Zbornik gozdarstva in lesarstva 88 (2009), s. 43 - 52

GDK: 811.4:174.7 *Picea abies*(497.4)(045)=111 Prispelo / *Recived*: 06. 02. 2009 Sprejeto / Accepted: 03. 06. 2009

Izvirni znanstveni članek Original scientific paper

STABLE CARBON ISOTOPES IN NORWAY SPRUCE (*Picea abies* (L.) KARST.) TREE RINGS AT TWO SITES IN SLOVENIA

Polona HAFNER¹ Tom LEVANIČ²

Abstract

Stable isotopes in tree rings are an important tool in climate reconstruction and in studies of tree response on environmental factors in the past, similar like maximum latewood density, tree ring width and other tree ring parameters. In this article we present stable carbon isotopes physiology and the first two Slovenian δ^{13} C chronologies for Norway spruce (*Picea abies* (L.) Karst.) growing at two sites with different ecological conditions. Five trees were sampled at Pokljuka alpine site and three at Sorško polje lowland plantation. δ^{13} C values were compared with CRU TS 1.2 meteorological data-set, which provides information on climate in Europe for the 1901-2000 period at a 10' spatial resolution. Results of correlation analysis for both sites revealed similar average δ^{13} C values, which are in strong negative correlation with amount of precipitation and positive correlation with temperatures in the growing period. Temporal stability of the correlation between precipitation and δ^{13} C values in July is significant for both sites, whereas the stability of δ^{13} C – temperature relationship is significant only for the Sorško polje site.

Key words: δ^{13} C, temporal stability, CRU TS 1.2 meteo data-set, climate

STABILNA OGLJIKOVA IZOTOPA V BRANIKAH SMREKE (Picea abies (L.) KARST.) NA DVEH RASTIŠČIH V SLOVENIJI

Izvleček

Stabilni izotopi v branikah dreves so, tako kot širine branik, maksimalna gostota kasnega lesa in drugi parametri branike, pomembno orodje pri rekonstrukciji klime v preteklosti in preučevanju odziva dreves na okoljske dejavnike. V prispevku predstavljamo fiziologijo stabilnih ogljikovih izotopov in prve slovenske δ^{13} C-kronologije smreke (Picea abies (L.) Karst.) na dveh lokacijah z različnimi ekološkimi razmerami. Na visokogorskem rastišču Pokljuka smo odvzeli vzorce petih, v nižinskem nasadu na Sorškem polju pa treh dreves. Vrednosti δ^{13} C smo primerjali s podatki CRU TS 1.2 meteorološke baze, ki vsebuje podatke o klimi za obdobje 1901-2000 v Evropi, s prostorsko ločljivostjo10'. Rezultati korelacijske analize so pokazali, da je na obeh rastiščih povprečna δ^{13} C-vrednost podobna in v močni negativni korelaciji s količino padavin ter v pozitivni korelaciji z visokimi temperaturami v vegetacijski sezoni. Stabilnost korelacijske povezanosti med padavinami v juliju in δ^{13} C je značilna za obe lokaciji, medtem ko se stabilnost odnosa δ^{13} C-temperature kaže le na Sorškem polju.

Ključne besede: δ¹³C, časovna stabilnost, CRU TS 1.2 meteorološki podatkovni set, klima

INTRODUCTION UVOD

Trees are long-living and permanent elements of the landscape. They germinate and grow at the same place, often for centuries. In their life period they produce new tree-rings every year, combining carbon from the air and oxygen and hydrogen from the soil water. Trees react to the varying abiotic and biotic factors influencing their growth and they respond by modifying the taken water and air. These small modifications are expressed as variations in isotopic ratios (MCCARROLL / LOADER 2004). These records can be therefore treated as terrestrial archives of high, annual temporal resolution, which contain valuable information on growth conditions at the time when a tree ring was formed (SAURER *et al.* 1997), which is of fundamental importance for the reconstruction of past environments (TREYDTE *et al.* 2001).

DEFINITION OF THE STABLE ISOTOPES DEFINICIJA STABILNIH IZOTOPOV

Isotopes are atoms of the same chemical element that comprise the same number of electrons and protons, but they differ in the number of neutrons and in atomic mass. Stable isotopes are defined as those that are energetically stable and do not decay; thus, they are not radioactive (SULZMAN 1994). All of the three main elements (carbon, oxygen and hydrogen) in wood have more than one stable isotope and they have almost identical chemical properties. Difference in mass is the main reason why isotopes are useful in environmental researches

¹ P. H., univ.dipl.inž. gozd., Gozdarski inštitut Slovenije, Večna pot 2, 1000 Ljubljana, polona.hafner@gozdis.si

² doc.dr. T. L., Gozdarski inštitut Slovenije, Večna pot 2, 1000 Ljubljana, tom.levanic@gozdis.si

- it allows physical, chemical and biological process to discriminate against one of the isotope and thereby imparting an environmental signal (MCCARROLL / LOADER 2004). The partitioning of isotopes between two substances or two phases of the same substance with different isotopic ratio is called "isotopic fractionation" and there are two main phenomena that lead to this process: isotopic exchange reactions and kinetic processes, determined by reaction rates of molecules (DAWSON / BROOKS 2001). The isotopic ratio is expressed in parts per thousand (‰) and it indicates the difference between standard and sample values. In case of "tree isotopes" there are two important standards - in case of carbon PDB (Pee Dee Belemnite) is used and for water, ice and plant material SMOW (Standard Mean Ocean Water). PDB and SMOW are now replaced by Vienna-PDB (VPDB) and VSMOW, as the original standards have been exhausted (PEZDIČ 1999; MCCARROLL / LOADER 2004) (Table 1).

