

学校编码: 10384

分类号\_\_\_\_\_密级\_\_\_\_\_

学 号: 21720091152185

UDC\_\_\_\_\_

厦 门 大 学

硕 士 学 位 论 文

福建省九龙江口外来种无瓣海桑扩散种群的遗传结构研究

Study on Population Genetic Structure of alien species  
*Sonneratia apetala* in the Jiulong River Estuary of Fujian

王 莹

指导教师姓名: 杨盛昌 副教授

专业名称: 细胞生物学

论文提交日期: 2012 年 05 月

论文答辩时间: 2012 年 05 月

学位授予日期: 2012 年 月

答辩委员会主席: 李振基 教授

评 阅 人: \_\_\_\_\_

2012 年 05 月

福建省九龙江口外来种无瓣海桑扩散种群的遗传结构研究

王 莹

指导教师: 杨盛昌 副教授

厦门大学



## 厦门大学学位论文原创性声明

本人呈交的学位论文是本人在导师指导下,独立完成的研究成果。本人在论文写作中参考其他个人或集体已经发表的研究成果,均在文中以适当方式明确标明,并符合法律规范和《厦门大学研究生学术活动规范(试行)》。

另外,该学位论文为“内源脱落和赤霉素在红树植物胎生过程中的协同作用”课题的研究成果,获得国家自然科学基金项目(NO. 30972334)的资助,在滨海湿地生态系统教育部重点实验室实验室完成。

声明人(签名):

年 月 日

厦门大学博硕



# 厦门大学学位论文著作权使用声明

本人同意厦门大学根据《中华人民共和国学位条例暂行实施办法》等规定保留和使用此学位论文，并向主管部门或其指定机构送交学位论文（包括纸质版和电子版），允许学位论文进入厦门大学图书馆及其数据库被查阅、借阅。本人同意厦门大学将学位论文加入全国博士、硕士学位论文共建单位数据库进行检索，将学位论文的标题和摘要汇编出版，采用影印、缩印或者其它方式合理复制学位论文。

本学位论文属于：

（      ） 1. 经厦门大学保密委员会审查核定的保密学位论文，  
于 年 月 日解密，解密后适用上述授权。

（      ） 2. 不保密，适用上述授权。

声明人（签名）：

年 月 日

2012 年 月



## 摘 要

无瓣海桑(*Sonneratia apetala*) 是红树林中优良乔木树种之一,天然分布于印度、孟加拉国、斯里兰卡等国盐度较低的泥质滩涂。

作为我国从国外引进的第一种红树植物,无瓣海桑具有生长快、生产力高等特点,在生产中被大量推广,并获得较好的生态、经济和社会效益。除此之外,无瓣海桑还被广泛地应用于控制外来入侵植物互花米草的蔓延。但无瓣海桑毕竟是外来种,是否会造成生物入侵并给滨海滩涂带来生态灾害目前尚无定论,但比较一致的看法是无瓣海桑有一定扩散和竞争能力,所以引进该树种应慎重。本实验通过研究福建九龙江口以及种源地海南东寨港的无瓣海桑种群的遗传多样性及其遗传结构,探讨该地无瓣海桑种群的扩散模式,为针对其合理的管理模式以及生态风险评估提出理论依据。

对福建九龙江口外来种无瓣海桑的种群生长特征进行分析,发现该地无瓣海桑个体较为集中,生长较为一致,并且小径级的植株较多,说明其种群自然更新较快,能更好地适应该地区的自然环境。

本研究参照 Chen Tao 设计的 14 对海桑属通用 SSR 引物,并从中筛选出了 12 对(Sca22、Sca23、Sca126、Sca224、Sca231、Sca234、Sca249、Sca266、Sca270、Sca284、Sca285、Sca286)可扩增出清晰、重复性好的条带用于 SSR-PCR 的扩增。对 166 份样品材料共扩增出 129 条带,扩增的片段大小在 100 至 350bp 之间,其中 107 条带具有多态性,其多态性百分率为 82.9%。不同引物检测到的多态性带数差异较大,变幅为 5(Sca285)-12(Sca249),平均为 9 条。无瓣海桑的遗传多样性及分化系数由 POPGENE 软件分析,结果显示该研究中无瓣海桑 Nei's 遗传多样性指数  $h$  为 0.2857, Shannon's 信息指数  $I$  为 0.4580。物种水平的遗传多样性( $H_t$ )为 0.7526,种群内遗传多样性( $H_s$ )是 0.4717,无瓣海桑分化系数为 0.6852,即 68.52%的变异存在于种群间,21.48%的变异存在于种群内,说明遗传变异主要存在于种群间。这与 AMOVA 分析,种群间变异量为 78.86%基本保持一致。另外,其种群间的基因流( $N_m$ )0.2793,明显小于 1,相对较小,说明种群间的生长有较强的独立性,并由随机漂变所引起。

