Effects of intraluminal distention and decompression on microvascular permeability and hemodynamics of the equine jejunum

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Objective—To determine whether intraluminal distention and subsequent decompression of the equine jejunum affects intestinal blood flow, hemodynamics, and microvascular permeability.

Animals—5 healthy adult horses.

Procedure—Horses were anesthetized and underwent exploratory laparotomy. Two jejunal segments were identified as sham-operated or instrumented segments. After baseline values were obtained, intraluminal distention was created in the experimental segment to induce an intraluminal pressure of 18 cm H2O. After 120 minutes of distention, the intestine was decompressed for 120 minutes. Mesenteric blood flow, oxygen delivery, oxygen consumption, microvascular permeability, wet weight-to-dry weight ratio, neutrophil infiltration, and vascular resistance were determined and comparisons made among control, sham-operated, and experimental segments.

Results—Mean jejunal blood flow was 21.4 ml/min per kg. There was a significant decrease in mesenteric blood flow to the distended intestine (13.4 ml/min per kg). Blood flow increased significantly during the decompression period (340% of baseline blood flow). Intraluminal distention and subsequent decompression resulted in a significant increase in microvascular permeability, as determined by the osmotic reflection coefficient. Oxygen delivery and oxygen content decreased significantly during the distention period and increased during decompression. Morphologic evaluation revealed a significant increase in edema and neutrophil infiltration after distention and decompression, compared with results for the sham-operated or control segments.

Conclusions and Clinical Relevance—Intraluminal distention and decompression of the equine jejunum results in low-flow ischemia and edema, which may contribute to adhesions and ileus in the postoperative period after surgery for obstructions of the small intestines. (Am J Vet Res 2001;62:225–236)

Horses with obstructions of the small intestines have a guarded prognosis because of commonly encountered postoperative complications such as continued necrosis, ileus, and adhesions. During small intestinal obstruction, intraluminal distention develops proximal to the obstructive site as a result of accumulation of ingesta, gas, and intestinal secretions. Current recommendations include removal of devitalized intestine and decompression of the remaining distended intestine.

The amount of intraluminal distention is associated with survival. Horses surviving surgery for small intestinal obstruction had a significantly lower intraluminal pressure in the segment proximal to the primary lesion than horses that did not survive. When pressures similar to those measured in affected horses were created in foals, 2 hours of distention caused severe morphologic serosal damage and adhesions 10 days after surgery.

Intestinal obstruction is initiated by mechanical obstruction or paralytic ileus. Once obstructed, an imbalance in intestinal secretion and resorption leads to progressive distention and increased pressures. As intraluminal pressure increases, it is transmitted through the interstitium to the veins, resulting in obstruction of venous outflow. Increased venous pressure increases capillary hydrostatic pressure with excessive capillary filtration, which causes edema, increased interstitial pressure, and, eventually, compromised arteriolar blood flow.

Effects of luminal distention on intestinal blood flow and hemodynamics have been studied in the small intestines of other species. Numerous authors have reported a decrease in intestinal blood flow and an increase in vascular resistance as intraluminal pressure increases. In addition, Granger et al reported that luminal distention increases transcapillary fluid exchange and lymph flow in the small intestines of cats. Few investigators have examined the effects of intraluminal distention on the microvasculature of the small intestines of horses. We previously documented that 120 minutes of intraluminal distention at a pressure of up to 23 cm H2O (18.4 mm Hg) followed by 120 minutes of decompression decreased the number of blood vessels perfused in the seromuscular layer. Although intraluminal distention and decompression caused minimal morphologic damage, the seromuscular layer had an excessive number of infiltrating neutrophils and edema. Because blood flow was not measured in that study, few conclusions could be drawn regarding hemodynamic events.

The purpose of the study reported here was to determine whether intraluminal distention and subse-
quent decompression of the equine jejunum affects intestinal blood flow or hemodynamics. We also attempted to determine whether intraluminal distention and decompression of the jejunum alters intestinal transcapillary fluid exchange and endothelial cell morphology.

Materials and Methods

Horses—Five healthy adult Thoroughbred geldings, 4 to 8 years old (mean age, 5.7 years), were used in the study. Horses did not have evidence of parasitic, gastrointestinal tract, or systemic disease as determined on the basis of results of physical examination and a CBC. Effects of intraluminal distention of the jejunum at a pressure of 18 cm H2O for 120 minutes and subsequent decompression on microvascular permeability, blood flow, hemodynamics, and intestinal morphology were determined. The experimental protocol was approved by the Institutional Animal Care and Use Committee at the Virginia Polytechnic Institute and State University.

