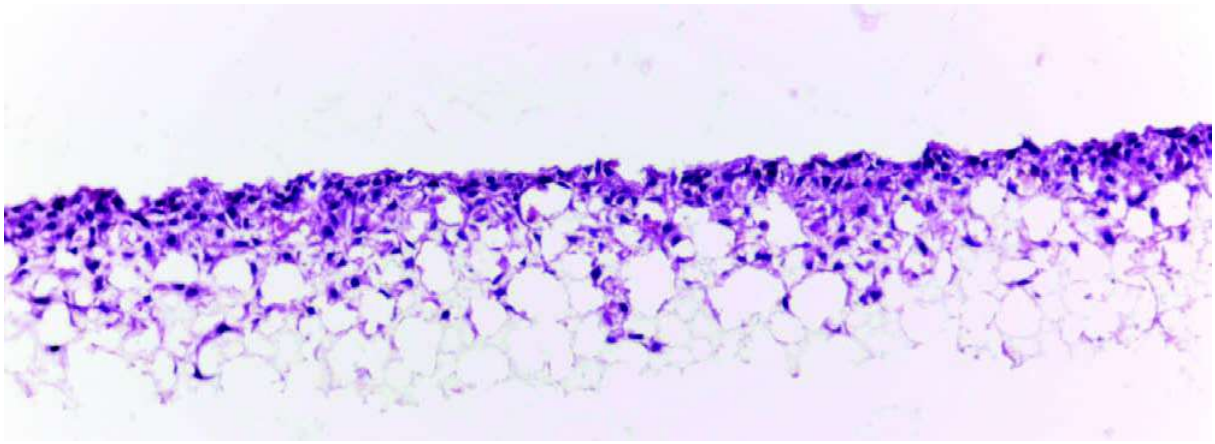
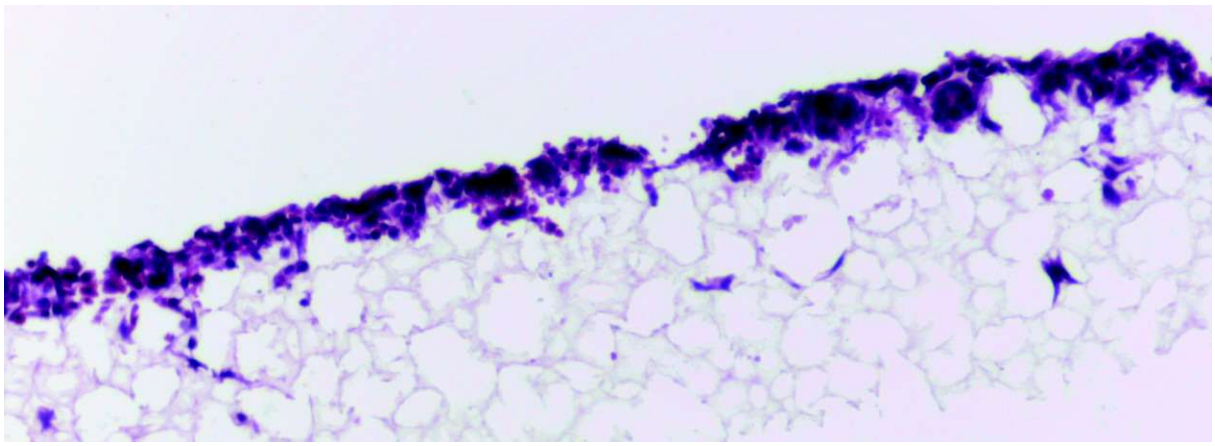
Comparison of 3D cell growth patterns on alvetex® 12-well plate format (AMS.AVP002)

Alvetex® in the 12-well plate format (AMS.AVP002) was treated with EtOH and washed with
complete medium prior to cell seeding. [Complete medium consisted of: DMEM, 10% FBS, 2 mM L-glutamine and 100 U/ml Penicillin & Streptomycin]. HaCaT cells (a human keratinocyte cell line) were plated at a density of $0.5x10^6$ cells in 150 µl per well while HepG2 cells (a human liver cell line) were seeded at $2x10^6$ cells in 150 µl per well. Plates were incubated for three hours before flooding with further media and maintained for 7 days. After preserving in Bouins fixative the discs were paraffin embedded, sectioned (10 µm) and counterstained with Haematoxylin and Eosin. HaCaT cultures demonstrated significant cell invasion into the matrix, while HepG2 cells remained resident in the top 25% of the matrix.



