



INDUSTRIAL TECHNOLOGY INSTITUTE



2ND BIENNIAL RESEARCH SYMPOSIUM

SCIENCE
TECHNOLOGY &
INNOVATION
for Knowledge based Economy

16-17
November 2015

PROCEEDINGS



**Biennial
Research
Symposium
2015**



Industrial Technology Institute

**Biennial Research Symposium
2015**

Science, Technology & Innovation for a
Knowledge Based Economy

ABSTRACTS

16th – 17th November 2015

Industrial Technology Institute

Biennial Research Symposium 2015

Science, Technology & Innovation for a Knowledge Based
Economy

16th – 17th November 2015

Main Auditorium

ABSTRACTS

Industrial Technology Institute
363, Bauddhaloka Mawatha
Colombo 07
Sri Lanka
www.iti.lk

This book contains the abstracts of papers presented at the Biennial Research Symposium 2015 of Industrial Technology Institute of Sri Lanka held on 16th -17th November 2015. No part of this publication may be reproduced, stored in a retrieval system or transmitted in any form without prior permission of Industrial Technology Institute of Sri Lanka.

The material in this publication has been supplied by the authors, and only minor copy editing, if relevant, has been done by the editors.

Editor of Proceedings - Dr. G.A.S. Premakumara
Editorial Committee - Dr. Ilmi Hewajulige
Ms. Purnima Jayasinha
Ms. K.H. Thushara Abeysekera

Published by Industrial Technology Institute
363, Bauddhaloka Mawatha
Colombo 07
Sri Lanka
Tel: +94-11-2379800
E-mail: info@iti.lk
Website: <http://www.iti.lk>

Published date: 16th November 2015

Symposium Chair Dr. Radhika Samarasekera

Symposium Secretary Dr. Ilmi Hewajulige

Organizing Committee Dr. G.A.S. Premakumara
Mr. A. Pannila
Ms. Manori Wijemanna
Ms. Purnima Jayasinha
Mr. Keerthi Fonseka
Dr. Iresha Kottegoda
Dr. P. Ranasinghe
Dr. W.M.S. Wijesinghe
Mr. Ruwan Weerasinghe
Dr. Maheshi Athapaththu
Dr. Yrosh Fernando
Ms. Hasanthi Disanayake
Ms. Nethra Tantrigoda
Ms. P. Perera

Review Committee Dr. I. R. M. Kottegoda
Dr. P. Ranasinghe
Dr. W. M. S. Wijesinghe
Mr. H. P. P. S. Somasiri
Dr. A. M. M. H. Athapaththu
Ms. Dushyanthi Ranpatige

Message from the Minister of Science, Technology and Research



I am pleased to send this message for the 2nd Biennial Research Symposium 2015 of the Industrial Technology Institute on “Science, Technology and Innovation for a knowledge based Economy”. The symposium will provide a platform for sharing knowledge and also to discuss issues related to science, technology and its impact on the economy of the country.

Science and technology must be used in a way that it will improve the quality of life of the mankind. By doing so, we can make a long-term sustainable development of the country. Also we are, from the historic times, are suppliers of quality raw material to the world. But now the time has come to convert these raw materials to end products and earn more foreign currency enriching the country’s economy.

I take this opportunity to convey my appreciation to the organizing committee of the symposium for their untiring effort to make this event a success.

Hon. A.D. Susil Premajayantha
Minister of Science, Technology and Research
LLB (Colombo), MPA (Sri Jayawardanapura)
Attorney-at-law & N.P., Commissioner for Oaths

Message from the Secretary, Ministry of Science, Technology and Research



It is a great pleasure to send this message for the 2nd Biennial Research Symposium 2015 of the Industrial Technology Institute. The theme of the symposium “Science, Technology and Innovation for a knowledge based Economy” is very suitable for the country at present as science, technology and innovations are interwoven with the economy in today’s world. The symposium will prove the dedication and responsibility of ITI towards the people of Sri Lanka.

The research symposium will create a platform for ITI’s expert researchers to communicate their scientific findings to the general public.

I take this opportunity to thank the organizing committee of the symposium and wish all the best for the presenters.

Mrs. R. Wijialudchumi
Secretary
Ministry of Science, Technology and Research

Message from the Chairman of ITI



The Industrial Technology Institute, for the second time, is successful in holding a Biennial Research Symposium under the theme “Science, Technology and Innovation for a Knowledge based Economy”.

As a leading Research Institution in Sri Lanka, ITI is mandated to elevate the level of technology and thereby be a strength to the industries. Thus, ITI has chosen the most appropriate theme for their Research Symposium.

It is expected that this symposium would be an event where the scientists and technologists at ITI will, disseminate their knowledge and experience to the general public. Therefore I am confident that the symposium would be of greater assistance to elevate the economy of our country by empowering the general public with scientific knowledge to enhance the value of their industries.

As the Institution is also celebrating its 60th Anniversary this year, I am confident that the symposium would be another milestone in their anniversary celebrations.

In conclusion, let me congratulate the organising committee for their efforts in organising a symposium of this nature and I sincerely hope that ITI will continue to organise and conduct similar activities to strengthen the Sri Lankan economy in the future.

Niroshana Perera
Attorney-at-Law
Chairman
Industrial Technology Institute

Message from the Director General of ITI



It is a great pleasure for me to give this message on the occasion of the 2nd Biennial Research Symposium of ITI. The theme of this symposium “Science, Technology and Innovation for a Knowledge based Economy” gives the opportunity to share the knowledge and the experience among the researchers and to introduce new innovations and technologies that would generate significant economic growth and

business opportunities in the country.

Industrial Technology Institute is a public entity and we have the responsibility towards the people of the country. It is our responsibility to share the scientific knowledge we created through public funds with the people of the country. The symposium is a forum where the knowledge created by ITI scientists is disseminated to the people.

The Symposium will contain key note address, plenary sessions, paper presentations and workshops.

I would like to take this opportunity to thank and congratulate the Chairman and the Organization Committee for their untiring effort to make this event a success.

I wish all the best for the symposium.

Dr. G.A.S. Premakumara

BSc, PhD, Dip (Psy), MIChemC, MIBiol, CChem, CBiol, FIBiol

Director General

Industrial Technology Institute

Message from the Symposium Organizing Chair



It is my pleasure and honor to welcome the scientists, engineers and technologists from research institutes, academia, other organizations and Industrial Technology Institute and other invitees to the second Biennial Research Symposium of ITI.

The second Biennial Research Symposium, which will be held from 16-17th November 2015, is a major event of ITI to be staged in Colombo during the Science Week. This event is expected to stimulate and foster the growth of scientific and technology research towards the development of the country. The research symposium will be declared open by the Honorable Susil Premajayanth, Minister for Science, Technology and Research.

The technical sessions of the symposium will be held on the 17th November 2015 at ITI with a special focus on Food, Herbal, Environmental, Electro, Material and Bio technology. A wealth of new knowledge is bound to unfold at the symposium and much collaboration and partnerships will begin.

As the Organizing Chair I am privileged and honored to organize the second Biennial Research Symposium on the theme “Science, Technology and Innovation for a Knowledge Based Economy”. My sincere thanks are due to Chairman & Board of Management, Director General and every member of the organizing committee for making the symposium a successful and memorable event.

I wish all of you a most enjoyable and interactive symposium.

Dr. Radhika Samarasekera

BSc (Hons), PhD (UK), FICChemC, MIChemC, CChem, FIBiol, CBiol, MIBiol

Organizing Chair & Additional Director General – R&D

Industrial Technology Institute

Message from Executive Director, COMSATS



It is a matter of pleasure for COMSATS to get the notification of the biennial Research Symposium of ITI to be held on 16-17 Nov., 2015. The theme of the symposium has been aptly chosen to reflect the current international trends and the focus of R&D activities at ITI. I am sure that this symposium will lead to better understanding of a knowledge economy and motivate researchers in ITI and other S&T organizations of Sri Lanka to apply their knowledge and skills towards economic development.

It is now well-established that economic development, competitiveness and wealth generation in today's world depends on the abilities of nations to create and harness knowledge-based technologies and services. Two essential ingredients of this prescription are: (i) the adoption of a Science, Technology and Innovation Policy; and (ii) putting in place a robust infrastructure for Research and Development. The second element of this approach further depends on the availability of financial and human resources which are generally in deficit in developing countries. The ultimate objective of generating globally competitive economic activity is further hampered by the lack of coordination among financial, legal, administrative, industrial and trade-related institutions. The state intervention is required in all sectors to generate a conducive innovation ecosystem. Once the local environment with respect to policies and practices becomes innovation friendly, eventually the international arena with its compulsion of Intellectual Property Rights, export controls, trade agreements, promotions, and market access come in the fore for wealth generating economic activity.

Keeping in view the complexity of creating and maintaining a viable knowledge-based economy, COMSATS has been holding region-specific workshops with generic title of “National Innovation System and Intellectual Property”. Three workshops have been held jointly by COMSATS and ISECO for the regions of Asia (in 2013), Africa (in 2014) and Arab (in 2015).

COMSATS recognizes the strong role ITI is playing in the socio-economic development of Sri Lanka, through scientific Research in its excellent laboratories and through creating awareness about issues relevant to knowledge commercialization.

I wish all the best for the successful organization of the ITI biennial Research Symposium and extend my full support to ITI in its role as COMSATS Centre of Excellence in Sri Lanka.

Dr. Imtinan Elahi Qureshi
Executive Director COMSATS

Contents

	No.
Message from the Minister of Science, Technology and Research	iv
Message from the Secretary, Ministry of Science, Technology and Research	v
Message from the Chairman of ITI	vi
Message from the Director General of ITI	vii
Message from the Symposium Organizing Chair	vii
Message from Executive Director, COMSATS	ix
Keynote address	xv

Technical Sessions

A comparative study on antioxidant properties of selected whole grain cereals: A preliminary <i>in vitro</i> study	1
Comparison of two packaging materials for the storage of symbiotic dairy beverage	3
Formulation of dietary fibre enhanced cracker using locally available whole grain cereals and legumes	4
Optimization of liquefaction and saccharification times for laboratory scale production of glucose syrup from Sri Lankan cassava starch	6
Study on amylose content, rheological properties and particle size distribution of Sri Lankan banana (<i>Musa spp</i>) flour	7
Effect of incorporation of rice bran on cooking, textural and sensory characteristics and nutritional composition of rice noodle	9

Efficacy of microfiltration and thermal process technology for development of super health drinks	10
Improving quality of mango (<i>Mangifera indica</i>) var. Karthakolomban by postharvest application of new edible wax formulations	12
Physicochemical and functional properties of Finger millet (<i>Eleusine coracana</i>) flour and its potential use in food product development	14
Evaluation of six wild fruit extracts as sources of antioxidant in nutraceutical and cosmetic products	16
Ethnopharmacological survey on medicinal plants used in snake bite treatments in Western and Sabaragamuwa provinces in Sri Lanka	18
Chemical composition of essential oils of different parts of <i>Myristica fragrans</i> (nutmeg) and <i>Syzygium aromaticum</i> (clove) grown in Sri Lanka	19
Anti-inflammatory volatile constituents from rhizomes and leaves of <i>Alpinia calcarata</i>	21
Chemical characterization of Ceylon cinnamon (<i>Cinnamomum zeylanicum</i> Blume) bark oils distilled from Galle and Ratnapura districts	23
Comparative identification of wild and domesticated bees honey	25
Determination and Isolation of marine derived Secondary Metabolites in Sri Lankan Marine Algae through Electron Spin Resonance, ABTS ⁺ on-line HPLC and High performance Centrifugal Partition	26

Chromatography (HPCPC)

