

13- cis- retinoic acid alters rate of apoptosis and extracellular matrix proteins in thyroid carcinoma cells under cultured microgravity

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Introduction

Several studies of our group have shown that culturing of human thyroid cancer cells at simulated microgravity (clinostat) induce apoptosis and increases expression of extracellular matrix proteins (Grimm et al. 2002 FASEB J. 16(6):604-6; Kossmehl et al. 2003 Endocrinology. 144(9):4172-9).

Retinoids are a group of natural and synthetic analogues of vitamin A. The therapeutic benefit of adding retinoids such as all trans retinoic acid (RA), 13-*cis*-RA to established single agent or combination immuno/chemotherapy regimens for the treatment of thyroid carcinoma cells were investigated.

Retinoids have been described to modulate apoptosis, extracellular matrix protein (ECMP) synthesis of fibronectin and laminin as well cell adhesion and migration.

Objective

The principal aim of this study was to investigate the effects of appropriate simulation of microgravity and treatment of retinoids in hormone producing human thyroid carcinoma cells.

Methods and Results

Clinorotation time was 24 h. We used short term (ST) treatment (RA) for 72h and long term treatment (RA) for 7 weeks before clinorotation at the final concentrations of 10⁻⁵ and 10⁻⁶ M. Immunofluorescence staining (Fig. 2) and flow cytometry (Fig. 3) were performed. Our data demonstrate that RA induces an inhibition of the production of collagen type I and III and as well as a decrease in apoptosis. 24h of simulated 0g induced a strong decrease of programmed cell death and ECMP (Fig. 3).

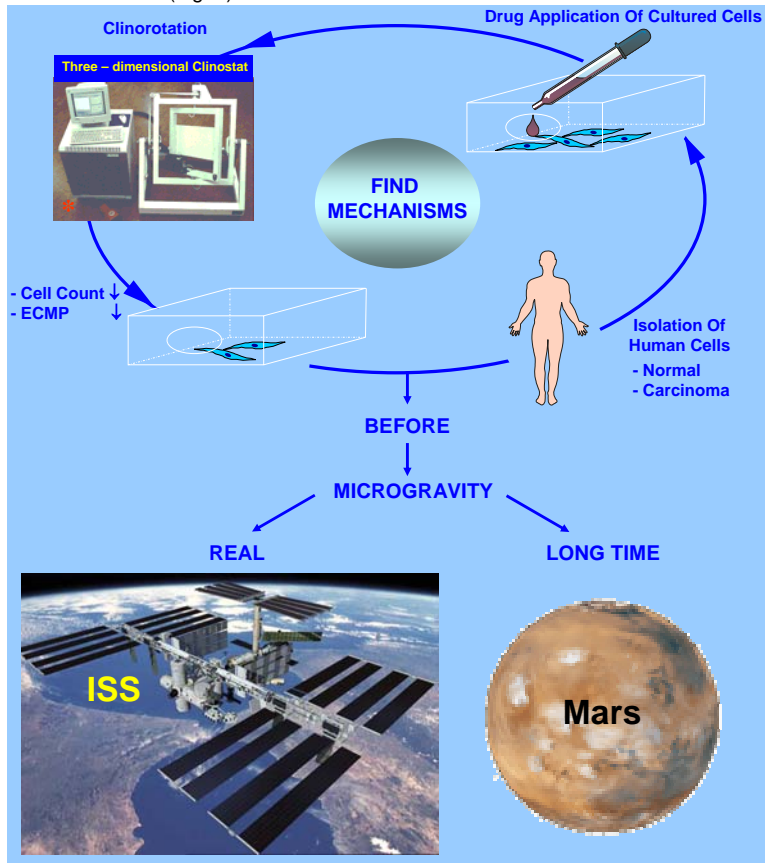


Fig. 1: Experimental pathway of the effects of microgravity on apoptosis and ECMP in combination with drug application and without animal tests.

