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Total Lipid and Fatty Acid Classes in Decomposing Mangrove Leaves of *Bruguiera gymnorrhiza* and *Kandelia candel*: Significance with respect to Lipid Input

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Changes in the concentration of total lipid and fatty acids (FAs) during the decomposition of mangrove leaves were investigated by field experiments using yellow leaves of *Bruguiera gymnorrhiza* (L.) Lamk. and *Kandelia candel* (L.) Druce, in order to quantify mangrove contribution to lipid and fatty acid inputs to marine sediments. Total lipid and total FA in the fresh (green and yellow) and decomposing leaves of both species were significantly higher during winter than summer. During decomposition, total lipid content and FA concentration, in particular branched chain fatty acids (BrFAs) and bacterial fatty acids (BFAs), increased to a maximum concentration in 45 days during winter and in 17 days during summer. Lipids were lost faster in *K. candel* leaf detritus than in *B. gymnorrhiza* leaf detritus in which >90% of the total lipid original weight was lost during the summer experiment and <60% during the winter experiment. The changes in the concentrations of total lipids and FAs in the decomposing leaves also indicate that mangrove leaves are significant sources of fatty acids and probably other lipid compounds to estuarine ecosystems and that tidal waters transport the lipids and FAs adsorbed to particulate matter from mangroves to adjacent estuarine sediments and the ocean.

Keywords:
· *Bruguiera gymnorrhiza*,
· *Kandelia candel*,
· lipid content,
· mangrove,
· decomposition,
· fatty acids.

1. Introduction

Mangrove leaf fall contributes more than 70% of total litter production in many mangrove forests of the world (Hardiwinoto *et al*., 1989; Twilley *et al*., 1997; Tam *et al*., 1998). A substantial amount of the leaf fall is exported from mangrove ecosystems by tidal waters, either fresh or in various stages of decomposition (Bano *et al*., 1997; Wafar *et al*., 1997). It is through the decomposition process that nutrients and other organic compounds such as lipids are released to estuarine waters and sediments via tidal transport. The lipids and nutrients are vital for the functioning of the estuarine communities e.g. crabs and fish.

There are few studies on the quantitative contribution of mangrove leaves as a source of lipids and FA to estuarine and coastal sediments. Earlier studies by Sassen (1977a, b) on mangrove peats, which involved the analysis of monobasic acids, and on degradation of short-chain fatty acids in leaf litter suggested a major input of mangrove leaf litter lipids to marine sediments. This study therefore aimed to examine in detail changes in total lipid and total FA concentrations (including those of major FA classes) and weight loss in mangrove leaves during microbial decay of two common mangrove species in the subtropics: *Bruguiera gymnorrhiza* and *Kandelia candel*. By studying decomposing mangrove leaves, we expected to determine how rapidly total lipids and FAs are removed from decaying leaves and therefore how much mangrove lipids and FAs might be contributed to the estuarine ecosystem by each species.

Since lipid and FA concentration in mangrove leaves may differ between species and seasons, we performed two decomposition experiments during winter and during summer, using senescent yellow leaves of the two species. Concentrations of total lipid and FAs in mature green leaves of both species were also measured in order to obtain the lipid and FA amounts before leaf fall.

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2. Materials and Methods

2.1 Study area

The leaf degradation experiment was carried out in a subtropical mangrove forest located in the northern part of Okinawa Island in southern Japan (26.5°N, 128°E). This mangrove forest is situated in an enclosed bay at the mouth of Oura River. It consists of mixed stands of *Bruguiera gymnorrhiza*, *Kandelia candel* and *Rhizophora stylosa* that cover an area of about 10 ha. The species *B. gymnorrhiza* dominates in the forest, followed by *K. candel* (Mfilinge *et al.*, 2002). *R. stylosa* is the least dominant species with few stands (<20) in the forest. The decay experiments used leaves of the most dominant species *B. gymnorrhiza* and those of the second dominant species *K. candel*. Sediments vary from mud to clay in the mangroves, and soft mud to sand in an adjacent intertidal flat. Annual mean air temperature is 22.9°C, the highest monthly mean occurs in July (28.0°C) and lowest in January (14.0°C) (Mfilinge *et al.*, 2002).

2.2 Experimental design

Two consecutive leaf degradation experiments were performed. The first was conducted during the winter season (between December 2001 and June 2002) while the second was conducted during the summer (between June and October 2002). In order to study comparative mangrove lipid and FA input to marine ecosystems, a mesh bag technique was used with slight modification. The mesh bags, made of nylon (1 mm mesh and 22 × 18 cm), were modified by cutting three small holes (each of 2 cm diameter) in each mesh bag. This was done to allow small crabs, gastropods and amphipods to access the decaying leaves, since it has previously been noted that not only microbes affect lipid and FA degradation and inputs to marine environments but also larger organisms (Sassen, 1977a).