<i>In vitro</i> Tyrosinase Inhibitory Activity of Some Selected Sri Lankan Medicinal Plants	28
Insecticidal indigenous <i>Bacillus thuringiensis</i> strains with potential Lepidopteran activity	30
<i>In vitro</i> anti-arachidonate 5-lipoxygenase activity of selected Sri Lankan medicinal plants	32
Development of a molecular assay to differentiate yellowfin tuna (<i>Thunnus albacares</i>) from other tuna species commonly found in Sri Lanka	34
Optimization of a DNA extraction method to isolate DNA from processed food samples for the detection of Genetically Modified (GM) Food	35
Amplification and cloning of thermo-stable alpha-amylase gene from <i>Geobacillus stercorophilus</i>	36
Development of a DNA rabies vaccine for dogs using glycoprotein gene of Rabies Virus	37
Trends of Dengue Incidence in Kundasale Medical Officer of Health (MOH) area, Central province, Sri Lanka	38
Optimization of Reverse Transcriptase Polymerase Chain Reaction based method for the detection of the Dengue Virus	40
Prevalence of Dengue Fever (DF) and Dengue Hemorrhagic Fever	41

(DHF) in the Gampaha District of Sri Lanka: A cross sectional study in a selected population of clinically suspected dengue patients	
Cloning of a Rabies Virus specific glycoprotein coding gene into a bacterial expression system for the expression of a recombinant protein	43
Determination of ethanol content in colognes from Sri Lankan market using a validated gas chromatographic-flame ionization detection (GC-FID) method	44
Characterization of banana fiber extracted from common banana varieties in Sri Lanka	45
Optimization of process of synthesis of graphite oxide from Kahatagaha graphite	46
Microbial Bioremediation of petroleum hydrocarbon contaminated soil and water	47
Advanced Air Dispersion Modeling of SO ₂ to Accommodate Air Polluting Industries in Proposed Industrial Zone at Hambantota	49
Evaluation of road traffic noise in Sri Lanka as a case study	50
Fabrication of a machine to calibrate the measuring tapes by mechanical comparison technique	51

Keynote Address - Abstract

Science and Technology have helped mankind to expand the frontiers of human civilisations from time immemorial. Yet, when market economy systems commenced ruling the world, science and technology alone could not serve mankind unless they are followed by innovation, a process, according to economists, of converting the products of science and technology to commercially viable goods and services. Hence, in today's world, it is the combination of three agents that would lead the world to prosperity: researchers who would come up with inventions, entrepreneurs who innovate them and governments that facilitate the working arrangements between the first two agents. Accordingly, to make the combination of science, technology and innovation a reality, research institutions should be connected to industry. Numerous examples are found in the Western world for such effective linking of research with industry. In this new model, scientific towns have been set up around famous research universities with active participation of world renowned private corporations. This is the new model which Sri Lanka should follow in order to create wealth and prosperity for Sri Lankans for gaining capacity to convert today's simple product economy to a complex product economy tomorrow. In the proposed Megapolis project of the government, special technopolis areas could be created with the participation of research institutions like ITI and universities, on one hand, and industrial firms, on the other. It is the duty of all government agencies to work together to help the nation to realise this objective.

W.A Wijewardena

Formerly Deputy Governor, Central Bank of Sri Lanka and

Presently, President, BMS, Sri Lanka and

Adjunct Faculty, Asian Institute of Technology, Thailand

A comparative study on antioxidant properties of selected whole grain cereals: A preliminary *in vitro* study

S.A.S Jayawardana¹, W.K.S.M. Abeysekera^{1*}, S. Yathursan¹, P. Ranasinghe²,

W.P.K.M. Abeysekera² and G.A.S. Premakumara²

¹*Food Technology Section, Industrial Technology Institute, Sri Lanka*

²*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

Cereals and cereal based foods have been the basis of the human diet since ancient times. Current dietary guidelines all over the world recommend the consumption of whole grain cereals as they have the ability to enhance health beyond basic nutrition. Antioxidants are secondary metabolites which have the ability to reduce the oxidative stress occurred in various diseases such as cardiovascular diseases, inflammatory diseases, ageing and cancer. Cereals are reported to have antioxidant properties (AP) but comparative studies on AP of different cereals are limited. Present study evaluates AP of some selected cereals.

Whole grains of rice (improved: BW 361 and Bg 359; traditional: Kalu Heeneti and Suwadal), corn, barley, finger millet and oat were used in this study. Ethanolic and methanolic extracts (MEs) of whole grain cereals were evaluated for Total Polyphenolic Content (TPC), Ferric Reducing Antioxidant Power (FRAP), Oxygen Radical Absorbance Capacity (ORAC), 1,1-diphenyl-2-picryl-hydrazyl (DPPH) radical scavenging and 2-azino-bis (3-ethylbenzothiazoline-6-sulfonic) acid (ABTS) radical scavenging AP *in vitro* (n=3 each).

Results showed significant differences (P<0.05) between extracts and among cereals for investigated for AP. MEs of cereals showed significantly high (P<0.05) antioxidant activity compared to ethanolic extracts of cereals (except TPC: Bg 359; FRAP: Bg 359; DPPH: Suwadal, Kalu Heeneti and Bw 361; ORAC: oat and Bg 359). Among the studied cereals finger millet exhibited the highest antioxidant activities for both extracts for all the investigated AP. The mean TPC, FRAP, ORAC, ABTS and DPPH radical scavenging AP of MEs of finger millet were 708.45 ± 11.72 mg Gallic acid equivalents and 1413.69 ± 1.61 , 1240.45 ± 67.38 , 955.76 ± 2.03 and 1225.22 ± 2.16 mg Trolox equivalents/100 g grain, respectively. The second highest AP for TPC and ABTS radical scavenging activity was observed for MEs of red rices Kalu Heeneti and BW 361 while

for FRAP it was ME of Kalu Heeneti. For ABTS and ORAC, MEs of barley and Suwadal showed the second highest AP, respectively. Lowest TPC, FRAP and ORAC activities were observed for MEs of oat and this was 41, 23, 24 times lower, respectively compared to MEs of finger millet. Ethanolic extract of corn and ME of Suwadal did not show ABTS and DPPH radical scavenging activities, respectively at tested concentrations. It is concluded that finger millet possesses the highest AP followed by red rices.

Acknowledgement: Treasury Grant TG 11/37

Comparison of two packaging materials for the storage of symbiotic dairy beverage

D.U.Rajawardana^{1*}, P.N Raju², H.M Rashmi² and S. Ganguli²

¹Food Technology Section, Industrial Technology Institute, Sri Lanka

²National Dairy Research Institute, Karnal, Haryana, India

The interest in developing probiotic functional food is increasing largely as they have shown positive effects on overall health and wellbeing. Production of probiotic foods is a challenge as many active cultures may destroy and product properties may change during storage. Therefore the aim of the present study was to compare two packaging materials for the storage of probiotic foods. *Lactobacillus plantarum* (Lp9) was used as the probiotic to develop oat and honey mixed synbiotic dairy beverage, optimized applying response surface methodology, using Design-Expert software v9. Survival ability of probiotics, chemical and organoleptic properties of the product stored in high gas impermeable EVOH (Ethylene vinyl alcohol) and LDPE (Low-Density Polyethylene) materials were assessed during refrigerated storage ($5\pm 1^{\circ}\text{C}$) in order to determine the shelf-life. The beverage contained $8.4 \log_{10}$ CFU mL^{-1} viable probiotic count, pH value of 4.48 ± 0.02 and 0.663 ± 0.029 titratable acidity as a percentage lactic acid. Viable probiotic counts of the samples stored in both packaging materials were within the acceptable range for a probiotic food ($>10^6$ CFU mL^{-1}) at the end of 14 days in refrigerated storage. According to the chemical and organoleptic scores obtained, the shelf-life of the beverage stored in high gas impermeable EVOH material was up to 10 days and LDPE material was only up to 6 days. Therefore, EVOH material could be a suitable packaging material for storing probiotic beverages to preserve the viability of probiotic organisms while safeguarding the organoleptic properties.

Formulation of dietary fibre enhanced cracker using locally available whole grain cereals and legumes

K.M.D.T.Rupasinghe¹, M.D.W.Samaranayake², D.M.K. Aponso², H.M.T. Herath^{2*}

¹*Department of Food Science and Technology, Faculty of Livestock, Fisheries and Nutrition, Wayamba University of Sri Lanka*

²*Food Technology Section, Industrial Technology Institute, Sri Lanka*

Documentary evidence suggests that there is an inverse relationship between intake of dietary fiber and risk for developing Non Communicable Diseases (NCDs) in adults. Present study focused on formulating a cracker with enhanced dietary fiber content by incorporating selected locally available whole grain cereals and legumes.

Flour composition of a normal cracker formulation (*i.e.* 100 % wheat flour) was substituted at levels of 50 %, 40 % and 30 % with a mixed-grain flour containing finger millet (FM), brown rice (BR) and legume; either mung bean (MB), chick pea (CP) or soya bean (SB) at different ratios. Since 50% substitution (BR: FM: legume=1.5:1.5:2) did not succeed in achieving required rheological properties of sheeting, the study was continued with 40 % and 30 % substitutions. The 40 % substitution of wheat flour with the mixed-grain flour was only feasible when incorporating chick pea as legume flour. Three formulations of cracker with 40% substitution were repeated at 1:1:2 (F1), 2:1:1(F2) and 1:2:1(F3) ratios of BR: FM: CP, respectively. The 30 % substitution was possible to formulate with all selected legumes (at 1:1:1 ratio of BR: FM:legume).

Nutritional composition (NC) and Dietary Fiber (DF) content of both raw materials and final products, glycemic response (*In-vitro*) and sensory attributes of formulated crackers were determined for crackers with 40% substitutions (F1, F2 and F3). Of the raw materials, SB had the significantly ($p \leq 0.05$) higher DF content of 24.88% while, CP, MB, FM and BR had a DF content of 11.27 %, 10.58 %, 9.57 % and 2.99 %, respectively. The highest DF content of 3.91 % was obtained in the 40% substitution of mixed-grain flour at ratio of 1:1:2 in BR: FM: CP(F1) and it was significantly ($p \leq 0.05$) higher than the control wheat cracker. However, no significant difference was observed among F1, F2 and F3 in DF content. From the formulated crackers, the lowest reducing sugar releasing rate in glycemic response assay ($0.1651 \text{ mgmL}^{-1}\text{h}^{-1}$) was obtained for F1 and this value was lower than that obtained for wheat cracker ($0.2325 \text{ mgmL}^{-1}\text{h}^{-1}$). The mean ranks for appearance, colour, flavour, crispiness, creaminess, mouth feel and

overall acceptability in sensory evaluation obtained by the F1 were not significantly ($p \geq 0.05$) different from ranks obtained by the wheat cracker. In conclusion, a cracker formulation with 40 % substitution of mixed-grain flour was the optimum level of substitution and formulated cracker with 1:1:2 (F1) ratio of BR:FM:CP was the best.

Acknowledgement: Treasury Grant (TG15/101)

Optimization of liquefaction and saccharification times for laboratory scale production of glucose syrup from Sri Lankan cassava starch

M.D.W. Samaranayake*, A.B.G.C.J. De Silva, H. M. T. Herath, K.V.T. Gunawardhana,
W. R. D. Fernando

Food Technology Section, Industrial Technological Institute, Sri Lanka

Glucose syrup is a viscous liquid product, widely used in food and pharmaceutical manufacturing industries. Increasing demand for glucose syrups created the necessity of substitute the import and therefore it is appropriate to establish an effective method for manufacturing glucose syrup using locally available starch based raw materials. Thus, the objective of present study was to optimize the process (*i.e.* enzymatic liquefaction time and saccharification time) to obtain high yield of glucose syrups with suitable Dextrose Equivalent (DE) values in relatively short enzymatic reaction time using commercially grown cassava variety MU-51.

