Effects of Gastric Distension on the Cardiovascular System in Rainbow Trout (*Oncorhynchus mykiss*)

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Summary

When animals feed, blood flow to the gastrointestinal tract increases to ensure an adequate oxygen supply to the gastrointestinal tissue and an effective absorption of nutrients. In mammals, this increase depends on the chemical properties of the food, as well as to some extent on the mechanical distension of the stomach wall. By using an inflatable nitrile balloon positioned in the stomach, we investigated the cardiovascular responses to mechanical stretch of the stomach wall in rainbow trout (*Oncorhynchus mykiss*). Distension with a volume equivalent to a meal of 2% of the body mass increased dorsal aortic blood pressure by up to 29% and central venous blood pressure increased transiently nearly five-fold. The increase in arterial pressure was mediated by an increased vascular resistance of both the systemic and the intestinal circulation. Cardiac output, heart rate and stroke volume did not change and only transient changes in gut blood flow were observed. The increase in arterial pressure was abolished by the α-adrenergic antagonist prazosin, indicating an active adrenergic vasoconstriction, whereas the venous pressor response could be the consequence of a passive increase in intraperitoneal pressure. Our results show that mechanical distension of the stomach causes an instantaneous increase in general vascular resistance, which may facilitate a redistribution of blood to the gastrointestinal tract when chemical stimuli from a meal induce vasodilation in the gut circulation. The normal postprandial increase in gut blood flow in teleosts is therefore, most likely, partly dependent on mechanical stimuli, as well as on chemical stimuli.
**Keywords:** Blood flow, coeliacomesenteric artery, postprandial, mechanical stimuli, teleost.
Introduction

In order for food to be effectively processed, several physiological responses must be coordinated. Proper adjustments of the cardiovascular system after food intake are of fundamental importance for the absorption and transport of nutrients, as well as for the supply of oxygen to the metabolically active gastrointestinal tract. In most animals so far studied the cardiovascular system does not have the capacity to fully perfuse all tissues with blood at any one time, and control mechanisms optimize blood flow through any given vascular bed, ensuring that proportionally more blood is distributed to active tissues with high oxygen demand (15).

In unfed fish, 16 to 40% of cardiac output (Q) is directed to the gastrointestinal tract. After feeding routine blood flow to the gastrointestinal tract can increase by as much as 78% in Chinook salmon (*Oncorhynchus tshawytscha*) (43), 42% in sea bass (*Dicentrarchus labrax*) (2), 100% in sea raven (*Hemitripterus americanus*) (3), 112% in the celiac artery ($q_{coa}$) and 94% in the mesenteric artery ($q_{mea}$) in red Irish lord (*Hemilepidotus hemilepidotus*) (6) and 72% ($q_{coa}$) and 42% ($q_{mea}$) in Atlantic cod (*Gadus morhua*) (5). In most cases the postprandial increase in gastrointestinal blood flow in fish is compensated for by an increase in Q, which seems to be more important than a redistribution of blood away from the systemic circulation (3). This is in contrast to the redistribution of blood, without major changes in Q, that has been reported in some mammals after feeding (49), but in concert with the increase in Q seen in other mammalian studies (17). The postprandial increase in Q in fish is regulated by the adrenergic control systems (3, 5, 6).
In rainbow trout (*Oncorhynchus mykiss*), as in most other teleost species, the gastrointestinal tract is supplied with oxygenated blood via the coeliacomesenteric artery (cma) which divides into two main arteries, the gastrointestinal artery which supplies the stomach, the spleen, the dorsal portion of the intestine and parts of the pyloric ceca; and the much larger intestinal artery which supplies the liver, the spleen, the pyloric ceca, and the ventral portion of the intestine (45) and possibly also portions of the stomach. In some older literature these vessels are referred to as the coeliac artery (*a. coeliaca*) and the mesenteric artery (*a. mesenterica*), respectively. Recent results using the corrosion cast technique have, however, revealed some major differences in the vascularization of the gastrointestinal system when comparing rainbow trout (unpublished result) and chinook (45). The coeliacomesenteric artery is, for example, much longer in rainbow trout and as a consequence the bifurcation occurs further downstream compared to chinook.

