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Some endemic plants of Borneo (photograph courtesy of Qammil Muzzammil)

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Dean's Message

Prof Dr Shabdin Mohd Long

Key Performance Indicator (KPI) for Public Universities in Malaysia has been seriously discussed at the national level. Our faculty must be prepared to revolutionize and be able to compete with other universities in Malaysia as well as at the international level in teaching, research and publications. It is envisaged that research results should be published, patented and commercialized by the faculty members in order to achieve recognition at national and international level. It is our hope that our faculty will be at par or even better compared to other faculties in other Malaysian universities and worldwide.

In order to enhance our research, recently Faculty of Resource Science and Technology (FRST) signed several MoAs and MoUs with government organizations, industries and NGOs. MoA between FRST and Malaysian Palm Oil Board (MPOB) resulted in the agreement to conduct research in plant diversity (amount of funding is RM 614, 190.00) and on the mammal diversity (amount of funding is RM 535,740.00) at MPOB Oil Palm Plantation at Sg Asap, Belaga, Sarawak. With the industrial counterpart, agreement with Handalas Sdn Bhd will be signed soon on technical and research cooperation of biotechnological applications in the commercial goat (Jamnapari breed) farming with the amount of funding RM688,148.40.

Several MoUs will be signed shortly between FRST and Field Museum of Natural History, Chicago, Illinois; Veterinary Research Institute, Ipoh; Institute for Medical Research, Kuala Lumpur and Institut Pasteur (IP), Cambodia. The MoUs are signed in order to promote joint research and development collaboration, training of staff, staff exchange and joint grant application. More MoAs and MoUs will be established in order to strengthen research, teaching and publication.

Please feel free to direct your enquiries at e-mail: Ishabdin@frst.unimas.my or to the editorial members for further information.

Happy reading

Overexpression of human *Wnt-2* gene in tumours of various origins

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The human *Wnt-2* gene is a member of the *Wingless type* (*WNT*) – gene family that is associated with the Wnt-signal transduction pathway (Miller, 2001). This pathway is believed to mediate mammalian developmental processes such as cellular proliferation and differentiation (Smalley and Dale, 1999), and may influence tumourigenesis when dysfunctional. Findings by McCoy *et al.* (2002) revealed evidence of growth factor-like proteins encoded by members of the *WNT*-gene family that may be involved in cancer development. We have also proven that *Wnt-4* is associated with the childhood kidney cancer, Wilms' tumour (Sim *et al.*, 2002). Products of the *Wnt-2* gene is a key component in the Wnt/b-catenin pathway (Fig. 1) (Miller, 2001), and is suspected to be linked to colon cancer (Katoh, 2001). In a recent study by us, *Wnt-2* was found to be significantly overexpressed in colorectal carcinoma biopsied tissues compared to their paired normal controls (Ma *et al.*, 2008), thus strongly implicating its direct involvement in tumourigenesis of the human colorectum. Despite increasing evidence of *Wnt-2*'s role(s) in colon cancer, little is known about its behaviour in other types of cancer. Here, we provide preliminary findings that implicate its association with three different types of human cancer, namely bladder carcinoma, prostate adenocarcinoma and nasopharyngeal carcinoma.

To evaluate expression levels of *Wnt-2* in the three types of human cancer, we employed Reverse-Transcriptase Polymerase Chain Reaction (RT-PCR) assay on biopsied tissues. Primers designed to identify specific region within the coding region of *Wnt-2* was used to detect and comparatively assess expression pattern between the tumour and normal tissues studied. Transcript profile evaluation was determined by eye-balling the intensity of PCR product bands on electrophoresed agarose gel.

Our results provide two important information. Firstly, they demonstrated for the first time, the presence of *Wnt-2* expression in human tissues from the organs of prostate and bladder, and the region of the nasopharynx (Fig. 2). The significance of this is that it provides novel clues on the presence of signalling pathways mediated by *Wnt-2* in the organs and tissues mentioned. Secondly, our data indicates that *Wnt-2* is overexpressed or up-regulated in all types of tumour relative to their normal controls (Fig. 2). This trend in differential expression is consistent with our previous findings (Ma *et al.*, 2008) of its endogenous activity in cases of colorectal carcinoma. Whether the activity/pathway regulated by *Wnt-2* (Fig. 1) during organogenesis of prostate, bladder and nasopharyngeal tissues is similar to colorectal system remains to be investigated.

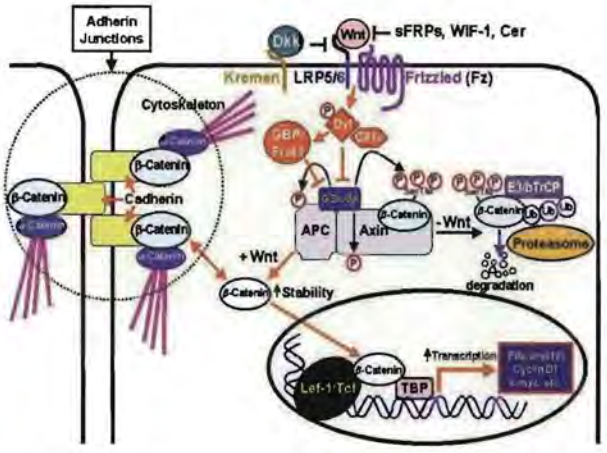

Fig. 1: The canonical Wnt/b-catenin pathway (Adopted from Howard *et al.*, 2003)

