Human colonic smooth muscle: spontaneous contractile activity and response to stretch

R C GILL, K R COTE, K L BOWES, AND Y J KINGMA

From the Departments of Surgery and Electrical Engineering, University of Alberta, Edmonton, Alberta, Canada

SUMMARY The length dependence of the spontaneous contractile activity of human colonic muscle was assessed *in vitro*. Muscle obtained from the right colon was more distensible than that of the left colon. This was true for all muscle layers. Maximum spontaneous active stress was exerted by both circular and longitudinal muscle layers of the right colon at greater degrees of stretch ($p < 0.001$) than those of the left colon. The contractile frequency of longitudinally oriented strips increased with length. The contractile frequency of intertaenial longitudinally oriented strips from the right colon was lower ($p < 0.001$) than that of strips from the left colon. The contractile frequency of circularly-oriented strips from the right colon (6.25 ± 0.38 min) was higher ($p < 0.001$) than that of strips from the left colon (3.35 ± 0.35 min). The human colon appears to consist of two distinct areas based on the mechanical behaviour of the smooth muscle during spontaneous contraction.

The human large intestine may be subdivided into right and left parts on the basis of differing embryology, blood supply, innervation and function.^{1,2} The right colon has a greater diameter than that of the left, both on abdominal radiographs³ and post-mortem,⁴ and also appears more distensible than the left, being the area of greatest distension in the face of a distal colonic obstruction. Colonic pathology may also show a particular pattern of localisation. Perforation of the colon occurs most frequently in the caecum⁵ whilst diverticular disease is most common in the descending and sigmoid colon.⁶ Diverticular disease has been associated with an abnormal response of the colonic muscle to stretch.^{7,8}

In view of these observations, it would seem probable that regional variations in colonic function and pathology reflect an underlying difference in the mechanical properties of muscle along the length of the human colon. Indeed, the spontaneous contractions of human colonic muscle and their modulation by intrinsic neural discharge and stretch appear to be of considerable importance as colonic function, albeit impaired, continues after extrinsic denervation.⁹⁻¹¹

The spontaneous contractile activity of colonic

muscle obtained from several animal species has been reported although little is known concerning the dependence of these spontaneous contractions upon muscle length. The majority of studies concerning the length dependence of colonic muscle have been conducted after the abolition of spontaneous contractions in order to free the contractile element and permit an analysis of the passive properties of the muscle.¹²⁻¹⁵ In these studies, the active contractile properties of the muscle were assessed by external stimulation either electrically,¹²⁻¹⁴ by K^+ -depolarisation¹³ or by drugs such as acetylcholine or carbachol.¹³ A recent *in vitro* study,¹⁶ however, established the length dependence of taenia from guinea-pig caecum during spontaneous contraction.

As far as human taenia coli is concerned, its spontaneous contractile activity *in vitro*¹⁷⁻¹⁹ appears similar to that of the colon from the guinea pig,²⁰ pig²¹ and even the non-taeniated longitudinal muscle layer of the dog.²² The length dependence of human taenia coli during spontaneous contraction is not known. The spontaneous contractile activity of the intertaenial longitudinal muscle layer of the human colon and its length-dependence have not been established. Spontaneous contractile activity of human colonic circular muscle layer *in vitro* is less clearly defined than that of the cat,²³ dog²² or pig.²¹ Contractions of the circular muscle layer of the human colon at a frequency of 2.4 min have been

Address for correspondence: Richard C Gill, GI Science Research Unit, The London Hospital Medical College, London E1 2AJ.

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reported by van Merwyk and Duthie¹⁹ who concluded that the mechanical response of this muscle layer to stretch was weak.

This present, *in vitro*, study was initiated to establish the length dependence of the longitudinal and circular muscle layers of the human colon during spontaneous contraction. Variations in the patterns of spontaneous contraction and their modulation by stretch have been assessed in muscle strips from different parts of the colon.

Methods

TISSUE

Strips of human colonic muscle obtained at surgery were studied *in vitro*. Their contractile force and frequency in response to stretch were assessed.