Main characteristics of the heavier isotopes are lower reactivity, higher density and viscosity, as well as higher melting- and boiling-point (PEZDIČ 1999). From this point on, we focused only on stable carbon isotope and its environmental signal.

THEORY OF STABLE CARBON ISOTOPES IN TREE RINGS

TEORIJA STABILNIH OGLJIKOVIH IZOTOPOV V BRANIKAH

As tree changes the components of CO_2 in response to environmental conditions, the isotopic records are not simply a sample of ancient air but a sensitive bioindicator of tree reaction in past environments (MCCARROLL / LOADER 2004). The isotope ratio is revealed as relative decline of internationally accepted standard in parts per thousand (‰) and is expressed in delta (δ) using the following equation:

$$\delta^{13}C = (R_{sample} / S_{standard} - 1) \ge 1000$$

where R_{sample} and $R_{standard}$ are the ratios of heavier (¹³C) to lighter (¹²C) isotope in a sample and standard, respectively (LOADER / ROBERTSON / MCCARROLL 2003). Higher (less negative) δ values show that sample contains more of heavier isotope, while more negative values indicate more lighter isotope in the sample (DAWSON / BROOKS 2001).

The isotope shift between air (R_{air}) and tree material (R_{sample}) is expressed by the discrimination (Δ) presenting isotope exchange between air and plant:

 $\Delta = (R_{air} - R_{sample}) / R_{sample} \cong \delta^{13}C_{air} - \delta^{13}C_{sample} \text{ (TREYDTE} et al. 2001).$

The ratio of ¹³C to ¹²C in CO₂ of air currently yields a δ^{13} C value of about -8‰ with respect to the standard (FARQUHAR / LEARY / BERRY 1982). Values in tree material are much lower, varying in range of -20% to -30%, demonstrating that trees are depleted in ¹³C relative to the air. This change in ratios is known as fractionation and there are more important points where it occurs. The first point is the transition of CO₂ through the stomata. ¹²CO₂ molecules can diffuse more easily, so internal air is depleted in ¹³C in comparison to ambient air and it results in "fractionation due to diffusion" -4.4‰. The second point of fractionation is estimated to be of about -27‰ and it occurs during the photosynthesis where the photosynthetic enzyme (rubisco in C₃ plants) tends to fix ¹²C in preference to ¹³C (MCCARROLL / LOADER 2004). Farquhar et al. (1982) introduced the equation expressing the isotopic composition of plant, considering these two fractionation points:

$$\delta = \delta_{atm} - a - (b - a) * c_i / c_a,$$

 Table 1:
 Elements, their stable isotopes and abundance in terrestrial environments, isotopic ratio measured, internationally accepted standards and their isotopic ratio (DAWSON / BROOKS 2001)

 Tabela 1:
 Elementi, njihovi stabilni izotopi in njihova zastopanost v naravi, merjeno razmerje izotopov, referenčni mednarodno sprejeti standardi in njihovo izotopsko razmerje (DAWSON / BROOKS 2001)

Element Element	Stable Isotopes Stabilni izotopi	Abundance (%)Ratio measuredZastopanost (%)Merjeno razmerje		Standard <i>Standard</i>	
Hydrogen	¹ H ² H (D)	99.985 0.015	² H/ ¹ H	VSMOW	
Carbon	¹² C ¹³ C	98.98 1.11	¹³ C/ ¹² C	VPDB	
Oxygen	¹⁶ O ¹⁷ O ¹⁸ O	99.759 0.037 0.204	¹⁸ O/ ¹⁶ O	VSMOW VPDB	

where δ_{atm} is isotopic composition of atmospheric CO₂, *a* is the fractionation occurring due to the diffusion in air, *b* is the fractionation caused by carboxylation, c_i [Pa] and c_a [Pa] are intercellular and ambiental CO₂ concentration, respectively. Water use efficiency (WUE) is related to both points of fractionation and thus in close relationship with δ^{13} C (WARREN / MCGRATH / ADAMS 2001). δ^{13} C values between different tissues (e.g. leaves and tree-ring) can differ greatly and this could be due to source-product transformation that occurs during dark respiration and photorespiration in leaves (GHA-SHGHAIE *et al.* 2003) or fractionation during transfer of photosynthates into tree rings (HELLE / SCHLESER 2004).

Several on tree-ring widths based spruce dendrochronological investigations were published in Slovenia, based on the study of growth at natural sites (LEVANIČ / ČUFAR / ZU-PANČIČ 1995; STANOVNIK 1998), response to pollution (FERLIN 1991; SLAPNIK 2006) or response to climate and site condition (LEVANIČ et al. 2006; HAFNER / LEVANIČ 2008). In this article we present the first two spruce δ^{13} C values chronologies for two sites in Slovenia, with aims (1) to determine correlations between δ^{13} C variations in spruce trees and climatic parameters at two different sites and compare the differences between the sites, (2) to test temporal stability of δ^{13} C signal, and (3) to check and test the difference between Slovenian and CRU TS 1.2 meteorological data-sets.