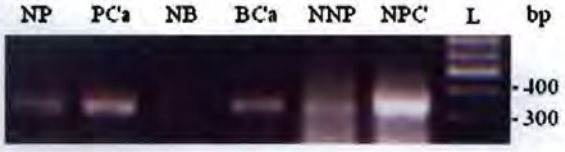

Fig. 2: Agarose gel electrophoretic diagram of RT-PCR test on *Wnt-2* expression in normal prostate (NP), prostate adenocarcinoma (PCa), normal bladder (NB), bladder carcinoma (BCa), normal nasopharyngeal (NNP), and nasopharyngeal carcinoma (NPC) biopsied tissues. The size reference marker (L) used is the 100 bp DNA ladder

Ongoing efforts in this study are the verification of expected expression patterns in an increased number of samples for the three types of cancer studied. Evaluation of WNT-2 protein levels in tissues/cell lines (diseased and normal) derived from human colorectal, bladder, prostate and nasopharyngeal regions has been proposed. The culmination of these findings will allow us to accurately map the composite pathway(s) mediated by *Wnt-2* during tumourigenesis of four types of cancer. This will have potential application in detection and diagnosis of the diseases involved.

Acknowledgements

The authors thank Dr. Ong Teng Aik and Dr. Tiong Thung Sing of the Faculty of Medicine and Health Science, UNIMAS for the provision of biopsied cancer tissue specimens.

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Induction of shoot proliferation for mass propagation of papaya (*Carica papaya* L.)

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Papaya is one of the most important export fruits in Malaysia contributing to RM120 million in export revenue annually (Lim and Siti Hawa, 2007) in recent years. The species has three sex types, i.e. staminate, pistillate and hermaphrodite. Commercial production of papaya in Malaysia and other tropical regions is based on hermaphrodite varieties because of consumers' preference for sweeter taste and thicker flesh. Papaya plants derived from seeds are not true-to-type due to the inherent hereozygosity and dioecious nature of this species. When seeds are used for planting, the population will consist of dioecious and hermaphrodite plants. The usual practice is to plant three to five seedlings together and to remove all but the most vigorous hermaphrodite plant at flowering. Such over-planting situation causes plant competition and resulted weak plant with poor rooting system, long inter-node and increased height to the first flower. Vegetative propagation of papaya by rooting of cutting and grafting can produce clones of selected mother trees but is inefficient in producing large amount of desired clones for commercial scale planting. Micropropagation using the *in vitro* culture technique has been developed in several countries to mass propagate selected papaya genotype however, efficiency of rooting still need to be improved. Micropropagated papaya plants have strong root systems with multiple primary roots, fruited earlier and at a lower height.

Outbreak of Papaya Ring Spot Virus and Bacterial decline (*Erwinia* sp.) in Peninsula Malaysia and Sabah in recent years had affected production of papaya. In the absence of disease resistant varieties, Malaysian growers are interested to plant tissue-culture-derived papaya which with reduced juvenile stage could bear some fruits before the plants finally succumb to diseases. More recently, the papaya seed garden in MARDI supplying one of the two export varieties was also wiped out by the diseases. This situation had prompted us to look into adapting the *in vitro* technique for mass propagation of planting stock of our local variety of papaya.

The field-grown, healthy and vigorously growing papaya trees of the variety, 'Sekaki' which have been fruited were selected (Fig. 1a). The trees were topped to remove the shoot apices. Foliar fertilizer was sprayed onto the stem to induce sprouting of side shoots. Side shoots 10-15 cm long were severed and submersed in fingicide solution for one minute, allowed to dry and then the bases were dipped in a commercial rooting powder before inserted into the sand bin to root. The shoots rooted in one month and were transplanted singly into polythene bag filled with sterilized potting mix of peat: sand: soil in the ratio 1:1:1 (v/v) and grown inside a plant-house (Fig. 1b). Watering was carried out carefully to avoid wetting of the leaves. Plant parts above the potting medium were kept dry except during the weekly application of foliar fertilizer and fungicide. The shoot tip and axillary buds (Fig. 1 c and d) of the stock plants were collected for use as the explants for *in vitro* culture.

The explants were disinfected by agitating in 1 ml/L mild detergent for 15 minutes, dipped in 70% Ethanol for 1 minute and submerged in 20% Clorox added with 0.1 ml/L of 'Tween 20' for 15 minutes. Then the explants were rinsed three times in sterilized distilled water before inoculated on modified DS medium (Drew and Smith, 1986) incorporated with 0.5 mg/L BAP and 0.2 mg/L NAA. A mean of 90% contamination-free culture could be established. The contamination-free explants were transferred to solid proliferation medium which is having the same composition as the medium used foe

establishment except the concentration of NAA was reduced to 0.1 mg/L.

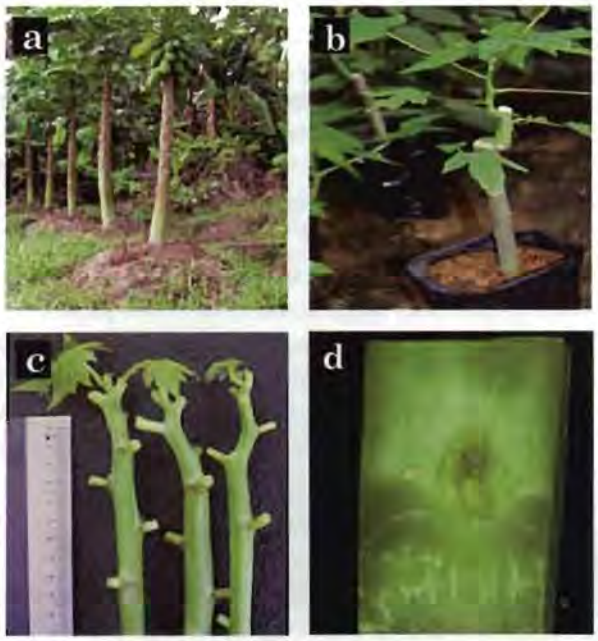

Fig. 1: Preparation of explants for *in vitro* culture

(a) Field grown selected papaya tree
(b) Rooted side shoots grown inside a plant house
(c) Side shoots to provide explant material
(d) An axillary bud from the side shoot used as the exlant for *in vitro* culture

