Joint Healthcare Infection Society and British Society for Antimicrobial Chemotherapy

SPRING MEETING 2012

Multi-drug resistant Gram negative infections

Tuesday 20 March
Royal College of Physicians, London

Programme & poster abstracts

CPD ACCREDITED
We are grateful for the support we have received from industry and wish to acknowledge the following companies.

Meeting sponsor:
Pfizer

Exhibitors:
AdvanDx
Astellas Pharma
Bard
bioMerieux
Bioquell
Eumedica
Forest Laboratories
Merck, Sharp & Dohme
Pall Medical
Pfizer Medical
Poster abstracts

Laboratory screening techniques and infection control procedures developed for patients transferred from African hospitals.
Aiken, ZA. and Taylor, G., Levi, K., Diggle, M., Snape, SE. Clinical Microbiology Department, Queen's Medical Centre Campus, Nottingham.

Analysis of Gram-negative bacteria collected during a longitudinal human antibiotic challenge study.

Identification of antibiotic resistance genes in the human microflora.
Card, R.1, Warburton, P2, Mullany, P.2 and Anjum, M.F.1 1 Animal Health and Veterinary Laboratories Agency, Weybridge, Surrey, UK. 2 UCL Eastman Dental Institute, University College London, London, UK

Davidson, J; Willocks, L; Eastaway, A and Henderson, D, Centre for Population Health Sciences, University of Edinburgh

Beta-lactamase negative amoxicillin resistant (BLNAR) Haemophilus influenzae in a paediatric hospital.
Drew, RJ; Barton, T; Gerrard, C. Department of Microbiology, Alder Hey Children's NHS Foundation Trust, Liverpool

A retrospective study of Pseudomonas aeruginosa isolates with reduced susceptibility to meropenem.
Dwyer, N, Hardiman, F. Department of Microbiology, Alder Hey Children's NHS Foundation Trust, UK

Extended-spectrum beta-lactamase (ESBL) producing Gram negative bacilli isolated form urines of patients in a Teaching Hospital Renal Unit – an epidemiological study.
FitzGerald R, Bukhari S, and Baines R. Department of Clinical Microbiology, Leicester Royal Infirmary and Department of Nephrology, Leicester General Hospital, University Hospitals of Leicester NHS Trust, Leicester, UK

Stokes test for direct sensitivity on urines: Was there a baby in the bathwater?
Gibb, AP, Bryce, D, St John's Hospital, Livingston, Scotland.

Antibiotic prescribing in the Acute Medical Unit of a UK teaching hospital: does competition between doctors improve prescribing practice?
Hopkins S, Grundy A., Barnardo A. Brighton and Sussex University Hospitals NHS Trust, UK

Routine antimicrobial surveillance across English regions using the AmSurv system.
Ironmonger, D; Bains, A; Edgheore, O; Reagan, M; Ward, S. Health Protection Agency

Assessing the efficacy of Pivmecillinam in the bacteriological urinary clearance of Extended Spectrum Beta Lactamase(ESBL) producing organisms.
Kiapi, G MBCHB MRCP MSC. Queen Elizabeth Hospital, Woolwich, South East London.

Detection of Plasmid-Mediated AmpC Resistance Using SYBR Green Real-Time PCR.
Lewis, JA., Lawrance, L., Moore, PC. and Arnold, DA. Department of Microbiology, Gloucestershire Royal Hospital, Gloucester

Are All Meropenem Prescriptions Justified?
Li, Mark; Banavathi K. Department of Microbiology, University Hospital of North Staffordshire NHS Trust, Stoke-on-Trent

An audit of Resistant Gram negative screening at a District General Hospital.
Peters JR, Legg JM. Microbiology Department, Worthing Hospital, Lyndhurst Road, West Sussex, BN11 2DH

Enhancement of Infection Prevention and Control Measures and Dynamic of Transmission During an Outbreak of Multi resistant Acinetobacter baumannii in a Critical Care Unit.

A prospective study of the incidence of resistant microbial flora associated with transrectal ultrasound guided prostate biopsy (TRPB).
Sheehan S, Nemadeh H, Khalifa E, Thompson P, Philpott-Howard J. King's College Hospital NHS Foundation Trust, King's College London School of Medicine

Predatory Bdellovibrio Invade and Kill Gram Negative Antibiotic-Resistant Bacteria:-Can This Be Translated Therapeutically?
Sockett RE1, Lambert C1, Aiken Z2, Diggle M2. 1School of Biology, University of Nottingham, Medical School and 2Clinical Microbiology, Nottingham University Hospitals NHS Trust, QMC, Derby Road Nottingham NG7 2UH UK.

