# Rice and Water: The Final Frontier<sup>1</sup>

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The Rockefeller Foundation

The First International Conference on Rice for the Future 31 August – 2 September 2004

Bangkok, Thailand

#### I. Introduction

## Brief history of rice culture and water use

Throughout the past 7-10,000 years of Asian history rice has been pivotal to all aspects of human activity whether fostering societal and community development, wagging war or seeking peace, creating wealth or enduring poverty, enjoying good health or surviving famine or providing a foundation for spiritual worship of deities. From the earliest records of rice domestication, irrigation water management has preoccupied Asian populations from rainfed peasant farmers to the mightiest emperors. Dr. Te-Tzu Chang, imminent rice historian, writing in the Cambridge World History of Food (Kiple and Ornelas, 2000) provides an excellent historical perspective on the intimate relationship between the history of water management and rice cultural practices as well as a reminder of its antiquity. He writes that early historical records of rice culture and flood control in the Yellow River flood plain of northern China date to approximately 4,000 years ago (2000 BC) and by 1400 BC dams, canals, and conduits were in operation there (Chang, 2000). The linage of Asian societies and governments over the past two millennia are replete with rulers who recognized the strategic importance of rice and created the necessary water management systems as the basis of their rule.

Water is essential to the growth and yield of all food crops and yet continuous flooding during much of the cropping period is uniquely associated with rice. In

Paper presented on the occasion of H.R.H. Princess Maha Chakri Sirindhorn's presentation of the Golden Sickle Award to Dr. John C. O'Toole for his contributions to the advancement of rice research over the past 30 years. September 1, 2004, Bangkok, Thailand.

addition to irrigation for the crop's water requirement, flooding rice fields plays a major role in soil chemistry and weed suppression. Annual flooding from river waters bring renewed soil fertility via silt deposits, cause anaerobic soil conditions that lead to a neutralizing effect on soil pH, suffocate noxious weed species and bring about a host of other rice cultivation factors unique to this crop species.

From this brief discussion it is obvious that irrigation of rice fields, as an intentional human activity, has been a part of Asian culture for many thousands of years. However, the ancients may never have suspected there would be so many of us to share the fresh water of 21<sup>st</sup> Century Asia!

## Current indicators of water resources in jeopardy

The media, from newspapers to scholarly journals, provide ample evidence that societies around the world are taking seriously the possibility that a combination of human population growth and climate change may soon impact the reliability of the globe's fresh water resources. Vörösmarty et al. (2000) used numerical experiments to combine global climate change model outputs with water budgets and socio-economic information to investigate future fresh water supply and demand scenarios up to 2025. The results leave little doubt that population growth is the primary driver of current and future strains on water resources given that many developing countries, such as China and India (combined population approx. 2.4 billion) appear set to grow their economies at unprecedented rates over this period. When population growth is coupled to the less well-predicted climate change parameters the scenario by 2025 is critical for many Asian countries where rice is the staple food crop (Vörösmarty et al. 2000)

The future imperative is clear---Asia cannot continue to depend on the quantity and quality of fresh water for rice culture in the traditional manner. Because changes of this magnitude will undoubtedly require major national level restructuring of agriculture, preparations are imminent for a radical change in the relationship between rice and water due to the current and increasing competition between urban, industrial and agricultural water use. (Vital Water Graphics www.unep.org/VitalWater/15.htm)

## II. The problem and its potential solutions:

The Problem--Economic and social dimensions of "drought" in rice

Whether due to inadequate irrigation or to direct deficits of rainfall, "drought" impacts rice farmers with high frequency and dramatic affects. Table 1 estimates these impacts in terms of the magnitude and economic value of average annual production lost due to drought in selected Asian locations and globally.

Table 1. Estimated Average Annual Rice Production Lost to Drought (Widawsky and O'Toole, 1990; Evenson, et al., 1996)

	Million metric tons lost	Million US\$ lost
China-rice	4.4	880
Eastern India-rice	2.9	580
Global-rice	18.0 (4% of total)	3,600

It is obvious that millions of rice farmers in Asia are disadvantaged partially due to the repeated shocks of drought on their pursuit of livelihoods earned from farming in "drought prone environments". The lack of water adequacy indicated by such terminology as "rainfed", "less-favored", "marginal", "drought-prone" and "poorly irrigated" crop production circumstances are implicitly associated with rural poverty. These farm families not only loose crop production due to periodic drought, they also suffer the additional costs related to drought induced "coping mechanisms", or "risk management strategies" they are forced to employ.

