

四川大头茶在不同群落中的遗传分化及适合度成分

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摘要:研究了四川大头茶在 3 个群落中的遗传分化和适合度成分。12 个引物的 RAPD 分析表明, 仅有 10% 左右的遗传多样性存在于种群间。四川大头茶纯林结实率(31.86%)显著高于针阔混交林(22.53%)和常绿阔叶林(24.52%)。每果种子数和每果种子均重各种群间差异均显著, 每果种子数和每果种子均重分别为常绿阔叶林(29.50, 0.0154g), 四川大头茶纯林(28.39, 0.0172g), 针阔混交林(27.44, 0.0195g), 表明二者间存在着负耦联关系(trade off)。而以 $R = \sum l_x b_x$ 表示的适合度却以常绿阔叶林最大。

关键词:四川大头茶; 群落; 遗传分化; 适合度

Genetic variation and reproductive fitness components of *Gordonia acuminata* in different communities

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Abstract: *Gordonia acuminata* is one of the dominant tree species of evergreen broad leaved forest and it occurs in succession series communities in Mt. Jinyun. Three populations at different successional stages, i. e., mixed coniferous evergreen broadleaved forest (MF), *Gordonia acuminata* forest (GF) and evergreen broadleaved forest (EF), and close to each other at geographic distance were selected to evaluate their genetic diversity and reproductive fitness.

12 samples were collected along a long diagonal transect in each population. RAPD (Random amplified polymorphic DNA) was used to detect genetic variation within and among populations. A total of 40 random primers were initially screened using 2 representatives from each of the three populations. 12 primers that gave distinct, repeatable fragment patterns were selected for final analysis. All 36 *Gordonia acuminata* individuals sampled were genetically different from each other. Based on the band frequency in terms of populations, estimation of Nei's gene diversity (H) and Shannon's index of diversity (I) was performed by using Popgene 1.31. Nei's index was 0.1734 (MF), 0.1821 (GF) and 0.2091 (EF), respectively for the three populations, with a genetic diversity of 10.21% among populations, while Shannon's index of diversity was 0.2695 (MF), 0.2780 (GF) and 0.3168 (EF), respectively, with a genetic diversity of 12.86% among populations, which indicated an increase with maturity of community.

10 reproductive shoots from each of 10 randomly selected individuals in each population were tagged. The number of flower at the peak of flowering period and that of fruit which matured but before dehiscence for each reproductive shoot were

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counted. Fruit-set ratio of GF population (31.86%) was significantly higher than those of MF population (22.53%) and EF population (24.52%).

10 matured fruits from 10 randomly selected individuals in each population were collected. The number of seeds and average seed weight in each fruit for the three populations were 27.44 and 0.0195g, 28.39 and 0.0172g, 29.50 and 0.0154g, respectively, which were significantly different and possibly indicated a trade-off relationship between those two traits.

2~4 plots of 400m² were established in each of three communities. Based on demographic studies, all vegetative individuals (including seedlings and saplings) with a DBH of less than 4 cm were assigned to one category, while reproductive individuals with a DBH of more than 4cm were assigned to seven classes with an interval of 5cm. Five to ten reproductive individuals were sampled from each class, and about one hundred shoots bearing matured fruits from different positions of crown were collected from each individual. Based on survivorship rate (l_x) and fecundity (b_x), reproductive fitness ($\sum l_x b_x$) was estimated to be 74.625, 64.572 and 92.102 for MF, GF and EF populations, respectively.

This study indicated that the difference of reproductive fitness components among three populations was attributed to environmental differences since its gene flow among populations was high and that the genetic diversity of population tended to increase with successional stage. Taking the available reports into consideration, the relationship between genetic variation and succession was discussed.

Key words: *Gordonia acuminata*; community; genetic variation; reproductive fitness
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种群间的遗传分化取决于繁育系统、种群历史和选择压力等^[1]。Linhart 和 Grant 曾在理论上阐述了群落生境差异引起遗传分化的可能性^[2]。研究表明,群落生境的差异对种群间遗传分化的影响因物种不同而有很大差异^[3~6]。而另一方面,许多报道指出,遗传多样性常与种群大小成正相关^[7~9]。通常认为,隔离的小种群由于基因流的不畅通,会引起遗传漂变和近交衰退,从而导致遗传多样性的降低,以及适合度成分的下降。然而,一些研究也表明,除了遗传变异外,许多其他因素,如种群密度^[10]、个体大小^[11]、生境质量^[12]等对适合度成分也有显著的影响。本研究主要探讨在种群相对较大的情况下,群落生境的差异对四川大头茶(*Gordonia acuminata*)种群间遗传分化和适合度成分的影响。

