Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

Pol. J. Environ. Stud. Vol. 33, No. 4 (2024), 1-7
DOI: *10.15244/pjoes/182892* ONLINE PUBLICATION DATE:

Original Research

Determination of Δc13 and Δc18 Isotope Ratios to Predict Carbon-Water Relationship in Different Forest Stands in Yenice Forest Hot Spot

Sevda Türkiş*

Ordu University, Faculty of Education, Department of Mathematics and Science Education, Ordu, Turkey

Received: 21 November 2023
Accepted: 20 January 2024

Abstract

The study is aimed to determine the C13 and O18 isotope ratios of plant species belonging to different life forms in different forest types in Yenice Forests hot spot. Carbon and oxygen isotope ratios are important in providing long-term information about plants. In our study, we grouped the plants by considering the life forms of Raunkiaer. We determined the C13 and O18 isotope ratios of life forms in different forest layers. We found highly positive correlations ($p<0.05$) between δ13C and δ18O isotope ratios in beech-fir and pure oak forest habitats. In the Yenice Forests, which have a megathermal climate type, plants in different forest layers have settled in the areas by choosing the life forms where water will be used most effectively. Geophyte (-27.38±2.27 $p<0.05$) is the life form with the highest C13 isotope ratio in terms of life forms. This situation proves that the group that felt the water deficiency the earliest is the geophyte form. For O18 isotope ratio, therophytes using surface water are the highest (24.06±3.92 $p<0.05$) life forms. It is important to examine the water deficiency in old forests such as Yenice Forests through life forms.

Keywords: Yenice Forests, carbon isotope ratio; oxygen isotope ratio; life forms; geophyte

Introduction

The importance of water scarcity as a factor limiting plant growth and terrestrial ecosystem productivity in future climate change scenarios may increase worldwide, especially in arid and semiarid regions. According to this, the study of intraspecific variability in growth and drought-related physiological responses is crucial for understanding the adaptation potential of forest trees to climate change. The carbon isotope composition (δ13C) in plant tissues provides an integrated measure of the ratio between chloroplastic and atmospheric CO_2 concentrations (Cc/Ca) and therefore the determination of carbon isotope ratios can be used to estimate the ratio between the net CO_2 assimilation rate (A) and stomatal conductance for water [1]. When plants are subjected to any environmental impact that causes a decrease in stomatal conductance or photosynthesis (drought, shade, etc.), atmospheric carbon dioxide concentrations will be reflected in a change in the 13C/12C isotope ratio in this organic matter (e.g., increased drought will lead to stomatal closure and a decreased ci/ca ratio). The 13C value is thus indicative of changes in photosynthesis and stomatal conductance in response to environmental

*e-mail: sevdaturkis@gmail.com
Tel.: +1-111-111-111; Fax: +1-111-111-112.

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

changes. A value of 13C is accepted as an indicator of water scarcity [2]. Since plant tissue is known to contain fewer carbon 13 isotopes than ambient air, studies of 13C sequestration by plants have become a powerful tool in plant ecology and global carbon cycle studies [3]. The isotope content of leaf tissue can give us predictions about plant water relationships [4].

Plant responses can only be modelled successfully when species are divided into life groups [5]. The most common and simplest method of classifying plants is by life forms. Raunkiaer proposed the term biological spectrum for plant life forms [6]. According to Raunkiaer (1934), the biological spectrum is an indicator of climates and the environment in general. Phanerophytes constitute more than 60% of tropical flora. In Mediterranean countries there are 30% phanerophytes and 40% therophytes, in temperate regions there are 50% hemicryptophytes, 30% cryptophytes and 20% therophytes. At the poles, phanerophytes and therophytes disappear and haemicryptophytes become dominant (60%). In general, the classification of plants is based on characteristics that are easy to identify but ineffective against environmental changes [7]. Ehleringer & Cooper (1988) found in their study that water use efficiency varies with life span: short-lived shrubs have lower water use efficiency than long-lived shrubs [8]. Ehleringer (1994) suggested that in desert environments, perennials that persist through multiple droughts should have lower hydraulic conductivity than short-lived annuals [9]. They also noted that life form diversity is positively correlated with the climatic diversity of a site and indicate some forms. Leaf carbon isotope ratio (δ13C) and carbon isotope partitioning (Δ) give information about how a plant regulates carbon dioxide and water fluxes in relation to its ratio to intracellular CO_2 (ci/ca) [10]. In addition, Δ is more stable over time than absolute fluxes, thus providing a better indication of whole-plant constraints rather than environmental constraints. For example, Cernusak (2020) examined how δ13C changes in plants along a rainfall gradient and found that the δ13C of a community reflects the long-term average of rainfall for that region [11]. Schubert et al. (2018) predicted that vegetation type would significantly influence carbon isotope discrimination on a global basis [12]. He et al. (2017) used carbon isotope ratios to examine elevational and longitudinal effects on plant physiological processes for a wide range of plants worldwide and found that δ13C values varied with elevation and latitude [13]. Natural resources and biodiversity are decreasing due to land use increase, human population pressure, and climate change. Over the last fifteen years, carbon isotopic analyses have been used as an effective tool to investigate the distributions of C3 and C4 photosynthetic pathways within different plant species [14]. Some researchers suggested that carbon isotope ratios in C3 plants indicate the average intercellular CO_2 concentration [15, 16]. There are studies that support this view [4, 9, 17]. Yenice Forests, located in the northwestern part of Anatolia, is one of nine different biodiversity hotspots in Turkey. These forests need to be protected, especially to protect monumental trees, deep canyons, in-forest streams and the rich animal and plant diversity they contain, old forests, and endemic species. Within the scope of the study, it is aimed to determine the C13 and O18 isotope ratios of plant species belonging to different life forms in different forest types in Yenice Forests hot spot, to determine water use efficiency and carbon and water relationships in forest ecosystems.