Cassava starch was extracted by gravity separation technique. Extracted starch was subjected to liquefaction under known, constant alpha-amylase concentration and DE value was measured at constant time intervals until the DE reached to the expected value of 8-15. Liquefied slurry obtained by optimized liquefaction time was subjected to saccharification under known, constant glucoamylase concentration at constant time intervals. The DE value of final product was calculated in order to optimize saccharification step to obtain glucose syrups with required intermediate or high DE values. Reducing sugar contents and DE values of liquefied slurry and glucose syrups were measured in triplicate.

According to the results, the optimum liquefaction time was 15 min and minimum saccharification times to obtain glucose syrups with intermediate (38-58) and high (58-73) DE values were 15 min and 75 min, respectively under the given conditions. In each case the total enzymatic reaction time spent for laboratory scale production of glucose syrup was less than 2 h.

Acknowledgement: National Research Council grant 13-095

Study on amylose content, rheological properties and particle size distribution of Sri Lankan banana (*Musa spp*) flour

A. B. G. C. J. De Silva^{1*}, S. Yathursan¹, S.A.S. Jayawardana¹, D.M.K. Aponso¹, J. K. R. R. Samarasekera², J. Gooneratne¹

¹Food Technology Section, Industrial Technology Institute, Sri Lanka

²Herbal Technology Section, Industrial Technology Institute, Sri Lanka

Green banana is a rich source of starch. Banana flour is a powder traditionally made of green bananas that is often used as a gluten free replacement to wheat flours. According to our previous studies matured green banana contains up to 70% of starch on dry basis. This study was focused on determining a correlation of maximum viscosity, which was taken from the brabender amylograph with amylose content present in banana flour. Amylose content, Rheological Properties (RP) and Particle Size Distribution (PSD) of Sri Lankan banana varieties, namely, *Seeni*, *Kolikuttu*, *Nethrapalam* and *Anamalu* were investigated. Banana varieties were peeled and cut into small pieces, soaked in the sodium metabisulphite solution (1%) for 10 minutes and dehydrated under controlled conditions in a tray dryer at 35 °C until the moisture content was less than 10%. The samples were ground using a centrifugal mill and sieved with size 0.5 mm mesh. Amylographs of banana varieties were compared with the amylograph of rice flour. Starch was extracted and amylose content was determined. Rheological properties were analyzed using a Brabender Amylograph. PSD was determined using sieve analysis. Particle size of banana complied with wheat flour standard of Codex Alimentarius. The variety, Nethrapalam containing the highest amylose content (37.09 ± 0.46) had the highest maximum viscosity and highest pasting temperature. There was a positive linear correlation of maximum viscosity of each variety with the variation of amylose content which inherently occurs in banana flour. There was no significant difference ($P > 0.05$) in amylose content (which ranged from 30.53 ± 0.20 to 37.09 ± 0.46) of four banana varieties. There was a significant difference ($P < 0.05$) in maximum viscosity of banana flour when compare with rice flour. There was no significant difference ($P > 0.05$) in pasting temperatures of four banana varieties (which ranged from 78.0 ± 0.0 to 81.0 ± 1.1) and rice flour (80.0 ± 0.7). These results further reveals that the presence of amylose tends to reduce the crystallinity of the amylopectin and influence the ease of water penetration into the granules and produce a higher viscosity with gelatinization. However, a noticeable change in the amylograph was observed near to maximum

viscosity with a small fluctuation of the maximum viscosity. Banana flours with low maximum viscosities are suitable for bakery products, such as bread and biscuits, while flours with high maximum viscosities can be used in preparation of desserts as gluten free flour.

Acknowledgement: Indian-Sri Lanka Inter-governmental science and technology programme

Effect of incorporation of rice bran on cooking, textural and sensory characteristics and nutritional composition of rice noodle

N.M. Rathnayaka¹, K.V.T. Gunawardhana^{2*}, I. Hewajulige², S. Ediriweera¹

¹*Faculty of Science and Technology, Department of Science and Technology,
Uva Wellassa University, Sri Lanka*

¹*Food Technology Section, Industrial Technology Institute, Sri Lanka*

Rice bran, a by-product of rice milling industry is rich in vitamins, minerals, amino acids, dietary fiber, essential fatty acids and plant sterols which provide wide range of health benefits. The main objective of this study was to determine the most acceptable level of rice bran incorporation to rice noodle securing its physico-chemical characteristics. This study examined the cooking, textural and sensory characteristics of 100% rice noodles and rice bran incorporated rice noodles which were produced by gelatinization of dough followed by cold extrusion. Rice bran incorporated rice noodles were prepared using rice flour with stabilized rice bran (variety AT 362) at different levels (10%, 15%, 20%, and 25%). Cooked rice noodle samples were evaluated for cooking loss, swelling ratio, tensile strength, extensibility, elastic recovery and firmness (n=3 each). Sensory properties of all treatments was evaluated by in-house trained panel (n=15) at ITI using nine point hedonic scale method. Further, proximate composition was estimated for the rice bran incorporated rice noodle selected from the sensory evaluation study (n=3 each).

There was a significance difference ($P < 0.05$) among treatments for the cooking loss of the noodle samples. The highest cooking loss was observed in the 10% rice bran incorporated rice noodle while the lowest in 25% rice bran incorporation. Firmness shows gradual decrease with increasing levels of bran incorporation, except in 25% bran incorporation. Elastic recovery of noodles ranged from $2.52 \pm 1.0\%$ to $9.73 \pm 6.56\%$. All treatments showed significant ($P < 0.05$) difference from each other for all the sensory attributes tested. The most acceptable bran incorporation level to the rice noodles was 15%, with respect to highest sum of ranks for the overall acceptability. Proximate composition of the 15% rice bran incorporated rice noodle was recorded as moisture content $12.07 \pm 0.33\%$, ash $1.27 \pm 0.01\%$, crude fat $1.51 \pm 0.4\%$, crude fiber $0.87 \pm 0.1\%$ and crude protein $6.85 \pm 0.2\%$. It is concluded that rice bran can be incorporated to rice noodle up to 15% while securing acceptable cooking, textural and sensory characteristics with enhance nutritional value.

Efficacy of microfiltration and thermal process technology for development of super health drinks

S.G. Walliwala¹, A.B.G.C.J. De Silva¹, P.N.R.J. Amunugoda^{1*}, G.D.D.R. Jayasinghe¹, W.K.S.M. Abeysekara¹, T.M.D.A. Jayawardane¹, I.M. Fernando²

¹*Food Technology, Industrial Technology Institute*

²*Faculty of Applied Science, University of Sri Jayewardenepura, Sri Lanka*

Microfiltration (MF) is an emerging non-thermal technique for clarifying and stabilizing of raw juices. There are limitations of microfiltration (non-thermal techniques) and thermal processing in achieving super health juice properties. The objective of this study was to test the efficacy of microfiltration and thermal processing technology for the preparation of Beet root (*Beta vulgaris*), Star fruit (*Averrhoa carambola* Linn) and Curry leaf (*Murraya koenigii*) juices for microbial sterilization, clarification and antioxidant capacity to improve juice property in order to develop super health drinks. Selected commodities were thoroughly cleaned with chlorinated water and potable water and blended. The resulting juice was strained, heated and sugar was added. Citric acid was added to maintain the pH (below 3.5). Sodium metabisulphite was used as a preservative agent. Total soluble solid, pH, colour and microbial content of these samples were evaluated. A sample from curry leaf juice was heated at 80° C and the resulting juice was filled into sterilized bottles, capped, kept in a water bath at 80° C for 20 minutes and cooled. Total soluble solid, pH, colour, microbial parameters and antioxidant activity of the sample were determined. Microfiltration was carried out in a laboratory scale membrane unit using polyvinylidene fluoride membrane (0.3µm) for selected juices under the pressure of 0.5 MPa. The permeate was collected into sterilized stomacher bags for microbial analysis and bottles to determine other physicochemical parameters. Microbial parameters and antioxidant activity of feed, retentate and permeate of curry leaf juices were determined. Microbiological analysis of the permeated juice showed that microfiltration was effective for the commercial sterilization compared to pasteurized (thermally processed) samples. Antioxidant activity of thermally processed curry leaf juice was significantly superior ($p < 0.05$) to that of micro filtrated samples indicating the antioxidant capacity increased with thermal processing. Results showed that reduce antioxidant activity of the permeated juice compared to feed and retentate samples. Clarification showed encouraging results of removing turbidity, viscosity and colour thus the possibility to develop single strength and blended super health drinks. Results

suggest that an integrated approach of both microfiltration and thermal processing techniques should be developed to improve the super health juice properties.

Acknowledgement: Treasury Grant TG 13/67

Improving quality of mango (*Mangifera indica*) var. Karthakolomban by postharvest application of new edible wax formulations

R.S. Wilson Wijeratnam, M.M.N.P Gunasekera , R.M.S. Gunathilaka, M.G.D.S. Perera, I.G.N. Hewajulige*

Food Technology Section, Industrial Technology Institute, Sri Lanka

Mango fruits are exposed to several exogenous factors that induce product deterioration from harvest to consumption. Phospholipase-D is a key enzyme involved in membrane deterioration that occurs during fruit ripening and senescence. Hexanal, an inhibitor of phospholipase-D has been successfully used as a pre- and postharvest treatment of fruits, vegetables and flowers.

A series of edible bio wax formulations incorporated with hexanal and cinnamon bark oil were developed and tested for application as a postharvest treatment to minimize incidence of disease, improve quality and enhance the storage life of mango var. Karthakolomban grown in Sri Lanka.

Quality evaluation of mango treated with wax formulations was carried out further to storage at low temperature (13.5 ± 1 °C and 80% RH) over 14 days. In order to select the best wax formulation capable of extending the storage life of mango, a series of 16 trials were conducted using 6 different wax formulations over three consecutive mango seasons. Storage studies revealed that two of the ITI wax formulations C5 (hexanal based) and C6 (cinnamon bark oil based) were best for reducing weight loss (27 ± 0.7 g) and preserving the quality of mangoes compared to the other wax formulations tested and the un-waxed controls (mean weight loss 33 ± 0.9 g). Fruits treated with C5 and C6 formulations showed significantly lower disease severity scores compared to controls. Both C5 and C6 treated fruits showed a high marketability (90 and 96%, respectively) compared to 83% marketability observed in the control fruits after two weeks storage at low temperature.

Fruits treated with wax formulations C5 and C6 showed greater firmness, 3.25 ± 0.17 Kpa; 3.65 ± 0.27 Kpa, low total soluble solids (TTS), $16.25 \pm 0.52\%$ and 16.5 ± 0.65 % and high pH values 3.79 ± 0.04 ; 3.66 ± 0.03 , respectively over 14 day storage at 13.5 °C, compared to the controls where firmness was 2.65 ± 0.72 Kpa and TTS was 17.25 ± 0.48 %, respectively. It is concluded that C5 and C6 wax formulations may have the

potential to enhance low temperature storage life and thus quality of Karthakolomban mangoes.

