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Siberian sturgeon (*Acipenser baeri*, Brandt JF 1869) gut: anatomic description

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Abstract

Efforts to refine rations for captive rearing are dependent on a clear understanding of gut anatomy and function. Gross anatomic and histological descriptions of the gut of the Siberian sturgeon *Acipenser baeri*, are provided in this manuscript to serve as a baseline characterization for comparison during basic research and health surveys of Siberian sturgeon populations. Histological evaluations were carried out on 200 individuals (body weight ranging from 50 to 8000 g), using gross anatomy and light microscopy. The most characteristic finding is about pancreas, which has three distinct lobes. The pancreatic duct ends in a papilla between the small intestine and the pyloric caecum. Eosinophilic zymogen granules characterize exocrine cells, and the endocrine cells are grouped together in structures that are similar to mammalian Islets of Langerhans with not homogeneous distribution. Morphometrical evaluation shows that the mean area of the islets is 8.32 $mm^2 \cdot 10^{-3}$. The Siberian sturgeon pancreas is a morphologically distinct organ functionally and histologically similar to that of mammals.

Keywords: Siberian Sturgeon, Gut, Pancreas, Anatomy, Histology

Introduction

Sturgeons (Acipenseriformes) have inhabited the planet for more than 200 million years. Thirty-one extant species remain in predominantly temperate areas around the globe but many species are endangered, or of special concern (WSCS 2007). Harvested both for their flesh and for their roe, commercial harvests have declined and efforts to sustain their availability as a food fish have focused on their propagation for both research, possible population augmentation and as food products. Sturgeons are also common residents of display aquaria where their unique physical appearance, lengthy natural history, and imperilled status play a role in the efforts of conservationists to promote environmental stewardship.

Conservation efforts to mitigate the imperilled status of wild sturgeon species and the captive propagation of sturgeon for the seafood market necessitate a sound understanding of sturgeon anatomy and physiology. Effective study of the pathophysiology of diseases that could contribute to the decline of wild sturgeon populations or captive stock are also dependent on our ability to differentiate between "normal" gross anatomy and histology and organ pathology. Efforts to refine rations for captive rearing are similarly dependent on a clear understanding of sturgeon pancreatic anatomy and function.

Optimizing captive cultivation of sturgeon requires a thorough understanding of their biology, and the basic attributes of sturgeon digestion and nutrition. In fish, as well as in mammals, the pancreas has both digestive and endocrine functions. Acini containing pancreatic exocrine cells secrete pancreatic enzymes that aid in digestion,

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and islets of Langerhans have endocrine functions. Early studies on the pancreatic structure of fish described a diffuse organ (Massari 1898) while more recent studies on white sturgeon gut anatomy (Buddington and Doroshov 1986) stated that "acinar cells are grouped into rosettes". Pancreatic clusters in fish are grouped around the duodenum and pyloric caeca, and in many species are interwoven with hepatic tissue to form a hepatopancreas (Guillame et al. 1999). In contrast, the pancreas of *Chondrichthyes* is an autonomous and compact organ, similar to that of higher vertebrates (Babkin 1929). In *Osteichthyes*, only a few species such as eels, catfish, lungfish, and coelacanths have a compact pancreas, while most species possess a diffuse gland located along the hepatic portal vein (Harder 1975). Comparatively little is known about the pancreas of sturgeon. Nicolas (1904) first described the embryonic development of the sterlet *Acipenser ruthenus* and demonstrated that the pancreas originates from three processes, two ventrally and one dorsal, distributed in the visceral cavity. Gawlicka et al. (1996) describe the presence of pancreatic tissue in the adult white sturgeon.
Studies of the sturgeon pancreas have generally focused on hormone and digestive enzyme characterisation with biochemical and proteomic techniques (Rusakov et al. 1998; Kim et al. 2000). The presence of pancreatic exocrine acini with zymogen granules was reported in a study conducted on the digestive system development of the green sturgeon larvae, *Acipenser medirostris* (Gisbert and Doroshov 2003). Although these studies represent a viable initial effort to understand the basic gastrointestinal physiology of sturgeon digestion baseline anatomic information is needed for fully understand the health benefits of various diets offered to sturgeon grown in aquaculture production ponds and display aquaria. These descriptive studies were conducted to enhance our understanding of basic sturgeon anatomy. This manuscript describes anatomically and histologically the gut and the pancreas of Siberian sturgeon *Acipenser baeri*, and provides a practical guide for the dissection of their abdominal organs. This visual gross morphologic and histology representation of the sturgeon gut organs can provides a comparison baseline for researchers conducting nutrition trials and for veterinarians working to ensure the health of captive display fish.