Anesthetic protocol—Horses were sedated with xylazine hydrochloride \( (0.2 \text{ to } 0.5 \text{ mg/kg of body weight), IV,} \) and ketamine hydrochloride \( (2.2 \text{ mg/kg, IV}) \), and anesthesia was maintained with halothane in oxygen, using intermittent positive-pressure ventilation. Mean blood pressure measured by direct arterial catheterization was maintained at 70 mm Hg or higher. Lactated Ringer's solution was administered IV at a rate of 5 to 10 ml/kg per h to maintain circulating volume and blood pressure. To maintain systolic arterial blood pressure between 100 and 120 mm Hg, dobutamine and blood pressure. To maintain systolic arterial blood pressure between 100 and 120 mm Hg, dobutamine was maintained with halothane in oxygen, using intermittent positive-pressure ventilation. Mean blood pressure measured by direct arterial catheterization was maintained at 70 mm Hg or higher. Lactated Ringer's solution was infused into the segment to induce an intraluminal pressure of 18 cm H2O. Intraluminal pressure was monitored, using a pressure transducer, and pressure in the segment was maintained by infusion of fluid (as needed) for 120 minutes. Mesenteric blood flow, mesenteric venous pressure, and intraluminal pressures were recorded every 15 minutes throughout the experiment.

At the end of the distention period, the bowel was decompressed by removing the intraluminal fluid. The segment was allowed to reperfuse for 120 minutes. Lymph samples were collected over a 15-minute period at baseline venous pressure (0 to 3 mm Hg). Once the lymph flow rate was stable, venous pressure was increased to 30 mm Hg. Lymph and mesenteric blood samples and lymph flow and mesenteric blood flow rates were obtained at 15-minute intervals until the lymph flow rate remained constant, which required 45 to 60 minutes.

After sample collection was completed and the intraluminal intestinal fluid was drained from the segment, the experimental, sham-operated, and control intestinal segments were weighed. Full-thickness intestinal biopsies were collected from the experimental, sham-operated, and control segments. A 10-cm section of each of the experimental, sham-operated, and control segments was weighed and stored at −70 °C. These sections were incubated at 60 °C for 24 hours, and the dry weight of each sample then was recorded. Data on dry weight were used to calculate lymph and blood flow per kilogram of tissue for each horse and to measure wet weight: dry weight (WW:DW) on full-thickness jejunum as an estimate of jejunal tissue edema. After completion of each experiment, anesthetized horses were euthanatized by IV administration of an overdose of sodium pentobarbital.

Laboratory analysis—Lymph volume was measured in calibrated micropipettes. Lymph and blood samples obtained for analysis of TP content were centrifuged to remove RBC, and protein values were measured by use of the modified biuret method. The pH, Pco2, and Po2 were measured in systemic and mesenteric arterial and jejunal venous blood samples. Blood gas and acid-base analysis of blood samples were determined, using an acid-base analyzer.

Osmotic reflection coefficient analysis—Microvascular permeability was determined by estimating the osmotic reflection coefficient (ORC), which was determined by using the steady-state relationship between lymphatic flow rate per kilogram of tissue and the lymphatic protein concentration-to-plasma protein concentration ratio (Cl: Cp).

Other reports have indicated that 1–(Cl: Cp) at filtration-independent lymph flow rates provides an accurate estimate of the ORC. Value for the Cl: Cp was plotted against the value for lymphatic flow rate per kilogram of intestinal tissue. Mean Cl: Cp for the 4 highest lymph flow rates per horse was calculated. Mean and SEM were calculated to determine flow-independent Cl: Cp, which was at the maximal lymph
flow rate. Comparisons of the minimal value of Cl:Cp at filtration-independent lymph flow rates were made between horses reported here and horses with normal jejunum reported elsewhere.11

Metabolic calculations—Arterial oxygen content (CaO₂) and jejunal venous oxygen content (JCVO₂) were calculated as the sum of oxygen bound to hemoglobin (Hb X percentage oxygen saturation) and oxygen dissolved in plasma (PO₂ X 0.003). Oxygen delivery to the experimental jejunal segment (Do₂jej,jjunum) was estimated as the product of CaO₂ X jejunal blood flow (Qjejunum). Jejunal oxygen consumption (Vo₂jej,jjunum) was estimated as the product of intestinal blood flow (Qe, Qjej,jjunum) times the difference between CaO₂ and JCVO₂. Jejunal oxygen extraction ratio (O₂extraj) was calculated as (CaO₂–JCVO₂)/CaO₂.

Intestinal vascular resistance (Rintvas) was estimated by dividing change in arterial and mesenteric venous perfusion pressure by intestinal blood flow.

