Acta Physiol Plant (2017) 39:191
DOI 10.1007/s11738-017-2489-3

ORIGINAL ARTICLE

Males and females of *Juniperus communis* L. and *Taxus baccata* L. show different seasonal patterns of nitrogen and carbon content in needles

Kinga Nowak-Dyjeta[1] · **M. J. Giertych**[1,2] · **P. Thomas**[3] · **G. Iszkuło**[1,2]

Received: 28 December 2016 / Revised: 3 March 2017 / Accepted: 24 July 2017 / Published online: 1 August 2017
© The Author(s) 2017. This article is an open access publication

Abstract Genders of dioecious species often show secondary sexual dimorphisms (not directly related to the sex organs), which may be related to reproductive demand for resources during the year. Our working hypothesis stated that phenology influences yearly sex-specific pattern of foliar nitrogen concentration in dioecious species. The concentration of carbon (C) and nitrogen (N) of last year's needles (on part of twigs where strobili are bearing) was measured in 1-month intervals from March to November in *Taxus baccata* L. and *Juniperus communis* L. separately for males and females. Seasonal C concentration of needles was unrelated to gender, probably due to storage elsewhere in the plants. Needles of *J. communis* females had higher N concentration in March and April but this dropped to the same level as males after flowering and vegetative growth began. This suggests that females of *J. communis,* a species of N-poor habitats, have a long-term strategy of N storage. Nitrogen concentration was not different between the sexes during the rest of the growing season. In *T. baccata*, N concentration was higher in males throughout the analysed period, but the highest differences were in the period of intensive shoot elongation and radial growth. The high demand for N in the period of intensive growth and female cone maturation may result in restrictions in the vegetative growth of females. The results indicate that the time (phenology) and species-specific strategy significantly affect the concentration of N in females and males.

Keywords Dioecy · Sexual dimorphism · Phenology · N storage

Introduction

The carbon (C) and nitrogen (N) contents of plant tissues change during the growing season in relation to resource uptake, storage and mobilization according to seasonal demand, combined with variability of resource availability in the environment (Millard and Grelet 2010). Carbon is stored throughout a tree (Millard and Grelet 2010) but N is stored in specific tissues and organs. Deciduous trees store N in stems, roots or wood, but evergreen trees store N mostly in the youngest needles (Dickson 1989; Millard et al. 2001; Pallardy 2008; Teitel et al. 2016). Storage of N in the leaves of evergreen trees is carried out after the completion of leaf growth. These resources are then mobilized from the start of the next growing season (Dickson 1989).

Resources (including C and N) are divided between three main functions: maintenance (including defence), growth and reproduction. These functions are often carried out at the same time and, because an organism has a limited supply of resources, allocation to one function leads to a decreased allocation to others (Levins 1968; Obeso 2002). The proportion of investment to each of these three basic functions in woody plants depends on many factors including plant age (young plants invest mostly in fast growth and defence, older plants in reproduction) and habitat richness. Most organisms on reaching maturity stop

---

✉ Kinga Nowak-Dyjeta
knd@man.poznan.pl

[1] Institute of Dendrology, Polish Academy of Sciences, Parkowa 5, 62-035 Kórnik, Poland

[2] Faculty of Biological Sciences, University of Zielona Góra, Prof. Z. Szafrana 1, 65-516 Zielona Góra, Poland

[3] School of Life Sciences, Keele University, Staffordshire ST5 5BG, UK

allocating resources to growth and start to allocate all the surplus energy (i.e. that not used for maintenance) to reproduction (Kozłowski and Wiegert 1986, 1987). Trees are an exception to this principle since after reaching sexual maturity, resources are still shared between primary and secondary growth and reproduction (Koenig and Knops 1998). In the case of dioecious trees with sexual dimorphism there may, therefore, be different strategies of resources allocation in males and females. This is a consequence of differences in life-history, morphological or functional traits (Agren et al. 1999; Dawson and Geber 1999; Delph 1999). Females and males have different strategies of resource investment to maximize reproductive success when resources are limited (Obeso 2002). Females usually support higher reproductive effort than males (e.g. Obeso 2002; Montesinos et al. 2006) and consequently allocate more resources to reproduction resulting in slower radial growth after reaching sexual maturity (Obeso 2002; Banuelos and Obeso 2004; Montesinos et al. 2006; Chen et al. 2014). Females are also often more demanding of habitat requirements (Obeso 2002; Chen et al. 2014) which may be a reason behind the spatial segregation of the sexes in some species (Bierzychudek and Eckhart 1988; Dawson and Ehleringer 1993; Matsushita et al. 2016). Females can, however, compensate for the higher reproductive investment (Dawson and Ehleringer 1993; Obeso 2002; Tozawa et al. 2009; Jiang et al. 2016). They can show greater gas exchange per unit area and mass in comparison to males (Dawson and Ehleringer 1993; Obeso 2002; Montesinos et al. 2012) and increase photosynthesis by, for example, increasing leaf area (Wallace and Rundel 1979; Meagher 1992; Kohorn 1994).