Materials and methods

Siberian sturgeons (*Acipenser baeri*, Brandt JF 1869) raised in a private fish farm (Azienda Pisani Dossi, Cisliano - Milano, Italy) with a body weight ranging from 50g to 8000g were used in this study. The observations started in 2003 and were carried out for three years on 200 sturgeons. During the migration, some sturgeon species stop eating (Dadswell 1979; Mason and Clugston 1993). Marked physiologic changes have been documented during period of food deprivation (Gillis and Ballantyne 1996; Hung et al. 1997). Accordingly, a group of 25 sturgeons was fasted for two weeks, in order to evaluate how ceased food intake may affect pancreatic morphology.

Fish were killed by anaesthetic overdose (benzocaine 100 mg/L) than the gastro-intestinal tract was carefully dissected to preserve the integrity of the organs. Incisions were made in the visceral cavity wall to allow quick formalin penetration through the intestinal organs. Tissue samples of the liver and intestine were fixed in buffered isotonic 4% formalin (pH 7.2) cooled at 4 °C; liver, pyloric caeca and intestine samples were taken from 50 to 300 g of fish body weight for histological examination. The samples were embedded in paraffin, sliced and stained with haematoxylin-eosin, Mallory AZAN and Crossmon tri-chromic stains, following the common histological procedures and techniques.

Sample slices were examined through stereo and light microscopy, and then digital images were taken to document morphology (Leica DFC 320 digital camera; Adobe Photoshop ver. 7.0 imaging software). Morphometrical evaluations were carried out on the digital pictures with Image-Pro Plus ver. 5.0 (Media Cybernetics Inc., Bethesda, USA).

Results

The natural aspect of the fresh sturgeon and visceral organs, after removal of the abdominal wall are showed in Fig. 1. It is easier to distinguish the different organs in fresh specimens, but their topographic arrangement is easily modified during sampling procedures (Fig. 2). In the visceral cavity, the Siberian sturgeon oesophagus is cranially located on the sagittal plane, just ventral to both the vertebral column and the head of the kidneys. Proceeding in a caudal direction, the oesophagus lies in the right side and at mid-height reaches the swim bladder which is joined, whereby the pneumatic duct junction and after that starts the stomach. In ventral position to the oesophagus, moving from cranial to caudal direction, are located the heart and liver followed by the stomach. The swim bladder is located dorsally to the oesophagus.

At the end of the swim bladder, near the origin of the spiral valve, the stomach forms a caudal curve, for which we coined the term "large gastric curve", and increases in diameter. Then, it passes to the left side of the visceral cavity and proceeds in a cranial direction, touches the liver, and forms a second curve that we indicate

as “small gastric curve”. Here, the stomach crosses the sagittal plane on the ventral wall of the visceral cavity, folds in a caudal direction, approaches the sagittal plane from the left and forms the pyloric sphincter (Figs. 3, 4). Schematically, the oesophagus and the stomach together form a horizontal spiral structure, laterally compressed.

