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Carbon: freshwater plants

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ABSTRACT

$\delta^{13}C$ values for freshwater aquatic plant matter varies from −11 to −50‰ and is not a clear indicator of photosynthetic pathway as in terrestrial plants. Several factors affect $\delta^{13}C$ of aquatic plant matter. These include: (1) The $\delta^{13}C$ signature of the source carbon has been observed to range from +1‰ for HCO_3^- derived from limestone to −30‰ for CO_2 derived from respiration. (2) Some plants assimilate HCO_3^-, which is −7 to −11‰ less negative than CO_2. (3) C_3, C_4, and CAM photosynthetic pathways are present in aquatic plants. (4) Diffusional resistances are orders of magnitude greater in the aquatic environment than in the aerial environment. The greater viscosity of water acts to reduce mixing of the carbon pool in the boundary layer with that of the bulk solution. In effect, many aquatic plants draw from a finite carbon pool, and as in terrestrial plants growing in a closed system, biochemical discrimination is reduced. In standing water, this factor results in most aquatic plants having a $\delta^{13}C$ value similar to the source carbon. Using Farquhar's equation and other physiological data, it is possible to use $\delta^{13}C$ values to evaluate various parameters affecting photosynthesis, such as limitations imposed by CO_2 diffusion and carbon source.

Key-words: aquatic plants; bicarbonate assimilation; C_4; CAM; isotope fractionation.

INTRODUCTION

The stable carbon isotope ratio ($\delta^{13}C$) of the total carbon in leaves of terrestrial plants is, within limits, a reasonable indicator of biochemical processes such as carboxylation pathway in photosynthesis and of physiological processes such as water use efficiency (Rundel, Ehleringer & Nagy 1988). In contrast to terrestrial plants, far less is known of the exact relationship between $\delta^{13}C$ and either biochemical or physiological processes in freshwater aquatic plants. It is clear from what is known that this relationship is far more complex in aquatic, than in terrestrial plants. Factors that contribute to this complexity include: (1) the carbon isotope signature of the source carbon is variable between aquatic environments; (2) the form of inorganic carbon assimilated is not the same in all aquatic species; (3) biochemical pathways of carbon reduction in photosynthesis are not as well understood in aquatic plants as they are in terrestrial plants; and (4) ambient diffusional resistances are massively greater in aquatic habitats and are markedly affected by natural conditions such as velocity of currents.

Table 1 presents all known $\delta^{13}C$ values for freshwater macrophytes and it is clear from these data that the range of $\delta^{13}C$ values is greater than that observed for terrestrial plants. Also, within a species, there is markedly greater variation than is typical for terrestrial taxa.

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FACTORS AFFECTING $^{13}C/^{12}C$ DISCRIMINATION

(1) Role of source carbon

The $\delta^{13}C$ of the carbon source can vary from approximately +1‰ for HCO_3^- derived from limestone, to approximately −7‰ for CO_2 dissolved in air-equilibrated water (however, under natural conditions, even fast-moving streams are unlikely to be in equilibrium with the air, Raven, Beardall & Griffiths 1982). Inorganic carbon, derived autochthonously through respiration of aquatic flora and fauna, or allochthonously through decomposition of litter deposited into the system, or passage through subterranean sites of heterotrophic activity, can markedly lower the $\delta^{13}C$ value of the inorganic carbon pool. $\delta^{13}C$ values for dissolved inorganic carbon of the aquatic environments listed in Table 1 show a range from approximately +1 to −21·2‰. The most negative numbers are almost certainly due to respiratory CO_2, which would have a signature of $\leq$−27‰ for C_3 plants. Sites where respiratory influence is likely to be greatest are shallow, rain-fed seasonal pools (e.g. Site 1 in Table 1). Such aquatic habitats are often densely vegetated, and due to the high PPFD (photosynthetic photon flux density) and daytime water temperatures >30°C, photosynthetic demand for CO_2 may exceed supply. Consequently, dissolved CO_2 is depleted early each day but replenished through respiration each night, although no diurnal change has been detected in the isotope value for the total inorganic carbon pool (−20·4 versus −21·2‰, for am and pm, respectively, Keeley, Sternberg & DeNiro

Table 1. Carbon isotope discrimination for plant cellulose from photosynthetic tissues of freshwater plants (all reports are for submerged foliage, unless otherwise indicated

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	Water DIC
CHLOROPHYTA							
(Characeae)							
Chara contraria					(No HCO_3^- uptake – xiv)		
ix	7	negl.				−15·7	–
i	1	negl.	6·4–9·6	15	(Apr.)	−15·8	–
i	1	negl.	6·2–8·3	15–30	(May)	−25·1	−20·4 (am) −21·2 (pm)
Chara sp.							
i	3	negl.	7·4	20		−30·6	−12·9
i	4	negl.	6·5	20		−27·1	−11·5
ix	4	negl.	6·5	20		−25·3	–
(Cladophoraceae)							
Cladophora glomerata					(HCO_3^- uptake – vii, xxi)		
vii	3	negl.	8·0	11		−30·6±3·2 (4?)[d]	−5·5
RHODOPHYTA							
(Lemaneaceae)							
Lemanea mamillosa					(No HCO_3^- uptake – vii)		
vii	35	fast	8·0	11		−38·9±1·6 (4?)	−5·5
BRYOPHYTA							
(Fontinalaceae)							
Fontinalis antipyretica					(No HCO_3^- uptake – xxii, xi, xiv; c.f. xv)		
i	4	negl.	6·5	20		−26·9	−11·5
ix	4	negl.	6·5	20		−27·1	–
v	34	fast	5·5	12		−49·4	−16·7
v	34	fast	5·5	12		−50·7	−21·2
v	18	fast	7·5	12		−43·9	−5·9
v	32	fast		n.g.[e]		−31·5	+1 (?)
v	32	fast		n.g.		−33·4	+1 (?)
(Hypnaceae)							
Amblystegium riparium					(HCO_3^- uptake – iii)		
iii	3	negl.	7·5	25	(Jun. '89)	−30·9	−8·6±0·5 (2)
i	3	negl.	7·2	20	(Jun. '83)	−34·7	–
i	3	negl.	7·5	25	(Aug. '83)	−34·3	−12·9
(Ricciaceae)							
Riccia fluitans							
v	30	negl.	6·0	12		−33·2	+1 (?)
Ricciocarpos natans							
v	30	negl.	6·0	12		−28·8	+1 (?)
LYCOPHYTA							
(Isoetaceae)							
Isoetes bolanderi							
i	4	negl.	6·5	20		−25·1	−11·5
ix	4	negl.	6·5	20		−24·1	–
I. echinospora							
v	24	negl.		n.g.		−21·2	–
I. howellii					(No HCO_3^- uptake – iii)		
ix	7	negl.		n.g.		−29.2	–
ix	7	negl.		n.g.		−24·3	–
i	2	negl.	6·6–7·6	20		−26·2	−16·3
ii	1	negl.	6·6–8·6	12–20	(Mar. '81)	−29·4	−16·3
ii	1	negl.	6·6–8·6	15–25	(Apr. '81)	−28·3	−15·5±0·1 (3)
ii	1	negl.	6·6–8·8	15–25	(May '81)	−28·0	−18·5±0·1 (3)

