Biology, Bikes, and Beida

My Summer Internship at Peking University

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Acknowledgments

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Finally, I must thank my family, friends, and members of my community, all of whom supported me during my trip, whether through a comment on my blog or by waking up at 2 AM to chat with me online. Because of your encouragement, halfway around the world didn't feel so far from home.

Introduction to the World Food Prize Foundation

I credit my initial exposure to the World Food Prize Foundation to my mother. As an employee of Pioneer Hi-Bred, she knew that the company planned to host a gathering of high school students speaking and collaborating on the issue of world hunger. Although I was still a little unclear about the details of the program, I agreed to attend one of the small-group discussion sessions.

What I saw impressed and intrigued me. The concern of these high school students regarding world hunger stirred them enough to make direct proposals for how to alleviate it. And even more amazingly, experts in this subject were listening carefully and encouraging the students' enthusiasm, rather than dismissing their inexperience. I resolved to investigate this program further.

After I educated myself about the wealth of opportunities offered through the Youth Institute and Borlaug-Ruan Internship, and the significance of the World Food Prize itself, I knew I had to be a part of it. In May of my junior year, I gained permission from my principal to represent our school, asked my biology teacher to be my sponsor, and sent in my application for the 2006 Youth Institute.

Since then, I have explained the aims and achievements of the World Food Prize Foundation to many people, from my teachers to people sitting next to me on the plane to my college roommate. But I never tire of describing the details because I gain great satisfaction from knowing that another person acknowledges this admirable program, and the serious issue it tackles.

In order to attend the Youth Institute, I wrote a paper speculating on the nature of a second Green Revolution, while focusing on a case study of Honduras and the prospects of agroforestry in that nation. Though I had spent much of my summer researching and reading about both these topics and the greater conversation on world hunger, I felt I only grasped a shadow of this complex and compelling issue. I longed to know what the statistics I was citing really meant, and whether the tentative suggestions I made had any significance in a practical context.

Once at the Youth Institute, however, excitement and curiosity replaced my frustrations. While listening to scientists and experts from around the world speak about

their work gave me a sense of the daunting extent of world hunger, I found remedy in the company of people passionate about finding solutions.

Those people included not only the experts but also my peers. By discussing the conclusions we had reached in our papers, I gained a small glimpse of how my own generation is coming to face world hunger.

I also mark my participation in the Youth Institute as the point when my perspective on eradicating poverty shifted from an altruistic wish to a moral obligation and important influence on my personal aspirations. (Of course, my internship further reinforced this view.)

With this new drive, I needed little convincing to apply for the Borlaug-Ruan Internship. I felt far less certain about my chances of receiving it. I had met so many well-informed and intelligent students at the Youth Institute that I tried hard not to get my hopes up. When I received a thin envelope from the World Food Prize in early March, I was afraid to open it. Luckily, it contained good news, and I dashed outside through the snow (forgetting my shoes in my excitement) to tell my family I was going to China.

Less than a year has passed since then. And yet so much has changed that it feels like much longer. I knew that China would be unlike anything I had experienced before, but I could not anticipate the ways that it would change me, and how I view the world. Something about those long summer days in Beijing forged an identity that I now carry as a Borlaug-Ruan intern.

Peking University

Peking University occupies the northwest section of Beijing known as the Haidian district, an area home to many universities and technology-related businesses. But perhaps no school in Beijing bears the renown of Peking University. Both its international prestige and historical significance as the first national university of China have built its stature as the leading university in Asia (1).

That said, I myself had never heard of Peking University. After learning of its distinction, I wavered between feeling excited and honored to visit such a place and bewildered at how I could be so oblivious to something that could be so famous to millions. This would not be the first time my ignorance of China's sheer size was brought to my attention.

The National Laboratory of Protein Engineering and Plant Genetic Engineering

Once I had spent a few days on campus, however, I quickly grasped both the size and the international breadth of the university. Classes there are still held during the summer, meaning that 15,000 undergraduates and nearly 1,800 international students were roaming the campus when I arrived (2). This meant that, as I was walking, I would see students from many other countries besides China, in addition to tourists of all ages and nations.

Academics aside, Peking University would garner attention based solely on the striking beauty of its setting. The campus emulates the palace gardens of Chinese emperors, so many of the buildings imitate traditional architecture - white walls with stark red trim and sloping, elaborately painted wooden roofs. Walks between these structures follow winding paths lined with ancient trees and exotic plants (exotic to me, anyway). Whenever I had free time, I loved exploring the expansive grounds or sitting by the peaceful Unnamed Lake.

