# Reply to the Letters to the Editor from Ballantyne and Wong

**In response:** We appreciate the concerted attempts to amplify mRNA from saliva by the groups of Drs. Ballantyne and Wong. However, we remain skeptical that the detected nucleic acid amplified in those studies is reliably RNA for all of the experimentally demonstrated reasons expressed in our article (1). These include multiple and redundant reciprocal RNase and DNase controls, no reverse transcription controls, two different commonly used platforms (reverse transcription-PCR and microarray), and a final confirmation using RNA-specific reverse transcription-PCR.

The articles by Wong and colleagues (2, 3) were indeed among the triggers for our attempts to amplify RNA from saliva; no attempt in those studies was made to prove RNA specificity of the gene expression findings. The articles by Ballantyne and colleagues (4, 5) were also among those stimulating our attempts to replicate saliva-based RNA amplification. The use of "no reverse transcription" controls is commendable, but the references to buccal "scrapings/saliva" imply that any RNA found could be derived from the scrape-exfoliated buccal cells, rather than from saliva itself. We are quite familiar with this possibility because we have previously reported quantitative mRNA expression signatures from brush-exfoliated buccal cells (6). However, saliva adjacent to unscraped mucosa, and spontaneously expectorated, is another matter. While we are impressed by the collective work of our colleagues, clear and more definitive, multilayered, and RNA-specific data are warranted before we lift our skepticism.

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## References

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