Feeding introduces two types of stimuli; the first immediate stimulus is the mechanical distension of the stomach, followed by a distension of various parts of the intestine, the second stimulus is the chemical influence of the digested food. In mammals, mechanical distension of the stomach or intestine increases myocardial contractility, arterial blood pressure and heart rate (7, 20, 25, 27, 28, 34, 37, 46, 47). The increase in arterial blood pressure is thought to primarily be mediated via an increase in the systemic vascular resistance, since the pressor response occurs without any obvious changes in heart rate or contractility (19). Afferent impulses in the reflex mediating the increase in vascular resistance are initiated by stretch-activated mechanoreceptors in the stomach wall and travel via the vagus nerve to the central nervous system (9). The efferent pathway is a sympathetic output via the splanchnic nerve (27, 46, 47), possibly in combination with
activation of the renin-angiotensin system (30). The mammalian literature suggests that the reflex-mediated increase in systemic vascular resistance, in combination with hydrolyzed products from the digested food, which induce local gastrointestinal vasodilation, are the primary mechanisms by which blood from the systemic circulation is directed to the gastrointestinal circulation following a meal (1, 10, 11, 13, 16, 22, 24, 29, 42). Ultimately, there seems to be a complex and precise regulation of the postprandial blood flow pattern in vertebrates, involving both mechanical as well as chemical stimuli. In fish, however, the precise regulation of the postprandial increase in blood flow to the visceral organs is still poorly understood. The relative contributions of the passive mechanical distension and the chemical composition of the ingested food to this regulation are largely unknown. Furthermore, no attempt has been made to record venous hemodynamic responses to feeding or gastric distension in fish. This is somewhat surprising, given the well-documented importance of the venous vasculature for cardiovascular homeostasis in fish (39).

The aim of this study was to establish the cardiovascular responses elicited by passive distension of the stomach wall in rainbow trout. In addition, experiments were conducted in order to elucidate the role of an α-adrenergic mechanism in the control of both the systemic and venous system during the mechanical distension

Material and methods

Experimental Animals
Rainbow trout (*Oncorhynchus mykiss*), ranging in size between 420-665 g (mean: 571±20 g, N=20), were purchased from a local hatchery (Antens laxodling AB). The fish were held in 2 m³ fibre-glass tanks supplied with aerated freshwater (10ºC) from the departmental re-circulating water system and fed dry trout pellets at regular intervals. The photoperiod was adjusted to 12:12 light:dark conditions. Upon arrival, fish were left for at least 1 week before any experimental procedures were performed. Ethical permit 64/2004 from the animal ethics committee of Göteborg, covered all experiments reported here.

