

CONTENIDO DEL INFORME TÉCNICO

FIA-PR-V-2006-1-A-006 IT

Fecha de entrega del Informe

21 Julio 2006

Nombre del coordinador de la ejecución

Patricio Arce-Johnson

Firma del Coordinador de la Ejecución

Patricio Arce Johnson

1. ANTECEDENTES GENERALES DE LA PROPUESTA

Nombre de la propuesta

Taller Internacional sobre Desarrollo de Tolerancia al Estrés Abiótico en Plantas

Código

FIA-CD-V-2006-1-A-006 - FIA-PR-V-2006-1-A-006

Entidad responsable

Pontificia Universidad Católica de Chile

Coordinador(a)

Patricio Arce-Johnson

Tipo de Iniciativa(s)

☐

Gira

☐

Beca

X Evento

☐

Consultores

☐

Documentos

Fecha de realización (inicio y término)

19 -21 de Junio 2006

2. RESUMEN DE LA PROPUESTA

Resumir en no más de una página la justificación, actividades globales, resultados e impactos alcanzados con la propuesta completa. Cuando exista más de una iniciativa, cada una de ellas debe ser resumida en forma específica. Estos resúmenes deben sintetizar los aspectos principales de la propuesta y cada una de sus iniciativas en forma general.

GLOBAL (Completar solo cuando existe más de una iniciativa)

GIRA TECNOLÓGICA

BECAS

CONSULTORES

EVENTOS

Antecedentes

Para enfrentar el desafío de alimentar a 3000 millones de personas más que habitarán la tierra en el año 2050, la agricultura mundial necesita usar todas las formas de aumentar la productividad en forma sustentable. Obviamente que una de las posibilidades que se deben considerar es de usar terrenos que actualmente no se cultivan por estar en climas o condiciones que no permiten su aprovechamiento. Sin embargo, la ciencia conoce de plantas que crecen y se desarrollan en condiciones extremas de aridez, salinidad, toxicidad o temperaturas. Esto quiere decir que la evolución ha podido encontrar adaptaciones genéticas que le permiten a esas plantas tolerar condiciones que normalmente no son adecuadas para su desarrollo fisiológico.

En Chile, tenemos una enorme extensión de zonas áridas o semiáridas, de terrenos con alta salinidad o con altas concentraciones de metales tóxicos y también tenemos regiones en las que las bajas temperaturas afectan el crecimiento vegetal. Adicionalmente, el cambio climático y calentamiento de la tierra está generando severos trastornos, cuyo efecto en la flora y fauna son inciertos. Por esta razón, el desarrollo de especies de interés agrícola resistentes al estrés abiótico sería de alta relevancia para el país.

En el país se han desarrollado o se están iniciando algunas iniciativas en el ámbito del estrés abiótico en plantas, en las que destaca la Universidad Católica de Chile, Universidad de Chile, Universidad de la Serena, Universidad de Tarapacá e INIA. Actualmente, uno de estos proyectos sobre desarrollo de cítricos con mayor tolerancia a la salinidad para la 3ª Región, cuenta con la asesorías del ICGB y la Universidad de

Cornell.

El ICGEB y la Academia de Ciencias del Mundo en Desarrollo (TWAS) han decidido propiciar un Programa Conjunto para impulsar la investigación en biotecnología de plantas relacionada con el estrés abiótico. Este programa abrió un concurso de ideas de proyecto (Letters of Intent) al que pueden acceder países en desarrollo. Una vez seleccionados los mejores pre-proyectos, los coordinadores de dichos proyectos serán invitados a participar en un Taller Internacional (Coordination Workshop), junto con investigadores en estos temas provenientes de países de bajo desarrollo científico-tecnológico para promover su participación en dichos proyectos. A ese Simposio también se invitará a 6-8 autoridades mundiales en el tema de resistencia a estrés abiótico. Posterior al Simposio se invitará a los mejores (20-25) pre-proyectos a presentar proyectos "in extenso" de entre los cuales se financiarán 5 con un aporte total de aproximadamente US\$ 60.000 cada uno por 3 años. Se pretende que estos 5 proyectos se constituyan en una Red de Investigación en este tema en el mundo en desarrollo y que sirvan de núcleo para capacitación de postgrado y que también puedan presentarse a otras instancias de financiamiento.

Con esta iniciativa, Chile ha sido elegido como sede para la realización del Taller Internacional sobre Estrés Abiótico en Plantas, gracias al apoyo de ICGEB, TWAS y distintas entidades y universidades chilenas. Al ser Chile sede de este Taller, nuestro país y sus investigadores en este campo pasarían a ocupar un lugar de liderazgo en un tema de gran relevancia nacional e internacional.

DOCUMENTOS TÉCNICOS

3. ALCANCES Y LOGROS DE LA PROPUESTA GLOBAL

Problema a resolver, justificación y objetivos planteado inicialmente en la propuesta

Al ser esta iniciativa una actividad de difusión científica y de colaboración nacional e internacional, los problemas a resolver se amplían a una realidad global que se encierra en problemas como la escasez de suelos cultivables, el análisis de los posibles cambios climáticos que se han producido, los efectos en las plantas y la posibilidad de utilizar la genética y biotecnología como una herramienta de mejora y los impactos que estos producirían para el sector agrícola nacional e internacional, destacando además el problema social que esto implicaría. Por lo tanto nosotros vemos como necesaria la convocatoria de importantes representantes del ámbito científico nacional e internacional debatiendo estos temas. Chile, debido a su geografía, no puede mantenerse alejado de esta temática, presentando cada vez más interés para científicos y Universidades, las cuales han tratado (con esta iniciativa) de reunir a todos los interesados en el tema de estrés abiótico en plantas y posicionar a Chile en un lugar competitivo en esta área.

El objetivo central de la iniciativa busca fomentar el estudio y la investigación sobre los factores genéticos y fisiológicos que otorgan tolerancia al estrés abiótico en plantas de relevancia económica e impulsar la colaboración internacional en estas investigaciones.

Objetivos alcanzados tras la realización de la propuesta

Con la realización de esta propuesta se logró difundir en el medio nacional la problemática mundial que existe frente al Estrés Abiótico en Plantas y cómo este problema podría relacionarse con la investigación científica y la agro-economía nacional. Se logró reunir importantes autoridades nacionales e internacionales, además de un importante contingente científico que, mediante a mesas redondas, pudo interactuar entre sí y con las autoridades y científicos internacionales invitados. Se logró fomentar la importancia del estudio de los factores genéticos y fisiológicos que otorgan tolerancia al estrés abiótico en plantas, en donde la investigación por parte de las Universidades mostró un nivel importante. Además se logró entender que los avances en la investigación científica, en el descubrimiento de nuevos factores genéticos y procesos fisiológicos ayudarían a resolver problemas agro-económicos de gran importancia para el país.

Resultados e impactos esperados inicialmente en la propuesta

Con esta iniciativa se buscaba promover la investigación de procesos biológicos y moleculares involucrados en la tolerancia frente al estrés abiótico en plantas, para lo cual el contacto con investigadores de otros países permitió ampliar los conocimientos, además la interacción con distintas autoridades, permitieron realzar la importancia de financiar y apoyar proyectos relacionados a estos temas. Se esperaba que la convocatoria reuniera a diversos investigadores y autoridades y que plantearan dudas y proyectos para avanzar en el tema.

Resultados obtenidos

Descripción detallada de los conocimientos y/o tecnologías adquiridos y/o entregados. Explicar el grado de cumplimiento de los objetivos propuestos de acuerdo a los resultados obtenidos. Para consultorías es necesario anexar el informe final del consultor.

