

# Human amniotic membrane homogenate: A novel biomaterial-based strategy to impede migration and invasion of bladder cancer cells

Aleksandar Janev<sup>1</sup>, Taja Železnik Ramuta<sup>1,2</sup>, Urška Dragin Jerman<sup>1</sup>, Hristina Obradović<sup>1,3</sup>, Maja Čemažar<sup>4</sup> and Mateja Erdani Kreft<sup>1</sup> \*

<sup>1</sup>Institute of Cell Biology, Faculty of Medicine, University of Ljubljana, Ljubljana, Slovenia

<sup>2</sup>Department of Synthetic Biology and Immunology, National Institute of Chemistry, Ljubljana, Slovenia

<sup>3</sup>Group for Hematology and Stem Cells, Institute for Medical Research, University of Belgrade, 11000 Belgrade, Serbia

<sup>4</sup>Department of Experimental Oncology, Institute of Oncology Ljubljana, Ljubljana, Slovenia

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**INTRODUCTION:** Bladder cancer ranks among the top ten most frequently diagnosed cancers globally, with approximately 25% of diagnosed cases presenting as initially aggressive muscle-invasive tumours, marked by poor prognosis and frequent metastasis [1]. Human amniotic membrane (hAM) is an extraembryonic membrane with anti-inflammatory, antifibrotic and antimicrobial properties [2], however, despite the growing number of studies, the cellular mechanisms underpinning its anticancer activity remain poorly elucidated. In our investigation, we delved into the mechanisms by which hAM homogenate influences bladder cancer and normal urothelial cells.

**MATERIALS AND METHODS:** The hAM homogenate was prepared from placentas collected from healthy donors after elective caesarean section according to the protocol described in [3]. Wound healing assay [4] was used to evaluate the influence of hAM homogenate on migration of normal porcine urothelial NPU cells, human papillary cancer urothelial RT4 cells and human muscle-invasive bladder cancer T24 cells. For the invasion of cancer cells, invasion assay [4] was performed using transwell chambers with membranes of 8 µm pore size coated with diluted Matrigel matrix. The effect of hAM homogenate on the migration pattern of T24 cells stably transfected with enhanced green fluorescent protein (eGFP) was examined using time-lapse confocal microscopy. Protein expression levels of molecules involved in cell migration were measured by Western blot analysis, while gene expression was quantified by RT-qPCR [3, 4]. To compare statistical differences between at two experimental groups, parametric unpaired two-tailed Student's t-test or the non-parametric Mann–Whitney test was used.

**RESULTS AND DISCUSSION:** Our study revealed that hAM homogenate impedes the migration rate of bladder cancer RT4 and T24 cells, but not of normal NPU cells. In addition to cell migration, hAM homogenate significantly decreased the invasion rate of muscle-invasive bladder cancer T24 cells. Furthermore, this antimigratory effect was associated with the downregulation of FAK and Rho GTPases expression—crucial proteins in actin cytoskeleton reorganization.

**CONCLUSIONS:** In summary, the findings highlight the significant potential of hAM homogenate as a valuable biomaterial in the field of medicine, particularly as a complementary component in the treatment of bladder cancer.

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\*Corresponding author E-mail: [mateja.erdani@mf.uni-lj.si](mailto:mateja.erdani@mf.uni-lj.si)

