Preface

Controlled sexual hybridization has been the most important approach for *Populus* breeding. Over the past decades, numerous hybrids have been generated, many of which have demonstrated desired features for forestry and/or ecology. However, there are several profound issues that are poorly understood. Here are some of the most frequently asked questions. What is the general mode of seed development in parent plants? What can we learn from an embryological perspective of successful cross combinations? And, most importantly, what are the barriers to other, unsuccessful combinations? Then, how to overcome these barriers? We have undertaken a research program on the hybrido-embryology of *Populus* since the early 1980s. Representative species from all five sections of the genus were included and trials covered as many as possible intersetional crosses. Our results showed that in intersectional crosses embryo abortion is a common barrier, although problems could happen at any stages of the hybridization process. Using an *in vitro* system for *in ovulo* embryo culture, we have successfully rescued hybrid embryos from abortion. Examples of hybrid plantlets obtained by this technique include *Populus simonii* × *P. pyramidalis*, *P. simonii* × *P. lasiocarpa*, and *P. simonii* × *P. euphratica*. We also worked on an intergeneric cross for the Salicaceae, *P. simonii* × *Salix matsudana*, and found that fertilization did take place in a few ovules, and embryo abortion was responsible for the small numbers of hybrid seeds.

In the recent years, we have extended our efforts into the embryology of distant hybridizations in conifers. *Cunninghamia lanceolata* is an important forestation species widely distributed in central and south China. In late 1950s, Professor Yieh Peichong reported successful distant crossings in both *Cunninghamia lanceolata* × *Cryptomeria fortunei* and *Cunninghamia lanceolata* × *Platycladus orientalis*, being intergeneric and interfamilial, respectively. Yet, this has raised a shadow of doubt among scientists. We thus decided to repeat the experiments. Our soft X-ray radiographic study has confirmed that the hybrid seeds from *Cunninghamia lanceolata* × *Cryptomeria fortunei* contained a few fully-developed embryos which could germinate into seedlings. On the contrary, investigation of *Cunninghamia lanceolata* × *Platycladus orientalis* revealed a post-fertilization failure.

In vitro fertilization has long been a dream for plant biologists. In 1991, Kranz, Bautor and Lörd reported the first successful case of *in vitro* fertilization with isolated, single gametes of maize. It was very encouraging for us to follow this "cell - cell" technique for overcoming fertilization failure in distant hybridization. We started out with the isolation of male and female gametes from both *Populus* and conifer species. We were very excited to obtain viable sperms and some female components (embryo sacs, egg apparates, or even single eggs). Fusion experiments were performed in the presence of polyethylene glycol, which revealed that the male and female gametes adhered to each other but had not fused yet. Further pursuit in this direction will definitely broaden the field of hybrido-embryology.

This book represents a pictorial summary of our research achieved over the past twenty

years. We have taken numerous photographs for publication. To write this book, we selected 700 images from which 80 plates were designed, including 34 in full-color. Some of the pictures were never published elsewhere. The information presented the sexual processes of controlled sexual hybridization, the secrets of hybridization barriers, the *in ovulo* embryo culture for overcoming hybrid embryo abortion, and the manipulation for isolating male and female gametes. This is an atlas of basic sexual reproductive biology of woody plants and experimental biotechnology for tree breeding. We hope this work will be helpful to both plant embryologists and tree breeders. It may also serve as a reference book for advanced students of plant biology and forestry.

Science is moving on at an unprecedented pace. Genome sequencing for *Populus trichocarpa* has recently been completed, and the *Pinus taeda* genome project is well underway. These two species, important economically and ecologically, are excellent models, one for angiospermatous trees and another for gymnospermatous trees. With these genomic resources, we are offered novel opportunities to study the molecular and genetic mechanisms of interspecific interactions during sexual hybridization. These opportunities represent a platform to display embryological and cellular events documented in this book in the context of molecular and genetic control and regulation. Discoveries in this direction will greatly improve our understanding of the recognition processes in compatible cross combinations. Fundamental progress is also anticipated in deciphering the machinery for the detected barriers in problematic combinations. These areas will be our interests for the many years to come.

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