MATERIAL AND METHODS MATERIALI IN METODE

SITE AND TREES SELECTION *RASTIŠČE IN IZBOR DREVES*

The main factor to distinguish sampling sites was different altitude with different ecological conditions. Typical alpine location at Pokljuka (1250 m a.s.l.) is a natural Norway spruce (*Picea abies* (L.) Karst) site, denoted by relatively cool and moisture conditions in summer - July and August are the warmest months, high amount of snow precipitation and low temperatures in winter. The majority of precipitation falls in October and November, however, it is abundant through the whole year. At Sorško polje lowland plantation (350 m a.s.l.), conditions are less favourable for spruce growth. The soils are shallow and just below the organic horizon lies a thick layer of gravel, which quickly drains water into deeper layers, out of reach for the spruce roots. Despite the relatively high amount of precipitation through the year, as well in growing period, spruce at Sorško polje therefore suffers from water stress in summer time, when temperatures are relatively high as well. Co-dominant spruces without any visible damage were included in this research - three trees from the Sorško polje and five from Pokljuka sites. Trees were of similar age, around 70 years and only last 40 years (tree-rings) were included in the research to avoid the "juvenile effect" (LIU *et al.* 2004; GAGEN *et al.* 2007).

METEOROLOGICAL DATA-SETS METEOROLOŠKI PODATKOVNI NIZI

So far, meteorological data from Slovenian local meteorological stations (Environment Agency of the Republic of Slovenia - ARSO) were used for similar dendro researches. Sometimes local meteo data-sets can be missing, short or erroneous especially in more remote places. The backup in such cases can be application of gridded CRU TS 1.2 database, developed by Tyndall Centre for Climate Change Research and the Climate Research Unit (CRU). The data-set includes time series of monthly observed precipitation, temperature, vapour pressure, diurnal temperature range and cloud cover for the 1901 – 2000 period for the European land surface at a 10 minute resolution (MITCHELL 2008). For the purpose of our research, climatic parameters were extracted from CRU database and used for the ensuing analyses. To compare local (Slovenian) and CRU data sets t-test and descriptive statistics, calculations were made in SYSTAT program.

SAMPLE ANALYSIS OBDELAVA VZORCEV

Stem disks of the chosen trees were used for stable carbon analysis. Samples were extracted manually using a scalpel. Each tree-ring sample was divided into early and late wood; only the latter was used for further proceeding. For each sample, α -cellulose was extracted through a series of chemical steps using a modified batch processing technique (LOADER *et al.* 1997; RINNE *et al.* 2005). α -cellulose was dried and between 300 to 350 µg of the sample were wrapped in tin capsules, which were combusted on-line to CO₂ at 1,000°C over chrome(III) oxide using an ANCA Elemental Analyser interfaced to a PDZ Europa 20/20 isotope ratio mass spectrometer. Results are presented as per mille deviations from the VPBD standard using the conventional delta δ^{13} C notation. The precision on replicate analyses of an internal cellulose reference material is typically better than 0.1 per mille (n = 15). All analyses were performed in the stable-isotope laboratory at the Swansea University in Wales, Department of Geography.

DATA PROCESSING OBDELAVA PODATKOV

Any change in isotopic composition of atmospheric CO_2 can potentially affect tree ring $\delta^{13}C$, as tree takes CO_2 from the atmosphere. To remove this influence, some corrections must be applied before construction of $\delta^{13}C$ chronology, otherwise they can mask the signal that is in our interest. Correction of isotope series was employed to remove the effect of anthropogenic changes in isotopic composition of atmospheric CO_2 (MCCARROLL / LOADER 2005), which is depleted in ¹³C because of fossil-fuel combustion and this change is reflected in products of photosynthesis, eventually in wood cellulose (GAGEN *et al.* 2007). Product of this correction is chronology, marked as "cor" in further text.

EPS - EXPRESSED POPULATION SIGNAL MOČ KLIMATSKEGA SIGNALA V IZBRANI POPULACIJI DREVES

In dendrochronological researches usually there is just a limited number of trees cored due to economic and ethic reasons. The results after handling and measurements of sampled trees are often averaged and they should be good representatives of the analysed site. Whenever sets of time series (tree rings) are averaged to enhance some common underlying signal or they are combined to produce spatial averages, the question of how well does the average of analysed series represent the population average is raised (WIGLEY / BRIFFA / JONES 1984). The answer to this question gives the calculation of the EPS value, which is dependent on number of trees and average correlation between analysed tree rings. There is no fixed value for EPS threshold, but $EPS \ge 0.85$ was excepted to be high enough to show that chosen tree ring series represent the population well while still containing qualitative climatic signal (WIGLEY / BRIFFA / JONES 1984).