HaCaT cells

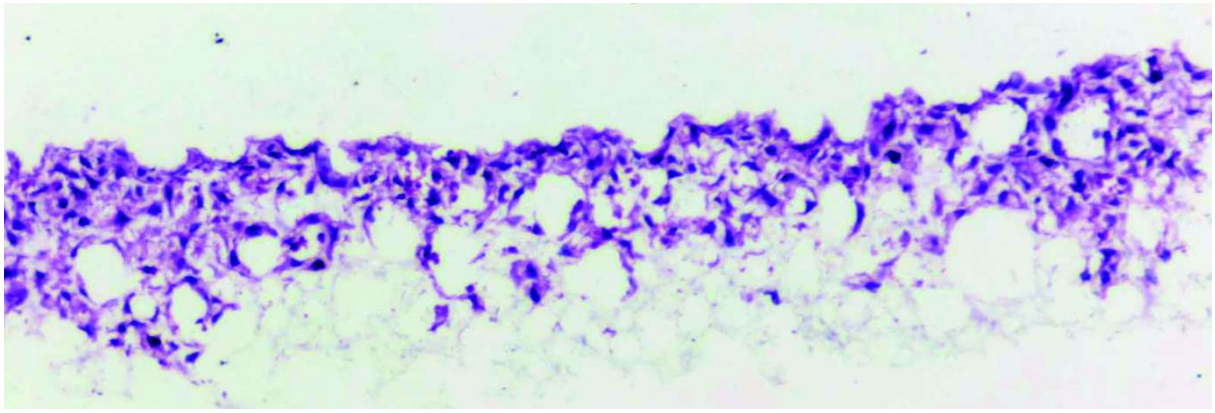


HepG2 cells

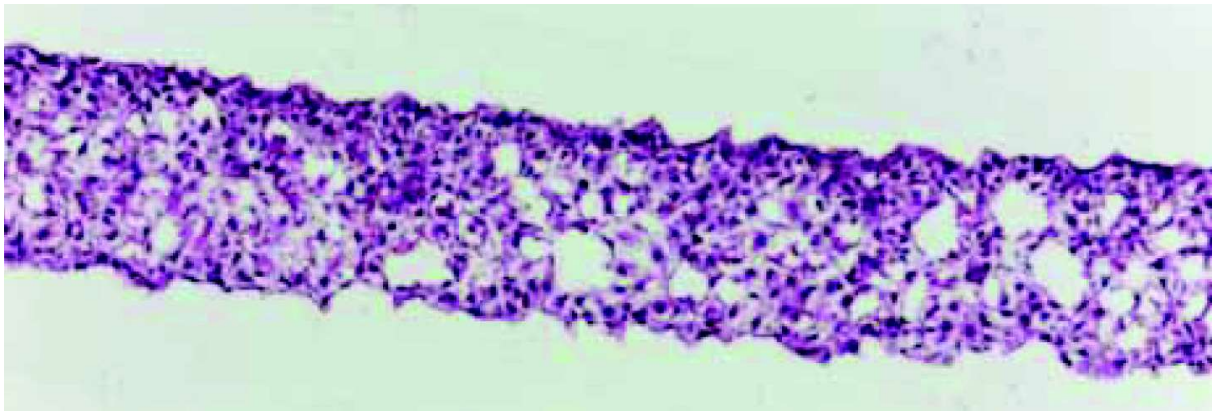
Figure 1. *Comparison of 3D cell growth patterns on alvetex® 12-well plate format (AMS.AVP002). Micrographs taken at 20x magnification.*

Comparison of 3D cell growth patterns of HaCaT cells on alvetex® presented in various formats

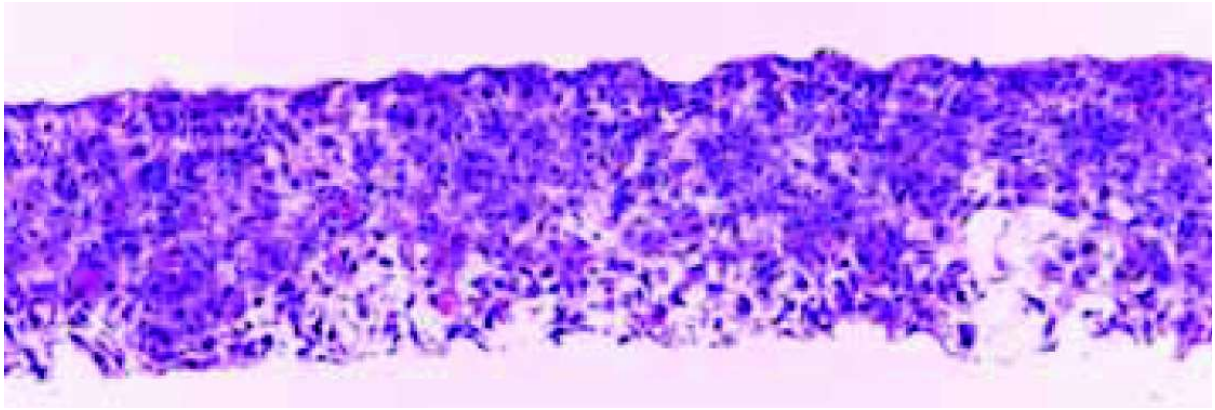
HaCaT cells (a human keratinocyte cell line) were seeded ($0.5x10^6$ cells in 150 µl per well) on EtOH-treated and complete medium washed alvetex® scaffolds in the following formats: 12-well plate (AMS.AVP002), 6-well inserts (AMS.AVP004-32) in 6-well plate and 12-well inserts (AMS.AVP005-34) in well insert holder in deep Petri dish (AMS.AVP015-2). Cultures were maintained for 7 days. [Complete medium consisted of: DMEM, 10% FBS, 2 mM L-glutamine and 100 U/ml Penicillin & Streptomycin]. After preserving in Bouins fixative the discs were paraffin embedded, sectioned (10 µm) and counterstained with Haematoxylin and Eosin.



12 well plate (AMS.AVP002)



6-well insert (AMS.AVP004-32) in a 6-well plate



12-well inserts (AMS.AVP005-34) in well insert holder

in deep Petri dish (AMS.AVP015-2)

Figure 2. *Comparison of 3D cell growth patterns of HaCaT cells grown on various alvetex® formats. Micrographs taken at 20x magnification.*

Note significantly more proliferation and cell invasion into the alvetex® scaffold in cultures grown in well-inserts, (media interconnected feeding regime) due to nutrient availability from above and below the scaffold. Thus, as media volume and availability increased so did cell proliferation and scaffold penetration. In the case of well inserts contained in a well insert holder in a deep Petri dish, this resulted in the formation of a slab of tissue-like material.