Luego de la realización del Taller Internacional de Estrés Abiótico en Plantas, se puede decir que concluyó con éxito esta actividad, gracias al apoyo de las Universidades participantes, ICGEB, TWAS, FDF, FIA, entre otras. Se logró convocar a destacados investigadores internacionales, los cuales presentaron sus investigaciones en esta área y su visión frente al problema de estrés abiótico en plantas. También se logró convocar a importantes investigadores y autoridades nacionales, las que mostraron la problemática nacional en este ámbito y los avances que se están gestando en este sentido. Finalmente, se lograron crear colaboraciones entre investigadores y se entendió que la conexión entre la problemática de estrés abiótico en plantas y la investigación científica, es necesaria para el avance científico y económico, para el país.

Resultados adicionales

Describir los resultados obtenidos que no estaban contemplados inicialmente como por ejemplo: formación de una organización, incorporación de alguna tecnología, desarrollo de un proyecto, firma de un convenio entre otros posibles.

Todos los resultados, estaban dentro del marco esperado, ya que era una actividad de difusión y de exposición de temas relacionados con los avances en ciencia y tecnología frente al estrés abiótico en plantas. Se superaron las expectativas en cuanto a la convocatoria y a la participación de personas interesadas, y se logró crear en Chile una conciencia frente a la necesidad e importancia que tiene el fomentar el estudio y la investigación frente a los mecanismos moleculares que regulan y permiten la tolerancia frente a distintos tipos de estrés abióticos en plantas.

Aplicabilidad

Explicar la situación actual del sector y/o temática en Chile (región), compararla con las tendencias y perspectivas presentadas en las actividades de la propuesta y explicar la posible incorporación de los conocimientos y/o tecnologías en el corto, mediano o largo plazo, los procesos de adaptación necesarios, las zonas potenciales y los apoyos tanto técnicos como financieros necesarios para hacer posible su incorporación en nuestro país (región).

La Pontificia Universidad Católica de Chile en conjunto con ASOEX y FDF, han constituido una sociedad anónima en el Consorcio Tecnológico de la fruta. Consorcio que tiene como objetivo general el desarrollo de nuevas variedades para la industria frutícola chilena. Adicionalmente, estas mismas instituciones participan en un Proyecto CORFO - Innova Chile sobre el desarrollo de portainjertos de cítricos tolerantes a la salinidad, el que está en el marco de la presente propuesta. El Dr. Jorge Allende (uno de los gestores de esta iniciativa) profesor de bioquímica de la Universidad de Chile, es consultor científico de FDF y participa en este carácter en todas las actividades vinculadas al sector frutícola. Chile es país miembro del ICGEB y el representante en la junta de gobernadores de esta

institución es el Dr. Jorge Allende, quién está activamente apoyando este Taller Internacional sobre estrés abiótico en plantas. Adicionalmente, nuestro país cuenta con más de 15 científicos miembros del TWAS. Actualmente, el Dr. Jorge Allende es Vicepresidente de esa academia de ciencias para el tercer mundo en representación de América Latina. Estos antecedentes, apoyaron de manera significativa que nuestro país haya sido el anfitrión de este importante evento internacional sobre estrés abiótico en plantas. Lo que proyecta a Chile en un país competitivamente importante para la realización de proyectos de investigación y para la formación de científicos en el área de estrés abiótico en plantas.

Detección de nuevas oportunidades y aspectos que quedan por abordar

Señalar aquellas iniciativas que surgen como vías para realizar un aporte futuro para el rubro y/o temática en el marco de los objetivos iniciales de la propuesta, como por ejemplo la posibilidad de realizar nuevas actividades.

Indicar además en función de los resultados obtenidos los aspectos y vacíos tecnológicos que aun quedan por abordar para ampliar el desarrollo del rubro y/o temática.

4. ASPECTOS RELACIONADOS CON LA EJECUCIÓN DE LA PROPUESTA

Programa Actividades Realizadas

Nº	Fecha	Actividad	Iniciativa
1	19-21 Junio 2006	Taller Internacional sobre Estrés Abiótico en Plantas	Evento de difusión

Detallar las actividades realizadas en cada una de las Iniciativas, señalar y discutir las diferencias con la propuesta original, y rescatar lo más importante de cada una de ellas. Por ejemplo, en el caso de Giras, discutir las actividades de cada visita, Becas, analizar las exposiciones más interesantes, Consultores, detallar el itinerario y comentarios del consultor, Eventos, resumir y analizar cada una de las exposiciones, y Documentos, analizar brevemente los contenidos de cada sección.

GIRAS

BECAS

CONSULTORES

EVENTOS

El Taller Internacional sobre Estrés Abiótico en Plantas, se realizó del 19 al 21 de Junio del 2006 en el centro de Extensión de la Pontificia Universidad Católica de Chile. Esta iniciativa se realizó a través de actividades de difusión (conferencias y charlas) y de discusión (mesas redondas).

Las actividades realizadas fueron las siguientes (se adjunta el programa del Taller, con el detalle de las sesiones) :

Día 1 (19 Junio) : Se hizo inicio a las actividades con la inauguración del Taller, para la cual se contó con la presencia de autoridades de la PUC, del Gobierno y de las instituciones participantes (TWAS, ICGEB, FDF, FIA, ASOEX, CORFO, RELAB). Luego se siguió con las conferencias del Dr. Marc Van Montagu, del Dr. Luigi Cattivelli y con las sesiones I, II y III de los participantes ICGEB-TWAS, en donde se presentaron distintas propuestas y proyectos relacionados con el tema.

Día 2 (20 Junio) : Se realizaron las conferencias del Dr. Sudhir Sopory y del Dr. Roberto Gaxiola. También, se realizó la IV sesión de los participantes ICGEB-TWAS y un simposio que contó con participaron distintos investigadores nacionales, lo que abordaron el tema de "Investigación en estrés abiótico de planas en Chile". Finalmente se concluyó con la exposición de algunos investigadores invitados (postulantes) para discutir temas sobre "estrategias de investigación en biotecnología de plantas, en países en desarrollo" .

Día 3 (21 junio) : Se realizaron dos actividades en paralelo. Una de las actividades reunió a autoridades nacionales (FIA, CORFO, FONDEF-CONICYT) junto a investigadores nacionales y representantes del sector empresarial y se discutieron temas relacionados con la posibilidad de financiamiento y apoyo para la investigación en áreas de estrés abiótico en plantas. La otra actividad reunió a las autoridades y representantes de ICGEB-TWAS, para evaluar las actividades de los días anteriores y sacar conclusiones sobre los proyectos a potenciar en esta área, en países en desarrollo.

DOCUMENTOS

Contactos Establecidos

Presentar los antecedentes de los contactos establecidos durante el desarrollo de la propuesta (profesionales, investigadores, empresas, etc.) de acuerdo al siguiente cuadro

Institución Empresa Organización	Persona de Contacto	Cargo	Fono/Fax	Dirección	E-mail

Material elaborado y/o recopilado

Entregar un listado del material elaborado, recibido y/o entregado en el marco de la propuesta. Se debe entregar adjunto al informe un set de todo el material escrito y audiovisual, ordenado de acuerdo al cuadro que se presenta a continuación.

También se deben adjuntar fotografías correspondientes a la actividad desarrollada. El material se debe adjuntar en forma impresa y en un medio electrónico (disquet o disco compacto).

Elaborado

Tipo de material	Nombre o identificación	Preparado por	Cantidad

Recopilado

Tipo de Material	Nº Correlativo (si es necesario)	Caracterización (título)
Artículo		
Foto		
Libro		
Diapositiva		
CD		

Programa de difusión de la actividad

En esta sección se deben describir las actividades de difusión de la actividad, adjuntando el material preparado y/o distribuido para tal efecto.

En la realización de estas actividades se deberán seguir los lineamientos que establece el "Instructivo de Difusión y Publicaciones" de FIA, que le será entregado junto con el instructivo y formato para la elaboración del informe técnico.

