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MESSAGE from Dr. Shahnaz Murad

It gives me a great privilege to be invited by the Malaysian Society of Parasitology and Tropical Medicine to write a message in the souvenir programme on the occasion of the 46th Annual Scientific Conference.

The Health Ministry, and in particular IMR has observed the growing scientific strength of the society over the 4 ½ decades. I take this opportunity to applaud your consistency and commitment throughout this period to disseminate knowledge especially in the field of Parasitology and Tropical Medicine.

It is inevitable that being in the tropical belt, Malaysia will continue to bear the burden of tropical diseases in particular, parasitic infection. Increasing HIV case, H1N1, dengue fever, shifting landscapes and changing climate can pose a consequence to disease transmission.

It is very encouraging, to note that the Malaysian Society of Parasitology and Tropical Medicine is organizing its 46th Annual Scientific Conference to focus the theme "Infectious Diseases: From Epidemic to Pandemic". This theme is timely and appropriate. We are faced with numerous epidemic which are becoming pandemic. I am happy to note the participants will be deliberating on the theme. The efforts by the government complimented by the contributions of "Scientific Non-Governmental Organization" similar to MSPTM can go a long way in helping create a healthy "One (1) Malaysia".

It is therefore, my sincere hope that, a conference such as this, which brings together the knowledge and expertise of distinguished scientist from Malaysia and abroad will result in an informed and beneficial exchange of ideas and lead to closer inter- regional cooperation and more realistic appreciation of the problems related to the effective management of epidemic diseases.

I am confident that the participants will find it useful and stimulating in their continuing research and knowledge to find effective and practical solutions to health problems facing Malaysia.

I wish all success in your deliberations, in particular your panel discussions where useful recommendations and resolutions arising, can help the Ministry of Health to formulate the way forward.

To all participants, I wish you the very best.

Mahner

Dr. Shahnaz Murad Director Institute for Medical Research



MESSAGE from Assoc. Prof. Dr. S Vellayan

With great pleasure I extend a warm welcome to all participants to the 46th Annual Scientific Conference and Annual General Meeting of the Malaysian Society of Parasitology and Tropical Medicine. The Organizing Committee has planned a very interesting program. The theme for this year is "Infectious Diseases: From Epidemic to Pandemic". The above theme is timeless, because of rapid development in Malaysia we encountered epidemic diseases transforming into pandemic affecting the world's population. The massive migration of population, both legal and illegal can bring its own share of problems from thus contributing to the epidemic to pandemic status. The two-day scientific deliberation will highlight some of the recent emerging epidemic and pandemic tropical zoonotic diseases and prepare us to face the challenges that lie ahead with great preparedness.

The highlight of the 46th Annual Scientific Conference will be the Panel Discussion, which will be chaired by Dr. Lee Han Lim, Head of Medical Entomology Unit, IMR. I would like to thank the members of the Panel, namely Dr. Kamaruddin Md Isa, Dr. Suresh Kumar, and Dr. Chong Chee Kheong.

The response to the call for free papers and posters has been overwhelming. A total of 128 papers comprising 63 oral presentations and 65 posters shall be presented during the two-day conference. The papers cover a variety of topics including Medical Entomology, Medical and Veterinary Parasitology, Molecular Biology, Immunology and Tropical Diseases. The subjects are clear indication of the wide range of professionals who form the membership of the society. It is with such support from the members of the society that has made all our annual conferences very successful.

I take the opportunity to thank Dr Shahnaz Murad, Director, Institute for Medical Research, Kuala Lumpur for her gracious acceptance to officiate the conference. Our sincere appreciation too to Dr. Shahnaz Murad, for her constant support on the society's activities. I would also like to thank all the speakers, chairpersons and the participants. The society would like to thank Prof Dato' Dr Khalid Yusoff, Dean, Faculty of Medicine, UiTM, Shah Alam to present the Honorary Membership Certificate to Prof Emeritus Dato' Dr C P Ramachandran. This year we are proud to announce that the winner of the Sandosham Gold Medal is Mr John Jeffery and the MSPTM Silver Medal winner is Dr Mohd Khadri Sahar. I would like to congratulate them

I would like to place on record and thank the council members and members of the organizing committee, subcommittees, the many sponsors during the economic turmoil period and the members of the society for ensuring the smooth running and success of the conference. Your active participation and contribution are essential to us. We will be documenting the two-day deliberation and forward it to the relevant ministries for implementation.

In closing, I wish all participants a rewarding and fruitful two-day conference and to the foreign participants, stay longer to enjoy the beauty of Malaysia. THANK YOU.

Assoc: Prof. Dr. S Vellayan President and Chairman of the Organizing Committee Malaysian Society of Parasitology and Tropical Medicine



SANDOSHAM GOLD MEDAL CITATION Mr. John Jeffery

It has been my privilege and pleasure to know Mr. John Jeffery since 1976 when I joined the Universiti Kebangsaan Malaysia (UKM) as a young and fledging lecturer, with little experience in teaching and research (outside of my Ph. D work). If I have risen to become a Professor of Parasitology (and served as editor of Tropical Biomedicine for 8 years) is in no small measure due to my association with John both personally and academically. He is inspired and inspires. He is unassuming, diligent and of high moral standard.

Research runs in John's veins, fuelled and inspired by his stint with American scientists from the Hopper's Foundation (HF) and the Arbovirus Research Unit (ARU) under Dr. Albert Rudnick in the 1970s. Even then his ability was noted by Dr. Rudnick: 'one of the best trained entomological technologist in all of Southeast Asia.' He is regarded highly by Prof. Sivaji Ramalingam and Dr. Bruce Harrison. He well known for his expertise in the field of taxonomy of mosquitoes. But his interest in research is diverse to encompass fields as protozoans, malaria, dengue, helminthes, parasitoids, zoonotic diseases, cockroaches, health education, forensic entomology etc. His broad interest has brought him in contact with researchers from many institutions in Malaysia. I dare say that some academics were inspired to do more research by his sheer enthusiasm. He is a prolific publisher of research papers, 201 todate! I belief that submission of his research publications alone would have earned him a postgraduate degree.

John loves field research, an activity that he picked up from his days with the HF and ARU. During field work he is a tireless person, often working late into the night when others have retired. He is a great team worker.

In UKM he was outstanding as researcher and senior medical laboratory technician. He is fondly remembered by many ex-medical students for his cheerful guidance given during parasitology practical classes.

John Jeffery has held all the major posts in the Malaysian Society of Parasitology and Tropical Medicine (MSPTM) except that of the President (reluctant to take it up!) and secretary. He is probably one of the few who knows all aspects of the MSPTM at tip of his fingers. He works quite tirelessly for the good of the association; in the Council, during annual seminars, mid-term seminars, Newsletters, Tropical Biomedicine etc. John has served as an Assistant Editor of Tropical Biomedicine for over 15 years.

John has received several accolade. In 1987 John was awarded the MSPTM in recognition of his contribution to research. The Malaysian Society of Pathologist awarded him the MSP Boehringer Mannheim Award in 1988. John is one of the few to receive the National Technology Award twice, in 1993 and 1996.

A truly exemplary scientist deserving of the prestigious Sandosham Gold Medal from our beloved Society.



MSPTM MEDALIST 2009 CITATION Dr. Mohd Khadri Shahar

Dr. Mohd Khadri Shahar had his tertiary education in three universities in Malaysia. Diploma in Sains from UiTM, Shah Alam, BSc. (Hons) and MSc from University Malaya and his PhD from University Sains Malaysia in 2005 and became the sole resource person of the country in providing ground information of sand flies studies. He joined Institute for Medical Research in 1993 during his MSc programme with University of Malaya in neuro-electro-toxico-physiology field and successfully demonstrates the effect of microwave on neuron. In 1997, he obtained his Diploma in Parasitology and Tropical Medicine. He served the School of Applied Parasitology and Entomology since 1995 as lecturer and also has served as supervisors for 28 DAP&E students of various nation.

He is a recipient of several grants namely TORAY Science Foundation for Phlebotomine study; National Institute of Health Malaysia for *Leptoconops* and *Aedes* pheromone projects; and SEAMEO TROPMED research projects for various entomological studies. He developed and disseminated guidelines for controlling the phlebotomine sand flies and *Leptoconops* biting midge for Ministry of Health Malaysia and has been great help to national tourism industry at beach resorts and chalets. Some of those research works has made him also as a sole resource person in bionomic and control measures of biting midge of sandy beaches in Malaysia. He has contributed over 35 publications in various disciplines of entomology in reputed local and international publications. In year 2005, IMR has awarded him the excellent service award.

Achievement of research works has make him been entrusted to lead research team on *Aedes* Pheromone for year 2006 to 2009 in collaboration with Universiti Malaysia Pahang. Currently, he and his team have been working closely with a BioNexus status company to commercialize their Autocidal Trap.

He has been an active member of MSPTM since 1994. Has played on active role in the council of the society. Has published numerous scientific articles in our Journal Tropical Biomedicine and presented his research findings at our conferences.

Dr Khadri has reached out to the public over the air, through the National Radio, Radio K.L., Radio 1. He has also interviewed in the Malaysian Television programmes on Mosquito biology and its diseases and species.

For his immense contributions to our society in particular to the progress of science in general, the Malaysian Society of Parasitology and Tropical Medicine takes great pleasure in awarding its prestige award - Silver Medal Award to Dr Mohd Khadri Shahar.

PROGRAMME 24 MARCH 2010 WEDNESDAY (DAY 1) Grand Ballroom Hotel Grand Seasons

TIME	VENUE : GRAND BALLROOM
	24 March 2010 MORNING
0730–1030	OPENING CEREMONY
0730–0845	Registration
0845–0855	Arrival of Guests
0855–0900	Arrival of YBhg Dr. Shahnaz Murad, Director, Institute for Medical Research, Kuala Lumpur
0900-0910	Welcome address by President MSPTM and Chairman Organising Committee – Assoc. Prof. Dr. S Vellayan
0910-0930	Opening Speech by YBhg Dr. Shahnaz Murad, Director, Institute for Medical Research, Kuala Lumpur
0930–1000	Sandosham Gold Medal Award MSPTM Silver Medal Award Presentation of the Honorary Membership Certificate
1000–1030	Visit to Exhibition Poster Arena / Tea Break
1030–1000	Presidential Address – Past, Present and Future of Veterinary Parasitology in Malaysia
1100–1300	Panel Discussion : "INFECTIOUS DISEASES: FROM EPIDEMIC TO PANDEMIC"
	Chairperson : DR. LEE HAN LIM (Head of Medical Entomology, Infectious Disease Research Centre, Institute For Medical Research, Kuala Lumpur)
1100–1130	Panel Speaker 1 Hemagglutinin (Ha) Gene Of The Highly Pathogenic Avian Influenza (HPAI) H5N1 Virus Has Not Changed. DR. KAMARUDDIN MD ISA (Director, Division of Farming Technological Resource Development, Department of Veterinary Services Malaysia, Ministry of Agriculture, Putrajaya)
1130–1200	Panel Speaker 2 H1N1 – The Final Verdict. DR. SURESH KUMAR Consultant Infectious Diseases Physician, Department of Medicine, Hospital Sungai Buloh.
1200–1230 1230–1300	Panel Speaker 3 Dengue Epidemiology and Challenges in Control. DR. CHONG CHEE KHEONG Sector Head, Vector Borne Disease Control, Ministry of Health, Putrajaya.
1230–1300	PANEL DISCUSSION
1300–1400	LUNCH

TIME	VENUE : GRAND BALLROOM
	24 March 2010 AFTERNOON
1400–1530	SESSION 1 BIOLOGY AND SURVEILLANCE OF AEDES VECTORS Chairperson : Prof. Sallehudin Sulaiman (Department of Biomedical Science, Faculty of Allied Health Sciences, UKM)
1400–1420	S1.1 Plenary Paper : Solving the dengue riddle with RIDL. Vasan SS (Adjunct Professor, Centre for Research in Biotechnology for Agriculture, University of Malaya, 50603 Kuala Lumpur)
1420–1430	S1.2 Effectiveness of a mosquito larval trap treated with <i>Bacillus thuringiensis</i> var <i>israelensis</i> or temephos for the control of <i>Aedes</i> mosquitoes. Khadri MS
1430–1440	S1.3 A Predictive Model for dengue outbreak in Malaysia. Suzilah I
1440–1450	S1.4 Ovitrap Surveillance as an effective tool in the integrated vector control management.Nawi S
1450–1500	S1.5 Effects of synthetic semiochemicals on Aedes aegypti (L.) behavior. Nurulhusna AH
1500–1510	S1.6 No evidence of dual infection of chikungunya and dengue 2 viruses in laboratory infected <i>Aedes aegypti</i> (L.) and <i>Aedes albopictus</i> Skuse. Rozilawati H
1510–1520	S1.7 Active mating time of Aedes aegypti (L.). Shuhaida MI
1520–1530	S1.8 The Skip Oviposition Behaviour of Laboratory, Field and RIDL strain of <i>Aedes aegypti</i> . Nazni WA
1530–1550	TEA BREAK
1550–1720	SESSION 2 MEDICAL PARASITOLOGY Chairperson : Prof. Rohela Mahmud (Head of Department of Parasitology, Faculty of Medicine, University Malaya, Kuala Lumpur)
1550–1610	S2.1 Plenary Paper : Water borne zoonosis. Suresh K and Tan TC (Department of Parasitology, Faculty of Medicine, University Malaya, Kuala Lumpur)
1610–1620	S2.2 Analysis of <i>Entamoeba histolytica</i> Antigens Specific for Amoebic Liver Abscess. Lim BH
1620–1630	 S2.3 Intestinal protozoa among humans and vegetables in Baghdad province – Iraq. Baha Latif
1630–1640	S2.4 The effects of the maturity levels of cysticercoid on the infectivity of <i>Hymenolepis diminuta</i> in the rats. Hairul Hafiz M
1640–1650	S2.5 Isolation Of Acanthamoeba spp. from river and water recreation In Perak.Putri Noradyani MH
1650–1700	S2.6 Parasites of frogs found in Sungai Pinang, Penang. Cheah SX
1700–1710	S2.7 Surgical Operation, As a Risk Factor for HCV Infection, A Study Testing Ab, RNA & Genotypes of HCV. Waqar AL- Kubaisy
1710–1720	S2.8 Knowledge, Attitude and Behaviors of students of tertiary institutions in north western Nigeria to HIV/AIDS. Magaji BA
1730	AGM (Members of MSPTM Only)
2030	DINNER

25 March 2010 Thursday (DAY 2) Grand Ballroom Grand Seasons Hotel

	25 March 2010 MORNING
0800-0830	Chairperson Assoc. Prof. Dr. Vellayan S (Faculty of Medicine, UiTM, Shah Alam)
	SANDOSHAM MEMORIAL LECTURE The Future of Parasitology and Tropical Medicine. Prof. Mak JW (Dean of Postgraduate Studies and Research, International Medical University, Kuala Lumpur)
0830–0930	SESSION 3 MALARIA VECTORS AND TREATMENT
	Chairperson: Prof. Dr. Fong Mun Yik (Department of Parasitology, Faculty of Medicine, University Malaya, Kuala Lumpur.)
0830–0850	 S3.1 Plenary Paper : Drug discovery for neglected diseases – Malaria & Filaria. Noor Rain A (Head of Bioassay Unit, Herbal Medicine Research Centre, Institute for Medical Research, Kuala Lumpur)
0850–0900	S3.2 Field evaluation of residual-sprayed deltamethrin Wg and deltamethrin Wp on different type of walls for the control of malaria vector in Serian, Sarawak. Rohani A
0900-0910	S3.3 Influence of red fruit oil against pathogenesis of malaria. Susy T
0910-0920	S3.4 PFCRT K76T mutation of <i>Plasmodium falciparum</i> Isolates In Sabah: First Evidence. Nor Azrina N
0920–0930	\$3.5 Composition of species and biting activities of adult mosquitoes in Balai Ringin, Serian, Sarawak. Malinda M
0930–1030	SESSION 4 FORENSIC AND MEDICINAL ENTOMOLOGY
	Chairperson: Prof. Baharudin Omar (Department of Biomedical Science, Faculty of Allied Health Sciences, UKM)
0930–0940	S4.1 Developmental times of forensically important flies in Malaysia. Kumara TK
0940–0950	S4.2 An analysis of different temperature variables on the growth of <i>Chrysomya megacephala</i> (Fabricius) (Diptera:Calliphoridae) at environmental condition. Raja Muhammad Zuha RK
0950–1000	S4.3 Assessment of morphology- and DNA-based identifications for entomological specimens of three selected forensic cases in Malaysia. Tan SH
1000–1010	S4.4 A preliminary study on cow dung diptera community in Sentul Timur, Kuala Lumpur. Heo CC
1010–1020	S4.5 <i>In vitro</i> antibacterial activity of medicinal <i>Lucilia cuprina</i> larvae (Diptera:Calliphoridae) against selected pathogenic bacteria. Teh CH
1020–1030	S4.6 Effect of protein on oogenesis of <i>Lucilia cuprina</i> (Wiedemann) and its utilization in maggot debridement therapy. Yeong YS
1030–1045	TEA BREAK
1045–1205	SESSION 5 CONTROL OF AEDES VECTORS
	Chairperson : Prof. Ridad Agoes
1045–1105	S5.1 Plenary Paper : Changing trends in measles and rubella incidence since inception of the measles-mumps-rubella immunization in Malaysia. Saraswathy TS (Virology Unit, Infectious Disease Research Centre, Institute for Medical Research, Kuala Lumpur)

1105–1115	S5.2 Biochemical detection of resistance mechanisms in field-collected <i>Aedes</i> (<i>Stegomyia</i>) <i>aegypti</i> (L.) in Shah Alam, Selangor. Loke SR
1115–1125	S5.3 Ovicidal effects of Bacillus thuringiensis israelensis on Aedes aegypti. Andy TW
1125–1135	S5.4 Development of permethrin resistance in several strains of <i>Aedes aegypti</i> (L.). Wan-Norafikah O
1135–1145	S5.5 Effects of temperature stress on development of <i>Aedes aegypti</i> (L.) and <i>Aedes albopictus</i> Skuse. Suhaiza H
1145–1155	S5.6 Simulated field performance of Spinosad DT against <i>Aedes aegypti</i> in Penang, Malaysia. Adanan CR
1155–1205	S5.7 Biological efficacy of natural Juvenile hormone III (JH III) from a weed plant. Fatemeh K
1205–1305	Chairperson : Dr. Mohd Khadri Shahar (Medical Entomology Unit, Infectious Disease Research Centre, Institute for Medical Research, Kuala Lumpur)
1205–1215	S5.8 Sub-lethal dose of diflubenzuron and susceptibility status of <i>Aedes albopictus</i> and <i>Aedes aegypti</i> towards diflubenzuron in Penang island. Chan HH
1215–1225	S5.9 Effects of atmospheric temperature on susceptibility of <i>Aedes (Stegomyia) aegypti</i> to vaporized acetone. Jahangir K
1225–1235	S5.10 Simulated field performance of Diflubenzuron against <i>Aedes aegypti</i> in Penang, in Malaysia. Muhamad Firdaus A
1235–1245	S5.11 Efficacy of aqueous extracts of <i>Areca Catechu</i> against larvae of <i>Aedes (Stegomyia)</i> aegypti. Prabadevi N
1245–1255	S5.12 Bionomic and susceptibility status of <i>Aedes</i> mosquito on selected insecticides in USM campus, Penang, Malaysia. Ang CY
1255–1305	S5.13 Knockdown effect of lemon (Citrus limon) peel extract as an insecticide towards <i>Culex</i> sp. mosquitoes with spraying method. Agustin I
1305–1400	LUNCH
1400–1530	SESSION 6 VETERINARY PARASITOLOGY
	Chairperson : Dr. Chandrawathani P
1/00_1/20	S6 1 Plenary Paper : Molecular phylogeny and Bio-geography of Food-borne Zoonotic
1400-1420	Helminths. Yukifumi N (Dept. of Helminthology, Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand)
1420–1430	S6.2 In vitro validation of anthelmintic activity of Butea monosperma and Calotropis procera. Zafar I
1430–1440	S6.3 Validation of FAMACHA eye score system in goat. Nor Azlina AA
1440–1450	S6.4 Oxyspiruriasis In Zoo Birds - Treatment and Control. Vellayan S
1450–1500	S6.5 Movements and home range of a common species of tree-shrew, <i>Tupaia glis</i> surrounding houses of otoacariasis cases in Kuantan, Pahang, Malaysia. Mariana A
1500–1510	S6.6 Severe anthelmintic resistance in commercial small ruminant farms in Perak, Malaysia. Imelda LV
1510–1520	S6.7 Diagnosis of leptospirosis, brucellosis & melioidosis disease in humans conducted in VRI. Naama T.

1520–1530	S6.8 The use of some common Malaysian herbs for worm control in goats in Malaysia. Theivanai J
1530–1545	TEA BREAK
1545–1830	SESSION 7 STUDENTS COMPETITION PRESENTATION
	Chairperson : Dr. Siti Nursheena Mohd Zain (Institute of Biological Sciences, Faculty of Science, University of Malaya, Kuala Lumpur) Best Student Oral Presentation Challenge Trophy. PH HENDRY
1545–1555	S7.1 Filariasis in Kuala-Lumpur & Selangor: Entomological, parasitological and molecular studies. Azdayanti M
1555–1605	S7.2 High prevalence of <i>Blastocystis</i> sp. subtype 4 among rural communities in Nepal. Lee li Li
1605–1615	S7.3 Effects of anthelmintic plant; <i>Terminalia catappa</i> towards nutritional and physiological aspects on Sprague-Dawley White Rats. Mohd Azrul L
1615–1625	S7.4 New predictive tools for pre-emptive dengue vector control in north Queensland, Australia. Aishah Hani A
1625–1635	S7.5 Does <i>Blastocystis hominis</i> exacerbate the growth of colorectal cancer cells? Chandramathi S
1635–1645	S7.6 Intestinal parasitic infections among children in Albania: current status and risk factors. Albana S
1645–1655	S7.7 Polytene Chromosome of the malaria vector <i>Anopheles arabiensis</i> Patton in Sudan. Mashair SM
1655–1705	S7.8 Prevalence and risk factors of protozoal infections among patients attending hospitals in Sana'a City, Yemen. Naelah Alyousefi A
1705–1715	S7.9 The comparison of artificial membrane feeding and direct feeding on <i>Culex quinquefasciatus</i> (Say) (Diptera: Culicidae). Siti Nasuha H
1715–1725	S7.10 Comparative study of the macroparasite communities of stray cats from four urban cities in Peninsular Malaysia. Norhidayu S
1725–1735	S7.11 Molecular characterization of <i>Blastocystis</i> sp. isolates from goats in Malaysia. Tan PC
1735–1745	S7.12 Better prediction of the development of liver cirrhosis in chronic hepatitis B infection using both HBV genetic and serum iron biomarkers. Chook JB
1745–1755	S7.13 Genotypic determination of <i>Toxoplasma gondii</i> strains by PCR-RFLP from clinical samples in Malaysia. Puviarasi M
1755–1805	S7.14 Variant surface glycoprotein (VSG) gene repertoires expressed by Malaysian isolates of <i>Trypanosoma evansi</i> . Mahira W
1805–1830	CLOSING CEREMONY
	By Assoc. Prof. Dr. Vellayan S (Immediate Past President) and Assoc. Prof. Dr. Stephen Ambu (President 2010-2011)

Infectious Diseases: From Epidemic To Pandemic ~

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Abstracts

Panel Speakers

Hemagglutinin (HA) gene of the highly pathogenic avian influenze (HPAI) H5N1 virus has not changed

Kamarudin M.I. Department of Veterinary Services Malaysia

Re-appearance of the HPAI H5N1 virus in 2003 in domesticated birds in Asian had resulted in the global spread of the disease in both animal and human population. More than 140 million birds died or were culled in more than 60 countries in the control of the disease in the animal population. The measures successfully controlled the disease in most of the affected countries. However, the disease remained active in some of the countries with less developed veterinary services. In ASEAN, seven of the Member States were affected by the disease. It was successfully eradicated in Malaysia and Thailand, but occurs sporadically in Myanmar, Lao PDR and Cambodia. The disease is endemic in Indonesia and Vietnam. The death of humans due to the same virus and the likelihood of human to human transmission prompted the WHO to declare the HPAI H5N1 virus a potential pandemic risk which threatens millions of human. Although the prediction so far is inaccurate, the threat still exists considering the fact that farmers and poultry handlers are continuously in contact with infected birds, especially in endemic countries. Similar to other influenza viruses, the H5N1 influenza virus undergoes frequent reassortment. Consequently, 7 of the 8 genes namely; PB2, PB1, PA, NP, NA, MP and NS of the 1996 early progenitor virus (Gs/GD/96), have been replaced. Fortunately, the HA gene remains the same. This allows for molecular and phylogenetic studies of H5N1 isolates to be made and the results provide valid comparitive and useful epidemiological information. Based on phylogenetic tree topology of HA analysis, the H5N1 viruses are grouped into 10 clades (0, 1, 2 (2.1, 2.2, 2.3.1, 2.3.2, 2.3.3, 2.3.4, 2.4 and 2.5), 3.4, 5, 6, 7, 8 and 9). Viruses in the same clades are considered to be of similar origin. The HPAI H5N1 viruses that caused outbreaks in Malaysia in 2004 were classified as Clade 1, while outbreaks in 2006 and 2007 were Clade 2.3.4. They were similar with some of the isolates in Thailand and Vietnam during the same period of outbreaks and all had originated from China. Viruses that caused outbreaks in Indonesia were Clade 2.1 and are different from Malaysian isolates. The clade studies suggested viruses that caused disease in Malaysia originated from China and might have entered Malaysia through infected chickens from Thailand (2004) and contaminated meat imported directly from China (2006 and 2007). As long as Thailand is able to maintain their freedom status and proper risk analysis is conducted prior to importation, the risk of re-occurrence of HPAI in Malaysia is considerably low and the pandemic possibility is almost negligible.

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WHO. 2007. Clades Nomenclature System.

H1N1 – The Final Verdict

Suresh Kumar

Consultant Infectious Diseases Physician, Department of Medicine Hospital Sungai Buloh, Jalan Hospital, 47000, Sungai Buloh, Selangor

In late March and early April 2009, a novel influenza strain was first detected in Mexico. Since then the virus has spread to every nook and corner of the world. More than 213 countries and overseas territories or communities have reported laboratory confirmed cases of pandemic influenza H1N1 2009. It has claimed over 16,000 lives. The hype and political activity that this virus created is unprecedented. This has made some quarters call the whole pandemic response as a hoax. Comparison of this pandemic with the seasonal flu epidemics reveals a different age group involvement even though the overall deaths and hospitalization may not be vastly different.

In European and Americas the influenza activity is decreasing and is below the seasonal baseline in these countries. Does this mean the threat of the pandemic is finally over? But south and Southeast Asia is seeing some heightened activity flu activity. Thailand is showing increased influenza activity. The implication of the increase in flu activity in the region is still unknown.

The new vaccine for H1N1 is currently available. It is now being used for patients with high risk of severe outcome with H1N1. The experience of promoting the vaccine to health care workers has not been good. The uptake of the vaccine is poor. More data is emerging on the safety and efficacy of the vaccines that we are currently using.

Dengue Epidemiology and Challenges in Control

Chong Chee Kheong Sector Head, Vector Borne Disease Control, Ministry of Health Putrajaya

In 2009 Malaysia managed to reduce the number of dengue cases by 16% to 41,486 compared to 49,335 in 2008. At present, the total number of cases this year is 12% less than that for the same period last year. However, the trend of the cases coming into year 2010 has been on the upwards and should this trend persist we will not be able to achieve a reduction of cases for this year. The number of deaths for 2010 has exceeded that of 2009 for the same period.

