M. Waycott · P.A.G. Barnes

AFLP diversity within and between populations of the Caribbean seagrass *Thalassia testudinum* (Hydrocharitaceae)

Received: 14 September 2000 / Accepted: 29 May 2001 / Published online: 12 September 2001
© Springer-Verlag 2001

Abstract Genetic variation was assessed in the seagrass *Thalassia testudinum* from three regions of the Caribbean and north Atlantic using allozyme electrophoresis and amplified fragment length polymorphism (AFLP) analysis. Very low allozyme variability was detected among the 196 shoots analyzed from a range of sites in the San Blas region of Panama. AFLP markers detected high similarity (0.87) among the population samples surveyed from Bermuda and Panama across six AFLP primer pairs and over 260 banding positions. High levels of gene flow were detected between all the sites analyzed (*Nm* > 1.7). Significantly complete genotypic similarity was observed between samples from Bermuda and Panama, indicating that long distance vegetative fragment dispersal is highly probable. Very low genetic differentiation between all sites, even Bermuda and Panama, some 2,700 km apart, agrees with other studies and is further evidence of a highly uniform gene pool in *T. testudinum*. High levels of genetic uniformity in *T. testudinum* may be related to long-term environmental change over its geographic range. While AFLP analysis proved useful in determining genetic variation in this seagrass, the application of co-dominant markers such as microsatellites will be more informative in determining the nature of genetic uniformity and its adaptive significance in *T. testudinum*.

Introduction

Seagrass communities are an essential component of global, nearshore marine habitats (Larkum et al. 1989). As marine angiosperms, seagrasses have adopted both sexual and vegetative methods of recruitment and survival (Inglis 2000). A greater understanding of seagrass population dynamics is essential to understanding long- and short-term factors in how these important communities are established and maintained (Duarte 1999; Waycott 2000a). The vegetative growth habit of seagrasses [via rhizomes in the guerilla habit of Lovett-Dougt (1981)] anchors the plants and provides stability and resistance to water currents and wave action (Arber 1920). The fine details of rhizome growth form adopted by various seagrass species are being understood through an integrated approach of modeling and empirical analyses (Marba and Duarte 1998). However, the long-term (i.e. evolutionary) importance of the guerilla vegetative growth strategy in seagrasses remains unexplored, particularly with respect to clonal dispersal and to survival following disturbance (Waycott 2000b).

Early observations among many hydrophilous aquatic plant species of low genetic variability led Les (1988) to suggest that the predominance of vegetative and/or asexual reproduction results from a greater fitness among genetically uniform offspring to preserve adaptive gene complexes. Obviously, testing this concept requires detailed knowledge of not only the genetic diversity of species but also the relationship of genetic diversity to survival and its adaptive significance. Clonal growth might be viewed, therefore, as a significant factor in the slow evolutionary rates among hydrophiles (Les 1988; Waycott 2000b). Genetic diversity studies of seagrass populations, in conjunction with analysis of evolutionary divergence among seagrass lineages, will improve our understanding of the impact of clonality on seagrass evolution.

Genetic variation, and the partitioning of that variation within and between populations, is an informative...
measure of population processes in plants (Hamrick and Godt 1989). To date, genetic diversity of seagrass populations has been assessed extensively in only a few species, with studies demonstrating a wide range in genetic variability from zero in Amphibolis antarctica (Waycott et al. 1996) to moderate in Thalassia testudinum (Kirsten et al. 1998; Davis et al. 1999) and Posidonia oceanica (Procaccini and Waycott 1998) to high in Posidonia australis (Waycott 1995; Waycott et al. 1997) and Zostera marina (Reusch et al. 1998; Ruckelshaus 1998). Partitioning of variation within and between populations is often high (e.g. P. australis) (Waycott et al. 1997), although in many studies to date inadequate population sampling reduces the interpretability of reported results. These observations, at least in part, conflict with earlier studies, thus emphasizing the need for detailed study before we can test models of adaptive gene complexes and the significance of genetic diversity among hydrophiles.

*T. testudinum*, commonly known as turtle grass, occurs throughout the Caribbean and Gulf of Mexico (den Hartog 1970). Research activities on this seagrass have tended to focus on productivity, community ecology, and ecophysiology with gaps in our understanding of population dynamics (e.g. Zieman et al. 1999). Of particular interest has been the observation of die-off in *T. testudinum* meadows over large areas in Florida Bay, Florida, USA (Robblee et al. 1991). This has led, in part, to the interest in population genetic studies of this species to determine if, perhaps, there is a genetic component to the turtle grass die-off (Kirsten et al. 1998; Davis et al. 1999). Our understanding of population genetic structure, the adaptive significance of observed genetic diversity and the ability of seagrasses to recover from different types of disturbances is only beginning to develop (e.g. Ruckelshaus 1998) and research into this area of seagrass biology is of critical importance.

In this paper, we describe the population genetic diversity of *T. testudinum* from one region in the Caribbean and one region in the north Atlantic using amplified fragment length polymorphism (AFLP) analysis. This relatively new DNA fingerprinting technique (Vos et al. 1995) was, prior to this study, untested in seagrass population genetic analysis. We use these data to describe the extent and distribution of clonal ramets over different spatial scales and to infer population dynamics from the pattern of genetic variation among regions observed.

**Materials and methods**

Field collections

Shoot samples of *Thalassia testudinum*, sampled quantitatively and considered representative of the population, were obtained from one location within the Caribbean (San Blas Archipelago, Republic of Panama) and one location in the north Atlantic (Bermuda). San Blas collections of *T. testudinum* were made in the northern sector of the San Blas archipelago among coral reefs adjacent to Punta San Blas (San Blas Point). Grab samples were collected from an additional site in the Caribbean: Florida Bay, Florida (Fig. 1).

![Fig. 1 Inset Distribution map of the seagrass *Thalassia testudinum* (dotted outline), land is shaded grey. The Caribbean and Atlantic collection locations are indicated. Main map Collection sites of *T. testudinum* in the San Blas archipelago, Panama, land is shaded grey, major coral reefs are outlined](image)