1 材料和方法

1.1 研究地区自然概况

缙云山自然保护区位于北纬 29°50',东经 106°26',重庆北碚区内,属典型的中亚热带温暖湿润的气候类型,其自然环境条件已有报道^[13]。各群落的基本情况见表 1。群落组成分别为,针阔交林(以下简称 P1):润楠(*Machilus pingii*) + 马尾松(*Pinus massoniana*) + 四川大头茶(*G. acuminata*) + 杉木(*Cunninghamia lanceolata*);四川大头茶纯林乔木层则主要以四川大头茶占绝对优势(以下简称 P2);常绿阔叶林(以下简称 P3):四川大头茶(*G. acuminata*) + 小叶栲(*Castanopsis corlesii* var. *spimulose*) + 川灰木(*Symplocos sedhunensis*) + 薯豆(*Elaeocarpus japonicus*) + 小叶青冈(*Cyclobalanopsis myrsinaefolia*)。其中常绿阔叶林(P3)和四川大头茶纯林(P2)相距 200m,与针阔混交林(P1)相距 1300 m,四川大头茶纯林(P2)与针阔混交林(P1)相距 1200 m。

1.2 种群基本情况调查

于花期(10 月),在所选的 3 个群落内逐株统计每个种群的开花植株数。种群密度为相邻开花植株个体距离的均值;透光度用 NZ-76 型照度计群落内(10 个点的均值)外同时测定;土壤酸度在室内用国产 pH S-2C 型精密级酸度计测定;土壤厚度每个群落用钻土法测定 10 个点取均值;土壤含水量取土深 10cm 处土样测定。

1.3 RAPD

1.3.1 样品采集 各群落沿样线各采样 12 株,样品为生殖株的当年生嫩叶。

表 1 四川大头茶 3 个种群的基本特征

Table 1 Basic features of <i>Gordonia acuminata</i> populations			
项目 Item	P1	P2	P3
地点 Location	乌龙冈 Wulonggang	杉木园 Shanmuyuan	青龙寨 Qinglongzhai
种群大小(开花植株) Population size (No. of flowering plant)	654	118	192
海拔 Altitude(m)	720	850	860
个体间距 Interindividual spacing(m)	3.38	3.62	4.19
群落透光度 Community transparence	9.8	19.1	11.5
土壤 pH(soil pH)	4.80	4.66	4.70
土壤有机质 Organic content of soil	4.14	6.08	4.67
土壤厚度 Soil depth(cm)	66	116	72
土壤含水量 Water content of soil(%)	8.71	14.70	13.10
P1 针阔混交林 Mixed coniferous evergreen broad-leaved forest; P2 四川大头茶纯林 <i>Gordonia acuminata</i> forest; P3 常绿阔叶林 Evergreen broad-leaved forest; 下同 the same below			

1.3.2 DNA 的提取、扩增 采集的当年生嫩叶,按照陈亮等^[14]的方法提取 DNA(另加蛋白酶 K 消化)。DNA 样品经分光光度计和琼脂糖凝胶电泳检测纯度和浓度后,稀释到 $10\text{ng} \cdot \mu\text{l}^{-1}$ 用于 RAPD 分析。用随意选取的 2 个样品 DNA 模板筛选 Sangon 系列引物 40 个($S_1 \sim S_{40}$),经 3 次筛选,确定扩增效果良好,可重复的引物 12 个,用于全部样品的扩增。扩增在 PTC-100™ Programmable Thermal Controller 扩增仪上进行,反应体积为 $25\mu\text{l}$,含 20ng 模板 DNA, $0.2\mu\text{mol} \cdot \text{L}^{-1}$ 引物, $1 \times \text{Taq}$ 聚合酶反应缓冲液, $1.0\mu\text{Taq}$ DNA 聚合酶, $2\text{mmol} \cdot \text{L}^{-1}\text{MgCl}_2$,各 $0.2\text{mmol} \cdot \text{L}^{-1}\text{dNTPs}$ 。扩增程序为 94°C 预变性 180s, 94°C 变性 60s, 38°C 退火 90s, 72°C 延伸 120s,共 45 个循环,最后 72°C 延伸 300s。PCR 产物用 1.5% 的琼脂糖凝胶在 $1 \times \text{TAE}$ 缓冲液电泳,EB 染色后在 BIO-RAD System 观察并保存结果。

