growth phase.

Research on the growth of floating macrophytes under conditions of unlimited nutrients showed that the biomass growth of floating macrophytes can be defined by a growth curve characterized by three phases: (i) a delay phase followed by exponential growth; (ii) a linear growth phase, and (iii) a slow exponential growth phase [9], which is in full agreement with the water lettuce growth regime obtained in this study.

Bearing in mind the increase in air temperature due to climate change, as well as the potential of the Tisza, Sava, and Danube rivers as corridors for the invasion of floating macrophytes from other countries to Serbia, there is a real risk of further expansion and establishment of permanent habitats of water lettuce. Considering the detrimental effects of water lettuce on the environment, it is necessary to establish regular monitoring and take urgent control measures, especially on rivers and canals near state borders. It is also essential to plan adequate preventive and active measures to control the propagation of water lettuce as an invasive plant and minimize negative impacts on waterbodies in Serbia. In this sense, understanding the dynamics of biomass growth of water lettuce in conditions of a moderate continental climate and defining a reliable mathematical model of the dynamics of biomass growth and stages of growth of water lettuce, as critical elements for planning adequate preventive and active measures, are of key importance.

CONCLUSIONS

Water lettuce is one of the most notorious weeds in the world, with numerous adverse effects on the environment. The results of this research provide valuable information/data on daily and annual water lettuce biomass production and the effects of air temperature on its biomass production, which facilitates the prediction of biomass growth in different climatic conditions while clearly defined growth stages of water lettuce can be used for planning adequate preventive and active measures to control the propagation of water lettuce as an invasive plant and to minimize its negative impacts on water bodies in Serbia and worldwide.

Based on this study, it can be concluded that water lettuce in moderate continental climate conditions can achieve an average productivity of around 5 to 6 kg/m^2, i.e., 50 to 60 t/ha in a season of 150 days, in partially controlled experimental conditions, while in natural conditions it can achieve even higher biomass quantities/production. Considering the potential risks of the propagation of water lettuce and the establishment of permanent habitats in the waterbodies of Serbia, there is a need for urgent regular monitoring and control measures but also setting up adequate preventive and active measures to control the propagation of water lettuce as an invasive plant. In light of new information about the occurrence of water lettuce and the risk of its further spread in natural water bodies in Serbia, the focus of this research was the analysis of different mathematical models of water lettuce growth dynamics that can be used to plan preventive and adequate active measures to control the spread of water lettuce as an invasive plant. Based on this research, a sigmoidal model has been proposed as the most effective for modeling the growth dynamics of water lettuce and defining growth phases, which is particularly important for planning measures to control the spread of water lettuce.

The obtained results can also be applied to future analysis of the possible use of water lettuce in the purification of wastewater in smaller settlements in temperate continental climate conditions. Further research is necessary on the application of different control methods for the spread of water lettuce as an invasive plant in moderate continental climate conditions, as well as on the risks the use of different control methods may pose to water quality or biodiversity in waterbodies.

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Data availability: The data underlying the reported findings have been provided with the submitted article and are available here: https://www.serbiosoc.org.rs/NewUploads/Uploads/Milicevic_8772_Raw%20Dataset.xlsx

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SUPPLEMENTARY MATERIAL

Supplementary Table S1. Meteorological data for the research period.