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	$^{13}C/^{12}C$ (‰) Water DIC
i	1	negl.	6·4–9·6	10–20	(Apr. '83)	−29·1	–
i	1	negl.	6·2–8·3	15–30	(May '83)	−28·4	−20·4 (am) −21·2 (pm)
ii	1	n.a.	n.a.		Emergent (May '81)	−29·7±0·5 (2)	n.a.
ii	1	negl.	6·6–7·6	20	(corm) (Mar. '81)	−29·9±0·5 (2)	−16·3
ii	1	negl.	6·6–7·6	20	(root) (Mar. '81)	−28·7±0·2 (2)	−16·3
ii	1	n.a.	n.a.		Emerg. (corm) (May '81)	−30·1±0·8 (2)	n.a.
ii	1	n.a.	n.a.		Emerg. (root) (May '81)	−29·4±0·6 (2)	n.a.
I. karstenii							
ii	6	negl.	5·2	10		−26·6	–
I. lacustris					(No HCO_3^- uptake – xxix)		
vi	37	negl.	4·0	8		−23·5±2·4 (4)	−17·5
vi	37	negl.	4·0	8	(root)	−23·1±1·8 (4)	−17·5
I. orcuttii					(No HCO_3^- uptake – iii)		
i	1	negl.	6·4–9·6	10–20	(Apr. '83)	−24·0	–
i	1	negl.	6·2–8·3	15–30	(May '83)	27·6	−20·4 (am) −21·2 (pm)
PTEROPHYTA							
(Parkeriaceae)							
Ceratopteris sp.							
ix	38	negl.	n.g.			−39·0	–
ANTHOPHYTA – Monocotyledoneae							
(Alismataceae)							
Alisma plantago-aquatica							
v	11	negl.	n.g.			−30·0	–
v	29	negl.	6·0	12		−29·2	+1 (?)
v	29	n.a.	n.a.		Floating	−27·5	n.a.
v	29	n.a.	n.a.		Emergent	−28·4	n.a.
Sagittaria cuneata							
i	4	negl.	6·5	20		−22·7	−11·5
S. sagittifolia							
v	11	negl.	n.g.			−36·0	−5·9+1·1 (2)
v	11	n.a.	n.g.		Floating	−28·8	n.a.
v	30	negl.	6·0	12		−28·8	+1 (?)
v	30	n.a.	n.a.		Floating	−27·9	n.a.
v	26	negl.	6·0	12		−25·0±0·4 (2)	+1 (?)
v	26	n.a.	n.a.		Floating	−25·8	n.a.
v	25	negl.	5·8	12		−25·7±0·3 (2)	+1 (?)
v	25	n.a.	n.a.		Floating	−25·5±0·5 (2)	n.a.
v	25	n.a.	n.a.		Emergent	−25·9±0·8 (2)	n.a.
(Cyperaceae)							
Eleocharis acicularis					(No HCO_3^- uptake – xxv)		
ix	7	negl.	n.g.			−25·6	–
i	2	negl.	6·6–7·6	20		−30·9	−16·3
i	1	negl.	6·4–9·6	10–20	(Apr. '83)	−25·0	–
i	1	negl.	6·2–8·3	15–30	(May '83)	−28·9	−20·4 (am) −21·2 (pm)
Eleocharis macrostachya							
i	1	negl.	6·2–8·3	15–30		−28·6	−20·4 (am) −21·2 (pm)
Schoenoplectus lucustris							
v	22	fast	7·0	12		−34·5	−4·7
v	22	n.a.	n.a.		Emergent	−27·9	n.a.

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	Water DIC
(Eriocaulaceae)							
Eriocaulon decangulare					(No HCO_3^- uptake – viii)		
viii	39	negl.		n.g.		−30·0±1·5 (5)	–
(Hydrocharitaceae)							
Elodea canadensis					(HCO_3^- uptake – xiv, xxvi, xvii, xviii)		
iv	36	negl.	7·8	12		−19·1±2·1 (17)	−8·3
iii	3	negl.	7·5	25	(Jun. '89)	−12·8±1·4 (2)	−8·6±0·5 (2)
i	3	negl.	7·2	20	(Jun. '83)	−19·1	–
i	3	negl.	7·5	25	(Aug. '83)	−18·2	−12·9
v	24	negl.		n.g.		−12·9	+1 (?)
v	12	negl.	5·5	12		−20·7	–
v	15	mod.	7·5	12		−23·9	–
v	17	mod.	7·5	12		−23·9	–
v	19	m-fast		n.g.		−31·3	−3·2
v	18	fast	7·5	12		−33·3	−5·9
v	33	fast		n.g.		−21·6	–
Hydrilla verticillata					(HCO_3^- uptake – xxiv)		
x	40	negl.		n.g.		−20·5	−15·1 (CO_2)
xxvii	n.g.	n.g.	7·5	?		−25·5	−10·6
Stratiotes aloides					(HCO_3^- uptake – xvi)		
v	28	negl.		n.g.		−24·3	+1 (?)
v	28	n.a.		n.a.	Emergent	−23·4	n.a.
Vallisneria americana					(HCO_3^- uptake – xxiii)		
iv	36	negl.	7·8	12		−18·2±1·6 (22)	−7·7
V. spiralis					(HCO_3^- uptake – xxii, xviii)		
ix	38	negl.		n.g.		−31·5	–
(Najadaceae)							
Najas flexilis					(No HCO_3^- uptake xxviii)		
iv	36	negl.	7·8	12		−22·5±0·6 (2)	−8·1
(Poaceae)							
Orcuttia viscida					(No HCO_3^- uptake – iii)		
iii	7	negl.	6·5	15		−14·8±0·8 (2)	−11·0
iii	7	negl.		n.a.	Floating	−15·9±0·7 (2)	n.a.
ii	8	n.a.		n.a.	Terrestrial (field)	−12·9	n.a.
iii	7	n.a.		n.a.	Terrestrial (greenh)	−15·1±0·6 (2)	n.a.
Neostapfia colusana					(No HCO_3^- uptake – iii)		
iii	7	negl.	7·0	15		−19·1±0·1 (2)	−6·3
ii	9	n.a.		n.a.	Terrestrial (field)	−13·7	n.a.
iii	7	n.a.		n.a.	Terrestrial (greenh)	−15·9±0·1 (2)	n.a.
Tuctoria greenei					(No HCO_3^- uptake – iii)		
iii	7	negl.	6·8	15		−18·3±0·2 (2)	−6·3
ii	10	n.a.		n.a.	Terrestrial (field)	−13·4	n.a.
iii	7	n.a.		n.a.	Terrestrial (greenh)	−14·6±0·2 (2)	n.a.
(Potamogetonaceae)							
Groenlandia densa							
v	11	negl.		n.g.		−23·2	–
Potamogeton alpinus							
v	29	negl.	6·0	12		−21·7	+1 (?)
v	29	n.a.		n.a.	Floating	−22·2	n.a.
P. crispus					(HCO_3^- uptake – xiv, xiii, xix)		
iv	36	negl.	7·8	12		−16·9±1·4 (11)	−7·8
P. illinoensis							
i	5	negl.	7·7	25		−25·3	−11·5