Materials and Methods

Karabük-Yenice area is located in the Western Black Sea Subregion of the Black Sea Region as a geographical area, between 41°00'14"-41°05'06" northern latitude and 32°21'06"-32°27'45" eastern longitude. Plant samples were collected from 3 different forest areas selected in Yenice Forests between 2017- 2018 vegetation seasons. Plant species collected from the field were identified according to the book titled "Flora of Turkey" by Davis [18]. Life-forms of the plant samples were defined according to the classification made by Raunkiaer (1934) on main life-forms and subdivisions [6]. Climate related data from the area were acquired from the climate station (TFA 35.1075) situated in the forest, collecting average forest temperatures and average precipitation of the forestland between 2017- 2018. Based on the data obtained, the climatic characteristics of the forest were determined according to the Thornthwaite formula (Table 1, Fig. 1). According to Thornthwaite, in addition to the amount of precipitation and temperature that play a role in precipitation efficiency, it is important to graphically show the precipitation efficiency with factors such as the water accumulation capacity of the soil and the latitude of the geographical region. In addition, the determination of the water balance of a site by this method provides the determination of important climatic characteristics of the study area [19]. Thornthwaite Drought Index (Ia) is calculated using the formula Ia=100d/n (d: annual water deficiency cm; n: annual PE cm) to measure the degree of drought in humid areas [20]. Annual PE (cm) in Yenice forests was calculated as 649.63 cm and annual water deficit was calculated as 39.22 cm. Accordingly, Yenice Forests represent the Megathermal climate type as climate type, and the drought index of 6.03 represents the area type with little or no water deficiency. The annual precipitation regime covers the winter, spring, autumn and summer seasons: The Eastern Mediterranean Precipitation Regime (type 2) prevails within the forest. We used the Emberger formula to determine drought intensity. Although biogeographically the flora of the area is in the auxin province of the European Siberian phyloristic zone, large forest locations under the influence of the Mediterranean climate can be found along microclimatic areas within the forest.

Stable Isotope Analyses

Carbon and oxygen stable isotope ratios of leaf dry matter were measured at the Stable Isotope Laboratory of Siirt University, Technology Application Laboratory. For

Table 1. Water balance of Yenice Forests according to Thornthwaite method.

	1	2	3	4	5	6	7	8	9	10	11	12
Monthly Average Temperature	2	2	5,9	7,7	14,8	16	19,6	20,6	13	12,1	8,2	5,8
Temperature Index	0,25	0,25	1,28	1,92	5,17	5,82	7,91	8,53	4,25	3,81	2,11	1,25
Uncorrected PE	6,63	6,63	23,43	31,97	68,53	75,05	95,11	100,8	58,9	54,17	34,4	22,97
corrected PE	5,57	5,5	24,13	35,49	84,98	93,81	120,79	118,94	61,26	52	28,55	18,61
Monthly Total Precipitation	81,5	72	89	86	78,5	90,1	57,8	52,9	71,7	76,5	59,4	73,2
Monthly Change of Accumulated Water	0	0	0	0	-6,48	-3,71	-62,99	-26,82	10,44	24,5	30,85	34,21
Accumulated Water	100	100	100	100	93,52	89,81	26,82	0	10,44	34,94	65,79	100
True Evapotranspiration	5,57	5,5	24,13	35,49	84,98	93,81	120,79	79,72	61,26	52	28,55	18,61
Lack of Water	0	0	0	0	0	0	0	39,22	0	0	0	0
Excess Water	75,93	66,5	64,87	50,51	0	0	0	0	0	0	0	20,38
Flow	48,16	71,21	65,69	57,69	25,26	0	0	0	0	0	0	10,19
Humidity Rate	13,63	12,09	2,69	1,42	-0,08	-0,04	-0,52	-0,56	0,17	0,47	1,08	2,93

the 13C and 18O analyses, approximately 3 mg powder samples were combusted in an NC2500 elemental analyzer connected to a Delta Plus isotope ratio (δ13C and δ18O) mass spectrometer (FinniganMAT, Bremen, Germany) operating in continuous flow mode. Isotope ratio values are expressed as (‰).