5. PARTICIPANTES DE LA PROPUESTA

GIRAS, BECAS: Ficha de Participantes

CONSULTORES: Ficha de(l) Consultor(es)

EVENTOS: Ficha de Expositores y Organizadores

DOCUMENTOS: Ficha de Autores y Editores

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E-mail	parce@bio.puc.cl
Nombre de la organización, empresa o institución donde trabaja / Nombre del predio o de la sociedad en caso de ser productor	Pontificia Universidad Católica de Chile
RUT de la organización, empresa o institución donde trabaja / RUT de la sociedad agrícola o predio en caso de ser agricultor	81.698.900-0
Cargo o actividad que desarrolla	Profesor Adjunto
Rubro, área o sector a la cual se vincula o en la que trabaja	Investigación científica

5. PARTICIPANTES DE LA PROPUESTA

GIRAS, BECAS: Ficha de Participantes

CONSULTORES: Ficha de(l) Consultor(es)

EVENTOS: Ficha de Expositores y Organizadores

DOCUMENTOS: Ficha de Autores y Editores

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Apellido Materno	
RUT Personal	
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Nombre de la organización, empresa o institución donde trabaja / Nombre del predio o de la sociedad en caso de ser productor	Universidad de Chile
RUT de la organización, empresa o institución donde trabaja / RUT de la sociedad agrícola o predio en caso de ser agricultor	
Cargo o actividad que desarrolla	
Rubro, área o sector a la cual se vincula o en la que trabaja	Investigación científica

5. PARTICIPANTES DE LA PROPUESTA

GIRAS, BECAS: Ficha de Participantes

CONSULTORES: Ficha de(l) Consultor(es)

EVENTOS: Ficha de Expositores y Organizadores

DOCUMENTOS: Ficha de Autores y Editores

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Apellido Materno	
RUT Personal	5.220.772-K
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Fono y Fax	231 6094 Fax: 231 7270
E-mail	direccion@fdf.cl
Nombre de la organización, empresa o institución donde trabaja / Nombre del predio o de la sociedad en caso de ser productor	Fundación para el Desarrollo Frutícola -FDF
RUT de la organización, empresa o institución donde trabaja / RUT de la sociedad agrícola o predio en caso de ser agricultor	72.173.800-0
Cargo o actividad que desarrolla	Director general
Rubro, área o sector a la cual se vincula o en la que trabaja	Agrícola

Participantes en actividades de difusión

Es necesario registrar los antecedentes de todos los asistentes que participaron en las actividades de difusión. El listado de asistentes a cualquier actividad deberá al menos contener la siguiente información:

Nombre	
Apellido Paterno	
Apellido Materno	
RUT Personal	
Dirección, Comuna y Región	
Fono y Fax	
E-mail	
Nombre de la organización, empresa o institución donde trabaja / Nombre del predio o de la sociedad en caso de ser productor	
RUT de la organización, empresa o institución donde trabaja / RUT de la sociedad agrícola o predio en caso de ser agricultor	
Cargo o actividad que desarrolla	
Rubro, área o sector a la cual se vincula o en la que trabaja	

6. EVALUACIÓN DE LA PROPUESTA

Evaluación de la actividad para cada INICIATIVA

En esta sección se debe evaluar la actividad en cuanto a los siguientes ítems:

a) Efectividad de la convocatoria (cuando corresponda)

Se logró reunir a autoridades e investigadores internacionales de alto nivel, además de autoridades e investigadores nacionales, relacionados con las temáticas a abordar.

b) Grado de participación de los asistentes (interés, nivel de consultas, dudas, etc)

Tanto las conferencias de los investigadores internacionales, como las mesas de conversación tuvieron gran convocatoria por parte de los invitados y del público en general.

c) Nivel de conocimientos adquiridos por los participantes, en función de lo esperado (se debe indicar si la actividad contaba con algún mecanismo para medir este punto y entregar una copia de los instrumentos de evaluación aplicados)

Se logró dar una visión completa sobre el tema de estrés abiótico en plantas a nivel mundial. No se realizó ninguna evaluación posterior.

d) Problemas presentados y sugerencias para mejorarlos en el futuro (incumplimiento de horarios, deserción de participantes, incumplimiento del programa, otros)

No hubo problemas de consideración.

Aspectos relacionados con la postulación al programa de Captura y Difusión

a) Información recibida por parte de FIA para realizar la postulación

☒ amplia y detallada

☐ aceptable

☐ deficiente

Justificar:

b) Sistema de postulación al Programa de Formación o Promoción (según corresponda)

☐ adecuado

☒ aceptable

☐ deficiente

Justificar:

c) Apoyo de FIA en la realización de los trámites de viaje internacionales (pasajes, seguros, otros) (sólo cuando corresponda)

☐ bueno

☐ regular

☐ malo

Justificar:

d) Recomendaciones (señalar aquellas recomendaciones que puedan aportar a mejorar los aspectos administrativos antes indicados)

7. Conclusiones Finales de la Propuesta Completa

En el caso de Giras Tecnológicas, en lo posible presentar conclusiones individuales por participante.

La realización del Taller Internacional sobre Estrés Abiótico en Plantas, fue una actividad positiva para el país, por haber sido una instancia de reunión, difusión y conversación entre investigadores y autoridades nacionales e internacionales, frente a un tema común y de gran interés para nuestro país.

I NVITED FOREIGN EXPERTS

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COL - didnot attend

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RESUMENES DE LAS CHARLAS REALIZADAS

Genomic approaches to dissect the molecular bases of cold and drought tolerance in barley and wheat

luigi cattivelli

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The molecular dissection of the abiotic stress response has revealed a complex situation, where the co-ordinated expression of many stress-related genes is associated with resistance. The lecture will present data obtained from several genomics studies focussed on the understanding of the genetic/molecular bases of cold and drought resistance in barley and wheat.

QTL mapping in segregant populations has led to the identification of few major QTLs controlling frost resistance in barley/wheat and the comparison of mapping data with the results deriving from expression studies has allowed the identification of several candidate genes (*Chf* and others) involved in the genetic bases of frost resistance.

Affymetrix microarray and Real Time Q-PCR have been used for analysis of gene expression in response to drought in wheat cultivars with contrasting level of stress resistance and in response to cold in barley chloroplast mutants showing a complete susceptibility to frost. We have shown that barley plants carrying a mutation preventing chloroplast development, beside the expected *albino* phenotype, are completely frost susceptible as well as impaired in the expression of several *cor* (cold-regulated) genes.

Based on the evidence that some molecular mechanisms involved in stress tolerance are conserved between species also phylogenetically distant, we developed a functional genomic study to define the role of several wheat genes involved in cold/drought response through the identification and the analyses of *A. thaliana* homozygous T-DNA knock-out lines carrying an insertion in sequences homologous to the genes isolated in response to stress in wheat. A physiological characterization of the insertional lines will be presented.

Development of Drought Tolerant Transgenic Cassava Varieties

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Abstract

Cassava (*Manihot esculenta*) is grown throughout tropical Africa, Asia, and the Americas for its starchy storage roots and it feeds an estimated 600 million people each day, being now the most important source of dietary calories in the tropics after rice and maize. Farmers choose it for its high productivity and its ability to withstand conditions in which other crops fail. Cassava is increasing its importance in the economy of most of the Brazilian northeastern states. Besides its traditional use as food and fodder, cassava demand from the poultry and bakery industries is increasing, especially due the high prices of maize and wheat flour. The need to increase production to match this increase in demand, have highlighted the need for more productive genotypes, adapted to the prevailing agro-ecological settings in the northeastern states. Until recently, tissue culture techniques (meristem culture, axillary proliferation, somatic embryogenesis, organogenesis, etc.) and genetic transformation protocols for the cassava genotypes presently used in the Northeast Brazil have not been developed. In order to meet this need, my laboratory have worked towards achieving these goals and we have succeeded in developing protocols for the regeneration (both by somatic embryogenesis and organogenesis) of ten major cassava genotypes and we have demonstrated the transient expression of reporter genes in several of these cultivars. We want now to use the experience we have acquired in the techniques for tissue culture and genetic transformation of cassava, to explore the possibility of obtaining transgenic cassava varieties with improved tolerance to drought stress. Although cassava is a drought-tolerant crop, yields are negatively affected by this stress. We will use friable embryogenic calli obtained from 2 Brazilian and 2 Nigerian genotypes of cassava, as targets for transformation with a genetic construction consisting of the gene coding for the transcription factor DREB1A that specifically interacts with drought responsive elements (DRE) and induces expression of stress tolerance genes. This project will be developed within the scope of a collaboration involving two laboratories in Brazil (Fortaleza and Brasília), one laboratory in Nigeria, and one laboratory in the USA.