Carbon demand increases at the time of formation and maturation of reproductive structures (McDowell et al. 2000). However, so far, no significant differences between C concentration between males and females have been found (Chen et al. 2014; Jiang et al. 2016). For example, no seasonal difference was found in non-structural carbohydrate concentration between males and females in *Juniperus thurifera* (DeSoto et al. 2016).

Variable differences in N concentration have, however, been found within dioecious species. The N concentration of *Rubus chamaemorus* leaves was found to be higher in males, particularly within 2–3 weeks after flowering (Agren 1988). *Nyssa sylvatica* stems were analysed four times a year and these studies demonstrated a significant effect of gender on N concentration. At fruit maturation (September) the concentration of N was significantly higher in males than females but only in one of two sites investigated (Cipollini and Stiles 1991). Lower N concentration was also demonstrated in females of *Populus cathayana* (Chen et al. 2014) and *Acer negundo* (Dawson and Ehleringer 1993). In other studies where the level of N was examined once in the season, no differences were usually found between the sexes (Agren et al. 1999; Jiang et al. 2016). However, the C and N concentration depends on the stage of plant growth and development; thus, the lack of differences seen in these studies may be due to analyses being performed just once a year. This is confirmed by a study of *Honckenya peploides* by Vilas and Retuerto (2016) who found a decrease of nitrogen concentration in females during growing season, whereas in males, the decrease was noted only from April to June.

In *J. thurifera*, Montesinos et al. (2012) claimed that females had a long-term strategy to increase N storage to compensate for massive reproductive masting events (synchronous production of many seeds at periodic intervals) because they found that an increase in shoot N was associated with increased photosynthesis in males but not in females. This suggests that females store surplus N while males are more reactive to current nutrient availability.

Although there have been a number of studies on *J. thurifera*, other gymnosperms and evergreen species have been little studied. European yew (*Taxus baccata* L.) and common juniper (*Juniperus communis* L.) are appropriate models to study dioecious species because males and females have been found to respond differently to their environments. Males of *T. baccata* grow taller (Iszkuło et al. 2009) and both *T. baccata* and *J. communis* males show greater radial growth than females after the beginning of sexual maturity (Cedro and Iszkuło 2011; Iszkuło and Boratyński 2011). Females of *T. baccata* have compensatory mechanisms allowing higher reproductive investment in the form of a greater area of needles and stomatal density, and these traits are highly correlated with gas exchange intensity (Iszkuło et al. 2009; Iszkulo et al. 2013). *T. baccata* and *J. communis* are evergreen gymnosperm species. Since evergreen species are known to store N in the youngest needles, the level of this element may be particularly sensitive to differences in phenology and different male and female reproductive functions. Analyses carried out throughout the growing season are still rare and, therefore, it is not known how the C and N concentration changes during the year in dioecious plants and whether indeed females have a different strategy of N-storage than males. If so, is it a general strategy of dioecious plants or is it limited to species growing under conditions of N deficiency? Therefore, the stated hypotheses are (1) phenology influences the sex-specific pattern of yearly foliar nitrogen concentration; (2) *T. baccata* and *J. communis* show the same sex-specific pattern of foliar nitrogen concentration.

Materials and methods

Study species

Juniper and yew have common features (gymnosperms, dioecy, evergreen, anemophily, zoochory), and yet have significant reproductive differences (Table 1). For example, yew strobili open in March, while juniper much later in May. Female cones of yew (seeds with arils) develop within one growing season, while juniper female cones ("juniper berries") mature over 2 or 3 seasons (Thomas and Polwart 2003; Thomas et al. 2007). These species also occur in different habitats; juniper is a pioneer species, occurring in dry and nutrient poor habitat while yew persists into late-successional stages where it grows in fertile habitats. Yew is an endangered species and certain woodland communities containing *T. baccata* have received priority habitat status EU Habitat Directive 92/43/EEC (Thomas and Polwart 2003; Thomas and Garcia-Marti 2015) although, as recently shown, juniper populations are also reducing (Thomas et al. 2007; Verheyen et al. 2009).

Study area and sampling

The study was conducted using *T. baccata* in the Kórnik Arboretum (52°14′27.93″N; 17°5′28.21″E) Poland and *J. communis* in the Torzym Forestry Commission land (52°18′36.93″N; 15°7′43.07″E), Poland. Juniper is a natural species in the latter area with a density of about 553 individuals per ha (Mandryk et al. unpublished data). They grow on poor, iron-rich soils (an outwash plain with sand and gravel) in a mixed oak–pine forest *Querco–Pinetum*. The yew population arose from spontaneous natural regeneration and forms the understorey in the Kórnik Arboretum on brown earth soils; there are about 2500 individuals >1 m tall (Giertych 2000). The potential vegetation is a rich, eutrophic variant of Central European lowland oak–hornbeam wood—*Galio sylvatici–Carpinetum* (Matuszkiewicz et al. 1995). Further details about this population are given in Iszkulo and Boratynski (2004, 2005).

