26-28th August 2014 (Tuesday – Thursday)
Basement II

POSTER PRESENTATIONS

P-AP01. Gene rearrangement using Fluorescence In Situ Hybridization (FISH) in the diagnosis of Burkitt lymphoma: preliminary result

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Introduction: Burkitt lymphoma is a solid tumor of B lymphoid cells. It is usually associated with translocations involving the c-myc gene of 8q24, particularly immunoglobulin heavy chain [IgH] t(8;14)(q24;q32), which is identified in approximately 80% of cases. The remaining 20% of cases demonstrate translocations of c-myc gene with immunoglobulin kappa light chain in t(2;8)(p11;q24) of immunoglobulin lambda (λ) light chain in t(8;22)(q24;q11). In Malaysia, diagnosis of Non-Hodgkin lymphoma (NHL) is generally based on morphological features and supported by immunohistochemistry findings. *Objective:* The aim of the study is to search for a molecular testing required for confirmatory diagnosis for Burkitt lymphoma in view of its unique molecular aberration. *Methods:* Retrospective study is carried out using archival paraffin tissue blocks of cases diagnosed as Burkitt lymphoma and Burkitt-like lymphoma collected from the Histopathology Unit, Department of Pathology, Hospital Kuala Lumpur (HKL) from 2006 to 2013. All patients aged less than 18 years old were included in this study. The cases were tested by Fluorescence in situ hybridisation (FISH) using 4 different probes which are LSI MYC Dual Colour, Break Apart Rearrangement Probe, LSI IGH Dual Colour, Break Apart Rearrangement Probe, LSI BCL6 Dual Colour, Break Apart Rearrangement Probe and LSI BCL2 Dual Colour, Break Apart Rearrangement Probe. 200 cells were counted and cut off threshold was set at 5% for each probes. *Results:* Currently two cases of Burkitt lymphoma and one case of Burkitt-like Lymphoma were analyzed. Out of two cases of Burkitt lymphoma, one case showed positive break apart for CMYC and IgH with no break apart of BCL6 and BCL2 gene. One case of Burkitt lymphoma and one case of Burkitt-like Lymphoma showed no break apart for all 4 probes tested. *Conclusion*: There is a possibility for Burkitt lymphoma cases to show non-translocation of CMYC and immunoglobulin chain (Ig). Further molecular tests need to be carried out for cases that showed no translocation of CMYC and Ig by FISH test to understand the mechanism involved at molecular level.

P-AP02. Extra gastrointestinal stromal tumours (EGIST) presenting as pelvic and vulvovaginal masses: A series of 3 case reports

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Introduction: Extra gastrointestinal stromal tumours (EGISTs) are mesenchymal tumours, which are identified outside the gastrointestinal tract with clinicopathological, immunohistochemical and molecular profiles similar to GIST. EGISTs are extremely rare. We reported 3 cases with the immunohistochemical profiles that support the diagnosis of EGISTs that were originally misdiagnosed as high-grade sarcoma. *Case presentation:* All the cases were retrieved from the histopathology files at the Department of Pathology Hospital Kuala Lumpur, from January 2005 to December 2013. Case 1: A 61 year-old Malay lady presented with a large nodular pelvic mass. Case 2: A 58 year-old Malay

lady who presented with a vaginal tumour adhered to the rectum. The mass is lobulated with well-circumscribed cream coloured cut surface. Case 3: A 72 year-old Iban post-menopausal lady presented with foul smelling pervaginal discharge. Intraoperatively, a vaginal tumour weighing 300g with solid cut surface is identified with no apparent attachment to the rectum. *Discussion:* Microscopically, all 3 cases show highly cellular malignant tumour composed of short intersecting fascicles of spindled cells with elongated plump nuclei and abundant cytoplasm. Immunohistochemistry revealed all the tumours were strongly positive for CD117 (c-kit), CD 34 and negative for smooth muscle actin. Thus, a diagnosis of EGIST was made. EGISTs that present as gynaecological masses are extremely rare. CD117 positivity is the hallmark in the diagnosis of EGISTs. Misdiagnosis may lead to inappropriate therapy therefore it is imperative to consider EGISTs in the differential diagnosis of any mesenchymal neoplasms in the pelvis and vagina.

P-AP03. Effect of leptin on the histopathology of the glomerulus in spontaneously hypertensive rats (SHR)

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Introduction: Leptin plays a role in stimulating fibrosis in renal tissue and may contribute to the pathogenesis of chronic renal disease. *Objective:* This study was designed to evaluate the effect of exogenously administered leptin on basic histopathological features in the kidneys of spontaneously hypertensive rats (SHR). *Methods:* SHR (n=16) were randomized to control and leptin treated groups. Animals in the control group were treated with saline by subcutaneous injection, animals in the treatment group were given leptin at a dose of 60 μg/kg/day by subcutaneous injection, for 6 weeks. On the 43rd day they were euthanized and kidneys were removed and immediately stored in neutral buffered formalin (10%). Subsequently they were processed and stained with hematoxylin and eosin. Slides were examined under light microscopy and 90 consecutive glomeruli were examined for i) cell counts based on the number of nuclei seen and ii) area of glomeruli. *Results:* Animals treated with leptin tended to have the least number of nuclei and lowest number of cells, though these differences did not reach statistical significance. *Conclusion:* Leptin may activate TGF β1 and SMAD pathway and excite inflammation; causing hypercellular and large glomeruli. However, histopathologically, our results did not reflect this finding. Recent reports imply that leptin may actually have a renoprotective role and thus suppress the process of inflammation; and our results appear to support this finding.

P-AP04. The expression of BCL2 in proliferating tumour cells in diffuse large B-cell lymphoma predicts tumour behaviour and its response to treatment

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Introduction: Diffuse large B-cell lymphoma (DLBCL) is a heterogeneous group of neoplasms with diverse morphology, molecular alterations and clinical behavior. Gene expression profiling divides it into two prognostically distinct subtypes. Its prognosis is also affected by the expression of anti-apoptotic

marker BCL2 and tumour proliferation fraction as assessed by Ki67 index. *Objective:* The study aims to determine BCL2 co-expression in Ki67-positive proliferating tumour cells in DLBCL and to analyse the correlation between the BCL2/Ki67 and its relationship with the clinicopathological features. *Methods:* Tissue micro-array of 90 DLBCL cases was immunostained for CD20, CD10, BCL6, MUM1, BCL2 and Ki67. BCL2 and Ki67 co-expression was determined using double-immunofluorescence labelling. *Results:* A total of 43/90 (48%) cases showed co-expression of BCL2 in proliferating cells (BCL2+/Ki67+) while in 47/90 (52%) cases, there was lack of co-expression (BCL2-/Ki67+). BCL2+/Ki67+ co- expression was seen in 13/32 (41%) of germinal centre B-cell (GCB) subtype cases and 30/58 (52%) of non- GCB-subtype cases. BCL2+/Ki67+ co-expression showed a positive trend towards the non-GCB subtype although it was not statistically significant (p=0.313). BCL2+/Ki67+ cases were associated with B symptoms (p=0.043) and primary extranodal tumours (p=0.019). The overall survival was lower in BCL2+/Ki67+ cases (mean 47m, median 19m) compared to BCL2-/Ki67+ cases (mean 48m, median 23m), although it was not statistically significant (p=0.605). In addition, complete response to chemotherapy was lower in BCL2+/Ki67+ cases (6/17, 35%) than in BCL2-/Ki67+ cases (13/23, 56%). *Conclusion:* BCL2+/Ki67+ cases were observed in almost half of DLBCL cases. This group showed a positive trend towards the prognostically worse non-GCB-subtype. It also showed poorer overall survival and lower response to treatment. It can be suggested that co-expression of BCL2 in proliferating cells prolong tumour cell survival and cause the tumour to behave more aggressively.

P-AP05. Reprogramming osteosarcoma cell lines through retrovirus-mediated transduction

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Introduction: Reprogramming of cancer cells have brought us closer to generate patient specific induced pluripotent stem cells (iPSC) that can be differentiated into any cell type for stem cell therapy, cancer vaccines or to produce larger cancer stem cell population for drug screening. *Methods:* We used Yamanaka factors, Oct4, Sox2, Klf4 and cMyc, to transduce osteosarcoma cells. Transduced cells were transferred to inactivated mouse embryonic fibroblast (iMEF) on Day 3 post transduction. Colonies were manually picked on Day 15 onwards. Reprogrammed osteosarcomas were characterised by observation on morphology, alkaline phosphatase and pluripotency markers expression, embryoid body formation and directed differentiation into adipocytes and osteocytes. *Results:* We managed to reprogram four osteosarcoma cell lines to pluripotent state. Embryonic stem cell (ESC)-like clusters started to appear between 15 to 20 days post transduction. Morphology of the colonies resembles ESC colonies with defined border and cells tightly packed with each other. We then characterised our reprogrammed osteosarcomas and found that all the reprogrammed osteosarcomas expressed Alkaline phosphatase and pluripotency markers, Oct4, SSEA4, TRA-1-60 and TRA-1-81, similar to ESC. In our observation, all reprogrammed osteosarcomas could form embryoid body-like spheres when cultured in suspension condition in a low attachment dish for up to 10 days. We further test the capacity of our reprogrammed osteosarcomas to differentiate by performing directed differentiation into adipocytes and osteocytes. Our directed differentiation results showed that all four reprogrammed osteosarcomas could differentiate into adipocytes as shown with Oil Red O staining. While, only Saos2-REP, MG63-REP and G292-REP could produce calcification as stained by Alizarin Red solution when they differentiate into osteocytes. *Conclusion:* These results support the ability of cancer cells to be reprogrammed. Reprogrammed osteosarcoma cells exhibit ESC-like characteristics.

P-AP06. High expression of FOXP1 is associated with poor prognosis in non-germinal centre B-cell (non-GCB) subtype of diffuse large B-cell lymphoma

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Introduction: Forkhead box protein P1 (FOXP1) is a transcription factor expressed in normal activated B cells and in a proportion of diffuse large B-cell lymphomas (DLBCL). FOXP1 is frequently associated with the less favourable non-germinal center subtype of DLBCL. *Objective:* This study aims to investigate the expression of FOXP1 in DLBCL molecular subtypes in correlation with patient's clinical parameters (age, performance status, LDH level, extranodal sites, stage and IPI scores). *Methods:* Immunohistochemistry for CD10, BCL6, MUM1 and FOXP1 was performed on 125 cases using tissue microarray. DLBCL subtype: germinal center B-cell (GCB) and non-germinal center B-cell (non-GCB) was determined from CD10, BCL6 and MUM1 expression according to Hans algorithms. The association of FOXP1 with DLBCL subtype and the clinicopathological parameters was analyzed using Chi square test. *Results:* A total of 39/125 (31.2%) cases are of GCB subtype and 86/125 (68.8%) cases are non-GCB subtype. In all, 95/125 (76.0%) cases were FOXP1 positive and 30/125 (24.0%) were FOXP1 negative. Among the positive cases, 70/95 (73.7%) was found to be significantly associated with non-GCB subtype (p=0.036). However, the non-GCB FOXP1 positive cases showed no significant correlation with clinical parameters in International Prognostic Index (IPI) including age (p=0.453), ECOG performance status (p=0.302), LDH levels (p=0.764), number of extranodal sites (p=0.842) and clinical stage (p=0.538). *Conclusion:* In our population, FOXP1 protein was predominantly expressed in non-GCB subtype suggesting its role in the development of this poorer prognosis subgroup of DLBCL. There was no association between FOXP1 and other clinical parameters in IPI suggesting its role as an independent prognostic factor in determining patient's outcome.

P-AP07. A rare entity of pure alveolar rhabdomyosarcoma of uterine corpus: a case report

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Introduction: Alveolar rhabdomyosarcoma accounts for only 20% of rhabdomyosarcomas. Rhabdomyosarcoma in middle-aged women is uncommon and occurence in the uterine corpus is an even rarer entity. Most of the cases are seen in the early to mid-teens but any age group can be affected. Typical sites are deep muscles of extremities, axial muscles and perineum. The usual behaviour is a rapidly growing tumour with advanced stage at presentation. *Case presentation:* A 57 year-old Indian lady presented with right iliac fossa pain for two weeks since early March 2014. There was pelvic mass felt on admission. Initial CT scan revealed a large solid cystic pelvic mass with intraabdominal lymphadenopathies. A total abdominal hysterectomy and bilateral salphingo-oopherectomy was perfomed. Intraoperatively, there was a large uterine tumour, which had breached the uterus. Histopathological examination of the tumour in the uterine corpus showed high grade round cells malignancy with typical alveolar pattern with no epithelial component seen. Similarly this tumour is noted at the paratubular region of the left fallopian tube. Immunohistochemical staining are positive for Desmin and Myogenin. It is negative for smooth muscle actin (SMA) and leucocyte common atigen (LCA). The omentum is uninvolved. It was concluded that this is a rare case of alveolar rhabdomyosarcoma of the uterine corpus. *Discussion:* Alveolar rhabdomyosarcoma as a pure form

needs to be established by adequate sampling as we need to exclude other differential diagnosis, which is of importance in terms of prognosis; namely adenosarcoma with sarcomatous overgrowth and carcinosarcoma. As expected, this is an aggressive tumour and patient is at Stage 4B, because when further evaluated there is also evidence of distant thoracic lymph nodes involvement. Patient is currently undergoing palliative chemotherapy.

P-AP08. Metastatic mullerian adenosarcoma to the lung: a case report

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Introduction: Mullerian adenosarcoma is a mixed tumour composed of benign, occasionally atypical glandular component and a sarcomatous stromal component, usually of low-grade in nature. Majority of cases occur after menopause and they commonly present with vaginal bleed. Recurrences are common but distant metastasis is rare. *Case presentation:* A 57 year-old lady presented with recurrent polypoidal tumours in the genital tract. In the first episode in 2008, she presented with polypoidal tumour in endometrial cavity, measuring 60mm in largest diameter and was diagnosed as an adenomyomatous polyp. After 4 years, she presented with recurrent polypoidal tumour in the vaginal vault measuring 85x60x55mm, which was associated with shortness of breath. CT thorax revealed a large mass in the upper lobe of right lung measuring 64x46x41mm and in the right main bronchus. Endobronchial biopsy was then performed. Histopathological examination from previous polypoidal tumour in endometrial cavity and recurrent tumour in vaginal vault as well as from endobronchial biopsy showed similar microscopic features. They were concluded as mullerian adenosarcoma. However, the recurrent polyp in the vaginal vault and the endobronchial tumour were solely composed of low-grade sarcomatous component with no evidence of sarcomatous overgrowth. *Discussion:* Recurrent polyp particularly in the endometrium or endocervix is uncommon and should therefore raise the possibility of an adenosarcoma. Correct diagnosis is important to ensure optimal management and long term follow-up. Recurrent adenosarcoma is usually composed solely of mesenchymal element. Proper sampling is important to exclude myometrial invasion and sarcomatous overgrowth, which are often associated with risk of recurrence and distant metastasis respectively. Distant metastasis is rare and has been reported in bone and is usually associated with sarcomatous overgrowth. In this case, sarcomatous overgrowth was not present in the biopsy sample and is yet to be scrutinized in the future excised sample.

P-AP09. Nasopharyngeal carcinoma with bone marrow infiltration: a case report

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Introduction: Bone marrow infiltration by nasopharyngeal carcinoma is very rare; making bone marrow assessment hardly necessary; whether at diagnosis or for staging of the disease. *Case presentation:* 44 year old Chinese gentleman was diagnosed with nasopharyngeal carcinoma stage IV (T2N1M1) with evidence of abdominal and mediastinal lymphadenopathy and multiple bone metastases in May 2013. Nasopharyngeal tissue biopsy showed non-keratinising nasopharyngeal carcinoma. He underwent one cycle of chemotherapy (Cisplatin and 5-Fluorouracil) in June 2013 which was then withheld for the second cycle in view of pancytopaenia. Bone marrow aspirates and trephine biopsy done on 10th July 2013 revealed hypocellular marrow with no evidence of tumour infiltration. Subsequently he received and completed palliative radiotherapy (45Gy/15#) to the postnasal space and neck on 6th August 2013.

Since then, he had several admissions due to symptomatic anaemia and was transfused with packed cells. In April 2014, he was readmitted for neutropaenic sepsis and symptomatic anaemia with symptoms of gum bleeding and easy bruising. Despite multiple platelet and packed cell transfusions, he was persistently anaemic and thrombocytopaenic. Full blood picture examination revealed pancytopaenia but no features of dysplastic changes or leucoerythroblastic picture was seen. Bone marrow aspirate done on the 23rd April 2014 was suboptimal due to dry tap. However trephine biopsy revealed sheets and clusters of malignant cells infiltrating the marrow spaces with areas of desmoplasia. These cells are moderate in size with vesicular pleomorphic nuclei, prominent nucleoli and positive for cytokeratin. Haemopoetic cell lines are markedly reduced and reticulin fibers are increased. Unfortunately he succumbed on the 16th May 2014. *Discussion:* This case illustrates the importance of careful examination of bone marrow aspirates and trephine biopsy in nasopharyngeal carcinoma patients whom presented with persistent pancytopaenia.

P-AP10. Prevalence of transitional cell/ urothelial carcinoma of bladder in Hospital Kuala Lumpur from year 2008 to 2013

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Introduction: Bladder cancer is the most common cancer in the urinary tract of both males and females. In the United States, 73,510 new cases of bladder cancer were diagnosed in 2012 with an approximately 14,880 deaths. In Malaysia, it is the ninth most common cancer in males. Males are affected 2 to 3 times more common than females. 90% of bladder cancer arises from the urothelium. *Objective:* This articles aims to describe the prevalence of urothelial carcinoma of bladder in Hospital Kuala Lumpur. *Methods:* This is a retrospective cross-sectional study in which all cases of transitional cell/ urothelial carcinoma diagnosed between the years 2008 and 2013 were included. *Results:* A total of 343 cases of bladder cancer data were recorded from year 2008 to 2013. Only transitional cell/ urothelial carcinoma cases were selected. 302/343 (88%) cases were male and 41/343 (12%) were female. 240/343 (70%) of cases were aged more than 60 years. The incidence of urothelial cancer was highest among Malay which constitute 182/343 (53.1%) followed by Chinese, 102/343 (29.7%) and Indian 46/343 (13.4%). Based on the 1973 WHO Classification of Bladder Cancer, 57/343 (16.6%) were grade 1 (G1), 118/343 (34.4%) were grade 2 (G2), 138/343 (40.2%) were grade 3 (G3) and 30/343 (8.8 %) of cases were not graded. 263/343 (76.7%) of patients presented with high grade disease. 216/343 (63%) of the cases fell into invasive stage (T_1-T_4) and 127/343 (37%) were non-invasive (T_a). *Conclusion:* Our data showed that cases of bladder cancer are common among Malays and are commoner in males. Bladder cancer cases in Malaysia occur predominantly in older age group (> 60 years) where majority of patients presented at invasive stage and high grade of disease.

P-AP11. Sociodemographic distribution of lymphoma in Hospital Kuala Lumpur from year 2008 to 2013

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Introduction: According to the 2007 National Cancer Registry of Malaysia, lymphoma is the sixth most common cancer. It is also the sixth most common cancer in males, whereas in females, it is the eighth most common cancer. *Objective:* This article aims to describe the sociodemographic distribution of lymphoma in Hospital Kuala Lumpur. *Methods***:** This is a retrospective cross-sectional study on retrieved Hodgkin lymphoma (HL) and Non-Hodgkin lymphoma (NHL) patients in HKL from 2008 to 2013. *Results:* Within those six years, 508 cases of lymphoma were diagnosed in HKL; 443 cases (87.2%) were NHL and 65 cases (12.8%) were HL, with an overall male predominance of 62.8%. 317 (71.5%) of NHL cases were aged below 60 years, 84 cases (19%) aged between 60 to 69 years and 42 cases (9.5%) were older than 70 years. Malays comprises 72% of NHL followed by Chinese, 17.2% and Indian, 8.3%. Based on the 2008 WHO Classification of Tumours of Haematopoietic and Lymphoid tissues, 222/443 cases (50.1%) of NHL were Diffuse large B cell type (DLBCL) followed by 35/443 cases (7.9%) were Anaplastic lymphoma, 24/443 cases (5.4%) were Burkitt lymphoma, 22/443 cases (4.9%) were Follicular lymphoma, 12/443 (2.7%) were T-cell lymphoblastic lymphoma, 10/443 (2.3%) were Marginal zone lymphoma and 9/443 cases (2.0%) of B-cell lymphoblastic lymphoma. HL occurs more commonly in males (56.9%). Majority of HL cases occur at age group of below 34 years (77%), 7 cases (10.7%) between 35-44 years and 8 cases (12.3%) ≥ 45 years. 34/65 cases (52.3%) were of Nodular sclerosis subtype, 11/65 cases (16.9%) were Mixed cellularity, 4/65 cases (6.2%) were Lymphocyte rich, 1/65 cases (1.5%) were Lymphocyte depleted. and 15/65 cases (23.1%) were not-otherwise-specified (NOS). *Conclusion:* DLBCL is the commonest type of NHL and Nodular sclerosis is the commonest subtype of HL. HL cases in Malaysia occur predominantly in younger age group and does not show significant bimodal peak.

P-AP12. Myxoid liposarcoma of the breast: a case report

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Introduction: Liposarcoma is one of the most common sarcoma in adults but the occurrence in the breast is very rare. Breast liposarcoma comprises 3-24% of all breast sarcomas. Liposarcoma is classified into 4 major histological subtypes: well-differentiated, myxoid/round cell, dedifferentiated and pleomorphic. *Case presentation:* We reported a 50 year old single nulliparous woman who presented with a breast mass for one year duration which was progressively increasing in size in the last three months. Examination revealed a huge firm smooth right breast mass measuring 5 x 8 x 6 cm. Mammogram showed a large lobulated soft tissue mass occupying the entire right breast with specks of calcification. A trucut biopsy showed a cellular tumour comprising small round cells with myxoid areas. A right modified radical mastectomy was performed which revealed a well demarcated lobulated solid yellowish tumour mass with haemorrhagic areas measuring 180 x 110 x 50 mm. Histologically the tumour is composed of a heterogenous population of cells comprising cellular round mildly pleomorphic nonlipogenic mesenchymal cells and a looser myxoid area containing small cells with surrounding arborizing 'chicken wire' capillary vasculature. Typical lipoblasts are seen. Eleven axillary lymph nodes sampled show no tumour involvement. The nipple is tumour free. A diagnosis

of a round cell/myxoid liposarcoma was made. *Discussion:* Breast liposarcoma is a rare disease which requires adjuvant chemotherapy and radiotherapy. However as the patient was mentally handicapped and was unable to communicate, the possible complications of chemotherapy could not be explained, therefore chemotherapy was deferred. This patient is currently still on radiotherapy.

