One of the vital function of skin is formation of a barrier that protects physical, chemical and bacteriological intrusion in the body, also prevents dehydration by evaporative water loss. Epidermis, the uppermost layer of the skin, is composed of keratinocytes, cells endowed with high proliferative potential and ability to differentiate. The loss of tight balance between keratinocyte proliferation and differentiation contributes to development of many skin diseases. It is estimated that in Poland about 5 million people suffer from various dermatological diseases. Understanding the molecular basis of the mechanisms leading to skin dysfunction is essential for the development of new therapeutic strategies.

It is known that epidermal homeostasis is maintained by activity of numerous genes. We believe that one of them is the *HSPA2* gene coding for the poorly characterized heat shock protein A2 (HSPA2). This protein belongs to the HSPA (HSP70) family of chaperone proteins with cytoprotective functions. In contrast to the other members of HSPA family, the expression of the *HSPA2* gene is not heat shock-inducible, but it can be directly modulated by the hypoxia-induced factor 1 (HIF-1) transcription factor, a master regulator of cell adaptation to oxygen deficiency (hypoxia). HIF-1 plays pleiotropic roles in skin biology, among others it controls epidermis integrity. So far, mostly due to high expression of the human *HSPA2* gene in the testes and various types of tumors, studies on HSPA2 protein focused on explaining its importance for male fertility and the acquisition of malignant phenotype by cancer cells.

Our recent finding have shown that HSPA2 is also highly expressed in various multilayered epithelia. Of particular interest is the pattern of HSPA2 expression in the skin, where the protein is present only in keratinocytes located in the basal layer of the epidermis. We have also observed that this pattern is altered in psoriasis, a chronic inflammatory skin disease manifested also by aberrant keratinocyte differentiation and function. The functions of HSPA2 in the skin are unknown. This project has been formulated on the basis of our preliminary findings suggesting that HSPA2 plays important role in maintaining epidermal homeostasis. Hereby, we hypothesize that HSPA2 may control the initiation of keratinocyte differentiation. We assume that HSPA2, as a molecular chaperone, may impact on keratinocyte phenotype through its ability to modulate activity of other key proteins (transcription factors, signaling proteins) for keratinocyte proliferation and/or differentiation. We presume that HSPA2, via direct interaction with HIF-1α, may impact on the activity of HIF-1, which is a pleiotropic modulator of skin homeostasis. The aim of this project will be to determine the role of HSPA2 in the maintenance of epidermal homeostasis and in the pathological processes of the skin. This goal is to be achieved by pursuing the following specific objectives:

- to determine the role of HSPA2 in controlling epidermal tissue homeostasis
- to find the HSPA2-interacting proteins in keratinocytes
- to examine the HSPA2 expression in psoriatic skin and to study of the effect of psoriasis-associated cytokines on the *HSPA2* gene expression in keratinocytes.

In this project we will use the *in vitro* tissue engineered techniques to construct three dimensional (3D) reconstituted human epidermis (RHE) model. This model, when combined with genetic manipulation methods, is considered a powerful tool for studying the effects of a particular protein on the integrity and function of human skin. Determining the impact of HSPA2 on keratinocyte biology we will characterize the cell phenotype resulting from genetic modifications in HSPA2 expression (overproduction and downregulation), and will examine the impact of introduced changes on keratinocyte ability to develop the full-thickness RHE. To accomplish the research goal we plan to use histological procedures, molecular and cell biology methods and techniques designed to analyze protein-protein interaction. To investigate how HSPA2 may contribute to keratinocyte physiology we will also use modern techniques of global gene expression analysis such as new generation sequencing and methods allowing identification of HSPA-binding partners using mass spectrometry.

Implementation of this project will improve our understanding of the homeostatic mechanisms underlying human skin integrity and function, as well as their alterations involved in the development of skin diseases (psoriasis, cancer). Results will expand current knowledge on the complex function of HSPA2 in the skin physiology, but also in skin pathology, we hope to disclose novel functions of HSPA2 and reveal its potential clients. These findings in turn may facilitate the discovery of new molecular targets for effective treatment of dermatological diseases, and to improve protocols for skin equivalents construction for the purposes of regenerative medicine. Moreover, results obtained in this project will be published in a high-impact journal, will be presented at national and international conferences, and will form a basis for preparation of doctoral dissertation