Before the experiments began, 20 mature green leaves of *Bruguiera gymnorrhiza* and *Kandelia candel* were picked from trees at random. Yellow leaves of *B. gymnorrhiza* and *K. candel*, which had just fallen on sediments at low tide, and those still attached to branches but ready to abscise, were then collected randomly and rinsed with distilled water in the field. In the laboratory ~10 g (equivalent to 5 g dry weight) of fresh weight yellow leaves was placed by sewing into the prepared mesh bags. This helped to avoid leaf loss but allowed grazing by invertebrates.

A sub-sample of the yellow leaves (20) was processed and analyzed immediately for lipids to obtain the total lipid, fatty acid concentrations and composition in leaves at time 0. In each experiment a total of 60 mesh bags containing fresh yellow *B. gymnorrhiza* and *K. candel* leaves were randomly deployed on the mangrove sediments. The experiment ran for 132 days during the winter through spring seasons and for 124 days during the summer season.

Three mesh bags (for each mangrove species) chosen at random were removed after 10 days when leaves of both species had turned brown, and thereafter every 17, 31, 45, 59, 73, 87 and 132 days during the winter season and 5, 17, 31, 45, 59, 73, 87 and 124 days during the summer. Mesh bags were washed with freshwater in the field to remove attached sediments. In the laboratory mesh bags containing detritus were rinsed again over a 0.5 sieve to remove small invertebrates such as crabs, gastropods and amphipods, fine sediments and unknown attached particles. A sub-sample of leaf tissue was removed and immediately stored at –20°C for lipid analysis. Water content in each sample was determined on separate sub-samples of fresh and decomposed leaves in three replicates.

2.3 Lipid extraction

Fresh weight samples of green, yellow leaves and leaf detritus were chopped into small pieces and milled in a miller. A sub-sample of 4 g and 2 g of the milled fresh leaves and leaf detritus, respectively, was used in each analysis. Samples were extracted following a slightly modified version of the method of Bligh and Dyer (1959). Two replicates were measured for each fresh leaf group and each retrieved mesh bag. Lipids were extracted by homogenization for 2 min followed by ultrasonication for 20 min with a mixture of distilled water:methanol:chloroform (1:2:1, 20 cm^3, v:v:v). Addition of distilled water:chloroform mixture (5:5 cm^3 v:v) formed an aqueous-organic two-layer system. The lipids migrated into the lower chloroform phase and separation was enhanced by centrifugation (2000 rpm 650 x g, 5 min). Each sample was extracted twice. The extracted lipids were filtered through pre-combusted GF/C filter paper to remove any fine sediments or particulate matter (Wilson *et al.*, 2001). The purified lipids were then concentrated by rotary evaporation. To estimate the total lipid content, the chloroform was evaporated completely under a stream of nitrogen, then on a hot plate set at 50°C. The remaining lipid was weighed on an electronic balance ER-182A. Total lipid content was calculated from an average of six vials and expressed as milligram of lipid per gram dry weight of leaves.

Fatty acid analysis The lipid extracts were saponified under reflux (2 h, 100°C) with a 2 mol dm^{-3} NaOH solution in methanol and distilled water (2:1, v:v). Saponification and methylation were performed according to Meziane and Tsuchiya (2000). Lipid groups were separated by Thin-Layer Chromatography (TLC) using Merck plates coated with silica gel 60 silica (Darmstadt, Germany). For further detail the reader is referred to Meziane

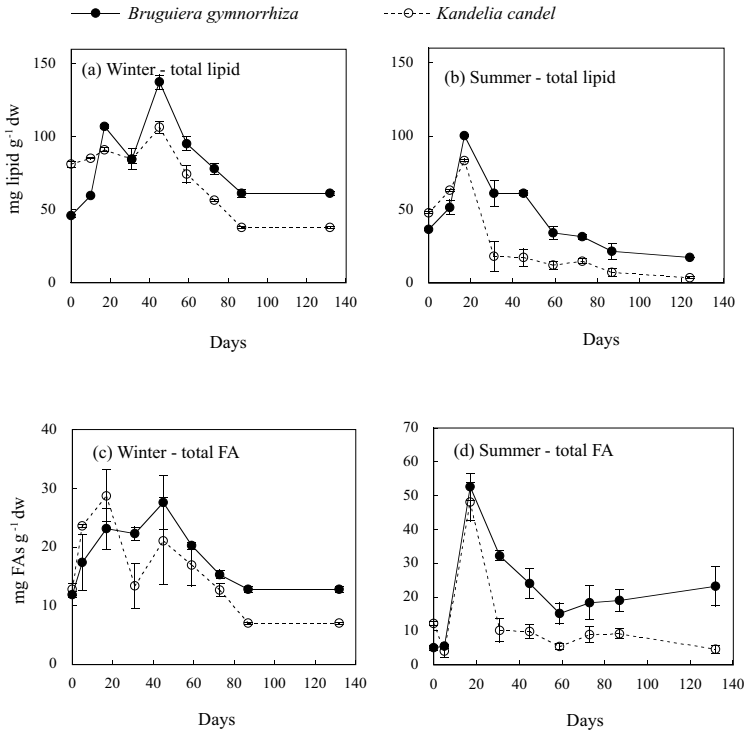


Fig. 1. Changes in total lipid and total fatty acid concentrations which occurred during the decomposition of *Bruguiera gymnorrhiza* and *Kandelia candel* yellow leaves during winter and summer. Data are means (±SD, n = 3).