Acinetobacter baumannii virulence is enhanced in Galleria mellonella following biofilm adaptation.
Wand M. E., Bock L. J., Turton J. F., Nugent P. G. and Sutton J. M. Health Protection Agency, Microbiology Services Division, Porton Down, Salisbury SP4 0JG, UK

Notes
## Programme

**0900** | REGISTRATION & COFFEE

**0955** | Welcome & introductory remarks  
*Professor Peter Hawkey, BSAC/HIS Working Party on MDR Gram Negative Infections*

### SESSION ONE

**Chair:** Dr Tim Boswell, Chairman, HIS

**1000** | Current epidemiology and new strains of multi-drug resistant Gram negative infections  
*Dr Neil Woodford, London*

**1030** | UK & European challenges in the definitions and laboratory diagnosis of multi-drug resistant Gram negative infections  
*Dr Christian Giske, Sweden*

**1100** | COFFEE & POSTER VIEWING

### SESSION TWO

**Chair:** Professor Laura Piddock, President, BSAC

**1130** | Multi-drug resistant Gram negative UTI infections: managing challenges and combination treatment in community settings  
*Dr Rod Warren, Shrewsbury*

**1200** | Multi-drug resistant infections - emerging strains: an international perspective  
*Professor Patrice Nordmann, France*

**1230** | BSAC AGM & LUNCH

### SESSION THREE: PARALLEL SESSIONS

**1345** | Managing outbreaks of multi-resistant Acinetobacter  
*Chair: Dr Peter Jenks, HIS*

Outbreaks in critical care and the and the implementation of effective control measures  
*Dr David Enoch, Peterborough*

High-throughput sequencing and infection control - what it means to you  
*Dr Tom Lewis, North Devon*

**1345** | Current treatment options - case based discussion  
*Chairs: Dr Nick Brown, BSAC, Dr Neil Woodford, London & Dr Robin Howe, Cardiff*

**Discussion topics:**  
- Treatment of *Stenotrophomonas maltophilia* infection; The increasing complexity of interpretation of susceptibility tests in Enterobacteriaceae with broad spectrum or combinations of beta-lactamases and the implications for treatment;  
- *Pseudomonas aeruginosa* infection, resistance and options for treatment

### SESSION FOUR

**Chair:** Dr Peter Wilson, BSAC/HIS Working Party on MDR Gram Negative Infections

**1430** | Parallel sessions feedback  
*Dr Peter Jenks, HIS & Dr Nick Brown, BSAC*

**1445** | National ARHAI guidance for the management of MDR Gram negative infections  
*Professor Peter Hawkey, Birmingham*

**1515** | Question time - which way forward now?  
*Discussion session led by Professor Peter Hawkey & Dr Peter Wilson*

**1600** | Closing remarks
Poster abstracts

Laboratory screening techniques and infection control procedures developed for patients transferred from African hospitals
Aiken, ZA. and Taylor, G., Levi, K., Diggle, M., Snape, SE. Clinical Microbiology Department, Queen's Medical Centre Campus, Nottingham.

Method
Four patients were transferred from healthcare facilities in Africa in October 2011. Following guidance from the Health Protection Agency and Centre for Disease Control, all patients were screened on arrival then weekly for carbapenem-resistant Enterobacteriaceae, multi-drug resistant Acinetobacter baumannii and methicillin-resistant Staphylococcus aureus. MacConkey plates with ertapenem discs were used for initial CRE screening with confirmation by the Modified Hodge test and a specially developed multiplex PCR. All isolates were subsequently sent to the Antibiotic Resistance Monitoring & Reference Laboratory (ARMRL).

Results
A number of multi-drug resistant organisms were isolated from this patient cohort including Klebsiella pneumoniae (OXA-48 positive), A. baumannii (OXA-23 positive) and a collection of other multi-drug resistant Enterobacteriaceae.

Patients were cohorted in the same bay. Three patients were bed bound and the fourth had a dedicated bathroom. Visitors were limited to 2 per patient and there was a dedicated nurse for this bay. Strict wound and skin precautions were adhered to. When 3 patients were discharged the remaining patient was placed in a side room and the bay cleaned with hypochlorite then hydrogen peroxide decontamination.

Each patient was last on the list for theatre. Post operatively, the theatre was cleaned with hypochlorite, ventilation switched off and hydrogen peroxide decontamination performed overnight.

There was no transfer of these organisms between patients in the bay, on the ward or in theatres.

Conclusion
Stringent infection control is needed to prevent cross contamination between patients. Guideline development and implementation is essential to ensure patients potentially harbouring multi-drug resistant organisms are screened using phenotypic and molecular methods and the results are fed back to appropriately guide management.

Analysis of Gram-negative bacteria collected during a longitudinal human antibiotic challenge study.

A longitudinal study was performed to investigate the emergence of antibiotic resistance in the human microbiota. Healthy volunteers were given doses of an antibiotic (Minocycline or Amoxicillin) or placebo. Faecal, skin and nasal samples were taken for up to 1 year after dosing. The isolates collected were subject to antimicrobial sensitivity testing and resistance gene analysis using a DNA-based miniaturised microarray capable of detecting over 70 AMR genes. The most common isolates recovered from faecal samples were Bacteroides spp. (Anaerobic) and Escherichia coli, Klebsiella spp. and Citrobacter spp (Aerobic). All isolates tested were Gram-negative with the exception of two aerobic isolates and in total twenty-three different bacterial spp. were recovered. To date 360 anaerobes and 706 aerobic isolates have been analysed with the AMR06 array and antibiograms performed. Analysis of the array data has found that 48.3% of the aerobic isolates were multi-resistant (carrying two or more genes). The most common genes detected among aerobic isolates tested thus far are: sul2, strB, blaTEM, catIII and tetB. Among the anaerobic bacteria 29.7% of the isolates had at least two AMR genes. Results have demonstrated that many bacterial spp present in the microbial population recovered from healthy individuals can carry a large variety and number of resistance genes; with several isolates carrying up to twenty genes including genes encoding Extended Spectrum Beta-Lactamases and AmpC-type enzymes.
Identification of antibiotic resistance genes in the human microflora.
Card, R.1, Warburton, P.2, Mullany, P.2 and Anjum, M.F.1
1 Animal Health and Veterinary Laboratories Agency, Weybridge, Surrey, UK
2 UCL Eastman Dental Institute, University College London, London, UK

