Biocompatibiliy study on a HydroBalanced wound dressing*

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C. Wiegand[1], M. Abel[2], A. Kramer[3], G. Müller[3], P. Ruth[2], U.-C. Hipler[1]

[1]Department of Dermatology, University Medical Center Jena, Germany
[2]Lohmann & Rauscher GmbH & Co. KG, Rengsdorf, Germany
[3]Institute for Hygiene and Environmental Medicine, Greifswald, Germany

Introduction

Biocompatibility is one of the main requirements for the safe use of medical devices. The *in vitro* cytotoxicity is often a qualitative analysis based on the examination of cell damage and growth after direct or indirect contact with the material [1]. According to the DIN EN ISO 10993-12 we prepared extracts of a HydroBalance wound dressing (HWD)* consisting of biocellulose. Cell proliferation under the influence of the extracts was determined by measurement of the cell ATP content. ATP (adenosine triphosphate) serves as the principal immediate donor of energy and is present in all metabolically active cells. Cell injury results in a rapid decrease in cytoplasmatic ATP. Measurement of ATP is therefore fundamental to the study of living processes [2].

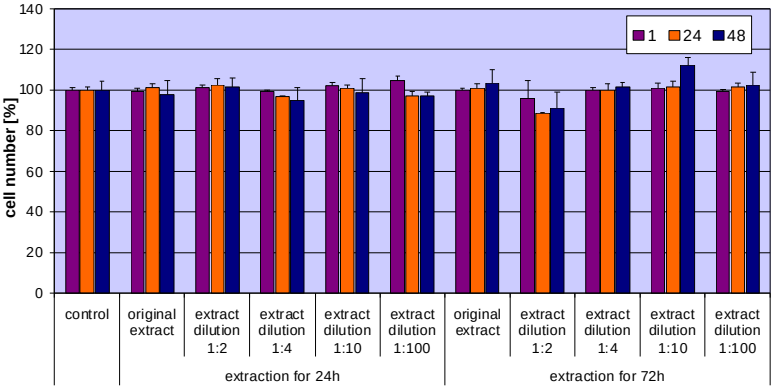

Fig. 1: Influence of HWD extract on HaCaT keratinocytes determined via measurement of the ATP content (mean ± SE).*

Material & Methods

HaCaT-cells, primary fibroblasts and keratinocytes were cultured with extract of the HWD* and increasing concentrations of the raw material chlorhexidine itself as reference. Cell proliferation was measured by means of the ATPLite™-M kit (Perkin Elmer, USA). The luminometric ATP assay is based on the detection of light generated by the ATP dependent enzymatic conversion of D-luciferin by luciferase. Interleukin release was determined via ELISA specific for IL-6 and IL-8 (Milenia biotec, Germany).

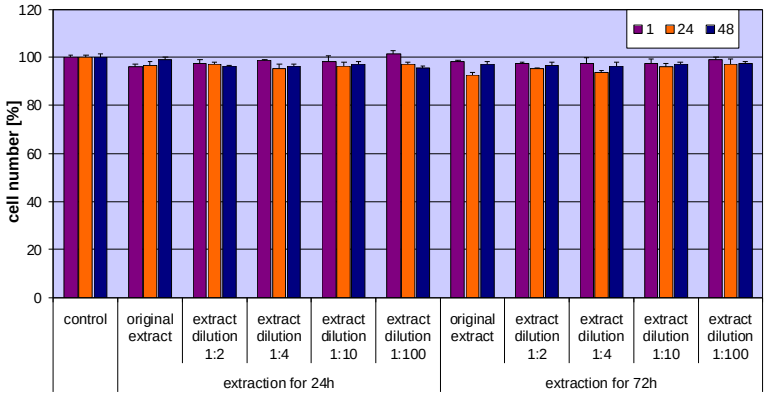

Fig. 2: Effect of HWD extract on fibroblast proliferation determined via measurement of the ATP content (mean ± SE).*

* HWD = Suprasorb® X (Lohmann & Rauscher, Germany)

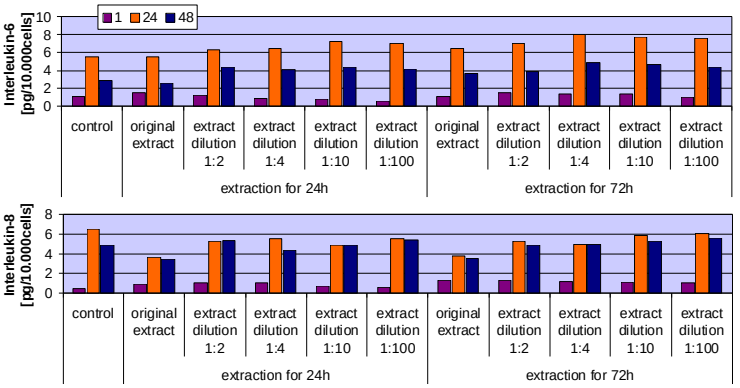

Fig. 3: Incubation with HWD extract does not change the release of IL-6 and IL-8 by keratinocytes compared to the control (mean)*

Results

No significant influence of the HWD* extract on the proliferation of human keratinocytes, HaCaT-cells (Fig. 1), and fibroblasts (Fig. 2) was found. The incubation of the cells with this extract did not change the release profile of IL-6 and IL-8 compared to the control (Fig. 3). The raw material chlorhexidine itself in different concentrations had a distinct negative effect on cell viability (Fig. 4).

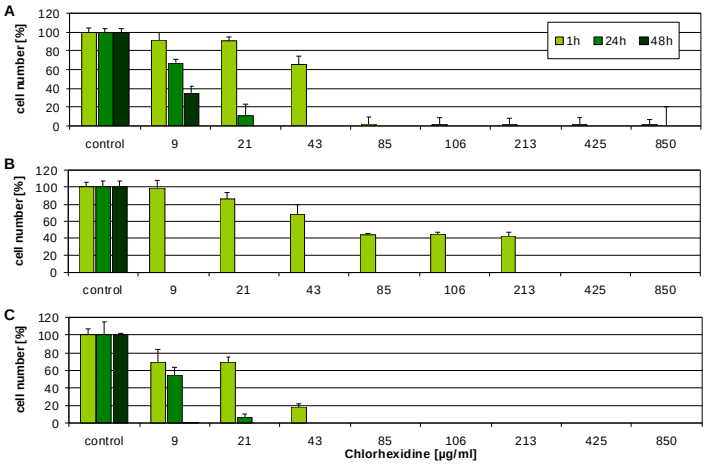

Fig. 4: Chlorhexidine has a negative effect on the viability of HaCaT-cells (A), keratinocytes (B) and fibroblasts (C) as determined by ATP measurement (mean ± SE).

Conclusions

In conclusion, the extract of HWD* does not exhibit a negative effect on the cells under the test conditions. The determination of ATP is expedient in cytotoxicity studies as it provides a stabile metabolic marker that enables direct monitoring of cell viability. A lot of methods have been used for ATP determination, but the most successful technique is the bioluminescent method, because of its sensitivity and the wide dynamic range [2].

References

1. *Johnson HJ et al. Biocompatibility test procedures for materials evaluation in vitro. I. Comparative test system sensitivity. J Biomed Mater Res 1983; 17:571-586.*
2. *Hipler U-C et al. The use of an ATP bioluminescence assay to quantify HaCaT cell cytotoxicity. In Bioluminescence and Chemiluminescence ed. by Roda A, Pazzagli M, Kricka LJ, Stanley PE. John Wiley & Sons Chichster, England 1999 pp169-72*

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