**Surgical procedures**

Fish were fasted for approximately one week prior to surgery. Individual fish were anaesthetized in water containing MS-222 (150 mg l⁻¹) buffered with sodium bicarbonate (300 mg l⁻¹) and transferred to an operating table covered with soft water-soaked rubber foam. The gills were continuously irrigated with aerated freshwater (10º C) containing MS-222 (75 mg l⁻¹) buffered with sodium bicarbonate (150 mg l⁻¹). To measure dorsal aortic blood pressure (P<sub>da</sub>), the dorsal aorta was cannulated with a tapered PE50 (polyethylene) catheter through the roof of the buccal cavity using a guide wire according to the technique described by Axelsson and Fritsche (4). The catheter was tunnelled through the snout. Central venous blood pressure (P<sub>ven</sub>) was measured by inserting a PE50 catheter approximately 10 mm into the duct of Cuvier towards the sinus venosus as described by Sandblom et al. (40). Both catheters were secured to the skin with sutures and filled with heparinized (100 IU ml⁻¹) saline (0.9%). To measure blood flow in the coeliacomesenteric artery (q<sub>ema</sub>), a Doppler flow probe with a diameter of 1.1-
1.2 mm was placed around the coeliacomesenteric artery, roughly 10 mm distal to the bifurcation of the coeliacomesenteric artery (45). To access the coeliacomesenteric artery, the fish was placed on its left dorso-lateral side and a 25 mm incision was made ventrodorsally from the base of the pectoral fin. The vessel was dissected free using blunt dissection, taking care not to damage any of the surrounding nerves. Once the cuff was positioned, the lead was secured with sutures to the skin and the incision was closed with uninterrupted sutures. Cardiac output (Q) was measured with a 1.8 mm Doppler flow probe placed around the ventral aorta. To place the cuff, the fish was positioned on its left side and a small incision was made at the base of the fourth gill arch dorsal to the ventral aorta. Connective tissue was removed until the ventral aorta was visible and there was enough room for the cuff. The cuff lead was secured with one suture close to the cuff and one to the back of the fish.

In one group of fish (n=11) the vascular capacitance responses to gastric distension was evaluated by measuring the mean circulatory filling pressure (MCFP). In these fish, the ventral aortic flow probe was modified to include an inflatable latex balloon which allowed for transient occlusions of blood flow as described previously (38). The MCFP was estimated from the venous plateau pressure between 5-7 s of occlusion. MCFP is dependent on total vascular compliance and smooth muscle tone, and on total contained blood volume (39). However, given the much higher compliance (at least twenty times compared to the arterial system, (12) and volume of the venous vasculature, it primarily reflects the capacitive state of the venous vasculature (39). Both flow probes were custom-made from Perspex and equipped with 20 MHz Doppler
crystals (0.5 mm; Iowa Doppler products, Iowa City, IA, USA) glued to a hole in the cuff.

Mechanical distension of the stomach, to mimic the mechanical pressure exerted by ingested food, was achieved by placing a balloon of fixed maximum size (approx. 15ml) into the most caudal part of the stomach (Fig. 1). The balloon was constructed from a non-distensible nitrile plastic material taken from disposable nitrile gloves (VWR International, Leuven, Holland). This material was chosen for its resistance to strong acids and proteolytic enzymes which characterize the intragastric environment. To place the balloon, a ~15 mm incision was made dorso-ventrally from the sagittal midline, approximately two thirds of the distance from the pectoral fin to the pelvic fin. A piece of plastic tubing, 6 mm in diameter, was introduced into the stomach via the mouth and a small hole was made in the posterior part of the stomach allowing a thin wire guide to be inserted through the hole and into the tubing. The end of a PE90 catheter, connected to the nitrile balloon, was then attached to the guide and retracted out through the hole in the stomach. The balloon was pulled into the stomach and the catheter was secured by sliding a small bell shaped rubber cap with a drilled hole over the catheter and against the stomach wall. Post-mortem analysis revealed that this prevented movement of the balloon and gastric juices from leaking out into the peritoneal cavity. The incision was closed with uninterrupted sutures and the catheter was secured to the skin by two additional sutures. Following surgery the fish were held in opaque plastic chambers supplied with aerated freshwater (10°C) from the departmental re-circulating water system, Fish were left to recover for 48 h before the experimental protocol was started.
Experimental protocols

Effects of distension volume and distension duration

Two experimental protocols were used to investigate the cardiovascular effects of distension volume and distension duration in untreated fish.

Protocol I: The distending volume was kept constant at 10 ml, which is approx 2% of the body mass in a 500 g fish. This volume is considered to be a fair sized meal and is equivalent to the volume of food used in previous feeding experiments in rainbow trout (18, 43). Four distension periods were tested (in chronological order): 5, 15, 40 and 60 min.