The shoot multiplication rate in the solid proliferation medium was 2.6 shoots per explant in three weeks. A temporary immersion system RITA® (Alvard *et al.*, 1993) was also used for shoot proliferation in the liquid medium (Fig. 2a-c). It was found that four shoots per explant were developed in the liquid proliferation system after two weeks. This showed that liquid culture in a temporary immersion system is more efficient for shoot proliferation. The multiple shoots were separated into individual shoot and cultured on the proliferation medium (Fig. 2d). Each of these shoots developed four new shoots in two weeks (Fig. 2e). Shoot growth in the temporary immersion system appeared more vigorous than in the solid medium. This may be due to the direct, constant and thorough contact of the liquid medium with the explants and aeration of culture. Roots were induced in medium containing IBA at 2mg/L (Fig. 2f). However, the roots are thick and short. Study is continued in production of normal roots as well as improving the shoot multiplication rate.

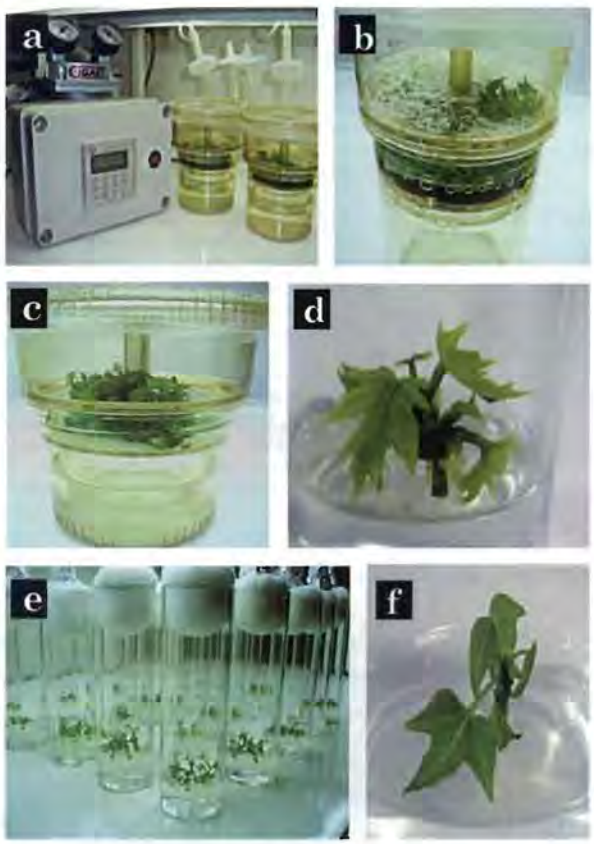

Fig. 2: *In vitro* culture of papaya in solid and liquid media

(a) The temporary immersion system with two culture vessels
(b) Shoots flooded with medium when the pump is "on"
(c) Shoots proliferated inside a vessel
(d) Proliferation of shoots in solid medium
(e) Shoots cultured in solid medium
(f) Individual shoot cultured in solid medium for rooting

For fruit trees, only those performance proven and usually field-grown and fruited trees are desired for multiplication. To clone these elite trees by *in vitro* technique, establishment of contamination-free culture is a prerequisite and is a great challenge. Explants obtained directly from the field-grown trees are loaded with micro-contaminants. In this study, we have successfully developed a system to generate 'cleaner' stock plants.

With explants obtained from these 'cleaner' stock plants, by adapting the reported protocols and using a liquid culture system which has not been reported for use in papaya, we have after ten months, established

a method of shoot proliferation for a local papaya variety called 'Sekaki'. Currently we are working on the rooting of these shoots and improving the shoot multiplication rate further. In addition, the shoots regenerated are to be grafted on seedling root stock with an aim to produce papaya tree that can fruit at lower height.

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CADAMOMICS: A genomic resource for wood formation in kelampayan, *Neolamarckia cadamba* (Roxb.) Bosser

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Forest trees represent the majority of terrestrial biomass production and a vital component of biodiversity. However, these slow growing trees are unable to meet current global demand for wood, resulting in the loss and degradation of forest. Plantation forests with the fast growing species have the potential to supply the bulk of wood needs on a long-term basis, and thus reduce the harvest pressure on natural forests for wood production to an acceptable level. *Neolamarckia cadamba* (Roxb.) Bosser or locally known as kelampayan (Fig. 1) has been identified as one of the potential fast growing species for planted forest development in Sarawak. Kelampayan is a large, deciduous and fast growing tree species that gives early economic returns within 8-10 years (Joker, 2000). It is one of the best materials for plywood industry and also serves as raw material for pulp and paper industry.

Despite the high economic value of tropical wood, little is known about the genetic control of wood formation or xylogenesis for this species compared to loblolly pine (59,797 expressed sequence tags, ESTs), poplars (25,218 ESTs) and spruce (16,430 ESTs) (Whetten *et al.*, 2001; Li *et al.*, 2009). Wood is manufactured through the process of cell division, cell expansion, secondary cell wall formation (involving cellulose, hemicellulose, cell wall proteins, and lignin biosynthesis and deposition) and programmed cell death (Li *et al.*, 2009). These processes are strongly interlinked and modulation of any one aspect of wood formation may affect many other aspects. As of July, 2009, no kelampayan EST information is available in the NCBI GenBank. Therefore, we applied genomics approaches to explore the molecular basis of wood formation in kelampayan. Here we report the generation and analysis of a genomic resource (expressed sequence tags, ESTs) database or Cadamomics for wood formation in kelampayan via high-throughput DNA sequencing of cDNA clones derived from developing xylem tissues.