Acknowledgement: IDRC Canada / DFATD Canada Research grant

Physicochemical and functional properties of Finger millet (*Eleusine coracana*) flour and its potential use in food product development

S. A. S. Jayawardana^{1*}, A. B. G. C. J. De Silva¹, J. K. R. R. Samarasekera²,
J. Gooneratne¹

Food Technology Section, Industrial Technology Institute, Sri Lanka

²Herbal Technology Section, Industrial Technology Institute, Sri Lanka

Physicochemical and functional properties of finger millet (*Elusine coracana*) flour were investigated with the objective of understanding its applications in formulating flour based food products. Sri Lankan finger millet varieties, recommended by the Department of Agriculture, namely Ravi, Rawana and Oshada were used in this study. Finger millet seeds were washed, dried, milled using a Fritsch mill (sieve size 0.5 mm) and used to determine the particle size distribution (PSD), amylose content, swelling power (SP), solubility index (SI) and rheological properties (RP). Starch was isolated from finger millet flour and amylose content was determined by the colourimetric measurement of the iodine binding capacity of amylose. A starch-water (1:50) suspension was heated at 95 °C for 30 minutes, cooled and centrifuged to separate the jell and the supernatant. SP was expressed using the weight of centrifuged swollen granules. SI was determined from the amount of dried solids recovered by evaporating the supernatant. RP were analyzed using a Brabender Amylograph. When considering the results of PSD, according to East African Standard for Millet Flour and Codex Standard for Pearl Millet flour, flour of all three finger millet varieties can be considered as fine flour. Amylose contents of three finger millet varieties ranged from 11.99 ±1.57 to 13.85 ±1.04 with no significant differences ($p>0.05$) among the varieties. Significant differences ($p<0.05$) were observed in SP and SI which ranged from 14.05 ± 0.12 to 17.73 ± 0.29 and 22.02 ± 0.30 to 24.83 ± 0.68 respectively. SP of all three varieties positively correlated with SI while negatively correlating with amylose content. These results indicated that as the amylose content increases, the swelling tends to be restricted. With the increase in SP, starch solubility similarly increased as a result of granule swelling permitting the exudation of amylose. There was a significant difference ($P<0.05$) in maximum viscosities which ranged from 505.0 ± 0.7 to 602.5 ±1.1 Brabender Units. The variety containing the highest starch percentage (65.16 ± 0.59), Oshada had the highest maximum viscosity. Highest maximum viscosity indicated the potential of using flour of Oshada in

preparation of desserts. Low maximum viscosities of Ravi and Rawana flour indicated the suitability for using in bakery products and as thickening agents.

Acknowledgement: Indian - Sri Lankan Inter-governmental Science and Technology Cooperation Programme

Evaluation of six wild fruit extracts as sources of antioxidant in nutraceutical and cosmetic products

L.M.P.R. Elamaldeniya¹, P. Ranasinghe*², P.K.Dissanayake¹, W.P.K.M Abeysekera²,
U.K.D.S.S Gunasekara²

¹Department of Export Agriculture, Faculty of Agricultural Sciences, University of Sabaragamuwa, Sri Lanka.

²Herbal Technology Section, Industrial Technology Institute, Sri Lanka

Freeze dried 95% ethanolic extracts of six wild fruits namely keena (*Calophyllum calaba* L; Guttiferae), madan (*Syzygium cumini* L; Myrtaceae), weera (*Drypetes sepiaria*; Euphorbiaceae), dan (*Syzygium caryophyllum* L; Myrtaceae), attikka (*Ficus racemosa* L; Moraceae) and kahata (*Careya arborea*; Lecythidaceae) were used in this study. Antioxidant Properties (AP) were evaluated using Total Polyphenolic Content (TPC), Total Flavonoid Content (TFC), Ferric Reducing Antioxidant Power (FRAP) 1, 1-diphenyl-2-picryl-hydrazyl (DPPH) screening, 2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) and Oxygen Radical Absorbance Capacity (ORAC) assays via 96-well micro plates *in vitro*

Results clearly revealed significant differences ($P < 0.05$) among six wild fruit extracts for the investigated AP ($P < 0.05$). Mean TPC, TFC, FRAP and ORAC, DPPH and ABTS radical scavenging activity of six wild fruits were in the range of 22.09 ± 1.88 – 590.95 ± 22.17 mg gallic acid equivalents (GAE)/g of extract, 2.87 ± 0.11 – 13.56 ± 0.26 mg quercetin equivalents (QE)/g of extract, 12.35 ± 0.10 – 455.91 ± 3.94 mg trolox equivalents (TE)/g of extract, 07.03 ± 0.88 – 25.54 ± 3.36 mg (TE)/g of extract, 30.49 ± 4.87 – 585.40 ± 6.43 mg (TE)/g of extract and 43.08 ± 02.89 – 435.30 ± 6.62 mg (TE)/g of extract respectively. Kahata fruit had the highest TPC (590.95 ± 22.17 mg (GAE)/g of extract), FRAP (455.91 ± 3.94 mg (TE)/g of extract) and DPPH (585.40 ± 6.43 mg (TE)/g of extract) antioxidant activity while dan fruit had the highest TFC (13.56 ± 0.26 mg (QE)/g of extract) and ABTS (435.30 ± 6.62 mg (TE)/g of extract) antioxidant activity. Keena, attikka and kahata fruits showed the highest ORAC (23.41 ± 2.68 , 22.20 ± 2.15 and 25.54 ± 3.36 mg (TE)/g of extract respectively) activity and activity among three fruits are statistically insignificant ($P < 0.05$).

In conclusion, keena, madan, weera, dan, attikka and kahata wild fruit extracts possess marked antioxidant properties and in general kahata fruit extract had the highest and

keena fruit extract had the lowest antioxidant activity. Further, Kahata, dan, weera and Madan extracts has higher potential to be used as natural antioxidants in herbal nutraceuticals and cosmetic products.

Ethnopharmacological survey on medicinal plants used in snake bite treatments in Western and Sabaragamuwa provinces in Sri Lanka

R.M. Dharmadasa^{1*}, G. C. Akalanka²

¹*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

²*Department of Natural Resources, Faculty of Applied Sciences, Sabaragamuwa University of Sri Lanka, Belihuloya, Sri Lanka*

Sri Lanka has great snake diversity and more than 95% of victims are relied on traditional snake bite treatments. However, the traditional treatments for snake bites are hindered by several reasons. Thus, in the present study we investigated types of medicinal plant materials required, parts of the plants used for different snake types, treatment types, frequency index, heavily used and rare materials, family wise distribution of plants and challenges faced by traditional practitioner and future prospects. Information was gathered from 74 traditional practitioners by distributing a well structured questionnaire and collected data were tabulated and analyzed. A total of 341 different plant species belonging to 99 families dominated by family Fabaceae (32 species), followed by Malvaceae (16 species), Asteraceae (15 species), Rutaceae (13 species) and Apocyanaceae (14 species) were recorded. Different parts of the plant such as leaf (53.67%), bark (26.10%), entire plant (14.08%), roots (10.26%), bulbs (8.80%), seeds (7.62%), fruits (6.45%), buds (5.87%), flowers (3.23 %) stems (2.93%) and latex (2.05%) were used for the preparation of nine different types of formulae. These formulae includes oral administration (172 plant species), external bandaging (167 plant species), oiling for external application (34 plant species), steaming (33 plant species), creaming for wounds (6 plant species), nasal treatments (40 plant species), head treatments (23 plant species), treatment for eyes (4 plant species) and washing of wounds (9 plant species). Further, documented plants together with traditional knowledge could be effectively utilized for isolation and characterization of antivenom for different snake species from the documented plants.

Chemical composition of essential oils of different parts of *Myristica fragrans* (nutmeg) and *Syzygium aromaticum* (clove) grown in Sri Lanka

S.Kathirgamanathar^{1*}, K.P.D.M.K.Pathirana²

¹*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

²*Department of Food Science, Faculty of Agriculture, University of Peradeniya, Sri Lanka*

Myristica fragrans (nutmeg) and *Syzygium aromaticum* (clove) are economically important spice crops grown in Sri Lanka. The essential oils of these plants contain complex mixtures of terpenoids and are widely used as flavour ingredients, cosmetics and in pharmaceutical industry. Since 1991, there is no reported detailed compositional study of essential oils of these plants. Therefore, the present study was carried out to develop an updated database for chemical composition of essential oils of different parts of these plants.

The seeds and mace of nutmeg and the flower buds of clove were purchased from spice shops in Kandy, Kegalle and Matale. The leaves and stems of nutmeg and clove were collected as fresh samples from the same area. The plant parts were air-dried and hydro-distilled using Clevenger apparatus and the yields of essential oils were calculated. These oils were analyzed using Gas Chromatography-Mass spectrometry (GC-MS).

The yields of essential oils from nutmeg seed kernel, mace, leaf and stem were 8.0, 12.8, 1.6 and 0.2% respectively. Clove flower bud, leaf and stem yielded 18.5, 5.9 and 4.7% of essential oils respectively. The eugenol is the major component in clove flower buds (75.38%), leaf (84.39%) and stem (84.83%) oils. Sabinene was the major component of nutmeg seed kernel (33.92%) and mace (28.85%). α -Pinene was the major constituent of leaf (27.05%) and stem oils (24.26%).

The GC-MS results revealed that 22, 21 and 24 compounds were identified from clove bud, stem and leaf essential oils and the nutmeg mace, seed kernel, leaf and stem essential oils contain 46, 34, 57 and 45 compounds respectively. In a previous study it was reported that sabinene is the major constituent of nutmeg seed kernel (28.60%) and mace (24.50%). Eugenol was the major constituents of clove bud (79.2%), stem (84.9%) and leaf (86.15%).

According to the results obtained, the chemical composition of the essential oils obtained

from different parts of clove and nutmeg were highly variable with the previously reported values.

Anti-inflammatory volatile constituents from rhizomes and leaves of *Alpinia calcarata*

M. Chandhrakanthan¹, S. Kathirgamanathar^{1*}, S.M. Handunnetti², G.A.S. Premakumara¹

¹*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

²*Institute of Biochemistry, Molecular Biology and Biotechnology, University of*

Colombo, Sri Lanka

Phagocytic cells, like macrophages, are known to be activated under oxidative conditions and the stimulated production of reactive oxygen species (ROS) by them results in the oxidative burst which causes tissue injury in chronic inflammatory conditions like Rheumatoid arthritis (RA). Rhizomes of *Alpinia calcarata* are popularly used in herbal medicine for its reputed bioactive properties. It has been reported that some herbal medicines contain rhizomes of *A. calcarata* as a major ingredient in treating arthritic conditions. The present study is aimed to investigate the anti-inflammatory activity of *A. calcarata* rhizome and leaf extracts by a chemiluminescence based kinetic assay and to identify the volatile constituents present in the fractionated extracts. Whole plants of *A. calcarata* were collected from Western province of Sri Lanka and extracted using solvent systems and fractionated to obtain volatile fractions. Gas chromatography-Mass Spectrometric (GC-MS) analysis was done to identify the components. Murine macrophage RAW 264.7 cells were activated by serum opsonized zymosan-A and changes in ROS production was determined by luminol based chemiluminescence. The leaves and rhizomes were sequentially extracted into petroleum ether, dichloromethane (DCM), ethyl acetate, methanol and water. The cytotoxicity of all the extracts (62.5, 31.25, 15.62, 7.81, 3.90, 1.95 $\mu\text{g/mL}$) was determined by trypan blue exclusion assay. Results obtained showed that all the extracts at 62.5 $\mu\text{g/mL}$ decreased ROS production, in which the hot petroleum ether extracts of rhizome and hot dichloromethane (DCM) extracts of leaf significantly inhibited ROS production ($88.65 \pm 2.24\%$ and $92.35 \pm 0.89\%$). Aspirin (62.5 $\mu\text{g/mL}$) was used as a standard drug and it showed inhibition of $80.65 \pm 1.12\%$. A dose response relationship was observed in the inhibition of all volatiles from extracts and the IC_{50} was found to be 11.57 ± 0.74 and 10.89 ± 0.28 $\mu\text{g/mL}$, for rhizome petroleum ether and leaf DCM extracts respectively. None of the extracts affected macrophages viability (90-95%) upon 1 h incubation at 250 $\mu\text{g/mL}$. GC-MS analysis of volatile fractions of *A. calcarata* rhizome and leaf showed reported anti-

inflammatory constituents like 1,8-cineole, α -pinene, β -pinene, camphor, carotol, and α -Phellandrene. Hexane and DCM fractions possess volatile and non-volatile constituents. In conclusion, volatile fractions of *A. calcarata* rhizome and leaf showed a significant inhibition of ROS production *in vitro* and could have a potential therapeutic effect on arthritis disease processes by inhibiting production of superoxide anions thus by preventing oxidative burst of macrophages.