Protocol II: The duration of the distension was kept constant at 30 min and four different distending volumes were tested (in chronological order): 2, 5, 10 and 15 ml, followed by 90-120 min without distension.

Each distension was preceded by 10 min of baseline recordings and the fish was left undisturbed for at least 1 h between distensions. The time used to inflate as well as deflate the nitrile balloon was kept constant, at approximately 1 and 2 min, respectively, during both experimental protocols. A randomised protocol was not used in these studies, since preliminary results revealed that smaller distension volumes that followed after a large distension exhibited very limited or no response, while by using increasingly larger distensions the effect of the smaller distension volumes (2 and 5 ml) could be effectively studied. Small distensions only seemed to attenuate the effects of the larger distensions.
marginally, but if anything, the reported responses to the large distensions may represent slight underestimates.

**Role of α-adrenergic mechanisms and venous capacitance responses**

To elucidate if α-adrenergic mechanisms underlie the cardiovascular responses observed in the first two protocols, a set of experiments were performed in another group of fish (n=11). In this group, the same cardiovascular variables were recorded with the exception of gut blood flow. Instead, MCFP was measured to elucidate the role of the venous vasculature in the observed cardiovascular responses.

After an initial baseline recording for 10 min the MCFP was measured. Cardiovascular variables were then allowed 30 min to return to baseline before the stomach was distended with a volume corresponding to ~2% of the body mass of the fish. After 5 min of gastric distension, MCFP was measured again. The distension was released after 10 min and the α-adrenoceptor antagonist prazosin (1.0 mg kg body mass⁻¹; Sigma, St Louis, MO, USA) was administered as a bolus injection via the central venous catheter. The fish was left for 3 h for the drug to exert its effect and since preliminary experiments had revealed that this recovery time was enough to remove any attenuating effect of repeated gastric distensions. Other reported cardiovascular variables were measured immediately before each MCFP measurement.

**Data acquisition**

Arterial, venous and gastric pressures (Pgas) were measured using pressure transducers (model DPT-6100, Smiths medizintechnik, Kirchseeon, Germany) connected
to a 4 channel amplifier (Somedic, Hörby, Sweden). Calibration was done against static columns of water. The Doppler flow probes were connected to a directional pulsed Doppler flow meter (model 545C-4, The University of Iowa, Iowa, USA). A PowerLab system connected to a PC running Chart5 (ADInstruments Pty Ltd, Castle Hill, Australia) was used for data acquisition.

Data analysis and statistics

Vascular resistances were calculated from raw data values as \( R_{\text{sys}} = \frac{(P_{\text{da}}-P_{\text{ven}})}{Q} \) for systemic resistance and \( R_{i} = \frac{(P_{\text{da}}-P_{\text{ven}})}{q_{\text{IA}}} \) for splanchnic resistance. Thus, \( R_{\text{sys}} = R_{i} + R_{\text{res}} \), where \( R_{\text{res}} \) is the vascular resistance in the system except for the splanchnic resistance. Two assumptions were made in these calculations; 1, that the driving force through the coeliacomesenteric artery is the dorsal aortic blood pressure and 2; that blood viscosity does not change during the experimental protocol. Heart rate (\( f_{\text{HR}} \)) was obtained from the pulsating pressure traces and cardiac stroke volume (SV) was calculated as \( SV = \frac{Q}{f_{\text{HR}}} \). All reported values are means ± S.E.M. Blood flows and resistances are presented as relative changes with the baseline set to 100%. The baseline value before distension was averaged and compared to values during and after the distension by repeated measures ANOVA followed by a Bonferroni post-hoc test to analyse individual points. In the group that received prazosin treatment, the effect of gastric distension and \( \alpha \)-adrenergic blockade was evaluated using a parametric, paired t-test. In the comparison between treatments, normalized \( R_{\text{sys}} \)-values were used. Significant difference from the baseline was assumed when \( p<0.05 \).
Results

Most animals remained calm throughout the entire protocol, but when the distension was released, some became agitated as indicated by brief (1-5 sec) struggles. However, this behavior was too short to be visible in the mean cardiovascular traces presented below.