Rapid diagnosis and response is the mainstay of our control activities but this difficult to maintain as it uses a lot of resources. The Ministry of Health is now highlighting the need for "Source reduction through community participation" as an adjuvant to the above. While public awareness is high public participation is still low. Integrated Vector Management will be introduced as well. The efficacy of "Lethal ovitraps" and transgenic mosquitoes is being studied.

The Ministry of Health is and should always be in the forefront in the war against dengue. The 5-year Dengue National Strategic Plan was implemented in April 2009 with the aim of reducing the cases by 10% every year. This was achieved last year but the challenges ahead are many.

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Abstracts

Session 1 Biology and Surveillance of Aedes Vectors

Solving the dengue riddle with RIDL

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Current control methods such as bed nets, ComBI, insecticides and larvicides have not been able to suppress *Aedes* mosquito populations in a sustainable manner below the threshold at which they spread diseases such as dengue and chikungunya. Thus, there is an urgent need to evaluate novel technologies which can fit into existing integrated vector control programmes and help combat *Aedes*-borne diseases. One such promising technology, called 'RIDL', is now available from Oxford University and its part-owned company Oxitec. When homozygous male mosquitoes of genetically sterile RIDL strains mate with wild type female *Aedes aegypti*, their progeny do not survive into biting adult mosquitoes. Sustained releases of large numbers of these sterile males can thus help suppress *Aedes aegypti* population, and in turn the diseases they transmit. Although mass-release of males sterilized by radiation (Sterile Insect Technique) has successfully controlled or eliminated several agricultural and public health pests, radiation damages male *Ae. aegypti* to the extent that they are unable to compete successfully for mates. RIDL has overcome this problem by eliminating the need for radiation – the males are made sterile by genetics. Two RIDL strains are currently showing promise: (i) a bi-sex lethal RIDL strain (codenamed 'OX513') is under evaluation in several disease endemic countries including India, Malaysia and Thailand, while (ii) a new female-specific flightless RIDL strain (codenamed 'OX3604) has shown promise in cage suppression trials conducted in USA. This paper summarizes the status of trials and regulatory progress in various countries on these two strains.

S1.2 Effectiveness of a mosquito larval trap treated with *Bacillus thuringiensis* var *israelensis* or temephos for the control of *Aedes* mosquitoes

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Dengue continues to be the most common vector-borne disease worldwide. Since 2001, the Kuala Lumpur City Hall had introduced the use of a mosquito trap known as Mosquito Larval Trap Device (MLTD) used to trap Aedes larvae. Further improvement of the efficacy of the trap was initiated by incorporating a biolarvicide Bacillus thuringiensis var israelensis (Bti) or temephos. The objective of this study was to determine the effectiveness of treated MLTD in laboratory and field studies. Under laboratory conditions, mosquito preference test on Bti- or temephos-treated MLTD was compared with untreated MLTD. Prior to this, optimal Bti dosage was determined by placing 2, 4, 6, 8 and 10 granules of a Bti corn-cob formulation separately into the MLTDs and placing all the traps into a mosquito rearing cage. In order to record the mosquito preference, sticky plastic cone was attached at the inner part of each MLTD funnel to trap landing mosquito adults. Subsequently, 200 lab-bred gravid Aedes aegypti were released into the cage and the number of mosquito trapped on the sticky surface of the funnel was recorded. The same method was tested on temephos-treated and untreated trap. For field testing, 30 terraced-houses at Taman Kenanga, Malacca were selected. Each selected house, monitored continuosly for Aedes breeding, was provided with 3 MLTDs (Bti-, temephos-treated and untreated) within the house compound. A conventional ovitrap was also placed in the house to monitor the Aedes population weekly. Results showed that 6 granules of Bti corn-cob (0.08g) attracted the highest number of adult mosquitoes in the laboratory indicating that Bti attracted more mosquito compared to temephos-treated (P<0.05) and untreated MLTD (P<0.05). However, 12 weeks of field study showed that untreated MLTD continuously trapped the highest number of larvae for the whole trial period. Bti- treated MLTD also trapped high number of larvae at 8th to 12th week compared to temephos-treated MLTD. Ovitrap index placed in the houses showed that very low index of Aedes breeding 0-3% at 1st and 2nd week and peaked at 5th week (20%) and decreased to 0-7% at 9th-12th week. No pupa was found in Bti- and temephos-treated MLTD but in untreated MLTD pupae were collected continuously. Thus, treated-MLTDs effectively controlled Aedes breeding in housing areas for the trial period of 12 weeks.

S1.3 A predictive model for dengue outbreak in Malaysia

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Early detection of a dengue outbreak is an important first step towards implementing effective dengue interventions resulting in reduced mortality and morbidity. A dengue mathematical model would be useful for the prediction of outbreak and evaluation of control measure. However, such model must be carefully parameterized and validated with epidemiological, ecological and entomological data. A field study was conducted to collect and analyse various parameters to model dengue transmission and outbreak. Dengue prone areas in Kuala Lumpur, Pahang, Kedah, and Johor were chosen for this study. Ovitraps were placed outdoor and used to determine the effects of metrological parameters on vector breeding. Vector population in each area was monitored weekly for 87 weeks. Weather stations, consisting of a temperature and relative humidity data logger and an automated rain gauge were installed at key locations in each study site. Correlation and Autoregressive Distributed Lag (ADL) model were used to study the relationship among the variables. Previous week rainfall plays a significant role in increasing the mosquito population, followed by maximum humidity and temperature. In several localities, the number of notified dengue cases and fogging positively related with the level of mosquito population. The secondary data of rainfall, temperature and humidity provided by the meteorological department showed insignificant relationship with mosquito population compared to the primary data recorded by the researchers. A well fit model was obtained for each of the locality which can be used as a predictive model to forestall possible outbreak.

S1.4

Ovitrap Surveillance as an effective tool in the integrated vector control management

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Effective dengue control measures are very important and the development of effective surveillance tools to detect dengue hot spots areas is crucial. In the integrated vector control management, the use of ovitraps as surveillance tools needs to be examined to show their effectiveness. The objective of this study is to monitor *Aedes* mosquitoes density and dengue incidences in hotspots localities in Kinta District. Seventeen (17) localities were selected as study sites from 1st July 2009 to 31st December 2009 where 16 localities were dengue hotspots and another locality was not a dengue problem area. From each locality, 30 residential premises were selected randomly for the placement of ovitraps. The ovitraps were placed outdoors and collected after 7 days. They were replaced with new ovitraps each time after collected. The ovitraps were further incubated for 5 days and examined subsequently. In the first half (3 months) of surveillance, control measures were carried out after dengue cases were notified whereas in the second half (3 months) of surveillance, control measures were proactively carried out when ovitraps were found positive. 29.6% of ovitraps were found positive in the second half of the study period compared to 23.1% of those in the first half of the study. However, the numbers of dengue cases were reduced by 53.9% in the second half of the study where control measures were carried out proactively. Ovitrap surveillance, together with the integrated vector control measures, may reduce dengue incidence and outbreak episodes in selected areas in Kinta District.

S1.5 Effects of synthetic semiochemicals on *Aedes aegypti* (L.) behavior

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Semiochemical is a substance or mixture of substances which is important in the insect communication system. This study was conducted to observe the behavior of male and female Aedes aegypti towards three synthetic semiochemicals which have the same properties as the natural semiochemicals identified from the mosquito. Three synthetic semiochemicals: tetradecanoic acid, trans-2-octenal and octadecane, which have different chemical structures were used in this study while hexane, an organic solvent was used as a control. Each chemical was prepared at 1 mg/L, 10 mg/L, 100 mg/L and 1000 mg/L. Five mosquitoes were released into the release chamber of a Y-tube, used routinely in such testing. Twenty µL of the prepared semiochemicals was pipetted onto a 30 mm Whatman Antibiotic Assay paper disc and placed in the test chamber arm. After 30 seconds, the door of the release chamber was opened and the mosquitoes were allowed to fly into the test or control chamber arms. Once all the mosquitoes were in the test and/or control chamber, the experiment was stopped. The number of mosquitoes in the test and control chamber was recorded and thirty replicates were conducted for each semiochemical. Male mosquito showed significant attraction towards tetradecanoic acid at 1000 mg/L, 100 mg/L and 10 mg/L (P < 0.05), while at 1 mg/L repelling activities were observed (P<0.05). Female mosquitoes showed significant attraction to 1 mg/L (P<0.05) of all semiochemicals but were repelled at 1000 mg/L (P<0.05). However at 100 mg/L no significant repellent activity was observed. At 10 mg/L both control and test obtained mean value of 3 ± 3.9 and 3 ± 0.67 , respectively, indicating absence of activity. Trans-2-octenal exhibited some degrees of attraction at 1000 mg/L (P<0.05) and 100 mg/L (P<0.05) on the male but was not significant on the female at the same concentration (P> 0.05) and 100 mg/L (P<0.05). At 10 mg/L and 1 mg/L, trans-2-octenal showed repellent effect for both sexes (P<0.05) for all tests. Both male and female were repelled by all octadecane concentrations. All the three semiochemicals exhibited some degrees of attraction and repellency on Ae. aegypti depending on their concentration.

S1.6 No evidence of dual infection of chikungunya and dengue 2 viruses in laboratory infected *Aedes aegypti* (L.) and *Aedes albopictus* Skuse

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Dengue and chikungunya infection, both arthropod-borne viral diseases are now commonly found in Africa and Southeast Asia and transmitted by *Aedes* mosquitoes. The clinical symptoms of chikungunya virus infection often mimic those of dengue and circulates in regions where dengue virus is also endemic. While dual infection of dengue and chikungunya viruses has been reported in the same patient, no such infection has been found in the mosquitoes. To determine if dual infection occurs in the same mosquito, laboratory studies were conducted to examine the susceptibility of two vector mosquito species, namely *Aedes aegypti* and *Aedes albopictus* to chikungunya and dengue virus. Multiplex PCR was used to detect the presence of both viruses in the infected mosquitoes. For each mosquito species, 200 females were orally fed with a human isolate of chikungunya virus and/or dengue-2 virus in human blood via a membrane feeding system and maintained on sugar solution for 14 days at room temperature. Each species were individually separated after the infection. Five mosquitoes were pooled daily and another five were dissected for salivary gland for testing. Multiplex PCR results indicated that both *Ae. aegypti* and *Ae. albopictus* were susceptible to either chikungunya or dengue virus until 14 days post -feeding. However, dual infection of chikungunya and dengue virus in the same mosquitoes was not detected.

S1.8

S1.7 Active mating time of *Aedes aegypti* (L.)

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The active mating time of *Aedes aegypti* was not well understood. The present study was carried out to determine the active mating time of the mosquito. Ten lab-bred virgin males and 10 virgin females aged between 3 to 5 days were released into a rearing cage. Sugar solution (10%) was provided *ad libitum*. Observation on the number of mating frequency at every 40 minute interval was carried out from 7 am to 7 pm for 10 days. The recording started immediately after the females were released into the cage. The peak frequency of the mosquito mating activities occurred from 3 pm to 4 pm averaging 90.6 \pm 20.1 of mating, while the lowest mating frequency was from 9 am to 10 am averaging 45.1 \pm 10.6. At every interval from 10 am to 2 pm, the mosquito mating frequency averaged 55.2 \pm 18.4, 56.0 \pm 14.5, 59.1 \pm 14.8, 70.5 \pm 17.0 and 65.6 \pm 16.6 times respectively. Besides obtaining more information on the mosquito mating behaviour, such information can also be useful in integrated mosquito control such as mass release of sterile male mosquito to compete with wild type male *Ae. aegypti*.

The Skip Oviposition Behaviour of Laboratory, Field and RIDL strain of Aedes aegypti

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Dengue is a major public health problem in many tropical countries world-wide. The principal vector *Aedes aegypti* is closely associated with the human habitats and breeds in man-made containers with clear water. The female *Ae. aegypti* prefers to breed in artificial containers both in indoors and outdoors. *Aedes aegypti* female normally does not lay her entire batch of eggs in one location but rather, distributes eggs in multiple containers, a behaviour known as "skip-oviposition". Surveillance of these mosquitoes is highly dependent on the egg laying behaviour. The objective of this study was to determine the oviposition behavior and egg-laying site selection of three strains of *Aedes aegypti*, namely laboratory strain, field strain and the RIDL strain. Ovitraps with low, intermediate and high number of eggs (con-specific eggs) were provided for the study of skip oviposition behaviour. Our study revealed that the newly emerged *Aedes aegypti* mosquitoes mated before consuming the first blood feeding. The time for the mating varied from 8 to 11 hours after emergence and adults rested for some period before blood seeking. The time for first blood meal is very much similar in the three strains. All three *Ae. aegypti* strains preferred to lay eggs in breeding sites with low egg density of 5-10 eggs/ovitrap. However, when no choice for selection of sites was offered, skip oviposition behaviour was not observed. Our study indicated that 95% of *Ae. aegypti* laid all the eggs within 72 hours. There was also no difference in oviposition when *Ae. aegypti* was provided with ovitraps containing eggs of different strains.

Abstracts

Session 2 Medical Parasitology

S2.1 Water borne Zoonosis – Factors influencing outbreaks

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In the last 20 years, it has been recognized that many disease are caused by emerging and reemerging diseases, 75% of which are zoonotic. Excreted material and other animal waste products are predominant sources of waterborne zoonotic pathogens. Materials such as wastewater /biosolids, faeces, urine and carcass contribute to pathogens that can be transmitted by rivers and other aquatic sources to humans. One third of world's population live in countries with water stress, a number that could increase to two thirds by the year 2025 making 3 billion people suffer from water shortage. Livestock revolution with an increase in animal breeding to meet global diet requirement will also be enhanced many fold causing an increase in water contamination and thus sensitive surveillance, intelligent risk management, coordinated efforts for effective monitoring, collaborative initiates to enhance water quality and the implementation of cohesive seamless cooperation from all concerned should take place. These and other factors will be discussed to highlight factors that contribute to outbreaks and in the light of emerging scenarios such as natural disasters, global warming and other uncontrollable events, discussion points highlighted in the presentation can help evolve recommendations for the future.

S2.2

Analysis of Entamoeba histolytica antigens specific for amoebic liver abscess

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Amoebiasis is caused *Entamoeba histolytica*. Localised infection at the large intestine is classified as intestinal amoebiasis whereas extraintestinal invasion of the colonic mucosal layer may lead to the fatal amoebic liver abscess (ALA). Ultrasound and other imaging techniques cannot differentiate ALA from pyogenic liver abscess. Polymerase chain reaction (PCR), culturing technique and antigen-detection system are reported to be more reliable but obtaining biopsied liver abscess is invasive, undesirable and even unsatisfactory as trophozoites are found mainly at the periphery of the abscess. Seven Syrian golden hamsters were each inoculated intraportally with ~1 x 10⁶ axenic HM-1:IMSS strain of *E. histolytica* to develop ALA, and subsequently obtained the serum sample. Western blotting was then performed by using *E. histolytica* crude soluble antigen (CSA) against each of the seven infected hamster serum samples. The controls included in Western blotting were the pooled serum samples from ALA hamsters; pooled pre-infected serum samples; pooled immunized serum samples and Tris-buffered saline (TBS). The immunized serum samples. Among the 19 antigenic bands, in which none was detected by 6/7 ALA serum samples and also the pooled ALA serum samples. The ~67 kDa and ~51 kDa bands were recognized by 6/7 and 4/7 ALA serum samples, respectively. The diagnostic potential of ~85 kDa polypeptide will be further tested against human ALA serum samples.

S2.3 Intestinal protozoa among humans and vegetables in Baghdad province – Iraq

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This study was conducted from January 5 to September 30, 2006 to detect intestinal protozoa in human stools and vegetables in Baghdad province - Iraq. Five hundred stool samples were collected from patients resided in two hospitals namely Al-Ermook Hospital (229 samples) and Central Child Hospital (271 samples). Different methods were used for diagnosis of protozoa in stool samples including: Direct wet, Sheather's sugar flotation, and Formalin – Ethyl Acetate sedimentation methods. Staining with Giemsa, Modified Ziehl – Neelsen, Safranan and Kinynoun's acid–fast stains. Five intestinal protozoa were detected: *Cyclospora cayetanensis, Cryptosporidium* spp., *Blastocyst hominis, Giardia lamblia* and *Entamoeba histolytica*. Out of 500 stool samples, 157 were positive (31.40%) for intestinal protozoa. Infection was most common among children 1 to 5 years old (44.55%). In regard to the seasonal variation, the highest rate of infection was in August (45.45%) and the lowest was in February (12.2%). Fifty four vegetable samples (lettuce, celery, basil) were collected at several local markets. Each sample was washed thoroughly with distilled water, the supernatant was collected and centrifuged at 3000 rpm for 10 minutes. The pellets were resuspended in normal saline and examined under the microscope. Out of 54 samples, 19(35%) harboured *Cyclospora cayetanensis, Cryptosporidium* spp, *G. lamblia* and *E. histolytica. Toxoplasma gondii* oocysts were also detected in two samples. This is the first report for *C. cayetanensis* in human stool and vegetables recovered in Iraq.

S2.4 The effects of the maturity levels of cysticercoid on the infectivity of *Hymenolepis diminuta* in the rats

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A study of the effects of maturity levels of cysticercoid on the infectivity of *Hymenolepis diminuta* in the rats was carried out at the Entomology Laboratory, University Malaysia Sabah. Three groups of six laboratory rats were used as hosts. Each group was orally infected with five to seven cysticercoid of different maturity levels; 7 days, 14 days and 21 days respectively. All cysticercoid were recovered from *Tribolium castaneum* which were infected with gravid *Hymenolepis diminuta* proglottids. Data were analysed using SPSS version 17. The maturity levels of cysticercoid had significant effects on the infectivity of *H. diminuta*; cysticercoid of 14 days old showed the highest significantly (P<0.05).

S2.5 Isolation of *Acanthamoeba* spp. from river and water recreation in Perak

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Acanthamoeba spp. are ubiquitous, free-living parasites that can cause Acanthamoeba keratitis, a type of corneal inflammation which is associated with contact lens wear. It has been established that one of the risk factors of this infection among contact lens users is swimming in fresh water and swimming pools with their contact lenses in. This study aimed to isolate Acanthamoeba spp. from river and recreation water in several area in Perak. An amount of 500 ml water was sampled from several different locations of Lubuk Timah, Sungai Gerik, Royal Belum Rain Forest and Tambun Water Park in Perak. Then, the water samples were filtered through nitrate cellulose membrane filter and cultured onto non-nutrient agar overlaid with *Escherichia coli*. All culture plates were examined under an inverted microscope for the presence of Acanthamoeba spp. and their morphological characteristics were confirmed under 'Image Analysis Software Video Test 4.0.' The results revealed that 20 percent of the samples from Lubuk Timah and Tambun Water Park were positive for Acanthamoeba. Acanthamoeba were isolated from 25 to 60 percent of the samples from three location of Sungai Gerik; Sungai Hulu Gerik, Sungai Hulu Kenderong and Sungai Kulim. Royal Belum rain forest was known as an isolated place from human activities but 14 percent of the water samples had this amoeba. These finding suggested that contact lens wearers should take extra caution on their practice of contact lens care as river water is a source of domestic water supply and also can be one of the risk of infection while doing water recreation activities.

S2.6 Parasites of frogs found in Sungai Pinang, Penang

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This study documents the parasite fauna found in anurans on Penang island. A total of 35 *Bufo melanostictus* Schneider from Sungai Pinang, Balik Pulau, Penang were collected and dissected in the laboratory for parasite examination. The toads were collected from November 2008 to January 2009 during night time using net or bare hand. Various organs including the blood, liver, lung, stomach, intestine and rectum were examined for presence of parasites. This study documents on the common parasites such as protozoa and helminth that infect Malaysian anurans. The prevalence, mean intensity and distribution of each parasite species along the digestive tract were recorded. Parasites found in anurans are protozoa (*Opalina ranarum* and *Nyctotherus cordiformis*), nematode (*Oxysomatium* sp., *Rhabdias* sp. and one unidentified species) and trematode (*Mesocoelium burti*). However, *Trypanosome* sp. and microfilarial worms which are normally found in the blood of anurans were not recorded in this research. Most of the parasites were found in the intestine and rectum. Blood and liver were negative for parasites. *Oxysomatium* sp. ranked the highest percentage infecting the toads which is 38.90% of the total parasites. *Rhabdias* sp. and *Oxysomatium* sp. are common parasites found in *B. melanostictus* with a prevalence rate of more than 50%. However, all the parasites found are not pathogenic to humans and therefore are of no medical importance.

S2.7 Surgical operation, as a risk factor for HCV Infection: a study testing Ab, RNA & Genotypes of HCV

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Medical risk factors associated with, such as blood transfusion and surgery, have been studied in many countries. Investigations were conducted to identify the role of previous surgical operation as a risk factor for acquiring HCV infection and to identify HCV genotypes among those with such a history. Serum samples of 3491 pregnant women, were initially screened for the detection of (anti-HCV). By third generation enzyme immunoassay reactive results were confirmed by third generation immunoblott assay. In addition 94 mothers' sera, were subjected to molecular analysis for the detection of and genotyping of HCV, using the RT-PCR and DNA enzyme immunoassay method. Anti-HCV seropositivity rate was found significantly three folds higher (7.69%) among women with a history of past surgery compared to (2.6%) in those having no such history p=0-0001. Women who underwent minor operation, showed higher rate of anti-HCV seropositivity (8.14%) than underwent major operation (7.11 those who underwent minor surgery at a governmental hospital exhibited significantly higher rate of anti-HCV, than women who underwent operations at a private hospital (11.3% Vs 4.12 respectively). Past surgical operation was detected as significant risk factor to exposure to HCV infection OR=3.1, particularly when this surgery was performed at a governmental hospital (OR 3.5). High prevalence but not significant HCV RNA was found (78.9%) among women with history of surgical operation, than 58.6% in their counter group. No association was found between HCV genotypes distribution and history of surgery. But 9 of the 15 positive HCV-RNA women were found harboring genotype HCV-4. Surgical operation may act as a risk factor for exposure to HCV infection. Therefore, we underscore for sterilization of instrument and the use of disposable materials, especially when a high turnover of patient is experienced.

S2.8 Knowledge, Attitude and Behaviors of students of tertiary institutions in north western Nigeria to HIV/AIDS

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The first reported case of HIV/AIDS in Nigeria was in 1986, which marked the beginning of the HIV/AIDS epidemic in Nigeria. The prevalence rate is now placed as 5.8%. This study is aimed at exploring the knowledge, attitude and practice of prevention of HIV/AIDS among an important risk group (students in tertiary institutions) with a view of making positive efforts in the control of the menace. A cross sectional analytic study was carried out among 422 students using a self-administered questionnaire. Significant level set at 0.05 CI. The respondents were found to have high knowledge of HIV/AIDS mean score of 83% and with 80% having a positive attitude. The gender was found to have a significant association with knowledge and the attitude of the respondents toward HIV/AIDS on the other hand was significantly associated with age. However, neither the gender nor the institution of study was found to be associated with the attitude. The high-risk behaviors' engaged by the students, which could place them at risk of contracting HIV/AIDS, are having multiple sexual partners (6%) and unprotected sexual intercourse (27%). This study revealed that students had an adequate knowledge of HIV/AIDS as well as positive attitude. However, a significant proportion still practices high risk sexual behaviors. Therefore, a collaborative effort is recommended between the institutions and other stakeholders in HIV/AIDS control for a programme targeting this group of people.

Abstracts

Session 3 Malaria Vectors and Treatment

Abstract

Sandosham Memorial Lecture

The Future of Parasitology and Tropical Medicine

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We often hear that parasitology as a discipline is fast losing its appeal and in danger of fading away. Indeed, parasitology is now often embedded in microbiology, tropical medicine, or infectious diseases in most medical curriculum. Should we welcome this as an inevitable evolvement of practice?

Advances in technology have influenced research and the practice of medicine. The convergence in the use of advanced technologies and molecular approaches to study infectious agents and cellular events has blurred the boundaries between disciplines. Most researchers have embraced these approaches in research but have neglected those generally known as 'classical techniques'. Thus, are we in fact, lamenting about the lack of classical approaches in parasitology and entomology research? Is there a need or are these no more relevant in the post-genomic era?

There are some exciting developments in research which have enhanced interest in parasites. These include opportunistic parasitic infections, 'parasites within parasites' and their role in nosocomial infections, outbreaks involving protozoan parasites, zoonotic infections, globalisation and travel related illnesses, and their role as indicators of environmental quality and health. These developments have shown the interdependence of disciplines and have underscored the relevance of parasites and arthropods in infectious diseases, allergies, public health practice, and occupational and environmental health issues.

Parasitology research is active but has shifted emphasis in approach. While most researchers are comfortable with molecular techniques in research there appears to be a need for training in classical techniques in parasitology and entomology research. There is healthy interdependence between disciplines, and this must be encouraged for the good of research.

Past practitioners in the field have laid extremely strong foundations through their research contributions in parasitology and entomology and are credited for the training and nurturing of current parasitologists and entomologists. We need to continue their exemplary work and through training and mentoring of future generations of practitioners, ensure that these disciplines continue to contribute to global knowledge for health promotion, prevention, and management of parasitic and associated diseases.

S3.1 Drug discovery for neglected diseases – malaria and filariasis

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Neglected Tropical Diseases (NTDs) is a group of tropical infectious diseases that are prevalent in the world's least developed countries. Drug for Neglected Diseases Initiatives (DNDi) defines neglected diseases as any, of a group of disorders that exclusively affect the poor and the powerless in rural and impoverished urban areas of developing countries. The diseases affect the poor who generally cannot afford treatment. After decades of research, some of NTDs are still responsible for human deaths and disability. Some do not cause death, nonetheless they can be an integral cause of poverty. Malaria causes death, the lymphatic filariasis, does not kill but it causes temporary and permanent disfiguring to the patient. It has major social and economic impact to the endemic countries. The emergence of multidrug resistant malaria parasite has reduces the efficacy of currently available antimalarial drugs. Effective drugs for lymphatic filariasis, is very limited to albendazole, diethylcarbamazine (DEC) and ivermectin. None of these is effective in killing the long-lived adult worms. The treatments are aimed at reducing transmission and pathology only. Discovery of new drugs is important towards the elimination for NTDs by 2020, as envisaged in the global campaign "End the Neglect 2020". Ministry of Health Malaysia, is one of the founding partners for DNDi. The Institute for Medical Research has taken up the challenge by initiating a Drug Discovery Program for Neglected Diseases. The program started with the two most common NTDs, malaria and lymphatic filariasis.

S3.2 Field evaluation of residual-sprayed deltamethrin WG and deltamethrin WP on different type of walls for the control of malaria vector in Serian, Sarawak

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Malaria control programme utilizing indoor residual spraying is only effective if a high coverage of targeted area is achieved and an insecticide that is effective against the specific mosquito vector is correctly applied. Efficacy and residual activity of most insecticides are also affected by the nature of the sprayed surfaces. The bioefficacy of indoor residual-sprayed deltamethrin wettable granule (WG) formulation for the control of malaria was compared with the current dose of deltamethrin wettable powder (WP) in malaria endemic areas in Serian, Sarawak. Doses of 20 mg/m² WP, 20 mg/m² WG, 30 mg/m² WG and 40 mg/m² WG were sprayed separately on brick, wooden, rough-bamboo and smooth-bamboo walls. Residual activity of WP and WG formulations were tested against *Anopheles maculatus* using WHO standard procedure. Deltamethrin at a dose of 30 mg/m² WG exhibited sustained level of effectiveness against *An. maculatus* up to 60 weeks post-spraying for both wooden and bamboo walls; but was only effective up to 41 weeks on brick wall. However, at the higher rate of application, i.e 40 mg/m² WG, >90% mortality of *Anopheles maculatus* was obtained for 41,34, 11 and 7 weeks on smooth-bamboo, rough-bamboo, brick and wooden wall, respectively. The results indicated that 30 mg/m² WG is ideal for controlling malaria vector up to 14 months. Where long-lasting residual spraying is envisaged, the usual two spraying cycles per year with 20 mg/m² deltamethrin may be replaced with 30 mg/m² deltamethrin WG every 12 months.
S3.3 Influence of red fruit oil against pathogenesis of Malaria

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Besides *Plasmodium falciparum* resistance against antimalarial drugs, malaria virulence caused by free radical overproduction correlating with high serum level of cytokines becomes a problem. Red fruit containing a lot of antioxidants grows in Papua, a malaria endemic area, and is considered to overcome these problems. The aim of this study is to explore whether red fruit oil can reduce the serum TNF α and Interleukin 6 level which participates in cytoadherence in mice suffering *Plasmodium berghei* malaria as a model of human falciparum malaria. Several doses of red fruit oil were given orally to these mice in five replications. Serum TNF α and Interleukin 6 level was examined using ELISA at the sacrifation/ fourth day. The data were analyzed using ANOVA and Tukey HSD with $\alpha = 0,05$. It was shown that at the optimum doses, red fruit oil could reduce TNF α and Interleukin 6 level as well. These will be reported.

