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Epithelial Cells and Macrophages of Aged Human Thymus Possess IGF-I Immunoreactivity

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The presence and distribution of insulin-like growth factor I (IGF-I) immunoreactivity in the aged human thymus were investigated both at light and electron microscopic levels. IGF-I immunoreactive cells were observed in the structurally preserved regions of the chronic involuted thymus. Presenting novel data for presence of IGF-I immunopositive epithelial cells and macrophages, we conclude that the aged human thymus is still capable to govern some "beneficial" microenvironment events, including IGF-I signalling mechanisms. The latter might be involved in the local regulation of T cell development and plasticity of thymocytes-epithelial cells interactions during aging.

Key words: IGF-I immunoreactivity, human thymus, chronic involution.

Introduction

Accumulating evidence shows that adult mammalian thymic cells express insulin-like growth factor I (IGF-I) immunoreactivity [1, 2]. Astonishingly, despite the generally acknowledged roles of IGF-I in the ontogeny [1, 7, 8], generation and survival of T-cells [3, 6, 10], little is known about the exact time course of IGF-I occurrence during the age-related thymic involution and the decline of immunoreactivity [4, 5, 9].

This is why, in the present study we concentrated our efforts to perform a detailed temporo-spatial analysis of IGF-I expression in the aged human thymus.

Material and Methods

Specimens from thymuses of old (aged 66-82 years) (n=14) and young (aged 2-27 years) (n=10) individuals were obtained from autopsy and thoracic surgery cases, and examined immunocytochemically at light and transmission electron microscopic level. The thymuses collected have had no pathological disorders. Three kinds

of antibodies (Ab), namely: Anti-human monoclonal IGF-I Ab (UBI/Biomol, Hamburg, Cat. Nr. 05-172); Anti-Pan cytokeratin monoclonal Ab (C 1801, Sigma Chemical Co.) and Anti-CD 14 monoclonal Ab (UCH-M1, sc-1182, Santa Cruz Biotechnology) were used. The immunoreactivity of IGF-I, cytokeratin and CD14 was studied.

Indirect immunoperoxidase staining, immunogold transmission electron microscopy and immunogold-silver staining procedures were applied [9, 11]. To define the nature of the thymic cell types which expressed IGF-I we stained serial tissue sections with Anti-cytokeratin Ab and Anti-CD14 Ab which reacted with epithelial cells and monocyte/macrophages, respectively (according to the manufacturer's instructions). Control experiments (negative and positive controls) were carried out in parallel. Labomikroscop Axioskop 20 (Fb Carl Zeiss Opton) and electron microscope Hitachi H500 were used.

Results

Thymuses from young individuals showed lobulated structure, distinct corticomedullary junction and prominent Hassall's corpuscles in the medulla. Aged thymuses displayed a large mass of adipose tissue containing scattered islands composed of epithelial cells, lymphocytes and reticular connective tissue. Most of the epithelial cells were organized into a framework that provided support for lymphoid cells.

Young thymus. All types of medullary epithelial cells, some subcapsular epithelial cells and macrophages displayed strong IGF-I immunoreactivity. Especially those medullary epithelial cells that were located in close proximity to the Hassall's corpuscles were very strongly positive (Fig. 1). They possessed IGF-I immunoreactivity with granular appearance diffusely distributed within the cytoplasm.

Aged thymus. IGF-I immunoreactivity was also present in the adult thymus. However, the immunocytochemical data showed a decreased amount of immunopositive epithelial cells and macrophages an attenuated IGF-I expression (Figs. 2, 3, 4). The cytoplasm of epithelial cells was only moderately immunopositive. The

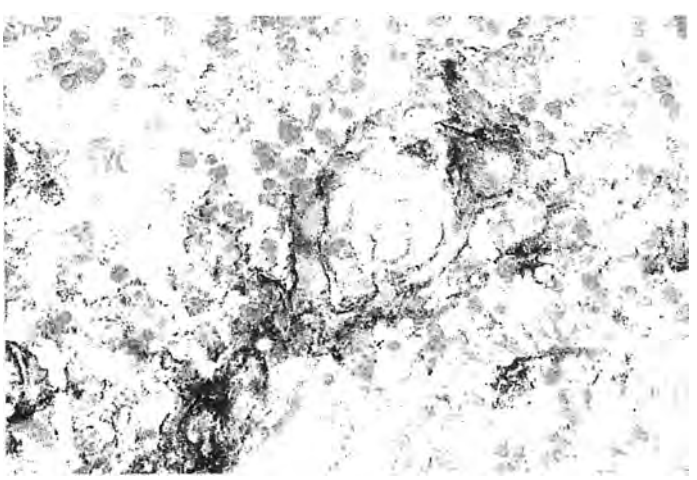

Fig. 1. Young human thymus (21 years old male) — strong IGF I-immunopositive medullary epithelial cells, immunoperoxidase staining, Mayer's hemalaun counterstaining). × 1000

