A007 Influence of Black Currant seed polysaccharides on normal human keratinocytes (NHK) and HaCaT

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Black Currant (*Ribes nigrum* L., Grossulariaceae) extracts are part of several pharmaceutical and cosmetical products because of the seed lipids and leaf anthocyanidines. No investigations have been done concerning Black currant seed polysaccharides.

As part of an examination on polysaccharide effects on normal human keratinocytes (NHK) and HaCaT cells Black Currant seed polysaccharides were isolated, purified and fractionated and subsequently tested on an influence on skin cell physiology on primary keratinocytes (NHK) and on a standarized keratinocyte cell line.

In vitro study parameters included quantification of the proliferation rate (determined by BrdU-incorporation), energy metabolism (determined by mitochondrial dehydrogenase), differentiation behaviour of NHK measured by involucrin- and keratin K1- and K10- synthesis, and direct cytotoxicity as derived from analysis of lactatedehydrogenase titers of keratinocytes supernatants.

Incubation of NHK and HaCaT cells with polysaccharides of Black Currant seeds enhanced proliferation rate and mitochondrial dehydrogenase activity significantly. No significant influence on differentiation parameters as involucrin and keratin K1 and K10 synthesis of NHK was observed. LDH titer measurement showed only basic cytotoxic effects in regard to untreated keratinocytes.

Seed polysaccharides might extend the spectrum of possible applications of Black Currant in cosmetic and dermatologic preparations or in cultures of keratinocytes because of their low cytotoxicity and their stimulating effects on proliferation and energy metabolism of keratinocytes.

A008 Influence of different mistletoe preparations on *in vitro* cell physiology of cancer cells

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Adjuvant cancer therapy with mistletoe extracts is mostly performed by evaluation of the respective cancer type and the status of the patient. In practice the receptiveness of the cancer cells to the mistletoe preparation is mostly not considered. For this reason a systematic investigation was performed to clarify if mistletoe preparations from different host trees have different influence on cell proliferation of different cancer cells in order to establish the first steps for an individualized adjuvant cancer therapy which could include an *in vitro* pre-investigation on receptiveness of the tumor cells against mistletoe preparations, followed by choosing the optimal preparation and concentration and then starting the therapy.

For this pre-investigation different cancer cell lines (HELA, MOLT-4, COR-L51, MFM-223, HEK-293, etc.) were treated with different commercial mistletoe preparations over a dose range between 1 and 1000 µg/ml. Subsequent determination of cell physiology parameters was performed (cell proliferation by cell counting and BrdU incorporation, mitochondrial activity, LDH levels, apoptosis induction). The investigation clearly proved that different mistletoe preparations influenced the cell proliferation and the mitochondrial activity of cancer cells to an different degree: maximum of cytotoxicity was shown for mistletoe originating from apple trees against MOLT-4 cells, while other cell lines were more susceptible to mistletoe from oak. Minor activity was shown for pine mistletoe against all cancer cells. It was shown that the apoptosis-inducing potential of the different mistletoe preparations correlates to the respective contents of lectin and viscotoxins.

In further studies similar investigations were performed on primary cancer cells from different cancer systems and the anti-proliferative effects of the different mistletoe preparations correlated to the cell status.

During these investigations it was shown that also the kind of *in vitro* cultivation may have an influence on the receptiveness of cells against the mistletoe: HEK-293 cells cultivated with serum supplements were influenced to a minor extend by mistletoe in the low-dose range (1 to 10 µg) compared to cells incubated in serum-free media.