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	$^{13}C/^{12}C$ (‰) Water DIC
P. gramineus							
v	25	negl.	5·7	12		−18·6	+1 (?)
v	25	n.a.	n.a.		Floating	−21·3	n.a.
v	26	negl.	6·0	12		−16·4	+1
v	26	n.a.	n.a.		Floating	−21·1	n.a.
v	31	n.a.	n.a.		Terrestrial	−27·1	n.a.
P. lucens					(HCO_3^- uptake – xvii, xviii)		
v	26	negl.	6·0	12		−14·3	+1 (?)
P. obtusifolius							
v	24	negl.	n.g.			−14·3	+1 (?)
v	33	fast	n.g.			−22·3	–
P. pectinatus					(HCO_3^- uptake – xix, xx, xxix)		
iv	36	negl.	7·8	12		−15·2±0·2 (2)	−6·5
i	5	negl.	7·7	25		−22·1	−11·5
v	23	negl.	8·0	12		−10·1	−5·4±1·1 (2)
v	17	mod.	7·5	12		−25·6	–
v	22	fast	7·0	12		−25·0	−4·7
P. perfoliatus					(HCO_3^- uptake – xiii, xiv)		
v	23	negl.	8·0	12		−11·7±1·3 (2)	−5·4±1·1 (2)
v	26	negl.	6·0	12		−13·9	+1 (?)
v	26	negl.	6·0	12		−15·4	+1 (?)
v	19	m-fast	n.g.			−32·5	−3·2
v	25	fast	5·8	12		−13·8	+1 (?)
v	33	fast	n.g.			−22·7	–
v	20	fast	n.g.			−28·5	–
P. richardsonii							
iv	36	negl.	7·8	12		−19·9±0·8 (2)	−6·9
P. robbensii							
iv	36	negl.	7·8	12		−19·5±1·1 (5)	−6·9
P. × nitens							
v	26	negl.	6·0	12		−15·9±0·2 (2)	+1 (?)
v	26	n.a.	n.a.		Terrestrial	−25·9	n.a.
P. × zizii					(HCO_3^- uptake – xiv)		
v	18	fast	7·5	12		−33·4	−5·9
(Sparganiaceae)							
Sparganium emersum							
v	15	mod.	7·5	12		−30·3	–
v	17	mod.	7·5	12		−32·9	–
v	19	m-fast	n.g.			−37·0	−3·2
S. gramineum							
v	29	negl.	6·0	12		−30·1	+1 (?)
v	29	n.a.	n.a.		Floating	−30·1	n.a.
(Zannichelliaceae)							
Zannichellia palustris					(HCO_3^- uptake – xxix)		
v	22	fast	7·0	12		−26·7	−4·7
ANTHOPHYTA – Dicotyledoneae							
(Acanthaceae)							
Hygrophila polysperma							
ix	38	negl.	n.g.			−24·9	–
ix	38	negl.	n.g.			−33·7	–
Synnema triflorum							
ix	38	negl.	n.g.			−36·5	–
ix	38	negl.	n.g.			−32·8	–

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes		$^{13}C/^{12}C$ (‰) Plant cellulose	Water DIC
(Apiaceae)								
Berula erecta								
v	11	negl.		n.g.			−28·5	−5·9±1·1 (2)
v	11	n.a.		n.a.	Emergent		−28·8	n.a.
v	16	mod.	7·0	12		(12 Jun.)	−32·4	−5·5±0·7 (3)
v	16	n.a.		n.a.	Emergent	(12 Jun.)	−27·2	n.a.
v	16	mod.	7·0	12		(21 Jul.)	−35·6	−5·5±0·7 (3)
v	16	n.a.		n.a.	Emergent	(21 Jul.)	−31·4	n.a.
v	16	mod.	7·0	12		(21 Aug.)	−36·1	−5·5±0·7 (3)
v	16	n.a.		n.a.	Emergent	(21 Aug.)	−31·3	n.a.
v	16	mod.	7·0	12		(13 Oct.)	−33·7	−5·5±0·7 (3)
v	16	mod.	7·0	12		(13 Oct.)	−37·4	−5·5±0·7 (3)
v	16	n.a.		n.a.	Emergent	(13 Oct.)	−28·9	n.a.
Eryngium aristulatum								
ii	1	negl.	6·2–8·3	15–30			−26·5	−20·4 (am) −21·2 (pm)
Oenanthe fluviatilis								
v	18	m-fast	7·5	12			−35·0	−5·9
v	22	fast	7·0	12			−33·2	−4·7
(Asteraceae)								
Megalodonta beckii								
iv	36	negl.	7·8	12			−19·0+1·2 (2)	−8·0
(Boraginaceae)								
Myosotis laxa								
v	14	negl.		n.g.			−36·9	–
M. scorpioides								
v	11	negl.		n.g.			−29·3	–
v	19	m-fast		n.g.			−31·8	−3·2
v	19	n.a.		n.a.	Emergent		−29·3	n.a.
Plagiobothrys undulatus					(No HCO_3^- uptake – iii)			
i	1	negl.	6·2–8·3	15–30			−27·4	−20·4 (am) −21·2 (pm)
(Brassicaceae)								
Cardamine amara								
v	34	negl.	5·5	12			−32·7±1·8 (2)	−19·0±3·2 (2)
Subularia aquatica					(No HCO_3^- uptake – xxix)			
v	24	negl.		n.g.			−23·9	–
(Callitrichaceae)								
Callitriche cophocarpa								
v	34	fast	5·5	12			−41·3	−19·0±3·2 (2)
C. longipedunculata								
i	4	negl.	6·5	20			−27·1	−11·5
ix	4	negl.	6·5	20			−24·0	–
C. hermaphroditica								
v	26	negl.	6·0	12			−15·9	+1 (?)
C. palustris								
v	29	negl.	6·0	12			−25·0	+1 (?)
C. cf. *obtusangula*								
v	11	negl.		n.g.			−32·8	–
C. sp.								
v	13	negl.		n.g.			−31·5	–
v	12	negl.	5·5	12			−28·5	–