The bacteria present in the human microflora can play an important role both in maintenance of health and in development of disease. They also serve as a reservoir for harbouring and exchanging genes of clinical relevance, such as those conferring resistance to antimicrobials. The aim of this study was to investigate the presence of antimicrobial resistance (AMR) genes present within the oral microflora of healthy adult volunteers from different European countries. Two culture-independent metagenomic approaches were employed, to obviate difficulties associated with the culture of many microfloral bacteria. In the first approach, a miniaturised microarray capable of detecting over 70 AMR genes was used. At least one gene was detected in each sample and in total 15 different AMR genes for five antibiotic classes were detected (beta-lactams, erythromycin, tetracycline, sulphonamides and aminoglycosides). The presence of selected AMR genes was confirmed using PCR and sequencing. For the second method, oral metagenomic DNA was cloned into a heterologous host and genes conferring resistance to ampicillin or sulphonamides selected by antimicrobial activity-based screening. Clones with resistance or reduced susceptibility to the antibiotics screened were recovered and inserts sequenced. All inserts originated from opportunistic bacterial pathogens. The ampicillin screen recovered BAC clones from Haemophilus parainfluenzae containing the acrRAB operon, encoding components of a multi-drug efflux pump. For sulphonamide, all clones contained the folP gene which encodes dihydropteroate synthase (the target of sulphonamide inhibition) and originated from Neisseria meningitidis, Veillonella parvula, or Streptococcus pneumoniae. The microbial community profile of the oral samples from each country was determined by 454 pyrosequencing of barcoded 16S rRNA gene amplicons. The genera identified in the activity-based screening were all represented in the oral profiles obtained. This study has enhanced our knowledge of the diversity of the microflora of healthy subjects, and described the presence of AMR genes from potentially pathogenic bacteria, which may interfere with therapeutic options following infection.

Davidson, J; Willocks, L; Eastaway, A and Henderson, D, Centre for Population Health Sciences, University of Edinburgh

Objective: To investigate trends in the incidence of Acinetobacter baumannii, Citrobacter freundii, Pseudomonas aeruginosa, Enterobacter cloacae and Serratia marcescens bacteraemia and their sensitivity to selected antibiotics; ceftazidime, ciprofloxacin, gentamicin and carbapenems recorded in the Scottish population between 2006 and 2010 calendar years.

Methods: Data on A.baumannii, C.freundii, P.aeruginosa, E.cloacae and S.marcescens bacteraemia reported from laboratories in Scotland to Health Protection Scotland (HPS) through the Electronic Communication of Surveillance in Scotland (ECOSS) between 2006 and 2010 were included in this epidemiological and statistical analysis of bacteraemia incidence and antibiotic sensitivity nationally and in NHS health boards.

Results: A total of 2705 episodes of bacteraemia caused by the selected organisms were reported to HPS between 2006 and 2010 calendar years. Significant time trends were found when Poisson regression was applied to the data, with an increase in rate of bacteraemia identified between 2006 and 2010 calendar years (IRR 2.239, 95% CI 1.646-3.045). As well as between yearly quarters (IRR 1.567, 95% CI 1.129-2.175) when quarter one, corresponding to the months of January to March, was compared to quarter four, October to December. Resistance was detected to all antibiotics tested, with yearly fluctuations in the percentage of resistant isolates but analysis using chi squared test for trends revealed no trends (ceftazidime p=0.4, gentamicin p=0.7 and ciprofloxacin p=0.1).

Conclusion: There has been an increase in Gram–negative bacteraemia reported within Scotland in recent years, a trend which reflects the global picture. Resistance does not appear to have increased, although the extent of resistance to ceftazidime is alarming, with gentamicin and ciprofloxacin resistance also prevalence.
Beta-lactamase negative amoxicillin resistant (BLNAR) Haemophilus influenzae in a paediatric hospital.
Drew, R; Barton, T; Gerrard, C. Department of Microbiology, Alder Hey Children’s NHS Foundation Trust, Liverpool

Aim: To determine the incidence of beta-lactamase negative amoxicillin resistant (BLNAR) Haemophilus influenzae in respiratory samples between January 2011 and December 2011. BLNAR Haemophilus influenzae are clinically significant as they may have reduced cephalosporin susceptibility.