For analysis, 20 individuals of yew (10 male and 10 female) and 20 individuals of juniper (10 male and 10 female) of similar height and diameter with visible reproductive structures (male or female strobili buds and/or female cones) were randomly chosen. Minimum distance between individuals was 3 m.

Sampling was repeated nine times in 2012, from March to November at intervals of 1 month. Phenological observations (beginning and end of flowering, growth, strobili maturation) were also made.

Twigs with last year's growth (determined using bud scale scars) were collected intact from the middle part of crown. A total of 30 needles from one tree were collected from last year's growth (2011 increment). Needles were removed from the twig; the area, length and width of fresh needles were measured using WinSeedle software with accuracy 0.0001 mm (Regent Inc.). Needles were then frozen before analysis. After thawing, needles were dried at 65 °C for 3 days and weighed to allow calculation of the specific leaf area (SLA: leaf area/leaf dry mass, $cm^2 g^{-1}$). Dried needles were finely ground and nitrogen (N) and carbon (C) concentrations in the needles were measured using an Elemental Combustion System CHNS-O (Costech Analytical Technologies Inc., Valentia, USA).

A multivariate approach (ANOVA, Obrien and Kaiser 1985) for repeated measures was used to analyse the morphological traits, and for N and C concentrations. All data were analysed using JMP 8.0 (SAS Institute Inc.).

Results

Morphological analysis

A significant effect of sex on the length of yew and juniper needles, and on the area of yew needles were found (Tables 2, 3). Needles of females were longer and had greater area in both species (Figs. 1, 2). These traits did not change significantly in each of analysed sampling dates (Tables 2, 3).

Table 1 Characteristics of yew and juniper

Species	*Taxus baccata*	*Juinperus communis*
Systematic group	Gymnosperm	Gymnosperm
Leaf life span	Evergreen	Evergreen
Mating system	Dioecious	Dioecious
Pollination system	Anemophily	Anemophily
Seed dispersal	Zoochory	Zoochory
Succession specialization	Late-successional	Pioneer
Female cones life span	One year	Two–three years
Strobili opening ("flowering")	Before the new leaves	Simultaneously with the new leaves

Table 2 ANOVA repeated measures analysis of *J. communis* needle area, length and specific leaf area (SLA), nitrogen (N) and carbon (C) content against time effect and sex

Source of variation	Needle length			Needle area			SLA			N			C		
	F	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*
Between subjects															
Sex	5.9590	1	**0.0259**	3.4072	1	0.0835	2.9908	1	0.1030	0.0064	1	0.9374	0.0137	1	0.6455
Within subjects															
Time	1.5129	8	0.2588	0.7513	8	0.6513	0.9970	8	0.4963	10.767	8	**0.0009**	12.472	8	**0.0005**
Time × sex	1.5060	8	0.2609	1.3623	8	0.3260	2.8534	8	0.0697	7.3779	8	**0.0035**	0.2278	8	0.9755

F F test statistic, *DF* degree of freedom, *P P* values in boldface indicate $P < 0.05$

Table 3 ANOVA-repeated measures analysis of *T. baccata* needle area, length and specific leaf area (SLA), nitrogen (N) and carbon (C) content against time effect and sex

Source of variation	Needle length			Needle area			SLA			N			C		
	F	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*	*F*	*DF*	*P*
Between subjects															
Sex	5.9818	1	**0.0256**	4.4955	1	**0.0490**	1.4207	1	0.2563	19.664	1	**0.0007**	1.9623	1	0.1847
Within subjects															
Time	2.6081	8	0.0753	2.2394	8	0.1159	1.9535	8	0.2389	46.187	8	**<0.0001**	6.158	8	**0.0199**
Time × sex	0.2235	8	0.9713	0.3171	8	0.9415	0.8118	8	0.6231	0.6228	8	0.7386	0.7312	8	0.668

F F test statistic, *DF* degree of freedom, *P P* values in boldface indicate $P < 0.05$