S3.4 PFCRT K76T mutation of *Plasmodium faciparum* isolates in Sabah: first evidence

Nor Azrina Norahmad, Noor Rain Abdullah, Norhayati Yaccob, Jenarun Jelip, Jiloris F. Dony, Khairul Faiz Ruslan, Lokman Hakim Sulaiman, Hasidah Mohd Sidek, Harald Noedl and Zakiah Ismail

Chloroquine (CQ) is the first line drug for the prevention and treatment of malaria in Malaysia. Resistant to CQ have been observed in Malaysia since 1996. It is known that CQ resistant has been associated with mutation in the *pfcrt* gene, which encodes a putative transporter located in the digestive vacuole. The substitution of lysine (K) to threonine (T) at amino acid 76 (K76T) in *pfcrt* gene of 32 *Plasmodium falciparum* positive samples from selected areas in Kalabakan Tawau. The blood samples were collected on 3mm Whatman filter paper and DNA was extracted using QIAmp DNA mini kit. Nested PCR specific mutation for the detection of resistant T76 genotype and sensitive K76 genotype were carried out. Primers used for the PCR were TCRP 1 and 2 followed by PCR with TCRP3 and TCRP4M or TCRP4W for indication of mutation and sensitive to CQ respectively. Both give a visible band of 366bp. For confirmation of polymorphism at codon 76 on the *pfcrt* gene, a nested PCR with TCRP 1 and 2 followed by CRTD 1 and 2 were carried out. The second PCR products were subjected to restriction enzyme digestion with *Apo* I. The PCR and the digested products were analyzed using Agilent DNA 1000 Kit. Our study showed that for both set of nested PCR demonstrated that 25 (78.12%) of the samples showed mutation at codon 76 and 18.75% are still wild type. Digestion of the PCR product with *Apo* I confirm the findings that the samples harboured parasites that were mutant at codon 76.

S3.5 Composition of species and biting activities of adult mosquitoes in Balai Ringin, Serian, Sarawak

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Knowledge of the composition and biting habits of mosquito associated in endemic areas are important in establishing sound vector control programmes and understanding the epidemiology of vector borne diseases. A total of 7057 mosquitoes comprising 27 species belonging to 5 genera were collected from Balai Ringin, Serian, Sarawak from April 2008 to August 2009. Collections were carried out indoors and outdoors for 12 hours from 6.00 pm to 6.00 am using human landing catch techniques. *Mansonia bonneae* (36%) was the predominant species caught in the study areas followed by *Culex vishnui* (32%), *Culex pseudovishnui* (30%) and *Anopheles letifer* (2%). A high rate of human biting by *Ma. bonneae* was detected during November but the activity was low during January. *Mansonia bonneae* biting activity peaked at 7.00 pm – 8.00 pm. *Culex vishnui* also exhibited similar biting activity peak while *An. letifer* exhibited biting activity peaked at 12.00 am – 1.00 am.

Abstracts

Session 4 Forensic and Medicinal Entomology

S4.1 Developmental times of forensically important flies in Malaysia

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The most common application of forensic entomology is in the estimation of the postmortem interval (PMI) based on the rate of development of the carrion flies. For accurate PMI estimation the larvae of four dominant forensic carrions flies namely *Chrysomya megacephala* (Fabricius) (Calliphoridae), *Chrysomya rufifacies* (Macquart) (Calliphoridae), *Sarcophaga* spp. (Diptera: Sarcophagidae) and *Synthesiomyia nudiseta* (Muscidae) which were collected from decomposed human corpses were studied. The emerged adult flies were kept as a stock colony and the duration of development under indoor fluctuating temperature regime was studied. The developmental times, rearing temperatures, and relative humidity were recorded twice daily from the time the eggs were collected until the adults emergence. An average of 5 larvae were randomly collected from the rearings twice daily, warm-water killed ($52 \pm 10^{\circ}$ C) and preserved in Kahle's solution. The larval instar stages were determined by observing the number of posterior spiracular slits and the larval length measured. When the immature stages completed, the measured larvae lengths for each sampling period averaged and were plotted against time at the confidence interval of 95%. The mean \pm s.d. (standard deviation) values were determined for each larval stages and their developmental times. The total duration of developmental of *C. megacephala*, *C. rufifacies*, *Sarcophaga* spp. and *Synthesiomyia nudiseta* were 9.0 \pm 0.5 days, 8.5 days, 12.3 \pm 0.6 days and 13.4 \pm 0.8 days, respectively.

S4.2 An analysis of different temperature variables on the growth of *Chrysomya megacephala* (Fabricius) (Diptera: Calliphoridae) at environmental condition

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The development of blowfly larvae (Diptera: Calliphoridae) growth model induced by temperature effect is an important function in forensic entomology, particularly in determining post mortem interval. Currently available blowfly growth data which established from laboratory were developed using only constant temperature but other thermal factors were not included to reflect the environmental effects impacting its growth. This study aims to determine the effect of different temperature variables in the growth of *Chrysomya megacephala* (Fabricius) using natural fluctuating ambient temperature, and temperature variables from larval mass, food substrate and postfeeding medium. A total of six replicates, each consisting 100 individuals of *C. megacephala* eggs were maintained in rearing containers using beef liver as food substrate. From this study, ambient temperature during the period of egg stage until adult emergence was ranged between 25.0 and 35.5°C (mean = 29.20, s.d. = 2.99), while other temperature values were recorded as: larval mass temperature (mean = 31.27, s.d. = 2.47) and postfeeding medium temperature (mean = 31.33, s.d. = 2.13). Total accumulative hour of development from eggs to pupa eclosion was approximately 187 hours. In a subsequent finding, there was no significant difference of mean temperature variables from the ambient temperature. In order to estimate larval mass temperature, multiple regression analysis was carried out and revealed high correlations between predictors (ambient, food substrate and postfeeding medium), rendering an unstable model. The correlation between temperature variables was further expressed by using structural equation modelling (SEM) diagram.

S4.3 Assessment of morphology- and DNA-based identifications for entomological specimens of three selected forensic cases in Malaysia

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Morphology-based identification of entomological specimens collected at the crime scenes is hindered by damaged or young (immature) specimens, and sometimes by the ambiguity of taxonomic identification. However, morphology-based identification is a faster method compared to DNA-based identification assay which have been used in many forensic entomology studies. In this study, we had assessed both morphology- and DNA-based identification methods in three selected forensic cases in Malaysia. DNA sequences of *cytochrome oxidase* subunit I and II from entomological specimens were successfully used for identification to the species level when the morphology-based identification was hampered. Both methods should complement each other to provide a robust and definite result for species identification, especially in postmortem interval estimation.

S4.4

A preliminary study on cow dung diptera community in Sentul Timur, Kuala Lumpur

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A preliminary survey of cow dung dipteran diversity was conducted in several locations in Sentul Timur, Kuala Lumpur. A total of five visits to a cattle farm were done within two months from November to December 2009. We examined about 50 cow dungs for the recovery of fly larvae and other dung associated arthropods. Adult flies were collected around the dung by using a sweeping net or plastic bag while the larvae were collected by using a pair of forceps. Some of the collected larvae were preserved in the 70% alcohol and the rest were raised to adult stage for identification. We collected 324 dipterans from 16 families namely Sepsidae, Muscidae, Calliphoridae, Sarcophagidae, Psychodidae, Ephydridae, Sphaeroceridae, Dolichopodidae, Lauxaniidae, Stratiomyiidae, Chloropidae, Neriidae, Tabanidae, Tephritidae, Chironomidae and Aphididae. Other than Diptera, we also collected ants (Formicidae), nymph of preying mantis (Acromantinae), leaf hopper (Cicadellidae), adult coleopterans (Scarabidae, Hydrophilidae) and a carabid larva, centipede (Chilopoda), earth worm (Lumbricus terrestris), Symphyla (Myriapoda), earwig (Dermaptera: Forticulidae), firebrat (Thysanura), tiny frog and spider. The sepsid were the most abundant dipteran community, followed by muscid such as Musca inferior, Stomoxys calcitrans and Musca crassirostris. Most muscid flies are bloodsuckers while the sepsid and other families were scavengers on the cow dung. We also bred the larvae found in the dung to the adult stage and subsequently identified as Allosepsis indica (Sepsidae) and Psychodidae. The above mentioned blood sucking flies seems to play an important role in zoonosis transmission. Further studies are important in investigating the biodiversity, bionomic and behavior of the disease vectors associated with cow dung for the future implementation of fly control measures in Malaysia.

S4.5 *In vitro* antibacterial activity of medicinal *Lucilia cuprina* larvae (Diptera: Calliphoridae) against selected pathogenic bacteria

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Maggot Debridement Therapy (MDT) is a type of biosurgery involving the intentional application of live, disinfected fly larvae into chronic non-healing wounds of human or animal to debride the necrotic wound. Many studies have demonstrated the potent antibacterial activity of Lucilia sericata larval excretions/secretions against bacteria, however, the antibacterial activity of the local strain of blowfly larva, Lucilia cuprina larval extract against bacteria has never been determined although MDT using L. cuprina was successfully conducted. The objective of this study is to determine the in vitro antibacterial activity of L. cuprina larval extract against selected pathogenic wound bacteria: Staphylococcus aureus, methicillin-resistant Staphylococcus aureus (MRSA), Staphylococcus epidermidis, Streptococcus pyogenes, Pseudomonas aeruginosa, Escherichia coli and Klebsiella pneumoniae. Larvae were sterilized using established procedures and homogenized. The larval homogenate was centrifuged and the methanol-containing supernatant was vacuum-concentrated to remove methanol. The end-product (larval extract) was kept at -70°C and re-suspended in 1ml sterile distilled water prior to use. Turbidometric (TB) and Colony-Forming Units (CFU) Assays were adopted to determine the in vitro antibacterial activity and properties (bactericidal and/or bacteriostatic) of larval extract against the seven selected bacteria. TB Assay has demonstrated significant growth inhibition of all bacteria tested. The CFU Assay further elucidated that the antibacterial activity of L. cuprina larval extract against S. aureus, MRSA, S. epidermidis, S. pyogenes was bacteriostatic but the effect was bactericidal against the Gram-negative bacteria of P. aeruginosa, E. coli and K. pneumoniae, particularly P. aeruginosa. We concluded that L. cuprina larval extract exhibited broad-spectrum antibacterial activity and was particularly potent against Gram-negative bacteria.

S4.6 Effect of protein on oogenesis of *Lucilia cuprina* (Wiedemann) and its utilization in maggot debridement therapy

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The main pre-requisite to practice maggot debridement therapy (MDT) is the ability to mass-produce sterile maggots. To achieve this, understanding of the reproduction of the flies is vital. Protein is essential for the oogenesis and development of ovaries of female flies. Egg production of female *Lucilia cuprina* (Wiedemann) is enhanced when female flies are fed with protein. The objective of this study was to investigate the effect of protein feeding on the oviposition pattern of *L. cuprina* and its relationship with the age of the flies. In each study, 20 females and 20 males were kept in a plastic container provided with granular sugar and water *ad libitum*. Protein (fresh cow liver, 5g) was introduced to newly emerged flies aged 0, 1, 2 to 7 days. Our study indicated that egg laying commenced after 4 days of feeding irrespective of the age of flies. Egg laying pattern was significantly correlated with protein feeding regardless of the age of flies. The outcome of this study is important as such information provides a better understanding of *L. cuprina* reproduction and therefore can be used successfully to predict fecundity timing of the flies. This is to ensure a constant and reliable supply of eggs to meet the increasing demand of the therapeutic maggots used in debridement of wounds.



Abstracts

Session 5 Control of Aedes Vectors

S5.1 Changing trends in measles and rubella incidence since inception of the measlesmumps-rubella immunization in Malaysia

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In 2002, the measles-mumps-rubella vaccine (MMR) was introduced in the Expanded Programme of Immunization and targeted to both boys and girls at 1 year and 7 years of age. The objectives of our study was to analyze the laboratory data available from 2004 till 2009 to examine the changing trends of measles and rubella cases in Malaysia since inception of the MMR. Samples for this study were received through the measles case based surveillance program or were hospital cases received for investigation of rash illness. Laboratory confirmed measles cases had dropped from 42.2% since 2004, when sporadic outbreaks were reported, to 3.9% in 2007, 2.2% in 2008 and 1.6% in 2009. Samples received for rubella had increased five fold from 365 in 2004 to 1522 in 2007. Positive rubella cases detected, increased from 4.1% in 2004 to 33.2% in 2007 and dropped to 19.2% in 2008 and 11.5% in 2009. The age group 11 to 20 years accounted for 73.6% of rubella cases confirmed in 2008, with a higher incidence among males than females. The measles elimination programme introduced by WHO in 2003 had contributed to significant progress in the control of measles. Rubella is not notifiable in Malaysia and the majority of rubella cases were detected through the integrated case-based surveillance for measles and rubella. This integrated surveillance with laboratory testing of all suspected cases with rash illness should be continued. However, to enhance the progress, specific targets should also be established in the national programme to eliminate rubella.

S5.2 Biochemical detection of resistance mechanisms in field-collected *Aedes* (*Stegomyia*) *aegypti* (L.) in Shah Alam, Selangor

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Biweekly ovitrap surveillance (OS) was conducted for a year (September 2007 – September 2008) in 2 different sites in a dengue endemic area in Shah Alam, Selangor. The field-collected *Aedes aegypti* from these 2 locations were colonized to F3 adults and subjected to WHO standard bioassay. The monthly insecticide susceptibility status was analyzed for 1 year. Populations with mortality rates below 80% in the bioassays were categorized as resistant. The field populations were susceptible to malathion (5%) and fenitrothion (1%). Resistance to propoxur (0.1%), bendiocarb (0.1%) and permethrin (0.75%) was detected in most months, while DDT resistance was detected throughout the year. Biochemical resistance enzyme assays were conducted to define the mechanisms involved in the resistance. Four enzymes, namely non-specific esterases (NSE), monooxgenases (MO), insensitive acetylcholinesterase (AChE) and glutathione-S-transferases (GST) were studied. High enzyme activities of both MO and NSE were correlated with the low mortalities in permethrin resistance (r>0.5; p<0.05). The application of permethrin for dengue control by the local authority in this area has resulted in resistance to this insecticide. However, there was no correlation of GST activity with DDT resistance (r<0.5; p>0.05). Increased enzyme levels of MO and NSE involved in cross-resistance between permethrin and DDT was detected. This study has provided baseline of insecticide susceptibility and the mechanism involved in *Ae. aegypti* from the locality. The data showed that there is a need to review current insecticide(s) used in dengue control programme and to implement resistance management strategy.

S5.4

S5.3 Ovicidal effects of *Bacillus thuringiensis israelensis* on *Aedes aegypti* (L.)

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Ovicidal effect of *Bacillus thuringiensis israelensis* (Bti), a biolarvicide was tested on *Aedes aegypti* eggs. A commercial Bti formulation (VectoBac® WG) was evaluated at various concentrations that are currently used in field operation. A Bti technical grade powder (VBC strain AM 65-52) was also included in this study. Blood- fed gravid females were allowed to oviposit on Bti treated and clean waters. Five replicates for each concentration were conducted. Eggs were collected and subjected to force hatching under vacuum. Three hundred eggs from each replicate were then observed under an imaging system to assess the hatching rate. Egg with intact operculum was considered as unhatched egg. Hatching rate was significantly reduced in eggs oviposited on Bti treated waters in comparison to untreated waters (p<0.05). The reduction rate was similar for both test samples, indicating that the ovicidal effect was due to Bti active component(s) and not by the inert substances in the commercial Bti formulation. Several of the unhatched eggs were dissected and dead embryos were observed. In another related study it was observed that Bti had no effect on eggs that were previously oviposited on Bti-free environment and had gone through the drying process. Thus, this will be the first report of Bti exhibiting ovicidal effects on *Ae. aegypti* eggs, confirming that Bti exerts additional effects on mosquito population.

Development of permethrin resistance in several strains of Aedes aegypti (Linnaeus)

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Aedes aegypti strains (laboratory, permethrin-selected and field-collected) were subjected to three standard laboratory testings: WHO larval bioassay; WHO adult mosquito bioassay; and mixed function oxidases (MFOs) enzyme microassay for detection of permethrin resistance among these colonies. The LC_{50} values among all field strains obtained from the WHO larval bioassay did not differ significantly. The highest LC_{50} value was from the Taman Melati field strain (0.39 mg/L). The resistance ratio for the permethrin-selected strain and the field strains of *Ae. aegypti* tested ranged between 1.86 – 5.57 folds. Exposure to piperonyl butoxide (PBO) prior to permethrin in WHO adult bioassay had reduced the LT_{50} values and affected the MFOs activities of tested mosquitoes as indicated by low mean optical density of oxidase activity (0.28 – 0.42) at 630 nm. The LC_{50} values or LT_{50} values and the oxidases level in these mosquitoes were highly correlated (r = 0.825; p < 0.05). This study indicated the existence of permethrin resistance in these mosquito strains. However, PBO was still effective in countering the resistance indicating the absence of knockdown resistance (kdr).

S5.5 Effects of temperature stress on development of *Aedes aegypti* (L.) and *Aedes albopictus* Skuse

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Aedes aegypti and Aedes albopictus are the mosquito vectors of dengue virus. Climatic factors such as temperature have strongly influenced the ecology, development and behavior of mosquitoes as well as the transmission dynamics of the diseases they transmit. Thus this study is to investigate the effects of temperature stress on development of these vectors. The development of *Ae. aegypti* and *Ae. albopictus* under 6 different temperatures (30°C, 32°C, 34°C, 36°C, 38°C and 40°C) was studied. The development time, survival, mortality and emergence rate was observed from day 1 to day 21. For each treatment, every stage of mosquitoes was separated according to the days of development. All data about egg hatching, survival, mortality, exuviated, death and ratio of male to female were recorded. Wing length was measured as a proportion of body size. Results showed that temperature stress influenced all the stages of *Aedes* mosquitoes. The eggs hatched faster at 40°C compared to at 30°C. At 40°C, the 4th instar larvae only need 1 day to pupate. Adults emerged faster when compared to mosquitoes exposed to 30°C. The egg development in gravid female was also faster. Female and male wing lengths decreased as temperature increased. We found that temperatures had significant effect on hatching and emergence time. Significant differences were observed between temperatures and wing length, body size and morphological characters.

S5.6 Simulated field performance of Spinosad DT against *Aedes aegypti* in Penang, Malaysia

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The efficacy and residual effect of Spinosad [DT (ready to use tablet)] against *Aedes aegypti* in earthen jars was conducted at the medium scale field trial in Penang, Malaysia. Two regimes were applied: full and ½ emptied and refill at each assessment interval and new food (0.5 g/jar) added. The efficacy and residual evaluation were conducted for a period of 16 weeks. The concentrations of Spinosad DT applied were 1.0 mg/l, 0.5 mg/l, 0.25 mg/l and control. Spinosad DT gave excellent larvicidal activity with more than 90% mortality at 72 hours and 100% Inhibition of adult emergence for both regime in earthen jars. For Spinosad DT tested against *Ae. aegypti* in plastic container, for non-replinish regime it gave excellent larvicidal activity with more than 90% mortality at 72 hours and 100% Inhibition of adult emergence up to 16 week while for half emptied and refilled regime it gave excellent larvicidal activity with more than 90% mortality at 72 hours and 100% Inhibition of adult emergence up to 16 week while for half emptied and refilled regime it gave excellent larvicidal activity with more than 90% mortality at 72 hours and 100% Inhibition of adult emergence up to 16 week while for half emptied and refilled regime it gave excellent larvicidal activity with more than 90% mortality at 72 hours and 100% Inhibition of adult emergence up to 16 weeks.

S5.7 Biological efficacy of natural Juvenile hormone III (JH III) from a weed plant, *Cyperus aromaticus*, against *Aedes aegypti*

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Cyperus aromaticus contains high levels of juvenile hormone III (JH III). Cell suspension cultures of the plant are able to produce JH III. Laboratory bioassays were carried out to evaluate the biological efficacy of chloroform extract of three lines of *C. aromaticus* cell suspension cultures produced by *in vitro* culture system (P4, ML, Z1) against the larvae of *Aedes aegypti*. The P4 line was the most effective compound against *Ae. aegypti*. Results indicated that inhibition of emergency values at the 50 and 90 percent level ($IE_{50} \& IE_{90}$) against *Ae. aegypti* were 61.22 and 171.52 ppm, respectively. However the ML line resulted in the lowest inhibition of emergence of *Ae. aegypti* with IE_{50} and IE_{90} values of 75.90 and 241.32 ppm, respectively. Z1 line induce 50 and 90 percent inhibition of adult emergence in *Ae. aegypti* at a concentrations of 65.02 and 184.35 ppm, respectively. These compounds may prove to be valuable insect growth regulators for control of mosquitoes to decrease the frequency of pathogen transmission to human. The use of juvenile hormone III (JH III) produced via *in vitro* culture system should be an effective, inexpensive and environmentally safe alternative for the management of vector mosquitoes.

S5.8 Sub-lethal dose of diflubenzuron and susceptibility status of *Aedes albopictus* and *Aedes aegypti* towards diflubenzuron in Penang island

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Laboratory-cultured *Aedes albopictus* and *Aedes aegypti* larvae were exposed to diflubenzuron at different concentrations to determine the EI_{50} and EI_{99} values. The EI_{50} of *Ae. albopictus* and *Ae. aegypti* were 0.00075 ppm and 0.00067 ppm, respectively whereas the EI_{99} were 0.00193 ppm and 0.00214 ppm respectively. In addition, field collected mosquitoes from different locations on Penang Island were exposed to diflubenzuron to determine their susceptibility status. Field collected *Ae. albopictus* and *Ae. aegypti* showed no resistance toward diflubenzuron solution. The longevity and fecundity of adults which survived exposure to a sub-lethal dose were studied. It was found that diflubenzuron did not influence the longevity and fecundity of *Ae. albopictus* and *Ae. aegypti*. Diflubenzuron also did not influence the hatchability of the eggs and the development of larvae produced by mosquitoes exposed to a sub-lethal dose of the insect growth regulator.

S5.9 Effects of atmospheric temperature on susceptibility of *Aedes (Stegomyia) aegypti* to vaporized acetone

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Atmospheric temperature is an important stimulus for mosquitoes in detecting host and its thermoreceptors exhibit maximum response at 25 to 28°C of atmospheric temperature with capability to a change of temperature as low as 0.05°C. Thus the study attempts to investigate whether mosquito susceptibility to acetone would vary at different atmospheric temperature. A female *Aedes (Stegomyia) aegypti* sugar-fed mosquito of age 2 to 8 days was confined in an airtight glass tube (15 mL) placed inside an environmental chamber with temperature conditioned at 25, 20, 15 or 10°C (± 2°C). Acetone was vaporized in the glass tube within the environmental chamber at an approximate dose of 500 mL/m³. Susceptibility of mosquito to vaporized acetone was based on knockdown behavior exhibited by the mosquito. The knockdown activity was observed immediately after the treatment of acetone up to a period of 10 minutes. Mean time of mosquito knockdown at 25, 20, 15 or 10°C from a 25 replicates of test were 122.6, 135.7, 148.2 and 136.7 seconds, respectively. It was believed that reduced temperature influenced the acetone vaporization, subsequently delaying knockdown time. But low temperature (10°C and below) also affected mosquito physiology, causing it to knockdown faster. Therefore atmospheric temperature does effects sensory response of *Aedes* mosquitoes to vaporized acetone.

S5.10 Simulated field performance of Diflubenzuron against *Aedes aegypti* in Penang, in Malaysia

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The efficacy and residual effect of Diflubenzuron [DT (ready to use tablet)] against *Aedes aegypti* in earthen jars and plastic containers were conducted at the medium scale field trial in Penang, Malaysia. Earthen jars and plastic containers with water holding capacity of 50 L were used in this study. For the earthen jars, the size measures 40 cm high with a rim diameter of 48 cm at the top, tapering to a diameter of 30 cm at the base, whereas the plastic containers measure 45 cm high with a rim diameter of 44 cm at the top, and a diameter of 27 cm at the base. For both earthen jars and plastic containers, a volume of 50 liters of water were used in each container and the water level was marked off (about 5 cm from the top). The efficacy and residual evaluation were conducted for a period of 16 weeks. The concentrations of Diflubenzuron DT applied were 0.4 mg a.i./l, 0.08 mg a.i./l, and control. A week after treatment, assessment of the earthen jars showed that Diflubenzuron DT at both 0.08 and 0.4 mg a.i./L were effective against *Aedes aegypti* up to week 16 with more than 95% larval mortality, whereas more than 75% mortality was recorded two days after treatment, it showed that Diflubenzuron DT at both 0.08 and 0.4 mg a.i./L were effective against *Aedes aegypti* larval mortality, whereas more than 75% mortality are effective against *Aedes aegypti* up to week 16 with more than 95% larval mortality, whereas more than 75% mortality up to week after treatment, it showed that Diflubenzuron DT at both 0.08 and 0.4 mg a.i./L were effective against *Ae. aegypti* up to week 16 with more than 95% larval mortality, whereas more than 58% mortality was recorded two days after treatment throughout the 16 weeks more than 58% mortality was recorded two days after treatment.

S5.11 Efficacy of aqueous extracts of *Areca catechu* against larvae of *Aedes (Stegomyia)* aegypti

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Betel nuts (*Areca catechu*) contains arecoline, which mimics acetylcholine, an important mosquito neurotransmitter. Though organic solvent-extraction was reported to yield better amount of arecoline from betel nuts, aqueous-extraction was selected for its environmental friendly processes. Betel fruits were segregated to four groups based on its colour to reflect its level of maturity; young (green), semi-mature (green-yellow), mature (yellow-orange) and over-mature (brown). Presence of arecoline in all four groups was confirmed using HPLC system with photodiode array detector. Relatively, over-mature and semi-mature betel nuts contained more arecoline compared to matured and young nuts. Aqueous extraction followed by freeze drying yielded reddish brown powder of extracts. *Aedes (stegomyia) aegypti* larvae were released into glass containers filled with solutions of betel extracts in distilled water at a selected dose of 50, 200, 1000, 2000, 5000 and 12500 ppm. The numbers of larvae that died at 24th hours of post treatment were recorded. The solutions of young, semi-mature, mature and over-mature betel extracts at the dose of 5000 ppm resulted in 92, 76, 36 and 80% of larval death, respectively. The findings suggested presence of other larvicidal actives besides arecoline in the betel extracts.