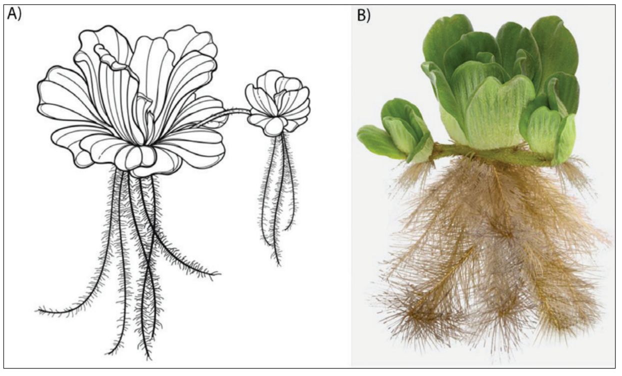
Day	Year	Month	Day	Air temperature 07 h (°C)	Air temperature 14 h (°C)	Air temperature 21 h (°C)	Mean daily air temperature (°C)	Minimal air temperature (°C)	Maximal air temperature (°C)	Precipitation (mm)	Mean daily air pressure (mb)	Mean daily relative humidity (%)	Mean daily cloud cover in 1/10	Sunshine (h)
0	2004	6	29	20.3	19.4	18.2	19.0	18.2	28.0	2.8	997.4	84	6.7	8.1
1	2004	6	30	17.2	24.8	16.5	18.8	14.1	26.8	5.6	997.6	65	1.3	12.4
2	2004	7	1	15.6	28.4	20.6	21.3	11.4	30.4	0	994.3	54	0.3	12.5
3	2004	7	2	19.6	32.0	25.6	25.7	14.0	33.5	0	989.8	57	1.0	11.9
4	2004	7	3	19.0	22.2	18.7	19.7	15.8	25.6	0	994.7	68	6.0	7.9
5	2004	7	4	17.2	29.7	22.7	23.1	13.5	30.7	0	996.1	53	1.3	12.3
6	2004	7	5	18.6	31.0	25.4	25.1	15.0	32.4	0	995.0	61	4.0	11.1
7	2004	7	6	23.6	33.4	24.9	26.7	17.0	34.7	0	993.7	47	0.3	12.3
8	2004	7	7	20.9	33.3	25.3	26.2	15.6	34.3	0	994.4	60	0.3	11.2
9	2004	7	8	22.1	34.3	27.0	27.6	18.0	35.7	0	994.5	53	0.0	12.3
10	2004	7	9	22.7	37.3	29.0	29.5	18.5	38.4	0	991.2	46	1.7	9.1
11	2004	7	10	25,0	37.9	27.0	29.2	20.0	38.6	0	987.1	49	4.3	11.5
12	2004	7	11	20.9	30.1	19.5	22.5	19.5	30.8	0	986.7	56	5.0	9.6
13	2004	7	12	17.7	23.0	16.0	18.2	15.0	25.0	1.9	985.8	80	9.0	4.4
14	2004	7	13	15.5	17.1	14.6	15.5	14.0	20.7	15.2	985.9	87	8.3	1.7
15	2004	7	14	14.7	21.4	16.7	17.4	12.5	22.5	1.8	991.7	68	7.7	4.9
16	2004	7	15	16.5	21.0	15.0	16.9	12.2	22.4	0	992.8	61	5.3	7.1
17	2004	7	16	15.7	24.5	17.7	18.9	12.9	26.0	0	996.0	67	5.3	7.0
18	2004	7	17	16.8	28.6	21.3	22.0	12.6	30.6	0	998.4	58	0.7	12.3
19	2004	7	18	18.9	31.4	23.3	24.2	13.2	33.2	0	998.1	55	0.7	12.8
20	2004	7	19	19.3	33.5	24.3	25.4	14.8	34.7	0	996.0	51	0.3	12.2
21	2004	7	20	21.4	34.2	24.5	26.2	15.7	35.0	0	994.6	55	1.0	12.1
22	2004	7	21	20.0	34.2	24.9	26.0	17.0	35.0	0	994.3	58	0.7	12.2
23	2004	7	22	21.0	34.8	24.6	26.3	17.5	36.3	0	994.0	61	1.7	11.4
24	2004	7	23	21.3	33.7	24.3	25.9	18.0	35.2	0	993.0	56	1.0	11.0
25	2004	7	24	23.7	30.4	24.5	25.8	19.4	31.5	0	990.6	57	6.3	11.6
26	2004	7	25	21.5	29.2	23.6	24.5	18.6	31.8	0	987.4	64	6.3	6.9
27	2004	7	26	22.5	26.1	18.2	21.3	18.2	28.4	0	986.0	73	7.3	5.3
28	2004	7	27	18.9	19.7	16.3	17.8	15.8	20.5	3.2	986.3	92	10.0	0
29	2004	7	28	16.2	18.4	14.8	16.1	14.8	19.9	19.5	991.1	84	9.0	0.6
30	2004	7	29	14.6	18.5	15.3	15.9	12.3	20.8	1.2	992.2	81	9.3	3.1
31	2004	7	30	15.0	21.2	17.0	17.6	13.0	24.0	0.6	993.7	78	8.0	6.6
32	2004	7	31	16.2	20.9	18.9	18.7	15.4	22.7	0.2	993.5	80	9.3	1.5
33	2004	8	1	18.2	27.1	19.3	21.0	16.6	28.2	3.1	991.4	74	3.3	9.2
34	2004	8	2	16.2	27.7	21.4	21.7	14.5	28.6	0	990.4	73	2.7	9.8
35	2004	8	3	17.0	29.3	20.9	22.0	14.5	30.2	0	988.6	67	5.7	10.6
36	2004	8	4	16.8	28.6	19.9	21.3	14.6	29.7	0	988.4	66	2.0	11.1
37	2004	8	5	16.9	28.8	22.0	22.4	14.6	29.9	0	989.0	67	1.7	11.9
38	2004	8	6	20.0	23.4	19.2	20.5	15.8	26.8	0	989.8	78	9.7	0.8
39	2004	8	7	19.1	27.8	21.1	22.3	17.5	29.0	0	990.9	71	4.3	7.0
40	2004	8	8	17.7	29.0	21.6	22.5	14.6	30.5	0	990.0	72	5.7	7.6