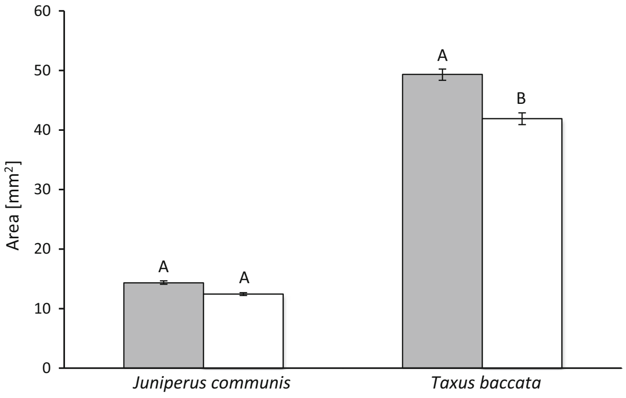

Fig. 1 Mean area (±SE) for the season (between March and November) of male and female *Taxus baccata* and *Juniperus communis* needles. *White bars* male individuals, *grey bars* female individuals. *Different letters* significant differences between male and female individuals (Student's *t* test)

No significant influence was found of sex and time on specific leaf area (SLA) (Tables 2, 3). Much higher average SLA was found in yew (31.03 $cm^2 g^{-1} \pm 0.43$ SE) compared with juniper (9.16 $cm^2 g^{-1} \pm 0.14$ SE).

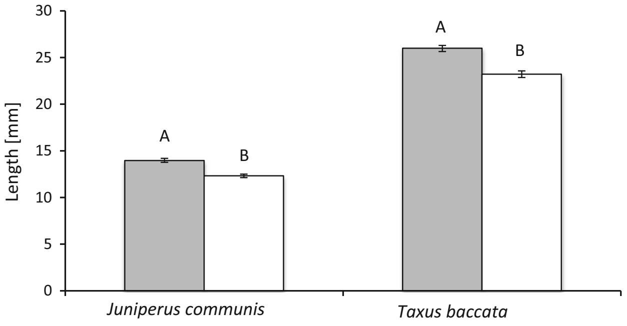

Fig. 2 Mean length (±SE) for the season (between March and November) of male and female *Taxus baccata* and *Juniperus communis* needles. *White bars* male individuals, *grey bars* female individuals. *Different letters* significant differences between male and female individuals (Student's *t* test)

Phenology

Both species began and ended vegetative growth at similar times (from mid-April to the end of July), but strobili opening of yew was observed before the development of vegetative growth (between 15 March and 6 April; Fig. 3)

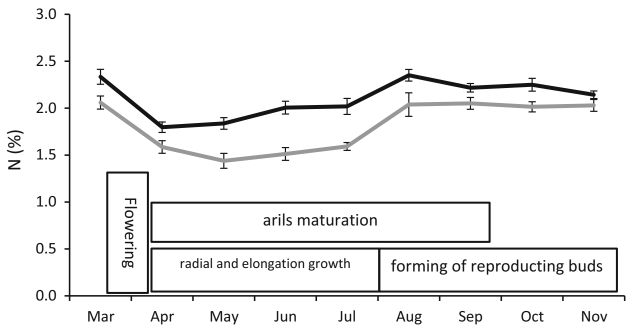

Fig. 3 Mean (±SE) annual nitrogen concentration of needles by gender in *Taxus baccata*. *Black line* male individuals, *grey line* female individuals

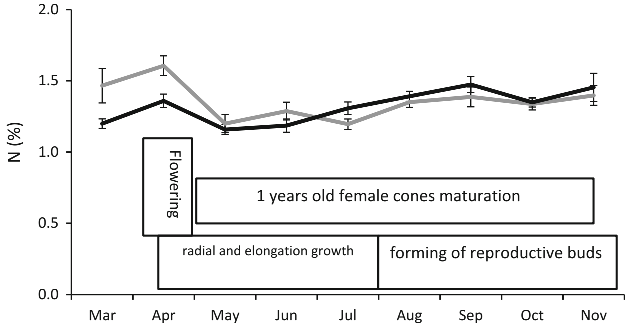

Fig. 4 Mean (±SE) annual nitrogen concentration of needles by gender in *Juniperus communis*. *Black line* male individuals, *grey line* female individuals

and in junipers the opening began in a similar period as the growth phase (between 6 April and 30 April; Fig. 4).

Nitrogen concentration

Yews were characterized by higher average levels of N (a mean of 1.92% ± 0.02 SE across all sampling dates) compared to the juniper (1.33% ± 0.01 SE). A significant effect of sex and time on the average of N concentration was found in yews (Table 3). Across all sampling dates the N concentration of yew needles was higher in males, with the largest differences shown in May, June and July (Fig. 3). In the needles of juniper, there was no overall effect of sex on the N concentration, but concentration varied significantly with time and with interaction of time and sex (Table 2). This was attributed to N concentration being higher in females in March and April just prior to flowering but not in the following months, and indeed after July there was a non-significant tendency for N to be higher in males (Fig. 4). In both species the N concentration of leaves decreased in early spring during the flowering period and then generally increased from May onwards as radial and elongation growth slowed and stopped (Figs. 3, 4).