It is difficult to develop techniques to estimate the actual costs related to how farmers cope with drought. However, using 1996 as a reference point (officially a "moderate" drought year) Pandey et al. (2000) tracked the costs associated with commonly described coping mechanisms in the East Indian state of Orissa. This study's results provide a measure of the prevalence and nature of specific coping mechanisms as demonstrated by the percentage of households in which the following *ex post* drought coping mechanisms were utilized--- selling of livestock (55%); selling of assets such as jewelry (25%) and land (16%); mortgaging jewelry (8%) and land (25%); reduced quantity of food consumption (87%); households that ate food not normally eaten (44%); postponed medical treatment (67%); curtailed children's education (5%); and the most desperate measure, permanent migration (2%). The true costs (monetary and personal/community social stresses) of these coping mechanisms are poorly understood at present although the few studies available indicate a very real and substantial burden on farm families and rural societies as a whole.

Thus we may conclude that annual yield losses of rice production due to drought are regionally and globally significant, and loss of opportunities and assets due to drought-induced coping mechanisms are a tremendous burden on farm families.

#### The Potential Solutions—new cultural systems and adapted rice varieties

Solutions can be visualized as beginning with changes in farm-level water management and concomitant changes in agronomic management. Ideally, the task of creating new rice varieties adapted to those conditions would proceed in tandem with development and testing of new agronomic technologies, because drastic modifications in water, soil fertility and weed management practices represent a significant selection pressure for new adapted cultivars.

Basically, the objective of these technologies, applied in concert, is to provide an answer for the following quandary. "How can we obtain more rice per unit of water, while safe guarding farm communities from the many and varied socioeconomic impacts of crop yield loses due to water deficit?"

New irrigation water management, soil fertility and weed management strategies

Alternatives to continuous flooding of rice have occupied considerable research over the past 25 years, especially in localities where water resources have already become physically and/or economically scarce. Tuong and Bouman (2003) present a review of water productivity in rice and survey the irrigation and associated technologies available given a water-limited future. Their assessment is broad in scope and couched in the realities of nationally variable government policies related to the economics of water management alternatives as well as the importance of environmental services from large watersheds. Periodic surface irrigation or "flush" irrigation methods are a viable alternative to continuous flooding. The major ramification of this alternative is partially or fully aerobic soils rather than the highly reduced flooded soils characteristic of present systems. This outcome greatly changes the traditional agronomic background of rice soil fertility and weed management and once again emphasizes the need for coordinated changes in water management, agronomic practices and adapted varieties.

New well-adapted rice varieties of rice for water-limited production systems.

The remaining sections of this paper address how the genetic modification of rice for various types of water-limited cultural scenarios is being pursued and what the future may hold.

Rice, *Oryza sativa* L., is a semiaquatic plant species. Many closely related species in the genus *Oryza* are adapted to habitats with low evaporative demand (shaded forest margins) and seasonal high-to-positive water tables such as the estuaries and marshes of major river flood plains (Chang, 1976). Given cultivated rice's phylogenetic origin it is not surprising that rice germplasm, in general, is best adapted to agricultural cultivation under flooded or saturated soil-water conditions. Indeed, the "rice bowls" of Asia are river flood plains and deltas where seasonal inundation creates semiaquatic conditions that no other major food crop can tolerate. However, within the world's cultivated rice germplasm there appears to be a great spectrum of native adaptation to hydrological backgrounds and associated soil physical/chemical conditions. Figure 1. illustrates the broad topographic/hydrologic range of rice ecotypic adaptation from upland/hill (aerobic soil) to drought-prone rainfed lowland (alternating flooded and aerobic soil) to adequately irrigated and the relatively rare deep-water rice areas.

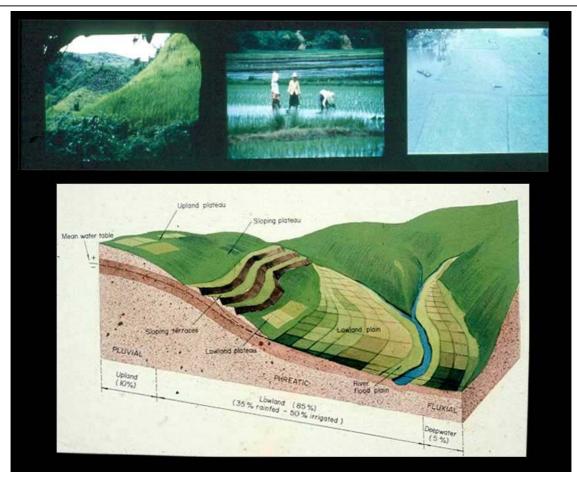


Figure 1. Rice is cultivated in a broad range of hydrological backgrounds. This figure illustrates the range of slope, topography and subsurface and surface water table conditions that underlie the terminology used to describe rice environments such as "upland" "lowland" and the common form of "irrigated" rice, that of river diversion to a reservoir or directly to a canal system. For a complete discourse on the definitions as well as contrasting nomenclature of rice cultural systems globally see IRRI, 1984)

