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LETTER

# Evaluation of the Performance of a Multiplex Real-Time PCR Assay for the Identification of *Aspergillus*, *Cryptococcus neoformans*, and *Pneumocystis jirovecii* Simultaneously from Sputum in Multicenter [Letter]

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## Dear editor

We appreciate the authors who have reported their research in the "Evaluation of the Performance of a Multiplex Real-Time PCR Assay for the Identification of Aspergillus, Cryptococcus neoformans, and Pneumocystis jirovecii Simultaneously from Sputum in Multicenter". Published in Infection and Drug Resistance 2022:15 6009–6017 This is very important information about the simultaneous identification of *Aspergillus*, *Cryptococcus neoformans*, and *Pneumocystis jirovecii* from sputum using real-time multiplex PCR assay and DNA sequencing methods. No cross-reactivity was detected for any bacteria or fungi. In this study, the authors report that in 40 patients, mixed infection by *Aspergillus* and/or *Cryptococcus neoformans* and/or *Pneumocystis jirovecii* was detected by real-time multiplex assay and the kit minimum detection limit for each of the three species was 1250 copies/mL. DNA sequencing was used as the gold standard; The performance of real-time multiplex assays to detect *Aspergillus*, *Cryptococcus neoformans*, and *Pneumocystis* were analyzed separately.[1]

In this study it was also reported that the detection performance of the real-time multiplex assay for *Aspergillus*, compared with DNA sequencing showed the difference between the two methods was not statistically significant, $P = 0.22 > 0.05$, the overall coincidence rate of the two methods was the same. However, if the purpose of this study is to see the detection performance of the real-time multiplex test, then we can compare it with the results of other studies that also examined the same bacteria with different test tools because currently there are several researchers who use multiple cross displacement amplification (MCDA) combined with a nanoparticle-based lateral flow biosensor (LFB) (MCDA-LFB), which proved fast, reliable and simple to detect the same type of bacteria.[2,3]

Unfortunately, this study reported different results between the multiplex real-time assay of *Aspergillus terreus* DNA sequencing results with negative results, and the results of the real-time multiplex test of *Aspergillus* spp. with positive results that make those results different.

Other studies have reported that in the case of 100 sputum samples, 20 (20%) and 15 (15%) samples were positive by MCDA-LFB and PCR methods, respectively. MCDA-LFB and traditional culture methods showed similar results. Compared with the culture method, the diagnostic accuracy of MCDA-LFB can reach 100%.[4] This shows that the MCDA-LFB method has a better detection ability than the PCR method because the entire

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process can be controlled within 60 minutes including DNA preparation (20 minutes), MCDA reaction (35 minutes) and reporting of results (2 minutes). So it can be concluded that this method has high specificity and sensitivity in detecting bacterial isolates with sputum samples.[5]

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## Disclosure

The authors report no conflicts of interest in this communication.

## References

1. Liu W, Li M, Xu Y, et al. Evaluation of the performance of a multiplex real-time PCR assay for the identification from sputum in multicenter. *Infect Drug Resist*. 2022;15:6009–6017. doi:10.2147/IDR.S379043
2. Ma Q, Yao J, Yuan S, et al. Development of a lateral flow recombinase polymerase amplification assay for rapid and visual detection of Cryptococcus neoformans/C. gattii in cerebral spinal fluid. *BMC Infect Dis*. 2019;19(1):1–9. doi:10.1186/s12879-019-3744-6
3. Tay E, Chen SCA, Green W, Lopez R, Halliday CL. Development of a real-time PCR assay to identify and distinguish between Cryptococcus neoformans and Cryptococcus gattii species complexes. *J Fungi*. 2022;8(5):4–11. doi:10.3390/jof8050462
4. Jiang L, Li X, Gu R, Mu D. Rapid detection of Aspergillus fumigatus using multiple cross displacement amplification combined with nanoparticles-based lateral flow. *Front Cell Infect Microbiol*. 2021;11:1–9. doi:10.3389/fcimb.2021.622402
5. Wu Y, Wang F, Wang C, et al. Detection of Pneumocystis jirovecii and Toxoplasma gondii in patients with lung infections by a duplex qPCR assay. *PLoS Negl Trop Dis*. 2021;15(12):1–15. doi:10.1371/journal.pntd.0010025



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