I spent most of my days in the National Laboratory of Protein Engineering and Plant Genetic Engineering. A short walk from my room in Shao Yuan, the international students' dormitory, took me past the bustling campus gate to the quiet corner which accommodates the lab. While the exterior of the building displays design as traditional as its neighbors, the interior exudes the innovation of scientific research. The entryway walls exhibit enlarged abstracts and diagrams of studies which have come from the department. Adjacent hallways lead to many rooms crowded with research projects in progress and busy students. Even on the occasions when I visited the lab late at night to send emails home, I could always find students hard at work.

My own work in the lab relied on the instruction of the enigmatic Dr. Dingming Kang. Dr. Kang was the first person to welcome me to Peking University – or Beida, as he and the other locals call it (short for Beijing Daxue). During my stay, he acted as my supervisor in the lab, as well as cultural guide and language tutor. Although his responsibilities as a professor both at PKU and the Chinese Agricultural University (CAU) kept him very busy, he still made time to demonstrate experimental procedures and show me around campus.

When Dr. Kang did leave the lab, however, he did not abandon me. While I was still becoming oriented with the daily rhythm of the lab, I would shadow students and observe their work. "Follow that boy" or "that small lady" Dr. Kang would say of graduate students several years my senior. Although my inexperience must have provided a challenge, all of the students were so kind and patient. Whatever challenges I encountered, they were always willing to help, or just to chat and share lunch.

One aspect of the lab that undoubtedly made my internship easier was the fact that many of the students had excellent English skills. This is largely because English is quickly becoming the standard language for discussion and publication of scientific research in the international community. Many students eagerly used me as an excuse to practice speaking English, and those less outgoing compensated with big smiles.

Overall, Peking University provided both a beautiful, sheltered oasis in the midst of a bustling city and a cosmopolitan center of culture and ideas. It turned out to be an ideal environment to work, make mistakes, learn, and triumph.

Research Projects

As an intern at Peking University, my duties were to assist in the ongoing studies in plant genetics at the lab in whatever capacity I could. After discussion of some of the lab's current initiatives, Dr. Kang determined those projects best suited for my abilities and interests. During the first week or two, I observed and contributed to experiments in small ways (measuring components of a gel, taking flasks to be autoclaved, etc.). As I became more familiar with the lab, and its occupants acquainted with me, I began to participate more fully. Over the course of the summer, I had significant involvement in three projects: 1) developing tissue culture methods for *Haworthia comptoniana* 2) running SDS-PAGE of maize protein 3) extracting RNA from rice.

I will describe these experiments in this order, as it is the sequence they were introduced to me. However, it must be noted that there was overlap in the time frames of each.

Haworthia Comptoniana Tissue Culture

Plant tissue culture is a general method commonly used in experiments concerning plant genetics because it allows one to produce an identical copy of a plant in a sterile environment.

The basic process of tissue culture is relatively straightforward. A sample of the plant (called the explant) is placed in a sterile flask on an agarose gel containing nutrients and hormones which will induce the growth of a callus. These calluses are undifferentiated cells, meaning that they can develop into tissue of any part of the plant (leaf, roots, etc.). When a callus is transferred to another gel, the components of that secondary gel will determine the plant tissue produced.

***Haworthia comptoniana* (3).**

I was involved with an experiment whose objective was to develop such a regeneration method for the plant *Haworthia comptoniana.*

Haworthia comptoniana is of the same family Asphodelaceae as aloe, and bears phenotypic resemblances, such as thick, semi-firm leaves growing

in clumps. It originates in South Africa, and has adapted to thrive in arid climates (3).

Our research focused on this ability to resist drought. Perhaps if the genes responsible for this trait could be identified, that knowledge would provide insight for developing more drought resistant hybrids of other plants, especially crops such as rice. The first step in pursuing this initiative was to develop a reliable procedure for regenerating *Haworthia comptoniana* specimens through tissue culture.

I would contribute to this effort by cultivating samples of *Haworthia comptoniana* and observing the resulting calluses.

Experiment Procedure

When beginning the tissue culture process, the solution for the gel was first mixed in a one-liter flask. This medium was composed of distilled water and agar to form the gel, and MS medium and sucrose to provide nutrients for the plant cells. Hormones were added to promote callus growth. Different combinations of hormones were tested in the *Haworthia comptoniana* study as part of the search for a reliable tissue culture procedure. (See Appendix A for further description of hormones used.)

After all the components of the gel medium were combined, sodium hydroxide or hydrogen chloride was used to adjust the pH. Throughout the adjustment of pH, the solution was mixed thoroughly using a magnetic stirrer.

The flask was then covered and autoclaved in order to sterilize the gel medium. After removal from the autoclave, the medium was quickly poured into smaller, 100-milliliter flasks. Each flask contained 40-60 milliliters of gel medium. It was important to divide the solution promptly because the liquid set to form a gel as it cooled.