Fig. 1: Morphological characteristics of *N. cadamba*

(a) The mature tree
(b) Twig with inflorescence
(c) The fruit

A total of 10,368 5' end reads were generated from the kelampayan cDNA library, yielding a total of 6,622 high quality ESTs of at least 100 bp in length. Average size of all high quality ESTs was 478 bp. The overall sequencing success rate was approximately 64 %. This was due to the presence of mononucleotide repeat motifs (polyA or polyT templates) in the sequences that causes sequence slippage problems. Assembly of 6,622 ESTs from 5' end sequences generated 4,728 xylogenesis unigenes, including 2,100 consensi and 2,628 singletons. The EST redundancy in the kelampayan EST database was 28.5%. However, the redundancy may increase during further EST sequencing (Li *et al.*, 2009). By comparison, the EST redundancy in the kelampayan

EST database is comparable to the estimated redundancy of 28 % in *Populus* (Aspeborg *et al.*, 2005) and the 28.8 % in *Pinus radiata* D. Don (Li *et al.*, 2009).

More than half (59.3 % or 3,930) of the 6,622 ESTs were assigned with putative functions using the blastx analysis (E-value $\leq 10^{-10}$) against the GenBank non-redundant protein database. However, about 24.0 % of all matches with blastx were either protein (20.4 %), predicted protein (3.0 %), hypothetical protein (0.4 %) or unnamed (0.2 %). There was a large percentage (40.7 %) of the sequences were classified as no hit or no significant similarity to sequences in GenBank (Fig. 2). This might probably due to the shorter average length (276 bp) of these sequences compared to the average length of the EST sequences showing significant similarities to the GenBank entries (616 bp). The high proportion of EST sequences did not match the known proteins indicate that novel genes can be identified in the developing xylem tissues of kelampayan.

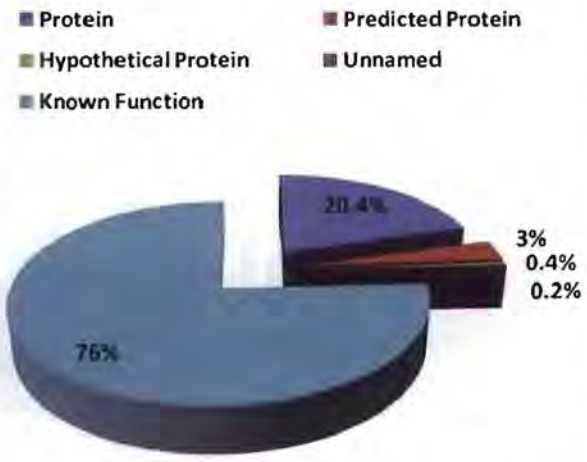

Fig. 2: Distribution of 3,930 ESTs with putative functions from *N. cadamba* developing xylem library

The most abundant protein in the ESTs whose putative function was inferred from sequence comparison was 60s ribosomal protein with 92 ESTs. Interestingly, all genes involved in lignin biosynthesis were detected in the kelampayan EST database, e.g. *cinnamate 4-hydroxylase* (*C4H*), *caffeoyl-coenzyme A O-methyltransferase* (*CCoAOMT*), *cinnamoyl-coenzyme A reductase* (*CCR*) and *cinnamyl alcohol dehydrogenase* (*CAD*). Also, several ESTs exhibiting homologies to cell-wall related proteins were also identified in the kelampayan EST database, e.g. alpha tubulin, arabinogalactan, cellulose synthase, sucrose synthase, expansin, UDP-glucose dehydrogenase and xyloglucan endotransglycosylase (Table 1). The gene ontology annotations and classifications of 4,735 xylogenesis unigenes are still in progress. Nevertheless, this study has generated an important genomic resource for wood formation in kelampayan. The identified genes in this study will provide a powerful means for identifying mechanisms controlling wood formation pathways and also will be candidates for association genetic studies in kelampayan aiming at the production of high value forests. Furthermore, comparison of kelampayan ESTs with sequences from angiosperms will also generate value added information about the evolution of higher plants.

Table 1: Thirty highly abundant genes or gene families involved in wood formation of kelampayan

Gene/ gene families	No. of ESTs
tubulin	42
arabinogalactan, AGP	30
caffeic acid O-methyltransferase, COMT	21
caffeoyl-CoA-3-O-methyltransferase, CCoAOMT	19
cinnamate 4-hydroxylase, C4H	18
cellulose synthase, CesA	13
cinnamyl alcohol dehydrogenase, CAD	11
sucrose synthase, SuSy	11
xyloglucan endotransglucosylase, XTH/XET	8
endo-β-glucanase (cellulose)	7
phenylalanine ammonia-lyase, PAL	6
ferulate 5-hydroxylase, F5H	6
expansin	6
glucan endo-β-glucosidase	6
fructokinase	4
pyrophosphate-dependent phosphofructo-kinase beta subunit	4
4-coumarate:CoA reductase, 4CL	3
cinnamoyl-CoA reductase, CCR	3
microtubule-associated protein	3
UDP-glucose dehydrogenase	3
α-expansin family protein	2
cytosolic phosphoglucomutase	2
galactosyltransferase family protein	2
pectate lyase	2
coumarate 3-hydroxylase (p-coumaryl shikimate/quinate 3-hydroxylase), C3H	1
hydroxycinnamoyl-CoA: shikimate/quinate hydroxycinnamoyl transferase, HCT	1
α-glucan-protein synthase	1
α-galactosidase	1
β-glucosidase	1

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Joker, D. (2000). Seed leaflet *Neolamarckia cadamba* (Roxb.) Bosser (*Anthocephalus chinensis* (Lam.) A. Rich. ex Walp.). URL: http://www.dfsc.dk (July 16, 2006).