Chemical characterization of Ceylon Cinnamon (*Cinnamomum zeylanicum* Blume) bark oils distilled from Galle and Ratnapura districts

H.D.Weeratunge*, S.K. Ganegamage and G.A.S.Premakumara

¹*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

Ceylon Cinnamon (*Cinnamomum zeylanicum* Blume) bark oil is especially known as the best quality fragrance essential oil with unique aroma, which is internationally important and valuable for perfumery, cosmetic and flavor industries. Commercially, cinnamon bark oil production is mainly done with heavy bark (Pathuru) and chips (Katta) collected from Galle and Ratnapura districts in Sri Lanka. The international market value of Ceylon cinnamon bark oil mainly depends on the *trans*-cinnamaldehyde content of the bark oil. The aim of this study was to investigate the variations in the chemical composition of Ceylon cinnamon bark oil extracted from heavy bark and chips collected from Galle and Ratnapura districts to identify superior quality cinnamon to produce best quality Ceylon cinnamon bark oil.

Heavy bark and chips of cinnamon bark were collected from Galle and Ratnapura districts (n =3, per district) and subjected to extraction of oils by hydro-distillation using Clevenger arm. Each sample of bark oil was analyzed using Gas Chromatography Mass Spectrometry (GC-MS) for α -pinene, β -thujene, α -phelandrene, o-cymene, linalool, β -caryophyllene, *trans*-cinnamaldehyde, cinnamyl acetate, eugenol and benzyl benzoate to study variation in the chemical composition with respect to geographical location and different form of cinnamon.

The highest percentage (66.90%) of *trans*-cinnamaldehyde was present in heavy bark collected from Ratnapura district and is significantly ($p < 0.05$) higher compared to the oil of heavy bark and chips from Galle district. The *trans*-cinnamaldehyde content in the heavy bark and chips in Ratnapura district were not significantly different ($p > 0.05$). Highest level of linalool (5.40%) and cinnamyl acetate (8.63 %) were observed in chips of Ratnapura district. Significantly ($p < 0.05$) higher percentage of eugenol (Galle-10.17% and Ratnapura - 11.04%) was observed in heavy bark compared to chips regardless of the district. The α -pinene were in the range of 0.08 - 0.39% and among districts and type of bark, they were significantly ($p < 0.05$) different. Levels of o-cymene and β -caryophellene did not show a significant ($p > 0.05$) difference between

districts and the types of bark. In future, these chemical composition data will be useful in producing high standard cinnamon bark oil by manipulating the ratios of chips and heavy bark during distillation.

Comparative identification of wild and domesticated bees honey

C. Wijayasiriwardena*, W.A.D.D Wasalamuni

Herbal Technology Section, Industrial Technology Institute, Sri Lanka

Unavailability of genuine wild bees honey leads to substitution with domesticated bees honey, and market samples are greatly adulterated with artificial invert sugars. Purity of wild and domesticated bees honey can be assessed according to Sri Lankan standard SLS 464; 1979. However, to assess its identity microscopical characters with some chemical tests will give the reliable information. Therefore, the present study was undertaken to develop a reliable test method to identify genuine bees honey.

Three bees honey samples from the wild and three domesticated samples were collected from different geographical regions of Sri Lanka in the month of September 2014. Sensory characters such as taste, odour, consistency, colour etc. were evaluated as per WHO guidelines. Microscopically identifiable components were observed under the Leica compound microscope and important characters were photographed. Parameters stated in SLS 464: 1979 were also investigated.

Results showed that the colour is lighter in domesticated samples, while taste has no difference, aroma is more prominent in wild honey, furfural test was negative for all the samples. Domesticated samples were found to have lesser number of pollen diversity and concentration and more often found to have flower parts, microscopically. Parameters stated in SLS for wild and domesticated bees honey were tested including moisture, percent by mass 20.93 ± 0.0013 and 17.93 ± 0.0071 , Total reducing sugars per cent by mass 57.89 ± 0.010 & 63.48 ± 0.9483 , Sucrose per cent by mass 2.32 ± 1.14 & 1.32 ± 0.0046 , Ash per cent by mass 0.53 ± 0.0007 & 0.58 ± 0.0014 , Acidity expressed as formic acid 0.03 ± 0.0003 & 0.02 ± 0.0007 and Fructose, glucose ratio 0.41 ± 0.928 & 0.69 ± 0.0600 for wild and domesticated bees honey samples respectively.

From the above results it can be concluded that the domesticated bee honey can be identified from wild bees honey with respect to pollen count and its diversity.

Acknowledgement: Treasury Grant TG 15/104

Determination and Isolation of marine derived Secondary Metabolites in Sri Lankan Marine Algae through Electron Spin Resonance, ABTS⁺ on-line HPLC and High performance Centrifugal Partition Chromatography (HPCPC)

K. W. Samarakoon^{1,2,*} H.H.C. Lakmal², You-Jin Jeon²

¹Herbal Technology Division, Industrial Technology Institute, Sri Lanka

²Department of Marine Life Science, Jeju National University, Jeju 690-756, Republic of Korea

Marine algae as an ‘untapped’ resource of natural products that can be used for the lead compounds in drug discovery. However, Sri Lankan seaweeds have not been studied extensively in this direction. Therefore, our objective of this study was to evaluate antioxidant activities of the selected marine algae species, *Ahnfeltiopsispygmaea*, *Chnoospora minima*, *Gracilariacorticata*, *Padinacommersonii*, *Sargassum sp. A* and *Sargassum sp. B* and further to isolate compounds, which responsible for antioxidant activities. Freshly collected algae samples from Hikkaduwa sampling site were lyophilized and homogenized. Then, each sample was extracted three times in 80% methanol by sonicating at 25°C for 90 minute period. The crude extracts were subjected to partition chromatography using organic solvents. Antioxidant activities were evaluated in terms of free radical scavenging activity against DPPH, hydroxyl and alkyl radicals by Electron Spin Resonance (ESR) spectrometer. Total Phenolic Content (TPC) was determined by Folin–ciocalteu method. Ethyl acetate (CME) and aqueous (CMW) extracts of *C. minima* showed the highest and significant TPC of 14.14% and 10.39%, respectively. However, the strongest radical scavenging activities were observed in *P. commersonii* ethyl acetate (PCE), aqueous (PCW) and *C. minima* aqueous (CMW) fractions against alkyl radical; IC₅₀ values 0.017, 0.02 and 0.05 mg mL⁻¹, respectively. *Sargassum sp. A* ethyl acetate (SAE), *C. minima* ethyl acetate (CME) and aqueous (CMW) fractions showed higher hydroxyl radical scavenging activity (IC₅₀ value 0.07, 0.11 and 0.12 mg mL⁻¹) compared to the fractions of other species fractions. The highest DPPH radical scavenging activity was identified from the CMW fraction (IC₅₀ value of 0.10 mg mL⁻¹). ABTS⁺ on-line High Performance Liquid Chromatography (HPLC) results showed that potent antioxidant activity on individual compounds from PCE and CME fractions based on the negative signals at 734 nm in ABTS⁺ chromatogram.

Subsequently, three compounds from PCE and two compounds from CME fractions were isolated by high performance centrifugal partition chromatography (HPCPC). The results confirmed that *P. commersonii* and *C. minima* contain natural products with promising antioxidant properties.

***In vitro* Tyrosinase Inhibitory Activity of Some Selected Sri Lankan Medicinal Plants**

G. D. Liyanaarachchi¹, R.Samarasekara¹, K.R.R. Mahanama², K.D.P. Hemalal³

¹*Herbal Technology Section Industrial Technology Institute, Sri Lanka*

²*Department of Chemistry, University of Colombo, Sri Lanka*

³*Rock Mountain Soap, Co.201-106, Bow Meadows Crescent, Canmore*

Tyrosinase (monophenol, dihydroxyphenylalanine: oxygen oxidoreductase) is a multifunctional, copper-containing enzyme that is involved in melanogenesis. Excessive melanin accumulation leads to human skin disorders, such as melasma, freckles, age spots and malignant melanomas. As plants are a rich source of bioactive chemicals that are mostly free from harmful side effects, interest in finding natural tyrosinase inhibitors is also a priority research area. Some potent tyrosinase inhibitors, such as anisaldehyde, quercetin and dalenin have been isolated from various plants. Therefore, the development of safe and effective tyrosinase inhibitors has become important for preventing pigmentation disorders, melanin-related human health problems and improving skin whitening properties.

Although research has been done in many other countries in the region on tyrosinase inhibitory activity of plant extracts, research published on that of *Vetiveria zizanioides*, *Rubia cordifolia*, *Nymphaea nouchali*, *Camellia sinensis*, *Elaeocarpus serratus*, *Nymphaea pubescens*, *Mesua ferrea*, *Saussurea lappa*, *Curucum romatica*, *Kokoona zeylanica*, *Coscinium feneatrarum* plants grown in Sri Lanka are limited. Hence the objective of this study was to determine tyrosinase inhibitory activity of ethanol extract of eleven medicinal plants including *Artocarpus heterophyllus* and *A. altilis* as changes in geographic condition of soil can lead to changes in plant metabolite composition affecting the biological activities. Cold ethanol extract of air-dried and powdered plants were evaluated *in-vitro* by an assay, based on the catalyzing ability of tyrosinase for the oxidation of L-DOPA compared to positive control, kojic acid.

Artocarpus heterophyllus bark extract exhibited the highest dose-dependent anti-tyrosinase activity having IC₅₀ value of 27.47±0.45 µg/ml. *A. altilis* was also found to be a good inhibitor of tyrosinase having IC₅₀ value of 125.17±6.75 µg/mL but showed less inhibition than the positive control kojic acid (IC₅₀ 76.61±0.80). Previous studies reported that *Artocarpus heterophyllus* and *A. altilis* contained several compounds such as

flavonols, flavanonandstilbenoids, reported to inhibit tyrosinase activity and 3-hydroxy-4-keto moiety of the flavonols has the ability to chelate Cu^{2+} in the enzyme. However, no activity was observed for extracts of *M. ferrea*, *C. fenestrarum*, *K. zeylanica*, *C. aromatica* and *S. lappa*. Although *E. serratus* bark extract exhibited 50% inhibition at 500 $\mu\text{g}/\text{mL}$ of extract *E. serratus* leaves and fruit extract showed moderate activity. *Eleocarpus serratus* reported to have flavonols, myricitrin, mearnssetin, 3-O- β -D-glucopyranoside, mearnsitrin, tamarixetin, 3-O- α -L-rhamnopyranoside are usually competitive inhibitors for the oxidation of L-DOPA by tyrosinase.

This study highlighted the tyrosinase inhibitory activity, which support the use of *A. heterophyllus*, *A. altilis* and *E. serratus* as an ingredient in cosmetic and pharmaceutical products.

Acknowledgement: Treasury grant TG 13/69

Insecticidal indigenous *Bacillus thuringiensis* strains with potential Lepidopteran activity

R.Y. Baragamaarachchi¹, O.V.D.S.J. Weerasena², R. Samarasekara^{1*}

¹*Herbal Technology Section, Industrial Technology Institute, Sri Lanka*

²*Institute of Biochemistry, Molecular Biology and Biotechnology, University of
Colombo, Sri Lanka*

Bacillus thuringiensis (Bt) is a naturally occurring, soil borne bacterium that has been used as a bio-insecticide since 1920's. *Bacillus thuringiensis* produces insecticidal crystal proteins that are toxic to different insect orders, especially to Lepidoptera, Coleoptera and Diptera. The objective of the present study was to isolate and identify insecticidal indigenous Bt strains with potential Lepidopteran activity.