*Clinostat: developed by T.Hoson et al. Bot Mag. 1992; 105:53. Manufactured by Dutch Space, Leiden, NL. Apparatus by A. Cogoli, ETH Zurich and Zero gLifeTec GmbH, Switzerland)

Conclusions

These findings indicate a new interplay between clinorotation and medical treatment. Moreover, these and further data could provide information leading to novel therapeutic or preventive programs for cancer utilizing retinoids that can offer an alternative in combination to the classic chemotherapy. In addition, our model helps to spare animal experiments. **Sponsored by DLR / Project number: 50 WB0524.**

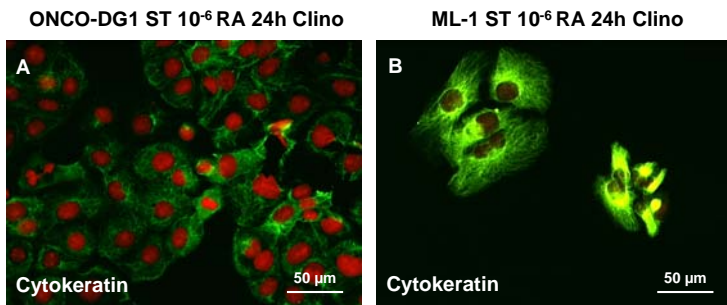


Fig. 2 Immunofluorescence : Cytokeratin (green) morphology was visualized using a Zeiss microscopy system. Propidium iodide staining (red) was observed concurrently.

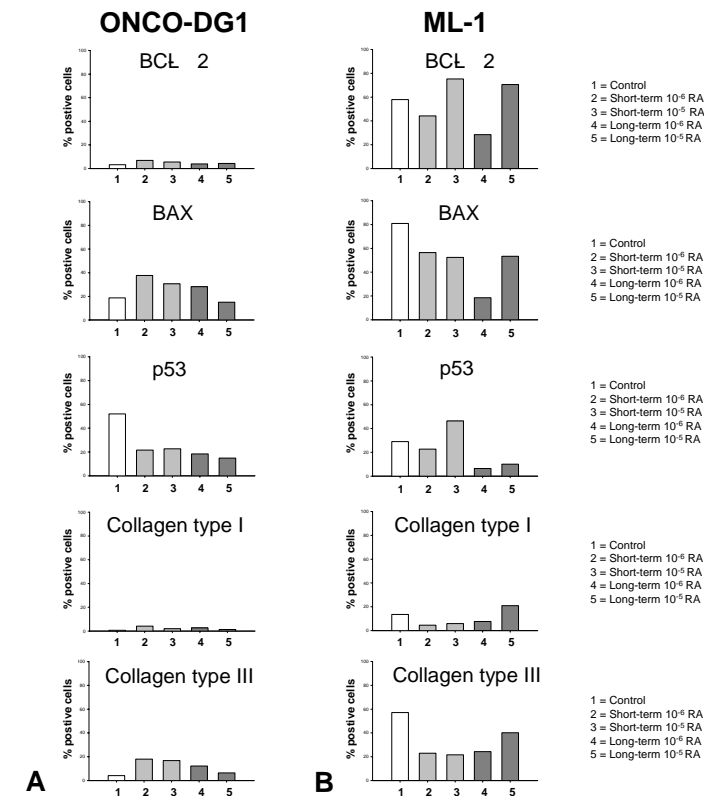


Fig. 3 Flow cytometry: Cellular antigens (A: ONCO DG1 and B: ML 1) were detected by flow cytometry by adding bcl 2bax, p53 and collagen type I and III to 10⁵ cells fixed in 70% ethanol using the antibody at saturation concentrations. The cell suspensions were analyzed with a Facscan flow cytometer (BD, Heidelberg, Germany) equipped with an argon laser. Cells exerting fluorescence intensities above the upper limit of the negative control distribution were considered positive.