Tissue preparation—Intestinal tissue specimens were collected at the start of each experiment and 300 to 360 minutes after induction of anesthesia. Jejunal tissue specimens were washed with sterile lactated Ringer’s solution and were pinned flat, with the serosal side down, on wooden tongue depressors. A 3 X 3-mm section of biopsied tissue was removed and placed in freshly prepared cacodylate-buffered 1% osmium tetroxide solution. Samples were dehydrated and embedded. Thick sections (500 µm) were stained with toluidine blue and basic fuchsin and examined, using light microscopy.12 Selected thin sections were stained with uranyl acetate and lead citrate for transmission electron microscopy to allow evaluation of endothelial cell structure in the serosa and mucosal integrity.9

The remaining section of tissue was placed in neutral-buffered 10% formalin for 48 hours. Fixed tissues were trimmed, embedded in paraffin, cut in 4-µm-thick sections, stained with H&E, and examined by use of light microscopy.

Quantitative morphology—Mucosal, submucosal, longitudinal, and circular muscle layers and serosal intestinal layers were measured, using a microscope with a calibrated cursor on the 10X objective. A 1.0-mm² grid was situated at the extraluminal surface of the serosal layer, and measurements were made of serosal thickness, using a 20X objective. Seromuscular edema or thickness was measured as the distance between the mesothelial cell layer of the serosa and the circular muscle layer. The circular and longitudinal muscle layers also were measured independently. Submucosal thickness was measured as the distance between the muscularis mucosa and circular muscle layer. Six views of each intestinal layer from each tissue section were measured, and mean thickness was calculated for each horse. Comparisons were made among the experimental, sham-operated, and control jejunal segments for each horse. Results for each horse were expressed as a proportion of the value calculated from the tissue section obtained from the control jejunum of that horse (baseline).

Quantitative assessment of neutrophil infiltration into the serosal layer was performed by 1 of the investigators (RMD), who was not aware of the origin of each jejunal segment. Number of neutrophils in a 0.01-mm² area were counted in the serosa directly below the mesothelium, using the oil-immersion objective of a microscope equipped with an ocular grid; the grid was positioned adjacent to the serosal mesothelium. For each horse, number of neutrophils was counted in 6 adjacent fields/section.

Statistical analysis—Mean Cl:Cp for the 4 highest lymph flow rates for each horse was determined. Mean and SEM were calculated to determine Cl:Cp for the flow-independent portion of the data. The Mann-Whitney U test was used to determine differences in Cl:Cp between values for the horses reported here and values for adult horses with normal jejunum reported elsewhere.11 Significance was set at a value of P ≤ 0.05.

Changes in jejunal blood flow, pH, PCO₂, base excess, HCO₃⁻, JCVO₂, Do₂jej,jjunum, Vo₂jej,jjunum, O₂extraj, Rintvas, and number of serosal neutrophils were compared with baseline values for all horses by use of single repeated-measures ANOVA. Differences between means were evaluated by use of the Student-Newman-Keuls procedure (significance defined as values of P < 0.05).

For each horse, ratios of thickness of the sham-operated and experimental segments to the thickness of the control segment of jejunum were calculated. Mucosal, submucosal, muscularis, and serosal thickness were compared among experimental, sham-operated, or control jejunal segments. Differences between thickness of the control, sham-operated, and experimental jejunal segments were calculated for each horse, and the hypothesis that the difference was 0 was tested by use of the Kruskal Wallis 1-way nonparametric ANOVA. When a difference was detected, the Mann-Whitney U test was used for comparison, with a value of P < 0.05 considered significant.

Results
Mean arterial pressure did not differ among horses during the 6-hour anesthetic period, and there was not a significant difference in mean arterial pressure over time during the experimental procedure in any horse. Measured systemic arterial and mixed-venous blood gas variables did not differ among horses or between baseline, distention, or decompression periods. The CaO₂ did not differ among horses or within any horse over time during the experiment.

All horses required periodic additional infusion of lactated Ringer’s solution into the experimental segment (approx 20 ml/30 min) to maintain an intraluminal pressure of 18 cm H₂O throughout the 120-minute distention period. Additions of fluid were made when the intraluminal pressure decreased by 1 or 2 cm H₂O. Mean ± SEM baseline blood flow to the experimental segment was 21.4 ± 0.8 ml/min per kg. There was a significant (P < 0.001) decrease in blood flow between 15 to 90 minutes after onset of intraluminal distention with the lowest mean flow 60 minutes after onset of distention (13.4 ± 1.5 ml/min per kg), which was 60% of baseline flow. Jejunal blood flow gradually increased during the distention period and approximated baseline flow by 120 minutes (20.4 ± 1.0 ml/min per kg). There was a significant (P < 0.001) increase in blood flow within 5 minutes after initiation of decompression to 65.0 ± 2.8 ml/min per kg (304% of baseline), with blood flow remaining increased but gradually returning to baseline values within 135 minutes after initiation of decompression (Fig 1). There was a significant (P = 0.003) decrease in blood flow 150 minutes after initiation of decompression (18.1 ± 1.0 ml/min per kg), which was 84% of baseline flow.

