

## **The chemical coding of zinc-enriched neurons in the intramural ganglia of the porcine jejunum.**

Joanna Wojtkiewicz<sup>1</sup>, Maciej Rowniak<sup>2</sup>, Robert Crayton<sup>3</sup>, Sławomir Gonkowski<sup>4</sup>.  
<sup>1</sup>*Department of Neurology and Neurosurgery, Stem Cell Research Laboratory, Faculty of Medical Sciences, University of Warmia and Mazury, Olsztyn, Poland, Olsztyn, Poland,*  
<sup>2</sup>*Department of Comparative Anatomy, Faculty of Biology, University of Warmia and Mazury, Olsztyn, Poland,* <sup>3</sup>*Department and Clinic of Urology, Faculty of Medical Sciences, Medical University of Warsaw, Warsaw, Poland,* <sup>4</sup>*Department of Clinical Physiology, Faculty of Veterinary Medicine, University of Warmia and Mazury, Olsztyn, Poland*

Zinc ions in the synaptic vesicles of “zinc-enriched neurons” (ZEN) have an important role in normal physiological and pathophysiological processes in target organ innervation. The divalent cation zinc transporter (ZnT3), is an excellent marker of ZEN, however, there is a paucity of data concerning the presence of ZEN neurons in the porcine jejunum.

Ten-micrometer thick cryostat sections of porcine jejunums were processed for routine double- and triple-immunofluorescence labeling for ZnT3 in various combinations of other antisera including pan-neuronal marker (PGP 9.5), substance P (SP), somatostatin (SOM), vasoactive intestinal peptide (VIP), nitric oxide synthase (NOS), leu-enkephalin (LENK), vesicular acetylcholine transporter (VACHT), neuropeptide Y (NPY), galanin (GAL) and calcitonin-gene related peptide (CGRP). Immunohistochemistry revealed that approximately 39%, 49% and 45% of all PGP9.5- positive neurons in the jejunal myenteric (MP), outer submucous (OSP) and inner submucous (ISP) plexuses were simultaneously ZnT3<sup>+</sup>. Moreover, the majority of ZnT3<sup>+</sup> neurons in all plexuses were simultaneously VACHT-positive. Both VACHT-positive and VACHT-negative ZnT3<sup>+</sup> neurons co-expressed a variety of active substances with different patterns of co-localization depending on the plexus studied. In the MP, the largest populations among ZnT3<sup>+</sup> neurons formed VACHT-negative but NOS-positive cells. In the OSP and ISP, substantial subpopulations of ZnT3<sup>+</sup> neurons formed VACHT-positive cells co-expressing SOM and GAL respectively. The broad-spectrum of active substances which co-localize with the ZnT3<sup>+</sup> neurons in the porcine jejunum suggest that ZnT3 takes part in the regulation of various processes in the gut, both in normal physiological and during pathophysiological processes.