et al. (2002). A second plate was prepared to estimate the proportion of fatty acid methyl esters (FAMEs) in the total lipids (Yamashiro *et al*., 1999). After drying, the plate was scanned (Epson GT-9000) and the image stored with Adobe Photoshop software (Adobe systems). An image analysis program (NIH image version 6) was used to estimate the relative contribution of the fatty acids, as a proportion of total lipid, by integrating the chromatogram.

The FAMEs were separated and quantified by a gas chromatograph (GC 14.B, Shimadzu) equipped with a flame ionization detector. Separation was performed with an FFAP-polar capillary column (30 m × 0.32 mm internal diameter, 0.25 μm film thickness). Helium was used as carrier gas. After injection at 60°C, the oven temperature was raised to 150°C at a rate of 40°C min^{-1}, then to 230°C at 3°C min^{-1}, and finally held constant for 30 min. The flame ionization was held at 240°C. Most FAMEs peaks were identified by comparing their retention times with those of authentic standards (Supelco Inc.) (Meziane *et al*., 2002).

2.4 Statistical analysis

A three-way ANOVA was performed, with species (2 levels), seasons (2 levels) and time (10 levels) were entered as fixed factors and used to compare change in lipid and FA concentration and mean percent weight loss of total lipid and total FA. The major FA classes were compared using MANOVA: saturated fatty acids (SAFAs), polyunsaturated fatty acids (PUFAs), monounsaturated fatty acids (MUFAs) and total branched chain fatty acids (BrFAs), and other FAs, even-long chain fatty acids (LCFAs), essential fatty acids (EFAs) and bacterial fatty acids (BFAs). Any significant species, season and time effect was further examined using Fisher's protected least significant difference (PLSD) test. The factor "time" in the statistical analysis represents age of the leaf from its green mature state, senescence and microbial decomposition. Raw concentration (mg g^{-1}) and percentage data (for total lipid and FA loss) were used in each analysis. The percent weight loss data could not be arcsine p-transformed because percentages greater than 100 were common in the data set. All statistical analyses were performed using StatView 5.0 software (SAS Institute Inc.). For all tests a criterion of $p < 0.05$ was used to determine statistical significance.

3. Results and Discussion

3.1 Dynamics of lipid content and total FA during decay

The pattern of change in lipid concentration and total FA (including the fatty acid classes) during decay varied between seasons and times for both species (signifi-