Mean ± SEM baseline venous pressure in the experimental jejunal segment was 2.0 ± 0.7 mm Hg and did not differ significantly among horses. Jejunal venous pressure increased significantly (P < 0.001)
during the first 60 minutes after initiation of distention (11.7 ± 0.8 mm Hg) then decreased slightly during the second 60 minutes after initiation of distention (7.75 ± 0.4 mm Hg) and during the initial 5 minutes after onset of decompression (8.0 ± 0.6 mm Hg). The $R_{\text{intvas}}$ in the experimental jejunal segment increased significantly ($P < 0.001$) to 193% of the baseline value within 60 minutes after initiation of distention, and it then gradually decreased to 79% of the baseline value by the end of the 120-minute distention period. Within 5 minutes after onset of decompression, $R_{\text{intvas}}$ decreased significantly ($P = 0.003$) to 31% of the baseline value, but it increased to 90% of the baseline value by the end of the decompression period (Fig 2).

Mean baseline values for lymph flow rate was 10.1 ± 0.9 µl/min per kg of jejunum for all horses at a baseline mesenteric venous pressure of 0 to 3 mm Hg. Lymph flow rate increased slightly (14.1 ± 0.3 µl/min per kg) during the distention period and increased significantly (23.1 ± 2.0 µl/min per kg) during the initial portion of the decompression period. After a constant lymph flow rate was established at a mesenteric venous pressure of 8 to 10 mm Hg, stepwise increases in venous pressure resulted in incremental increases in lymph flow rate (up to 7.5 times the baseline lymph flow rate). In all horses, Cl:Cp decreased rapidly as lymph flow rate increased, and Cl:Cp became stable at high lymph flow rates.

Intraluminal distention to a pressure of 18 cm H$_2$O for 120 minutes followed by decompression resulted in a mean Cl:Cp of 0.36 ± 0.07 for the filtration-independent portion of the curve, which was a significant ($P < 0.001$) increase, compared with mean Cl:Cp (0.19 ± 0.06) in the normal jejunum of horses reported elsewhere (2).

Baseline blood gas values for mesenteric arterial and venous blood samples did not differ significantly among horses. Mean jejunal venous pH decreased significantly ($P = 0.002$) 120 minutes after initiation of intraluminal distention (Fig 4). Jejunal pH began to increase during the decompression period, although it did not reach baseline values. Jejunal venous PCO$_2$ increased significantly ($P = 0.006$) from a baseline value of 35 mm Hg to a value of 41 mm Hg 120 minutes after initiation of distention, and it continued to increase to a value of 47.2 mm Hg after the 120-minute decompression period. Jejunal venous PO$_2$ decreased significantly ($P = 0.003$) from a baseline value of 40.2 mm Hg to a value of 27.1 mm Hg 120 minutes after initiation of intraluminal distention, and it then increased to 35.0 mm Hg 120 minutes after onset of decompression. Jejunal venous HCO$_3^-$ ($P = 0.005$) and base excess ($P = 0.018$) decreased significantly 120 minutes after initiation of intraluminal distention (from 23.9 to 17.3 mEq/L and from $–5.3$ to $–7.7$, respectively). Jejunal venous HCO$_3^-$ and BE increased to 23.3 mEq/L and $–2.9$, respectively, after decompression.

Values for jejunal oxygen metabolism were determined (Table 1). The $JCV_{O_2}$ decreased to 70% of the baseline value at the end of the distention period; it then increased after decompression. Value for $DO_{2\text{jej}}$ decreased significantly ($P < 0.001$) to 46% of baseline within 60 minutes after initiation of distention, and it then decreased further to 12.3% of baseline by 120 minutes after initiation of distention. Within 5 minutes of baseline of decompression, $DO_{2\text{jej}}$ increased significantly ($P = 0.005$) by 288% and was at 93% of baseline by 120 minutes after onset of decompression. The $VO_{2\text{jej}}$ decreased significantly ($P = 0.003$) to 30% of baseline during the distention period, but it increased significantly ($P = 0.006$) by 208% within 5 minutes after onset of decompression and was 64% of baseline 120 minutes after onset of decompression. The $O_2$ decreased significantly ($P = 0.003$) from a baseline value of 40.3% to a value of 25% 120 minutes after initiation of distention, but it increased to 31.5% at the end of the decompression period.