ACQUISITION

Tissue was obtained from 22 patients undergoing partial colectomy for carcinoma. After resection of the diseased portion, a ring of colon (approximately 2.5 cm long) was removed from either the proximal or distal margin of the future anastomotic site; the blood supply of the segment was maintained until the moment of excision. Specimens were immediately placed in oxygenated Krebs' Ringer solution. Six specimens from ascending, five from transverse, four from descending and seven from sigmoid colon were obtained.

PREPARATION

The colonic segments were opened along the border of a taenia, carefully cleaned to prevent soiling of the muscle layers with faecal content, and pinned to the surface of a dissecting dish filled with oxygenated Krebs' Ringer solution. The mucosa and submucosa were cut off. Strips of muscle, approximately 4 mm wide, were cut with the long axis lying in the direction of the muscle fibres. Longitudinally-oriented strips (10–15 mm long) were obtained both from the taenia and the region between taeniae. Circularly oriented strips (8–10 mm long) were taken from the intertaenial region. Strips were pinned at one end to the Sylgard surface of a horizontal tissue chamber maintained at $37 \pm 1^\circ\text{C}$ and continuously superfused with oxygenated Krebs' Ringer solution. Care was taken during this procedure to avoid stretching the preparations.

RECORDING TECHNIQUE

Contractile activity of the muscle strips was recorded with a force displacement transducer (Grass FT03). Each muscle strip was attached to a force transducer with surgical 4-0 silk thread. Recordings were made of the contractile activity on a Beckman R511A polygraph.

SOLUTIONS

The superfusate used was a modified Krebs' Ringer solution containing the following (mM): Na^+ , 139.2; K^+ , 5.4; Ca^{2+} , 2.5; Mg^{2+} , 1.2; Cl^- , 125.1; HCO_3^- , 22.0; H_2PO_4^- , 1.2; glucose, 10.1. When equilibrated with 95% O_2 , 5% CO_2 gas mixture at 37°C , this solution had a pH of 7.3–7.4.

EXPERIMENTAL PROTOCOL

At the start of each experiment, strips were mounted slack (<0.05 mN force) in the tissue chamber and allowed to equilibrate for a period of 30 min. The length of the strips was then measured. Subsequently, the length of the strips was increased, stepwise, by increments of 1.0 mm each 15 min. The experiment was concluded either when the muscle strip ceased to contract spontaneously or the upper limit of the force transducer was reached (approximately 250 mN). After the experiment, the strips were removed from the tissue chamber, firmly blotted without crushing, and weighed on a balance (Mettler B5) to the nearest 0.1 mg.

CROSS SECTIONAL AREA

The cross sectional area (A) of each muscle strip was calculated from the equation $A = (M/pL) \cdot f_T$. M is the mass (mg), p is the density (mg mm^{-3}) and L is the length (mm) of the strip determined at rest. The density of the tissue was taken as 1.056 mg mm^{-3} .¹² f_T is the proportion of the muscle strip contributed by the longitudinal (taenial and intertaenial longitudinally oriented strips) or circular (intertaenial circularly oriented strips) muscle layer. A value for f_T was obtained from both histological and non-fixed transverse sections of unstretched muscle from all parts of the colon.

ANALYSIS OF DATA

Recordings of contractile activity were analysed visually. Three variables were assessed at each increment of length increase for each of the strips; active force, baseline force and contractile frequency. Active tension was taken to be the difference between maximum and minimum force during a cycle of spontaneous contraction. Data were obtained only from the last 10 min recording at each increment of length increase to allow a period of five minutes for the strip to accommodate the increase in length. This was found to be necessary as once the baseline tension of the strip was greater than zero, further increases in length produced an initial increase in baseline tension which decayed curvilinearly to a constant value. The major part of this decay was complete within two minutes. The length of the strip at which an increase in baseline tension was first recorded was measured and referred to as

the initial length, L_i . Subsequent changes in the length of the strip were normalised with respect to this initial length.

The number of spontaneous contractions during the 10 minute recording period were counted and the contractile frequency determined.

The active and baseline stresses in the longitudinal axis of the muscle strip were calculated by expressing the active and baseline tensions, respectively, per unit cross sectional area of the appropriate muscle layer.

STATISTICAL ANALYSIS

Results are expressed as the mean $\pm$ SD, and analysed statistically using Student's *t* test.

