# Crossing Continents:
## The Scientific and Human Elements of Hybrid Rice




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2011 Borlaug-Ruan International Internship
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## ACKNOWLEDGEMENTS

I would like to express my deepest gratitude to the **World Food Prize Foundation** (WFP), **Dr. Norman Borlaug**, humanitarian, Nobel Peace Prize Laureate, and founder of the WFP, **Mr. John Ruan,** Chairman of the WFP, and **Ambassador Kenneth M. Quinn**, former US Ambassador to the Kingdom of Cambodia and President of the WFP, for the incredible opportunity they have granted me and the other Boralug-Ruan Interns. Their vision has impacted me in more ways than I can count; my experience has empowered me to be the researcher making the difference for millions. Such exposure to the issues of food insecurity inspires thousands of young adults, like myself, to lift millions of people out of food insecurity and to educate my fellow peers.

Special thanks to **Mrs. Lisa Fleming,** Director of Youth and Education Programs of the WFP, for being a fierce friend, advisor, and coordinator. Thank you for choosing me as a 2011 Borlaug-Ruan Intern, tirelessly coordinating my internship, and always looking out for helping me adjust to the new environment in Changsha. Without your help, constant support, and kind words I would not have had the global experience of helping the world.

Thank you to **Professor Yúan Lóngpíng,** the father of hybrid rice, Director-General of CNHRRDC, and 2004 World Food Prize Laureate, for allowing me to be part of the center and conduct research in the lab as well as inspiring me to complete such ground-breaking research on rice hybridization. I would like to thank **Dr. Yèyún Xīn** for arranging and coordinating the logistics of my internship at CNHRRDC. Thank you to my mentor, **Dr. Lìjié Zhōu,** for her hospitality from day one and superior instruction in the field of hybrid rice. Without her, I could not have conducted the experiment, analyzed the results on a global scale, and had the experience of a lifetime in Changsha, China. Special thanks to **Bīn Bāi**, **Chén Lǐ**, **Fùgēng Wǔ,** and **Fěi'tíng Kuàng,** graduates students at CNHRRDC, for teaching me Chinese, helping me to understand the research, and for always being a friend. To **Dr. Xiǎoxía Hán** and **everyone** at CNHRRDC, thank you for teaching me all about hybrid rice and the constant struggle between quality and quantity. Thank you all for your continual patience, kindness, and helpfulness throughout my two months in Changsha, China.

My sincere thanks to all my teachers at Rivermont Collegiate in Bettendorf, IA for their constant support. Special thanks to **Mrs. Linda Hampton**, my science teacher, for her dedicated encouragement of all of my pursuits and ambitions since the sixth grade. I would also like to thank, **Mrs. Gwen Livingstone Pokora**, my English teacher at Rivermont Collegiate, for being a constant mentor and supporter of my expeditions. Without such dedicated, supporting, and inspiring teachers, it would not have been possible for me to reach for the stars.

Lastly, I would like to express my endless love and gratitude to my family. A sincere thanks to my brother, **Ramana Gorrepati**, for constantly being my role model and a positive influence in my life, and my father and mother, **Dr. Krishnarao Gorrepati and Mrs. Pramilarani Gorrepati**, for their endless support of my dreams and endeavors – no matter where it takes me.

Gratefully,

## BACKGROUND INFORMATION: CNHRRDC

China National Hybrid Rice Research and Development Center (CNHRRDC) was founded in 1984, becoming the first institute to formally specialize in hybrid rice. Located in Changsha, Hunan Province, China, CNHRRDC concentrates on six distinct divisions of hybrid rice production: scientific research, international cooperation and development, technology development, administration, personnel, and finance. The Center conducts a variety of research pertaining to hybrid rice from hybrid rice and molecular breeding to observational journaling. Research at CNHRRDC has attained more than 50 national, provincial, and ministerial achievements; researchers have also developed over 40 commercially viable hybrid rice varieties, which have had a tremendous socioeconomic impact throughout China.

CNHRRDC has published several books such as *Breeding and Cultivation of Hybrid Rice, Hybridriceology, Technology of Hybrid Rice Production,* and the journal *Hybrid Rice* – one of the only technical journals specializing in hybrid rice. CNHRRDC dedicates itself to teaching the world about hybrid rice and sharing its wealth of information and knowledge. It has formed relationships with the International Rice Research Institute in Los Baños, Laguna, Philippines and other organizations in the US, Vietnam, Indonesia, Pakistan, Bangladesh, Israel, and Malaysia. Over 600 scholars and officials from 30 different countries have visited and studied at CNHRRDC (China National Hybrid Rice Research and Development Center).

Professor Yuan Longping, "Father of Hybrid Rice" and 2004 World Food Prize Laureate, is currently the Director-General of CNHRRDC. A debilitating famine caused by a chain of natural disasters and unsuitable policies in the 1960s inspired Professor Yuan Longping to hybridize rice. A big idea in the 1960s has turned into a big success by yielding up to 20% more than inbred varieties (Li, 2001). Today, approximately 50% of China's rice fields grow Professor Longping's various hybrid rice species – feeding an additional 70 million people annually. For his work, determination, and success, Professor Longping has received numerous accolades including the 2004 World Food Prize, the 2004 Wolf Prize in Agriculture, 2001 Magsaysay Award, China's State Supreme Science and Technology Award, and the United Nations Food and Agriculture Organization Medal of Honor for Food Security (Barclay, 2010).

### Longping High-tech International Exchange Center

International Training Courses on Hybrid Rice Technology 1999-*Present*

CNHRRDC places great importance on not only producing different strains of hybrid rice, but also informing and educating the rest of the world through training courses on hybrid rice technology. Located within the campus grounds, the Longping High-tech International Exchange Center hosts agricultural experts from all over the world for a four-month training course on growing, developing, and implementing hybrid rice in those countries where the utilization of hybrid rice would provide a world of relief for the bourgeoning population. Through modern training facilities, translators, experts in the field, modern management ideas, and advanced agricultural technologies, Longping High-tech International Exchange Center has trained over 700 people across the globe.

The Longping High-tech International Exchange Center has hosted agricultural experts from all over the world: Mozambique, Guinea Bissau, Angola, Brazil, Indonesia, Thailand, Cuba, East Timor, Sierra Leone, Sri Lanka, Philippines, Bangladesh, Nepal, Myanmar, Uganda, Vietnam, Sudan, Rwanda, Fiji, Cote D'Ivorie, D.P.R. of Korea, and Congo.

The program has helped to rapidly communicate and to apply hybrid rice technology worldwide and facilitate trade cooperation between China and the world. The program has proven to be a great success and importance in the struggle to obtain worldwide food security.

# INTRODUCTION

## Rice

Do you have a craving for some Vietnamese pho or some Spanish paella or maybe some Indian palau? All of these dishes utilize rice as their base. It is the one food that seems to transcend all cultural and geographic boundaries. And it seems that all people on this planet have a reason to appreciate its value. But what is it about this little grain that makes it so critical to the success of the world economy and to the health and nutrition of billions of individuals around the globe? Why does it play such an integral role in so many ancient cultures? Why have we become so reliant on this little grain of rice? Why is it readily available, and yet we incessantly try to improve upon it?

### *Culture, Origin, Planting, and Nutrition of Rice*

Beyond the obvious notion that rice is a crucial element of many meals and recipes passed down from one generation to the next, it also plays an important cultural role such that many rice-derived products are used in a number of different purposes including as fuel, thatching for the roofs of houses and huts, and rope that can be put together in an elegant patterns to form sandals, mats, rugs, brooms, and even raincoats. Even the hulls of the rice are put to good use for polishing, or as fertilizer, or for making paper. The role that rice plays in culture depends heavily on where you are in this world. In the United States, rice is simply a commodity, but in other nations like Japan, rice is much more. It is symbol of independence and tradition. In the Shinto religion in Japan, “honorable rice” is used to make sacred offerings of sake and rice cakes. Moreover, in this nation that needs to import most of its food, the Japanese government makes it a principle not to import any rice as long as they are able to sustain their needs through domestic cultivation (Krock, 2000).

Every culture has rituals and myths related to rice that unifies their heritage through this common thread. Myths concerning the origin of rice can take many forms and many cultural rituals are integrated into the preparation of rice crops and their harvest. An aboriginal myth from the Miao people of Sichuan, China recounts a time when their ancestors did not have the seeds to plant their rice paddy fields for the year. And so, they set free a green tropical bird, which flew up to the rice granary of the heaven god and returned with the heavenly rice seed. Similarly, a myth of the Minahassa region of Indonesia tells of a man who traveled to heaven and returned to earth with unhulled rice hidden in a wound in his leg. Another well-known aboriginal tale coming from Malaysia tells of time when all the sky was laid flat on the surface of the Earth in the form of rice, until one day when the women of the tribe cast it upwards in the first harvest. As the legend goes, from that time onward rice has fallen back to Earth for each year’s harvest (UNESCO, 1984). Similar stories are found not just in Southeast

Asia, but also among the agricultural peoples of Africa and the Americas. In China, it is believed that rice has been a stable cultivation for farmers for between 3000-4000 years, where it gradually became an important part of aristocratic life. In modern times, the country's rural culture has developed around the growing of rice, and foods made from rice are the basis of festivals such as the Land Opening Festival, which celebrates the start of the rice cultivation season and the Spring Festival (Jinhui, 2002).

The beginnings of rice cultivation dates back thousands of years, though the exact origin still remains an issue of fiery debate among countries each with strong claims of rice in their rich traditions. One theory suggests that the rice genus Oryza originated on the supercontinent Gondwanaland about 200 million years ago, which encompassed modern day South America, Africa, Antarctica, Australia, Madagascar, and India. The rice genus is a grass that belongs to the same family as barley, oats, rye, and wheat. Its robustness allows it to be cultivated in tremendously varied environments from the scorching deserts of Egypt and the Middle East to the cool Himalayan climates of Nepal down to the fertile, rain-filled fields in Brazil. Today, the genus Oryza includes over 20 major cultivated species including black wild rice, red wild rice, brown rice, mixed wild rice, green pandanus leaf flavored, and many more. The wild varieties of Oryza are widely distributed in the subtropical regions of the world including South America, Africa, and Asia (Chang 1985). There are two commonly cultivated species of rice, the first called *Oryza glaberrima* is limited to a specific region in West Africa while the other known as *Oryza sativa* is the more widespread and is the preferred commercial variety (Bertin et al. 1971). All in all, nearly 11% of the world's cultivable land, approximately 358 million acres, is used for rice cultivation according to the International Rice Research Institute (IRRI) in the Philippines (IRRI, 1997) Rice was originally brought to the pre-colonial American settlements in the mid-17th century and soon became an important staple. In the United States, while rice cultivation trails that of other major grains such as wheat and corn, the domestic productions still exceeds domestic consumption, so a considerable percentage is exported to Europe and parts of South America. Globally, rice production is geographically centered in Western and Eastern Asia with China and India accounting for more than one-half of global supply. World production has demonstrated tremendous growth in recent years, almost solely from increasing production in Asia.

The planting of rice can often times be an arduous year-round process whether grown as a dry-land crop in Brazil or as a paddy crop watered by monsoon rains and floodwater in China. The methods of growing differ greatly according to local tradition but in most Asian countries the traditional hand methods of cultivating and harvesting rice are still practiced. The fields will be prepared by initially tilling and aerating the ground with the help of water-buffalo drawn plows and then subsequently fertilized usually with manure or sewage. The rice seedlings, which are first planted in seedling beds for the first 30-50 days, are transplanted by hand to the fields, which have been flooded by rain or river water. During the growing season, irrigation of the fields is maintained by inflowing canals or by hand-watering. After they are cut, the rice, which is still covered by the brown hull and properly, referred to as paddy, is threshed to loosen the hulls. The hulls are then put through of process of drying, winnowing, dehulling and finally milling. Rice cultivation in the modern day has advanced considerably and become a primarily mechanized process.

Rice is oft considered the most important foodstuff for much of the world population, after all, according to the Manila-based International Rice Research Institute it is estimated that one out of every two individuals on this planet depends on rice as a staple food. Moreover, it represents a significant source of both calories in developing countries, providing over 21% of the calorific needs of the world's population and up to 76% of the calorific intake in South East Asia (Seck, 2011). But rice is also quite nutritious, especially when compared to other cereals. Rice has about 2.0 to 2.5 mg. of protein per 1/2 cup of cooked rice and is thus higher in protein quality than wheat and corn and comparable to lentils, peanuts, and oats, which enjoy a reputation as protein-rich foods (Houston and Canler, 1970). Rice is not often considered a good source of protein and is instead often maligned in Westernized countries for being an overly starchy and carbohydrate-rich food. A measure of protein content also serves as a good indicator of not just nutritional content, but also in determining functional characteristics important to food scientists such as cooking texture. The rice grain is also a good source of critical B-complex vitamins and iron and is also low in sodium and cholesterol (Juliano and Bechtel, 1985).

### ***More than Just a Grain for China***

China has a tremendously rich culture with arts, beliefs, customs, and institutions that date back thousands of years. Agricultural is fundamental to this country's history, and central to this is rice which has had a tremendous influence of China's history as well as on developments in ideology, society, politics, and economics. Chinese farmers have been diligently cultivating their land in China for thousands of years and millions still rely on the land thus substantiating its strong rural presence in society. This emphasis on rice has led the Chinese to pay special attention to advancing irrigation technologies and cultivation practices. Rice cultivation thus led to a distinct economic lifestyle in the ancient Chinese economy. Life revolved around the seasons with most Chinese working the land in certain ways during the different seasons with spring ploughing, summer weeding, and autumn harvests.

Traditional Chinese culture may very well be considered a "rice culture." Archaeologists have documented evidence that China began planting rice at least 3,000 to 4,000 years ago. In ancient China, vast amounts of land along the reaches of the Yangtze River were suitable for planting rice. To date, traces of rice have been located all across China from the Hanshui Basin in the northwest to the Guangxi Province in the far south. Rice was first grown at a time when people lived in hunter-gatherer societies in which most or all food was obtained from hunting, fishing, and fruit collecting. It is speculated that the birth of rice cultivation was a chance event when women at the time happened to leave seeds lying in lowland area (Amano, 1979)

By the rise of the Zhou dynasty from 1027 BC to 221 BC, rice had become well accepted in society and central to aristocratic banquets. During the Autumn Period in Chinese history, rice became a more central part of the diet for Chinese commoners. And by the time of the Han Dynasty around 206 BC to 220 AD, rice occupied an important cultural role for the commoner. It had become a specialty food that was used to brew wines and was offered as a sacrifice to the Gods (Amano). Moreover, rice was used in a variety of foods that became synonymous with several yearly celebrations.