The Siberian sturgeon small intestine begins next to the pylori and its right wall immediately is widely anastomosed to the pyloric caecum, which is located on the left visceral side between the small gastric curve and the small intestine. In caudal direction, the small intestine forms a short tract with two elbow curves, for which we coined the name of “S curve” (Fig. 4), this intestinal structure is in tight relation to the spleen, all of them are visually evident on the left side. The last tract of the gut contains the spiral valve followed by the rectum in the visceral caudal region. Lateral observation on the left side (Fig. 3A) shows, ventrally to back muscular columns, the gonad, the kidney, and the pyloric caecum, which is located under the stomach. The pancreatic tissues have an ivory colour, the surface is rough and shiny and they are pulpy and friable in consistency. The pancreas with its pancreatic veins, which collects the blood from the spiral valve and from the small intestine, can be clearly observed on the right side, the gall bladder can be observed on the bottom curvature of the liver (Figs. 2, 3B).

A conspicuous part of the pancreas is found between the small gastric curve, the pyloric caecum and the small intestine (Figs. 5B; 6B,D), we consider it “pancreatic body”. Moreover, most of the pancreas is attached to the dorsal face of the pyloric caecum and has several small ramifications that extend along the stomach (Fig. 7) and the liver serosa (Fig. 8A,C). The pancreas strikingly presents as three large lobes.

The biggest pancreatic lobe starts from the pyloric caecum, run along the right side of the visceral cavity endings on the upper side of the spiral valve between the first and second coil. We call this the “pancreas long lobe” that divides into several small branches, and take rapports with: the spleen body (Fig. 9), the spiral valve on the right side and the dorsal ligament of spiral valve. The pancreas and the spleen (Fig. 9A, D), near the spiral valve, appear as a single organ without any clear separation of their connective tissue (Fig. 5C).

The second pancreatic lobe, “left lobe”, is on the left and follows the small intestine inside the ligament that joins it to the spleen (Fig. 5C). This lobe terminates close to the S curve of the small intestine upper side (Fig. 9A). The third pancreatic lobe, “right lobe” is smaller and runs along the right wall of the small intestine (Fig. 5C).

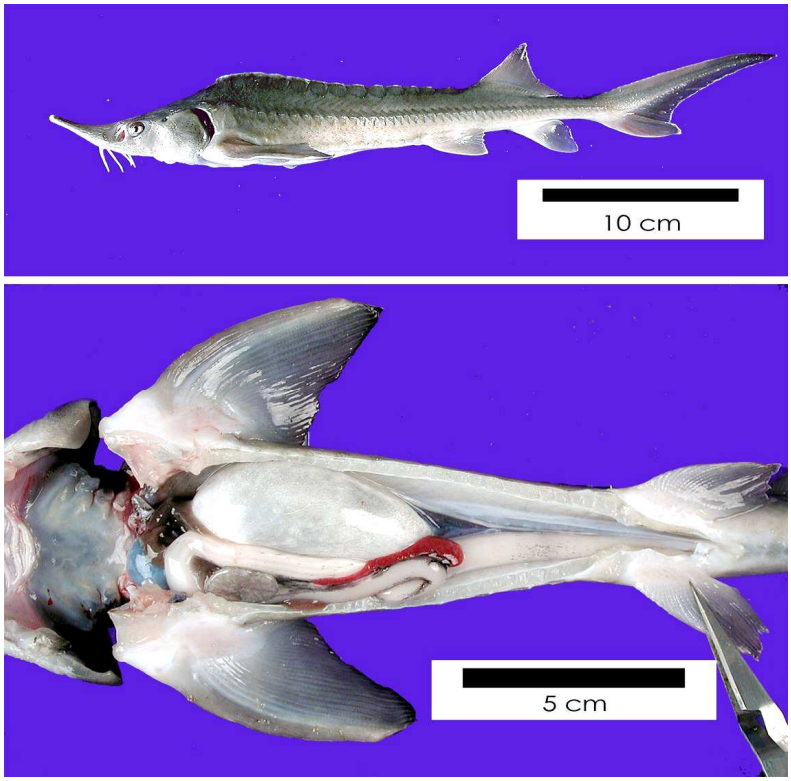

Fig. 1. Fresh specimens view. Upper photograph show the lateral view of the whole sturgeon; lower photograph show ventral view after ablation of the abdominal and mouth ventral wall. It is possible to see the vivid colour of the spleen and pale colour of the intestine (fasted sturgeon).