Results

LENGTH STRESS CHARACTERISTICS

Spontaneous contractile activity was recorded from all the 46 longitudinally-oriented strips of taenia (Fig. 1a), 40 inter-taenial longitudinally-oriented strips (Fig. 1b) and 32 inter-taenial circularly-oriented strips (Fig. 1c, d).

In general, the spontaneous active force generated increased with length up to a maximum. Further increases in length were associated with a decrease, or abolition, of active force generated and a marked increase in baseline stress (Fig. 2).

Differences were observed in the response of each muscle layer to stretch.

(a) Taenial longitudinally-oriented strips

Maximum active force generated occurred at $2.40 \pm 0.15 L_i$ in taenial longitudinally-oriented strips

Fig. 1 Spontaneous contractile activity recorded from taenial (A) and intertaenial (B) longitudinal strips obtained from the transverse colon at $1.15 L_i$. Note the different pattern of spontaneous contractions recorded from intertaenial circular strips obtained from the left (C) and right (D) transverse colon at $2.2 L_i$.

Table 1 Maximum active stress, and length at which this was observed, for longitudinal and circular muscle layers of human colon in vitro

Colon region	Maximum active stress (mN mm ⁻²)	Length at maximum active stress (L_i)
Ascending		
T L-O*	43 $\pm$ 11	2.40 $\pm$ 0.15
I-T L-O†	91 $\pm$ 17	2.35 $\pm$ 0.15
I-T C-O‡	5.5 $\pm$ 1.3	3.60 $\pm$ 0.20
Transverse		
T L-O	16 $\pm$ 6	2.35 $\pm$ 0.20
I-T L-O	118 $\pm$ 29	2.35 $\pm$ 0.15
I-T C-O	6.4 $\pm$ 0.8	3.60 $\pm$ 0.40
Descending		
T L-O	22 $\pm$ 7	1.80 $\pm$ 0.20
I-T L-O	120 $\pm$ 23	1.90 $\pm$ 0.15
I-T C-O	5.5 $\pm$ 0.6	2.20 $\pm$ 0.15
Sigmoid		
T L-O	23 $\pm$ 9	1.80 $\pm$ 0.10
I-T L-O	128 $\pm$ 18	1.90 $\pm$ 0.10
I-T C-O	11.8 $\pm$ 2.0	2.15 $\pm$ 0.10

Values are mean $\pm$ SD.

*Taenial longitudinal muscle layer.

†Intertaenial longitudinal muscle layer.

‡Intertaenial circular muscle layer.

obtained from the ascending and transverse – that is, right, colon, this being at a greater length ($p < 0.001$) than that of strips obtained from the descending and sigmoid – that is, left, colon ($1.80 \pm 0.15 L_i$). The maximum active force generated by the taenia of the ascending colon was greater ($p < 0.001$) than that of the taenia obtained from the transverse, descending or sigmoid colon (Table).

(b) Inter-taenial longitudinally oriented strips

Maximum active stress was recorded at $2.35 \pm 0.15 L_i$ in inter-taenial longitudinally-oriented strips from the ascending and transverse colon, this being a greater length ($p < 0.001$) than that of strips from the descending and sigmoid colon ($1.90 \pm 0.15 L_i$). These values are not significantly different ($p > 0.1$) from those of taenia obtained from corresponding regions of the human colon. The maximum active stress exerted by the inter-taenial longitudinal muscle layer did not vary significantly ($p > 0.25$) between strips obtained from different parts of the human colon but was, in all cases, greater ($p < 0.001$) than that of the taenia (Table).

(c) Circular strips

Maximum active stress was recorded at $3.60 \pm 0.30 L_i$ in inter-taenial circular strips obtained from the ascending and transverse colon, this being at a

Fig. 2 Active (solid circles) and passive (solid squares) stress of spontaneously contracting longitudinal and circular muscle layers of human colon as a function of their initial length (L_i). Standard deviations of the mean values are shown as bars except when smaller than the symbol.

greater length ($p < 0.001$) than that of strips from the descending and sigmoid colon ($2.20 \pm 0.10 L_i$). The maximum active stress exerted by the circular muscle of the sigmoid colon was greater ($p < 0.001$) than that of the ascending, transverse or descending colon. In all parts of the colon, the active stress exerted by the circular muscle was less ($p < 0.001$) than that of either the taenial or inter-taenial longitudinal muscle (Table).

