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CARBON ISOTOPE (д^{13}C) SIGNALS: INVASIVE PLANT RESPONSE TO CHANGING ENVIRONMENT

Проць Б.Г. **Сигнали ізотопу вуглецю (д^{13}C): реакція інвазійного виду рослин на зміну довкілля** // Наук. зап. Держ. природознавч. музею. – Львів, 2008. – **24**. – С. 77-82.

Шляхом експериментальних досліджень встановлено індикаційну здатність високоінвазійного виду рослин *Impatiens glandulifera* Royle реагувати на зміни природних умов за результатами вуглець-ізотопного (^{13}C %) аналізу. Нами виявлено взаємозв'язок між концентрацією ізотопу вуглецю у листях і стеблах цього виду та затіненням середовища, родючістю ґрунту, а також генеративними параметрами рослини. Між різними популяціями виду не встановлено достовірної різниці у накопиченні ізотопу вуглецю.

Проць Б.Г. **Сигналы изотопа углерода (д^{13}C): реакция инвазионного вида растений на изменения среды** // Науч. зап. Гос. природоведч. музея. – Львов, 2008. – Вып. **24**. – С. 77-82.

Путем экспериментальных исследований нами установлена индикационная способность высокоинвазионного вида растений *Impatiens glandulifera* Royle реагировать на изменения природной среды за результатами углерод-изотопного (^{13}C %) анализа. Выявлена взаимосвязь между концентрацией изотопа углерода в листьях, стеблях этого вида, а также затенением среды, плодородием почвы и генеративными параметрами растения. Между разными популяциями вида не установлено достоверной разницы в накоплении изотопа углерода.

Plant performance along environmental gradients offers one way to evaluate potential plant responses to climate change. Biochemical processes and diffusion during photosynthetic CO_2 assimilation lead to discrimination against the heavier ^{13}C isotope in plants [4]. The resulting fractionation is reflected in the stable carbon isotope signature (д^{13}C) of organic plant components. Plant performance along environmental gradients offers one way to evaluate potential plant responses to climate change. Isotopic measurements linked to plant performance (e.g., д^{13}C of leaves or whole plant biomass) allow multiple observations and replications along distinct environmental gradients. Light, air humidity, precipitation and temperature are environmental factors known to influence photosynthesis through their effect on stomatal conductance and CO_2 fixation [6, 12,].

The isotopic studies along environmental gradients (especially altitudinal changes) have consistently shown a shift towards increasing д$^{13}C_{leaf}$ at upper elevations [7, 8, 9, 14]. This pattern is correlated with physiological and morphological changes such as leaf thickness [17], leaf nitrogen content [11] and stomatal density [9]. In addition, correlations are reported for abiotic factors such as soil moisture [16], air temperature [13], and gradients of atmospheric pCO_2 and pO_2 [10, 11].

The Farquhar photosynthesis model combines photosynthesis with carbon isotope discrimination at the leaf level [5], but excludes downstream biochemical processes modifying the stable isotope ratio. The lack of understanding of the д^{13}C-signal chain limits the use of carbon isotopes in many ecological applications. So far, д^{13}C values and their variations in different cellular components and plant parts have been observed, but

interactions between them are not well understood. It is especially related to highly invasive species, which have got a rich amount of adaptation properties. Such ability of the plants to survive and expand in different environmental conditions might be interesting model for studying of carbon-isotope reactions to environment.

The study addresses the following questions: (1) Does exist any carbon-isotope ($д^{13}C$) response in Himalayan Balsam to environmental changes? (2) Does any difference in such response for local and distant populations? (3) Are any varieties of responses for one morphological part of the plant compare to other part?

Materials and methods of the study

The study object

The highly invasive *Impatiens glandulifera* Royle (*Balsaminaceae*) has been selected as a main object of the study. It is the tallest spontaneous annual plant in Europe, which makes a strong competitor with other species. The dominance of *I.glandulifera* along riverbanks has been repeatedly reported to cause problems in stream management [3]. Furthermore, the species is able to reduce the fitness of native flora [1] and to replace it in invaded sites [2, 3]. *I.glandulifera* is included into the PLANTLIFE's HIT LIST: the top 10 most harmful invaders. Crawley [2] considered it to be one of the "top twenty" British aliens. Native to the Western Himalayas, this plant occurs in 26 countries of Europe, also in the Far East, Japan and United States now. The species is currently expanding within the Eurasian and North American ranges [3].

