$\delta^{13}C$ values, photosynthetic pathways, and plant functional types for some plants from saline meadows, Northeastern China

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Abstract

Based on stable carbon isotope ratio ($\delta^{13}C$) measurements, photosynthetic pathway types were determined for 61 species in 54 genera and 24 families of flowering plants from the saline meadows of Northeastern China. Of these total vascular plants, 18 species in 17 genera from 6 families were found to have C_4 photosynthesis; 43 species in 38 genera from 20 families had C_3 photosynthesis. Six dicotyledonous species exhibited C_4 pathway, 12 monocotyledonous species were found with C_4 photosynthesis. The dicotyledonous C_4 species had relative greater mean $\delta^{13}C$ value and less total carbon content than both monocotyledonous C_4 and C_3 species. Most dicotyledonous C_4 species were annual forbs and halophytes. Some C_4 species had been previously documented, but their $\delta^{13}C$ values varied remarkably from those of the present study. Even though there are some fluctuations for the $\delta^{13}C$ values of some C_4 species, $\delta^{13}C$ value was still more reliable for C_3 and C_4 identification than the use of the enzyme ratio method and of low CO_2 compensation concentration.

Additional key words: C_4 photosynthetic pathway; saline meadow species; stable carbon isotope ratio ($\delta^{13}C$); total carbon content.

Introduction

The C_4 photosynthetic pathway is present in plants from a wide range of genera and families of both the monocotyledonous plants (hereafter monocots) and dicotyledonous plants (hereafter dicots) (Black 1971, Downton 1975, Raghavendra and Das 1978, Mateu Andrés 1992, Li 1993). The species known to have C_4 photosynthesis are predominantly found in hot and/or arid ecosystems, especially in deserts and grasslands (Winter 1981, Ziegler *et al.* 1981, Wang 2002b), while C_4 plants do occur in cool and moist ecosystems, *e.g.* salt meadows, alpine and wet forests. It is estimated that approximately only 1/3 of the 6 000 C_4 species were identified worldwide and the other 2/3 remains unclear (Hattersley and Watson 1992, Ehleringer *et al.* 1997, Wang 2006). Even though this number has increased since this date, it is still very low when compared with about 220 000 known species of angiosperms (Kennedy *et al.* 1980, Mateu Andrés 1992). The knowledge of photosynthetic pathway classifications is fundamental for a true understanding of the evolution and eco-physiology of C_4 plants, hence for their taxonomic and geographical distribution under global changes (Teeri and Stowe 1976, Raghavendra and Das 1978, Teeri *et al.* 1980, Redmann *et al.* 1995, Collatz *et al.* 1998, Pyankov *et al.* 2000, Wang 2004).

Saline meadows, one of the main vegetation types, cover a large area in the central part of Northeastern China, including Inner Mongolia, Jilin, and Heilongjiang provinces. This type of ecosystem is the eastern end of the Eurasian steppe zone, on average 140 m above sea level. Most of the meadow regions have meadow chernozem soil, with 3.5 to 6.0 % organic matter in the surface layer and relatively high soil pH (ranging from 7 to 10). More than 820 vascular plant species in 344 genera and 89 families have been identified, while the $\delta^{13}C$ values for the species from the meadows have not been studied in detail. The objective of the present study was to determine $\delta^{13}C$ values in the species from the meadow ecosystems, Northeastern China, and to relate $\delta^{13}C$ to their plant functional types and total carbon content. The results will be important for studying changes in photosynthetic pathways with climate.

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Materials and methods

Plant samples were collected from species growing in nature meadows and adjacent farm fields located near the Grassland Ecology Field Station of Northeast Normal University, on the Changling Horse Breeding Farm (44°42'–44°50' N, 123°42'–123°51' E), Jilin, China. Shoot and leaf tissues were collected from growing plants, dried at 80 °C for 48 h, and ground. Stable carbon isotope ratio (δ^{13}C) in the plant tissue was determined by using *Delta^{plus} XP* mass spectrograph. Plants with δ^{13}C values above –19 ‰ were considered to have C_4 photosynthesis, and those with δ^{13}C values lower than –21 ‰ to have C_3 photosynthesis (Redmann *et al.* 1995). Plants identified in Table 1 were classified into 5 plant functional types, *i.e.* shrubs (SHR), high perennial grasses (HPG), annual grasses (ANG), annual forbs (ANF), and perennial forbs (PEF) by morphological attributes.