It used to make rice dumplings that are eaten on the first day of the full moon each new year and also to make *zongzi* during the Dragon Boat Festival and rice porridge to celebrate the day that the Buddha attained enlightenment. Most notably, rice is major part of the lunar New Year celebration dinner where Chinese families make a New Year’s cake called “*gao*” in Chinese from flour milled from rice. Chinese make this cake in the hope of garnering blessings for the upcoming year’s harvests and wishes for a better future. In addition to eating rice for the New Year, they express good wishes in terms of rice. For instance, the Chinese say, "May your rice never burn!" in addition to "Happy New Year" (Newman, 2004). So rice has come to mean a great deal for the Chinese in culture, in celebrations, with religion, and for livelihoods.

### *Hybrid Rice*

With rice being such as staple in the Asian diet, it became exceedingly clear in the early 1960’s that the rate of rice production would be overwhelmed by the booming population growth in Southeast Asia, and that this could only result in widespread famine and soaring rice grain prices. Therefore, the need to increase rice production plays a fundamental role in maintaining food security and alleviating poverty and famine. This was the impetus behind the transformation of the rice plant. The newly established International Rice and Research Institute (IRRI) worked to overcome this potentially crippling obstacle by breeding a new variety of rice that was shorter in height to better support the increased grain clusters, that had a shortened growing period of 110 days down from the typical 160 days so that farmers could now cultivate two to three crops each year, and that was more resistant to devastating crop diseases and insects. According to the IRRI, the world’s cumulative rice production doubled in the period between 1967 and 1992, a time notably referred to as the Green Revolution (Krock).

The basis for hybridization is not a novel idea, the idea of seed selection and domestication to select for the desirable traits dates back to the first century B.C. in China. The trend in selection according to historical records in the *Ch'i-Min-Yao-Shu* encyclopedia mentioned that farmers in the field were focused on earlier maturity and the selection of bigger grains in order reduce cold damage and to yield a larger harvest (Chang, 1979).

It has been estimated that by the year 2030, the world will need to produce about 60% more rice than was produced during the mid-1990s. In theory, there is still great potential to be tapped in the quest for improved rice production including establishing better irrigation systems in developing countries, improving soil conditions and fertilizer use, and breeding higher yield hybrid varieties. China, in particular, is making significant progress in developing and promoting hybrid rice technology. Over 50% of the total land area in China, or nearly 15 million hectacres is devoted to rice cultivation. It has been consistently shown that hybrid rice improves yield by at least 20% in comparison to native domesticated rice varieties, with hybrid rice yielding about 7 tons per hectare versus an average of 5.6 tons per hectares for domesticated varieties. This increase of 1.4 tons per hectare feeds an estimated 60 millions additional people each year, therefore it becomes evident that hybridization is an essential tool that China is using to counter future food insecurity (Li, 2009)

Not surprisingly, the issue of hybridization and genetic manipulation of our food supply is quite polarizing and has drawn great ire and created controversy about the risks and benefits of genetically modified foods. Notoriously referred to as “Frankenfoods," genetically modified foods have become the target of consumer advocacy groups who fear that modifying the genetics of the food we eat will have significant downstream repercussions on our health and on the environment. But food scientists closely involved with rice hybridization argue that unlike other genetic modifications such as soybeans made resistant to glyphosate herbicides or tomatoes which have an enzyme suppressed to retain fruit hardness, rice hybridization serves to yield food that is more nutritious such as the widely-touted Golden Rice that is engineered to make its own vitamin A. With nearly a million children suffering from severe vitamin A deficiency every year and 350,000 going blind because of it, Golden Rice was seen as nothing short of miraculous because it could prevent blindness and even death among the world's poorest (Potrykus, 2001).

### *Why Hybrid Rice?*

Not only are there the obvious health benefits, but hybrid rice has proven to also benefit the economic prosperity of its farmers. Hybrid rice is particularly critical with the demand of rice surging with the recent population boom, especially in developing countries. This is compounded by a dawning reality that more rice has to be cultivated on far less land and with far less input than in the past. Unfortunately, hybrid rice production costs have also increased owning to higher seed, fertilizer, pesticide, and labor costs The hybrid rice varieties that have already been developed have documented a 15–20% higher yield potential than domesticated rice varieties, and they additionally have the ability to perform better under more adverse conditions. This increased yield translates to higher gross incomes for farmers, which on average, has been 15-19% and is a boon for many struggling farmers (Sebastian and Bordey, 2004).

### *What Is Hybrid Rice?*

Hybrid rice is a rice crop that is grown from a cross between two parents that are genetically unlike, to yield progeny that offer greater vigor, yield, etc. over their parents, a phenomenon known as heterosis. Rice in the wild is a self-pollinating crop; therefore, for commercial development, a male sterile line is key to ensure that pollen is unviable to produce seeds through self-pollination and diminish the genetic pureness. A male sterile line is then used as the female parent and grown side by side with a pollen parent to produce the hybrid seed through cross-pollination. When China initially made inroads in hybrid rice technology starting in 1964, it developed the three-line hybrid rice methods. A subsequent two-line hybrid rice was successfully developed in 1995 that has a 5%-10% yield advantage higher than the existing three-line hybrid rice. More recently, a "super rice" research program was initiated in China and India starting in 1996 that strives to develop super high-yielding rice hybrids from crosses involving indica/japonica parents; indica and japonica being the two most common subspecies of *Oryza sativa* in Asia. The indica and temperate japonica subspecies are the most different in their agronomic traits and thus show maximum heterosis (Virmani, 2003). So far, the results from the program have been favorable.

### *Current Situation of Hybrid Rice*

The yield heterosis of hybrid rice combinations is still limited and quite variable from year to year in various planting seasons. In fact, the biggest concern for hybrid rice programs going forward is the long-term maintenance of pure genetic lines, which if compromised, tends to decrease the yield advantage. It seems that this problem is so compelling to food scientists that they fear that it could cause the collapse of any national hybrid rice program. Several reports are already out that indicate that purity in certain lines has already deteriorated and have thus affected the characteristics that these lines were selected for originally (Ikehashi, 2000). The future of hybrid rice appears promising though. While government has predominantly paved the way thus far, the recently held Third National Workshop on Hybrid Rice envisions that the private sector will play a greater role in the commercialization of hybrid rice and that government must work more on establishing a favorable policy environment for hybrid rice (Sebastian and Bordey).

## The Struggle: Quantity and Quality

The idea has often been perpetuated that the purpose of hybrid rice is to increase the yield of crops. And while this is true to some extent, there is another entire camp that argues that new hybrid varieties of rice need to focus as much on quality and taste as it does on yield. In 1964, Yuan Long Ping put forth the idea of utilizing hybridization in China. Hyrbidization of rice has since been a wildly successful initiative in China. Hybrid rice planting area rose dramatically in the first year from 7 ha in 1974 to 373 ha in 1975 all the way up to 139,000 ha in 1976. In 1991, the area of land devoted to hybrid rice cultivation reached 17.6 million ha, making up 54% of total rice area. A more telling quantitative measure during this same time span is the nation's average yield in rice production, which increased from 4.2 tons/ha to 6.7 tons/ha (Yuan, 1994). So the popularity of hybrid rice is clearly substantial and its widespread adoption in the whole of China from Shanghai in the east to Yunnan in the west speaks to its wide adaptability in addition to its high yield.

But the other side to this story is that since the yield of hybrid rice reached new highs in the early 90s, it has stalled for several years, and it appears that our current knowledge and technology as well as our drive to improve upon prior success has plateaued. Without new methods being developed or different genetic resources being discovered, it will be difficult to further increase the yield of hybrid rice. Similarly, the numbers also show that farming adoption of hybrid rice varieties, while initially achieving monumental increases, has reached its peak in China and has since fallen to a level around 15.5 million ha. The main reason seems to be the decrease in the acreage allotted to early cropping of hybrid rice. Since, rice crops can be grown in about 3 months time, farmers tend to grow two cycles of crops per year on the same land, one early crop and one late crop. In a recent study looking at this exact issue, it was noted that only about 20% of early cropping rice area in southern China was used for hybrid rice, whereas over 90% of late cropping rice area is allocated for hybrid rice (Virmani, 2003). The explanation may be attributable to a lack of high yielding combinations with short growth duration, and more notably a commonly held perception among farmers and consumers of poor grain quality in hybrid varieties. And therein lies the crux of the struggle between quality and quantity.

With increasing living standards across China, more has come to be expected from food quality. In contrast to hybrid rice, conventional rice shows better grain quality across all quantifiable measures. Therefore, grain quality is a major limiting factors going forward for increased hybrid rice development and consumer adoption. In a recent survey in 2001, the main weaknesses of hybrid rice were reported as: (1) its quality was subpar compared to conventional rice, (2) its price was lower, and (3) its low milling quality led to more broken grains (Rao and Desai, 2001). In another survey, farmers reported that hybrid rice varieties had an unpleasant smell after cooking and had poorer grain quality, and as a result the selling price was 11% lower than conventional commercial rice varieties (Janaiah and Hossain, 2002). The lower price will be a detriment to the selling power of hybrid rice and will surely reduce its favor among farmers even further, impeding any societal benefits that further enhancements of yield could have.

Having a better understanding of the features that contribute to the overall grain quality of rice will lay the groundwork for creating new breeding strategies for combining high quality with high yield. In Asia where rice consumption is particularly high, rice quality traits such as physical appearance and cooking properties dictate market value and play a pivotal role in the consumer acceptance of new varieties (Chávez-Murillo et al., 2011). Other traits such as shape, grain length, and whiteness also play a role, but are more region and culture dependent, and consequently much more difficult to homogenize. Our current knowledge of factors is incomplete but with recent developments in genomics and the elucidation of the rice genome, we can enhance our understanding of the pathways that determine individual quality traits and pave the way toward targeted grain improvement and ultimately the creation of a more appealing rice grain. This is a necessity in order to meet the growing global demand for high quality rice, and to keep the door open for future improvements in yield that can only come through hybrid rice. Therefore, how we balance developing rice hybrids with both high yield and good grain quality will be the major challenge for food scientists in the coming years.

# RESEARCH

## Impact of allelic diversity of *Wx* and *ALK* genes on the nutritional characteristics of rice and the evaluation of rice starch biosynthesis in the hybrid Y58S male sterile breeding line

### 1. Introduction

Rice *(Oryza sativa)* is a critical cereal crop that is by far the most economically important food crop in many developing countries, as well as a major crop in many developed countries where its consumption has increased significantly in recent years. Now, rice provides the staple food for more than half of the world's population (Van Tran, 2010). It provides as much as 80% of the daily energy intake in some Asian countries and is the single most important source of protein in the world because of the quantity consumed (Hill and Hardy, 1999). It has therefore become essential to the meet the demands of the world's increasing population by maximizing rice productivity. At present, many leading rice-producing countries, including China and India, still have large disparities between their potential output and their present yield.

Since the first successful application of hybrid rice technology in 1974, hybrid grain yield has greatly increased and now accounts for over 55% of the annual rice-planting area in China (Yang et al., 2007). But while hybrid technology of rice has been growing rapidly because of its higher yield and good adaptability, grain quality of hybrid rice remains to be improved in China. Much of the problem stems from the poor cooking and eating quality of many widely grown varieties, particularly the *indica* subspecies of *Oryza sativa*, which predominates Southern China. Therefore, grain quality has now become a primary consideration for farmers and rice breeders alike. Characteristics of grain quality dictate market value and have a pivotal role in the adoption of new hybrid varieties (Champagne et al. 1999). In general, quality traits encompass physical appearance, cooking and sensory properties, and more recently, nutritional value. The traits most obvious to rice consumers in defining cooking and sensory quality include cooking time, texture of cooked rice, aroma and fragrance retention after cooking, and softness of the cooked rice (Fitzgerald et al., 2009).

The nutritional quality of rice is collectively as important as cooking and eating quality, and correlations do exist between certain nutritional traits and certain characteristics that determine the essential cooking and processing characteristics of rice such as protein content and amylose content. The nutritional value of rice is usually determined by its protein content, but could also be measured in terms of amylose content, which is correlated with two important areas of nutritional interest: glycemic index and resistant starch content (Hill and Hardy). Glycemic index is a means of classifying carbohydrates according to the rate at which they raise the blood-sugar level, with low levels proving to be beneficial to human health through the control of blood sugar, lipid levels, and weight reduction. Resistant starch, likewise promotes health, and in particular gastrointestinal health by reducing the risk of bowel cancer and increasing the bowel's advantageous microflora (Jenkins et al., 1998).

Our current knowledge of the individual elements that factor into grain quality is incomplete, and this gap in knowledge is made apparent by the persistence of benchmark varieties in China in spite of the yield gains in hybrid varieties. Grain quality is a complex multifactorial characteristic because of the various environmental elements, such as temperature, water supply, and soil fertility, which all play an important role in quality determination. Knowledge amassed in the past decades indicate that the poor cooking and eating quality is directly related to three attributes of the physicochemical characteristics of starch in the endosperm – namely, amylose content (AC) (Juliano, 1985), gel consistency (GC) (Cagampang et al., 1973), and gelatinization temperature (GT) (Little et al., 1958).

Rice quality is traditionally benchmarked on amylose content, which is determined in part by the waxy (Wx) gene which codes for the granule bound starch synthase (GBSS) enzyme (Sano, 1984; Webb, 1991; Shure et al., 1983). In rice, numerous functional alleles of the *Wx* gene have been found to exist that correspond to specific AC levels. A molecular analysis by Dr. Zy Wang revealed that the splicing pattern of the first intron of the *Wx* gene is highly correlated with AC level. Transcripts with the intron spliced out would produce high AC, while those with the intron unspliced would produce grains with no amylose (Wang et al., 1995)

Besides amylose content, gelatinization temperature (GT) is a key factor for evaluating the cooking and eating quality of rice. It marks the critical temperature at which the starch granules start to lose their crystalline structure. Water and time required for rice cooking are highly correlated to GT, which means that rice varieties with low GT need less water and shorter cooking time than those with high GT (Gao et al., 2003). The inheritance of the genes affecting GT has been widely studied and is thought to be regulated by a major gene, *ALK,* which encodes the soluble starch synthase II (Gao et al., 2011).

At present, there is an outstanding problem in breeding for rice eating quality in China. Many recently released hybrid rice varieties, male sterile lines, or restorers of hybrid rice have low apparent amylase content (AC) and fail to meet the demand of rice market. Previous studies have shown that male sterile lines and restorer lines had a great effect on the hybrid rice quality and that amylose content depends heavily on the male sterile lines (Dai et al., 2006). Great attention must be paid to the improvement of starch characteristics in hybrid rice breeding. AC is mainly controlled by the *Wx* gene. GC is also controlled by the *Wx* gene or by one locus tightly linked to this gene. GT is not significantly related to AC and the *Wx* genotype, but is instead controlled by the *ALK* locus (Tian et al., 2009). By using the $Wx_{in}$ gene molecular marker and assisted selection, the objective of the present study will attempt to successfully up-regulate the amylose content of the hybrid rice male sterile line, Y58S, to improve rice's cooking and eating quality, and to promote greater adoption of this hybrid variety. The second objective of the present investigation is to examine allelic diversification at the *Wx* and *ALK* locus that affects ECQ (eating and cooking quality) traits in Asian rice and to clarify the still unresolved question of which of the many major and minor genes control grain ECQs.