Table S1 continued

Day	Year	Month	Day	Air temperature 07 h (°C)	Air temperature 14 h (°C)	Air temperature 21 h (°C)	Mean daily air temperature (°C)	Minimal air temperature (°C)	Maximal air temperature (°C)	Precipitation (mm)	Mean daily air pressure (mb)	Mean daily relative humidity (%)	Mean daily cloud cover in 1/10	Sunshine (h)
41	2004	8	9	19.8	24.9	18.6	20.5	18.5	24.9	0.4	989.9	83	5.3	1.8
42	2004	8	10	17.3	21.6	20.4	19.9	16.5	24.1	6.3	991.6	83	8.3	2.8
43	2004	8	11	17.9	28.3	20.5	21.8	15.3	29.7	3	991.6	71	3.0	11.2
44	2004	8	12	16.9	31.3	22.9	23.5	14.0	32.4	0	990.2	60	2.0	11.9
45	2004	8	13	20.5	33.2	25.5	26.2	18.0	34.0	0	986.6	58	4.3	8.8
46	2004	8	14	21.4	19.4	17.1	18.8	16.7	25.5	0	989.2	83	8.7	0
47	2004	8	15	16.5	22.5	17.2	18.4	13.9	23.6	0	993.0	78	7.7	4.5
48	2004	8	16	16.1	25.8	19.1	20.0	12.9	27.1	0	993.9	65	3.0	9.9
49	2004	8	17	14.9	28.3	20.0	20.8	11.4	30.0	0	994	53	0.7	11.9
50	2004	8	18	14.9	31.4	21.6	22.4	10.8	32.3	0	995.1	59	0	11.8
51	2004	8	19	18.2	34.0	23.5	24.8	14.7	34.6	0	993.7	57	0	12.7
52	2004	8	20	19.4	35.5	25.3	26.4	16.5	36.8	0	988.0	51	0	11.8
53	2004	8	21	20.4	35.9	25.7	26.9	18.0	36.6	0	985.5	50	0	11.6
54	2004	8	22	19.1	21.9	15.6	18.1	15.6	25.7	0	993.9	70	5.7	7.4
55	2004	8	23	16.6	24.8	18.4	19.6	14.5	26.0	0.2	999.1	63	4.7	7.9
56	2004	8	24	13.4	27.9	19.7	20.2	10.8	29.5	0	997.7	60	0	11.1
57	2004	8	25	16.8	33.8	24.3	24.8	12.5	34.3	0	992.2	47	2.3	10.0
58	2004	8	26	20.8	34.7	21.9	24.8	17.5	35.6	0.7	985.1	49	4.7	9.1
