Annals of Anatomy xxx (2012) xxx–xxx

Contents lists available at SciVerse ScienceDirect

Annals of Anatomy

journal homepage: www.elsevier.de/aanat

Research article

Morphometric analysis of the human anterior pituitary's folliculostellate cells during the aging process

Miljana Pavlović[a], Ivan Jovanović[a,*], Slađana Ugrenović[a], Ljiljana Vasović[a], Miljan Krstić[b], Mirjana Bakić[c], Vladimir Živković[a], Vesna Stojanović[a]

[a] University of Niš, Medical Faculty, Department of Anatomy, Blvd. Dr Zoran Djindjić 81, 18000 Niš, Serbia
[b] University of Niš, Medical Faculty, Department of Pathology, Clinical Center Niš, Blvd. Dr Zoran Djindjić 48, 18000 Niš, Serbia
[c] University of Podgorica, Medical Faculty, Department of Anatomy, Kruševac bb, 81000 Podgorica, Montenegro

ARTICLE INFO

Article history:
Received 19 October 2012
Received in revised form
10 November 2012
Accepted 13 November 2012

Keywords:
Folliculostellate cells
Aging
Morphometry

ABSTRACT

Folliculostellate cells represent non-endocrine cells of the anterior pituitary which influence the function of the endocrine cells via paracrine action. Though there is a lack of literature data on their presence during human aging, the aim of this research was to perform the quantification of anterior pituitary folliculostellate cells by the application of immunohistochemical and morphometric methods. The material for the study consisted of 15 anterior pituitaries taken from cadavers at routine autopsy. Their tissue was processed by standard histological procedure and the obtained histological slices were stained by S100 polyclonal antibody. Digital images of stained histological sections were analyzed by morphometric method with ImageJ system. The volume density of S100 positive cells was measured for each case. Results of morphometric and statistical analysis showed a significantly positive correlation between folliculostellate cell volume density and the age of the evaluated cases. Linear regression additionally showed that the age significantly predicts folliculostellate cells volume density in our sample. Further, all cases were classified into three age groups and One Way ANOVA showed that the volume density of folliculostellate cells was significantly higher only in the third age group in relation to the first and the second group, respectively. Volume densities of the first and the second age groups were not significantly different. So, the results of our study pointed to the conclusion that folliculostellate cells presence generally increases with age, but this increase is significant only in the oldest cases and might represent the modality of successful anterior pituitary aging.

© 2012 Elsevier GmbH. All rights reserved.

1. Introduction

Agranular, non-endocrine, stellate-shaped cells, which may also form the lining of follicle-like cavities are named, on the basis of their appearance, folliculo-stellate (FS), or follicular cells (FC) and, were mostly considered to be supporting structures for the endocrine cells. It is certain that these cells do not produce any of the established hormones, but neither their origin nor their functional significance is well understood (Bilezikjian et al., 2003; Allaerts and Vankelecom, 2005; Yamashita et al., 2005; Acosta et al., 2010).

Researchers supposed that these cells could be involved in trophic and catabolic processes and in macromolecular transport. The majority of the earlier performed studies indicated that FS cells might serve to modulate hormone production and secretion in the anterior pituitary via local paracrine actions (Vankelecom et al., 1997; Acosta et al., 2010). Interdigitations between them and hormone-producing cells might support the presence of such intercellular communication. Further, it was found that they could be a source of several growth factors and peptides such as basic fibroblast growth factor, vascular endothelial growth factor, follistatin and activin A, lipocortin 1 as well as of cytokines such as leukemia inhibitory factor, interleukin-6 and macrophage migration inhibitory factor (Soji et al., 1994; Bilezikjian et al., 2003; Allaerts and Vankelecom, 2005). Moreover, Bilezikjian et al. (2003) postulated that these cells have been reported to respond to a number of factors such as TGFβ, activin, pituitary adenylatecyclase-activating polypeptide (PACAP), calcitonin, IL-1β, IL-6, glucocorticoids and lipopolysaccharide (LPS), which probably exert effects on the anterior pituitary endocrine cells by the modulation of the FS cells paracrine factors production.