S5.12 Bionomic and susceptibility status of *Aedes* mosquito on selected insecticides in USM campus, Penang, Malaysia

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A research was conducted to study the bionomic and susceptibility status of *Aedes* mosquito in the campus of USM, Penang, Malaysia. Three study sites were selected which included Durian Valley, Desasiswa Aman and the Sports Complex, USM. Mosquitoes were sampled using the ovitrap and the result showed that there were three species of *Aedes* mosquito in USM campus, *Aedes albopictus, Aedes aegypti* and *Aedes niveus* subgroup. *Aedes albopictus* was the most abundant mosquito in USM campus. The data collected showed that there were no correlation between the eggs collected from the campus and the abiotic factors including mean temperature, total rain and relative humidity. A 24 hours bare leg catch was performed in Durian Valley to study the biting cycle of the *Aedes* species. It was found that *Aedes* mosquito catch. A bioassay test was conducted on the *Aedes* mosquito collected from the USM campus against the diagnostic dose of malathion and permethrin. The result showed that almost all *Aedes* mosquito in the three study sites had high possibility of developing resistance towards malathion 5.0% and permethrin 0.75%. Only *Ae. aegypti* from Durian Valley showed no resistance towards permethrin 0.75%. The KT₅₀ and KT₉₅ of permethrin 0.75% were lower than malathion 5.0%.

S5.13 Knockdown effect of lemon (*Citrus limon*) peel extract as an insecticide towards *Culex* sp. mosquitoes with spraying method

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Culex sp. mosquitoes are biological vectors of filariasis, chikungunya and encephalitis. One of the methods used to control the growth and development of these mosquitoes is by using insecticide against them. Although syntethic insecticides is effective it could however cause adverse effects to the environment and human's health. Organic insecticides is considered to be the alternative way due to its safety for the environment because its compound can breakdown easily. Lemon peel (*Citrus limon*) containing *limonene* which is proven effective as insecticide against *Culex* sp. as it is shown to have knockdown effect towards adult *Culex* sp. mosquitos. The purpose of this reasearch is to determine the knockdown effect of lemon peel extract as an insecticide towards *Culex* sp. mosquitoes. This study was experimental laboratoric with *true experimental-post test only control group design*. There were 5 different treatments, using malathion 0,28% as positive control, aseton 1% as negative control, and lemon peel extract in 1,25%, 2,5%, and 5%. This research used 25 *Culex* sp. as samples. Treatments were observed every 0, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60 minutes. The result of this research is that the lemon peel extract 5% has the fastest KT50 (less than 5 minutes). And then, from the result of the KT50, we can know the efectivity of lemon peel extract using insecticide score. The conclusion of this experiment is that lemon peel extract at 2,5% and 5% has knockdown effect as an insecticide towards adult *Culex* sp. mosquitoes.

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Abstracts

Session 6 Veterinary Parasitology

S6.1 Molecular phylogeny and bio-geography of food-borne zoonotic helminths

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Food-borne parasitic zoonoses (FBPZ) are rather neglected because of their localized distribution due to the traditional eating habits. However, recently FBPZ have become a global public health issue, because they are spreading all over the world by international traveling and immigration of humans, and also by global trading of raw food materials. FBPZ are not familiar to the physicians of non-endemic countries. Ignorance or unawareness of FBPZ often results in misdiagnosis or inappropriate treatment. Many FBPZ pathogens have species- or intra-species variations depending on their geographic distribution and such variants often show variable pathogenicity. Along with the popularization of molecular genetic methods in medical sciences, identification of FBPZ or any other pathogens has gradually shifted from morphology to molecular method. Incongruence between morphological and molecular speciation or even within molecular speciation by different genetic markers urged us to explore new insights on phylogeny and geo-biology of FBPZ. One such example is the genus Paragonimus. Based on morphology, this genus was considered to consist of about 50 species. However, molecular phylogenetic analyses of ITS2 and CO1 genes revealed that this genus would be classified into 5 clades of species complex; namely Paragonimus westermani and Paragonimus skrjabini species complex. Each species complex is composed of several genetically distinct subspecies with variable pathogenicity and geographical distribution. In case of Fasciola, Fasciola hepatica and Fasciola gigantica are easily distinguished by morphology, but they are genetically proven to be very close and even their natural F1 hybrid was discovered. Similar story can be applied for Taenia saginata and Taenia asiatica. For the genus Gnathostoma, each known species are genetically homogenous, without obvious variations. However, by molecular phylogenetic analyses, surprisingly the Latin American species fall into the middle of the Asian species clades. Angiostrongylus cantonensis seemed to be divided into several subspecies by molecular data. In such molecular era, genetic analysis of Malaysian unique species or isolates would provide valuable information as the missing ring of evolution to construct more accurate phylogenetic trees of those parasite species.

S6.2 *In vitro* validation of anthelmintic activity of *Butea monosperma* and *Calotropis* procera

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Development of resistance in parasites to several families of drenches and chemical residues and toxicity problems has posed a threat in effective chemotherapeutic parasite control programme. These considerations have revived interest in exploiting the potential of medicinal plants for their use as anthelmintics. This paper validates use of *Butea monosperma* bark and *Calotropis procera* stem and bark as anthelmintics in the traditional medicine in Pakistan. *In vitro* evaluation for anthelmintic activity of crude methanol extracts (CME) of *B. monosperma* and *C. procera* was carried out using egg hatch test (EHT), larval development assay (LDA) and adult motility assay (AMA) using *Haemonchus contortus* eggs and worms. Both *B. monosperma* and *C. procera* exhibited broad range of effects as they were effective in all the three *in vitro* tests. In EHT, LC₅₀ of *B. monosperma* and *C. procera* was 239.88 and 316.23 µg/mL; whereas the respective LC₅₀ values for LDA were 100 and 141.25 µg/mL. In AMA, all the *H. contortus* worms were found dead at 3 and 12 hours post-exposure to CME of *B. monosperma* and *C. procera* at 8000 µg/mL, respectively. Both *C. procera* and *B. monosperma* exhibited dose-dependent effects in all the three tests as evident from R2 values being 0.9767 and 0.8552 for EHT and 0.9831 and 0.9805 for LDA, respectively and increasing mortality of worms with increasing doses of plant extracts. The plants validated for their anthelmintic activity may be subjected to further scientific studies for drug development.

S6.3

Validation of FAMACHA eye score system in goat

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The FAMACHA[®] system was developed in South Africa to classify animals into categories based upon the level of anemia. The ocular mucous membrane of sheep and goats are classified by comparison with a laminated color chart bearing pictures of conjunctiva classified into five categories ranging from normal red through pink to practically white in severe anemia. FAMACHA[®] has been extensively tested in South Africa and United States. To be used in Malaysia, it is important that the system be tested. The objective of the current study was to validate FAMACHA[®] in correlation with Packed Cell Volume (PCV) and Fecal Egg Count (FEC). A total of 230 goats from six farms in Terengganu were chosen for the study. Eye color based on FAMACHA[®] grading, blood and fecal samples were taken and subjected to PCV and FEC respectively. Two separate FAMACHA[®] scores defined as anemia were ≥ 3 and ≥ 4 . The correlation between PCV and FAMACHA[®] eye score, and PCV and FEC were highly significant (P< 0.01), but the correlation between FEC and FAMACHA[®] eye score were not significant. Sensitivity was 100% when FAMACHA[®] scores of 3 and above were considered as anemic, but specificity was low (15.89%). Specificity increased (63.08%) when FAMACHA[®] scores of 4 and above were considered as anemic, concurrently sensitivity was lowered (81.25%). The percentage of false negative (anemic animals but not identified by FAMACHA[®]) cut off points. The data obtained strongly suggest that FAMACHA[®] method is a valuable diagnostic tool for identifying anemic goats.

S6.4 Oxyspiruriasis in zoo birds - treatment and control

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Oxyspirura mansoni (Cobbold, 1879) Ransom, 1904 is a widespread nematode parasite occurring under the nictitating membrane of birds. Zoo birds off-feed, sick or found dead were examined for the parasite. Oxyspiruriasis caused by this nematode was diagnosed in three species of pheasants: 3 *Chrysolophus pictus* (golden pheasant), 7 *Lophura nycthemera* (silver pheasant) and 9 *Phasianus colchicus* (common pheasant) in Zoo Negara Malaysia are reported here. Treatment and control measures, against the intermediate host, *Phcnoscelus surinamensis* (Surinam cockroach) are outlined. The golden pheasant is a new host for *O.mansoni* in peninsular Malaysia.

S6.5 Movements and home range of a common species of tree-shrew, *Tupaia glis* surrounding houses of otocariasis cases in Kuantan, Pahang, Malaysia

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This is the first radio telemetry study in Malaysia to monitor movements of a common species of tree-shrew, Tupaia glis (with ticks on their body) surrounding houses of otoacariasis patients in Felda Bukit Goh, Kuantan, Pahang. The area was chosen because of its high number of new and repeat otoacariasis cases, and suspected factors contributing to the cases. The objective of this study is to document their activities and movement patterns, home range, nesting behaviour and social organization. These are the basic vital information to understand behaviour of T. glis before any effort to control them and their habitat. In this study, 5 treeshrews (3 males and 2 females) were monitored. Each tree-shrew was fitted with a transmitter chip radio collar which operates between the frequencies 154.13 MHz to 154.21 MHz. Each transmitter was tracked with a Portable Telemetry Receiver (Sirtrack, New Zealand) and fitted with a 3-element Yagi antenna. Collared tree-shrews were located using standard methods of ground based triangulation. Each location was taken from at least 2 directional fixes and a minimum of 3 compass bearings. Fixes were taken hourly for each collared individual from the time of emergence from nest (beginning of activity) till time of entry into the nest (end of activity) every day for 5 to 7 continuous days. Three series of radio telemetry observations were carried out i.e from 14-19 April; 27 April – 4 May and 2–7 June 2008. The bearings, time and positions of observer were recorded and later plotted on a graph paper in order to derive coordinates of the collared animal. These coordinates were then analysed using Ecological Software Solutions (Biotas Version 1.03). Besides jack fruit tree and long bushes, nests shared by 2 shrews of different sex were found in 2 houses. They were suspected to be mating couples. All tree-shrews emerged from and returned to their nests at 0700 hours and 2000 hours, respectively. Both the time of exit and entrance into nest were the same between sexes (p>0.05). Their average total active period was 4.9 to 7.0 hours with a total daily travel distant of 270 m to 382 m. A male and female tree-shrew can move as far as 3285 m and 4591 m, respectively. Active movements of T. glis were during daytime. In this study, T. glis was observed to spend whole of their total active periods moving around their ranges i.e from trees in fruit orchards or from a nearby secondary forest to trees in compound of 4 to 7 houses and vice versa. They were regular 'visitors' of the houses day and night except for one individual which visited during daytime only. Home range and core area size for the tree-shrews were 2.0-3.4 ha and 0.05-0.42 ha, respectively. Generally, the mean home range size of females was 20.8% larger than that of males. Females covered a 15.4% slightly higher daily movement range compared to male tree-shrews. At the end of study, only 2 radio-collared tree-shrews were detected. One was captured alive and the other one was found dead. Status of the others was unknown.

S6.6

Severe anthelmintic resistance in commercial small ruminant farms in Perak, Malaysia

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Worm infection is one of the most prominent causes of mortality and morbidity in Malaysia. It causes financial losses to the livestock industry. This is mainly due to grazing activities of livestock on pasture contaminated with third stage infective larvae of parasitic nematodes, which perpetuates the infection. A study of the anthelmintic resistance status on four commercial sheep and goat farms in Perak was conducted. The Faecal Egg Count Reduction Test (FECRT) is critical for evaluating the resistance status of each farm as well as worm species that exists in the farm so that further action can be taken to manage the problem. The four drug groups tested in this study are Ivermectin, Oxfendazole, Levamisole and Closantel. Faecal samples were subjected to the standard procedures of McMaster for worm egg estimation and also larval culture for third stage larvae identification. Results of Farm 1 showed resistance to the 3 groups of anthelminitic except Ivermectin and the worm population of this farm was predominantly *Trichostrongylus* sp.(74%). The results of Farm 2 showed resistance to all 4 groups of anthelmintic that were tested and the major worm population of this farm was *Haemonchus contortus* (79%). The results of Farm 3 and Farm 4 showed resistance to 3 groups of anthelmintic except Closantel and Ivermectin respectively. However, the worm populations for the two farms were *H. contortus* (58%) and *Trichostrongylus* sp (54%) respectively. These farms have used the four anthelmintic groups frequently over the past few years, thus resulting in the severe case of anthelmintic resistance. Recommendations have been made to control helminths using alternative approaches such as cut and carry feeding, herbal medication like Neem (*Azadirachta indica*) leaves and rotational grazing as well as improvement in management of animals to increase their immunity.

S6.8

S6.7 Diagnosis of leptospirosis, brucellosis and melioidosis disease in humans conducted in VRI

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The Veterinary Research Institute diagnoses diseases such as leptospirosis (caused by *Leptospira* sp.), melioidosis (caused by *Burkhorlderia pseudomallei*), and brucellosis (*Brucella abortus*) in humans as requested by the Health Department, Malaysia. In 2009, a total of 71 samples were received by the Serology Unit requesting for diagnosis of these diseases. A total of 53 samples were tested for leptospirosis, one for melioidosis and 17 for brucellosis. All samples were human sera collected by the Ipoh General Hospital from patients suspected to harbour these diseases. Results indicate 7 samples were positive for leptospirosis using microscopic agglutination test (MAT). All samples submitted for melioidosis were negative using Complement fixation test (CFT). For brucellosis, the tests conducted were *B. abortus* CFT, Brucella Coomb's Test, Brucella Rose Bengal Plate Test (RBPT), Brucellosis Serum Agglutination Test (SAT) and all were negative. The highest number of samples received was in April and October which is 14 samples and 16 samples respectively and most of these samples were tested for leptospirosis. These diseases are zoonotic and diagnosis will aid in early detection and treatment of the diseases in the human population. Subsequently, samples from animals especially livestock are also tested to help eradicate the diseases.

The use of some common Malaysian herbs for worm control in goats

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Helminthiasis caused by strongyles is an important constraint to small ruminant farming in Malaysia. With the advent of anthelmintic resistance, there is increased interest to look for alternative worm control options such as using herbal remedies. An *in vivo* study was conducted at a government goat farm to assess the anthelmintic effects of some common herbal plants such as neem leaves (*Azadirachta indica*), noni leaves (*Morinda citrifolia* Linn) and bitter gourd fruit (*Momordica subangulata* Blume). Seven groups of 5 adult Boer goats each were fed with 100g of neem, noni leaves, bitter gourd fruits individually pureed as well as a combination of the leaves and fruit, two times per week. In addition, all goats received fixed amounts of molasses (100gm), effective microorganisms (EM)(4ml) and ad lib water. The animals had semi intensive management with grazing and cut grass feeding in the stalls. The animals that had high feacal egg counts were monitored over a period of 1 month and results indicate a drop in the faecal egg count in animals fed with neem leaves (70%) and bitter gourd fruit (94%). No significant differences in faecal egg counts were observed in the animals fed with noni leaves or combination of herbal plants. One of the reasons for this could be due to the low dosage of the herbal plant fed to the animals. This preliminary work paves the way for more detailed study of common herbs which can be used for worm control in small ruminants. This information is useful to encourage farmers to go towards green farming with less dependence on drugs for worm control.

Abstracts

Session 7 Students Competition Presentation

S7.1 Filariasis in Kuala Lumpur and Selangor: entomological, parasitological and molecular studies

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The National Programme strategy to eliminate filariasis in Malaysia was implemented with the aim of ensuring that Malaysia is free from this disease by 2012. The present study provides new information based on a surveillance study regarding the current status of filariasis, focusing on areas within Kuala Lumpur and Selangor. Five sites were chosen in this study. Two of these sites were reported to have cases of human filariasis due to *Brugia pahangi*, a common parasite of cats and dogs. *Armigeres subalbatus* mosquito was found to be the vector. The infection and infective rates of the mosquitoes with nematode larvae in three of the study areas were 6% and 3% respectively. The larvae were confirmed to be *B. pahangi* by both parasitological and molecular examinations. Blood samples were taken from possible reservoir hosts such as cats and monkeys, and were examined for the presence of microfilariae. Cat was identified as a potential reservoir host as it was found to be heavily infected with *B. pahangi* within the same areas where infected *Ar. subalbatus* was found and cases of human filariasis were reported. Another species, *Dirofilaria repens* was also found in one study area. Thus, from these results, zoonotic *B. pahangi* infection is a real possibility, especially in areas where human and animal reservoir populations are living closely along with the presence of the mosquito vector.

S7.2 High prevalence of *Blastocystis* sp. subtype 4 among rural communities in Nepal

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Blastocystis sp. has been reported to be the most prevalent organism found in any stool survey. Though two studies carried out in Nepal showed Blastocystis sp. infection in various groups, the organism has not gained sufficient attention to warrant an inclusion in the periodic stool surveys carried out in Nepal. Therefore, the prevalence and molecular characterization of Blastocystis sp. in rural communities in Nepal was studied. The presence of Blastocystis sp. was examined in faecal samples collected from 241 individuals from two rural villages (Bahunipati in Sindhupalchowk and Bolde Phediche in Kavrepalanchowk), Nepal. Faecal samples were cultured in Jones' medium supplemented with 10% horse serum followed by incubation at 37°C and examined daily for vacuolar Blastocystis sp. using light microscope for the next three days consecutively. Faecal samples that were positive for Blastocystis sp. by in vitro cultivation were further genotyped using sequenced-tagged site (STS) primerpolymerase chain reaction (PCR). Prevalence for Blastocystis sp. infection was 26.1% and was significantly (p=0.027) higher in Bahunipati (36.4%) than Bolde Phediche (22.3%). Blastocystis sp. subtype 4 (84.1%) was the predominant genotype followed by subtype 1 (63.5%). Prevalence of mixed subtypes Blastocystis sp. infection was high (65.1%). This might be due to poor sanitary facilities and consumption of food or water contaminated with Blastocystis sp. cysts. 81.0% and 47.6% of the Blastocystis sp. infected individuals were found to consume untreated drinking water and had frequent contact with animals respectively. Hence, this is the first to report the molecular characterization of Blastocystis sp. in rural communities in Nepal which suggest its potential human-to-human, waterborne and zoonotic transmissions. Therefore, an intervention strategy should be taken to drive public health and hand hygiene awareness among the villagers.

S7.3 Effects of anthelmintic plant; *Terminalia catappa* towards nutritional and physiological aspects on Sprague-Dawley white rats

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A range of local plants has been chosen as alternative anthelmintic. Based on ethnoveterinary medicine, *Terminalia catappa* is believed to have the potential for worm control in goats. In this study, Sprague-dawley white rats were used as lab animal. Crude extract of *T. catappa* were daily administered to the rats for fourteen days to investigate the effects of this plant towards nutritional behavior and any physiological changes on the animals. The quantity of feed and water that was consumed by the rats were recorded on day-1, day-7 and day-14, while daily clinical observation was conducted two times per day during the treatment period. As a result, the nutritional behavior is normal, which is determined by the rate of animal's diet that was significant due to the increment of the animal's body weight. From the daily clinical observation, there were no serious physiological changes occurred in the animals. In conclusion, this potential anthelmintic plant does not affect the nutritional behavior and also the physiological aspect of the animals.

S7.4 New predictive tools for pre-emptive dengue vector control in north Queensland, Australia

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Vector forecast (i.e. the act of predicting the fluctuations of vector abundance) is rarely being used to guide control operations of dengue vectors. In this study, the relationship between meteorological data and female *Aedes aegypti* collection data from BG Sentinel mosquito traps placed in eleven monitoring sites in Cairns, north Queensland, Australia was characterised using multiple linear regression. Longer- and shorter-term factor models reveal daytime temperature and relative humidity as the most significant weather factors to be associated with adult vector abundance. A considerable regression coefficient ($R^2 = 0.61$) and validation of the shorter-term factor model have illustrated the potential of simple meteorological-based modeling to be used as a predictive tool for dengue vector abundance. Such a tool would enable timely vector control measures to be executed prior to the occurrence of dengue outbreak. The influences of non-meteorological factors and mosquito collection techniques on vector abundance data are also discussed.

S7.5 Does *Blastocystis hominis* exacerbate the growth of colorectal cancer cells?

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Blastocystis hominis is one of the most common intestinal protozoan parasites in humans. Numerous studies have shown that blastocystosis is coupled with intestinal disorders. Previously, researchers have used *B. hominis* culture filtrates to investigate its ability in triggering inflammatory cytokine responses and gene transcription factors in human colonic epithelial cells. Past studies reported that parasitic infections initiate carcinogenesis through inflammatory processes. Therefore, the chances of *B. hominis* infection in facilitating cancer growth should not be ruled out. However such investigations are still lacking. Thus, it is important to further examine on cytophatic effect, cellular immunomodulation and apoptotic responses of *B. hominis* particularly in malignancy. In present study, we assessed the effect of solubilized antigen from *B. hominis* on cell viability of peripheral blood mononuclear cells (PBMCs) and human colorectal carcinoma cells (HCT116). The effect of the antigen on gene expressions of cytokines namely interleukin-6 (IL-6), interleukin-8 (IL-8), tumour necrosis factor alpha (TNF- α), interferon gamma (IFN- γ), nuclear factor kappa-light-chain-enhancer of activated B cells (NFkB, a gene transcription factor) and pro-apoptotic genes namely protein 53 (p53) and cathepsin B (CTSB) of PBMCs and HCT116 were also studied. Results obtained favours the fact that antigen from *B. hominis*, at a certain concentration, could facilitate the proliferation of HCT116 while having the ability to down-regulate immune cell responses (PBMCs). Therefore, it is essential to screen colorectal cancer patients for *B. hominis* infection as it possesses the ability to increase the tumour growth.

S7.6 Intestinal parasitic infections among children in Albania: current status and risk factors

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Intestinal parasitic infections among children remain a global issue. There is lack of information regarding common intestinal infections among children in Albania. Therefore, a cross-sectional study was carried out involving stool samples collected from 321 children of two main Albanian districts, Tirana (152) and Elbasan (169), comprising of 143 males and 178 females, aged <6 months – 15 years old, with 168 clinical-based and 153 community-based. Pre-tested standard questionnaire was used to gather personal and clinical data. Stool samples were examined for ova and (oo)cysts of all parasites using formalinether concentration method and permanent stains. The overall prevalence of intestinal parasites was 61/321 (19%). The most common were protozoa (11.5%), followed by soil-transmitted helminthes (STH) (8%). *Giardia duodenalis* was the most common parasite (10.9%), followed by hookworm (5.6%), *Ascaris lumbricoides* (1.9%), *Trichuris trichiura* (0.6%), *Cryptosporidium* (0.3%) and *Entamoeba histolytica/dispar* (0.3%). Univariate analysis showed that Tirana district (12.5%) had higher prevalence of STH compared to Elbasan district (4.1%) (OR=3, 95% CI (1.30 – 6.97), p<0.05). Higher prevalence of STH was also noted in community-based (10.5%) compared to clinical-based (6.0%). Diarrhoea was five fold higher among children infected with STH compared to those not infected (OR=5.5, 95% CI (1.63 – 18.20), p<0.005). Abdominal pain (OR=3.3, 95% CI (0.90 – 12.45), p = 0.054) and diminished appetite (OR = 4.95%, 95% CI (1.18 – 13.41), p<0.05) were other clinical manifestations among those acquiring helminthes.

S7.7 Polytene chromosome of the malaria vector *Anopheles arabiensis* Patton in Sudan

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Anopheles arabiensis is the most wide spread and the main malaria vector in Sudan. A study was carried out to develop a cytogenetic map of the vector mosquito using its salivary gland and ovarian nurse cells polytene chromosomes. The cytogenetic map is important for characterizing the various chromosomal inversion polymorphisms, deletion, puffs and ectopic pairing in *An. arabiensis* collected from three different sites in Sudan. The detailed cytogenetic map of *An. arabiensis* revealed distinct chromosomal inversions especially paracentric inversions in many of the specimens collected, reflecting an inversion polymorphism in natural population of the vector mosquito.

S7.8 Prevalence and risk factors of protozoal infections among patients attending hospitals in Sana'a City, Yemen

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Intestinal protozoal infections are public health problems in developing countries including Yemen. The aim of this study was to determine prevalence and risk factors of protozoal infections (Giardia duodenalis, Entamoeba histolytica/dispar and Cryptosporidium) among patients attending hospitals in Sana'a, the capital of Yemen. Stool samples were collected from 503 patients aged between 1 and 80 years old, 219 were males and 284 females. Biodata were collected via pretested standard questionnaire. Faecal samples were processed and examined for (oo)cysts using wet preparation after formalether concentration techniques. Cryptosporidium oocysts were detected using Ziehl-Neelsen staining technique. The overall prevalence of intestinal protozoal infection was 30.9%. The infection rates of G. duodenalis, E. histolytica/dispar and Cryptosporidium were 17.7%, 17.1% and 1%, respectively. Univariate analysis identified eight predictors of intestinal protozoal infections which include living in rural areas (OR = 1.518, 95% Cl 0.999-2.305, p < 0.05), rearing animals (OR = 1.748, 95% CI 1.168-2.617, p < 0.05), not washing hands (OR = 1.466, 95% CI 0.991-2.169, p < 0.05), not washing fruits and vegetables before eating (OR = 1.661, 95% CI 1.060-2.601, p < 0.05), not drinking filtered/boiling water (OR = 1.507, 95% CI 1.009-2.251, p < 0.05), not bathing two times weekly (OR = 1.820, 95% CI 1.192-2.779, p < 0.05), watering plants using untreated water (OR = 1.850, 95% CI 1.067-3.207, p < 0.05) and working mother (farmers) (OR = 2.255, 95% CI 1.220-4.169, p < 0.05). Multivariate analysis using forward stepwise logistic regression confirmed rearing animals and not bathing two times weekly as significant risk factors of intestinal protozoal infections. Based on these factors there is a probable risk of zoonotic transmission and this requires further investigation.

S7.9 The comparison of artificial membrane feeding and direct feeding on *Culex quinquefasciatus* (Say) (Diptera: Culicidae)

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The potential of artificial membrane feeding method in laboratory was studied by using *Culex quinquefasciatus* vcru strain. Bloods used were from cattle and goat to represent zoophilic classes, human to represent arthopohilic and chicken to represent ornithopilic. These simple artificial membrane feeding techniques were measured as successful in laboratory where 72% to 90% mosquitoes were observed engorged in this study. From one-way ANOVA with post-hoc comparison statistic analysis, the engorgement of *Cx. quinquefasciatus* were not influenced by blood classes by using these feeding techniques in laboratory [F(3,16)=3.549, p<0.05]. Consequently, most of the mosquitoes that have been exposed with this artificial feeder showed the positive response. The mean percentage of engorged females that laid eggs ranged from 86.4% to 95.9% by using all type of blood classes. An almost similar pattern of fecundity and sex ratio of *Cx. quinquefasciatus* were determined between the direct feeding and artificial feeding techniques.

S7.10 Comparative study of the macroparasite communities of stray cats from four urban cities in peninsular Malaysia

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A study on diversity and distribution of macroparasites from stray cats in peninsular Malaysia was carried out from August 2008 to September 2009. Post-mortem examination was conducted on 425 stray cats captured from four localities representing the west (Kuala Lumpur), east (Kuantan), northern (Georgetown) and southern (Malacca) states of peninsular Malaysia with each location representing a unique habitat. High prevalences of macroparasitic infection were observed in all localities with Kuantan (80.8%) exhibiting highest, followed by Malacca (76.2%), Kuala Lumpur (73.4%) and Georgetown (71.1%). In total, 8 endoparasite helminthes were recorded consisting of five nematode species (*Toxocara malaysiensis, Toxocara cati, Ancylostoma braziliensis, Ancylostoma ceylanicum, Physaloptera praeputialis*) two cestode species (*Taenia taeniaeformis, Dipyllidium caninum*) and one trematode specie (*Platynosomum fastosum*). Most helminthes were present in all study sites except for, *P. praeputialis* that was not present in Kuala Lumpur. Species prevalence differed between locations, with *Ancylostoma* sp. exhibited the highest in Malacca (38.1%) followed by Kuala Lumpur (36.5%) while, *T. malaysiensis* in Georgetown (36.67%) and Kuantan (40.38%). Only three ectoparasites were recovered from this study namely, *Ctenocephalides felis, Felicola subrostrata* and *Haemaphysalis* sp. Distribution of *Haemaphysalis* sp. was found in all cities except for Kuantan. It was observed that parasite distribution was also influenced by host-age and gender.

