Carla Costa¹ ², Ana Catarina Alves¹, Solange Costa¹ ², Amadeu M.V.M. Soares¹, Marta S. Monteiro¹, Susana Loureiro³ and João Paulo Teixeira¹ ²*
¹Portuguese National Institute of Health, Portugal ²EPIUnit - Institute of Public Health, University of Porto, Portugal ³Department of Biology & CESAM, University of Aveiro, Portugal

The Comet assay is a valuable tool for the detection of DNA damage in genotoxicity and human biomonitoring studies. Throughout the years, this biomarker has undergone several adaptations in their protocol in order to increase its sensitivity and the possible outcomes. By including an additional step of DNA digestion with lesion-specific endonucleases, the comet assay can provide information regarding the type of DNA damage detected in cells. The use of these enzymes has also allowed the development of a methylation-sensitive modified version of the comet assay. This version enables the routine measurement of global methylation, as well as CpG island DNA methylation in a variety of cells while simultaneously determining the genetic integrity of examined cells (Wentzel, 2012). Briefly, it makes use of isochromatid restriction enzymes HpaII and MspI (that display differential sensitivity to DNA methylation) to characterize methylation outside CpG islands and restriction enzyme NotI to determine DNA methylation in CpG islands. The technique has been recently adapted to a medium-throughput version (Lewies, 2014) that allows the simultaneous analysis of a larger number of samples and overcomes some technical problems. Nevertheless, this technique has not yet been carried out in human biomonitoring studies. In this context, the aim of this work was to make use of this version of the comet assay to characterize global DNA methylation in approximately 50 human samples. Samples were analysed by the methylation-sensitive modified version of the comet assay (medium-throughput) and by ELISA based assay. Data obtained with both methods were compared and reproducibility of the methylation-sensitive modified version of the comet assay determined. Results obtained contribute to knowledge on the feasibility of this version of the comet assay and its possible usage in human biomonitoring studies as an epigenetic biomarker.

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