

PP-247 Cloning and expression of *msp2* protein of *Anaplasma phagocytophilum*

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Background: *Anaplasma phagocytophilum* is an obligatory intracellular bacterium that causes human granulocytic anaplasmosis (HGA), which is an emerging tick-borne disease of public health concern in many areas of the world, and the first case of nosocomial human-to-human transmission was reported in China, 2006. The biggest challenge for prevention and control of Anaplasmosis is the rapid diagnosis of the disease during the early process of illness. Here we reported on cloning and expression of *msp2* protein of *A. phagocytophilum*, which is the foundation of developing rapid diagnosis reagents and vaccine and studying on pathogenesis of *A. phagocytophilum*.

Method: 400bp fragments (133 aa) of *msp2* outside of cell membrane, which included important antigenic determinants were selected according to the results of Bioinformatics analysis and amplified by PCR as DNA of *A. phagocytophilum* strain Webster as template. The expression vector (*pET30a(+)-msp2*) was constructed by linking the PCR products of *msp2* gene fragment into the *pET30a(+)* and then transforming it into *E. coli* BL(*plys*).

Result: An expected protein band was observed by IPTG inducing the constructed expression vector and identifying by SDS-PAGE electrophoresis. Time-of-Flight mass spectrometry analysis showed that the amino acid sequences of the recombinant protein were 100% homology with that of the goal protein in the study. Western-Blot analysis demonstrated that the protein positively reacted with rabbit serum immunized with *A. phagocytophilum*.

Conclusion: A highly expression and purified recombinant *msp2* proteins of *A. phagocytophilum* was obtained and a similar natural immunogenicity of *A. phagocytophilum* was noticed.

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PP-248 Pleural effusion in dengue: a cross-sectional study from Karachi

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Background: To study the presentation pattern of Pleural Effusion (PEf) among dengue patients in Karachi. Also to explore limitations faced by the physicians of a developing country in identifying it.

Methods: A cross-sectional study was conducted on patients being admitted at Aga Khan University Hospital in between January 2005 to September 2008. Information on presentation patterns and radiological investigation used to confirm PEf were noted. Data was entered and analyzed in SPSSv16 using Chi Square Test.

Results: Out of total 663 patients, 50 (71%) had PEf as diagnosed by radiographic modalities. Among them were 31 (62%) males and 19 (38%) females. Mean age of presentation was 32±15 years. Majority 23 (46%) had right-sided PEf. Only 309 patients showed respiratory clinical signs and symptoms, 299 (97%) had their Chest X-ray and 10 (3%) had Ultrasound Abdomen studied. Severity of Dengue to PEf and clinical features are shown in Table I and II respectively. Among the radiological confirmed dengue patients with

PEf, 44 (90%) showed altered haem-concentration with an insignificant odd ratio of 0.7 (p value = 0.56, 95% CI: 0.263–2.066).

Conclusion: In comparison to others less number of our patients had PEf. Diagnostic modality preferred by our physicians was Chest X-ray.

Table 1. Comparison of severity of dengue to pleural effusion

Variable	Dengue severity frequency (%)		
	DF	DHF	DSS
Presence of effusion (n = 45)	43 (96%)	1 (2%)	1 (2%)
Severity of effusion (n = 37)			
Mild	35 (97%)	1 (3%)	–
Moderate	1 (100%)	–	–
Side of effusion (n = 43)			
Right	22 (96%)	1 (4%)	–
Left	7 (100%)	–	–
Both	12 (92%)	–	1 (8%)

DF, dengue fever; DHF, dengue hemorrhage fever; DSS, dengue shock syndrome.

Parameter (units)	Mean±SD
Temperature (°C)	38±9
Systolic BP (mmHg)	113±19
Diastolic BP (mmHg)	73±14
Pulse pressure (beats/min)	39±16
Hct (gm/dl)	42±9
Plt (×10 ⁹ /L)	37±31
PT (s) (n = 243)	13±3
APTT (s) (n = 241)	44±15

mmHg: millimeters of mercury; Hct: hematocrit; Plt: Platelets; PT: prothrombin time; APTT: activated partial thromboplastin time.

PP-249 Characteristics of febrile thrombocytopenia during dengue epidemic 2010 in Rawalpindi, Pakistan

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Background: This study aims to determine clinical and laboratory profile along with disease outcome of all cases admitted in Holy Family Hospital with febrile thrombocytopenia during an outbreak of dengue fever in Rawalpindi, Pakistan. We also wish to highlight the need of an effective prevention-control program.

Method: This cross-sectional study included 500 patients admitted in the isolation ward of Holy family Hospital, a public tertiary care hospital, presenting with acute febrile illness during dengue outbreak 2010. Only patients who full filled the WHO criteria for acute dengue (fever and +2 of the following symptoms: headache, eye pain, myalgia, arthralgia, leukopenia, rash, and bleeding) were included.

Results: A total of 500 patients were admitted with suspected diagnosis of dengue fever, of which 34 (6.8%) cases were confirmed with IgM serology. 452 (90.4%) were clinically classified as DF, 47 (9.4%) as DHF and 1 (0.2%) as DSS. 359 (72%) of the cases were from the age group 15–44 years, with 335 (67%) males and 165 (33%) females. Maximum number of patients 294 (62%) were seen in October. The most prevalent symptoms were fever 500 (100%), Bleeding 290 (58%) Body Aches 237 (47.5%), Nausea 210 (42%) and Headache 103 (20.6%). Most common bleeding presentation was gum bleed 76 (25.6%). Main hematological abnormalities were thrombocytopenia