由 UPGMA 法获得的基于 Nei's 无加权计算无瓣海桑各种群的遗传距离聚类图。在所研究的 5 个无瓣海桑种群中,浮宫的两个无瓣海桑种群(林内与林缘)

之间的遗传距离最小（0.0960），海南东寨港与福建九龙江口两个无瓣海桑种群间的遗传距离最大（0.9760）。从总体趋势上看种群间遗传关系可以得出，种群的自然分布情况和遗传上关系大致相同，浮宫的两个无瓣海桑种群（FG（林内）和 FG（林缘））最先聚为一类，然后与东园无瓣海桑种群（DY）聚在一起，最后三者与玉枕无瓣海桑种群（YZ）聚在一起，而海南东寨港的无瓣海桑（DZG）在聚类图的根部。

本文研究表明，福建九龙江口外来种无瓣海桑种群的遗传多样性较大，这使得其具有较强的环境适应能力；而可能受到该地环境与气候的影响，种群间的基因交流较小，而该地无瓣海桑的扩散能力受到限制。

**关键词：**无瓣海桑；SSR；遗传结构



## Abstract

*Apetala* ( *Sonneratia apetala*) is one of the excellent tree species in the mangroves, naturally distributed in lower salinity and muddy tidal flats of India, Bangladesh, Sri Lanka and other countries.

*Apetala* with fast growth and high productivity is the first kind of mangrove plants, which was imported from foreign countries. *Apetala* was promoted widely in production, and acquired good ecological, economic and social benefits. In addition, *Sonneratia apetala* is widely applied to the control of invasive plant *Spartina alterniflora*'s spread. But after all, *Sonneratia apetala* is the alien species. So whether it can cause the biological invasion and ecological disaster brought to the coastal beach is still unclear. But a more consistent view is that *Sonneratia apetala* has a certain diffusion and competitive ability. So the introduction of the species should be cautious. Here through the study of genetic diversity and genetic structure of the *apetala* populations in the Jiulong River Estuary of Fujian as well as a source of Hainan Dongzhai Harbor, we explore the *Sonneratia apetala* populations diffusion model, which is expected to supply theory basis for its reasonable management mode and ecological risk assessment.

Through growth characteristic analysis of alien species *Sonneratia apetala*, we found that the *Sonneratia apetala* individuals gathered up and grew relatively consistently. Besides, individuals with small DBH occupied the majority. It showed that *Sonneratia apetala* populations here updated fast, which made them can adapt to the local natural environment better.

This study referred to the 14 pairs of *Sonneratia apetala* general SSR primers, which was designed by Chen Tao. And we selected 12 pairs ( Sca22, Sca23, Sca126, Sca224, Sca231, Sca234, Sca249, Sca266, Sca270, Sca284, Sca285, Sca286 ) that may expand clearly, duplicate well for SSR amplification. The 12 pairs of SSR primers was used to amplify all 166 materials to increase 129 bandings, which weight between 100-350bp, 107 bandings had the polymorphism, its polymorphism

percentage was 82.9%. Amplified bands number was from 5( Sca285 )-12( Sca249 ), and equally 9. The *Sonneratia apetala* heredity multiplicity and differentiation coefficient obtained by the POPGENE software, Nei's heredity multiple index  $h$  was 0.2857, Shannon's information  $I$  was 0.4580. Species level heredity multiple (  $H_t$  ) were 0.7526, occupied in the group to inherit multiple (  $H_s$  ) is 0.4717. The *Sonneratia apetala* (  $G_{st}$  ) value was 0.6852, namely 68.52% variations existed in occupy the group, 21.48% variation existed in occupying in the group which is consistent with 78.86% by AMOVA analysis. In addition, the gene flow among populations (  $N_m$  ) 0.2793 is significantly less than 1, which is relatively small. It shows that the growth among populations has strong independence, which was caused by random drift.