P-AP13. Histopathological and biochemical effects of *allium sativum* oil administration in type 1 diabetic rats

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Introduction: Allium sativum (AS) commonly known as garlic, has been widely recognized as hypoglycemic agent against type2 diabetes mellitus, however, little is known about its effect on damaged pancreatic tissue in insulin dependant type1 diabetes (T1DM). *Objective:* The study was carried out to experimentally investigate the biochemical effects of AS oil administration in T1DM with an attempt to find a relation between these findings and histological effects on damaged pancreatic tissue. M*ethods:* We evaluated with ELISA kits the levels of serum insulin in male Sprague-Dawley rats with Streptozocin-induced T1DM in addition to measuring of blood glucose and body weights. All biochemical results were compared with AS effects on pancreatic histological changes. The four groups (6 rats each) under study received different intraperitoneal doses of AS oil for a period of 30 days. *Results:* Daily intraperitoneal administration of AS oil (either low dose 10 mg/kg or high dose 20 mg/kg) for up to 30 days to T1DM rats effectively reduces levels of blood glucose with significant effect on rat's body weights. Meanwhile, level of serum insulin due to damaged Langerhans islet cell was significantly changed. *Conclusion:* These results suggest that AS oil treatment has therapeutic effects against biochemical changes which occur in T1DM with significant repairing histological effects. The data may provide new strategies for using AS to be recommended as excellent candidate in the clinical management of T1DM.

P-AP14. *Nigella sativa* oil has significant histological and immunological effects on dermatitis herpetiformis associated with refractory coeliac disease

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Introduction: Dermatitis herpetiformis (DH) is an autoimmune skin disorder associated with coeliac disease (CD). *Objective:* This clinical-trial was carried out to assess the histological and immunological profiles of DH caused by refractory CD after *NS* oil administration with GFD to prove its potential immunomodulatory effect in DH. *Methods:* Fourteen DH patients diagnosed as CD cases were recruited in the study. Three of them were considered as refractory CD after 12 months period of GFD. Treatment by *NS* oil capsules (450mg; twice a day) was started with GFD to these 3 patients for additional period of 6 months. Duodenal biopsies before and after treatment were interpreted according to modified Marsh criteria, while skin biopsies were assessed by direct immunofluorescent technique. Moreover, their sera were tested for antigliadin, anti-endomysium, and anti-tissue transglutaminase

antibodies. *Results:* There were clear histological remissions in 11 patients (78.6%) treated with GFD after 12months; however, 3 patients (21.4%) showed MarshIIIa criteria. The skin biopsies of these 3 patients showed deposits of IgA and complement at the junction of the dermal and epidermal layers of the skin, in addition they have at least one positive antibody test. However, treatment with *NSO* in addition to GFD resulted in complete duodenal and skin histological remission with significant absence of all immunological antibodies. *Conclusion:* Administration of *NS* oil with GFD in treatment of DH associated with refractory CD effectively lead to complete clinical remission due to complete duodenal and skin histology remission in addition to absence of serological CD antibodies. Our preliminary findings demonstrated the immunomodulatory effect of *Nigella sativa oil 'NSO'* treatment with gluten free diet (GFD) in treatment of intestinal tissue injuries of refractory CD. Ultimately, these results may substantially improve the immunotherapeutic application of *NS* in clinical management of autoimmune diseases.

P-AP15. Effect of *Nigella sativa* oil administration on damaged pancreatic tissue in Type 1 diabetic rats

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Introduction: Type 1 diabetes mellitus (T1DM) is a chronic disease that impairs production of insulin due to an autoimmune destruction of pancreatic islet cells. *Nigella sativa* oil (NSO) was known as a hypoglycemic agent in both types of diabetes but little is known about its ability to repair the damaged pancreatic tissue in T1DM. *Objective:* The study was carried out to investigate the potential effect of NSO administration on repairing the damaged pancreatic tissues in T1DM. *Methods:* T1DM was induced by intraperitoneal injection of streptozocin single dose (65 mg/kg). Four equal sized groups of 24 male Sprague-Dawley rats were fed different doses of NSO. After completion of the experimental protocols (30 days), pancreatic tissues were collected and dissected using routine histological preparation. Blood samples were also collected and serum was assayed for serum insulin using an ELISA kit. *Results:* The normal control group showed normal cells in pancreatic islet of Langerhans while the diabetic control group with no treatment showed shrunken islets of Langerhans displaying degenerative and necrotic changes. Meanwhile, the treatment by low dose (0.2 mg/kg) of NSO protected the majority of cells in the islets of Langerhans, however the high dose (0.4mg/kg) of NSO treatment showed similar result as in normal control group. *Conclusion:* These experimental results suggested that NSO treatment has a therapeutic protective effect against pancreatic histological damage, which occurred in T1DM.

P-AP16. Reprogramming cancer cells derived from human oral squamous cell carcinoma (OSCC)

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Introduction: Oral Squamous Cell Carcinoma (OSCC) is one of the leading causes of death worldwide if not diagnosed in time. Consequently, screening and diagnosis of OSCC are faced with unlimited challenges among clinicians, as there are no defined symptoms that could be identified. Existing treatments are unable to eradicate OSCC as the solid tumors are heterogeneous in nature and tend to resist cancer treatments followed by disease recurrences. Previous studies have shown that the application of induced pluripotent stem cells (IPSCs) technology may be utilized as an alternative tool to study cancer. IPSCs derived from OSCC may be applied to assess its biological properties to halt the resistant tumors and ease recurrence. *Objective:* We attempted to reprogram oral cancer cells into IPSCs to serve as a disease model for the study of tumorigenecity effects of oncogene activation and onco-suppressor gene inactivation, suggesting a novel therapeutic strategy. *Methods:* H103, Oral Squamous Cell Carcinoma cells (OSCC) were tranduced into induced pluripotent cells (iPSCs) via Retro Virus using the Yamanaka OSKM factors (OCT4, SOX2, Klf4, c-Myc). Putative colonies were preliminarily characterised for morphological changes. Immunofluorescence staining identifies presence of endogenous gene expressions. *Results:* Transduction efficiency via GFP expressions were detected at 48 hours confirming the uptake of transgenes. Embryonic Stem Cells (ESCs) like colonies appeared at day 15 and were morphologically different from that of the parental cell lines. Specific pluripotency markers were detected in suspected colonies using immunofluorescence staining. *Conclusion:* OSCC cells were successfully re-programmed into IPSCs using Yamanaka's approach, as evidenced by morphology of the colonies and presence of pluripotency markers.

P-AP17. Invasive thymoma with discontinuous spread to diaphragm: a case report

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Introduction: Thymomas are rare tumours and are characterized by indolent growth pattern. However, these tumours are known to cause invasion and dissemination. *Case presentation:* We report a case of invasive thymoma with discontinuous spread to diaphragm (so-called drop metastasis) in a 30-year-old Malay lady. She presented with fever, cough and shortness of breath for one week. Immediately after admission, her condition deteriorated and she passed away within 24 hours. Autopsy showed an 8cm anterior mediastinal mass with adhesion to the lung. Also identified were smaller tumours in the left diaphragm. Histological features and immunohistochemical study support the diagnosis of WHO type B1 thymoma with lung invasion and diaphragmatic implants. *Discussion:* Despite its rarity, the possibility of thymoma has to be considered when encountering a patient with mediastinal and diaphragmatic lesion.

P-AP18. Plasma cell myeloma initially presenting with solitary pleural nodule, diagnosed via video-assisted thoracoscopic surgery (VATS)

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Introduction: Plasma cell myeloma, also known as multiple myeloma (MM), is a disease of clonal B cell proliferation. Some patients develop extramedullary (extraosseous) (EM) plasmacytoma. EM manifestations can be detected either at the time of diagnosis or during the course of disease. EM involvement confers poor prognosis. *Case presentation:* We hereby report a case of plasma cell myeloma in a 65-year-old male, who first presented with incidental finding of solitary peripheral nodule in chest X ray. No pleural effusion was found. Biopsy via video-assisted thoracoscopic surgery (VATS) was performed. Diagnosis of solitary plasmacytoma of the pleura was established by histology findings with associated immunostains of CD138, CD56 and Kappa light chain restriction. Subsequent study showed Ig A Kappa paraproteinemia and marrow plasmacytosis. He received chemotherapy and high dose therapy with autologous stem cell rescue. The patient is well after 2 years of diagnosis and achieved a complete response. *Discussion:* EM plasmacytoma should be included in the differential diagnosis of a pleural tumour. Investigations to exclude the possibility of MM with concurrent pleural involvement are essential. Correct identification of this tumour is important for patient care.

P-AP19. The prevalence of hydatidiform moles (HM) and the subtypes – comparison between routine H&E and immunohistochemistry p^{57kip} staining

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Introduction: The distinction of hydatidiform moles (HM) subtypes are important to determine its' clinical behavior. It is made on the basis of morphological examination. The absence of a standardized morphological criteria, very early gestation and atypical cases had caused major difficulties in diagnosis. Immunohistochemistry (IHC) $p57^{kip2}$ staining has been reported to aid the diagnosis of difficult cases. *Objective:* The aim of the study is to determine the prevalence of HM subtypes and usefulness of IHC P^{57kip}. *Methods:* Paraffin embedded HM tissues (2007 - 2012) were selected from UKMMC and HKL. Only 82 cases (55.6%) fulfilled the inclusion criteria for IHC $p57^{kip2}$ analysis. HM subtypes were reclassified based on H&E and IHC $p57^{kip2}$. *Results:* A total of 147 HM cases were collected. There were 51 (34.6%) cases of complete hydatidiform mole (CHM), 54 (36.7%) partial mole (PHM) and two (2) (1.36%) persistent trophoblastic diseases. Forty cases (27.2%) were diagnosed as only molar pregnancy. The mean age of presentation was 25 to 29 years old (± 8.56). Malay ethnicity revealed the highest distribution. Histopathological examination of the selected 82 cases, revealed the diagnosis of CHM in 39 (47.5%) cases, PHM in 41 cases (50.0%) and two (2.43%) cases were diagnosed as molar pregnancy. However with IHC P^{57kip} analysis, 66 cases (80.5%) were re-diagnosed as CHM, 14 cases (17.1%) as PHM while another two cases (2.4%) were actually decidual and cystic tissues. IHC P^{57kip} analysis showed a discrepancy of 33% from the initial routine H&E diagnosis. *Conclusion:* The IHC staining using P^{57kip} monoclonal antibody improves the characterization of the hydatidiform subtypes and it is useful to complement routine H&E staining especially in the early gestations with less distinctive morphologies. Thus, immunohistochemistry (IHC) $p57^{kip2}$ should be considered as one of the routine test in diagnosing HM subtypes.

P-AP20. Clear cell sarcoma of the kidney: 3 case reports

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Introduction: Clear Cell Sarcoma of the Kidney (CCSK) is an uncommon malignant renal neoplasm of childhood. However, it represents one of the most common tumors with "unfavorable histology" listed in the National Wilms Tumor Study Group (NWTSG) clinical protocol. It has a peak incidence in children aged 3-5 years, with a male:female ratio of 2:1. CCSK is extremely rare in infants younger than 6 months and in young adults. We report 3 cases of CCSK clinically diagnosed as Wilms tumour in Hospital Kuala Lumpur within a period of 5 years. *Case presentation:* Case 1: A 3 year-old Malay boy presented with a right abdominal mass and a right foot mass. The histological diagnosis was CCSK with soft tissue metastasis. Case 2: A 3 year-old Malay girl presented with left abdominal mass and underwent left nephrectomy with lymph node dissections. The histological diagnosis was CCSK. Case3: A 3 year-old Malay boy presented with right abdominal mass. He underwent right nephrectomy with para-aortic lymph nodes dissections. The histological diagnosis was CCSK with lymph nodes metastasis. *Discussion:* CCSK has distinctive histopathology features with propensity to bone metastasis (14%) and has poor clinical outcome. Most CCSKs have the classic histological pattern (90%) as in these case reports, typically monomorphic population of clear, round to polygonal cells separated by characteristic delicate vascular septa. It is important to recognise variant patterns of CCSK and distinguish them from other paediatric renal neoplasm because of the more aggressive clinical course and selection of therapy. CCSK is an uncommon malignant childhood neoplasm and can be misdiagnosed as other renal neoplasms. This further emphasises the major role of histopathologists in tissue diagnosis to ensure appropriate patient management and treatment.

P-AP21. Paraoxonase-1 (PON1): bridging the chronic organophosphate exposure to atherosclerosis

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Introduction: Organophosphate (OP) is hydrolyzed by paraoxonase (PON1), an antioxidant enzyme that prevent atherosclerosis by inhibiting oxidative modification of low density lipoprotein (LDL). Low PON1 activities have been observed among individuals chronically exposed to OP while their decreased activities were reported in individuals with atherosclerosis related disease. However, a relationship between chronic OP exposure, PON1 and the development of atherosclerosis has not yet been reported. *Objective:* The aim of this study was to investigate the effects of chronic OP exposure on the development of atherosclerosis in rat model. *Methods***:** Twenty male Sprague-Dawley rats were divided into 3 groups; Group 1 did not receive any injection, both Group 2 and Group 3 received subcutaneous injection of vehicle and injection of 18.0 mg/kg of chlorpyrifos (CPF) respectively every other day for 180 days. Blood were analyzed for paraoxonase enzyme activities and ox-LDL. Aorta were harvested and stained for light and electron microscopic examination. *Results:* The paraoxonase activities, oxidized LDL and PON1: ox-LDL ratio were found to be significantly lower in OP exposed rats. The OP exposed rats also showed positive early atherosclerosis changes microscopically with VCAM-1 expression. The electron microscopic (EM) examination showed evidence of vascular damage with disruptions of the intimal layer of aorta, irregularly oriented and morphologically changed endothelial cells and numerous endothelial gaps with areas of deendothelialization. *Conclusion:* This study highlighted that chronic OP exposure leads to the development of atherosclerosis, which is confirmed microscopically

and further affirmed by positive VCAM-1 expression. The basis for the above observation could be explained by low PON1 activities and low PON1: ox-LDL ratio. In conclusion, chronic intermittent low dose of OP chlorpyrifos induced the development of early atherosclerosis, which could be explained by inability of the PON1 to hydrolyze oxidized-LDL.

P-AP22. CD24 expression in adenocarcinoma of colon

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Introduction: Colon cancer is the most frequent cancer in Malaysian men and second in women with 13.2% prevalence in 2006. CD24 is a mucin-like glycoprotein anchored to cell membrane by two chains of glycophosphatidylinositol (GPI). It directly binds to tyrosine kinases in intracellular signalling and acts as ligand to P-selectin. It forms tumour thrombi in lymphovascular vessels and assists in rolling and adhesion of cancer cells to vascular wall during cancer cell metastasis. *Objective:* This study aims to study the expression of CD24 in adenocarcinoma of colon. *Methods:* 176 adenocarcinoma of colon were included in this study. The paraffin embedded tissues were retrieved from the departmental archives. The tissues were cut at 4 mm thick and were immunohistochemically stained with CD24 monoclonal antibody (LabVision, USA). CD24 expression was indicated by cytoplasmic membrane staining and scored using H-score system. *Results:* There were 111 (63.1%) men and 65 (36.9%) women with median age of 63 years old. Ethnic Malays formed 71 (40.3%) cases; Chinese 75 (42.6%) cases and Indian 25 (14.2%) cases. Of 176 cases, there were 10 (5.7%) grade 1 cases, 161 (91.5%) grade 2 cases and five (2.8%) grade 3 cases. There were 27 (15.3%) stage I, 56 (31.8%) stage II, 90 (51.1%) stage III and 3 (1.7%) stage IV. 93 (52.8%) cases had lymph nodes metastases. 87 (49.4%) cases and 67 (38.1%) cases showed strong and weak CD24 expression respectively. There was significant difference in CD24 expression between tissues with lymph node metastases and those without. Significant correlation was seen between grade (p=0.036) with CD24 expression. However, no significant correlation seen between CD24 expression with adenocarcinoma stages and demographic factors. *Conclusion:* CD24 expression is prominently increased with grade and stage of adenocarcinoma. Significant positive associations can be explained by nature of CD24 in regulating cell-cell interaction and in assisting cancer cells metastases. Thus CD24 has a potential as future prognostic marker in adenocarcinoma of colon.

P-AP23. CD166 expressions in colon adenocarcinoma

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Introduction: Colon adenocarcinoma shows increasing trend worldwide, with the highest mortality rate being in elderly. CD166 is a type 1 transmembrane glycoprotein known as activated leukocyte cell adhesion molecule (ALCAM). It mediates homophilic (ALCAM-ALCAM) and heterophilic (ALCAM-CD6) cell-cell communication leading to cell clustering and migration. Pathophysiologically, aberrant CD166 expression inhibits clustering of tumor cells and alleviates them to migrate for tissue invasion. *Objectives:* We aim to study the expression of CD166 in colon adenocarcinoma. *Methods:* 176 colon adenocarcinomas were included in this study. The paraffin embedded tissues were retrieved from the departmental archives and immunohistochemically stained with CD166 monoclonal antibody (Abnova, UK). CD166 expression was indicated by cytoplasmic membrane staining and scored using H-score system. *Results:* There were 65 (36.9%) women and 111 (63.1%) men with median age of 63 years old. Ethnic Chinese proportionated 75 (42.6%) of cases while Malays and Indian were 71 (40.3%) and

25 (14.2%) cases accordingly. Well-differentiated adenocarcinomas formed 10 (5.7%) cases, while 161 (91.5%) and 5 (2.8%) cases represented moderately and poorly differentiated adenocarcinomas respectively. 27 (15.3%) cases constituted stage I adenocarcinoma, while 56 (31.8%) and 90 (51.1%) cases formed stage II and stage III adenocarcinoma respectively. Only 3 (1.7%) cases were of stage IV. 93 (52.8%) cases were positive for lymph nodes metastases. Weak and strong CD166 expression were found in 79 (44.9%) and 87 (49.4%) cases respectively. There was no significant correlation between CD166 expression with stage (p=0.076), lymph node metastases (p=0.240), grade (p=0.502) and demographic factors. However, significant correlation was drawn between CD166 expression and inflammatory infiltration (p=0.039).*Conclusion:* No significant correlation was found between CD166 expression and essential clinicopathological factors. This warrants a comprehensive prognostic study on CD166 since other studies stated otherwise. Significant aberrant CD166 expression with inflammatory infiltration might suggest the role of CD166 in modulating relevant inflammatory cells in carcinogenesis and metastasis.

P-AP24. Colon adenocarcinoma and CD133 expression

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Introduction: Colon cancer is the most frequent cancer in Malaysian men and second in women with 13.2% prevalence in 2006. CD133 also known as prominin-1 and AC133 is a transmembrane glycoprotein of five domains. It is located at structures of highly curved plasma membrane protrusions and invaginations, where it binds to the cholesterol of plasma membrane. CD133 is directly involved in developmental framework by acting as scaffold to maintain cancer stem cell structure, hence making it resistant to treatment. *Objective:* This study aims to observe the expression of CD13 in colon adenocarcinoma. *Methods:* 176 paraffin embedded colon adenocarcinoma tissue were retrieved from the departmental archives and stained with CD133 polyclonal antibody (Abnova, UK). CD133 expression. The intensity and percentage of positive cells were scored using H-score. *Results:* There were 65 (36.9%) women and 111 (63.1%) men with median age of 63 years old. Ethnic Chinese formed 75 (42.6%) cases while Malays and Indian were 71 (40.3%) cases 25 (14.2%) cases respectively. Cases with grade 1 were 10 (5.7%), grade 2 cases were 161 (91.5%) and grade 3 cases were five (2.8%). Additionally, there were 27 (15.3%) stage I, 56 (31.8%) stage II, 90 (51.1%) stage III and three (1.7%) stage IV. Cases with lymph nodes metastases were 93 (52.8%) cases. 72 (40.9%) cases and 84 (47.7%) cases exhibited weak and strong CD133 expression correspondingly. Surprisingly, there was no significant correlation between grade (p=0.975), stage (p=0.851) and lymph node metastases (p=0.974) with CD133 expression. There was also no significant correlation with demographic factors. *Conclusion:* No significant association between CD133 expressions with clinicopathological parameters strongly suggesting that the marker is unsuitable as cancer stem cell marker for colon adenocarcinoma. Since there are other studies saying otherwise, advance prognostic study have to be carried out to weigh the role of CD133 in carcinogenesis and metastasis.

P-AP25. Analyses of SYT-SSX fusion-transcripts of synovial sarcoma using one-step real-time polymerase chain reaction

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Introduction: Synovial Sarcoma is a rare cancer and account for 5-10% of adult soft tissue sarcomas. Despite establishment of some immunohistochemistry staining, making a definitive diagnosis of synovial sarcoma remains a challenging task. The t(X;18) (p11.2;q11.2) has been demonstrated to be highly characteristic of synovial sarcomas, and the resulting *SYT-SSX* fusion transcripts have been shown to be useful diagnostic markers. *Objective:* The current study aims to apply a one-step real-time, reverse transcriptase polymerase chain reaction (RT-PCR) multiplex assay for detection of either *SYT-SSX1* or *SYT-SSX2* fusion transcripts. This is an upgrade method than conventional RT-PCR employed in our previous study. *Methods:* Paraffin-embedded fixed-tissue (PEFT) block samples of 3 confirmed and 15 possible cases of Synovial Sarcoma were subjected to RNA purification using the standard spin column protocol and a one-step direct RT-PCR using multiplex primers and Taqman probe for detection of either SYT-SSX1 or SYT-SSX2 transcripts. *Results:* Our analyses showed positive SYT-SSX fusion transcript in all confirmed cases and 6 possible cases of synovial sarcoma**.** *Conclusion:* One-step direct RT-PCR is a rapid test method for identification of SYT-SSX fusion-transcript and useful in establishing the diagnosis of Synovial Sarcoma.

P-AP26. A preliminary study of Anaplastic Lymphoma Kinase (ALK) expression and clinicopathological relationship in neuroblastoma of Malaysian patients.

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Introduction: Neuroblastoma is one of the most common solid tumors diagnosed in infancy and childhood. In Malaysia, this childhood cancer is listed as the sixth most common brain and nervous system cancer in 2007. Studies have shown ALK as the major predisposing gene in neuroblastoma as ALK gene alteration can be spontaneously regressed or inherited. *Objective:* This study aims to investigate the ALK expression by immunohistochemistry and gene alteration by FISH in neuroblastoma of Malaysian patients and correlates the prognostic relevance of ALK with the clinicopathological findings. *Methods:* We investigated ALK gene aberration by fluorescence *in situ* hybridization (FISH) using ALK Break Apart FISH Probes and ALK protein expression by immunohistochemistry (IHC) using ALK monoclonal antibodies CD246 on 17 samples. The clinicopathological association with the patient outcome was also analysed. *Results:* There were slight female to male predominance in Malaysian population with ratios 0.9% (9/8) with a median age of 4 years. Among neuroblastoma cases, 41.1% of patients are younger than 18 months. IHC data showed that 17.6% (3/17) were positive with membranous staining and no nuclear staining was observed. FISH data showed that 11.7% (2/17) of neuroblastoma samples were found to have copy number gain. A three year overall survival rate was also found in ALK gain tumour patients. *Conclusion:* Screening for ALK gene alteration (amplification and copy number gain) using IHC and FISH technique is possible and can be adopted as a baseline investigation in stratification risk for neuroblastoma patient in Malaysia.