Li, X., Wu, H. X., Dillon, S. K. and Southerton, S. G. (2009). Generation and analysis of expressed sequence tags from six developing xylem libraries in *Pinus radiate* D. Don. *BMC Genomics* 10: 41.

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Study on the complexity of *Costus globosus* Bl. in Sarawak

Meekiong Kalu, Norazima Ali Hossein, Isa Bin Ipor and Cheksum Supiah Tawan

Department of Plant Science and Environmental Ecology

The genus *Costus* or locally known as setawar hutan or spiral ginger was previously classified under the family Zingiberaceae (Schumann, 1904) until lately has been separated as a distinct family known as Costaceae (Specht *el al.*, 2001 Kress *et al.*, 2002). The diagnostic characteristics which have been used for the separation were based on absence of aromatic oils, the branched aerial stems with spiral monostichous phyllotaxy and one-sided spiral leaves arrangement or by, some merely a special branch compared to the traditional members of the family Zingiberaceae. The family Costaceae is notably pantropical, represented in the Neotropics (*c.*60 species), tropical Africa (*c.*25 species) and only five species in Asia (Maas, 1979). Hitherto, only three species have been recorded in Sarawak (Meekiong *et al.*, 2006).

This study is a part of on-going research on the Costaceae in Sarawak, focusing on the *Costus globosus* Bl. complex. Few species such as *C. acanthocephalus, C. tonkinensis, C. globosus, C. kingii, C. kunstleri, C. ridleyii* and *C. velutinus* or so called *Costus* species from Asia were assembled together as *C. globosus* complex (Holttum, 1950; Maas 1979; Specth and Stevenson, 2006). This group of *Costus* has not been extensively studied and classified. This study is designed to describe morphological and anatomical variations within the *Costus globosus* complex. Other approach, molecular techniques were applied to verify the status of each taxa within the complex. The aim of this study is to unravel taxonomic concern on *C. globosus* (Fig. 1) complex and to pursue appropriate classification between conventional Holttum's classification (Holttum, 1950) and molecular approaches by Specht and Stevenson (2006).

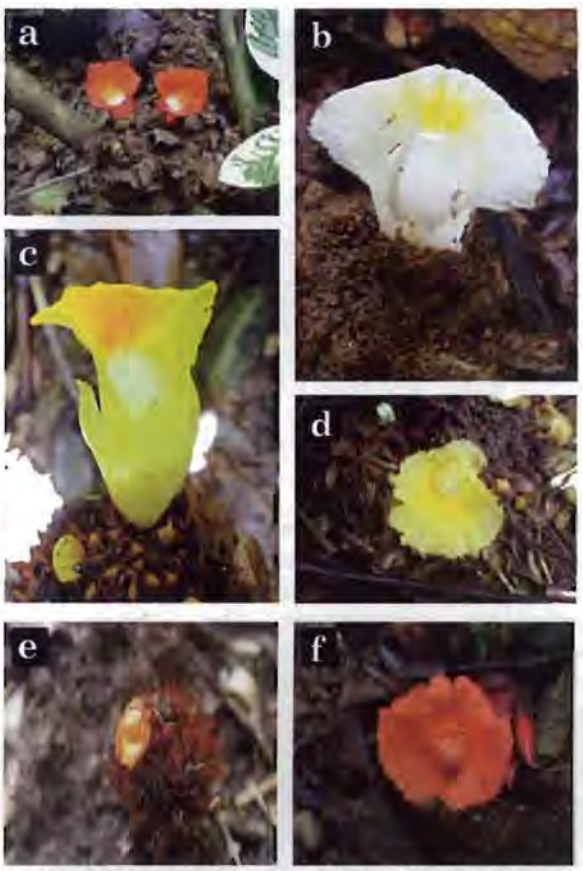

Fig. 1: Flowers of *Costus globosus* found in Sarawak

(a) red form
(b) white form
(c) yellow form
(d) hairy yellow form
(e) orange form
(f) hairy orange form

Prefacing data from field studies based on morphological and anatomical has initiated a remarkable variation within the *C. globosus*. Colour variations and hairiness were the two distinctive features spotted during the studies. Eight variants so far have been recognized among the *C. globosus* complex and among these two of them were identified and described as different species. As to solve *C. globosus* complex of Sarawak, molecular biological techniques will be applied to support morphological data and verify the status of each taxa within the complex.

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Application of synthetic pathway for organotin(IV) derivatives using the pyruvic acid isonicotinoyl hydrazone as the chelating agent

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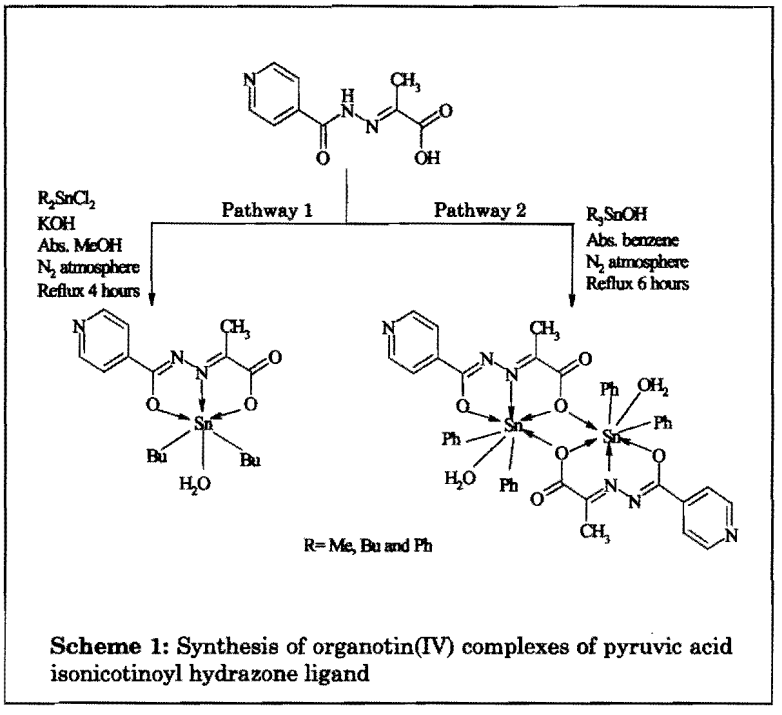
Department of Chemistry