Carbon concentration

The average C concentration in the needle was slightly higher in yew (49.4% ± 0.15 SE) compared with juniper (48.6% ± 0.11 SE). In neither species there was a significant difference between the sexes but in both juniper and yew C concentration varied significantly over time (Tables 2, 3). The lowest C concentration of needles was observed in May, June and July in both species, and in March in the case of juniper (Fig. 5). The C concentration was considerably higher in yew than in juniper in March and then from July to November. In the intervening months of April, May and June the trend was reversed with N being higher in Juniper (Fig. 5).

Discussion

N concentration of leaves varied significantly through the year in both species but only in *T. baccata* there was a significant difference between male and female trees; males exhibited higher N concentration than females throughout the year. In *J. communis* differences between the sexes were apparent only in early spring and at this time females had higher level of N concentration. These differences can be linked to phenology. There was, however, no effect of sex on the carbon concentration.

Nitrogen concentration

The changes of N concentration observed in our study are consistent with annual fluctuations in other evergreen plants (Matyssek 1986; Silla and Escudero 2003) where the N concentration drops during early spring and then storage starts after the vegetative growth and increases until the end of the season (Dickson 1989). The decrease of N concentration during the growth of new shoots in both species is caused by the translocation of resources from the last year's needles to emerging vegetative and reproductive

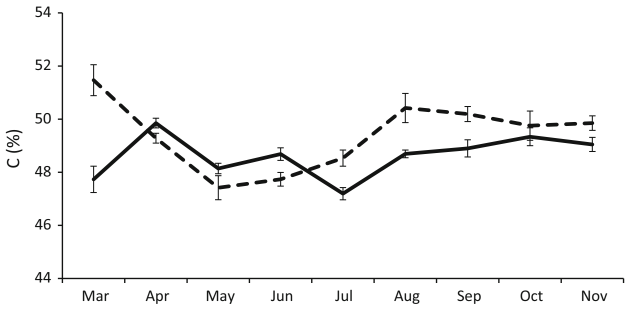

Fig. 5 Mean (±SE) annual carbon concentration of needles by species. Data for male and female have been aggregated in each species, *solid line* represents *Juniperus*, *dotted line* represents *Taxus*

tissues (Bryant et al. 1983; Grelet et al. 2001; Millard et al. 2001; Teitel et al. 2016). However, yew and juniper showed different patterns for N concentration fluctuations.

Taxus baccata

In the case of yew, males had a higher concentration of N throughout the entire period of measurement (Fig. 3). Nitrogen levels reduced in both sexes during flowering. Thereafter, N began to increase after flowering in the male, but in females it carried on decreasing, leading to the greatest differences between sexes from April to August. This accords with previous studies on *Rubus chamaemorus* and *Populus cathayana* where N concentration was frequently higher in male than in female plants (Agren et al. 1999; Chen et al. 2014), and partly with observations on *Honkenya peploides* where the N concentration of females decreased from April to August, while in males a decrease was only found from April to June (Vilas and Retuerto 2016). The data in our study, however, demonstrate these changes for the first time in an evergreen dioecious woody species, and that the greatest differences in N concentration are during intensive plant growth.

This difference between male and female trees corresponds with the period of most intensive vegetative growth, when females allocate resources not only for the vegetative growth but also for the formation and growth of seeds and arils. Yew strobili open before the new leaves, but development of arils and seeds takes place simultaneously with vegetative growth. This is the probable reason for the greater decrease of the N level in females as they invest N simultaneously in the growth of new shoots and maturing cones. This is compounded by the seeds and arils of *T. baccata* completing their development in one growing season which thus requires more resources in a short period of time in comparison with juniper that takes longer (Table 1). This greater N demand in yew may be related to the greater reproductive effort of females found in many species (e.g. Obeso 2002).

Since N level in female yews declines during periods of high demand, there was, therefore, no evidence of a compensatory mechanism connected with the storage of N in yew. This may result from the fact that yew typically grows in rich soils (see "Materials and methods" section) with less pressure of N deficiency than juniper. It should also be taken into consideration that flowering and vegetative growths do not overlap (Fig. 3) which probably helps to reduce the temporal demand for N (Delph 1990; Milla et al. 2006).

Juniperus communis

Changes in the N concentration of juniper followed a different pattern. Nitrogen concentration was higher in female junipers before the start of strobili opening in March and April (Fig. 4) which suggests the existence of a long-term strategy for increasing N storage before the growing season (Dawson and Geber 1999; Montesinos et al. 2012). Juniper had a lower overall N concentration than yew (Figs. 3, 4) and grows on soils poorer in N, thus it appears that female junipers are more deficient in N than male junipers and the females put greater emphasis in accumulating and storing N for the start of the growing season. A similar strategy has been demonstrated in another species of the genus, *J. thurifera*, which also grows on poor N habitats in Mediterranean area (Montesinos et al. 2012). The rapid decrease of N at the beginning of growth in the female juniper may also be exacerbated by vegetative growth and strobili opening overlapping in this species (Fig. 3) (Delph 1990; Milla et al. 2006). The fact that the female cones mature over 2–3 years may be a compensatory mechanism for the slow accumulation of N from growing in poor sites. In a similar way, lower N level in male needles, compared to female needles from the beginning of sampling until after the start of strobili opening could also be connected with production of pollen, which is very costly in terms of N (Ashman 1994; Vilas and Pannell 2011).