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	Water DIC
v	14	negl.		n.g.		−33·5	–
v	19	m-fast		n.g.		−33·2±1·1 (2)	−3·2
v	16	mod.	7·0	12	(12 Jun.)	−30·2	−5·5±0·7 (3)
v	16	n.a.		n.a.	Floating (12 Jun.)	−27·3	n.a.
v	16	mod.	7·0	12	(21 Aug.)	−33·7	−5·5±0·7 (3)
v	16	mod.	7·0	12	(13 Oct.)	−35·5	−5·5±0·7 (3)
v	16	n.a.		n.a.	Floating (13 Oct.)	−30·9	n.a.
v	14	n.a.		n.a.	Floating	−30·0	n.a.
(Campanulaceae)							
Lobelia dortmanna					(No HCO_3^- uptake – xxix)		
vi	37	negl.	4·0	8		−31·7+0·8 (4)	−17·5
vi	37	negl.	4·0	8	(root)	−30·0+1·2 (4?)	−17·5
v	29	negl.	6·0	12		−33·2+0·8 (2)	+1 (?)
v	29	negl.	6·0	12	(green stem)	−30·2	+1 (?)
v	29	n.a.		n.a.	Emergent	−29·6	n.a.
(Ceratophyllaceae)							
Ceratophyllum demersum					(HCO_3 uptake – xxiv)		
i	5	negl.	7·7	25		−29·8	−11·5
v	30	negl.	6·0	12		−32·3	+1 (?)
v	17	mod.	7·5	12		−27·9	–
v	22	fast	7·0	12		−26·6	−4·7
(Crassulaceae)							
Crassula paludosa							
ii	6	negl.	5·2	10		−24·0	–
(Elatinaceae)							
Elatine hydropiper							
v	26	fast	6·0	12		−22·0	+1 (?)
E. triandra							
v	24	negl.		n.g.		−19·6	–
(Haloragaceae)							
Myriophyllum alterniflorum					(HCO_3^- uptake – xiv)		
v	26	negl.	6·0	12		−16·1	+1 (?)
M. brasiliense					(No HCO_3^- uptake – xvii)		
i	5	negl.	7·7	25		−28·4	−11·5
M. spicatum					(HCO_3^- uptake, lvs. only – xiv, xvii, xxiii)		
iv	36	negl.	7·8	12		−15·7±1·8 (32)	−7·5
v	18	fast	7·5	12		−30·5	−5·9
v	22	fast	7·0	12		−27·9	−4·7
M. verticillatum					(No HCO_3^- uptake –xvii)		
v	30	negl.	6·0	12		−28·7	+1 (?)
v	32	mod.		n.g.		−27·3	+1 (?)
v	33	fast		n.g.		−27·5	–
(Hippuridaceae)							
Hippuris vulgaris					(No HCO_3^- uptake – xiv)		
v	11	negl.		n.g.		−33·7	−5·9±1·1 (2)
v	11	n.a.		n.a.	Emergent	−30·6	n.a.
(Lamiaceae)							
Mentha arvensis							
i	2	negl.	6·6–6·7	20		−25·3	−16·3
(Lentibulariaceae)							
Utricularia vulgaris					(No HCO_3^- uptake – xiv)		
v	30	negl.	6·0	12		−31·3	+1 (?)
v	31	negl.		n.g.		−33·7	+1 (?)

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	$^{13}C/^{12}C$ (‰) Water DIC
(Lythraceae)							
Lythrium hyssopifolium							
i	1	negl.	6·2–8·3	15–30		−30·7	−20·4 (am) −21·2 (pm)
(Nymphaeaceae)							
Nuphar lutea					(No HCO_3^- uptake – xiv)		
v	29	negl.	6·0	12		−26·0	+1 (?)
v	15	mod.	7·5	12		−30·8	–
v	15	n.a.	n.a.		Floating	−27·0	n.a.
v	13	negl.	n.g.			−33·0	–
v	13	n.a.	n.a.		Floating	−28·2	n.a.
v	27	negl.	6·5	12		−26·5+1·1 (3)	+1 (?)
v	27	negl.	6·5	12		−26·0	+1 (?)
v	27	n.a.	n.a.		Floating	−26·4+2·0 (2)	n.a.
Nymphaea alba							
v	27	negl.	6·5	12		−27·1	+1 (?)
v	27	n.a.	n.a.		Floating	−26·6	n.a.
(Onagraceae)							
Ludwigia natans					(No HCO_3^- uptake – xviii)		
ix	38	negl.	n.g.			−32·5	−20·8
(Plantaginaceae)							
Littorella uniflora					(No HCO_3^- uptake – xiv)		
ii	7	negl.	n.g.			−25·0+0·0 (2)	–
(Ranunculaceae)							
Ranunculus aquatilis					(HCO_3^- uptake – xiv)		
i	1	negl.	6·4–9·6	10–20	(Apr. '83)	−14·5	–
i	1	negl.	6·4–8·3	15–30	(May '83)	−20·7	−20·4 (am) −21·2 (pm)
i	2	negl.	6·6–7·6	20		−24·0	−16·3
ix	7	negl.	n.g.			−13·4	–
ix	7	negl.	n.g.			−16·8	–
v	21	fast	n.g.			−37·4	–
R. baudotti					(HCO_3^- uptake – xxix)		
v	23	negl.	8·0	12		−11·6±0·6 (4)	−5·4±1·1 (2)
R. calcareus-peltatus							
v	19	m-fast	n.g.			−29·3	−3·2
R. flammula							
i	2	negl.	6·6–7·6	20		−27·7	−16·3
v	12	negl.	5·5	12		−28·1	
v	12	n.a.	n.a.		Emergent	−26·8	n.a.
R. fluitans							
v	20	fast	n.g.			−25·0	–
v	20	fast	n.g.			−30·2	–
R. lingua							
v	34	fast	5·5	12		−36·7	−19·0±3·2 (2)
R. peltatus							
v	26	negl.	6·0	12		−17·9	+1 (?)
v	24	negl.	n.g.			−14·7	–
R. penicillatus					(HCO_3^- uptake – vii)		
v	18	m-fast	7·5	12		−29·0±0·1 (2)	−5·9
v	18	m-fast	7·5	12		−29·5	−5·9
R. reptans							
v	26	negl.	6·0	12		−22·1	+1 (?)
v	26	n.a.	n.a.		Terrestrial	−25·5	n.a.