1.3.3 遗传参数 运用 POPGENE 1.31 软件^[15]计算 Nei 指数和 Shannon 指数,再分别求各种群平均值和遗传分化系数 G_{ST} 。

1.4 适合度成分

1.4.1 结实率、每果种子数和种子均重 于盛花期(10 月份),在各群落内随机选取 10 株个体,借助于楼梯随机选取 10 个花枝(每花枝有 4~12 朵花)挂牌,翌年 7 月份果实成熟但未开裂之前,统计各株的结实率。果实成熟期,在各群落内随机选取 10 株个体,每个体随机选取 10 个果实,统计每果种子数,风干后用万分之一天平称得每果种子的平均重量。

1.4.2 适合度或净增殖率 在每个群落内各设置 2~4 个 400m^2 的样地,将胸径在 4cm(四川大头茶开始生殖的起始胸径)以下的个体作为一个径级,胸径在 4cm 以上则以 5cm 为一径级,统计各径级的个体数,并分别对 4cm 以上的各径级选择平均标准木 5 株以上(高径级不足 5 株时,以实存株数统计),记录全株的结实枝条数,对每株平均标准木,分别在树冠的东、南、西、北、上、下 6 个部位各采集结实平均标准枝 15~20 枝,计算每个结实平均标准枝上的结实数,再乘上由 1.3.1 中所得的各种群平均每果种子数,即得生育力 b_x ,并用 $R = \sum l_x b_x$ 计算出各种群的适合度。

1.4.3 数据分析 用 SPSS 统计软件对结实率、每果种子数和种子均重进行统计分析。

2 结果与分析

2.1 遗传分化

Nei 指数和 Shannon 指数计算的四川大头茶 3 个种群群内、种群间的遗传多样性,以及各自在总变异中所占的比例,结果见表 2。从表 2 中可以看出,Nei 指数和 Shannon 指数计算的结果一致,即均为:常绿阔叶林>四川大头茶纯林>针阔混交林。两种指数计算的种群间遗传分化分别为 10.21% 和 12.86%。Shannon 指数估算的遗传多样性和遗传分化均高于 Nei 指数,这与两种统计方法各自的统计原理有关。Shannon 指数只根据扩增产物的有无来确定某一 DNA 片段的表型频率,而 Nei 指数则假定某一特定位点上有两个等位基因,根据各自的基因频率来计算遗传多样性。从遗传学角度出发,Nei 指数的方法似乎更具有生物意义。而 Shannon 指数只是一种表型参数,但它避免了对 RAPD 显隐性位点的讨论。

2.2 适合度成分

2.2.1 结实率、每果种子数和种子均重 四川大头茶各种群的结实率、每果种子数和种子均重见表 3。从表中可以看出,四川大头茶纯林的结实率最高,极显著高于针阔混交林和常绿阔叶林,而针阔混交林和常绿阔叶林的结实率差异不显著。四川大头茶各种群的每果种子数两两之间差异极显著,每果种子数依次为:常绿阔叶林>四川大头茶纯林>针阔混交林。四川大头茶各种群的每果种子均重也两两之间差异极显著,种子均重依次为:针阔混交林>四川大头茶纯林>常绿阔叶林,和每果种子数正好相反。

2.2.2 适合度或净增殖率 适合度的两个基本成分是生殖和生存,四川大头茶各种群适合度 $R = \sum l_x b_x$ 见表 4,其大小顺序为:常绿阔叶林>针阔混交林>四川大头茶纯林,与上述四川大头茶各种群结实率、每果种子数和种子均重大小均不一致。

3 讨论

(1)群落生境与遗传分化 种群间的遗传分化取决于繁育系统、种群历史和选择压力等^[1]。群落生境的差异导致遗传分化是可能的。首先,群落类型的差异意味着各群落在植物组成和稳定性方面(即演替)存在差异,而且不同的群落类型会在一系列生境因子上存在差异,如土壤、光照等,这些因子可能单独或综合起来造成不同的选择压力,从而导致同一物种在不同群落类型

表 2 四川大头茶的遗传多样性

Table 2 Genetic diversity of <i>Gordonia acuminata</i> populations						
	P1	P2	P3	H_S	H_T	G_{ST}
Nei index	0.1734	0.1821	0.2091	0.1882	0.2096	0.1021
Shannon index	0.2695	0.2780	0.3168	0.2881	0.3306	0.1286

