<u>*Title of Exchange*</u>: Evaluating the Genetic and Genomic Basis for Different Levels of Blight Resistance among Native Chinese Chestnut Trees (*Castanea mollissima, C. henryi, and C. seguinii*)

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Country visited and dates of visit: China. September 5 - 18, 2011

Summary: Our group deems the cooperative exchange trip a success. Our main objective – that of initiating research collaborations and collecting samples from three chestnut tree species (*Castanea mollissima, C. henryi, and C. seguinii*) to use in collaborative research was accomplished. Our long-term goal is to determine which genes in these Chinese species are involved in chestnut blight resistance and apply what we learn to the restoration of the American chestnut (*Castanea dentata*).

Description: After initial confusion about our objectives and the schedule, Mr. Lin Luogeng (Associate Advisor) was very gracious and helpful about changing the schedule to meet our research needs. We understand this was difficult and we appreciate his efforts. The exchange began with a visit to the Chinese Ministry of Agriculture where we meet with Dr. Shidong Li and Dr. Li Shifang and several other research scientists and watched an overview presentation on key agricultural research in China. We then met with our primary collaborators, Dr. Xiangning JIANG (Professor, Associate Dean) and Dr.

Wei HE (Professor) at the Beijing Forestry University. We went on tours of the facilities and discussed their respective research in plant physiology and plant pathology.

We took a flight on September 7th to Xi'an to begin our sample collection part of the trip. Upon arrival we meet Mr. Wang who coordinated the trip through Zhengan County. During this time we met many gracious and helpful officials. Mr. Chen Yu-Zhao (Director of Forest Regeneration in Ankang) was extremely helpful in finding the wild *Castanea mollissima, C. henryi, and C. seguinii* trees that we needed to sample. Our trip came during a rainy season and there were many mud and rockslides that blocked roads. We appreciated the skill of the driver and our guides so that we were able to collect all the samples safely. While collecting the samples, we were able to take GPS readings at each tree so that we could return to them for future sampling during our collaborative research.

On September 10th we flew back to Beijing with our samples. We spent much of the rest of the time in Dr. Xiangning JIANG's lab extracting DNA, RNA, and metabolites from the samples. Dr. Xiangning JIANG had graciously purchased all the supplies we need ahead of our visit and we have since reimbursed him for these. The extractions were a success and we made cDNA from the RNA. All these samples were critical for the future collaborative research. While in the lab, Sara Fitzsimmons, Katie D'Amico, and Bill Powell also helped review Dr. JIANG's students papers before they were submitted for publication. We also discussed opportunities for U.S. students and postdoctoral fellows to do six to twelve month research internships at Beijing Forestry University. Those opportunities are now being advertised at SUNY-ESF. During this week, Dr. Powell gave a seminar describing the chestnut research going on in his lab at SUNY-ESF in the U.S.

The official part of the visit ended on September 18th. The members of the team either arrived a few days early or left a few days later so that we could visit many of China's historical sites. Even during the official time, Mr. Lin treated us to the diverse cuisine of China as well as visiting some of the most interesting sites such as the Great Wall and Summer Palace, among others. All in all, the team enjoyed the visit.

Specific objectives accomplished:

- 1. Establish collaborative opportunities with Dr. Xiangning JIANG (Professor, Associate Dean) and
- Dr. Wei HE (Professor) at the Beijing Forestry University.
- Identified six each of "wild-type" Castanea mollissima, C. henryi, and C. seguinii trees, took height estimates, DBH, and health ratings. GPS coordinates were taken so that we could return to the same trees in the future. Samples were taken with a cork borer from the trucks of all trees (Fig. 1) and leaf samples were taken for selected trees. Very few cankers were found and most were healed over. Some of these were sampled.



Figure 1. 100-year-old C. mollissima sampled (M1).

3. DNA was isolated from all the woody plugs and leaves collected. The leaf DNA was more pure, so it will be used in further studies. Three DNA samples from what was deemed the healthiest trees were sent to Dr. John Carlson at Penn State University for genome sequencing. The DNA

sequences, when complete, will be available to all researchers at http://www.fagaceae.org/home . The DNA samples were M1L-D1 (11ug) and M1L-D2 (10.9ug) from the M1 Castanea mollissima tree (Fig. 1); S3L-D1 (5.8ug) and S3L-D2 (6.3ug) from the S3 C. seguinii tree (Fig.2); and H5L-D1 (4.1ug) and H5L-D2 (3.6ug) from the H5 C. henryi tree (Fig. 3).



Figure 2. C. seguinii sampled (S3).



Figure 3. C. henrvi sampled (H5).

- 4. RNA was isolated from all the cork borer samples and made into cDNA. This cDNA will be used for gene expression studies in future research. Additional cDNAs were made from each species from healthy tissue and from canker margin tissue (some of which were healed). These are being made into full-length cDNA libraries for gene sequencing and for future cloning of genes for use in producing transgenic American chestnut trees at SUNY-ESF and our collaborators, Dr. Scott Merkle and Dr. Joe Nairn at UGA. Copies of the finished cDNA libraries will be sent back to our collaborators in China.
- 5. Metabolites were isolated from selected samples and sent to Dr. Tim Tschaplinski at Oak Ridge National Labs, TN. These will be used to compare differences in metabolite accumulation between species. We are not sure how well this will work because typically these types of samples need to be kept frozen, which was not possible when shipping from China.



Figure 4. Plug and leaf samples in RNAlater solution (left) and precipitated DNA from these samples (right).

Pictures from trip:



Figure 5. Sara's GPS tracking of the drive to collect samples from the chestnut tree species.



Figure 6. Research group in Zhengan County.



Figure 7. Mr. Chen showing Kathleen and Katie a



Figure 8Research group collecting Cmollissima samplesFigure 9Andy taking a cork hore sampleEvaluating the Genetic and Genomic Basis for Different Levels of Blight Resistance among Native ChineseChestnut Trees (Castanea mollissima, C. henryi, and C. seguinii)Page 4 of 5



Figure 10. Katie and Sara recording data on trees.



Figure 11. Amelia extracting metabolites.



Figure 12. Amelia & Kathleen working in the lab.



Figure 13. Andy & Bill grinding samples.