VISCERAL PERCEPTION

Prevertebral ganglia and intestinofugal afferent neurones

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Intestinofugal afferent neurones (IFANs) are a unique subset of myenteric ganglion neurones that regulate normal gastrointestinal function. The IFANs relaying mechanosensory information to sympathetic neurones of the prevertebral ganglion (PVG) function as volume detectors. It is possible that mechanosensory information arriving in the PVG via axon collaterals of visceral spinal afferent nerves can be modulated entirely within the PVG itself.

SUMMARY

Intestinofugal afferent neurones (IFANs) are a unique subset of myenteric ganglion neurones which relay mechanosensory information to sympathetic prevertebral ganglion (PVG) neurones. They function as slowly adapting mechanoreceptors which detect changes in volume. IFANs are arranged in parallel to the circular muscle fibres and respond to circular muscle stretch rather than tension. When activated by colonic distension, IFANs release acetylcholine at the PVG, and evoke nicotinic fast excitatory postsynaptic potentials (F-EPSPs), which are amplified by a parallel release of vasoactive intestinal peptide (VIP). A subset of IFANs respond to colonic distension by releasing gamma aminobutyric acid (GABA) in PVG which in turn facilitates the release of acetylcholine from cholinergic IFANs. This reflex arc formed by IFANs and sympathetic PVG neurones provides a protective buffer against large increases in tone and intraluminal pressure. IFANs with Dogiel type II morphology, which have dendritic processes "in parallel" to the circular muscle layer, provide synaptic input to sympathetic PVG neurones, and they also innervate Dogiel type I IFANs which project to PVG neurones. Visceral spinal afferent neurones have axon collaterals which form en passant synapses with PVG neurones. They release substance P (SP) and calcitonin gene related peptide (CGRP) in prevertebral ganglia, and evoke slow excitatory postsynaptic potentials (S-EPSPs) in sympathetic neurones. The release of SP is facilitated by release of neurotensin from central preganglionic neurones and is inhibited by central preganglionic neurones that release enkephalins. This raises the possibility that the mechanosensory information arriving in PVG via axon collaterals of visceral spinal afferent nerves can be modulated entirely within the PVG itself.

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INTRODUCTION

The gastrointestinal tract is equipped with several different sets of mechanosensory afferent neurones (fig 1). Vagal mechanosensitive afferent nerves are low threshold tension detectors activated at physiological levels of distension.[1 2] They play an important role in controlling normal intestinal function. Two specialised types of vagal afferent endings have been identified within the muscularis externa.[3–6] Intramuscular arrays (IMAs) are fine varicosed nerve fibres running parallel to longitudinal muscle fibres. Although the IMAs are arranged in parallel to muscle fibres, they appear to function as "in series" receptors,[3] and are sensitive to contraction and tension.

Intraganglionic laminar endings are varicose nerves found within the myenteric plexus. Although they appear to be chemosensitive and have a local effector role,[3 4] they may also function as tension receptors.[4 7 8] Spinal afferent mechanosensory nerves with cell bodies in the spinal dorsal root ganglia have terminal fields in the mesentery, serosa, mucosa, and muscle layers.[9 10] Mechanosensory spinal afferent nerves are high threshold tension detectors that respond dynamically across a wide range of stimulus intensity extending to the noxious range.

INTRINSIC PRIMARY AFFERENT NEURONES (IPANS)

IPANs in the myenteric plexus comprise an important group of mechanosensory neurones that regulate normal gastrointestinal function.[11 12] Mechanosensory IPANs are entirely contained within the intestinal wall. The cell bodies of mechanosensory IPANs are located in the myenteric ganglia and their processes extend within the myenteric plexus and also into the mucosa. Mechanosensory IPANs have Dogiel type II morphology, and electrophysiological IPANs have properties of the AH type neurones.[12] Intracellular recordings have shown that mechanosensory IPANs respond to stretch in the longitudinal direction of the bowel wall even though their processes primarily project circumferentially.[12] As the action potential discharge persists after blocking synaptic discharge, it has been suggested that IPANs are primary afferent neurones. They are also known to interconnect with each other to form a self reinforcing network.[11]

Abbreviations: CGRP, calcitonin gene related peptide; F-EPSP, fast excitatory postsynaptic potential; GABA, gamma aminobutyric acid; IFAN, intestinofugal afferent neurone; IMA, intramuscular array; IMG, inferior mesenteric ganglion; IPAN, intrinsic primary afferent neurone; PVG, prevertebral ganglion; S-EPSP, slow excitatory postsynaptic potential; SMG, superior mesenteric ganglion; SP, substance P; VIP, vasoactive intestinal peptide.