S7.11 Molecular characterization of *Blastocystis* sp. isolates from goats in Malaysia

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This represents the first study to report the prevalence as well as the genetic diversity of *Blastocystis* sp. in goat in Malaysia. A total of 188 faecal samples were collected from four enclosed farms (UPM1, UPM2, Kuala Klawang and Ulu Langat) and were directly genotyped using sequenced-tagged site (STS) primers-PCR. The overall prevalence of *Blastocystis* sp. in goat samples in Malaysia was 34% with Ulu Langat farm showed the highest prevalence (41.7%). 64 isolates were classified into four distinct *Blastocystis* sp. subtypes. The most predominant *Blastocystis* sp subtype was subtype 1 (54.7%) followed with subtype 2 (46.9%), 4 (46.9%) and subtype 3 (12.5%). Interestingly, 34 goat *Blastocystis* isolates from three farms (UPM1, UPM2 and Kuala Klawang) belonged to subtype 1. Meanwhile, all goat *Blastocystis* isolates from Ulu Langat farm showed mixed subtype infection of subtype 1, 2, 3 and 4. This finding indicates an active transmission of *Blastocystis* sp. among the goats. However, the zoonotic potential of these goat isolates remains to be elucidated.

S7.12 Better prediction of the development of liver cirrhosis in chronic HBV infection using both HBV genetic and serum iron biomarkers

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Hepatitis B virus (HBV) and high liver iron deposits have both been associated with the development of liver cirrhosis (LC). However, the relationship between LC and HBV genotype, basal core promoter (BCP) and precore mutations has been inconsistent. In this study, sera from participants of chronic HBV infection with and without LC were used for whole genome analysis for HBV genotype and mutations in the core, surface, X and polymerase regions as well as for estimation of serum iron status. Statistical analyses showed that precore wild-type, high serum iron and high ferritin were associated with LC respectively (P values = 0.001, 0.020 and 0.008). Moreover, the combined use of these two biomarkers would give rise to the better association with LC (P value < 0.001). This is the first study to provide a more promising approach to assess the risk of LC development in chronic HBV patients which is crucial in patient management and intervention.

S7.13 Genotypic determination of *Toxoplasma gondii* strains by PCR-RFLP from clinical samples in Malaysia

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The population genetic structure of *Toxoplasma gondii* in Europe and North America is highly clonal with three predominant types (I, II, and III). While strains from all three lineages were isolated from human, the majority of human toxoplasmosis cases were associated with strains of a type II. We determined genetic analysis of *SAG2* and *GRA6* loci in eleven patients presenting with ocular or psychotic disorders by nested polymerase chain reaction followed by restriction fragment length polymorphism (PCR-RFLP) in Malaysia. Their sera were confirmed for the presence of IgG and IgM antibodies against *T. gondii* by using a standard commercial enzyme-linked immunosorbent assay (ELISA, Trinity Biotech, USA) and enzyme immunoassay for IgG avidity test (Nova Tec Immundiagnostica GmbH, Dietzenbach, Germany). Type II strains of *T. gondii* were found in a majority of the samples, accounting for nine (82%) of the 11 patients. In contrast, type III strain was found in only two (18%) patients and none was found for type I strain. The results of this study support the previous finding that type II strains are most often associated with human toxoplasmosis. The PCR-RFLP analysis described here will be useful for analyzing the correlation of specific clonal lineages with *Toxoplasma* infection in man.

S7.14 Variant surface glycoprotein (VSG) gene repertoires expressed by Malaysian isolates of *Trypanosoma evansi*

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Trypanosoma evansi has the ability to evade the host immune modulators through a complex process of antigenic variation. This mechanism involves switching of the variant surface glycoprotein (VSG), a dense monolayer that covers the cell surface of the parasite. The VSGs of 24 *T. evansi* isolates from cattle, buffalo and deer hosts from peninsular Malaysia were amplified following inoculation into mice. Total RNA was extracted and RT-PCR amplification was performed. Three to five clones of each isolate were sequenced in order to obtain a consensus amino acid sequence. The VSG sequences were aligned and phylogenetic analyses were carried out to determine similarities between sequences. Six of the 24 isolates expressed more than one VSG, with a number of sequences showing high similarities to VSGs expressed by *Trypanosoma brucei*. The phylogeny obtained revealed that there was no distinct pattern of VSG clustering based on either geographical location or hosts. However, a number of identical VSGs were sequenced from geographically distinct isolates. The present data indicates that the VSGs expressed by local *T. evansi* strains are highly variable, with no apparent geographical localization or hosts.



PUNCAK MINDA SERVICES

Abstracts

Poster Presentations
Trichomonas vaginalis in the Malaysian population - prevalence and implication

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P1

P2

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Trichomoniasis is the most common curable sexually transmitted non-viral disease mainly causing symptoms in men and women. World-wide, approximately more than 200 million people are infected with this parasite annually. This study was undertaken to determine the prevalence of *Trichomonas vaginalis* in Malaysia especially in Klang valley, also to determine the possible association with life style risk factors and cervical neoplasia. A total of 695 high vaginal swabs were taken from those patients who came for routine Pap smear and sexually active women with complaints of vaginal discharge, were included in this study from June 2007 to December 2009. 67.19% (n=467) of patients were asymptomatic and 32.81% (n=228) were symptomatic. This study demonstrates a cytological and culture method of detection of *T. vaginalis* infection. 11 positive cases (1.58%) of *T. vaginalis* were detected with six patients having cervical intraepithelial neoplasia (CIN3). The contributory risk between the exposed subjects and the study population is 54.55% and 0.86% resepctively. The other 5 cases (1.32%) showed inflammatory changes with moderate leucocytosis in the Pap smear. Inflammation was significantly more common in the symptomatic group (p=0.000). *Trichomonas vaginalis* infection was more prevalent in patients who were sex workers. The prevalence rate of trichomoniasis in our study population is 1.58%. The study confirms that life style does play a very important role in the transmission of this infection which emphasizes that preventive measures is mandatory in this group of people.. This study shows that there may be an association between trichomoniasis and cervical neoplasia.

Modified Fields' staining – A rapid stain for Trichomonas vaginalis

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Trichomonas vaginalis, a flagellate protozoan parasite commonly found in the human genitourinary tract is transmitted primarily by sexual intercourse. Diagnosis is usually by *in vitro* culture method and staining with Giemsa stain. There are laboratories that use Gram staining as well. We report for the first time that Modified Fields' stain apart from being a rapid fast stain (20 seconds) confers sharper staining contrast which differentiates the nucleus and the cytoplasm of the organism when compared to Giemsa and Gram staining especially on xenic and parasite spiked urine samples showing that bacterial accompaniments and contents in urine would not influence MF staining. The alternative staining procedure, offers in a diagnostic setting, a rapid stain that can easily visualize the parasite with sharp contrasting characteristics between organelles especially the nucleus and cytoplasm. Vacuoles are more clearly visible in parasites from xenic isolates when compared to Giemsa staining and this offers an opportunity to study better transformational changes during the parasite's life cycle.

P3

P4

A multiple mode of reproduction in Trichomonas vaginalis

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Trichomonas vaginalis, a flagellated protozoan parasite is commonly found in the genitourinary tract. Binary fission is the only plausible mode of reproduction shown thus far where the nucleus of the parasite divides into two but this does not account for the rapid growth rates seen *in vitro* cultures within a short time. We provide evidence for a multiple asexual mode of reproduction seen in Giemsa, Modified Fields', acridine orange and DAPI stained *Trichmonas vaginalis* smeared from *in vitro* culture samples. The nucleus of the parasite is seen to divide as many as four within the formed cells. The size of cells seem to increase with many prominent nuclei seen within a large cluster-like body of cells. We observe that this mode of reproduction is not commonly seen and postulate that it could be triggered to increase progeny formation due to stress as they were viable when tested with trypan blue dye test. The study confirms that multiple mode of asexual reproduction does exist in *T.vaginalis* and its role should be further studied.

In vitro cultivation of human Plasmodium knowlesi: Challenges and limitations

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Plasmodium knowlesi is a primate malaria commonly found in Southeast Asia and now it is recognized as the fifth species of human malaria. Successful in vitro cultivation is vital for better understanding of the biological aspects of the organisms. Hence, in this study we attempted to culture P. knowlesi from patients that were admitted to University Malaya Medical Centre (UMMC) from September 2009 till March 2010. Five nested-PCR confirmed cases of P. knowlesi were used. Thick and thin blood smear were made and stained with Giemsa. Both slides were examined under light microscope at 1,000x oil immersion magnification to detect the presence of the parasite and also to determine the percentage of parasitemia level before culture. Static candle jar method was applied and P. falciparum K1 was used as a control. Two ml of EDTA blood were used to initiate the culture and maintained in HEPES-buffered RPMI-1640 media supplemented with 15% AB human serum, fresh human O+ erythrocytes and sodium bicarbonate. Infected human erythrocytes were incubated in a culture flask, allowing the parasites to grow at 37°C in an incubator with 24 hours changing medium. The percentage of parasitemia for each sample was examined daily. Sample with highest percentage of parasitemia (4.1%) at the initial inoculums could survive up to 3 days whereas sample with lowest percentage (1.2%) died on the next day of culture. The preliminary results suggest that high percentage of parasitemia of initial inoculums was desired for a better chance of successful in vitro cultivation of *P. knowlesi*. The major limitations of the study are all the blood samples were obtained from patients after treatment with antimalarial drug; the delay in sample processing for more than twenty four hours and the potential harmful effect of EDTA to the parasites. Hence, in the future study, we may need to obtain the blood before the patients are being treated and inoculate the samples within twenty four hours.

P5 Biodiversity of Geohelminth Eggs from Urban and Suburban areas in Peninsular Malaysia

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Soil contamination with geohelminth eggs is a potential source of infection to human thus, poses a threat to the public especially to children with parasites namely; Ascaris spp., Toxocara spp., Trichuris spp and Ancylostoma spp. Many reports have highlighted the geohelminth impact on human worldwide in relation to diversity of parasite infection and prevalence, however, very little is known of the status here in Malaysia. This study was conducted to determine the distribution and biodiversity of soil-transmitted helminthiasis in 3 localities i.e. west (Shah Alam, Klang and Petaling Jaya), northern (Georgetown) and southern (Malacca) states of Peninsular Malaysia, in particular the contamination of sandpits surrounding children's playgrounds. To date, a total of 175 soil samples were taken from the sandpits of 35 playgrounds between August 2009 and January 2010. This study employed the centrifugal floatation technique with the use of saturated NaCl (SGI.25) to determine egg counts (EPG). Among the sites studied in this report the eggs of Toxocara spp. were observed to be higher in the soil of an urban and suburban area, followed by Ascaris spp., Ancylostoma spp. and Trichuris spp. The percentage of Toxocara spp. eggs recovered in Penang's playground ranged from 10.74% to 42.88%, Ascaris spp. is 10.63% to 31.94%, Trichuris spp. is 2.48% to 9.46% and Ancylostoma spp. is 7.46% to 19.42%. The percentage of Toxocara spp. eggs recovered in Malacca's playground ranged from 6.86% to 14.58%, while Ascaris spp. is 2.40% to 6.66%, Trichuris spp. is 1.26% to 5.0% and Ancylostoma spp. is 2.92% to 5.66%. The percentage of Toxocara spp. eggs recovered in Kuala Lumpur's playground ranged from 9.06% to 34.26%, while Ascaris spp is 2.88% to 10.34%, Trichuris spp is 1.78% to 5.06% and Ancylostoma spp. is 2.12% to 4.14%.

P6 Macroparasitic distribution and biodiversity study in wild rodent populations of Kuala Lumpur

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A survey of wild rodent population in Kuala Lumpur was carried out from the months of September through to November 2009 to determine their diversity and distribution of ectoparasites and endoparasites. A total of 100 rodents were captured comprising of 5 species; Rattus rattus diardii, Rattus annandelei, Rattus norvegicus, Rattus argentiventus, and Rattus tiomanicus. The most dominant species was Rattus rattus diardii (64%). Rodent population observed showed higher number of female (52%) compared to male (48%) and adults (71%) compared to juveniles (29%). Infestation prevalence showed a total of (92%) rats were infected with ectoparasites while (100%) with endoparasites. Two main factors were investigated to determine infestation in the rodent population, i.e. age-related and sex. Study showed that ectoparasites recovered fell under 3 broad groups, namely lice (Polyplax spinulosa), mites (Laelaps nuttali and Laelaps echidninus) and flea (Xenopsylla cheopis) while endoparasites recovered were Taenia taeniaformis, Rodentolepis nana, Hymenolepis diminuta and Nippastrongylus brasiliensis. Rats generally harbored more than one group of ectoparasites and endoparasites on their bodies. Laelaps nuttali and Laelaps echidninus attributes to the highest infestation rate of (89%) followed by Polyplax spinulosa (76%) and the lowest being (49%) for Xenopsylla cheopis. Overall prevalence was (79%) with Nippostrongylus brasiliensis parasites being the dominant helminth followed by Taenia taeniaformis was (43%), Hymenolepis diminuta was (26%) and the lowest being (20%) for Rodentolepis nana. One of the ectoparasites found, Xenopsylla cheopis is a known vector of murine typus and plague, thus may pose potential health risks to human. This study provides a much needed baseline for the ecology of urban area and showed current data for further research.

P7 A survey of bacterial and parasitic infections of rats caught in the Veterinary Research Institute (VRI), Ipoh, Malaysia

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A postmortem survey on ten rats (*Rattus* spp.) caught in the Veterinary Research Institute (VRI) was done to evaluate the parasite, viral and bacterial status. The rats were trapped in the laboratory animal facilities in VRI. The results show that seven rats harboured mites (*Demodex* sp) from skin samples. Parasite eggs such as strongyle and *Strongyloides* sp. were also recovered from the intestines of four rats. Bacterial species isolated from the organs include *Mycoplasma arthritidis, Corynebacterium* sp., *Staphylococcus epidermis, Escherichia coli, Enterococcus* sp., *Leptospira canicola, Leptospira celledoni* and *Leptospira pyrogenes*. Many of these organisms are zoonotic especially leptospirosis which can cause severe disease in humans. No pathogenic viruses were recovered from the rats. Control and eradication of pests like rats is essential to safeguard the health of humans.

P8

Detection of blood protozoan in the urban rat population of peninsular Malaysia using Quantitative Buffy Coat (QBC) technique

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A study was conducted to detect blood protozoan in urban rats using Quantitative Buffy Coat (QBC) technique. A total of 234 blood samples were collected from rats via heart puncture trapped from various locations in peninsular Malaysia; Kuala Lumpur (west), Kuantan (east), Malacca (south) and Georgetown (north). A total of 55-110 UI of blood was drawn into a QBC tube coated with Acridine Orange (AO-fluorescent dye) and centrifuged. Parasites when present was detected concentrated below the buffy coat in QBC tube viewed under Paralens microscope adaptor. A total of 3 blood protozoan were identified to genus level; *Plasmodium* sp. (20.1%), *Babesia* sp. (17.9%). and *Trypanosoma* sp. (12.8%) with 44.9% blood samples positive for infection in the whole population. Of these, rodent from Kuantan had the highest numbers of rats infected (47.9%), followed by Georgetown (43.48%), Malacca (44.9%) and Kuala Lumpur (31.8%). Infection observed by rat species showed the highest, *Rattus norvergicus* (30.3%), followed by *Rattus rattus diardii* (12%), *Rattus exulans* (1.3%) and *Rattus argentiventer* (1.3%). In terms of sex, females recorded higher 23.1% than males 21.8% among those positive with infection. This study showed that QBC technique proved to be a fast and useful screening test for detection of blood protozoan in rodents.

Human Blastocystis sp. subtype 2 and subtype 4 in recreational waters in Selangor

P9

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Although Blastocystis sp. has been reported previously in recreational waters, its potential for waterborne transmission has not been elucidated. Hence, the molecular characterization of Blastocystis sp. in recreational waters was determined. 90 samples were collected from 5 rivers (Congkak River, Gabai River, Tekala River, Kanching River and Rangkap River) and 2 lakes (Jaya Lake and Titiwangsa Lake) in the Selangor state, Malaysia. Various volumes of water samples (0.5L, 1.0L, 5.0L and 10.0L) were collected from each collection point and filtered through a membrane filtration system. Collected water sediment was layered on ficoll-paque and centrifuged for the isolation of Blastocystis sp. cysts. The cysts were cultured in Jones' medium supplemented with 10% horse serum followed by incubation at 37°C and examined daily for vacuolar Blastocystis sp. using a light microscope for the next three days consecutively. Blastocystis sp. cysts were enumerated using haemacytometer with trypan blue as the background dye. All cultures that were positive for Blastocystis sp. were genotyped using sequenced-tagged site (STS) primer-polymerase chain reaction (PCR). The presence of Blastocystis sp. was detected in three samples collected from Congkak River (up, mid and down stream) and one sample from Java Lake. The minimum volume detectable for Blastocystis sp. was determined to be 10.0L. The range for Blastocystis sp. cysts was 4x10³-1.2x10⁴ cysts/L. All four recreational water samples have Blastocystis sp. subtype 2 and subtype 4. Since these subtypes were previously reported in humans, there is a high possibility that all four recreational water samples were contaminated with Blastocystis sp. excreted by humans. Therefore, it could pose as a potential hazardous threat to visitors who frequent these recreational waters.

P10 Possible zoonotic and waterborne transmissions of *Blastocystis* sp. in a rural community in Bahunipati, Nepal

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The presence of *Blastocystis* sp. was examined in faecal and river water samples from Bahunipati, a rural village in Nepal. *Blastocystis* sp. was examined in faecal samples collected from 65 farm animals (buffalos, cows, goats and pigs) and 66 humans. Briefly, faecal samples were cultured in Jones' medium supplemented with 10% horse serum followed by incubation at 37°C and examined daily for vacuolar *Blastocystis* sp. using light microscope for the next three days consecutively. Faecal samples positive for *Blastocystis* sp. by *in vitro* cultivation were further genotyped using seven subtype-specific sequence tagged site (STS) primer-polymerase chain reaction (PCR). Four river water samples (collected from Sindukhola River and Indrawati River), one litre each, were centrifuged to obtain water sediment. *Blastocystis* sp. that might be present in the water sediments was further genotyped. *Blastocystis* sp. infected 36.4% of humans and 15.4% of animals. *Blastocystis* sp. subtype 4 was the predominant genotype for humans and animals infecting 95.8% and 40.0% of them respectively. All four river water samples were contaminated with *Blastocystis* sp. with subtype 1 (100%) being the predominant genotype. Two individuals from two separate families (family A and B) were infected with *Blastocystis* sp. subtype 4. A buffalo (family A) and pig (family B) that they reared nearby their house were also infected with subtype 4. Human *Blastocystis* sp. subtype 1 and subtype 4 were found in all river samples. Hence, these findings suggest possible zoonotic and waterborne transmissions within this village.

P11 Triosephosphate isomerase (TPI) protein phylogeny supports Stramenopiles grouping of *Blastocystis*

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The taxonomic position of *Blastocystis* sp. remains controversial. The small subunit ribosomal RNA (SSU rRNA) gene phylogeny demonstrated that *Blastocystis* belongs to Stramenopiles In contrast, a protein phylogeny of elongation factor-1á (EF-1 α) suggested that *Blastocystis* be placed under the Subphylum Conosa together with *Entamoeba*. In the present study, the phylogenetic affinities of *Blastocystis* was re-evaluated using another highly conserved gene, triosephosphate isomerase (TPI). A total of 15 clinical isolates of *Blastocystis* sp. were used. TPI-based phylogentic tree showed five distinct groups of isolates corresponding to the subtype 1 to 5 based upon the small subunit ribosomal RNA (SSU rRNA) gene phylogeny. The TPI protein phylogeny supported the Stramenopiles grouping of *Blastocystis* with a bootstrap value of 93%. Hence this study further confirms that *Blastocystis* belongs to the Stramenopiles.

P12 Assessment of urinary hyaluronidase activity in rats infected with *Blastocystis* hominis

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The fact whether *Blastocystis hominis* can invade has always been in question. Apart from a few sporadic studies such as that done on gnotobiotic guinea pigs which showed surface invasion and mucosal inflammation of the host's intestine caused by *B.hominis* infection, no real documentation of invasion has been proven. Studies have shown that hyaluronidase is secreted during the penetration into the host's skin and gut by nematode parasites. Here, we attempt to determine hyaluronidase in urine samples of rats infected by *B.hominis*. The presence of hyaluronidase in urine may provide an indirect evidence of invasion by *B.hominis* into colonic epithelium causing the degradation of extracellular matrix proteins (e.g.: hyaluronic acid). In current study, urinary hyaluronidase levels in Sprague-Dawley rats infected with *B.hominis* were observed for 30 days. Hyaluronidase levels in the infected rats were significantly elevated on Day 28 and Day 30 compared to the day before inoculation (p< 0.01 and p<0.05 respectively). During this stage, parasitic burden in infected rats' stool was high. Histological examination of cecum and colon from infected rats showed that although there was no evident damage seen, intense inflammatory-cell infiltration at the mucosal layer shows that *B.hominis* has pathogenic potential. Detection of *B.hominis* at the edge of mucosa layer of cecum suggests that it is invasive and supports the possible reason for the elevation of HAase. This preliminary research has verified that the organism is capable of having invasion or penetration activity in order to facilitate its' growth in the hosts' intestine.

P13 Blastocystis hominis infection increased levels of oxidative stress and proinflammatory cytokines in sprague-dawley rats

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Numerous studies have suggested the presence of oxidative stress in humans and animals infected with parasites. However most of these studies are based on blood parasites and research on intestinal parasites concerning to oxidative stress is still limited. Previously, we have reported that oxidative stress is elevated in Malaysians infected with intestinal parasites. Majority of these humans had asymptomatic infection *Blastocystis hominis*. Generally when a host's immune system is triggered by parasitic infection, oxidative burst is activated by macrophages which in turn affect the inflammatory system. However, not much is known about the *B.hominis* infection on oxidative stress and inflammatory response *in vivo*. In this study, we compared the levels of oxidative stress biomarkers in urine and blood samples between uninfected and *B.hominis* infected rats. The pro-inflammatory cytokines (IL-6 and IL-8) in the rats' serum were also measured. Infected rats had high levels of oxidative indices namely advanced oxidative damage level was higher. Ferric reducing antioxidant power (FRAP) was elevated at the early stage of infection but decreased significantly during the last week of study period indicating that the host's antioxidant status may be overwhelmed by oxidative damage. High levels of serum IL-6 and IL-8 in the infected rats indicate that inflammation at the site of infection may have taken place. To date, this is the first *in vivo* study to confirm that *Blastocystis* infection can result in significant oxidative burst which may lead to inflammation.

Effect of Blastocystis hominis on oxidative stress and cancer

P14

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Parasitic infections initiate carcinogenesis through inflammatory processes which produce free radicals (e.g.: reactive oxygen species, ROS) to kill the invading parasites. Excess of ROS can cause DNA mutations leading to carcinogenesis. Thus, we aimed to investigate the effect of intestinal parasites namely *Blastocystis hominis* in exacerbating colorectal cancer. The study involves 3 areas: assessment of oxidative stress level in A) cancer patients with and without parasitic infection and B) *in vivo* model with *B. hominis* infection; C) evaluating the effect of *in vitro B. hominis* infection in promoting cancer cell growth. Results showed that colorectal cancer patients (majority with *B. hominis* infection) exhibited high level of oxidative protein damage. Non-cancerous individuals with intestinal parasitic infections (majority with asymptomatic *B. hominis* infection) had high levels of oxidative damage to lipid and protein. Asymptomatic *B. hominis* infection in rats showed that the parasite could trigger oxidative damage to lipid and protein and elevate the pro-inflammatory cytokine levels. These findings imply that intestinal parasites especially *B. hominis* can result in significant oxidative burst which may lead to inflammatory processes, an initial step in triggering carcinogenesis. Furthermore, *in vitro* study using antigen isolated from asymptomatic *B. hominis* confirmed that the parasite possesses the ability to enhance the growth of colorectal cancer cells. In conclusion, the study has proven that *B. hominis* can elevate levels of oxidative stress and promote the growth of colorectal cancer cells. Thus, there is a vital need to include treatment protocols in patients with asymptomatic blastocystosis.

Effect of ultraviolet radiation on methicillin-resistant Staphylococcus aureus

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Staphylococcus aureus has been identified as one of the commonest causes of hospital and community acquired infections. With the emergence of methicillin-resistant *S. aureus* (MRSA), methicillin which is was one of the strongest in the arsenal of antibiotics to treat staphylococcus infections is no longer effective. Elimination of this organism on wounds and work surfaces would be an important step in providing a chemical-free sanitizing alternative. This study was carried out to determine the bactericidal and morphological effects of ultraviolet (UV) radiation exposure on MRSA. Approximately 2.0 x 10⁶ colony forming unit (CFU) of MRSA were exposed to UV radiation of 253.7 nm wavelength for a duration that ranged from 30s to 180s. Results showed that there were 97% (Log₁₀6) reduction in the number of CFU of MRSA within 30s of exposure to UV. However at 60s exposure, 100% of the MRSA were killed. Antibiotic susceptibility test performed on the UV-exposed MRSA against penicillin, oxacillin, erythromycin and vancomycin showed that their antibiotic susceptibility was not affected. Similarly, the morphology of MRSA did not change, as observed by Grams' staining and confirmed by scanning electron microscopy (SEM). It can be concluded that a minimum of 60s exposure to UV light at 253.7 nm wavelength is bactericidal to MRSA and exposure to UV has no affect on the morphology as well as the susceptibility of MRSA against the antibiotics tested.

P16

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Potential home remedies for methicillin-resistant Staphylococcus aureus infections

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Since the emergence of methicillin-resistant *Staphylococcus aureus* (MRSA), many natural products have been explored as alternative medical approach on treating against infections caused by this bacterium. Most of these remedies were inspired through traditional practices that have been passed down from one generation to another. The high success rates claimed by the folklore provoke an interest in investigating the antimicrobial properties of some food and natural products commonly available in most homes. Antimicrobial activities of honey, *Allium sativum* (garlic), *Curcuma longa* (tumeric), *Nigella sativa* (black seed), *Aloe barbadensis* (aloe vera) and *Holothuria scabra* (sea cucumber) against MRSA were determined using disk diffusion method. It was observed that honey, fresh *A. sativum*, extracts of *N. sativa* and *H. scabra* produced zones of inhibition whereas the other samples failed. *Nigella sativa* extract exhibited the largest zone of inhibition which was ten times larger than that produced by honey, *A. sativum*, and *H. scabra*. Agar dilution method used to measure the minimum inhibition concentration showed that a concentration of <10% of the *N. sativa* extract was able to inhibit the growth of MRSA. The findings of this preliminary study indicate that the extract of *N. sativa* has high inhibitory effect on MRSA in comparison to honey, *A. sativum*, and *H. scabra*. This observation warrants further investigation into the potential use of *N. sativa* for treatment against MRSA infections.

