Laboratory Studies

Chair: H Reid and L Young-Martin

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Assessment of trace element concentrations in human placenta by portable handheld X-ray fluorescence analyser

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Objective: To explore the use of a portable handheld X-ray fluorescence (XRF) analyser in detecting trace elements in human placenta tissue.
Methods: Twelve placenta samples were randomly selected from mothers at the University of the West Indies (UHWI) labour ward from January to March 2013. Each participant answered a questionnaire on their fish eating patterns. The samples were dried and pelletized. The concentrations of trace elements were determined using Niton portable handheld X-ray fluorescence analyser.
Results: The ages of the mothers were from 19–44 years old, and the fetal weight ranged from 2.27–4.56 kg. The mean dry weight concentration for the ion was 531.1 ± 81.9 ppm and zinc was 50.5 ± 14.9 ppm. The XRF analyser recorded a range of 835 ± 112 ppm to 195 ± 56 ppm for iron and 41 ± 21 ppm to 64 ± 21 ppm for zinc. Zinc concentrations were within close range for analysed samples. The obtained values were similar to the literature from other analytical techniques. They are also within range of dietary requirements. Manganese, copper, arsenic and selenium were below the detection limit. There was no significant trend in maternal fish consumption and iron and zinc concentrations in placenta.
Conclusion: The portable handheld XRF analyser is suitable to detect concentrations of iron and zinc in placenta samples. It can be used as an efficient screening tool for trace element analysis.

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Influenza viraemia and its significance among Jamaican patients, 2009 to 2012

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Objective: To determine the prevalence of influenza viraemia among patients with laboratory diagnosed (respiratory swabs) influenza infection in Jamaica during 2009 to 2012.
Method: Sera and respiratory swabs were randomly submitted from patients diagnosed with acute respiratory tract infection (ARI) and severe acute respiratory tract infection (SARI) to the National Influenza Centre. Sociodemographic data were obtained from laboratory and hospital records and influenza virus detected using real-time polymerase chain reaction (PCR) [CDC primers]. Respiratory swabs were first tested for the presence of the influenza virus and sera of the corresponding positive swabs tested subsequently for the virus. The Statistical Package for the Social Sciences (version 17) was used for data analysis.
Results: A total of 65 swabs and corresponding sera were tested for the presence of influenza virus. Males and females accounted for 52.6% and 47.4%, respectively, with 52.3% of cases classified as SARI and 47.7% ARI. The distribution of influenza types and subtypes was as follows: 46.2 % (30/65) influenza A (H1N1) pdm09, 38.5% (25/65) influenza A (H3N2), 15.4% (10/65) influenza B and 0% each for seasonal influenza A (H1N1), avian A (H5N1) and A (H3N2) variant. There were 10.8% (7/65) of all cases that were fatal and specifically 22.6% (7/31) in SARI cases. Viraemia was detected in 42.8% (3/7) of fatal cases compared to non-fatal cases where no viraemia was found (Fisher's exact test, $p = 0.001$). There was a positive correlation between viraemia and fatality (rs = 0.633, $p = 0.0001$) and viraemia and influenza A (H1N1) pdm09 (rs =

0.238, 0.002). The mutation D222G which is documented to be associated with increased severity, was detected only among SARI influenza A (H1N1) pdm09 cases.
Conclusion: Influenza viraemia is associated with fatality and may be a predictor of poor prognosis.

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Influenza vaccine composition and circulating influenza viruses in Jamaica, 2010 to 2012

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Objective: To compare the influenza viruses in Jamaica with Northern Hemisphere Influenza vaccine components from 2010 to 2012.
Method: Swabs and aspirates systematically submitted from patients seen at sentinel sites with respiratory infections were included in this study. Influenza virus was detected using real-time polymerase chain reaction (CDC-rRT-PCR). Antigenic characterization of virus isolates was by haemagglutination inhibition (HI) and sequence analysis (CDC).
Results: A total of 1803 specimens were tested of which 13.26% were positive for influenza. Among influenza A viruses detected, the prevalence of influenza A (H1N1) pdm09 (antigenically characterized as A/California/07/2009-LIKE (pandemic H1N1) and which has been a component of the Northern Hemisphere (NH) vaccine since 2010) was: 10% (8/80) in 2010, 86.67% (13/15) in 2011 and 46.15% (12/26) in 2012. Influenza A/H3 accounted for 85% (68/80) in 2010, 13.33% (2/15) in 2011 and 36.1% (13/26) in 2012 and has been included in the NH vaccine as A/Perth/16/2009-like (H3N2) in 2010, A/Victoria/210/2009 in 2011 and A/Victoria/361/2011 (H3N2)-like virus in 2012. Seasonal influenza A/H1N1 which has not been a component of the NH vaccine since 2009–2010 was detected in 2012 at a prevalence of 3.85%. Influenza B viruses increased from 3.53% (3/85) in 2010, to 68.75% (33/48) in 2011 to 77.36% (82/106) in 2012. Influenza B Victoria (B/Brisbane/60/2008-like) and Yamagata lineage viruses co-circulated in Jamaica from 2010 to 2012, with the predominant Yamagata lineage-virus type antigenically characterized as B/Wisconsin/1/2010–like. Yamagata lineage-viruses detected did not have corresponding components in the 2010–2011 or 2011–2012 NH vaccines. Influenza A (H1N1) pdm09 strain designations resulting from viruses characterized include A/Jamaica/641/2012 and A/Jamaica/764/2012 and the Yamagata lineage-viruses B/Jamaica /2988/2012 and B/Jamaica/70/2012.
Conclusion: The majority of circulating viruses in Jamaica from 2010 to 2012 had similar virus components to the NH vaccine, with the exception of the B /Yamagata-lineage viruses detected in 2012.