All roads lead to Rome; in order to achieve the ultimate goal in no time we need to lead the right route. A right synthetic pathway initially developed for the preparation of coordination compounds by many researchers (Pellerito and Nagy, 2002; Raveendran and Pal, 2005). Chemists always look into several aspects when come to the selection of synthetic approach; feasibility, economic, greener, high yield, faster reaction and different product yield are the key features that have to take into consideration. The preparation, characterization and application of coordination complexes have become a very important aspect in coordination chemistry in the past two decades (Davies, 1997). The increasing interest in this field is due to the potential relevance of such compounds in academic and applied research. For coordination chemistry, the ligand is interesting due to its potential multiple coordinate possibilities and the same ligand can produce different types of coordination complexes in high yield by employing a different synthetic routes. Yamamoto and Yanagi (1982) showed that 2,6-*bis* (trimethyltin)pyridine could be synthesized by reacting sodium trimethylstannane with 2,6-dicholro pyridine nearly two decades ago. Later, Ulrich and Eschbaumer (1999) have synthesized similar 2,6-*bis*(trimethyltin)pyridine starting with 2,6-dichloro pyridine after modification and optimization of the published procedure the yield was increased to 69%.

The above considerations stirred our interest in some detailed synthesis and patterns for organotin(IV) complexes of the pyruvic acid isonicotinoyl hydrazone. To our surprise, we obtained a series of organotin(IV) compounds with different coordination modes by reaction of the pyruvic acid isonicotinoyl hydrazone with R_3SnOH or R_2SnCl_2 (R= Me, Bu and Ph) under N_2 atmosphere. KOH was used as base with R_2SnCl_2 in the formation of complex compound.

When using a 1:1:1 mole ratio of ligand:R_2SnCl_2:KOH in anhydrous MeOH, we obtained a group of monomeric five coordinated organotin(IV) complexes. However, when the ligand and R_3SnOH reacted in absolute benzene, seven coordinated dimeric complexes were obtained (Scheme 1).

From the organotin(IV) complexes reported here, we can see that two types of organotin(IV) derivatives obtained from the new synthetic pathway. We can conclude that the pyruvic acid isonicotinoyl hydrazone ligand play important roles in the formation of monomeric and dimeric organotin(IV) complexes and it form coordinate covalent bonds to tin(IV) centres through the enolic-O, azomethine-N and hydroxyl-O atoms.



Scheme 1: Synthesis of organotin(IV) complexes of pyruvic acid isonicotinoyl hydrazone ligand

Based on the organotin(IV) complexes reported, two types of organotin(IV) derivatives obtained from the new synthetic pathway. It could be concluded that the pyruvic acid isonicotinoyl hydrazone ligand play important roles in the formation of monomeric and dimeric organotin(IV) complexes and it form coordinate covalent bonds to tin(IV) centres through the enolic-O, azomethine-N and hydroxyl-O atoms.

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Alcohol dehydrogenase of sago palm

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Alcohol dehydrogenases (ADH, EC 1.1.1.1) is an enzyme that converts pyruvate into ethanol with the concomitant regeneration of NAD+ in the fermentation pathway of yeast, higher plants and in some bacteria. It is a readily assayed enzyme whose activity has been detected in vast number of higher plants consisting *Arabidopsis*, maize, pearlmillet, sunflower, wheat, palms, rice, tomato and pea. The role of ADH is not just restricted for plant cell survival during low oxygen stress in waterlogged roots but also involved in various other reactions such as in the production of aromatic compounds, acetaldehyde detoxification, carbon reutilization, interconversion of aldehydes and alcohols in tomato plant to enhance the flavor of the fruit and in seed germination of rice crop.

The work in detecting the expression of *Adh* in sago palm have been undertaken for the past six years. We have shown that the ADH protein is present in various tissues such as the dry and submerged roots, young and mature leaves. Expression pattern (Fig. 1) also differs between the tissues with highest ADH detected in young leaf, followed by submerged and dry leaves, and finally mature leaf (Roslan *et al.*, 2008).

Following on the protein results, cDNA work was conducted in order to isolate the *Adh* cDNA from young leaves. Differential display work have also indicated several fragments that were differentially expressed in the young and mature leaves (Fig. 2) although the work involving submerged and dry roots remained inconclusive as further analyses is still required. Through 3' and 5' rapid amplification of cDNA ends (RACE) technique (Frohman *et al.*, 1988), we have isolated a complete cDNA of *Adh1* gene from sago palm with approximately 1351 base pairs (bp) of cDNA including the 3' untranslated region that corresponds to 380 amino acids (Fig. 3).

