

The salivary proteome in relation to oral mucositis

S.J.M. van Leeuwen¹, G.B. Proctor², A. Staes^{3,4,5}, A.M.G.A. Laheij^{6,7}, C.M.J. Potting⁸, M.T. Brennan⁹, I. von Bültzingslöwen¹⁰, F.R. Rozema^{6,11}, M.D. Hazenberg¹², N.M.A. Blijlevens^{8*}, J.E. Raber-Durlacher^{6,11*}, M.C.D.N.J.M. Huysmans¹

¹Radboud university medical center, Radboud Institute for Health Sciences, Department of Dentistry, Nijmegen, The Netherlands

²Centre for Host Microbiome Interactions, King's College London Dental Institute, London, United Kingdom

³VIB-UGent Center for Medical Biotechnology, Ghent, Belgium

⁴Department of Biomolecular Medicine, Ghent University, Belgium

⁵VIB Proteomic Core, Ghent, Belgium

⁶Department of Oral Medicine, Academic Centre for Dentistry Amsterdam, University of Amsterdam and VU university, Amsterdam, The Netherlands

⁷Department of Preventive Dentistry, Academic Centre for Dentistry Amsterdam, University of Amsterdam and VU university, Amsterdam, The Netherlands

⁸Radboud university medical center, Radboud Institute for Health Sciences, Department of Hematology, Nijmegen, The Netherlands

⁹Department of Oral Medicine, Atrium Health's Carolinas Medical Centre, Charlotte, NC, United States of America

¹⁰Department of Oral Microbiology and Immunology, University of Gothenburg, Gothenburg, Sweden

¹¹Department of Oral and Maxillofacial Surgery, Amsterdam UMC, University of Amsterdam, Amsterdam, The Netherlands

¹²Department of Hematology, Amsterdam Infection and Immunity Institute, and Cancer Center Amsterdam, Amsterdam UMC, University of Amsterdam, Amsterdam, The Netherlands

Objective: Decreased salivary flow rate and/or changes in protein composition reported after autologous hematopoietic stem cell transplantation (ASCT) reduces the protective function of saliva. Oral mucositis (OM); inflammation of oral mucosa resulting of preparative chemotherapy for ASCT is a risk factor for systemic infections and affects patients' quality of life. In this study, a TMT-labelled proteomics experiment and a label-free quantification (LFQ) proteomics experiment were used to identify the salivary proteome in ASCT recipients and to explore whether differences in the salivary proteome occur between patients with ulcerative OM (uOM; WHO score ≥ 2) and those without (nOM).

Methods: In both experiments, salivary proteins were analyzed using liquid chromatography and tandem mass spectrometry. For the TMT-labelled experiment, saliva samples of 5 uOM and 5 nOM patients were pooled at different time points: baseline, 1, 2, and 3 weeks after ASCT and 3 months after ASCT. Principle component analysis was used to explore patterns between the uOM and nOM pools at different time points. For the LFQ experiment, an uOM pool (consisting of saliva from 9 uOM patients and 6 time points (same time points as TMT experiment and 12 months after ASCT)) was generated next to a nOM pool (10 nOM patients and 6 time points). Unique and up-regulated proteins of the uOM and nOM pools were further investigated with gene ontology.

Results: A different salivary proteome was indicated with a distinct clustering of the uOM pools at baseline, week 2 and week 3 after ASCT from the other pools. In the LFQ experiment, unique and up-regulated proteins of the uOM pool consisted of more intracellular proteins. Those proteins of the nOM pool were more involved in immune system-related processes.

Conclusion: The salivary proteome was suggestive of extra mucosal damage in uOM patients and extra protection in nOM patients.