The experimental jejunal segment had a thin wall and appeared pinkish-gray 120 minutes after initiation of dis-
Within 5 minutes after onset of decompression, the experimental segment became slightly hyperemic with increased spontaneous motility. Jejunal histomorphology of control samples were normal in all horses. Jejunal samples obtained from the sham-operated segment after 6 hours outside the abdomen had normal mucosal morphology with a mild increase in submucosal and serosal edema and serosal mesothelial cell loss (Fig 5). After the 120-minute distention period and 120-minute decompression period, the mucosa appeared normal but had dilated central lacteals in the villi. There was moderate-to-severe edema in the submucosa and seromuscular layers with increased WBC infiltration and hemorrhage in the serosal layer (Fig 6).

We did not detect a significant difference between WW:DW of full-thickness control segments among horses. There was an increase, but not significant, for WW:DW in the sham-operated segments (60% increase over control segments). There was a significant \( P < 0.001 \) increase in WW:DW (150%) in the experimental jejunal segment after distention and decompression, compared with control and sham-operated segments (Fig 7).

Mean thickness of the mucosal, submucosal, circular, or longitudinal muscle layers or serosal layer did not differ significantly between the control or sham-operated segments. There was a pattern for increased submucosal thickness in the sham-operated jejunal segments, compared with values for control segments. There was a significant \( P < 0.001 \) increase in thickness of the submucosal layer (250%), circular muscle layer (160%), and serosal layer (270%) in experimental segments, compared with control and sham-operated segments (Fig 8).

Number of WBC in the serosa was significantly \( P = 0.03 \) increased in the sham-operated segments (9.3 cells/0.01 mm\(^2\)), compared with control segments (1.0 cells/0.01 mm\(^2\)). There was a significant \( P < 0.001 \) increase (39.5 cells/0.1 mm\(^2\)) in number of neutrophils in the serosal layer of the experimental segment after distention and decompression, compared with control or sham-operated segments (Fig 9).

Evaluation of the mucosa after distention and decompression by use of transmission electron microscopy revealed minimal gaps between adjacent luminal epithelial cells with an intact microvilli surface, but there was edema and WBC infiltration at the mucosal basement membrane (Fig 10). Muscle cells in the circular and longitudinal muscular layers were swollen and separated by edema. After distention and decompression, muscle cells had mitochondrial swelling and cellular vacuolation (Fig 11), which was not evident in muscle cells of the control segment.
Figure 5—Photomicrographs of a section of submucosa (SM; left) and serosa (S; right) from a sham-operated jejunal segment of a horse. Notice the dilatation of the central lacteal in the mucosal villi and the amount of submucosal edema. H&E stain; bar = 15 µm.

Figure 6—Photomicrographs of a section of submucosa (SM; left), circular muscle (CM) layer, longitudinal muscle (LM) layer, and serosa (S; middle), and serosa (right) from a segment of equine jejunum in the experimental group after distention and decompression. Notice the increased edema in the submucosa, compared with the sham-operated segment. In the serosa, increased cellular infiltrate is evident, especially at the outer serosal surface (arrows). H&E stain; bar = 15 µm.
Ultrastructure of capillaries in the control segments appeared normal (Fig 12). After distention and decompression, the capillaries had swollen endothelial cells, which decreased the capillary lumen (Fig 13). Endothelial cells had evidence of vacuolation with endoplasmic reticular dilation and mitochondrial swelling.

**Discussion**

Constant intraluminal pressure of 18 cm H₂O in the equine jejunum caused a significant decrease in blood flow to the segment during the period of intraluminal distention. Numerous studies in other species have documented a decrease in blood flow with increased intraluminal distention, although the magnitude of intraluminal pressure needed to alter mesenteric blood flow has not been consistent.6-8,15,16 A stepwise increase in intraluminal pressure reduced blood flow in a stepwise manner in the jejunum of cats,7 but other reports indicate that mesenteric blood flow is not
altered until an intraluminal pressure of 100 mm Hg is reached. The intraluminal pressure required to alter blood flow is dependent on experimental methods used. The intraluminal pressure required to alter mesenteric blood flow is much higher in preparations of isolated denervated intestinal loops, compared with intestines that have an intact blood and nerve supply.

