

# 分子生物学科・環境科学研究センター・ SUPER FORUM 共催セミナーのご案内

3月26日(木) 16:20～17:50 理学部3号館二階11番教室

## Development of SSR markers for effective applications in *Phalaenopsis* orchid variety identification

時間: 16:20-17:05 (講演30分、質疑応答15分)  
主講: Prof. Wen-Luan Wu (台湾 国立成功大学)

## Orchid genomics and functional genomics research

時間: 17:05-17:50 (講演30分、質疑応答15分)  
主講: Prof. Hong-Hwa Chen (台湾 国立成功大学)

問い合わせ先:  
高木 優 (環境科学研究センター)



# Development of SSR markers for effective applications in *Phalaenopsis* orchid variety identification



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### Research Focus

Molecular marker and gene expression in  
*Phalaenopsis*

### Abstract

Simple sequence repeats (SSRs) or microsatellites, which are short (1-6 bp) tandemly repeated DNA sequences. SSRs are preferred and powerful markers of choice for variety identification and genetic diversity analysis. *Phalaenopsis* is one of the most interesting genera of orchids due to the members are often used as parents to produce hybrids. The development of very efficient SSR markers would be useful for orchid variety identification and served as a complementary tool in the system of DUS (distinctness, uniformity and stability) testing to enhance the plant breeders' rights (PBRs) protection. To reach the purposes described above, developing highly informative SSR markers for *Phalaenopsis* orchids is essential. In this study, over 6000 SSR markers have been characterized from cDNA, small-insert, large-insert genomic libraries and genome-wide sequences of *P. aphrodite* subsp. *formosana* and *P. equestris*. Among 800 primer pairs tested for amplification and polymorphism in 12 *Phalaenopsis* wild species. A set of SSR markers with higher polymorphism were labeled with fluorescent dye and exploited for genotyping of commercial valuable *Phalaenopsis* varieties and individuals of the intraspecific population. The results revealed that these markers are highly effective to distinguish varieties and could be uniquely assigned an allele code for each variety, even which derived from the same parental cross and suggested that the SSR markers developed in this study would be of immense use for various genotyping applications in orchids.

# Orchid genomics and functional genomics research



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## Research Focus

Orchid Biology and Biotechnology, Genomics,  
Cell Biology, Molecular Biology, Virology

## Abstract

Containing more than 25,000 species, the Orchidaceae family, classified in class Liliopsida, order Asparagales, is one of the largest angiosperm families. Orchidaceae is one of the most diverse plant families and renowned for its spectacular flowers with highly evolved petals, labellum and fused androecium and gynoecium (gynostemium) to attract diverse pollinators for effective pollination. Because of these, and other specific reproductive and ecological adaptations as well as obligate orchid-mycorrhizal interactions, orchids are important species for evolutionary biology. The genus *Phalaenopsis*, a beautiful and popular orchid, comprises approximately 66 species. The nuclear DNA contents from 18 *Phalaenopsis* species have been estimated by using flow cytometry. The 1C values ranged from 1.37 pg for *P. sanderiana* to 8.3 pg for *P. parishii*. Analysis of karyotypes of nine *Phalaenopsis* species revealed that *P. aphrodite*, *P. stuartiana*, *P. equestris*, *P. cornu-cervi* and *P. lueddemanniana* are with small and uniform chromosomes (1–2.5 mm). Both *P. equestris* and *P. aphrodite* subsp. *formosana* have a relatively small genome size with a 2C-value of 2.95 pg and 2.81 pg, respectively. OrchidBase 2.0 collects the transcriptomics data of ten species distributed in the five subfamilies. The whole genome sequence of the tropical epiphytic crassulacean acid metabolism (CAM) orchid, *P. equestris*, a frequently used parent species for orchid breeding has been completed. The assembled genome contains 29,431 protein-coding genes. Due to self-incompatibility, the *Phalaenopsis* genome is highly heterozygous. Heterozygous regions of the genome seem to be enriched for genes for apoptosis, programmed cell death and defense response, which are likely candidates for self-incompatibility loci. Genes involved in CAM photosynthesis were found to evolve by gene duplication and positive selection. Finally, we find expanded and diversified MADS-box C/D-class, B-class AP3, and AGL6 class genes, which may contribute to the highly specialized morphology of orchid flowers. To study the function of orchid genes, it is not applicable with genetic transformation in *Phalaenopsis* due to their long life cycle for 3 years. Instead, virus-induced gene silencing (VIGS) strategy has been adopted by using *Cymbidium* mosaic virus as the infectious vector. So far, we have used VIGS to unravel the functions of genes involved in floral morphogenesis, floral scent and floral pigmentation patterning.