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JOINT EVENTS

THE 13TH
CONFERENCE
OF THE
**INTERNATIONAL
LEPTOSPIROSIS
SOCIETY**

THE 4TH
MEETING
OF THE
**EUROPEAN
LEPTOSPIROSIS**
AND OTHER RODENT-BORNE HAEMORRHAGIC FEVERS
SOCIETY

ABSTRACT BOOK

2-4 SEPTEMBER, 2024
ART & HISTORY MUSEUM, BRUSSELS

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DETECTION OF *LEPTOSPIRA* spp. FROM LEFT-OVER SAMPLES USING NOVEL PROBE-BASED MULTIPLEX QUANTITATIVE POLYMERASE CHAIN REACTION (qPCR)

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Introduction: Leptospirosis is a common infectious disease in several tropical countries. Laboratory investigations are crucial for diagnosis of leptospirosis. This study aims to evaluate clinical utility of *leptospira* diagnostic tests including hydrolysis probe-based multiplex real-time PCR and specific IgM detection for patients in a rural endemic area.

Materials and methods: Specific hydrolysis probes and primer sets were designed to simultaneously detect the *LipL32* gene and *16S rRNA* of *Leptospira* spp. PCR had been evaluated previously in another cohort. Blood samples obtained from suspected cases of leptospirosis at Phrom Khiri (rural district) Hospital during January-February (endemic season) in 2024. EDTA whole blood was collected within 24 hours of symptom onset. DNA was extracted using the Qiagen DNA Blood Mini Kit. Probe-based qPCR was performed using the BioRad CFX96 real-time thermal cycler. *Leptospira*-specific IgM was performed using rapid immunochromatography test (LEPkit).

Results: Blood samples were collected from 117 suspected cases of leptospirosis, among which 33 patients (28%) were IgM-positive, whereas 87 patients (72%) were IgM-negative. Among 33 IgM-positive cases, only 16 cases (48%) were positive by real-time PCR. Among 87 patients with negative IgM, second blood samples could be obtained from 17 patients for repeated diagnostic tests. Interestingly, 5 out of 17 patients (29.41%) showed IgM seroconversion in second specimens, and PCR was positive in all of these 5 patients.

Conclusions: The results suggest that this probe-based qPCR is useful for diagnosis of leptospirosis. The specificity of the IgM rapid test in this endemic area is still questionable because about half of patients with IgM-positive were PCR-negative even at the early stage of infection. The combination of gene detection and serologic testing is recommended to increase diagnostic accuracy. PCR and commercial serologic tests from different companies should be further evaluated in larger clinical samples.

References

- 1 - Podgoršek D, Ružić-Sabljić E, Logar M, Pavlović A, Remec T, Baklan Z, Pal E, Cerar T. Evaluation of real-time PCR targeting the *lipL32* gene for diagnosis of *Leptospira* infection. BMC Microbiol. 2020 Mar 11;20(1):59. doi: 10.1186/s12866-020-01744-4. PMID: 32160864; PMCID: PMC7066766.
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