

POSTER ABSTRACTS

AGRICULTURAL SCIENCES

1. GENETIC MAPPING USING RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) MARKERS IN A *Vigna radiata* x *V. mungo* INTERSPECIFIC CROSS*

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To further saturate the genetic linkage map of mungbean, random amplified polymorphic DNA (RAPD) analysis in an F₂ population of a *Vigna radiata* var. NCM x *V. mungo* var. Acc. 25 cross was conducted. DNA isolation and RAPD conditions were standardized for optimum resolution and to ensure reproducibility. The optimized procedure was used to survey for polymorphism between two parents. Out of the 146 Operon primers tested during screening, 131 well-resolved and strongly amplified putative polymorphic markers were produced. The inheritance and linkage relationships of 19 RAPD markers in the F₂ (NCM X Acc. 25) population were determined. Fifty-eight percent of the markers followed the expected dominance ratio of 3:1 and two RAPD linkage groups were formed. These markers will be subsequently anchored in the existing RFLP map of mungbean.

Key words: Mungbean, blackgram, RAPDs, genetic mapping

*Best Poster Paper Award in the Agricultural Sciences.

2. GENETIC LINKAGE AND QUANTITATIVE TRAIT LOCI ANALYSIS IN POTATO II. MOLECULAR MAPPING OF GENES FOR RESISTANCE TO THRIPS

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Among the staple food crops, potato ranks highest in nutritive value and productivity. Its major production constraint in the Philippine lowlands, however, is the severe occurrence of pests including thrips. Breedings for resistant varieties still provides the long term most effective strategy of control. Two *Solanum tuberosum* x *S. berthaultii* backcross populations (BCB and BCT) were developed and evaluated for resistance to thrips under greenhouse conditions. Appropriate statistical analysis was carried out and putative quantitative trait loci (QTLs) were located using the RFLP/RAPD map of potato. Interval mapping by MAPMAKER/QTL software was employed and further verified by one-way ANOVA ($P < 0.05$). One QTL on chromosome 8 in BCB, and one QTL on chromosome 1 in BCT were found to influence resistance. The QTL accounted for 027% of the phenotypic variation observed, with the locus in BCB consistently identified for the resistance traits evaluated. Further multiple regression analysis of the identified QTL is underway and field screening will be set up to verify the resistance reaction under natural growing conditions.

Key words: Potato, *Solanum tuberosum*, *S. berthaultii*, interval mapping, one-way ANOVA, QTLs

3. MOLECULAR MAPPING OF FIBER QUALITY IN COTTON (*Gossypium* sp.)

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Nine cotton accessions were identified as parents for the improvement of fiber quality traits (length, strength, and fineness). These include three of the currently recommended varieties, UPLC-2, CRDI-1, and CRDI-2 as the recurrent parents, and five outstanding accessions in terms of fiber quality traits, CRDN 174, PR-1, SI-A, SI-SAID, SI-Neris, and SI-J, as donor parents. Random amplified polymorphic DNA (RAPD) analysis was used to identify markers that are polymorphic between the recurrent and donor parents. The DNA extraction and RAPD procedures were initially optimized based on the protocols by Igbal et al. (1997). Using the optimized conditions, survey for polymorphism was done. The polymorphic markers identified will be subsequently mapped in an F2 population.

Key words: Random amplified polymorphic DNA (RAPD), cotton mapping

4. CLONING AND CHARACTERIZATION OF A MELANIN BIOSYNTHETIC *THR1* REDUCTASE GENE ESSENTIAL FOR APPRESSORIAL PENETRATION OF *Colletotrichum lagenarium*

