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On the Superiority of LAMP Over PCR for Tick-Borne Relapsing Fever Surveillance

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Dear Editor

In a recently published study, Houmansadr and Soleimani provided strong evidence that loop-mediated isothermal amplification (LAMP) targeting the *glpQ* gene performs better than conventional PCR for the identification of relapsing fever borreliae in *Ornithodoros tholozani* ticks.

The reported detection frequencies, 18.44% for *glpQ*-LAMP versus 12.62% for *glpQ*-PCR, underscore the superior analytical sensitivity of the LAMP assay. This advantage is likely attributable to the use of six primers targeting eight distinct regions of the DNA template (1). In addition, the substantial intermethod agreement ($\kappa = 0.779$, according to the classification of Landis and Koch [1977] [2]), together with the statistically significant difference in sensitivity ($P < 0.001$; the original authors' report of $P = 0.000$ is a rounding artifact, as a true probability of exactly zero is mathematically impossible), supports the authors' conclusion that LAMP is a reliable and sensitive option for field-based surveillance.

One major strength of this investigation is its real-world applicability. As the authors correctly noted, tick-borne relapsing fever (TBRF) remains underrecognized in rural endemic regions of Iran, owing in part to the short-lived and intermittent nature of spirochetemia, as well as the expense of PCR instrumentation. LAMP, by contrast, operates under isothermal conditions (65°C), provides a rapid turnaround time of approximately 60 minutes, and enables visual readout through turbidity or color change, making it particularly suitable for resource-limited settings. Although Yang and colleagues (3) reported that LAMP was more sensitive than PCR for detecting *Borrelia burgdorferi* sensu lato in Chinese tick samples, it is important to note that *B. burgdorferi* sensu lato, the Lyme disease group, is phylogenetically distinct from relapsing fever borreliae such as *Borrelia persica*. The *glpQ* gene itself is a recognized marker for differentiating these two major groups. Accordingly, sensitivity findings cannot be assumed to generalize automatically across these divergent genogroups, and such comparisons should therefore be interpreted with caution.

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Nevertheless, several aspects merit further consideration. First, although the authors noted that six LAMP-positive specimens were negative by PCR, the possibility of false-positive LAMP results due to nonspecific amplification or primer-dimer formation, although reduced in LAMP, cannot be entirely excluded. Crucially, the original study did not apply a gold-standard confirmatory method, such as Sanger sequencing of amplicons, to these discordant samples. Without such confirmation, it remains unclear whether these six specimens represent true low-burden infections or nonspecific amplification events. Cross-validation using an independent genetic marker, for example *flaB* or *rrs*, or sequencing of a subset of discordant samples would strengthen the reliability of the findings. Second, the paper did not report the limit of detection (LoD) of the *glpQ*-LAMP assay in tick-derived matrices; inclusion of this parameter would be valuable for harmonizing future surveillance protocols. Third, the analytical specificity of the *glpQ*-LAMP assay was not assessed. The original study does not appear to have evaluated potential cross-reactivity with other tick-borne pathogens known to co-circulate in *Ornithodoros tholozani* populations in the study region. In the absence of specificity data, the epidemiological value of positive findings without orthogonal confirmation remains uncertain.

Conclusion

Houmansadr and Soleimani (2025) have provided valuable field-derived evidence supporting *glpQ*-LAMP as a pragmatic, economical, and sensitive approach for TBRF monitoring in tick vectors (4). Given that TBRF cases continue to be documented in Iran, with 1,415 cases recorded between 1997 and 2006, especially in the provinces of Ardabil, Hamedan, Zanjan, and Kurdestan, integrating LAMP into regional health laboratories could improve vector surveillance and ultimately reduce diagnostic delays (5). Future studies should therefore focus on validating this technique in clinical specimens obtained from febrile patients living in the same endemic foci, while also incorporating sequencing-based confirmation of discordant samples and systematic assessments of analytical specificity against co-circulating tick-borne pathogens.

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