Carbon concentration

Lack of differences in C concentration between the sexes in both species should be considered in the context of morphological and physiological adaptations. Carbon demand is very high during the formation and maturation of reproductive structures (McDowell et al. 2000) and yet no difference was found in the C concentration of the leaves between the sexes. A similar result was observed, however, in *Populus cathayana* (Chen et al. 2014) suggesting that this is a more general relationship in dioecious plants. Greater reproductive effort in females is probably compensated for by a higher rate of gas exchange and photosynthesis. A second reason for the lack of differences in the level of C in the leaves of different sexes may be due to storage of C in all parts of the plant including roots and shoots, not just the needles. This is especially pertinent since remobilization is closely linked to the intensity of photosynthesis and growth (Millard and Grelet 2010). Thus, C concentration of the needles is not strictly linked to the maturation of the reproductive structures because the transport of C may be largely from the roots and shoots at the beginning of the growing season. Indeed, in the case of *J. thurifera*, a similar lack of differences was found between sexes in total non-structural carbohydrates, but females stored more non-soluble sugars in the wood than did males (DeSoto et al. 2016). On the other hand, it has to be taken into consideration that developing seeds and associated structures (cone berries and arils) also carry out photosynthesis and are partly independent in terms of the C economy (Dickson 1989; Ashman 1994).

Morphological adaptations

Greater area and length of female *T. baccata* needles may be a morphological adaptation for supporting greater reproductive effort of females. This confirms the trend shown in many studies in different dioecious plant species (Wallace and Rundel 1979; Meagher 1992; Kohorn 1994) as well as *T. baccata* (Iszkuło et al. 2009, 2013). There is a close correlation between the assimilation apparatus area and gas exchange such that larger leaves are associated most often with a higher rate of gas exchange in females (Dawson and Bliss 1989; Dawson and Ehleringer 1993). This phenomenon is probably a consequence of a greater reproductive effort of females connected with the production of seed and accompanying structures. Females are selected to have higher rates of leaf gas exchange (Dawson and Ehleringer 1993; Montesinos et al. 2006). Larger size of needles in the female vs. male is more distinct in the yew—which may be related to the differences in habitat. Junipers often grow in full light exposure but yews grow in shade conditions and light is a limiting factor.

Conclusions

So far, to our knowledge, this is the first demonstration of changes in N concentration in dioecious evergreen woody plants in short time sequences. We confirmed the hypothesis that phenology influences the sex-specific pattern of yearly foliar nitrogen concentration. Our other hypothesis was rejected as these two species had different sex-specific pattern of foliar nitrogen concentration. Juniper and yew are ecologically similar in many aspects: they both are gymnosperms, dioecious, anemophilous and zoochorous (Table 1). However, both species have opposite traits that are likely to significantly affect the species-specific N management. Juniper growing in N-poor pioneer habitats makes it necessary to have a long-term strategy for N storage by females. This is compounded by vegetative and reproductive growth overlapping in juniper. Yew grows in richer, late-successional habitats in mainland Europe and does not show such adaptation. The longer maturation time of juniper strobili over 2–3 years may be a compensatory mechanism to reduce the demand for N within a growing season.

Author contribution statement Study conception and design: KND, MJG, GI; acquisition of data: KND, GI; analysis and interpretation of data: KND, MJG, PT, GI; drafting of manuscript: KND, MJG, GI; critical revision and final version: KND, MJG, PT, GI.

Acknowledgements This study was financed by the Polish National Science Centre award number DEC-2012/07/B/NZ9/01314, and partly the statutory works of the Institute of Dendrology Polish Academy of Sciences and Faculty of Biological Sciences, University of Zielona Góra. We would like to thank Torzym Forestry District for help during the fieldwork.

Open Access This article is distributed under the terms of the Creative Commons Attribution 4.0 International License (http://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made.