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Melanin biosynthesis of *Colletotrichum lagenarium* is essential for appressorial penetration of the host plant. A melanin-deficient mutant 9141 (Thr) has a defect in the conversion of 1,3,8-trihydroxynaphthalene to vermeline in the

melanin biosynthetic pathway. The mutant formed nonmelanized appressoria and had little infectivity on cucumber leaves. A cosmid clone pCR1 was selected from a heterologous probe *BRM2*, one of the clustered genes involved in melanin biosynthesis of *Alternaria alternata*, pCR1 transformed the Thr mutant 9141 to wild type phenotype. A DNA fragment (*THR1*) homologous to *BRM2* was subcloned from pCR1 and the nucleotide sequence determined. *THR1* contains one open reading frame that encodes a protein of 282 amino acids. A transformant resulting from gene disruption showed a light brown phenotype different from the dark brown phenotype of the wild type 104-T. The transformant formed nonmelanized appressoria and had little infectivity. The *THR1* amino acid sequence contains a region highly similar to the *Ver1* gene involved in the conversion of versicolorin A to sterigmatocystin in aflatoxin biosynthesis by *Aspergillus parasiticus* and to the T₄HN reductase gene involved in the conversion of 1,3,6,8-tetrahydroxynaphthalene to scytalone and 1,3,8-trihydroxynaphthalene to vermeline in melanin biosynthesis by *Magnaporthe grisea*. Expression of the *THR1* gene during spore germination of *C. lagenarium* was detected by RNA blotting. We propose that the *C. lagenarium* *THR1* gene encodes a reductase involved in the conversion of 1,3,8-trihydroxynaphthalene to vermeline.

Key words: Reductase gene, cloning, *Colletotrichum lagenarium*, fungal transformation, gene disruption, melanin biosynthesis, appressorial penetration, cucumber, sequencing, melanin-deficient mutant

5. GENETIC LINKAGE AND QUANTITATIVE TRAIT LOCI ANALYSIS IN POTATO III. EVALUATION AND MAPPING OF GLANDULAR TRICHOMES UNDER THE PHILIPPINE LOWLAND and HIGHLAND CONDITIONS

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The resistance reaction conferred by *Solanum berthaultii* to a wide range of insects was associated with the presence of glandular trichomes (GT). Quantitative trait loci (QTLs) have already been analyzed for this valuable character under

temperate conditions using the *S. tuberosum* x *S. berthaultii* backcross populations (BCB and BCT). In the Philippines, these populations have been further verified for resistance against thrips. To determine if this newfound resistance is also GT-based, trichome expression and QTLs were evaluated under the Philippine lowland (IPBUPLB, Laguna) and highland (NPRCRTC-BSU, La Trinidad, Benguet) conditions. In both elevations, trichome types A and B expression in BCB and trichome A in BCT were confirmed. Proper statistical analysis was carried out and the RAPD/RFLP map of potato was used to validate the QTLs. MAPMAKER/QTL and one-way ANOVA were employed. The same interval on chromosomes 5 and 2 was verified for trichome B density in BCB, while on the same chromosome different intervals were identified for trichome A. A putative QTL, which was not previously mapped, was further located in chromosome 2 in BCB along the interval of TG 276 and TG 306 markers. Further multiple regression analysis and correlation with QTLs for thrips resistance is underway.

Key words: Potato, *Solanum tuberosum*, *S. berthaultii*, glandular trichomes, interval mapping, one-way ANOVA, QTLs

6. MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF TALL AND DWARF POPULATIONS OF COCONUT, *Cocos nucifera* L.

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Selected tall and dwarf varieties of coconut collected from the Institute of Plant Breeding – Fiber and Industrial Crops Division, Philippine Coconut Authority (PCA) – Zamboanga Research Center, and PCA – Davao Research Center were characterized for horticultural and morphological traits. Genomic DNA extraction, random amplified polymorphic DNA (RAPD), and microsatellite (SSRs) conditions were optimized. Preliminary putative molecular markers were generated and are currently being correlated with the agro-morphological characterizations. Informative markers will be used to construct a molecular linkage map of coconut and to tag morpho-agronomic traits of interest. Ultimately, tightly linked markers will be identified and designed/converted into easily scorable PCR-based markers for use in marker-assisted (MAS) breeding in coconut.

