Qualitative and quantitative analysis of the developing enteric nervous system

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The enteric nervous system (ENS) is an anatomically and neurochemically complex division of the peripheral nervous system (Furness and Costa 1987) with unique embryonical origins (Doyle et al. 2004). In the present studies, chicken embryos, hatched chicks, and human embryonal tissue samples were used. The human studies always fulfilled the requirements of the declaration of the Medical World Federation proclaimed in Helsinki in1964. From the gut samples whole-mount preparations were made in each study. NADPH-diaphorase histochemistry was used to visualize nitrergic neurons in the developing chicken gut on embronic days 12, 13, 14, and 19 (Bagyánszki et al. 2000). Between the examined time points, both the desity of ganglia and nitrergic neurons decreased significantly, the later showing a delay in the colon. At the first three examined time points, the neurons/ganglion ratio showed a wave-like pattern with higher values in the proximal part of the small intestine and the colon. This pattern changed into a more balanced one by embryonal week 19. In our next study we attempted to develop a staining method to label the entire population of myenteric neurons in the intestine of developing chickens (Román et al. 2001). Histochemical staining with cuprolinic blue and immunostaining against neurofilament (NF) were used to stain the total number of neurons. Cuprolinic blue stained a huge number of neurons in the myenteric plexus. Double-staining with cuprolinic blue and NF revealed that NF staining labelled only approximately 35% of the total myenteric neuronal population. Double-staining made it possible to intensify both stainings, which allowed a more accurate morphological classification of NF-labelled neurons. Further on, the spatial distribution of nitrergic neurons was investigated in the developing human fetal myenteric plexus (Román et al. 2004). NADPH-diaphorase-stained segments of human fetal intestine were photographed, and the borders of the myenteric plexus and nuclei were digitalized. Plexus Pattern Analysis programme designed in our laboratory was used to perform randomization analyses. The results showed that nitrergic neurons were aggregated within the ganglia. The developmental dynamics of changes in the pattern of nitrergic neurons exhibited regional differences. These results suggest that, in addition to the gut wall and the plexus, intraganglionic constituents might also contribute to the spatial distribution of nitrergic neurons.

References

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