P17 Plasmodium vivax gene polymorphism in field samples in Tawau, Sabah

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It is known that Plasmodium vivax does cause morbidity. Recent study has shown that they can be fatal. Therefore analyzing the P. vivax population structure is important in order to understand the role of genetic diversity in the transmission of the disease. Several polymorphic genes, such as the circumsporozoite protein (Pvcsp) and merozoite surface proteins (Pvmsp) have been used for evaluating the genetic diversity of P. vivax. The present study is to investigate the genetic diversity of P. vivax in blood samples collected from general population in Kalabakan, Tawau, Sabah. Twenty seven samples were found positive for P. vivax by rapid diagnostic test (ParaMax 3). DNA from blood spots on filter paper of these samples were extracted using QIAamp DNA mini kit (Qiagen, USA). Three genes used were Pvcs coding for circumsporozoite protein and msp-1 and msp- 3α coding for the merozoite surface protein 1, and 3α . The two sequence types for Pvcsp gene were, VK210 and VK247. Nested PCR were performed followed by restriction enzyme digestion. Pvcsp nested PCR products, were digested with Alu I and Bst NI to determine the P. vivax family type VK210 and VK247 respectively. The PCR and the digested products were analyzed using Agilent DNA 1000 Kit (Agilent, Germany) and agarose gel electrophoresis. The nested PCR for msp-1 gene was to indicate the presence of region F1, F2 and F3 high polymorphic region. The amplification of msp-3 a gene was to indicate the parasite belongs to Type A (Belem strain (Brazil)) or Type B and C for strains other than Belem. The present study showed that the Pvcsp gene belonged to type VK 247 (51.85%) only. The amplification of the msp-1 gene revealed that 100% of the samples tested have F3 region, 14 (51.9%) with F2 region and 5 (18.5%) with F1 region. Study on Msp-3α gene showed presence of Type A (Belem strain) and B (unknown) with 25.92% and 22.22% respectively. The P. vivax strain typing for pvcsp gene in Kalabakan has the VK247 type, which is similar to Thailand and Papua New Guinea where VK247 was found to be predominant. The sample tested were absent for VK210 type. VK247 type signifies mixed infection while VK210 signifies pure infection.

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Survey of Eimeria species in broiler farms in the west coast of peninsular Malaysia

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The intracellular protozoan parasites *Eimeria* species are the causative agents of the economically important disease coccidiosis in chickens. There are seven distinct *Eimeria* species that are known to cause avian coccidiosis, namely *Eimeria acervulina, Eimeria brunetti, Eimeria maxima, Eimeria mitis, Eimeria praecox, Eimeria necatrix* and *Eimeria tenella*. The aim of this study was to identify the diversity of *Eimeria* species present in commercial broiler farms in the west coast of peninsular Malaysia using morphological characterisation. In this study, fecal materials with signs of possible infection were sampled from six poultry farms from five different states in the west coast of peninsular Malaysia. Oocysts were purified using salt flotation and differentiated by their shape and dimension into the respective species. Five *Eimeria* species were identified from the samples but with different infection levels. The characterisation showed that *E. tenella* and *E. maxima* were found in all six farms, *E. mitis* and *E. brunetti* in five farms, and *E. acervulina* in only one farm. The results of this study also indicate the presence of multiple *Eimeria* species infection in the collected samples. Understanding the occurrence and distribution of various *Eimeria* species in broiler farms is necessary to develop effective controls against coccidiosis.

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Identification of Eimeria species in free range chickens in Muar, Johor

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Coccidiosis is recognised as the parasitic disease that has the greatest economic impact on poultry production. There are seven species of *Eimeria* that are known to cause coccidiosis in chickens, namely *Eimeria maxima, Eimeria tenella, Eimeria acervulina, Eimeria brunetti, Eimeria mitis, Eimeria praecox* and *Eimeria necatrix*. A survey was carried out in 11 different locations in Muar, Johor where 3 to 5 chickens aged between 2 to 3 weeks were kept overnight in a cage prior to litter collection. Following washing and salt flotation, samples from four of the locations were found to contain *Eimeria* oocysts and were designated as S1, S2, S3 and S4. Each of the samples were then sporulated, and subsequently amplified in specific pathogen free chickens. Chicken litters were collected at days 4-5, days 6-7 and days 8-10. The results showed that for S1, oocysts were detected at days 4-5 and days 8-10, while for S2, oocysts were present at days 6-7. For S3 and S4, the presence of oocysts was observed at days 6-7 and days 8-10 respectively. Genomic DNA was then extracted from each of the positive samples and subjected to PCR amplifications using seven different pairs of primers, each capable of identifying a specific *Eimeria* species infecting the chicken. Results of this study showed that out of the seven avian *Eimeria* species, only two species, specifically *E. tenella* and *E. acervulina* were found infecting the free range chickens.

P20 Expression of *Toxoplasma gondii* dense granule antigen (GRA2) in the yeast *Pichia pastoris* for the use in serodiagnosis of toxoplasmosis

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Toxoplasma gondii is an obligate intracellular protozoan parasite that infects a wide range of animals, including humans. Routine diagnosis of toxoplasmosis is based on antibody detection in an infected person. Antigens used in immunodiagnosis assays are usually total protein extracted from *T. gondii* cells propagated in mouse or *in vitro* culture.GRA2 is a 28kDa hydrophobic protein, highly immunogenic and plays an important role during *in vivo* infection. The recombinant protein can be incorporated as antigen in ELISA to detect specific IgG and IgM. The *Pichia pastoris* expression system is appropriate since it can yield very high levels of secretion and be scaled-up without loss of yield. RNA of *T.gondii* was extracted and GRA2 gene was amplified using specific primers. GRA2 gene was cloned into pPICZ α A and transformed into *P. pastoris*. Positive recombinant clones were confirmed by sequencing and selected for expression. The purified protein will be tested in Western blot and ELISA to evaluate the sensitivity and specificity of recombinant GRA2 protein. GRA2 gene was amplified, cloned into PPICZ α A and transformed in *P. pastoris*. Sequencing result showed 100% similarity of recombinant GRA2 with GRA2 gene in GeneBank. To date, purification of recombinant GRA2 is on-going and evaluation test for sensitivity and specificity will be carried out once the recombinant GRA2 is purified.

P21 Vaccination against toxoplasmosis by *Toxoplasma gondii* recombinant dense granular DNAs and proteins

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Toxoplasma gondii is an obligate intracellular protozoan parasite which infects all warm- blooded animals, including human, and causes toxoplasmosis resulting in serious public health problems, especially to immunosuppressed patients and pregnant women. It also causes great economic loss in livestock industry. Therefore, a safer means of vaccine is desperately needed. This study focuses on dense granule antigens, GRA2 and GRA5 as vaccination candidates due to their immunogenicity properties. DNA fragments encoding GRA2 and GRA5 were amplified through PCR, and were cloned into pcDNA3.1 C and pRSET B vectors. Preliminary study of DNA vaccination was carried out in BALB/C mice with pcGRA2 and pcGRA5. Serums collected were screened through Western Blotting but did not show promising results. Further study on the DNA vaccination together with adjuvant will be followed. Expression of recombinant GRAs-pRSETB was done in *Escherichia coli* BL21 (DE3) pLysS. The expressed proteins were collected and purified through The MagneHis[™] Protein Purification System. The non-purified and purified products of both GRA2 and GRA5 recombinant proteins were analyzed with SDS-PAGE and Western Blotting, showing the sizes of approximately 33 kDa and 27 kDa respectively. Immunogenic properties of these recombinant proteins will be performed.

P22 *Toxoplasma gondii*: Molecular cloning and expression of full length rhoptry protein 2 (ROP2) gene in yeast

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Toxoplasmosis is a world-wide infection caused by *Toxoplasma gondii*. *Toxoplasma gondii* is an obligate intracellular protozoan parasite which belongs to the phylum Apicomplexa. In immunodeficient individuals such as AIDS patients, reactivation of the chronic infection can lead to Toxoplasmic encephalitis (TE). Rhoptry protein 2 (ROP2) is one of the rhoptry secreted protein which plays critical role in parasitophorous vacuole membrane (PVM) formation and in turn helps the parasite to invade into host cell. High immunogenicity of ROP2 makes it a candidate for diagnosis as well as vaccination against toxoplasmosis. Current diagnosis of toxoplasmosis with commercial kits is not specific and sensitive enough as total proteins of *T. gondii* tachyzoite were used for detection. In this study, the full length ROP2 gene of *T. gondii* was cloned into pPICZ α A expression vector and extracellularly expressed in the yeast *Pichia pastoris*. The secreted recombinant ROP2 (rROP2-F) was analyzed by SDS-PAGE and Western blotting. A 64kDa recombinant protein was detected by toxoplasma-antibody positive human serum.

P23 Protection of mice from fatal *Toxoplasma gondii* infection by immunization with recombinant SAG1 (P30) antigen in alum

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Whereas it is not currently foreseen that a vaccine against *Toxoplasma gondii* will be used to prevent congenital toxoplasmosis in humans, immunizations of sheep and swine may prevent abortion and cyst formation respectively and thereby interrupt an important route of transmission to humans. *Toxoplasma gondii* infection is a problem for sheep farmers, causing considerable economic loss, and a live vaccine based on an attenuated 7: *gondii* isolate which do not form tissue cysts is at present used to prevent 7: *gondii* induced abortion in sheep. We have cloned and expressed the 7: *gondii* major surface antigen of the tachyzoite, SAGI, in an *Escherichia coli* vector. The antigen is highly immunogenic and antibodies against the recombinant SAG1 also recognize native *T. gondii* tachyzoite antigen. Immunization with alum and rSAG1, enhanced the protection (75% survival). A significantly higher survival of newborns from immunized outbred mice exposed to infection during gestation was observed (4.25±3.77 live pups/litter) in comparison to non-immunized mice (1.08±2.15 live pups/litter) without preventing parasite vertical transmission. Analysis of the immune response showed that protected animals developed a specific humoral and cellular Th1 response to recombinant SAG1 and native *T. gondii* tachyzoite antigens. Immunization with the recombinant SAG1 is therefore expected to induce protective immunity, limiting clinical and pathological consequences of *T. gondii* infection.

P24 Seroprevalence of toxoplasmosis among stray cats in Kuala Lumpur suburban city areas

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Infection by the protozoan parasite, *Toxoplasma gondii* is widely prevalent in humans and animals throughout the world. Transmission takes place mainly by ingestion of raw or undercooked meat that contains parasite cysts or by ingestion of oocysts excreted in cat feaces, which can contaminate water and raw vegetables. The incidence of toxoplasmosis in urban areas can thus be also related to environmental contamination with oocysts. A direct measure of this environmental contamination by oocyst counting is unfeasible for technical reasons. An interesting alternative for measuring *T. gondii* urban spreading is the seroprevalence in free-living urban animals, used as sentinels, once they are exposed to similar risks of *Toxoplasma* infection-like humans. With this aim, we tested serum samples from stray cats for antibodies to *T. gondii* by a recombinant antigen (rSAG1) based antigen enzyme-linked immunosorbent assay (ELISA). Antibodies to *T. gondii* were found in 50.5% (101 of 200) of these stray cats (P < 0.05). Other test system for comparision with rSAG1 ELISA showed low resolution and concordance, precluding their use for diagnosis of *T. gondii* infection. *Toxoplasma gondii* seroprevalence in stray cats could be an indirect indicator of the parasite spreading in urban areas.

P25 A preliminary screening on the potential of *Curcuma longa* (Tumeric) extracts for the treatment of malaria

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Curcuma longa (tumeric) have been used as a traditional medicine for the treatment of a variety of sicknesses including parasitic diseases. Anti-malarial drug resistance has become one of the greatest challenges against malaria control. Drugresistance to chloroquine and more recently quinine was responsible in the spread of malaria to new areas and recurrence of malaria in areas where the disease has been eradicated. To test whether the above claim has a sound medical basis, experiments were performed to show the effect of *C. longa* extract on the survival of malaria parasite, *Plasmodium berghei* in mice. Intraperitoneal and oral administrations of ethanol, chloroform and aqueous seed extracts (50, 100, 200 and 400 μ L kg⁻¹) of *C. longa*, were screened in the 4-day suppressive assays for their anti-malarial properties against *P. berghei* in mice. Both intraperitoneal and oral treatments of the seed extracts showed suppression activities in all groups of mice with the highest values were noted by the 100 and 200 μ L kg⁻¹ doses of the ethanol extract and by the 100 μ L kg⁻¹ dose of the chloroform extract which significantly (p <0.05) decreased the parasitaemias and increased the survival times of the infected mice. On the other hand, the aqueous extract showed a dose-dependent suppression activity by which the 200 and 400 μ L kg⁻¹ extracts doses showed significant degrees of suppression activities in the infected mice. The results of this study confirm the usage of this plant as remedy for malaria and open a new opportunity to further investigate the potentials of this plant a novel antimalarial in the future. However, the active responsible principles are yet to be identified, which need further studies to elucidate the anti-malarial mechanism of its action.

P26 Species abundance and infection of *Trypanosoma evansi* in biting flies from four selected animal farms in Peninsular Malaysia

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Animal trypanosomiasis due to *Trypanosoma evansi* is an important disease that affects a wide range of domestic and wild animals and is mechanically transmitted by several blood-sucking flies. This study was conducted to assess the abundance and infection in biting flies from four selected animal farms in Peninsular Malaysia. Samples of biting flies were caught by using a sweep net. The flies were then transferred into a cage. All catches were recorded accordingly and identification of each biting fly was done in the laboratory under a stereo microscope with reference to keys of Greenberg (1971). A total of 11,676 flies were caught. As many as nine species were collected from the family Muscidae namely *Lyperosia exigua*, *Stomoxys calcitrans*, *Musca inferior*, *Musca domestica*, *Musca sorbens*, *Musca crassirostris*, *Musca cf asiatica*, *Musca ventrosa* and *Musca cf conducens* and six others from Family Calliphoridae (*Chrysomya megacephala*), Family Sepsidae, Family Platystomatidae (*Scholastes* sp), Family Sphaeroceridae (*Leptocera* sp), Ulidiidae (*Physiphora* sp), and Hymenoptera (*Trigona* sp). Among them, *Lyperosia exigua* (95.47 %) was the predominant species found at the four sampling sites followed by *Musca domestica* (2.61%), Family Sepsidae (0.39%), *Stomoxy calcitrans* (0.37%), *Musca crassirostris* (0.30%), Family Sphaeroceridae (*Leptocera* sp) (0.13%), *Musca cf. conducens* (0.09%), *Chrysomya megacephala* (0.03%) and others (0.07%). A total of 1157 pools of biting flies were tested using polymerase chain reaction method to detect the presence of *T. evansi*. The PCR results indicated that none of them were infected with *T. evansi*. *T. evansi* infection in biting flies is probably very low in these farms.

P27 Physiological and biochemical changes associated with *Trypanosoma evansi* infection in Boer goats

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Trypanosoma evansi is a widely distributed haemoprotozoa due to its ability to be transmitted mechanically by biting flies. The dynamics of T. evansi infection and associated physiological and biochemical changes were investigated in 18 female Boer goats between 8-10 months of age. The animals were divided into three equal experimental groups, of which two were infected with different field strains of T. evansi (G1 and G2) and one non-infected control (C). A total of 1x10⁴ T. evansi were inoculated intravenously into each goat in the infected groups and blood was collected via venipuncture every alternate day for 60 days. The infected goats were treated after 60 days with diminazene diaceturate, and recovered from the infection. Changes in weight and core body temperature were monitored throughout the infection. Plasma was obtained from whole blood and screened for globulin, aspartate transaminase (AST), creatinine, total protein (TP) and creatine kinase (CK). Core body temperature raised to 40.1°C in the infected goats. The infected animals also showed a 16.3% reduction in body weight gain compared to the uninfected controls. Creatinine and CK decreased by 20.1% and 43.0%, respectively in the infected goats, while the concentration of TP increased gradually over the course of the infection by 34.5% and 26.5% in groups G1 and G2, respectively. In addition, globulin levels increased by 67.8% (G1) and 52.9% (G2), indicating an active immune response to the infection. The levels of AST were also markedly elevated in the infected groups, suggestive of hepatocellular involvement. Although T. evansi infection in goats has not been reported to be acute or fatal, the long term biochemical changes caused by the parasite pose a serious health threat. In addition, the poor weight gain has important implications on productivity.

P28 Evaluation of the Card Agglutination Test for Trypanosomiasis (CATT) and its effectiveness in assessing the dynamics of *Trypanosoma evansi* infection in goats

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The Card agglutination test for trypanosomiasis (CATT) is a standard serological test used for detection of African human and animal typanosomosis and *T. evansi* infection (surra). The diagnosis of *T. evansi* infection in goats is difficult using standard parasitological techniques due to low and erratic parasitaemia. The aim of this experiment was to assess the performance and usefulness of CATT in monitoring *T. evansi* infection in goats. Three groups, each comprising of 6 young female Boer goats were individually inoculated *via* the jugular vein with approximately 1x10⁴ *T. evansi*. The trypanosomes were originally isolated from cattle and a deer from Peninsular Malaysia. Parasitaemia and serology were monitored every other day for 62 days post-infection (PI) and the animals were treated with diminazene diaceturate once thereafter. The goats were examined for 24 days post-treatment (PT) as above. All the animals became positive for *T. evansi* by haematocrit centrifugation technique (HCT) and CATT/ *T. evansi* from days 4 to 6 and days 8 to 10 post-inoculation, respectively. After two major parasitaemia peaks observed around days 6 and 16 PI, the parasite counts were maintained at low levels (ca.100 parasites/ml) until the animals were treated. The mean CATT scores were moderately high during infection but were drastically reduced in the subsequent days PT. The result showed adequacy of CATT/ *T. evansi* for assessing and monitoring of *T. evansi* infection in goats.

P29 Dynamics of parasitaemia and haematological changes in goats infected with two different field isolates of *Trypanosoma evansi*

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The dynamics of parasitaemia and associated haematological changes between two different field isolates of *Trypanosoma evansi* were investigated in 12 female Boer goats aged 6-8 months. The isolates originated from two distinct geographical locations in peninsular Malaysia (Johor and Pahang). A total of 1 x 10⁴ *T. evansi* were inoculated intravenously in each goat and blood samples were collected *via* venipuncture every alternate day for 60 days for haematological evaluation. The infected goats were treated after 60 days with diminazene diaceturate, and a full recovery was observed. The parasite density fluctuated throughout the infection with several peaks, the first apparent at day 6 (13.6 x 10⁶ and 2.5 x 10⁶ parasites per ml for the Johor and Pahang strains, respectively). Haematological parameters examined included packed cell volume (PCV), haemoglobin concentration (Hgb), and total red blood cell counts (RBC). The PCV reduced by 9.0% in the infected group compared to the uninfected controls by day 16 post-infection. The trend continued until the PCV was 30% lower than the controls at the end of the 60 day trial period. The Hgb and RBC showed significant decrease from day 6 onwards and attained a 20% reduction. Moderate eosinophil and neutrophil reduction was apparent from day 6 onwards, while monophil counts increased three-fold in the infected group. Other haematological indices including MCV, MCH and MCHC, leucocyte and basophil counts showed no significant differences. The most significant outcome of *T. evansi* infection in goats with regards to the haemogram was haemolytic anaemia and monocytosis.

P30 Activity and turnover of eosinophil and neutrophil granulocytes are increased in visceral leishmaniasis

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Visceral leishmaniasis (VL) is a disease of public health importance in Sudan and many other tropical countries. The roles of inflammatory cells such as neutrophil and eosinophil and their markers Eosinophil Cationic Protein (ECP) and Myeloperoxidase (MPO) had been investigated in both serum and plasma. Plasma levels mainly reflect cell turnover and serum levels mainly reflect secretory activity. Our aim was to investigate the involvement of eosinophils and neutrophils as reflected by the serum/plasma levels and findings were compared with the levels of several cytokines believed to be involved in the production and activation of these cells. Sera and plasma were collected from 125 VL patients with a mean age of 25 years (18-30) and male to female ratio 91/34 (72%/28%) and 181 healthy Sudanese controls with a mean age of 11 (7-21) and male to female ration 97/84 (51%/49%). Ethical approval for this study was obtained from the ethics committee of Ministry of Health, Khartoum, Sudan and from the ethics committee of Uppsala University. Results showed lower eosinophils and neutrophils count in the VL group (p=0.0001 and p=0.002 respectively). The ECP levels were highly elevated in the control serum as compared to VL (p<0.0001) while plasma levels of MPO highly elevated among VL (p<0.0001). Turnover of these cells in VL were highly increased as compared to controls (5 and 8-fold, respectively). Levels of IL-5, GM-CSF and IL-17 were significantly increased among VL (p<0.0001, p=0.017 and p=0.03 respectively), whereas the eotaxin and IL-8 levels were significantly lower (p<0.0001 and p=0.002 respectively). Positive correlations were found between IL-8 and the two cell markers (p<0.0001). We conclude that eosinophil and neutrophil turnover and activity are increased in subjects living in the rural areas of Sudan and those with VL, the turnover was even further increased. The relatively low secretary activity of eosinophils and neutrophils in patients with VL may relate to the reduced production and availability of the chemokines eotaxin and IL-8.

P31 Circulating immune complexes (IC) and IC-induced levels of GM-CSF are increased in Sudanese patients with acute visceral *Leishmania donovani* infection undergoing sodium stibogluconate treatment: implications for disease pathogenesis

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Infection with *Leishmania donovani* is associated with IL-10 as well as with GM-CSF. Immune complexes (IC) exert important functions by stimulation of monocytes/macrophage-mediated production of pro- and anti-inflammatory cytokines in rheumatic diseases. In this investigation, we have explored IC-induced cytokine production during *Leishmania* infection. Sera from 43 patients with visceral leishmaniasis (VL), 17 patients with post-kala-azar dermal leishmaniasis, and 20 healthy Sudanese controls were precipitated with polyethylene glycol (PEG). The PEG precipitates were added to serum-free PBMC for 20 h,whereupon supernatant levels of IL-1beta, IL-6, IL-10, IL-1 receptor antagonist protein, TNF-alpha, TNF receptor p75, and GM-CSF were investigated using ELISA. Circulating levels of C1q-binding IC were also measured in the serum samples. PEG precipitates from *Leishmania*-infected patients induced significantly higher levels of GM-CSF (p = 0.0037) and IL-10 (p < 0.0001), as well as of IL-6 (p < 0.0001) and IL-1 receptor antagonist (p = 0.0238) as compared with PEG precipitates from controls. Patients with acute VL as well as VL patients receiving sodium stibogluconate treatment displayed significantly increased levels of PEG precipitate-induced GM-CSF. The induction of GM-CSF by circulating IC was especially prominent in acute VL patients receiving sodium stibogluconate treatment for PEG precipitate-induced levels of GM-CSF (disease activity, p = 0.0006; treatment, p = 0.0005; interaction, p = 0.0046). Parallel associations were determined for C1q-binding immune complexes, but not for any cytokine other than GM-CSF. The importance of IC-induced GM-CSF in leishmaniasis warrants further study.

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RNA extraction from Lates calcarifer tissues post-infection by Cryptocaryon irritans

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Lates calcarifer (Asian sea bass) is one of the most vastly farm-cultured fish species in Malaysia due to its high demand and corresponding market price. Breeding the fish in sea cages may lead to infection by various marine pathogens, in particular, *Cryptocaryon irritans*. Cryptocaryonosis or 'white spot disease' may be fatal to most marine fish and we are attempting to understand the fish response to infection. In a preliminary study, fish (500-700g) were obtained from the sea cages with previous incidences of *C. irritans* outbreak and induced with ice twice a day to confer stress upon the fish. 200 cysts were utilized for the infection of each fish within the aquarium and approximately 3-14 days later, white spots were visible on the fish body. The fish were killed and liver, spleen, gills and kidney were removed and snap- frozen in liquid nitrogen. All the organs were then homogenized in TriReagent[®] and RNA was extracted following the manufacturer's protocol. Approximately 1.2 µg of total RNA was obtained from 0.1g of sample. The total RNA of all organ samples will subsequently be used in Quantitative Real-Time PCR assays whereby the expression of immune-related genes such as cytokines, chemokines, acute phase response proteins (APR) and antimicrobial peptides will be profiled.

P33 Effect of climate on helminthiasis in small ruminants in Perak

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A retrospective study of the faecal samples in small ruminants diagnosed for helminthiasis in relation to the weather conditions in eight Perak districts was done in 2008. Helminthiasis in Perak is commonly diagnosed at the Parasitology Unit of Veterinary Research Institute throughout the year. There is an urgent need to know the effects of climate, if any, on the increasing trend of helminthiasis samples diagnosed in the districts of Perak. The McMaster, floatation and larvae culture methods were conducted to diagnose helminths in faecal samples from sheep and goats. Animals with faecal egg counts (FEC) of 500 eggs per gram and above are classified as positive for helminth infection which would require treatment. More than 90% of the samples received showed helminthiasis caused by strongyles with the predominant species being *Haemonchus contortus*. Data on rainfall and humidity are correlated to the farm location in each district and further analysed according to parasitological status. Generally, weather data in the various districts of Perak, does not show any significant variations in mean annual rainfall (2584 mm) or humidity (85%) in a dristrict as they are high throughout the year. The infective larval stage of helminthias scases in some districts in Perak. Results show that the highest number of helminthiasis from Perak districts is 1239 samples. Efforts are also taken to help farmers reduce the helminthiasis infections in the farms by conducting anthelmintic resistance testing and providing extension services to alleviate the worm problem.

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Soil-transmitted helminthiasis in Malaysia: burden, associated risk factors and the need for a school based intervention

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Soil-transmitted helminths including Ascaris lumbricoides, Trichuris trichiura and hookworm continues to be prevalent and a public health concern in Malaysia despite efforts to control and reduce the worm burden of infections. Helminthiases result in iron deficiency anaemia, growth retardation, protein energy malnutrition and poor intellectual development on the infected individuals. Like in other parts of the developing world, children of school-age (6-15 years) are more prone to infection by STHs than other groups. Higher prevalence rates continue to persist among the rural aborigines, estate workers and in urban slums and squatters. The prevalence of *T. trichiura* remained higher (2.1%-98.2%) than other STHs. Ascaris lumbricoides followed closely with a prevalence rate of 4.6%-86.7%. Hookworm is the least with a prevalence rate of 0%–37%. Factors leading to higher prevalence include: poor socio-economic status, illiteracy, poor personal hygiene, environmental factors, overcrowding and sharing house with animals. To ensure an effective worm control in the country, it is suggested that, a national worm control programme should be introduced with a special emphasis on school based intervention.

P35 Chikungunya virus infection and transovarian transmission in laboratory strain of *Aedes aegypti* (L.) and *Aedes albopictus* Skuse

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Infection due to chikungunya virus has now become a public health threat to the Malaysian population since the national outbreak in 2008. *Aedes albopictus* was identified as the main vector in Johor. In order to investigate the ability of local vector mosquitoes to transmit this virus, laboratory studies were conducted to examine the susceptibility of local laboratory strain of *Aedes aegypti* and *Ae. albopictus* to chikungunya virus and the possibility of transovarian transmission of the virus. Reverse Transcriptase-Polymerase Chain Reaction (RT-PCR) was used to detect the presence of the virus in the mosquito. For each mosquito species, 200 females were orally fed with a human isolate of chikungunya virus in human blood via a membrane feeding system and maintained on sugar solution for 14 days at room temperature. Each species were dissected for salivary gland. RT-PCR results indicated that both *Ae. aegypti* and *Ae. albopictus* were infected by chikungunya virus up to 14 days post-feeding. *Aedes aegypti* is therefore a potential vector of chikungunya virus, while the vector status of *Ae. albopictus* was re-confirmed. The eggs from each mosquito species were hatched and screened for the presence of transovarian transovarian transovarian transovarian transovarian transovarian transovarian transovarian transmission of chikungunya virus by RT-PCR. No virus infection in larvae was detected, indicating absence of transovarian transmission of chikungunya virus in both mosquito species.

P36 Efficacy study of mosquito plant (*Pelargonium citrosum*) as repellent for *Aedes* mosquito

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Mosquito plant, *Pelargonium citrosum* from the family Geraniaceae is well known to repel mosquitoes. This study showed that *P. citrosum* is efficient significantly in repelling mosquitoes when the P value was 0.028 (P>0.05) by employing Pearson Chi-Square test. There was a difference in the landing rates of mosquitoes between areas with mosquito plant and areas without mosquito plant. Thus, this plant has of great potential to be used as an alternative control measures towards mosquito bites. This study also indicated that the *Aedes* mosquitoes show some degree of repellency to the plant during its peak biting period.