Ahmed Nada

**ISOLATION AND CHARACTERIZATION OF SALT RELATED GENE(S)
FROM MANGROVE**

Abstract

Wherever they grow in large regions of the earth, such as the arid zones, and lands with high salts, plants are subjected to a great variety of stresses tending to restrict their chance of development and survival. Salt stress is one of the most serious environmental factors limiting the productivity of the crop plants. Salinity imposes ionic stress, osmotic stress, and secondary stresses such as nutritional disorders and oxidative stress. This will lead to whole plant senescence and plant death. According to Food and Agriculture Organization (FAO) Seven percent of the land surface area is affected by salinity and fifty percent of cultivated lands are affected by salinity. Egypt has practically no rain and its agriculture depends on irrigation from the Nile river. In Egypt The major part of the country is covered by desert, arable land is very limited. On the other hand Eritrea is a mostly arid mountainous country with dramatic degree of land degradation. A century ago, about 30% of the total land area of Eritrea was covered by forest. This figure dwindled to 11% in 1952 and in 1960 it was estimated to be 5%. Drought and salinity are the biggest problems which faced Cultivation. Thus, production of salt tolerance plants are necessary for cultivation of new reclaimed land. Biotechnology is considered as a method for overcoming this obstacle and fighting against desertification. Recombinant DNA techniques not only allow the identification, mapping and purification of some genes but also permit isolation of genes from plants which can adapt the saline conditions (Halophytes) and their introduction into genome of another organism. This study will focus on identification of gene expression patterns in halophytes (mangroves) that will help us to reveal the molecular mechanisms of salt tolerance in halophytes and also to isolate and characterize these gene(s) like transcription factors. These gene(s) can be used to transform plants of economical value to help them to survive under highly saline conditions. These transgenic plants can help in the breeding and land reclamation programs.

Identification of key genes involved in drought and salt stress tolerance in the model plants

Physcomitrella patens and *Prosopis strombulifera*

Dra. Sabina Vidal Macchi, Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias, Universidad de la República

Abiotic stress, especially the osmotic stress caused by drought, freezing or high salinity, is an important cause for suboptimal yields in agriculture. The understanding of the molecular mechanisms underlying stress tolerance in plants will provide aid in selection strategies within breeding programs as well as tools for biotechnological approaches, where the overproduction of key proteins could render a substantial improvement in stress tolerance.

This project aims at the characterization and identification of new genes involved in abiotic stress tolerance in two plant model systems: the moss *Physcomitrella patens*, and the halophyte *Prosopis strombulifera*.

P. patens has recently become a model plant to study gene function taking advantage of two characteristics not found in other plant models: high frequency of homologous recombination, which allows successful targeted gene disruption, and the haploid dominant stage of the plant life cycle, which facilitates the phenotypic characterization of mutants. Recent studies from Frank et al 2005 (*Planta* 220, 384-394) and from our laboratory (Saavedra et al, 2006, *Plant J.* 45, 235-249) demonstrated that this plant exhibited a high tolerance against drought, salt and osmotic stress. Our studies implicated a fundamental role of a dehydrin-like gene (*PpDHNA*) from *P. patens* in stress tolerance. A knockout mutant of this gene was severely impaired in osmotic and salt-stress recovery, providing the first direct genetic evidence in any plant species for a dehydrin exerting a protective role during cellular dehydration. Taken together, these results make *P. patens* an excellent model to study abiotic stress adaptation in plants.

P. strombulifera is a salt tolerant legume that has optimal growth conditions in up to 500 mM NaCl. The group of Dr. Virginia Luna has been able to perform a thorough study on the physiological responses of *P. strombulifera* in different salt and osmotic stress conditions and their results indicate that this species constitutes an excellent model for studying salt tolerance in halophytes.

While a substantial amount of information is present concerning the regulation of stress-induced genes, little is known about the function of the corresponding proteins in stress tolerance. Consequently, the cloning and functional analysis of a set of osmotic-stress induced genes from two stress tolerant plants will extend the possibilities to generate targets for genetic engineering stress tolerance in crop species.

The overall objective of this research proposal is to identify key components of the molecular mechanisms underlying osmotic stress tolerance in *P. patens* and in *P. strombulifera*, with the long-term goal of generating tools for the development of strategies for possible crop yield improvement.

Specifically, we aim to:

a) Identify genes induced during salt and osmotic stress in these two species; b) Analyze the contribution to osmotic stress tolerance of the identified genes in *P. patens* by targeted gene disruption; c) Analyze the correlation of the expression pattern of the identified genes in *P. strombulifera*, with the hormonal (abscisic acid) and stress responses to extreme stress conditions; d) Perform functional studies of the selected genes by evaluating the abiotic stress tolerance of transgenic plants (initially *Arabidopsis*), overexpressing such sequences from *P. strombulifera* and *P. patens*; e) Assessing a possible protective role in higher plants of a *P. patens* dehydrin-like protein (DHNA), previously isolated and characterized in our laboratory.

The functional analysis of target genes and proteins will also allow to address fundamental questions in plant biology, such as the role of many response genes in stress protection or stress recovery in plants, the degree of conservation or the molecular responses between higher and lower plants, and will contribute to a deeper insight in extreme salt tolerance in halophytes.

This project is intended to be carried out in collaboration with Dr. Aldo Rojas's group (Biotechnology Program, Instituto Nicaraguense de Tecnología Agropecuaria, NICARAGUA) and Dr. Virginia Luna's group (Universidad Nacional de Río Cuarto, Facultad de Ciencias Exactas, Físicas y Naturales, Río Cuarto, ARGENTINA).

Root Engineering: A Strategy for Agriculture in Marginal Areas of Cultivation

Water availability is a major concern for agriculture in both developed and developing countries. Global water strategies focus on reducing overall agricultural use and increasing availability for human consumption. However, population growth and global warming are driving regional shifts in production and increased demand for irrigation. Improvement of plant water utilization is therefore critical. Some drought resistant plants develop deep and dense root systems. These natural adaptations suggest that manipulation of developmental mechanisms to enhance lateral root proliferation can be an effective strategy to engineer drought resistant plants. Despite their obvious role in water uptake, roots have not been targeted in genetic engineering strategies to improve crop performance under drought conditions.

We have shown that overexpression of the H^+ -pyrophosphatase (H^+ -PPase) AVP1 results in salt and water stress tolerant *Arabidopsis* plants (1). Both phenotypes were initially explained by an enhanced uptake of ions into their vacuoles. Significantly, further characterization of these AVP1 overexpressing plants revealed a dramatic enhancement of their root development with obvious implications for their ability to withstand drought. Moreover, root and shoot development in *avp1-1* loss-of-function mutants was impaired (2).

Our data indicated that although classically thought of as a tonoplast resident proton pump responsible for acidifying the vacuole, AVP1 also contributes to the regulation of apoplastic pH and to auxin transport, likely by mediating the trafficking of the PM P-ATPase and associated proteins, including PIN1 (2). Changes in intracellular auxin levels are also known to alter the expression of *P-ATPase* genes, establishing a feedback loop where AVP1 activity can regulate both targeting and level of the PM proton pump. Thus, in addition to its established role in the maintenance of vacuolar pH, our new data reveal a novel role for AVP1 in facilitating auxin transport and the regulation of auxin-related processes, such as root development (2). Of note, the high degree of identity at the amino acid level among the type I H^+ -PPases in the plant kingdom (3), together with the striking phenotypes that its overexpression produced in *Arabidopsis* (1, 2), suggested that the H^+ -PPase AVP1 could be a potential target for genetic engineering of root systems in agriculturally important crop plants.