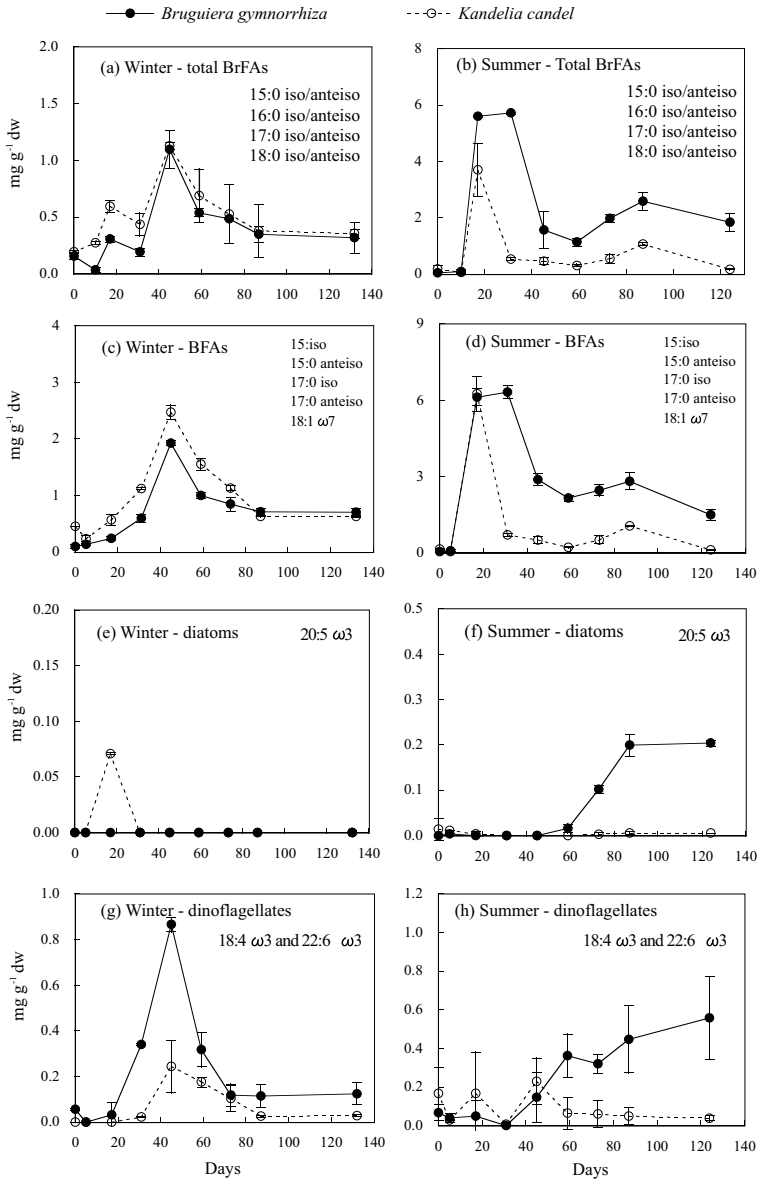


Fig. 2. Changes in concentration of branched chain fatty acids (BrFAs), bacterial fatty acids (BFAs) and fatty acid markers for diatoms and dinoflagellates which occurred during the decomposition of *Bruguiera gymnorrhiza* and *Kandelia candel* yellow leaves during winter and summer. Data are means (±SD, n = 3).

cant, species × season × time interaction in ANOVAs), which indicates that season and type of species had an effect on the concentrations of the lipid and total FAs in the fresh and decaying leaves (Fig. 1). In both species the concentration of lipid and total FAs and fatty acid classes, in particular BrFAs and BFAs, increased to a maximum in 45 days during winter and in 17 days during the summer experiment. Thereafter concentrations declined gradually until the end of the experiments (Figs. 1 and 2a–d). A secondary maximum was noticed for total lipid and total FA on the 17th day during winter. Apart from chemical changes in the lipids, the dramatic increase in lipid concentrations can probably be attributed to weight loss resulting from the removal of more labile compounds e.g. sugars and starches, by leaching, microbial colonization and input of organics from external sources. A time lag to maximum lipid and FA concentration during winter was probably a consequence of low temperatures, which affected microbial activity and possibly delayed microbial colonization.

Phospholipids are important chemical constituents of the membrane lipids and play a major structural role

Table 1. Total lipid, total fatty acid and fatty acid class concentrations (mg g^{-1} dry weight) in the green and yellow leaves of *Bruguiera gymnorrhiza* and *Kandelia candel* during winter and summer seasons. Lower case letters indicate post-hoc Fisher's PLSD results. A lower case letter "a" indicates significant difference between species and "b" indicates significant difference between seasons. Values without lower case letters are not significant different.

Species	Lipid	Winter				Summer			
	Fatty acid	Green		Yellow		Green		Yellow	
Bruguiera gymnorrhiza	Total lipid	32.3	(ab)	45.7	(ab)	35.8	(ab)	36.4	(ab)
Kandelia candel	Total lipid	50.8		81.2		41.6		47.7	
Bruguiera gymnorrhiza	Total FAs	13.6	(ab)	11.9	(ab)	4.3	(ab)	5.0	(ab)
Kandelia candel	Total FAs	15.9		12.8		12.2		12.1	
Bruguiera gymnorrhiza	SAFAs	5.1	(ab)	7.4	(ab)	3.9	(ab)	4.2	(ab)
Kandelia candel	SAFAs	4.7		8.6		9.6		8.8	
Bruguiera gymnorrhiza	MUFAs	1.9	(ab)	1.3	(b)	0.2	(ab)	0.3	(b)
Kandelia candel	MUFAs	1.5		1.5		1.0		0.7	
Bruguiera gymnorrhiza	PUFAs	6.3	(ab)	2.8	(ab)	0.2	(a)	0.4	(ab)
Kandelia candel	PUFAs	9.4		2.0		1.3		1.8	
Bruguiera gymnorrhiza	Total BrFAs	0.2		0.2		0.1		0.1	
Kandelia candel	Total BrFAs	0.2		0.2		0.2		0.2	
Bruguiera gymnorrhiza	even-LCFAs	0.6	(a)	0.8	(a)	0.1		0.3	
Kandelia candel	even-LCFAs	0.3		0.2		0.3		0.5	
Bruguiera gymnorrhiza	EFAs	6.3	(ab)	2.8	(ab)	0.1	(ab)	0.3	(ab)
Kandelia candel	EFAs	9.3		4.0		1.2		1.5	
Bruguiera gymnorrhiza	BFAs	0.1		0.1	(ab)	0.0		0.0	(b)
Kandelia candel	BFAs	0.2		0.5		0.1		0.1	