P-AP27. Placental mesenchymal dysplasia: a rare but significant pathology

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Introduction: Placental mesenchymal dysplasia (PMD) is a rare placental vascular anomaly characterized by dysplastic enlarged stem villi with vascular malformations. Most of the cases are associated with IUGR and fetal death as well as with Beckwith- Wiedemann syndrome (BWS) although normal fetal outcome has been reported.We would like to share this rare case diagnosed through placenta examination. *Case presentation:* A 22-year-old lady in her second pregnancy had recurrent painless pervaginal spotting during the second trimester. Her ultrasound scan showed presence of subchorionic haematoma, cystic lesions in the placenta and a growing fetus. There was no history of trauma or abdominal pain and she was normotensive throughout the pregnancy. Her first daughter was born 2 years ago by spontaneous vaginal delivery and is currently alive and well. She presented with leaking liquor at 35 weeks gestation and underwent emergency caesarian section due to fetal distress. Grossly the placenta is enlarged with cystic areas and histopathological findings include abnormally large stem villi with cisterns accompanied by collections of malformed large fetal vessels and thrombosis. *Discussion:* PMD is unfamiliar to pathologists and in early pregnancy often clinically mistaken as partial hydatidiform mole. Unlike partial mole, the trophoblastic proliferation is absent and often of normal karyotype. Persistent fetal hypoxia may stimulate placenta remodeling with stromal hyperplasia seen in dysplastic villi. The variations of PMD findings with the gestational age suggest that the vascular malformations develop progressively secondary to circulatory imbalance and poor vascularization of the dysplastic villi. Awareness of this entity and recognizing the characteristic pathological features are important for early diagnosis and detection of fetal complications.

P-AP28. The utility of Galectin-3 as immunohistochemical marker for thyroid malignancies

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Introduction: Thyroid cancer is the most common endocrine malignancy. Despite World Health Organization (WHO) guidelines for diagnosis of thyroid cancer, diagnosis is still susceptible to observer variations in some cases. Galectin-3 is a potential immunohistochemical marker for diagnosis of thyroid malignancy. *Objective:* The aim of this study is to determine the diagnostic utility of galectin-3 as an immune marker for malignant thyroid lesions. *Methods:* Immunohistochemical expression of galectin-3 is tested on 18 thyroid specimens (from a sample size of 170), excised at Universiti Kebangsaan Malaysia Medical Centre (UKMMC) from January 2010 to December 2013. The cases are comprised of 10 benign lesions (five goitres, five follicular adenomas) and eight malignant thyroid lesions (five papillary thyroid carcinomas, one follicular carcinoma, two anaplastic carcinomas). *Results:* Our preliminary results show galectin -3 immunopositivity in all eight (100%) malignant thyroid lesions while only two out of 10 (20%) benign lesions were positive (one goiter, one follicular adenoma). *Conclusion:* Galectin-3 shows promise as a potential immune marker for thyroid malignant lesions, while its role for differentiating the follicular lesions remains to be seen upon completion of the study.

P-AP29. COX-2 overexpression in invasive breast carcinoma

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Introduction: COX-2 is an enzyme that mediates the metabolism of arachidonic acid, leading to production of prostaglandin. It is thought to play an important role in carcinogenesis through the prostaglandin synthesis. COX-2 expression has been demonstrated in various malignancies such as the colon, lung, stomach, liver and breast in many previous studies. Its expression has been shown to correlate significantly with adverse prognostic factors in breast cancer. *Objective:* The purpose of this study is to assess the expression of COX-2 in invasive breast carcinoma of ductal and lobular subtypes and to correlate it with various clinicopathological parameters. *Methods:* Expression of COX-2 was evaluated immunohistochemically in 178 cases of invasive breast carcinoma of ductal (N=174) and lobular (N=4) subtypes. The staining intensity between the two tumor subtypes was also reviewed. Its expression was then correlated statistically with various clinicopathological parameters such as age, tumor size, grade, lymphovascular and lymph node invasion as well as the hormone receptor (ER, PR) and HER2 status. *Results:* We detected a moderate to high cytoplasmic expression of COX-2 in 165 (94.8%) cases of ductal and all four (100%) lobular carcinomas. Increase COX-2 expression was more commonly observed in tumours with larger size (> 20 mm), higher histological grade, positive lymph node metastasis and lymphovascular invasion, negative ER status and HER2 gene amplification. However, there was no statistically significant correlation between COX-2 expression with tumour subtypes and the above clinicopathological parameters. *Conclusion:* Our data showed a higher expression of COX-2 in invasive breast carcinoma (94.9%) especially the higher grade tumours, tumours with lymph node metastasis and triple negative cases. These findings may suggest the possible role of COX-2 expression in determining treatment options and as prognostic indicator.

P-AP30. CD5 expression in diffuse large B-cell lymphoma amongst Malaysian patients

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Introduction: Diffuse large B-cell lymphoma (DLBCL) is the largest subtype of malignant lymphoma and is potentially curative with rituximab plus cyclophosphamide, doxorubicin, vincristine and prednisolone (R-CHOP). De novo CD5-positive DLBCL is seen in 10 – 15% of DLBCL cases. This unique subtype is associated with elderly onset, high risk of IPI, female predominance, abundant extranodal involvement and poor prognosis as well as frequent CNS relapse rate. CD5-positive DLBCL is likely to arise from somatically mutated CD5-positive progenitor B-cells via the genetic analysis. *Objective:* The aims of the study were to investigate the incidence of CD5-positive DLBCL and to further characterize this distinct subtype in a Malaysian population. *Methods:* Immunohistochemical staining was performed to detect the presence of CD5 positivity in 102 cases of DLBCL, on paraffin sections. *Results:* A total of 11 cases (12%) were CD5-positive DLBCL in which 10/11 cases (90.9%) belong to non-germinal centre B cell subtype (Non-GCB). In addition, 9/11 cases (81.8%) showed poor International Prognostic Index (IPI). BCL-6 gene rearrangement was present in two CD5-positive cases. All CD5-positive DLBCLs were negative for cyclin D1. The outcome of CD5-positive DLBCL in response to chemotherapy were significantly inferior (P= 0.01) in comparison to CD5-negative DLBCL since nine patients died of the disease. *Conclusion:* Our results were in keeping with previous studies

in which CD5+ DLBCL cases account for approximately 10-15% of DLBCL. The prognosis of the disease is significantly poorer in comparison to CD5- DLBCL. The expression of this biomarker is associated with high risk IPI group. Further studies with larger population of CD5+ DLBCL patients are required for better characterization of the disease.

P-AP31. Differential expression of cellular senescence marker p14arf in the assessment of breast cancer development

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Introduction: Breast cancer is a hormonally driven age-related disease in women. Cellular senescence is an age-related irreversible cell cycle arrest at the G1 phase upon intrinsic or extrinsic induction. *Objective:* This study was conducted to assess and characterize the expression of epithelial senescence marker of p14 during breast cancer development. *Methods:* We retrieved retrospective cohorts of 1072 invasive ductal carcinoma cases for 11 years from six provinces in South Sydney west area. Tissue microarray blocks consist of two cores of each normal, benign, premalignant and malignant feature. We, therefore, performed immunohistochemical staining for P14 on formalin fixed-paraffin embedded samples. Covariates such as age, tumour size, lymphovascular and nodal involvement, tumour grade, stage, ER, PR and HER2 status were obtained. *Results:* We detected increasing p14 positive staining from 33.9% in normal, 34.9% benign and 53.9% premalignant to 59.9% in malignant. There were significant correlations of normal, benign, premalignant and malignant features ($p<0.0001$). There was a significant association between P14 expressions with tumour grade and lymphovascular invasion ($p<0.05$), but no significant association with age, lymph node status, tumour size, ER, PR, HER2 and staging. There was no difference in recurrent-free survival ($p=0.098$) or overall survival ($p=0.59$). Multivariate cox regression analysis showed significant prognostic value of p14 in predicting the outcome of the disease ($p<0.05$). C*onclusion:* These findings suggest P14 plays an important role in the progression to invasive breast cancer. Positive normal tissue have tendency to have high expression in more progressive features. It appeared p14 may suppress breast tumour from metastasis and is an independent prognostic factor for better survival.

P-AP32. Human Telomerase RNA (hTERC) gene amplification is a marker of disease progression in squamous lesions of the cervix

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Introduction: The human telomerase RNA (hTERC) gene is located at chromosome 3q involved in the maintenance of chromosome length and stability. The amplification of hTERC gene is associated with tumorigenesis. *Objective:* The objective of this study is to determine the amplification of hTERC gene in normal and abnormal cervical lesions. *Methods:* Eighty five liquid-based cytology (LBC) samples were collected from Universiti Kebangsaan Malaysia Medical Centre (UKMMC) and Hospital Kuala Lumpur (HKL), which include negative for intraepithelial lesion or malignancy (NILM) (n=5),

Atypical Cells of Undetermined Significance (ASC-US) (n=18), Low Grade Squamous Intraepithelial Lesion (LSIL) (n=26), High Grade Squamous Intraepithelial Lesion (HSIL) (n=29) and Squamous Cell Carcinoma (SCC) (n=2). hTERC amplification was determined using dual colour Fluorescence In-Situ Hybridization (FISH) probe. *Results:* hTERC gene amplification was detected in 34.1% of cases; 0% in NILM, 17% in ASC-US, 23% in LSIL, 62% in HSIL and 100% in SCC. Amplification of hTERC was significantly associated with cytologic grade ($p<0.05$). There was a marked increase of hTERC amplification in cases with HSIL and above lesions ($p<0.05$). *Conclusion:* The amplification of hTERC is an indicator of disease progression from low grade to high grade cervical lesions.

P-AP33. Prevalence of CHEK2 gene mutations in multiethnic cohort of breast cancer patients in Malaysia

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Introduction: The cell-cycle checkpoint kinase 2 gene (CHEK2) is a candidate breast cancer susceptibility gene that encodes a protein kinase that is involved in cell-cycle checkpoint control after DNA damage. *Objective:* The aim of this study is to identify and characterise the CHEK2 gene sequence changes among Malaysian high risk breast cancer patients. *Methods:* A total of 59 high-risk breast cancer patients were selected from Universiti Kebangsaan Malaysia Medical Centre (UKMMC), Hospital Kuala Lumpur (HKL) and Hospital Putrajaya (HPJ). Mutational screening on all coding sequence of the CHEK2 gene was performed using direct DNA sequencing. Sequence variants identified were screened further in case-control cohort consisting of 878 unselected invasive breast cancer patients (180 Malays, 526 Chinese and 172 Indian) and 270 healthy individuals (90 Malays, 90 Chinese and 90 Indian). DNA damage assay was done on yeast to determine the effect of the variants on normal function of CHEK2. *Results:* We identified two missense mutations (p.I160M and p.R180C) and two synonymous mutations (p.E84E and p.L129L) in this study. Analysis in a case-control cohort unveiled the variant p.I160M in 2/172 (1.1%) Indian cases and 1/90 (1.1%) Indian controls, and the variant p.R180C in 2/526 (0.38%) Chinese cases and 0/90 controls, and in 2/180 (1.1%) Malay cases and 1/90 (1.1%) Malay controls. Consistent with in silico analysis which predicted these variants to be likely pathogenic, we found in the DNA damage assay that both variants are more sensitive in response to DNA damage compared to the wild type. *Conclusion:* In conclusion, two potentially pathogenic missense mutations were identified among high-risk breast cancer patients in this cohort (3.4%). The results of this study indicate that CHEK2 germline mutations are rare among high risk breast cancer patients and may play a minor role in increasing risk to breast cancer among Malaysian population.

P-AP34. A rare encounter of chronic hydatid cyst in Malaysia

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Introduction: A cystic lesion of the liver may present a diagnostic and therapeutic challenge to the attending physician. One of the rare differentials of liver cyst is hydatid cyst. Hydatid cyst is an infection caused by the Echinococcus granulosus tapeworm that is endemic in the Mediterranean region, the Middle East, Australia and South America. Hydatid cysts are not endemic in Malaysia and are rarely seen. *Case presentation:* We hereby report a case of hydatid cyst of the liver in a 55-year-old Chinese-Australian woman who presented with a calcified liver cyst and negative hydatid serology. A liver segmentectomy was performed and revealed a well-circumscribed, calcified liver cyst containing only creamy whitish material without the typical daughter cyst. A histological examination revealed different layers of the cyst wall and the presence of loose, calcified scolices without a daughter cyst. *Discussion:* This case highlights the importance of considering hydatid cyst in the differential diagnosis of liver cyst even in non-endemic areas, as the ease of travelling and migration allows the condition to be seen outside the endemic region.

P-AP35. MicroRNA sponges as a potential therapeutic agent in cancer therapy

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Introduction: MicroRNA sponges are decoy mRNAs that compete with endogenous mRNA for base pairing with microRNAs. MicroRNAs are about 19-25 nucleotides in length and are now known to have important post-transcriptional roles in almost every cellular process such as development, metabolism and cancer. Sponges are potentially more efficient than antisense oligonucleotide to inhibit specific microRNAs. *Objective:* We aim to describe the use of microRNA sponges as a mean to inhibit the function of microRNAs. *Methods*: Two sponges were designed and constructed with the intention to soak up either miR-9 or isomiR-9 separately. The 3' UTRs of CDH1 containing the target site of miR-9, and DNMT3B with the target site of isomiR-9 were used as the templates for the construction of these sponges. The initial sponges that were created contained 6 multiple miRNA binding sites that are driven by a CMV promoter. Fixed amounts of pGL3-DNMT3B-3'UTR and isomiR-9 were transfected together with either pcDNA-miR-9 sponge or pcDNA-isomiR-9 sponge at different concentrations into HEK293 cells. The experiment was repeated using pGL3-CDH1-3'UTR and miR-9. *Results*: The 0 ng columns show the results for the inhibition of the pGL3 vector by miR-9 or by isomiR-9. The repression caused by miR-9 was alleviated by the introduction of 100 ng of miR-9 sponge but not by the isomiR-9 sponge. By contrast, isomiR-9 sponge partially alleviated the repression of isomiR-9 on DNMT3B-3'UTR but sponge miR-9 did not. *Conclusion*: MicroRNA sponge can effectively specifically inhibit microRNA function even with only one nucleotide difference in the microRNA. This shows that miRNA sponge could potentially be used to knockdown specific microRNAs. It would be interesting to investigate whether these sponges could be of therapeutic important in cancer therapy.

P-AP36. Construction of a lentiviral vector that expresses embryonic stem cells specific microRNA

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Introduction: Cloning and deep sequencing of human embryonic stem cells (hESCs) consistently identified the miR-302 cluster as the most abundant and specific miRNA in stem cells. The polycistronic miR-302 cluster encodes five miRNA genes that have an important role in the regulation of embryonic stem function. These five miRNA genes include miR-302a, b, c and d, and miR-367. Forced expression of the miR-302 cluster can reprogram somatic cells to pluripotent stem cells or can enhance the production of stem cells by OSKM factors. *Objective:* The aim of this study was to construct a lentiviral system that expresses the microRNA 302 cluster and to reprogram adult fibroblasts. *Methods*: MiR-302 cluster comprising of miR-302b, miR-302c, miR-302a, miR-302d and miR-367, accompanied by 120bp upstream and 150bp downstream of the cluster was amplified by PCR from human genomic DNA. As this vector has a red fluorescent protein (RFP) marker, the pTRIPz-302 cluster lentivirus was first tested by infecting HEK293 (human embryonic kidney) and MRC5 (human lung fibroblasts) cells to observe for RFP. Subsequently, northern blots of total RNAs collected from the infected HEK293 and MRC5 cells showed miRNA expressions from members of the miR-302 cluster, i.e., miR-302a, miR-302a* and miR-367. *Results*: These cells expressed RFP, formed colonies and were faintly positive toward alkaline phosphatase. In addition, RT-PCR showed these cells expressed low level of DNMT3B and Nanog, equivocal Oct4 and Lin28, but they did not express Sox2, suggesting that they were not fully reprogrammed. *Conclusion*: We showed that lentiviral vector is an effective way to introduce microRNA into fibroblasts. The low level of miR-302 cluster expression by pTRIPz-302 cluster in MRC5 cells might be the reason of the partial reprogramming.

P-AP37. Variable responses upon reprogramming of osteosarcoma cells

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Introduction: Osteosarcoma is a form of rare but most common primary malignancy to arise from bone. Cancer cells reprogramming has been studied due to its possibility to alter cancer properties which can be used to generate cancer-specific pluripotent cells for disease modeling and subsequent development of therapeutic strategies. *Objective:* In this study, we aim to assess the reprogramming response of different osteosarcoma cell lines. *Methods*: In this study, induced pluripotent cancer cells were generated from four osteosarcoma cell lines, Saos-2, G-292, U-2 OS and MG-63 using Yamanaka's four transcription factors, Oct3/4, Sox-2, c-Myc, and Klf-4. Characterisation studies of the reprogrammed osteosarcomas were carried out by observation of morphology changes, immunofluorescence and quantitative real time PCR detection of pluripotent markers and teratoma formation *in vivo*. Embryonic stem cells (ESC) were used as positive control. *Results*: All the reprogrammed osteosarcoma cell lines expressed the

pluripotent markers of Oct-3/4, SSEA-4, TRA-1-60 and TRA-1-81 comparable to ESC. U-2 OS and MG-63 were the most responsive to reprogramming with formation of highest number of ESC-like colonies. However, reprogrammed U-2 OS and MG-63 lost the ESC-like morphology after passage 15 while reprogrammed Saos-2 and G-292 were able to maintain their ESC-like morphology in prolonged culture. ESC-like morphology for reprogrammed Saos-2 and G-292 was observed up till passage 30. From the four reprogrammed osteosarcoma cell lines, only reprogrammed G-292 maintained the aggressiveness of cancer and formed benign teratoma *in vivo*, while Saos-2 and U-2 OS appeared to have lost the aggressiveness of cancer upon reprogramming with no teratoma formed. *Conclusion*: We found that the efficiency of reprogramming did not correlate with the sustainability of the ESC-like morphology. Thus, the variable responses displayed by osteosarcoma cell lines upon reprogramming could be due to the heterogeneity in osteosarcoma cell lines.

P-AP38. A case of Thyroid Transcription Factor-1 (TTF-1) positive primary neuroendocrine carcinoma of oesophagus: a potential diagnostic caveat

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Introduction: Thyroid transcription factor-1 (TTF-1), a 38-kDa homeodomain-containing nuclear protein of the *Nkx-2* gene family, plays an important role in regulating genes in the developing thyroid, lung and brain tissue. It is expressed in almost 90% of pulmonary small cell carcinoma. Therefore, it is routinely used to determine the possibility of primary lung in cases of metastatic neuroendocrine carcinoma (of small-cell type). We described a rare case of primary neuroendocrine carcinoma of the oesophagus which was TTF-1 positive. *Case presentation:* The patient, a 37-year-old man, presented with a three-month history of progressive dysphagia, associated with significant loss of weight and loss of appetite. Oesophagogastroduodenoscopy (OGDS) showed a friable fungating tumour at the upper mid oesophagus located 21 to 28 cm from incisura. Biopsy examination revealed nests and sheets of malignant small round blue cells displaying hyperchromatic nuclei with inconspicuous nucleoli and scanty cytoplasm. Nuclear molding was occasionally noted. Numerous mitosis and apoptotic bodies were seen. Immunohistochemically, the malignant cells expressed neuroendocrine markers (chromogranin, synaptophysin and CD56). Of interest is the immunostaining for TTF-1, which was initially performed to exclude lung primary, was found positive in the present case. However, CT thorax showed no lung lesion apart from a few enlarged mediastinal nodes which were suspicious of metastasis. In the light of the clinical, endoscopic and radiological findings, a diagnosis of primary neuroendocrine carcinoma of the oesophagus was rendered. *Discussion:* TTF-1 expression is not unique to pulmonary small cell carcinoma, and its expression in other extrapulmonary neuroendocrine carcinoma (of small-cell type) has been previously reported. Hence, we proposed that TTF-1 immunopositivity should be cautiously interpreted in the light of clinical context in cases of extrapulmonary neuroendocrine carcinoma.

P-AP39. Pap smear screening history in urban women with invasive cervical cancer: An 8-year observational case-control study

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Introduction: Despite the success of cervical (Pap) smear screening in preventing invasive cervical cancer (ICC) as reported in many developed countries, the incidence of ICC in developing countries remains worryingly high. This may reflect the failure of screening programme initiated in these countries including Malaysia. *Objective:* The aim of our study was to investigate the Pap smear screening histories among patients diagnosed with ICC in Universiti Kebangsaan Malaysia Medical Centre and the optimum Pap smear screening interval for it to be effective. *Methods:* A hospital-based case-control study which included 192 cases of histologically confirmed ICC diagnosed over a period of eight years (2006 – 2014) and 192 randomly selected control groups with similar birth years. Their respective Pap smear screening histories were recorded. *Results:* Pap smear screening uptake for both case and control subjects during the study period were alarmingly low, with only 26.0% (n = 50) and 39.6% (n = 76) respectively reported to have previous Pap test. Of these 50 case subjects, 45 (90.0%) had Pap smears following a clinical presentation and only 11 (22.0%) had had prior (negative) screening smears. There was significant protective benefit if the screening interval was less than 36 months and not thereafter (for duration 7 – 12 months: OR = 0.07, 95% CI = 0.01 – 0.95; 13 – 36 months: OR = 0.07, 95% CI = 0.01 – 0.97). In addition, we also found that Chinese women had twice the odds of getting ICC compared to Malay women. *Conclusion:* Pap smear screening uptake in Malaysia is still low. Current screening programme has to be enhanced and improved to increase the Pap smear screening coverage. This study reinforces the importance of compliance to a screening interval of at most three years to reduce the incidence of largely preventable ICC among Malaysian women.

P-AP40. Papillary squamo-transitional cell carcinoma of the uterine cervix: a case report

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Introduction: Papillary squamo-transitional cell carcinoma (PSTC) is a rare cervical malignancy. It is a distinctive subcategory of squamous cell carcinoma of the uterine cervix. Its exact clinicopathological features are still unknown. Less than 150 cases have been reported worldwide, none from Malaysia. *Case presentation*: A forty year old Malay lady presented with a 2 months history of persistent per vaginal bleeding and post coital bleeding. An irregular fungating growth was noted arising from posterior cervical lips on speculum examination. Pap smear showed atypical squamous cell suspicious of high-grade lesion while the cervical biopsy revealed squamous cell carcinoma. A bulky, heterogeneously enhancing mass involving the cervix and lower portion of the uterus was found on radiological imaging. Further clinical staging concluded a fungating growth on the posterior lip extending into the internal os and lower part of the uterus. Histopathological examination diagnosed a papillary squamo-transitional cell carcinoma. The carcinoma was characterised by papillomatous architecture of neoplastic cells with fibrovascular core indistinguishable from the transitional cell carcinoma in the urinary tract. However, the immunohistochemical findings were similar to squamous cell carcinoma with associated HPV infection (CK7+/ CK20-/ p63+/ CK5/6+/ HPV+/ p53+/ p16+). She subsequently had a radical hysterectomy and similar histology findings were concluded. *Discussion:* The papillary squamo-transitional cell

carcinoma of the uterine cervix is indistinguishable from the commoner benign papillary lesions or its counterpart in the urinary tract. Thorough examination, knowledge and suspicion of this potentially aggressive tumour are essential as it poses a diagnostic challenge clinically and histologically.