To test if the water stress resistance phenotype triggered by the overexpression of AVP1 in *Arabidopsis* could provide a more universal strategy to improve the performance of crops under water deficit conditions, we engineered plants of a commercial cultivar of tomato (*Lycopersicon esculentum*) to express the *Arabidopsis* AVP1 H^+ -PPase. This approach resulted in a) greater pyrophosphate-driven cation transport into root vacuolar fractions, b) increased root biomass, and c) enhanced recovery of plants from an episode of soil water deficit stress (4). We concluded that more robust root systems allowed transgenic tomato plants to take up greater amounts of water during the imposed water deficit stress, resulting in a more favorable plant water status and less injury. This study documents a novel general strategy for improving drought resistance of crops. Furthermore, it is tempting to speculate that enhanced

development of root systems will also have a positive impact in plant's mineral nutrition. We are currently testing the latter hypothesis.

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Novel Ultraviolet-B resistant mechanisms revealed by Arabidopsis mutants

Yoshihiro Hase (Japan Atomic Energy Agency)

The UV-B is the most harmful radiation in sunlight. The damages to DNA have been recognized as an important consequence of exposure to UV-B light. Studies on many Arabidopsis mutants that are hyper-sensitive to UV-B light indicated that the DNA repair and sun screening pigment are the main factors involved in protection mechanism to UV-B light. In order to find other unidentified mechanisms to cope with UV-B light, we tried to isolate Arabidopsis mutant that show hyper-resistance or hyper-sensitivity towards UV-B light, using ion beams as a mutagen. To date, four lines of UV-resistant and six lines of UV sensitive mutants have been obtained. In this Workshop, I will talk about novel mechanisms to cope with UV-B light revealed by the analyses of these mutants.

1) *uvi1* (*UV insensitive 1*) mutant

The *uvi1* mutant shows hyper-resistance to UV-B light. The fresh weight of *uvi1* plants grown under supplemental UV-B light was around twice that of the wild type. We found that the increased resistance correlated with enhanced capacity for DNA repair. The photoreactivation for CPDs and dark repair for (6-4) photoproducts was faster than the wild type. The transcript level of *PHR1* encoding CPD photolyase was at higher level than in the wild type. Although the mutation has not been linked to the gene, these results suggest that the *UVI1* works as a negative regulator of these DNA repair pathways.

2) *uvi4* (*UV insensitive 4*) mutant

The *uvi4* mutant shows hyper-resistance to UV-B light. The DNA repair ability of *uvi4* was not significantly different from that of the wild type. The *UVI4* gene was mapped on the bottom of chromosome 2 and was found to encode a novel basic protein. We found that the *uvi4* mutant has increased ploidy level due to the stimulated endoreduplication. The hypocotyl cells of *uvi4* underwent one more round of endoreduplication and the leaf cells also showed increased ploidy level. Tetraploid Arabidopsis was hyper-resistant to UV-B compared to diploid Arabidopsis. These results suggest that the enhanced polyploidization is responsible for the increased UV-B tolerance of the *uvi4* mutant and that the ploidy is one of the important mechanisms to cope with UV-B light.

3) *suvi1* (*sensitive to UV I*)/*rev3-1*

The root elongation of *suvi1* mutant is inhibited by UV-B light in both light and dark condition. The DNA repair ability of *suvi1* was not significantly different from that of the wild type. The positional cloning revealed that the *suvi1* has a mutation in *AtREV3* that is a homologous gene of yeast REV3. The yeast REV3 is a subunit of DNA polymerase ζ (zeta) that is known to be involved in error-prone DNA translesion synthesis (TLS). The error-prone TLS is one of the damage-tolerance pathways, in that, the DNA damage is bypassed by specialized TLS-type DNA polymerases. A BrdU incorporation experiment showed that the DNA synthesis after UV-B exposure was highly repressed in *rev3-1/suvi1* mutant due to the disruption of TLS pathway. Our result is the first report showing the presence of TLS system in higher plants.

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Genomics approaches for improving drought, salt, and cold tolerance in citrus
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Brazil is the largest world producer of citrus fruit, with over 1 million hectares of citrus trees in its territory and fruit production at about 18 million tons. The country is also the main exporter of frozen concentrate orange juice (FCOJ), contributing with 53% of all FCOJ produced in the world. The production is concentrated at São Paulo State, located at Southeastern Brazil, which is responsible for about 80% of the oranges that Brazil produces. Northeastern Brazil is the second largest producer of citrus fruit, providing about 10% of the total nation's production mainly concentrated (90%) at Bahia and Sergipe States. The Northeastern citriculture has a great potential for implementing its growth especially by the absence of pests and diseases which are commonly found at Southeastern, including citrus canker and CVC. However, the main challenge is to overcome the constraints imposed by drought, since majority Northeastern areas are semiarid zones where the annual rainfall is less than 750 mm and it is concentrated at a period of two or three months. To directly address the drought stress problem confronting citrus production, we propose to employ the genomic approaches of EST sequencing, transcript profile, physical mapping, genetic engineering, and marker-assisted selection. Our research group is currently using such genomic approaches in order to improve citrus fruit quality (carotenoid content) and we could easily expand our analyses for primarily drought stress tolerance and further salt and/or cold stress tolerance depending on international collaboration. As result, we expect (i) to perform large-scale identification and mapping of citrus genes involved in drought, salt, and/or cold stress tolerance and (ii) to transfer them to otherwise acceptable citrus varieties that are deficient in such characteristics by genetic engineering and/or conventional breeding. Minimization of the abiotic stress effects in citrus based in genomics is the key for the expansion and sustainability of the citrus production world-wide.

An Agronomic View of Drought Resistance ^{1/}.

Edmundo Acevedo.

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SUMMARY

The agronomic view of drought resistance is usually related to the ability that a given crop species or genotype within a species has to yield under drought. Biomass production, hence carbon gain, under stress is central to this concept. In this work, the grain yield of a group of 100 wheat genotypes (*Triticum aestivum* L.) originating from CIMMYT, ICARDA and INIA grown in 10 environments (including Obregon in NW Mexico and Santiago, irrigated and droughted) will be presented. The environmental conditions fluctuated across environments. The mean yield of the genotypes varied from 6.9 to 3.4 T/Ha according to the environment. A strong genotype x environment interaction accounted for more than 34% of the yield variation observed. The yield potential varied among sites mainly due to differences in the photo thermal quotient during the ear growth period which in Obregon was $1.0 \text{ MJ m}^{-2}\text{d}^{-1}\text{C}^{-1}$ and in Santiago was $1.8 \text{ MJ m}^{-2}\text{d}^{-1}\text{C}^{-1}$. There was a significant association between PTQ and the yield potential ($r = 0.61^{***}$). The major variation in yield potential was due to number of grain per square meter and the grain filling rate which conformed a first principal component ($r = 0.84^{***}$). The genotypes originating from INIA had the higher mean yield potential. The differences in yield under water stress of the genotypes were associated to a principal component ($r = -0.86^{***}$) determined mainly by phenology (the number of days to anthesis and the number of days to physiological maturity). Most of the variation in yield under water stress was attributed to a residual yield index, associated significantly to biomass yield, number of grains m^{-2} and grain filling rate. The genotypes originating from ICARDA had the higher yield under water stress. One genotype having the highest yield stability along with high mean yield originated from CIMMYT. It was observed that ^{13}C discrimination, crop to air temperature difference and osmotic adjustment may assist in selecting genotypes under drought.

^{1/}Financial support for this work was obtained from CIMMYT, ICARDA and FONDECYT.

Identification of key genes involved in drought and salt stress tolerance in the model plants

Physcomitrella patens and *Prosopis strombulifera*

Dra. Sabina Vidal Macchi, Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias, Universidad de la República

Abiotic stress, especially the osmotic stress caused by drought, freezing or high salinity, is an important cause for suboptimal yields in agriculture. The understanding of the molecular mechanisms underlying stress tolerance in plants will provide aid in selection strategies within breeding programs as well as tools for biotechnological approaches, where the overproduction of key proteins could render a substantial improvement in stress tolerance.

