

The enteric nervous system: New developments and emerging concepts

Rosa Serio, Maria Garzia Zizzo, Mariangela Mastropaolo

Abstract

The enteric nervous system (ENS) is an integrative neuronal network, organized in two ganglionated plexuses, myenteric and submucosal, composed of neurons and enteric glial cells, controlling the activity of the smooth muscle of the gut, mucosal secretion and blood flow. The ENS contains as many neurons as the spinal cord, and the functional and chemical diversity of enteric neurons closely resembles that of the central nervous system. This highly integrated neural system is also referred to as the 'brain-in-the-gut', because of its capability to function in the absence of nerve inputs from the central nervous system.

Keywords

Enteric nervous system; neurotransmission; Parkinson's disease

The ENS controls gut motility and secretion via local reflexes that are triggered by local distension of the intestinal wall, distortion of the mucosa, and chemical contents in the lumen. This neuronal regulation of GI functions is due to the liberation of specific neuromediators synthesized by functionally defined enteric neurons. In addition, ENS is involved in the control of immune and inflammatory processes throughout the gut. Thus, it is not surprising that any damage to ENS circuitries and in the neurotransmitters systems results in a wide array of gut disorders, including motor impairments, which are characterized by high morbidity. In the future, challenges are to properly understand the molecular and cellular changes that underlie enteric neuropathies, to utilize knowledge of the normal neurochemistry, pharmacology and physiology of the ENS to devise strategies to treat disorders of motility and to develop effective therapeutic compounds.

Introduction

The enteric nervous system (ENS) may be defined as the system of neurons and their supporting cells (glia) that is found within the walls of the gastrointestinal tract, including the pancreas and gall bladder. An "enteric neuron" is any neuron whose cell body is within the ENS. The enteric nervous system regulates gastrointestinal functions including motility, secretions, blood flow, and the immune system. It is unique in its ability to function independently of the central nervous system (CNS) in the control of the functions of the digestive tract. For this reason, the ENS is considered to be a "second brain in the gut".¹ However, the CNS is able to modulate, but not entirely control, the GI function by sending instructions via the two components of the extrinsic autonomic nervous system: the sympathetic and parasympathetic nervous system.

The CNS and the ENS share a common origin and have functional and chemical similarities: i) the ENS in fact derives from neural crest cells that migrate to the cranial portion of the gut and subsequently move

Rosa Serio MSc, PhD *

Dipartimento di Scienze e Tecnologie Molecolari e Biomolecolari (STEMBIO), Viale delle Scienze, I-90128 Palermo, Università di Palermo, Italia
Email: rserio@unipa.it

Maria Grazia Zizzo MSc, PhD

Dipartimento di Scienze e Tecnologie Molecolari e Biomolecolari (STEMBIO), Viale delle Scienze, I-90128 Palermo, Università di Palermo, Italia
Email: mariagrazia.zizzo@unipa.it

Mariangela Mastropaolo, BSc, MSc

Dipartimento di Scienze e Tecnologie Molecolari e Biomolecolari (STEMBIO), Viale delle Scienze, I-90128 Palermo, Università di Palermo, Italia

* corresponding author

caudally to reach the entire GI tract; ii) the ENS contains a number of neurons similar to those found in the spinal cord (approximately 80–100 million neurons); iii) ENS express all neurotransmitters so far known in CNS (more than 30 neurotransmitters). These include classical neurotransmitters such as acetylcholine (ACh), noradrenaline, serotonin, GABA and glutamate, but a great number of other neurotransmitter and hormones also participate in the regulation of functions in the GI tract: vasoactive intestinal polypeptide (VIP), nitric oxide, galanin, motilin, adenosine triphosphate, tachykinins, etc.; iv) the ENS is an integrative neural network including afferent neurons, interneurons, and efferent neurons; v) the enteric glial cells are remarkably like astrocytes in structure and biochemistry and a diffusion barrier around the capillaries surrounding ganglia, similar to the cerebral blood-brain barrier.

