# Investigation of FGFs Role in Human Keratinocyte Cell Line HaCaT Expressing H-RasV12 Oncogene

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We recently reported (Costa et al, 2008) that FGF2 (fibroblast growth factor 2) inhibit proliferation of Ras-dependent malignant mice cell lines by triggering senescence. To investigate this phenomenon in human cells, we transfected HaCaT (an immortalized non-tumorigenic human keratinocyte line that displays a functionally impaired p53 pathway) with the retroviral vector pBABEneo containing an insert coding for the fusion protein estrogen receptor biding domain + H-RasV12 (ER:RasV12), a construction activated by the estrogen agonist 4OH-tamoxifen (4OHT). A number of clonal HaCaT-ER:RasV12 sublines were isolated and proved to be sensitive to FGF2 and FGF1 toxicity only in the presence of 4OHT. However this phenotype was not stable because the ER:RasV12 sequence was not maintained by these HaCaT-ER:RasV12 sublines. In addition, HaCaT cells were also transfected with pDCR-RasV12, yielding clonal sublines that constitutively expressed the RasV12 oncoprotein, but were not sensitive to FGF2 and FGF1 toxicity. Thus, for still unknown reasons, HaCaT sublines expressing ER:RasV12 or RasV12 presented apparently contradictory responses to FGF2. To solve this and to determine the importance of p53 and pRb pathways, we are now testing other keratinocytes cell lines which express E6 and E7 HPV oncoproteins.

Key-words: FGF2, keratinocytes, H-RasV12, growth, senescence.
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