The overall aim of the proposed research is to improve understanding of the molecular basis of rice starch quality and improved hybrid rice quality. Accordingly, we conduct this research to: (1) Estimate the effects of two major starch-synthesis genes *Wx* and *ALK* on the AC, GC, GT of several hybrid rice and conventional rice varieties; (2) Determine that the $Wx_{in}$ gene has been introgressed into the Y58S hybrid rice male-sterile by using the developed molecular marker; and (3) Determine if the AC is up-regulated for improved cooking and eating quality in the male sterile line Y58S through physical-chemical quality assays of backcrossed progeny.

## 2. Methods and Materials

### 2.1. Plant materials

Y58S is the predominant male sterile line that occupies the largest number of acres in China. This cultivar is predominantly used to establish hybrid lines and is notable for low amylose content (AC), intermediate gel consistency (GC) and gelatinization temperature (GT), Basmati 370 is a commercially important rice cultivar found predominantly in South Asia. It was originally cultivated in the foothills of the Himalayas. Y58S and Basmati 370 are quite distinct in their agronomic characteristics, yield, and morphological features. For other experiments these plant varieties were used: Nanjing 11, Starbonnet 99, Fresco, Brazilian upland rice, n-300, American glabrous rice, Nipponbare, Guanglingxiangnuo, hua 7, Guihuahuang, Y58S, 9311, Zhenshan 97, Teqing, Peiai 64S, Basmati 370, IR64, and P88S. These represent a variety of rice breeding lines cultivated in China.

### 2.2. Introgression of $Wx_{in}$ conferring intermediate amylose from Basmati 370 into Y58S.

In order to improve the AC of a key male sterile line, Y58S, a rice variety with $Wx_{in}$ gene, Basmati 370, was backcrossed with a key male sterile line. As shown in Fig. 1, the Basmati 370 rice strain was crossed with the Y58S, using Y58S as the female parent and Basmati as the male parent, to develop the F1 generation. The F1 plants were backcrossed to Y58S using mixed pollen to generate the BC1F1 generation. Consecutive backcrossing was conducted until the BC3F1 generation was produced. Marker assisted selection (MAS) was carried out using 50 individuals in each generation of backcrossing. The plants that carried heterozygous alleles of certain markers were selected and crossed with Y58S. The selected plants in BC3F1 were self-pollinated to generate the BC3F2 population. Twelve BC3F2 lines possessing homozygous Basmati 370 alleles were successfully developed. SSR and SNP markers will be used for the genotyping. Both phenotypic selection and AS-PCR and PCR-Acc I marker assistant selection were conducted in segregated population to determine whether the plant has elite agronomic traits and $Wx_{in}$ genotype. In this study, 40 BC3F3 breeding lines of the key two-line male sterile line/the rice variety with $Wx_{in}$ gene are used. 10 varieties (or lines) are used to study on the genetic relationship among *Wx* gene, *ALK* gene, AC, GC and GT.

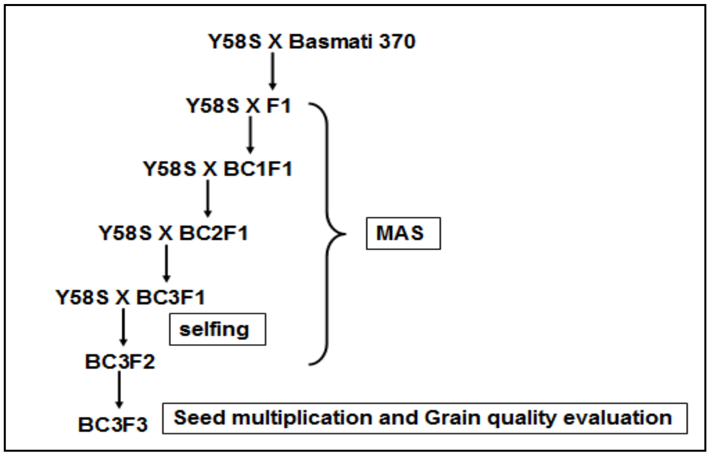


Fig. 1. Schematic representation of gene introgression of $Wx^{in}$ into Y58S using marker assisted selection and backcross breeding.

## 2.3. DNA marker analysis

DNA extraction of the seedling of several hybrid progeny and their parents was isolated from 0.5g of leaf tissue according to the DNA trap method (Xia et al., 2011). The PCR reaction was performed according to specifications outlined in *Composition and functional properties of rice* (Xiao et al., 1996). *Wx* genotype is tested by AS-PCR marker and PCR-Acc I marker (Qaioquan et al., 2006). *Alk* genotype is tested by CAPS marker (Gao et al., 2003). (CAPS marker modification, S020-F: 5'-GGTTCTCGGTGAAGATGGC-3', S020-R: 5'-GTGGTCCCAGCTGAGGTCC-3', $SS\ II\text{-}3^{a}$, $SS\ II\text{-}3^{C}$:Ban Ⅱ(cut site) 326 = 284 + 42; $SS\ II\text{-}3^{b}$: Ban Ⅱ (no cut site) 326). SSR markers used to genetic background analysis.

## 2.4. Grain quality evaluation

The physical-chemical properties of grain quality were measured according to the protocols provided by Hybrid Rice Molecular Breeding Laboratory. The AC, GC, and GT in mature seeds are measured by the colorimetric method with iodine-potassium iodide, as described previously by Juliano, the length of the gel according to the method of Cagampang, and the alkali spreading value (ASV) as described by Little (Little et al., 1958). Two duplicates were performed, and the mean value was taken as the final result.

*AC (Tian et al., 2005)*

The formation of a helical complex between amylose and iodine gives rise to the typical deep blue color of starch dispersions stained with iodine and forms the basis for quantitative determination of amylose content. The formation of these complexes is determined by colorimetry. One hundred grams of rice grains were de-hulled and milled using a miller manufactured by the Jiading Food and Oil Machinery Factory, Shanghai, China, according to the National Standards NY 147–88. The milled rice was ground into powder with a Udy Cyclone Sample Mill (Udy Corporation, Colorado, USA), and was then sieved through a 0.5mm thick 100-mesh sieve into a 100 ml flask. Exactly 25 mg rice flour was gelatinized overnight in 1 ml of 1.0 N NaOH and 1 ml 95% ethanol in a water bath set at 50°C. The solution was boiled in the water bath for 10 min and then cooled to room temperature. The cooled solution was extracted three times with 1 ml I N acetic acid and then 1.5 ml iodide solution to remove the lipid. The AC was determined with an ART-3 Automatic Titrator according to the manufacturer's instruction (Hirama Laboratories, Japan) at 720 nm. Standard amylose solutions (2.8%, 12.8% and 26%) were prepared as checks by dissolving pure amylose and amylopectin in distilled water. Amylose content of the sample was determined in reference to the standard curve and expressed on a percent basis. Based on the amylose percentage, the varieties can be grouped as per criteria seen in Table 1.

| **Category** | **Amylose Content (%)** |
|---|---|
| Waxy | 1-2 |
| Very low amylose | 2-9 |
| Low | 10-20 |
| Intermediate | 20-25 |
| High | 25-30 |

Table 1. Amylose percentage classification table used to categorize the AC of rice progeny and the parental lines.

*GC (Cagampang et al. 1973)*

The GC was measured according to the method of Cagampang (Cagampang et al. 1973). Briefly, 100 mg rice flour was weighed in 11 x 100 mm culture tubes, to which 0.2 ml of 95% ethanol containing 0.025% thyml blue was added to prevent clumping of the powder during gelatinization. Two ml of 0.2 N KOH were added and dispersed through the mixture. The tubes were covered with glass marbles and boiled vigorously in a water bath for 8 min. The samples were removed and placed at room temperature for 5 minutes; the tubes were put on ice for 20 minutes, and then laid down horizontally over a ruled paper, graduated in

millimeters. The length of gel migration from the bottom of the test tube was measured after 60 minutes. The gel length was measured 1 hour later as the distance from the bottom of the tube to the front of the gel migration. The gel length obtained provides a measurement of gel consistency as per Table 2.

| **Category** | **Consistency (mm)** |
|---|---|
| Soft | ≧61 |
| Medium | 41-60 |
| Hard | ≦40 |

Table 2. Gel consistency classification table used based on gel length obtained

*GT (Little et al. 1958)*

The GT was measured on the basis of individual grains expressed as the alkali spread value (ASV) using the method of Little, with minor modifications (Little et al. 1958). Six whole milled rice grains were taken in Petri plates and 10 ml of 1.7% of KOH was added. The grains were carefully separated from each other using a forceps and incubated at room temperature for 23 hours to allow spreading of the grains. Then the alkali spreading value was calculated as low, intermediate, or high. Each grain was visually examined for its level of intactness and assigned a numerical score according to Table 3.

| Score | Level of intactness |
|---|---|
| 1 | not affected kernel |
| 2 | swollen kernel |
| 3 | swollen kernel, with incomplete or narrow collar |
| 4 | swollen kernel, with complete and wide collar |
| 5 | split or segmented kernel, with complete and wide collar |
| 6 | dispersed kernel, with merging collar |
| 7 | completely dispersed and intermingled kernel |

Table 3. Above: Classification table to determine level of intactness. Below: Gelatinization temperature classification based on ASV score.

| Classification | ASV | Gelatinization temperature |
|---|---|---|
| High | 1-3 | > 74 |
| Intermediate | 4-5 | 70-74 |
| Low | 6-7 | < 70 |

2.9. Analysis of data

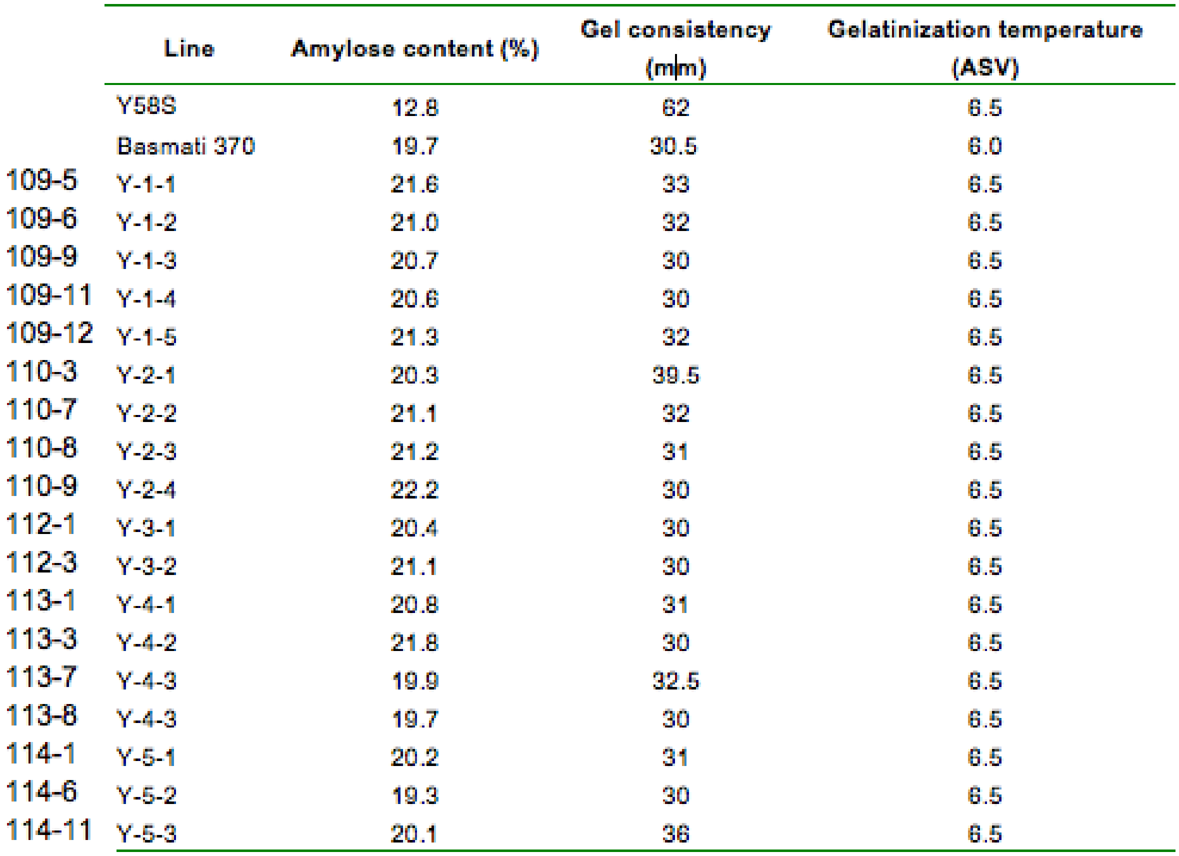
One-way analysis of variance of the quality traits AC, GC, and GT across all varieties and the BC3F3 progeny was conducted using SPSS computer statistical software.

## 3. Results

### 3.1. Grain quality

Three quality traits of eighteen improved lines from the BC3F3 generation, the original Y58S and the donor parent, Basmati 370, were measured using seeds harvested from paddy fields in Changsha, Hunan Province, China. The results of grain quality are presented in Table 4. The effects of starch synthesis-related genes (SSRGs) were tested against each ECQ property (i.e., AC, GC, or GT) across the panel using a mixed model method. Nutritional characteristics of glycemic index and resistant starch, both of which are health benefits, were evaluated by determining amylose content percentage using numerical scores delineated into evaluative categories (see Table 1). The two parents differed significantly in their amylose content percentage; Basmati 340 was 12.8%, whereas Y58S was measured at 19.7%. Although there is a measureable difference, both parents are subjectively classified in the low category. The sampling of the progeny generation showed that a majority, instead had an intermediate AC, which varied from 19.3% (Y-5-2) to 22.4% (Y-2-2) (See Table 4). The average AC in the BC3F3 progeny generation was 20.7 +/- 0.8, statistically different ($p = 8.7 \times 10^{-20}$) from the Y583 parent. The mean is also greater than the AC of the Basmati 370 parent; although not significant ($p = 24.3$), it represented an increasing trend. This suggests a synergistic effect among several minor genes and the role of several modifiers that boosted AC in the progeny beyond what the allelic makeup of the parents would suggest based on our current theory of *Wx* being the singular locus for altering amylose content.