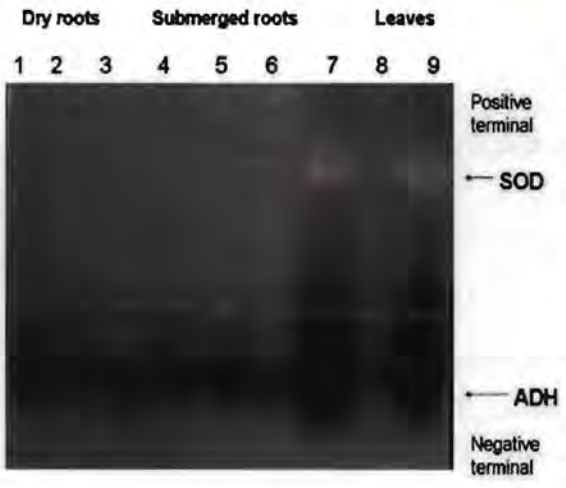

Fig. 1: Polyacrylamide gel with specific protein staining of Adh and superoxide dismutase (SOD). The dry (lanes 1-3) and submerged roots (lanes 4-6), young leaves (lanes 7 and 9) and mature leaf (lane 8) showing different intensities of ADH protein (Roslan *et al.*, 2008)

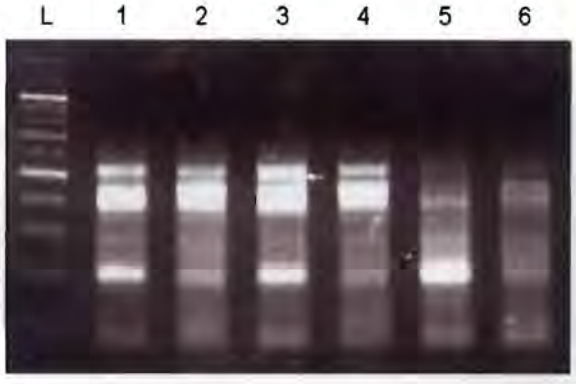

Fig. 2: Agarose gel electrophoresis of the products from differential display for young (lanes 1, 3 and 5) and mature (lanes 2, 3 and 6) leaves showing bands that were differentially expressed in the two tissues, L is 1kb DNA ladder

MASTVGQVIKCRAAVSWEAGKPLVMEEVEVAPPQAME
VRMKILYTSLCHTDVYFWEAKGQTPVFPRIFGHEAGGII
ESVGEGVTELAPGDHVLPIFTGECKECAHCKSEESNMC
DLLRINTDRGVMINDGKSRFTINGKPIYHFLGTSTFSEY
TVVHVGCVAKINPLAPLDKVCVLSCGISTGFGATVNVAK
PPKGSTVAVFGLGAVGLAAAEGARASGASRIIGVDVNPK
RFEEAMKFGCTEFVNPMDHDKPVQEVIAEMTNGGVDR
SVECTGNINAMISAFECVHDGWGVAVLVGVPHKEAEFK
THPMNFLNERTLKGTFFGNYKPRSDIPAVVEKYMNKEL
ELEKFITHSVPFSEINKTFDYMLKGESLRCIIHMDG

Fig. 3: The deduced amino acid sequence for *Adh* isolated from sago palm.

Current work analyzing the nucleotide sequence and in isolating the factors that regulate the expression of *Adh* is on-going. In the future, the *Adh* cDNA will be heterologously expressed in order to determine its function in plants.

Acknowledgements

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Determination and morphology of leaf starch granules from trunking and non-trunking sago palm leaves

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Sago palm is from the genus *Metroxylon* which is widely distributed from Thailand, Malaysia and Indonesia to Micronesia, Fiji and Samoa. In Sarawak the areas around Mukah and Dalat are among the places that have sago palms grown for commercial plantations. The sago palms found in plantations and grown wildly vary from 9 to 33 m.

Sago palms main product is starch and the storage organ for starch in sago palm is the trunk or bole of the plant. Sago palms that produce starch are termed as trunking sago palm while those that do not produce starch are called non-trunking sago palm. In plantations, both type of trunking can be found. According to Siong *et al.* (2005), the trunking palm is grown on the shallow peat which contains elevated ranks of ash such as minerals and nutrients such as nitrogen, phosphorus and potassium which are required to sustain the natural development and trunking of palms. On the other hand, the non-trunking sago palm is grown on the deep peat which is deficit of nutrient which is required to sustain the natural development and trunking of the palms (Wan Sulaiman *et al.*, 2005).

Starch that is found in the trunk of sago palm is initially produced in the leaves during the day and degraded into smaller molecules such as glucose and maltose by enzymes at night. The glucose and maltose produced is then transported to the trunk. In the trunk of sago palm, storage starch is produced and stored. There has been many research conducted previously to study the starch granules from the storage area of sago palms. One of the research conducted by Nozaki *et al.* (2004), studied the starch granule size from starch storage area which is the trunk of sago palm grown on different types of soil especially the acidic sulfate and mineral soil.

Besides that, there has also been a research conducted recently by Tie *et al.* (2008) to study the starch morphology from the trunk of sago palm at different growth stages. There has not been any research conducted as up to date to study the starch granule morphological structure from the starch synthesis area which is the leaves of sago palms. Therefore, this work presents the initial finding in determining and comparing the starch granule structure and morphology from both the trunking and non-trunking sago palm leaves.

From the study, the starch granules from the trunking sago palm leaves showed that shape of the extracted starch granules are oval and the size of the starch granules are 10 μm (Fig. 1). In addition, many different sizes of starch granule from the trunking sago palm leaves can also be observed which ranges from 1 μm to 5 μm (Fig. 2). The shapes of the starch granules from the trunking sago palm leaves observed (Fig. 2) were truncated and oval.