Results and Discussion

Changes in carbon and oxygen isotope ratios were analyzed based on life form differences in forest layers in Yenice Forests (Table 2). In terms of life forms, ANOVA test results show significant differences at $p<0.05$ level for both carbon isotope ratios and oxygen isotope ratios. According to Tukey test, the groups causing the difference were 1st (hemicryptophyte) and 4th (geophyte) groups for carbon ratio and 1st (hemicryptophyte) and 5th (terophyte) groups for oxygen isotope ratio. In terms of these life forms in different forest habitats, the group with the highest carbon 13 isotope ratio was the geophyte life form with -27,38±2,27. The lowest carbon isotope ratio was the hemicryptophyte life form with -31,29± 0,98. The groups that showed a difference in terms of oxygen isotope 18O ratios were hemicryptophytes with the lowest amount (19,58±2,32) and therophyte life form with the highest amount (24,06±3,92) (Fig. 2, 3). When the areas were examined in terms of forest layers, significant positive correlations were found between the oxygen and carbon isotope ratios of the species in beech-hornbeam forest areas (0.819*) and pure oak forests (0.776*) at $p<0.05$ level. No correlation was detected in beech-fir mixed areas (Fig 4).

Stable carbon isotope ratios of terrestrial plants have the potential to provide unique insights into physiological processes and interactions between plants and the environment [21]. The study of stable isotopes of carbon, nitrogen, and oxygen composition provides

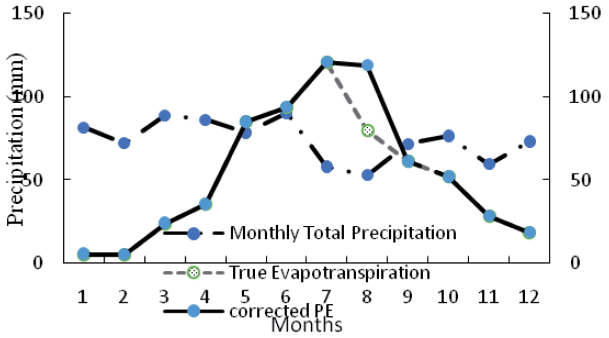

Fig. 1. Water balance graph of Yenice Forests according to Thornthwaite method.

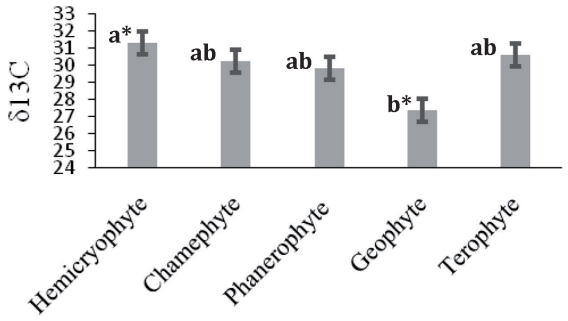

Fig. 2. Carbon isotope ratios of life forms in Yenice Forests.

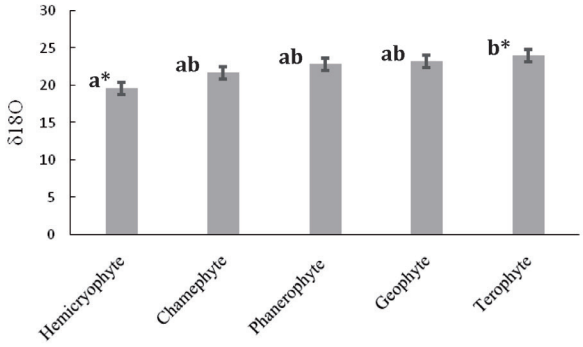

Fig. 3. Oxygen isotope ratios of life forms in Yenice Forests.

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

Table 2. Carbon isotope ratio, Oxygen isotope ratio, life form of plants from the Yenice Forests.