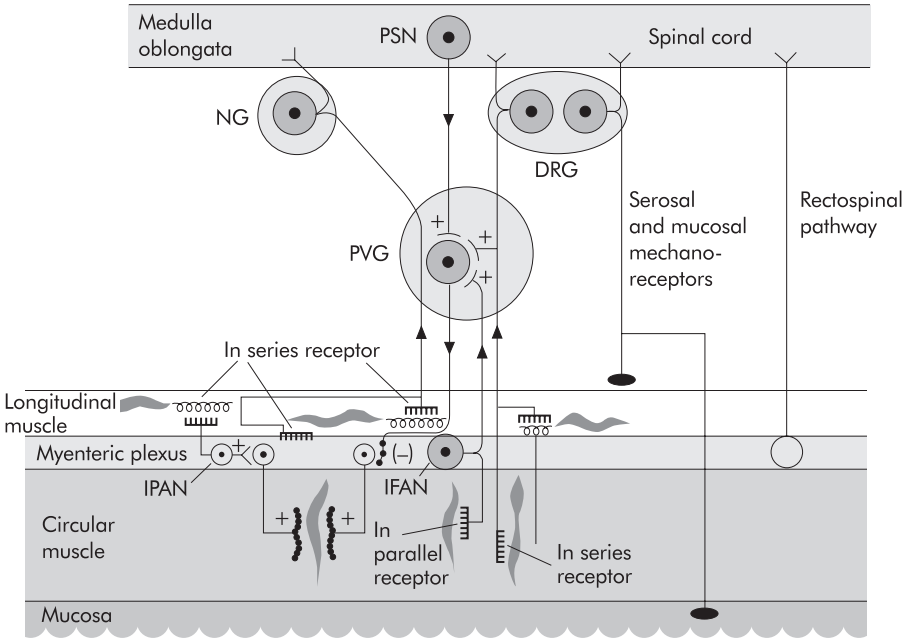

Figure 1 Schematic diagram of the possible arrangement of mechanosensory afferent neurones. The intestinofugal afferent neurones (IFANs) function in an "in parallel" arrangement to the circular muscle fibres whereas intrinsic primary afferent neurones (IPANs) function in an "in series" arrangement. The physiological stimulus of the rectospinal neurones has not been identified.[41 42] They may participate in the defecation reflex and are included here for completeness. DRG, dorsal root ganglion; NG, nodose ganglion; PSN, preganglionic sympathetic neurone; PVG, prevertebral ganglion.

INTESTINOFUGAL AFFERENT NEURONES (IFANS)

IFANs are another set of mechanosensory nerves that regulate normal gastrointestinal function (fig 1; fig 2, pathway 1). IFANs are a unique subset of myenteric ganglion neurones. Their cell bodies and dendritic neurites lie within the intestinal wall but their axons leave the bowel wall to form synapses in the inferior mesenteric ganglion (IMG) and superior mesenteric ganglion (SMG) and the coeliac ganglion.[13–17] Collectively, these three ganglia are referred to as PVG. IFANs function as slowly adapting mechanoreceptors[17 18] which are functionally arranged in parallel to the circular muscle fibres.[17] They respond to circular muscle stretch but not tension.[17] When activated by colonic distension, IFANs release acetylcholine in the PVG and evoke nicotinic F-EPSPs (fig 2, pathway 1).[19] Mechanosensory IFANs also release the neuropeptide VIP[19] which functions as a neuromodulator that amplifies fast nicotinic transmission. A subset of IFANs respond to colonic distension by releasing GABA in the IMG of the guinea pig.[19 20] GABA, released by colonic distension, acts on $GABA_A$ receptors to facilitate release of acetylcholine from cholinergic IFANs projecting from the colon.[19] The sympathetic IMG and SMG and the coeliac ganglion neurones that receive excitatory synaptic input, reflexly release noradrenaline in the gastrointestinal wall to modulate smooth muscle contraction and intrinsic based reflexes.[21 22] The majority of the IFANs supplying the coeliac ganglion and the SMG and IMG arise from the colon and rectum.[21 23] This contrasts with the approximately even distribution within the gastrointestinal wall of the noradrenergic inhibitory neurones that project from these ganglia indicating that the colon and rectum reflexly modulate their own motor activity as well as motor activity in regions of the intestine lying more proximal.[21 24] The functional significance of this reflex arc is that it helps maintain the colon wall in a relaxed condition during filling, to oppose the tendency of colonic smooth muscle cells to depolarise and contract, as it is distended with intraluminal content. This regulates the motor activity of the intestine and ensures that the bulk and fluid content of material in more proximal regions of the gut is appropriate, and that it arrives at the colon on time. The reflex arc therefore provides a protective reflex buffer mechanism against large increases in tone and intraluminal pressure.

