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Short communication

A simple and cost-effective method for cable root detection and extension measurement in estuary wetland forests

Alejandra G. Vovides [a, *], Beatriz Marín-Castro [b], Guadalupe Barradas [c], Uta Berger [a], Jorge López-Portillo [b]

[a] Department of Forest Biometry and Systems Analysis, Institute of Forest Growth and Forest Computer Sciences, Technische Universität Dresden, Pienner Str. 8, 01735 Tharandt, Germany
[b] Functional Ecology Network, Instituto de Ecología A.C. (INECOL), Carretera Antigua a Coatepec 351, Xalapa 91070, Veracruz, Mexico
[c] Social Cooperative on Sustainable Tourism "La Mancha en Movimiento S. de S.S.", La Mancha s/n, Actopan 91496, Veracruz, Mexico

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ABSTRACT

This work presents the development of a low-cost method to measure the length cable roots of black mangrove (*Avicennia germinans*) trees to define the boundaries of central part of the anchoring root system (CPRS) without the need to fully expose root systems. The method was tested to locate and measure the length shallow woody root systems. An ultrasonic Doppler fetal monitor (UD) and a stock of steel rods (SR) were used to probe root locations without removing sediments from the surface, measure their length and estimate root-soil plate dimensions. The method was validated by comparing measurements with root lengths taken through direct measurement of excavated cable roots and from root-soil plate radii (exposed root-soil material when a tree tips over) of five up-rooted trees with stem diameters (D_{130}) ranging between 10 and 50 cm. The mean CPRS radius estimated with the use of the Doppler was directly correlated with tree stem diameter and was not significantly different from the root-soil plate mean radius measured from up-rooted trees or from CPRS approximated by digging trenches. Our method proved to be effective and reliable in following cable roots for large amounts of trees of both black and white mangrove trees. In a period of 40 days of work, three people were capable of measuring 648 roots belonging to 81 trees, out of which 37% were found grafted to other tree roots. This simple method can be helpful in following shallow root systems with minimal impact and help map root connection networks of grafted trees.

1. Introduction

The accessibility to below-ground biomass has limited our knowledge on structural-functional aspects of root systems, especially for large plants (Danjon et al., 2013). Most existing methodologies are destructive and either require the full excavation of root systems (Danjon et al., 2005; Smith et al., 2014), or pulling trees until up-rooted (Blackwell et al., 1990; Coutts, 1983; Crook and Ennos, 1998; Gasson and Cutler, 1990; Ray and Nicoll, 1998; Sapijanskas et al., 2014), an irreversible disturbance and destructive strategy that in many cases cannot be performed with species enlisted in the IUCN red list. Strategies to study roots *in situ* other than excavating the whole root system have been developed more recently, like rhizotrons, ground penetrating radar (GPR), and the use of medical instrumentation such as X-ray computed tomography (CT, Taylor et al., 1991; Perez et al., 1999; Butnor et al., 2001) and magnetic resonance imaging (MRI, Fang et al., 2012). Rhizotrons are structures with glass windows that allow the direct measurements of roots growing in the soil. (Taylor et al., 1991). Ground Penetrating radar technology is a fully non-destructive method that operates transmitting electromagnetic waves through the soil and records times of reflection to 3D images of the buried materials (Nadezhdina and Čermák, 2003; West, 2009). Finally, the use of medical instrumentation such as the CT and MRI allow for 3D reconstruction of fine root structure within intact core samples (Fang et al., 2012).

While rhizotrons are effective to estimate below ground biomass, root growth-rates and rhizosphere dynamics, they are unsuitable for mechanical stability studies because measurements can only be performed on root tissues that come in contact with the

* Corresponding author.
E-mail address: vovides@forst.tu-dresden.de (A.G. Vovides).

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glass (Burke and Raynal, 1994; Taylor et al., 1991). On the other hand the CT, GPR and MRI point to a promising non-invasive methods for detailed studies on root structure of plants, nevertheless these are technologies of high economical costs (no less than USD 10,000 for GPR), and are still under development (Fang et al., 2012). To date, GPR has only been used to estimate stand level below ground biomass (Barton and Montagu, 2004; Butnor et al., 2003; Danjon et al., 2013), while CT and MRI can only be performed on soil cores extracted from the field and are highly sensitive to water content, making them inappropriate for wetland forested system studies (Butnor et al., 2001; Fang et al., 2012; Luo et al., 2008; Perez et al., 1999).

Studies on anchoring systems of large plants, to date, still relay in complete excavation of root systems to perform structural analysis through the use of terrestrial laser scanning (Danjon et al., 2013, 2005; Smith et al., 2014), or to pulling and up-rooting mechanisms to characterize the strength of root-soil plates (Blackwell et al., 1990; Coutts, 1983; Coutts et al., 1999; Cucchi et al., 2004). A trees root-soil plate, referring to the section of woody roots and soil that get exposed after mechanical failure of the stem, is the object of most studies dealing with tree mechanical stability and resistance to wind damage (Coutts, 1983). For standing stems, this region is known as the "central part of the anchoring root system", (hereafter CPRS) and represents the main area of plant anchorage (Coutts, 1983; Danjon et al., 2005; Stokes et al., 2005). While tree stability depends on root structure, the latter is influenced by soil structure; trees growing on deeper soils will have more vertical root growth than on shallow soils (Ray and Nicoll, 1998; Stokes et al., 2005) or at sites with a high water table that creates anoxic condition (Coutts, 1983; Keeley, 1988; Ray and Nicoll, 1998), thus limiting the development of deep roots. Wetland trees, like the black mangrove (*Avicennia germinans*), lack a tap root, or vertical sinker roots to increase anchorage, and root development is limited to the first 20–30 cm below ground surface (López-Portillo et al., 2005; McKee, 2001), thus, trees must compensate stability by growing longer horizontal woody roots. Still, the lack of a deep rooting system makes trees more vulnerable to windthrow (uprooting due to wind forces), and it represents a particular risk in water-saturated soils (Coutts, 1983; Krause et al., 2014).