Methods: All Haemophilus spp. isolates from respiratory samples were identified using the Laboratory Information System. Respiratory samples included bronchoalveolar lavages, sputa, cough swabs, as well as ear, nose and throat swabs. One sample per patient was included each day and repeat isolates were removed. Susceptibility testing was performed using the BSAC method.

Results: 345 samples were identified, 325 (94%) were H.influenzae and 20 (6%) were H. parainfluenzae. Of the 325 H. influenzae isolates, 257 (79%) were amoxicillin and co-amoxiclav susceptible, 59 (18%) were amoxicillin resistant and co-amoxiclav sensitive (beta-lactamase positive) and nine (3%) were amoxicillin and co-amoxiclav resistant. Of these nine isolates, six (2% overall) were BLNAR, 2 were beta-lactamase positive and co-amoxiclav resistant (BLPACR) and one was unknown. Cefotaxime and meropenem MICs were performed on three BLNAR H. influenzae isolates and five fully sensitive H.influenzae isolates. The median MIC of the BLNAR group (median MIC=0.094) was higher than that of the isolates that were amoxicillin sensitive (median MIC=0.023). There was no notable difference in the meropenem MIC between the two groups.

Conclusion: Only 2% of all H. influenzae isolates were BLNAR, but these isolates had a higher cefotaxime MIC than fully susceptible H. influenzae isolates. This emphasises the need to perform cefotaxime MICs when treating patients with infections due to BLNAR H.influenzae.

A retrospective study of Pseudomonas aeruginosa isolates with reduced susceptibility to meropenem.
Dwyer. N, Hardiman. F. Department of Microbiology, Alder Hey Children’s NHS Foundation Trust, UK

This study was undertaken at Alder Hey Children’s hospital over a seven month period to determine the incidence and mechanism of meropenem resistance in isolates of Pseudomonas aeruginosa from respiratory samples in non-cystic fibrosis children.

From July 2011 to February 2012, our department received 1680 requests for culture on paediatric respiratory specimens. These samples included sputum, cough swabs, endo-tracheal aspirates, broncho-alveolar lavages and naso-pharangeal aspirates. Of this total, 646 were from children with cystic fibrosis, these samples were removed from the study to avoid skewing of results, leaving a total number of 1034.

266 (26%) of the 1034 specimens grew Paeruginosa within this time frame and following removal of duplicates (one sample only per patient was included). Repeat isolates were disregarded) we were left with 77 isolates. Antibiotic susceptibilities were performed according to BSAC guidelines. Of the 77 Paeruginosa isolates, 8 (10%) were found to be either resistant or intermediate to meropenem. We then performed meropenem MICs and in house testing for metallo-beta lactamase (MBL) activity.

4 of our isolates (50%) tested positive for MBL and were referred to the HPA for confirmatory testing. All of the referred isolates were subsequently found to be negative for MBL. The reduced carbapenem susceptibility was consistent with OprD porin loss with or without efflux.

In conclusion, the incidence of meropenem resistant P.aeruginosa from non-CF respiratory samples is low within Alder Hey, and the isolation of an MBL positive strain has yet to occur.
Extended-spectrum beta-lactamase (ESBL) producing Gram negative bacilli isolated form urines of patients in a Teaching Hospital Renal Unit – an epidemiological study
Rosemarie FitzGerald, Sayed S Bukhari, and Richard Baines.
Department of Clinical Microbiology, Leicester Royal Infirmary and Department of Nephrology, Leicester General Hospital, University Hospitals of Leicester NHS Trust, Leicester, UK

Introduction and objectives:
ESBL-producing Gram negative bacilli (GNBs) are emerging as an increasing challenge in the treatment of urinary tract infections (UTIs) of renal patients, as they are becoming more prevalent and increasingly resistant to multiple classes of antibiotics, including beta-lactams, fluoroquinolones and aminoglycosides. The emergence of resistance to carbapenems is of particular concern, as this will further limit the number of therapeutic options for these pathogens in a time when there is already a lack of antimicrobials for Gram negative organisms coming to the market.

With this background, we studied the epidemiology of ESBL-producing GNBs isolated from urines of patients attending our renal service, to evaluate the current epidemiology and gain insight into what may be expected in the future.

Methods:
A data-based search was carried out on the microbiology computer system (iLab) which included all urines sent from the renal unit to the microbiology laboratory during the period from July 2010 to September 2011 which grew ESBL-producing GNBs. Patients who had documented bacteraemia with an ESBL-producing GNB isolate with similar antibiogram as the urinary isolate were also included.

The data was recorded onto an Excel spreadsheet which included GNB isolates and antimicrobial susceptibility tests conducted according to the British Society for Antimicrobial Chemotherapy (BSAC) standard methods for beta-lactams including carbapenems, fluoroquinolones and aminoglycosides.

Results:
There were a total of 157 ESBL-producing GNB isolated from urines and six from blood cultures. Of the urinary isolates 128 (82%) were resistant to ciprofloxacin, 12 (8%) to ertapenem, 52 (33%) to gentamicin, 37 (24%) to nitrofurantoin, 141 (90%) to trimethoprim and 3 (2%) to meropenem/imipenem.

