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Rapid specific detection of oral pathogens using CRISPR-based diagnostics

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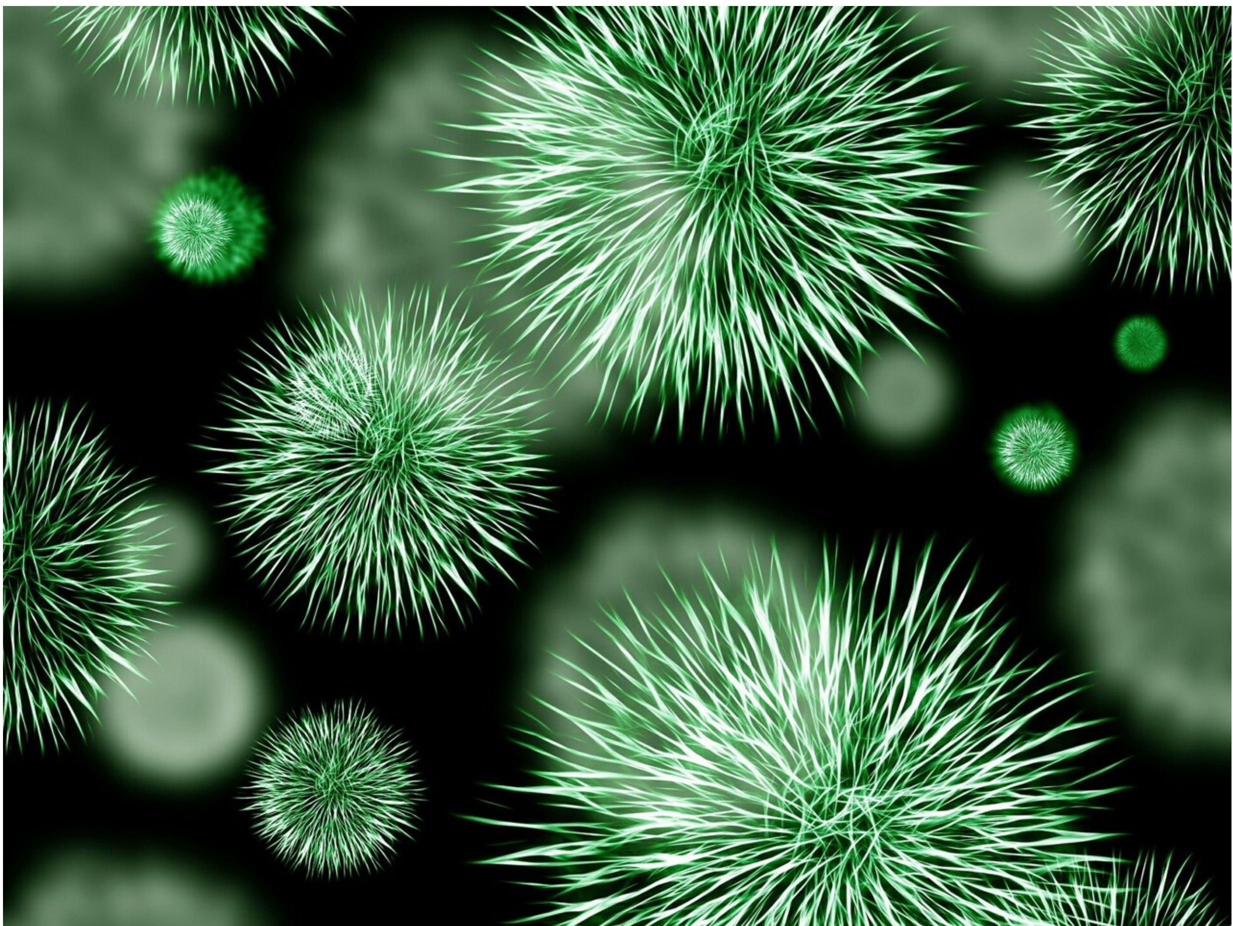

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A study aiming to develop a low-cost, rapid detection technique for the widescale detection and screening of oral microorganisms suitable for point-of-care settings was presented at the 102nd General Session of the IADR, which was held in conjunction with the 53rd Annual Meeting of the American Association for Dental, Oral, and Craniofacial Research and the 48th Annual Meeting of the Canadian Association for Dental Research, on March 13-16, 2024, in New Orleans, LA, U.S..

The abstract, "Rapid Specific Detection of Oral Pathogens Using CRISPR-Based Diagnostics" was presented during the "Craniofacial Diagnostic Sciences" Oral Session that took place on Friday, March 15, 2024 at 8 a.m. Central Standard Time (UTC-6).

The study, by Batbileg Bor of the ADA Forsyth Institute, Cambridge, MA, U.S., tailored the novel CRISPR-Cas-based diagnostic platform Specific High-Sensitivity Enzymatic Reporter Unlocking (SHERLOCK) for the species-specific detection of oral bacterial pathogens and human papillomavirus (HPV) nucleic acids.

The investigators developed a computational pipeline capable of generating guide-RNAs and species-specific gene primers suitable for SHERLOCK. These constructs were synthesized by cell-free biosynthesis systems, and their specificity and sensitivity were experimentally validated by fluorescence readings of reporter RNAs.

The study achieved the detection of oral bacteria within the single-molecule range that remained specific in the presence of off-target DNA found within saliva. Furthermore, the assay was refined for detecting common oral pathogens (e.g., P. gingivalis, F. nucleatum) directly from unprocessed saliva samples. The results of the detection, when tested on 30 patient saliva samples, fully aligned with those of other detection

methods such as qPCR and 16S rRNA sequencing. As a proof of principle, investigators also specifically detected HPV strains 6, 11, 16, 18, 33, and 35 from gDNA background.

This method of detecting oral pathogens is highly scalable and can be easily optimized for implementation at point-of-care settings. The detection takes approximately 35 minutes or less, is extremely low cost, and requires no special skills or techniques to run. Because SHERLOCK targets nucleic acid sequences specifically, future assays can be developed to detect other microorganisms (fungi and archaea) as well as human gene products.

Provided by International Association for Dental, Oral, and Craniofacial Research

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