TEMPORAL STABILITY OF CLIMATE SIGNAL ČASOVNA STABILNOST KLIMATSKEGA SIGNALA

Although limiting factors controlled tree ring parameters in the past just as they do today, it is possible that the role of different factors at a single location or over an entire region could change over time. DENDROCLIM2002 software (BIONDI / WAIKUL 2004) was used to analyze the dynamic response functions, which provide an information about temporal stability of relationship between tree rings δ^{13} C and climate variables. The calculation of response and Pearson's correlation coefficients using bootstrapped method is based upon 1000 repetitions with random selection from analyzed data and the results are tested to their significance at the 0.05 level (BIONDI / WAIKUL 2004). In our research, "forward intervals" option was used, with base length of 25 years. Coefficients were exported to statistical program R 2.5.1 where graphs of temporal stability were made. Other graphs were drawn in SigmaPlot 10.0.

RESULTS REZULTATI

CLIMATIC DATA-SET COMPARISON

PRIMERJAVA METEOROLOŠKIH PODATKOVNIH SETOV

Two sample t-test analysis showed no significant difference between CRU and Slovenian annual average temperature (t=-0.878 ns, df=198) but statistically significant difference between annual sum of precipitation (t=-2.602 **, df=198) for Sorško polje. There is visible agreement between temperature sets and clear offset between precipitation data-sets at Sorško polje, with higher values for Slovenian data-set particularly in April, June, October and November with the exception of August, where CRU values are higher (Figure 1).

Local precipitation data for Pokljuka are incomplete and a part of values is obtained by interpolation, so they should be treated with caution. At Pokljuka, there is a significant difference between local and CRU temperatures (t=7.534***, df=92). Offset is approximately 1°C and is constant for all months. However, the curve course is similar for both datasets. The comparison between precipitation data-sets has also shown a significant difference (t=-6.682***, df=92). The amount of precipitation retrieved from CRU data-set is consistently lower, except in October and November, where the difference between two data-sets is significantly higher (Figure 2). For both Pokljuka and Sorško polje, a decreasing trend of precipitation and an increasing trend of temperatures (approximately 0.03°C per year) was observed in the 1960 – 2000 period.



Fig. 1: Comparison between CRU and Slovenian meteorological station meteorological data-set for monthly average temperatures and monthly sum of precipitation at Sorško polje for the 1901-2000 period

Slika 1: Primerjava med CRU in lokalnimi (Ljubljana) meteorološkimi podatki o povprečnih mesečnih temperaturah in mesečnih količinah padavinah na Sorškem polju za obdobje 1901-2000



Fig. 2: Comparison between CRU and local (Mrzli studenec) meteorological station data-set for average monthly temperatures and monthly sum of precipitation at Pokljuka for the 1954-2000 period

Slika 2: Primerjava med CRU in lokalnimi (Mrzli studenec) meteorološkimi podatki o povprečnih mesečnih temperaturah in mesečnih količinah padavin na Pokljuki za obdobje 1954-2000

$\delta^{13}\text{C}$ VALUES OF INDIVIDUAL TREES AND SITE RECORDS

δ¹³C VREDNOSTI POSAMEZNIH DREVES IN LOKACIJ

The average corrected δ^{13} C values for both sites are similar, -22.8 ‰ at Sorško polje and

-22.46 ‰ at Pokljuka. The same is true for absolute minimum and maximum values of trees per site - the values for individual trees ranged in interval from -25.8 ‰ to -20.4 ‰ at Sorško polje and between -24.1 ‰ and -20.9 ‰. at Pokljuka.

Although the average values are close for both sites, there is a difference in variability of individual trees. At Sorško polje, an average absolute difference between minimum and maximum within the tree's δ^{13} C value is 4.6‰, and only 2.2‰. at Pokljuka. T-test confirmed statistically significant differences between chronologies (t = 2.042***, df=80) (Table 2). Year-to-year variations are large at Sorško polje; records from Pokljuka also follow the same trend, just on a smaller scale. There are some years when the same reaction of trees can be observed, e.g. increase of δ^{13} C in the years 1967, 1976, 1983, and decrease of δ^{13} C in the years 1980 and 1989 are characteristic of both sites (Figure 3 and 4). The mean June to August temperature follows the curve of δ^{13} C chronologies for both sites and has the same peaks in the above mentioned



Fig. 3: Sequences of individual tree corrected ("cor") δ¹³C series and local "cor" chronology at Sorško polje
Slika 3: Zaporedja korigiranih ("cor") δ¹³C-vrednosti posameznih dreves in lokalna kronologija "cor" na Sorškem polju



Fig. 4: Sequences of individual tree corrected ("cor") δ^{13} C series and local "cor" chronology at Pokljuka Slika 4: Zaporedja korigiranih ("cor") δ^{13} C vrednosti posameznih dreves in lokalna "cor" kronologija na Pokljuki

years. In spite of apparently small number of sampling trees and the difference in variability, the EPS test confirmed that trees are yielding high common signal (EPS ≥ 0.85) and are thus suitable for further analysis.

CLIMATIC SIGNAL IN δ¹³C RECORDS KLIMATSKI SIGNAL V δ¹³C ZAPISIH

Pearson's correlation coefficients were calculated between mean climate variables and δ^{13} C values. The correlation analysis revealed a clear and significant positive relationship between δ^{13} C values and temperatures at both sites (Figure 5). Also the combinations of all summer months, including May, give high correlation values, up to r = 0.55 at Sorško polje and up to r = 0.57 at Pokljuka for July-August combination.