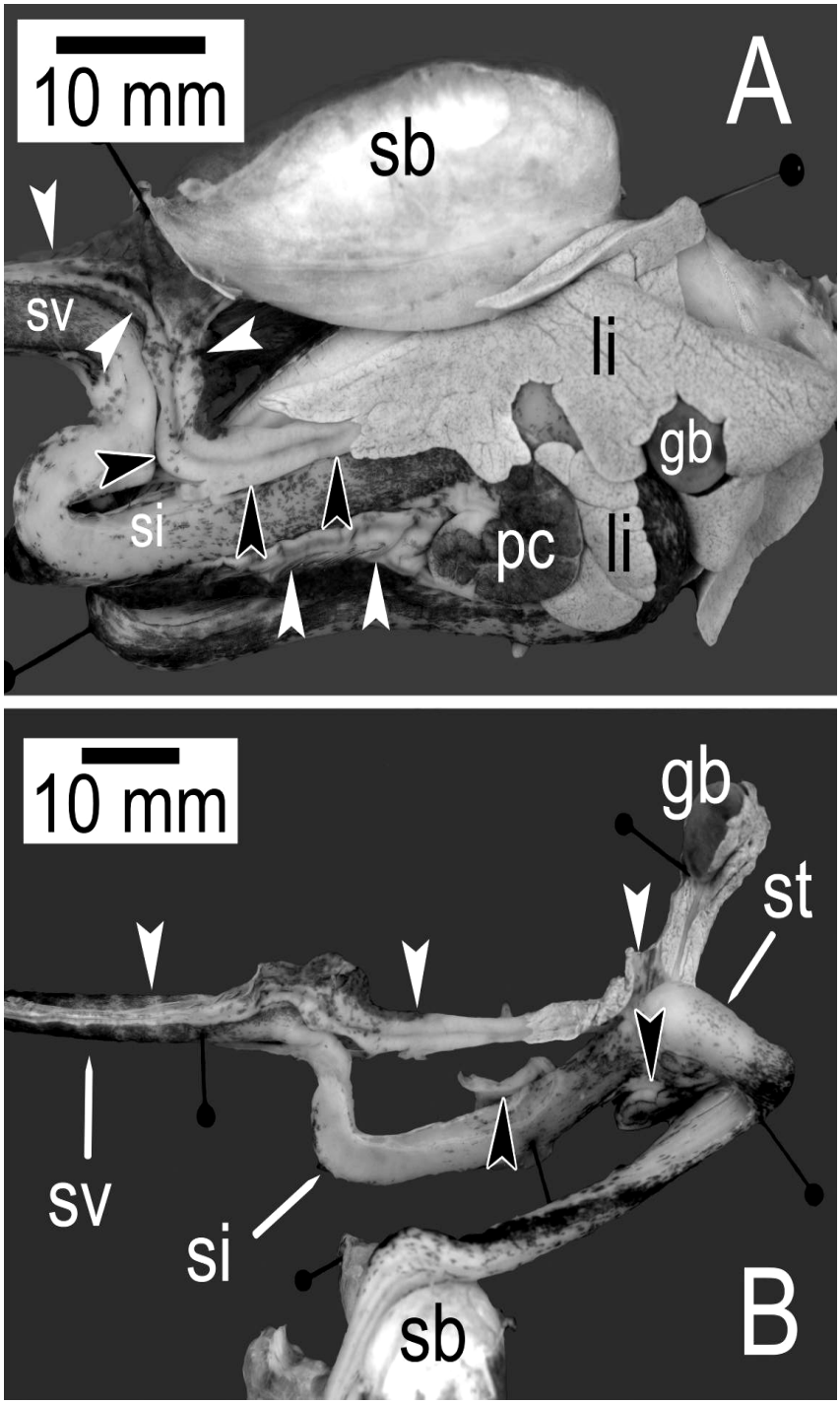

Fig. 2. Photograph A: intestinal package isolated from the fish (fresh tissue): visible pancreatic tissue (black and white arrowheads), liver (li), gall bladder (gb), swim bladder (sb), pyloric caecum (pc), small intestine (si), and spiral valve (sv). Photograph B: the gut package unfolded and the liver removed. Stomach (st), small intestine (si), spiral valve (sv), and swim bladder (sb), the pancreatic tissue (black and white arrowheads) is visible, the gall bladder (gb) is partially enveloped by a column of liver around the bile duct.