Gel consistency of Basmati 370 and Y58S was 30.5 mm and 62 mm, respectively (See Table 4). In all progeny, the hard gel consistency (GC) value (< 40 mm) was not significantly different among the progeny and the Basmati 370 parent strongly suggesting allelic transfer of the progeny with only the Basmati 370 gene variant. Both Basmati 370 and Y58S parental lines also had similar alkali spreading values (ASV) of 6.0 and 6.5, respectively (See Table 4). The mean ASV of BC3F3 progeny was 6.5 +/- 0.0 ASV, matching the Y58S parent's recorded ASV and validating the effort to backcross the progeny to singularly isolate the allelic variant of the *ALK* gene that expresses a phenotype that matches the desired low gelatinization temperature that was inherent in the original Y58S parent. Introduction of increased amylose content into the germplasm of an the Y58S male sterile breeding line now makes this agronomically important trait available for conventional rice hybrid breeding programs, while maintaining the desired ASV and gel consistency. In all 40 BC3F3 lines, AC was improved to intermediate level and GC was made harder. This addresses the problem that has plagued Chinese hybrid rice adoption by substantiating the more culturally accepted form of hybridization through natural backcrossing crossing as means of isolating and selecting the desired alleles into the Y58S male sterile line. Natural backcrossing avoids any highly culturally stigmatized transgenic methods to improve rice quality and improve the cooking and eating qualities of rice to promote its adoption among rural Chinese.



| | Line | Amylose content (%) | Gel consistency (mm) | Gelatinization temperature (ASV) |
|---|---|---|---|---|
| | Y58S | 12.8 | 62 | 6.5 |
| | Basmati 370 | 19.7 | 30.5 | 6.0 |
| 109-5 | Y-1-1 | 21.6 | 33 | 6.5 |
| 109-6 | Y-1-2 | 21.0 | 32 | 6.5 |
| 109-9 | Y-1-3 | 20.7 | 30 | 6.5 |
| 109-11 | Y-1-4 | 20.6 | 30 | 6.5 |
| 109-12 | Y-1-5 | 21.3 | 32 | 6.5 |
| 110-3 | Y-2-1 | 20.3 | 39.5 | 6.5 |
| 110-7 | Y-2-2 | 21.1 | 32 | 6.5 |
| 110-8 | Y-2-3 | 21.2 | 31 | 6.5 |
| 110-9 | Y-2-4 | 22.2 | 30 | 6.5 |
| 112-1 | Y-3-1 | 20.4 | 30 | 6.5 |
| 112-3 | Y-3-2 | 21.1 | 30 | 6.5 |
| 113-1 | Y-4-1 | 20.8 | 31 | 6.5 |
| 113-3 | Y-4-2 | 21.8 | 30 | 6.5 |
| 113-7 | Y-4-3 | 19.9 | 32.5 | 6.5 |
| 113-8 | Y-4-3 | 19.7 | 30 | 6.5 |
| 114-1 | Y-5-1 | 20.2 | 31 | 6.5 |
| 114-6 | Y-5-2 | 19.3 | 30 | 6.5 |
| 114-11 | Y-5-3 | 20.1 | 36 | 6.5 |

Table 4. Summary of the 18 progeny sampled from the BC3F3 generation. The amylose content of the progeny was increased compared to the parent lines. Gelatinization temperature in the progeny was equal to the Y85S line. Gel consistency more closely represented the Basmati 370 line.

### 3.2 *Waxy* gene allelic association with ECQ properties in Diverse Rice Varieties

To examine continuous variation of amylose levels in *Oryza sativa* landraces, the four *waxy* gene putative alleles ($Wx_a$, $Wx_{in}$, $Wx_b$, and *wx)* were investigated in their native lines. Apparent amylose levels ranged from 1.7% to 23.3% in the various lines, showing a positive relationship between AC content and allele type, with AC trending according to the alleles as such: $Wx_a > Wx_{in} > Wx_b > wx$. The average value of AC in $Wx_a$-type varieties was about 22.4%, about 8% greater than that of $Wx_b$ varieties (14.5%), while the $Wx_{in}$-type varieties (19.0%), lies in-between the values of $Wx_a$ and $Wx_b$ varieties (See Supplemental Fig. 1). In the 18 varieties, the lines with $Wx_a$ gene have higher AC and harder GC, and the materials with $Wx_{in}$ gene have intermediate AC, while the materials with $Wx_b$ gene had lower AC and softer GC (See Supplemental Fig. 2). The *wx* allele, studied only in the Guanglinxiangnuo subspecies exhibited aberrantly low amylose content and elevated GT. When correlation was conducted collectively across the panel, it revealed highly correlated ECQs: AC is negatively correlated with GC (r = −0.94) and GT value (r = −0.68), whereas GC is positively correlated with the GT value (r = 0.33). With this negative correlated association between AC and GC, it would appear that *Wx* locus may function as the major gene for both amylose content and gel consistency, and also as a minor gene affecting gelatinization temperature.

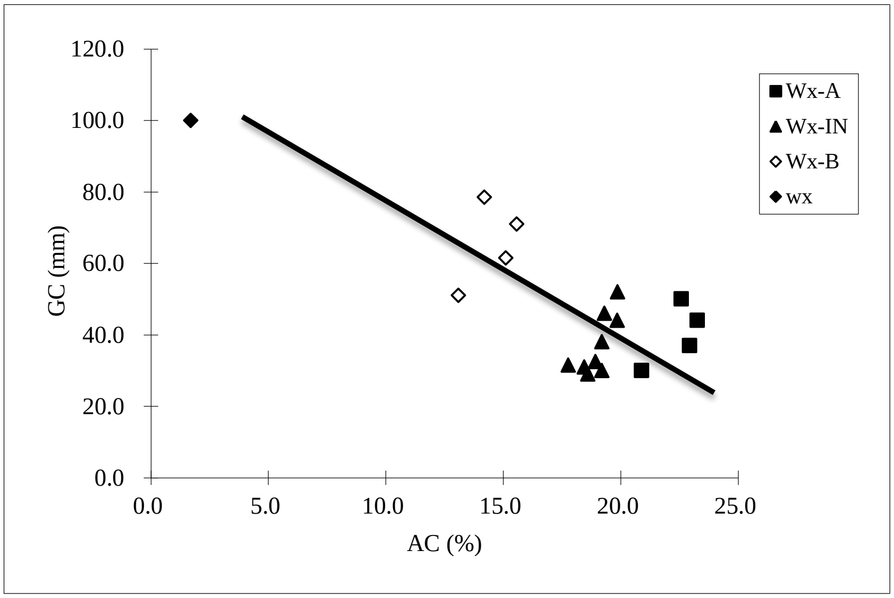
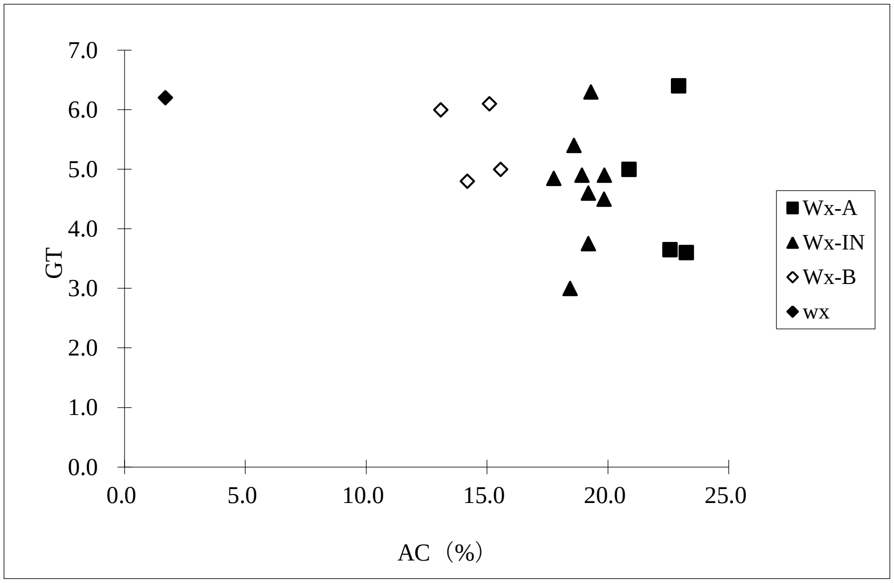


Figure 2. Above: Plot of amylose content against gel consistency in 18 different rice lines with varying *Wx* alleles. AC is negatively correlated with GC (r = −0.94) Below: Plot of amylose content against gelatinization temperature estimated with ASV in 18 different rice lines with varying *Wx* alleles. AC is negatively correlated



with GT value (r = −0.68),

### 3.3 Allelic diversities of *ALK* and ECQ effects

To examine the properties of the allelic variations in *ALK,* the three presumed alleles (*ALK-I*, *ALK-II*, and *ALK-III)* were investigated in their native lines. For the properties of GT, *ALK-II,* has a significant p-value suggesting that this allele meaningfully increases GT (Figure 3). The effect of the different *ALK* alleles was not as striking when looking only at its direct role on GC and GT (Supplementary Figure 2). When plotted against GT, correlation studies showed a moderately negative relationship with AC, confirming what we had seen previously in evaluating the effect of the *waxy* allele (Figure 3).

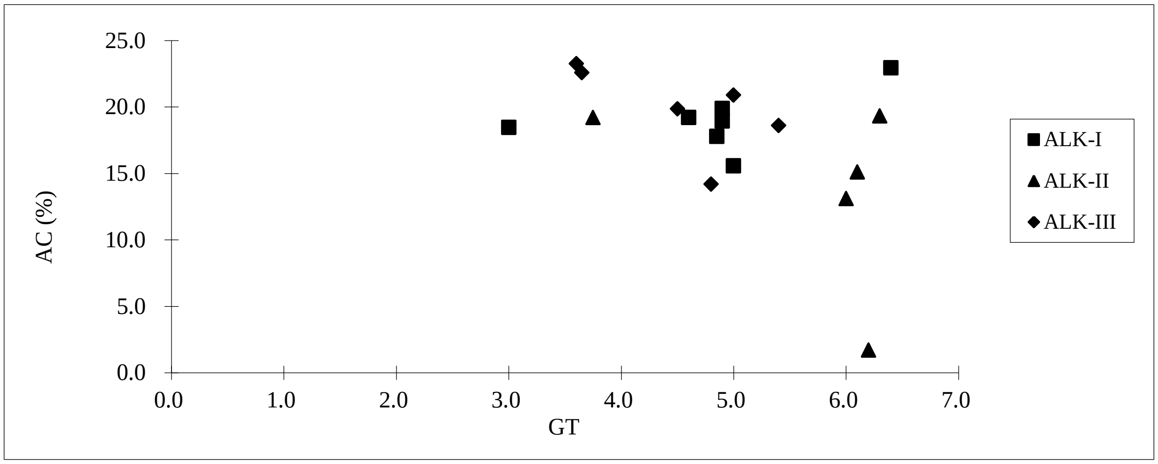
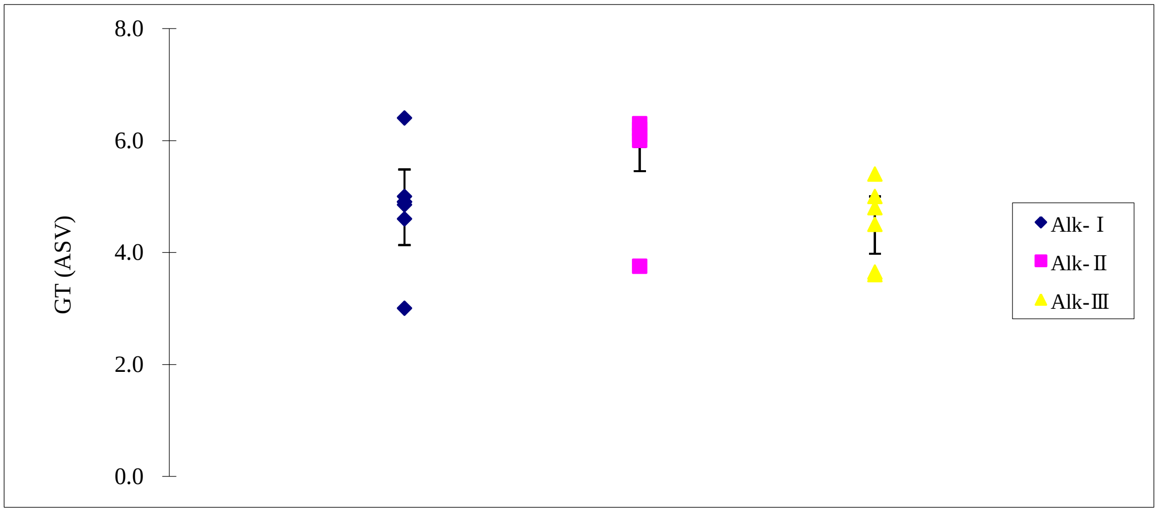


Figure 3. Above: Plot of amylose content against gelatinization temperature in 18 different rice lines with varying *ALK* alleles. AC is moderately negatively correlated with GC (r = −0.52) Below: Comparison of gelatinization temperature estimated with ASV in three different *ALK* alleles in 18 native rice lines.



## 4. Discussion

### 4.1 Elucidation of starch synthesis gene alleles in regulating rice quality

Over the past few decades, scientists have extensively evaluated and experimented with properties of rice grain quality and nutrition, but clear data wasn't always available. Today, the picture is rapidly evolving and we are discovering more about how certain *starch* synthesis-related *genes* work at different synthesis steps during starch development to affect grain quality. Researchers have long recognized that amylose content is a major determinant for cooking and eating quality. Cooked rice with high AC is dry and fluffy and has significant health benefits of lowering post-prandial blood sugar, while rice with low AC is moist, tender, and sells better, but has a high glycemic index to quickly raise post-prandial blood sugar (Juliano, 1985; Denardin et al., 2007). Generally speaking, the AC of commercialized varieties varies from 14% to 25% depending on the subspecies of rice. Our study provides strong evidences that *Wx* not only affects AC, but that it also regulates GC as a major gene and GT as a minor one.

In addition, we have also shown that other minor genes additively affect AC in particular, and possibly also GT and GC, and that all these associated genes form a fine complex network controlling ECQs of rice grains. Even more revealing that our picture is incomplete are recent findings through hereditary studies concerning the *Wx* gene that currently suggest the basis for 16 kinds of *Wx* alleles based on polymorphism analysis (Wan et al., 2007). The correlations between grain quality properties, as demonstrated in the current study, further complicate the process of elucidating the exact genetic features that modify ECQs.

Moreover, our results also demonstrated that *ALK* alleles are important in altering GT, but may also act on AC and GC. This is especially relevant to nutrition since intermediate and high GT rice offer more resistant starch (Eggum et al., 1993) and lower glycemic index (Panlasigui et al., 1991). Although our study was limited in its ability to precisely interpret the extent to which genes influenced quality traits, primarily because we sought to study allelic variations in natural lines and thus without near-isogenic lines (NILs), our analysis can be confounded by minor genes; secondly, our sample size, though varied, was restricted in size. Our conclusions, however, are confirmed by studies that have shown that when *Wx* alleles were uniform across lines, *ALK-I* contributed to higher AC, whereas *ALK-II* led to lower AC (Tian et al., 2009). This would mean that varieties with the *ALK-I* allele would show higher AC and GT values, and varieties with *ALK-II* allele would have lower AC and GT values. This is only resolved with our findings of a negative correlation between AC and GT values because of haplotype findings that found that predominant clustering of haplotype combination that resulted in such phenotypes. The findings described herein should implicate methods of transgenic modification that could vastly improve nutrition and grain quality in our hybrid rice varieties by selecting for those alleles that work in combination to give us the desired traits.