While our knowledge on mangrove wetlands has increased dramatically in the last few decades (Alongi, 2008, 2002; Field et al., 1998; Srikanth et al., 2015; Twilley, 1988), our understanding of their root system is limited to areal structures, biomass estimations and functional anatomy and physiology (Angeles et al., 2002; Brooks and Bell, 2005; Castañeda-Moya et al., 2011; Komiyama et al., 2000; Mendez-Alonzo et al., 2015; Ohira et al., 2012; Srikanth et al., 2015), while knowledge of the structure of the anchoring system becomes urgent to better understand and predict their mitigation effect on surges and ecosystem responses to environmental change (Srikanth et al., 2015). As previous studies on terrestrial forests show that the length of lateral roots, and thus the CPRS, increases with tree size (Smith et al., 2014), this study proposes a low-invasive method based on the application of the Doppler effect to detect and measure woody root lengths without digging trenches. The Doppler effect referrers to the change in the frequency of a wave, for an observer moving relative to the source of the wave (Maulik, 2006). This principle was first described for light wave movements by Christian Doppler in 1842, and latter verified with sound waves in 1844 (Maulik, 2006). Using this principle, a simple method was developed to measure the length of cable roots to approximate CPRS diameters of the species *A. germinans* with the use of a few steel rods (hereafter SR) and a portable ultrasonic fetal Doppler (hereafter UD).

The portable UD holds a transducer, a receiver and an amplifier; the transducer sends out an ultrasonic signal (a frequency higher than humans are capable of hearing), which travels through the surface it is in direct contact with. When the emitted high frequency waves encounter movements (i.e. the blood flowing in an artery or a heart beating), the waves bounce back modified by the frequency of the encountered waves, then the received frequency is further amplified into an audible signal (Maulik, 2006). The Doppler effect system can help in the detection of woody roots connected to a stem without digging trenches; if a sound wave is created on a given root by gently hitting on it with a SR, and the probe of the UD is located in the collar ring of a stem, the ultrasonic waves traveling from the UD through the stem and roots, will bounce with the waves generated by the SR and travel back to the UDs receiver, causing a positive signal in the UD, expressed as an audible sound and a frequency equal to that of the SR hitting on the root. The sensitivity of the UD is high enough to monitor the heartbeat of a five to 7.6 cm long (8–12 weeks) human embryo (Papaioannou et al., 2010), and has been successfully employed to measure the heart rate of wrasse fish (*Notolabrus celidotus*) and small crab species with heart rates twice as high than a human heart rate at 13 weeks of development (Iftikar and Hickey, 2013; Iftikar et al., 2010; Papaioannou et al., 2010).

This work shows the ability to effectively measure the length of horizontal woody roots and further approximate the size of the CPRS polygon with a major reduction on costs and time investment through the use of the UD. Our hypothesis is that the CPRS, in wetland trees with cable root development, is mainly delimited by woody cable roots, thus the estimated CPRS radius will be similar to the radius of root-soil plates of uprooted trees of the same species. To test the accuracy of the developed method, data were compared between the measurements taken with the UD and (1) root-plate radius of uprooted trees found in the field; (2) lengths taken through the use of SRs without UD; and (3) through the excavation and direct measurements of roots. The potential applications of this method, for wetland forest woody root research, is discussed.

2. Methods

2.1. Study site

The method was developed between October and November 2015 and validated during the month of July 2016, in a mangrove ecosystem form the central Gulf Coast of Mexico, in the La Mancha Lagoon (19°35′N, 96°22′W). This region has an average annual precipitation between 1200 and 1500 mm and a mean annual temperature of 25 °C, with minimum and maximum temperatures of 22° and 28 °C in January in May, respectively (López-Portillo et al., 2005). The lagoon is surrounded by 300 ha of mangrove forest co-dominated by *Avicennia germinans* (black mangrove), *Rhizophora mangle* (red mangrove) and *Laguncularia racemosa* (white mangrove). Two main mangrove geomorphic habitats are recognized in the area: Mangrove-vegetated mudflats and interdistributary basins. The first is characterized by the accumulation of clay and loam sediments, and the latter is dominated by organic-rich sediments related to a marked fresh-water influence (Thom, 1967; Vovides et al., 2014). Salinity within these sediments can range between 600 and 1200 mM NaCl (Vovides et al., 2014), while soil compaction in the area ranges between 1.13 and 4.8 kg cm^{-2} (Vovides et al., 2016), which means sediments are soft and easily penetrable.

2.2. Root length measurement with a portable Doppler

To approximate the CPRS polygon for the mangrove species *A. germinans*, 51 trees were selected, with stem diameters measured at 130 cm of height (D_{130}) ranging between 10 and 96 cm.