Arterial pressor response

When the stomach was distended, an initial and very transient (~1min) bradycardia was typically observed (also this effect was too short to be visible in the mean cardiovascular traces presented below). The most notable cardiovascular response to the mechanical distension was a pronounced increase in dorsal aortic blood pressure (Figs. 2a-c and Table 1). For example, with the 10 ml distension for 5 min, $P_{da}$ increased significantly to a peak value 29% higher compared to the routine $P_{da}$ of 3.4±0.1 kPa (Table 1). It was evident that the duration of the pressor response increased with the length of the distension (Fig. 2b and Fig 2c, respectively), and for the shorter distensions, $P_{da}$ remained significantly elevated for as long as the stomach was distended. Since $Q$ did not change, the increase in $P_{da}$ was entirely due to an increased $R_{sys}$. The increase in vascular resistance was mediated by an increased $\alpha$-adrenergic tone as the arterial pressor response was abolished by prazosin pre-treatment (Fig. 3).

The $P_{da}$ response was also dependent on the volume of the distension, which is evident when comparing Fig. 2a with Fig. 2b. Distending the stomach with 2 ml for 30
min (not shown) gave no significant response, while 5, 10 and 15 ml distensions induced increasingly larger pressor responses (+8.0, +20.2 and +21.1% from routine Pda, respectively). Although the Pda responses to longer distensions were qualitatively similar (Figs. 2b and c), repeated and/or prolonged distensions seemed to attenuate the magnitude of the response (Fig. 4). For example, this was evident by the lack of a statistically significant response in the 60 min distension which had been preceded by the 5, 15 and 40 min distensions (Table 1). Figure 4 illustrates how the peak Pda response increases with the logarithm of the distension volume and declines with the power of the number of distensions. The non-linear correlation between Pda-response and distension volume is explained by the fact that the response declines with the number of distensions.

**Venous pressor response**

Also venous blood pressures increased significantly with stomach distension, although the venous responses were somewhat more variable than the arterial responses (data not shown). Similar to the arterial responses, the peak Pven response increased with distension volume (Table 1). With the 2, 5, 10 and 15 ml distensions, the peak increase from routine Pven (0.03-0.04 kPa) was +64, +76, +180 and +419%, respectively (Fig. 2a-c and Table 1). MCFP also increased significantly from a routine 0.04±0.04 kPa to 0.07±0.04 kPa (Fig. 3). Administration of prazosin increased routine MCFP to 0.13±0.04 kPa, but did not abolish the increase in Pven or MCFP during gastric distension, indicating that the venous pressor responses were not mediated via α-adrenoreceptors and may be passive effects of the distension per se (Fig. 3). This was further corroborated by the very rapid increase in venous pressure during distension and the immediate return to baseline
following release of the distension as opposed to the arterial response that showed a slower return to baseline (compare Fig. 2b and 2c).

**Coeliacomesenteric artery blood flow**

The change in coeliacomesenteric artery blood flow was limited to a transient increase in blood flow when the balloon was deflated (Fig. 2c and Table 1). The lack of an increase in q_{ia} during the distension, despite the concomitant increase in P_{da}, can partially be explained by the associated increase in vascular resistance of the intestinal circulation (Fig. 2b and c). The coeliacomesenteric artery blood flow rapidly returned to the initial baseline after it had reached its peak value following the release of the stomach distension. However, with the transient increase in q_{ia}, there is a tendency for the response to attenuate as the duration of the distension is prolonged. For example, the peak increase after a distension of 10 ml for 15 min was 22% above the initial baseline, and only 17% for the 10 ml distension for 40 min. The response in q_{i} to the other distension times and volumes were qualitatively similar (Table 1).