59	2004	8	27	16.1	17.2	15.5	16.1	14.7	21.9	3.5	989.1	90	9.7	0
60	2004	8	28	15.9	23.0	16.6	18.0	14.6	24.5	7.6	992.7	70	6.0	4.3
61	2004	8	29	13.8	26.8	19.4	19.9	12.0	28.5	0	994.7	69	0.7	10.6
62	2004	8	30	15.3	30.2	20.0	21.4	13.6	31.8	0	993.7	66	1.0	10.9
63	2004	8	31	16.4	32.0	22.4	23.3	14.0	32.6	0	991.6	57	4.3	10.1
64	2004	9	1	17.0	26.2	19.2	20.4	15.8	27.4	0	995.8	67	3.0	10.1
65	2004	9	2	15.9	28.2	22.1	22.1	14.5	29.0	0	996.6	67	0.3	10.1
66	2004	9	3	17.5	28.4	21.5	22.2	15.4	29.6	0	999.5	65	6.0	8.4
67	2004	9	4	14.9	26.9	19.6	20.3	14.0	27.5	0	999.8	63	2.7	10.0
68	2004	9	5	15.5	24.7	19.2	19.7	13.8	25.5	0	999.7	65	5.0	1.2
69	2004	9	6	14.6	25.3	17.6	18.8	11.3	26.1	0	1002.3	51	3.3	7.6
70	2004	9	7	14.6	25.1	16.2	18.0	9.7	25.7	0	1002.3	41	0.3	10.9
71	2004	9	8	7.4	24.9	19.6	17.9	5.7	26.0	0	998.5	56	2.7	9.5
72	2004	9	9	13.9	20.5	12.0	14.6	12.0	21.5	0.9	1001	52	3.3	9.5
73	2004	9	10	7.6	17.9	10.0	11.4	5.5	19.1	0	1004.5	51	3.7	9.3
74	2004	9	11	5.4	21.8	12.3	13.0	2.5	24.5	0	1003.5	52	0	10.7
75	2004	9	12	6.2	27.9	16.2	16.6	4.0	29.0	0	997.9	52	1.7	10.0
76	2004	9	13	13.4	26.8	18.5	19.3	8.5	28.6	0	996.5	56	2.0	9.2
77	2004	9	14	12.8	29.8	19.0	20.2	11.6	31.5	0	996.4	61	2.3	9.9
Average				17.3	27.4	20.2	21.3	14.3	29.0	1.0	993.3	63.8	3.8	8.6
Min				5.4	17.1	10.0	11.4	2.5	19.1	0.0	985.1	41.0	0.0	0.0
Max				25.0	37.9	29.0	29.5	20.0	38.6	19.5	1004.5	92.0	10.0	12.8

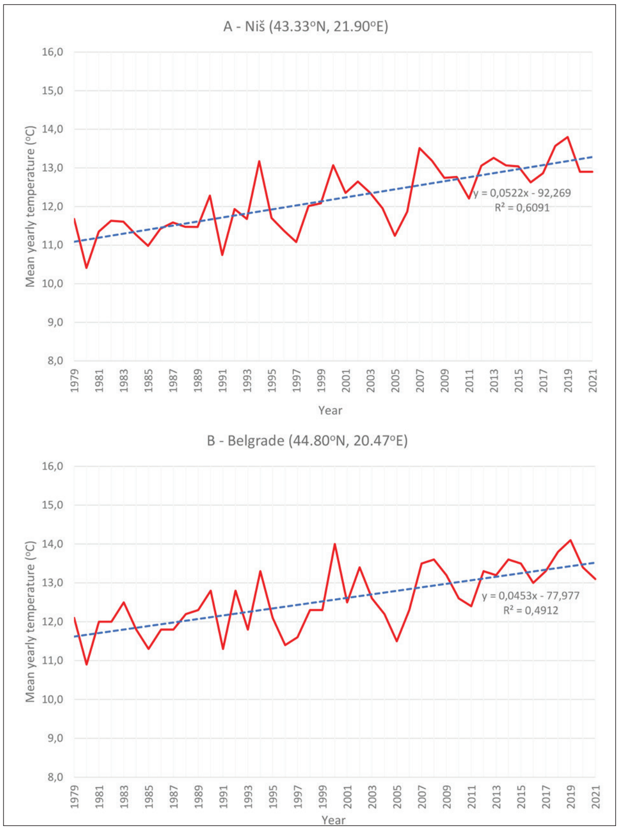
Supplementary Table S2. Mean values of meteorological data by weekends during the research period.