Intra-abdominal pressure also is a factor, because it requires less intraluminal pressure to decrease mesenteric blood flow when the intestine is maintained within the abdomen, which is attributable to the added extraluminal pressure of the peritoneal cavity. We chose an extra-abdominal intraluminal pressure of 18 cm H₂O in an intact intestinal model. This intraluminal pressure was chosen because similar extra-abdominal intraluminal pressures were observed in clinical cases involving horses with small intestinal obstruction. Also, other studies of distention in foals and adult horses revealed morphologic and microvascular abnormalities at intraluminal pressures of 25 cm H₂O measured within the abdomen, which decreases to 18 cm H₂O when the intestines are removed from the abdomen.

Increased intraluminal pressure resulted in a decrease in mesenteric blood flow that slowly recovered with time. Maximum decrease in blood flow was detected within the first 15 minutes after initiation of distention (46% of baseline blood flow), after which there was a progressive increase in blood flow such that 120 minutes after initiation of distention, mesenteric blood flow recovered to 93% of baseline flow (Fig 1). Other studies of intestinal function designed to maintain a constant intraluminal pressure over time have revealed similar findings. This stress relaxation or delayed compliance is an intrinsic mechanism by which the intestine responds to additional luminal fluid or gas accumulations. Each increment of added fluid causes a steep increase in intraluminal pressure and decrease in blood flow; the intestinal wall then relaxes and stretches to accommodate the increased volume, thereby reducing the intraluminal pressure.
This relaxation is followed by a recovery of blood flow, which is less complete as distention progresses. Delayed compliance probably developed in our study because we had to repeatedly add fluid to the intestinal lumen to maintain a constant intraluminal pressure. This mechanism explains the recovery of blood flow during the distention period in our study.

Intestinal blood flow is regulated by intrinsic, extrinsic, and humoral factors. The intrinsic ability of the intestines to control perfusion and oxygenation during changing tissue demands is attributed to metabolic and myogenic influences on the local resistance and exchange vessels. Myogenic control of intestinal autoregulation is based on the assumption that vascular resistance is directly related to arterial transmural pressure attributable to smooth-muscle receptors detecting stretch of the vessel. Increased venous pressure attributable to smooth-muscle receptors causes vasoconstriction. Therefore, increases in venous pressure cause increased vascular resistance and a reduction in number of perfused capillaries.

In the study reported here, mesenteric venous pressure increased significantly within the first 60 minutes after initiation of intraluminal distention, and it then gradually decreased 120 minutes after initiation of distention. The mesenteric vascular resistance (Fig 2) paralleled venous pressure during the distention period in this study. Another study of equine intestines revealed that 120 minutes of intraluminal distention at 25 cm H2O caused a significant decrease in the number of perfused blood vessels in all intestinal layers. The increased vascular resistance and decreased capillary perfusion associated with intraluminal distention would result in the decreased blood flow seen during the distention period in our study.

Within 5 minutes after onset of decompression, mesenteric blood flow to the segment increased to 303% of baseline flow; it then gradually decreased to 84% of baseline flow 235 minutes after onset of decompression. Others have reported an initial increase in blood flow after deflation of distended small intestine in other species; however, blood flow never returned to predistention values, and vascular resistance remained higher than control values. Reactive hyperemia describes the increased blood flow seen after periods of arterial occlusion in ischemic intestine, which is an attempt to repay the oxygen debt sustained during the ischemic period. In a previous study, we observed an immediate 231% increase in jejunal blood flow following 60 minutes of low-flow ischemia (25% of baseline blood flow), but flow then decreased to 60% of baseline values 4 hours after onset of reperfusion. It appears that blood flow in the equine small intestine responds to intraluminal distention and subsequent decompression in a manner similar to that after low-flow ischemia and reperfusion.

The increased blood flow observed after decompression is explained by the metabolic theory of intestinal autoregulation, which predicts that inadequate oxygenation of parenchymal cells causes production of vasodilator metabolites responsible for dilation of resistance vessels and opening of precapillary sphincters that increase the number of perfused capillaries. Metabolites that have vasodilator effects on mesenteric circulation include hydrogen ions, potassium ions, PCO2, adenosine, purine metabolite accumulations, and nitric oxide. Capillary recruitment increases the surface area for oxygen exchange and decreases the blood-to-cell diffusion distance. The decreased pH and P O2 in jejunal venous blood and increases in jejunal PCO2 during distention suggest the development of tissue hypoxia (Fig 4). The significant decrease in HCO3- and base excess observed during the intraluminal distention period may represent the buffering capacity of the intestine in response to decreasing pH. The DO2jejunum also decreased significantly from baseline during distention, indicating the possibility for an oxygen debt in the tissues. Similar results have been described in the large colon of horses after a 3-hour period of low-flow ischemia and in the large colon of ponies after application of a 720° volvulus. Acidemia, hypercarbia, and hypoxia observed during the distention period in the study reported here may have induced vasodilation and capillary recruitment, causing the increased blood flow that we observed in the initial portion of the decompression period. The return of blood flow to baseline values seen during the latter part of the decompression period coincided with a gradual increase in jejunal pH and decrease in PCO2. This also has been reported for the equine large intestine during reperfusion after an ischemic insult. According to the metabolic theory, return of blood flow washes out accumulated metabolites (hydrogen and potassium ions, PCO2, adenosine), thereby removing the signal for vasodilation and returning the blood flow to baseline values.

