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Mangrove propagule size and oil contamination effects: Does size matter?

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ABSTRACT

Three mangroves species with differential propagule size, *Avicennia marina* (2.5 ± 0.3 cm), *Bruguiera gymnorrhiza* (16 ± 2 cm) and *Rhizophora mucronata* (36 ± 3 cm), were subjected to oil contamination. In a series of glasshouse and field experiments, the sediment, propagules, leaves and stems were oiled and growth monitored. Oiling of the propagules, leaves, internodes or sediment reduced plant height, leaf number, leaf chlorophyll content index and induced growth abnormalities, leaf abscission and mortality, with effects being greatest in *A. marina*, intermediate in *R. mucronata* and least in *B. gymnorrhiza*. The results suggest that the greater susceptibility of *A. marina* to oil is due to early shedding of the protective pericarp and rapid root and shoot development after detachment from the parent tree and not to propagule size. After seedling emergence, micromorphological factors such as presence of trichomes, salt glands and thickness of protective barriers influence oil tolerance.

1. Introduction

Oil spills occur throughout the world, primarily from ships, but also from oil wells pipelines and other petrochemical infrastructure. Crude oil is composed of up to 17,000 organic compounds, of which about 75% are hydrocarbons (Bjorlykke, 2011). The light and medium compounds in oil remain at the surface and volatilize or degrade over time, while those with high molecular weight are deposited on sediments and retain toxic properties for years (Reed et al., 1999). Despite their hydrophobic nature, polycyclic aromatic hydrocarbons (PAHs) can bio-concentrate and are toxic and mutagenic to organisms (Duke et al., 1998; Meudec et al., 2006).

Mangroves, which inhabit low wave energy, sheltered locations in intertidal zones of tropical and subtropical regions, are highly vulnerable to oil spills (Mille et al., 2006). Oil can penetrate into soft sediments and coat aerial roots (Getter et al., 1985; Proffitt et al., 1995) leading to oxygen deficiency, suffocation and growth irregularities in the short-term and mortality in the long-term (Zhang et al., 2007; Cavalcante et al., 2009). Oil can persist in the mangrove environment under typical reduced conditions. Several studies have documented the effect of different types of crude oil on mangrove species under laboratory (Youssef and Ghanem, 2002b; Zhang et al., 2007) and field conditions (Tam et al., 2005; Naidoo et al., 2010).

Studies on oil contamination have frequently concentrated on the mass mortality of plants after large-scale crude oil spills (Getter et al., 1985; Duke et al., 1998). Some studies focused on the effects of oil contamination on seedling establishment and growth under sub-lethal concentrations (Proffitt et al., 1995). Responses of mangroves to oil treatment include reduction in foliage production and total plant biomass (Mille et al., 2006; Ke et al., 2011), reduction in the rate of photosynthesis and respiration (Getter et al., 1985; Naidoo et al., 2010), increased mutation (Cavalcante et al., 2009), development of anomalous growth forms (Tam et al., 2005), damage to root cell membranes (Watts et al., 2006; Kang et al., 2010), impairment of transpiration (Youssef and Ghanem, 2002a), and increased mortality (Tam et al., 2005). The effects of oil on germination and growth of mangroves under sub-lethal concentrations are poorly understood. In addition, there is little or no information on the effects of mangrove propagule size on oil contamination effects. This work focuses on oil effects on the establishment and early development of seedlings, a critical life stage for mangroves. Many mangrove species have large propagules and the reserves contained within them support growth for an extended period of time (Tomlinson, 1994). The maternal reserves of the propagules may thus exert a large influence on early seedling growth under stressful conditions.

The aim of this study was to determine the effects of oil contamination on the growth of three mangrove species (*Avicennia marina*, *Bruguiera gymnorrhiza* and *Rhizophora mucronata*) with distinct differences in propagule size. Species with large propagule size and greater maternal reserves, such as *R. mucronata* and *B. gymnorrhiza*, would be

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expected to be more tolerant of oil contamination than those with smaller propagules, such as *A. marina*. This hypothesis was tested by determining the effects of oiling of the propagules, stems and leaves, as well as of the sediment, in glasshouse and field experiments.

2. Materials and methods

2.1. Glasshouse study

Propagules of *A. marina* and *B. gymnorrhiza*, were collected from the Beachwood Mangroves Nature Reserve (29°48′ S, 31°02′ E) and those of *R. mucronata* from the Isipingo Estuary (29°59′ S, 30°56′ E). After collection, *A. marina* propagules were placed in water and pericarps allowed to shed naturally for 24 h. Propagules of *A. marina* were planted in 17 cm diameter × 15 cm height and those of *B. gymnorrhiza* and *R. mucronata* in 24 cm × 21 cm plastic pots. All pots contained a mixture of sand, potting soil and compost (1:2:1 volume based). Pots were watered daily with tap water, once monthly with 10% seawater and maintained in a glasshouse for 13 months at 25 °C (day) and 18 °C (night).

