In situ characterization of glycans in the horse bladder urothelium

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The urinary bladder cavity is lined by a highly specialized epithelium, the urothelium, which prevents permeation of solutes and noxious agents back into the bloodstream and underlying tissues, serving also as a sensor and transducer of physiological and nociceptive stimuli [1]. In addition, in physiological conditions the urothelium can also function as a secretory tissue [2]. A mucous layer protects the urothelium of the urinary bladder from potentially harmful environmental substances, attachment of bacteria and proteolytic enzymes present in the urine [3]. Moreover, the glycan composition of urothelial mucous layer could have a role in the intravesical pharmacological treatments [4]. Despite their considerable importance in urinary bladder physiology, few studies are available about the glycoconjugates expressed in non-human species. In this study the glycoconjugate pattern of urothelium lining the horse urinary bladder was investigated. Tissue fragments from three horse stallions in good health status, aged 2.5-4 years, were fixed in 4% (w/v) PBS-buffered paraformaldehyde, embedded in paraffin wax and stained with a panel of twelve lectins, in combination with saponification and sialidase digestion (Ks). The urinary bladder urothelium has three distinct layers from the basal zone to the lumen consisting of basal, intermediate and superficial cells (umbrella cells). Cytoplasm of basal cells showed glycans ending with Neu5AcGal1-3GalNAc, GlcNAc and with terminal/internal Man (Ks-PNA, GSA II, and Con A II reactivity). A sub-population of intermediate cells also displayed terminal Neu5Ac2-6Gal/GalNAc, NeuNacα2-3Galβ1-4GlcNAc, Gal1-3GalNAc, Gal, terminal and sialic acid-linked GalNAc, internal GlcNAc and Fucα1-2Galβ1-4GlcNAc (MAL II, SNA, PNA, GSA I-B_4, SBA, ks-SBA, Ks-WGA, and UEA I reactivity). The cytoplasm of umbrella cell population contained all the above cited sugar residues. Moreover, LTA-reactive fucosylated glycans and Ks-DBA-positive sialoderivatives were found in some scattered umbrella cells. These sialoglycans were secreted in the bladder lumen. The bladder luminal surface stained with MAL II, SNA, PNA, Ks-PNA, and GSA I-B_4 displaying a coating of sialo- and galactose-terminating glycoconjugates. These findings show that different glycosylation patterns exist along the horse bladder urothelium, and different sub-populations of umbrella cells are present secreting the sialoglycans which constitute the protective gel layer lining the bladder. Compared to results of a similar study carried out onthe donkey bladder urothelium [2], the present research reveals a species-specific glycan pattern and could contribute to a better understanding of the differences between domesticated odd-toed ungulate mammals via comparative glycopattern investigation.

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