The experiment

The Burgholz forest massif and the riverside of the Weisse Elster river channels (surroundings of Halle city, Eastern Germany) has been chosen as a study polygon. Six experimental plots have been established inside of the forest (shaded site; vegetation community of *Querco-Ulmetum minoris* Issler 1953) and on near the open site (*Impatienti glanduliferae-Convolvuletum sepium* Hilb. 1972). They are three for each site. Two populations (local/Halle/Germany and distant/Umea/Sweden) have been used. The seeds of these populations had germinated in the climate controlled chamber, after grown in the open greenhouse (Bad Lauchstдdt experimental station of the UFZ Research Centre, Leipzig/Halle, Germany) and finally placed on the Burgholz site's experimental plots during May-June (Fig. 1). The total duration of the field experiment has been 4,5 months (June-October). Total duration of the study was 8 months. The plants were growing in the highly fertilised soil and poor (sand) soil conditions. The pots of the plants with sand soil have been located in plastic sacks to avoid penetration of rich river soil on the experimental field (Fig. 2). Totally we used four treatments for each population with the next combinations: 1) open site + high fertilised soil; 2) open site + low fertilised soil; 3) shaded site + high fertilised soil; 4) shaded site + low fertilised soil. Total number of replicates was 5 per treatment and per population. Total number of plants was 40. The total number of samples, which has been analysed was 80 (2 populations x 4 treatment x 5 plants/per treatment x 2 morphological parts per plant). The above ground and inflorescence biomass as well as plant leaves and stem material have been collected and measured at the end of the experiment.

The leaves and stems were dried at 65°C for 48 h and ground in a steel ball mill (Mixer Mill, Retsch MM2000, Germany). The powder was weighed into small tin cups (0.6-0.8 mg

for $^{13}C/^{12}C$ measurements) and combusted to CO_2 for carbon isotope analysis with the elemental analyser (EA-1108, Carlo Erba, Italy), which is connected via a variable open split (Conflo II, Finnigen MAT, Germany) to a continuous flow mass spectrometer (DELTAS Finnigan MAT, Germany). The isotope signature is expressed in the delta notation д $^{13}C=(R_{sample}/R_{standard}–1)Ч1,000$ (‰), relative to the international standard (PDB for carbon), where R_{sample} is the $^{13}C/^{12}C$ ratio of the sample and $R_{standard}$ that of the standard.

Fig. 1. View of the experimental plot fragment on open site of the riverside of the Weisse Elster (Germany)

Fig. 2. Plants in poor and rich soil conditions on open site

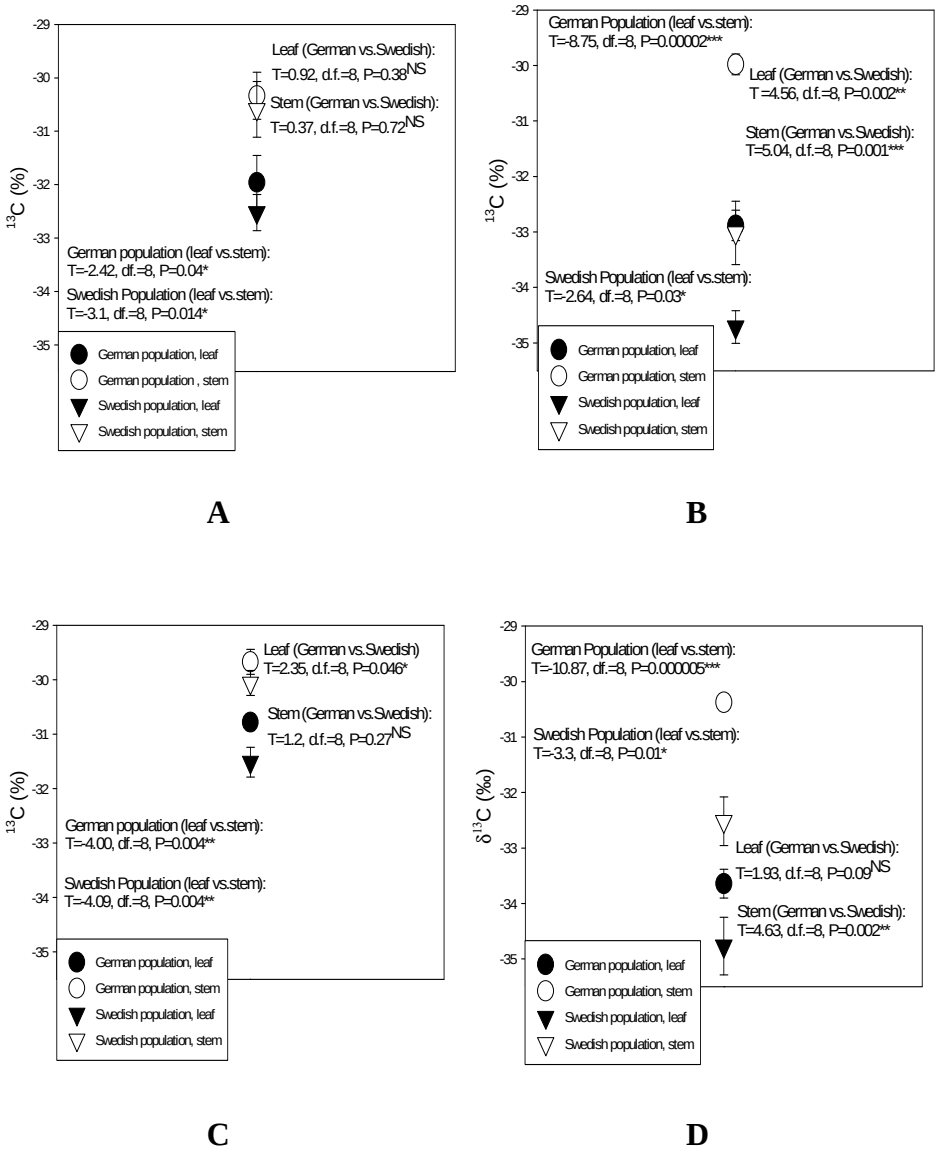
In the field experiment, treatment effects on д ^{13}C values were tested with ANOVA [15]. The correlations were tested with a simple regression analysis [18]. All statistical analyses were carried out with the program SigmaPlot 2001 for Windows Version 7.101 (1986-2001 SPSS Inc.).