If rice has evolved primarily as a semiaquatic species over millions of years and yet some "ecotypes" and near relatives (*Oryza* species) are adapted to the margins of that ecophysiological domain, then how can modern high yield rices be modified to take advantage of that genotypic variation for cultivation in water-limited environments? Perhaps it is by identifying and introgressing particular genetic traits into the best commercial varieties existing today?

A number of studies have shown that the basic morphological and physiological traits associated with better adaptation to non-flooded rice culture are simply the plant's water uptake system (root system) and factors that limit transpiration or nonproductive water loss from the shoot system (cuticular resistance, stomatal closure and leaf rolling). Genotypic variation exists among rice ecotypes for all these traits and related physiological responses to water deficit (root systems/depth allocation, Lafitte et al. 2001; cuticular resistance to water vapor/leaf surface wax, O'Toole and Cruz, 1983; Haque et al. 1992; capacity to osmotically adjust leaf tissue, Lilley and Ludlow 1996). In recent years there has been a resurgence in physiological studies related to the impact of water deficits on physiological processes that cumulatively result in crop yield under water-limited conditions. These studies also partially explain the substantial genotype X environment interaction that confounds and frustrates this research area (Fukai and Cooper, 1995; Pantuwan et al. 2002b; Lafitte and Courtois, 2002; Kamoshita et al. 2002b;).

Physiological traits are interesting, but the rice breeder and farmer are most interested in the grain yield performance of new varieties under drought stress conditions regardless if that water deficit is a function of rainfed culture or new irrigation water management systems.

# III. The past, present and future of rice genetic improvement for "water-limited production environments"

Past in context: The Green Revolution

In the context of the Green Revolution, beginning in the late 1960s and early 1970s, great pride was experienced by national research organizations by raising yields from 1-2 t/ha to 3-5 t/ha in extensive irrigated areas across south and southeast Asia. Given this atmosphere it is easy to see why traditional rainfed or poorly irrigated sectors were thought to be "backward" and not worthy of

investment of either the limited human or financial resources of the day. In the light of these successes in the irrigated sector, enhancing rice production through breeding for rainfed zones was associated with a low probability of success and resulted in low priority for research support.

In the late 1970s when breeding and releasing the new "high yield varieties" or HYVs reached its zenith and yet population growth continued to keep pace with cereal production (or visa versa), decision and policy makers looked more seriously at the imperative of investing in rice production technologies that "fit" rainfed and poorly irrigated areas. As might be expected the better-prepared disciplines of pathology and entomology, with "known breeding and selection methods", received rice breeders' immediate attention. MacKill (1986) illustrated conclusively that rice breeders were well aware of the importance of drought as a major constraint to yield and its stability. However, when surveyed they reported making very few specific crosses or selections for this trait. It was clear that without knowledge of and confidence in drought screening and selection protocols, breeding for drought tolerance was simply not a part of their mainstream rice improvement programs. Studies of drought tolerance were left to a few poorly supported geneticists and breeders dependent on natural occurrence of drought annually and without the "tool box" found in the recognized disciplines focused on breeding for disease and insect resistance. Given the magnitude of the problem, and its recognition by breeders, administrators and economists, it is amazing that for decades, more effort was not channeled toward breeding solutions for a problem of this economic magnitude and social importance.

Evenson and Gollin (2003) assessed the impact of the Green Revolution between 1960-2000 with regard to productivity impacts of international crop genetic improvement research on developing countries. They describe the significant role of the centers in providing modern varieties and breeding lines to public and private sectors. However, their analysis also identifies the significant differences in the impacts of those varieties based on their agroecological "suitability". The adequacy of water was identified as one of the primary criteria for expression of the new genetic technology's potential. Hence the introduction and adoption of modern varieties for the water-limited sector was slow and more limited. These authors divide the Green Revolution into early (1961-1981) and late (1982-2000) phases. They noted that varieties released in the late phase began to show the influence of major national and international research centers' change of focus as those centers turned their attention to "marginal environments". From this analysis, as well as Mackill's (1986) survey noted above, we are left with the impression that the Green Revolution varieties of rice, although well adapted to fully irrigated conditions or the best hydrological class of rainfed rice areas, were far less suited to water-limited environments.