Obtained by the UPGMA occupies the group based on the Nei's non-weighting computation heredity from different *Sonneratia apetala* populations. Among the 5 studied *Sonneratia apetala* populations, the minimum genetic distance ( 0.0960 ) is between the two *apetala* populations in Fugong (inside the forest and forest edge), and the maximum genetic distance ( 0.9760 ) is between Hainan Dongzhai harbour and the Jiulong River Estuary of Fujian *apetala* populations. Look from overall genetic relationships, what can be derived is that populations of natural distribution is roughly consistent with genetic relationships. The two *apetala* populations in Fugong ( FG ( Linnaeus ) and FG ( forest ) ) first clustered together, and then with the *Sonneratia apetala* population in Dongyuan ( DY ). Together, the above three go together with Yuzhen *apetala* population ( YZ ), while Hainan Dongzhai Harbor *apetala* (DZG) is in the cluster map root.

This study shows that the genetic diversity of the alien species *Sonneratia apetala* population in Jiulong River Estuary of Fujian is large. So it has the strong ability to adapt to the environment; and probably due to the effects of the environment and its climate, gene exchange between populations is small, and the capability of *Sonneratia apetala* diffusion is limited.

**Key Words:** *Sonneratia apetala*; SSR; genetic structure

# 目录

<b>第一章 前言</b> .....	<b>1</b>
<b>一、研究背景</b> .....	<b>1</b>
1.1 无瓣海桑的形态学及生态学特性.....	1
1.2 无瓣海桑传播特性的研究.....	2
1.3 引种无瓣海桑的影响.....	3
1.4 无瓣海桑的引种概况.....	4
<b>二、植物种群遗传结构的研究进展</b> .....	<b>5</b>
2.1 种群遗传多样性的研究概述.....	6
2.2 植物种群空间遗传结构研究现状及研究方法.....	11
<b>三、DNA 分子标记技术及其在植物种群遗传结构和多样性研究中的应用</b> ....	<b>13</b>
3.1 DNA 分子标记发展简史.....	13
3.2 常用的分子标记方法及其特性的比较.....	14
3.3 SSR 分子标记技术.....	17
<b>四、本研究的目的、内容和意义</b> .....	<b>20</b>
<b>第二章 实验材料与方法</b> .....	<b>21</b>
<b>一、样品采集</b> .....	<b>21</b>
1.1 样地概况.....	21
1.2 样品的采集与保存.....	23
<b>二、实验仪器与试剂</b> .....	<b>24</b>
2.1 实验仪器.....	24
2.2 实验试剂与溶液.....	24

<b>三、实验方法 .....</b>	<b>26</b>
3.1 基因组 DNA 提取方法 .....	26
3.2 基因组 DNA 的电泳检测 .....	26
3.3 基因组 DNA 的纯度分析 .....	27
3.4 SSR 引物筛选及 SSR-PCR 反应体系的建立 .....	27
3.5 数据分析方法 .....	29
<b>第三章 结果及分析 .....</b>	<b>31</b>
<b>一、福建九龙江口无瓣海桑的种群生长特征分析 .....</b>	<b>31</b>
1.1 福建九龙江口无瓣海桑个体的空间分布 .....	31
1.2 浮宫无瓣海桑种群的胸径级结构 .....	32
1.3 浮宫无瓣海桑种群的株高级结构 .....	34
1.4 浮宫无瓣海桑个体株高与胸径的关系 .....	36
<b>二、无瓣海桑种群遗传多样性的 SSR 结果分析 .....</b>	<b>36</b>
2.1 基因组 DNA 的提取结果 .....	36
2.2 SSR 电泳结果及多态位点分析 .....	37
2.3 无瓣海桑种群内的遗传多样性分析 .....	39
2.4 种群间遗传分化与基因流分析 .....	40
2.5 种群间遗传距离及遗传关系分析 .....	41
<b>第四章 讨论与结论 .....</b>	<b>43</b>
<b>一、福建九龙江口无瓣海桑的种群生长特征分析 .....</b>	<b>43</b>
<b>二、引物的获得 .....</b>	<b>43</b>
<b>三、基因组 DNA 的提取 .....</b>	<b>43</b>
<b>四、福建九龙江口无瓣海桑种群的遗传多样性 .....</b>	<b>44</b>