P-CP01. 'Rack & Ruler' Improves Laboratory Specimen Rejection Rate

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Introduction: The comprehensive laboratory testing process must be monitored to ensure reliable test results. Preanalytical phase contributes to 32%-70% of the total errors in laboratory. It is crucial to detect the root cause of the problem and implement necessary corrective action for patient diagnosis, treatment and total cost of the medical treatment. The aim of this project is to reduce rejection rate due to haemolysis by using simple sample collection tools. *Materials & Methods:* Baseline specimen rejection data was collected for three months. The main cause and site of rejection was identified. A simple gadget; JPMD EZY RULER which contains a "reminder" of the pre analytical DOs and DON'Ts and JPMD EZY RACK which provides guidance on the draw of orders and mixing of samples were designed to minimise sampling errors. Training sessions were carried out to improve the preanalytical knowledge gadgets introduced to help in minimising sampling errors. The rejection rate was monitored and compared with baseline data. *Results:* The baseline rejection rate was 1.38%. The main cause of rejection was haemolysis and the main sites of rejection were the Neonatal Intensive Care Unit and Emergency Department. The specimen rejection rate was reduced significantly to 0.74% post intervention and consistently maintained at ≤ 0.8 at present which fulfilled the National Indicator Standard for rejection rate (0.3 - 0.8%). *Discussion:* Haemolysis mainly occurs during the preanalytical phase of sample collection. The main factor that contributed to this is lack of knowledge of phlebotomists on preanalytical factors. With these innovations, the phlebotomists are educated on the correct ways of blood sampling and this has resulted in the reduction of rejection rates. In conclusion, these innovations have improved the rejection rate, reduced the rate of re-bleed and provided results in a timely manner for overall management of patients.

P-CP02. Effect of Handedness and Sample Collection Types on Blood Lactate Measurements

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Introduction: Blood (plasma) lactate levels estimated from samples collected from different body sites may vary. Lactic acid is produced by skeletal muscle, and since skeletal muscle mass has been shown to be higher on the dominant upper limb, we hypothesised that blood lactate levels from the "dominant" upper limb will be higher than that of the "non-dominant" side at rest. This study was aimed to assess the effect of handedness on blood lactate levels drawn from the right and left upper limb. *Materials & Methods:* We compared lactate levels in venous blood samples from the right and left antecubital vein and an arterial sample collected from the right upper limb from 14 men and six women. A mandatory 30-minute rest period was maintained prior to the collection. Samples were collected in fluoride/EDTA tubes without the application of tourniquet and were kept on ice at 4°C. Samples were then immediately centrifuged at 4°C and lactate was analysed immediately using a spectrophotometric method. *Results:* The mean arterial lactate level was 6.33 ± 1.91 mmol/l, while the average venous levels were 6.33 ± 1.82 mmol/l. Lactate from blood sampled from the left arm was

6.35 ±1.89 mmol/l while from the right arm it was 6.31± 1.80 mmol/l. The values when compared with paired student's t test showed no significant difference between the lactate levels from the three collection points. *Discussion:* We found that blood lactate levels between the right and left upper limb sites were comparable. They were also comparable with the arterial blood lactate levels. These findings were observed in both male and female subjects. Thus, we conclude that in our sample of subjects' handedness or site of collection of blood sample had no effect on the lactate levels and handedness was not influenced by the sex of the subjects.

P-CP03. Investigation of Urine Crosslinked N-Telopeptides of Type I Collagen (NTX) Levels of Obese and Non-obese Postmenopausal Women

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Introduction: Osteoporosis is an important health problem in postmenopausal women resulting in increase in bone fragility and fractures. In previous studies, urinary cross linked N-telopeptides of Type I Collagen (NTX) levels have been investigated for their potential use as a parameter and marker of bone resorption. This study aimed to investigate urine NTX levels in obese and non-obese postmenopausal women to determine if it can be used as a bone turnover biomarker in these subjects. *Materials & Methods:* Forty obese postmenopausal women (BMI>30 kg/m^2, 53.3 ± 5.1 years old) and 26 non-obese postmenopausal women (BMI between 18 to <24.9 kg/m^2, 55.9 ± 6.5 years old) were included in the study. Urinary NTX and creatinine levels of all subjects were measured by ELISA and Jaffe methods, respectively. The ratio of urinary NTX levels to urinary creatinine levels was determined. *Results:* Urinary NTX:creatinine levels were significantly higher (p=0.009) in obese than in non-obese postmenopausal women. *Discussion:* The underlying relationship between obesity and bone is complex and continues to be an active research area. It is traditionally believed that obesity is beneficial to bone health because of well-established positive effect of mechanical loading conferred by body weight on bone formation. However, whether the mass derived from obesity or excessive fat accumulation is beneficial to bone remains controversial. In this study, our finding of increased urinary NTX levels in postmenopausal obese women suggests that obesity plays a role towards osteoporosis in postmenopausal obese women. Thus, we concluded that urinary NTX level may be used as a marker to investigate bone turnover in postmenopausal obese women.

P-CP04. A Review of Serum Potassium Concentration during Aldosterone Renin Ratio Blood Sampling in Hospital Pulau Pinang

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Introduction: With the introduction of the aldosterone renin ratio (ARR) as a screening test, the detection rate of primary aldosteronism has increased considerably. Although the ARR is a reliable screening test for primary aldosteronism, false positives and negatives occur. Optimal performance of this test consists of eliminating factors that can affect renin and aldosterone concentration such as correction of hypokalaemia with potassium chloride supplements and cessation of certain antihypertensives for at least two weeks before blood sampling. This is a retrospective study to determine the serum potassium concentration when ARR was requested in Hospital Pulau Pinang and its effects on the ARR result. *Materials & Methods:* Results of serum potassium concentration for each patient requested for ARR in the year 2013 were obtained from Laboratory Information System, Hospital Pulau Pinang. *Results:*

There were 65 cases of ARR requested in 2013 from Hospital Pulau Pinang and sent for analysis in Hospital Putrajaya. There were nine cases of raised ARR and 56 cases of normal ARR. From the 56 cases of normal ARR, 54% of the requests showed serum potassium concentration of less than 4 mmol/L during the blood sampling. *Discussion:* The ARR is only a screening test and should be repeated once or more before deciding whether to proceed to confirmatory suppression testing. Because potassium regulates aldosterone, uncorrected hypokalaemia may lead to false negative results. It is recommended to optimise the potassium level to at least 4 mmol/L before sampling for ARR. In this study, the 54% cases of normal ARR could not be confirmed as the low potassium level may lead to false negative results. It is important for pathologists to advice clinicians on the optimisation of factors affecting patient's results before the sampling of ARR. This is to prevent the misinterpretation of results that may impact patient outcome.

P-CP05. A Comparison of Atherogenic Markers in Metabolic Syndrome

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Introduction: Metabolic syndrome (MS) is a constellation of risk factors that increases an individual's cardiovascular risk. Small dense low density lipoprotein cholesterol (sdLDL), non-high density lipoprotein cholesterol (non-HDL), and the ratio of triglyceride (TG) to high density lipoprotein cholesterol (HDL) [TG/HDL ratio] are markers indicating atherogenicity and increased risk for development of coronary heart disease. We compared the levels of these atherogenic markers in subjects with and without MS, and determined the correlation of the markers with MS components. *Materials & Methods:* Anthropometric measurements and fasting blood samples were obtained from 63 subjects who were then divided into MS, central obesity without MS (COBXMS) and lean normal control (NC) groups. Fasting plasma glucose (FPG) and lipid profile were analysed using standardised enzymatic assays. Serum sdLDL concentration was measured using a homogenous enzymatic assay (Randox sLDL-EX"SEIKEN" on Roche Cobas Integra 400); non-HDL was calculated by subtracting HDL from total cholesterol, while TG/HDL ratio was obtained by dividing TG with HDL. Levels of the atherogenic markers were compared between the three groups and evaluated for correlations with the components of MS (waist circumference, blood pressure, TG, HDL and FPG levels). *Results:* The levels of sdLDL, non-HDL and TG/HDL ratio were significantly higher in the MS group compared to the COBXMS and NC groups. TG/HDL ratio demonstrated significant correlation with all components of MS, while sdLDL was significantly associated with all MS components except one (diastolic blood pressure). In contrast, non-HDL was correlated with only two MS components (waist circumference and TG). The atherogenic markers showed significant correlation with each other. *Discussion:* Subjects with MS have significantly elevated atherogenic markers compared to those without MS, which may partially explain the increased cardiovascular risk in MS. TG/HDL ratio demonstrated the strongest association with MS components.

P-CP06. A Comparison between Spot Urine Protein and 24-Hour Urine Protein as Biomarkers for the Detection of Urinary Paraprotein in Multiple Myeloma

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Introduction: Multiple myeloma (MM) is a haematological malignant disease characterised by plasmacytosis leading to excess secretion of abnormal monoclonal immunoglobulin called paraprotein. In the urine, it is termed Bence-Jones protein (BJP). The gold standard for detection and quantification of BJP is based on a 24-hour urine (24-HU) sample which is inconvenient and frequently unreliable due to its heavy dependence on patient compliance. Spot urine sample has been used instead. This study compared the correlation between spot urine in the form of early morning sample (EMU) and 24-HU collection in determining BJP in patients with MM. *Materials & Methods:* This was a prospective cross sectional study. Seven MM patients were recruited within a period of two months. Patients were instructed to collect 24-HU and EMU. All serum and urine samples were analysed quantitatively for protein and creatinine on Cobas 702 (Roche). Electrophoresis (SUEP) and densitometric scanning of electrophoretic patterns were then performed on Hydrasys (Sebia) platform. Immunofixation (IFE) was done on Hyrys (Sebia) platform to confirm the results of SUEP. *Results:* Only six patients had presence of BJP in the urine confirmed by IFE. Three patients had BJP detected in EMU which was non-detectable in their 24-HU samples. One patient had BJP detected in 24-HU sample which was not detected in EMU. There was a weak linear relationship between the total urine protein concentration assayed using both methods ($y=0.2479x + 68.667$, $R^2 = 0.1003$). The BJP concentrations between the methods however, were positively correlated ($y= 1.9932x – 0.0117$, $R^2 = 0.9645$). *Conclusion:* We propose the use of EMU as an alternative for 24-HU collection in terms of quantification of BJP in monitoring disease progress. However, more prospective studies with larger sample numbers would need to be done to confirm this pilot study and to assess its diagnostic sensitivity and specificity in MM.

P-CP07. The Effect of *Channa Striatus* (Haruan) Extract on Total Antioxidant Status (TAS) during Wound Healing in Post Lower Segment Caesarean Section (LSCS) Women

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Introduction: Channa striatus (Haruan) is widely consumed in Malaysia to promote wound healing. Wound healing is a dynamic process that involves three overlapping phases; inflammation, new tissue formation and tissue remodeling. During inflammatory phase, large amount of reactive oxygen species (ROS) are produced resulting in cellular damage which in turn delays wound healing. *Channa striatus* has been proposed to have antioxidant properties counteracting ROS and enhancing wound healing. This study was done to compare the level of total antioxidant status (TAS) in patients receiving *Channa striatus* extract and placebo during wound healing of post Lower Segment Caesarean Section (LSCS) women. *Materials & Methods:* This was a randomised; double blinded, placebo-controlled study conducted in Hospital Universiti Sains Malaysia (HUSM). The treatment group consumed 500mg of freeze dried *Channa striatus* extract daily while the placebo group consumed 500mg of maltodextrin daily for 4 weeks. Venous blood was taken from each subject postoperatively at day 1, week 2, week 4 and week 6 and were analysed for TAS level using Selectra E machine. Data analysis was done

using SPSS Version 20. *Results:* A total of 73 patients were studied, 39 patients consumed *Channna striatus* and 34 consumed maltodextrin. The TAS levels in patients who consumed *Channa striatus* were significantly different within group between day 1 and week 2, week 4 and week 6. Meanwhile in the placebo group, TAS level were only significant in week 4 and week 6. There were no significant differences in the TAS levels between both groups in all the weeks. *Discussion:* Although there were no significant difference between *Channa striatus* and placebo group, patients who were supplemented with the *Channa striatus* extract had increased TAS levels from week 2 onwards. The increment in the TAS levels might be involved in the enhancement of wound healing process in patients, however further studies are required to confirm it.

P-CP08. Diagnostic and Prognostic Value of Procalcitonin in Patients with Septic Shock admitted to the General Intensive Care Unit, Hospital Kuala Lumpur, Malaysia

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Introduction: Sepsis is a common cause of morbidity and mortality in intensive care units (ICU). The diagnosis of bacterial infection is challenging because clinical presentations of severe infection and systemic inflammation due to non-infectious causes, can be very similar. The parameters of infection such as body temperature, heart rate and leukocyte counts lack sensitivity and specificity. Procalcitonin (PCT) is a promising marker for early diagnosis of sepsis. This study aims to assess the diagnostic value of PCT by comparing its level in septic shock and non-septic shock patients and to assess its prognostic value by comparing its level in survivors and non-survivors. *Materials & Methods:* A cross-sectional study was carried out in General Intensive Care Unit, Hospital Kuala Lumpur for a period of nine months. 47 patients who presented with clinical evidence of shock according to American College of Chest Physician/Society of Critical Care Medicine 1991 Consensus Guidelines were recruited and divided into septic and non-septic shock groups. *Results:* PCT level was significantly higher in septic compared with non-septic shock patients [31.62 ng/ml (8.08-86.23) vs 1.35 ng/ml (0.61-6.17) respectively]. As a screening test, a PCT cut-off value of 1.28ng/ml yielded 100% sensitivity and 50% specificity (NPV of 100%, PPV of 88%) for discriminating septic and non-septic groups. As a diagnostic test, PCT cut-off value of ≥12.40ng/ml yielded 100% specificity and 68% sensitivity (PPV of 100%, NPV of 46%). The area under the ROC curve was 0.914 (95% Confidence Interval, 0.83-1.00). In a multivariate logistic regression analysis, PCT did not independently predict the mortality of septic shock patients. *Discussion:* In this study, we found that PCT has a role in ICU setting in terms of ruling out sepsis in patients presented with shock. However, its role in determining the survival of patients with sepsis was not established in this study.

P-CP09. Assessing HbA1c Knowledge among Type 2 Diabetic Patients in Hospital Putrajaya

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Introduction: The glycosylated haemoglobin (HbA1c) test is used for evaluating long term glycaemic control. The aim of this research was to assess HbA1c knowledge among type 2 DM patients in a tertiary endocrine centre in Malaysia. *Materials &Methods:* This was a cross-sectional descriptive study among Type 2 DM patients undergoing routine follow up in the endocrine clinic of Hospital Putrajaya. Patients were invited to answer a validated questionnaire which assessed their knowledge on HbA1c. Their last HbA1c results were retrieved from the laboratory information system. *Results:* A total of 92 participants were recruited. Fifty-six (60.9%) were aware of the term HbA1c. Out of those who were aware of the term HbA1c, 37 (66.1%) knew the correct indication for HbA1c measurement. Fifty one (91.1%) knew their HbA1c target goal out of which 30 (58.8%) had achieved this target. *Conclusion:* The level of HbA1c knowledge was acceptable. However, continuing efforts must be made to improve patients understanding of their disease and clinical disease biomarkers.

P-CP10. Prevalence of Vitamin D Deficiency in Women in North Sumatera, Indonesia determined by Chemiluminescent Immunoassay Method

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Introduction: Previous studies reported low levels of serum 25-hydroxy vitamin D [25(OH)D] in women from temperate and tropical countries but there are few such studies in Indonesia. This study aims to assess the serum vitamin D status of Indonesian women and its associated factors. *Materials & Methods:* A cross-sectional study was conducted on 156 apparently healthy women during the dry season in North Sumatera. The parameters studied include duration of sun ray exposure, intake of vitamin D food source, reported physical activity, percentage of body fat (by bioelectrical impedance analysis), and serum 25(OH)D levels (by chemiluminescent immunoassay). Subjects were categorised into deficient (<20 ng/mL), insufficient (20-32 ng/mL), sufficient (32-54 ng/mL), and normal (54-90 ng/mL) based on serum 25(OH)D levels. Pearson correlation, independent t-test, and one way Anova tests were used for statistical analysis. *Results:* The mean age of subjects was 35.6±7.7 years, with about 70% of them working indoors. Fifty percent (n=78) of the women were obese. Majority (82.7%, n=129) had low vitamin D intake (<10mg/day). More than half (52.6%, n=82) of the subjects had sun ray exposure < 1 hour whereas 62.2% (n=97) wore "hijab" covering their arms and legs. A low physical activity level was reported in 65.4% (n=102). There were 148 (95%) women with vitamin D deficiency-insufficiency whilst 5% in the sufficiency category. The mean serum 25(OH)D level was 17.71 ng/mL (95% confidence interval 16.22-19.34 ng/mL). There were positive associations between ethnicity, occupation, vitamin D food source intake, sun ray exposure and physical activity with serum 25(OH)D concentration. *Discussion:* Majority of women in North Sumatera, Indonesia were found to be vitamin D deficient-insufficient by chemiluminescent immunoassay method. It can be concluded that vitamin D deficiency can occur in women living in a tropical country, especially with sun-avoiding lifestyle, indoor occupation, and low intake of vitamin D food source.

P-CP11. Real-Time PCR Detected Silent Mutation of Vitamin D Receptor Gene in Indonesian Women

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Introduction: Studies have shown that low serum 25-hydroxy vitamin D [25(OH)D] levels may lead to an increase in mortality and morbidity, especially in women. Many factors are linked to vitamin D deficiency in tropical countries. We aimed to examine factors associated with serum 25(OH)D levels, including single nucleotide polymorphisms of vitamin D receptor/VDR genes (*Taq*I and *Bsm*I), lifestyle (sun exposure, intake of vitamin D and physical activity), and percentage of body fat in Indonesian women. *Materials & Methods:* This was a cross sectional study, conducted in government institutions, private institutions, and private residences in the City of Medan, North Sumatera, Indonesia. Based on the serum 25(OH)D levels (by chemiluminescent immunoassay method), subjects were categorised into deficient(<20 ng/mL), insufficient (20-32 ng/mL), sufficient (32-54 ng/mL) and normal (54-90 ng/mL). Deoxyribonucleic Acids (DNA) isolation was conducted using 'salting out' method. Both VDR genes, rs731236 and rs1544410, were evaluated in regard to allele discrimination using *StepOnePlus*™*Real Time* PCR device, with added TaqMan probes. *Results:* There were 148 subjects categorised as deficient/insufficient and eight as sufficient 25(OH)D levels. However, none of the subjects achieved normal 25(OH)D values. All subjects were heterozygous (TC for *Taq*I and AG for *Bsm*I) using Real Time PCR and had low sun ray exposure, high body fat and low intake of vitamin D. Additionally, vitamin D deficiency/insufficiency was associated with occupation, sun exposure, physical activity, vitamin D intake and body fat. *Discussion:* This study found that from all varieties of single nucleotide polymorphism, only one heterozygous cluster was found. There were no homozygous wildtype or homozygous mutant gene found in association with low serum 25(OH)D level. This silent mutation form eventually may cause an evolution in human, which affects the susceptibility against diseases. *Conclusion:* Vitamin D deficiency may occur in women with polymorphisms of the vitamin D receptor genes *Taq*I and *Bsm*I.

P-CP12. Association between Glycaemic Control and Dyslipidaemia in Type 2 Diabetic Patients in Hospital Putrajaya

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Introduction: The prevalence of diabetes mellitus (DM) in Malaysia is increasing drastically. Patients with diabetic dyslipidaemia are at a greater risk of developing cardiovascular disease. There is limited data on the association between glycated haemoglobin (HbA1c) and dyslipidaemia in type 2 DM (T2DM) patients in multiethnic Malaysian population. The aim of this study was to determine the association between glycaemic control and dyslipidaemia in dyslipidaemic T2DM patients in selected population in Malaysia. *Materials & Methods:* This was a cross-sectional study using retrospective data from January 2009 to December 2012 of 214 T2DM patients with dyslipidaemia, carried out at the endocrine clinic, Hospital Putrajaya, a tertiary endocrine centre in Malaysia. Data was analysed using IBM SPSS Statistics version 21.0 for Windows. *Results:* Fasting blood glucose (FBG) and HbA1c were significantly correlated with total cholesterol (TC), triglyceride (TG), low density lipoprotein cholesterol (LDL), non-high density lipoprotein cholesterol (non-HDL), LDL/HDL ratio and TC/HDL ratio. The magnitude of correlation for all these lipid parameters was greater with HbA1c than FBG.

The correlation coefficient of HbA1c was higher with TC/HDL ratio than other lipid parameters. The level of TC, TG, non-HDL and TC/HDL ratio were significantly higher in patients with HbA1c ≥ 6.5% as compared to patients with HbA1c < 6.5%. In patients on statin treatment, non-HDL, LDL/HDL ratio, TC/HDL ratio and HbA1c were significantly lower than non-treated patients. *Conclusion:* These findings indicate significant association between glycaemic control and dyslipidaemia in T2DM patients. Thus, in addition to glycaemic control, HbA1c can be potentially used as a biomarker for dyslipidaemia and as an indirect marker for predicting CVD risk in T2DM patients.

P-CP13. Subclinical hypothyroidism among patients with depressive disorders

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Introduction: Subclinical hypothyroidism (SHT) is a biochemical diagnosis, defined as an elevated thyroid stimulating hormone (TSH) with normal free thyroxine (FT4). It affects 4-10% of the adult population and is more prevalent in elderly women. Its commonest cause is autoimmune thyroiditis, detected by anti-thyroid peroxidase antibody (TPO-Ab). About 2-5% of SHT patients progress to overt hypothyroidism annually. The SHT prevalence among depressed patients ranges between 3% and 17%. This study aimed to determine the prevalence of SHT and TPO-Ab positivity among patients diagnosed with depressive disorders. *Materials & Methods:* It was a cross sectional study carried out in Universiti Kebangsaan Malaysia Medical Centre over a 12 months period. Serum TSH, FT4 and TPO-Ab were measured. *Results:* Results showed that 82% of depressed patients were euthyroid, 4% had SHT, 11% had subclinical hyperthyroidism and 2% had discordant thyroid function. TPO-Ab positivity among the subjects was 7%. Among patients with SHT, only one had positive anti-TPO. *Conclusion:* The prevalence of SHT and TPO-Ab positivity in the study population, at 4% and 7%, respectively, were comparable to previous findings.

P-FP01. Pattern Of Completed Suicides In Johor As Reported To National Suicide Registry Malaysia (NSRM)

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Introduction: Suicide is defined as a conscious act of self- induced annihilation occuring in a life situation in which death is felt to be the best possible solution. Factors associated with suicidal behaviour are multifaceted that interact in complex ways. We conducted this study to analyse the epidemiology, circumstances of death and significant life events data of suicide victims in Johor for the year 2013, as reported to National Suicide Registry Malaysia (NSRM). *Materials & methods:* Completed suicide cases reported to NSRM in Johor during one year study period from 1st January 2013 until 31st December 2013 were recruited in the study. Background epidemiology, significant life events and detailed circumstances of death data were collected and analysed. *Results:* There were 87 cases of suicide deaths occured during this study period which brings the suicide incidence rate in Johor to 2.69 per 100,000 population. Local Malaysian citizen represented 73 (83.9%) of total victims. Suicides was higher among male (85.1%, n=74) and of Chinese ethnicity (47.1%, n=41). The majority of victims belonged to the age group of 31 – 35 years (23%, n=20) Victims' background education level were mostly up to secondary school (40.2%, n=35). The most common suicide method used was hanging (59.8%, n=52), followed by poisoning (21.8%, n=19) and jumping from a height (9%,

n=8). Muar district registered the highest incidence rate (7.53 per 100,000) followed by Segamat and Johor Bahru. A high proportion of victims (71.3%, n=62) experienced significant emotional life events prior to committing suicide. *Discussion:* Johor recorded incidence rate of suicide at 2.87 per 100,000 population. Muar and Segamat registered the highest rate amongst all districts suggesting that people living in rural regions are at a higher risk of suicide than their more urbanized area counterparts.