## Present situation

Conventional breeding enhanced by new knowledge and selection technologies.

During the past five to seven years a number of institutions in China, India, Thailand and the International Rice Research Institute, Philippines have launched rice genetic improvement programs to address the loses attributed to current and anticipated water-limited rice culture.

Serious progress in conventional breeding of new rice varieties for water-limited environments is, in this author's opinion, relatively new among rice research institutions in Asia and Africa. Although a few dedicated breeders attempted over the past four decades to make crosses involving drought tolerant donors, lack of a systematic capacity to screen the resulting segregating populations as well as poor financial support frustrated those efforts.

A systematic survey of experts with global expertise and experience in a wide range of disciplines and crop species during 1997-1999 (Ito et al. 1999; Ribaut and Poland, 2000) indicated that rice research institutions and researchers across Asia lacked both the basic knowledge of water as an experimental parameter (its controlled application and measurement in soils and plants) and the rudimentary equipment to initiate repeatable, science-based field phenotyping/screening of domestic or exotic rice germplasm. Thus rice physiologists, geneticists and breeders had practically no chance of either reliably identifying donor germplasm or routinely conducting genetic studies and breeding line selection, the basis of all crop genetic improvement.

I am very pleased to share with you some of the good news in this respect. To some degree this shortage of trained personnel and equipment has now begun to change in China, India and Thailand with work progressing in Cambodia and Lao PDR as well as West Africa. Several international workshops and training courses have dealt with the theory and practice of science-based screening of rice for drought tolerance (Ito et al. 1999; Saxena and O'Toole, 2002; IRRI, 2002; www.plantstress.com). One particular publication has partially filled the knowledge gap and provided updated information to rice breeders regarding the theory and practice of breeding for drought tolerance in rice (Fischer et al. 2003). The demand for this manual has outpaced even the editor's expectations. It is available on IRRI's Knowledge Bank web site (<a href="http://www.knowledgebank.irri.org/webboard/upload/drought.pdf">http://www.knowledgebank.irri.org/webboard/upload/drought.pdf</a>).

The rice research programs of China are perhaps the most aggressive in Asia in dealing with this challenge due to China's looming water crisis. In the late 1990s assessments of China's future options for fresh water resources illustrated the dire consequences with regard to water and rice (World Bank, 1997). In March 2000 an international workshop was held at Hainan Island, China in which researchers from several Chinese institutions and the International Rice

Research Institute formally took stock of efforts to genetically modify rice for future water-limited production scenarios and planned collaborative research. Several outcomes from that event are noteworthy. Facilities to conduct "managed or controlled stress" screening have been constructed in Eastern and Central China at Shanghai and Wuhan, respectively (Figure 2 & 3) as well as field drought screening facilities developed on Hainan Island where temperatures allow winter-spring rice crops to be field screened for drought tolerance thus adding one selection cycle per year to the breeding process.



Figure 2. In 2001 and 2002 the Shanghai Agrocbiological Gene Center-Shanghai Agriculture Academy of Science constructed over 2000 sq meters of specialized plastic greenhouses (A&B). The facilities include overhead sprinkler and surface drip irrigation capacity, deep (1.8m) drainage systems, and air ventilation capacity. Early experience illustrates the importance of managing the "microclimate" over the crop to simulate realistic field level evapotranspiration as well as the soil water status. C. Field screening facilities on Hainan Island allow large scale off-season (winter-spring) field screening for drought tolerance.



Figure 3. Researchers at the National Key Laboratory of Crop Genetic Improvement, Huazhong Agriculture University, Wuhan, China have constructed perhaps the world's first large scale "rainout shelter". This facility assures control of the water regime to field screen rice for drought tolerance. The structure has an experimental area of 1,800 sq meters and incorporates rain sensors to close the double-layer roof thus protecting experiments. Unlike rainout shelters for other crop species, this structure incorporates deep soil and ground water table management and drainage (2.0 m deep concrete valved-drains) and surface and sprinkler irrigation facilities to simulate water deficits under large-scale rice cultivation.