Working on tissue culture

I must pause to emphasize the necessity of a sterile working environment in performing tissue culture. After the gel medium is autoclaved, it is essential that it and all objects in subsequent steps are uncontaminated by bacteria. This meant that I performed tissue culture in a laminar flow cabinet, which circulated air outward

in order to prevent spores in the air from contaminating the gel or plant sample. Other necessary precautions were passing the mouth of each flask and all blades and forceps used through a flame, as well as frequently soaking my hands with 75% ethanol.

The process of sterilizing the plant samples themselves was especially stringent. The explant was immersed in 75% ethanol 3 times, rinsed with distilled water, soaked in 20% hyposodium chloride for 20 minutes, rinsed with distilled water 5 times, and placed on sterilized paper to dry. Once sterilized, the explant is sliced into small (1-3 cm) pieces, 3-5 of which were placed in a single flask.

Completed set of *Haworthia comptoniana* tissue culture

In the first weeks of my internship, I gained practice in this procedure by performing tissue culture for *Arabidopsis thaliana*, for other projects in the lab, under Dr. Kang's supervision. Once I was comfortable with the method, I was allowed to do two sets of tissue culture using *Haworthia comptoniana* (one of 28 100mL flasks, and another of 19 100mL flasks).

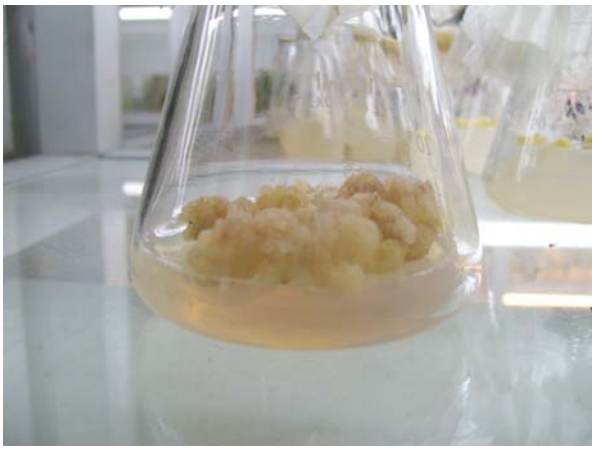
In order for calluses to be induced, the flasks were kept in darkness at 22 degrees Celsius. However, it was uncertain how long it would take for calluses to develop. The time frame varies, with rice and tobacco generally taking 15-20 days, and other plants much longer. I knew I had no guarantee that I would see the results of my experiment within the eight weeks I had in China.

My first set of *Haworthia comptoniana* tissue culture did not yield results, because of human error rather than time constraints. Despite my careful defenses otherwise, 22 of the 28 flasks I assembled on June 28 showed signs of contamination two weeks later. Sprawling white fungal growths surrounded my meticulously cut samples, rendering them useless for experimentation.

Contaminated Tissue Culture

Needless to say, I was disappointed and frustrated, but Dr. Kang was unfazed. "Don't worry! You are learning!" he told me. However, I was still apprehensive about the results of my second attempt.

Fortunately, that set, done on July 3, fared much better, and only 2 of the 19 flasks were contaminated. Dr. Kang surprised me by beckoning me downstairs on August 1, just a few days before I left, and showed me the flasks that had been tucked away for nearly a month. I was thrilled to see each flask blooming with calluses!



Haworthia comptoniana **calluses (see Appendix B)**

I was able to use these calluses to perform the next step in tissue culture. This time, five gel media were prepared, each with a different combinations of hormones to test which might promote growth of different specialize cell types. (see Appendix C)

Each medium was autoclaved and divided into ten flasks. These were then taken back to the laminar flow cabinet, where I transferred small pieces (again, 3-5 of 1-3 cm in size) of callus material to each of the flasks containing new medium.

This is the stage at which I left the project in August, when I had to depart for the U.S. Tissue culture is a time consuming process, and my two months at Peking University only allowed me to contribute to a few steps in the development of *Haworthia comptoniana* tissue culture. The results from my experiment suggested that the D1 medium used was slightly more effective at inducing calluses, but more repetitions of the experiment would be necessary to confirm this. In addition, many other variables of

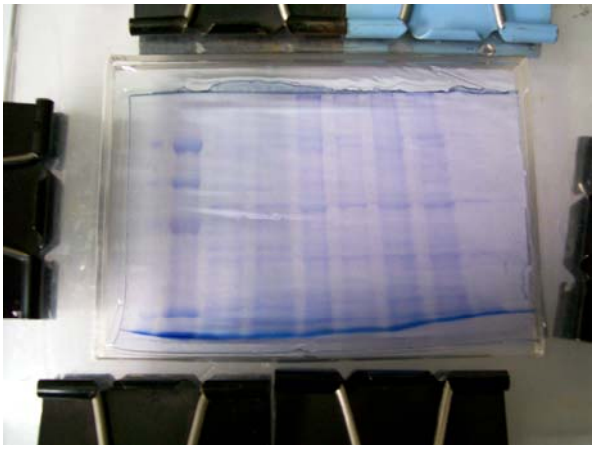
process, such as sterilization techniques, hormones used in the medium, and exposure to light, may be modified to yield a more or less effective method.