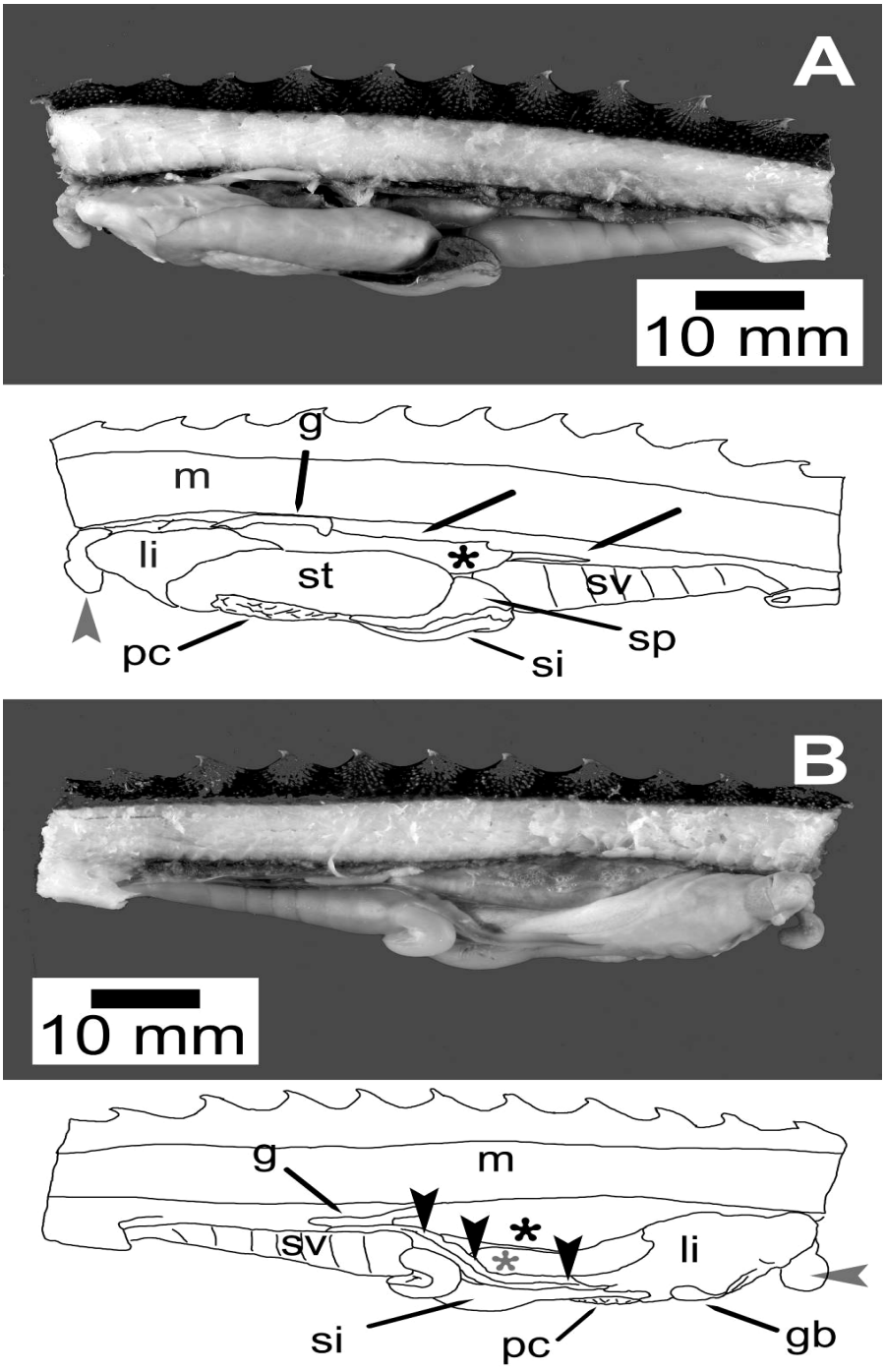

Fig. 3. Sturgeon (formalin fixed) after removal of the head, tail and abdominal wall. The organs were kept in place by the fixation process before abdominal wall ablation. The bone plaques with their typical saw blade profile are visible on the sturgeon's back. Photograph A: left side of the fish. The sketch below the photograph shows: dorsal muscles (m), left gonad (g), the left kidney (the two black rods), heart (grey arrowhead), liver (li), stomach (st), swim bladder (black asterisk *), pyloric caecum (pc) below the stomach, small intestine, and the S curve (si), body of the spleen (sp) in black, and the spiral valve (sv) followed by the rectum on the right. Photograph B: right side of the internal organs. The sketch below shows, from right to the left, heart (grey arrowhead), liver (li), gall bladder (gb), oesophagus (grey *), swim bladder (black *), pyloric caecum (pc), small intestine (si) with S curve, right gonad (g), spiral valve (sv), the long branch of the pancreas (black arrowheads), and the vein inside (visible as a small dark line).