In Thailand's tropical climate breeders have a dry-season in which low probability of rainfall allow several months to conduct large-scale field screening. Over the past ten years Thai physiologists and breeders have developed three field sites for dry-season mass screening primarily at the vegetative stage (Figure 4) and in addition use late-sown wet-season screening to evaluate stress response at the critical reproductive stage. This combination of facilities, equipment and key locations has allowed systematic progress for the national rainfed rice-breeding program (Fukai et al. 1999, Pantuwan et al. 2002a; Jongdee, 2003)

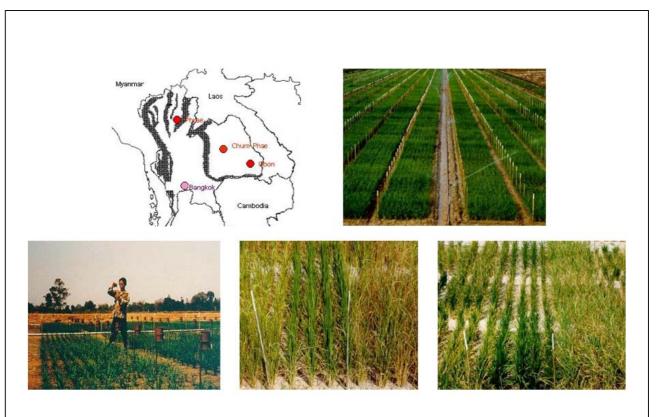


Figure 4. Thailand's Rice Research Institute has selected three locations where soil and climatic conditions favor both dry-season mass screening at the vegetative stage and wet-season reproductive stage screening. The key to operations is access to and control of water resources and training in operation of water management practices to accomplish the desired coincidence between developmental stage and water stress period.

Rice scientists in India have long viewed genetic improvement for "drought-prone" or water-deficit environments as a challenge verging on impossible. However, over the past three decades the national and state multilocation testing system (All India Coordinated Rice Improvement Program-AICRIP) has made slow but steady progress in varietal development for marginal areas that includes drought tolerance. Today a number of researchers in the Indian states of Tamil Nadu, Karnataka and in five states of Eastern India, are using new "managed stress environment" facilities to operate field-oriented selection practices (IRRI, 2002; Poland et al. 2004).

In all three cases above--China, India and Thailand, two innovations characterize a new and successful approach. First, providing physiologists, geneticists and breeders control of water stress severity, duration and coincidence with yield determining growth stages, gave rise to development and utilization of effective selection measures. Second, by employing farmer's participatory selection groups as the final evaluators (Witcombe et al. 2002), real and lasting progress is now within reach (Virk et al. 2002; Virk et al. 2004). These innovations when taken together bode well for the large -scale dissemination of new drought tolerant rice varieties across Asia in the very near future. However, if the considerable power of biotechnological tools are considered, then these new varieties may be only the beginning of our ability to create rice varieties that will eventually lead to "more rice for less water" (slogan of the Indian National Rice Drought Tolerance Network) and "water saving rice" (slogan of the Chinese national effort).

#### Conventional breeding with marker assisted selection (MAS)

In recent years much has been published regarding the utility of DNA marker assisted selection (MAS) for drought tolerance in cereals (Ito et al. 1999; Ribaut et al. 2002; www.plantstess.com). A great deal of effort and expense has been invested in this technology. Initially MAS in rice improvement was most applicable to single gene disease resistance such as that directed toward resistance to rice bacterial blight and leaf blast diseases (Hittalmani et al. 2000; Toenniessen et al. 2003). However, because drought tolerance is acknowledged to be a quantitative trait and thus "polygenic" in nature, the role of MAS in improvement of rice for drought tolerance is still equivocal. Research to correlate particular traits (phenological, physiological and morphological) with yield under stress (preferable managed stress environments) is relatively new (see reviews Fukai and Cooper, 1995; Nguyen et al. 1997). In the past five years the capacity of several national programs to conduct high quality field-level screening has grown significantly in tandem with their in-house capacity to apply DNA molecular markers for genetic research.

Since 1995 we have seen numerous publications that identify DNA molecular markers related to secondary traits correlated with yield under drought

conditions. The most exhaustively researched traits assessed to date have been: root/root system morphology (Champoux et al. 1995; Yadav et al. 1997; Price and Courtois, 1999; Shen et al. 2001; Zhang et al. 2001; Toorchi et al. 2001; Kamoshita et al. 2002a; Babu et al. 2003); and osmotic adjustment of the shoot (Lilley et al. 1996, Nguyen et al. 1997; Robin et al. 2003). However, the salient traits, biomass production and grain yield and its components, measured under managed stress environments, have received attention only in very recent years. This was due primarily to the time required for the above-mentioned mastery of high quality water management in experimental field conditions and concomitant development of suitable mapping populations that were truly segregating for drought tolerance and related physiological traits/mechanisms.