表 3 四川大头茶各种群的结实率、每果种子数和种子均重

Table 3 Fruit set, number of seeds per fruit and mean seed weight of <i>Gordonia acuminata</i> populations			
种群 Population	结实率(%) ($n=10$) Fruit set	每果种子数 ($n=100$) No. seeds/ fruit	每果种子均重 (g)($n=100$) Mean seed weight/fruit
P1	22.53 ± 3.65^a	27.44 ± 1.31^a	0.0195 ± 0.0009^a
P2	31.86 ± 3.49^b	28.39 ± 1.30^b	0.0172 ± 0.0008^b
P3	24.52 ± 3.92^a	29.50 ± 1.28^c	0.0154 ± 0.0008^c

数据为平均值 $\pm$ SE(n 为样本数),同一列字母不同者表示差异显著($p<0.01$) Valules are mean $\pm$ SE (n is sample size), data followed by different letters within a column differ significantly($p<0.01$)

中种群遗传组成上的空间分化;其次,种群的遗传多样性是在长期演化过程中积累起来的,同一物种在不同群落类型中(即可能意味着演替阶段的不同)种群的发展史是不一样的,早期入侵者在刚刚侵入某一领地时,所面临的是一种生境条件,随着演替的进行,光照、湿度、竞争、植食等一系列因子会发生变化,其后代面临的是另一种环境压力,因此在不同的演替阶段会留下不同的基因型,从而导致各演替阶段种群遗传组成上的差异。但从现有的研究结果来看,群落生境导致的遗传分化多在 10%以下^[3,4,6],只有锥栗(*Castanopsis chinensis*)达到了 24.6%^[5]。本研究,四川大头茶 3 个种群相对较大(开花植株>100),种群间的遗传分化只有 10%左右,表明种群间强烈的基因流在一定程度上抵消了生境的选择压力。

表 4 四川大头茶各种群的生殖力表
Table 4 The fecundity table of *Gordonia acuminata* populations

径级(cm) Size class	标准化存活数 Standard survivor No.			存活率 Survivorship rate(l_x)			生育力 Fecundity(b_x)			$l_x b_x$		
	P1	P2	P3	P1	P2	P3	P1	P2	P3	P1	P2	P3
0~4	1000	1000	1000	1.0000	1.0000	1.0000	0	0	0	0	0	0
4~9	2.226	2.906	2.205	0.0022	0.0029	0.0022	2137	2346	2315	4.756	6.818	5.104
9~14	2.910	1.300	2.123	0.0029	0.0013	0.0021	5428	6203	6783	15.798	7.415	14.401
14~19	1.712	1.530	0.572	0.0017	0.0015	0.0006	9053	11022	10957	15.499	16.829	6.263
19~24	0.571	0.382	0.817	0.0006	0.0004	0.0008	18016	21354	21172	10.281	8.166	17.289
24~29	0.514	0.535	0.490	0.0005	0.0005	0.0005	27905	28037	28847	14.322	15.010	14.134
29~33	0.171	0.076	0.327	0.0002	0.0001	0.0003	34569	36236	35528	5.918	2.771	11.605
>33	0.228	0.153	0.572	0.0002	0.0002	0.0006	35222	45187	40773	8.040	6.912	23.306
Σ										74.625	64.572	92.102

(2)适合度成分 适合度的估测是十分复杂的,尤其是多年生多次生殖植物,迄今用于估计植物适合度成分的有种子萌发率、生长速率、繁殖的时间、花或花序的数量、花粉萌发率和花粉管的生长、结实率、果实数量和重量、种子数量和重量、存活等^[1]。一般认为隔离的小种群由于基因游的不畅通,常会导致遗传漂变,近交衰退,进而导致适合度的下降。然而,近年来的一些研究表明,适合度成分还受其他许多因素的影响,如小种群对传粉者吸引力小,导致植株授粉不足,种子产量下降^[10]。种群密度也影响着授粉行为和繁殖成功^[16,17]。还有生境质量等^[12]。