Distribution of dengue virus in *Aedes aegypti* mosquitoes by using RT-PCR in Medan, **P37** North Sumatera

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Dengue infection, whose transmission depends on several factors, i.e. virus, vector, environment and social cultures, is still a serious health problem in many tropical regions. Considering the profile of the vectors, Aedes mosquitoes are therefore available almost throughout Indonesia, mostly in urban areas. This study is conducted in 5 subdistricts in Medan, from where cases had been reported, to map the distribution of the four types of the virus in the field caught Aedes aegypti by using RT-PCR, a method that can immediately, sensitively, and specifically detect the type of the virus. The descriptive study was conducted from September to November 2008. From 328 samples, 8 are positive for serotype 1 (DEN-1) and 25 DEN-2. No DEN-3 and DEN-4 were detected.

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Simulated field thermal application of a synergised pyrethroid formulation against Aedes (Stegomyia) aegypti (L.) and Culex guinguefasciatus Say

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The use of water based pyrethroid formulations for space spray is becoming more popular because they are odorless, exhibit quick knock down and requiring only low dosage. This study was conducted to determine the effectiveness of a water based synergised pyrethroid formulation containing s-bioaalethrin-0.8% w/w, permethrin-18.7% w/w and piperonyl butoxide-16.8% w/w against Aedes aegypti and Culex guinguefasciatus by using thermal fogger. All the concentration with 33, 50 and 100 times dilution of synergised pyrethroid in water against Ae. aegypti and Cx. guinguefasciatus showed mortality ranged from 96.67 - 100% and 70.00 - 100%, respectively. The efficacy of synergised pyrethroid in all series of dilution against larvae of Ae. aegypti and Cx. quinquefasciatus after 24 hours continuous exposure ranged from 0 - 100% and 52.00 - 100%, respectively. This study shows that the synergised pyrethroid was effective against both species of mosquitoes, especially Ae. aegypti. However, the formulation shows better larvicidal effect against larvae of Cx. quinquefasciatus in comparison to Ae. aegypti.

P39 Comparative bioefficacy of the repellency of DEET and plant essential oil against dengue vector, *Aedes aegypti* (L.): A laboratory study

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This study was conducted to evaluate the effectiveness of a commercial repellent cream containing plant essential oil of citronella (0.20% w/w) and thyme (0.48% w/w). The evaluation was conducted according to the standard method of SIRIM. In this method, a standard repellent containing DEET (10% w/w) was used to compare the efficacy of repellent containing plant essential oils. Both human forehands (baits) treated with repellent containing plant essential oils and DEET, respectively, were exposed to *Aedes aegypti* in the cage for 3 minutes. The number of mosquitoes landing and/or biting was recorded during this period. The assessment period was 1, 2, 4, 6 and 8 hours post-application of repellent. At least 3 human baits were used and the tests were triplicated. The percentage means reduction of *Ae. aegypti* landing on human forehands treated with repellent containing plants essential oils and DEET were 82.50 \pm 3.39% and 88.48 \pm 1.04%, respectively. There was statistically no significant difference between the percentage reductions of both repellents (p > 0.167). This study indicated that repellent cream containing citronella oil and thyme oil was as effective as DEET and the repellent effects lasted for the test period of 8 hours under laboratory conditions.

P40 Evaluation of Insect Growth Regulators (IGRs) against field collected *Aedes aegypti* (Linnaeus) from Kuala Lumpur and Selangor, Malaysia

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The larvicidal activity of the 2 groups of insect growth regulator (IGR) namely, juvenile hormone mimic (methoprene and pyriproxyfen) and chitin synthesis inhibitor (diflubenzuron, novaluron and cyromazine) was evaluated against field collected *Aedes aegypti* larvae in the laboratory. Ovitrap collection was initiated in Kuala Lumpur (Lembah Pantai) and Selangor (Ampang and Serdang Jaya) to obtain field strains of *Ae. aegypti*. The IGR bioassay was performed according to the protocol of WHO. The larvae were exposed to a series of concentrations to obtain 50% and 90% emergence inhibition (EI). Novaluron was most effective against both strains of *Ae. aegypti* larvae, with El₅₀ ranged from 0.00004 - 0.00006 mg/L, followed by diflubenzuron (0.00015 - 0.00043 mg/L), methoprene (0.0049 - 0.019 mg/L;), pyriproxyfen (0.023 - 0.093 mg/L) and cyromazine (0.086 - 0.14 mg/L). The resistance ratio of novaluron, diflubenzuron and cyromazine against field strain *Ae. aegypti* was less than 1, implying absence of resistance in field *Ae. aegypti* against these 3 chitin synthesis inhibitors. However, resistance ratio for methoprene and pyriproxyfen ranged from 19.00 – 72.42 and 1.49 – 6.06, respectively, indicating presence of resistance in field *Ae. aegypti* against these 2 juvenile hormone. However, further study should be conducted to confirm the effectiveness of IGRs against *Aedes* larvae under field conditions.

P41 *Aedes aegypti* larval competition for space and nutrients in artificial containers: wild type versus RIDL OX513A and OX3604C strains

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Aedes mosquito-borne fevers such as dengue and chikungunya threaten over half the world's population including Malaysia. As current vector control programmes and ComBI cannot stop the spread of these diseases, scientists around the world are looking into the feasibility of deploying RIDL® Sterile Insect Technique (RIDL-SIT) as part of integrated vector control programmes. Two promising strains of RIDL Aedes aegypti - 'OX513A' and 'OX3604C' were used in this study. One feature of these strains is that affected progeny of released RIDL males and wild type females die late in development; these 'doomed' larvae potentially suppress other larvae in the same container by competition. We set out to test whether the strains are capable of inducing such suppression. We compared the RIDL strains with wild type in larval competition experiments, to quantify the effect of competition for space and nutrients in artificial containers. We placed varying numbers of heterozygous or homozygous RIDL-SIT and wild type larvae in small containers representing typical breeding sites and monitored their development. We found no significant reduction in competition for resources in heterozygous larvae of either strain, relative to wild type. Thus larvae of the RIDL-SIT strain would be able to compete with wild counterparts in the field, adding to the vector population reduction achievable using this technique. Some reduction in competitiveness of homozygous OX513A larvae was indicated, though further experiments would be required to quantify this. Since homozygous larvae would not arise in the field this would not affect field efficacy, though there may be implications for mass rearing of this strain prior to release. This study indicates that the RIDL strains examined are competitive as heterozygous larvae; theoretical studies have indicated that a late-acting lethal RIDL system could significantly improve programme cost-effectiveness via competition of 'doomed' heterozygous larvae with wild type. Our data indicate that the RIDL strains examined are likely to be able to achieve this beneficial effect.

P42 Efficacy effect of *Litsea elliptica* essential oil aerosol against *Aedes aegypti* (L) (Diptera: Culicidae) in the laboratory

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Incident of dengue fever have been reported dramatically over the past decade. The only way to decrease this incident is via eradication of *Aedes aegypti*, vector of dengue fever. The control of *Ae. aegypti* still basically depends on usage of synthetic insecticides. Now, most of the synthetic insecticides become less efficient due to resistance that occurs to the mosquitoes. It is timely to acquire insecticides that are less expensive, target specific, and easily biodegradable. This study has been done to determine the efficacy of *Litsea elliptica* essential oil that has been formulated against *Ae. aegypti* female mosquitoes. The essential oil was obtained by hydrodistallation method. Peet- Grady method with modification was performed to determine the efficacy of the aerosol. Results indicated the LC_{50} , LC_{90} and LC_{99} for *L. elliptica* aerosol at 20 minute (1753.540%; 64475.240%; 1218439%); 1 hour (21423.030%; 2888168%; 157437000%); 24 hours (2067743%; 85091520000%; 4922996x 10⁸%); 48 hours (67995.430%; 593682000%; 970303400000%); 72 hours (9413.091%; 88.053%; 12831240000%) respectively. The result showed significant different between the test aerosol and the positive control aerosol (p<0.05). It shows those essential oil that has been formulated and tested the efficacy is less efficient compared to the positive control. The tested aerosol is less efficient after the formulation into the aerosol.

P43 Repellency activity of essential oil from mosquito plant, *Pelargonium citrosum* and their major chemical compounds

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In an effort to protect man from being bitten by insects, a few methods have to be considered. The methods include the physical method such as using bed nets and wearing long sleeve clothes and/or chemical protection such as using repellent or cream products which are available in the market. These methods are totally against the adult mosquito. For this concern, repellency activity was to be taken into consideration. In this study, essential oil of *Pelargonium citrosum*, or locally known as Jeremin was tested its repellency property against *Aedes aegypti*. The repellency activity was assessed by using the test cage described in the American Society for Testing and Materials (ASTM) Standard E951-83 Laboratory testing of non-commercial mosquito repellent formulations on the skin. The essential oil was also examined by GC and GC/MS leading to the identification of their major chemical constituents. The result showed that the median effective concentration (EC₅₀) value of the essential oil was 0.0310 mg/cm². From our previous study of the same species, this result showed weaker repellency property than before, and we assumed it's due to the less yield percentage of the active chemical constituents that contribute to the repellency property.

P44 Vertical oviposition patterns of *Aedes* mosquitoes in urban high-rise apartments in Kuala Lumpur

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The vertical oviposition pattern of *Aedes aegypti* and *Aedes albopictus* was studied using ovitraps located at every floor of a high rise apartment which is 60 meters in height. The ovitraps were positioned outdoors in staff quarters of Kuala Lumpur Hospital. Of the 429 ovitraps set up, 115 (26.8%) were positive with *Ae. aegypti* (1194) and *Ae. albopictus* (245) larvae. Data from different elevations showed that significantly more *Ae. aegypti* eggs were collected at 0-2.8 meter (ground floor) elevations than any other elevation. *Aedes aegypti* larvae were collected from every floor of the apartment up to 20th floor. The results suggested that the invasion of high rise ecosystems by *Ae. aegypti* can enhance transmission of dengue. This ecological shift in the *Ae. aegypti* population exploiting new habitats associated with human activity, suggested that strategies should be developed to educate householders as well as creating appropriate vector control measures to prevent future threats of dengue transmission in high rise buildings.

P45 Larviciding practices in the prevention and control of dengue fever in urban community

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Despite the effortless control activities done by the local council and the District Health Office, continuous outbreak of dengue cases still occurs in the area of PJS3 and PJS4, Petaling Jaya, Selangor. This has prompted a study to be done in this area to look at the larviciding practices by the community to determine the larviciding practices among the urban community in the prevention and control of dengue fever in the area of PJS3, Taman Sri Mania, Petaling Jaya, Selangor. A Cross sectional comparative study design was conducted among 524 respondents aged 18 to 80 years old. A stratified systematic random sampling was used in this study in which the area of PJS3 was divided into 3 strata and systematic random sampling was used for each stratum in which every 2 houses was selected. Structured close ended self administered questionnaires were used to collect data. Almost three quarter of respondents have heard about laviciding and majority of them (67.35%) knew about larviciding from the media. Only 60.46% of the respondents know how to use the larvicide and most of them (89.12%) know the location of larva breeding site. About 79.09% of respondents will use the larvicide if it is easy to get. More than half of the houses of respondent (59.73%) have not been checked for larvae. Furthermore, about 75.76% of the respondents thought that the larvicide was useful. The prevalence of larvicide use was 26.9 %. There was a significant difference in the knowledge (K), attitude (A), practice (P) and total KAP score between user and non-users of larvicides. Awareness level among the community was still low: the community was not well informed about the use of larvicide and no proper knowledge of larvicide and larviciding. The practice of larviciding in the community of PJS3, Taman Sri Manja, Petaling Java as a method of mosquito breeding control and prevention of dengue fever was not satisfactory. Furthermore, larvicide was not easily available to the community.

A pilot study on knowledge, attitude and practice of larviciding in the prevention and control of dengue fever in Shah Alam 2009

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Increasing dengue cases were reported nationwide in the first guarter of 2009 and Section 7, Shah Alam, Selangor reported the highest number of cases in Malaysia for two consecutive weeks which has prompted a study to be done in this area to assess the knowledge, attitude and practice among the community about larviciding in the prevention and control of dengue fever. A Cross sectional comparative study design was conducted among 534 respondents aged > 18 years old. A stratified random sampling was used in this study in which the area of Section 7 is divided into 3 stratas and systematic random sampling was used for sampling. A structured close-ended, self administered questionnaire was used to collect data. About 70% of respondents have knowledge of larvicide use. More than half of the respondents (53%) expressed difficulty in getting larvicide. About 67% of respondents agreed that larviciding is effective in control and prevention of dengue fever. Majority (91%) of the respondents thought that larvicide was useful and most of them would use larvicide if it was easy to get or buy. Respondents suggested that larvicide should be sold in shops (55%), pharmacy (17%) and supermarket (14%). However, only 35% of them have ever used larvicide and among them, less than half were able to properly demonstrate how to use larvicide. As a summary, only 15% of respondents are practicing proper larviciding in order to eradicate mosquito breeding site inside and around their houses. The prevalence of larvicide use was 35%. There was a significant difference between non-larvicide user and larvicide user in the knowledge (K), practice (P) and total KAP score but not in the attitude (A) score. It is concluded that appreciation and practice of larviciding in the community of Section 7 Shah Alam as a method of eradication of mosquito breeding site and prevention of dengue fever are not satisfactory. The community was not adequately educated with proper knowledge of larvicide and larviciding, and larvicide was not easily available to the community.

P47 Repellency of Mostique EGX-101^(R) lotion on *Aedes albopictus* (Skuse) (Diptera: Culicidae) in the laboratory

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The repellent activity of Mostique EGX-101^(R) against *Aedes albopictus* was investigated under laboratory conditions with human volunteers. The percentage repellency increased from 24.6% to 81.4% as the Mostique EGX-101^(R) concentrations increased from 1% to 10% respectively. The positive controls, neem oil 2% and deet 0.1% indicated percentage repellency of 86.3% and 93.9% respectively. The percentage protection of EGX-101^(R) applied at 10% indicated a 100% against *A. albopictus* after 1 hour and 88.1% after 2 hours post-treatment. At 7.5% percentage protection was 94.3% after 1 hour and 78.6% after 2 hours post-treatment. The positive controls, neem 2% indicated 100% protection after 1 hour and 92.9% after 2 hours post-treatment; deet 0.1% indicated a protection of 100% after 1 hour and 95.2% after 2 hours post-treatment. In conclusion EGX-101^(R) has the potential to be an effective repellent against dengue vector *Aedes albopictus*.

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Investigating differentially expressed proteins of dengue virus 2- infected vector cells

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Dengue has been recognized as a critical global endemic emerging worldwide throughout tropical and subtropical regions; the natural habitat of its principal transmission vector, *Aedes* mosquitoes. Gaining insight on the cellular mechanism of the disease vector will prove to be invaluable in the hopes of eradicating dengue. The study of viral pathogenesis has been revolutionized with the advent of proteomics and its development. We present here our preliminary testing for the potential application of a non gel-based proteomics approach to study the cellular mechanisms of dengue virus infection in the *Aedes albopictus* mosquito (C6/36) cell lines. Dengue virus serotype-2 were propagated and harvested from C6/36 cell lines. Tissue culture infective dose (TCID₅₀) of the virus stock was then determined for ensuing virus infection analyses. C6/36 cells were then subjected to either infection or mock infection of the dengue virus and cell viability was determined through MTS assay in order to ascertain the optimum infection incubation period while excluding apoptotic and necrotic factors. Whole cell protein extracts from infected and mock infected cells will then be subjected to protein characterization by liquid chromatography linked tandem mass spectrometry (LC-MS/MS) and matrix assisted laser desorption ionization–time of flight (MALDI-TOF/TOF).

P49 Survey of mosquito larvae distribution in container habitats in dengue outbreak areas in Malaysia

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Rapid urbanization in Malaysia has resulted in the creation of conducived habitats for cosmopolitan vector mosquitoes. Artificial containers are the most important factors determining the breeding of mosquito larvae especially *Aedes* since such containers are the major larval habitats in and near human habitation. The objective of this study was to investigate physico-chemical parameters of the water in the containers and to identify mosquito larvae found in various container habitats. The survey covered container breeding habitats ranging from tyres, plastic containers, tins, polystyrene to other discarded containers surrounding the housing estates and shop houses in dengue outbreak areas. Waste tyres are a well known major breeding site of both *Ae. albopictus* and *Ae. aegypti*, followed by discarded plastic containers and tins. Both *Ae. albopictus* and *Ae. aegypti* was selective in selecting oviposition sites compared to *Ae. albopictus* and preferred partially shaded areas. Water in the artificial containers with moderate pH (6.4-7.6), relatively high dissolved oxygen (72.2-91.9%), conductivity (453-459 µScm⁻¹) and temperature (25-31°C) are suitable breeding habitats for these mosquito species.

P50 Microalgal species associated with the potential mosquito vector breeding habitats in Pos Lenjang, Kuala Lipis, Pahang

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Most phytoplankton are suitable food for many species of mosquito larvae, however, some are harmful. A study was carried out to investigate the correlation between presence of specific microalgal species and potential vector larval breeding habitats. The water samples were collected from the larval breeding sites during the mosquito larval surveys. A total of 92 species of microalgae were identified. *Micrasterias* sp., *Euglena* sp., *Chorella* sp. and *Synechococcus* sp. were particularly common. *Anopheles maculatus* was recorded in association with most algal species except in *Arthrospira* sp., *Botryococcus* sp., *Eunotia* sp., *Genicularia* sp., *Melosira* sp. and *Surirella* sp. infested breeding sites. Only *Anopheles* species were found in samples containing *Anabaena* sp. The physico-chemical parameters for the breeding habitats range a between 9-171 µScm⁻¹ for conductivity, 1.9–8.8 mgL⁻¹ for dissolved oxygen, 4.75–8.25 for pH and 22-32°C for temperature.

P51 The effect of *Piper aduncum* Linn. (Fam: Piperaceae) essential oil as aerosol spray against *Aedes spp*

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The efficacy of *Piper aduncum* L. essential oil formulated in aerosol cans was evaluated against dengue vectors in a room. The aerosol spray test was based on Malaysian Standard (MS 1221:1991UDC 632.982.2) for aerosol spray testing methodology and examined the knockdown effect within 20 minutes exposure. Mortality rate after 24 hour of holding period was also determined. Our result showed that the knockdown effect of commercial aerosol spray (0.09% prallethrin 0.05% d-phenothrin) and *P. aduncum* essential oil spray (8% and 10% concentrations) was significantly higher in *Aedes (Stegomyia) albopictus* (Skuse) adult females, when compared with that of *Aedes (Stegomyia) aegypti* (L.) adult females (p<0.05). There was a significant difference in knockdown between commercial aerosol spray and essential oil spray for both *Aedes spp* (p<0.05). The mortality was significantly higher in *Ae. aegypti* which gave 80% mortality, when compared to *Ae. albopictus* which indicated 71.6% mortality (p<0.05). The commercial aerosol spray indicated 86.5% and 97.7% mortalities against *Ae. albopictus* and *Ae. aegypti* respectively (p<0.05). Based on these data, *P. aduncum* essential oil has potential to be used as aerosol spray against *Aedes spp*.

P52 A study on 48 hours ovitrap to measure the effectiveness of integrated vector control in Kinta District

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The number of dengue cases in Kinta District was the highest in Perak. Thus, several measures were conducted to curb the problem. Among those was 48 hours ovitrap which was implemented to determine the effectiveness of the integrated vector control dengue localities. The objective of this study was to ensure the integrated vector control were carried out correctly by the detection of *Aedes* mosquito eggs within 48 hours after the initial fogging either indoor or outdoor. Thirty-seven (37) localities were selected randomly in these studies. It was conducted throughout the year 2009. All selected localities are the dengue hot spot areas. From each locality, 20 residential premises were selected randomly for the placement of ovitraps. The ovitraps were placed outdoors after fogging activities was done and collected after 2 days. They were replaced with new ovitraps each time after collected. Ovitraps were further incubated for 5 days and would be examined subsequently. Only 1 locality was found positive for Aedes after 48 hours of the initial fogging (2.7%). However, 22 localities (59.5%) were found positive after 1 week of the initial fogging. The median numbers of positive ovitraps were 2 in each locality. The integrated vector control was found more effective in the first 48 hours after the initial fogging compared to a week later after the initial fogging.

P53 Community awareness related to transmission, treatment and prevention of malaria in aboriginal and rural endemic areas, Peninsular Malaysia

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Focus on participation of community with improvement of their awareness of transmission, treatment and prevention of malaria can greatly increase the realization and sustainability of malaria control program. This was a cross-sectional study, and households were randomly selected. In total, 100 (aboriginal) and 123 (rural) households respondents were interviewed either house-to-house or in outpatient clinics in the forest and rural areas, respectively, using semi-structured questionnaires. Knowledge about malaria transmission was significantly higher among the rural respondents (x^2 =10.006, P=0.002). There is no significant difference between both communities in treatment-seeking behavior (x^2 =2.935, P=0.087). However, the use of alternative medicine, in particular, medicinal plants and beliefs in witchcraft and sorcery in the treatment of febrile diseases were significantly higher among aboriginal population (x^2 =16.980, P<0.001). As for the understanding of effective preventive measures, the use of mosquito bednets was not significantly influenced by type of population (x^2 =0.034, P=0.853). However, the knowledge and practice of different preventive measures to combat malaria such as insecticide and elimination of breeding areas was significantly higher among rural population (x^2 =23.136, P<0.001). In conclusion, both communities were aware of malaria as a disease, but knowledge, practices and attitudes were inadequate. Providing efficient health education to people residing in malaria endemic areas would improve their understanding about malaria prevention in order to achieve the elimination of malaria from the country.

P54 Cloning and expression of *Plasmodium knowlesi* merozoite surface protein (MSP-1₁₉) in yeast *Pichia Pastoris*

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Among the several simian malaria species, *P. knowlesi*, is emerging as an important human pathogen. The objective of this study is to obtain pure recombinant protein for diagnosis purposes. We are targeting to express the C-terminal part of the Merozoite Surface Protein-1 (MSP-1₁₉) of *P. knowlesi* in yeast *Pichia pastoris* expression system to obtain the recombinant protein. MSP-1 plays a role in the invasion of red blood cells and antibodies against MSP-1 of *Plasmodium falciparum* was found to implicate protective immunity. The target gene was amplified by PCR and the 330bp product was cloned into pPICZáA vector. The construct was then transformed into *P. pastoris* using EasyComp Kit (Invitrogen). The transformant was screened and positive clones were put into expression. A 12kDa protein was observed in the Western Blot after expression for 4 days.

P55 Prevalence of *Plasmodium knowlesi* among wild macaques (*Macaca fasicularis* and *M. nemestrina*) from Selangor Malaysia determined by nested-PCR of the SSU rRNA

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The present study aimed at employing molecular tools to detect the presence of *Plasmodium knowlesi*, a zoonotic pathogen, among wild urban macaques (*Macaca fasicularis* and *Macaca nemestrina*) in the state of Selangor, Malaysia. We used *Plasmodium* genus-specific primers for initial amplification of the SSU rRNA and a subsequent nested-PCR with specific primers which targeted *P. knowlesi*. Blood samples were collected from 125 wild macaques (18 *M. nemestrina* and 107 *M. fasicularis*) from various areas in Selangor, Malaysia. Giemsa-stained thin blood films (TBF) were prepared, and nested-PCR was performed. The molecular prevalence of *Plasmodium*, determined by PCR, was 64.5% for *M. fasicularis* and 100% for *M. nemestrina*. When *P. knowlesi*-specific amplification was carried out, the pathogen was detected in 23.3% and 5.6% of the blood of *M. fasicularis* and *M. nemestrina*, respectively. These results indicate that local wild macaques harbor a high rate of infection with *Plasmodium*. In addition, the prevalence of *P. knowlesi*, the zoonotic malaria parasite is higher than previously assumed. This warrants further investigation as the wild urban macaques which are in close contact with humans, may be important reservoirs of human malaria in Malaysia.

Current situation of malaria in Yemen

P56

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Malaria remains a major public health problem causing mortality and morbidity in tropical and subtropical countries. A crosssectional study was carried out to determine malaria prevalence and its clinical pattern during malaria season in Yemen. Blood samples were collected from 511 symptomatic patients, aged between 1 and 85 years, who voluntarily participated in this study, of them 268 were males, 242 were females and one whose sex was not stated. Malaria was screened using Giemsa-stained thick and thin blood films. Clinical profile was recorded through physical and laboratory examinations and biodata were collected by pre-tested standard questionnaire. The overall prevalence was 15.3%. Three malaria species (*Plasmodium falciparum, Plasmodium vivax* and *Plasmodium malarae*) were detected with the predominance of *Plasmodium falciparum* (83.33%). People living in the rural areas had higher infection rate compared to urban areas (P < 0.005). Children were at higher risk of developing severe malaria compared to adults (P < 0.05). Severe anaemia was significantly associated with young malaria patients (P < 0.05). Respiratory distress, jaundice, convulsion and bleeding were non-significantly more apparent among younger age groups of malaria cases. The study indicates that malaria is still a public health problem with children being at higher risk of developing severe malaria which may lead to death.

P57 Live *Brugia pahangi* immature female worm recovered from the subconjunctiva of a patient in Malaysia

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This is a first diagnosed case of human eye infection with *Brugia pahangi* in Malaysia. The patient was a 24-year-old Malay female sales representative from Sg. Buloh, Selangor. She presented with complaints of persistent right eye redness for one month, associated with mild pain. Eye examination showed her visual acuity was 6/6 in both eyes; the right eye conjunctiva was hyperemic with no discharge. Slit-lamp examination revealed multiple entangled tiny transparent thread-like mobile worms at the superior subconjuctival level. Two live worms were removed from the eye through a small incision of the conjunctiva under topical anesthesia. Systemic examinations were all normal. Her full blood count revealed no eosinophilia. Blood examination for microfilaria at midnight was negative. Serological test for filariasis was negative. The worm was identified as immature *Brugia* species on microscopic examination. The species of the worm was confirmed as *B. pahangi* by PCR targeted at the cytochrome oxidase I (COXI) gene. DNA sequencing of the amplicon showed 99% similarity with *B. pahangi* COXI sequence (Genbank accession no EF406112.1). The patient was cured three days after worm removal. On further questioning, she said that she had four cats at home. One of the cats was found to be positive for microfilariae of *B. pahangi*. The present case suggests that *B. pahangi* can cause ocular infection. As to whether zoonotic *B. pahangi* could cause lymphatic filariasis in human as observed in zoonotic sub periodic *Brugia malayi* remains a puzzle.

P58 Parity status and filarial infection in field-collected *Armigeres kesseli* Ramalingam and *Armigeres salbabatus* Coquillett from forested and suburban areas in Malaysia

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The ability of *Armigeres* to transmit diseases such as animal filiariasis and lymphathic filiariasis has been well documented in many parts of the world. The present study was to detect the presence of filiarial worm and parity rate in Malaysian *Armigeres* in a sub-urban area, Sungai Penchala, Kuala Lumpur (3°10'0"N, 101°38'0"E) and a forested area, Taman Alam, Selangor (3°20'24.69"N, 101°14'33.87"E) in Malaysia. Human landing catches (HLC) were conducted for 24 hours (0700-0700) during the study for three different nights. The *Armigeres* obtained from both sites were identified and dissected to determine their parity status. A total of 451 *Armigeres* adults were obtained from November 2009 to January 2010. After dissection, 63.40% of *Armigeres* obtained from Sungai Penchala were parous (*Ar. kesseli* = 34.31%; *Ar. salbabatus* = 29.09%). On the other hand, 75.87% of *Armigeres* obtained from Taman Alam were parous (*Ar. kesseli* = 48.28%; *Ar. salbabatus* = 27.59%). No filarial worm was detected in this study, but the potential for these mosquitoes to harbour pathogen is still high due to the high densities of parous mosquitoes obtained from the population.

