L.8: Influence of tobacco consumption on the Raman diagnosis of oral neoplasia

There is enough epidemiological evidence that exposure to tobacco is one of the significant etiological factors for the development of oral cancers and pre-cancers. It is, therefore, expected that use of tobacco may cause changes in the spectral signatures of the oral cavity mucosa. A clinical study was carried out by us (at Laser Biomedical Application Section) in collaboration with Tata Memorial Hospital, Mumbai for investigating the tobacco consumption induced changes in the *in-vivo* Raman spectra of oral mucosa of healthy volunteers and its effect on the differential diagnosis of oral lesions [*Journal of Analytical Oncology*, 2016, 5, 110-123].

The clinical in-vivo study involved 28 healthy volunteers and 171 patients with oral cavity lesions. The oral lesions investigated belonged to any of the three histopathologic categories: 1) squamous cell carcinoma (SCC), 2) oral sub-mucosal fibrosis (SMF) or 3) leukoplakia (LPK). Twenty of the healthy volunteers had habits of either smoking and/or of chewing tobacco while the rest did not have any tobacco consumption habits.

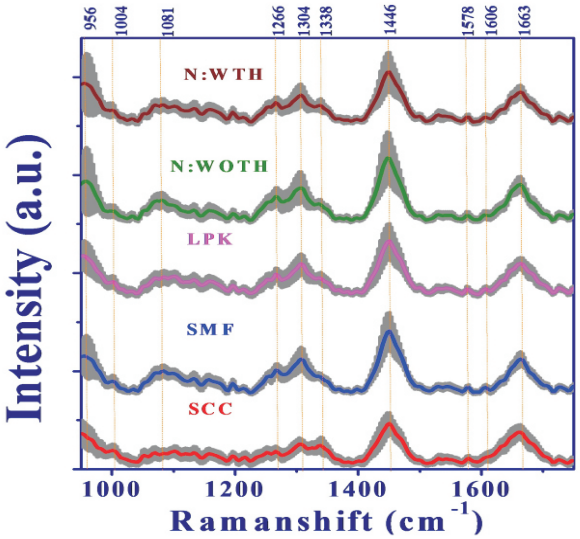

Fig. L.8.1: Mean, normalized Raman spectra of SCC, SMF, LPK, N:WOTH, and N:WTH. The error bars (gray) represent ±1 standard deviation.

The sets of Raman spectra (Fig L.8.1) measured from the healthy volunteers with tobacco (N:WTH) and without tobacco habits (N:WOTH) were observed to have a number of statistically significant differences between the different Raman bands. For example, the intensities of the Raman bands in the wavenumber regions of 1244-1272 cm^{-1}, 1297-1313 cm^{-1}, 1434-1456 cm^{-1} and 1643-1672 cm^{-1} were found to be considerably higher in the spectra of tobacco non-user as compared to those of tobacco users (Fig L.8.2) indicating changes in the collagen and lipid contribution. It was found that the Raman spectra of healthy volunteers with tobacco consumption habits could be separated from the spectra of those without any habit of tobacco consumption with an accuracy of over 95%, using the earlier developed supervised NLMRDF-SMLR algorithm in the leave-one-subject-out cross validation mode.

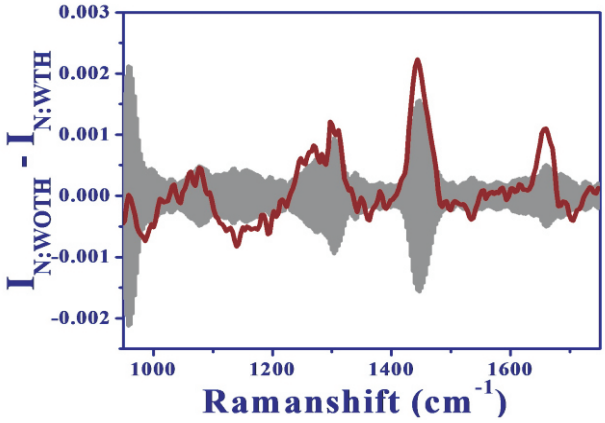

Fig. L.8.2: Mean difference spectra showing statistical differences between oral tissue Raman spectra of healthy volunteers without any tobacco consumption habit (N:WOTH) and with tobacco consumption habit (N:WTH). Gray bands indicate 95% confidence intervals of the difference determined by standard error confidence intervals.

Further, it was found that exclusion of the spectral data of the healthy volunteers with the tobacco consumption habits from the reference normal database considerably improved the overall classification accuracy (92% as against 86%) of the algorithm in separating the oral lesions (SCC, SMF, LPK) from the normal oral tissues. The spectra belonging to SCC, SMF, LPK and healthy volunteers were seen to be correctly classified with ~89%, 85%, 82% and 85%, accuracies respectively, when the reference normal included the spectra of tobacco users. However the situation was seen to drastically improve with the corresponding accuracies improving to ~ 90%, 97%, 92% and 96% when the spectra of tobacco consuming healthy volunteers were excluded from the spectral data of reference normal.

The results of the clinical study demonstrate the potential of Raman spectroscopy in screening tobacco users who are at an increased risk of developing dysplasia or malignancy. Further, the results also show that for accurate discrimination of oral lesions based on their Raman spectra, the reference normal database should exclude spectral data of tobacco using healthy subjects.

Reported by:
H. Krishna and S.K. Majumder (shkm@rrcat.gov.in)