Validity of Messenger RNA Expression Analyses of Human Saliva

To the Editors: In the September 1, 2006 issue of *Clinical Cancer Research*, Kumar et al. (1) cast doubt on the potential efficacy of saliva as a noninvasive source for the analysis of clinically important gene transcripts. The investigators claim that their data indicate that the cell-free portion of saliva contains no bona fide mRNA species. The investigators report a failure to detect mRNA transcripts from the *36B4*, *β-actin*, and *GAPDH* housekeeping genes. Instead they detected pseudogene signals originating from contaminating genomic DNA.

Previously, we have reported the presence of a number of housekeeping and tissue-specific gene transcripts in saliva, including *GAPDH*, *statherin*, and *histatin 3* (2, 3). As forensic scientists, we are interested in detecting the presence of tissue-specific mRNAs in dried body fluid stains, including saliva, as a means of positively identifying the tissue source of origin of a physiologic stain recovered from the crime scene (2–4). We have not expressly determined whether the varieties of mRNA species that are routinely detected in saliva originate from the cellular or noncellular "supernatant" fractions of whole saliva because, in the forensic context, the cellular versus noncellular source is immaterial. Nevertheless, the presence of mRNA species in whole saliva is irrefutable and we routinely isolate mRNA of sufficient quality and quantity from whole saliva for routine reverse transcription-PCR and quantitative reverse transcription-PCR analyses. The persistence and stability is such that we can detect and analyze mRNA from small quantities of dried saliva stains collected weeks or even months after the initial deposition.

Given our own experience, it is surprising, therefore, that Kumar et al. failed to detect mRNA either in the centrifuged pellet (i.e., cellular fraction) or in whole saliva (which contains cellular components). This may suggest a problem with their experimental technique that puts the validity of their conclusions about the nonpersistence of mRNA in the cell-free portion of saliva in doubt.

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References

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