LENGTH-FREQUENCY CHARACTERISTICS

In order to compare the effect of increasing length on the contractile frequency of strips obtained from different parts of the colon, the contractile frequency has been expressed in relation to the length at which the maximum active stress was recorded, referred to as L_{max} .

(a) Longitudinal strips

The spontaneous contractile frequency of taenial and intertaenial longitudinal muscle obtained from all colonic regions increased with length between 0.5 – $1.0 L_{max}$ (Fig. 3a, b). The contractile frequencies of strips obtained from the ascending and transverse, ie right, colon were not significantly different ($p > 0.4$); these data are thus combined. Similarly, no significant difference ($p > 0.4$) was observed in the descending and sigmoid, ie left, colon, and these data are also combined. No consistent difference ($0.05 < p > 0.4$) was observed between the contractile frequencies of taenial strips obtained from the right or left colon between 0.5 – $1.0 L_{max}$. Further, the contractile frequencies of intertaenial longitudinal strips obtained from the left colon were not significantly different ($p > 0.1$) from those of taenial longitudinal strips obtained from either the right or left colon at lengths of 0.5 – $1.0 L_{max}$. The contractile frequencies of intertaenial longitudinal strips from the right colon were, however, lower ($p < 0.001$) than those of either intertaenial strips from the left colon or taenia from right and left colon at all lengths studied.

(b) Circular strips

The contractile frequency of intertaenial circular strips obtained from all parts of the colon did not vary significantly ($p > 0.4$) with increases in length between 0.3 – $1.0 L_{max}$ (Fig. 3c). The contractile frequency of strips obtained from the right and left parts of the colon showed marked regional variation (Fig. 4).

Above L_{max} , the contractile frequency of the intertaenial circularly-oriented strips increased. This increase in frequency could not be accurately quantified as contractions were observed at frequencies of 3–7 min, 8–15 min and 14–27 min; on

Fig. 3 Spontaneous contractile frequency of taenial (A) and intertaenial (B) longitudinal strips as a function of length. Shaded areas represent the mean ± 1 SD of data obtained from ascending and transverse (▨) and descending and sigmoid (▩) colon respectively. The spontaneous contractile of intertaenial circular strips (C) obtained from the ascending and right-transverse (upper panel ▨), left transverse (lower panel ▨) and descending and sigmoid (▩) colon are similarly expressed.

occasions these contractions summated and gave rise to contractions of prolonged duration which sometimes recurred at a frequency of 1–3 min (Fig. 5).

Fig. 4 Contractile frequency of circular strips obtained from different parts of the human colon at strip lengths $< L_{max}$.

Fig. 5 Spontaneous contractile activity of intertaenial circular strip from the ascending colon at lengths exceeding L_{max} . Note contractile activity at frequencies of 8–15 min and 14–27 min; these could summate and give rise to contractions of long duration.

Discussion

Spontaneous contractions of both circular and longitudinal muscle layers of the human colon were, in general, only observed when the baseline stress exceeded zero at lengths equal to or slightly greater than the initial length L_i . This suggests that some degree of stretch is required in order to elicit spontaneous contractile events from human colonic isolated muscle.

The active and baseline stress curves of both circular and longitudinal muscle layers of the right (ascending and transverse) colon were less affected by length changes than were the left (descending and sigmoid) colonic muscles. This would be consistent with the right colon being more distensible than the left and functioning as an accommodative reservoir in which the temporary storage, absorption and bacterial fermentation of content could proceed.² This marked difference between the right and left regions may be related, to their differing innervation, blood supply and embryological origin.¹ A further distinction could be made between the contractile properties of the right and left colon. Circular strips obtained from the right colon contracted at nearly twice the frequency of strips from the left colon at lengths less than L_{max} . A similar observation has been made *in vivo*.²⁴ This provides some evidence of a right-to-left colonic gradient.