Because oxygen delivery is the volume of oxygen delivered to a vascular bed per minute and is a function of blood flow, it was not surprising that DO2jejunum decreased during intraluminal distention and then increased during decompression. Similarly, oxygen consumption decreased significantly during the initial 60 minutes after initiation of intraluminal distention, and it then gradually increased by the end of the distention period to parallel oxygen delivery and local blood flow. Oxygen consumption is the amount of oxygen that diffuses from the capillaries to the intestinal tissue and is a function of tissue activity, viability, and blood flow.

Oxygen consumption is relatively constant until a minimal flow rate is reached, after which it decreases linearly with blood flow, indicating that oxygen is being consumed. The decreased JCVO2 observed during the distention period suggests that tissue oxygenation was compromised during intraluminal distention. Decreased intracellular P O2 should elicit arteriolar vasodilation and capillary recruitment by intrinsic metabolic mechanisms. The increase in blood flow seen within 5 minutes after onset of decompression was associated with a significant increase (148% above baseline) in oxygen consumption, which then gradually decreased and was 70% of baseline at the end of the decompression period. The increased oxygen consumption after decompression probably was attributable to vasodilation that facilitates replacement of the oxygen debt that developed during the low-flow period.
Oxygen extraction significantly decreased from a baseline value of 40% to 25% 120 minutes after initiation of intraluminal distention, and it then increased to 31.5% after the decompression period (Table 1). Other studies have revealed reduction in oxygen extraction during intraluminal distention of the small intestines. This accumulation activity of circular muscle and may be an important mechanism in postoperative ileus. The accumulation of neutrophils in the small intestine, an increase in venous pressure secondary to intraluminal distention results in increased vascular resistance and reduction in capillary density, which also was found in the study reported here. Because oxygen extraction is directly proportional to the number of perfused capillaries (capillary surface area), reducing the number of perfused capillaries or increasing capillary-to-cell diffusion distance with edema would cause decreased oxygen extraction.

The ORC describes the fraction of the total oncotic pressure generated across a capillary membrane. The ORC determined by Cl:Cp is representative of capillary leakage. In the study reported here, we observed a significant decrease in the ORC to 0.64 120 minutes after initiation of intraluminal distention followed by a 120-minute decompression period, compared with ORC of normal equine jejunum, which is 0.81. Changes in microvascular permeability can result from many factors, including vasoactive mediators, endotoxins, and reactive oxygen metabolites generated from local tissue hypoxia or activated neutrophils. There is substantial evidence to suggest that reactive oxygen metabolites such as xanthine oxidase generate superoxide ion-mediated changes in capillary permeability in postischemic intestinal injury. Other studies in horses have documented significant increases in xanthine oxidase after ischemia-reperfusion of the small intestine. We did not measure xanthine oxidase or reactive oxygen metabolites in our study, but we did induce an ischemic injury with intraluminal distention-decompression such that these factors may have caused the changes in capillary permeability that we observed.

We detected a 40-fold increase in neutrophils in the serosal layer after distention-decompression, compared with values for the control segments (Fig 5). Ischemic injury of endothelial cells can activate nearby neutrophils resulting in chemotaxis, adherence to postcapillary venules, and eventual migration into surrounding tissues. WBC interact with plasma membrane receptors resulting in the release of large amounts of reactive oxygen metabolites and enzymes (myeloperoxidase and catalase), causing damage to vascular endothelium that results in protein leakage. Granulocyte adherence to postcapillary venules can increase vascular resistance and decrease mesenteric blood flow in other species. Experimentally, the accumulation of leukocytes within the small intestine of rodents was accompanied by reduction in contractile activity of circular muscle and may be an important mechanism in postoperative ileus. The accumulation of neutrophils suggests that neutrophils contribute to the permeability changes seen after distention-decompression in a manner similar to that observed in ischemia-reperfusion models used for investigating intestinal permeability. It also is possible that the increased vascular resistance and decreased mesenteric blood flow seen at the end of the decompression period was attributable to leukocytes adhered to the vascular endothelium. Whether it was reactive oxygen metabolites altering endothelial cells or the cellular inflammatory response, it appears that, regardless of the mechanism, intestinal distention sufficiently compromises blood flow to cause reperfusion injury.