Results

Sixty-one species in 54 genera and 24 families from saline meadows of Northeastern China were examined (Table 1). The δ^{13}C values of all species examined fall into two distinct groups, those of more than –15.01 ‰ (C_4 photosynthesis) and those of less than –24.74 ‰ (C_3 photosynthesis). Of the total 61 species, carbon isotope ratios indicative of C_4 photosynthesis were found in 18 species (30 % of 61 species) from 17 genera and 6 families; 43 species (70 % of 61 species) from 38 genera from 20 families had C_3 photosynthesis. 1/3 of the C_4 species was weed dicots, *e.g. Amaranthus retroflexus* L., *Kochia sieversiana* (Pall.) C.A. Mey., *Salsola collina* Pall., *Euphorbia humifusa* Willd., *Portulaca oleracea* L., and *Tribulus terrestris* L. C_4 monocots species were mainly grasses [*e.g. Arundinella hirta* (Thunb.) Tanaka, *Chloris virgata* Sw., *Digitaria ciliaris* (Rotz.)]. Generally, the dicotyledonous C_4 species had relative higher mean δ^{13}C value (ranging from –11.40 ‰ to –12.93 ‰) than the monocotyledonous C_4 species (ranging from –10.86 ‰ to –15.01 ‰). But the mean δ^{13}C value for C_3 dicots (–28.63 ‰) was less than for C_3 monocots (–26.75 ‰). The photosynthetic pathways for 36 species (*Apocynum lancifolium* Russan., *Cynanchum amplexicule* Hemsley, *Eurotia ceratoides* (L.) L.A.M.) were not reported in recent studies, and 2 species [*Euphorbia humifusa* Willd. and *Miscanthus sacchariflorus* (Maxim.) Hack.] were not included in the recent list of C_4 species in China. The δ^{13}C values for some C_4 monocots [*e.g. Arundinella hirta* (Thunb.) Tanaka, *Hemarthria sibirica* (Gand.) Ohwi, *Setaria viridis* (L.) Beauv.] were much greater than those in earlier references. The mean total carbon contents for C_4 dicots (38.0 %) was significantly less than that for C_4 monocots (43.5 %), C_3 dicots (43.8 %), and C_3 monocots (44.1 %), respectively.

Table 1. δ^{13}C values, photosynthetic pathways (C_3 or C_4), carbon content, CC [% of dry mass], and plant functional types (PFTs) in species from saline meadows of Northeast China. PFTs: SHR = shrubs, HPG = high perennial grass, ANG = annual grass, ANF = annual forbs and perennial forbs (PEF).

Family	Species	δ^{13}C [‰]	C_3/C_4	CC [%]	PFTs
Dicotyledoneae					
Amaranthaceae	*Amaranthus retroflexus* L.	–11.92	C_4	36.91	ANF
Apocynaceae	*Apocynum lancifolium* Russan.	–26.96	C_3	47.06	SHR
Asclepiadaceae	*Cynanchum amplexicule* Hemsley	–26.41	C_3	44.44	PEF
	Metaplexis japonica Makino	–27.60	C_3	40.45	PEF
Bignoniaceae	*Incarvillea sinensis* Lamark.	–29.37	C_3	42.16	ANF
Campanulaceae	*Adenophora stenophylla* Hemsley	–27.16	C_3	45.62	PEF
Chenopodiaceae	*Chenopodium acuminatum* Willd.	–29.82	C_3	38.62	ANF
	Eurotia ceratoides (L.) L.A.M.	–28.22	C_3	39.27	SHR
	Kochia sieversiana (Pall.) C.A. Mey.	–11.42	C_4	40.37	SHR
	Salsola collina Pall.	–11.98	C_4	35.89	ANF
	Suaeda corniculata (C.A. Mey.) Bunge	–27.96	C_3	37.03	ANF
	S. glauca Bunge	–27.01	C_3	36.70	ANF
Compositae	*Artemisia anethifolia* Weber	–27.57	C_3	46.27	SHR
	A. anethoides Mattf.	–27.60	C_3	40.45	PEF
	A. mongolica Fisch.	–24.54	C_3	47.91	SHR
	Heteropappus altaicus (Willd.) Novopokr.	–27.80	C_3	45.16	PEF

Table 1 (continued)