本项研究中,四川大头茶各种群间结实率、每果种子产量和每果种子均重都有显著差异,这种差异不大可能是由近交衰退引起的,同时所研究的 3 个种群也都较大(开花植株>100)。四川大头茶纯林结实率显著高于其他两个种群,很可能和其较优越的土壤、光照条件有关(表 1),而种群密度通常只有低于某一阈值时才会影响结实率^[18]。一些研究也发现结实率与可利用性资源有关^[11,19,20]。四川大头茶两两种群间每果种子数、每果种子均重差异显著,而且每果种子数和每果种子均重变化趋势正好相反,表明四川大头茶每果种子数量和每果种子单粒重之间的负耦联关系(trade off),这是因为种子之间对资源存在竞争,种子数量的减少使每粒种子的可利用资源增加,结果产生较大的种子^[21,22]。尽管用于表示植物适合度成分的指标很多,但通常认为只用生长、存活和生育力对适合度有直接的影响^[23]。因此, $R(= \sum l_x b_x)$ 作为一个综合指标,能够较真实地反映各个种群的适合度,在所研究的 3 个四川大头茶种群中,常绿阔叶林四川大头茶种群适合度最大,野外调查也发现四川大头茶在常绿阔叶林分布最广,长势最好,表明了常绿阔叶林可能最适合于四川大头茶生长。

4 结语

常绿阔叶林是我国最大的森林生态系统,壳斗科、樟科、山茶科是其主要的建群种和优势种。目前有关其遗传多样性在各演替阶段或群落类型中的分布可总结如下:锥栗(*Castanopsis chinensis*),针叶林 0.197,针阔混交林 0.297,常绿阔叶林 0.311^[5];荷木(*Schima superba*),针叶林 0.353,针阔混交林 0.336,常绿阔叶林 0.304^[4];黄果厚壳桂(*Cryptocarya concinna*),针阔混交林 0.3459,常绿阔叶林 0.3564^[6];四川大头茶,针阔混交林 0.2695,四川大头茶 0.2780,常绿阔叶林 0.3168(Shannon 指数)(本研究);灌木层优势种川鄂连蕊茶(*Camellia rosthorniana*),毛竹林 0.3148,针阔混交林 0.3085,常绿阔叶林 0.3345(Shannon 指数)^[3]。虽然上述研究都是在局部范围的少数种群中进行的,其结果似乎表明,亚热带常绿阔叶林优势种随演替等级或群落稳定性的提高,种群遗传多样性有增加的趋势。这可能与种群在进展演替过程中,仍是该种群的内源性发展,很少有外种群的基因流进入有关。四川大头茶以 $R = \sum l_x b_x$ 表示的适合度也以常绿阔叶林最高。因此可以预期,当常绿阔叶林被破坏后,群落发生逆行演替,种群的遗传多样性和适合度下降,种群适应未来环境变化的能力减弱,如果是这样,上述研究就为亚热带常绿阔叶林的保护提供了初步的理论支持。

References:

[1] Booy G, Hendricks R J, Smulders M J M. Genetic diversity and the survival of populations. *Plant Biology*, 2000, 2: 379~395.

