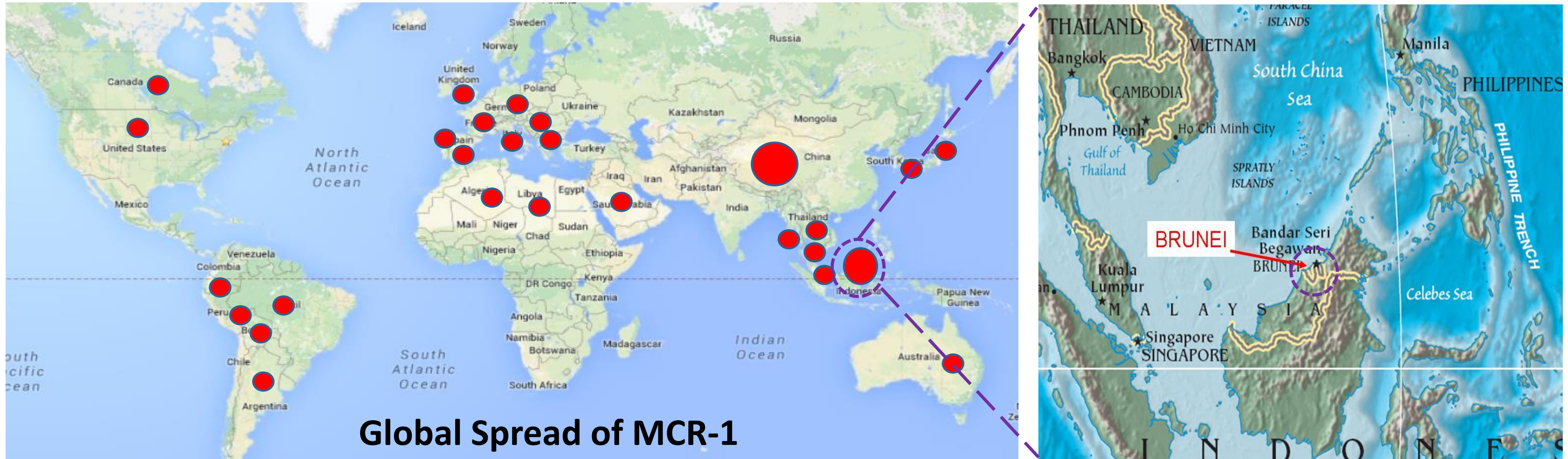


INTRODUCTION

Colistin resistance in Enterobacteriaceae, due to plasmid mediated phosphoethanolamine transferases (MCR-1-5)^{1,3-5}, was identified in late 2015. Most reports are of carriage in healthy food-producing animals (chickens, pigs, calves), particularly in South East Asia, but there is limited data on the prevalence in humans. Acquisition of the *mcr-1* gene by pathogenic multidrug-resistant (MDR) Gram-negative bacteria could seriously compromise treatment. Here, we report a high rate of MCR-1 producing *E. coli* in human faecal samples as part of a period prevalence study in Brunei Darussalam.



AIMS

To determine the prevalence of plasmid mediated colistin resistant MCR-1 *E. coli* in clinical human faecal samples in Brunei Darussalam.

MATERIALS AND METHODS

Sample collection

- A period prevalence surveillance study using stool samples from RIPAS Hospital microbiology laboratory was undertaken from July to August 2017.
- Stool samples (n=124) submitted for routine diagnostic analysis from various locations (general wards, outpatient clinics, ICU) were included.

Screening for Polymyxin Resistance Isolates

- Samples were enriched with colistin (5 mg/L) in Trypticase Soy Broth (TSB) and incubated for 24 h at 37°C without shaking. Turbid cultures were plated directly onto CHROMagar COL-APSE (Figure 1).

Bacterial Identification and Antibiotic Susceptibility Testing

- Identification of red/pink colonies were confirmed by MALDI-TOF MS (Bruker, Coventry, UK).
- Susceptibility testing was performed by disc diffusion and included antibiotics used only in veterinary medicine and MICs by Etest. Resistance was interpreted according to currently available breakpoints proposed in EUCAST, CLSI, and VET-EUCAST guidelines.

Detection of MCR-like genes Clonality and Phylotypes

- PCR amplification was performed using primers targeting *mcr* genes (1-5) as reported previously.²⁻⁵
- Clonal relatedness was determined by Random Amplified Polymorphic DNA PCR (RAPD-PCR).
- Phylotypes A, B1, B2 and D were determined by multiplex PCR.

RESULTS

- Colistin-resistant *E. coli* were recovered from 61 out of 124 (**49.2%**) of the stool samples.
- All of the 61 colistin-resistant *E. coli* isolates were positive for *mcr-1* gene. The MICs for Polymyxins were in the range of 3-32 mg/L .
- RAPD analysis (**Figure 2**) revealed considerable diversity, with 26 indistinguishable profiles.
- Co-resistance** was highest with **quinolones (pefloxacin, 85%), phenicols (florfenicol, 85%, chloramphenicol, 87%), β-lactams (amoxicillin/clavulanic, 80%)** antibiotics (**Figure 3**).
- Phylotyping identified that **50 %** (13/26) belonged to **phylogroup D** and only **1** isolate belonged to **phylotype B2**.