The ENS is organized in two ganglionated plexuses, myenteric and submucosal, composed of neurons and enteric glial cells. The myenteric plexus (or Auerbach's plexus) lies between the external longitudinal and internal circular muscle layers. The submucosal plexus (or Meissner's plexus) lies between the circular muscle layer and the mucosa. Neurons of the myenteric plexus control the activity of the smooth muscle of the gut, whereas those in the submucosal plexus regulate mucosal secretion and blood flow. The ENS controls gut motility and secretion via local reflexes that are triggered by local distension of the intestinal wall, distortion of the mucosa, and chemical contents in the lumen. These reflexes involve parallel circuits of synaptically interconnected ENS neurons, which include primary intrinsic afferent neural cells, ascending and descending interneurons, excitatory and inhibitory motoneurons, vasomotoneurons and secretomotoneurons. For example, in the myenteric plexus, activation of ascending interneurons and excitatory motoneurons results in the release of excitatory neuromediators (acetylcholine, substance P) onto smooth muscle fibers causing circumferential contraction of the circular muscle layer upstream of the bolus. Activation of descending interneurons and inhibitory motoneurons results in the release of inhibitory neuromediators (VIP - vasoactive intestinal polypeptide, nitric oxide) causing relaxation of circular muscle downstream of the bolus.

The basic electrical rhythms in the gut are fairly constant and characterized by slow waves,

rhythmical oscillations of the smooth muscle membrane potential, which are responsible for the contractions of the muscles. These threshold events represent true smooth muscle action potentials. Spikes are not essential for excitation–contraction coupling in GI smooth muscle, but their occurrence is associated with more forceful contractions. Smooth muscle cells lack the

ionic mechanisms necessary to regenerate electrical slow waves. The search for the origin of rhythmicity in intestinal contraction has identified pacemaker regions of the slow waves located at the myenteric and submucosal borders of circular muscles and contains a network of cells known as the interstitial cells of Cajal.² These interstitial cells of Cajal are distinctive populations of muscle-like cells. They make contact with each other and with muscle cells and nerve terminals and function as pacemakers in gastrointestinal muscles by initiating rhythmic electrical activity. Excitatory agonists, such as ACh, stimulate intestinal phasic motor activity by increasing the amplitude of slow wave activity and thereby enhancing spike potential activity, resulting in a contractile wave passing down the gut. Inhibitory transmitters, such as VIP or NO, causes reduction of the amplitudes of slow waves and the membrane potential at the peak of the slow wave is too far below threshold for an action potential to occur and the smooth muscle relaxes. Besides acetylcholine, vasoactive intestinal peptide and nitric oxide, other chemical agents may act as modulator of gastrointestinal function acting at different levels, smooth muscle cells, ICCs, nerve terminals, interneural synapses. So, the gastrointestinal smooth muscle contractility is regulated not by one or another of the transmitters, but by at least two and often more, pharmacologically coupled and working in close coordination. These complex functional interactions have some variations depending on the species, organ system and region of the gut.

In addition, there is evidence that the ENS is involved in the control of immune and inflammatory processes throughout the gut. Immuno-neural integration progresses sequentially, beginning with immune detection followed by signal transfer to enteric neural microcircuits, followed by neural interpretation and then selection of a specific neural program of coordinated mucosal secretion and motor propulsion that effectively clears the antigenic threat from the intestinal lumen.

Thus, it is not surprising that any damage to ENS circuitries results in a wide array of gut disorders, including motor impairments, which are characterized by high morbidity, with a markedly compromised patient's quality of life and occasional fatal outcomes. Besides a few exceptions, the mechanisms through which neural diseases cause gastrointestinal dysfunction, including motor abnormalities, remain poorly understood.