IFANS AND SENSORIMOTOR ACTIVITY

Simultaneous recordings of circular muscle contraction (fig 3, top), intraluminal pressure (fig 3, middle), and synaptic activity (fig 3, bottom) recorded from a sympathetic ganglion neurone of the mouse SMG illustrate the relationship between IFAN activity and colonic motor activity. It can be seen that mechanosensory afferent synaptic input to the SMG neurone was virtually absent when the circular muscle layer was contracted, intraluminal pressure was at its highest, and when the circumference of the colon was decreased. In contrast, there was an increase in mechanosensory afferent synaptic input during "receptive relaxation" (decrease in intraluminal pressure) and increase in the volume and circumference of the colon wall prior to contraction. These data suggest that the mechanosensory IFANs that synapse with PVG neurones are arranged "in parallel" with the circular muscle layer. As a result of this arrangement, the frequency of synaptic input to PVG neurones would be expected to increase during increases in the circumference of the colon wall during filling. Emptying of the colon during circular muscle contraction decreases colonic circumference, and unloads the mechanoreceptors resulting in a decrease in synaptic input.

The "in parallel" arrangement of the mechanosensitive IFANs distinguishes them from vagal and spinal mechanosensitive afferent nerves. Distension sensitive visceral mechanoreceptors in the vagus, splanchnic, and pelvic nerve trunks function as "in series" receptors.[25–27] They behave as if mechanically connected with the longitudinal muscle elements because their discharge frequency increases during longitudinal stretch of the viscus and during active longitudinal muscle contraction. These mechanoreceptors are thought to act as sensors of visceral wall tension developed both passively by distension and actively by contraction.[26] "In series" tension receptors are not regarded as monitors of visceral volume because the relationship between volume of the

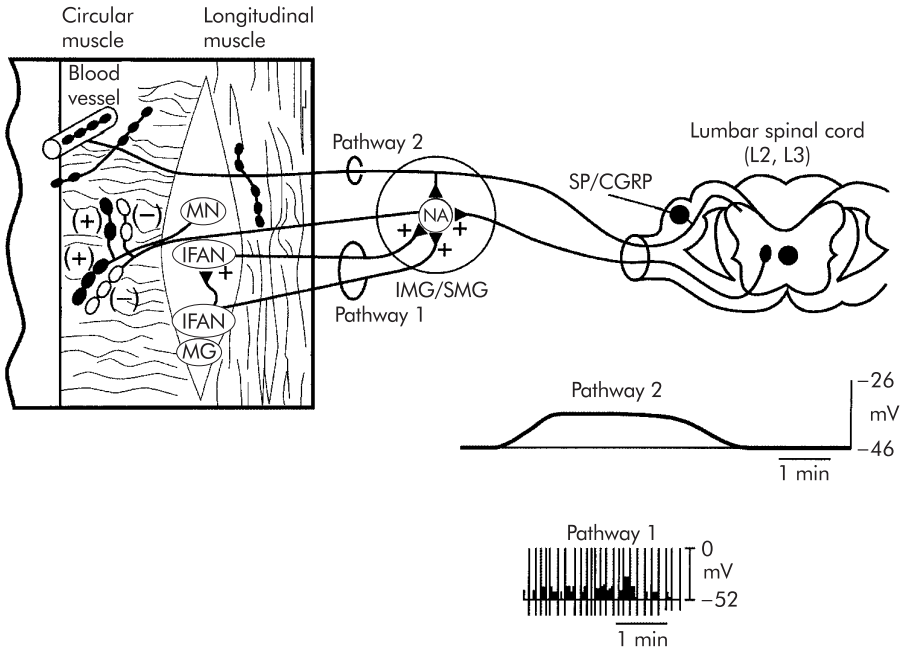

Figure 2 Schematic diagram of mechanosensory afferent neurones synapsing with prevertebral ganglion neurones. Pathway 1 is comprised of neurones with cell bodies in the myenteric plexus. Activation of these neurones evokes fast nicotinic excitatory postsynaptic potentials and vasoactive intestinal peptide dependent slow excitatory postsynaptic potentials in inferior mesenteric ganglion/superior mesenteric ganglion (IMG/SMG) neurones. Pathway 2 is comprised of neurones with cell bodies in the dorsal root ganglia. Activation of this pathway leads to release of substance P (SP) and calcitonin gene related peptide (CGRP) from the axon collaterals,[20] and to slow excitatory synaptic potentials. IFAN, intestinofugal afferent neurone; MG, myenteric ganglion; MN, motor neurone; NA, noradrenergic neurone.