All correlations between precipitation and δ^{13} C values are negative with the exception of precipitation in October at Sorško polje, however, this correlation is not significant (Figure 6). At Sorško polje, correlations with precipitation are even stronger than those with temperature. Precipitation in June has significant and strong impact on δ^{13} C values, in contrary to non-significant correlation with temperature. Influence of

Table 2: Number of analysed tree-rings (N), minimum (Min) and maximum (Max) δ^{13} C values per tree, difference between Min and Max δ^{13} C value (Max- Min), mean δ^{13} C value per tree (Avg) and standard deviation (St.dev). All values are expressed in [‰].

Tabela 2:Število analiziranih branik (N), minimalna (Min) in maksimalna (Max) $\delta^{13}C$ vrednost analiziranega drevesa,
razlika med minimalno in maksimalno $\delta^{13}C$ vrednostjo (Max-Min), povprečna $\delta^{13}C$ -vrednost (Povp.) v drevesu
in standardni odklon (Std. odkl.). Vse vrednosti so izražene v [%].

	SOR1	SOR2	SOR3	POK1	POK2	POK4	POK5	POK6
N	41	41	41	41	41	41	41	41
Min	-25.1	-25.8	-25.0	-23.7	-22.5	-23.9	-22.9	-24.1
Max	-20.8	-21.0	-20.4	-21.2	-21.2	-21.8	-21.3	-20.9
Max-Min	-4.4	-4.9	-4.5	-2.5	-1.4	-2.1	-1.6	-3.2
Avg. / Povp.	-23.0	-23.4	-22.5	-22.8	-21.9	-22.9	-22.1	-22.5
St.dev. /Std. odkl.	±1.0	±1.0	±1.0	±0.6	±0.3	±0.5	±0.4	±0.8



Month / Mesec

- Fig. 5: Pearson's correlation coefficients between δ^{13} C values and monthly temperatures and their combinations for the Sorško polje and Pokljuka sites ($r_{sig} = 0.23$)
- Slika 5: Pearsonovi korelacijski koeficienti med δ^{13} C-vrednostmi in mesečnimi temperaturami ter mesečnimi kombinacijami temperatur na Sorškem polju in na Pokljuki ($r_{znac} = 0.23$)

precipitation in all summer months and their combinations is significant and negative in case of Sorško polje, but for the Pokljuka site this is only true for July's precipitation and summer months combinations.

TEMPORAL STABILITY IN δ¹³C VALUES – CLIMATE RELATIONSHIP

ČASOVNA STABILNOST ODNOSA MED δ¹³C VREDNOSTMI IN KLIMO

Temporal analysis of δ^{13} C signal stability showed that the δ^{13} C values at Sorško polje are significantly connected with high temperatures in July. After 1993, this relationship has not been significant anymore, however, the correlation of δ^{13} C with combination of warm summer months including May appears. Stable positive relationship between δ^{13} C values and July – August temperatures can be observed for the

whole analysed period (1960 - 2000). On the other hand, negative correlations are visible for all combinations of summer months precipitation sums. The $\delta^{13}C$ – climate relationship is less pronounced at the Pokljuka site. Warm temperatures in July and August are significantly positively correlated with $\delta^{13}C$ values, but the lack of consistency is evident. There is a strong negative correlation with precipitation in July for the last decade, otherwise the precipitation signal is quite variable (Figure 7).

DISCUSSION RAZPRAVA

The CRU TS 1.2 temperature data-set proved to be a reliable replacement for temperature records in Slovenia. However, comparison between both precipitation data-sets revealed significant differences between sums of precipitation





Slika 6: Pearsonovi korelacijski koeficienti med δ^{13} C-vrednostmi in mesečnimi padavinami ter mesečnimi kombinacijami padavin na Sorškem polju in na Pokljuki ($r_{max} = 0.23$)



Fig. 7: Temporal stability of precipitation and temperature signal in δ^{13} C at Sorško polje and Pokljuka Slika 7: Časovna stabilnost signala temperature in padavin v δ^{13} C-vrednostih na Sorškem polju in na Pokljuki

with higher values for Slovenian data. This is probably due to the complex orography and specific precipitation regime in Slovenia, which is characterized by heavy local showers. Since the course of precipitation curves is similar and the major difference in amount of precipitation is recorded in autumn months, when the growing period ceases, we concluded that it is possible to use gridded CRU precipitation data-set in places where local data are not available, erroneous or with many missing records (e.g. local data-set for Pokljuka).