References

Agren J (1988) Sexual differences in biomass and nutrient allocation in the dioecious *Rubus chamaemorus*. Ecology 69:962–973. doi:10.2307/1941251

Agren J, Danell D, Elmqvist T et al (1999) Sexual dimorphism and biotic interactions. In: Geber MA, Dawson TE, Delph LF (eds) Gender and sexual dimorphism in flowering plants. Springer, Heidelberg

Ashman T (1994) aA dynamic perspective on the physiological cost of reproduction in plants. Am Nat 144:300–316. doi:10.1086/285676

Banuelos MJ, Obeso JR (2004) Resource allocation in the dioecious shrub *Rhamnus alpinus*: the hidden costs of reproduction. Evol Ecol Res 6:397–413

Bierzychudek P, Eckhart V (1988) Spatial segregation of the sexes of dioecious plants. Am Nat 132:34–43

Bryant J, Chapin F, Klein D (1983) Carbon nutrient balance of boreal plants in relation to vertebrate herbivory. Oikos 40:357–368. doi:10.2307/3544308

Cedro A, Iszkuło G (2011) Do females differ from males of European yew (*Taxus baccata* L.) in dendrochronological analysis? Tree Ring Res 67:3–11

Chen L, Dong T, Duan B (2014) Sex-specific carbon and nitrogen partitioning under N deposition in *Populus cathayana*. Trees 28:793–806. doi:10.1007/s00468-014-0992-3

Cipollini M, Stiles E (1991) Costs of reproduction in *Nyssa sylvatica*: sexual dimorphism in reproductive frequency and nutrient flux. Oecologia 86:585–593. doi:10.1007/BF00318326

Dawson T, Bliss L (1989) Patterns of water-use and the tissue water relations in the dioecious shrub, *Salix arctica*—the physiological-basis for habitat partitioning between the sexes. Oecologia 79:332–343. doi:10.1007/BF00384312

Dawson T, Ehleringer J (1993) Gender-specific physiology, carbon isotope discrimination, and habitat distribution in boxelder, *Acer negundo*. Ecology 74:798–815. doi:10.2307/1940807

Dawson TE, Geber MA (1999) Sexual dimorphism in physiology and morphology. In: Geber MA, Dawson TE, Delph LF (eds) Gender and sexual dimorphism in flowering plants. Springer, Berlin, pp 175–215

Delph LF (1990) Sex-differential resource allocation patterns in the subdioecious shrub *Hebe subalpina*. Ecology 71:1342–1351. doi:10.2307/1938271

Delph LF (1999) Sexual dimorphism in life history. 149–173. In: Geber MA, Dawson TE, Delph LF (eds) Gender and sexual dimorphism in flowering plants. Springer, Berlin, pp 149–173

DeSoto L, Olano JM, Rozas V (2016) Secondary growth and carbohydrate storage patterns differ between sexes in *Juniperus thurifera*. Front Plant Sci 7:723. doi:10.3389/fpls.2016.00723

Dickson RE (1989) Carbon and nitrogen allocation in trees. Ann Sci For 46:631s–647s. doi:10.1051/forest:198905ART0142

Giertych P (2000) Factors determining natural regeneration of yew (*Taxus baccata* L.) in the Kórnik Arboretum. Dendrobiology 45:31–40

Grelet GA, Alexander IJ, Proe MF et al (2001) Leaf habit influences nitrogen remobilization in *Vaccinium* species. J Exp Bot 52:993–1002. doi:10.1093/jexbot/52.358.993

Iszkulo G, Boratynski A (2004) Interaction between canopy tree species and European yew *Taxus baccata* (Taxaceae). Pol J Ecol 52:523–531

Iszkulo G, Boratynski A (2005) Different age and spatial structure of two spontaneous subpopulations of *Taxus baccata* as a result of various intensity of colonization process. Flora 200:195–206. doi:10.1016/j.flora.2004.03.001

Iszkuło G, Boratyński A (2011) Initial period of sexual maturity determines the greater growth rate of male over female in the dioecious tree *Juniperus communis* subsp. *communis*. Acta Oecol 37:99–102. doi:10.1016/j.actao.2011.01.001

Iszkulo G, Kosinski P, Hajnos M (2013) Sex influences the taxanes content in *Taxus baccata*. Acta Physiol Plant 35:147–152. doi:10.1007/s11738-012-1057-0

Iszkuło G, Jasińska AK, Giertych MJ, Boratyński A (2009) Do secondary sexual dimorphism and female intolerance to drought influence the sex ratio and extinction risk of *Taxus baccata*? Plant Ecol 200:229–240. doi:10.1007/s11258-008-9447-5

Jiang H, Zhang S, Lei Y et al (2016) Alternative growth and defensive strategies reveal potential and gender specific trade-offs in dioecious plants *Salix paraplesia* to nutrient availability. Front Plant Sci. doi:10.3389/fpls.2016.01064

Koenig WD, Knops JMH (1998) Scale of mast-seeding and tree-ring growth. Nature 396:225–226. doi:10.1038/24293

Kohorn L (1994) Shoot morphology and reproduction in jojoba—advantages of sexual dimorphism. Ecology 75:2384–2394. doi:10.2307/1940892

Kozłowski J, Wiegert R (1986) Optimal allocation of energy to growth and reproduction. Theor Popul Biol 29:16–37

Kozłowski J, Wiegert RG (1987) Optimal age and size at maturity in annuals and perennials with determinate growth. Evol Ecol 1:231–244