Week	Year	Air temperature 07 h (°C)	Air temperature 14 h (°C)	Air temperature 21 h (°C)	Mean daily air temperature (°C)	Minimum daily air temperature (°C)	Maximum daily air temperature (°C)	Precipitation (mm)	Mean cloud cover in 1/10	Sunshine (h)
1	2004	18.69	28.79	22.06	22.91	14.40	30.59	0.80	2.03	11.49
2	2004	20.69	30.43	22.63	24.10	17.23	31.93	2.44	4.09	8.54
3	2004	17.61	27.80	20.40	21.57	13.41	29.20	0.26	3.00	9.77
4	2004	21.27	29.73	22.34	23.94	17.79	31.24	0.46	4.76	8.34
5	2004	16.20	23.30	18.23	19.00	14.44	24.91	3.51	6.76	5.91
6	2004	18.23	26.30	20.40	21.34	16.01	27.84	0.96	5.29	6.14
7	2004	17.74	26.97	20.33	21.36	14.60	28.90	0.43	4.20	8.31
8	2004	17.43	30.20	21.40	22.63	14.41	31.64	0.03	1.49	10.61
9	2004	16.44	28.24	20.01	21.19	14.13	29.89	1.69	4.10	7.86
10	2004	15.71	26.40	19.34	20.21	13.50	27.26	0.00	2.94	8.33
11	2004	9.53	24.23	15.37	16.14	7.11	25.74	0.13	2.24	9.73

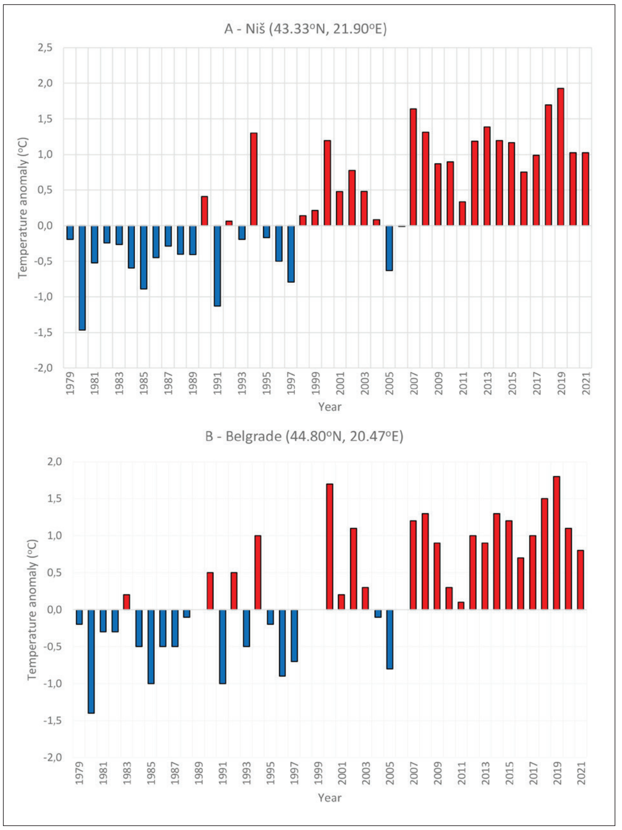


Supplementary Fig. S1. Schematic representation of plant parts (A); the appearance (B) of water lettuce.

Supplementary Fig. S2. Experimental tubs with plants.



Supplementary Fig. S3. Variations and trend change of mean yearly air temperature in the period 1979 to 2021. for Niš (A) and the wider region of Belgrade (B).



Supplementary Fig. S4. Yearly air temperature anomalies in the period 1979 to 2021 for Niš (A) and the wider region of Belgrade (B).