Family	Species	Life Form	Carbon isotope ratio (parts per thousand)	Oxygen isotope ratio (parts per thousand)
ACERACEAE	*Acer campestre*	Phanerophyte mes p scap	-31,098	21,266
ACERACEAE	*Acer platanoides*	Phanerophyte mes p scap	-31,041	22,289
APIACEAE	*Sanicula europae*	Hemicryptophyte scap	-32,255	19,911
APIACEAE	*Aeopodium podagraria*	Geophyte rhiz	-32,104	19,3425
ASPARAGACEAE	*Lilium pedicellare*	Geophyte bulb	-28,212	25,202
ASTERACEAE	*Anthemis aciphylla*	Hemicryptophyte scap	-31,63	15,783
ASTERACEAE	*Cirsium hypoglossum*	Hemicryptophyte	-31,468	21,662
BUXACEAE	*Buxus sempervivens*	Chamaephyte	-28,722	18,544
FABACEAE	*Trifolium pratense*	Hemicryptophyte scap	-32,006	21,183
FABACEAE	*Lathyrus laxiflorus*	Hemicryptophyte rept	-30,923	21,791
FAGACEAE	*Quercus petrea* subsp. *petrea*	Phanerophyte scap	-29,791	22,001
HYPERIDACEAE	*Hypericum bithynicum*	chamaephyte	-31,951	17,727
LABIATAE	*Stachys thirkei*	Hemicryptophyte scap	-29,733	21,578
MALVACEAE	*Tilia rubra*	Phanerophyte	-27,764	22,822
ONAGRACEAE	*Circae lutetiana*	Geophyte rhiz	-30,482	22,781
ORCHIDACEAE	*Cephalanthera longifolia*	Geophyte rhiz	-27,985	22,036
ORCHIDACEAE	*Cephalanthera rubra*	Geophyte rhiz	-25,118	24,462
ORCHIDACEAE	*Epipactis pontica*	Geophyte rhiz	-25,146	25,512
POACEAE	*Bromus sp.*	Terophyte t scap	-33,07	22,733
POACEAE	*Melica uniflora*	Hemicryptophyte scap	-28,138	26,099
ROSACEAE	*Rubus hirtus*	Chamaephyte	-30,934	15,912
ROSACEAE	*Mespilus germeniaca*	M Phanerophyte scap	-29,581	21,971
ROSACEAE	*Crataegus pentagyna*	Chamaephyte	-29,383	21,77
RUBIACEAE	*Galium odoratum*	Hemicryptophyte scap	-29,555	25,052
SCROPHULARIACEAE	*Veronica leoicarpa*	Hemicryptophyte scap	-31,237	20,763

a better understanding of the biogeochemical cycle of plant ecosystems [22]. To date, various techniques have been used to investigate the mechanisms and strategies adopted by plants. Numerous studies have used plant WUE (the ratio of carbon assimilated by the plant to water excreted by the plant) inferred from δ13C signatures (i.e. the 13C to 12C ratio) of plant leaves and other materials to analyze plant performance under varying water stress conditions [21, 23, 24]. The advantage of measuring carbon 13 isotope ratios in plant tissues is that it provides information on ci/ca values over time rather than instantaneous ci/ca estimation. Hussain et al. (2019) stated that the simultaneous analysis of carbon and oxygen isotope ratios of leaf material in tropical rainforests is important for long-term information on assimilation and evapotranspiration regimes for plants [22]. Transpiration is a critical physiological mechanism involved in water conservation strategies and allows plants to grow and survive [25].

It has been reported that the forest understory is more humid and has higher levels of respirable carbon dioxide than the forest top [22]. Within the scope of this study carried out in different forest habitats, the change in isotope values for plant species belonging to different life forms in different forests was investigated. In this study, highly positive correlations ($p<0,05$) were found between C13 and O18 isotope ratios in beech-hornbeam and pure oak forest habitats. There was no correlation between C13 and O18 isotope ratios in beech-fir forests. Plants growing under high humidity have about 2.7% more negative C13 values than those growing under low humidity due to higher stomatal conductance [22]. This condition has led to correlations between relative humidity and the uptake of isotopically spent carbon dioxide in different forest habitats [13]. Similar results were also observed in our study. Different forest habitats allow variation in soil moisture content due to different biochemical and morphological characteristics of different species of trees. Beech-hornbeam, oak, and beech-fir have the highest variation in moisture content, respectively. Forests with a positive correlation were found in habitat areas with high moisture content. The impact of global changes on forest ecosystem processes depends on the species-specific responses of trees to the combined effect of multiple stressors and the capacity of each species to adapt and cope with environmental change [26]. Furthermore, seasonal cycles linked to different leaf phenology, as well as the presence of periods of stress (e.g. summer drought) during the growing season, can also lead