Key words: Coconut, *Cocos nucifera*, RAPD, DNA marker, genome map, MAS

7. GENETIC ENGINEERING FOR RICE IMPROVEMENT AT PHILRICE

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The increasing population rate and shrinking area devoted to rice production in the Philippines pose a big challenge to plant breeders. Genetic engineering at PhilRice is being conducted to support the existing strategies to increase the yield potential of rice, such as the improvement of pest resistance in the high-yielding varieties, in the new plant type elite lines, and in the cytoplasmic male sterile lines used in hybrid rice breeding. A P3 laboratory and CL2 screenhouse established at PhilRice following the National Committee on Biosafety of the Philippines Guidelines was constructed. Gene introgression into selected varieties and lines is being conducted through the use of the natural vector *Agrobacterium tumefaciens*. Preliminary results of the tissue culture experiments on the determination of the most suitable explant, selection procedure, tissue culture media for transformation; evaluation of the use of different binary vectors in transforming rice; and transformation strategies will be presented.

Key words: Genetic engineering, rice, crop improvement, binary vectors, *Agrobacterium tumefaciens*, plasmids, selection, transformation, natural vector

8. MOLECULAR MARKER-AIDED DEVELOPMENT OF RICE VARIETIES FOR DIRECT-SEEDING

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Seedling vigor (SV) is important under direct-seeded rice culture. Quantitative trait loci (QTLs) for SV-related traits have been mapped to different rice chromosomes in both indica and japonica rice. Molecular markers flanking these QTLs could be useful as diagnostic probes for introgressing and/or pyramiding favorable SV alleles. Forty-nine promising cultivars for breeding direct-seeded varieties at 37 RAPD loci flanking 11 SV QTLs were assayed and their respective genotypes compared with:

(a) that of *Italica Livorno* (IL), a cultivar used to map SV QTLs in japonica rice, (b) among themselves, and (c) actual SV performance based on slantboard tests. Relatively little genetic similarity was observed between IL and the cultivars assayed suggesting the presence of other SV QTLs in the rice genome. This was supported by results from cluster analysis using 67 randomly selected RAPD markers that revealed a different pattern compared with groupings based on RAPDs flanking SV QTLs. Slantboard tests identified consistently superior varieties for the four SV traits measured – length of shoots, roots, mesocotyl, and coleoptile – and other high-SV donors, in addition to those already known (IL and Black Gora) were identified. Crosses between alternative genetic donors for SV to develop new populations that would segregate for the maximum number of high-SV alleles could facilitate the development of superior direct-seeded rice varieties.

Key words: Seedling vigor, QTL, molecular marker, RAPDs, rice

9. CHROMOSOMAL SEGMENT WITH MAJOR AND MINOR GENES FOR BLAST RESISTANCE IDENTIFIED USING DNA MARKERS

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The combination of major and minor genes has been associated with durability of resistance. Two durability resistant varieties of rice *Oryza sativa L.*, ‘Lemont’ and ‘Teqing’ have four major genes tagged using DNA markers. The recombinant inbred population of these varieties was evaluated at the blast nursery to identify minor genes and their association with major genes mapped in this population. Data on area under disease progress curve (AUDPC), percent diseased leaf area (%DLA), and Standard Evaluation System for blast (SES rating), analyzed both at interval mapping and single marker analysis of 167 molecular markers revealed 13 putative quantitative trait loci (QTLs) for blast resistance distributed in nine chromosomes. Three of the QTLs were associated with major genes tagged in this population, five were near the reported QTLs and/or major genes, and five were apparently new QTLs for blast resistance. QTLs mapped at or near the major genes were identified consistently at the seedling stage and had good resistance compared with those mapped to the regions not associated with major genes. These suggest the importance of both major and minor genes in breeding for durable resistance to

blast. Molecular markers will be useful in identifying these lines that are generally difficult to identify during classical selection for disease resistance.

