Human Embryonal Tissues of all Three Germ Layers can Express the CD30 Antigen. An Immunohistochemical Study of 30 Fetuses Coming after Therapeutic Abortions from Week 8th to week 16th of Gestation

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Summary

Originally, expression of the CD30 antigen was shown to be typical of the tumor cells of Hodgkin disease and of anaplastic large cell lymphomas. In reactive lymphoid tissue, CD30 is expressed only in a small population of activated lymphoid blasts. Since then, several reports have been published describing CD30 expression in non lymphoid tissues and neoplasms, such as embryonal carcinomas, seminomas, cultivated macrophages, histiocytic neoplastic cells, deciduals cells, and mesothelioma cells. In order to gain insight into the functions of CD30, given that it can mediate signals for cell proliferation and apoptosis, we studied the distribution of the antigen in different fetal archival paraffin-embedded tissues from week 8th to 16th of gestation.
We investigated the immunohistochemical expression of CD30 in 30 paraffin-embedded tissue samples representing all three germ layers, using the monoclonal antibody Ber-H2
CD30 is expressed early in human fetal development (8th–10th week) in a wide variety of tissues, with the exception of the skin and thymus in which it is expressed later on. This is consistent with the observation that these organs are not fully differentiated before 10th and 13th week, respectively. No expression was observed in the cardiovascular and respiratory systems.
The finding of CD30 expression in the terminal period of organogenesis, period, which is highly hormone related, implies that the antigen has an important role in cell development, maturation, and pathway to terminal differentiation in almost all fetal tissues and structures.

Key words: CD30 antigen – fetal tissues – 8th–16th week of gestation

Souhrn

Lidské embryonální tkáně všech tří zárodečných listů mohou exprimovat CD30 antigen. Imunohistochemická studie 30 plodů z léčebných potratů v 8.–16. týdnu gestace

Exprese antigenu CO30 byla považována za typickou pro nádorové buňky Hodgkinovy nemoci a anaplastického velkobuněčného lymfomu. V reaktivní lymfatické tkáni je CD30 exprimován pouze aktivovanými lymfoblasty. V poslední době se však množí zprávy o expresi CD30 nelymfatickými tkáněmi a nádory, jako jsou embryonální karcinom, seminom, kultivované makrofágy, nádorové histiocty, buňky deciduy, a buňky mezoteliomu. Vzhledem ke skutečnosti, že CD30 může zprostředkovávat signály pro buněčnou proliferaci a apoptózu, sledovali jsme pomocí monoklonální protilátky Ber-H2 distribuci tohoto antigenu v archivních parafinových bločcích tkání všech tří zárodečných listů plodů stáří 8.–16. týdnu gestace.
CD30 je exprimován již v časném fetálním období (8.–10. týdnu) v nejrůznějších tkáních orgány, s výjimkou kůže a thymu, v nichž dochází k expresi později. To odpovídá skutečnosti, že tyto nejsou

před 10., resp. 13. týdnem ještě diferencovány. V kardiovaskulárním a dýchacím systému jsme expresi neprokázali.
Závěr: Průkaz exprese CD30 v období končící organogeneze, které je výrazně hormonálně závislé, ukazuje, že tento antigen hraje důležitou roli v buněčném vývoji, vyzrávání a přechodu k plné diferenciaci téměř všech fetálních tkání a struktur.

Klíčová slova: CD30 antigen – fetální tkáně – 8.–16. týden gestace

Čes.-slov. Patol., 42, 2006, No. 1, p. 9–15

The CD30 antigen is a 120 kD cytokine receptor which belongs to the tumour necrosis factor receptor (TNFR) superfamily (1, 2). Currently, it is known that the expression of CD30 antigen is not limited to lymphoid tissue and lymphoproliferative disorders. For instance, strong CD30 expression has been documented in embryonal carcinoma and human decidual cells (3-5). Cytoplasmic Ber-H2 immunohistochemical staining has been reported in occasional carcinomas, many sarcomas, and vascular tumours (3). In addition, Ber-H2 immunohistochemical staining can be demonstrated in pancreatic tissue (3), some salivary gland tumours, and normal salivary gland tissue (3). The fact that the CD30 molecule can mediate signals for cell proliferation or apoptosis (6) prompted us to perform a systematic investigation of the antigen distribution in embryonal tissues using immunohistochemistry, from week 8^{th} onwards, in an effort to uncover patterns of expression that may elucidate the potential role of the marker during development stages.

MATERIALS AND METHODS

Tissue procurement

The tissue material (30 fetuses) used in this study was obtained from the files of the Department of Histology – Embryology at the University of Thrace. Samples representing a wide variety of tissues from all systems were collected from 30 fetuses: 15 males and 15 females (15 at 8^{th} to 10^{th} week of gestation and 15 at 12^{th} to 16^{th}, respectively) after therapeutic abortion. The organs used did not show any evidence of morphological abnormality. The Regional Ethics Committees approved the study. Written informed consent was obtained from all individuals and the procedures followed accorded with institutional guidelines.

Sections of tissue roughly 3-mm thick were fixed in 10% buffered formaldehyde for 7 hours then subjected to routine processing and paraffin embedding. Slides were obtained in all cases, and stained with hematoxylin-eosin (H-E), PAS, Giemsa and Gomori for morphological evaluation.

Monoclonal antibody and immunohistochemical staining

Antigen retrieval from formalin-fixed, paraffin-embedded tissue was performed by heating unstained sections immersed in DAKO Target Retrieval Solution (DAKO, Carpintera, CA) according to the manufacturer's instructions. A modified labeled avidin-biotin immunohistochemical staining was performed with the use of the LSAB-2 System Peroxidase Kit (DAKO) on DAKO Autostainer, according to the manufacturer's instructions. In short, deparaffinized sections were incubated with 3% hydrogen peroxide for 5 min., followed by 10-min. incubation with 1:20 solution of Ber-H2 MAb (Novocastra Laboratories Ltd., Newcastle, UK). That was followed by sequential 10-min. incubations with a biotinylated link antibody and peroxidase-labeled streptavidin. Staining was completed after a 10-min. incubation with DAKO Liquid 3,3'-diaminobenzidine Substrate-Chromogen System utilizing 3,3'-diaminobenzidine (DAB) chromogen. Biopsied lymph nodes of Hodgkin's disease were used as controls. All cases were coded, and the grading of the immunostaining was performed on a sliding scale of 1+ to 4+ according to the percentage of reactive cells (0 = no staining, 1+ = 1% to 25%; 2+ = 26% to 50%; 3+ = 51% to 75%; 4+ > 75%) Staining intensity was not the same in each one case. The scores represent the immunostained cells observed in the majority of cases.

RESULTS

Five microscopic fields of each tissue were evaluated in each case. The results of the immunostaining are summarized in table 1. Two observers examined the sections independently, and positive cellular staining was manifested as