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Original Article

Alpha-chlorohydrin effects on the epididymis of adult albino rat: A histological and immunohistochemical study

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ABSTRACT

α-Chlorohydrin (ACH) is a well-known food contaminant, has shown anti-fertility activity in males. In this research we studied the histological and immunohistochemical changes in the epididymis of adult albino rat. Twenty adult male albino rats were divided into control (group 1) (10 rats) and experimental (groups 2 and 3) (10 rats). The experimental rats received α-chlorohydrin at a single oral dose of 100 mg/kg body weight. After 6 h (group 2) and 24 h (group 3) of ACH treatment, the specimens were submitted for the light, electron microscopic and β-tubulin immunohistochemical evaluations. Light microscopy after 6 h showed focal separation of lining epithelium, while EM examinations showed dilated rER, and Golgi complex. After 24 h LM demonstrated different stages of cell exfoliation; however, EM proved multilamellar bodies and dilated rER besides the granules. Immunohistochemical examinations revealed decreased reactions in experimental groups. However, the mean total optical density of the β-tubulin immunostaining revealed statistical nonsignificant difference between the three groups. Thus ACH as an undesirable unavoidable food contaminant induced rapid histological alterations on caput epididymis with duration-dependent manner at the level of the cell and tissue, that puts another obstacle on the road of fertility.

1. Introduction

α-Chlorohydrin [3-chloro-1,2-propanediol, ACH], is an organic chemical compound which is carcinogenic, highly suspected to be genotoxic in humans and has male anti-fertility effects [1]. α-Chlorohydrin also has harmful effects in many organs such as testis, kidney, liver and brain. ACH or [3-monochloropropane-1,2-diol or 3-chloropropane-1,2-diol] crosses the blood testis barrier and the blood brain barrier and distributed widely in the body fluids [2].

It is primarily created in foods by protein hydrolysis by adding hydrochloric acid to speed up the reaction of the [soya] protein with lipids at high temperatures, in foods contact with materials containing epichlorohydrin- as used in the production of some tea bags and sausage casings. A wide range of household food products from sliced bread to crackers, beefburgers, popular brands such as Mother's Pride and cheese are found with [ACH] above safe limits [3].

The epididymis is the site of accumulation and storage of spermatozoa of mammals. It is an excellent target for the development of a male contraceptive. This is because the process of sperm maturation occurs in this organ; the immature spermatozoa undergo changes in the morphology, surface properties and biochemical composition during epididymal transit then, spermatozoa become

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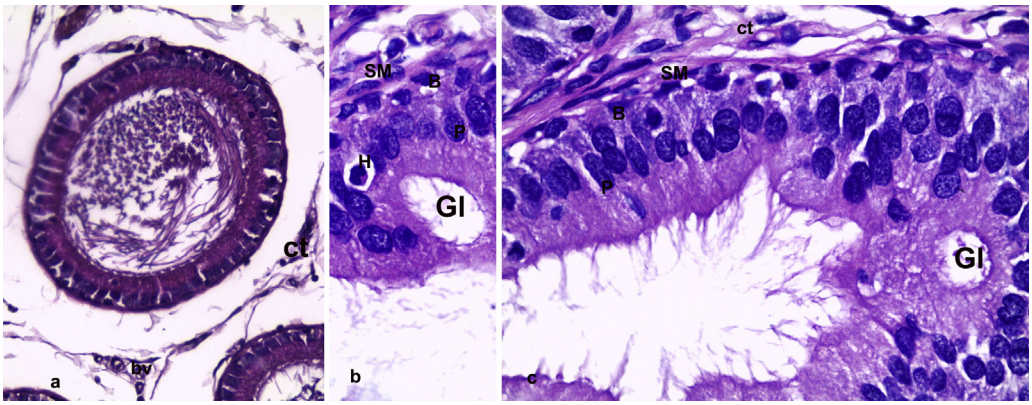

Fig. 1. Photomicrographs of the control epididymis caput tubules appear as tubules of smooth muscle (SM) separated by intertubular connective tissue (ct) with blood vessels (bv), lined by a tall pseudostratified columnar epithelium and filled with spermatozoa. There are short basal pyramidal cells (B) and tall principal cells (P) exhibited apical stereocilia. Intraepithelial glands (Gl) and hallo cells (H) are noticed. H&E, Mic. Mag. ×400 (a), ×1000 (b and c).

motile and are able to recognize and fertilize an egg once they traverse the epididymal duct. These changes depend on the epididymal microenvironment which is formed by the absorptive and secretory functions of the epididymal lining epithelium [4–6].

