Abstract
In acne different pathogenetic factors contribute to the inflammation process, defect in keratinisation, increased sebaceous gland activity and increased colonisation of Propionibacterium acnes. The results of in vitro and in vivo studies confirm the previous reports on strong anti-microbial effectiveness of phytosphingosine in vitro and in vivo. In addition, phytosphingosine shows excellent clinical results in the context of skin care in acne, based on the two properties, i.e. anti-inflammatory and anti-microbial activity. These results demonstrate the potential of phytosphingosine to enhance or complement existing acne therapies acting as an active cosmetic ingredient.

Introduction
The earliest subclinical acne ‘lesion’ is a microcomedone, hyperproliferation of the follicular epithelium being its characteristic feature. Recently significant pro-inflammatory factors, such as interleukin-1, have been identified around clinically normal pilosebaceous follicles from uninvolved skin in acne patients prior to hyperproliferation of the follicular epithelium[1]. This contributes to the concept that acne vulgaris should be classified as an inflammatory skin disease.

Materials and methods
I In vitro studies
Methods
Anti-microbial activity
Using a methodology similar to that previously described(2) the inhibitory effect of phytosphingosine on growth of different micro-organisms was tested.

Release of interleukin-1α by UVB irradiated human skin on culture
The effect of UVB was investigated using human skin explants in culture as a model. Phytosphingosine (0.2% and 1.0%) and dexamethasone (10M-6) were applied to human skin explants in culture to test their anti-inflammatory potential. The products were applied one hour before and immediately after irradiation (20 minutes of UVB 2 J/cm²). The interleukin-1α secretion was measured using an ELISA kit at 24 hours.

Effect on the Artificial Human Epidermis after Irritation with SDS
The efficacy of phytosphingosine on a 3D artificial skin model (SkinEthic™) was investigated after damage with the irritant surfactant sodium laurylsulfate (SDS). After thawing of the artificial human epidermis followed by controlling their viability, a 0.25% SDS solution (dissolved in PBS) has been added to the skin models for 40 minutes to induce chemical stress.

Afterwards the skin slides were washed and a cosmetic O/W formulation (vehicle, formulation containing 0.145% phytosphingosine) was applied.

After 24 hours different parameters as cell death represented by lactate dehydrogenase (LDH), viability according to the XTT assay, inflammatory response judging from interleukin-1α (IL-1α) expression.

II In vivo studies
Topical in vivo study on anti-microbial efficacy
The anti-microbial efficacy of topical phytosphingosine within an emulsion-based format was determined in an in vivo test. Two products (phytosphingosine and phytosphingosine-salt) were compared against a control formulation, and a frequently used anti-microbial, triclosan, as a positive control. The formulations were tested on the unwashed hands of 12 subjects based on bacterial counts. The total microbial count was redetermined on the skin at zero time, after 1 hour and after 4 hours.