A significant 150% increase in WW:DW was detected in the experimental segments after distention-decompression injury, compared with values for control segments (Fig 7). Partial increase in weight was attributed to the experimental model, because sham-operated segments had a 60% increase in weight, compared with control segments. This was expected, because other investigators of equine intestines have reported mild intestinal edema in control segments after prolonged anesthesia and have suggested that inflammation from an open peritoneal cavity was the cause. Using this model for calculating the ORC in normal equine jejunum, we found a 73% increase in weight of experimental segments, compared with control segments, which was not significantly different from the sham-operated segments. The model itself apparently causes some edema. If 60 to 70% of the fluid accumulation was caused by the model, the distention-decompression injury still resulted in an additional 90% increase in weight. This is similar to but less than the 400% increase in WW:DW observed after a 60-minute period of low-flow ischemia and reperfusion in equine jejunum.

Morphologic changes seen after distention and decompression correlate with the WW:DW findings. Similar to results of other intestinal distention studies in horses, minimal mucosal damage was seen, but a substantial amount of submucosal and seromuscular edema, as well as increased neutrophil infiltration and mesothelial cell loss in the serosa, was observed. The increased tissue thickness was localized to the submucosa (250% increase over baseline), circular muscle layer (160%), and serosal layer (270%; Fig 8). These morphologic changes were primarily located in the subserosal layers, which could be associated with peritoneal adhesions described after intraluminal distention and decompression in foals of another report.

Edema is a manifestation of excess accumulation of fluid in the interstitium and frequently is reported as a gross and histologic finding after decompression of previously distended small intestine in horses. Edema is caused by lymphatic obstruction, alterations in capillary hydrostatic and oncotic pressures, or increased microvascular permeability. If capillary pressure increases secondary to increased venous pressures (as a result of intraluminal distention), fluid moves from the vascular system to the interstitium. Granger et al documented an increase in capillary filtration rate in the jejunum of cats during increased luminal distention that was attributed to enhanced capillary permeability and changes in capillary hydrostatic pressure. We did not measure capillary hydrostatic pressures, but we did observe increased capillary permeability and increased venous pressure secondary to increased luminal distention in our study. The intestinal edema observed in this study was probably...
attributable to a combination of changes in capillary permeability and alterations in capillary hydrostatic and oncotic forces.

When capillary hydrostatic pressure or permeability increases, interstitial fluid accumulates and eventually leads to exudation of interstitial fluid into the intestinal lumen. Many studies in other species describe increased secretion of fluid and electrolytes into the small intestines during or after increased luminal distention. The additional fluid accumulation within the intestinal lumen further increases intraluminal pressure, which causes additional vascular compromise. This cycle leads to increased tissue damage, which emphasizes the importance of decompressing the distended bowel at the time of surgery for horses with small intestinal obstructions.

Damage to endothelial cells observed in the serosal capillaries and venules suggested that the capillary permeability changes seen were mediated by damage attributable to cellular hypoxia and reperfusion. Retraction of endothelial cells and gap formation are caused by various inflammatory mediators such as histamine, prostaglandins, leukotrienes, platelet-activating factors, or cytokines. These mediators can be generated from leukocytes that adhere to the vascular endothelium or by stimulated endothelial cells. Tumor necrosis factor and interleukin-1 activate the endothelium and cause cells to change shape, which causes increased vascular permeability. Damage to endothelial cells and cell leakage, observed by the use of transmission electron microscopy, after distention-decompression is indicative that hypoxic cell damage was a cause of the increased capillary permeability.

Longitudinal and circular muscle cells had mitochondrial cell damage and edema observable by transmission electron microscopy. Mitochondrial swelling within the cells is indicative of hypoxic damage and inadequate energy. Because mitochondria provide enzymes and a structural framework for generation of ATP, cellular injury causes suppression of oxidative phosphorylation and limits generation of ATP. Inadequate amounts of ATP cause the energy-dependent Na⁺,K⁺-ATPase membrane pump to fail, leading to influx of intracellular sodium, which is accompanied by an osmotic gain of water and results in cellular swelling and dilation of the endoplasmic reticulum.

Depletion of ATP is primarily responsible for acute cellular swelling and is 1 of the earliest manifestations in ischemic cell injury. Lack of tissue oxygenation and subsequent cellular energy deficits were probably responsible for the ultrastructural damage seen in the muscle cells in the study reported here.

References