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Investigation of the mechanism of the anti-inflammatory action of the fluid from the bud of *Spathodea campanulata* (African tulip)

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Objective: To investigate the mechanism of the anti-inflammatory action of the fluid from the bud of *Spathodea campanulata* in Sprague-Dawley rats.
Method: Sprague-Dawley rats (male, 200–250 g) were divided randomly into three groups of six. Animals were used as their own control and were administered 0.9% saline orally immediately before the induction of inflammation with bradykinin 10 mg/kg ip (group 1), prostaglandin E2 (group 2) and histamine (group 3). Paw volumes were measured at fifteen minutes intervals for two hours. After one week, the groups were administered ip a selective bradykinin antagonist 0.4 mg/kg, mefenamic acid 3.6 mg/kg and diphenhydramine 0.7 mg/kg, respectively, as treatment 15 minutes before the induction of inflammation. After two weeks, each animal was administered African tulip extract 50 mg/kg ip 15 minutes before inflammation was induced in the paw. The results were analysed by analysis of variance (ANOVA) and Student's *t*-test and statistical significance taken at a p value of 0.05.
Results: After the sub-plantar injection of 0.1 ml of the pro-inflammatory agents (bradykinin, histamine and prostaglandin E2), marked increases in paw volumes were seen over two hours (in the presence of saline). In the presence of their respective antagonists, increased swelling was also seen but to a lesser extent compared to the controls. The results showed that the African tulip extract (50 mg/kg) significantly inhibited the inflammation induced by bradykinin ($p = 0.016$), prostaglandin E_2 ($p = 0.041$) and histamine (p 0.001).
Conclusion: The extract from the fluid in the bud of the African tulip plant reduced the inflammation induced by bradykinin, prostaglandin E2 and histamine. The mechanism may involve inhibition of the release of these mediators or receptor antagonism.

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The prevalence of potential drug-drug interactions on prescriptions filled at the University Health Centre Pharmacy between November 2012 and February 2013

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Objective: To identify potential drug-drug interactions (DDIs) on prescriptions filled at the University Health Centre Pharmacy, Mona campus, Jamaica, between November 2012 and February 2013.

Methods: This investigation utilized a cross-sectional analysis on all prescriptions with more than one drug that were filled at the Health Centre Pharmacy during the study period. Potential DDIs were identified using an online drug interactions checker database.

Results: During the period of the study, a total of 2814 prescriptions were analysed for potential DDIs. The prevalence of potential DDIs found during the study period was 49.82%. A total of 1367 drug-drug interacting pairs were identified, with the three most frequently interacting pairs being aspirin and losartan (94), hydrochlorothiazide and metformin (84) and diclofenac tablets and diclofenac gel (68). Major potential DDIs accounted for 14% of the total number of interactions detected, while moderate potential DDIs and minor potential DDIs were 81% and 5% respectively.

Conclusion: With the prevalence of potential DDIs at approximately 50%, this study has highlighted the need for educational initiatives to ensure that physicians and pharmacists collaborate in an effort to minimize the risks to the patients. These interactions are avoidable for the most part, as the use of online tools can facilitate the selection of therapeutic alternatives to reduce these risks as well as guide decisions in light of the need for closer patient monitoring.

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Currently recommended breakpoints for drug susceptibility testing of *Mycobacterium tuberculosis* to fluoroquinolones may be erroneous and lead to the misclassification of resistant and susceptible strains

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Objectives: (i) To investigate if the currently recommended breakpoints for drug susceptibility testing (DST) of *Mycobacterium tuberculosis* to fluoroquinolones can adequately separate susceptible strains from resistant, and (ii) to elucidate the level of cross-resistance between older and newer fluoroquinolones.

Methods: In total, 75 *M tuberculosis* strains with a varying previously reported resistance pattern were included. Determinations of the minimum inhibitory concentration (MIC) were done on standard MIddlebrook 7H10 agar plates containing serial two-fold dilutions of ofloxacin and levofloxacin and the existence of mutations conferring resistance was detected by sequencing of the gyrA gene.

Results: As expected, the MICs of strains without resistance mechanisms were normally distributed; however, the highest MICs among those strains were considerably lower than the currently recommended DST breakpoints of 2 mg/L for the tested fluoroquinolones. In addition, the MICs of strains with resistance mechanisms, particularly those with mutations in codon 94, were close to or even overlapping the upper part of the normal distribution of susceptible strains. Nevertheless, cross-resistance between ofloxacin and levofloxacin was complete.

Conclusion: Our findings suggest that the present DST breakpoints for fluoroquinolones are too high, which means that resistant strains may be misclassified as susceptible. The close relation between MICs of strains with and without mutational resistance mechanisms may be compensated by using an "I" (intermediary) category when reporting DST results. Previous papers have proposed that ofloxacin-resistant strains may be susceptible to levofloxacin, but we hypothesize that these conclusions may be partly (or wholly) due to the use of erroneous DST breakpoints.