Through a coordinated effort between researchers at the International Rice Research Institute and the Rice Research Institute of Thailand, a single mapping population (219 doubled haploid lines (DHL) from the cross CT9993-5-10-1-M X IR62266-42-6-2) was systematically made available to scientists in India, Israel, Philippines, Thailand, USA and elsewhere for multi-environment drought tolerance testing and comparison of the resulting quantitative trait loci (QTL) identified. Initially the population was assessed in Israel, outside the normal range of rice diseases and pests that frequently confound field screening for drought tolerance. Blum et al. (1999) confirmed that indeed this population was segregating for "true" drought tolerance in field-level screening of the 219 DH lines. This set the stage for a number of studies that allowed comparison of a single population in varied natural and managed stress environments.

For the sake of brevity I refer here to only the published results from two research groups that used this particular mapping population referred to CT9993/IR6266. Both research groups were well qualified in their phenotypic capacity as well as genotypic characterization and analysis. The research group in southern India (Babu et al. 2003) provides an integrated view of plant water stress indicators, DH line phenology, and production traits linked to QTL. As is often the case there are far too many QTL (47) from this multilocation-multiyear study to present a synthesis here. However, the reader is referred to the paper as its discussion provides excellent links to previous reports on the same DH line population. Babu et al. (2003) discuss the relationship of specific QTL for root traits (from previous published reports) with rice yield and biomass from their field experiments conducted under varying levels of water stress. Their discussion centers on segments of chromosome 4 between the markers RG939-RG476-RG214. They persuasively document the growing literature linking this QTL to root morphology, rooting depth and confirm the parental origin (CT9993) of the chromosome segment associated with particular root morphological traits. They also identified QTLs of interest on chromosome 1 as being related to the plant water status (% Relative Water Content-RWC); chromosome 3 related to biomass yield under stress and chromosome 9 related to plant water status (RWC) and lack of stress-delayed flowering.

The second research group located in Thailand (Lanceras et al. 2004) found QTL related to grain yield and lack of stress-delayed flowering under stress treatments at the same location on chromosome 3 as reported in India by Babu et al. (2003). In addition, the Thai group identified QTL for grain yield and biological yield on chromosome 4 in the interval RZ69-RZ565. Their experimental technique used a variable irrigation gradient that provided four irrigation treatments or "environments" under the same soil and weather conditions and this may have provided greater phenotypic resolution among the DH lines. In addition to those two important QTL locations (Chromosome 3 & 4) in common between the two studies, Lanceras et al. (2004) identified a QTL on chromosome 8 in the interval G187-RG997 linked to biomass yield and percent spiklet sterility. In both reports the researchers provide us with an excellent history of these QTL-trait associations beginning with Champoux et al. (1995) and following through the many and varied physio-mophological traits mapped by Zhang et al (1999) and Zhang et al. (2001). The creation and international deployment of a common experimental genetic population among researchers with related research objectives has promoted international collaboration and spurred rapid progress in identifying and confirming QTL of interest.

These two studies (Babu et al. 2003 and Lanceras et al. 2004) confirm much of the previous identity/location of drought tolerance QTLs in rice, especially those linked to secondary traits. In addition, and as is to be expected, they show new and independent QTL identification specific to their location specific soil physical-chemical and crop weather conditions. However, what is of interest to this author is that both groups, working in Southern India and Northeast Thailand, located common QTL for biomass and yield and yield components under drought conditions and over several crop seasons/irrigation treatments in the same intervals on chromosomes 3, 4, and 9 (Figure 5).