Among the plethora of neural systems which control GI motility I wish to focus on the dopaminergic system since it seems to be a major candidate for the impairment of GI function frequently encountered by patients affected by Parkinson's disease (PD).³

Parkinson's disease is the second most common neurodegenerative disease after Alzheimer's disease. It is well-known that PD is an extrapyramidal motor disorder which is caused by the massive loss of dopaminergic neurons in the substantia nigra pars compacta. Nevertheless, it has become increasingly evident in recent years that Parkinson's disease is a multicentric neurodegenerative process that affects several neuronal structures outside the substantia nigra, among which is the enteric nervous system.⁴ Remarkably, recent reports have shown that the lesions in the enteric nervous system occur at a very early stage of the disease, even before the involvement of the central nervous system, and abnormalities or disturbances in function at virtually all levels of the GI system have been identified.⁵ Weight loss, dental deterioration, salivary excess, dysphagia, impaired gastric emptying, decreased bowel movement frequency, and difficulty with the act of defecation itself all may occur in the setting of PD.³ This led to the postulate that the enteric nervous system could be critical in the pathophysiology of Parkinson's disease, and that the pathological alterations within the enteric nervous system could be involved in the gastrointestinal dysfunction frequently encountered by Parkinsonian patients. In addition to being the source of significant clinical symptoms and dysfunction in individuals with PD, it has now been suggested that the ENS may actually be the port of entry of the pathologic process that eventually culminates in the clinical picture of PD.⁵ Animal models of PD are essential tools with which to identify novel therapeutic targets and test potential therapies. However, appropriate animal models to investigate in depth the GI impairment in PD is lacking due to the scarce information of the physiological role of DA in the gut, necessary also to solve the respective roles of intrinsic and extrinsic innervations in GI dysfunction.

Although the gut contains dopamine,⁶ this catecholamine has only recently been confirmed as an intrinsic neurotransmitter of the enteric nervous system (ENS).^{7,8} In fact, enteric dopaminergic neurons, which express tyrosine hydroxylase (TH) and the dopamine transporter (DAT) but lack dopamine β -hydroxylase, enzyme that converts dopamine to norepinephrine, have been identified in mouse, guinea pig⁸ and human.⁷ In mouse, all five classes of dopamine receptors D1-like family, including D1 and D5 receptors, and D2-like family, including D2, D3, and D4 receptors have been identified throughout the digestive tract.⁹ Dopamine receptors appear

decreased in the myenteric plexus of PD patients. However, the function of enteric dopaminergic neurons in the regulation of GI motility is still far from clear and the mechanism of dopamine action and location of dopamine receptors are controversial. Moreover, conclusions regarding the role of dopamine in modulating GI tract motility have been confounded by the ability of dopamine agonists to activate adrenergic receptors. Therefore, the role of DA in bowel motility needs to be identified and considered for potential treatments of GI dysfunction in PD.

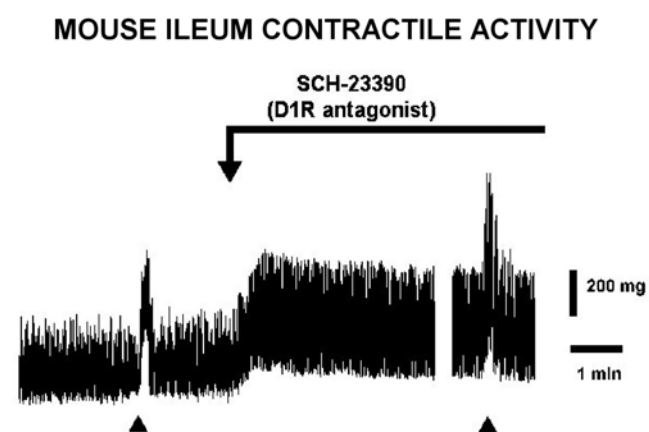


Figure 1- Typical recording of muscular tension developed in mouse ileum under control conditions and following D1 receptor blockade. Arrows indicate the initiation of cholinergic neural stimulation.

Note that smooth muscle tone, spontaneous phasic contraction amplitude and neurally-induced contraction were enhanced following D1 receptor blockade indicating that dopamine-mediated inhibitory input is tonically supplied to mouse ileum.