OF the blood culture isolates 5 of 6 (83%) were resistant to ciprofloxacin and gentamicin, 6 of 6 (100%) were resistant to trimethoprim and none were resistant to meropenem, imipenem or ertapenem.

Conclusion:
These results indicate that ESBL producing isolates carry mechanisms that render them resistant to multiple classes of common used antibiotics, thus limiting therapeutic options for clinical infections. It was reassuring that 100% of our blood isolates and 98% of our urinary isolates were susceptible to Meropenem/imipenem.

Stokes test for direct sensitivity on urines: Was there a baby in the bathwater?
Gibb, AP, Bryce, D, St John’s Hospital, Livingston, Scotland.

Aim: To compare Stokes test directly on urines with Vitek2 sensitivity on isolated bacteria.

Method: Urines were examined by wet-film microscopy. Stokes sensitivity test was set up on samples judged to be positive by microscopy, with 6 disks (amoxicillin, co-amoxiclav, cefalexin, ciprofloxacin, trimethoprim, nitrofurantoin). Stokes test is a method where zone sizes are compared to those obtained with a control E. coli plated circumferentially on the same plate. Zones >= 3mm radius smaller are scored as resistant. Conventional culture and sensitivity by Vitek2, using CLSI criteria, was run in parallel.

Results: 77 isolates were compared. There was 100% agreement in results for trimethoprim, ciprofloxacin, cefalexin, and amoxicillin. With nitrofurantion there were 5 discrepancies where Stokes was sensitive by Vitek2 was intermediate. In these 5 cases the MIC was 64, which would be sensitive by EUCAST criteria. With co-amoxiclav there were 9 discrepancies: 8 were sensitive by Stokes but intermediate by Vitek2, (MICs of 8 in seven, 16 in one). One isolate was co-amoxiclav sensitive by Stokes but resistant by Vitek2. All the isolates resistant to 3rd generation cephalosporins by Vitek2 were cefalexin resistant by Stokes. Stokes results would typically be available a day before Vitek2.

Conclusions. There was a single “very major” error with co-amoxiclav out of 462 comparisons. Stokes test direct on urine may be a reasonable strategy to provide sensitivity results on low-risk urine samples, if a protocol is put in place to refer more resistant organisms for formal testing. We would refer for formal testing if Stokes indicated resistance to cefalexin, or if there was sensitivity to only one class of antibiotic. Formal testing would be required for all patients likely to have upper tract infection or to require intravenous treatment.
Antibiotic prescribing in the Acute Medical Unit of a UK teaching hospital: does competition between doctors improve prescribing practice?
Hopkins S, Grundy A, Barnardo A. Brighton and Sussex University Hospitals NHS Trust, UK

Background: Poor antibiotic prescribing is detrimental for many reasons: increased mortality, unnecessary drug costs and toxicity, and selection of multiresistant pathogens.

To improve antibiotic prescribing, the HPA recommends development of local trust antibiotic guidelines. Compliance with these remains a national problem. Strategies to increase compliance include physician education, audit and feedback, and healthcare system changes e.g. electronic prescribing. However, interventions can be ineffective and costly. A novel approach to improving prescribing is to use competition as an incentive.

Objectives: To audit compliance with trust antibiotic guidelines in a UK teaching hospital. To improve antibiotic prescribing through competition.

Methods: Drug charts were audited until 50 antibiotic prescriptions had been evaluated for compliance with trust empirical antibiotic guidelines and documentation of antibiotic indication and duration.

The intervention was then instigated: a competition between medical teams and grades of doctor. The competition was announced at teaching sessions and the Grand Round, on posters and screen-savers, and by daily reminders to the on-call team. 10 days post-intervention, prescribing was re-audited by the same method.

Results: Pre-intervention, 124 drug charts were reviewed: 40% of patients were on antibiotics, 80% complied with trust empirical guidelines, 52% had a documented indication, and 46% had a documented duration.

After the intervention, a further 138 records were reviewed. 36% of patients were on antibiotics. Of these, 84% complied with trust empirical guidelines, 58% had a documented indication, and 46% had a documented duration. These changes were not statistically significant.

Conclusion: This audit shows that antibiotic prescribing in this teaching hospital remains imperfect despite years of national & local campaigns. Using a competitive incentive is inexpensive and novel but in this small study no significant evidence of improvement was found. If physicians were rewarded individually or the incentive was greater, the intervention may have had an effect. “Carrot & stick” strategies, e.g. competition, may have a role as one component of a multifaceted approach, however in isolation the competition “carrot” per se does not seem to work.

Routine antimicrobial surveillance across English regions using the AmSurv system
Ironmonger, D; Bains, A; Edeghere, O; Reagan, M; Ward, S. Health Protection Agency

Summary
We describe the development, current uses and future of the HPA’s antimicrobial surveillance system (AmSurv) across the English regions and a web-based portal (AmWeb) for user generated outputs in operation in the West Midlands.

Introduction
The national roll-out of the HPAs Antimicrobial Surveillance System (AmSurv) commenced in October 2009 with the aim of monitoring trends and detecting the emergence of new antimicrobial resistance (AMR) across England using information obtained from the network of NHS Trust and HPA laboratories.