in the cytoplasmic membranes of animals and plants, including bacteria and fungi. For example, membrane lipids in most bacteria consist of the glycerophospholipids phosphatidyl-glycerol and phosphatidylethanolamine (Lopez-Lara *et al.*, 2003). A recent study indicates that rapid bacterial growth results in rapid increase in phospholipids in degrading organic matter in the marine environment (Goutx *et al.*, 2003). The rapid increase in total lipid content early in decay can be partly attributed to rapid growth of bacteria (Figs. 2c and 2d), probably fungi, and the products of their metabolic activity. Although detritus was thoroughly cleaned before analysis, other external sources including phytoplankton, zooplankton and tiny invertebrates, could have remained in the cleaned detrital samples and might have contributed to elevated lipid concentration. The detection of phytoplankton markers: diatoms 20:5ω3 and dinoflagellates 18:4ω3 and 22:6ω3 (Carrie *et al.*, 1998) indicated that diatoms and dinoflagellates colonized the decaying leaves (Figs. 2e–h).

The increase in total lipids could also be attributed to increases in other lipid compounds, such as sterols, during microbial decay. A study of marsh grass found that new free sterols, e.g. triterpenol and cholesterol, are produced in large amounts in aged detritus due to the breakdown of bound sterols present in the detritus (Lee *et al.*, 1980). Cholesterol is the most abundant sterol in many phytoplankton, zooplankton, benthic organisms and tiny invertebrates such as crabs (Nes and McKean, 1977). Although the invertebrates were entirely removed from the detritus, some microorganisms could have remained and might have accounted for the increase in sterol, and consequently total lipid concentration.

Ergosterol is a major constituent of fungi membranes (Olsson *et al.*, 2003) and has been used as a biomarker of saprophytic fungi in decaying plant material (Newell *et al.*, 1988; Newell, 1996; Mudge and Norris, 1997). Ergosterol concentration in the detritus was not measured in this study, but a previous study in the same mangrove forest, using mangrove leaves confined in mesh bags (inaccessible to invertebrates), indicated that fungal colonization increased rapidly early in decay (Mfilinge *et al.*, 2003), a similar time frame found for total lipids during the winter and summer experiments in this study, suggesting that fungal colonization also contributes to the increase in total lipid and FA in the decomposing mangrove detritus.

Total fatty acid (fatty acid methylated esters "FAMEs" and free fatty acids) concentration were significantly higher in the *Kandelia candel* fresh leaves than in *Bruguiera gymnorrhiza* during winter and summer (Fisher's PLSD, $p < 0.01$). The concentration of fatty acids in the fresh leaves (13–17 mg g^{-1}dry wt) during winter is comparable to values previously reported for Australian mangrove leaves by Wannigama *et al.* (1981) and Hogg and Gillan (1984) (Table 1). Of the major FA classes