Two δ^{13} C chronologies were constructed for two different spruce sites in Slovenia. Main distinctive factors are different altitudes and ecological conditions. Average δ^{13} C values are almost the same at both sites. Taking into account the results of leaf nitrogen content analyses at different altitudes, which revealed that leaves of trees growing at higher altitudes usually contain less negative δ^{13} C values (HULTINE / MAR-SHALL 2000; SAH / BRUMME 2003), one would expect the same difference reflected in tree ring's δ^{13} C values. This is not the case of our results, which are in agreement with conclusion of Treydte *et al.* (2001), that δ^{13} C signal in subalpine spruce appears to be independent of altitude. These findings are contrary to the maximum density results, where significant lag between values at different altitudes can be observed (HAFNER / LEVANIČ 2008). δ^{13} C values are thought to be tree-ring parameter, strongly linked to climate, less sensitive to ecological conditions and not varying greatly between sites (GAGEN / MCCARROLL / EDOUARD 2004). But although mean δ^{13} C values are similar for both sites, there is a difference in standard deviation between them, with higher variations at Sorško polje. Standard deviations values explain that there is different response to micro-site conditions which is, at least in this case, masked in average δ^{13} C site values. Spruce in Slovenia naturally grows at higher altitudes (above 600 m a.s.l.) and does not need much summer warmth but demands good soils, high relative air humidity and well distributed precipitation through the whole year. Drought can significantly decrease spruce's vitality (KOTAR / BRUS 1999). Total annual sum of precipitation at Sorško polje is about 1,400 mm - so the reason for higher $\delta^{13}C$ is probably not the lack of precipitation but moisture stress caused by fast drainage of water into deeper layers. Trees at Sorško polje react more sensitively in comparison to trees at Pokljuka, where ecological conditions for spruce growth are optimal. Water condition at dry sites seems to be more important than temperature (SAURER et al. 1997; GAGEN / MCCARROLL / EDOU-ARD 2004). The dominant controls of stable carbon isotopes ratio are stomatal conductance, which responds to drought and air humidity, and photosynthetic rate, which responds to

temperature and sunlight (MCCARROLL / LOADER 2006). Correlation analysis revealed significant positive connection between summer temperatures and δ^{13} C values at both site. On the other hand, correlation between precipitation and δ^{13} C values is negative and more pronounced at Sorško polje. Hot and dry conditions in July and August lead to higher δ^{13} C values in tree-rings. This indicates the closure of stomata as a response to reduced internal concentration of CO₂ and finally in ¹³C enriched tree-ring's cellulose (GAGEN / MCCARROLL / EDOUARD 2004). δ^{13} C provides an information on CO₂ uptake and vapour loss during photosynthesis (WUE) and consequently registers drought stress under limiting conditions (BARBER / JUDAY / FINNEY 2000).

Temporal stability of the climate-isotope ratio has a significant meaning specially in climate reconstruction as it is based on the assumption that the climate-isotope relations are stable over time (REYNOLDS-HENNE et al. 2007). Our time series were quite short for the temporal stability analysis and the main reason is that trees were rather young and that the first decades of chronology were excluded from the analysis. In this way we avoided the "juvenile effect", characterized by ¹³C depletion, which is caused by incorporation of respired CO, and changes in hydraulic conductivity as trees gain height (after GAGEN et al. 2007), or because of lack of light (SAURER et al. 1997). Different authors suggested different periods of juvenile phase - 20 to 50 years (LIU et al. 2004; GAGEN et al. 2007). In our case, only the last 40 years were appropriate for analysis, as incorporation of longer tree ring series was still showing the age trend. At Sorško polje, all summer months combinations of precipitation exhibit stable negative correlation with δ^{13} C, with exception of May-June combination. The most consistent is correlation with July included, when the average precipitation amount in the last 50 years is the lowest in the vegetation period, while the demand for water supply in this period is high. Temporal stability of precipitation signal could be related to the assumption that due to recent climate warming, drought may have been an important factor limiting carbon uptake and isotopic composition of trees (BARBER / JUDAY / FINNEY 2000). On the other hand, there is a great lack of consistency of climate signal in δ^{13} C values at Pokljuka. Researches on longer δ^{13} C time series of pine revealed changes in $\delta^{13}C$ - climate relationship: a shift in the dominance of temperature from August to July is in possible relation to the longer growing season. Reynolds-Henne et al. (2007) also concluded that long-terms δ^{13} C climate trends are not well represented by the 20th century relationships, as sensitivity to climate condition seems to increase strongly during this period of heightened anthropogenic influences.