We aim to describe the current structure of AmSurv, including a web-based reporting tool (AmWeb); uses of AmSurv in public health surveillance activities from a regional perspective; and future plans and developments.

Methodology
AmSurv is a passive reporting system that collects and collates antimicrobial susceptibility reports from all bacterial isolates grown from a range of human specimens collected by hospital laboratories in all HPA regions. Patient demographic information is included to support the characterisation of the affected population.

Current status and outputs
- AmSurv currently collects 75% of AMR reports in England.
- AmWeb provides graphical (Drug/Bug) and tabular reports of organisms vs. antibiotics
- AmSurv data has been used to support the investigation of two outbreaks of Carbapenemase producing organisms in the West Midlands

Discussion
The monitoring of antimicrobial resistance trends is an important component of the UK strategy for preventing and controlling infectious diseases in the population. Detailed knowledge and understanding of endemic and emerging antimicrobial resistance patterns is needed to support the development of evidence-based interventions and plans as well as monitoring the effectiveness of control measures.
Assessing the efficacy of Pivmecillinam in the bacteriological urinary clearance of Extended Spectrum Beta Lactamase(ESBL) producing organisms

Kiapi, G MBCHB MRCP MSC. Queen Elizabeth Hospital, Woolwich, South East London.

Introduction:
A major challenge today is the treatment of urinary tract infections caused (ESBL)coliforms in the community. Pivmecillinam is a semi synthetic amino-penicillin derivative licensed for the treatment of uncomplicated urinary tract infections in the UK. Pivmecillinam is a pro drug of mecillinam which is administered orally.

Mecillinam has been previously shown to have in vitro activity against a range of E.coli expressing beta lactamase activity, some with the production of multiple beta lactamases. However, little evidence exists for the bacteriological clearance of urinary ESBL coliforms following therapy with mecillinam

Methods
ALL urinary coliforms resistant to ceftazidime were tested for evidence of (ESBL) production. A sample of 53 urinary ESBL E.coli and Klebsiella sp were checked for mecillinam sensitivity using the 10ug mecillinam disc by British Society of Antimicrobial Chemotherapy (BSAC) method.
Oral Pivmecillinam was then recommended for the treatment of urinary tract infections in the community. Inclusion and exclusion criteria were applied. Clearance urine samples from patients who had completed a recommended course of pivmecillinam were then checked for reduction in urinary white cells, organism count and evidence of bacteriological clearance of ESBL coliforms.

Results
Ninety eight percent of Ecoli ESBL isolates showed sensitivity to mecillinam. Eighty two percent of Klebsiella sp ESBL isolates showed sensitivity to mecillinam. Urinary clearance was demonstrated in eighty percent of patients treated with mecillinam.

Conclusion
Mecillinam may be an effective antibiotic for the treatment of ESBL Ecoli and Klebsiella Urinary tract infections in the community averting the need for inpatient treatment.

Detection of Plasmid-Mediated AmpC Resistance Using SYBR Green Real-Time PCR

Lewis, JA., Lawrance, L., Moore, PC. and Arnold, DA. Department of Microbiology, Gloucestershire Royal Hospital, Gloucester, GL1 3NN

Objective
The objective of the study was to develop a simple, rapid real-time PCR assay for the detection of plasmid-mediated AmpC resistance.

Methods
Control strains for AmpC plasmids were obtained from ARMRL (HPA, Colindale). The six control strains were representative of the plasmid AmpC groups (CIT, ACC, DHA, FOX, MOX and ENT/EBC).

The assay was optimised for use on a Smart Cycler (Cepheid), using the Quantifast SYBR Green master mix kit (Qiagen). Each reaction comprised of 12.5ul mastermix, 2.0ul of DNA template and 0.1uM of each primer; made up to a final volume of 25ul with water. A negative control was included in each run, using water in place of the DNA template.

DNA templates were prepared using a mechanical lysis method from overnight cultures, and cycling parameters were set according to the mastermix manufacturer’s recommendations. Positive reactions were determined as runs with a Ct value of <30.

Melting curve analysis was completed at the end of each amplification. The use of melting curve analysis enabled the differentiation of target products from any non-specific amplification resulting from the use of the SYBR Green dye.

All PCR products were sequenced using the forward amplification primer (Eurofins MWG Operon). Sequence data were compared with the relevant GenBank entries for each control strain.

Results
The assay run time on the Smart Cycler was approximately 44 minutes. The preparation stage took approximately 5 minutes for each isolate, giving an analysis time per isolate of less than 50 minutes.

All six control strains demonstrated amplification of a specific product. The melting point temperatures between runs were consistent (within 0.5oC) for each strain. All sequenced amplification products demonstrated excellent homology when analysed using ClustalW.