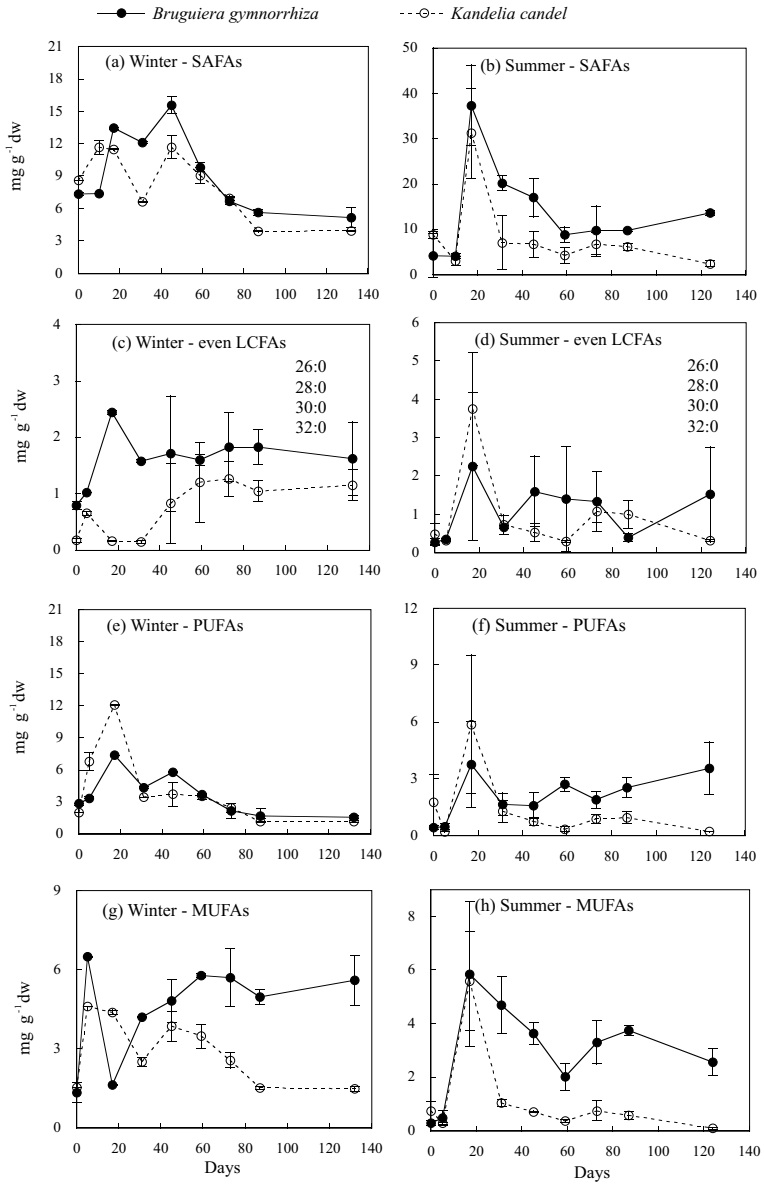


Fig. 3. Changes in concentration of saturated fatty acids (SAFAs), even long chain fatty acids (even-LCFAs), polyunsaturated fatty acids (PUFAs) and monounsaturated fatty acids (MUFAs) which occurred during the decomposition of *Bruguiera gymnorrhiza* and *Kandelia candel* yellow leaves during winter and summer. Data are means (±SD, n = 3).

detected in the fresh leaves, SAFAs dominated the FA compounds, followed by PUFAs in both species and seasons. There was significantly higher concentration of SAFAs in *B. gymnorrhiza* than *K. candel*, and significantly higher concentration of PUFAs in *K. candel* than in *B. gymnorrhiza* fresh leaves in both seasons (Table 1). This could be related to differences in leaf chemistry between the two species, because *K. candel* leaves contain higher N and lower C:N ratios than those of *B. gymnorrhiza* (Mfilinge *et al.*, 2002), which also indicates that *K. candel* leaves are of significantly higher nutritional quality than *B. gymnorrhiza* leaves.

The nutritional quality in these leaves was also indicated by their significantly higher concentrations of essential fatty acids (EFAs) ω3 and ω6 (Sargent *et al.*, 1990) than those of *B. gymnorrhiza*, which indicates that *K. candel* leaves are more nutritious than *B. gymnorrhiza* leaves and that they are likely to contribute more EFAs to the estuarine ecosystem and the ocean than those of *B. gymnorrhiza* leaves (Table 1). On the other hand, the higher concentration of PUFAs, in particular the EFAs, in the fresh leaves during winter than summer possibly

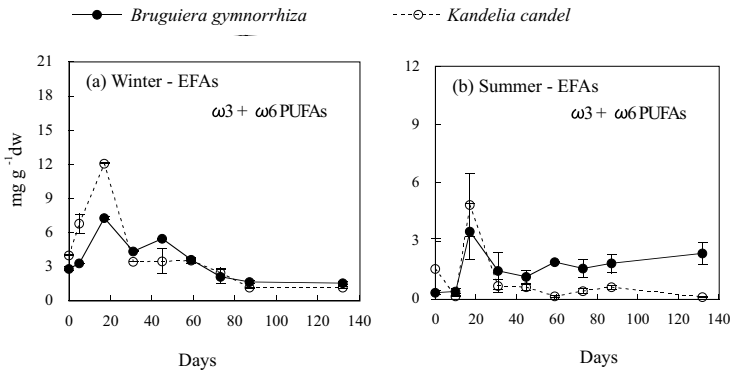


Fig. 4. Changes in concentrations of essential fatty acids (EFAs) which occurred during the decomposition of *Bruguiera gymnorrhiza* and *Kandelia candel* yellow leaves during winter and summer. Data are means (±SD, n = 3).

suggests an adaptation to the cold temperatures during winter, by biosynthesizing large quantities of PUFAs from SAFAs through desaturation reactions in order to maintain membrane fluidity and other cell functions such as photosynthesis at low environmental temperatures (Sargent *et al.*, 1990). The same reason could probably explain the observed higher lipid and total FA concentrations during winter than during summer.

During decomposition, total fatty acid including the major classes (SAFAs, PUFAs, MUFAs and total BrFAs) and markers for bacteria (BFAs), EFAs and even-long chain fatty acids (LCFAs) increased during the first 17 days and 45 days (for BrFAs and BFAs) (Figs. 3 and 4). The concentration of SAFAs in the detritus declined constantly with detritus age. However, there was no marked decline in concentrations of the long chain SAFAs, particularly those with >24 even numbered C atoms (even-long chain fatty acids, LCFAs) (Figs. 3c and 3d). These acids are synthesized only by vascular plants (Volkman *et al.*, 1980) and consequently have been used as markers of material of vascular plant origin, including mangroves in sediments, and animal tissue (Meziane and Tsuchiya 2002; Meziane *et al.*, 2002). These markers occur more abundantly in the mangrove sediments than in mangrove leaf detritus (Mfilinge *et al.*, 2003), suggesting that the acids are more resistant to microbial degradation than the short chain saturated FAs, and especially unsaturated fatty acid. This indicates that LCFAs may be preserved in mangrove sediments and nearby ecosystems for a long period without undergoing further transformation, and therefore may be useful as indicators of the contribution of mangrove lipid to the estuarine ecosystem and the ocean.