SDS-PAGE of Maize Protein

In early July, I began work on a small side project with Dr. Kang. He was studying two strains of maize which were being grown in a small plot behind the lab. One strain was fertile, but the other was sterile (unable to self-pollinate) because of a genetic mutation. Unsure of the exact genetic cause, Dr. Kang recruited my help in running SDS-PAGE on protein taken from leaf cuttings of each strain.

SDS-PAGE stands for sodium dodecyl sulfate polyacrylamide gel electrophoresis, a process which sorts proteins according to size. This is done by using an electric charge to propel the proteins through the web-like structure that agar forms in a gel. Proteins of the same size are stopped in the same space, with smaller proteins traveling farther along the gel. Thus, each protein type forms a distinct band.

By running SDS-PAGE on samples from the two strains of maize, differences in proteins may be identified, suggesting the genetic cause for one strain's sterility. As was the case in the tissue culture project, Dr. Kang and I would be performing the initial step of an extended experiment. Running 1-dimensional gels of the proteins would determine whether the quality of the samples was sufficient for further 2-dimensional SDS-PAGE analysis (sorting by charge in addition to size).



Most distinct results from SDS-PAGE

The first step was to obtain the samples of the maize. A cutting of leaf from each strain was brought into the lab and soaked in liquid nitrogen while being ground with a mortar and pestle. This created a very fine powder which could then be mixed with a running buffer.

The gel used in this procedure was approximately 7 x 12 cm and paper thin. It was composed of two parts, a lower and upper gel, with a higher concentration of agar in the lower gel for finer separation of the

proteins. The gel was kept clamped in a vertical frame and submerged in the running buffer. Electrophoresis was applied, and a long wait ensued. It would take up to five hours for our gels to finish running. (See Appendix D for gel and buffers used.)

After a few repetitions, we obtained a gel whose bands were distinct enough to justify further analysis with 2D SDS-PAGE, but unfortunately time constraints did not allow me to proceed to this second step. However, SDS-PAGE did give me excellent practice with grinding plant samples and preparing solutions - skills necessary for my next project: RNA extraction.

Extraction of RNA from Rice

Extracting RNA from rice was the final project I worked on, as well as the most complex, delicate, and time-consuming. Even with two students from the lab guiding me, Dr. Kang emphasized that this was a difficult process, and I felt honored that I was trusted enough to help.

The samples of rice we used were obtained from fields belonging to the CAU, located just outside of Beijing. I visited these fields my first week at Peking University, since my upbringing in Iowa had given me no introduction to rice paddies. A few weeks later we returned to cut samples from the leaves, which were immediately placed in test tubes and frozen with liquid nitrogen. This prevented the RNA from degenerating, keeping it in optimal condition for extraction.

Grinding rice samples into powder

As in SDS-PAGE, the samples were first ground to a powder while being constantly doused with liquid nitrogen. But because we would be analyzing RNA instead of proteins, it was much more important to keep the samples sterile. This meant wearing two layers of gloves and a face mask and being extremely cautious to make sure nothing touched the samples.

Once the samples were ground, we proceeded to use the heat phenol method for RNA extraction. Because of the chemicals used from this point on, it was also necessary to wear a lab

coat.

Each day we conducted RNA extraction we were able to use samples from two varieties of rice. This meant we were able to take six 100-200 mg samples of tissue from each variety. Each of the twelve samples were placed in test tubes and mixed with 600 microliters of extraction buffer, which had been mixed immediately beforehand. This step was especially sensitive because the rice samples had to be kept as cold as possible to avoid RNA degeneration, but if the test tubes were too cold, the extraction buffer would freeze and expand, causing the entire mix to burst open.

This was followed by a long series of steps, which consisted of repeatedly placing the test tubes in a centrifuge or vortex, reserving either the supernatant or pellet remaining, and incubating the sample at 4 degrees Celsius. Each step further isolated the RNA in the plant sample. (A full description of the process used may be found in Appendix E.) After these steps had been completed, the final pellet was cleansed with 100% ethanol and dried, then dissolved in 10 microliters of DEPC water.

This solution was then added to the RNA loading buffer, incubated for 3-5 minutes at 65 degrees Celsius, chilled on ice, and loaded onto the gel. The gel used was a formaldehyde agarose gel, and was much thicker (1 cm) than that used in SDS-PAGE. It also contained ethidium bromide, used to stain the RNA so that it becomes fluorescent under an ultraviolet light. Because ethidium bromide is a mutagen, I was not allowed to mix and pour the gel. However, I did assist in the sensitive task of loading the RNA into the tiny wells in the gel. The gel would run for about an hour, after which the results could be photographed and analyzed.