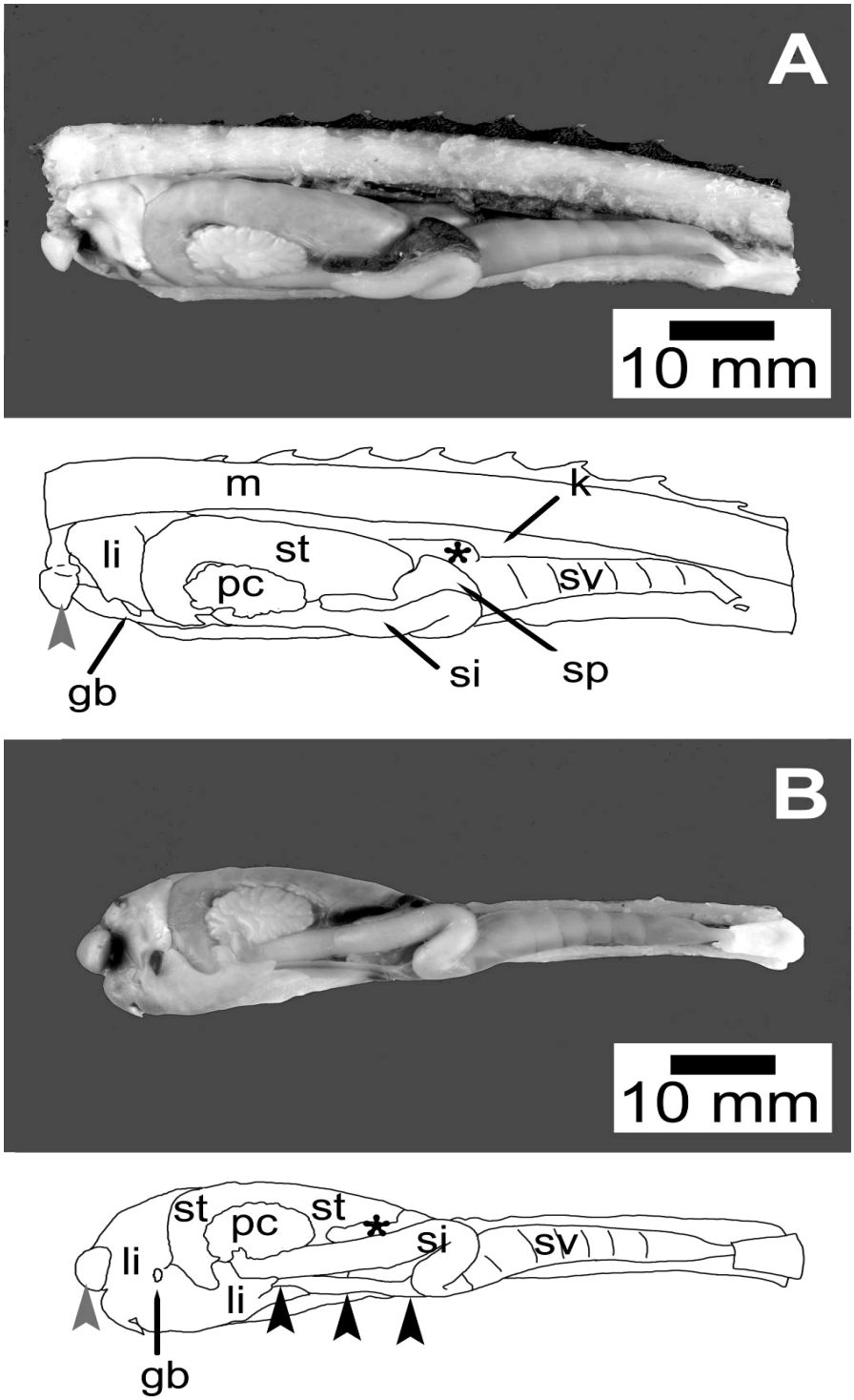

Fig. 4. Photograph A: left oblique view (45 ° angle). The sketch shows the dorsal muscles (m), kidney (k), heart (grey arrowhead), liver (li), gall bladder (gb), stomach (st) and pyloric caecum (pc) followed by the small intestine (si) and spleen (sp), the caudal dome of the swim bladder (black *) with the spiral valve (sv) on the right. Photograph B: ventral view of the internal organ package (ventral face). The left side of the photograph is the cranial side the fish, left side of the fish at the top of the photograph. In the sketch below, it is possible to see: heart (grey arrowhead), liver (li), gall bladder (gb) and the stomach (st) with the pyloric caecum (pc), the small intestine with its S curve (si) and the spiral valve (sv). The long branch of the pancreas is highlighted with black arrowheads.