P-FP02. Group B Streptococcus Pneumonia In Sudden Unexpected Infant Death

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Introduction: Group B Streptococcus (GBS) is a common cause of infection in newborn and early infancy. However, GBS infection in an infant older than three months is infrequently reported in the literature. We reported a case of sudden death of an apparently well six month old infant due to GBS pneumonia, diagnosed at autopsy. This case highlighted the importance of a detailed autopsy in sudden unexpected death in infancy (SUDI) and the crucial role of post-mortem microbiological study in such cases. Relevant autopsy protocols that need to be employed during microbiological sampling are briefly discussed. *Case:* A six month old of apparently well, male infant was brought in dead to the Emergency Department of Hospital Sultanah Aminah. He underwent medicolegal autopsy four hours after death, as part of an overall SUDI investigation. Internal examination showed generalised hyperinflation with patchy consolidation of the upper and middle lobes of lungs. Multiple matted mesenteric lymphadenopathy were also detected. Blood and lung tissue specimens collected under aseptic technique yielded growth of GBS. Virology studies from the heart and lung tissues were negative for common viral infections. Postmortem histology from consolidated lungs confirmed pneumonic features while mesenteric lymph nodes showed reactive changes in-keeping with underlying infective process. Death was attributed to GBS pneumonia. *Discussion:* GBS infection in infant older than three month old is associated with various predisposing risk factors with primary viral infection leading to secondary GBS sepsis as one of the suggested risk factors. Proper autopsy protocols including detailed microbiological investigations should be part of overall SUDI investigation. Postmortem should be preferably performed as early as possible and multi-site samplings are highly recommended. Microbiology results must be interpreted in conjunction with clinical history, histology findings, antibiotic sensitivity pattern and pathogenicity of organisms isolated in the livings.

P-FP03. Sudden death among young adults: Is postmortem troponin-T analysis helpful?

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Introduction: Sudden unexplained death among young adults is commonly encountered at autopsy. Cardiac causes may be attributed to congenital heart defects, genetic abnormalities causing lethal arrhythmia, cardiomyopathies and other rare non-ischemic causes. The use of cardiac troponin T, a marker for myocardial injury has been recommended and refuted in the evaluation of the presence and extent of myocardial damage in various types of death. The objective of this study is to describe the findings of cardiac troponin T post mortem serum in different groups of causes of death. *Methods:* Sixty nine medico-legal autopsy cases were recruited from Department of Forensic Medicine, Hospital Sungai Buloh, Selangor and Department of Forensic Medicine, Hospital Sultanah Aminah, Johor Bahru,

Johor for 8 months duration from Jun 2013 until February 2014. Inclusion criteria were adults of 18-50 years of age and refrigerated body with post mortem interval < 48 hours. Autopsy cases were divided into five groups; cardiovascular diseases (CVD), thoracic trauma (TT), non-thoracic trauma (NTT), sudden unexplained death (SUD) and other diseases (OD). Blood samples were collected and stored at -20 ºC. Troponin T analysis was performed by using electro-chemiluminescence immunoassay method on an automated analyzer (Elecsys 2010, Roche Systems, Germany). *Results:* A total of 69 (male=63, female=6) autopsy cases were selected; CVD (n=21), SUD (n=5), TT (n=12), NTT (n=28), OD (n=3). Troponin T level (mean±SD) in cases of CVD, SUD, TT, NTT, and OD are 14.7±47.1 µg/L, 24.7±42.3 µg/L, 6.7±83.2 µg/L, 21.9±53.1µg/L, 13.7±14.0 µg/L respectively. There was no significant different of troponin T level in different causes of death (p≥0.05). *Conclusion:* The result of this study generally indicates that post mortem cardiac troponin T is not a helpful biomarker to support the diagnosis of myocardial injury in the various types of deaths.

P-FP04. Effectiveness of Bone Cleaning Process Using Chemical and Entomology Approaches: Time and Cost

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Introduction: The objective of this study was to determine the most cost effective and efficient method for the process of bone cleaning and comparison between chemical and entomology approach which can be used dynamically in the setting of National Institute of Forensic Medicine Malaysia HKL. *Methods:* A total of 10 subjects with ages ranging from 20 to 40 years old were chosen in this descriptive cross sectional research. Normal intact sternum bones were taken from subjects of uncomplicated postmortem cases and subjected to the bone cleaning procedures. A total of four methods were used including the two chemical approaches i.e. laundry detergent and the combination of 6% hydrogen peroxide and powder sodium bicarbonate; whilst the other two entomology approaches were by using 2nd instar maggots of *Chrysomyia rufifacies* and *Ophyra spinigera*. *Results:* Based on the average weight reduction 1.4 gm per day, median number of 11.3 days to achieve the desired score and an average cost MYR 4.10 per case used for each method, bone cleaning using maggots was the most efficient and cost effective method. Even though laundry detergent would be slightly faster with a median number of 7.5 days for the cleaning process, its decalcification effect had made the bone become brittle with misleading weight reduction. However, the number of maggots used could be increased to achieve a faster cleaning process without increasing the cost if compared between detergent and entomology method. *Conclusion:* The outcome of this study was supported by the analysis conducted by the Forensic Specialists using blind validation with the achievement of 77.8% of chosen entomology method. Furthermore, Emission Scanning Electron Microscope (SEM) evaluation profile also revealed that bone cleaning using entomology methods especially maggots *Chrysomyia rufifacies* species preserved the original condition of the bones better in terms of the clarity of the injuries and morphological features on the bones which was consistent with the results of this study.

P-FP05. Postmortem CT Scan in Sudden Natural Death: A Case Report Of Pneumothorax Due To Metastatic Lung Carcinoma

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Introduction: Lung cancer is the leading cause of cancer deaths in Malaysia. Deaths in patient with lung cancer can be due to wholly by the primary lung tumour burden or complications from the extensive lung cancer metastases. Spontaneous pneumothorax is a rare manifestation of lung cancer and prognosis is poor because most often it is diagnosed at advanced stage of lung cancer. More common presentation of advanced lung cancer includes bony metastases. Postmortem Computed Topography (CT) scan is useful before autopsy of natural deaths to show certain pathological processes, such as pneumothorax and bony metastasis which may go undetected during routine dissection. *Case:* We are reporting a case of fatal right pneumothorax as manifestation of advanced primary lung cancer. A 42 years old Indonesian lady with no known medical illness was found dead at home. She had loss of appetite, loss of weight and became activities of daily living (ADL)-dependent for few months prior to her death. Postmortem CT scan prior to conventional autopsy showed evidence of right pneumothorax with consolidations of bilateral lungs and hyperdense lesions at the lumbar vertebra. External examination revealed a cachexic adult female with chest asymmetry, in which the right chest appeared higher than the left chest and was resonant on percussion. The right pneumothorax was proved during autopsy. Internal examination also showed extensive lung cancer with metastases to regional lymph nodes, liver, adrenal glands and lumbar vertebra. Histological examination with special stains confirmed presence of primary lung adenocarcinoma with distant metastases. *Discussions:* This case illustrates the importance of Postmortem CT Scan as adjunct or supplement to guide the conventional autopsy, especially in detecting certain pathological processes that may go undetected during routine dissection. Therefore, more comprehensive and detailed autopsy reports can be achieved. However, conventional autopsy techniques consist of external evisceration, dissection of major organs with macroscopic and microscopic identification of pathological processes remains gold standard in natural deaths.

P-FP06. An Autopsy of a Case of Tuberculosis Pericarditis

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Introduction: Tuberculous (TB) pericarditis is a rare but life-threatening condition. It may lead to diastolic heart failure in constrictive pericarditis. *Case:* This is a case of a 31 year-old vagabond man, found dead at a pavement in the city. Generally, he had good built with body mass index of 25 but unkempt with poor hygiene. Externally there were injection marks at the right inguinal region and right side of neck. There was also distended neck veins and bilateral pedal edema. On internal examination, there was symmetrical serous pleural effusion and ascites. However the pleural fluid was negative for tuberculosis and the viral screening for hepatitis and HIV were nonreactive. Internal examination showed the lungs were grossly edematous and the pericardium was thickened, calcified and intensively adherent to the sternum, diaphragm and posterior chest wall. Other organs were unremarkable. The heart was removed together with the pericardium because it was inseparable. Surprisingly, the thickness of the ventricle walls was within the normal limits and the valves were unremarkable. Microscopic examination of the pericardium revealed multiple central caseation necrosis surrounded by elongated epithelioid cells with lymphocytes infiltration at the peripheries. The diagnosis of congestive cardiac failure secondary to chronic constrictive pericarditis as a consequence of tuberculous pericarditis was made. *Conclusion:* Tuberculosis was the most common cause of constrictive pericarditis in the developed

world before development of effective drug therapy. It remains important in developing countries. Constriction of the heart can follow an initial insult by as little as several months, it usually takes years to develop. The end result is dense fibrosis, often calcification, and adhesions of the parietal and visceral pericardium. The pathophysiological consequence of pericardial scarring is markedly restricted filling of the heart. This results in elevation and equilibration of filling pressures in all chambers and the systemic and pulmonary veins. Congestive heart failure can develop as consequence of constrictive pericarditis which is lethal.

P-FP07. Fatal Measles Related Pneumonia of 2 Toddlers: A Case Report

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Introduction: Fatality due to Measles are uncommon and rare in Malaysia ever-since the measles vaccination program was included as part of the Expanded Programme on immunisation in 1982. Beginning in 2002 MMR has been given at the age of 1 year old for all Malaysian Citizens. Since the introduction of measles vaccination in Malaysia, the occurrence of measles reduced with the increase coverage of the measles vaccination. There were 4 reported deaths due to measles in Malaysia in the period of 2008-2012. *Case:* We report cases of fatal measles pneumonia involving 2 noncitizen toddlers. Both toddlers were from two different families known to each other. Gross examination and histology of the lungs of both cases were consistent with pneumonic features. Virology studies for both cases were positive for Measles virus and bacteriology investigations were positive for Bordetella pertussis bacteria. *Conclusion:* Measles is a serious and extremely contagious disease. It is caused by a virus that spreads easily through the air, in which it can survive for several hours. Simply by sharing space with a contagious person, even for a short time, an individual can contract the infection and develop the disease. Measles remains a major cause of infant mortality worldwide despite the existence of a safe and effective vaccine. In addition to the normal symptoms of the disease, measles can lead to complications, long-term effects, and, more rarely, death. We are highlighting the diagnostic aspect in an autopsy case of fatal measles related pneumonia and the importance of immunisation programme to prevent measles.

P-FP08. Investigation outcome following forensic autopsy of disinterred body in Malaysia: a report of two cases

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Introduction: Disinterment or exhumation of body from its burial place falls under a special provision in Malaysian Criminal Procedure Code (Section 335, Chapter XXXII). Granted by relevant authority, disinterment of remains and followed by autopsy, may shed important lights towards answering relevant medicolegal issues. We reported two cases of disinterment and highlighted their outcome of investigation. *Case:* A body of 75 year-old lady was disinterred five days after burial following suspicion of missing jewellery and valuable personal belongings, and late recollection of unexplained injuries around mouth seen by relatives during bathing ritual prior to burial. At autopsy, identification of deceased was verified but no suspicious injuries concluded in view of decomposition changes. Circumstantial evidences, however, warranted homicidal death investigation. Two suspects, a live-in step-daughter and another accomplice were remanded and later charged in court. Following trial, they were found guilty and sentenced to death upon conviction. The second case involved a three year-old

child who stayed with his father following legal separation of the parent. He was often been left under the care of his father's new girlfriend. One day, he was found dead at home by his father. The body was initially released for burial by the local investigative authority. However, the act of quick, private burial arrangement by his father without informing his death to the biological mother who lived a far distance, had led the event to be investigated as homicide. Disinterment of the body was performed two hours after burial. Autopsy detected classic non-accidental injuries and death was attributed to blunt head trauma. Both father and his girlfriend were later charged for murder and currently awaiting trial. *Conclusion:* Despite its rare occurrence, disinterment of bodies with detailed forensic autopsy may able to provide crucial information towards the right direction of police investigation and proper administration of justice.

P-FP09. Livor mortis provides valuable clue in an autoerotic death

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Introduction: Death related to sexual asphyxia or autoerotism is an uncommon encounter in Malaysia. Determining the cause and manner of death in such cases can be a challenging task to forensic pathologist and the investigative authority especially if family members have altered the scene to disguise the actual circumstances of the victim's death prior to the attendance of investigators at the scene. In this case, we present an autoerotic death with prominent impressions of clothing detected on the livor mortis distribution area that provides valuable information in the death investigation. *Case:* A 34 year-old married man was found dead at home by his wife upon coming back from her workplace. The body was found inside their bedroom with no evidence to suggest suspicious or homicidal nature. At autopsy, the body was only clad in long pant and male underwear. Female sanitary pad was however found covering the genital upon removing of the underwear. The face and upper chest were deeply congested with presence of petechial hemorrhages over skin and conjunctivae of eyes. There were also restraint marks over both wrists and ankles. An impression of brassiere can be clearly seen over chest on the background of distributed livor mortis. Internal organs appeared markedly congested. On further questioning following autopsy, the wife confessed that she apparently found her husband unresponsive in a hog-tie position with hands and ankles tied with wool yarn, wearing her underclothes. A rubber ball dog toy was found inside his mouth. Due to panic and confusion, she removed her clothes and rubber ball toy and put him back on his long pant before notifying the death to the investigative authority. She denied discovery of any pornographic paraphernalia. *Conclusion:* Livor mortis not only useful in assessment of time since death and body position after death but it may also assist forensic pathologist in the investigation of the manner of death as in this autoerotic death case illustrated.

P-FP10. Forklift- assisted decapitation, a rare method of suicide

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Introduction: Within context of daily forensic autopsy practice, discovery of decapitated bodies are often associated with either homicidal or accidental deaths. Decapitation seldom related to suicide either in Malaysia or worldwide. We reported a rare case of suicide by decapitation, assisted by forklift. *Case:* A beheaded body of a 34 year-old security guard was found at an open-spaced workshop by a co-worker. The decapitated head was found nearby with a piece of metal cable in vicinity. The body appeared to be slumping to his left side behind a stationary forklift. There was another fork-lift facing

the on the foot pedal. A longer and slightly bigger diameter metal cable was tied to the back of the second forklift which was also in the direction of the body, opposite direction a few meters away. The gear of the second forklift was tied with a nylon rope in which the free end was in the direction of the deceased while a block of rubber was found. Further investigation revealed that the deceased had earlier expressed his intention to end his life over financial grudge. Three days prior to his death, he was prevented by few colleagues from jumping onto fast moving traffic at a busy highway. He was also noticed to diligently enquire from his workmates on how to operate forklift, of which was clearly not amongst job scope of a security-officer. The night before discovery of his body, he had purportedly locked his housemate from outside before left for his so-called 'night-shift' duty. *Conclusion:* A tactically planned and motivated suicide method via decapitation can still be made possible in a distressed man with previous suicidal act. Due to its rarity, this case initially caused a big mystery to the death investigating team. However, vigilant circumstances evidences consideration, scene investigation and forensic post mortem examination brought the truth to reality.

P-HE01. Comparison of collection efficiency between Cobe Spectra & Spectra Optia apheresis machine

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Introduction: Peripheral blood stem cells (PBSC) have been used for transplantation since the early 1990s and different models of apheresis machines have been used to harvest these PBSC. However, different machines will have different collection efficiencies. The objective of this study was to study the collection efficiency of a newer fully automated Spectra Optia MNC protocol as compared to the retrospective semi-automated MNC protocol for Cobe Spectra. *Materials & methods:* This study was conducted from April 2010 to May 2014. Pre and post apheresis peripheral blood $CD34^+$ cells as well as the harvested $CD34^+$ cell yield were analysed using FACSCalibur flowcytometer. The variables that were recorded for the calculation of collection efficiency were: total $CD34^+$ cells in the apheresis products, pre and post apheresis peripheral blood $CD34^+$ cells and total blood volume processed, and were compared with the retrospective data collected for Cobe Spectra between January 2007 – December 2009. *Results:* A total of 60 apheresis with Spectra Optia protocol were collected and compared with 77 apheresis performed using the COBE Spectra MNC protocol. The median pre and post apheresis PB $CD34^+$ count were 32.7/ul and 21.9/ul versus 33.6/ul and 28.5/ul, the median total stem cell yields were 201 x 10^6 cells versus 137.7 x 10^6 cells and the median product volume were 240ml versus 156ml respectively. The overall collection efficiency was superior for Spectra Optia protocol (80.2%) compared to Cobe Spectra MNC protocol (63.7%). *Discussion:* Spectra Optia protocol had shown better collection efficiency as compared to Cobe spectra MNC protocol. These could be explained by the improved technology of this fully automated system which had a better control of the stem cell collection. However, the larger volume of product collected and the longer duration of collection procedure were the two main drawbacks of this Spectra Optia.

P-HE02. Plasma-derived microparticles in patients with polycythaemia vera

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Introduction: Microparticles (MPs) are small membrane-bound vesicles, measuring less than 1.0 μm, which are released during cellular activation or during apoptosis. One of the important features of microparticle is the exposure of the anionic aminophospholipid phosphatidylserine on its outer surface. These microparticles are present in low concentration in normal plasma. Many studies have reported the role of microparticles in haemostasis and thrombosis as well as their importance in cancer cell survival, invasiveness and metastasis. Increased levels of circulating microparticles have been observed in myocardial infarction, atherosclerosis and essential thrombocythaemia. The objective of this study was the evaluate the levels of MPs in patients with polycythaemia vera. *Materials and methods:* Plasma samples from 15 patients with polycythaemia vera (PV) and 15 healthy controls were obtained and the levels of platelet as well as endothelial derived microparticles were analyzed using flowcytometry and compared. *Results:* Patients with PV were found to have a significantly higher percentage of platelet derived microparticles compared to healthy controls ($P<0.05$). However, we observed a higher level of endothelial derived microparticles in the control group compared to our patients' group, but the differences were not statistically significant ($P>0.05$). We also observed a higher median haemoglobin and haematocrit level, white cell count, red cell count and platelet counts in our patients' group ($P<0.05$). No correlation was observed between platelet derived microparticles and the platelet counts in both the patients and the control group. *Conclusion:* The median levels of positive events for platelet derived microparticles were found to be higher in patients with PV compared to normal controls. In view of the small sample size, further study needs to be done to evaluate the significance of MPs in this patient group.

P-HE03. Alloantibody and autoantibody immunization in repeatedly transfused thalassaemia patients: Hospital Ampang experience

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Introduction: Regular blood transfusion is the major treatment for thalassaemia patients. Repeated exposure to donor red cells put them at risk of sensitization. This will induce the development of alloantibodies and autoantibodies towards red cell antigens, which complicates management of transfusion therapy especially in transfusion-dependent thalassaemia patients. The pattern of red blood cell alloantibody and autoantibody and its associated factors were studied amongst these patients. *Materials & methods:* Clinical and serological data of 637 patients who were registered at the Thalassaemia Clinic Ampang Hospital from 2006 – 2013 were collected and analysed. Transfusion dependent was defined as patients receiving a blood transfusion at least every 4 weeks or earlier. *Results:* Ninety four patients (14.8%) were transfusion-dependent; 49 were thalassaemia major, 40 were thalassaemia intermedia and 5 were Hb H disease. A total of 22.3% were positive for antibodies towards red cell antigens, whereby 16.0% of the patients had alloantibodies, 1.1% had autoantibodies while 5.3% had both allo- and autoantibodies. The average number of packed cells transfused before the development of antibody was 5.58 bags. We found that 38.1% of these patients had already developed antibodies upon referral to Hospital Ampang. Anti-E had the highest frequency (12.8%) followed by non-specific auto-IgG (6.4%), anti- S (5.3%) and anti-Jkb (4.3%). *Discussion & Conclusion:* The

antibodies of the highest frequency detected are all clinically significant antibodies that may induce a haemolytic transfusion reaction. Anti-E presents as the most detectable antibody, which corresponds with the fact that the CDe haplotype is highly prevalent amongst the Malaysian population. Incomplete patient records were the biggest limitations in this study. A more detailed and comprehensive database is also needed to provide better care and management of the blood transfusion requirement of these patients.

P-HE04. Evaluation of an automated coagulation analyser: the ACL TOP 500 CTS

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Introduction ACL TOP 500 is a fully automated coagulation analyzer using optical clot detection, chromogenic and immunologic method. ACL Top 500 CTS is used for in vitro hemostasis and fibrinolysis diagnostic testing in the assessment of thrombosis and hemostasis. The major aim of this evaluation is to establish and construct the validity of the performance characteristics of the ACL TOP 500 CTS. *Materials & methods* The evaluation was performed in the Haematology Laboratory, Department of Pathology Hospital Tuanku Jaa'far Seremban. Control materials from the manufacturer and 3.2% sodium citrated plasma were used to perform the coagulation testing throughout the study. The evaluation includes precision, carryover study, linearity, establishment of reference intervals and INR validation. The correlation for PT, APTT and Fibrinogen with Stago-R Evolution were also evaluated. *Results* Imprecision study of PT, APTT and fibrinogen in terms of %CV were found to be within the manufacturer's imprecision performance specifications, ranging from 0.6 to 3.2%. Fibrinogen assay exhibited a great linearity range on ACL TOP 500 CTS with R value of 0.999. Sample carryover was negligible. Good correlation was observed in PT, APTT and fibrinogen between ACL TOP 500 CTS and Stago-R Evolution. The reference intervals established for PT and APTT were shorter as compared to the intervals established for Stago R Evolution. This could be due to different type of thromboplastin and activator used in the testing. *Conclusion* The findings confirmed that the technical performances of ACL TOP 500 CTS met with the standards claimed by the manufacturer. Despite its extensive features, the analyzer requires minimal maintenance thus easy to operate.