Key words: Resistance gene, rice blast, molecular markers, mapping, disease resistance, major gene, minor gene, *Oryza sativa*

10. INHERITANCE OF TUNGRO RESISTANCE GENES FROM TWO TUNGRO-RESISTANT DONOR RICE VARIETIES

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The inheritance of RTSV resistance from two donor varieties – Utri Merah and Utri Rajapan – was studied using 111 and 131 F₃ families derived from the crosses TN1 x Utri Merah and Utri Rajapan – was studied using 111 and 131 F₃ families derived from the crosses TN1 x Utri Merah and TN1 x Utri Rajapan, respectively. One-week old seedlings were inoculated using 5 RTBV+RTSV-viruliferous *Nephotettix virescens* (GLH) per seedling. Visual tungro incidence scores and ELISA were made 30 days after inoculation. Three inoculation trials were conducted for each family to determine the infection due to RTBV and RTSV. Visual tungro incidence scores and the ELISA results for RTSV and RTBV were not correlated. Some lines of both crosses were observed to have high infection of RTBV but low disease incidence indicating tolerance to RTBV. Frequency distribution of percentage infection by RTSV among F₃ families for both crosses indicates that recessive genes control RTSV resistance from both donor varieties. Frequency histograms of percentage infection by RTSV among F₃ families from both crosses also indicate bi-modal distribution of resistant and susceptible lines and support the hypothesis that one recessive gene and two recessive genes control RTSV resistance in Utri Rajapan and Utri Merah, respectively. The information gained in this study will be very useful in the systematic transfer of these resistance genes into elite rice cultivars and in molecular mapping.

Key words: Rice, tungro, Utri Merah, Utri Rajapan, genetics, resistance, ELISA, GLH, RTSV, RTBV, *Oryza*

11. NUCLEAR AND ORGANELLAR DNA APPLICATION IN *Oryza*: TAXONOMY AND PHYLOGENE

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Conventional taxonomy and phylogeny of germplasm are based on the tedious characterization of morphological variation. The ability to assay DNA variation that underlies morphological variation offers great promise as a convenient alternative for the genetic characterization of germplasm. Restriction fragment length polymorphism (RFLP) was used to survey DNA variation in 22 species of the genus *Oryza*. At the ribosomal DNA (rDNA) multigene family, 15 rDNA spacer length (*sl*) variants were identified using restriction enzyme *Sst*I and wheat rDNA unit as probe. Particular *sl* variants predominated in certain isozyme groups of *O. sativa*, indicating a potential of *sl* polymorphism in varietal classification. The distribution of *sl* variants supports the origin of *O. sativa* and *O. nivara* from *O. rufipogon*, and that *O. spontanea* arose from introgressions among *O. sativa*, *O. nivara*, and *O. rufipogon*. The distribution also suggests that the CC genome, of all the genomes in the *Officinalis* complex, may be the closest to the *Sativa* complex genomes, and it affirms the genetic position of the *Officinalis* complex intermediate between the *Sativa* and *Ridleyi* complexes. Variation at the *Oryza* organelle genomes was probed with a maize mitochondrial gene, *atpA*, a wheat chloroplast inverted repeat segment, *p6*. Results indicated that the complexes can be differentiated by their mitochondrial genome, but not their chloroplast genome when digested by *SSt*I or *Bam*HI. Therefore, the natural DNA variation in the nuclear and mitochondrial genomes has demonstrated great potential in complementing the conventional basis of taxa classification and phylogeny in the genus *Oryza*.

Key words: *Oryza sativa*, *O. nivara*, *O. rufipogon*, *O. spontanea*, *O. officinalis*, RFLP, ribosomal DNA, *Ridleyi*, maize *atpA*

12. INDUCTION OF SALT TOLERANCE IN RICE BY SILICA TREATMENT

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The objective of the study was to investigate the possibility of inducing salt tolerance in rice by silica treatment. A total of ten varieties namely, salt-tolerant

Kala Rata 1-24, Hsieh-Tso 12, Bhura Rata, SEW 273-5-13, IR4595-4-1-13, Joalbanga, and salt-susceptible Karti Gotak, Mangasa, Jyothi, and IR 28 were used. The 50 ppm Na_2SiO_3 treatment started at transplanting time and lasted until the 14th d of salt treatment. The 50 mM NaCl treatment started 40 d after sowing (DAS). The dry weight increase, amount of Na^+ and Cl^- in the different plant parts, transpiration stream concentration factor (TSCF) of Na^+ , and changes in the cell wall constituents were observed.