Intestinal motility can be studied *in vitro* using the organ bath technique, which allow the recording, as a change in isometric tension, of spontaneous contractile activity and neurally evoked responses. Thus, using such an approach we can distinguish effects of various chemical agents on GI motility which occur through modulation of the enteric nervous system from effects mediated by receptors located on smooth muscle cells. Our analysis of the effects of dopamine on mouse ileum contractility indicates that intestinal muscle is under basal inhibitory control by a dopamine-mediated system (Fig. 1). D1 receptors play a major role being located both at postjunctional level and at prejunctional level, inhibiting acetylcholine release from enteric motor neurons.¹⁰ We have clarified whether the source of dopamine is from neural or non-neural cells. Moreover, the results presented showed that there is an agonistic interaction of the dopaminergic receptor subtypes (D1 and D2 receptors)

in the regulation intestinal contractility. It remains unclear why the loss of DA neurons in the gut determines inhibition of contractility and constipation. Furthermore, data are sparse for the role of the enteric dopaminergic nervous system in the pathophysiology of the gastrointestinal symptoms including the relative contribution of D1 and D2 receptors in the presence of different DA concentration (which varies from the healthy state to the disease) and the eventual changes in the pattern of expression of DA receptor subtypes following DA denervation. Moreover, the possibility that more than one neurotransmitter system is actually affected might be considered.

In conclusion, investigative evidence and emerging concepts implicate dysfunction in the nervous system as a significant factor underlying patient symptoms in functional gastrointestinal disorders. The future challenges are, then, to properly understand the molecular and cellular changes that underlie enteric neuropathies, to utilize knowledge of the normal neurochemistry, pharmacology and physiology of the ENS to devise strategies to treat disorders of motility and to develop effective therapeutic compounds.

References

1. Gershon, M. D. 1998. *The Second Brain: The Scientific Basis of Gut Instinct and a Groundbreaking New Understanding of Nervous Disorders of the Stomach and Intestine*. New York, NY: HarperCollins Publishers.
2. Takaki M, Suzuki H, Nakayama S. Recent advances in studies of spontaneous activity in smooth muscle: ubiquitous pacemaker cells. *Prog Biophys Mol Biol*. 2010;102(2-3):129-35.
3. Pfeiffer RF. Gastrointestinal dysfunction in Parkinson's disease. *Parkinsonism Relat Disord*. 2011;17(1):10-5.
4. Lebouvier T, Chaumette T, Paillusson S, Duyckaerts C, Bruley d, V, Neunlist M, et al. The second brain and Parkinson's disease. *Eur J Neurosci*. 2009;30(5):735-41.
5. Lebouvier T, Neunlist M, Bruley d, V, Coron E, Drouard A, N'Guyen JM, et al. Colonic biopsies to assess the neuropathology of Parkinson's disease and its relationship with symptoms. *PLoS One*. 2010;5(9):e12728.
6. Eaker EY, Bixler GB, Dunn AJ, Moreshead WV, Mathias JR. Chronic alterations in jejunal myoelectric activity in rats due to MPTP. *Am J Physiol*. 1987;253(6 Pt 1):G809-15.
7. Anlauf M, Schafer MK, Eiden L, Weihe E. Chemical coding of the human gastrointestinal nervous system: cholinergic, VIPergic, and catecholaminergic phenotypes. *J Comp Neurol*. 2003;459(1):90-111.
8. Li ZS, Pham TD, Tamir H, Chen JJ, Gershon MD. Enteric dopaminergic neurons: definition, developmental lineage, and effects of extrinsic denervation. *J Neurosci*. 2004 ;24(6):1330-9.
9. Li ZS, Schmauss C, Cuenca A, Ratcliffe E, Gershon MD. Physiological modulation of intestinal motility by enteric dopaminergic neurons and the D2 receptor: analysis of dopamine receptor expression, location, development, and function in wild-type and knock-out mice. *J Neurosci*. 2006;26(10):2798-807.
10. Zizzo MG, Mule F, Mastropaolo M, Serio R. D1 receptors play a major role in the dopamine modulation of mouse ileum contractility. *Pharmacol Res*. 2010;61(5):371-8.