**Discussion**

Previous studies on postprandial hemodynamic responses in fish have predominantly involved force feeding with commercial fish pellets or fish meat, and have addressed the effects of time or different cardiovascular challenges such as exercise and hypoxia (2, 3, 5, 6, 15, 43, 44). As far as we are aware, no studies have previously investigated the cardiovascular responses to passive mechanical gastric distension in fish. In the present study we show that when the stomach is mechanically distended in rainbow...
trout using a balloon, there is a pronounced increase in $P_{da}$, without any significant changes in $Q$ or $q_{ia}$. This pressor response is mediated via increased systemic and gastrointestinal vascular resistances due to an increased $\alpha$-adrenergic tone. The result is in agreement with previous similar studies on mammals (29), and feeding studies on Atlantic cod (5). The total systemic resistance is the sum of the gastrointestinal resistance and the resistance in the rest of the vascular system. Since we were not able to obtain absolute values for $Q$ and $q_{ia}$ we cannot determine the exact contribution of $R_i$ to the total $R_{sys}$. However, given that roughly 1/3 of $Q$ is directed to the gastrointestinal tract in other teleost species (2, 3, 5, 6, 44), it is reasonable to assume that the gastrointestinal vascular circulation of rainbow trout also receives 1/3 of $Q$ and therefore accounts for approximately 1/3 of the total systemic resistance. Thus, since $Q$ and $q_{ia}$ is unaffected by the distension, the increase in total $R_{sys}$ is dependent on both an increase in $R_i$ as well as an increase in $R_{res}$.

In mammals, studies on the effects of gastric mechanical stimuli on cardiovascular function are limited to a few papers where the stomach has been distended using a fluid filled balloon (26-28, 46, 47) and where undigested food constituents, which cause no chemical stimulation as opposed to hydrolysed food constituents, have been used to distend various parts of the gut (10). An increased arterial blood pressure after feeding and gastric distension was reported in several studies in cats as well as rats (26, 27, 31, 35). However, most of these studies have been performed on anaesthetised animals (23, 34, 48).

In the present study, blood flow to the gastrointestinal tract was largely unaffected by the mechanical distension, except for a transient increase when the distending pressure
was released. However, in a normally feeding fish, the reflex mediated increase in $R_{sys}$ and $P_{da}$ caused by the passive distension may be an important initial mechanism which serves to maintain $q_{ia}$, even though $R_i$ increases when the stomach is distended.

Subsequently, chemical stimuli from the food will induce a vasodilation in the gut, possibly enhanced by vascular escape mechanisms (36), i.e. decreases in the vascular resistance despite a persistent $\alpha$-adrenergic tone. This ultimately increases gastrointestinal blood flow. Such biphasic responses have been observed in dogs (17). Thus, we hypothesize that the initial increase in systemic resistance and arterial blood pressure following stomach distension might be important to facilitate the subsequent increase in gut blood flow. In mammals, chemical stimuli such as bile, glucose and fat from digested food induce vasodilator responses and are the main determinants of the postprandial hyperemia (1, 8, 11, 13, 22, 25, 41, 49). Data for gut blood flow responses to stomach distension in mammals are mainly based on studies using micro-spheres. Some of these studies indicate that a small increase in blood flow occurs following distension (26), while others show little effect on gut blood flow (10).