The epididymal epithelial cells were versioned into four major cell types: principal, basal, clear, and halo cells. The narrow cells were added by Robaire et al. [7]. Moreover Domeniconi et al. [8] furthered two other cell types to that categorization; apical cells and dark cells. The migratory cells demonstrated crossing the epithelium, as intraepithelial lymphocytes and macrophages are thought to be component of the lining epithelium [9].

Principal cells [PCs] are the most abundant cells and play a major role in secretion and absorption [10]. Tight junctions between principal cells form the blood–epididymis barrier [5,6]. This barrier, in prolongation with the blood–testis barrier, is critical for the avoidance of autoimmune responses against antigenic germ cells [12]. Basal cells are flat elongated cells that may have a defensive role, both by detoxifying electrophiles [8,9] or by acting like macrophages [11]. It is believed that the basal cells function as stem cells, regenerating themselves as well as the other cells as the need arises [4]. Clear cells participate in the uptake of luminal components and the discarding of cytoplasmic droplets detached from spermatozoa [12].

The halo cells are rounded or irregular in shape with dark, small nuclei surrounded by a light halo of pale cytoplasm. Halo cells have been postulated to be lymphocytes or monocytes and are supposed to play a role in the immunological barrier of the male reproductive duct [13]. The apical cells were found to be fewer in number than any other cell type. They have a wide apical portion with apically located spherical nucleus and narrow stem not extending to the basal lamina [7]. Dark cells [DCs] appear among the PCs as narrow, tall, and darkly stained cells, extending from the basement membrane to the lumen [7]. The narrow cells are narrower than the principal cells, attenuated, and send a thin process of cytoplasm to reach the basement membrane. They are characterized by numerous apically located cup-shaped vesicles that are

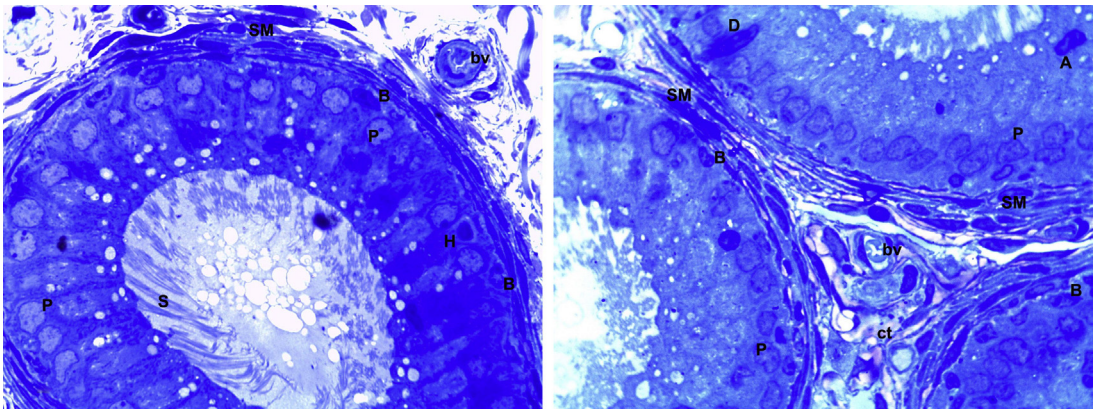

Fig. 2. Photomicrographs of semithin sections for control group show tubules of smooth muscle (SM) cells, regularly arranged cells of the lining pseudostratified epithelium. Basal cells (B) appear as pyramidal cells with dense nuclei. The principal (P) cells are tall cells, have irregular, oval basal nuclei with one or two nucleoli, paler than those of the basal cells, apical stereocilia (S) and noticeable vacuoles. Hallo (H) cell with its narrow rim of clear cytoplasm, apical (A) cell with its characteristic apically located spherical nucleus and dark (D) cell with its dark, elongated fusiform nuclei are forcibly defined. Connective tissue (ct). Blood vessels (bv). Toludine blue, Mic. Mag. ×1000.