CONCLUSION

- This is the **first surveillance study** of the carriage of **plasmid-mediated colistin resistant *E. coli*** in clinical samples in **Brunei Darussalam**.
- A previous study highlighted endemic rates (100%) of MCR-1 in healthy chickens sampled on Brunei poultry farms.⁶
- The high rate of carriage in humans suggests that MCR-1 may be silently disseminating in the community.
- Comparative genomic analysis of human and poultry isolates is underway to determine links between the human and animal strains. This is critical to inform on any interventions aimed at tackling and controlling further spread of MDR bacteria and route cause of the problem.

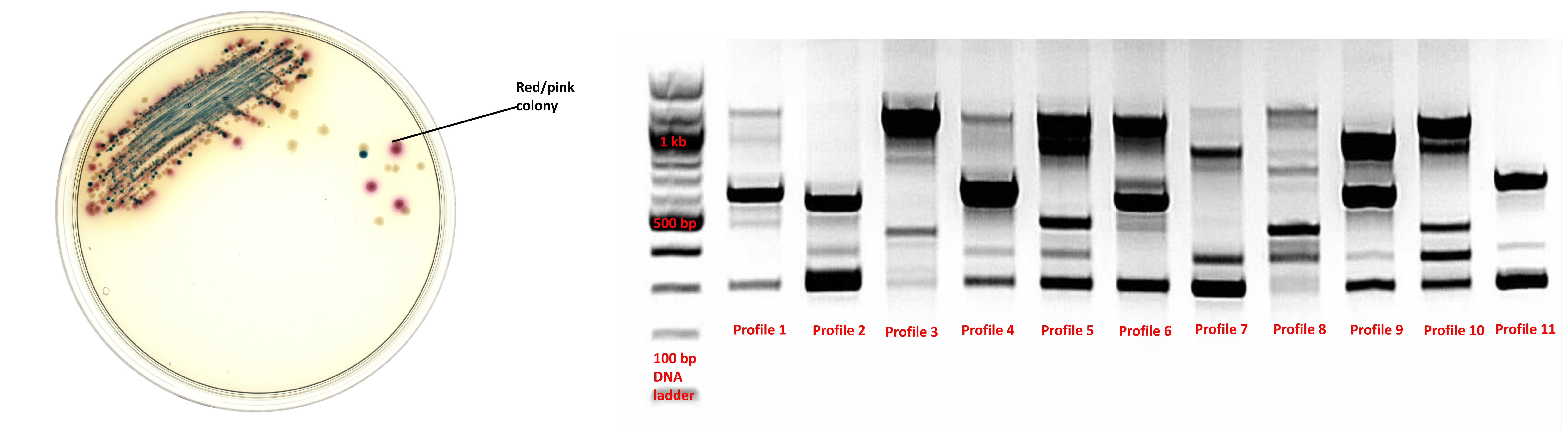


Figure 1: Growth of red/pink colonies on CHROMagar COL-APSE

Figure 2: RAPD profiles of MCR-1 *E. coli* isolates

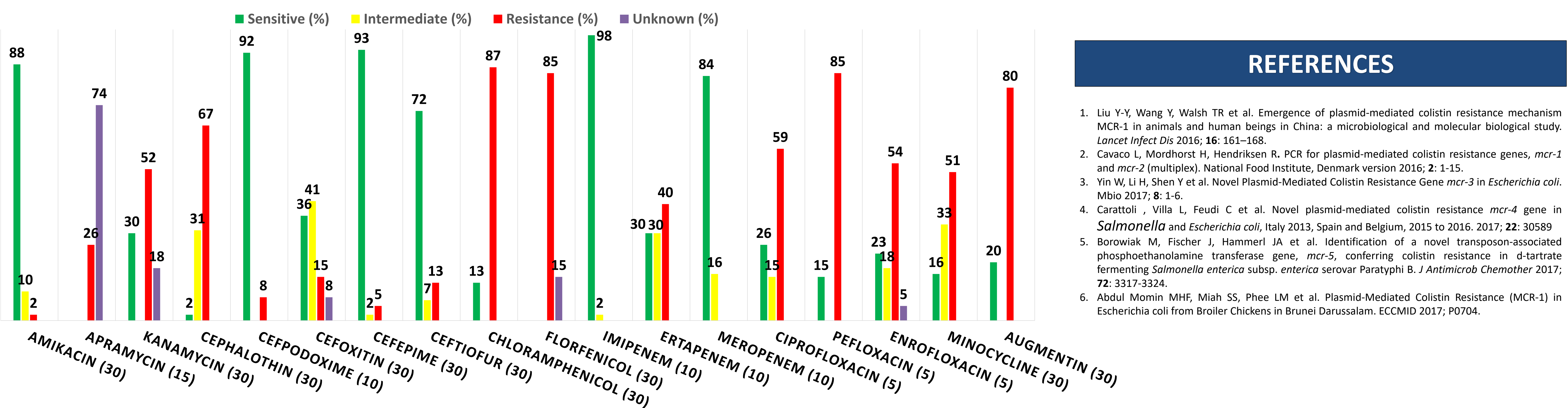


Figure 3: Percentage of co-resistance in MCR-1 *E. coli* isolates

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