The spontaneous active stress exerted by both circular and longitudinal muscle layers increased with length until an optimal length, L_{max} , was reached, and further length increases decreased the force generated. This is similar to findings reported by others in caecum^{17, 25} blood vessels,^{26, 27, 28} urinary bladder,²⁹ small intestine,³⁰ uterus³¹ and colonic^{13–15} smooth muscle preparations in which spontaneous contractile events have been arrested. The marked increase in baseline stress beyond L_{max} may serve, physiologically, to prevent overdistension of the colon and subsequently ensure that the contractile elements are not stretched beyond the point at which they can exert maximum active stress. This might be of importance in certain pathological conditions which result in over distension of the viscus, particularly as others have shown that stretching smooth muscle preparations to lengths greater than L_{max} is associated with an irreversible impairment in the ability to contract.^{32, 33} We defined the baseline stress as the minimum stress exerted by the smooth muscle during a cycle of spontaneous contraction. This definition is similar to that of Price *et al.*¹⁷ who concluded that the minimum stress exerted by the guinea pig taenia caecum during a cycle of spontaneous contraction was comparable to the passive stress exerted by a

preparation in which the spontaneous contractions had been arrested with adrenaline. We did not determine the passive properties of human colonic muscle after the abolition of spontaneous contractions and cannot exclude the possibility that the contractile element, as well as connective tissue and non-contractile elements, contributed to the baseline stress.

The absolute values for the maximum active stress of spontaneously-contracting human colonic muscle reported in this paper are not likely to reflect the maximum active stress that this muscle is capable of generating. Gabella³⁴ has shown that the active contractile stress exerted by guinea-pig taenia caecum in response to carbachol is greater than the active stress of spontaneous contraction at the same muscle length. Thus, we cannot assume that all the contractile elements of human colonic muscle are completely, or even partially, activated during spontaneous contraction. The maximum active stress exerted by the intertaenial longitudinal muscle layer was considerably greater than that of the taenia. This may reflect either a greater activation of the inter-taenial longitudinal muscle layer during spontaneous contraction or a higher muscle-to-connective tissue ratio in the inter-taenial region; histological assessment of the relative proportion of smooth muscle fibres to collagen and elastin is needed.

In conclusion, this study shows that spontaneous contractions of human colonic muscle are dependent upon length. The human colon appears to consist of two distinct areas, based on the mechanical behaviour of the muscle during spontaneous contraction *in vitro*, which may be related to their differing embryological origin and function.