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

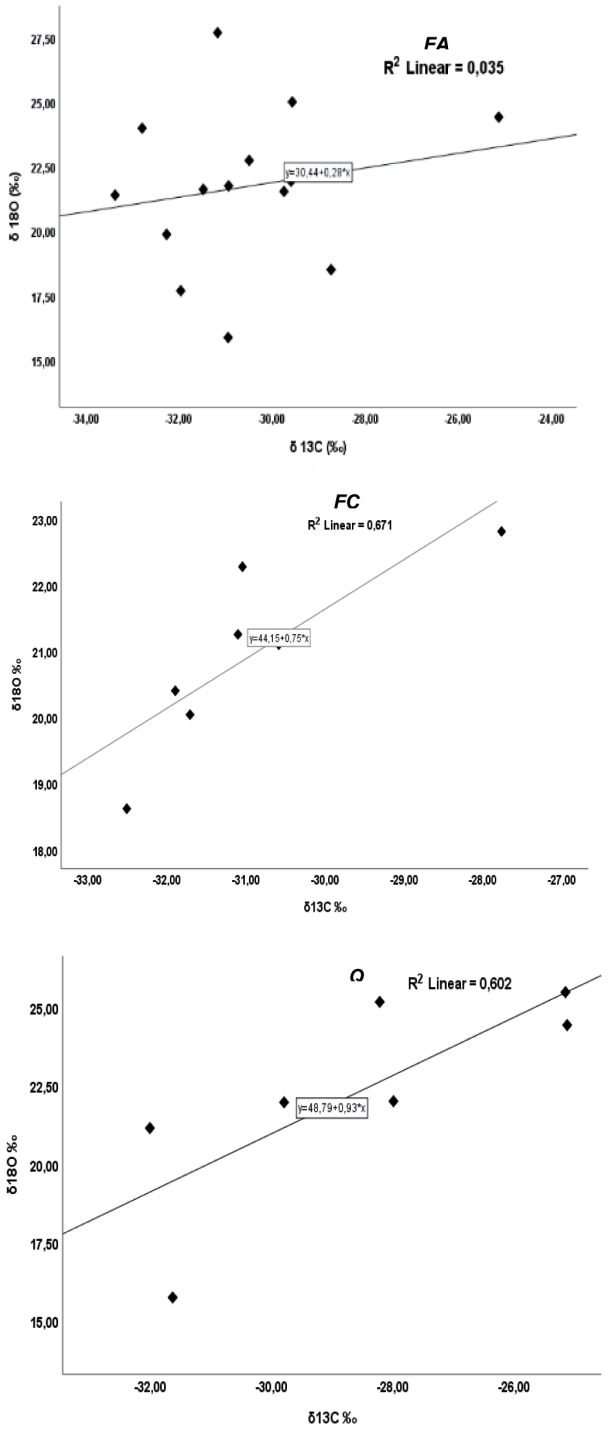

Fig. 4. Relationship between carbon (δ13C) and oxygen (δ18O) isotope ratios for different forest layers in Yenice Forests. FA: Fagus--Abies Forest; FC: Fagus-Carpinus Forest; Q: Quercus Forests

to vastly different rates of dependence on stored carbon [27-29]. The rapid development of deep roots seems to be an important strategy for evergreen tree species to obtain water during the dry season, while deciduous trees during drought minimize water loss from the remaining leaves, in addition to losing some of their leaves [30, 31].

Determination of the carbon isotopic ratios of leafs have proved useful in ecological research because they can provide insight into long-term carbon-water flux rates integrated over the entire lifetime of a leaf or needle. All plant water relationships can be studied by analyzing carbon isotope abundances in leaf tissue [10]. Plants experiencing water stress typically close their stomata to minimize water loss through transpiration, thus reducing the inner CO2 ratio (ci/ca). Under conditions of reduced ci/ca, plants are largely indiscriminate towards 13C during photosynthesis, and thus plant tissue becomes relatively enriched in 13C. It is stated that a heavier δ value under stress indicates stomatal closure for almost all species [32].

The implication of this for water resource studies is that abundant and reliable water sources (e.g., soil water in the upper layers) are likely to become enriched in 13C as drought conditions progress. Reducing the internal CO2 ratio (ci/ca) is the reason why stomata are closed to reduce water lost through transpiration. Under reduced ci/ca conditions, the 13C used in photosynthesis increases [1]. There is an increase in C13 when distress conditions occur with water [32.]. Within the scope of the study, the highest 13C isotope ratios were found in geophyte species in different forests. This clearly shows the reason why the large genome size and increased stomatal size in the geophyte life form forms a measure against drought even in humid temperate forests. In other words, geophyte species show sensitivity (measure of protection) to drought in tolerance ranges narrower than other life forms. Raunkiaer (1934) selected buds according to their longevity [6]. This is the degree and measure of protection provided to meristematic tissues during the unfavorable season. Monitoring the variation of carbon isotope ratios according to life forms allows the identification of the most sensitive groups within the areas. Thus, it is possible to predict the plant groups that will be first affected by global climate change in the three major different temperate humid forests in Yenice Forest, which is defined as a hot spot. Therefore, the fact that geophytes keep the carbon 13 isotope ratio high makes them indicator groups. There was a significant difference between the 18O oxygen isotope ratio at the $p<0.05$ level in terms of therophytes. The therophyte life group sample *E. farctus* has determined values with low oxygen isotope ratio in species [33]. Similar results in our study also revealed low oxygen isotope values as an indicator of low water use efficiency, since the use of surface water is dependent on surface water. The frequency of therophytes existing in a given area manifests itself after the completion of the vegetative developmental stage in the short period before the onset of the dry period [34]. This is an indication that Δ13C variation in the same growth conditions may also be due to genotypic differences [32].