The entire process, from grinding the rice samples to running the gel, took an entire day's work. The last few weeks of my internship were spent duplicating this procedure and recording which samples taken from the field provided the clearest results on the gel (meaning most distinct bands). RNA that withstood this process would be less likely to degenerate, and thus more suitable for further experimentation.

Reflections on Research

I traveled to Peking University for the purpose of observing and working alongside scientists concerned with doing research which may ultimately improve food security. While I have always admired the work that such researchers do, I now have

even greater respect for the patience and diligence required, having witnessed it firsthand. Each of the projects I took part in (all offshoots of Dr. Kang's or students' work) constituted a small step on the long road toward important agricultural innovations. And yet the procedures were painstaking and time-consuming, obliging one to stay fully focused on the task at hand. It is a commendable feat indeed to remain inspired and bear in mind the ultimate goal of this work, so that each step, from running gels to sterilizing forceps, feels relevant and satisfying. This attitude clearly appeared in the lab, where students spent long days and nights with their projects, and professors were continually meeting and discussing their latest findings. In the midst of all I learned about biotechnology and plant genetics, I also absorbed this focused mindset and found it as vital to research as any experiment or equipment.

Cultural Experiences

In addition to observing the scientific process which leads to progress in food security, I sought to experience and try to understand a country and culture other than my own. These two objectives were very much connected. Through exposure to daily life in Beijing and by speaking with Chinese citizens from various ways of life, I gained some small sense of the complex history and culture of China, and how food security initiatives must take these into account. On the other hand, the scientific progress I witnessed in the lab revealed some of the tools available for developing nuanced solutions.

Because of this relationship, the abundant cultural experiences that I enjoyed this summer bore enormous importance.

Life in Beijing

Beijing itself is an environment very different from the one in which I grew up. Having spent most of my life in rural Iowa, visiting cities only slightly bigger than Des Moines, the transplant to a metropolis of fifteen million delivered quite a shock. When driving through the city, I constantly tried to absorb both the crowds of people and the advertising bombarding me from every storefront. Luckily, the garden-like setting of Peking University softened my abrupt change in habitat. Another urban feature, air pollution, also called for rapid adaptation on my part. Descending in the plane over the city, I could soon see the thick haze encasing it. Although I gradually became

accustomed to the lackluster air quality, the hordes of cars filling the streets continually reminded me of Beijing's smoggy predicament.

Because I lived on campus, I only traveled by taxi on a few occasions. For trips across the city, a student would usually accompany me on the subway, and shorter distances called for a ride on the bus. I became familiar enough with the route between PKU and the CAU that Dr. Kang sometimes let me take the bus by myself. The shout of "Beijing Daxue!" and a nudge from my neighbor let me know that I was at my stop.

However, I most frequently moved through the city on the wheels of a bike. Dr. Kang is an avid cyclist and insisted that I get a bike of my own for the summer. Within a few days of my arrival, we had gone to the mall, and on the way back I had my first experience biking the streets of Beijing. It was terrifying, to say the least, to ride inches away from gigantic buses and honking cars. However, I willingly agreed each time Dr. Kang suggested we go for a bike ride, because I felt it gave me a uniquely authentic perspective of Beijing. Citizens of all ages rode bikes in the street, and when I rode alongside them, I felt less like a conspicuous American, and a little more like a native.

If biking through Beijing traffic did not break me in to a Chinese lifestyle, eating Chinese food certainly did. Honestly, the food was one of the things I had most looked forward to about the trip. I consider myself extremely fortunate to have sampled a wide variety of foods in the span of two months. Most days I ate at one of the many dining halls on campus (by my own count, there were at least a dozen). I tried to make sure that I ate something new as often as possible, a goal easily accomplished at the majority of my meals. I developed quite an affection for the rice, steamed bread, dumplings, noodles, and many dishes of spicy meats and vegetables that I tasted. Although I did pick up several Chinese words for food, my vocabulary was not quite extensive enough for the array of options that I faced each day. Luckily, the smiling servers were quite accomplished at interpreting my gestures. I tried to make up for my inexperience with Chinese food by becoming comfortable with chopsticks. Fortunately, experts surrounded me at every meal, and I learned through observation much more quickly than I had expected.

Although the rhythm of the city and different food set a vibrant backdrop to my daily life, my cultural experience was shaped most by the community of people with

whom I interacted on campus. My appearance marked me plainly as a foreigner even at a school as international as Peking University. As a result, I had to grow accustomed to the stares, and occasional picture-taking, that came with this designation. However, the attention also gave me the opportunity to meet many more students than I might have otherwise. Once assured of my nationality, they eagerly tested their English slang or asked questions about my country.