Family	*Species*	δ13C [‰]	C3/C4	CC [%]	PFTs
Compositae (cont.)	*Inula japonica Thunb.*	–28.83	C_3	41.49	PEF
	Saussurea glomerata Poir.	–29.28	C_3	35.78	PEF
	S. runcinata DC.	–28.46	C_3	43.28	PEF
	Taraxacum mongolicum Hand.	–28.47	C_3	39.55	PEF
	Xanthium strumarium L.	–27.63	C_3	42.08	ANF
Convolvulaceae	*Calystegia hederacea* Wallich	–28.13	C_3	41.53	ANF
Euphorbiaceae	*Euphorbia humifusa* Willd.	–12.93	C_4	41.70	ANF
Fabaceae	*Astragalus adsurgens* Pall.	–28.37	C_3	40.18	PEF
	Glycyrrhiza uralensis Fisch.	–27.59	C_3	46.71	PEF
	Lathyrus quinquenervius (Miq.) Litv.	–27.94	C_3	44.42	PEF
	Lespedeza davurica Schindler	–26.99	C_3	45.41	SHR
Labiatae	*Dracocephalum moldavica* L.	–28.50	C_3	41.84	ANF
	Leonurus japonica Houtt	–25.65	C_3	41.71	ANF
	Lycopus lucidus Turcz.	–28.46	C_3	39.38	ANF
	Stachys chinensis Bunge	–28.05	C_3	42.23	PEF
Linaceae	*Linum stelleroides* Planchon	–28.70	C_3	43.79	ANF
Plantaginaceae	*Plantago asiatica* L.	–28.54	C_3	41.96	PEF
Plumbaginaceae	*Limonium bicolor* O. Kuntze	–29.78	C_3	46.03	PEF
Polygonaceae	*Polygonum sibiricum* Laxm.	–27.99	C_3	42.96	AEF
Portulacaceae	*Portulaca oleracea* L.	–11.40	C_4	34.97	ANF
Ranunculaceae	*Thalictrum simplex* L.	–26.29	C_3	45.61	PEF
Rosaceae	*Potentilla flagellaris* Willd.	–28.07	C_3	43.56	PEF
	Sanguisorba officinalis L.	–26.76	C_3	46.72	PEF
Ulmaceae	*Ulmus pumila* L.	–28.40	C_3	44.05	SHR
Umbelliferae	*Siler divaricatum* Benth. *et* Hook.	–27.21	C_3	42.35	PEF
Zygophyllaceae	*Tribulus terrestris* L.	–11.71	C_4	38.31	ANF
Monocotyledonae					
Gramineae	*Arundinella hirta* (Thunb.) Tanaka	–11.10	C_4	43.18	HPG
	Calamagrostis epigeios (L.) Roth	–28.86	C_3	42.27	HPG
	Chloris virgata Sw.	–15.01	C_4	44.28	ANG
	Digitaria ciliaris (Retz.) Koel	–14.10	C_4	42.25	ANG
	Echinochloa crus galli (L.) Beauv.	–11.59	C_4	41.84	ANG
	Eragrostis poaeoides Beauv.	–12.42	C_4	43.61	ANG
	Hemarthria sibirica (Gand.) Ohwi	–10.86	C_4	45.98	HPG
	Leymus dasystachys (Trin.) Nevski	–25.50	C_3	44.64	HPG
	Miscanthus sacchariflorus (Maxim.) Hack.	–12.23	C_4	45.31	HPG
	Panicum miliaceum L.	–12.98	C_4	44.38	ANG
	Pennisetum flaecidum Griseb.	–11.71	C_4	42.87	HPG
	Phragmites communis Trin.	–24.74	C_3	42.86	HPG
	Puccinellia chinampoensis Ohwi	–25.90	C_3	45.44	HPG
	P. tenuiflora (Turcz.) Scrib. *et* Merr.	–25.58	C_3	44.90	HPG
	Setaria italica (L.) Beauv.	–11.61	C_4	41.47	ANG
	S. viridis (L.) Beauv.	–13.22	C_4	42.67	ANG
	Spodiopogon sibiricus Trin.	–11.54	C_4	44.34	HPG
Liliaceae	*Allium odorum* L.	–27.01	C_3	42.01	PEF
Typhaceae	*Typha minima* Funk	–29.66	C_3	46.34	PEF

In the saline meadow regions, the total 61 species fall within 5 plant functional types, *e.g.* 33.4 % (21 of 61) in PEF, 26.2 % (16 of 61) in AEF, 16.4 % (10 of 61) in HPG, and 11.5 % (7 of 61) in each of SHR and ANG, respectively (Table 1). No C_4 species was found in PEF, but all ANG species, 50 % of HPG, 31.3 % of ANF, and 14.3 % SHR were identified with C_4 photosynthesis, suggesting that the occurrence of C_4 photosynthesis was very common in ANG, HPG, and ANF functional types in the saline meadows. The mean total carbon contents for both ANF (39.7 %) and ANG (42.9 %) were less than for PEF (43.1 %), HPG (44.2 %), and SHR (44.3 %), but there were no significant differences among the PEF, HPG, and SHR types.