An increase in branched fatty acid concentrations (e.g. 15:0, 17:0 iso and anteiso), and MUFAs is indicative of significant bacterial production (Gillan and Hogg 1984), as was previously observed in decomposing mangrove leaves of *B. gymnorrhiza* and *K. candel* (Mfilinge *et al.*, 2003) and dead leaves of *Rhizophora mangle* (Findlay *et al.*, 1986). The present study found an increase in BrFAs, MUFAs and BFAs (odd-BrFAs + 18:1ω7) bacterial markers (Jeffries, 1972 and Volkman *et al.*, 1980) after 45 days during winter and 17 days during summer. This corresponds to significant increases in total FAs and total lipid in the detritus of both species, suggesting that bacterial activity plays a significant role in lipid and fatty acid transformation.

Diatom and dinoflagellate colonization in the detritus may have contributed to the increases in PUFAs (Figs. 2e–h and 3e) and EFAs, particularly in the *B. gymnorrhiza* detritus during summer (Fig. 4b). This might have increased the nutritional quality in the aged *B. gymnorrhiza* detritus making it more nutritious than its fresh leaves, as previously noted by Mfilinge *et al.* (2003).

3.2 Weight loss and contribution of mangrove leaves to lipids

The pattern of total lipid and total FA weight loss during decay varied between seasons and times for both species (significant, species × season × time interaction in ANOVAs), indicating that total lipid content and fatty acid were lost at different rates between the two species, and that season had an effect on the weight loss during decay. However, the total lipid content and FA in the detritus remained over 100% for some time, despite the detritus weight loss, possibly due to rapid microbial growth, breakdown of bound lipids originally present in the yellow leaves, diatom colonization and input of lipids from the surrounding sediment. The total lipid content and FA was lost faster in *Kandelia candel* than *Bruguiera gymnorrhiza* leaves due to high initial N content (low C:N ratio) in the yellow leaves that enhanced microbial growth and decay (Mfilinge *et al.*, 2002), and that the loss in weight was faster during summer than during the winter experiment because warm temperatures enhanced micro-

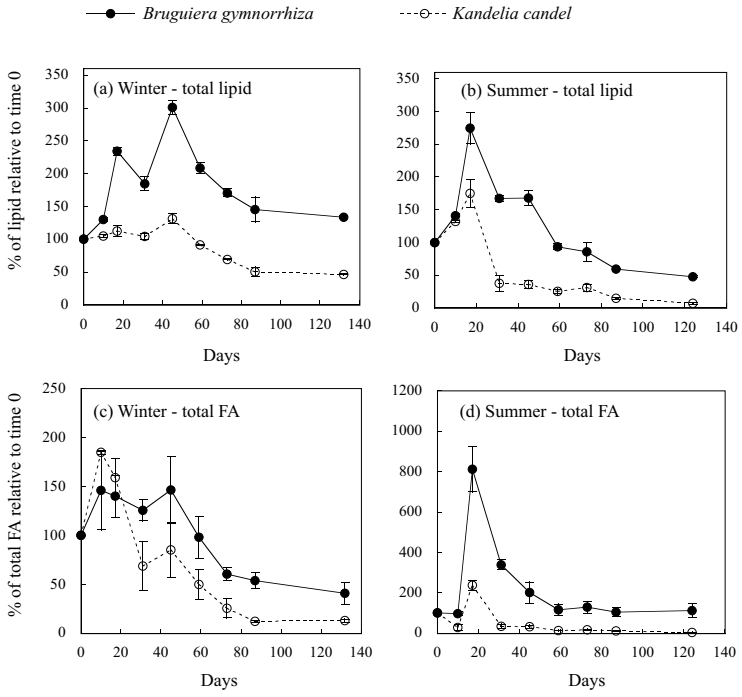


Fig. 5. Percent of lipid and total FAs remaining relative to time 0 in the decomposing leaf detritus of *Bruguiera gymnorrhiza* and *Kandelia candel* during winter and summer. Data are means (±SD, n = 3).

bial activity and decay (Fig. 5).