P-HE05. Utility of ISTH-BAT questionnaire and ELISA-VASP-P assay for screening of mild platelet-based bleeding disorder

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Introduction: Mild platelet-based bleeding disorder is relatively rare in the population. It is very challenging to distinguish clinically significant mild bleeding symptoms from normal bleeding symptoms. This study aims to explore the utility of ISTH-BAT questionnaire and ELISA-VASP-P assay in identifying mild bleeding disorder. *Materials and methods*: This study was conducted at Universiti Putra Malaysia (UPM) and Pusat Darah Negara (PDN) from June – October 2013. Informed consented respondents were blood donors (n=25) and patients (n=14) from PDN. Patients suggestive of platelet ADP receptor defect were selected based on previous screening of platelet aggregation test. Respondents were interviewed according to the ISTH-BAT questionnaire to assess their bleeding symptoms and were given scores. Blood was drawn into 3.2% sodium citrate tube for ELISA-VASP-P assay. The

assay assessed platelet P_2Y_{12} receptor activity and reported as platelet reactivity index (PRI). Statistical analysis with $p<0.05$ was considered significant. *Results*: Median of total bleeding score among healthy donors was 0 (IQR=0). Among the patients, median score was 0.5 (IQR=4). Total bleeding score was significantly higher in the patients (p=0.002), with a mean rank of 25.7 as compared to healthy donors' mean rank of 16.8. This illustrated that the bleeding symptoms were significantly present in the patients. Symptoms that were significantly present in the patients were cutaneous bleeding, menorrhagia and muscle hematoma. The PRI analysis revealed no significant difference between patients and healthy donors (p=0.069). Median PRI of healthy donors was 93 %(IQR=11.4%) and median PRI of patients was 87.6 %(IQR=24%). *Discussion & conclusion*: These results demonstrated that the patients did not suffer from platelet P_2Y_{12} receptor defect despite significant presence of the bleeding symptoms. Thus, ISTH-BAT questionnaire is useful to evaluate mild bleeding symptoms and ELISA-VASP-P assay has an advantage of confirming platelet ADP receptor defect (specifically P_2Y_{12} receptor) with minimal blood volume requirement.

P-HE06. A case report: Non-deletional Hb H – Hb Quong Sze disease

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Introduction: Haemoglobin (Hb) Quong Sze is a low incidence alpha (α) thalassaemia mutation seen in less than 1% of the Malaysian Chinese population, with the missense mutation at codon 125 of the α2 globin gene (CTG → CCG or Leu → Pro). Interaction of Hb Quong Sze with Southeast Asian double α-globin gene deletion (--SEA) results in a non-deletional Hb H disease. In this case report, we describe a child with HbH-Hb Quong Sze who presented to us with features of thalassaemia intermedia. *Case report*: An 8-year-old Chinese boy was referred to UKM Medical Centre six years ago for pallor and jaundice since birth and failure to thrive. Both parents were of Chinese descent. On examination, he had jaundice and hepatosplenomegaly as well as frontal bossing. Full blood picture showed hypochromic microcytic red cells with anisopoikilocytosis and Hb level of 8.5g/dL. Multiplex gap polymerase chain reaction (PCR) amplification done at that time only detected the common (--SEA). He was treated conservatively and never had a blood transfusion. His Hb was maintained between 8.3g/dL and 9.6g/dL. However, his weight and height was constantly below the third centile. Six years later, after the introduction of multiplex-PCR amplification, DNA analysis was repeated and he was found to have Hb Quong Sze (αα125) as well. *Discussion and conclusion*: Non-deletional Hb H- Hb Quong Sze is present at a low incidence in Malaysia. The incidence may be higher than reported due to lack of awareness and facilities in the laboratory to detect this mutation. The mutation results in a highly unstable Hb and is undetectable by routine Hb electrophoresis. Interaction of this Hb with (--SEA) will cause severe haemolytic anaemia. Accurate detection can only be made by molecular analysis. Establishment of the diagnosis is necessary as these mutations show marked phenotypic variability.

P-HE07. DNA methylation of tumor suppressor genes in acute myeloid leukemia with normal and abnormal karyotype

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Introduction: Acute myeloid leukemia (AML) is a heterogeneous disease in terms of clinical features, treatment outcomes, cytogenetic abnormalities and gene mutations. Chromosome abnormalities are powerful prognostic indicators in AML. In leukemia, the balance of methylation status in normal cells is lost. The promoters of tumor suppressor genes (TSGs) which are unmethylated, become methylated and gene expression is lost. The objective of this study is to identify the TSGs that are methylated in AML with normal and abnormal karyotype. *Materials and methods:* Cytogenetic studies were performed according to standard techniques on the bone marrow aspirate of AML patients at diagnosis. Based on cytogenetic findings, 30 AML patients with normal karyotype (intermediate risk group) and 15 AML patients with chromosome translocations in the favorable risk group [t (8; 21) or t (15; 17)] were selected for DNA methylation studies using quantitative real time PCR array. Blood with normal cell counts were used as normal controls. *Results:* The three frequently methylated TSGs were SLC5A8, DRD2, and HOXA7. The frequencies of the methylated genes in normal karyotype vs abnormal karyotype were as follows: SLC5A8: 50% vs 86.7%, DRD2: 40% vs 80%, and HOXA7: 10% vs 40%.*Discussion:* The frequency of methylated TSGs was higher in patients with chromosome translocations compared to normal karyotype. SLC5A8 was the most frequently methylated TSG. SLC5A8 is involved in the transport of butyrate that has antiproliferative activities through histone acetylation and gene expression. The role of TSG methylation in AML requires further investigation.

P-HE08. Copy number abnormalities characterization by multiplex ligation-dependent probe amplification (MLPA) in *BCR-ABL1* positive adult acute lymphoblastic leukemia patients

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Introduction: Philadelphia chromosome; t(9;22)(q34;q11) results in the translocation of the *BCR* gene at chromosome region 22q11 to *ABL1* gene at region 9q34. This translocation occurs in 2% - 3% of children and 25% of adults with B-cell precursor ALL (BCP-ALL). *BCR-ABL1* positive acute lymphoblastic leukemia (ALL) is associated with a shorter duration of remission and with an inferior long time survival of less than 20% in adults. *Materials & methods:* Genomic DNA and RNA were extracted from bone marrow or peripheral blood samples at initial diagnosis or relapsed ALL cases, using standard methods in Ampang Hospital. Karyotyping, RT-PCR for t(9;22) and multiplex ligation-dependent probe amplification (MLPA) were used to detect genetic alterations. *Results:* In total, 38 patients were included in this study. Frequency age of Ph+ ALL diagnosed was between 14 to 20 years old (6/38, 15.8%), followed by 60.5% (23/38) at age of 21 to 50 and 23.7% (9/38) at age 51 to 70 years and no information of age was in 2.6% (1/38) of cases. Approximately 55.3% (21/38) of these patients were identified having e1a2 transcripts, 44.7% (17/38) were identified with major transcripts (28.9% (11/38) b3a2 and 15.8% (6/38) b3a2 fusion transcripts). Cytogenetic abnormalities in addition to t(9;22) were detected in most of the samples by karyotype analysis. Incidence of copy number abnormalities (CNA) tested by MLPA for targeted gene aberrations showed 26.3% (10/38) of cases at least had one abnormality with aberrant IKZF1, 50% (19/38) had more than two and 23.7% (9/38) had no CNA.

Discussion: This study had shown more than 45% of adult Ph+ ALL patients harbored the p210 *BCR-ABL1* fusion transcripts. *IKZF1* deletions were highly involved in adults with Ph+ ALL.

P-HE09. Red cell characterisation in rheumatoid arthritis patients presenting with anaemia

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Introduction: Anaemia is a prevalent finding in patients with rheumatoid arthritis. Although 60% of anaemia is due to anaemia of chronic disease (ACD), many cases are related to iron deficiency (IDA). *Materials & methods*: 111 patients with rheumatoid arthritis and anaemia were recruited from the Rheumatology Clinic, Selayang Hospital and segregated into microcytic hypochromic (Mi), normocytic (No) and macrocytic (Ma) groups. *Results*: There were 39 (35%) patients in Mi group, 70 (63%) patients in No group and only 2 (1.8%) patient in Ma group. Moderate anaemia was found in 6 (8.6%) patients in No group, as compared to 5 (12.8%) in Mi group. There were only 2 patients with severe anaemia and both were in Mi group. Age did not show any association with the severity of anaemia. *Discussion*: In this cohort of patients, more than 60% are likely to have anaemia of chronic disease (ACD) based on the red cell indices. However, both ACD and IDA can co-exist and can be difficult to identify. In conclusion, based on these results, patients with microcytic anaemia have more severe anaemia compared to normocytic anaemia, irrespective of age.

P-HE10. The effect of polymorphisms of γ-glutamyl hydrolase gene on methotrexate serum levels and related toxicity in children with acute lymphoid leukemia

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Introduction: Methotrexate (MTX) is an important drug for the treatment of childhood acute lymphoblastic leukemia (ALL). However, related toxicity occurs in many organs which may cause interruption of treatment, morbidity, and mortality. Single nucleotide polymorphisms (SNPs) of gammaglutamyl hydrolase (GGH) are known to alter their enzymatic activity and thus affect the metabolism of MTX and influence the effectiveness. Therefore, this study evaluated the association of −401C/T and +452C/T polymorphisms of γ-glutamyl hydrolase and the risk of cytotoxicity of MTX to acute lymphoblastic leukemia in children. *Materials and methods:* The subjects were children diagnosed with ALL at Department of Pediatrics, Faculty of Medicine Aliasghar Hospital, Tehran, Iran. Genotyping was performed using RFLP-PCR and association of genotypes with toxicity of methotrexate was evaluated by HPLC method and common terminology criteria for adverse events respectively. *Results:* We studied 83 children with ALL with a median age of 6 years. The predominant gender was male with 57.8% while female were 42.2%. The most frequent genotype of the −401C/T and +452C/T polymorphisms was the −401C/C and +452C/C which were found in 54.2% and 74.6% respectively. An association between the −401C/T polymorphism and the risk of toxicity of MTX was found [(thrombocytopenia ($p = 0.010$) and leukopenia $p=0.040$)], and patients with the −401C/C genotype had 33.73% (95% CI 0.009-0.019) higher chance of toxicity. No association was found between the +452C/T polymorphism and the risk of thrombocytopenia and leukopenia. *Discussion:* Our investigation suggests that the −401C/T polymorphism in the γ-glutamyl hydrolase may be a factor involved in the generation of MTX toxicity and has to be determined in patients with ALL.

P-HE11. Hb Barts hydrops foetalis: an unusual scenario

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Introduction. Hb Barts hydrops foetalis is the most severe form of a-thalassaemia, a fatal condition incompatible with life in absence of early diagnosis and intrauterine transfusions. It is seen more commonly in Chinese-Malaysians than in Malays. A 29-year-old Malay female presented at 30 weeks gestation with preeclampsia and a hydropic foetus. *Materials and Methods.* Blood collected in EDTA was available from the parents and intracardiac blood from the hydropic foetus for full blood picture, Hb analysis and DNA molecular studies. *Results.* The findings in the couple were compatible with HbE-α-thalassaemia. Both had inherited HbE and α^0-thalassaemia. The HbE levels were <25 % and the α^0 was the –SEA deletion. The intracardiac blood was in keeping with Hb Barts hydrops foetalis (homozygous α^0 / α^0). The C-grams generated on the BioRad Variant II had a characteristic pattern. In homozygous α^0 /α^0 the predominant Hb is Hb Barts with absence of HbA and HbF. *Discussion and Conclusion* Homozygosity of α^0-thalassaemia is an uncommon cause of Hb Barts hydrops foetalis in the Malays. HbE is a common Hb variant in the Malays. In screening for thalassaemia, when HbE levels are less than 25% the possibility of concurrent alpha thalassaemia should be considered. Carriers who are presumptively identified as HbE α-thalassaemia require comprehensive DNA molecular studies and counselling. Ideally, this should be done prior to any conception.

P-HE12. Inhibition of U266 cell growth by human mesenchymal stromal cell-mediated siRNA silencing of interleukin-6

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Introduction: Studies demonstrated that mesenchymal stromal cells (MSC) from bone marrow stroma produced high concentration of interleukin-6 (IL-6) that promoted multiple myeloma growth in a paracrine manner. In view of the failure of IL-6 blocking antibodies to demonstrate substantial clinical responses in early clinical trials, more effective methods are needed to disrupt the favourable microenvironment provided by the bone marrow. In this study, we evaluated the silencing of IL-6 in MSC post siRNA transfection and the efficacy of these MSC on multiple myeloma cell growth and IL-6 production inhibition. *Materials & Methods:* MSC ($1x10^5$) were transfected with 100 pmol IL-6 siRNA (SASI_Hs01_00155911) using 5µL Lipofectamine 2000 with untransfected MSC as control. IL-6 transcript and protein levels were determined using Real Time qPCR and ELISA assay respectively. Transfected MSC were subjected to viability, immunophenotyping and trilineage differentiation studies. Multiple myeloma cells, U266 ($3x10^2$), were co-cultured with supernatant collected from MSC expressing IL-6 siRNA. The level of IL-6 production and growth of U266 cells post co-culture were determined on days 3 and 5 respectively using ELISA and MTS assays. *Results:* IL-6 mRNA and protein were significantly suppressed by 5.9-fold and 63% relative to untransfected MSC by 72 hours. The suppression was sustained up to 120 hours with 2.1-fold mRNA reduction and 71.8% protein reduction. IL-6 siRNA transfection did not affect the viability, surface markers and trilineage differentiation capacity of MSC. U266 co-culture results showed cell growth and IL-6 production when

co-cultured with MSC expressing IL-6 siRNA were inhibited by 2-fold compared to untransfected co-culture wells up to 5 days. *Discussion:* When co-cultured together, MSC expressing IL-6 siRNA inhibited U266 cell growth and IL-6 production significantly supporting the potential usage of MSC for siRNA-mediated IL-6 gene silencing to treat multiple myeloma.

P-HE13. Effective silencing of NAMPT and LYST with siRNA in human myeloma RPMI8226 cells

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Introduction: Multiple myeloma (MM) is the malignancy of differentiated B lymphocytes. It accounts for 10% of all hematological cancers and approximately 2% of all cancer deaths. Previous array comparative genomic hybridization analysis on 63 multiple myeloma patients revealed copy number gains at chromosomal regions 7q22.3 and 1q42.3 in approximately 90% and 60% of myeloma cases studied, respectively. NAMPT and LYST were localized at chromosome 7q22.3 and 1q42.3, respectively. This study aims to investigate the effects of small interfering RNA (siRNA) of NAMPT and LYST on the proliferation and apoptosis of human myeloma RPMI8226 cells. *Material & methods:* Human myeloma RPMI8226 cells were transfected with three different siRNAs targeting NAMPT and LYST with the Amaxa nucleofection technique. Transcription and expression of NAMPT and LYST were checked by real-time RT-PCR analysis. *Results:* Our results showed that the NAMPT and LYST siRNA fragments could be successfully transfected into RPMI8226 cells, which resulted in the significant inhibition of transcription and expression of these genes in the myeloma cells. Expression of NAMPT and LYST were knockdown up to approximately 72% and 88% respectively in myeloma cells when compared to the negative control siRNA. *Conclusion:* Future work is aim to evaluate the proliferation and apoptosis of the transfected cells and the protein expression of NAMPT and LYST after transfection.

P-HE14. High-resolution analysis of chromosomal alterations in adult acute lymphoblastic leukemia

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Introduction: Chromosomal alterations occur frequently in acute lymphoblastic leukemia (ALL), affecting either the chromosome number or structural changes. These alterations can lead to inactivation of tumour suppressor genes and/ or activation of oncogenes. The objective of this study was to identify recurrent and/ or novel chromosomal alterations in adult ALL. *Materials & methods:* We studied 41 cases of adult ALL compared with healthy normal controls using Single Nucleotide Polymorphism (SNP) array. *Results:* Our analysis revealed 43 copy number variant regions, of which 44% were gains and 56% were losses. The most frequent copy number gains were on chromosome regions 8p23.1 (71%), 1q44 (66%), 1q23.3 (54%), 11q23.3 (54%), 12p13.33 (54%) and 8q24.21 (51%). On the other hand, copy number losses were most frequently seen on chromosomes 1p31.1(76%), 3q26.1(68%), 11p11.12 (63%), 4q12 (59%), 19p13.2 (59%), 7q11.21 (56%), Xp22.33 (54%), 7q11.21 (51%) and, 19p13.11 (51%). *Discussion:* Gain of 8p23.1 and loss of 1p31.1 were the most frequently found alterations in

this study. These chromosomal regions contain genes such as *SPAG11B*, *DEFB104A*, *DEFB105A*, *DEFB107A*, *DEFB106A* and *SPAG11A*. These potential genes may contribute to leukemogenesis in adult ALL. The cytogenetic and molecular mechanisms underlying these chromosome changes deserve further investigations.

P-HE15. Factors influencing temperature of blood and blood products during transportation between blood donation centre and a medical centre in the Klang Valley

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Introduction: Malaysia is a hot and humid country which its temperature ranges around 32-39°C. This temperature range is unsuitable for blood and blood products storage but somehow its transportation between National Blood Centre in Jalan Tun Razak Kuala Lumpur and Clinical Training Centre in UiTM Sg Buloh must be made enable to maintain the quality of the blood and blood products. Concern about the cold chain integrity of blood and blood products drove us to document and analyse detailed temperature records in the entire trip to and fro the two locations. *Materials & Methods:* We analysed 219 temperature readings between January and May 2014 using a calibrated Microlite data logger (*Fourtec USA*). The blood and blood products were packed in an expanded polypropylene insulated transport box (*Delta T, Germany)* with a capacity of 20L. The system comes with an either a 4°C or -30°C temperature shell depending on the type of blood product that will be carried. The transport will depend on the availability of a sedan, van or ambulance. The time of transportation is between 9 am to 1 pm. *Results:* The mean temperature (T_m) for packed red cell transport is 5.69°C (0.5-9.2°C). The standard deviation (SD) is 1.37 and coefficient of variation (CV) is 24.0. The T_m for platelet is 21.7°C (19.2-28.1°C). The SD is 1.85 and CV is 8.5. The T_m for FFP and cryoprecipitate is -18.7°C (-30.62-(-11.9)°C). The SD is 3.37 and CV is -18.0. The mean time travelled was 2.5 hours (1.5-5 hours). *Discussion:* The packed cell temperature variation was the most difficult to maintain whereas platelet was the easiest to maintain within range. The contributing factors to temperature variation are duration of travel, ice-pack preparation, many stopovers, transport type, quantity of blood products carried in a container and ambient temperature of the traffic!

P-HE16. Immunophenotype and PML gene breakpoints in classical and variant acute promyelocytic leukaemia

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Introduction: Acute promyelocytic leukaemia (APL) is a distinct subtype of acute myeloid leukaemia, characterized by a high response rate to all-trans retinoic acid (ATRA). Rapid diagnosis and prompt treatment with ATRA is critical in the management of APL. Cytomorphological diagnosis is fast and easy but misclassification of the APL subtypes, usually with the microgranular variants (M3v) type of APL can occur. CD34 and CD2 positivity has been reported to be associated with M3v and bcr3 isoform. We aimed to study the immunophenotypic features and PML gene breakpoints in classical APL and M3v cases in UKM Medical Centre and Hospital Kuala Lumpur. *Materials & Methods:* Haematological profile was performed on peripheral blood samples of APL. Morphological diagnoses of bone marrow aspirate smears (MGG stained) were determined by two haematologists. Flowcytometry was performed using a panel of 32 monoclonal antibodies and a polyclonal anti-Tdt antibody on these samples. PML

gene breakpoints analysis was confirmed by RT-PCR. *Results:* A total of 48 APL cases were studied. Leucocytosis was significantly associated with M3v (p=0.023). Morphogically, 34 cases (70.8%) were diagnosed as classical APL and 14 (29.2%) as M3v. The 'typical' APL immunophenotypic features (CD34 and HLA-DR negative; MPO, CD117, CD13, CD33 positivity) were seen in 33 cases (68.8%): 8 M3v and 25 classical APL. CD 33 positivity occurs in 100% of APL cases. The immunophenotypic profile; concerning the presence or absence of these antigens, including absence of HLA-DR and CD34 was indistinguishable in the two APL subtypes. Molecular analysis revealed bcr1 isoform was more common than the bcr2 or bcr3. However, there was no significant difference between bcr isoforms and APL subtypes. *Conclusion:* Immunophenotypic features of classical APL and M3v were very similar. Thus the 'typical' APL immunophenotypic profile is diagnostic for M3v and contributes to prompt management of APL cases.

P-HE17. STR analysis for chimerism status of allogeneic peripheral blood stem cell transplantation

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Introduction: Analysis of donor chimerism is important to document engraftment after allogeneic peripheral blood stem cell transplantation (PBSCT). The PCR-based method for the amplification of highly polymorphic short tandem repeat (STR) sequences has emerged as the reference tool for chimerism monitoring of PBSCT. We report our experience in chimerism monitoring with STR analysis for the PBSCT cases in UKM Medical Centre. *Materials & Methods:* DNA was extracted from 3-5 ml peripheral blood (EDTA) of patients and donors before and after PBSCT. PCR amplification of STR loci for receipients and donors were performed using AmpFISTR Identifiler Plus Kit (Applied Biosystem) according to manufacturers' protocol. The locus was used to identify and quantify informative markers between donor and receipient. Capillary electrophoresis of PCR product was performed using Genetic Analyzer ABI 3130. Informative STR loci for donor-receipient were determined. *Results:* A total of 71 cases (patient-donor pairs) were studied since 2005; 42 were Malays, 20 were Chinese and 9 from other races. Analysis (day-100) showed a complete chimerism in 53 cases (74.65%), mixed chimerism in 15 cases (21.13%) and no chimerism in 3 cases (4.22%). In Malays, both D21S11 and FGA loci showed the highest informativity (71.43%) and TPOX (44.0%) was the lowest. In Chinese, both TH01 and D8S1179 loci (>80.0%) showed highest informativity and the lowest was D19S433 (28.56%). A few loci showed consistent informativity (>50%) in all groups: D3S1358, vWA, FGA, TH01, D21S11, D13S317, D7S820, DS16S539 and D2S1338. TPOX showed a low informativity in all ethnic groups. *Discussion:* The STR analysis of the PBSCT cases has provided a panel of useful and informative STR markers. Thus, selecting only a few suitable, useful and high informative candidate loci from this STR-panel, would be adequate and cost effective for chimerism analysis to determine engraftment status of the PBSCT cases in our Malaysian population.

P-HE18. Hb H disease (alpha-thalassaemia intermedia): genotype and phenotype correlation in Hospital Kuala Lumpur

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Introduction: Haemoglobin H (Hb H) disease is the commonest cause of thalassaemia intermedia. In the majority of cases, Hb H disease results by compound heterozygosity for alpha(0)-thalassemia due to deletions that remove both linked alpha-globin genes on chromosome 16, and deletional alpha(+)-thalassemia from single alpha-globin gene deletions. These are known as deletional Hb H disease (--/-α). In a smaller proportion of patients, Hb H disease may occur from interactions between alpha(0)-thalassemia with non-deletional mutations (alpha(T)alpha or alpha alpha(T)) or with abnormal hemoglobins such as Hb Constant Spring, Hb Quong Sze or Hb Adana. These are known as the non deletional Hb H disease (--/$\alpha^T\alpha$) or (--/$\alpha\alpha^T$). Mostly patients with Hb H disease will have compensated haemolytic anemia with average haemoglobin levels between 7-10 g/dL. The manifestations include thalassaemia facies, jaundice and hepatosplenomegaly. Patients with non deletional Hb H disease usually are more symptomatic i.e anaemic with hepatosplenomegaly, and more likely require transfusions especially during acute infection, fever or stress. *Materials & Methods:* All samples presumptively identified as alpha-thalassaemia intermedia from screening tests were recruited. Retrospectively all their clinical history with haematological data from full blood count and haemoglobin analysis results were analyzed. Results from multiplex PCR-based technology performed in Molecular Haematology Laboratory Hospital Kuala Lumpur which incorporates both GAP-PCR for deletion detection and ARMS-PCR for non deletional detection were characterized. Results of molecular characterization (genotypes) of Hb H disease, haematological parameters and its clinical phenotypes were then tabulated. *Results & Conclusion:* From these findings, patients with Hb H non deletional type showed more severe clinical phenotype than patients with Hb H deletional type. However, patients with identical genotype may not necessary show the same severity.