### 4.2 Enhanced male sterile lines for improved nutrition and grain quality in hybridized rice

Y58S, developed by crossing Annong S-1 with Changfei 22B and Lemont and Pei'ai 64S is a new male sterile line with wide ecological adaptability to hybridization. Because it has characteristics of good plant type, low critical sterility-inducing temperature, good flowering habits, strong stress and disease resistances, and fine grain quality, it is favorable for safe hybrid seed production and high-yielding rice lines in China (Deng, 2005). In terms of quality, Y58S is a non-fragrant cultivar, with low AC, intermediate GC, and intermediate GT, that occupies the largest number of acres in China. At present, there is an outstanding problem in breeding for rice eating quality in China. Great attention must be paid to the improvement of starch characteristics in hybrid rice breeding. Backcrosses were made using a Basmati 370 rice variety with *Wx*$_{in}$ gene as the donor parent, and the elite *Wx* gene was introgressed from donor parent through backcross with marker-assisted selection. After successive backcrosses, progeny were developed with improved AC, showing herein that through gene molecular marker and assisted selection, rice breeders can successfully up-regulate the AC of hybrid rice. In theory, this improvement should boost demand for Y58S derived rice lines, increase use of hybridized rice crops, and serve as an economic boon to farmers who can sell higher quality rice at a great premium. Thus, with the development of biotechnology and linkage maps in rice, and with the roles of SSRGs being elucidated, the long-term prospects of creating the ideal rice grain through the combination of both molecular and conventional approaches is promising.

## 5. Conclusions and Future Work

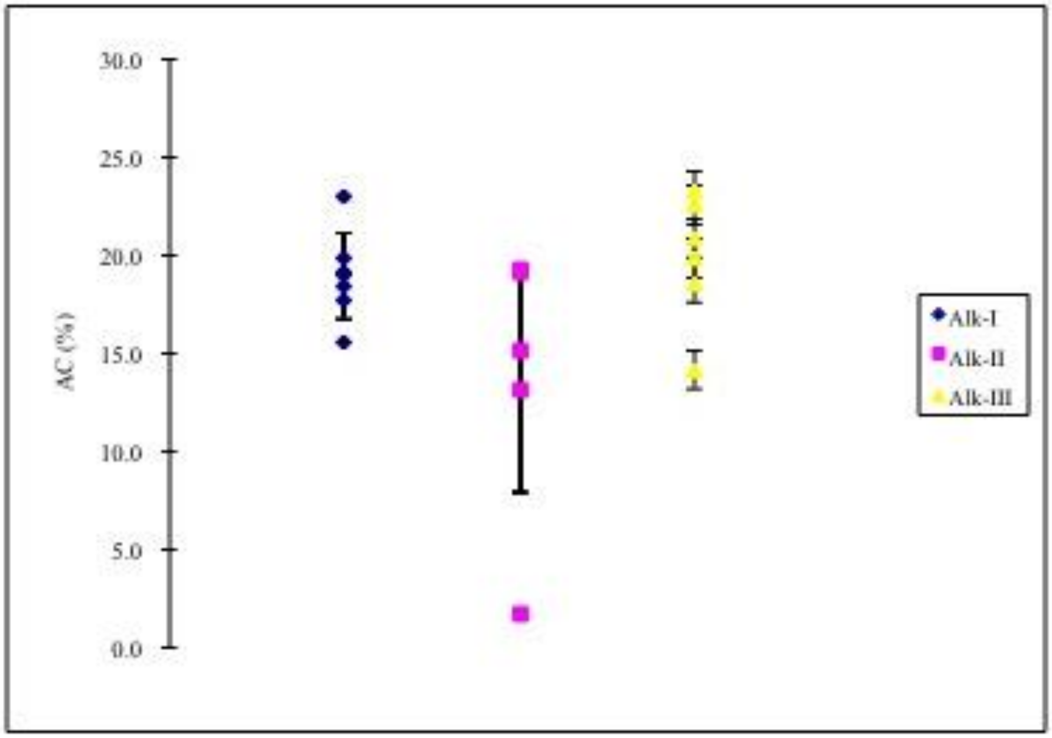
The outcome of this study shows that there were negative correlations between AC and GC, and both AC and GC were closely related to *Wx* genotype, which confirm that the AC of rice grain is controlled mainly by Wx gene. It also implicates a major role for *ALK* alleles in altering GT, but also a minor role on AC and GC. This work also addresses the feasibility of successfully up-regulating the AC of hybrid rice through conventional breeding methods as opposed to transgenic means. It suggests that we now have a better understanding of some of the principal processes and genetic traits that impact ECQs. Moreover, we now have advanced technology to such a degree through linkages maps, molecular marker assisted selection, etc. It is now more feasible that ever to modify grain quality on a genetic level.

The conclusions drawn from our study was limited in sample size and the extent to which natural breeding lines was confounded by our use of natural breeding lines, but the validity of our conclusions is strongly supported by our own results as well as by outside studies that have affirmed through tests with near-isogenic lines that the trends that we observed in our data were in fact widely supported. The findings described prove that we could vastly improve nutrition and grain quality in our hybrid rice varieties by selecting for those desired alleles. To refine our current conclusions we would need to continue to deduce the contributions of major starch-synthesis genes to these three traits in order to further understand the genetic basis of rice eating and cooking quality. With newly released rice genome sequence information, the expanding data on metabolic pathways, genotype and phenotype associations, and gene regulatory networks, we can perform more tailored experiments. Looking ahead to the future, I would work toward using techniques of genetic engineering or molecular marker-assisted selection to create lines that would utilize microRNAs that will effectively turn off and on genes in the process, so that I could systematically approach the question of which genes truly play a role in advancing grain quality. Using this approach in combination with systems biology techniques (i.e. microarrays) I can tackle many challenging questions. For instance, how many major and minor genes control grain ECQs? What is the relationship among the genes that control AC, GC, and/or GT? What is the limit to our ability to breed elite varieties with desired ECQ properties by genetic manipulation of AC, GC, and/or GT.

## 6. Appendix

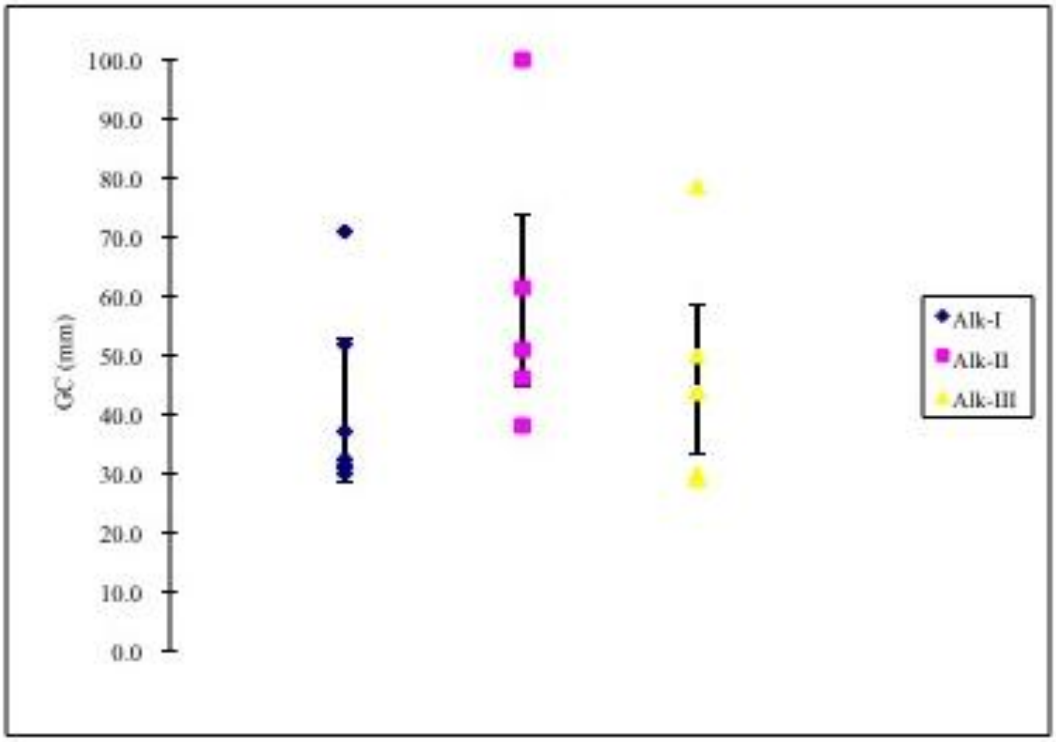
|  | AC(%) | GC(mm) | GT (ASV) |  |
|---|---|---|---|---|
| **Zhenshan 97** | 20.9 | 30.0 | 5.0 | Wxa |
| **Teqing** | 22.6 | 50.0 | 3.7 | Wxa |
| **nanjing 11** | 22.9 | 37.0 | 6.4 | Wxa |
| **Peiai 64S** | 23.3 | 44.0 | 3.6 | Wxa |
| **Basmati 370** | 18.6 | 29.0 | 5.4 | Wxin |
| **hua 7** | 19.2 | 38.0 | 3.8 | Wxin |
| **Guihuahuang** | 19.3 | 46.0 | 6.3 | Wxin |
| **starbonnet 99** | 19.2 | 30.0 | 4.6 | Wxin |
| **Fresco** | 18.5 | 31.0 | 3.0 | Wxin |
| **Brazilian upland rice** | 19.9 | 52.0 | 4.9 | Wxin |
| **n-300** | 17.8 | 31.5 | 4.9 | Wxin |
| **American glabrous rice** | 18.9 | 32.5 | 4.9 | Wxin |
| **IR64** | 19.9 | 44.0 | 4.5 | Wxin |
| **Nipponbare** | 15.6 | 71.0 | 5.0 | Wxb |
| **Y58S** | 13.1 | 51.0 | 6.0 | Wxb |
| **P88S** | 14.2 | 78.5 | 4.8 | wxb |
| **9311** | 15.1 | 61.5 | 6.1 | wxb |
| **Guanglinxiangnuo** | 1.7 | 100.0 | 6.2 | wx |

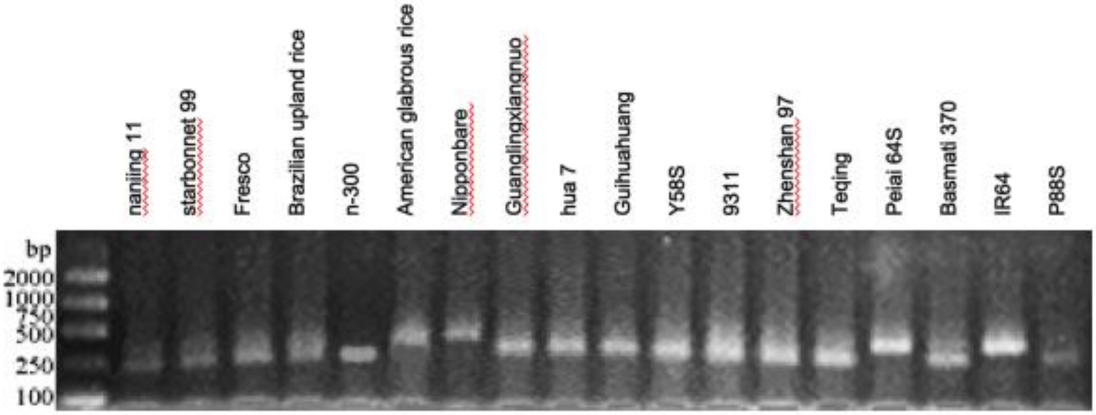
Supplementary Fig.1. Summary of the 18 native rice lines that were assessed for ECQs of AC, GT, and GC. These lines were distinguished based on their allele at the *Wx* locus



Supplementary Fig. 2. Above: Comparison of amylose content in 18 native rice lines each with one of three unique *ALK* alleles

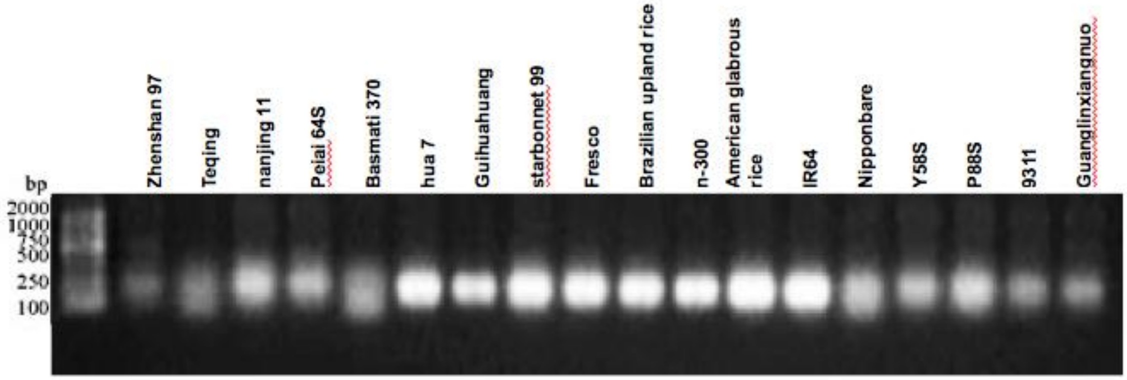
Below: Comparison of gel consistency in 18 native rice lines each with one of three unique *ALK* alleles





Supplementary Fig. 3. Above: Gel electrophoresis of PCR products to identify various *Wx* in 18 rice lines

Below: Gel electrophoresis of PCR products to identify various *ALK* in 18 rice lines



## Analysis: Socioeconomic Effects

Rice has a tremendous economic and sustaining role in society. Worldwide, only 5-6% of rice made in rice-growing countries is exported as a result of high domestic consumption in these countries. For instance, Japan only needs to import about 8% of their rice per year (Pinstrup-Anderson, 2002). Consequently, these nations are particularly susceptible to natural disasters and catastrophes such as crop failures, increments weather, disease outbreaks, and pest invasions.

Before World War II, records on global rice production were sparse. The United States Department of Agriculture first collected agriculture statistics in the 1930s and by the early 1950s the Food and Agriculture Organization of the United Nations (FAO) started keeping track of their own data. Today, the International Rice Research Institute (IRRI) publishes a comprehensive report periodically on aspects of import and export, production, price trends, etc. This increased awareness has come to reflect the increasing economic importance that rice plays in our world today. With the current world's population expected to reach 8 billion by 2030, the severe situation of population growth pressure becomes obvious. The IRRI estimates that rice production must increase nearly 60% by the year 2020 or come to face steep shortages and skyrocketing grain prices. Increases will happen primarily in in rice consuming areas such as Asia, Africa, Central America, and the Middle East. But according to recent projections, total rice production is projected to increase at a slower pace than demand since total crop area is estimated to decrease slightly over the next 10 years (FAPRI, 2009).