Propagules of *A. marina* (2.5 ± 0.3 cm) were subjected to one of five treatments:

i. Control (C): propagules with intact pericarps, no oil applied;
ii. SO: propagules with intact pericarps and 50 ml of oil carefully poured onto the soil surface;
iii. 1/2PO: 1/2 of the propagule with an intact pericarp was dipped in oil;
iv. PO1: propagules with intact pericarps were completely dipped in oil;
v. PO2: propagules without pericarps were completely dipped in oil.

Propagules of *B. gymnorrhiza* (16 ± 2 cm), and *R. mucronata* (36 ± 3 cm) were subjected to one of three treatments:

i. Control (C): no oil applied;
ii. SO: 50 ml of oil carefully poured onto the soil surface;
iii. 2/3PO: 2/3 of the propagule from the radical end was dipped in oil.

Each treatment was replicated seven times. Bunker fuel oil 150, obtained from FFS Refineries (Pty) Ltd., Durban was used in this study. The properties of this oil were described previously (Naidoo et al., 2010). Plant height and leaf number were determined monthly for the duration of the study (13 months). Chlorophyll content index was determined on the top leaf four months after commencement of treatments and monthly thereafter with a hand-held chlorophyll absorbance meter (CCM-200, Opti Sciences, Tyngsboro, MA, USA).

2.2. Field study: leaf and internode oiling

Field investigations were undertaken in the Beachwood Mangroves Nature Reserve and Isipingo Estuary. The average daily maximum and minimum temperatures for Beachwood are 26 °C and 17 °C, and for Isipingo, 27 °C and 16 °C, respectively. The mean annual rainfall is 1228 mm for Beachwood and 1040 mm for Isipingo. Soil characteristics for the sites are provided in Table 1. Selected individuals of each species, growing naturally in the field, were selected at random and tagged for measurements.

Healthy, naturally occurring, one year old seedlings of *A. marina* (46.5 ± 7 cm) and *B. gymnorrhiza* (48.5 ± 8 cm) in Beachwood and *R. mucronata* (66.5 ± 9 cm) in Isipingo were subjected to one of three treatments:

i. C: control plants were untreated;
ii. IO: the second internode from the base of the stem of *A. marina* (the first internode was too short) and the first internode of *B. gymnorrhiza* and *R. mucronata* were oiled with a brush;
iii. LO: both surfaces of the last pair of mature leaves, directly below the shoot tip of each plant, were oiled; Internodes and leaves of *A. marina* and *B. gymnorrhiza* were oiled at the commencement of the experiment and again after 7, 15, 19, 27, 32, and 48 weeks, while those of *R. mucronata* were oiled at the commencement of the experiment and again after 5, 14, 18, 25, 30 and 48 weeks. Initially, leaves of all three species were oiled on the adaxial surface only but after15 weeks, both surfaces were oiled. Each treatment was replicated seven times for each treatment-species combination. Plant height and leaf number were determined.

2.3. Field study: sediment oiling

Field sediment oiling was undertaken in the Isipingo Estuary. Healthy one to two year old seedlings of *A. marina* (110 ± 5 cm), *B. gymnorrhiza* (65 ± 6 cm) and *R. mucronata* (76 ± 8 cm) were selected. Groups of seedlings of each species were enclosed by 0.5 m radius × 0.5 m height perspex cylinders which were inserted into the soil to a depth of 5 cm to prevent the escape of oil from the enclosures. Control plants were enclosed in a similar fashion, but no oil was applied. Each treatment was replicated four times. Oil dosage was 5 l m^{-2} for all plots and monitoring continued for 12 months. Measurements of plant height and leaf number were taken 16, 26, 38, 46 and 53 weeks after oiling.

2.4. Soil analysis

Soil samples from a depth of 0–15 cm were collected from the experimental sites in Beachwood and Isipingo, air dried, crushed with a wooden mallet and passed through a 1 mm sieve. Soil pH was measured

Table 1
Soil characteristics of the study sites at Beachwood Mangroves Nature Reserve and Isipingo Estuary. Values are means ± SE, n = 3.

		Beachwood	Isipingo
Salinity (psu)		19 ± 3	26 ± 4
Redox potential (E_h) mV		−40 ± 4	−32 ± 3
Clay content %		27.7 ± 2	23.3 ± 3
pH		6.2 ± 0.2	6.6 ± 0.4
P	$g\ l^{-1}$	0.06 ± 0.003	0.03 ± 0.001
K		0.8 ± 0.02	0.5 ± 0.01
Ca		1.3 ± 0.01	1.8 ± 0.02
Mg		1.7 ± 1.9	1.2 ± 0.05
Zn		0.08 ± 0.002	0.32 ± 0.003
Mn		0.05 ± 0.002	0.03 ± 0.001
Cu		0.01 ± 0.001	0.02 ± 0.003
N		4.7 ± 0.1	5.4 ± 0.05
C		47.0 ± 4.3	51.7 ± 3.8

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