Conclusions
The data obtained demonstrated that plasmid-mediated AmpC resistance can be detected in a range of organisms using this assay. The assay is quick and easy to introduce in a routine clinical laboratory.
Are All Meropenem Prescriptions Justified?
Li, Mark; Banavathi K. Department of Microbiology, University Hospital of North Staffordshire NHS Trust, Stoke-on-Trent ST4 7PX

The increase in carbapenem resistant gram-negative isolates and the emergence and spread of carbapenemases in UK hospitals in the recent years is a cause of concern. The use of meropenem in our hospital was audited during the first 3 weeks of November 2011 against indications in the Trust antimicrobial guidelines to inform ongoing antimicrobial stewardship programme. The list of patients prescribed meropenem was obtained from pharmacy or from the wards that stocked meropenem. Patient notes and prescription charts were reviewed. Seventy-one patients were prescribed meropenem during this period of which notes for 58 patients could be reviewed. The median age was 62.5 yrs (range 0 to 100 yrs). The median duration of treatment was 7.5 days (range 1-45 days). Most meropenem use was in medical patients (62%). Highest numbers of patients on meropenem were in renal, endocrine, respiratory and haematology wards. The highest proportion of meropenem use was for respiratory infections (27%) followed by sepsis (21%). Only 21% of patients on meropenem had a current infection with multi-drug resistant gram-negative organism including ESBL and AmpC producers. Penicillin allergy was the indication in 12%, while in 17% meropenem was started as an escalation of treatment. Forty-eight percent of meropenem prescriptions were not according to our Trust antimicrobial policy. Of these 96% of the cases were not discussed with microbiology or infectious diseases. Several recommendations including adherence to DoH Secondary Care Prescriber’s Checklist, development of criteria for escalation of antibiotic treatment to meropenem and avoiding meropenem use in penicillin allergy have been proposed.

An audit of Resistant Gram negative screening at a District General Hospital.
Peters JR, Legg JM. Microbiology Department, Worthing Hospital, Lyndhurst Road, West Sussex, BN11 2DH

Introduction
All patients admitted to the 12 bedded intensive care department or 4 bedded enhanced surgical unit at our hospital are screened for resistant Gram negative organisms on day of admission and at weekly intervals thereafter. This practice was commenced following an outbreak of multi-resistant Klebsiella in the ITU a few years ago. A swab is taken from up to 3 sites and cultured on ISO-Sensitest media. Six antibiotic discs are then applied to detect resistance. Resistance to Cefpodoxime, Ciprofloxacin, Gentamicin or Augmentin is treated as potentially significant and formal identification of the isolate and further sensitivities are performed.

Objectives
The aim of this study was two fold. Firstly, to prospectively collect data on the organisms captured by this practice and record any subsequent clinical specimens positive for this organism. Secondly, to audit whether screening is occurring appropriately in this patient group.

Methods
All Gram negative screens were accessed from the microbiology computer system over a period of six weeks. Results of the initial and subsequent screening swabs were recorded. Of those positive for a resistant organism, the microbiology records of the patient were searched and any clinical specimen which cultured the same organism in the patient episode was recorded.

Results
In total 133 patients with gram negative screens were reviewed. 18 patients had a positive Gram negative screen and 19 organisms were identified. Serratia marcescens, Enterobacter cloacae, ESBL producing E.coli and Klebsiella pneumoniae were most frequently isolated. The multi-resistant Klebsiella that initiated screening practice at our hospital was not isolated in this study period. 4 of the 18 patients had definite infection with the resistant organism and 4 had possible infection. Of those with definite or possible infections, 25% had a positive screening swab before the clinical specimen was positive and 75% of screening swabs were positive after or at the same time the clinical specimen was positive. Of the patients who were screened, 82% had appropriate swabs taken and repeated at the correct interval. In the remaining patients, the majority had too many swabs taken or too frequently during their admission.

Conclusions
A relatively low percentage (14%) of our patient group had a positive screening swab. Of the patients with a positive screen and infective episode with this organism, screening may have aided clinical decision making in just 25% of cases. This study does not assess the impact of this screening practice on infection control measures however. Theaudit showed that a reasonable percentage of people were appropriately screened but there may be potential cost savings if this is improved. In the era of increasing multi-resistant Gram negative organisms, it may be of value to reaudit this patient group in the future to see whether the nature and frequency of isolated multi-resistant organisms will change.
A prospective study of the incidence of resistant microbial flora associated with transrectal ultrasound guided prostate biopsy (TRPB)
Sheehan S, Nemade H, Khalifa E, Thompson P, Philpott-Howard J.
King’s College Hospital NHS Foundation Trust, King’s College London School of Medicine

Objectives
Optimum prophylaxis for TRPB remains a challenge. Ciprofloxacin is the most commonly prescribed antibiotic worldwide; however in the setting of the increasing incidence of multi-drug resistant Gram negative infections, such prophylaxis may no longer be optimal. In this prospective study, the organisms to which patients are exposed at the time of the procedure and their sensitivities are reviewed.

Methods
67 patients undergoing TRPB were enrolled. All patients received ciprofloxacin and metronidazole prophylaxis. Pre and post biopsy mid-stream urine samples and blood cultures at 5 mins, 60 mins and 24 hours post biopsy were collected for culture. Washings from the biopsy needle were also submitted for culture.