During the first half of the last century, production growth was brought on by the increase in farming wetland and the expansion of irrigation systems and the use of nitrogen fertilizer. In the late 1960s, with the advent of the “Green Revolution,” improvement came through higher grain yield. Rice production especially in Asia consequently increased from 240 million metric tons in the mid-1960s to 474 million tons in the late 80s and early 90s (IRRI). Despite these steep increases in the 1960s, however, two important problems still exist: (1) food production per capita has advanced slightly ahead of the growth of the population and, (2) the cost of farming equipment, fertilizers, labor and chemicals has continued to increase whereas the wholesale price of rice has slumped and in some cases has dipped below the 1960s level.

The forecasted deficit in rice is particularly troublesome in terms of a food security standpoint, especially in nations where undernourishment is prevalent and rice remains the predominant caloric source. Achieving food security will require guaranteeing access and affordability to safe and nutritious food. Furthermore, since both the farmers and the consumers of rice tend to be quite poor, there are countering pressures to keep the price high from farmers and to keep the price low from consumers. Developing the technology requires significant investment in a seed program by government or private entities. At the farm level though, this technology might represent financial constraints especially for small farmers in the poorest countries due to higher seed cost. The real price of rice is forecasted to increase significantly over the next few years, doubling by the end of the next decade. Adopting hybrid rice will work to only slightly decrease prices. Prices are estimated to double the current price in India and China, and almost double in other countries in South East Asia. Consumers will thus have to spend twice as much of their income as they do currently to attain the same amount of sustaining rice. Assuming that gross domestic product of these countries continues, though, rice should theoretically become more affordable, however, the gap between the income classes is expected to widen meaning that the population that relies the most on rice will perceive lower income gains and the increase in rice price will make it less affordable for this segment of the population throughout the world. Cheap input and water is also an obstacle. The fates of rice and water are inseparable. Because rice requires a large amount of ground water, the cost of water can lead to radical changes in rice cultivation. Water, like oil, is a depleting resource critical for our future that will experience tremendous increases in demand, but unlike oil, water is irreplaceable. The outlook appears troubling and points to shrinking rice supplies, and significant price increases that will likely worsen the food security situation in Asia and around the world (Harrison, 2002).

The only foreseeable way to solve the crisis seems to be through advances in science and technology. The adoption of hybrid rice has the potential to change production, trade, and availability of this commodity. One possible method of greatly enhancing the yield of food crops involves the addition of stalks to each plant, allowing for more panicles per plant. The IRRI's Genetic Resources Center has a cold-storage unit that houses some 80,000 rice samples waiting to be crossbred into new varieties. This allows IRRI scientists to make finite genetic changes to plant embryos in test tubes instead of crossbreeding plants in the field, thereby reducing the breeding time and allowing for isolation of specific traits. And despite low adoption rates of hybrid rice in many countries, the contribution to the aggregate supply is substantial. Without hybrid varieties, global production would have been 2.3% lower (Pinstrup-Andersen, 1997). The higher prices would have filtered through the production chain and resulted in higher consumer prices, which would have resulted in contracted consumption particularly for poor households, and a loss of income for all rice farmers.

So if hybrid is the key in the coming years, the question becomes what rate of adoption is necessary over the next several decades to improve productivity and support current per capita consumption rates. Even if countries can afford to boost hybrid rice adoption to the estimated rates, consumer prices are nevertheless likely to increase, and the poorest segments of the population, nearly half of the global population will endure serious strife. Unfortunately, this troubling scenario seems to already be unfolding. It was noted by the global bank recently that the price of rice has risen by 7.2% over the past six months and so too have the costs of all other consumer commodities; and, they will continue to do so. The current catalyst for the jump can be traced to the new domestic policy in Thailand, a country that supplies 1/3 of export demand. The new Prime Minister, Yingluck Shinawatra, is following through on a campaign promise to push up the international price of rice. Her plan includes buying up current supplies from her farmers at a price 40% above current market cost, in anticipation of a higher payout in the future once prices have risen. A second catalyst this year was the Fukushima nuclear disaster in Japan following the devastating March 11th earthquake and tsunami. An estimated 1.5 million tons of rice was lost to the irradiation, and Japan, which largely relied on domestic production to fill consumption requirements, will soon delve into the international market for more imports. So this demand in combination with more expensive rice from Thailand may cause hoarding in other rice-producing countries (Richardson, 2011).

The countries most susceptible to price increases are those that have limited local cultivation and depend heavily on importation such as West Africa Central America, Somalia, and Haiti. USAID made a dire prediction recently that prices were likely to climb 20 percent over the next three months (IRIN, 2011). The last time we saw such a dramatic spike was in the spring of 2008, when the price went to more than $1000 per metric ton. At that time, Vietnam chose to restrict its exports to keep domestic prices low, and consequently cut down on global supply. While rice is not expected to trade up to those levels, it currently stands $615.55 per metric ton, an 19-month high. In response to this threat, African countries have been investing heavily in local rice production to guard against increased volatility. And recently a government ban in India was lifted to free up two million tons of rice for international trade. But many countries still heavily depend on importation, not to mention the World Food Programme, which relies on rice for 1/10 of its total food purchases (IRIN). Volatile food prices are a big challenge amid this new world. This uncertainty and skepticism will surely increase the number of people in crisis in 2012 and beyond.

## Food Insecurity

The 1996 World Food Summit in Rome defined adequate food security as existing “when all people, at all times, have physical and economic access to sufficient, safe and nutritious food to meet their dietary needs and food preferences for an active and healthy life” (Rome Declaration, 1996)

Food insecurity undermines the future resilience of any country. The ramifications of food insecurity are widely felt in health, economics, national security, education, and much more. There is probably no issue more central to human life and global stability than access to food. More than fallen currencies, or high unemployment, people without access to food are truly facing a life or death situation. It undermines all of our futures in one form or another. While enough food is being produced today to feed the entire population, and food production continues to increase at the same rate as the population is growing with major yield more than doubling over the past thirty years, it remains grossly unequally distributed and it leaves 700 million in the developing world chronically hungry. What's more is that the world is estimated to be challenged with an additional 100 million more people each year, the largest annual increase ever noted (WHO, 2003), and is expected to pass the 7 billion mark on October 31 of this year. Moreover, with burgeoning populations demanding more food, water, and land at a time when climate change threatens our agricultural resources and vast farmland is being diverted from food production, we are brewing the perfect storm for a major collapse in food security. And so the question becomes what can we do to improve the dismaying prospects of food insecurity.

No one should have to go hungry today especially considering that over the past decade or so, food prices in the world market have been at all-time lows, in part because of the increases in yield derived from hybrid varieties that have intensified this downward price pressure. But historical food prices were not necessarily indicative of access. More than 1 billion poor individuals worldwide live on less than a dollar per day and clearly will still have tremendous difficulty securing food and expressing effective market demand. At one time it seemed reasonable that we as a society could expect to halve the number of chronically hungry people in this world by 2015, at least this was the goal set out in the World Food Summit of 1996 and the first Millennium Development Goal7 (Rome Declaration). But over the past few years, the world has been crippled by compounding economic, financial and food crises that have reversed efforts to reduce famine. Moreover, global food prices have spiked since 2006 and the rise has been a continued trend thanks in part to a worsening disparity between food demand and supply poorly functioning agricultural markets.

We are hit with unprecedented fluctuations in price based predominantly on speculation and hype and also the strengthening link to energy prices and the world's move to a more affluent meat-based diet. Additionally, for the first time ever, we are witnessing first-hand the exacerbating effects of climate change on global food production. Recent aberrant weather worldwide, including the unprecedented drought striking the Horn of Africa are cause for concern. In normally temperate regions, we have seen the hottest seasons on record. High seasonal temperatures is a dramatic detriment to agricultural productivity and food security. Global climate models shows a greater than 90% probability that by the end of this century, we will see extremes in temperature that we have not witnessed in our lifetimes. Without sufficient investments and legitimate attempts to curb climate change, we will experience an unimaginable magnitude of damage to our food systems (Coveney, 2009).

## Discussions, Conclusions, and Recommendations

And while promoting food security has for this reason become central to international agendas, this is not adequately reflected in public investment, which has been in steep decline. In 1979, official development assistance aid (a term created by the Organization for Economic Cooperation and Development to measure aid) to agriculture programs made up 18% of total assistance, in 2009 that number has plummeted to 6% (FAO, 2010). Government investments also fell by 1/3 in Africa and by almost 2/3 in Asia. Failure to stimulate investment in agricultural development will result in mass starvation. The last time our world was faced with a similar situation was in the 70s and 80s and the only reason that our world avoided a catastrophe at that point in time was because of the development and utilization of hybrid technology to increase yield. That foresight is no longer present and continued, sustainable agricultural development seems a thing of the past.

With just under 1 billion people going hungry every day and growth figures projecting that the world's population will top 9 billion by 2050, we still have a long way to go. To address these challenges, new investments are needed. Public instruments, particularly in developing countries, must be re-established and new mechanisms much be enabled involving the private sector, which thus far has been slow to emerge. Likewise, more focus needs to be placed on enacting policies for private agricultural investment on an individual scale. Small-scale farmers need to be at the leading edge of any new agenda. There are about 500 million small-scale farms worldwide, the majority of whom are family-run, and they produce most of the food consumed locally. Small-scale farmers are the biggest gross investors in agriculture in the world, and can thus have significant influence on economic growth as well ensuring adequate food security and reducing poverty. But in order to thrive, small-scale farmers need to have better access to water and financial loan services to pay for yearly costs for seed, fertilizer, and equipment. Moreover, food security can be better insured if there is improved infrastructure to ensure that the increased yield makes it to the free market. Agricultural research and technology is also needed to tackle future problems of land degradation and climate change.

Small-scale farmers also need incentives to reinvest and they require a long-term commitment from their government to support small-scale agriculture. For instance, a proven model that could be initiated is the Grameen Bank project, which was launched in Bangladesh by former Professor of Economics at Chittagong University and former Nobel Peace Prize Winner, Dr Muhammed Yunus. The project sought to establish a credit program to make financial resources available to landless and poverty-stricken people in Bangladesh through an idea known as microfinancing. It gave individuals the potential to create employment for themselves, to increase their income, self-esteem, and standard of living. The project was highly successful and Grameen Bank members have witnessed improvements in income, standard of living, and employment generation (Yunus, 1999).

Lastly, if we are to restore food security, there first needs to be a re-establishment of global economic stability and prosperity. Without a return, recovery efforts and ultimately growth will be undermined by the rapid population growth.  Measures must be put in place that complement growth-focused policies such as investment in infrastructure and research. World grain production per person must also increase steadily. Recent trends suggest that there has been a leveling off for the world as a whole and there is no indication that the per capita world food production will cease its recent downward trend (Pinstrup-Anderson, 1994). The recent stagnation and possible fall in world grain production per person should be of serious concern because factors other than population growth will continue to push grain demand upwards.

Future food security will depend not on the progress made by the private sector and the government, but through progress of minor players like the small-scale farmer. We must watch for changes in particular in the global economic climate and economic growth, population growth and urbanization, improvements in technology and rural infrastructure, and environmental awareness. If we continue this troubling trend that started several decades ago, we are doomed to suffer from greater food insecurity and global volatility.

## INDEPENDENT RESEARCH

### Analysis of the Affect of Climate, Principles and Urbanization, and Acceptance of Hybrid Rice in Changsha, China

*Introduction*

China is changing, as is much of the rest of the world. A culture that used to be dominated with individual family farms and an agricultural economy is giving way to a more industrialized way of life as China tries to modernize its economy. But while there is this great wave of change, there are individuals missing out on this historic rise in prosperity. Farmers are being left at the wayside and left to struggle in underdeveloped and underserved regions of country.

Since my project was so intimately related to the farmers in China and the work of farming rice that is the essence of their livelihood, I wanted to use some of my time in Changsha to speak directly with these farmers and get to know them on a more personal level. I wanted to understand how they are coping with this change and what their mindset is at this point. I wanted to get a sense of their lives as farmers, where they felt farming in China was headed and how secure they felt about their future.

Secondly, I wanted to understand how my work in the lab affected these farmers. I sought out their understanding of hybrid rice and what affected the adoption rate in China. We had come up with our own conclusions in the lab and by reading research papers about why hybrid rice was not as popular and well-accepted as conventional rice lines, but I needed to know firsthand what the farmers thought. I wanted to hear it straight from their mouths about what it was about the quality of the rice that they hated.

Finally, much of my life has been dedicated to fighting climate change and promoting environmental conservation through my non-profit organization, Warning about Warming, and so naturally I was very curious about the interplay between farming and climate. Was it even on the minds of farmers here in China?, because it definitely was in other parts of the world. On March 31 of this year, the Food and Agriculture Organization warned of "potentially catastrophic" impacts on food production from climate changes that are expected to hit the developing world. Climate change is making farming more difficult, even in our own backyard. The US National Climatic Data Center announced in June that the weather extremes experienced thus far in the year were "unprecedented" and "never before" seen (Jamail, 2011). Severe weather events such as the severe, life-threatening drought we see in Australia or the incomparable flooding happening right now in Thailand will increase, even in parts of the world that aren’t accustomed to weather extremes. Changes worldwide in temperature, rainfall, and related pest and disease outbreaks will drive up food costs around the world and make farming more challenging. And so, I wanted to get a sense of what Changsha farmers have experienced because they are at the forefront of this battle. They see and feel the impact of global warming before anyone else on this planet.