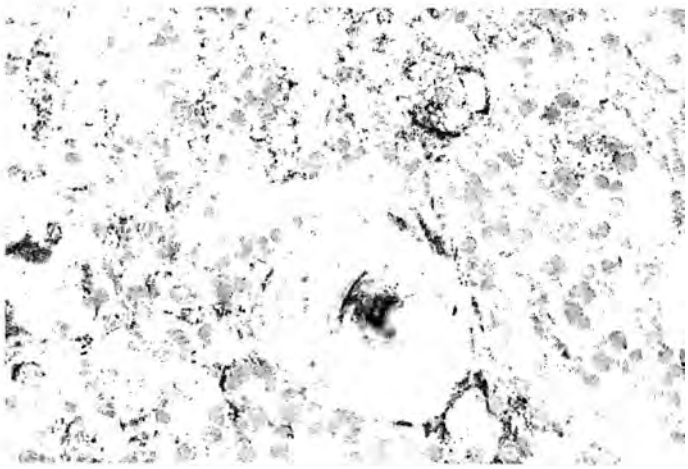

Fig. 2. Aged human thymus (69 years old male)-attenuated IGF I immunoreactivity of medullary epithelial cells around to Hassall's corpuscle; immunoperoxidase staining; Mayer's hemalaun counterstaining. × 1000

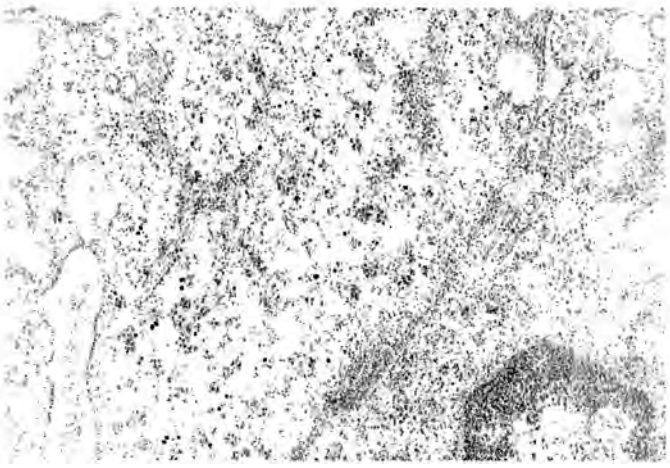

Fig. 3. Aged human thymus (69 years old male) — IGF I-gold granule complexes with cytoplasm localization in part of immunopositive medullary epithelial cell; immunogold transmission electron microscopy. × 26 000

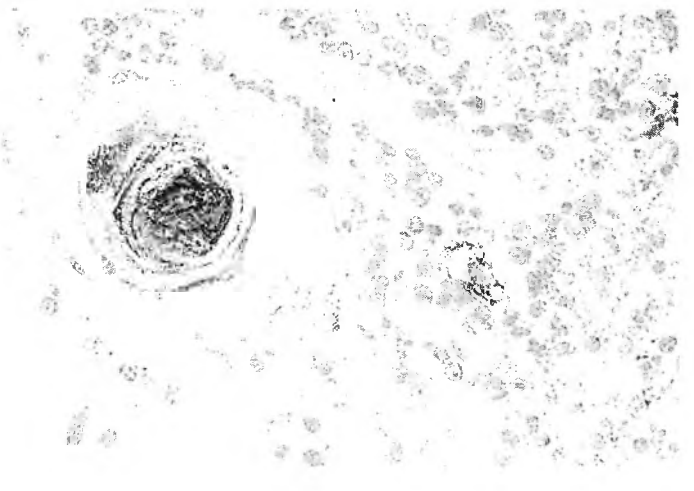

Fig. 4. Senile human thymus (77 years old male) — single IGF I-immunopositive medullary epithelial cells and Hassall's corpuscle with central immunopositive part; immunoperoxidase staining; Mayer's hemalaun counterstaining. × 1000

labelling intensity of the Hassall's corpuscles was heterogeneous. The detailed analysis by electron microscopy showed cytoplasm localization of the IGF I-gold granules complexes in the thymic epithelial cells and macrophages.

Discussion

The present study is the first attempt to investigate the presence and distribution of IGF-I immunopositive epithelial cells and macrophages at both light and electron microscopic levels during the chronic involution of human thymus. Our results are in good correlation with literature data about the immunocytochemical characteristics of animal thymic cells [1, 3, 10].

We found that the age-involuted human thymus retains IGF-I immunoreactivity in its structurally preserved regions. Most probably, the aged human thymus is still capable to govern some "beneficial" microenvironment events, including IGF-I signalling pathways [2, 7] that might be involved in the local regulation of T cell development and in the plasticity of thymocytes-epithelial cells interactions during aging.

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