

New Methods for Testing Skin Protection Against Noxae Using the Bovine Udder Skin (BUS)

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Keywords: BUS Model, Skin Protection, Free Radicals, ESR Spectroscopy, Skin Irritation, Coolant, Water-soluble Noxa, Lipid-soluble Noxa, AWMF Guideline »Berufliche Hautmittel«

Parts of this publication were presented at the 58th SEPAWA Congress, October 12-14, 2011, Fulda, Germany

ABSTRACT

Quantitative and qualitative determination of skin irritation and skin protection through market formulations using an *in vitro* skin model, the perfused bovine udder (BUS) and different analytical methods, was the objective of the present research. Various noxae that are percutaneously absorbed cause skin irritation and damage through different mechanisms. Cell vitality, signs of inflammation, and the presence of inducible free radicals are among the analytical parameters that enable quantification of skin damage. In this study cytotoxic effects were determined using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay, and tissue levels of the pre-inflammatory mediator prostaglandin E₂ in treated/untreated whole skin biopsies were assessed using an enzyme immunoassay. The inducibility of free radicals was determined by electron resonance spectroscopy tech-

niques. Noxae (standardized chemicals and products) like detergents, coolants, toluene, and n-octane caused reduced cell vitality and metabolic activity, inflammation as evidenced by the presence of prostaglandins, and enhanced values in the inducible Free Radical Status in *in vitro* bovine udder skin. The effect of the noxae is dose-dependent and permeation time-dependent, allowing construction of dose-effect curves. The induced cytotoxicity and free radical status show a positive correlation, while the inflammatory actions analyzed by the prostaglandin E₂ tissue concentration do not.

As a consequence, the skin compatibility of noxae and performance of skin protection formulations can be evaluated by the measurement of different cell physiological and biophysical parameters at the very early stages of skin damage.