viscus and wall tension is probably not constant as there are numerous nervous and humoral factors which can alter gastrointestinal smooth muscle tone and therefore influence the relationship between volume and wall tension. On the other hand, mechanosensory input from IFANs to sympathetic PVG neurones behaves as if the sensory transducers are monitoring intracolonic volume during filling and emptying of the colon by sensing changes in stretch or tension of the circular muscle layer. This raises the possibility that the afferent projections of mechanosensory IFANs which make synaptic contact with PVG neurones carry a message qualitatively different from that transmitted to the central nervous system by vagal and spinal mechanosensory afferent nerves.

MORPHOLOGICAL CLASSIFICATION OF IFANS

Retrograde labelling experiments suggest that two populations of IFANs monitor circular muscle activity and intraluminal volume. These are primary intestinofugal neurones that project without synaptic interruption to PVG neurones and second or higher order neurones that receive cholinergic synaptic input from primary IFANs (fig 1, pathway 1).

Using retrograde labelling methods, we have identified IFANs with Dogiel type I and type II morphology (fig 4A, B, respectively). These neurones were found in the myenteric plexus of the guinea pig colon 30 days after application of DiI crystals to the lumbar colonic nerve. In our experience, IFANs with Dogiel type II morphology, identified by retrograde labelling, have their dendritic processes arranged parallel to the circular muscle layer. Based on electrophysiological and pharmacological data, we suggest that the IFANs with Dogiel type II morphology provide synaptic input to sympathetic PVG neurones and to IFANs with Dogiel type I morphology, and that the latter project to PVG neurones. Thus PVG neurones receive synaptic input from primary and secondary or higher order IFANs.[22] The data also suggest that both types of neurones are mechanosensory. Our observation that a subset of mechanosensory IFANs have Dogiel type II morphology is not in agreement with the data obtained by Lomax and colleagues[15] or Sharkey and colleagues[28] who used retrograde labelling methods to identify the morphology of IFANs. The morphological and electrophysiological data obtained by Lomax and colleagues[15] and Sharkey and colleagues[28] indicate that IFANs are Dogiel type I neurones with electrical properties consistent with myenteric S type neurones or myenteric type 3 neurones.

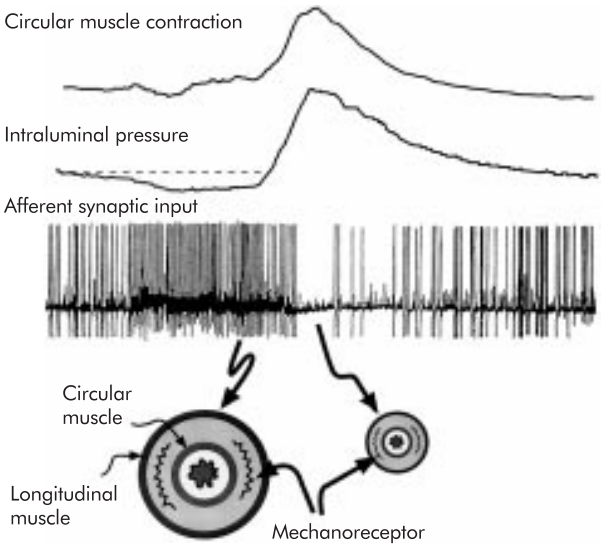

Figure 3 Relationship between spontaneous circular muscle contraction, intraluminal pressure, and mechanosensory afferent synaptic input to a sympathetic neurone in the mouse superior mesenteric ganglion. Note that during "receptive relaxation" prior to contraction, there is an increase in the frequency of excitatory synaptic input whereas the frequency of synaptic input markedly declines at peak intraluminal pressure and contraction. These data suggest that the mechanosensory nerve fibres function as "in parallel" receptors. All recordings were made simultaneously in vitro.