The estimate of amount of total lipid and total FA (based on the total lipid content and total FA initially present in the 5 g dry wt of yellow leaves) lost from yellow leaves of *B. gymnorrhiza* and *K. candel* during summer and winter experiments indicated a greater release of total lipids and fatty acids (in terms of total amounts contributed by both species in one season) to the estuarine environment during winter than summer (Fig. 6). This is because leaves, which fall during the winter season, contain larger amounts of lipid and fatty acid than those falling during summer (Table 1), and during decomposition these leaves are likely to contribute more lipids to the estuarine environment than leaves which fall during summer. Nevertheless, because the total litter production is the lowest during the winter season (~1 g $m^{-2}day^{-1}$ vs. 6 g $m^{-2}day^{-1}$ dry weight during summer and autumn) in Oura Bay mangrove (Mfilinge and Tsuchiya, unpubl. data), the contribution of lipids and fatty acids to estuarine sediments may be less than that of summer and autumn when leaf production is the highest.

The results also indicate that *K. candel* leaves may contribute more lipids (and fatty acids) than *B. gymnorrhiza* leaves, and therefore this species is likely to contribute more FA compounds to the surrounding sediments (Fig. 6). However, there is much less leaf litter production by *K. candel* in Oura Bay than by *B. gymnorrhiza* (Hardiwinoto *et al.*, 1989; Mfilinge and Tsuchiya, unpubl. data), and therefore its lipid contribution to the estuarine environment may not be significant compared to *B. gymnorrhiza*.

The estimated total leaf litter production by *Bruguiera gymnorrhiza* in Oura Bay mangroves during the study was 9.09 t $ha^{-1}yr^{-1}$ (Mfilinge and Tsuchiya, unpubl. data). Assuming that all leaf litter decayed at the same rate, the estimated amounts of total lipid and total FA released per year to the marine environment from *B. gymnorrhiza* decomposing leaves, ignoring contribution by external sources, would be: 0.33 t $ha^{-1}yr^{-1}$ of total lipids and 45.3 kg $ha^{-1}yr^{-1}$ of total fatty acids. Of the total fatty acid; SAFAs may contribute 38.2 kg $ha^{-1}yr^{-1}$, PUFAs 3.8 kg $ha^{-1}yr^{-1}$, MUFAs 2.6 kg $ha^{-1}yr^{-1}$, EFAs 2.9 kg $ha^{-1}yr^{-1}$ and BrFAs 0.5 kg $ha^{-1}yr^{-1}$. Contributions were estimated by:

$$T_{LF} \times L_{WT}/5g,$$

where T_{LF} = total annual leaf fall in the 10 ha mangrove forest; L_{WT} = weight of a lipid or FA, originally present in the 5 g dry weight of yellow leaves. The estimated total lipid amount (0.33 t $ha^{-1}yr^{-1}$) is much less than the estimate for C contribution (2.24 t ha^{-1} per 4 months) from *B. gymnorrhiza* mangrove leaves in the same mangrove forest during the period of highest leaf litter fall (July to

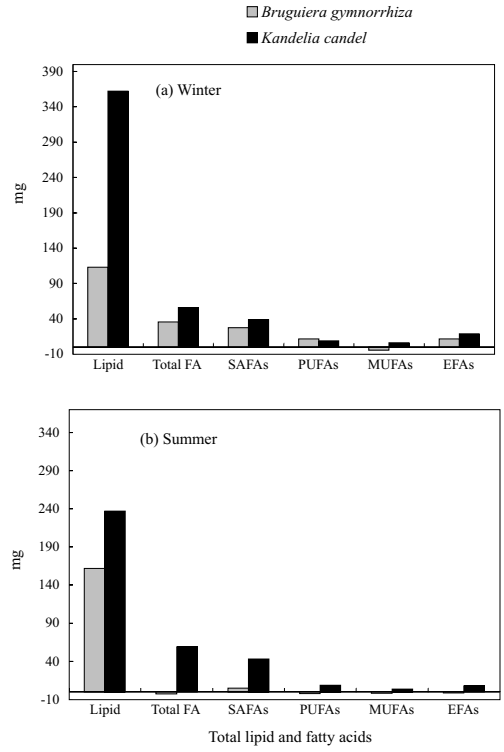


Fig. 6. Absolute amounts of lipid, total FA, major fatty acid classes and EFAs lost from mangrove leaves during the winter and summer experiments based on amounts initially present in the 5 g dry weights of yellow leaves and the last day of the experiment, 132nd for the winter and 124th for the summer. SAFAs (Saturated fatty acids); PUFAs (Polyunsaturated fatty acids); MUFAs (Monounsaturated fatty acids) and EFAs (essential fatty acids).

October) on Okinawa Island (Mfilinge *et al*., 2002).

In conclusion results indicate that lipid present in mangrove leaves is lost faster in *Kandelia candel* than *Bruguiera gymnorrhiza*, more rapid in the summer season than winter season and that from a qualitative point of view *K. candel* may contribute more to the lipid input into marine sediments than *B. gymnorrhiza*, but from a quantitative point of view *B. gymnorrhiza* is the principal source of lipid in the marine sediments of Oura Bay estuary. This quantitative comparison of total lipid and total FA weight loss between species and season during mangrove leaf decay is likely to provide base-line data important to our understanding of lipid inputs to estuarine sediments.

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