P-HE19. Molecular analysis of the intermedia alpha thalassemia of heterozygote compound alpha thalassemia

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Introduction: Alpha Thalassemia is caused by a lacking or no production of alpha chain synthesis resulting from alpha globin gene mutation such as deletion or non deletion. Deoxyribonucleic Acid Analysis is important to find out the type of mutation to predict prognosis and therapy, also for genetic counseling. Purpose of this case report is to show that Thalassemia Intermedia could be caused by double heterozygote non deletion mutation. This is the first case reported in Medan, North Sumatera, Indonesia. *Case report:* A nine month baby girl brought by her parents to Clinical Pathology Department of Haji Adam Malik Hospital, Medan, examined full blood count as a procedure for regular transfusion. Hematology reports: Hb 9.6 g/dL, MCV 72.1 fL, MCH 24.1 pg, peripheral blood smear showed: hypochromic microcytic red cells. Haemoglobin Analysis: HbA_2 1.7%, HbF 4.6%. Hematology features (mother): Hb 13.2 g/dL, MCV 76.1 fL, MCH 25.3 pg, HbA_2 2.7%, HbF: not found, suspected: Thalassemia alpha trait carrier. Hematology features (father): normal range, HbA_2 2.6%, HbF: not found, HbCS: 0.3%. Molecular analysis examined at Molecular Biology Eijkman Research Institute, Jakarta, DNA analysis (baby): double heterozygote mutation at codon at 59 gene globin- $alpha_2$ ($GGC^{glycin} \rightarrow GAC^{Aspartat}$), called Hb Adana and heterozygote mutation at Cd142 gene globin-$alpha_2$

($TAA^{Stop} \rightarrow CAA^{Glutamin+30aa}$) called Hb Constant Spring. Molecular Analysis of the mother: heterozygote mutation at codon 59 gene globin-alpha$_2$ ($GGC^{glycin} \rightarrow GAC^{Aspartat}$) and father's mutation is heterozygote at codon 142 gene globin- alpha$_2$ ($TAA^{Stop} \rightarrow CAA^{Glutamin+30aa}$). *Discussion and Conclusions:* The baby is Thalassemia Alpha/HbH disease based on double heterozygote mutation or Hb Adana and Hb constant spring from both parents. Her mother has a heterozygote mutation, called Hb Adana and her father has a heterozygote mutation, called Hb Constant Spring. Clinical manifestation showed thalassemia intermedia, probably caused by non genetic factors or oxidant compound exposures.

P-HE20. Quality assessment of irradiated red blood cell products: experience in Universiti Kebangsaan Malaysia Medical Centre

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Introduction: Quality assessment of red cell products after donation to transfusion is very important to ensure the products are of good quality. This is also applicable to the blood unit that has undergone irradiation process. The national standard requirement for quality assessment is: haemolysis should be <0.8% of the red-cell mass, total haemoglobin (Hb)/unit should be >45 g/unit. Haematocrit (HCT) for red cell in additive-solution should be 0.5-0.7 with volume 340ml±38. At least 95% of blood units' sampled should meet the assessment specification. The aim of this study was to evaluate the quality of the irradiated blood products as compared to prior irradiation. *Materials and Methods:* This experimental study was conducted on 39 red-cell in additive-solution units at the Blood-Bank unit, UKMMC in 2013. Pre and post-irradiated samples were collected. Full blood count was done to estimate the Hb level, HCT level and total Hb/unit was calculated. Plasma-Hb was measured and percent haemolysis was calculated. Total volume of the blood bag was measured. All the parameters were documented and analysed. *Results:* For the 39 units of red cells in additive-solution, the average volume was 338ml. The pre and post irradiated results were as follow: HCT was 60.44% and 58.74%, Hb/unit was 64g/U and 62 g/U and %haemolysis was 0.30 and 0.34 respectively in the pre and post-irradiated samples. *Discussion:* This study showed that although there is some reduction of HCT and Hb/unit in the blood units following irradiation process, the quality of the products remained within the standard requirements of national standards. In conclusion from this study, although irradiation process has some effects on the red cell products, the effects are not significant and the products remained in acceptable quality to be used following irradiation process for indicated patients.

P-HE22. Evaluation of glucose-6-phosphate dehydrogenase stability in stored EDTA blood samples

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Introduction: Glucose-6-Phosphate Dehydrogenase (G6PD) deficiency among newborn is the commonest cause of neonatal jaundice in Malaysia. OSMMR2000-D G6PD Assay Kit has been used to quantitate the level of G6PD activity in newborns delivered in UKMMC and some of government hospitals. The duration of sample storage prior to analysis is one of the matters concerned. This study is conducted to identify the maximum days of blood sample that can be stored in EDTA tube and give a constant reading. *Materials & methods:* A total of 77 cord blood sample of normal term neonates

delivered at UKM Medical Centre were recruited for this study. These cord bloods were collected in EDTA tubes and the samples were measured for G6PD enzymes levels daily for 7 days using the OSMMR2000-D G6PD Assay Kit. The samples were stored in the same EDTA tube and refrigerated at 2-8°C in between the assays. Mean of G6PD activity was compared between days of analysis using paired t-test. *Results:* In this study, 67 out of 77 cord blood samples showed normal enzyme levels while another 10 had severe enzyme deficiency. Daily mean of G6PD activity of all normal samples showed a sudden drop in the enzyme level after day 4 of analysis while severe deficient samples showed a sudden drop after day 2. There was statistically significant difference in mean of G6PD activity between Day 1 and Day 5; and Day 4 and Day 5 ($p < 0.05$). The statistically differences in the mean of G6PD activity between Day 1 and Day 4; and Day 3 and Day 4 were not significant ($p = 0.476$ and 0.056 respectively). *Discussion and conclusion:* The maximum time that can give a constant reading for G6PD enzyme levels is within 4 days and blood samples must be refrigerated at 2-8°C in EDTA tube.

P-HE23. Diagnostic performance of reticulocyte haemoglobin equivalent (Ret-He) in detecting iron deficiency anaemia in haemodialysis patient

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Introduction: Iron deficiency anaemia is common in chronic kidney disease (CKD) but is difficult to diagnose due to underlying systemic inflammation. Reticulocyte Haemoglobin (Ret-He) reflects the amount of iron available for haemoglobin production in the bone marrow and it is less affected by inflammation compared to other biochemical markers. The aim of this study is to evaluate the clinical usefulness of Ret-He in diagnosing iron deficiency in CKD patients and to compare the diagnostic performance between Ret-He and traditional biochemical parameters. *Materials and methods:* Peripheral blood samples from 110 haemodialysis patients were collected in EDTA anti-coagulant and Ret He was analysed using Sysmex XE-2100 haematology analysers. Their serum iron, TIBC, ferritin and transferrin saturation were also measured and diagnostic performance of Ret-He was evaluated towards these biochemical parameters. *Results:* Based on Receiver Operating Characteristics (ROC) curve analysis, Ret-He has a significant ($p < 0.05$) and greater area under the curve (AUC) towards transferrin saturation (82.9%) as compared to other biochemical parameters. Optimal cut-off value for Ret-He based on transferrin saturation was 31.6 pg and; sensitivity and specificity of 84.1% and 78.8%, respectively. This study also showed moderate significant correlation (based on Pearson correlation) between transferrin saturation and Ret-He, with correlation coefficient, $r = 0.413$. ROC curve analysis revealed that by using a Ret-He cut-off level of 31.6 pg iron deficiency could be diagnosed with a sensitivity and specificity of 75% and 63.4% respectively. The area under the curve was 73.3%. *Discussion and conclusion:* Ret-He is a reliable marker of cellular hemoglobin content and can be used to identify the presence of iron-deficient states and the cut-off value 31.6 pg is appropriate for the assessment and monitoring of iron deficiency.

P-HE24. RBC G6PD activity measurement – a comparison between whole blood EDTA samples and dried blood spot

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Introduction: Glucose-6-Phosphate Dehydrogenase (G6PD) deficiency is an important cause of neonatal jaundice. Routine screening for G6PD deficiency employs the use of semi quantitative methods on dried blood spots on filter paper. Our laboratory has established the quantitative enzyme assay, a more sensitive method to detect partially-deficient individuals. The stability of G6PD in dried blood spots has been reported to be affected by humidity and temperature, especially when low-value samples are evaluated. This study aimed to determine whether dried cord blood spotted on filter paper samples give comparable enzyme activity reading to that of EDTA whole blood samples. *Materials & Method:* Blood samples from 30 normal term neonates delivered at UKM Medical Centre were studied. We compared red blood cell G6PD activity in EDTA blood samples and blood spotted on filter paper dried at room temperature and 10 spotted blood samples dried using hair dryer. The blood samples in EDTA tube were refrigerated at 2-8°C while blood samples on filter papers were kept in the box at room temperature between the assays. The G6PD enzyme levels were measured in the different types of samples daily from day 1 until day 7 using the OSMMR2000-D G6PD Assay Kit. The mean of G6PD activity in EDTA tube and filter paper were compared using paired t-test. *Results:* All samples showed normal enzyme level. The mean of G6PD activity for samples in filter paper were lower compared to EDTA tube for all days. There was a statistically significance difference in the mean of G6PD activity between samples in EDTA tube and filter paper dried using a dryer on day 1 to day 3 ($p < 0.05$). *Discussion:* A dried blood sample show lower enzyme activity measurement and, hence is not suitable for G6PD enzyme activity determination. The enzyme activity in dried blood samples is probably adversely affected by humidity and temperature.

P-HE25. Karyotyping and fluorescence *in situ* hybridization (FISH) analysis of multiple myeloma

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Introduction: Multiple myeloma (MM) is a malignant monoclonal plasma cell proliferation with a wide genetic heterogeneity. Cytogenetics is an important prognostic factor of MM but the information is limited due to the malignant cells which have a low spontaneous proliferative activity. However, with the advent of fluorescence *in situ* hybridization (FISH) more abnormalities can be identified as the method is more sensitive and can be applied on the interphase and metaphase cells. *Objective:* This study aims to identify chromosome abnormalities in MM using the conventional cytogenetics (karyotyping) and FISH analysis. *Materials and methods:* Aspirated bone marrow cells from 17 MM patients were cultured and harvested following standard procedure. Chromosomes were stained using trypsin-Giemsa method. FISH using four probes were applied – IGH(14q32 rearrangement), D13S319/13q34 (-13/13q deletion), IGH/FGFR3 (t(4;14)), IGH/MAF(t(14;16)) and TP53(-17/17q deletion). These probes are associated with unfavorable prognosis in MM. *Results:* Abnormal karyotype was identified in three

cases (two cases with hyperdiploidy and one with deletion 7q31). MM patients with hyperdiploidy generally have better prognosis with better overall survival. FISH identified seven abnormal cases – IGH/FGFR3 (2 cases), 14q32 rearrangement (1case), deletion 13q34 (2 cases), deletion FGFR3 (1 case), deletion 7q31 (1 case). The remaining showed no abnormality by FISH and/or karyotyping. *Discussion:* As shown in this study and in the previous reports, most MM malignant cells harbor chromosome abnormalities. We have identified ~59% of MM cases with chromosome abnormalities using FISH and karyotyping. It is also evident from this study that FISH is more sensitive than karyotyping in detecting the abnormalities but numerical abnormalities such as hypo/hyperdiploidy are more readily observed by karyotyping. *Conclusion:* Therefore, both the conventional and FISH methods are essential for the detection of chromosome abnormalities in MM as it can assist the clinicians to decide the best treatment and management for their patients.

P-HE26. A case report of weakened A antigen expression due to acute myeloid leukaemia

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Introduction: Blood group antigens are an inherited character of the red blood cell (RBC) surface and their expression should be persistent throughout the life of an individual. However, weaken or missing antigens expression has been reported in patients with haematological malignancies. *Case report*: A 40-year-old gentleman was diagnosed with acute myeloid leukaemia (AML). He had no history of blood transfusion. A blood sample was sent for cross match. His ABO grouping has shown a discrepancy between the forward grouping (O) and the reverse grouping (A). Repeated testing on a new sample to exclude pre-analytical error remained unchanged. Further testing by forward grouping performed after 30 minutes incubation at room temperature displayed as O whilst at 4ºC, it showed weak positive reaction at anti-A, anti-A,B. The reaction became stronger (2+) with enzyme treated RBCs. The adsorption and elution test confirmed that this patient was group A, and the most likely diagnosis was weakened A antigen expression secondary to AML. *Discussion & Conclusion* : The alteration of ABO antigens expression in haematological malignancies is usually resulted from a number of mechanisms that affect the ABH genes within the malignant stem cells. There is inactivation of the genes that produce A/B transferase and/or H transferase. The effect of lost or weakened expression of ABO antigens depends on the size of malignant clone. If malignant clone size is significant, the effect will be a mixture of antigen positive and negative RBCs. If the malignant clone has totally replaced the normal clone, the effect will be complete loss of the respective antigens.It is crucial to recognize the weakened ABO antigen expression in patients with haematological malignancies. Failure to identify this may lead to wrong grouping of the patients and subsequently lead to disastrous outcome.

P-HE27. Iron status among the institutionalised elderly in Sarawak

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Introduction: Iron deficiency (ID) is the most common nutritional disorder and an important public health problem in South East Asia. It is generally assumed that, on average, about half of the cases of anaemia are caused by ID. Although iron stores appear to increase with advancing age, signs of ID still occur in the elderly in many countries. This study aimed to assess the iron status among the institutionalised elderly in Kuching, Sarawak; and to determine the correlations between iron status with gender, body mass index (BMI) and functional status. *Materials & methods:* This was a cross-sectional and experimental study that involved a private nursing home (Lions Nursing Home) and a government old folk home (Rumah Seri Kenangan). A total of 45 institutionalised elderly (48.9% males, 51.1% females) with written consent were recruited into the study. Anaemia was assessed by measuring the haemoglobin (Hb), haematocrit, red cell count, red cell indices and blood film examination. Iron status was determined by serum ferritin levels using enzyme immunoassay. BMI was calculated based on the height and weight whereas functional status was assessed using Barthel Index (BI) through a standard questionnaire. *Results:* Elderly males showed significantly higher Hb levels (12.6±1.5 g/dL) as compared to females (11.6±1.4 g/dL; p=0.028). However, the difference in their ferritin levels were not statistically significant (males: 104.2±64.0 ng/mL; females: 120.4±93 ng/mL; p=0.91). Prevalence of anaemia and iron deficiency anaemia (IDA) among the elderly was 53.3% and 8.9% respectively, with higher prevalence of IDA in the males (9.1%) than females (8.7%). All the ID subjects (8.9%) were found to be anaemic. In addition, iron overload was detected in 8.9% of the elderly. No significant correlation was noted between serum ferritin and BMI or functional status. *Conclusions:* Anaemia is very common among the institutionalised elderly in Sarawak. However, ID and IDA was uncommon, and do not correlate with BMI or functional status. Thus, anaemia of the elderly is mainly due to other causes which need further investigation (e.g. chronic illnesses and gastrointestinal bleeding).

P-HE28. Bone marrow necrosis in a chronic myeloid leukaemia patient receiving imatinib treatment

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Introduction: Bone marrow necrosis (BMN) is a rare pathologic entity associated with a wide variety of diseases. A paper published in the year 2000 reported there were 240 cases of BMN and 13 of them were CML cases. Most of the reported CML cases showed association between BMN and the blast transformation state of the disease. Here we report a case of BMN following imatinib mesylate therapy in Philadelphia chromosome positive CML in chronic phase. *Case report:* A 47-year-old woman was diagnosed to have CML in chronic phase after presented with 2 years history of constitutional symptoms, leucocytosis (WBC 408 x10^9/L) and hepatosplenomegaly. She was treated with hydroxyurea 500mg bd for a week before it was withheld due to rapid reduction of WBC before it was restarted 12 days later with 500mg od dose for a week before converted to Imatinib mesylate 400mg od. While on the thyroxine kinase inhibitor she had symptomatic anaemia with increasing frequency of blood transfusion. Serial laboratory investigation showed anaemia but her white cell and platelet count were normal. Her bone marrow aspirate examination showed no sign of blastic transformation. However, her

trephine biopsy showed substantial areas of necrosis. *Discussion and conclusion:* Studies have shown that imatinib mesylate might play a role in BMN especially if the case was in blast crisis. Overgrowth of leukaemic cells during blast crisis may cause stress that might lead toward necrosis of the cells. However, in this patient, there is a possibility of the BMN caused by imatinib mesylate as the patient had very high white cell count at presentation.

P-HE29. Cold AIHA in primary bone marrow follicular lymphoma

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Introduction: The association between autoimmune hemolytic anemia (AIHA) and non-Hodgkin lymphoma (NHL) is well known. However, the occurrence of AIHA with extranodal lymphoma is exceedingly rare and the majority of reported cases had advanced disease. Primary lymphoma especially primary follicular lymphoma arising from the bone marrow microenvironment is very rare. Here we report a case of primary bone marrow follicular lymphoma (PBMFL) with severe cold AIHA. *Case report:* A 65-year-old man presented with symptomatic anaemia for one week. Physical examination revealed no lymphadenopathy or hepatosplenomegaly. His full blood count showed pancytopenia with Hb of 6g/dl, WBC of 3.1×10^9/l and platelet count of 105×10^9/l. His bone marrow smears were haemodiluted with peripheral blood. However, the trephine biopsy findings were consistent with follicular lymphoma. CT scan showed moderate splenomegaly but no lymphadenopathy noted elsewhere. He was started with R-CHOP. At day 2 of chemotherapy, his peripheral blood smear showed presence of red cells agglutination. Direct Coombs test was positive with significant cold agglutinin titer and high serum LDH. Nevertheless, the cold agglutinin titer had reduced by 2nd cycle of R-CHOP. He achieved complete remission after completed 4 courses of R-CHOP followed by 2 courses of R-CVP. *Discussion and conclusion:* Here we highlight a case of cold AIHA as a complication of PBMFL. The rationale of using rituximab in cases such as this can be explained by the pathogenetic mechanism of NHL related AIHA due to antibody production directly or indirectly mediated by the neoplastic clone.

P-HE30. Evaluation of the rapid test screening method and pretest clinical score (4 T's) for the diagnosis of heparin-induced thrombocytopenia in Universiti Kebangsaan Malaysia Medical Center

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Introduction: Heparin-induced thrombocytopenia (HIT) is a pro-thrombotic disorder caused by heparin. Thrombocytopenia is a common scenario in hospitalized patients. When it occurs in patients receiving heparin, it would be useful to have a reliable rapid screening test coupled with a clinical scoring system that could differentiate patients with HIT from those with other reasons for thrombocytopenia. *Materials and methods:* There were 8 patients referred to the laboratory for the investigation of possible HIT in Universiti Kebangsaan Malaysia Medical Center following the clinical scoring system, the '4T's' of high (score 6-8) and intermediate (score 4-5) probability groups. The laboratory screening for HIT antibodies was STic EXPERT HIT rapid test kit which designed for qualitative detection of IgG antibodies against PF4/polyanion complexes using the lateral flow immunoassay principle. *Results:* A total of eight patients were screened for anti HIT antibodies. Six of them were end stage renal disease (ESRD) patients on hemodialysis, one with stage IV thyroid cancer and another patient with NSTEMI on clexane. 5 patients have intermediate 4T's score and 3 patients have high 4T's score. Out

of the eight patients screened, only two patients showed positive result with the HIT rapid test, each has a high and intermediate 4T's score respectively. The others showed negative results. *Conclusion:* A negative HIT rapid test together with low to intermediate 4T's scores is useful to exclude cases of HIT, and the diagnosis of HIT can be made for patients with high scores and positive screening result. However, in cases with intermediate scores and positive screening results and cases with high scores but negative screening results, serotonin release assay is important to exclude the diagnosis of HIT in these cases.

P-MB01. Percentage of True Positive and False Positive Blood Cultures and Factors Related with Contamination and True Bacteraemia at Clinical Training Centre, UiTM Sungai Buloh Campus

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Introduction: Blood culture is the most important method in detecting bacteraemia and fungaemia. However, the clinical significance of blood culture isolates may be doubtful and unreliable due to contamination. The proper blood taking technique, diagnosis and identification of bacterial and fungal infection is very important to produce accurate and reliable blood culture reports. The study aims to determine the percentage (%) of the true positive and false positive blood cultures and factors related with contamination and true bacteraemia. *Materials & methods:* The clinical data was obtained and extracted from medical record manually from year 2012 and 2013. *Results:* There were total 124 blood culture bottles collected from 62 patients at Clinical Training Centre, UiTM Sungai Buloh campus for 2012 and 2013. Of the total blood cultures, 30 (24.2%) were positive blood cultures and 94 (75.8%) were negative blood cultures. From the total number of positive blood cultures, 6 (4.8%) of it were considered as false positive due to contamination. Based on patient clinical diagnosis, chart and organisms isolated, all patients considered to have a true bloodstream infection showed increased temperature which is >38°C, heart rate >90 beats/minute and white cell count exceeding normal range (4.0 – 11.00 $10e^3$/µl). These parameters show the significance of systemic inflammatory response syndrome (SIRS) which lead to sepsis. Meanwhile, patients who show normal temperature, heart rate and normal white blood cell count but came out with positive culture coagulase-negative staphylococcus or other skin microflora were considered as false positive blood culture due to contamination. *Discussion:* Percentage of contamination rates of blood culture is above the benchmark of 3%. This contamination rate can be reduced by having a well trained phlebotomist and good aseptic technique when collecting blood from patients. Furthermore, clinical diagnosis, patient condition, and other specific finding from laboratory test are very important as indicator in the evaluation of sepsis.

P-MB02. *Rhodococcus equi* Infection: A Report of 2 Cases

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Introduction: Rhodococcus equi (formerly known as *Corynebacterium equi*) is a zoonotic infection which has emerged as an important pathogen in immunocompromised patients. It is a facultative, nonmotile, intracellular organism which has the ability to form a red (salmon-colored) pigment, can be weakly acid-fast, has a similarity to diphtheroids and is currently grouped under aerobic actinomycetes.

Case report: We described two cases of *Rhodococcus equi* infection with different outcome. The first case was a 33-year-old man living with HIV. He presented with a chronic cough of one month duration, which was non-productive, associated with high-grade fever, night sweats, excessive loss of weight and increasing dyspnoea. Chest radiograph showed consolidation at the left lung up to mid-zone. Blood and sputum cultures isolated *Rhodococcus equi.* The patient eventually succumbed to the infection and died 3 days after admission. The second case was a 22-year-old man, post-renal transplantation from a living-non-related donor on tacrolimus. He presented with tonic clonic seizure and was diagnosed with cryptococcal meningitis. He developed low grade fever after 2 weeks in the ward. Computed tomography (CT) of thorax revealed features in keeping with infective process. Bronchio-alveolar lavage culture isolated *Rhodococcus equi*. He was treated with three weeks of intravenous imipenem and oral rifampicin and his symptoms resolved. He was discharged well after three weeks on treatment and was continued with oral rifampicin and oral clarithromycin for 6 weeks. *Discussion & conclusion: Rhodococcus equi* is a gram-positive coccobacillus, and it may be misidentified as diphtheroids. The organism can be difficult to eradicate, making treatment challenging. Morbidity is related to complications and chronicity of the infection. Standard treatment regimens have not been established and should be guided by antibiotic susceptibility testing. Increased awareness of the organism and its infection may help with early diagnosis and timely treatment.