Bacillus thuringiensis was isolated from soil samples collected from different climatic zones in Sri Lanka. Sodium acetate / heat treatment method was performed to isolate Bt from soil samples. Isolated Bt was grown on chromogenic *Bacillus* agar to differentiate Bt like colonies. Phenotypic characterization was carried out to confirm the isolates as Bt. Gyrase B gene of Bt was sequenced to identify the subspecies of the strains. Lepidopteran specific *Cry* genes; *Cry1*, *Cry2* and *Cry9* of Bt were evaluated by PCR analysis.

Bacillus thuringiensis was visualized as blue/green colonies on *Bacillus* agar and all Bt isolates gave positive results for gram staining. Endospores of Bt were stained as green elliptical structures. Crystals were visualized in dark blue colour when stained with Coomassie blue. Gyrase B gene sequencing revealed that Bt strains isolated from soil samples in Sri Lanka are *Bt kurstaki* (*Btk*, Bt AB8 and Bt AB15), *Bt graciosensis* (*Btg*, Bt AB6, Bt AB7, Bt AB10, Bt AB11, Bt AB12, Bt AB13, Bt AB14 and Bt AB16), *Bt poloniensis* (*Btp*, Bt AB17), *Bt Canadensis* (*Btc*, Bt AB19) and *Bt konkukian* (*Btkn*, Bt AB21). PCR analysis revealed the presence of Lepidopteran toxic *Cry1* gene in *Btg* (Bt AB6, Bt AB7, Bt AB10, Bt AB14, Bt AB16), *Btk* (Bt AB8, Bt AB15) and *Btp* (Bt AB17). *Cry2* gene in *Btg* (Bt AB16), *Btk* (Bt AB15) and *Btp* (Bt AB17). *Cry9* gene in *Btg* (Bt AB6, Bt AB7, Bt AB10, Bt AB11, Bt AB12, Bt AB13), *Btp* (Bt AB17), *Btc* (Bt AB19) and *Btkn* (Bt AB21) isolates. These findings suggest that these Bt strains possess Lepidopteran specific insecticidal *Cry* genes; *Cry1*, *Cry2* and *Cry9* and therefore have

potential Lepidopteran activity to be used as biological controlling agents against Lepidopteran insects found in agricultural crops as an alternative for synthetic pesticides. However, bio-efficacy of these Bt strains need to be evaluated at laboratory and field level against lepidopteran insects.

Acknowledgement: National Science Foundation Grant RG/2011/BT/05

***In vitro* anti-arachidonate 5-lipoxygenase activity of selected Sri Lankan medicinal plants**

H.D.S.M. Perera¹, R. Samarasekera¹, S. Handunnetti², O.V.D.S.J. Weerasena²

¹*Herbal Technology Section Industrial Technology Institute, Sri Lanka*

²*Institute of Biochemistry, Molecular Biology and Biotechnology, University of Colombo, Sri Lanka*

Arachidonate 5-Lipoxygenase (A5-LOX) is commonly found in human cardiovascular system and central nervous system. The catalytic products of A5-LOX are potent mediators of oxidative and inflammatory reactions. These compounds and mediators elicit diversified biological activities leading to cardiovascular and neurodegenerative diseases including carcinogenesis. Therefore A5-LOX enzyme inhibitors are better candidates in the preventive intervention of A5-LOX mediated diseases. Medicinal plants used in traditional medicine are potential sources for new A5-LOX inhibitors. The objective of the present study is to evaluate *in vitro* anti-A5-LOX potential of selected Sri Lankan medicinal plants, *Spondias dulcis* (Ambarella), *Cynometra cauliflora* (Naminam), *Symplocos cochinchinesis* (Sewala bombu), *Melaleuca leucadendra* (Lothsumbul), *Sphaeranthus indicus* (Mudumahana) and *Bacopa monieri* (Lunuwila).

Air-dried and powdered leaves of selected plants were extracted with ethanol using cold extraction technique. *In vitro* anti-A5-LOX activity of ethanol leaf extracts was determined by kinetic A5-LOX enzyme inhibitory assay using a spectrophotometric method in 96-well micro-plates. The conversion of linoleic acid to 13-hydroperoxylinoleic acid was monitored at 234 nm at 25 °C for 10 minutes to determine maximum linear velocity of the reaction. Percentage inhibition was calculated in comparison to control and IC₅₀ values were determined. Baicalein was used as the reference standard.

In the anti-A5-LOX assay, IC₅₀ values of extracts ranged from 48.71 ± 1.15 to 346.56 ± 6.84 µg/mL indicating significant, dose-dependent anti-A5-LOX activities as opposed to the reference standard baicalein (IC₅₀: 1.55 ± 0.24 µg/mL, p<0.05). The ethanol leaf extract of *M. leucadendra* showed the highest activity (IC₅₀= 48.71 ± 1.15 µg/mL), followed by *S. cochinchinesis* (IC₅₀=64.10 ± 0.22 µg/mL), *S. dulcis* (IC₅₀ =66.14 ± 4.39 µg/mL) and *C. cauliflora* (IC₅₀ =77.21 ± 3.14 µg/mL). The ethanol leaf extract of *S. indicus* (IC₅₀ = 137.07 ± 9.27 µg/mL) and *B. monieri* (IC₅₀ = 346.56 ± 6.84 µg/mL)

showed moderate activity.

The present study reveals *in vitro* anti-A5-LOX inhibitory activity of the leaves of the selected plants indicating the potential of these extracts as A5-LOX inhibitors.

Acknowledgement: NRC Grant No: 12-100

Development of a molecular assay to differentiate yellowfin tuna (*Thunnus albacares*) from other tuna species commonly found in Sri Lanka

D.R.C. Perera¹, P.A.D.H.N. Gunathilaka^{2*}, W.W.P. Rodrigo², A.M.M.H. Athapaththu²,
H.H.K. Achala²

¹*Aquatic Resources Technology Degree Program, Uva Wellassa University, Sri Lanka*

²*Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

Exports of chilled large tunas, such as yellow fin tuna (*Thunnus albacares*) and bigeye tuna (*Thunnus obesus*), have become an attractive venture in recent years. In Sri Lanka differentiation of tuna species prior to processing is achieved through morphological identification, which is not a reliable method. Therefore, the aim of this study was to develop a Polymerase Chain Reaction (PCR) based diagnostic method to differentiate yellowfin tuna from other tuna species commonly found in Sri Lanka, in order to help the fish processing industries and fish exporters for species confirmation. Muscle tissues of *T. albacares* (n= 08), *T. obesus* (n= 08) and *Katsuwonus pelamis* (Skipjack tuna, n=08) were analyzed to evaluate this molecular assay. Amplification of DNA from tuna samples were carried out using primers flanking a 558 bp region of the cytochrome b gene. The PCR amplified DNA products of tuna samples were digested with *Eco*NI restriction enzyme. Products having band sizes of 187 bp and 371 bp was observed from *T. albacares* after digesting with *Eco*NI. The restriction fragment pattern was used to differentiate *Thunnus albacares* from other adulterated tuna species in fish processing industry. Therefore, the current study carries a reliable approach to identify and distinguish *T. albacares* from the other tuna species which are commonly found in Sri Lanka.

Optimization of a DNA extraction method to isolate DNA from processed food samples for the detection of Genetically Modified (GM) Food

V.N. Talwatta¹, W.W.P. Rodrigo², H.H.K. Achala^{2*}, W.T.G.S.L. Withana²,
A.M.M.M. Athapaththu², and P.A.D.H.N. Gunathilaka²

¹*Animal Science Degree Programme, Uva Wellassa University, Badulla, Sri Lanka*

²*Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

Genetically modified (GM) food is an important application in food improvement. This has become a significant problem regarding authentication of GM food because non-labeled GM foods exist in the market. Polymerase Chain Reaction (PCR) technique is one of the important and reliable methods in detecting GMO originated food items. Deoxyribonucleic acid (DNA) extraction from different food matrices is a critical step in PCR analysis. The aim of the present study was to optimize an efficient modified Cetyl Triethyl Ammonium Bromide (CTAB) method to extract DNA from processed food samples. The method was assessed by the yield and purity of the extracted DNA and suitability for PCR amplification. Spectrophotometric results of the extracted DNA samples from processed food items indicate 150 mg to be the optimum sample weight to extract DNA. Sodium Dodecyl Sulfate (SDS) can be used as a protein removal agent instead of the Proteinase K enzyme. Since the use of SDS is more economical than the use of Proteinase K, the accepted procedure can be considered as successful and effective. During analysis of post PCR products using 1 % agarose gel, both samples (Processed cereal mixture and Soya meat) successfully amplified chloroplast DNA. Therefore, the DNA extraction method which was optimized in this study can be used in the detection of GM food using PCR.

Amplification and cloning of thermo-stable alpha-amylase gene from *Geobacillus stearothermophilus*

M.S. Thiwanka¹, W.W.P. Rodrigo^{1*}, H.H.K. Achala¹, A.M.M.H. Athapaththu¹,
P.A.D.H.N. Gunathilaka¹

¹*Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

Alpha Amylase is a hydrolase type of enzyme which aids in the breakdown of starch into maltose. Thermo-stable α -amylases have many commercial applications in starch processing, brewing and sugar production and in detergent manufacturing processes. *Geobacillus stearothermophilus* bacteria produces thermo-stable alpha amylase enzyme but it is not an efficient producer of this enzyme since the enzyme yield is low. Hence, using genetic modifications of α -amylase as a recombinant protein is a good solution to improve the yield of the enzyme to the level required in the industry. The objective of this research was to clone the thermo-stable alpha amylase gene from *G. stearothermophilus* into cloning vector of pGEM[®]-T Easy, in order to express the enzyme in the highly efficient protein expression system of *Pichia pastoris*. *G. stearothermophilus* was grown on Nutrient broth medium (NB) at 60 °C overnight and genomic DNA was extracted by low cost laboratory developed protocol. Primers were designed to contain *Xho*I restriction site in both primers and 6His tag coding sequence in reverse primer for the purification of the recombinant protein. Alpha amylase gene was amplified by PCR and expected sized band was observed (1670 bp) on an agarose gel. The purified PCR product was cloned into pGEM[®]-T Easy vector (pGEM[®]-TEasy-Amy) and correct recombinant construct was screened by rapid screening method, colony PCR and restriction digestion. Currently work is underway to confirm correct clone by sequencing and cloning of α -amylase gene into expression vector pPIC9.

Development of a DNA rabies vaccine for dogs using glycoprotein gene of Rabies Virus

K.G.W.W. Bandara^{1*}, G.P.W.K. Withanage², A.M.M.H. Athapaththu¹

W.T.G.S.L. Withana¹, N. V. Chandrasekharan², S.G. Senaratne¹

¹ *Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

² *Department of Chemistry, Faculty of Science, University of Colombo, Sri Lanka*

Rabies is a zoonotic disease that causes acute encephalitis. Rabies is caused by the rabies virus which is transmitted by rabid animals to humans. It has become a serious public health issue in many developing countries, with over 60 % of human deaths occurring in Asia. Sri Lanka is a rabies-endemic country. Vaccination is the only way to combat the disease for pre and post exposure. Currently the dog rabies vaccine is imported by the government. The inability of currently used imported vaccines to provide highly potent, cost-effective, safe and sustained protection has led to the production of a DNA rabies vaccine for dogs to eliminate rabies in dogs as the most cost effective way for preventing rabies in people.

A study conducted in Sri Lanka reported that the Sri Lankan rabies viruses are distinct and probably originated from a single clone. Because the glycoprotein protein induces a full spectrum of antiviral immune responses glycoprotein gene sequences of local rabies virus strains were obtained from NCBI and analyzed. Since neutralizing antibodies recognize only, one of the five distinct antigenic sites on the rabies virus glycoprotein, the amino acid changes at antigenic sites of the local rabies virus strains were carefully analyzed. A glycoprotein gene sequence of local rabies virus strain H-08-1320 (Gen bank Acc. No: AB569299) which has the minimum amino acid changes at antigenic sites was selected and used for the production of DNA rabies vaccine.