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References

- Gray H. The large intestine. In: Williams PH, Warwick R. eds. *Gray's Anatomy*, 36th ed; Vol. 8. *Splanchnology*. Edinburgh: Churchill Livingstone, 1980: 1350-68.
- Phillips SF, Devroede GJ. Functions of the large intestine. In: Crane RK, ed. *International review of physiology, gastrointestinal physiology III*. Baltimore: University Park Press, 1979: 263-90.
- Meschan I. The large intestine. In: Meschan I, ed. *An atlas of normal radiographic anatomy*, 2nd ed. *The intestine beyond the duodenum; the biliary tract*. Philadelphia: W B Saunders Company, 1963: 635-4.
- Torsoli I, Fomorino ML, Cruciani V. The relationships between anatomy and motor activity of the colon. *Am J Dig Dis* 1968; **13**: 462-7.
- Ellis H. *Intestinal obstruction*. New York: Appleton-Century-Crofts, 1982.
- Eastwood MA, Watters DAK, Smith AN. Diverticular disease - is it a motility disorder? *Clin Gastroenterol* 1982; **11**: 545-61.
- Parks TG, Connell AM. Motility studies in diverticular disease of the colon. *Gut* 1969; **10**: 538-42.
- Smith AN, Shepherd J, Eastwood MA. Pressure changes after balloon distension of the colon wall in diverticular disease. *Gut* 1981; **22**: 841-4.
- Connell AM, Frankel H, Gottman L. The motility of the pelvic colon following complete lesions of the spinal cord. *Paraplegia* 1963; **1**: 98-115.
- Sleisinger MH, Silverberg M. Anatomy and developmental anomalies. In: Sleisinger MH, Fordtran JS, eds. *Gastrointestinal disease: pathophysiology, diagnosis, management*. Section 7. *The colon*. Philadelphia: W B Saunders Company, 1978: 1505-23.
- Truelove SC. Movements of the large intestine. *Physiol Res* 1966; **46**: 457-512.
- Gordon AR, Siegman MJ. Mechanical properties of smooth muscle. I. Length-tension and force-velocity relations. *Am J Physiol* 1971; **221**: 1243-9.
- Aberg AKG, Axelsson J. Some mechanical aspects of an intestinal smooth muscle. *Acta Physiol Scand* 1965; **64**: 15-27.
- Lowy J, Mulvany MJ. Mechanical properties of guinea pig taenia coli muscles. *Acta Physiol Scand* 1973; **88**: 123-6.
- Price JM, Patitucci PJ, Fung YC. Mechanical properties of resting taenia coli smooth muscle. *Am J Physiol* 1979; **236**: 211-20.
- Price JM, Patitucci PJ, Fung YC. Mechanical properties of taenia coli smooth muscle in spontaneous contraction. *Am J Physiol* 1977; **233**: 47-55.
- Duthie HL, Kirk D. Electrical activity of human colonic smooth muscle in vitro. *J Physiol* 1978; **283**: 319-30.
- Vanasin B, Ustach TJ, Schuster MM. Electrical and motor activity of human and dog colon in vitro. *John Hopkins Med J* 1974; **134**: 201-210.
- van Merwyk AJ, Duthie HL. Characteristics of human colonic smooth muscle in vitro. In: Christensen J, ed. *Gastrointestinal motility*. New York: Raven Press, 1980: 473-8.
- Golenhofen K. Slow rhythms in smooth muscle (minute rhythm). In: Bulbring E, Brading AF, Jones AW, Tomita T. *Smooth muscle*. Baltimore: Williams and Wilkins 1970: 316-42.
- Huizinga JD, Diamant NE, El-Sharkawy TY. Electrical basis of contractions in the muscle layers of pig colon. *Am J Physiol* 1983; **245**: 482-91.
- El-Sharkawy TY. Electrical activities of the muscle layers of the canine colon. *J Physiol* 1976; **263**: 199-213.
- Christensen J, Caprilli R, Lund GF. Electric slow waves in circular muscle of cat colon. *Am J Physiol* 1969; **217**: 771-6.
- Kerlin P, Zinsmeister A, Phillips S. Motor responses to food of the ileum, proximal colon, and distal colon of healthy humans. *Gastroenterology* 1983; **84**: 762-70.
- Bulbring E, Kuriyama H. The effect of adrenaline on

- the smooth muscle of guinea-pig taenia coli in relation to the degree of stretch. *J Physiol* 1963; **169**: 198–212.
- 26 Dobrin PB. Influence of initial length on length-tension relationship of vascular smooth muscle. *Am J Physiol* 1973; **225**: 664–70.
 - 27 Herlihy JT, Murphy RA. Force-velocity and series elastic characteristics of smooth muscle from the pig carotid artery. *Circ Res* 1974; **34**: 461–6.
 - 28 Paul RJ, Peterson JW. Relation between length, isometric force, and O₂ consumption rate in vascular smooth muscle. *Am J Physiol* 1975; **228**: 915–22.
 - 29 Uvelius B. Isometric and isotonic length-tension relations and variations in cell length in longitudinal smooth muscle from rabbit urinary bladder. *Acta Physiol Scand* 1976; **97**: 1–12.
 - 30 Meiss RA. Some mechanical properties of cat intestinal muscle. *Am J Physiol* 1971; **220**: 2000–2007.
 - 31 Csapo A, Goodall M. Excitability, length-tension relation and kinetics of uterine muscle contraction in relation to hormonal status. *J Physiol* 1954; **126**: 384–95.
 - 32 Paul RJ. Smooth muscle: mechanochemical energy conversion relations between metabolism and contractility. In: Johnson LR, ed. *Physiology of the digestive tract*. New York: Raven Press, 1981: 269–88.
 - 33 Peterson JW, Paul RJ. Effects of initial length and active shortening on vascular smooth muscle contractility. *Am J Physiol* 1974; **227**: 1019–24.
 - 34 Gabella G. The force generated by a visceral smooth muscle. *J Physiol* 1976; **263**: 199–213.