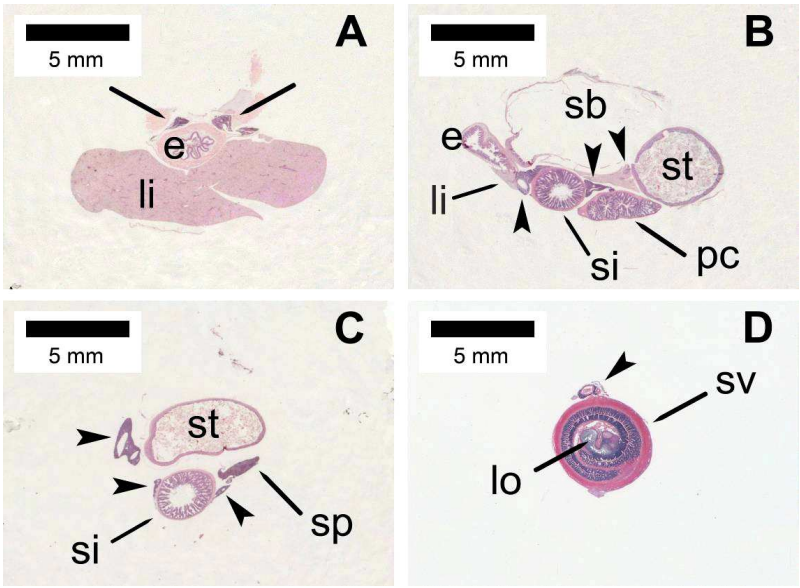

Fig. 5. Transversal histological sections, stained with haematoxylin-eosin. Photographs A to D are in cranio-caudal sequence. The left side of the fish is on the right side for each photograph. Photograph A: the section just caudal to the heart, oesophagus (e) in the sagittal position, head of kidneys (black rods), and liver (li). Photograph B: section passing through the pyloric caecum, and the pancreatic tissue (black arrowheads). The point where the oesophagus becomes the stomach (e), swim bladder (sb). The stomach (st) after the caudal curve, pyloric caecum (pc), small intestine (si). The pancreas comes into contact with the right lobe of the liver (li). Photograph C: the section passes through the large curve of the stomach (st), immediately before the S curve of the intestine, where it then becomes larger in diameter. The small intestine (si) has pancreatic tissue cords on both sides (black arrowheads); it is possible to see the long branch of the spleen (sp). The long lobe of the pancreas (black arrowheads) is on the right side of the stomach curve. Photograph