The pcDNA3.1 mammalian expression vector was selected to clone the rabies virus glycoprotein gene for the production of DNA vaccine that will express rabies virus glycoprotein in vaccinated dogs. The codon optimized gene sequence was cloned to pcDNA3.1 commercially. The clone designated RVG-Dog-opt-pcDNA3.1 was transformed into *Escherichia coli* DH5 α cells, transformants were screened and stored at -80 °C for future work. Synthesized glycoprotein gene was sequenced and the sequence analysis revealed the 100 % homology with codon optimized rabies virus glycoprotein gene sequence. Work is underway to carry out the clinical experiments using dogs to analyze the immune potential of the plasmid DNA vaccine in dogs.

Trends of Dengue Incidence in Kundasale Medical Officer of Health (MOH) area, Central province, Sri Lanka

N.W.B.A.L. Udayanga¹, P.A.D.H.N. Gunathilaka^{2*}, M.C.M. Iqbal³, W. Abeyewickreme¹

¹ *Molecular Medicine Unit, Faculty of Medicine, University of Kelaniya, Sri Lanka.*

² *Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

³ *National Institute of Fundamental Studies, Kandy, Sri Lanka*

Dengue is one of the prominent health issues in Sri Lanka as the average number of dengue cases recorded each year remains increasing over time due to recent outbreaks throughout the country. Thus, a statistical and geo informatics based analysis of the recent trends in dengue distribution was carried out to identify spatial and temporal trends in distribution patterns of dengue and to determine the susceptibility of a population to dengue infection in the Kundasale Medical Officer of Health (MOH) area. Monthly records of reported dengue cases from 2010 to 2014 of the Kundasale MOH area were collected and spatial maps of the recorded dengue case distribution in each Grama Niladhari Division (GND) on a monthly basis for the entire study period by using Arc GIS 10.2. Normal Chi square test coupled with Paired-Chi square test in MINITAB (version 14.12.0) were derived to investigate the impact of gender and age on the incidence. Ambakote, Neththarampotha, Kundasale North and South localities indicated relatively high susceptibility to dengue outbreaks throughout the study period, while most of the GNDs (n= 24) indicated less (null) susceptibilities. As suggested by the results of the Paired-Chi square test [$>X^2(79, 0.95) = 100.744$], the emergence of dengue outbreaks indicated a significantly declining trend in recorded dengue cases in all of the GNDs (except for Balagolla East, Kengalla and Malpana) during recent years. The Percentage Infected Male: Female Ratio (PIMFR) remained significantly altered throughout the study period ($p < 0.05$ at 95% of significance). Males were relatively more susceptible to dengue infection than females (with 55.2: 45.0 of average PIMFER). The vulnerability of age groups for dengue infection was analyzed among different age groups; Year 0 - 5 (16.43 %), 6 - 10 (14.00 %), 11 - 20 (29.75 %), 21 - 30 (14.57 %), 31 - 40 (7.90 %), 41 - 50 (11.98 %), 51 - 60 (5.39 %), and > 61 (0 %). According to the Paired-Chi square test, the vulnerability of age groups tend to shift significantly throughout the study period [$>X^2(7, 0.95) = 14.067$]. In conclusion, males tend to

indicate relatively high susceptibility to dengue. Age group of 11-20 was observed as the highest vulnerable group, while the group >50 age was identified as least vulnerable.

Optimization of Reverse Transcriptase Polymerase Chain Reaction based method for the detection of the Dengue Virus

S.A. Kumar¹, A.M.M.H. Athapaththu^{2*}, P.A.D.H.N. Gunathilaka², W.W.P. Rodrigo²,
H.H.K. Achala²

¹*Animal Science Degree Programme, Uwa Wellassa University, Sri Lanka*

²*Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

The major public health problem in the tropical countries including Sri Lanka is the Dengue (DEN). Diagnosis of the disease as early as possible would improve the patient management, vector controlling, and lower the fatality rate. Thus, early diagnosis of DEN has become a major requirement in clinical setup. Therefore, the aim of this study was to optimize a RT-PCR based method for the detection of the DENV and serotypes.

Different extraction methods namely; Trizol, Silica and commercial kit based methods were used to determine the suitable extraction method for the DENV from clinical samples (NS1 positive). The optimization of RT-PCR including synthesis of cDNA was performed. In addition, the optimization of nested PCR was carried out from cDNA to differentiate DENV serotypes.

A commercially available RNA extraction kit (CEYGEN) showed successful results for isolating RNA from viral samples. Complementary DNA (cDNA) was synthesized using DC-2C primer, incubating at 80°C for 4 min. The RT-PCR amplification conditions were optimized as denaturation at 94°C for 5 min, followed by 40 cycles at 94°C for 15 sec, 55°C for 15 sec, 72°C for 30 sec, and the final extension was performed at 72°C for 10 min using DC-1S and DC-2C primers. The nested PCR was successfully optimized with proper amplification conditions and different annealing temperatures 50°C, 42°C, 50°C and 50°C for different serotypes D1, D2, D3 and D4 respectively.

Altogether, three samples were confirmed as positive for DENV diagnostics and were identified as D1, D4 and D2 DEN serotypes with the band sizes at 490 bp, 398 bp and 230 bp respectively. Therefore, in this study, RT-PCR and nested PCR methods were optimized to diagnose DENV as well as serotypes of DENV. Analysis of large number of clinical samples is needed for the evaluation of the method.

Prevalence of Dengue Fever (DF) and Dengue Hemorrhagic Fever (DHF) in the Gampaha District of Sri Lanka: A cross sectional study in a selected population of clinically suspected dengue patients

M.M.E. Munasinghe¹, A.M.M.H. Athapaththu¹, W. Abeyewickreme², W.W.P. Rodrigo², L. K. Fernando³, P.A.D.H.N. Gunathilaka¹

¹ *Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

² *Molecular Medicine Unit, Faculty of Medicine, University of Kelaniya, Sri Lanka*

³ *Centre for Clinical Management of Dengue & Dengue Hemorrhagic Fever, Negombo, Sri Lanka*

Dengue is one of the most serious mosquito borne viral infections mainly affecting tropical and subtropical countries of the world. In Sri Lanka, dengue has currently become a major public health issue, with high rates of morbidity and mortality. In the absence of specific treatment and a vaccine for dengue fever, vector control is the only method by which spread of dengue can be prevented. As effective control and prevention programmes depend upon improved surveillance data, this study was carried out to report the characteristics of Dengue infection among a selected population of patients in the District of Gampaha. A cross-sectional study was conducted from November 2014 to March 2015 among clinically suspected dengue patients recorded at the Centre for Clinical Management of Dengue & Dengue Hemorrhagic Fever, General Hospital, Negombo. Information such as age and gender of the patient and number of days after the infection was recorded. Patients were screened using the NS1 antigen detection test kit, and their clinical features were also recorded during the treatment period. Collected data were analyzed retrospectively for demographic features and results of the NS1 antibody test. A total of 181 serum samples from dengue suspected patients were analyzed, of which 128 samples (70.7 %) screened positive for dengue virus infection on the NS1 antigen test. A majority of the infected individuals were males [54.69% (n= 70)] while 45.31% (n= 58) were females. The mean age of the study population was 26.5 years (SD = 6.4). Dengue Fever (DF) was the predominant disorder type (96.88 %, n= 124) among the study population of which 54.83 % (n= 68) was represented by males. Dengue Hemorrhagic Fever (DHF) represented only 3.1 % (n= 4) among the dengue infected population. The most affected age group was 0 to 15 years (35.94 %, n= 46) (Pediatric population), followed by 16 - 30 year group (35.16 %, n= 45), 21.85 % (n= 28) 31 - 45 year group, 6.25 % (n= 8) of 46 - 60 years and only 0.78 % (n= 1) of > 60

years. The present study emphasizes the need for continuous epidemiological surveillance for timely formulation and implementation of an effective dengue control programme.

Cloning of a Rabies Virus specific glycoprotein coding gene into a bacterial expression system for the expression of a recombinant protein

H.S. Sewwandi¹, W.W.P Rodrigo^{2*}, A.M.M.H. Athapaththu², P.A.D.H.N. Gunathilaka²,
K.G.W.W. Bandara², H.H.K. Achala²

¹*Animal Science Degree Program, Uva Wellassa University, Badulla, Sri Lanka*

²*Biotechnology Unit, Industrial Technology Institute, Sri Lanka*

Rabies is an infectious disease characterized by an acute and profound dysfunction of the central nervous system caused by Lyssa virus of family Rhabdoviridae. It recognized as one of the oldest diseases of human kind throughout the world. According to World Health Organization (WHO), about 50,000 human deaths report every year worldwide due to rabies infections. Tests for rabies antibodies are occasionally ordered to determine if people have been successfully immunized against the disease. Currently, three tests have been approved by the WHO for determining the levels of rabies neutralizing antibodies. However, these tests require lots of expertise and are generally carried out in reference laboratories at a high cost. Therefore, it vital to develop and standardize simple techniques such as Enzyme Linked Immunosorbent Assay (ELISA) for determining the level of antibodies against rabies virus. Development of such a diagnostic kit would make this testing more widely available at a lower cost. For that it is essential to purify rabies virus specific recombinant protein antigen. Hence, the aim of present study was to clone rabies virus specific glycoprotein gene (RVG) into bacterial expression vector for the production of rabies virus specific recombinant protein in order to explore the feasibility of developing a diagnostic kit for detecting anti-rabies antibodies. Initial attempts were made to isolate plasmid DNA of pET28a(+) and pcDNA 3.1-RVG vectors. The *Bam*HI and *Xho*I restriction sites were used to digest the plasmids. The purified Rabies Virus Glycoprotein gene was cloned into pET28a(+) bacterial expression vector. The pET28a(+)-RVG plasmids were successfully transformed into TOP10F' competent cells through electroporation. Transformants were screened and selected by rapid screening method.

Determination of ethanol content in colognes from Sri Lankan market using a validated Gas Chromatographic-flame ionization detection (GC-FID) method

M.R.P. Dassanayake*, J.K.A.B. Wijegunasekara, G.V.V. Liyannarachchi,
A.L.S Malvenna, S.L.O.E. Samarathunga, K.R.M. Gunarathne
Chemical and Microbiology Laboratory, Industrial Technology Institute, Sri Lanka

Colognes are the most frequently used cosmetic in Sri Lanka. The ethanol content of adult colognes and baby colognes are regulated by the national standards in the country and are stipulated to be within 50-90% (v/v) in adult colognes and 55-65% (v/v) for baby colognes respectively. However, method given in the local standards for the determination of ethanol is based on gravimetry, and therefore is less sensitive and time consuming.

A gas chromatographic method with flame ionization detection was developed using n-propanol as the internal standard. The method was then validated using standard procedures. The method had good precision, accuracy, and reproducibility over a wide linear range. The precision of the test method was determined at three ethanol levels of 99, 55 and 10% (v/v) and the relative standard deviations of repeatability and reproducibility were in the range of 0.004 % to 0.028 % respectively. Method also showed good recovery at three concentration levels within the range of 99.8-103.5% (v/v) and was reliable from 0.01% (v/v) to 99% (v/v) with a linearity of $r=0.999$. The limit of detection and limit of quantification of the method were 0.01 and 0.03% (v/v) respectively, indicating the applicability of the developed method for analysis of nonalcoholic perfumes as well. The method also had good specificity with recovery at three concentration levels ranging from 98.4 to 99.6% (v/v) over interfering alcohols such as methanol and iso-propanol. This is the first market survey on ethanol content of colognes in the country, and colognes samples ($n=262$) including baby colognes ($n=162$) and adult colognes ($n=100$) from ten different brands, representing over eighty percent of market share for cologne in the local market, were analyzed using the GC-FID method. All the baby cologne samples tested except one were within the local regulated levels while out of 100 adult cologne samples studied, only six samples from four different brands, exceeded the acceptable ethanol level.