- [2] Linhar Y B, Grant M C. Evolutionary significance of local genetic differentiation in plants. *Annual Review of Ecology and Systematics*, 1996, **27**:237~277.
- [3] Cao G X, Zhong Z C, Xie D T, et al. RAPD analysis of *Camellia rosthorniana* populations in different communities in Jinyun Mountain. *Acta Ecologica Sinica*, 2003, **23**(8):391~397.
- [4] Wang Z F, Wang B S, Li M G, et al. Molecular ecology of lower subtropical species, *Schima superba* and *Castanopsis chinensis* in three succession series communities. *Acta Botanica Sinica*, 2000, **42**(10):1082~1088.
- [5] Wang Z F, Wang B S, Li M G, et al. Genetic differentiation of *Castanopsis chinensis* population in three communities in Dinghu Mountain, China. *Acta Ecologica Sinica*, 2001, **21**(8):1208~1213.
- [6] Zhang J L, Wang Z F, Wang B S, et al. Aflp analysis in *Cryptocarya concinna*, a dominant population in lower subtropical evergreen broad-leaved forests at Heishiding Natural Reserve. *Acta Ecologica Sinica*, 2001, **21**(3):391~397.
- [7] Fischer M, Matthies D. RAPD variation in relation to population size and plant performance in the rare *Gentianella germanica*. *American Journal of Botany*, 1998, **85**(6):811~819.
- [8] Fischer M, Husi R, Prati D, et al. RAPD variation among and within small and large populations of the rare clonal plant *Ranunculus reptans* (Ranunculaceae). *American Journal of Botany*, 2000, **87**(8):1128~1137.
- [9] Schmidt K, Jensen K. Genetic structure and AFLP variation of remnant populations in the rare plant *Pedicularis palustris* (Scrophulariaceae) and its relation to population size and reproductive components. *American Journal of Botany*, 2000, **87**(5):678~689.
- [10] Agren J. Population size, pollination limitation, and seed set in the self-incompatible herb. *Lythrum salicaria*. *Ecology*, 1996, **77**(6):1779~1790.
- [11] Goubitz S, Werger M J A, Shmida A, et al. Cone abortion in *Pinus halepensis*: the role of pollen quantity, tree size and cone location. *Oikos*, 2002, **97**(1):125~133.
- [12] Hogbin P M, Ayre D J, Whelan R J. Genetic variation and reproductive success of road verge populations of the rare shrub *Grevillea barklyana* (Proteaceae). *Heredity*, 1998, **80**:180~186.
- [13] Zhong Z C. Study on the vegetation geography of major forests in Sichun. In: Zhong Z C ed. *ecological study on evergreen broad-leaved forest*. Chongqing: Southwest Normal University Press, 1988. 142~156.
- [14] Chen L, Chen D M, Gao Q K, et al. Isolation and appraisal of genomic DNA from tea plant (*Camellia sinensis*(L)O. kuntze). *J. Tea. Sci.*, 1997, **17**(2):177~181.
- [15] Yeh F C, Yang R C, Boyle T B. POPGENE1. 31. 1999. ftp://ftp. microsoft.com /Softlib /MSLFIES/H PG L. EXE.
- [16] Roll J, Mitchell R J, Cabin R J. Reproductive success increases with local density of conspecifics in a desert mustard (*Lesquerella fendler*). *Conservation Biology*, 1997, **5**:738~746.
- [17] Van Treuren R, Bijlsma R, Ouborg N J. Relationships between plant density, outcrossing rates and seed set in natural and experimental populations of *Scabiosa columbaria*. *Journal of Evolutionary Biology*, 1994, **7**:287~302.
- [18] Knight T M. Floral density, pollen limitation, and reproductive success in *Trillium grandiflorum*. *Oecologia*, 2003, **137**:557~563.
- [19] Kato E, Hiura T. Fruit set in *Styrax orassia* (Styracaceae); the effect of light availability, display size, and local floral density. *American Journal of Botany*, 1999, **86**(4):495~501.
- [20] Niesenbaum R A. Light or pollen-seasonal limitations on female reproductive success in the understory shrub *Lindero benzoin*. *Journal of Ecology*, 1993, **81**:315~323.
- [21] Eriksson O. Seed size variation and its effect on germination and seedling performance in the clonal herb *Convallaria majalis*. *Acta Oecologica*, 1999, **20**(1):61~66.
- [22] Kery M, Matthies D, Spillmann H H. Reduced fecundity and offspring performance in small populations of the declining grassland plants *Primula veris* and *Gentiana lutea*. *Journal of Ecology*, 2000, **88**(1):17~30.
- [23] David P. Heterozygosity-fitness correlations; new perspectives on old problems. *Heredity*, 1998, **80**:531~537.

参考文献:

- [3] 操国兴, 钟章成, 谢德体, 等. 缙云山川鄂连蕊茶在不同群落类型中的 RAPD 分析. *生态学报*, 2003, **23**(8):1583~1589.
- [4] 王峥峰, 王伯荪, 李鸣光, 等. 南亚热带森林优势种群荷木和锥栗在演替系列中的分子生态学研究. *植物学报*, 2000, **42**(10):1082~1088.
- [5] 王峥峰, 王伯荪, 李鸣光, 等. 锥栗种群在鼎湖山三个群落中的遗传分化研究. *生态学报*, 2001, **21**(8):1208~1213.
- [6] 张军丽, 王峥峰, 王伯荪, 等. 黑石顶南亚热带常绿阔叶林优势种群黄果厚壳桂的 AFLP 分析. *生态学报*, 2001, **21**(3):391~397.
- [13] 钟章成. 四川主要森林植被地理学. 见: 钟章成主编. 常绿阔叶林生态学研究. 重庆: 西南师范大学出版社, 1988. 142~156
- [14] 陈亮, 陈大明, 高其康, 等. 茶树基因组 DNA 的提纯与鉴定. *茶叶科学*, 1997, **17**(2):177~181.