Stomach distension also increased $P_{ven}$ and MCFP. However, in contrast to the arterial pressor response, the increase in venous pressures was not abolished by the administration of prazosin, indicating that the response was not mediated via $\alpha$-adrenoreceptors and could possibly represent passive effects of an increased intraperitoneal pressure which compress the venous vessels. In addition, the initial increase in $P_{ven}$ was most likely enhanced by the transient bradycardia that occasionally occurred at the beginning of the distension. The increase in $P_{ven}$ may be of physiological importance in normally feeding animals since an increased cardiac preload will increase
Q via the Frank-Starling mechanism (14, 39). In this study, there was no increase in Q although P\textsubscript{ven} increased during the relatively short period of stomach distension. Even though a simultaneous increase in P\textsubscript{ven} could counteract the increased P\textsubscript{da} and thus maintain a constant Q, it is unlikely that the increased preload or afterload would significantly affect Q considering the magnitude of these responses. A β-adrenergically mediated change in the myocardial contractility, as seen in mammals after distension (27), could also influence this relationship. Previous studies on the red Irish lord (\textit{Hemilepidotus hemilepidotus}) have reported an increase in Q after feeding which is large enough to support the increase in gut blood flow (6). Our results do not exclude that Q would have increased with a longer distension time, but we find it more likely that the postprandial increase in Q that is observed in many teleosts after feeding is due to a reduced vascular resistance in the gastrointestinal circuit mediated by chemical stimuli from the ingested food.

There appears to be a correlation between the magnitude and duration of the mechanical stimuli and the magnitude and duration of the pressor response in rainbow trout (Fig. 2a,b,c and Fig. 4). This is in contrast to results from mammals (34, 35) and the physiological importance of this is uncertain. There is also an attenuating effect of multiple and/or prolonged distensions on the peak response of P\textsubscript{da}. This is a response which is also commonly observed with repeated stimulation of other physiological systems involving sensory receptors, such as touch receptors in the skin (9), but other factors could also be involved such as muscle/nerve fatigue or enzyme exhaustion.

Based on the assumption that 2% of the body weight is a normal size meal for rainbow trout (18, 43), the range of distension volumes used in the present study were
selected to mimic both sub- and supraphysiological volumes. Larger volumes would possibly have produced more pronounced responses, but stimulation of gastric nociceptors with larger volumes was a substantial concern (21). It could be argued that the observed pressor response was a general stress response caused by the potentially noxious stimuli as the balloon was distended. However, we find this possibility unlikely because the distension volumes were in the physiological range and the fish did not appear distressed by the distension as they typically remained calm in their holding tanks during the distension. In addition, the pressor response also declined rapidly as the distension was released. Depending on the method used to distend the stomach, gastric distension can generate different responses in mammals (28, 34, 35). For example, when a balloon is used to distend the stomach, rapidly adapting mechanoreceptors in the stomach mucosa are stimulated. On the contrary, these receptors are not stimulated by distension with fluid, where the stomach is tied off at both ends and filled using a catheter (32-34). However, we believe that a fluid filled balloon represents a less surgically invasive and more physiologically relevant alternative, since on the one hand ingested food is unlikely to fill the entire stomach as uniformly as injected fluid, and on the other hand tying of the stomach at both ends most likely causes damage to both the blood and nerve (efferent and afferent) supplies.

**Perspectives and significances**

To our knowledge this is the first study to investigate the cardiovascular responses associated with mechanical stomach distension in fish. This has previously been studied in mammals and our results suggest that the cardiovascular response to distension is
conserved throughout evolution since it comprises a sympathetic response with an increase in arterial blood pressure and systemic vascular resistance in both mammals and fish. In some mammalian studies there is also an increase in $Q$ which was not seen in this study. The reason for this discrepancy remains to be resolved. The significance of the increase in sympathetic tone after mechanical stomach distension is not entirely clear but it appears to be similar to the initial response to feeding in several mammalian studies. We believe that this cardiovascular response facilitates the shunting of blood to the gastrointestinal vascular system during a meal, but further studies are needed to make a conclusive statement about this. From an evolutionary perspective, it would be interesting to study other species with, for example, different feeding habits such as ambush predators that, in contrast to the rainbow trout, can devour large meals in a short period of time and therefore distend the stomach more rapidly and with large volumes. The methodology developed in this study will allow such comparisons to be made, and will allow further studies on the subject of vertebrate gastrointestinal physiology and feeding adaptations.