More recent works suggest a role for FS cells in reciprocal communication between the immune and endocrine systems

* Corresponding author. Tel.: +381 692351980; fax: +381 184238770.
E-mail addresses: pavlovic_miljana@yahoo.com (M. Pavlović), ivanjov@sbb.rs (I. Jovanović), sdugren@sbb.rs (S. Ugrenović), likica@medfak.ni.ac.rs (L. Vasović), krstic.miljan@gmail.com (M. Krstić), mirjana.bakic@gmail.com (M. Bakić), vladimir029@gmail.com (V. Živković), vesna@medfak.ni.ac.rs (V. Stojanović).

0940-9602/$ – see front matter © 2012 Elsevier GmbH. All rights reserved.
http://dx.doi.org/10.1016/j.aanat.2012.11.002

Please cite this article in press as: Pavlović, M., et al., Morphometric analysis of the human anterior pituitary's folliculostellate cells during the aging process. Ann. Anatomy (2012), http://dx.doi.org/10.1016/j.aanat.2012.11.002

(Herkenham, 2005). FS cells act as phagocytes of both cellular debris and apoptotic bodies, which support kinship between them and cells of the mononuclear phagocytic system (Vankelecom et al., 1993). FS cells respond to β-adrenergic stimuli and display enhanced cAMP-accumulation upon it. Their production of the radical nitric oxide (NO) brought them into the limelight of the pituitary hormone secretion regulation through the role of NO (Allaerts and Vankelecom, 2005).

Older studies have established that FS cells form gap junctions mutually, which allow synchronized excitability within and between them via propagated Ca^{2+} currents, suggesting a mechanism by which long-distance communication in the anterior pituitary could occur via this electrically coupled network and, consequently, pulsatile hormone release even in denervated pituitaries (Soji et al., 1994; Bilezikjian et al., 2003; Herkenham, 2005). New hormone-secreting cells might be produced either by mitosis of existing differentiated cells and, to a greater extent by possible maturation of undifferentiated stem cells. It has more than once been suggested that FS cells could represent local anterior pituitary stem cells (Bilezikjian et al., 2003; Vankelecom, 2007). The finding of thyrotropin (TSH) receptors expressed in a subpopulation of FS cells opened a possibility of ultra-short feedback loop presence for the regulation of TSH secretion by FS cells (Allaerts and Vankelecom, 2005).

The origin of these non-hormone secreting anterior pituitary interstitial cells, according to their markers expression might be neuroectodermal and also possibly be glial in nature. On the other hand, their phagocytic capacities, IL-6 secretion, and the fact that they share some immunohistochemical, ultrastructural and functional characteristics with lymphoid dendritic cells, suggests that probably their subpopulation could be derived from the monocyte-dendritic cells-macrophage lineage (Allaerts et al., 1997).

In spite of the fact that the anterior pituitary is very important for the homeostasis maintenance of the human body, studies dealing with its structural changes during the aging process are very rare. Sano et al. (1993) described some of such changes in their paper. They evaluated age related changes of the anterior pituitary connective tissue and endocrine cells using histological and semi-quantitative methods. However, there is a lack of literature data on the aging changes of FS cells. Studies evaluating this problem were performed mostly on animal material (Weiss and Lansing, 1953; Sasaki, 1988; Allaerts et al., 1997; Cónsole et al., 2000). The latter cited facts and the significance that FS cells might have for the endocrine cell function, which, based on the literature data, lead us to perform their quantification in the anterior pituitary samples at different ages and in an indirect way to try to establish their possible impact on anterior pituitary functioning during human aging.

2. Materials and methods

Pituitary glands were removed from 15 cadavers (7 males and 8 females), during routine autopsies performed at the Department of Forensic Medicine at Medical Faculty in Niš, according to the guidelines of the faculty's Internal Ethical Committee. The age of the cadavers ranged from 44 to 89 years. Pituitary tissue was sampled within 12 h after the time of death. There were no data of previously diagnosed neurological or endocrine disorders in medical documentation on the respective cadavers. Also, during the autopsies, no visible brain damage was observed. Pituitary glands were then cut into horizontal sections which included the tissue of both anterior and posterior lobes. These sections were fixed in 10% buffered formalin for 24 h and then embedded in paraffin. Afterwards, 5 μm thick tissue sections were made and stained with polyclonal S100 antibody (Dako, polyclonal rabbit anti-cow S-100; code no. Z0311).