Results

Carbon isotopic values distinctly differ between the two photosynthesis systems. In C3 plants, д $^{13}C_{leaf}$ values of whole tissue are reported from – 20‰ to – 35‰ whereas C4 plants range from – 7‰ to – 15‰ [4]. However, within these groupings intra- and inter-plant and species variability remains high.

Our д $^{13}C_{leaf}$ values of studied *Impatiens glandulifera* Royle are registered between – 30,47‰ and – 35,78‰. Some values are even higher compare to previously recorded in literature [4], however all means are within the mentioned range. The д $^{13}C_{stem}$ values are lower (between 29,26‰ and 34,5‰).

The received data (Figs. 3-6.) based on use of four treatments for two populations (local and distant) show the strong isotopic responses of leaves and stems of German and Swedish populations to all studied environmental conditions. The leaves are much more sensitive (P = 0.004** - 0.0005***) to environmental changes compare to stems (P = 0.03* - 0.002**). Probably, such response is weaker in stressful conditions, like low fertilizer and shaded site.



Figs. 3-6. Carbon isotope concentration (д ^{13}C) in leaf and stem of two populations of *Impatiens glandulifera* Royle for four treatments: A – open site/low fertilized soil condition, B – shaded (forest) site/high fertilized soil condition, C – open site/high fertilized soil condition, D – shaded (forest) site/low fertilized soil condition.

Especially high difference between leaf carbon isotope concentrations (д 13C) of open and shaded sites has been received on high fertilizer soils (T = 6,73, P = 0,00009***, d.f. = 9) and low fertilizer soils (T = 2,59, P = 0,029*, d.f. = 9).

A strong linear regression has been recorded between carbon-isotope concentrations (д 13C) of leaves and stems versus above ground biomass and reproductive effort (Figs. 7, 8). The highest level of correlation on environmental changes has been noticed between leaves and reproductive effort (biomass of inflorescence).

It is no clear identified difference for carbon-isotope response between local (German) and distant (Swedish) populations ($P = 0.09^{NS}$).

The present carbon-isotope studies need to be considered as a first step on the path of understanding of interactions between different cellular components and parts in the plant. *Impatiens glandulifera* Royle can be treated as useful model for this type of studies.

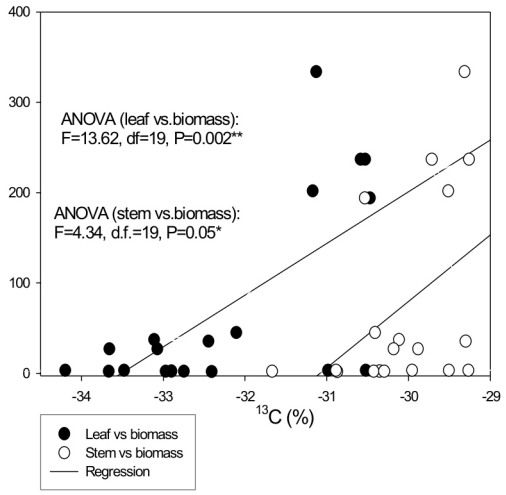


Fig. 7. Carbon isotop concentration (д ^{13}C) in leaf and stem versus above ground biomass in *Impatiens glandulifera* Royle

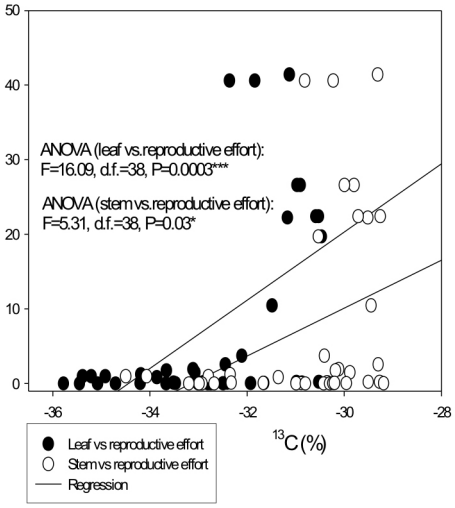


Fig.8. Carbon isotop concentration (д ^{13}C) in leaf and stem versus reproductive effort in *Impatiens glandulifera* Royle

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