This project aims at the characterization and identification of new genes involved in abiotic stress tolerance in two plant model systems: the moss *Physcomitrella patens*, and the halophyte *Prosopis strombulifera*.

P. patens has recently become a model plant to study gene function taking advantage of two characteristics not found in other plant models: high frequency of homologous recombination, which allows successful targeted gene disruption, and the haploid dominant stage of the plant life cycle, which facilitates the phenotypic characterization of mutants. Recent studies from Frank et al 2005 (*Planta* 220, 384-394) and from our laboratory (Saavedra et al, 2006, *Plant J.* 45, 235-249) demonstrated that this plant exhibited a high tolerance against drought, salt and osmotic stress. Our studies implicated a fundamental role of a dehydrin-like gene (*PpDHNA*) from *P. patens* in stress tolerance. A knockout mutant of this gene was severely impaired in osmotic and salt-stress recovery, providing the first direct genetic evidence in any plant species for a dehydrin exerting a protective role during cellular dehydration. Taken together, these results make *P. patens* an excellent model to study abiotic stress adaptation in plants.

P. strombulifera is a salt tolerant legume that has optimal growth conditions in up to 500 mM NaCl. The group of Dr. Virginia Luna has been able to perform a thorough study on the physiological responses of *P. strombulifera* in different salt and osmotic stress conditions and their results indicate that this species constitutes an excellent model for studying salt tolerance in halophytes.

While a substantial amount of information is present concerning the regulation of stress-induced genes, little is known about the function of the corresponding proteins in stress tolerance. Consequently, the cloning and functional analysis of a set of osmotic-stress induced genes from two stress tolerant plants will extend the possibilities to generate targets for genetic engineering stress tolerance in crop species.

The overall objective of this research proposal is to identify key components of the molecular mechanisms underlying osmotic stress tolerance in *P. patens* and in *P. strombulifera*, with the long-term goal of generating tools for the development of strategies for possible crop yield improvement.

Specifically, we aim to:

a) Identify genes induced during salt and osmotic stress in these two species; b) Analyze the contribution to osmotic stress tolerance of the identified genes in *P. patens* by targeted gene disruption; c) Analyze the correlation of the expression pattern of the identified genes in *P. strombulifera*, with the hormonal (abscisic acid) and stress responses to extreme stress conditions; d) Perform functional studies of the selected genes by evaluating the abiotic stress tolerance of transgenic plants (initially *Arabidopsis*), overexpressing such sequences from *P. strombulifera* and *P. patens*; e) Assessing a possible protective role in higher plants of a *P. patens* dehydrin-like protein (DHNA), previously isolated and characterized in our laboratory.

The functional analysis of target genes and proteins will also allow to address fundamental questions in plant biology, such as the role of many response genes in stress protection or stress recovery in plants, the degree of conservation or the molecular responses between higher and lower plants, and will contribute to a deeper insight in extreme salt tolerance in halophytes.

This project is intended to be carried out in collaboration with Dr. Aldo Rojas's group (Biotechnology Program, Instituto Nicaraguense de Tecnología Agropecuaria, NICARAGUA) and Dr. Virginia Luna's group (Universidad Nacional de Río Cuarto, Facultad de Ciencias Exactas, Físicas y Naturales, Río Cuarto, ARGENTINA).

Overexpression of genes encoding ion transport proteins (Na^+/H^+ antiporter and H^+ -Pyrophosphatase pump) as a strategy to improve salt-and drought tolerance in wheat and barley.

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Drought and salinity are major constraints on crop production and food security, and have adverse impact especially on socio-economic aspect in the Middle East and North Africa (MENA) region. Adaptation of plants to salt stress (i.e. resumption of growth after exposure to high soil salinity) requires cellular ion homeostasis involving net intracellular Na^+ and Cl^- uptake and subsequent vacuolar compartmentalization without toxic ion accumulation in the cytosol. Sequestration of Na^+ ions into the vacuole through the action of a vacuolar membrane Na^+/H^+ antiporter and H^+ -Pyrophosphatase pump is one mechanism that confers salt tolerance to these organisms. The full-length cDNAs of the wheat Na^+/H^+ antiporter *TNHX1* and the H^+ -pump pyrophosphatase *TVPI* were cloned and sequenced using a wheat cDNA library (GenBank accession no. AY296910 and AY296911 for *TNHX1* and *TVPI*, respectively). Transgenic *Arabidopsis* plants overexpressing the wheat vacuolar Na^+/H^+ antiporter *TNHX1* and H^+ -PPase *TVPI* are much more resistant to high concentrations of NaCl and to water deprivation than the isogenic wild-type strains. These transgenic plants grow well in the presence of 200 mM NaCl and also under water deprivation regime, while wild type plants exhibit chlorosis and inhibition of growth. Leaf area decreased much more in wild type than in transgenic plants subjected to salt or drought stress. The water potential was less negative in wild type than in transgenic plants. Moreover, these transgenic plants accumulate more Na^+ and K^+ in their leaf tissue than the wild type plants. Water loss rate under drought or salt stress was higher in wild type than transgenic plants. Increased vacuolar solute accumulation and water retention could confer the phenotype of salt and drought tolerance of the transgenic plants. Overexpression of the isolated genes from wheat in *Arabidopsis thaliana* plants is worthwhile to elucidate the contribution of these proteins in the tolerance mechanism to salt and drought. A similar strategy could be one way to develop transgenic staple crops with improved tolerance to these important abiotic stresses.

To introduce the selected genes into wheat and barley crops we will adapt genetic engineering technologies (biolistic particle bombardment and also *Agrobacterium*-mediated transformation) to the wheat and barley cultivars that are representative of the germplasm in the MENA region. The genes will be cloned into binary vectors under constitutive maize Ubiquitin-1 or inducible (Rd29A, CHX17) promoters. The use of the Rd29A promoter has been shown to minimize the adverse effects caused by overexpression of stress-associated genes in the absence of stress. The wild type and transformed wheat and barley genotypes will be evaluated for physiological response to salinity and soil water deficits, and also for growth and yield in salinity-affected fields in the MENA region. The results of the proposed research program will contribute to the evaluation of newly emerging biotechnologies in terms of their efficacy for ameliorating abiotic salinity stress effects on crop production in the MENA region. Genetically engineered drought- and salt-tolerant plants could provide an avenue to the reclamation of farmlands lost to agriculture because of salinity and a lack of rainfall.

Stability Increasing of Agricultural Crops in Azerbaijan

Dr.Zeynal Akparov

Genetic Resources Institute's Director, National Co-ordinator on GR
Baku, Azerbaijan

Plant a crops world of Azerbaijan is very various. Valuable varieties and forms, which are folk selections product their wild relatives distinguishing with their stability are valuable genetic source for investigations in the direction of high productive scientific selection varieties establishment here. In the Azerbaijan environment conducting of scientific researches directed on entire realization of update variety potentiality as well improvement of the existed varieties and forms are of the most important priorities. Approximately 30% of the overall crop area of the country (50%) is under draught stress, 42% of it is under salinity stress and entirely is under a stress of a high temperature. As a result of these the loss of cereal products is 25-70%, loss of protein is 30-45%.

Beginning science the 1980-th agricultural biotechnology in Azerbaijan has being accepted as a progressive one. It could be indicated, for example especially: investigations have been conducted by using plant tissue culture methodologies as: study of morpho-physiological process hormonal regulators; study of high temperature effect to morphogenetic and adaptive processing at wheat cell culture; study of local hard wheat and soft wheat varieties stability to chloride and sulphate salinity. Plants stability to stresses has been revealed by: (1) the genetic analyses like genome activeness, variations occurred in labile DNA fraction which is able to transcribe the chromatin, intensity of the RNA synthesis, genetic systems activity of mitochondria and chloroplast as well by: (2) the physiological methods like generation possibilities of seeds in osmotic fluids, stress-depression degree of the chlorophyll, water routine and etc. As a result of these researches plant gene types stable to the abiotic stress factors as: draught, salinity and high temperature have been found.