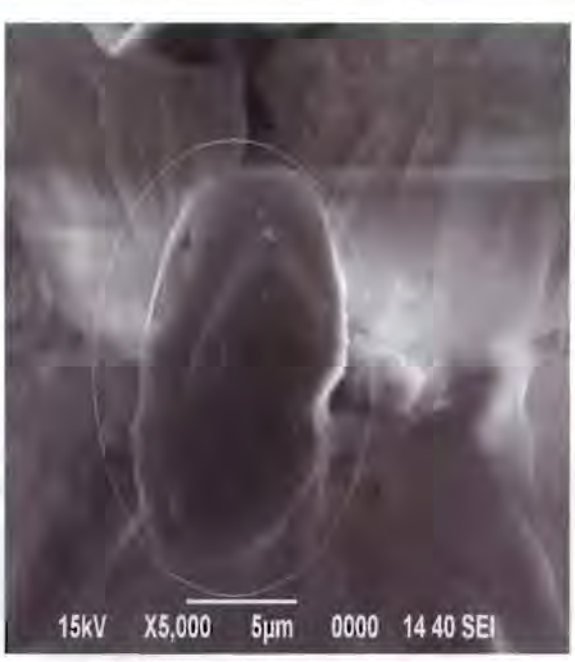

Fig. 1: The starch granules extracted from trunking sago palm leaves which have an oval shape

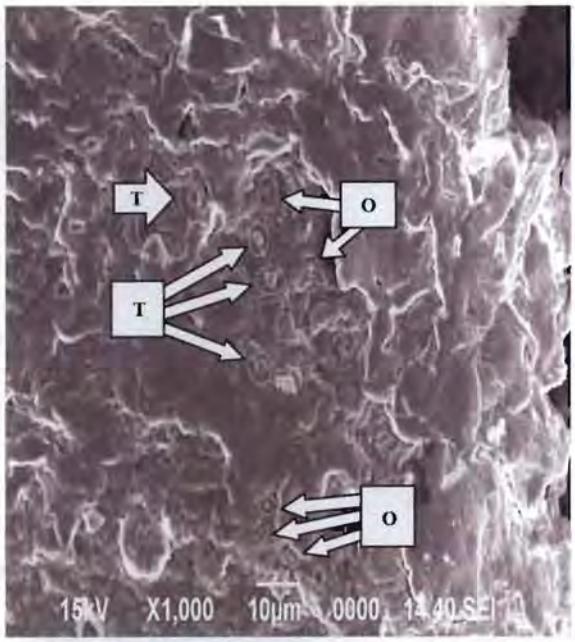

Fig. 2: Many different sizes of the starch granule from the trunking sago palm leaves (T: truncated, O: oval)

The results obtained from the non-trunking sago palm leaves sample shows that the structure of starch granule from non-trunking sago palm leaves are oval and truncated. The size of the granules from the non-trunking sago palm leaves range from 2-6 µm(Fig. 3 and Fig. 4).

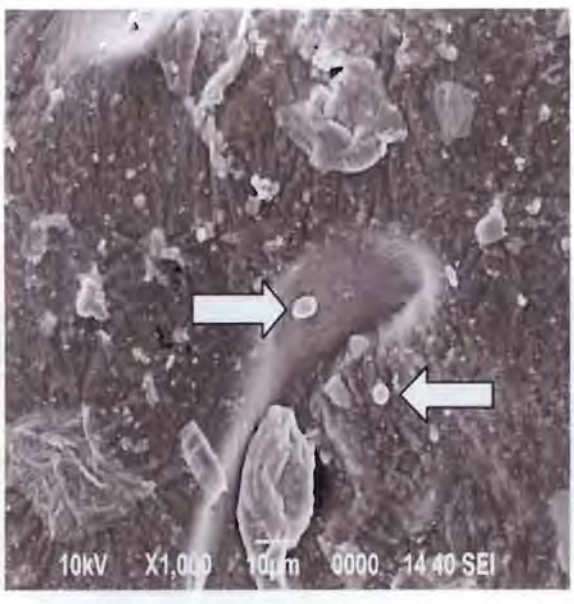

Fig. 3: Starch granule structure from the non-trunking sago palm leaves

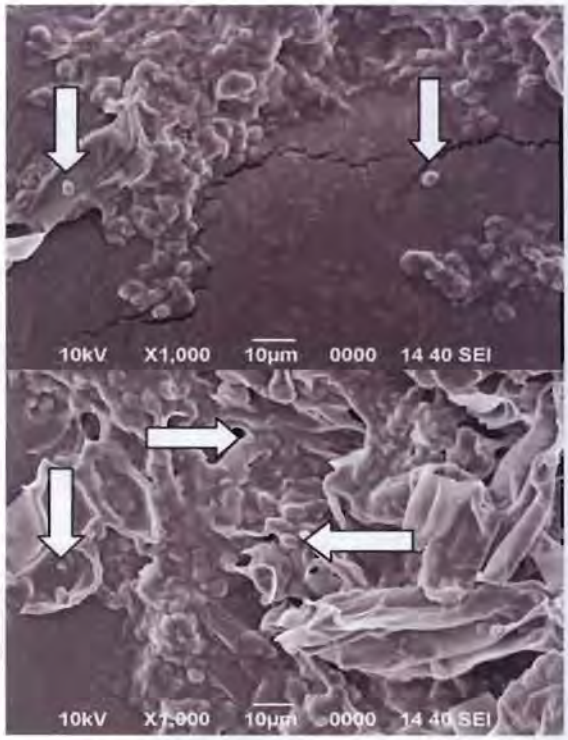

Fig. 4: Starch granules structure from non-trunking sago palm leaves with truncated and oval shape

Initial findings of this work showed that there are no observable differences between the shape of the starch granule from trunking and non-trunking sago palm leaves. In addition, it was shown that the size of starch granules from trunking and non-trunking sago palm varies between 1 to 10 µm with no definitive size to differentiate between the two different groups of plants. Further work will be done to determine the size, number of granules at different time throughout the day.

Acknowledgements

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