I remember in particular one encounter I had in the courtyard at the center of campus. I had finished the day's work in the lab and planned to relax with some Chinese ice cream and watch students playing frisbee. Within a few minutes, a young woman approached me with the greeting, "Hello, are you an American?" After giving my usual spiel on who I was and what I was doing in China, she began to tell me about the circumstances that had brought her to Beijing. She frankly described her upbringing in rural China in a largely illiterate village and how she had been able to attend a university and obtain a degree in German philosophy. Now, after teaching for several years, she was feeling the pressure from her family to get married, and had traveled to Beijing in hopes of finding a husband. As we chatted, two Peking University students who had overheard our English joined us. One student had English homework, which I gladly examined, and the other had many questions about American politics. By the time we all dispersed, the moon had risen, and my ice cream had long ago melted.

I meandered back to my dorm, considering the lives I had stumbled upon, especially that of the young woman. We had both come to Beijing as visitors for two months, and were both curious about other cultures. "To see the world is my dream," she had said to me. And yet our situations were very different. I had come to Beijing for eight weeks of untroubled exploration and education. She came urgently seeking a future for herself. As an American, I had the privilege to pass easily between the US and China and nearly any other country in the world. She, on the other hand, asked me earnestly if I had any tips for gaining permission to travel to America. These unjust differences between us gnawed at me, and gave me a glimpse of the many privileges I had hitherto taken for granted.

Field Trips

While I did quite literally take field trips to the rice paddies of the CAU, I had several other opportunities to travel outside of Peking University and Beijing. Many were site-seeing trips, taken on weekends with either students from the lab or Dr. Kang as my guide. Although visiting such monuments of Chinese history as the Great Wall and Forbidden City proved fascinating and awe-inspiring, I most valued the trips with purposes other than tourism.

For instance, on June 21 and 22 I was able to attend the 18th International Conference on Arabidopsis Research, held that summer outside of the U.S. for the first time. Dr. Gu arranged for me to attend the conference, and I appreciated this act fully once I mentioned it to some students. They gestured emphatically while trying to express the importance of this conference and their excitement that it would be held in Beijing. The conference took place in an elegant hotel, and included dozens of presentations given by scientists from all over the world on Arabidopsis research. This area of study has wide-ranging applications because *Arabidopsis thaliana* has a small genome and is easily experimented upon, and is therefore suitable for many fields of research. Because I, unlike 95% of the conference attendees, did not have a collegiate degree, I missed some of the finer details of the presentations. However, this did not detract from the unique chance to view a multinational conference, conducted in English, from a Chinese perspective.

Further diversification of my understanding of the Chinese culture came through a weekend expedition made by Dr. Kang, JoAnn Kirsch, and myself, to a rural village outside Beijing. A few days away from the city gave us the opportunity to stay with a young woman and her daughter, and explore the village. Nestled in a mountain valley, the town consisted of clusters of simple one-story brick houses. Our host lived in one of the clearly nicer homes, which had several rooms and running water. A small round table sat in a central, roofless room where our host served the "home-cooking" Chinese meals which JoAnn and I helped to prepare. However, not all the villagers had these comforts. While we wandered the dusty streets, we visited the home of one of the older villagers. The small woman led us into her house comprised of two tiny rooms with dirt floors and newspaper plastered on the walls. My eyes were quickly drawn to the only source of

color – intricately cut red paper symbols of the Chinese zodiac. The woman grinned as she gestured, with obvious pride, that she had made them by hand. She had some type of speech impediment, and could only make strained, indistinct noises. So, whatever information she could convey was spoken to our host, whose Chinese was then translated to English by Dr. Kang. But the message of poverty did not need to be spoken; it was communicated quite clearly by her surroundings. And I felt the strongest implications came from what I did not see or hear – the millions of others in China living in conditions like hers and worse.

Personal Reflections

When considering the ways in which my trip to China has affected me, I sometimes think that it would be easier to count the ways in which it has not. My mother recently asked me if remembering the trip felt like recalling a dream, distant and unreal. On the contrary, my memories of China permeate my life daily. I am constantly comparing my daily life in America to that I which I led in China, and my conversations regularly lead to stories I can relate from my experience.

Many of these changes come in concrete forms. I am now quite skilled at using chopsticks, a fearless cyclist, and rather unimpressed by American versions of Chinese food. Also, I am studying Chinese language as one of my courses at Grinnell College, and find it elating to piece together the characters and sounds I encountered while in China. My summer experience will undoubtedly influence all of my choices about future studies. Now more than ever, I am determined to do work which will contribute directly to poverty alleviation, especially regarding world hunger. In order to accomplish this, I must carefully construct my pursuits of anthropology, biology, economics, language study, political science, and economics, as well as decisions about where to study abroad.