POVZETEK SUMMARY

Vsi trije glavni gradniki lesa (ogljik, kisik in vodik) so v naravi zastopani z več kot enim stabilnim izotopom, ki imajo skoraj identične lastnosti, razlikujejo pa se v atomski masi, ki posledično povzroča diskriminacijo proti težjemu izotopu v fizikalnih, kemičnih in bioloških procesih. Izotopsko sestavo snovi (branike) izražamo z vrednostjo delta (δ) v tisočinkah deleža [‰], podajamo pa jo kot relativni odklon od mednarodno sprejetega standarda (VPDB za ogljik). Vrednosti izotopske sestave branik so občutljiv bioindikator reakcije drevesa na okoljske razmere v preteklosti in kot take pomemben podatek v dendroekoloških in dendroklimatoloških raziskavah. Izračun δ¹³C-vrednosti temelji na podatkih o izotopskem razmerju CO₂ v atmosferi (-8‰), frakcionaciji pri difuziji CO₂ skozi listne reže (-4,4‰), frakcionaciji med procesom fotosinteze (-27‰) ter razmerju medcelične in okoliške koncentracije CO₂. Prvi cilj naše raziskave je bil preveriti razlike med lokalnimi slovenskimi (ARSO) meteorološkimi podatki ter CRU TS 1.2 meteorološke podatkovne baze, ki je osnovana na zbranih podatkih z 10' prostorsko ločljivostjo za celotno Evropo. Nadalje smo predstavili prvi dve smrekovi kronologiji δ¹³C-vrednosti za dve rastišči v Sloveniji, izračunali korelacije med δ^{13} C-vrednostmi in klimatskimi parametri ter primerjali razlike med obema rastiščema in testirali časovno stabilnost klimatskega signala v δ^{13} C-vrednostih. CRU TS 1.2 (CRU) meteorološke podatkovna baza se je v dendroklimatološki analizi izkazala kot razmeroma dobra zamenjava za temperaturne podatke lokalnih meteoroloških postaj, pri podatkih o padavinah pa prihaja do večjih, statistično značilnih razlik. CRU podatke smo uporabili za korelacijsko analizo, ki je pokazala dobre, negativne korelacije δ^{13} C s poletnimi temperaturami in negativne korelacije s padavinami. Največji vpliv imajo julijske temperature in količina padavin, visoke korelacije z δ13C-vrednostmi pa obstajajo tudi med padavinami in temperaturami drugih poletnih mesecev in njihovih kombinacij. δ^{13} C-vrednosti se na obeh rastiščih gibljejo okoli podobnega povprečja, to je -22.8 ‰ na Sorškem polju in -22.46 ‰ na Pokljuki. Kljub temu da je trend poteka krivulje na obeh lokacijah podoben, med kronologijama obstaja statistično značilna razlika, ki jo je potrdil tudi t-test ($t = 2.042^{***}$, df=80). Variabilnost v δ^{13} C-vrednostih v branikah dreves je na Sorškem polju večja kot na Pokljuki. Analiza stabilnosti odnosa med δ^{13} C in klimo na Sorškem polju pokaže značilno pozitivno povezanost δ^{13} C z visokimi temperaturami v juliju in avgustu ter s povprečno temperaturo od maja do julija. Prav tako so značilne negativne in stabilne korelacije s količino padavin v vseh poletnih mesecih, na Pokljuki pa je zaznati pomanjkanje zveznosti klimatskega signala v δ^{13} C-vrednostih.

REFERENCES VIRI IN LITERATURA

- BARBER, V. A. / JUDAY, G. P. / FINNEY, B. P., 2000. Reduced growth of Alaskan white spruce in the twentieth century from temperature-induced drought stress.- Nature 405: 668-673.
- BIONDI, F. / WAIKUL, K., 2004. DENDROCLIM2002:A C++ program for statistical calibration of climate signals in tree-ring chronologies.-Computers & Geosciences 30: 303-311.
- DAWSON, T. E. / BROOKS, P. D., 2001. Fundamentals of Stable Isotope Chemistry and Measurement.- In: *Stable Isotope Techniques in the Study* of *Biological Processes and Functioning of Ecosystems*. Unkovich, M. / Pate, J. / McNeill, A. / Gibbs, D. J. Dordrecht, **40**, p. 1-18.
- FARQUHAR, G. D. / LEARY, M. H. O. / BERRY, J. A., 1982. On the Relationship between Carbon Isotope discrimination and the Intercellular Carbon Dioxide Concentration in Leaves.- Australian Journal of Plant Physiology
- FERLIN, F., 1991. Nekatere značilnosti pojava umiranja smreke in njenega prirastnega odzivanja na emisijske strese.- Zbornik gozdarstva in lesarstva 37: 125-156.
- GAGEN, M. / MCCARROLL, D. / EDOUARD, J.-L., 2004. Latewood Width, Maximum Density, and Stable Carbon Isotope Ratios of Pine as Climate Indicators in a Dry Subalpine Environment, French Alps.-Arctic, Antarctic, and Alpine Research 36, 2: 166-171.
- GAGEN, M. / MCCARROLL, D. / LOADER, N. J. / ROBERTSON, I. / JALKANEN, R. / ANCHUKAITIS, K. J., 2007. Exorcising the segment length curse summer temperature reconstruction since AD 1640 using non-detrended stable carbon isotope ratios from pine trees in northern Finland.- The Holocene 17, 4: 435-446.
- GHASHGHAIE, J. / BADECK, F.-W. / LANIGAN, G. / NOGUÉS, S. / TCHERKEZ, G. / DELÉENS, E. / CORNIC, G. / GRIFFITHS, H., 2003. Carbon isotope fractionation during dark respiration and photorespiration in C3 plants.- Phytochemistry Reviews 2: 145-161.
- HAFNER, P. / LEVANIČ, T., 2008. Vpliv klime na maksimalno gostoto kasnega lesa v branikah smreke (*Picea abies* (L.) *Karst*) na dveh rastiščih v Sloveniji.- Zbornik gozdarstva in lesarstva 85: 3-10.
- HELLE, G. / SCHLESER, G. H., 2004. Beyond CO2-fixation by Rubisco – an interpretation of 13C/12C variations in tree rings from novel intraseasonal studies on broad-leaf trees.- Plant, Cell and Environment 27: 367-380.
- HULTINE, K. R. / MARSHALL, J. D., 2000. Altitude trends in conifer leaf morphology and stable carbon isotope composition.- Oecologia 123: 32-40.
- KOTAR, M. / BRUS, R., 1999. Naše drevesne vrste.- Ljubljana, Slovenska matica, 320 p.
- LEVANIČ, T. / ČUFAR, K. / ZUPANČIČ, M., 1995. Kronologije letnih prirastkov jelke in smreke v Sloveniji.- Les 47, 9: 259-261.