Characterization of banana fiber extracted from common banana varieties in Sri Lanka

M.D.Y. Milani^{1*}, D.S. Samarawickrama¹, G.P.C.A. Dharmasiri²

¹ *Materials Technology Section, Industrial Technology Institute, Sri Lanka*

² *University of Cambridge, Cambridge, United Kingdom*

The structure, the morphology and the thermal behavior of fibers extracted from common banana varieties ('Ambun' (Cavendish AAA), 'Ambul' (Mysore AAB), 'Kolikuttu' (Silk AAB), 'Seenikesel' (Pisang Awak ABB), 'Alukesel' (ABB) found in Sri Lanka were investigated using Fourier Transform Infrared spectroscopy (FTIR), Scanning Electron Microscopy (SEM) and Differential Scanning Calorimetry (DSC). All the samples exhibited similar FTIR spectrums and DSC thermograms. Morphological studies of fiber surface using SEM analysis revealed that there is a rough surface with filaments in all five varieties and it further revealed that there is a hollow structure in the aforesaid fiber varieties. Present study suggesting that any of these fibers and /or mixture of these fibers can be used for appropriate application based on the similar properties of these different banana varieties.

Acknowledgement: DFATD Canada / IDRC Canada research grants

Optimization of process of synthesis of graphite oxide from Kahatagaha graphite

L.C.D. Nayanajith¹, I.R.M. Kottegoda¹, S.R.D. Rosa²

¹Materials Technology Section of Industrial Technology Institute, Sri Lanka

²Department of Physics, University of Colombo, Sri Lanka

Graphite oxide (GO) is a derivative of graphite that consists of carboxyl, hydroxyl and epoxy groups attached to the carbon atoms of graphite. GO is regarded as a value added product of graphite. Usually, GO is synthesized using the Hummer method, where graphite is oxidized using KMnO_4 and H_2SO_4 at controlled conditions. In this effort, Kahatagaha graphite (KGr) in powder form was used as the precursor for the synthesis of GO. The conventional Hummer method was modified by varying its reaction temperature and time in order to obtain the highest yield of GO from KGr with the optimal functionalization. The oxidation time employed was 1 h, 4 h and 24 h and the temperature applied was 35 °C and 90 °C (Products obtained by varying the reaction conditions were labeled as ${}^{\text{Time}}\text{GO}^{\text{Temperature}}$). The resulting products from each experiment were characterized with FTIR and XRD spectroscopic techniques and the yields of GO were estimated gravimetrically. The purpose of performing XRD was to check the presence of unreacted graphite in the synthesized GO where 2θ peak of graphite oxide and graphite appears at about 11° and at about 26° respectively. XRD indicated that unreacted graphite was not present in the ${}^{24}\text{GO}^{35}$ and ${}^{24}\text{GO}^{90}$. FTIR was used to identify the functional groups attached to GO which indicated as peaks at characteristic frequencies in the spectrum (C=O around 1700 cm^{-1} O-H around 3200 cm^{-1} and C-O-C around 1200 cm^{-1}). Their intensities indicated the amount of functional group attached to the structure. FTIR results of GO showed the peak intensities of their functional groups varying in a manner that ${}^{24}\text{GO}^{90} > {}^{24}\text{GO}^{35} > {}^4\text{GO}^{90} > {}^1\text{GO}^{90} > {}^4\text{GO}^{35} > {}^1\text{GO}^{35}$. Also, gravimetric analysis indicated the yields of the synthesized GO as ${}^{24}\text{GO}^{35} > {}^4\text{GO}^{90} > {}^{24}\text{GO}^{90} > {}^1\text{GO}^{90} > {}^4\text{GO}^{35} > {}^1\text{GO}^{35}$. The optimal result was shown by the product ${}^{24}\text{GO}^{35}$ which does not retain unreacted graphite, shows the highest yield and possesses the optimal functionalization.

Acknowledgements: NRC grant 12-022

Microbial Bioremediation of petroleum hydrocarbon contaminated soil and water

E.Y. Fernando*, C.K. Hemachandra, C.U. Patabendi, S. Prabagar, W.R.K. Fonseka
Environmental Technology Section, Industrial Technology Institute, Sri Lanka

Soil and water contamination with petroleum products is a chronic problem faced by many countries around the globe including Sri Lanka. Many constituents in petroleum hydrocarbons are persistent in nature. Hence, in chronically exposed localities such as refineries and storage/bunkering facilities, heavy petroleum contamination of soil and water takes place. In Sri Lanka, there have been instances in the recent past where accidental release of petroleum products into the environment causing a public outcry and raised serious concerns by the relevant authorities. Contamination of the environment by such recalcitrant petroleum products could potentially induce devastating effects on humans and all other exposed ecosystems. Many high-molecular weight aromatic hydrocarbons are especially known for their acutely carcinogenic and mutagenic nature. Therefore, in Sri Lanka, it is imperative that adequate research efforts are made in order to alleviate the adverse effects of environmental contamination by petroleum products. The specific objective of the current study was to apply the environmentally friendly approach of microbial bioremediation to address this persistent environmental problem.

Several highly contaminated sites were selected and sampled during the study. Soil and water samples were taken in sterile vials and the petroleum hydrocarbon exposed microbiota were cultured in the laboratory. The minimal medium used for culturing these environmental isolates was the specially formulated Bushnell-Haass Minimal Medium (BHMM), where the selected hydrocarbon contaminant of choice was supplemented to the medium as the sole source of carbon for microbial growth.

The preliminary results of the study indicated that microbiota isolated from four contaminated localities were capable of growth within the BHMM when the medium was supplemented with a heavy diesel fuel mixture. The turbidity of the cultures (at 600 nm) rapidly increased compared to an un-inoculated control, within a time-span of 120 hours. This implies that the cultured bacterial isolates from the contaminated localities were capable of rapidly degrading the heavy-diesel fuel mixture. Currently, work is

under way to screen the environmental isolates for degradation of different petroleum hydrocarbon mixtures and to conduct molecular identification of bacterial species responsible for hydrocarbon degradation.

Acknowledgement: Treasury Grant TG 15/99

**Advanced air dispersion modeling of SO₂ to accommodate air polluting industries
in proposed industrial zone at Hambantota**

D M H S Dissanayake*, K A N Kumarasinghe

Environmental Technology Section, Industrial Technology Institute, Sri Lanka

This air dispersion modeling study was done using 'ISC-AERMOD View' software 'Version 8.5.1' to predict the effect of the point source emissions of the proposed industrial zone adjacent to the Magam Ruhunupura Mahinda Rajapaksha (MRMR) Port at Hambantota on the ambient air. The study was carried out for three air quality parameters; sulphur dioxide (SO₂), oxides of nitrogen (NO_x) and particulate matter as total suspended particulate (PM as TSP) under different scenarios (based on the fuel type and options for locating industries). The predicted ambient concentrations of SO₂ and NO_x were averaged for 1 h, 8 h and 24 h; while that for PM was averaged for 24 h and annual in order to compare the predicted maximum emission concentrations with their corresponding ambient air quality (AAQ) standard.

According to the model output, except predicted maximum 1-hr average SO₂ concentrations, all other time averaged maximum SO₂ and NO_x and PM concentrations comply with the respective AAQ standard. Hence 1-h average SO₂ concentration in the ambient air was considered as the limiting factor (*i.e.* decision making factor) for this modeling study. Under this circumstance, four options for locating industries and three SO₂ monitoring locations were recommended.

Evaluation of road traffic noise in Sri Lanka as a case study

C.M.Kalansuriya^{1*}, D.U.J.Sonnadara², A.S.Pannila¹

¹ *Electro Technology Laboratory, Industrial Technology Institute, Sri Lanka*

² *Department of Physics, University of Colombo, Sri Lanka*

The main objective of the study was to determine the existing road noise levels in different types of roads in Sri Lanka and relate to the flow of vehicles. Thus, the traffic noise level variations were evaluated with respect to vehicle composition during the day time and night time.

The study was carried out covering different type of roads with different widths and number of vehicles in the outskirts of Colombo, the commercial capital of Sri Lanka. Measurement location of the road selected was from approximate 30 km from the city of Colombo. At this distance congestion due to traffic is less and the flow of traffic is quite smooth. Noise measurements were carried out approximately 5 m from the edge of the traffic route and 1.5 m above the ground level. In each location, noise measurements were taken for duration of 13 h on weekdays categorized as, early morning, late morning, afternoon, evening and night.

The results of the survey showed that the average noise levels L_{Aeq} did not change significantly throughout the study. According to the results analyzed, marked difference was observed between L_{Aeq} and L_{90} values. A large variation was observed between L_{90} , L_{50} and L_{10} values. Noise level variation was not constant and significantly fluctuated from time to time.

The overall mean value of category A road L_{eq} was 73.7(A) with a standard error of 0.14 dB(A). For category B road, the overall mean value of L_{eq} was 67.8 dB(A) with a standard error 0.14 dB(A). It can also be observed that for local roads, nearly 99% measured noise levels were below 70 dB(A). However, for category A roads level below 70 dB(A) was only 2% which is a significant variation. Over 75% exceeded 72 dB(A) and 13% exceeded 75 dB(A). Category B road nearly 95% measured noise levels were below 70 dB(A). This needs to be addressed through policy decision for the control of traffic noise.

Fabrication of a machine to calibrate the measuring tapes by mechanical comparison technique

N. P. Liyanawaduge^{1*}, A. Sooriyaarchchi²

¹*Industrial Metrology Laboratory, Industrial Technology Institute, Sri Lanka*

²*Engineering Services, Industrial Technology Institute, Sri Lanka*

This describes the newly developed tape calibration facility of Industrial Metrology Laboratory of Industrial Technology Institute. The system is capable of calibrating measuring tapes up to 10 m by direct comparison method. The system is constructed on stainless steel wall which is fixed to a series of parallel L-irons, fixed to the cement floor. The system includes several important parts namely measuring head, tape supporting arms and tape tensioning system. The system operates on the mechanical comparison method to calibrate the tapes. The reference tape is moved while keeping the test tape unmoved by rotating a micrometer head. This movement is measured by a digital dial gauge. Both the reference and test tapes are supported by a series of horizontal supporters having circular cross section. These supporters are placed 0.5 m distance apart along the system. The tapes are stretched out up to the appropriate tensions by tensioning weights. A Closed-Circuit Television (CCTV) camera is used for clear vision of scale marks. Coincidence of scale marks is done by viewing the image, which is sent by the camera to a LCD display. To compensate the temperature variation from the reference temperature, the temperature of the tapes is measured by a digital thermometer. The deviation in length from zero of the test tape is measured with 1 μm resolution. The correct length assigned to the scale mark is determined by considering temperature and sag correction. A model to calculate combined standard uncertainty of the measured length is also formulated. This comprises the uncertainties from reference standard, dial gauge, vertical and horizontal parallelism, coincidence, temperature effect and tension of tapes. The performance of the system was tested by a calibrated tape. It was observed that the test results are in agreement with the calibration curve, given in the calibration certificate of the tape.

Acknowledgement: Treasury grant TG 12/00/04

Ministry of Science, Technology and Research



4Ever Skin Naturals (Private) Limited

Hemsons International (Private) Limited



Analytical Instruments
(Private) Limited

Nature's Beauty Creations Ltd



Javana Graphics (Pvt) Ltd. Manufactures
of Panther brand products.

PrintXcel (Private) Limited.
Manufactures of Promate brand products





Industrial Technology Institute

363 | Bauddhaloka Mawatha

Colombo 07 | Sri Lanka

Tel: +94112379800 | email: info@iti.lk