Continued.

Table 1. (Continued)

Data-source[a]	Site[b]	Water flow rate[c]	pH	°C	Notes	$^{13}C/^{12}C$ (‰) Plant cellulose	Water DIC
v	32	mod.	6·0	12		−28·8	+1 (?)
v	32	n.a.	n.a.		Terrestrial	−28·2	n.a.
R. trichophyllus							
iv	36	negl.	7·8	12		−17·0±0·8 (3)	−8·2
v	26	negl.	n.g.			−15·5	+1 (?)
v	11	negl.	n.g.			−25·4±4·7 (2)	–
R. sp.							
v	12	negl.	5·5	12		−25·3	–
vii	35	fast	8·0	11		−22·5±1·8 (4?)	−5·5
(Scrophulariaceae)							
Veronica anagallis-aquatica							
v	14	negl.	n.g.			−28·5	–
v	14	n.a.	n.a.		Emergent	−29·2	n.a.
v	18	fast	7·5	12		−42·2	–
v	22	fast	7·0	12		−31·3	−4·7
v. comosa							
i	2	negl.	6·6–7·6	20		−26·4	−16·3

[a]Data sources:
i=Keeley *et al.* (1986). ii=J.E. Keeley, J.A. Raven, C.B. Osmond & L. Sternberg, unpublished data. iii=J.E. Keeley & D.R. Sandquist, unpublished data. iv=LaZerte & Szalados (1982). v=Osmond *et al.* (1981). vi=Richardson *et al.* (1984). vii=Raven *et al.* (1986). viii=Raven *et al.* (1988). ix=Sternberg, DeNiro & Keeley (1984). x=Wong *et al.* (1984). xi=Bain & Proctor (1980). xii=Elzenga & Prins (1988). xiii=Kadono (1980). xiv=Maberly & Spence (1983). xv=Penuelas (1985). xvi=Prins & DeGuia (1986). xvii=Prins *et al.* (1982). xviii=Prins *et al.* (1980). xix=Sand-Jensen (1983). xx=Sand-Jensen & Gordon (1984). xxi=Simpson & Eaton (1986). xxii=Steeman-Nielsen (1947). xxiii=Titus & Stone (1982). xxiv=Van, Haller & Bowes (1976). xxv=Morton & Keeley (1990). xxvi=Madsen & Sand-Jensen (1987). xxvii=Benedict (1978). xxviii=Wetzel (1969). xxix=Spence & Maberly (1985).

[b]Sites:
1=Mesa de Colorado pool (CA, USA). 2=Mather pool (CA, USA). 3=Birch Lake (CA, USA). 4=Siesta Lake (CA, USA). 5=Searsville Lake (CA, USA). 6= Sumapaz Lake (Colombia). 7=Greenhouse (CA, USA). 8=Sacramento pool (CA, USA). 9=Jepson Prairie pool (CA, USA). 10=Chico pool (CA, USA). 11–34=respectively, sites B1–B12 (UK) and F1–F12 (Finland) from Osmond *et al.*, 1981. 35=Dichty Burn (Scotland, UK). 36=Lake Memphregog (Quebec, Canada). 37=Loch Brandy (Scotland, UK). 38=Palm Beach sloughs (FL, USA). 39=Laboratory (CA, USA). 40=lake (TX, USA).

[c]Water flow: negl.=negligible; mod.=moderate (<10 m min^{-1}); fast (>10 m min^{-1}).

[d]$\bar{X}$±S.D. (N).

[e]n.a.=not applicable to aerial foliage; n.g.=data not given.

1986). This diurnal process of photosynthetic depletion and respiratory addition of CO_2 results in a cyclic enrichment of ^{12}C through the season; $\delta^{13}C_{water}$=−15·5 to −21·2‰ from early to late spring (see *Isoetes howellii*, Table 1 and Keeley *et al.* 1986).

There is evidence that differences in source carbon can account for differences in $\delta^{13}C$ of plant biomass. For example, Osmond *et al.* (1981) found that the site to site differences in $\delta^{13}C$ of the moss *Fontinalis antipyretica*, from fast moving streams in Finland, could be accounted for by the $\delta^{13}C$ of the source carbon. The site with the lowest plant $\delta^{13}C$ values (−49·4 to −50·7‰) were approximately 17 to 18‰ lower than the $\delta^{13}C$ for plants from another site. This difference in plant matter was similar to the estimated difference in $\delta^{13}C$ of the source carbon between the two sites (17–22‰, see Table 1). Additionally, LaZerte & Szalados (1982) showed that with a mixture of species from different sites there was a statistically significant correlation between $\delta^{13}C_{plant}$ and $\delta^{13}C_{water}$.

Despite these demonstrations, species from the same site, and exposed to the same source carbon, may differ markedly in $\delta^{13}C$; for example, *Elodea canadensis* and *Amblystegium riparium* from Birch Lake consistently differed by 16–17‰ (Site 3, Table 1). Thus, chemical and physical factors, other than source carbon, are clearly involved.

(2) Inorganic carbon species

Unlike terrestrial plants, certain submerged aquatic plants may use HCO_3^- in addition to CO_2 (Raven 1970; Bain & Proctor 1980; Maberly & Spence 1983). Across the range of temperatures commonly encountered by aquatic plants, the $\delta^{13}C_{HCO_3^-}$ will be 7–11‰ less negative

than $\delta^{13}C_{CO_2}$ (Mook, Bommerson & Staverman 1974). Therefore, the proportion of carbon assimilation arising from active uptake of HCO_3^- will affect the $\delta^{13}C$ of the plant material. CO_2 is commonly described as the 'preferred' form of carbon, which is an anthropomorphic way of saying that the Km for HCO_3^- uptake is typically much higher than the Km for CO_2 uptake. The proportion of these two carbon species that is assimilated is dependent upon species-specific differences in capacity for active transport of the HCO_3 ion and on the proportion of CO_2 and HCO_3^- in the boundary layer of the leaf. The ratio of CO_2 to HCO_3^- is a function of ambient pH (e.g. at pH 5·5, 80% of the inorganic carbon is as CO_2, whereas at pH 8·5, CO_2 is <1%), total carbon level, photosynthetic rate and level of turbulence.