Si treatment changed the plant dry weight at 40 DAS of K. Gotak and Mangasa. Based on the dry weight increase, Si treated plants performed better than with no Si treatment after 14 d in 50 mM NaCl. Si treated plants have lower amounts of Na^+ and Cl^- in the different plant parts, especially in the leaf blades, except for the Na^+ content of SEW and Joalbanga. Si treatment also improved the TSCF Na^+ which implies improvement of the membrane selectivity for Na^+ . The modification in the cell wall constituents by Si treatment before the imposition of salt stress may have contributed to lower TSCF Na^+ which reduced Na^+ accumulation and consequently improved the dry weight increase of plants.

Key words: Adaptation, cell wall constituents, rice, salt tolerance, varietal differences

13. POTENTIAL METHODS FOR RAPID ASSAY OF LIPOXYGENASE

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Potential methods for the rapid assay of sweet corn lipoxygenase (LPO) were evaluated and compared. Both model system and homogenate system studies were carried out. Partially purified LPO had maximum activity at pH 6.0 and 2.0 mM linoleic acid. Methylene blue bleaching (MBB), carotene bleaching (CB), potassium iodide-starch (KI-S) methods were evaluated both spectrophotometrically and visually. Both the MBB and KI-S methods worked well for the model system. The three methods were then evaluated for vegetable homogenate systems. The vegetables studied were sweet corn and green beans. The MBB method indicated positive results for both sweet corn and green beans whereas the CB method did not work for either vegetable. On the other hand, the KI-S method worked very well for green beans but not for sweet corn, due to the presence of carotenoid compounds which are oxidized preferentially. The MBB method is suitable for both

carotenoid-containing and non-carotenoid-containing vegetables. It is also more sensitive than the KI-S method. The use of this rapid assay is recommended for determination of LPO activity in sweet corn.

Key words: Lipoxygenase, rapid assay, methylene blue bleaching, carotene bleaching, potassium iodide-starch, corn, green beans, spectrophotometry, model system, homogenate system

14. GENERATION OF PHOTOPERIOD-INSENSITIVE VARIANTS FROM PHOTOPERIOD-SENSITIVE RICE VARIETY WAGWAG THROUGH ANTHER CULTURE

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Anther culture is basically utilized in crop breeding as a tool for rapid generation of breeding lines. However, as with other in vitro culture technologies, anther culture may result in the generation of variants that may have useful agronomic traits, or that may serve as source of genetic variation. In this study, plants were generated and cultured from anthers of rice, cv. Wagwag. The Wagwag variety, which was used as anther source, flowers and produces grains only during the wet season, but not in the dry season planting. From a total of 96 Wagwag lines generated via anther culture, 16 individual plants were identified, which flower and produce grains during both dry and wet season planting. These putative variants were screened for salt tolerance, and evaluated for their morpho-agronomical traits.

Key words: Photoperiod sensitive, putative variants, anther culture, Wagwag variety, salt tolerance, morpho-agronomical trait

15. DISCRIMINATING THE QUALITY OF PHILIPPINE RICE VARIETIES WITH THE NEW TENSIPRESSER

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The objective to breed intermediate-amylose (AC) rices in the Philippines has resulted in the commercial release of rice varieties with a narrow range of starch properties. Conventional physicochemical tests can hardly differentiate among these rices and yet their eating qualities still differ based on sensory evaluation. Alternative objective techniques for grain quality evaluation are necessary. The applicability of the New-Tensipresser in evaluating the quality of Philippine rice varieties was looked into. Three modes of measurements were tried, namely: low compression (LC), high compression (HC), and continuous compression (CC) tests. In the LC test, a wider range of difference was evident for the cooked rice texture data of low-AC and intermediate-AC rices compared to the glutinous and high-AC samples. This suggests that low-AC and intermediate-AC rices can be discriminated well using the LC mode. The data for HC test varied appreciably for all amylose classes. For the CC test, a wider range of difference was noted on the toughness data among and within amylose classes. Likewise, glutinous and low-AC samples varied noticeably in tenderness value; high-AC and intermediate-AC samples did not. The texture data obtained with the apparatus highly correlated with apparent amylose content, iodine blue value, and sensory evaluation scores. The results indicate the good potential of the New-Tensipresser as a tool for routine assessment of rice grain quality in the Philippines. It can differentiate cooked rice texture of cultivars with wide to narrow range of starch properties, gives quick results, and requires a small sample size.