P-MB03. Significance of anti-HCV signal-to-cutoff (S/CO) ratios in blood donors: an experience from Regional Referral Hospital in Malaysia

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Introduction: Routine screening of donated blood for hepatitis C virus antibody (anti-HCV) is a recommended practice by WHO to ensure safety of blood products. As a result of highly sensitive anti-HCV enzyme immunoassays, false-positive results are common in low-prevalence population. The purpose of this study was to determine whether anti-HCV signal-to-cutoff (S/CO) ratios could be used to identify false-positive and true-positive results in ≥95% of the time. *Materials & methods*: We reviewed the serological results of individuals donated between 2008 and 2012. Monolisa anti-HCV Ag-Ab enzyme immunoassay was used for screening. Line immunoassay (LIA) was used for confirmation in anti-HCV positive donors. Optimal cutoff points that identify ≥95% of false-positive and true-positive results were determined by using receiver operating characteristic curve analysis. *Results*: Of 120,674 specimens, 257 were anti-HCV positive with S/CO ratio of ≥1. S/CO ratios of <3.5 and ≥4.5 were the optimal cut-off points to identify false positive and true positive results respectively. 111 of 115 (96.5%) with S/CO ratio of <3.5 were false-positive with negative/intermediate LIA and 4 (3.5%) were true positive. 128 of 133 (96.2%) with S/CO ratio of ≥4.5 had true HCV infection with positive LIA and 5 (3.8%) were false positive. *Discussion*: Our study showed that S/CO ratios were highly predictive of confirmatory HCV test results in ≥95% of the time. S/CO ratios could potentially be used as "stand-alone" tests for exclusion and inclusion of HCV infection. Confirmatory HCV tests should be limited for anti-HCV positive individuals whose S/CO ratios could not be used to discriminate false-positive and true-positive results accurately. This can reduce HCV testing cost significantly and improve the reliability of reported test results. If confirmatory tests are not performed, laboratories need to establish optimal S/CO ratios which can predict confirmatory HCV test outcomes in ≥95% of the time.

P-MB04. Multidrug-resistant Tuberculosis (MDR-TB): Smear Positive Acid Fast Bacilli (AFB) as Independent Risk Factor; Uitm Experience

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Introduction: Recently dengue outbreak may have out-numbered TB cases in Malaysia, but not for mortality rates, which still belongs to TB. In tuberculosis, MDR-TB contributed to the significant increase of mortality rates. WHO estimated that more than 2 million people will contract MDR-TB in 2015. In Malaysia, the MDR-TB rate was 0.3% in 2005, raised to 1.3% in 2011 and reduced slightly to 0.7% in 2013. Hence, this study conducted with the aim to identify smear positive AFB and its correlation with development of MDR-TB. *Materials & Methods:* One hundred sputum samples were collected, stained and cultured from January 2011 to April 2014. Kinyoun stain was used to detect AFB in the sputum. All sputum were submitted to the University Malaya Medical Centre for identification and susceptibility testing. *Results:* Of 100 sputum collected, 78% (n=78) were culture negative and 22% (n=22) culture positive. Of 22% culture positive, 77% (n=17) isolates were identified as *Mycobacterium tuberculosis* complex and 23% (n=5) as non-tuberculous *mycobacterium* (n=3, *Mycobacterium abcessus*; n=1, *Mycobacterium avium intracellulare* complex; n=1, *Mycobacterium fortuitum*). Of 77% *Mycobacterium tuberculosis* complex isolates, 71% (n=12) were AFB positive and 29% (n=5) AFB negative respectively. Among 71% AFB positive there was no MDR-TB detected, but there was 24% (n= 3, isoniazid; n=1, ethambutol) monoresistant to anti-TB drug detected. *Conclusion:* MDR-TB was not detected at our centre. However monoresistant to anti-TB drug were identified with high rate among smear positive sputum that predicts more occurrence of MDR-TB. The MDR-TB cases may be detected with increased number of specimen than employed in this pilot study. Nevertheless, these finding may serve as preliminary data for further MDR-TB projects.

P-MB05. Time-to-report Analysis of Vitek 2 System, an Automated Phenotypic Method, for the Identification of Medically Significant Gram-Negative Bacteria

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Introduction: A shorter time taken for identification of microorganism facilitates earlier susceptibility testing as well as accurate and timely reporting of microbiology results. The objective of this study is to retrospectively analyse time-to-result performance of VITEK 2 system's (bioMerieux) gram-negative identification (GN-ID) card. *Materials and methods:* One hundred and sixty gram-negative bacteria which were isolated from sterile and non-sterile clinical samples were collected between 2011 and 2014. These isolates were identified using VITEK 2 GN colorimetric identification card, in the microbiology laboratory of Centre for Pathology & Diagnostic Research Laboratories (CPDRL), Faculty of Medicine, University Teknologi MARA, Sungai Buloh, Malaysia. *Results:* The VITEK 2 system successfully identified eight Enterobacteriaceae and nine non-Enterobacteriaceae up to their species and subspecies level. The two most common Enterobacteriaceae, *Klebsiella pneumoniae* (n = 40) and *Escherichia coli* (n = 34); as well as those most common from the non-Enterobacteriaceae group, *Pseudomonas aeruginosa* (n = 26) and *Acinetobacter baumanii* (n = 17) were closely analysed. Mean time-to-results for the identification of those isolates were as follows; *K pneumoniae* 5.3 hours (range 4 to 10.25 hours), *E coli* 4.7 hours (range 2.75 to 7 hours), *P aeruginosa* 6.4 hours (range 5

to 8 hours) and *A baumanii* 6.1 hours (range (5 to 7 hours). This study demonstrated that in general, VITEK 2 system has the capacity to identify most of the common, medically significant gram-negative bacteria in between 4 to 6 hours' time. *Discussion:* VITEK 2 is an automated phenotypic identification system which is able to produce results within the same day of performing the assay, in comparison to the more traditional, manual version that typically requires an overnight incubation for completion. This translates to VITEK 2 system's superiority in expediting antibiotic susceptibility testing and eventually faster result reporting to the clinicians for better patients' care.

P-MB06. The Accuracy of Bacterial Identification in External Quality Assurance Programmes Using VITEK 2 Identification System

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Introduction: Automated microbial identification in clinical microbiology has emerged as a diagnostic tool to rapidly identify the causative agents in patients with sepsis. The rapidity in acquisition of results however must not compromise its accuracy. The main aim of the study is to evaluate the accuracy of VITEK 2 system (bioMérieux) in bacterial identification. *Materials & Methods*: A total of 50 bacterial isolates received from the External Quality Assurance Programmes (EQAP) between January 2012 to December 2013 were subjected to identification using the VITEK 2 system at the Microbiology Diagnostic Laboratory, Faculty of Medicine, Universiti Teknologi MARA. The EQAP providers were the Royal College of Pathologists of Australasia (RCPA) and Institute for Medical Research (IMR) Malaysia. The isolates consisted of 27 gram-negative organisms, 14 gram-positive organisms and 6 *Neisseria-Haemophilus* group and 3 anaerobic organisms. The accuracy of the identification of the isolates was based on the answers obtained from the EQAP report. *Results*: VITEK 2 accurately identified 89.5% and 91.7% at the genus level of the isolates from RCPA and isolates from IMR respectively. While at the species levels, the accuracy was 84.2% for the RCPA and 83.3% for the IMR isolates. Four isolates were incorrectly identified as the types of the organisms are not included in the VITEK 2 database. One organism failed to be identified. *Discussion*: In the study, the sensitivity of VITEK 2 in the identification of the bacterial isolates obtained from the EQAP was 84.0%. Since our evaluation was mainly limited to EQAP strains and small sample size, it is predicted that the VITEK 2 system should be used in conjunction with other identification system for optimal results in a routine clinical microbiology laboratory.

P-MB07. Inhibitory Effects of Selenium Nanoparticles on Nanobacteria Isolated From Urinary Stones of A Northeastern Iranian Population

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Introduction: Nanobacteria (NB) or calcifying nanoparticles (CNP) has certain characteristics such as small size (0.1 to 0.5 microns) and high resistance to heat. NB has received good attention as one of the agents that has most probably main role in initiation of pathologic calcification in vivo. *Material and*

methods: Urinary stones from patients were collected from pathology laboratories. After powdering the stones, these powders demineralized with HCl and neutralized with Tris buffers. Culturing of these organisms was performed after filtering the solution through 0.22 μm membrane filters in gamma-irradiated DMEM, fetal bovine serum (FBS) and selenium nanoparticles. Stone chemical analysis was performed by XRD and commercial kit methods. Presence of nanobacteria was investigated with Scanning Electron Microscopy (SEM), Transmission Electron Microscope (TEM), microbiologic and spectrophotometric methods. *Results:* the nanobacteria growth was observed using spectrophotometer. Results of scanning electron microscope (SEM) showed a spherical shape in size of 33 to 380 nm in medium without selenium nanoparticles. Energy Dispersive X-ray spectroscopy (EDS) also showed that these organisms have a calcium and phosphor coating around itself. After addition of selenium nanoparticles, calcium and other minerals decreased. *Conclusions:* Nanobacteria have spherical form and they grow very slowly in medium without selenium nanoparticles. Selenium nanoparticles may play an inhibitory role on nanobacteria growth due to its impediment effects on calcium oxalate crystal growth an aggregation.

P-OT01. Prevalence of overweight/obesity and its associated risk factors amongst the primary school children in a suburban area of Malaysia

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Introduction: Childhood obesity increases the risk of obesity and other non-communicable diseases in adulthood. This study aimed to determine the prevalence of overweight/obesity and its associated risk factors amongst primary school children in a suburban area of Malaysia. *Materials & methods:* This cross-sectional study was conducted amongst children aged 8 and 9 years old in five randomly selected primary schools in suburban Selangor. Body weight and height were measured to determine the body mass index (BMI). Data on the socio-demographics, dietary habits, physical activity and duration of sleep were obtained through interview. A 41-items food frequency questionnaire was used to determine the type of diet taken by the children. *Results:* A total of 335 (mean age 9.18±0.28 years) children were recruited. Sixty-percent (n=191) were females. The mean BMI was 16.39±3.58 kg/m^2. The prevalence of overweight and obesity was 11.3% and 12.2%, respectively. There was no significant difference in the socio-demographic characteristics and duration of sleep between overweight/obese and non-overweight children. A significantly higher proportion of overweight/obese children (60.8%) took unhealthy diets containing high fat and high sugar than non-overweight children (43.8%) (p<0.05). The duration of watching TV and playing electronic gadgets per week was significantly longer in the overweight/obese (14.92±13.33hour versus 11.54±11.46 hour) (p<0.05). *Discussion:* Taking an unhealthy diet and spending longer time watching TV or playing electronic gadgets were significantly associated with overweight/obesity in the Malaysian primary school children surveyed. This finding is consistent with that reported in other studies and highlights the need for lifestyle intervention to prevent future obesity or related diseases amongst obese children.

P-OT02. Risk factors associated with increasing waist circumference among Malaysian adolescents in the Klang Valley

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Introduction: An increasing prevalence of abdominal obesity (AO) in adolescents is a worldwide trend. The significance of this observation is the association between abdominal obesity and an increased morbidity and mortality. The aim of this study was to determine the prevalence of AO and the risk factors associated with increasing waist circumference among adolescents (15-17 years old) in the Klang Valley. *Methods & materials***:** A cross-sectional study was carried out among adolescents in five randomly selected secondary schools. Body mass index (BMI) and waist circumference (WC) were obtained. Information on socio-demographic characteristics, dietary habits, level of physical activity, and sleeping duration were obtained via interview-administered questionnaires. A 73-item food frequency questionnaire (FFQ) was asked to determine the habitual food intake of the participants. *Results:* Of the 832 adolescents examined, 56.1% were female. The median age of participants was 16 (IQR=1) years. The ethnic distribution was as follows: Malays 48.3%, Chinese 40.6%, and Indian and other ethnic groups 11%. The overall prevalence of abdominal obesity (AO) was 11.3%. Linear regression analysis showed that (i) skipping dinner (β=4.535; SE=1.035; $p<0.001$), (ii) lower cereals/tuber FFQ score (β= -0.293; SE=0.098; $p<0.05$), and (iii) higher sweet drink/dessert FFQ score (β=0.244; SE=0.4097; $p<0.05$) were significantly associated with WC after adjustment for gender. However, after adjustment for BMI and gender, these three factors were not significantly associated with WC. Instead, Indian ethnic group was a significant independent risk factor associated with WC (Malays versus Indian: β =-1.608; SE=0.559; $p<0.05$; other ethnicity versus Indian: β =-4.072; SE=1.823; $p<0.05$). *Discussion:* Lifestyle intervention that emphasizes on changes in dietary habits is suggested for the management of abdominal obesity in the adolescents.

P-OT03. HPV 16 E2 as an apoptosis-inducing protein *in vitro*: a systematic review

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Introduction: High risk human papillomavirus type 16 (HPV-16) is a well known etiological factor for cervical cancer and several other oropharyngeal cancers. One of the protein produced by early gene in HPV 16 is E2, which is postulated to cause death of cancerous cells via both p53-dependent and independent pathways. The discovery has potential to widen the therapeutic window for a novel vaccine development. This systematic review aims to elucidate the current state of knowledge on E2 as an apoptosis inducer *in vitro. Method:* Medline and EMBASE were searched up to April, 2014 using * HPV or Human Papillomavirus and * Apoptosis or Cell death and **in vitro* as the search terms. There were no language and year restrictions and only studies involving E2 protein were chosen. Information was collected on the study design, vector and cell line used, method involved to verify the cell death, and the apoptosis level. *Results:* There are many *in vitro* studies on HPV16 proteins causing cell death on the cancerous cell lines, but limited studies focusing on E2 protein. Only one paper directly studied E2 as an apoptotic agent. The paper reported that a combined radiation treatment with recombinant adenovirus-M5 with E2 protein is claimed to increase the potency with $p<0.01$. The study showed the M5 is able to act as HPV 16 E2 delivery platform for targeting the SiHa and HeLa cancerous cells while enhancing the radiation treatment for cervical cancer. Apoptosis events are identified using Annexin V-FITC and propidium iodide staining while the significance level is measured by flow cytometry.

Conclusion: E2 protein may provide a new insight for targeting HPV 16-associated cancerous cells by apoptosis. However, there are very few studies studying E2 protein. Therefore, more studies are required to elucidate the significance of E2 protein in apoptosis while widen the possibility of developing new vaccine therapy.

P-OT04. Emotional work, mental resilience and psychological distress among female medical-oncology nurses

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Introduction: The work of oncology nurses can be emotionally taxing, arising from the 'emotional work' (EW) they perform, such as expressing care, concern and empathy in the course of managing and improving the well-being of the patients. This makes them vulnerable to developing major psychological distress, which might affect their work performance. However, this relationship between EW and distress may be moderated by the nurses' personal level of mental resilience and demography. Therefore, research in local settings is recognized as a necessary step to improve oncology nurses' well-being and, ultimately, patient care. *Methods and materials:* Self-report measures of demography, psychological distress (General Health Questionnaire-12), mental resilience (Resilience Scale-14), and emotional work (Emotional Work-6) were administered to oncology nurses in a general hospital in Singapore (response rate = 92%). Data from all female respondents in the medical-oncology department (N = 172; 76% registered nurses, 45% with oncology specialty training, 65% working in inpatient settings) were analyzed. *Results:* EW was positively related to mental resilience, but not to distress. Distress was negatively correlated with mental resilience. Similarly, participants scoring above the GHQ-12 bimodal threshold of 2 (caseness of distress; 32% of cohort) had lower mental resilience than those with low to normal levels of distress. Registered nurses, and those who had received oncology-specialty training, had lower mental resilience scores and higher distress scores than their enrolled nursing colleagues. There were no differences in EW across demography or caseness of distress. *Conclusion:* Although oncology nurses reported psychological distress, it appeared that their mental resilience was a protective factor. Future research could focus on how to increase nurses' mental resilience. This is especially important for registered nurses, whose greater distress could have arisen from being tasked with more emotionally demanding responsibilities.

P-OT05. The reduction of psychiatric symptoms in newly diagnosed cancer patients through a Nurse-led Psychosocial Intervention Program

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Introduction: Psychiatric co-morbidity is common in cancer patients, making it necessary to provide psychosocial support. This is especially so for newly diagnosed patients, as there is a lack of evidence-based guidelines on psychosocial interventions for them. To address this need, the National University Cancer Institute, Singapore (part of the National University Hospital) developed a nurse-led psychosocial intervention program, as part of a Healthcare Quality Improvement Project. The aim of this study is to evaluate the effectiveness of this intervention program in reducing psychiatric symptoms, through

the use of pre- and post-intervention measures. *Methods and materials:* 111 participants were offered the intervention; 51 (46%) were not keen in participating and formed the control group. Self-report questionnaires consisting of the Distress Thermometer (DT) and the Hospital Anxiety and Depression Scale (HADS) were administered at baseline and 2 months, to measure the levels of distress, anxiety and depression. A 20-minute intervention was delivered face-to-face by trained oncology nurses monthly (2 visits). It included psycho-education on symptom recognition and management, cognitive and behavioral relaxation techniques, as well as counseling and supportive therapy. *Results:* Overall, there was a significant decrease in all three types of symptoms from baseline to 2 months. Further analyses revealed significantly greater decreases for symptoms of anxiety ($F = 7.039$, $p < 0.05$) and distress ($F = 6.628$, $p < 0.05$) for the intervention group as compared to the control group, but not for symptoms of depression ($F = 2.647$, $p > 0.05$). *Conclusion:* Although the psychiatric symptoms appear to decrease over time, this study has shown that nurse-led psycho-education and patient support are effective in reducing these symptoms by a larger extent as compared to the symptoms in patients who did not receive the intervention. Future research could investigate into other intervention programs that could facilitate the reduction of depressive symptoms.

P-OT06. Gene silencing effect of single walled carbon nanotubes (SWNT) conjugated with siRNA

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Introduction: Carbon nanotubes (CNTs) are a potential candidate for drug, antigen and nucleic acid delivery vehicle in nanomedicine. The large surface of CNTs provides a structural advantage and allows loading of functional groups or therapeutics such as nucleic acid, drugs and proteins. Our study is to deliver siRNA to cells using single walled carbon nanotube (SWNT) to achieve gene silencing effect. *Materials & methods:* SWNT was functionalized by dissolving one mg of HiPCo® SWNT and 5 mg of PL-PEG-NH_2 or PL-PEG-maleimide with different molecular weights in 5 ml of water, sonicated for 60 min at room temperature and centrifuged for 6 h (24,000*g* at room temperature). The supernatant were collected and measured their concentration at 808nm by UV–VIS–NIR spectrometer. The resulted non-covalent functionalized SWNTs were further conjugated with a 5'-thiolated siRNA against GFP (siGFP) and RFP (siRFP). The siRNA target sequence against GFP is 5'-CGGCAAGCTGACCCTGAAGTTCAT-3' and that of against RFP is 5'-GUGGGAGCGCGUGAUGAACTT-3'. *In vitro* silencing of GFP and RFP expression by SWNT-siRNA were evaluated in stable expression cell lines by fluorescence spectroscopy. *Results:* A range of 50-80% GFP expression knocked down was observed in H1299, HeLa, MCF-7 and 293T cells by SWNT-siGFP. SWNT conjugated with both siGFP and siRFP were also shown knocking down both GFP and RFP simultaneously in H1299 stable co-expression cell line. *Discussion:* The successful knockdown of GFP expression in different cell lines indicated that siRNA were capable to be released from the conjugated SWNT-siRNA in the cytoplasm and silent the gene expression. It is also indicated that two different types of siRNA targets could be conjugated with SWNT and achieved two different gene silencing effects simultaneously.

P-OT07. Genetic engineering of immortalized human keratinocytes (NIKS) with human papillomavirus oncogenes

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Introduction: High-risk human papillomavirus (HPV), type particularly HPV 16 and 18 are strongly associated with pathogenesis of cervical cancer. In addition, high-risk HPV are also associated with other epithelial cancers such as head and neck cancers. Introduction of viral oncogenes such as HPV16 E6 and E7 has been shown to induce early neoplastic changes in immortalized human keratinocytes mimicking the cellular neoplastic transformation of cervical cancer. Such manipulation of cultured cells can provide useful in vitro models for drug screening and disease modeling. The aim of our study is to establish a research platform for the aforementioned studies using Normal Immortalized Human Keratinocytes (NIKS), which is derived from human foreskin. *Materials and methods:* The NIKS were transfected with recombinant plasmids expressing HPV oncogenes; these include plasmids pLXSN HPV16 E6, pLXSN HPV16 E7, pLXSN HPV16 E6/E7 expressing the HPV16 E6, E7 and E6 plus E7 oncogenes respectively. The transfected NIKS clones were selected using G418 antibiotics and the expression of E6 and E7 oncogenes were validated by both the RT-PCR and Western blot. Cell cycle profile of the transfected and non-transfected (control) cells were determined by cell cycle analysis. *Results:* Transfected NIKS were positive for HPV16 E6 and E7 oncogenes at both mRNA and protein levels. Transfected NIKS showed higher G2/S phase events compared to the non-transfected cells. *Conclusion:* HPV16 E6 and E7 transfectants were successfully produced.

P-OT08. Generation of primary cervical keratinocytes culture from hysterectomy sample

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Introduction: Cell lines commonly employed for experimental work involving screening and/or testing of chemotherapeutic agents are immortalized cancer cell lines. Numerous limitations are associated with the use of such test systems, the most notable being the extensive and dynamic changes (mutations) and variations in their genomes. Primary cells, on the other hand are more physiological and representative of cells in vivo, and therefore may serve as a better in vitro experimentation and exploration platform for drug screening as well as for various other research studies. Here, we sought to establish primary cervical keratinocyte culture as a platform for drug testing studies. *Materials and methods:* Hysterectomy specimens were collected during surgery from patients with informed consent. Cervical tissue from the specimens were harvested and processed immediately to isolate primary cervical keratinocytes by the enzyme digestion method. The cells were maintained and expanded on irradiated 3T3 feeder layer in complete medium supplemented with epidermal growth factor, cholera toxin and insulin to develop primary cell line. Proliferation potential and doubling time of the primary keratinocytes were determined by cell cycle analysis and MTT assay, respectively. All tissue specimens were tested for the human papillomavirus (HPV) using primers specific to the virus in a polymerase chain reaction. Only HPV negative samples were used for establishing the primary cell cultures. *Results:* We have been successful in establishing the cervical cell line over several passages. The primary cervical keratinocytes were polygonal in shape and grow in colony form on 3T3 feeder cells. Proliferation capacities of the primary keratinocytes progressively declined after a few passages and became quiescent after passage eight. *Conclusion*: Primary cervical keratinocytes cultured from hysterectomy sample can be generated, albeit for a limited number of passages. These primary normal cells along with cells isogenic except for defined changes engineered (eg. over-expression of oncogenes or ablation of anti-oncogenes) can be used as an experimental model or a drug screening platform.