P59

Isolation and identification of mites from an animal holding facility

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Some mites are known to elicit allergic diseases such as asthma, rhinitis and atopic dermatitis in humans. Animal models such as rats are used to mimic human disease processes. Environmental monitoring of experimental animal models exposure to dust mite allergens, for example in an animal holding facility, is important for the understanding of the relationship between exposure and development of allergic diseases. This study aimed to determine the distribution of different mite population densities and species composition in an animal holding facility, and to isolate and identify the mites based on morphological characteristics. In November 2009 to February 2010, dust samples were collected from floor, table, animal bedding, animal, food pellets, food storage container and air conditioner unit with a vacuum cleaner, which is adapted with a chamber that collects dust onto a filter paper. Dust samples were obtained by vacuuming an area of 900 cm² for 2 minutes. Mites on the filter paper were observed under a stereomicroscope and picked up with a fine needle. The mites were cleared in lactic acid and mounted on microscope slides in Hoyer's medium. Mites were then identified based on morphology and taxonomy keys. Mites that were found in animal holding facility were Aleuroglyphus ovatus, Austroglycyphagus malaysiensis, Suidasia species, Tarsonemidae, Cheyletidae and Mesostigmata. No report on A. ovatus was documented so far in Malaysia. Aleuroglyphus ovatus was reported in Australia, Argentina, Brazil, China, Egypt, India, Nigeria, Russia, Spain and USA. Aleuroglyphus ovatus, A. malaysiensis, Suidasia sp. are storage mites and are known to be important allergens. Tarsonemidae mites are fungal-feeding mites while mites such as Cheyletidae are predators to other mites and cause sensitization. Mesostigmatid mites are predators or parasites, which cause allergies as well.

P60

Potential use of lemon grass (*Cymbopogan nardus*) and neem (*Azadirachta indica*) as anti dust mites

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This experiment was undertaken to examine the acaricidal effects of essential oil of lemongrass (*Cymbopogon nardus*) and ethanolic neem leaf extract (*Azadirachta indica*) at different concentrations (50%, 25%, 12.5%, 6.25% and 3.125%) and exposure times (24 hrs, 48 hrs, 72 hrs and 96 hrs) on house dust mites *Dermatophagoides farinae* and *D. pteronyssinus*. The topical and contact activities of the two herbs were studied. In topical activity, 2 µl of herbal extracts were applied direct to the dust mites while in contact activity, mites were exposure to Whatman No.1 filter paper impregnated with herbal extract solution. Lemongrass extract was better than neem for both topical and contact activities. At 50% concentration, both 24 hrs topical and contact exposures to lemongrass resulted in more than 90% mortalities for both species of mites. Mortalities decrease with decreasing concentrations of lemongrass but increasing the exposure time did not apparently increase mortalities. 50% neem at 24 hrs topical exposure produce higher mortalities in *D. pteronyssinusi* than *D. farinae;* however the mortalities were similar for 24 hrs contact exposure at that concentration. Mortalities generally decrease with decrease with topical and contact activities. Increasing exposure times, increased mortalities for both activities for both topical and contact activities.

P61 Effect of a commercial air ionizer on dust mites *Dermatophagoides pteronyssinus* and *Dermatophagoides farinae* (Acari: Pyroglyphidae) in the laboratory

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This study evaluated the effect of a commercial ionizer on 2 species of house dust mite, *Dermatophagoides pteronyssinus* and *Dermatophagoides farinae* in a laboratory, using a special designed test set-up. Mortality of dust mites was assessed after 6, 16 and 24 hours for direct exposure and after 24, 36, 48, 60 and 72 hours for exposure in simulated mattress. New batches of mites were used for each exposure time. After 6 hours exposure, 39 and 7% mortalities were recorded for *D. pteronyssinus* and *D. farinae*, respectively. About 63% and 45% mortalities were recorded for *D. pteronyssinus* and *D. farinae*, respectively. About 63% and 45% mortalities were recorded for *D. pteronyssinus* and *D. farinae*, respectively after 16 hours exposure. Mortalities then increase to 82% and 71% for *D. pteronyssinus* and *D. farinae*, respectively after 24 hours exposure. Exposure after 24 hours in simulated mattress shows 6 and 7% mortalities for *D. pteronyssinus* and *D. farinae*, respectively. About 16 and 21% mortalities were recorded for *D. pteronyssinus* and *D. farinae*, respectively after 36 hours, and then followed by a gradual increase in % mortalities with each succeeding 3 days interval. At the end of 72 hours exposure in simulated mattress, 29 and 65% mortalities were recorded for *D. pteronyssinus* and *D. farinae*, respectively. Controls for both species of mites have 0% mortality throughout the study. LT₅₀ for direct exposure of ionizer was achieved after 10 hours and 18 hours for *D. pteronyssinus* and 72 hours (3 days) for *D. farinae*.

P62 First report of *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelidae) as a museum insect pest in Malaysia

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This paper reports the infestation of psocid, *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelidae) in the insect museum at Medical Entomology Unit, Institute for Medical Research, Kuala Lumpur. These tiny organisms were recognized as museum insect pest and found frequently in the insect boxes containing mosquitoes, flies, cockroaches and butterflies. They feed on dead insect specimens and cause severe physical damages to the valuable reference specimens many collected in the early 20th century. Hence, it is important to control their population immediately to prevent them from causing further deterioration to the museum collection.

P63 Morphological description of second and third instar larvae of *Hypopygiopsis violacea* Macquart (Diptera: Calliphoridae), a forensically important fly in Malaysia

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Hypopygiopsis violacea, a species of fly of forensic importance, was recovered from a corpse and described for the first time. The morphological structures of the second and third instar larvae of four specimens were examined using light microscope. Observations were focused on three main morphological characters: cephalopharyngeal skeleton, anterior and posterior spiracles. Cephalopharyngeal skeleton of second instar larva is darkly pigmented and without accessory sclerite below the mouth hook. The anterior spiracles of second and third instar larvae have 8-9 papillae each, arranged in a single row. The posterior spiracle of second instar larva has two spiracular slits with no thickening of peritreme. This differentiates it from the third instar, whereby the latter has three slits for each posterior spiracle. Cephalopharyngeal skeleton of third instar larva is heavily pigmented. An accessory sclerite is found below the hook part of third instar larva but is absent in second instar. Peritreme of the posterior spiracle of third instar larva is thick almost completely encircling a button. The intersegmental spines of the cuticular surface are dome-shaped and unicuspid. Third instar larva of this species is large with size approximately 15 mm long. These findings provide important identification features of immature stages of *Hy. violacea* which could be useful in forensic entomology.

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The ecology of forensically important flies in Iran and Malaysia

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Forensic entomology has become relatively common in criminal investigation, but gaps in knowledge of local dipterans taxonomy have become apparent. The blowfly (Calliphoridae) and fleshfly (Sarcophagidae) are two forensically important flies. This is a preliminary study which was conducted to ascertain and compare forensically important fly in Iran and Malaysia. Four species were forensically important and identified in Iran: *Lucilia sericata, Calliphora vicina, Sarcophaga crassipalpis* and *Sarcophaga (Bercaea) africa. Sarcophaga (Bercaea) africa* is a new record for species of Sarcophagidae in Iran. Likewise, the dipteran fauna found during this study in Malaysia were *Chrysomya megacephala, Chrysomya rufifacies* (Macquart), and Sarcophagidae. Another purpose of this study was to investigate seasonal and regional differences in maggot growth rate (PMI) in carrion blow fly and flesh fly of Iran and Malaysia. The blowfly, *Lucilia sericata*, in Iran showed a longer PMI (6-7 days) compared to *Chrysomya megacephala* (3-4 days) for it to grow up to L3 stage in Malaysia.

P65 Development rates of blowflies, *Chrysomya megacephala* (Fibricius) and *Chrysomya rufifacies* (Macquart) feeding on gasoline-exposed tissue: Effect of gasoline in estimation of postmortem interval

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Analysis of the insect colonizers of corpses can be a valuable forensic tool in the determination of post-mortem interval (PMI) which can eventually aid in criminal investigations. Until recently, most developmental data have been obtained from the Diptera (true flies), particularly blow flies of Calliphoridae since they are believed to be the first to be found on a decomposing corpse. In addition, the study of these fly larvae found on corpse can also contribute to the identification of drugs or toxins present in the corpse. This study was conducted to examine the effect of gasoline on the growth of blowflies, *Chrysomya megacephala* and *Chrysomya rufifacies*. A total of 36 adult Sprague-Dawley rats were divided into 6 equal groups which were given orally 0 (control), 9.5, 18.85, 37.7, 75.4 and 150.4 ml/kg body weight of gasoline, respectively. Both *C. megacephala* and *C. rufifacies* larvae were then allowed to grow on the liver of carcasses. Results indicated that control larvae from both species developed more rapidly than larvae feeding on tissue containing gasoline. The time required for adult emergence of both species was greater for gasoline-exposed colonies. Control larvae of *C. megacephala* and *C. rufifacies* and *C. rufifacies* also had the highest survival rate compared to larvae exposed to the five different doses of gasoline. From GC-MS analysis, principle components of gasoline were also detected in *Chrysomya* larvae samples. It is concluded that the presence of gasoline altered the development rate of *C. megacephala* and *C. rufifacies* and thus disrupt normal postmortem interval (PMI) estimation.

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Influence of larval density and food substrates on the development of blowfly, *Chrysomya megacephala* (Diptera: Calliphoridae)

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In forensic entomology, the developmental time of fly is always used to estimate the post mortem interval (PMI). Therefore, factors which may affect the larval size and growth rate may have implications for reliable PMI determinations. This study investigates the competitive effects of larval density on forensically-important blowfly, *Chrysomya megacephala* reared on four types of beef tissues. Cultures with different degrees of larval density were established by transferring 5, 50 or 100 newly hatched larvae to different food substrates either beef's liver, lung, spleen or muscle. An indication of developmental time was obtained by recording the length of time from egg to adult emergence. Analysis of the data from all sets of food substrates indicated that the development of larvae was significantly faster in the cultures contained 100 larvae, followed by cultures of 50 larvae and 5 larvae (p<0.05). However, there was no significant difference in developmental time from eggs to emergence of flies fed on different food substrates. In conclusion, the competitive feeding environment within the more crowded larval cultures resulted in increased development rates. We therefore recommend forensic practitioners to note the presence of maggot density from which larvae are collected as this has obvious implications for post-mortem interval determinations.

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Prevalence of protozoan diseases in local horses in peninsular Malaysia

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A prevalence study on the protozoan diseases of equines such as Surra (*Trypanosoma evansi*), anaplasmosis (*Anaplasma phagocytophila*) and piroplasmosis (*Babesia equi* and *Babesia caballi*) was carried out on local horses in Peninsular Malaysia by the Veterinary Research Institute. Blood and serum samples of equines were received from 12 states throughout Malaysia. This study was done to establish the incidence of positive infections by the thin smear, microhaematocrit centrifugation and serological techniques such as competitive ELISA and Indirect Flourescent Antibody Test. A total of 887 blood and 238 serum samples were tested for Trypanosomiasis and results showed that all blood samples were negative by thin blood smear and buffy coat examination. However, 12% were positive serologically. All blood samples were also tested for piroplasmosis and the results were negative. A total of 380, 180 and 100 serum were tested for *Babesia equi*, *Babesia caballi and Anaplasma phagocytophila* respectively. There were 20%, 1% and 0% positive for *B. equi*, *B. caballi and Anaplasma phagocytophila* respectively. There were 20%, 1% and 0% positive for *B. equi*, *B. caballi and Anaplasma phagocytophila* respectively. There were 20%, 1% and 0% positive for *B. equi*, *B. caballi and Anaplasma phagocytophila* respectively. This information forms the basis for disease control programs for international events whereby imported horses are assured of a safe and comfortable stay in Malaysia especially when competing with local horses. Consistent screening and early prevention in local horses can be carried out to minimize the risk of disease. As the vectors and reservoir hosts for these diseases are widely distributed in Malaysia, eradication of these diseases in not possible but with stringent precautions, the disease outbreaks or spread can be minimized.

P68 Evaluation of antioxidant activities of *Hibiscus sabdariffa* variety Arab, UKMR-1 and UKMR-2

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Hibiscus sabdariffa is a member of the Malvaceae family. It has abundance of natural phenolic compounds such as anthocyanin and is believed to have the potential as a natural source of antioxidant. The origin of *H. sabdariffa* is not fully known, but in Malaysia it was introduce in the early 1990s by the Department of Agriculture in Terengganu. Universiti Kebangsaan Malaysia in cooperation with Malaysian Nuclear Agency has initiated mutation breeding programme to generate new genetic variability of *H. sabdariffa*. New varieties named UKMR-1 and UKMR-2, have been produced using Arab variety as the parent variety in a mutation breeding. The study to evaluate the antioxidant activities of these new varieties were conducted to determine whether the new varieties also have potential as a natural source of antioxidant. The antioxidant activities of UKMR-1 and UKMR-2 variety were determined using three assays, namely, the DPPH radical scavenging assay (to analyze the capabilities of the extracts to scavenge DPPH radicals), reducing power assay (to analyze the capabilities of the extracts to reduce Fe3+/ ferricyanide to ferrous form, Fe2+) and metal chelating assay (to determine the abilities of the extracts to chelate Fe2+ ion). Results from this study showed that both UKMR-1 and UKMR-2 are potential sources of antioxidant. However, between the two, UKMR-2 showed a better antioxidant activity. Investigation need to be carried out to further evaluate their antioxidant potential with different assays.

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Screening of Ervatamia coronaria medicinal plant for antioxidant activity

Heida Nadia Zulkefli and Nurhayati Zainal Abidin

Division of Biohealth Science, Biological Science Institute, Faculty of Science, 50603, University Malaya

In recent years the use of natural antioxidants has been promoted because of concerns regarding the safety of synthetic ones. Antioxidants play an important role protecting against damage by reactive oxygen species. This study focused on flowering plants called *Ervatamia coronaria* from family Apocynaceae which is widely distributed in tropical country. This plant exhibited anticancer, ant-inflammatory and antimicrobial activity but not many researches have been carried out regarding anti-oxidant activity of this plant. Attention has been drawn to screen the antioxidant activity of roots, stems and leaves of *E. coronaria* using various solvent systems such as petroleum ether, methanol, chloroform and water. DPPH radical scavenging assay, metal chelating assay and reducing power assay were carried out to evaluate antioxidant potential of the extracts. The IC₅₀ for standard reference ascorbic acid in DPPH radical scavenging assay was 0.004 mg/ml. The assay showed that the crude chloroform extracts for roots, crude methanol extracts for stems and leaves exhibited active scavenging activity on the stable 2,2-diphenyl-1-picrylhydrazyl with IC₅₀ 0.160, 0.176 and 0.162 respectively. In the metal chelating assay, crude water extracts for roots and chloroform extracts for stems showed active inhibition of the formation ferrozine-Fe²⁺ complex in 1 mg/ml with 92.62% and 85.34% respectively when compared to standard reference ethylenediaminetera acetic acid which was 98.51%. However, all the extracts showed low activity in reducing power assay when compared to standard reference ethylaned to standard reference butylated hydroxyanisole. The extracts of *E. coronaria* that showed active antioxidant activity are worth of further investigation in order to identify the active compounds.

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Growth profile of selected actinobacteria isolate and their antitrypanosomal activity

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Two selected actinobacteria isolates were grown in two different fermentation media (M3 and M2) and the culture broth were extracted with buthanol solvent and harvested for *in vitro* antitrypanosomal activity against *Trypanosoma brucei brucei* strain BS221. Growth profile of isolates A032 and A039 which exhibited strong antitrypanosomal activity (IC_{50} value 0.22 ug/ml) and moderate (IC_{50} value 4.14 ug/ml) were studied. The fermentation process was monitored for ten days and duplicate flasks were harvested every day. Data on wet weight biomass, pH and bioactivity were also collected each day. The cell growth reached its maximum on day 3 for isolate A032 and day 7 for isolate A026. The broth pH profile for both isolates which initially were neutral became alkaline during the growth phase until the end day of fermentation. Antitrypanosomal activity was observed after 32 h of fermentation for isolate A032 and 96h for isolate A039. Results from growth profile study will be used as a basis for selection of harvest day during large scale fermentation for bioactive compounds purification.

No	Title	Name and Address
PS1	Hemagglutinin (Ha) Gene Of The Highly Pathogenic Avian Influenza (HPAI) H5N1 Virus Has Not Changed	DR. KAMARUDDIN MD ISA Director, Division of farming Technological Resource Development, Ministry of Agriculture, Putrajaya.
PS2	H1N1 – The Final Verdict	DR. SURESH KUMAR Consultant Infectious Diseases Physician, Department of Medicine, Hospital Sungai Buloh, Jalan Hospital, 47000, Sungai Buloh, Selangor.
PS3	Dengue Epidemiology and Challenges in Control	DR. CHONG CHEE KHEONG Sector Head, Vector Borne Disease Control, Ministry of Health, Putrajaya.

LIST OF PANEL SPEAKERS
No	Title	Name and Address
PL1	Solving the dengue riddle with RIDL	Vasan SS Director & Chief Executive Officer, Oxitec SB, Plaza See Hoy Chan, Suite 1502, Jalan Raja Chulan, Kuala Lumpur, 50200, Malaysia.
		Adjunct Professor, Centre for Research in Biotechnology for Agriculture, Level 5, Block B, IPS Building, University of Malaya, 50603 Kuala Lumpur, Malaysia.
PL2	Water Borne Zoonosis	Suresh K and Tan TC Department of Parasitology, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur.
PL3	Drug Discovery For Neglected Diseases- Malaria & Filaria	Noor Rain A Head Bioassay Unit, Herbal Medicine Research Centre (HMRC), Institute for Medical Research,Jalan Pahang, 50588, Kuala Lumpur.
PL4	Changing trends in measles and rubella incidence since inception of the measles-mumps-rubella immunization in Malaysia	Saraswathy TS Virology Unit, Infectious Disease Research Centre (IDRC), Institute for Medical Research, Jalan Pahang, 50588 Kuala Lumpur.
PL5	Molecular phylogeny and Bio-geography of Food-borne Zoonotic Helminths	Yukifumi N Dept of Helminthology, Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand.

LIST OF PLENARY SPEAKERS

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No	Title	Name and Address
C1	Infectious Diseases: From Epidemic to Pandemic	Dr. Lee Han Lim Head Entomology Unit, Infectious Diseases Research Centre (IDRC), Institute for Medical Research, Jalan Pahang, 50588, Kuala Lumpur.
C2	Biology and Surveillance of Aedes Vectors	Prof. Sallehudin Sulaiman Lecturer, Department of Biomedical Science, Faculty of Allied Health Sciences, National University of Malaysia, Jalan Raja Muda Abdul Aziz, 50300, Kuala Lumpur.
C3	Medical Parasitology	Prof. Rohela Mahmud Head, Department of Parasitology, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur.
C4	Malaria Vectors and Treatment	Prof. Dr. Fong Mun Yik Lecturer Department of Parasitology, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur.
C5	Forensic and Therapeutic Entomology	Prof. Baharudin Omar Lecturer, Faculty of Allied Health Sciences, National University of Malaysia, Jalan Raja Muda Abdul Aziz, 50300, Kuala Lumpur.
C6	Control of Aedes Vectors: 1	Prof. Ridad Agoes Indonesia
C7	Control of Aedes Vectors: 2	Dr. Khadri MS Senior Research Officer, Infectious Diseases Research Centre (IDRC), Institute for Medical Research, Jalan Pahang, 50588, Kuala Lumpur.
C8	Veterinary Parasitology	Dr. Chandrawathani P Deputy Director, Veterinary Research Institute, 59, Jalan Sultan Azlan Shah, 31400, Ipoh, Perak.
C9	Students Competition Presentation	Dr. Siti Nursheena Mohd Zain Senior Lecturer, Institute of Biological Sciences, Faculty of Science, University of Malaya, 50603, Kuala Lumpur.

LIST OF ORALS

No	Title	Name
	SESSION 1 BIOLOGY AND SURVEILLANCE OF AEDES VECTORS	
OR1	Effectiveness of a mosquito larval trap treated with <i>Bacillus thuringiensis</i> var <i>israelensis</i> or temephos for the control of <i>Aedes</i> mosquitoes	Khadri MS
OR2	A Predictive Model For Dengue Outbreak In Malaysia.	Suzilah I
OR3	Ovitrap Surveillance as an effective tool in the integrated vector control management	Nawi S
OR4	Effects of synthetic semiochemicals on Aedes aegypti (L.) behavior	Nurulhusna AH
OR5	No evidence of dual infection of chikungunya and dengue 2 viruses in laboratory infected <i>Aedes aegypti</i> (L.) and <i>Aedes albopictus</i> Skuse	Rozilawati H
OR6	Active mating time of Aedes aegypti (L.)	Shuhaida MI
OR7	The Skip Oviposition Behaviour of Laboratory, Field and RIDL strain of Aedes aegypti	Nazni WA
	SESSION 2 MEDICAL PARASITOLOGY	
OR8	Analysis of Entamoeba histolytica Antigens Specific for Amoebic Liver Abscess	Lim BH
OR9	Intestinal protozoa among humans and vegetablesin Baghdad province - Iraq	Baha L
OR10	The effects of the maturity levels of cysticercoid on the infectivity of <i>Hymenolepis diminuta</i> in the rats.	Hairul Hafiz M
OR11	Isolation of Acanthamoeba spp. From River and Water Recreation in Perak	Putri Noradyani MH
OR12	Parasites of frogs found in Sungai Pinang, Penang	Cheah SX
OR13	Surgical Operation, As a Risk Factor for HCV Infection: A Study Testing Ab, RNA & Genotypes of HCV	Waqar AL- Kubaisy
OR14	Knowledge, Attitude and Behaviors of students of tertiary institutions in North western Nigeria to HIV/AIDS	Magaji BA
	SESSION 3 MALARIA VECTORS AND TREATMENT	
OR15	Field Evaluation of Residual-Sprayed Deltamethrin WG and Deltamethrin WP on Different Type of Walls for the Control of Malaria Vector in Serian, Sarawak	Rohani A
OR16	Influence of Red Fruit Oil against Pathogenesis of Malaria	Susy T
OR17	PFCRT K76T Mutation of <i>Plasmodium falciparum</i> Isolates in Sabah: First Evidence	Nor Azrina N
OR18	Composition of species and biting activities of adult mosquitoes in Balai Ringin, Serian, Sarawak	Malinda M

	SESSION 4 FORENSIC AND THERAPEUTIC ENTOMOLOGY	
OR19	Developmental times of forensically important flies in Malaysia.	Kumara TK
OR20	An analysis of different temperature variables on the growth of <i>Chrysomya megacephala</i> (Fabricius) (Diptera:Calliphoridae) at environmental condition	Raja Muhammad Zuha RK
OR21	Assessment of morphology- and DNA-based identifications for entomological specimens of three selected forensic cases in Malaysia.	Tan SH
OR22	A preliminary study on cow dung diptera community in Sentul Timur, Kuala Lumpur	Heo CC
OR23	<i>In vitro</i> antibacterial activity of medicinal <i>Lucilia cuprina</i> larvae (Diptera:Calliphoridae) against selected pathogenic bacteria	Teh CH
OR24	Effect of protein on oogenesis of <i>Lucilia cuprina</i> (Wiedemann) and its utilization in maggot debridement therapy	Yeong YS
	SESSION 5 CONTROL OF AEDES VECTORS	
OR25	Biochemical detection of resistance mechanisms in field-collected Aedes (Stegomyia) aegypti (L.) in Shah Alam, Selangor	Loke SR
OR26	Ovicidal effects of Bacillus thuringiensis israelensis on Aedes aegypti (L.).	Andy TW
OR27	Development of permethrin resistance in several strains of Aedes aegypti (Linnaeus)	Wan Norafikah O
OR28	Effects of temperature stress on development of <i>Aedes aegypti</i> (L.) and <i>Aedes albopictus</i> Skuse	Suhaiza H
OR29	Simulated field performance of Spinosad DT against <i>Aedes aegypti</i> in Penang, Malaysia	Adanan CR
OR30	Biological efficacy of natural Juvenile hormone III (JH III) from a weed plant, <i>Cyperus aromaticus,</i> against <i>Aedes aegypti</i>	Fatemeh K
OR31	Sub-lethal dose of diflubenzuron and susceptibility status of <i>Aedes albopictus</i> and <i>Aedes aegypti</i> towards diflubenzuron in Penang island	Chan HH
OR32	Effects of Atmospheric Temperature on Susceptibility of <i>Aedes (Stegomyia) aegypti</i> to Vaporized Acetone	Jahangir K
OR33	Simulated field performance of Diflubenzuron against <i>Aedes aegypti</i> in Penang, in Malaysia	Muhamad Firdaus A
OR34	Efficacy of Aqueous Extracts of Areca Catechu against Larvae of Aedes (Stegomyia) aegypti	Prabadevi N
OR35	Bionomic and Susceptibility Status of <i>Aedes</i> mosquito on selected insecticides in USM campus, Penang Malaysia.	Ang CY
OR36	Knockdown Effect of Lemon (Citrus limon) Peel Extract as an Insecticide towards <i>Culex</i> sp. Mosquitoes with spraying method	Agustin I

	SESSION 6 VETERINARY PARASITOLOGY	
OR37	In Vitro Validation of Anthelmintic Activity of Butea monosperma and Calotropis procera	Zafar I
OR38	Validation of FAMACHA eye score system in goat	Nor-Azlina AA
OR39	Oxyspiruriasis in Zoo Birds – Treatment and Control	Vellayan S
OR40	Movements and Home Range of a Common Species of Tree-Shrew, <i>Tupaia glis</i> Surrounding Houses of Otoacariasis Cases in Kuantan, Pahang, Malaysia	Mariana A
OR41	Severe Anthelmintic Resistance in Commercial Small Ruminants Farms in Perak, Malaysia	Imelda LV
OR42	Diagnosis of Leptospirosis, Brucellosis & Melioidosis Disease in Humans Conducted in VRI	Naama T
OR43	The Use of Some Common Malaysian Herbs for Worm Control in Goats in Malaysia	Theivanai J

LIST OF STUDENTS COMPETITION

No	Title	Name
S1	Filariasis in Kuala Lumpur & Selangor: Entomological, Parasitological and Molecular Studies	Azdayanti M
S2	High prevalence of Blastocystis sp. subtype 4 among rural communities in Nepal	Lee IL
S3	Effects of Anthelmintic Plant; <i>Terminalia catappa</i> towards Nutritional and Physiological Aspects on Sprague-Dawley White Rats	Mohd Azrul L
S4	New predictive tools for pre-emptive dengue vector control in north Queensland, Australia	Aishah Hani A
S5	Does Blastocystis hominis Exacerbate the Growth of Colorectal Cancer Cells?	Chandramathi S
S6	Intestinal parasitic infections among children in Albania: current status and risk factors	Albana S
S7	Polytene Chromosome of the malaria vector Anopheles arabiensis Patton in Sudan	Mashair SM
S8	Prevalence and risk factors of protozoal infections among patients attending hospitals in Sana'a City, Yemen	Naelah Alyousefi A
S9	The comparison of artificial membrane feeding and direct feeding on Culex quinquefasciatus (Say) (Diptera: Culicidae)	Siti Nasuha H
S10	Comparative study of the macroparasite communities of stray cats from four urban cities in Peninsular Malaysia	Norhidayu S
S11	Molecular Characterization of <i>Blastocystis</i> sp. isolates from goats in Malaysia	Tan PC
S12	Better prediction of the development of liver cirrhosis in chronic hepatitis B infection using both HBV genetic and serum iron biomarkers	Chook JB
S13	Genotypic determination of <i>Toxoplasma gondii</i> strains by PCR-RFLP from clinical samples in Malaysia	Puviarasi M
S14	Variant Surface Glycoprotein (VSG) gene repertoires expressed by Malaysian isolates of <i>Trypanosoma evansi</i>	Mahira W

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