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P43 Consequences of mandatory screening in corneal transplantation

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Background: Human T-cell lymphotropic virus (HTLV) infection is associated with adult T-cell lymphoma (ATL), tropical-spastic-paraparesis (TSP) and HTLV-associated-myelopathy (HAM). In 2004 mandatory screening of corneal donors for HTLV was introduced. We audited HTLV screening of cadaveric corneal donors at the CTS-Manchester Eye Bank.

Patients and Methods: A retrospective analysis of laboratory and transplantation records of corneal donations from August 2004 to December 2005 was carried out. Results of HTLV initial reactivity and subsequent confirmation of test reactivity was compared with Eye Bank data on corneal tissue use.

Results: 1009 corneal donors were tested. 73 (7.2%) were initially reactive. Ethnic origin of all initially reactive samples was Caucasian (100%). 22 (2.1%) were negative on repeat testing. 51 (5%) were referred for confirmation. Of those referred, no samples confirmed positive. 7 (0.7%) samples were insufficient for testing. This equates to a potential donor loss of 160 corneas.

Conclusions: We observed unacceptable wastage of donor tissue on the basis of a screening test with low specificity and poor reproducibility, for a disease of low endemicity in the UK and low risk of subsequent development of disease if transmitted. We question whether mandatory HTLV testing should be suspended until an alternative screening test is available. We suggest that the decision to use corneal tissue should be considered in the light of clinical history and risk factors for HTLV disease.

P44 Greek measles epidemic strain, 2005–2006

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Aim: The purpose of this work was the molecular study of the virus strain that caused the last measles outbreak in Greece.

Methods: Twenty-four saliva specimens were obtained from selected patients serologically confirmed as measles cases between December 2005 and March 2006. Measles virus detection was performed by a nested RT-PCR. The 560 bp segment of the N gene of these MV strains was used for genotyping.

Results: The N gene sequences of the Greek MV strains were identical to each other, so a phylogenetic tree was constructed using one representative MV (ThesGRE/06).

Conclusion: Our data confirmed that the measles virus strain which caused the 2005–2006 outbreak in Greece belonged to genotype D6, had a highly homology with Mvi/Ankara.TUR/29.04 strain and differed by 5.5% from the Edmonston B vaccine strain.

P45 Evaluation of the bioMérieux easyMAG automated extraction system

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Background and Aims: The aim of the evaluation was to compare DNA and RNA quality that had been extracted using the bioMérieux easyMAG system with the automated Roche MagnaPure and manual Qiagen and Gentra methods. To assess the stability of viral RNA extracted using easyMAG.

Methods: The evaluation compared cycle/threshold (CT) or load values of 'real-time' PCR assays (HSV1/2, VZV, CMV, BK, HBV, Enterovirus, Adenovirus, Norovirus genotypes 1/2, Influenza A/B, Neisseria meningitidis) using samples and isolates extracted by

easyMAG plus one of Roche MagnaPure, Gentra or Qiagen methods. Samples included EDTA whole blood, plasma, serum, CSF, faeces, NPA, swabs (genital, throat and nose) and urine (n = 181). Stability of Enteroviral RNA was tested by storage at ambient temperature, +4°C, -20°C and -70°C and tested at intervals of zero, two, four and seven days.

Results: Samples extracted by easyMAG, when tested by 'real-time' PCR assays had a comparable level of sensitivity to MagnaPure, Qiagen or Gentra methods. There was no change in the sensitivity of the Enteroviral RT-PCR when tested using RNA extracted by easyMAG and stored at ambient temperature, +4°C, -20°C and -70°C and over the different time intervals.

Discussion and Conclusion: EasyMAG processes different sample types of different input and elution volumes with DNA and RNA extracted concurrently. DNA and RNA quality was comparable with automated and manual extraction methods. A disadvantage of easyMAG is that samples cannot be processed overnight. EasyMAG extracted RNA had excellent stability with no evidence of degradation when stored at ambient temperature for seven days.

P46 Improved laboratory diagnosis of HTLV in corneal transplant donor specimens

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Background: Human T-cell lymphotropic virus (HTLV) infections are reported worldwide, but with specific geographic foci. Maximum seroprevalence ranges from 3–6% in the Caribbean to 30% in rural parts of southern Japan. In the UK the infection is extremely rare. Screening of corneal donors for HTLV has been mandatory since 2004 in the UK, usually using the only CE marked EIA test available in the UK. Testing is normally carried out on cadaveric samples, and a high frequency of repeatedly-reactive results has been observed that have not been confirmed when specimens have been referred to a specialist laboratory.

Aims: In a recent audit of 1009 donors at the CTS-Manchester Eye Bank over a period from August 2004 to December 2005, none of 73 reactive samples were confirmed as HTLV positive. During this time up to 160 corneal transplants were lost as a result of these false positive tests. To determine whether modification of the EIA used for HTLV antibody screening, the use of a gel particle agglutination assay for HTLV antibodies and/or a line immunoassay could significantly reduce the number of reactive samples which need confirmation we conducted a study both using retrospective samples, and prospectively.

Results and Discussion: An improved algorithm using two tests dramatically reduces the number of corneas that have to be discarded thereby improving the service offered to the transplant centre.

P47 The first detection of bovine coronavirus in calves diarrhea in west of Iran

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Background: Bovine coronavirus (BCoV) is associated with severe diarrhea in newborn calves, neonatal calf disease (NCD), winter dysentery (WD) in adult cattle, and respiratory tract infections in calves and feedlot cattle. The BCoV was first recognized as a cause of potentially fatal diarrhea of neonatal calves in 1972. Economically important NCD and WD outbreaks were reported. There is not studying about importance of this virus in calf diarrhea in Iran. Therefore, a study was performed to determine the extent to which BCoV is present in calves with diarrhea from farms in west of Iran.

Method: A total of 108 fecal samples from diarrheic calves were collected and then RNA extracted by QIAamp virus RNA mini kit (Qiagen, UK) as instructed by the manufacture then BCoV RNA was detected by reverse transcription-PCR (RT-PCR) method.

Results: BCoV RNA was detected in 13 of the 108 diarrheic samples (12%) by RT-PCR targeting a 730 bp fragment of the