Results

<table>
<thead>
<tr>
<th>Site</th>
<th>Gram positive</th>
<th>Gram negative</th>
<th>Anaerobes</th>
<th>Yeast</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prostate needle washings*</td>
<td>31/67 (76%)</td>
<td>23/67 (34%)</td>
<td>11/67 (16%)</td>
<td>1/67 (1.5%)</td>
</tr>
<tr>
<td>Blood cultures*</td>
<td>6/67 (9%)</td>
<td>0**</td>
<td>1/67 (1.5%)</td>
<td>0</td>
</tr>
<tr>
<td>Post-TRPB urinanalysis</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>63%</td>
<td>20%</td>
<td>0%</td>
<td>-</td>
</tr>
<tr>
<td>Metronidazole</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

* Prostate needle washings and blood cultures had one or more microbial species.
** During the study period, two patients (who declined to enter the study) had Gram negative septicaemia post TRPB, one of whom had an extended spectrum beta lactamase producing E.coli that was also ciprofloxacin resistant in blood cultures.

61 patients had positive cultures from the washings of the biopsy needle.

The biopsy needle washings grew:
- Gram positive bacteria: 16 E. faecalis, 17 streptococci including 4 Group B, 1 Group G and 1 S.milleri group. 18 had other genera of Gram positive bacteria.
- Gram negative bacteria: 18 E.coli, 4 other Enterobacteriaceae, and 1 Acinetobacter spp.

Continued...
**Predatory Bdellovibrio Invade and Kill Gram Negative Antibiotic-Resistant Bacteria: Can This Be Translated Therapeutically?**

Sockett RE1, Lambert C1, Aiken Z2, Diggle M2
1School of Biology, University of Nottingham, Medical School and 2Clinical Microbiology, Nottingham University Hospitals NHS Trust, QMC, Derby Road Nottingham NG7 2UH UK.

Bdellovibrio bacteriovorus are tiny Gram negative predatory bacteria that naturally occur in soils, water, and can be isolated from human and animal saliva and faeces. Bdellovibrio invade the periplasm of other larger Gram negative bacteria that have a lipo-polysaccharide outer membrane. They kill the prey and digest their contents from within; finally lysing the original prey outer membrane to escape and seek new prey. Unlike bacteriophages Bdellovibrio have a very broad prey range and no specific receptors for prey entry. We have used timelapse microscopy and enumeration to study the invasion of a range of human clinical isolates of Klebsiella and Acinetobacter strains in vitro, carrying OXA48 and other antibiotic resistance markers. We find that these strains are rapidly invaded and killed by the Bdellovibrio. We have also studied the effects of Bdellovibrio on the wellbeing of live animals when ingested, finding that it causes beneficial, but short-lived effects upon gut inflammation and Salmonella colonization, importantly without negative effects on wellbeing. Thus Bdellovibrio could be tested as topical agents for wound infections, without fears about consequences of accidental ingestion. However many questions remain about the idea of using live bacteria as a potential medicine – these issues are discussed on the poster. Bdellovibrio should not be overlooked in the war against antibiotic-resistant pathogens; as they have co-evolved as predators of such pathogens, without themselves having any pathogenic activity. Their genome encodes a wide range of anti-Gram-negative enzymes with a broad antibacterial activity. We are widening their testing against problematic antibiotic-resistant strains and welcome dialogue with the conventional chemotherapeutic community.

**Acinetobacter baumannii virulence is enhanced in Galleria mellonella following biofilm adaptation**

Wand M. E., Bock L. J., Turton J. F., Nugent P. G. and Sutton J. M.
Health Protection Agency, Microbiology Services Division, Porton Down, Salisbury SP4 0JG, UK

The opportunistic nosocomial pathogen A. baumannii is responsible for a growing number of infections; however, few of its potential virulence factors have been identified, and how this organism causes infection remains largely unknown. Bacterial biofilms are often an important component in infection and persistence but there is no conclusive evidence to link biofilm formation with virulence and severity of infection in A. baumannii. To investigate this link, several clinical isolates were assessed in biofilm culture models and were tested for virulence in the insect model Galleria mellonella. In both systems, the profiles showed significant differences between strains, but no correlation was observed between virulence and the ability to form biofilms. In contrast, A. baumannii cells from a biofilms produced higher mortality rates than an equivalent numbers of planktonic cells. Relative to planktonic cells, A. baumannii biofilm cultures also showed reduced sensitivity to antibiotics normally used in the treatment of A. baumannii, especially colistin. This model, therefore, provides a suitable system to investigate the link between biofilm growth and various factors influencing virulence during A. baumannii infection.

**Continued...**

The blood cultures grew:
- 2 E. faecalis, 1 Bifidobacterium spp, 2 Corynebacterium spp, 1 Lactobacillus spp, 1 coagulase negative staphylococcus, 1 Propionibacterium spp.

**Conclusions**
This study showed a high rate of ciprofloxacin resistance among bacteria associated with TRPB. Post biopsy sepsis is a well-recognised complication and the rising incidence of multi-drug resistant Gram negative organisms presents a significant challenge for antimicrobial prophylaxis. Alternative approaches such as transperineal biopsy will avoid transrectal contamination with resistant faecal flora.