Other changes are less quantifiable, but far more revelatory of the importance of my internship. One critical, albeit unsurprising, transformation was of the way I view China. Before my journey to that country, I thought of China in the context of the news stories by which it entered my life. As a result, ‘China’ to me connoted an immense and foreboding country, characterized by an authoritarian government and rapid economic growth. In other words, I regarded China as a single entity, and although I knew it

contained 1/5 of the world's population, I could not comprehend that as anything more than a number. For me, traveling to China and interacting with its people propelled the quantum leap to appreciating a country on a deeply human level. Although the fact that a country is made up of individuals is a simple conclusion to draw, a world of difference lies between understanding it and believing it.

As I mentioned, I expected the internship to illuminate my views of China. What I did not anticipate to shift were my own perspectives of the United States, and my identity as an American. Throughout my life, I viewed my upbringing in rural Iowa as pleasant, but also modest and commonplace. The more time I spent in China, the more I began to fathom the extent to which my life had been privileged and carefree. I found it quite unsettling to realize how lucky I was to have been provided with food, water, a spacious home, a quality education, and political freedoms in addition to numerous material luxuries.

When discussing these discrepancies with my Chinese friends, the effect sometimes seemed merely ridiculous, such as when I admitted that my family owned five vehicles. On other occasions, the evidence had more somber effect. One day, while strolling through the fields of the CAU with Dr. Kang, I happily recounted how often I had wandered through the rolling fields of my Iowa home. Dr. Kang said that he, too was familiar with this practice, but because he had gathered bits of unharvested grain from the fields for his family, as a child during the great famine of 1958-1960.

Understanding the true circumstances in which I had grown up helped me to fully recognize my ignorance of lifestyles other than my own. Although living in Beijing greatly broadened my horizons, it also instilled in me a desire to learn more. The life I led in China was not by any means fully representative of all Chinese, or even all Beijing citizens. I am extremely eager to continue my exploration of China, and the rest of the world.

My summer in China changed me in ways both expected and unexpected, introducing me to scientific innovations, proving that I could adapt to an unfamiliar environment, and altering my understanding of the world in which I live. These are the results of my internship, the ways that my experience has affected me. I hope that they will serve as valuable tools for ways in which I, in turn, will impact the world.

Appendix A

Media Used in Callus Induction

Medium C1

1000 mL distilled water

4.4 g MS

7 g agar

30 g sucrose

hormones: 6-BA (2 mg/L)

NAA (0.2 mg/L)

KT (1mg/L)

Medium D1

1000 mL distilled water

4.4 g MS

7 g agar

30 g sucrose

hormones: 6-BA (3 mg/L)

NAA (0.2 mg/L)

Explant Sterilization Procedure

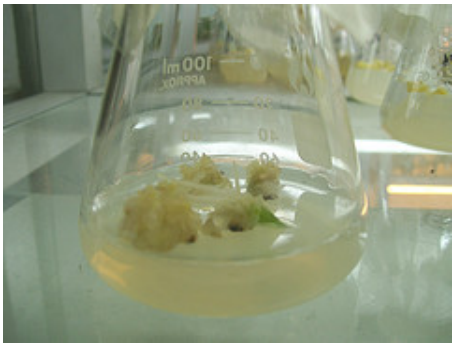
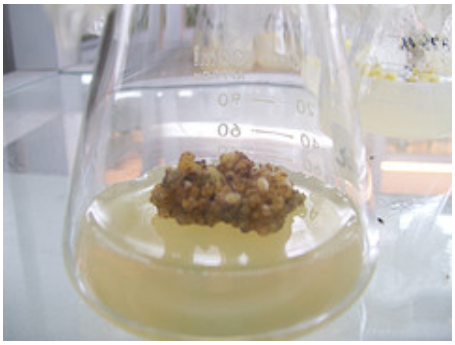
1. Soak explant in 75% ethanol, shaking flask gently for one minute. Repeat twice.
2. Rinse explant once in distilled water.
3. Soak explant in 20% hyposodium chloride, shaking gently for 20 minutes.
4. Rinse explant in distilled water 4-5 times.
5. Dry explant on sterilized paper filter.