Discussion

Classifications of plant species into photosynthetic pathway types are the fundamental knowledge for a true understanding of the differences in evolution and ecophysiology between C_3 and C_4 plants, C_4 species geographical distribution, as well as C_3 and C_4 responses to global changes. The $\delta^{13}C$ value of plant tissue is diagnostic for photosynthetic pathways and is widely used as a criterion for C_3 and C_4 classifications (Downton 1975, Redmann *et al.* 1995, Pyankov *et al.* 2000). 61 species from saline meadows were classified to their types of photosynthesis (Table 1), 18 species were found with C_4 photosynthesis, and the other species with C_3 photosynthesis. Relatively more C_4 species were identified in this study, indicating that the occurrence of C_4 species was most common in the saline meadow regions. This proved that C_4 species have greater capacity to tolerate saline stresses in the regions, for many C_4 species were C_4 halophytes, *e.g. Chloris virgata* Sw., *Hemarthria sibirica* (Gand.) Ohwi, and *Kochia sieversiana* (Pall.) C.A. Mey., and some can form large patches of consociations. Unlike in deserts and steppes (Winter 1981, Pyankov *et al.* 2000, Wang 2002b, 2003), two thirds of C_4 species identified in Table 1 were grasses (12 of 18), suggesting that high salinity and moisture of the saline meadows favour the presence of the C_4 grasses. This explanation is also supported by the observations that Gramineae was the leading family with C_4 photosynthesis in the grasslands of Northeast China (Wang 2004). Studies of Winter (1981), Pyankov *et al.* (2000), and other authors proved that deserts (especially the Middle East deserts) represent centres for the evolution of Chenopodiaceae C_4 flora, for Chenopodiaceae C_4 species have higher tolerances to drought, high temperature, and high irradiance than the other types of plants. The $\delta^{13}C$ values for some C_4 monocots [*e.g. Arundinella hirta* (Thunb.) Tanaka, *Hemarthria sibirica* (Gand.) Ohwi, *Setaria viridis* (L.) Beauv.] were about 41.5, 37.6, and 26.1 % greater than those in earlier measurements by Redmann *et al.* (1995). There is no direct evidence to explain the significant differences in $\delta^{13}C$ values for these C_4 monocots between the present study and the earlier measurements (Redmann *et al.* 1995), but the variations may reflect the responses of C_4 species to the differences in sample sites, geographical distribution, and climatic changes. Even though there are some fluctuations for the $\delta^{13}C$ values with conditions (*e.g.* sites, moisture, and temperature), $\delta^{13}C$ was still more reliable for C_3 and C_4 identification than the use of the enzyme ratio method and low CO_2 compensation concentration.

Of the 5 plant functional types, more than 1/3 of the 61 species was found in PEF, but no C_4 species was identified in this type (Table 1). Two thirds of C_4 species was annuals (*e.g.* 5 ANF and 7 ANG), 50 % of HPG, was identified with C_4 photosynthesis, indicating that the occurrence of C_4 photosynthesis was very common in ANG, HPG, and ANF types in the saline meadows. Annual plant species can use seasonal precipitation efficiently in the saline meadow regions where the precipitation mainly falls between middle-June and middle-August (70 % of total rain fall), and these species can withstand severe spring droughts (from early May to middle June) as seeds (Wang 2004). Few SHR C_4 species (*Kochia sieversiana*) from the saline meadow regions indicated the C_4 flora was significantly different from that from steppes and deserts (Wang 2002a,b). More abundance of annual C_4 species in the saline meadows suggested that this kind of ecosystem was not stable, for most of these species grow only in the rainy seasons, but their occurrences may improve the soil conditions at salinity stages (Wang 2004). Therefore, the rangeland management in the regions must take into account occurrence of C_4 species and their dynamics, which are closely related to land conditions and climate variations.

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