For some aquatic species, there is substantial evidence that little or no capacity exists for active uptake of HCO_3^- (Table 1). For other species, evaluating the effect of CO_2 versus HCO_3^- assimilation on total plant $\delta^{13}C$ is complicated by the fact that the ratio of CO_2 to HCO_3^- varies diurnally and seasonally and an integrated measure of the contribution of each carbon species on the total carbon assimilation is unavailable for any aquatic species. As a matter of speculation, perhaps with increased understanding of factors controlling carbon discrimination, the $\delta^{13}C$ value might one day provide just such an integrator of CO_2 and HCO_3^- uptake.

(3) Photosynthetic pathways

Apparently, all three photosynthetic pathways defined for terrestrial plants are present in freshwater habitats. As is the case with land plants, the C_3 pathway appears to be widespread and CAM and C_4 limited to special situations.

A sample of species with different photosynthetic pathways is shown in Table 2. In order to minimize the effect of other factors, the only species included were those largely lacking bicarbonate uptake and from habitats with negligible water flow. Although the three C_4 species listed exhibit the C_4 biochemical pathway, as evidenced by carbon fixation into organic acids followed by rapid turnover to PCR pathway products, only *Neostapfia colusana* has aquatic foliage with well developed kranz anatomy (J.E. Keeley, unpublished data).

While Table 2 is not an exhaustive list of all information on aquatic plant photosynthetic pathways, these data illustrate that, even if one takes into account the source carbon, isotope ratio does not distinguish aquatic C_3, C_4 and CAM plants. Whereas terrestrial C_3 plants have a $\Delta^{13}C$ ($=\delta^{13}C_{plant}-\delta^{13}C_{carbon\ source}$) between −20 and −25‰, similar to the $\Delta^{13}C$ observed for the floating leaf of *Nuphar* (Table 1), aquatic C_3 plants are markedly less negative, and fall within the range observed for aquatic C_4 species (Table 2). Aquatic CAM species range from −4 to −8‰ (Table 2), which is also similar to that observed for many terrestrial CAM species (Griffiths 1992, in this issue, p. 1051). The most negative value reported for an aquatic CAM species is in *Isoetes bolanderi* and it is likely that this is affected by the fact that much of the carbon comes via the roots from the organic-rich substrate (Sandquist & Keeley 1990). Such sediments are likely to be rich in respiratory CO_2 from decomposition.

Table 2. Photosynthetic pathway and carbon isotope value for aquatic macrophytes selected for their lack of bicarbonate uptake and sampled from habitats with negligible water flow rate (data from Table 1). Photosynthetic pathway based on published and unpublished data (see Keeley & Busch 1984; Keeley 1990; J.E. Keeley, unpublished data; Keeley *et al.* 1986; Raven *et al.* 1987; Salvucii & Bowes 1981; Beer & Wetzel 1982)

Species	Photosynthetic pathway	$\delta^{13}C$ (‰) Plant	Source (total DIC)	Plant-source*
Fontinalis antipyretica	C_3	−26·9	−11·5	−10·0
Plagiobothrys undulatus	C_3	−27·4	−20·4	−3·2
Myriophyllum brasiliense	C_3	−28·4	−11·5	−8·3
Najas flexilis	C_3	−22·5	−8·1	−4·3
Isoetes howellii	CAM	−26·2	−16·3	−3·9
		−28·5	−15·6	−6·6
		−28·4	−20·4	−4·2
(emergent)	C_3	−29·4	−7·0 (air)	−22·4
I. lacustris	CAM	−23·5	−17·5	−6·0
I. orcuttii	CAM	−27·6	−20·4	−3·4
I. bolanderi	CAM	−25·1	−11·5	−8·2
Eleocharis acicularis	C_3/C_4	−30·9	−16·3	−8·6
		−28·9	−20·4	−4·7
Orcuttia viscida	C_4	−19·0	−11·0	−3·8
(terrestrial)	C_4	−12·9	−7 (air)	−5·9
Tuctoria greenei	C_4	−18·4	−6·5	−4·7
(terrestrial)	C_4	−13·4	−7 (air)	−6·4
Neostapfia colusana	C_4	−15·4	−6·5	−0·5
(terrestrial)	C_4	−13.7	−7 (air)	−6·7

*Assumes dissolved CO_2 (not HCO_3^-) as the source. $\delta^{13}C$ for CO_2 fraction calculated as described in Mook *et al.* (1974).

The lack of differentiation in $\Delta^{13}C$ between C_3, C_4 and CAM photosynthetic modes, and the observation that aquatic C_3 species are less negative than terrestrial C_3 species, suggests other factors, such as the greater diffusive resistance of the aquatic milieu, apparently over-ride the large fractionation (−30‰) imposed by Rubisco.

(4) Diffusional resistances

The diffusion coefficient of CO_2 in water is about 10000 times smaller than in air so that diffusion through the unstirred boundary layer around the leaves of aquatic macrophytes is an important rate limiting step in photosynthesis. Although $\delta^{13}C$ fractionation may occur due to diffusion (Smith & Walker 1980), the primary consequence of diffusive resistance created by the boundary layer is that it counteracts biochemical discrimination by Rubisco. Decreased discrimination between the plant and the carbon source arises if the carbon source is finite, as in the boundary layer around the leaf, and fixation of carbon leads to an accumulation of the discriminated isotope (^{13}C), resulting in a less negative $^{13}C/^{12}C$ ratio for the source carbon. When all the available carbon is fixed, the $^{13}C/^{12}C$ ratio in the synthesized products will be the same as in the source; i.e. discrimination is zero. This effect is similar to the elimination of ^{13}C discrimination by terrestrial C_3 species when maintained in a closed system of recycled CO_2 (Berry & Troughton 1974).

Not surprisingly, the degree of isotope discrimination is greatly affected by the extent of mixing of the bulk solution and this can be seen in data for several species; for example, *Fontinalis antipyretica, Elodea canadensis* and *Potamogeton perfoliatus* — all three exhibited much less discrimination against ^{13}C in habitats of standing water than in fast moving streams (Table 1). For a C_3 species lacking bicarbonate uptake (e.g. *Fontinalis antipyretica*), it can be shown that under conditions where ambient diffusional resistances are minimal (such as a fast moving stream), the $\Delta^{13}C$ [=−49·4‰−(−17·9‰ for CO_2)]=−31·5‰ is remarkably close to the biochemical fractionation of Rubisco (Osmond *et al.* 1981). Based on this analysis, it is likely that fractionation differences would be apparent between aquatic C_3, C_4 and CAM species in habitats where diffusional resistances are minimal, such as rapidly moving streams. Although plants with C_4 or CAM photosynthesis are largely unknown from such habitats, this hypothesis could be tested under artificial conditions.

