Despite significant progress in biomedical research, many important diseases are still difficult to treat successfully. Numerous new drug candidates have shown promises in preclinical studies, but failed in clinical studies. This raises a question on the predictability of preclinical studies. Too often the promising results obtained in animal studies are not reproduced in clinical studies. Recently, this problem has been most visible in the area of nanoparticle formulations for cancer treatment. A recent trend to overcome the lack of translation from small animal studies to the clinic is to utilize human-derived cells and tissues to improve the success rate in clinical trials. Human cells grown in monolayer cultures have been traditionally used. Human cells grown in a routine 2D culture with low-calcium medium, however, do not differentiate and do not crosstalk with other types of differentiated cells, which is mandatory for normal tissue function.

Organotypic 3D disease models based on human cells have been developed for improved transferability of preclinical results to clinical success. Recently, significant advances have been made in reconstruction of human skin. One impetus of such advances was that the European Union decided to accept validated protocols for toxicity testing based on reconstructed human epidermis in place of animal studies [1]. Based on reconstructed human full-thickness skin, disease models have been used for drug efficacy testing, e.g., a psoriasis model and a non-melanoma skin cancer (NMSC) model. The latter model and an ichthyosis model served for early studies on nanoparticle-enhanced drug delivery [2]. Yet, morphology and skin barrier function have not been related to skin penetration and local biotransformation as well as the efficacy of drugs.

Professor Monika Schäfer-Korting and her colleagues, who tried to improve preclinical research on NMSC treatment, systematically adapted the ratio of transformed to normal human keratinocytes to establish advanced organotypic models for the most frequent cancer in humans (cutaneous squamous cell carcinoma) [3]. Before dermal tumor invasion or metastasizing, tumor cells grow within the epidermis, usually referred to as actinic keratosis. Despite disappointing cure rates, in particular in immunodeficient patients, actinic keratosis is subject to topical therapy. In contrast, cutaneous squamous cell carcinoma is subject to surgery with an often disfiguring outcome. This underpins the need for therapeutic improvement, inhibiting the progression of actinic keratosis to the malignant tumor. The models, with varied seeded number of tumor cells, reflect the morphological hallmarks of actinic keratosis and cutaneous squamous cell carcinoma, lesions confined to the epidermis only or spreading into the dermis, respectively. Morphological alterations are detected not only by immuno-histology but also by non-invasive reflectance confocal microscopy. In the normal model, the location of tight junction proteins (claudin-1, claudin-4, occludin, ZO-1) is in accordance to normal human skin. In the tumor model there is a loss of occludin and claudin-4 and a heterogeneous distribution of claudin-1. This widely reflects the alterations in tumor patients.

In the NMSC models, the reduced fraction of orthorhombic lateral stratum corneum lipid packing also indicates impaired skin barrier function. Less dense lipid packing is accompanied by reduced cholesterol and increased phospholipid levels and by a shift in ceramide subclasses to higher levels of phytoceramides. The amounts of cholesterol and phospholipids in the stratum corneum resemble the carcinoma microenvironment in patients, which are depleted from cholesterol probably due to its high consumption by the carcinoma. Whereas the proportion of ceramide subclasses in normal models is in accordance with normal human skin, the absolute ceramide content in the stratum corneum is lower. These lipid changes in the NMSC model result in significantly increased caffeine permeability compared to the normal model, the enhancement being strongly correlated to the carcinoma cell count. Although studies in patients are needed to verify enhanced permeability of skin in NMSC, the results indicate that the hyperkeratosis may not impair drug access to the tumor cells in actinic keratosis.

Consistent with the clinical outcome, ingenol mebutate commercial gel caused abundant epidermal cell necrosis in both NMSC models and normal models as indicated by decreased Ki-67 indices and enhanced LDH release. The effects are close to rapidly induced cell death in previous ingenol mebutate studies. A clear trend to increased skin surface pH values in NMSC models, however, may improve the benefit/risk ratio by the use of pH-cleavable drug delivery systems. Thus, the study described here may pave the way not only to improved preclinical testing but also to the development of disease-adjusted drug carrier systems. Understanding the underlying mechanisms allows a faster route to the cure of the disease.

References


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