学校编码：10384

学号：21720091152222

厦门大学

硕士学位论文

模拟酸雨对不同土壤钙条件下四种木本植物生理及钙相关基因表达的影响

Effects of simulated acid rain on physiological indexes and Ca-related genes expression of four tree species under different calcium levels in soil

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专业名称：植物学

答辩日期：2012年6月

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摘 要

酸雨在我国尤其在西南部地区已成为十分严重的环境问题。本研究中，我们选取了四种在我国南方受酸雨危害严重地区广泛分布的木本植物作为实验材料，分别为枫香(Liquidambar formosana Hance)，木荷(Schima superba Gardn et Champ.)，马尾松(Pinus massoniana Lamb.)及红豆杉(Taxus wallichiana var. mairei)。研究土壤中不同的钙水平对四种木本植物在应对酸雨胁迫中的各种响应，包括酸雨对植物叶片的伤害，叶绿素含量，叶片净光合速率，叶片钙含量和钙相关基因的相对表达水平等。采用实时荧光定量PCR技术(quantitative real-time PCR，qRT-PCR)检测4种木本植物8个钙相关基因的表达水平在不同钙水平和酸雨影响下表达变化，这些基因是钙调素（calmodulin 1，CaM1）基因，钙调神经磷酸酶B亚基蛋白（calcineurin B-like calcium sensor protein 1，CBL1）基因，钙依赖蛋白激酶（calcium-dependent protein kinase 1，CDPK1）基因，谷氨酸脱氢酶（glutamate dehydrogenase 2，GDH2）基因，呼吸爆发氧化酶（respiratory burst oxidase homolog A，RbohA）基因，接触诱导蛋白（touch 3，TCH3)基因，钙网蛋白（calreticulin 3，CRT3）基因和钙联蛋白（calnexin 1，CNX1)基因，发现四个物种的钙相关基因的表达水平均随土壤钙浓度的增加而提高。其次，在酸雨胁迫下，两个抗性物种（木荷和红豆杉）的基因表达水平比两个敏感物种（枫香和马尾松）的基因表达更稳定，变化的幅度更小，说明与敏感物种枫香和马尾松相比，木荷和红豆杉有更强的抗酸雨能力。由此揭示在酸雨胁迫下，不同酸雨耐受性的物种在不同土壤钙浓度下的分子响应的差异。此外，我们还测定了酸雨处理下四个物种的一些相关的生理指标的变化，包括叶片钙含量，叶绿素含量及叶片净光合作用速率。结果表明，与钙相关基因的表达变化一致，与抗性物种相比，酸雨胁迫对两个敏感物种（枫香和马尾松）的影响较大，伤害更严重。同时，我们也探讨了不同钙浓度对于不同物种缓解酸雨胁迫的作用和影响。酸雨条件下，钙相关基因整体上都表现表达水平随着钙浓度的增加而相对减小的趋势，说明在基因水平上钙的增加能缓解植物对于酸雨胁迫的响应强度。除此之外，补钙对敏感物种的各项生理指标和生长均有较明显的改善和促进作用，而对两个抗性物种没有显著影响。

上述结论说明，在酸雨胁迫下，不同木本植物钙相关基因及相关生理响应不仅取决于植物对酸雨耐受能力的差异，并且不同的土壤钙浓度对其也有十分重要的影响。

关键词：模拟酸雨；钙；木本植物；叶片伤害；生理指标；钙相关基因表达

Abstract

Acid rain has been a serious environmental problem in southern China. Four selected tree species, Liquidambar formosana Hance, Schima superba Gardn et Champ., Pinus massoniana Lamb. and Taxus wallichiana var. mairei, are widely distributed in southern China, where acid deposition is becoming more and more serious. In this study, we investigated the differential responses to simulated acid rain (SiAR) at different calcium (Ca) levels for four tree species. In order to understand the differences in expression levels of Ca-related genes between acid rain-tolerant and acid rain-sensitive species, quantitative real-time PCR (qRT-PCR) was used to determine the effects of Ca on the expression patterns of eight Ca-related genes in four selected tree species including calmodulin 1 (CaM1), calcineurin B-like calcium sensor protein 1 (CBL1), calcium-dependent protein kinase 1(CDPK1), glutamate dehydrogenase 2 (GDH2), respiratory burst oxidase homolog A (RbohA), touch 3 (TCH3), calreticulin 3 (CRT3) and calnexin 1(CNX1). We found the mRNAs of eight Ca-related genes were up-regulated significantly by increasing Ca concentrations in all four selected species. Furthermore, it was shown that the changes of genes expression in tolerant species, S. superba and T. wallichiana var. mairei, were smaller than sensitive species, L. formosana and P. massoniana under SiAR treatment. This result indicated that compared with L. formosana and P. massoniana,the SiAR-tolerance of S. superba and T. wallichiana var. mairei were higher. The changes of Ca-related genes indicated that different tree species responded to SiAR was different at various Ca levels in molecular level. Moreover, we investigated the effects of three Ca levels on leaf necrosis,total chlorophyll content, net photosynthetic rate (Pn) and Ca content in leaves of four species under SiAR. It was indicated that SiAR had more negative effects on total chlorophyll content, Pn and Ca content in leaves of the two sensitive species, which were consistent with the expression changes of eight Ca-

related genes. However, the growth of seedlings could be ameliorated in two sensitive species by adding Ca. In contrast, the physiological processes of the two tolerant species were much less affected by both SiAR and Ca treatments. This conclusion implies that the changes of Ca-related genes expression and physiological responses caused by SiAR may be attributed not only to the sensitivity of tree species to acid deposition, but also to their different Ca requirement.

Keywords: Simulated acid rain; calcium; tree species; physiological indexes; Ca-related genes expression

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