Another important factor affecting the degree of discrimination would be photosynthetic rate; at high rates, the carbon source in the boundary layer is more likely to be finite, thus reducing discrimination by Rubisco. Therefore, we might expect $\delta^{13}C$ values to be lowest in C_3 plants from oligotrophic conditions, where photosynthetic rates are likely to be slow enough that carbon in the boundary layer is better mixed with carbon in the bulk solution. However, this difference is potentially offset by the fact that carbon levels are substantially higher in eutrophic environments, and thus, not as readily depleted in the boundary layer.

MODELING AQUATIC MACROPHYTE FRACTIONATION

Unlike terrestrial studies, $\delta^{13}C$ values are of very limited use in aquatic plant studies unless there is available information on the physiology and biochemistry of photosynthesis. Having such data, however, Raven *et al.* (1982) and Raven, MacFarlane & Griffiths (1987) suggest that $\delta^{13}C$ values may be used to evaluate the relative limitations to photosynthesis attributable to diffusion of CO_2 in aquatic plants. With a modification of the equation of Farquhar, O'Leary & Berry (1982) it was proposed that:

$$\frac{c_i}{c_s} = \frac{(^{\delta}\text{ plant} - ^{\delta}\text{ solution}) - a}{(b-a)} \qquad (1)$$

where:

c_i=the CO_2 concentration at the site of Rubisco activity during steady-state photosynthesis (mol cm^{-3});

c_s=the CO_2 concentration (mol cm^{-3}) in the bulk medium;

$^{\delta}$ plant=the $\delta^{13}C$ value of the plant material (‰ relative to PDB);

$^{\delta}$ solution=the $\delta^{13}C$ value of the dissolved carbon dioxide in solution or bicarbonate;

a=the δ value associated with CO_2 diffusion in solution from a source to a sink (‰ relative to source CO_2, taken as equal to zero, O'Leary 1981); and

b=the δ value associated with CO_2 fixation by Rubisco (‰ relative to the CO_2 supplied to the enzyme active centre, equal to −30‰, O'Leary 1981).

Using Eqn 1 and data presented in Table 3, Raven *et al.* (1982, 1987) calculated that for *Lemanea mamillosa*, a C_3 plant with no significant HCO_3^- assimilation, in a rapidly moving stream, c_i=23 mmol m^{-3} or c_i/c_s=0·77. This was interpreted to mean that 23% of the limitation on photosynthetic rate is associated with CO_2 diffusion from the bulk phase to the carboxylase, with the remaining 77% imposed by biochemical restrictions. Using this estimate of c_i and laboratory measurements of maximum photosynthetic rates, they calculated that, for a cylindrical organ of 450 μm diameter, the thickness of the unstirred layer around it equalled 11 μm. For *Cladophora glomerata*, they calculated 164 μm for the unstirred layer around the thallus branches, a distance at least half of the mean distance between the branches in the thallus. Subsequent work has shown that, under conditions of high c_i/c_s or significant bicarbonate uptake, $\delta^{13}C$ values cannot readily be used to estimate the unstirred layer thickness (Raven & Farquhar 1990).

An interesting aquatic system for evaluating models of isotope discrimination is Birch Lake, where the moss

Table 3. Photosynthetic characteristics of macrophytes from Dichty Burn, Scotland, UK (from Raven *et al.* 1982, 1987)

Parameter	Water			*Lemanea mamillosa*	*Cladophora glomerata*
DIC (mol C m^{-3})	0·03				
pH	8·0				
Temperature (°C)	11·0				
Water flow (m s^{-1})	>1				
	(DIC)	(HCO_3^-)	(CO_2)		
$\delta^{13}C$ (‰)	−5·5±1·0	−5·2±1·0	−15·9±1·0	−38·9±2·0	−30·6±1·6
Photosynthetic pathway				C_3	C_3
HCO_3^- uptake				No	Yes
Photosynthetic rate					
(μmol mg^{-1} chl h^{-1})					
pH 6·5 (@ 10 mmol m^{-3} CO_2)				42·6±2·9 (3)	31·6±2·4 (9)
pH 8·0 (@ 10 mmol m^{-3} CO_2)				40·3±7·8 (4)	35·2±7·2 (9)
(pmol cm^{-2} s^{-1})					
pH 6·5 (@ 10 mmol m^{-3} CO_2)				68·5±4·8 (3)	7·31±0·33 (5)
pH 8·0 (@ 10 mmol m^{-3} CO_2)				64·8±14·0 (4)	8·14±0·99 (9)

An interesting aquatic system for evaluating models of isotope discrimination is Birch Lake, where the moss *Amblystegium riparium* coexists intertwined with *Elodea canadensis* (Table 4). Both are C_3 plants, but $\delta^{13}C$ is consistently 16–18‰ more negative in *Amblystegium*. *Elodea canadensis* is known to be a bicarbonate-user (Madsen & Sand-Jensen 1987; Elzenga & Prins 1988), whereas it is unclear to what extent *Amblystegium* can utilize bicarbonate; preliminary results suggest it is not a HCO_3^--user but solid evidence is lacking (J.E. Keeley, unpublished data). For *Elodea*, the proportion of CO_2 uptake versus HCO_3^- uptake over the season is unknown; however, calculations of $\Delta^{13}C$ shown in Table 4 could be interpreted as evidence that HCO_3^- uptake does represent a substantial portion of the carbon gain in *Elodea*. For example, if we assume no HCO_3^- uptake, the $\Delta^{13}C$ is calculated to be +3 to +4‰; such numbers indicate ^{13}C enrichment of the plant over the source carbon. This could only occur if high photosynthetic rates resulted in a chemical disequilibrium between

Table 4. Photosynthetic characteristics of macrophytes from Birch Lake, California, USA (from Keeley *et al.* 1986, unpublished data)