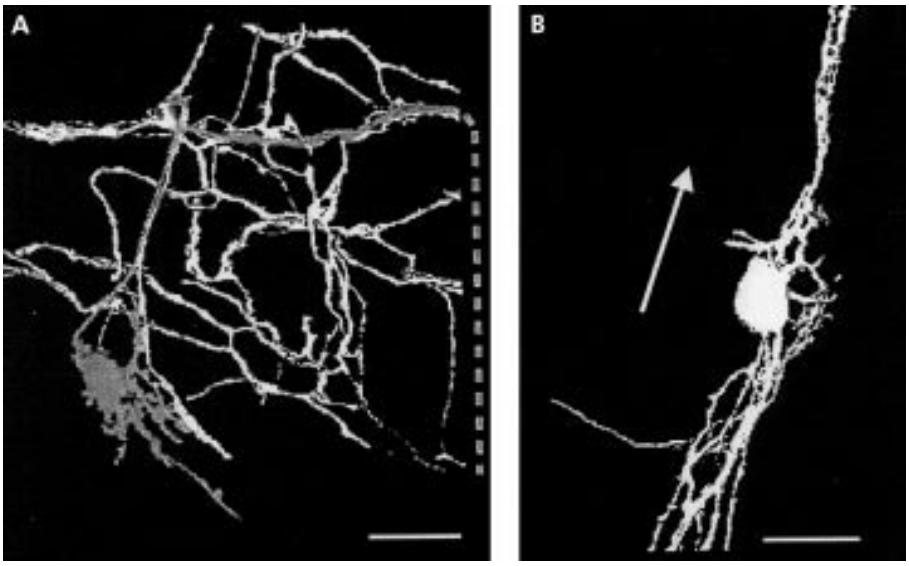

Figure 4 Examples of the shapes of two retrogradely labelled colonic myenteric neurones revealed by application of the fluorescent dye DiI to the lumbar colonic nerve of the guinea pig. Neurones were imaged with a confocal laser scanning microscope and reconstructed using ANALYZE image processing software. Fifty optical sections in 1 μm step increments were made to reconstruct the neurone in (A), and 41 sections in 1 μm step increments were made to reconstruct the neurone in (B). The arrow in (B) indicates the orientation of the circular muscle layer. Calibration bars are 50 μm.

VISCERAL SPINAL AFFERENT NEURONES

PVG neurones also receive mechanosensory synaptic input from visceral spinal afferent neurones that have axon collaterals which form en passant synapses with PVG neurones (fig 2, pathway 2).[13–16 21–23 29–35] The mechanosensory colon spinal afferent nerves that form axon collaterals and make en passant synapses with PVG neurones are mechanosensory, and have cell bodies which are small in diameter (<20 μm).[36] The mechanosensory colon spinal afferent nerves release SP and CRGP in PVG and evoke S-EPSPs in sympathetic neurones (fig 2, pathway 2).[20 37] This pathway has a higher (>15 cm H_20) threshold for activation compared with IFANs (<15 cm H_2O).[19 20 37]

The peripheral terminals of this high threshold pathway in the intestinal wall signal wall tension and are arranged in series with both longitudinal and circular muscle layers.[17 20 37] When the terminals are activated, they evoke S-EPSPs in PVG neurones, amplifying fast cholinergic synaptic input arriving from IFANs, thereby increasing sympathetic inhibitory drive to innervated segments of the intestine. The findings that the dorsal root ganglion neurones belonging to this pathway are of small (<20 μm) diameter, that colon distension releases SP and CGRP[20 37] from axon collaterals in the PVG, and that their release is abolished following lumbar dorsal root rhizotomy and by capsaicin pretreatment in vivo,[38] are important observations as they raise the possibility that the mechanosensory colon spinal afferent nerves might be the same as the visceral nociceptive pathway.

Release of SP and CGRP from en passant synapses in the IMG can be modulated by peptidergic central preganglionic nerves. Central preganglionic nerves release neurotensin which facilitates release of SP[38–40] whereas preganglionic nerves release enkephalins that inhibit colonic distension induced release of SP from en passant synapses.[20] This raises the interesting possibility that mechanosensory information arriving in the PVG via axon collaterals of mechanosensory spinal afferent nerves can be modulated separately in the PVG without alteration of the signal referred centrally via the central extension of the same mechanosensory spinal afferent nerve.

CONCLUSIONS

It is apparent from this brief overview that a large array of mechanosensory afferent nerves regulate normal gastrointestinal function, and that the PVG forms an extended neural network which connects the lower intestinal tract to the upper gastrointestinal tract. The IFANs relaying mechanosensory information to sympathetic neurones of the PVG function as volume detectors. The physiological function of the IFANs will be much better understood when the ion channels that confer mechanosensitivity are identified.

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