*Results*

| **Analysis of the Affect of Climate, Urbanization, and Acceptance of Hybrid Rice in Changsha, China** | |
|---|---|
| ***POPULATION SNAPSHOT*** | |
| **Gender** | |
| **Male** | **23 (77%)** |
| **Female** | **7 (23%)** |
| **Age Distribution** | |
| **< 20** | **0 (0%** |
| **20-29** | **3 (10%)** |
| **30-39** | **6 (20%)** |
| **40-49** | **8 (27%)** |
| **50-59** | **5 (17%)** |
| **>60** | **8 (27%)** |
| **Education Level** | |
| **None** | **12 (40%)** |
| **Primary** | **18 (60%)** |
| **Secondary** | **0 (0%)** |
| **Marital Status** | |
| **Yes** | **30 (100%)** |
| **No** | **0 (0%)** |
| **Number in Household (incl. children, grandparents, etc)** | |
| **1** | **0 (0%)** |
| **2** | **3 (10%)** |
| **3** | **17 (57%)** |
| **4** | **5 (17%)** |
| **5+** | **5 (17%)** |

### *PRINCIPLES & URBANIZATION*

| | | |
|---|---|---|
| **Land status** | | |
| | **Own** | **30 (100%)** |
| | **Lease** | **0 (0%)** |
| **Crops grown** | | |
| | **Rice** | **30/30** |
| | **Corn** | **1/30** |
| | **Peanuts** | **1/30** |
| | **Sweet potato** | **1/30** |
| | **Rapeseed** | **1/30** |

### *PRINCIPLES & URBANIZATION*

| | | |
|---|---|---|
| **Desire to leave farming for city?** | | |
| | **Yes** | **19 (63%)** |
| | **No** | **11 (37%)** |
| **Feel that government hears your concerns?** | | |
| | **Yes** | **22 (73%)** |
| | **No** | **8 (27%)** |
| **Does govt. control of prices hurt you?** | | |
| | **Yes** | **14 (47%)** |
| | **No** | **16 (53%)** |
| **Are rice prices competitive?** | | |
| | **Yes** | **17 (57%)** |
| | **No** | **13 (43%)** |

| | **Men** | **Women** |
|---|---|---|
| Own land | 23/23 (87%) | 7/7 (0%) |
| Favorable govt. opinion | 17/23 (74%) | 5/7 (71%) |
| Leave for the city | 16/23 (70%) | 3/7 (43%) |
| Are prices competitive | 12/23 (52%) | 5/7 (71%) |

| | **20-29 yrs.** | **30-39 yrs.** | **40-49 yrs.** | **50-59 yrs.** | **60+ yrs.** |
|---|---|---|---|---|---|
| Own land | 3/3 | 6/6 | 8/8 | 5/5 | 8/8 |
| Favorable govt. opinion | 0/3 | 2/6 | 8/8 | 4/5 | 8/8 |
| Leave for the city | 3/3 | 6/6 | 7/8 | 3/5 | 0/8 |
| Are prices competitive | 1/3 | 3/6 | 5/8 | 3/5 | 5/8 |

## *PRINCIPLES & URBANIZATION*

**Biggest problems facing your crops?**

1. **low income crops**
2. **area for farming is not enough**
3. **flooding, drought, other bad weather conditions**

**Biggest concern for the future?**

1. **rice prices**
2. **fertilizer prices**

**Main causes of poverty in China?**

1. **fields are dispersed**
2. **lot of poverty in mountain areas, because there is no transportation to sell their goods**
3. **lot of people, not enough technology**
4. **lots of people, less land**

**How could livelihoods be improved?**

1. **government should improve policies to support agriculture**

**What role should government play in reducing food insecurity?**

1. **more responsibility**
2. **should play major role in supervision of food security and agriculture**

## *ACCEPTANCE OF HYBRID RICE*

**Do you currently farm hybrid rice? Why or why not?**

1. **Yes, high yield**

**What are the advantages of hybrid rice, what are the disadvantages?**

**ADV: high yield**
**DISADV: cost of replanting every year, conventional rice is higher quality and easier to manage, susceptible to insects**

**How would you suggest rice seeds be changed?**

1. **better taste**
2. **more nutritional value**
3. **better cooking quality to make it more like conventional rice**

| *CLIMATE* | | |
|---|---|---|
| **Do you use pesticides and fertilizers?** | | |
| | **Yes** | **30 (54%)** |
| | **No** | **0 (46%)** |
| **Do you try to conserve water?** | | |
| | **Yes** | **0 (0%)** |
| | **No** | **30 (100%)** |
| **Do you experience water shortages?** | | |
| | **Yes** | **23 (77%)** |
| | **No** | **7 (23%)** |
| **Do you think climate change exists?** | | |
| | **Yes** | **25 (83%)** |
| | **No** | **5 (17%)** |
| **Is the climate affecting your farming?** | | |
| | **Yes** | **19 (63%)** |
| | **No** | **11 (37%)** |

| ***CLIMATE*** |
|---|
| **How do you feel the temperature has changed in the last 20 years?** |
| **1. over the years it has steadily increased**<br>**2. temperature has only gotten warmer** |
| **Have you experienced flooding? When? How much? Did it ruin your crops?** |
| **1. no recent flooding**<br>**2. yes, several years ago, it was very serious, it ruined my crops** |
| **Have you experienced drought? When? How much? Did it ruin your crops?** |
| **1. yes, several years ago** |
| **Is the soil quality good?** |
| **1. alright, not as good as before, more fertilizer made soil bad**<br>**2. doesn’t care**<br>**3. alright, not bad** |
| **What do you think about the pollution in China?** |
| **1. industrial sewage is affecting rice planting and agriculture**<br>**2. needs treatments**<br>**3. air pollution**<br>**4. water resources affect agriculture**<br>**5. pollution worsened by industry, but agricultural pollution is little** |

*Discussion*

The results of the survey gave me a clear impression that the farmers in Changsha felt distinctly threatened by the onset of technology and industry in China. The group was predominantly older and was married and had families to take care of. They were moderately educated and yet all individuals I interviewed owned their own land. Most of the individuals that I spoke to had thoughts of leaving for the city; this was especially notable among the male segment of the group. The women were split fairly evenly between wanting to stay in their home or uprooting their livelihood and heading for the city for higher paying jobs. Also, when broken down by age groups, the younger individuals are more eager to leave the countryside, as they have not yet established a permanent life and a family for themselves in Changsha. They see increased hope in the city and increased opportunities for jobs that will provide them more income than they currently have. Even a portion of the older population, although smaller, still preferred to go to the city and leave everything behind, but the majority was afraid to seek out new employment when they were already established in farming. This is particularly concerning to see adults wanting to leave behind what has been their life for 40+ years. The group was split on whether rice prices are competitive, but it appears definitive that the younger generations are less optimistic and hold a far less favorable opinion of the current government. They feel that the government should take responsibility and play a major role in the supervision of food security and in improving policies to support farmers. For the future, they are concerned that rice prices will not provide the adequate income for their families and they are concerned about the cost of inputs such as fertilizers.

Concerning climate change, a surprisingly large proportion of farmers believe that global warming is a genuine problem. They acknowledge that the temperature has steadily increased over the past few years. Moreover, some have reported that they have experienced flooding and drought in the past. But while they acknowledge climate change, few believe that it is affecting their crops and even fewer consciously choose to conserve water and natural resources. So it would appear, that the message of global climate change has reached rural China, but they are not yet compelled to take action. This will have to change in the near future if we as a society are to successfully curb the oncoming climate change.

On the issue of hybrid rice, all those interviewed grew hybrid rice and found it beneficial for its high yield. Yet they do acknowledge some drawbacks and for the most part, they confirm our understanding that hybrid rice is poorer in quality. Importantly, they also indicated that hybrid rice is additionally troublesome because it is more difficult to manage and more susceptible to insects. Therefore, the research that we are working on in the lab to improve hybrid rice quality is validated by the opinions that I heard from these farmers.

Through working in the lab and my independent research working in the field, I learned that it is not just the work of the researcher that makes the difference; it is the work of the farmer as well. Through the combined cooperation of the two, food insecurity can truly be alleviated.

## A Global Perspective: Food Insecurity and the Utilization of Hybrid Rice

### *Introduction*

The second independent research project I started stemmed from the unique circumstances surrounding my visit to Changsha. For the entire two months that I was working in the lab at the China National Hybrid Rice Research and Development Center, there was a special group of individuals there who were learning about hybrid rice so that they could bring their newfound knowledge and wisdom back to their home countries. This group was a diverse set of individuals coming from several countries in Africa, many of which are faced right now with drought and famine. I took to interviewing these individuals to learn why they were here, why they thought learning about hybrid rice was important for their community, and what their most important concerns about food, food supply, and rice were back at home. It was a once in a lifetime opportunity. I was exposed to the culture that these men and women brought to our housing complex and I learned a lot about their views on agricultural and the unique privilege they had to come to China to learn about rice cultivation practices. Food security is a truly global issue and this made it ever more apparent to me.

*Results*

| A Global Perspective: Food Insecurity and Hybrid Rice | | |
|---|---|---|
| ***POPULATION SNAPSHOT*** | | |
| **Gender** | | |
| | **Male** | **8 (80%)** |
| | **Female** | **2 (20%)** |
| **Age Distribution** | | |
| | **< 20** | **0 (0%)** |
| | **20-29** | **3 (30%)** |
| | **30-39** | **4 (40%)** |
| | **40-49** | **3 (30%)** |
| | **50-59** | **0 (0%)** |
| | **>60** | **0 (0%)** |
| **Education Level** | | |
| | **None** | **0 (0%)** |
| | **Primary** | **10 (100%)** |
| | **Secondary** | **0 (0%)** |
| **Marital Status** | | |
| | **Yes** | **2 (20%)** |
| | **No** | **8 (80%)** |
| **Number in Household** | | |
| | **1** | **8 (80%)** |
| | **2** | **0 (0%)** |
| | **3** | **1 (10%)** |
| | **4** | **0 (0%)** |
| | **5+** | **1 (10%)** |
| **Occupation** | | |
| | **Farmer** | **9 (90%)** |
| | **Engineer** | **1 (10%)** |

| ***A GLOBAL PERSPECTIVE: FOOD INSECURITY AND HYBRID RICE*** |
|---|
| **Why are you here?** |
| **1. to learn about hybrid rice** |
| **Why is learning about hybrid rice important for your community?** |
| **1. employ new techniques**<br>**2. bring back new information to our communities**<br>**3. to combat food insecurity**<br>**4. to combat poverty**<br>**5. rice can help the ailments of the world** |
| **What did you expect to learn from this experience before you came?** |
| **1. more about hybrid rice and how I can grow it** |
| **How will what you learn here help your community back home?** |
| **1. improve production**<br>**2. we can employ new techniques in hybrid rice production and increase production in our communities** |
| **What are your most important concerns about food, food supply, and rice at home?** |
| **1. lack of food causes sickness**<br>**2. food supply is not enough for family at home**<br>**3. food at home needs to be greater quantity and better quality** |
| **How do you think we can solve these problems?** |
| **1. more inventions**<br>**2. more world cooperation**<br>**3. solving the problems together**<br>**4. greater advanced technologies** |
| **What do you think is the biggest problem facing rice production in the future?** |
| **1. increased population**<br>**2. not enough food production** |

### *Discussion*

Staying at the Longping Hightech International Exchange Center gave me the inspiration to conduct interviews with the agronomists who were learning about hybrid rice. I wanted to learn more about hybrid rice in different parts of the world. Five countries were represented during my summer at the center: Angola, Guinea-Bissau, Brazil, Mozambique, and East Timor. I was interested to learn about hybrid rice production in other countries and the difficulties with poverty throughout the world. Everyone there had a different story to tell and different experiences with food insecurity in their communities, but they all came to the center for one reason – to learn about hybrid rice and to bring knowledge back to their communities.

Hearing their responses, I learned that the major concern that they all have is increasing the quantity of rice to feed their villages and towns. Never did I realize how much one crop can mean to a community. Talking and interacting with them truly made me understand that it is not just research in the lab; that research will help millions who depend daily on the quantity and quality of the food grown.

Through working with and interviewing the participants of the Longping Hightech International Exchange Center, I was able to not only travel to China to learn about food insecurity, but I was also able to travel to Angola, Guinea-Bissau, Brazil, Mozambique, and East Timor.

## PERSONAL JOURNEY

### Culture & Travels

Shǒuyè. Home. What more is a home than a place where you learn, live, and love? This summer I discovered a new home: a place where I gained more knowledge in two months than ever before, a place where I lived and explored all that my surroundings had to offer, a place where I was surrounded by love and hospitality on a daily basis.

When I first arrived in Changsha, I was greeted with warm smiles and waving hands. My mentor constantly worried and took care of me, reminding me of my own mother. During my first week in Changsha, I was immediately engulfed by culture. Since my mentor was preparing for a dance for a festival at the Scientific and Technological Exchange Center, I was able to join in and learn the dance as well. It was a great experience, and I learned how to dance with the *shànzi* (paper fan)!

Throughout my time, I experienced new food, a new language, and a new culture. Difficult is probably an understatement when it comes to learning the language, but after a few weeks, I was able to pick up on words easily. When I came to China, I had one rule for myself – try everything and let nothing hold me back. I did just that. And that was the best advice I have ever given myself. I was able to experience so many new foods and activities, such as pig's blood, chicken's foot, and badminton. Surprisingly, I got quite good at badminton, beating one of the best players at the center. Now whether he threw the game, I will never know, but for the record I won.

Whether I went to Shaoshan to see Chairman Mao's birthplace, climbed Yuelu Mountain, or explored Martyr Memorial Park, I always learned something new about Chinese history. It was remarkable to be immersed in thousands of years of history. I gained a greater respect for the culture, customs, and traditions as each day passed. Although each destination I traveled to taught me something new, my last destination and my favorite was Zhangjiajie National Forest Park. Just learning about my opportunity to travel to Zhangjiajie excited me because a few weeks earlier I talked to Allison Zhao, a 2009 Borlaug-Ruan Intern, who was familiar with the area. She said that if there was one place that I should go and see it was Zhangjiajie.

Within the park, we traveled up Tianmen Mountain, which was simply breathtaking. It is said to be the mountain that leads to the gates of heaven. It look us 4 buses, 3 aerial cableways, a two hour walk through the clouds, and 1 ski lift to get to the top, but it was amazing and well worth the travel. However, once all the buses, aerial cableways, paths, and the ski lift were taken, there was just one leg of the journey left and just one thing in our way – 999 steps to the top. Arriving at the top, however, was indescribable. I must say that there is something so magical about mountains – the giants in the sky. The natural beauty of the mountain is something that can never be replicated. Mist and clouds surround the majestic mountain, as if for protection. This time, unfortunately, pictures do not do the mountain justice. It is one of those times where you have to be there in order to truly appreciate the beauty and magnificence of such a place. I truly was in heaven. As I sat at the top of the mountain and looked down at all the hills and clouds, I thought about my time in Changsha. I thought about all the friends I had made and all the inspirational people that surrounded me. I realized that hunger is like a mountain, a daunting and difficult endeavor to climb and defeat, but one that is possible.

## Reflections

I came to the Global Youth Institute as a ninth grader, knowing virtually nothing about food insecurity. I came with a passion for the environment; I left with a greater understanding of how the environment directly affects food insecurity for millions around the world. I learned that my passion for the environment could be used to help make a difference in matters of hunger. I left the Youth Institute informed, astounded, and empowered to make a difference. That motivation did not end right after the institute, instead it continued on. I conducted a Hunger Banquet at my own school to inform kids my own age why it is such an important issue and how almost every discipline whether it is science, business, or law is intertwined in the issue of hunger.

I wanted to know and experience more about the scientific side of food insecurity, and that led me to apply for the Borlaug-Ruan International Internship. Never could I have imagined an opportunity more perfect and enlightening as this internship. With my passion and previous experience of being a scientific researcher, working this summer at the China National Hybrid Rice Research and Development Center was the opportunity of the lifetime. I was able to gain a cross-cultural experience through my two months in Changsha. I was able to gain insight on not only the scientific element of food insecurity but also the human element.

On one of my last nights in Changsha, eating dinner with all of my friends from CNHRRDC, I thought to myself, “These are the hunger-fighters that will make the difference. These are the people who are going to make the biggest difference for humanity, whether they know it or not.” I knew that I wanted to be a part of this – a part of something greater. It was incredible being surrounded by people with the same dedication and motivation for the same cause, that kind of inspiration and ambition cannot be faked.

