Summary

Altered glycosylation is a universal feature of cancer cells and certain types of glycan structures are well-known markers for tumor progression. Thomsen-Friedenreich (TF) antigen an established carbohydrate antigen consisting of Galβ1→3GalNAc-ser/thr, expressed during metastasis on mucosal epithelial cells. This antigen is also part of predominantly occurring O-linked glycans of mucins, and is called mucin core I glycan. Saliva being rich in mucins, aberrant glycosylation could be expected in the salivary constituents. Monitoring the cancer associated glycosylation changes has become an important and integral part of prognosis and detection of cancer. Tumor antigen specific lectins have become potential tools replacing monoclonal antibodies to study the expression of different antigens expressed during oncogenesis and metastasis.

In the recent past Sclerotium rolfsii lectin (SRL), a TF antigen binding lectin has been purified and characterized for its fine sugar specificity and is being currently studied intensively for its properties in this laboratory. One of the objectives of these investigations is to investigate cancer associated glycosylation changes by exploiting TF antigen binding property of SRL. Present study was aimed at identifying unique cancer associated antigens expressed in saliva and oral squamous epithelial tissues during oral Squamous Cell Carcinoma (SCC).
In all the investigations carried out in the present study involve saliva samples from 10 SCC confirmed patients and compared with 10 samples from normal, healthy individuals.

Preliminary investigations carried out to determine quantitative differences in the salivary components of normal and oral cancer patients indicate significant differences in the expression of total amounts of total proteins, total sugars, hexosamines and sialic acid levels. Salivary components from cancer patients showed higher levels compared to normal healthy ones.

Lectin precipitation assays carried out to understand the nature of changes in glycans, using SRL, indicated major differences with respect to carbohydrate chains occurring in salivary glycoproteins. Much smaller concentrations of salivary glycoproteins of cancer patients are required for precipitation of SRL compared to normal saliva. Results indicate the presence of SRL-recognizing unique glycans in SCC patients, but not present or present in very small quantities in normal saliva.

Comparative SDS PAGE patterns of normal and SCC saliva glycoproteins showed totally different and distinct glycoproteins in the saliva of all the SCC patients. Specific SRL binding glycoproteins were identified by lectin blot assay after SDS PAGE and compared with well-known cancer antigen marker lectin, PNA. Interestingly PNA and SRL interact with one common glycoprotein band of 45 kDa, but SRL with two additional glycoprotein bands of 29kDa and 27 kDa. However no such
lectin binding glycoproteins were found in any of the saliva samples of normal healthy individuals. Thus results confirm the expression of TF antigen in saliva of SCC patients.

Histochemical binding studies, using biotinylated SRL and PNA with human normal and malignant oral tissues revealed interesting differences. Biotinylated SRL showed specific binding sites on malignant cells. These findings strongly indicate that the TF antigen is expressed not only on the squamous epithelial cells but also in salivary glycoprotein fractions secreted by these cells during squamous cell carcinoma. Thus it warrants for careful investigations using large number of tissue samples to develop SRL as a potential histochemical probe in cancer biology.

To confirm the specificity of lectin to intrinsic antigens expressed during malignancy, interaction studies of SRL with Candida albicans, isolated from oral cavity of cancer patients, were carried out. These binding studies of FITC-SRL with Candida albicans showed no binding and agglutination and confirmed that SRL binds to the salivary glycoproteins uniquely expressed during SCC of oral cavity but not the remnants of C. albicans.

Thus considering the TF antigen binding property of SRL, our study indicated the occurrence of unique glycoproteins with altered glycosylation resulting in the expression of TF antigen during squamous cell carcinoma of oral cavity.