Parameter	Water			*Amblystegium riparium*			*Elodea canadensis*		
DIC (mol C m^{-3})	2·1								
pH	7·5								
Temperature (°C)	25								
Water flow (m s^{-1})	negligible								
$\delta^{13}C$ (‰)	(DIC)	(HCO_3^-)	(CO_2)	(Plant)	(Plant -HCO_3)	(Plant -CO_2)	(Plant)	(Plant -HCO_3)	(Plant -CO_2)
June 1989	−8·6	−8·0	−17·0	−30·9	−22·9	−13·9	−12·8	−4·8	+4·2
August 1983	−12·9	−12·3	−21·3	−34·3	−22·0	−13·0	−18·2	−5·9	+3·1
Photosynthetic pathway				C_3			C_3		
Dark fixation (% of light fixation):				1–2			1–2		
Initial fixation products:				82% PGA			70% PGA		
Rubisco (μmol mg^{-1} chl h^{-1})				195±60 (4)			241±33 (9)		
PEPcase (μmol mg^{-1} chl h^{-1})				1±2 (4)			19±3 (4)		
HCO_3^- uptake				?			Yes		
Carbonic anhydrase									
(E.U.×10^6 mg^{-1} chl)				524±147 (3)			not detectable		
Chlorophyll (mg g^{-1} fresh weight)				2·35±0·37 (9)			1·08±0·14 (20)		
Leaf area (cm^2 mg^{-1} chl)				194±70 (2)			125±26 (2)		
Photosynthetic rate									
(μmol mg^{-1} chl h^{-1})									
pH 4·0 (@ 500 mmol m^{-3} CO_2)				49·0±3·3 (3)			73·4±6·9 (3)		
pH 5·0 (@ 500 mmol m^{-3} CO_2)				50·1±2·8 (3)			–		
pH 8·0 (@ 500 mmol m^{-3} CO_2)				73·1±4·9 (3)			81·9±8·6 (3)		
(pmol cm^{-2} s^{-1})									
pH 4·0 (@ 500 mmol m^{-3} CO_2)				70·0			163·1		
pH 8·0 (@ 500 mmol m^{-3} CO_2)				104·5			182·0		

Table 5. Photosynthetic characteristics of macrophytes from Mesa de Colorado seasonal pool (from Keeley *et al.* 1986, unpublished data)

Parameter		Water		*Isoetes howellii*		*Plagiobothrys undulatus*		*Ranunculus aquatalis*		
DIC (mol C m^{-3})		0·7								
pH		6·2 (am) 8·3 (pm)								
Temperature (°C)		15 (am) 30 (am)								
Water flow		negligible								
$\delta^{13}C$ (‰)	(DIC)	(HCO_3^-)	(CO_2)	(Plant)	(Plant -CO_2)	(Plant)	(Plant -CO_2)	(Plant)	(Plant -HCO_3)	(Plant -CO_2)
(am)	−20·4	−14·1	−24·2	−28·4	−4·2	−27·4	−3·2	−20·7	−6·6	+3·5
(pm)	−21·3	−21·2	−29·7	−28·4	+1·3	−27·4	+2·3	−20·7	+0·5	+9·0
Photosynthetic pathway				CAM		C_3		C_3		
Acid accumulation (μmol mg^{-1} chl $night^{-1}$)				200–400		not detectable		not detectable		
Initial light fixation products:				PGA & O.A.		PGA		PGA		
Rubisco (μmol mg^{-1} chl h^{-1})				229±24 (5)		335±47 (2)		245±45 (2)		
PEPcase (μmol mg^{-1} chl h^{-1})				28±8 (5)		16±5 (2)		23±15 (2)		
HCO_3^- uptake				No		No		Yes		
Carbonic anhydrase (E.U.×10^6 mg^{-1} chl)				10±14 (3)		11±12 (3)		26±21 (3)		
Photosynthetic rate (μmol mg^{-1} chl h^{-1}) pH 6·0 (@ 500 mmol m^{-3} CO_2)				10·2±2·3 (3)		10·0±1·9 (3)		74·1±21·2 (3)		

HCO_3^- and CO_2 in the boundary layer, and due to dehydroxylation of HCO_3^- the leaf was supplied with CO_2 enriched in ^{13}C, relative to bulk CO_2. However, if we assume a proportion of carbon uptake is through both CO_2 and HCO_3^- uptake, the calculated $\Delta^{13}C$ (plant−source) would be between +4·2 and −5·9‰.

Using Eqn 1, it is determined that c_i ranges from 0·97 to 1·60 mol m^{-3} for *Amblystegium*, dependent upon whether the carbon source is CO_2 or HCO_3^-. For *Elodea*, using CO_2 as the carbon source gives a negative C_i, whereas HCO_3^- gives a c_i of 0·34–0·41 mol m^{-3}. This suggests that the carbon pool from which Rubisco directly draws is more likely finite in *Elodea*, and consequently, the $\delta^{13}C$ of the plant matter is likely to be similar to the source carbon. In the case of *Amblystegium*, the much greater c_i may provide a large enough carbon pool so that the biochemical discrimination by Rubisco can be expressed, resulting in a more negative $\delta^{13}C$ for plant biomass.

Another factor that may contribute to differences in $\delta^{13}C$ between these species is the observed difference in activity of carbonic anhydrase (Table 4). This enzyme catalyses the reversible hydration of CO_2 and is present in *Amblystegium* but absent from *Elodea*. However, the exact location of carbonic anhydrase in *Amblystegium* is unknown, and thus, little can be said about its role in accounting for the differences in isotope ratio between these two species.

A final comparison of $\delta^{13}C$ values is for vernal pool plants of differing photosynthetic characteristics (Table 5). Despite the fact that one-third to a half of the total carbon gain in *Isoetes howellii* is derived from the CAM pathway, this species has a relatively negative $\delta^{13}C$. However, the source carbon is among the most negative observed for aquatic environments, and the $\Delta^{13}C$ is similar to that observed for terrestrial CAM plants (Table 1).

As mentioned above, these shallow seasonal pools fluctuate during the day in pH and this has a marked effect on the $\delta^{13}C$ for both forms (HCO_3^- and CO_2) of the source carbon (Table 5). Previous studies showed that species in these pools exhibited a depression in carbon uptake, which paralleled the morning depletion of CO_2 from the bulk solution (Keeley & Sandquist 1991). This pattern would be indirectly predicted by the $\Delta^{13}C$ since carbon uptake in the afternoon would result in $\delta^{13}C$ values more positive than the source carbon. Calculations of $\Delta^{13}C$ would also predict that *Ranunculus aquatalis* is a bicarbonate user, since CO_2 uptake would result in $\Delta^{13}C$ more positive than $\delta^{13}C_{\text{source carbon}}$. Further, these data suggest carbon uptake is concentrated in the morning (as observed for other vernal pool species, Keeley & Sandquist 1991), since afternoon uptake of either CO_2 or HCO_3^- would result in a positive $\Delta^{13}C$.

This discussion gives some examples of the possible uses for $\delta^{13}C$ values of aquatic plants. It is clear that much remains to be learned from application of carbon isotopes to aquatic plant studies.

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