**Acknowledgements**

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References


Figure legends

Figure 1. Schematic illustration of rainbow trout (*Oncorhynchus mykiss*) showing placement of the nitrile balloon (A) into the most caudal portion of the stomach. The balloon is secured using a plastic cap (B), which also prevents leakage of stomach contents into the body cavity. The PE90 catheter (C) is tunnelled through the body wall via an incision (D). The pyloric ceca (E) are the thin sacs at the transition between the stomach and the proximal intestine. Hatched arrows indicate the oral-anal route taken by ingested food particles.

Figure 2. Mean (±S.E.M.) dorsal aortic blood pressure (P_{da}), venous pressure (P_{ven}), cardiac output (Q), coeliacomesenteric artery blood flow (q_{cma}), systemic vascular resistance (R_{sys}), coeliacomesenteric artery vascular resistance R_{cma} and heart rate (f_{H}) in rainbow trout (*Oncorhynchus mykiss*) during distension of a balloon in the stomach (n=7). Arrows indicate start and stop of distension. Three different distensions are shown: in (A) the stomach is distended for 30 min using 5 ml; in (B) the stomach is distended for 30 min but the distending volume is increased to 10 ml and in (C) the duration of the distension is increased to 40 min while the distending volume is kept at 10 ml. * (p<0.05), ** (p<0.01) and *** (p<0.001) denote significant difference from baseline.

Table 1. Mean (±S.E.M.) dorsal aortic blood pressure (P_{da}), venous pressure (P_{ven}), cardiac output (Q), coeliacomesenteric artery blood flow (q_{cma}), systemic vascular resistance (R_{sys}), coeliacomesenteric artery vascular resistance R_{cma} and heart rate (f_{H}) in
rainbow trout (*Oncorhynchus mykiss*) during distension of a balloon in the stomach (n=7). The baseline values are measured just before the distension and consecutive values are shown for 10, 20, 30 and 40 min after the initiation of the distension. Peak values illustrate the maximum response attained during the distension, with the peak occurring anywhere between 0 to 40 min after the initiation of the distension. * (p<0.05) denote significant difference from baseline.

Figure 3. Peak (±S.E.M.) change in dorsal aortic pressure (P_{da}), venous pressure (P_{ven}), systemic vascular resistance (R_{sys}) and mean circulatory filling pressure (MCFP) before and after prazosin treatment when distending the stomach for 30 min using 10 ml in 450-665 g (599±27 g) rainbow trout (*Oncorhynchus mykiss*) (n=11). * (p<0.05) denotes significant difference from baseline. ¶ indicates a significant difference between treatments. N.S (p>0.05) denotes no significant effect of prazosin treatment.

Figure 4. Non-linear regression analysis illustrating the relationship between the volumes used to distend the stomach in rainbow trout (*Oncorhynchus mykiss*) and the dorsal aortic (P_{da}) peak response (A) and the attenuation in peak response with increasing number of distensions and/or prolonged distensions (B). The peak response increases with the logarithm of the number of distension, with an R^2-value of 0.9494, and decreases with the power of the number of distensions with an R^2-value of 0.9382.
Table 1. Mean cardiovascular parameters, compared to baseline and peak response, when distending the stomach in rainbow trout (Onchorhyncus mykiss). Values are means ± SEM.

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<td>Q (%)</td>
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<td>fH (bpm)</td>
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<td>Pda (kPa)</td>
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<tr>
<td>Pven (kPa)</td>
<td></td>
<td>0.01±0.03</td>
<td>0.07±0.04</td>
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<td>0.00±0.02</td>
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<td>fH (bpm)</td>
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<td>Pven (kPa)</td>
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<td>0.04±0.01</td>
<td>0.07±0.02</td>
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Fig. 3
Fig. 4

A

\[ y = 0.925x^{-0.5263} \]
\[ R^2 = 0.9382 \]

B

\[ y = 0.337\ln(x) - 0.2174 \]
\[ R^2 = 0.9494 \]