An empty stomach has so many devastating effects – illiteracy, poverty, and war. Everyone on Earth has the same needs; it truly is life’s Chance Card that puts us in the circumstances we live in, through no choice of our own. Although the battle will be difficult, being in that room brought me to the realization that no person, no object, and no boundary can stop the passion of a person making a difference. It truly was this internship that led me to this realization. It was this experience that changed my life.

I can genuinely say, that ever since my experience this summer, I have never been the same. I see the issues of world hunger and the solutions that need to be made through a whole different set of eyes. I see the future of food insecurity revolutionized through new scientific approaches and improved leadership in developing countries. I now see a world full of possibilities, possibilities that I will be a part of.

# Photos


杂交水稻
国家重点实验室
State Key Laboratory of Hybrid Rice
中华人民共和国科学技术部


旗颂


湖南农业大学


超级杂交水稻

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# APPENDICES

Appendix 1:

## Analysis of the Affect of Climate, Urbanization, and Acceptance of Hybrid Rice in Changsha, China

Name: ____________________ Age: ____ Education Level: ________________

Gender: _______ Date: __________

Are you married? Yes ________ No ______

How many people live in your household/home? ________

***Principles & Urbanization:***

1. Do you own your own land or lease land? ______________________

2. What crops do you grow? _____________________________________________

3. What is your total income per farming year? ___________ Yuan

4. Do you use modern farming machines? Yes ________ No ______
   If no, why not?

________________________________________________________________________

________________________________________________________________________

5. What do you grow during winter, non-rice growing time?

________________________________________________________________________

________________________________________________________________________

6. Who helps you farm? How many people?

________________________________________________________________________

________________________________________________________________________

7. Is industry ruining/impeding your farmland? Yes ________ No ______

8. Do you want to leave farming to work in the city? Why? Yes _______ No ______

________________________________________________________________________

________________________________________________________________________

9. Do you feel your concerns are addressed in government? Yes _______ No _______

10. Because the government holds grain prices down, does this hurt your farming? Yes ____ No ___

11. What is the biggest problem you face with your rice crops?

_______________________________________________________________________

_______________________________________________________________________

12. What is your biggest concern in the future?

_______________________________________________________________________

_______________________________________________________________________

13. Is there a great cost of producing rice? How much a month?

___________________________________ ________ Yuan

14. Are rice prices competitive for you? Yes _______ No _______

15. What do you believe are the main causes of poverty in China?

_______________________________________________________________________

_______________________________________________________________________

16. How could the livelihoods of the people in China be improved?

_______________________________________________________________________

_______________________________________________________________________

17. What role should the government play in reducing food insecurity in China?

_______________________________________________________________________

_______________________________________________________________________

18. How has the yield of your crops been in recent years? Increase or decrease?

_______________________________________________________________________

_______________________________________________________________________

19. Did you know that a policy approved by the Communist Party in October 2008 that aims to end rural poverty gives farmers the right to trade, rent, sublet, subcontract, engage in joint stock ownership and transfer their land rights? Yes _______ No________

***Acceptance of Hybrid Rice:***

20. Do you currently farm hybrid rice? Why or why not? Yes______ No______

_______________________________________________________________________

21. If not, have you tried farming hybrid rice in the past? Why or why not? Yes______ No______

______________________________________________________________________

22. What are the advantages of hybrid rice, what are the disadvantages?

Advantages:

_________________________________________________________________

Disadvantages:

_______________________________________________________________

23. How would you suggest rice seeds be changed?

______________________________________________________________________

---

24. Do you think hybrid rice seeds are expensive or cheap? Yes______ No______

25. Is hybrid rice similar to conventional rice? List differences you find? Yes______ No______

______________________________________________________________________

---

a) Did this affect why or you are not using hybrid rice? ______________________________

26. Does the quality of rice affect what types of rice you grow? What kind of qualities appeal most to you when selecting rice? Aroma? Translucency? Yes______ No______

______________________________________________________________________

---

***Climate:***

27. Do you use pesticides and fertilizers? Yes______ No______

28. If so, do you try using natural fertilizers, i.e. manure? Yes______ No______

29. Do you try to conserve water? Yes______ No______

30. Do you experience water shortages? Yes______ No______

31. Do you think climate change exists? Yes______ No______

32. Is the climate affecting your farming? Yes______ No______

33. How do you feel the temperature has changed in the last 20 years?

______________________________________________________________________________

34. Have you experienced flooding? When? How much? Did it ruin your crops?
______________________________________________________________________________

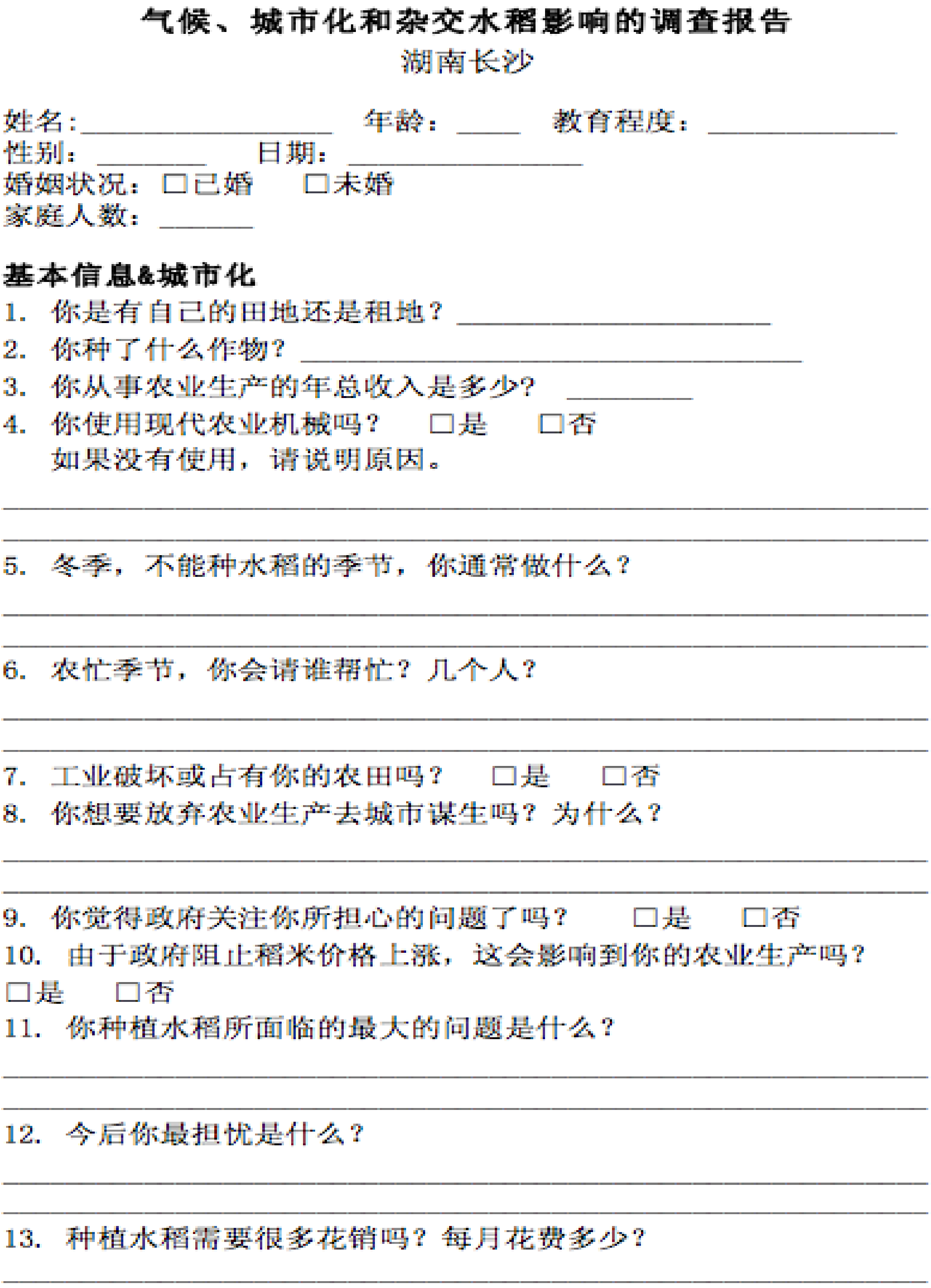
35. Have you experienced drought? When? How much? Did it ruin your crops?
______________________________________________________________________________

36. How much land have you found to be not farmable? For what reason?
______________________________________________________________________________

37. Is the soil quality good? Yes? No? Why not? Do you need lots of fertilizers? Yes____ No____
______________________________________________________________________________

38. What do you think about the pollution in China?
______________________________________________________________________________

Appendix 2
Analysis of the Affect of Climate, Urbanization, and Acceptance of Hybrid Rice in Changsha, China in Chinese



## 气候、城市化和杂交水稻影响的调查报告

湖南长沙

姓名：____________________ 年龄：_____ 教育程度：_______________
性别：_________ 日期：___________________
婚姻状况：□已婚 □未婚
家庭人数：_______

**基本信息&城市化**

1. 你是有自己的田地还是租地？_________________________
2. 你种了什么作物？________________________________________
3. 你从事农业生产的年总收入是多少？ __________
4. 你使用现代农业机械吗？ □是 □否
   如果没有使用，请说明原因。

____________________________________________________________________________
____________________________________________________________________________

5. 冬季，不能种水稻的季节，你通常做什么？

____________________________________________________________________________
____________________________________________________________________________

6. 农忙季节，你会请谁帮忙？几个人？

____________________________________________________________________________
____________________________________________________________________________

7. 工业破坏或占有你的农田吗？ □是 □否
8. 你想要放弃农业生产去城市谋生吗？为什么？

____________________________________________________________________________
____________________________________________________________________________

9. 你觉得政府关注你所担心的问题了吗？ □是 □否
10. 由于政府阻止稻米价格上涨，这会影响到你的农业生产吗？
□是 □否
11. 你种植水稻所面临的最大的问题是什么？

____________________________________________________________________________
____________________________________________________________________________

12. 今后你最担忧是什么？

____________________________________________________________________________
____________________________________________________________________________

13. 种植水稻需要很多花销吗？每月花费多少？

____________________________________________________________________________

14. 种植水稻你认为划算吗？ □是 □否

15. 你认为中国贫穷最主要的原因是什么？

________________________________________

________________________________________

16. 怎样才能改善中国百姓的生活？

________________________________________

________________________________________

17. 中国政府在减少食品安全问题上应担任什么职责？

________________________________________

________________________________________

18. 近几年你所种植的作物的产量怎么样？是增加还是减少？

________________________________________

________________________________________

19. 2008年10月党中央出台了一个惠农政策，允许农民以转包、出租、互换、转让、股份合作等形式流转土地承包经营权，你知道这个政策吗？ □是 □否

## 杂交水稻

20. 你目前种植杂交水稻吗？为什么？

________________________________________

________________________________________

21. 如果目前你没有种植杂交水稻，过去你尝试过种植杂交水稻吗？为什么？

________________________________________

________________________________________

22. 杂交水稻的优点和缺点是什么？

优点：________________________________

缺点：________________________________

23. 对水稻品种的改良，你有什么建议？

________________________________________

________________________________________

24. 你认为杂交水稻价格贵还是便宜？ □是 □否

25. 杂交水稻与普通栽培稻相似吗？ 列出两者之间的不同。

________________________________________

________________________________________

a）杂交水稻与普通栽培稻的不同会影响你种或是不种杂交水稻吗？为什么？

________________________________________________

________________________________________________

26. 当你选择水稻品种时你会考虑稻米品质吗？哪种稻米品质是你优先考虑的？香味？透明度？

________________________________________________

________________________________________________

**气候：**

27. 你使用农药和化肥吗？ □是 □否
28. 如果是，你会使用农家肥吗，例如人畜粪便？ □是 □否
29. 你会设法储水灌溉吗？ □是 □否
30. 农田灌溉时会出现缺水的情况吗？ □是 □否
31. 你认为气候在变化吗？ □是 □否
32. 气候变化会影响你的农业生产吗？ □是 □否
33. 过去二十年，你认为气候发生了什么变化？

________________________________________________

________________________________________________

34. 你经历过洪灾吗？什么时候？严重吗？洪水是否破坏了你的农作物？

________________________________________________

________________________________________________

35. 你经历过旱灾吗？什么时候？严重吗？旱灾是否破坏了你的农作物？

________________________________________________

________________________________________________

36. 你发现有多少农田不能耕种了？什么原因引起的?

________________________________________________

________________________________________________

37. 土壤肥力状况是否良好？什么原因引起的？你需要施很多肥料吗？

________________________________________________

________________________________________________

38. 你怎么看待中国的污染问题的？

________________________________________________

Appendix 3

## A Global Perspective: Food Insecurity and Hybrid Rice

Name: ________________ Age: ____ Education Level:____________ Gender: ________
Date: ___________ Country: _________________ Village/City:_____________
Are you married? Yes _________ No ______ Occupation: ___________________
How many people live in your household/home? _________

Why are you here?
______________________________________________________________________________
______________________________________________________________________________

How long will you spend here? _________________

Describe your life in your country.
______________________________________________________________________________
______________________________________________________________________________

Why is learning about hybrid rice important for your community?
______________________________________________________________________________
______________________________________________________________________________

What did you expect to learn from this experience before you came?
______________________________________________________________________________
______________________________________________________________________________

How much did you know about hybrid rice and rice cultivation before coming here?
______________________________________________________________________________
______________________________________________________________________________

How will what you learn here help your community back home?
______________________________________________________________________________
______________________________________________________________________________

What are your most important concerns about food, food supply, and rice at home?
______________________________________________________________________________
______________________________________________________________________________

How do you think we can solve these problems?
_________________________________________________________________________________
___________________________________________________________________________

What do you think is the biggest problem facing rice production in the future?
______________________________________________________________________________
______________________________________________________________________________

Appendix 4:
A Global Perspective: Food Insecurity and Hybrid Rice in Portuguese for the Agronomists from East Timor, Angola, Mozambique, Brazil, and Guinea-Bissau

**A perspectiva global sobre a insegurança alimentar e o arroz híbrido**

Nome:_____________________________ Idade:_______ Nível académico_____________
Sexo:________________ Data____/____/_____ País:__________________________
Província/Cidade:___________________ Estado Civil:__________________________
Profissão:_______________________________ Agregado familiar:________________

1. Porque está na China?____________________________________________________

2. Quanto tempo ficará na China?__________________________________________
3. Indica as principais actividades económicas no seu país.

4. Porque acha que aprender o arroz híbrido é importante para a sua comunidade?______________________________________________________________

5. O Que espera aprender com a sua vinda a esta experiência?

6. Antes de vir a China o que sabia sobre o arroz híbrido e as formas de cultivo?

7. Como ajudará a sua comunidade com o que aprendeu na China?

8. Qual é o maior interesse na alimentação em casa, são as fontes de alimentos ou o arroz?

9. Como acha que poderemos resolver esses problemas?

10. Qual acha que será o maior problema a enfrentarmos no futuro na produção de arroz?