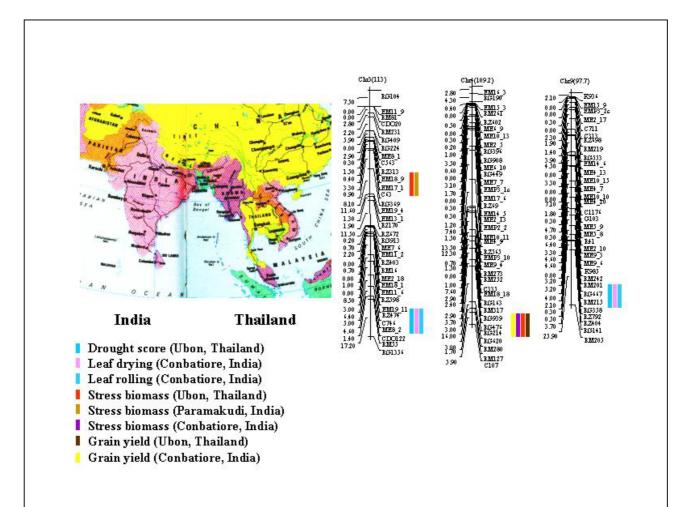


Figure 5. This simplified representation, adapted from the unpublished data of R. C. Babu and T. Toojinda (personal communication), illustrates the congruence found in visual drought scoring parameters and the salient traits biomass and yield under stress treatments in two distant environments--- Southern India and Northeast Thailand. Trait QTL intervals of interest are represented by the colored bars next to molecular marker maps of rice chromosomes 3, 4 & 9. (Personal communication R. C. Babu and T. Theerayut and Babu et al. 2003; Lanceras et al. 2004)

Another research group in India has focused primarily on root system parameters and have pursued QTL and their molecular markers for nearly ten years (Hemamalini, et al. 2000; Venuprasad et al. 2002). They are currently close to combining root traits and leaf blast disease resistance in a commercially acceptable background (Shashidhar et al. 2004). These three research groups, using a number of different mapping populations and screening protocols have come to the conclusion that they can use from one to five QTL markers to select, via MAS, for a specific root system trait (maximum root system length) or yield performance in drought-prone rainfed or poorly irrigated target environments. At this time, the three groups are actively conducting experiments to unequivocally

test the effectiveness of individual and pyramided QTL to select for drought tolerance in their specific target environments.

An independent effort to search for drought tolerant QTL and introgress them into commercial varieties was initiated by scientists at the International Rice Research Institute in 1998 entitled the "International Rice Molecular Breeding Program" (Yu et al. 2003). Although not originally focused on drought tolerance, as confidence grew in the dry-season field screening to reliably select drought tolerant backcross families (Lafitte et al. 2002; Lafitte et al. 2004) drought tolerance became one of the major traits of interest. At present 17 QTL have been identified in relation to severe reproductive stage field screening for drought tolerance (Zheng et al. 2004). Like the Indian and Thai groups, these Chinese researchers intend to pyramid QTL of greatest significance into commercially desirable genetic backgrounds. Their goal is to combine both high yield potential and drought tolerance in new rice varieties suited for China's current and future water-limited production systems.

One of the continuing problems with quantitative trait introgression by MAS is the shear magnitude of the task of selecting for 3-5 QTL markers in a breeding program. When this is coupled with the relatively low proportion of the variance accounted for by any single QTL, many are skeptical of the potential for effective MAS of drought tolerance in rice. Regardless of the "interesting" and "provocative" results to date, the efficacy of MAS for drought tolerance in rice is still a work in progress. Nevertheless, the broad interest in both the genetic control of drought tolerance and location of related QTLs is already contributing to identification and map-based cloning of candidate gene(s) involved in this complex phenomenon.

## Future scenarios

DNA Molecular Marker Assisted Selection & Genetic Engineering for Enhanced Drought Tolerance

This section will not delve deeply into the full spectrum of biotechnology - bioinformatics tools available today and their potential role in rice breeding for drought tolerance. Instead I refer the reader to an excellent comprehensive review by Bennett (2003) in which he appraises their role in plant breeding for increased "water productivity". I will limit this section to a continuation of the discussion initiated in the previous section on MAS and a brief extension to the role of genetic engineering for drought tolerance with regard to rice only.

Experimental pyramiding of "major QTLs" is already underway, however it is still an open question whether this approach is viable in large breeding programs due to expense (Dreher et al. 2003) and strong genotype X environment interactions of some important QTL (Kamoshita et al. 2002b; Lanceres et al. 2004). Should

any of the "major" QTL now being researched prove useful in a particular target environment of scale or of economic significance, we may see limited use of MAS in the later stages of breeding programs after other more conventionally screened traits, or those with single gene/QTL selection protocol (diseases, insects, quality), have already resulted in highly adapted superior breeding lines.