3. Immunohistochemistry

The immunohistochemical procedure included the deparaffinization of tissue sections with xylene (2× 5 min) and hydration in decreasing concentrations of ethanol (100% 2× 2 min, 96% 2 min and 70% 2 min) up to distilled water. Antigen retrieval was performed by proteinase K digestion for 10 min at room temperature. After incubation for 10 min in a solution of 3% H_2O_2 in water in order to inhibit endogenous peroxidase activity, sections were washed in distilled water (5 min) and (3× 10 min) in phosphate-buffered saline (PBS, 0.1 M, pH 7.2). They were further incubated with the primary antibodies (1:400). The slides were subsequently washed (3× 3 min) in PBS and then incubated for 30 min at room temperature inside a wet chamber with biotinylated anti-rabbit IgG secondary antibody (Novocastra, UK). After that they were washed (3× 3 min) in PBS. Novocastra™ streptavidin-peroxidase conjugate (Streptavidin HRP, Novocastra Peroxidase Detection System, Novocastra, UK) was applied for 30 min in a wet chamber and it bound to the biotin present on the secondary antibody. Sections were further incubated with the AEC+ substrate (chromogen, 3,3′-diaminobenzidine – DAB) 3–5 min at room temperature, washed in running water, counterstained with Mayer's hematoxylin for 1 min, washed again 10 min in running water, dehydrated in increasing graded ethanol (70%, 96% i 100%), cleared in xylene and mounted with DPX mounting media. To confirm the specificity of the immunoreactions, omission of primary antibody was performed. No positive structures or cells were found in these sections. Stained sections were histologically analyzed with light microscope under the 10×, 40× and 100× lens magnifications.

4. Morphometric analysis

Morphometric analysis was performed on single S100 stained anterior pituitary histological sections in each of all 15 evaluated cases. Forty fields of vision of each of stained section (case) were randomly selected (approximately 10 at the level of intermediate and 15 at the level of each of its two lateral parts) and captured with digital camera under the 100× lens magnification. The 24 bit RGB images obtained were then processed and analyzed with ImageJ software (http://rsb.info.nih.gov/ij/) according to the following procedure: images were firstly split into the red, green and blue 8 bit channels; S100 positive regions were the most clearly distinguishable from other anterior pituitary's tissue components on blue channel's images and this channel was used for the image segmentation; image segmentation resulted in a binary images in which S100 positive zones were represented by black pixels. FS cells volume density (V_{VFSC}) represents their volume in the unit of volume of the anterior pituitary tissue (Russ, 2004). Its measurement was performed, according to the latter cited author, by measurement of S100+ zones area fraction in obtained 2D binary images. FS cells volume density for a single case was obtained as the mean value of its 40 analyzed fields of vision volume density.

5. Statistical analysis

Statistical analysis was performed with NCSS – PASS 2007 statistical package. It included linear correlation and linear regression analysis in order to establish the dynamics of FS cell volume density during the aging process. More detailed analysis of the age related FS cell volume density changes included classification of the cadavers into three age groups: I (40–59 years old), II (60–79 years old) and III (80 years and older cases). Significance of the differences between analyzed age groups was evaluated with One Way ANOVA. Mutual differences between age groups were additionally established with Tukey–Kramer post hoc test. Gender differences were

Please cite this article in press as: Pavlović, M., et al., Morphometric analysis of the human anterior pituitary's folliculostellate cells during the aging process. Ann. Anatomy (2012), http://dx.doi.org/10.1016/j.aanat.2012.11.002

Download English Version:

https://daneshyari.com/en/article/8461372

Download Persian Version:

https://daneshyari.com/article/8461372

Daneshyari.com