Appendix B

Selected Photographic Documentation of Calluses





Appendix C

Media Used in Callus Transfer

Medium C2	Medium D2
1000 mL distilled water	1000 mL distilled water
4.44 g MS	4.44 g MS
30 g sucrose	30 g sucrose
7 g agar	7 g agar
1 6-BA	3 KT
1 KT	0.2 NAA

Medium A8	Medium A9
1000 mL distilled water	1000 mL distilled water
4.44 g MS	4.44 g MS
30 g sucrose	30 g sucrose
7 g agar	7 g agar
8 6-BA	3.5 BA
1 KT	0.5 KT
0.1 NAA	0.1 NAA

Medium C3

1000 mL distilled water

4.44 g MS

30 g sucrose

7 g agar

0.5 BA

0.5 KT

0.1 NAA

Appendix D

SDS-PAGE Gels and Buffers

Upper Gel (total volume 2 mL)

Deionized water	1.4 mL
30% acrylamide	0.33 mL
1.0 mol/L Tris (pH 6.8)	0.25 mL
10% SDS	0.02 mL
10% ammonium persulfate	0.02 mL
TEMED	0.002 mL

Lower Gel (12%) (total volume 5 mL)

Deionized water	1.6 mL
30% acrylamide	2.0 mL
1.5 mol/L Tris (pH 8.8)	1.3 mL
10% SDS	0.05 mL
10% ammonium persulfate	0.05 mL
TEMED	0.002 mL

Running Buffer (5x)

Tris-Cl	15.1 g
Glycine	94 g
10% SDS	50 mL

add deionized water to total volume of 1L

Sample Loading Buffer (2x)

100 mM TrisCl (pH 6.8)

200 mM DTT (dithiothreitol)

4% SDS

0.2% bromophenol blue

20% glycerol

Appendix E

RNA Extraction Method (heat phenol)

1. Grind 100-200 mg tissue into fine powder in liquid N_2 and add 600 microliters extraction buffer. Mix immediately. Incubate in H_20 at 80 degrees Celsius for 5 minutes, mixing occasionally.
2. Add 600 microliters 24:1 chloroform: isopropyl alcohol and vortex for 30 seconds to 1 minute.
3. Centrifuge at 12,000 rpm for 5 minutes (at room temperature). Save supernatant.
4. Add 4M LiCl equal to the volume of remaining supernatant.
5. Incubate at 4 degrees Celsius for 30-60 minutes.
6. Centrifuge at 12, 000 rpm at 4 degrees Celsius for 20 minutes. Save pellet.
7. Add 0.5 mL 100% ethanol. Centrifuge at 12, 000 rpm at 4 degrees Celsius for 5 minutes.
8. Save pellet, dry at room temperature for 5 minutes.
9. Resuspend the pellet in 400 microliters DEPC H_20. Centrifuge at 4 degrees Celsius for 5 minutes at 12, 000 rpm. Save supernatant.
10. Add 40 microliters 3M NaOAc pH 5.2.
11. Add 2.5 volume 100% ethanol. Keep at -20 degrees Celsius for one hour or overnight.
12. Centrifuge at 12,000 rpm at 4 degrees Celsius for 20 minutes. Save pellet.
13. Add 0.5 mL 70% ethanol. Centrifuge at 12,000 rpm at 4 degrees Celsius for 10 minutes.
14. Save pellet, add 0.5 mL 100% ethanol. Centrifuge at 12,000 rpm at 4 degrees Celsius for 10 minutes. Dry pellet.
15. Dissolve pellet in 10 microliters DEPC H_20.

Composition of FA Gel Buffers (taken from the "RNeasy Mini Handbook" published by QIAGEN)

10x FA Gel Buffer

200 mM 3[N-morpholino] propanesulfonic acid (MOPS) (free acid)

50 mM sodium acetate

10mM EDTA

pH to 7.0 with NaOH

1x FA Gel Running Buffer

100 mL 10x FA gel buffer

20 mL 37% (=12.3 M) formaldehyde

880 mL RNase-free water

Note: Equilibrate gel in 1xFA gel running buffer for at least 30 minutes before starting electrophoresis.

5x RNA loading buffer

16 microliters saturated bromophenol blue solution

80 microliters 500 mM EDTA pH 8.0

720 microliters 37% (12.3 M) formaldehyde

2 mL 100% glycerol

3084 microliters formamide

4 mL 10x FA gel buffer

Add RNase-free water to 10mL

Protocol for Formaldehyde Agarose (FA) Gel Electrophoresis (taken from the RNeasy Mini Handbook published by QIAGEN)

1.2% FA gel (100 mL)

1.2 g agar

10 mL 10xFA gel buffer

RNase-free water to 100 mL

Microwave to melt agarose. Cool to 65 degrees Celsius in a waterbath. Add 1.8 mL of 37% (12.3 M) formaldehyde and 1 microliter ethidium bromide (10 mg/mL). Mix thoroughly and pour onto gel support. Prior to running the gel, equilibrate in 1xFA gel running buffer for at least 30 minutes.

RNA sample

Add 1 volume of 5xRNA loading buffer per 4 volumes of RNA sample. Incubate for 3-5 minutes at 65 degrees Celsius, chill on ice, and load onto the equilibrated FA gel.

Gel Running Conditions

Run gel at 5-7 V per cm with 1xFA gel running buffer.

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