Significantly, the search for drought tolerance QTL and subsequent fine mapping mean that given the readily available rice genomics resources and bioinformatics platforms available today, the search for candidate genes can be rapidly expedited. This leads directly to the proposition of transferring and effecting regulation of key genes/alleles such as those controlling stress induced signaling cascades or transcription activators into adapted commercial varieties. The determination of the new transgenic rice plant's expression of enhanced "drought tolerance" in whole plant and/or crop level systems--- brings our discussion full circle. In the earlier parts of this paper the paramount need for high quality phenotyping/screening was noted as perhaps the major impediment to success in conventional and/or MAS breeding. Transgenic rice with experimental promoters and genes of interest will be no less in need of unequivocal phenotypic evaluation for drought tolerance. At the risk of being repetitive, transgenics will require rigorous assessment of the wild type and transgenic plants under high quality science-based "managed stress environments" protocols. If this extra effort or collaboration is not conducted, journal photos of seedlings in growth chambers and greenhouses will continue to plague the interpretation of "drought tolerance" claims from plant molecular biology laboratories.

Finally, what about Arabidopsis and rice? The extraordinary amount and quality of recent outputs on physiology, biochemistry, genetics and molecular biology of abiotic stress response of Arabidopsis genotypes is a rich resource available to rice researchers. A very timely publication, "Arabidopsis Special Issue: Translational Biology" (Plant Physiology, June 2004, Vol 135, No. 2) provides several review articles addressing this topic. Rensink and Buell (2004) review progress with rice functional genomics in light of the lessons learned from Arabidopsis that have paved the way for more efficient and rapid progress on the sequencing and analysis of the rice genome. On the subject of drought tolerance, Rensink and Buell illustrate significant corollaries between rice and Arabidopsis. They demonstrate the utility of using Arabidopsis information as a knowledge base for rice research by citing the work of Rabbani et al. (2003). In Rabbani et al. (2003), analysis of microarray expression profiling identified 73 abiotic stress inducible genes in rice, 51 of which had already been observed in Arabidopsis. This type of confirmation builds confidence and demonstrates the value of consulting the Arabidopsis databases to guide rice research, and through synteny relationships among genomes, that of all major cereals.

In the same special issue Zhang et al. (2004) provide an indicative review of the recent literature on cold and drought stress research in Arabidopsis. Their discussion focuses on progress in abiotic stress tolerance with emphasis on

applications in genetic improvement of crop plant species. Although the literature is rapidly advancing with "promising" reports of "increased drought tolerance" from constitutive expression and increasingly stress inducible or organ specific expression of a large number of "candidate genes", these authors look forward to the efforts being extended to crop species in realistic evaluations. To this author's knowledge, testing *in planta* of numerous "candidate genes" derived from: consensus mapping of rice drought tolerance QTLs; Arabidopsis signal transduction and functional genomics research; and a combination of the above from bioinformatics-based modeling *in silico*, are now under way in both public (China) and private sectors (India) of the Asian rice world. In both cases, the efficacy of the transgenic rice events produced will depend on the quality and perception of field-level efficacy testing. The first results are expected in 2-4 years in China (Xiao et al. 2004).

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# IV. The Challenges Ahead

The primary challenge before us is to <u>sustain the momentum</u> of current activities. This will require effective communication of "the problem" and commitment of financial resources for a decade or more. To achieve this outcome we must not limit our attention to the scientific arena, but become more proactive as scientists in the public awareness, policy and decision-making arenas of the media and government. Political time frames are notoriously short and yet the process of rice breeding that we are now involved in will require a decade or more of sustained support to reach the ultimate success. Currently campaigns in India and China are in progress to raise public awareness. They intend to call state/provincial and national attention to the water-rice conundrum facing their nations and emphasize the existence of new facilities and accompanying expertise with which to meet the farmer's need for new varieties in a water-limited future.

With regard to the physiological or molecular biology challenges, there is one trait that has proven to be extremely difficult to address. When we recall the goal of decreasing nonproductive (extrastomatal) water loss from the shoot, it is increased cuticular resistance to water vapor loss from the shoot system, (water proofing the rice plant) which has proven recalcitrant in research efforts. The parameter used as a proxy for selection, the amount of leaf surface wax, demonstrates a strong genotype X environment interaction while direct measurement of leaf diffusive resistance to water vapor has not been reliable with regard to sources of genotypic variation. Given this level of phenotyping difficulty and trait plasticity, the answer may lie in genetically engineering greater resistance to water vapor flow through the shoot's epidermis. In keeping with the previous discussion, perhaps the key to this goal may reside in the array of

Arabidopsis mutants and related information base on molecular biology and control of genes in the leaf surface wax biosynthesis pathways?

The End September 28, 2004 Bangkok, Thailand

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