五、福建九龙江口无瓣海桑种群的遗传分化 .....	46
六、无瓣海桑的引种与管理 .....	47
七、结论与展望 .....	47
参考文献 .....	49
致 谢.....	58

厦门大学博硕



# Catalogue

<b>Chapter 1 Introduction .....</b>	<b>1</b>
<b>Section 1 Research background.....</b>	<b>1</b>
1.1 Morphological and ecological characteristic of <i>Sonneratia apetala</i> ....	1
1.2 Study on transmission characteristics of <i>Sonneratia apetala</i> .....	2
1.3 The effects of <i>Sonneratia apetala</i> introduction .....	3
1.4 The outline of <i>Sonneratia apetala</i> introduction .....	4
<b>Section 2 The research progress of plant population genetic structure .....</b>	<b>5</b>
2.1 Brief outline of population genetic diversity .....	6
2.2 The current study situation and methods of plant population spatial genetic structure .....	11
<b>Section 3 DNA molecular markers and the application in plant genetic structure and genetic diversity .....</b>	<b>13</b>
3.1 The development of DNA molecular markers .....	13
3.2 Common molecular markers and comparison .....	14
3.3 Brief outline of SSR technology .....	17
<b>Section 4 This research goal content and significance .....</b>	<b>20</b>
<b>Chapter 2 Experimental materials and methods.....</b>	<b>21</b>
<b>Section 1 The collerction of experimental samples .....</b>	<b>21</b>
1.1 Basic information of sampling sites.....	21
1.2 Sample collection and preservation .....	23
<b>Section 2 Experimental apparatus and reagents.....</b>	<b>24</b>
2.1 Experimental apparatus.....	24
2.2 Reagents and solutions.....	24

<b>Section 3 Experimental methods .....</b>	<b>26</b>
3.1 Extraction methods of genomic DNA.....	26
3.2 Electrophoresis of genomic DNA .....	26
3.3 Purity analysis of genomic DNA .....	27
3.4 SSR primer screening and SSR-PCR reaction system.....	27
3.5 Data analysis methods.....	29
 <b>Chapter 3 Results and Analysis .....</b>	 <b>31</b>
 <b>Section 1 Growth characteristic analysis of <i>Sonneratia apetala</i> populations in the Jiulong River Estuary of Fujian.....</b>	 <b>31</b>
1.1 Spatial distribution for <i>Sonneratia apetala</i> individuals in the Jiulong River Estuary of Fujian .....	31
1.2 Diameter at breast height classes distribution of <i>Sonneratia apetala</i> populations in Fugong.....	32
1.3 Plant height classes distribution of <i>Sonneratia apetala</i> populations in Fugong .....	34
1.4 The relationship between plant height and diameter at breast height of <i>Sonneratia apetala</i> individuals in Fugong .....	36
 <b>Section 2 Heredity multiplicity results and analysis of <i>Sonneratia apetala</i> ...</b>	 <b>36</b>
2.1 The result of genome extraction.....	36
2.2 Analysis of SSR electrophoresis and polymorphism sites .....	37
2.3 Genetic Diversity within <i>Sonneratia apetala</i> populations.....	39
2.4 Genetic differentiation and flow between populations .....	40
2.5 Heredity distance and relations between populations .....	41
 <b>Chapter 4 Discussion and Conclusion .....</b>	 <b>43</b>



Degree papers are in the "[Xiamen University Electronic Theses and Dissertations Database](#)". Full texts are available in the following ways:

1. If your library is a CALIS member libraries, please log on <http://etd.calis.edu.cn/> and submit requests online, or consult the interlibrary loan department in your library.
2. For users of non-CALIS member libraries, please mail to [etd@xmu.edu.cn](mailto:etd@xmu.edu.cn) for delivery details.

廈門大學博碩