Key words: Rice, tensipresser, cooked rice texture, grain quality, amylose, starch, sensory evaluation, compression test, tenderness, toughness

16. EFFECT OF GAMMA IRRADIATION ON MILLED RICE QUALITY

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Gamma irradiation as a technique for preserving cereal grains and other foodstuffs is now gaining prominence. It has been proven to reduce post-harvest losses from insect infestation and microbial action. To validate its effects on storage, physicochemical, cooking, and sensory qualities of milled rice, this study was then conducted. Batches of milled rice stored in three different packaging materials (polypropylene sack, polyethylene bag, and polypropylene sack lined with polyethylene bag) were irradiated with 0, 0.5, and 1.0 kilogray of Co⁶⁰. Treated samples were stored at room temperature and their grain qualities were evaluated monthly for a period of nine months. Irradiation decreased grain whiteness, gel consistency, water uptake ratio during cooking, and sensory ratings. It increased iodine blue value and percent soluble solids. The magnitude of change was influenced by the dose of irradiation. The effect was more apparent with 1.0 kGy dose. Differences in sensory quality between irradiated and non-irradiated samples became less evident with time. Irradiated rice stored in a polypropylene sack lined with a polyethylene bag was more organoleptically acceptable than those packed in polypropylene sacks and polyethylene bags.

Key words: Irradiation, preservation, grain quality, cereals, storage, water uptake ratio, gel consistency, soluble solids, iodine blue value, sensory quality

17. IN VITRO RESPONSE OF IMMATURE RICE INFLORESCENCE TO PHENYLACETIC ACID

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Genotype, plant growth regulator, explant type, and culture conditions are some parameters that can be exploited to optimize in vitro culture of crops, such as rice. In this study, the response of two varieties of indica rice, IR 64 and PSBRc 46, cultured in vitro, to phenylacetic acid (PAA) at 10 and 40 mg/L, as compared with

2,4-dichlorophenoxyacetic acid (2,4-D) at 2 mg/L culture medium was evaluated. Spikelets from two sizes, 1 and 2 cm long, of immature inflorescence were used as explants. Separate sets of cultures were maintained in the dark and under light conditions. Morphological and developmental responses, such as explant growth, callus formation, callus differentiation, and plant regeneration, as affected by different culture parameters will be presented.

Key words: Rice, immature inflorescence, in vitro, phenylacetic acid, 2,4-dichlorophenoxyacetic acid

18. ENHANCING PLANT REGENERATION FROM ANTHER CULTURE-DERIVED CALLUS OF INDICA RICE BY DESICCATION

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Green plant regeneration is a limiting factor for efficient utilization of anther culture in rice breeding, especially for indica rice. In this study, the effect of simple desiccation on the enhancement of plant regeneration from anther culture-derived calli of three (one inbred and two F₁ crosses) indica rice genotypes was investigated. Calli were air-dried for 24 h and 48 h resulting in approximately 11% and 16% water loss from the tissue, respectively. Desiccation for 24 h enhanced callus differentiation, as manifested by callus greening or presence of green specks on the tissue, in all genotypes, while with 48 h desiccation treatment, only two genotypes exhibited improved differentiation. Desiccation for 24 h enhanced more shoot elongation from differentiated calli in the inbred PSBRc 1 compared with 48 h desiccation. While with F₁ cross, PRIA x IR 64, higher frequency of shoot elongation from differentiated calli was obtained with 48 h desiccation. Desiccation decreased frequency of rhizogenesis (unipolar germination). No reduction in the frequency of albinism was obtained with desiccation. However, desiccation markedly reduced the incidence of callus necrosis.