The major goal of the proposed project is arrangements implementation as follows: establish biotechnology laboratory as well provide with the necessary equipment and educated technical personnel; reveal plant varieties forms and stability genes stable to abiotic stress factors as well their choose; establish high stable/ uninterruptedly productive varieties and forms in unfavourable environment by using natural gene sources as a donor; arrange activities in the directions of the stress genes isolation of the revealed stable to stress plants, their identification and transformation; assistance to the agricultural investigators in the biotechnology research; establish relevant gene pools and DB-s; organize training on proteome applying in cultivation for stability to abiotic stress etc.; develop training text-books and manuals, questionnaires, WEB site on biotechnology; relevant models establishment with the goal of bio-safety and controle mechanism development.

We intend to collaborate with the relevant international/regional organizations in these activities' implementation as: International Centre on Genetic Engineering and Biotechnology (ICGEB), FAO, ICARDA, Kansas University of the USA and the relevant universities of Turkey.

***Solanum chilense* as a source of genes involved in tolerance to salt stress.**

Simón Ruiz Lara. Instituto de Biología Vegetal y Biotecnología. Universidad de Talca.
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Saline stress, low temperatures and drought are abiotic stresses that cause severe damage in plant growth. The growing salinity and drought in soils caused by inadequate irrigation practices and climatic factors became the study of natural tolerance mechanisms of native species a study area highly relevant.

The native plant *Lycopersicon chilense* is a wild tomato that inhabits the Atacama Desert. In its natural habit the plant is exposed to severe drought and salinity conditions and to wide temperature fluctuations. Its phenotypic plasticity allows this plant to adequate to the restrictions imposed by the environment and turns it into an interesting model to study the stress tolerance mechanisms operating in glicophyt species.

In this work, it has been established that, after an acclimatizing period, *L. chilense* triggers its stress tolerance mechanisms. Acclimatizing process involves several changes in gene expression as well as in sugar accumulation, photosynthesis, leaf morphology and cellular ultrastucture.

Genes that are differentially expressed under drought, saline stress and low temperatures have been identified and classified in fourth groups according to the process in which they are involved: general anti-stress response, recovering of ionic-osmotic homeostasis, control of cell growing-expansion and genes that codify detoxifying or repairing enzymes to recover cellular damage.

Representative cDNAs of this four groups of genes have been cloned in binary vectors and transgenic plants of *Nicotiana tabacum* var. Xanthi and *Lycopresicon esculentum* var. money maker have been obtained. Phenotypic effects of the over-expression of these transcripts in transgenic plants and their tolerance to drought stress have been evaluated.

Financed by:

Fundación para la Innovación Agraria (FIA), Proyecto Biot-01-A-065.

Programa de Biotecnología en Solanáceas. DIAT-UTALCA.

Centro de investigación en Biotecnología Silvoagropecuaria (CIBS)

Meeting at Santiago: June 19-21, 2006

Abstract :

Genetic Manipulation Towards Developing Salinity Tolerance in Crop Plants: gene cloning, promoter analysis and raising transgenics

S.K.Sopory

Plant Molecular Biology, International Centre for Genetic Engineering and Biotechnology
Aruna Asaf Ali Road, New Delhi 110067

Plant productivity is greatly decreased in the presence of abiotic stress like , drought , salinity, temperature extremes etc. Presently conventional breeding has yielded limited success towards development of resistant varieties for abiotic stress tolerance in crop plants. A better understanding of the molecular basis of stress tolerance will be a prerequisite for undertaking genetic manipulation of crop plants for developing stress tolerance .

Abiotic stress leads to an enhanced expression of a number of genes and results in the modulation of various physiological and metabolic factors. We have cloned a large number of stress up-regulated EST's in rice, and Pennisetum glaucum and have obtained from these a number of full length cDNAs .The expression of these genes in response to various abiotic stresses has been checked by northern and RT- PCR.. A detailed work on some of the genes encoding compoments in the signal transduction, DNA metabolism , transcription factors and those involved in calcium and glutathione homeostasis, has been undertaken. In addition upstream elements of some of these genes have been cloned and analyzed using promoter:: reporter assays. We have also cloned some of the identified genes in plant transformation vectors and transgenic plants have been raised for their functional validation.A detailed analysis has been done for glyoxalase gens, NHX and rab 7 . Based on our studies on model plants, we have now found that the over - expression of genes encoding for glyoxalase I and II, and Vacuolar sodium proton antiporter confer stress tolerance to rice and Brasica juncea..

INTERNATIONAL WORKSHOP "INCREASING TOLERANCE TO ABIOTIC STRESS IN PLANTS"

Santiago, Chile, June 19-21, 2006

Organized by: - Pontificia Universidad Católica de Chile (PUC)
- Foundation for Fruit Development of Chile (FDF)

Sponsored by : The International Center for Genetic Engineering and
Biotechnology (ICGEB)
The Academy of Sciences of the Developing World (TWAS)
Foundation for Agricultural Innovation of Chile (FIA-Chile)
The Latin American Network of Biological Sciences (RELAB)

Objectives of the Workshop

- 1- To stimulate research in plant biotechnology in the field tolerance to abiotic stress
- 2- To facilitate collaboration in research in this topic among developing countries, especially involving scientifically lagging countries.
- 3- To provide information to selected researchers about the goals and objectives of the joint ICGEB/TWAS Program on Tolerance to Abiotic Stress in Plants and to coordinate their applications to this program..

Venue: *Aula Magna Eliodoro Matte*, Centro de Extensión de la Pontificia Universidad Católica de Chile. Av. Bernardo O'Higgins 380, Santiago, Chile (19 – 21 June)
Sala Matte, Centro de Extensión de la Pontificia Universidad Católica de Chile. Av. Bernardo O'Higgins 380, Santiago, Chile (21 June, morning)

PROGRAM

Monday, June 19th, 2006

9:00 Inauguration – Authorities of the Ministry of Agriculture of Chile, TWAS, RELAB, Pontificia Universidad catolica de Chile, FDF, FIA

Mr. Ronald Bown - ASOEX

Mr. Rodrigo Vega – Executive Director FIA

Mr. Jaime Lavados G. – Chairman of the Board (FDF)

Dr. Pedro Pablo Rosso – Head of P. Catholic University of Chile.

Dr. Alvaro Rojas M. – Minister of Agriculture (Chile)

Dr. Jorge E. Allende – President RELAB Corporation

9:30 Conference of Marc Van Montagu (Belgium)
"Plant Biotechnology, an imperative topic for developing countries"
Introduced by Dr. Jorge E. Allende

First ICGEB/TWAS Session – Chairperson: Dr. Edmundo Acevedo

- 10:15 Dr. Gabor Galiba (Hungary)
“Molecular breeding for abiotic stress tolerance in cereals”
- 10:35 Prof. Jennifer Ann Thomson (South Africa)
“Development of maize and other crops Tolerant to abiotic stress”
- 10:55 Discussion
- 11:10 Coffee Break

Second ICGEB/TWAS Session – Chairperson: Dr. Roberto Gaxiola

- 11:30 Prof. Nicolay V. Ravin (Russia)
“Use of bacterial H⁺ pyrophosphatases for development of salt tolerant plants”
- 11:50 Prof. Khaled Masmoudi (Tunisia)
“Overexpression of genes encoding ion transport proteins (Na⁺/H⁺ antiporter and H⁺ pyrophosphatases) as a strategy to improve salt and drought tolerance in wheat and barley”
- 12:10 Dr. Sabina Vidal (Uruguay)
“Identification of key genes involved in osmotic stress tolerance in the model plants. *Physcomitrella patens* and *Prosopis strombulifera*”
- 12 :30 Discussion
- 12 :45 Lunch
- 14 :30 Conference by Dr. Luigi Cattivelli (Italy)
“Genomic approaches to dissect the molecular bases of cold and drought tolerance in barley and wheat”
Introduced by Dr. Jorge E. Allende