Levins R (1968) Evolution in changing environments: some theoretical explorations. Princeton University Press, Princeton

Matsushita M, Takao M, Makita A (2016) Sex-different response in growth traits to resource heterogeneity explains male-biased sex ratio. Acta Oecol 75:8–14. doi:10.1016/j.actao.2016.06.009

Matuszkiewicz W, Faliński JB, Kostrowicki R, Wojterski T (1995) Potential natural vegetation of Poland—General Map, Sect. 6

Matyssek R (1986) Carbon, water and nitrogen relations in evergreen and deciduous conifers. Tree Physiol 2:177–187

McDowell SCL, McDowell NG, Marshall JD, Hultine K (2000) Carbon and nitrogen allocation to male and female reproduction in Rocky Mountain Douglas-fir (*Pseudotsuga menziesii* var. *glauca*, Pinaceae). Am J Bot 87:539–546. doi:10.2307/2656598

Meagher T (1992) The quantitative genetics of sexual dimorphism in *Silene latifolia* (Caryophyllaceae). 1. Genetic variation. Evolution 46:445–457. doi:10.2307/2409863

Milla R, Castro-Díez P, Maestro-Martínez M, Montserrat-Martí G (2006) Costs of reproduction as related to the timing of phenological phases in the dioecious shrub *Pistacia lentiscus* L. Plant Biol 8:103–111. doi:10.1055/s-2005-872890

Millard P, Grelet G (2010) Nitrogen storage and remobilization by trees: ecophysiological relevance in a changing world. Tree Physiol 30:1083–1095. doi:10.1093/treephys/tpq042

Millard P, Hester A, Wendler R, Baillie G (2001) Interspecific defoliation responses of trees depend on sites of winter nitrogen storage. Funct Ecol 15:535–543. doi:10.1046/j.0269-8463.2001.00541.x

Montesinos D, Luís MD, Verdú M et al (2006) When, how and how much: gender-specific resource-use strategies in the dioecious tree *Juniperus thurifera*. Ann Bot 98:885–889. doi:10.1093/aob/mcl172

Montesinos D, Villar-Salvador P, García-Fayos P, Verdú M (2012) Genders in *Juniperus thurifera* have different functional responses to variations in nutrient availability. New Phytol 193:705–712. doi:10.1111/j.1469-8137.2011.03982.x

Obeso JR (2002) The costs of reproduction in plants. New Phytol 155:321–348. doi:10.1046/j.1469-8137.2002.00477.x

Obrien R, Kaiser M (1985) MANOVA method for analyzing repeated measures designs—an extensive primer. Psychol Bull 97:316–333. doi:10.1037//0033-2909.97.2.316

Pallardy SG (2008) Nitrogen metabolism. Physiology of woody plants, 3rd edn. Academic Press, San Diego, pp 233–254

Silla F, Escudero A (2003) Uptake, demand and internal cycling of nitrogen in saplings of Mediterranean *Quercus* species. Oecologia 136:28–36. doi:10.1007/s00442-003-1232-5

Teitel Z, Pickup M, Field DL, Barrett SCH (2016) The dynamics of resource allocation and costs of reproduction in a sexually dimorphic, wind-pollinated dioecious plant. Plant Biol 18:98–103. doi:10.1111/plb.12336

Thomas PA, Garcia-Marti X (2015) Response of European yews to climate change: a review. For Syst 24:eR01. doi:10.5424/fs/2015243-07465

Thomas PA, Polwart A (2003) Biological flora of the British Isles: *Taxus baccata* L. J Ecol 91:489–524. doi:10.1046/j.1365-2745.2003.00783.x

Thomas PA, El-Barghathi M, Polwart A (2007) Biological flora of the British Isles: *Juniperus communis* L. J Ecol 95:1404–1440. doi:10.1111/j.1365-2745.2007.01308.x

Tozawa M, Ueno N, Seiwa K (2009) Compensatory mechanisms for reproductive costs in the dioecious tree *Salix integra*. Botany 87:315–323. doi:10.1139/B08-125

Verheyen K, Adriaenssens S, Gruwez R et al (2009) *Juniperus communis*: victim of the combined action of climate warming and nitrogen deposition? Plant Biol 11:49–59. doi:10.1111/j.1438-8677.2009.00214.x

Vilas JS, Pannell JR (2011) Sexual dimorphism in resource acquisition and deployment: both size and timing matter. Ann Bot 107:119–126. doi:10.1093/aob/mcq209

Vilas JS, Retuerto R (2016) Sexual dimorphism in water and nitrogen use strategies in *Honckenya peploides*: timing matters. J Plant Ecol. doi:10.1093/jpe/rtw072

Wallace C, Rundel P (1979) Sexual dimorphism and resource-allocation in male and female shrubs of *Simmondsia chinensis*. Oecologia 44:34–39. doi:10.1007/BF00346394