Plant stem growth is achieved by the rate of cell division and rapid growth of the geophytic stem by pumping water into the non-photosynthetic subterranean divided tissues. Geophytes overcome unfavorable growth conditions by persisting in the form of an underground organ capable of accumulating sufficient nutrients available for the rapid development of the aboveground body. The reserves stored during annual cycling periods in

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy

favorable conditions [35, 36] enable geophytes to develop successfully even at very low initial external energy supply. This feature provides them with a favorable strategy in steppe vegetation in the seasonally arid Mediterranean climate or in temperate, deciduous forests where the light phase is limited to cold seasons characterized by a low initial level of sunlight. The genomes of geophytes are very large, and their evolution is closely related to specific life forms. Since a large genome leads to a large stomatal structure, the regulation of carbon dioxide uptake by stomata is related to stomatal length, as large stomata are known to increase plant sensitivity to drought [37]. In higher plants, stomatal guard cell length is known to be very closely related to genome size [38]. Life forms are closely related to environmental factors, and this is interpreted as a strategy for acquiring resources [39]. Life forms reflect the adaptation to climate and distribution of plants and translate these forms into micro and macro climate indicators [40]. This means that they are the group that uses water most economically, even if there are no restrictions on the use of water in the environment, and this group actually proves efficient water use with the type of climate it represents. Soil C models are mainly driven by soil temperature and moisture. These factors are generally excellent predictors of soil respiration rate R over short temporal scales and under certain conditions [41]. However, over larger spatio-temporal scales, plant activity and other factors may become increasingly important. Therefore, taking advantage of plant-soil relationships in further studies may help to accurately represent seasonal variation in plant C allocation among different functional groups, by modelling regional or global scales over longer time scales [42]. Climate change forecasts in Europe are unanimous in predicting significant warming for every season. The most important impact of climate change is that it affects the water cycle by changing precipitation patterns and evapotranspiration processes at various scales.

In the coming years, this indicates that less soil water will be available for plant growth in forest interior areas during the summer months [43, 44].

Conclusions

Determining the life forms that can exhibit the most strategic behavior against the decreasing amount of water in climate change is important in terms of the measures that can be taken. Determination of isotope ratios preferred by life forms can be an indicator for sensitive areas such as Yenice Hotspot forests in climate change monitoring.

Acknowledgements

I would like to thank Mesut Gök for her unwavering support for the analysis at Siirt University Technology Application Laboratory.