Key words: Anther culture, rice breeding, indica rice, desiccation, water loss, plant regeneration, callus differentiation, rhizogenesis, albinism, necrosis

19. MULTIPLE SHOOT DIFFERENTIATION FROM EXCISED SHOOT TIPS AND NODAL SECTIONS OF CORN (*Zea mays* L.)

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In vitro techniques have been developed to regenerate clumps of multiple shoots at high frequency from excised shoot tips and nodal sections of in vitro germinated seedlings of corn (*Zea mays* L.). Each shoot tip produced about 7 shoots after 4 weeks of culture on MS medium supplemented with 2 mg/L BAP, 500 mg/L casein hydrolysate, and 20 g/L sucrose. Clumps of very small shoots were also produced from the nodal sections. Shoots developed into plantlets and formed roots when transferred onto MS medium supplemented with 1 mg/L IBA. Shoot tip cultures of 9 inbred lines (8 yellow corn and 1 white corn) formed multiple shoots, with frequency ranging from 5 to 100%. Inbred line P22 gave the best multiple shoot response. This technique of axillary/adventitious shoot differentiation offers an efficient and high frequency regeneration system suitable for corn genetic transformation work.

Key words: *Zea mays*, multiple shoot clumps, axillary/adventitious shoot, organogenesis, regeneration.

20. CALLUS INDUCTION IN *Ipomoea muricata* JACQ. (CONVOLVULACEAE) HYPOCOTYL AND LEAF EXPLANTS

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Recent studies employed the technique of tissue culture to produce secondary metabolites from plant sources. Initially, calli are formed from plant parts, grown in artificial medium and later, tested for the presence of bioactive compounds. Callus was induced from *Ipomoea muricata* Jacq. (Convolvulaceae), a medicinal plant active against various types of skin ailments. Hypocotyl and leaf explants were

culture on agar-solidified Murashige and Skoog (MS) medium supplemented with 3% (w/v) sucrose and various combination of auxins [α -naphthalene acetic acid (NAA), indole-3-acetic acid (IAA), 2,4-dichlorophenoxyacetic acid (2,4-D)], and cytokinins [benzylaminopurine (BAP), kinetin (K), adenine hemisulfate (AH)]. Cultures were incubated in the dark for 2-4 weeks at 28-30°C. Whitish to pale yellow calli were observed on both explants with 0.1, 0.5, 1.0 mg/L NAA with BAP and in 2,4-D with AH in factorial combinations. When grown on MS medium with 0.1, 0.5, 1.0 mg/L IAA and 0.1, 0.5, 1.0 mg/L K in factorial combinations, callus was induced on the explants in all treatments. When tested for the presence of secondary metabolites, particularly alkaloids, the callus derived from hypocotyl and the hypocotyl explant gave a negative result with the Culvenor-Fitzgerald test. The leaf explant gave a positive result with the test.

Key words: Callus, callus induction, tissue culture, *Ipomoea muricata*, Convolvulaceae, secondary metabolites, alkaloids

21. VEGETATIVE PROPAGATION OF *Paraserianthes falcataria* THROUGH TISSUE CULTURE

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The vegetative propagation of *Paraserianthes falcataria* through tissue culture was developed. Tissue culture was developed from trials conducted to determine the appropriate sterilization technique of the tissues and the medium and incubation conditions appropriate for multiple shoot induction and for regeneration of plantlets by rooting these shoots. The best source of tissues for culture was also determined from the comparative response of tissues collected from trees (mature tissues) and developing seedlings (juvenile tissues) to the culture media tested.

Results of experiments of *P. falcataria* showed that plantlets can be produced from both juvenile and mature tissues cultured aseptically under laboratory conditions of 18 h light and temperature range of 24-29°C. More plantlets can be produced from juvenile nodal sections. These juvenile sections are best collected from aseptically germinated seeds of this species sterilized by 3-min dipping in 2% sodium hypochlorite. Multiple shoot induction of cultured tissues was best in modified MS medium with 5 ppm BAP and 0.5 ppm IAA (M8). Rooting of excised shoots was observed in same MS medium but with 2 ppm IBA (M20).