Third ICGEB/TWAS Session – Chairperson Dr. Shudir Sopory

- 15:15 Francisco A. P. Campos (Brazil)
“Development of drought tolerant transgenic Cassava varieties”
- 15:35 Andrés Zurita (CEAZA – Chile)
“Tolerance Strategies of Quinoa plants under salt stress”
- 15:55 Ahmed M.K.A. Nada (Egypt)
“Isolation and Characterization of salt related gene(s) from Mangrove”

- 16:15 Discussion
- 16:30 Coffee Break

Tuesday, June 20th, 2006

- 9:00 Conference by Dr. Sudhir Sopory (ICGEB, India)
"Experimental Development of Transgenic Plants Resistant to Abiotic Stress"
Introduced by Dr. Rafael Vicuña
- Fourth ICGEB/TWAS Session – Chairperson: Dr. Marc Van Montagu
- 9:45 Prof. Marcio Costa (Brazil)
"Genomics approaches for improving drought, salt and cold tolerance in citrus"
- 10:05 Prof. Patricio Arce (PUC – Chile)
"Increasing salt tolerance in citrus rootstocks"
- 10:25 Dr. Eisa El Gaali (Sudan)
"Biotechnological approaches for improvement of date palm"
- 10:45 Discussion
- 10:55 Coffee Break
- 11:15 Conference of Dr. Roberto A. Gaxiola (USA)
"Root Engineering. A Strategy for Agriculture in Marginal Areas of Cultivation"
Introduced by Dr. Patricio Arce
- Research in Abiotic Stress Resistance in Chile
Chairperson: Dr. Eugenio Dusselin (U. de Tarapaca)
- 12:00 Dra. Maria Teresa Pino (INIA)
"Transcription Factors (CBF/DREB) and Cold Tolerance in two Solanum species"
- 12:15 Dr. Erwin Krauskopf (Universidad Andrés Bello)
Characterization of genes involved in abiotic stress response in Eucalypts globulus.

12:30 Lunch

Research in Abiotic Stress Resistance in Chile

Chairperson: Dr. Eugenio Dusselin (U. de Tarapaca)

14:30 Dr. Edmundo Acevedo (U. de Chile)
"An Agronomic view of drought resistance"

14:45 Dra. Claudia Ortiz (Univ. de Santiago Chile)
"Copper accumulation and tolerance mechanisms in Chilean rabbitsfoot grass (*Polypogon australis*)"

15:00 Dr. Simon Ruiz (U. de Talca)
"Solanum chilense as a source of genes involved in tolerance to salt stress".

15:15 Dra. Ana Gutierrez (U de la Frontera)
"Characterization of stressoxidative and cold tolerance gene in *Deschampsia antarctica* Desv"

15:30 Dra. Roxana Ginocchio (CIMM)
"Importance of copper bioavailability on plant toxicity "
Centro de Investigaciones Mineras

15:45 Dr. Claudio Pastene (Universidad de Chile)
"Paraheliotropism in beans: a tradeoff between water stress resistance and yield"

16:00 Coffee Break

16:30-18:00 ICGEB/TWAS Round Table "Strategies for research in plant biotechnology in developing countries"

Chairperson: Dr. Mohamed H.A. Hassan

Participants: Dr. Inuwa S. Usman (Nigeria)

Dr. Jorge Quezada (Bolivia)

Dr. Tadesse Mehari Hagos (Eritrea)

Dr. Zeylan Akparov (Azerbaijan)

Dr. Esteban Falconi (Ecuador)

Dr. Zafar Ismailov (Uzbekistan)

Dr. Dhalia Garwe (Zimbabwe)

Wednesday, June 21st, 2006

9:00 *Aula Magna Eliodoro Matte*
Meeting of Group 1 (Language: English) – Discussion of the goals and activities of the ICGEB/TWAS joint Research Program on Plant Tolerance to Abiotic Stress

Chaired by Dr. Decio Ripandelli.

9:00 *Sala Matte*
Meeting of Group 2 (Language: Spanish) – Discussion of Chilean Authorities (FIA, CORFO, FONDEF-CONICYT) with researchers and productive sector business enterprises about the interest and possibility of supporting a special line of research in this topic in Chile.

Chaired by Mr. Edmundo Araya – General Director of FDF (Chile).

1) Conference: "Climatic change in Chile: Current Evolution and Projections"
Dr. Patricio Aceituno – Fac.Cs.Físicas y Matemáticas (U. de Chile).

2) New research topics on Abiotic Stress in Plants in Chile.

- 2.1) Sr. Eugenio Doussellin – Universidad de Tarapacá.
- 2.2) Sr. Roberto Ipinza – Instituto Forestal
- 2.3) Dr. Patricio Arce – Fac. Ciencias Biológicas (PUC)
- 2.4) Dr. Luis Gurovich – Fac. Agronomía y Cs.Forestales (PU)
- 2.5) Dr. Andrés Zurita – CEAZA IV Región
- 2.6) Dr. Carlos Muñoz - INIA

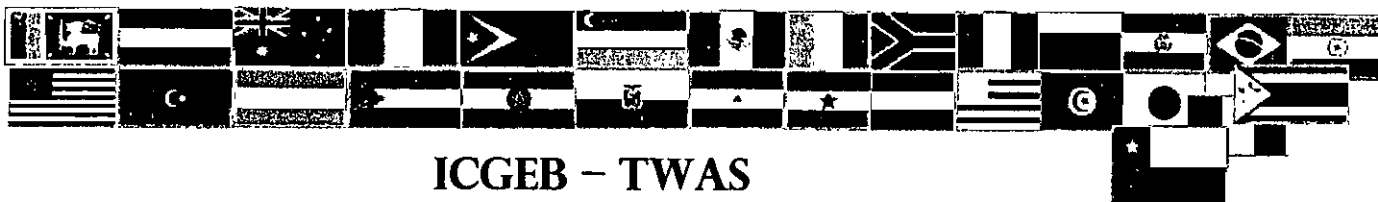
3) Funding for Research Projects on Abiotic Stress in Plants.

- 3.1) Sr. Rodrigo Vega A. – Director Ejecutivo de la FIA
- 3.2) Sr. Gonzalo Herrera – Director Ejecutivo del FONDEF
- 3.3) Sr. Jean Jacques Duhart – Director Ejecutivo de INNOVA-Chile
- 3.4) Sr. Jaime Lavados - – Chairman of the Board (FDF)

11:00 Coffee Break

11:30 *Aula Magna Eliodoro Matte*
Conference Dr. Yoshihiro Hase (Japan)
"Novel Ultraviolet-B resistant mechanisms revealed by Arabidopsis mutants"
Introduced by Dr. Jorge E. Allende

12:15 Closing Ceremony – Conclusions
Dr. Jorge E. Allende



ICGEB – TWAS
JOINT PLANT BIOTECHNOLOGY PROGRAMME
INTERNATIONAL WORKSHOP ON
PLANT TOLERANCE TO ABIOTIC STRESS

International Experts

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Institute Plant Biotechnology for Developing Countries,
Department of Molecular Genetics, Ghent University, Belgium

Shudar V. Sopory

International Center for Genetic Engineering New Delhi, India

Luigi Cativelli

Agricultural Research Council of Italy, Centre for Genomic Research
and Centre for Cereal Research, Italy

Roberto A. Gaxiola

Plant Molecular Genetics, Department of Plant Science, University
of Connecticut, USA

Yoshihiro Hase

Director General of Quantum Beam Science Directorate, Japan
Atomic Energy Agency, Japan

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the joint ICGEB/TWAS
Program on Tolerance of
Abiotic Stress in Plants and
to coordinate their
applications to this program.

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2006

JUNE 19 – 21

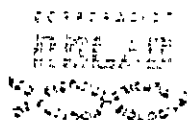
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