References

1. FARDUSI M.J., FERRIO J.P., COMAS C., VOLTAS J., DE DIOS V.R., SERRANO L. Intra-specific association between carbon isotope composition and productivity in woody plants: a meta-analysis. Plant Science, **251**, 110, **2016.**
2. SIEGWOLF R.T., LEHMANN M.M., GOLDSMITH G.R., CHURAKOVA O.V., MIRANDE-NEY C., TIMOVEEVA G., WEIGT R.B., SAURER M. Updating the dual C and O isotope—Gas-exchange model: A concept to understand plant responses to the environment and its implications for tree rings. Plant, Cell & Environment, **46** (9), 2606, **2023**.
3. GAT J., MOOK W.G., MEIJER H.A.J. Environmental Isotopes in the Hydrological Cycle: Principles and Applications. Volume II: Atmospheric Water. International Atomic Energy Agency, **2001.**
4. FARQUHAR G.D., BALL M.C., VON CAEMMERER S., ROKSANDIC Z. Effect of salinity and humidity on δ 13C value of halophytes—evidence for diffusional isotope fractionation determined by the ratio of intercellular/ atmospheric partial pressure of CO2 under different environmental conditions. Oecologia, **52**, 121, **1982.**
5. BOX E.O. Factors determining distributions of tree species and plant functional types. Vegetatio, **121,** 101, **1995.**
6. RAUNKIAER C. The Life Forms of Plants and Statistical Plant Geography. Oxford University Press, **1934.**
7. SCHULZE E.D. Plant life forms and their carbon, water and nutrient relations. In Physiological Plant Ecology II. Encyclopedia of Plant Physiology; Lange O.L., Nobel P.S., Osmond C.B., Ziegler H., Springer, Berlin, Heidelberg, Volume **12,** pp. 615, **1982.**
8. EHLERINGER J.R., COOPER T.A. Correlations between carbon isotope ratio and microhabitat in desert plants. Oecologia, **76**, 562, **1988.**
9. EHLERINGER J. Annuals and perennials of warm deserts. In Physiological Ecology of North American Plant Communities; Chabot B.F., Mooney H.A. Springer Dordrecht, Netherlands, pp. 162, **1985.**
10. WANG A., SIEGWOLF R.T., JOSEPH J., THOMAS F.M., WERNER W., GESSLER A., RIGLING A., SCHAUB M., SAURER M., LI M., LEHMANN M.M. Effects of soil moisture, needle age and leaf morphology on carbon and oxygen uptake, incorporation and allocation: a dual labeling approach with 13CO2 and H218O in foliage of a coniferous forest. Tree Physiology, **41 (**1), 50, **2021.**
11. CERNUSAK L.A. Gas exchange and water-use efficiency in plant canopies. Plant Biology, **22,** 52, **2020.**
12. SCHUBERT B.A., JAHREN A.H. Incorporating the effects of photorespiration into terrestrial paleoclimate reconstruction. Earth-Science Reviews, **177**, 637, **2018.**
13. HE Z., ZHAN S., WANG W., HU L., WU S. Different patterns of changes in foliar carbon isotope composition along altitude. Polish Journal of Ecology, **65** (2), 227, **2017**.
14. XIE Q., HUETE A., HALL C.C., MEDLYN B.E., POWER S.A., DAVIES J.M., MEDEK M.D., BEGGS P.J. Satellite-observed shifts in C3/C4 abundance in Australian grasslands are associated with rainfall patterns. Remote Sensing of Environment, **273,** 112983, **2022.**
15. FARQUHAR G.D., O'LEARY M.H., BERRY J.A. On the relationship between carbon isotope discrimination and the intercellular carbon dioxide concentration in leaves. Functional Plant Biology, **9** (2), 121, **1982.**
16. SCHUBERT B.A., JAHREN A.H. Incorporating the effects of photorespiration into terrestrial paleoclimate reconstruction. Earth-Science Reviews, **177**, 637, **2018.**

