GENE ACTIVITY AND TROPHOBLAST INVASIVENESS: DDRT-PCR-STUDIES. G. Huch, H.-P. Hohn, H.-W. Denker, Institute of Anatomy, University of Essen, Medical School, D-45122 Essen, Germany

Differentiation of human trophoblast during placentation is characterized by a reduction of invasiveness and acquisition of endocrine and transport functions. The majority of molecular mechanisms controlling this process, however, are still unknown.

In a systematic search for altered gene activity during trophoblast differentation we analyzed by DDRT-PCR (differential display RT-PCR) trophoblast cell types with different invasive potential.

In JAR choriocarcinoma cells, a malignant counterpart of human trophoblast, differentiation was induced experimentally with dibutyryl-cyclic AMP (cAMP) and phorbol-12-myristoyl-13 acetate (PMA): invasiveness is reduced by cAMP but enhanced by PMA. Normal human trophoblast cells were isolated from first trimester (high invasiveness) and term placentae (low invasiveness). Total RNA of treated and non-treated JAr and trophoblast cells was extracted (first trimester and term placentae) and used to perform DDRT-PCR.

Minor differences were seen in PCR-amplifications of JAr cells treated or not with cAMP or PMA. Considerable differences were detectable between invasive (first trimester) and "non"-invasive (term) trophoblast cells and between normal and malignant (JAr) trophoblast cells respectively. PCR-products representing major differences between the trophoblast cell types were amplified with the primers used in the DDRT-PCR and cloned in the vector pCRII (Invitrogen). Following sequencing and data base research (EMBL) one of the fragments isolated from DDRT-PCR-gels of first trimester placentae shows 98% homology to the ß1-integrin subunit. This molecule is known to be involved in the invasive pathway of the cytotrophoblast (Damsky et al.,1994).

These preliminary data suggest that altered gene expession (in addition to posttranscriptional regulation) can be detected by DDRT-PCR in invasive and noninvasive trophoblast cells. Further studies are performed to identify molecules involved in the control of differentiation and invasion of the human trophoblast. Altered gene regulation will be examined with northern blots and in situ hybridisations.

Supported by Dr.Mildred Scheel Stiftung für Krebsforschung