- LEVANIČ, T. / JALKANEN, R. / GRIČAR, J. / GAGEN, M. / OVEN, P., 2006. Use of different growth parameters of Norway spruce (*Picea abies* (L.) Karst.) to study tree response to climate.- Zbornik gozdarstva in lesarstva 80: 97-109.
- LIU, Y. / MA, L. / LEAVITT, S. W. / CAI, Q. / LIU, W., 2004. A preliminary seasonal precipitation reconstruction from tree-ring stable carbon isotopes at Mt. Helan, China, since AD 1804.- Global and Planetary Change 41: 229-239.
- LOADER, N. J. / ROBERTSON, I. / BARKER, A. C. / SWITSUR, V. R. / WATERHOUSE, J. S., 1997. An improved technique for the batch processing of small wholewood samples to α-cellulose.- Chemical Geology 136: 313-317.
- LOADER, N. J. / ROBERTSON, I. / MCCARROLL, D., 2003. Comparison of stable carbon isotope ratios in the whole wood, cellulose and lignin of oak tree-rings.- Palaeogeography, Palaeoclimatology, Palaeoecology 196: 395-407.
- MCCARROLL, D. / LOADER, N. J., 2004. Stable isotopes in tree rings.-Quaternary Science Reviews 23: 771-801.
- MCCARROLL, D. / LOADER, N. J., 2005. Isotopes In Tree Rings.- In: Isotopes in Palaeoenvironmental Research. Leng, M. J., Springer, p. 67-116.
- MCCARROLL, D. / LOADER, N. J., 2006. Isotopes in tree rings.- In: Isotopes in Palaeoenvironmental Research. Leng, M. J., Springer, p. 303.
- MITCHELL, T., 2008 CRU TS 1.2 http://www.cru.uea.ac.uk/~timm/index. html
- PEZDIČ, J., 1999. Izotopi in geokemijski procesi.- Ljubljana, Naravoslovnotehniška fakulteta, Oddelek za geologijo, 269 p.
- REYNOLDS-HENNE, C. E. / SIEGWOLF, R. T. W. / TREYDTE, K. S. / ESPER, J. / HENNE, S. / SAURER, M., 2007. Temporal stability of climate-isotope relationships in tree rings of oak and pine (Ticino, Switzerland).- Global Biogeochemical Cycles 21: GB4009.
- RINNE, K. T. / BOETTGER, T. / LOADER, N. J. / ROBERTSON, I. / SWITSUR, V. R. / WATERHOUSE, J. S., 2005. On the purification of α-cellulose from resinous wood for stable isotope (H, C and O) analysis.- Chemical Geology 222: 75-82.
- SAH, S. P. / BRUMME, R., 2003. Altitudinal gradient of natural abundance of stable isotopes of nitrogen and carbon in the needles and soil of a pine forest in Nepal.- Journal of forest science 49, 1: 19-26.
- SAURER, M. / BORELLA, S. / SCHWEINGRUBER, F. / SIEGWOLF, R., 1997. Stable carbon isotopes in tree rings of beech: climatic versus siterelated influences.- Trees 11: 291-297.
- SLAPNIK, A., 2006. Dendroekološka analiza debelinskega prirastka navadne smreke (*Picea abies* (L.) Karsten) v okolici termoelektrarn Šoštanj in Trbovlje. Magistrsko delo, Maribor, Univerza v Mariboru, Pedagoška fakulteta, Oddelek za biologijo, 124 p.
- STANOVNIK, M., 1998. Dendroekološka analiza rasti smreke (*Picea abies* Karst.). Diplomsko delo, Ljubljana, Univerza v Ljubljani, Biotehniška fakulteta, Oddelek za lesarstvo, 54 p.
- SULZMAN, E. W., 1994. Stable isotope chemistry and measurement: a primer.- In: *Stable Isotopes in Ecology and Environmental Science*. Lajtha, K. / Michener, R. H. Boston, Blackwell Publishing, p. 23.
- TREYDTE, K. / SCHLESER, G. H. / SCHWEINGRUBER, F. H. / WINIGER, M., 2001. The climatic significance of δ¹³C in subalpine spruces (Lotschental, Swiss Alps) - A case study with respect to altitude, exposure and soil moisture.- Tellus 53B: 593-611.
- WARREN, C. R. / MCGRATH, J. F. / ADAMS, M. A., 2001. Water availability and carbon isotope discrimination in conifers.- Oecologia 127: 476-486.
- WIGLEY, T. M. L. / BRIFFA, K. R. / JONES, P. D., 1984. On the Average of Correlated Time Series, with Applications in Dendroclimatology and Hidrometeorology.- Journal of Climate and Applied Meteorology 23: 201-213.