17. HAN K., ZHANG J., WANG C., YANG Y., CHANG Y., GAO Y., LIU Y., XIE J. Changes in growth, physiology, and photosynthetic capacity of spinach (Spinacia oleracea L.) under different nitrate levels. Plos one, **18** (3), e0283787, **2023**.
18. DAVIS P.H. Flora of Turkey and the East Aegean Islands.; Ediburgh University Press: Edinburgh, The United Kingdom, Volume **1-10, 1965-1988**.
19. KILINÇ M, KUTBAY H.G., YALÇIN E, BILGIN A. Bitki Ekolojisi ve Bitki Sosyolojisi Uygulamaları. Palme Yayincilik: Ankara, Turkey **2006** [In Turkish].
20. YILMAZ E. Türkiye'de Thornthwaite iklim indislerindeki eğilimler. Coğrafya Dergisi, **40**, 163, **2020** [In Turkish].
21. CERNUSAK L.A., UBIERNA N. Carbon isotope effects in relation to CO2 assimilation by tree canopies. In Stable Isotopes in Tree Rings. Tree Physiology; Siegwolf R.T.W., Brooks J.R., Roden J., Saurer M., Eds., Springer, Cham, Volume **8**, 291, **2022**.
22. HUSSAIN M.I., EL-KEBLAWY A., MITTERAND TSOMBOU F. Leaf age, canopy position, and habitat affect the carbon isotope discrimination and water-use efficiency in three C3 leguminous Prosopis species from a hyper-arid climate. *Plants*, **8** (10), 402, **2019.**
23. NIE Y., CHEN H., WANG K., DING Y. Seasonal variations in leaf δ13C values: implications for different water-use strategies among species growing on continuous dolomite outcrops in subtropical China. Acta Physiologiae Plantarum, **36**, 2571, **2014.**
24. DING Y., NIE Y., CHEN H., WANG K., QUEREJETA J.I. Water uptake depth is coordinated with leaf water potential, water-use efficiency and drought vulnerability in karst vegetation. New Phytologist, **229** (3), 1339, **2021**.
25. MUÑOZ-VILLERS L.E., HOLWERDA F., ALVARADO-BARRIENTOS M.S., GEISSERT D.R., & DAWSON T.E. Reduced dry season transpiration is coupled with shallow soil water use in tropical montane forest trees. Oecologia, **188** (1), 303, **2018.**
26. COCOZZA C., PAOLETTI E., MRAK T., ZAVADLAV S., LEVANIČ T., KRAIGHER H., GIOVANNELLI A., HOSHIKA Y. Isotopic and water relation responses to ozone and water stress in seedlings of three oak species with different adaptation strategies. Forests, **11** (8), 864, **2020.**
27. ROG I., JAKOBY G., KLEIN T. Carbon allocation dynamics in conifers and broadleaved tree species revealed by pulse labeling and mass balance. Forest ecology and management, **493**, 119258, **2021**.
28. KLEIN T. Carbon allocation dynamics in Mediterranean pines under stress. Pines and Their Mixed Forest Ecosystems in the Mediterranean Basin, 117, **2021**.
29. LÜTTSCHWAGER D., JOCHHEIM H. Drought primarily reduces canopy transpiration of exposed beech trees and decreases the share of water uptake from deeper soil layers. Forests, **11** (5), 537, **2020.**
30. ADAMS R.E., WEST J.B. Functional Groups Mask Inter- and Intraspecific Variation in Water Use Strategies in a Seasonally Dry Tropical Forest. Frontiers in Water, **4**, 950346, **2022.**
31. SANTIAGO L.S., SILVERA K., ANDRADE J.L., DAWSON T.E. Functional strategies of tropical dry forest plants in relation to growth form and isotopic composition. Environmental Research Letters, **12** (11), 115006, **2017.**
32. GOUVEIA C.S., GANANÇA J.F., SLASKI J., LEBOT V., DE CARVALHO M.Â.P. Variation of carbon and isotope natural abundances (δ15N and δ13C) of whole-plant sweet potato (Ipomoea batatas L.) subjected to prolonged water stress. Journal of Plant Physiology, **243**, 153052, **2019**.
33. ZUNZUNEGUI GONZÁLEZ M., ESQUIVIAS SEGURA M.D.L.P., GALLEGO FERNÁNDEZ J.B. Spatial and seasonal patterns of water use in Mediterranean coastal dune vegetation. Plant and Soil, **477** (1-2), 807, **2022.**
34. HASSANI S.M, YAZDANSHENAS H., NAZARPOOR FARD K., BASSIRI R., PUR REZAEE J. Study of physiognomy and origin of plant species in Sarshiv area of Marivan, Iran. Journal of Rangeland Science, **4** (4), 270, **2014.**
35. POPOVIĆ Z., VIDAKOVIĆ V. Ecophysiological and Growth-Related Traits of Two Geophytes Three Years after the Fire Event in Grassland Steppe. Plants, **11** (6), 734, **2022.**
36. TRIBBLE C.M., MARTÍNEZ-GÓMEZ J., HOWARD C.C., MALES J., SOSA V., SESSA E.B., CELLINESE N., SPECHT C.D. Get the shovel: morphological and evolutionary complexities of belowground organs in geophytes. American Journal of Botany, **108** (3), 372, **2021**.
37. VESELÝ P., ŠMARDA P., BUREŠ P., STIRTON C., MUASYA A.M., MUCINA L., HOROVÁ L., VESELÁ L., ŠILEROVÁ A., ŠMERDA J., KNÁPEK O. Environmental pressures on stomatal size may drive plant genome size evolution: evidence from a natural experiment with Cape geophytes. Annals of Botany, **126** (2), 323, **2020**.
38. KROGMEIER K.R. Impacts of Polyploidy and Subsequent Natural Selection on the Ecophysiology of Solidago Altissima (Doctoral dissertation, Appalachian State University), **2021.**
39. VOLAIRE F.A unified framework of plant adaptive strategies to drought: crossing scales and disciplines. Global change biology, **24** (7), 2929, **2018.**
40. TÜRKİŞ S., ELMAS E. Safranbolu-Tokatlı Kanyonu'nun Floristik Kompozisyonu, Hayat Formu ve Korotip Özellikleri. Kahramanmaraş Sütçü İmam Üniversitesi Tarım ve Doğa Dergisi, **21** (6), 817, **2018** [in Turkish].
41. DAVIDSON A. Permafrost and wetland carbon stocks. Science, **330** (6008), 1176, **2010**.
42. NYBERG M., HOVENDEN M.J. Warming increases soil respiration in a carbon-rich soil without changing microbial respiratory potential. Biogeosciences, **17** (17), 4405, **2020**.
43. VAR DER LINDEN E.C., HAARSMA R.J., VAN DER SCHRIER G. Impact of climate model resolution on soil moisture projections in central-western Europe. Hydrology and Earth System Sciences **23**, 191, **2019.**
44. CSÁKI P., GYIMÓTHY K., KALICZ P., SZOLGAY J., ZAGYVAINÉ K.K.A., GRIBOVSZKI Z. Multi-model climatic water balance prediction in the Zala River Basin (Hungary) based on a modified Budyko framework. Journal of Hydrology and Hydromechanics **68** (2), 200, **2020.**

Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy • Author Copy