D: section of the proximal third of the spiral valve (sv). In this tract it is possible to see the pancreatic tissue (black arrowhead), and lymphoid organ (lo) in the spiral valve axis.

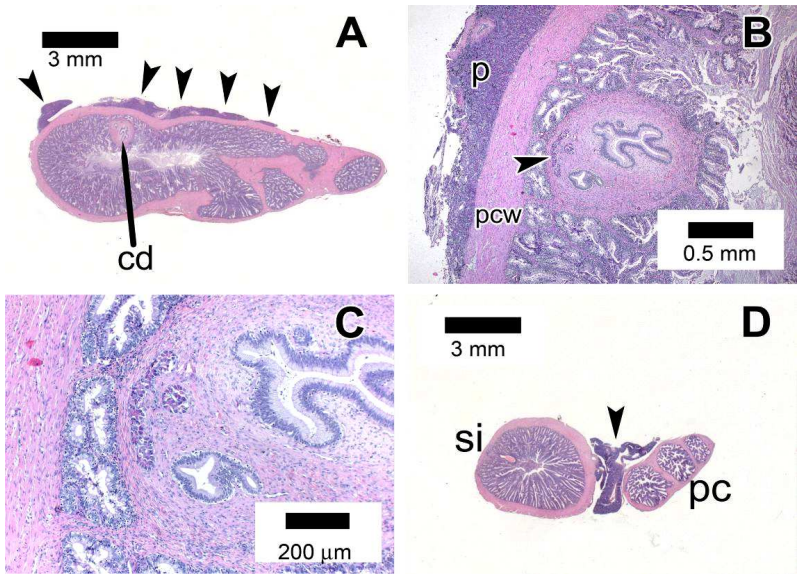

Fig. 6. Sections stained with haematoxylin-eosin. Photograph A: oblique section of the small intestine and pyloric caecum and the dorsal face with pancreatic tissue (black arrowheads). A structure similar to the major duodenal papilla with a common bile duct and, probably, the pancreatic duct (cd), is visible between the dorsal wall and the dorsal mucosa of the small intestine. Photograph B: magnification of the duct zone. Pancreatic tissue (p) on the pyloric caecum wall (pcw) ducts with some pancreatic acini (black arrowhead). Photograph C: details of the duct in photograph B. Photograph D: transversal section through a caudal portion of the pyloric caecum (pc) and small intestine (si). Pancreatic tissue can easily be seen (black arrowhead). It is also possible to distinguish the body of the pancreas and a small branch of the pancreas attached to the small intestine.

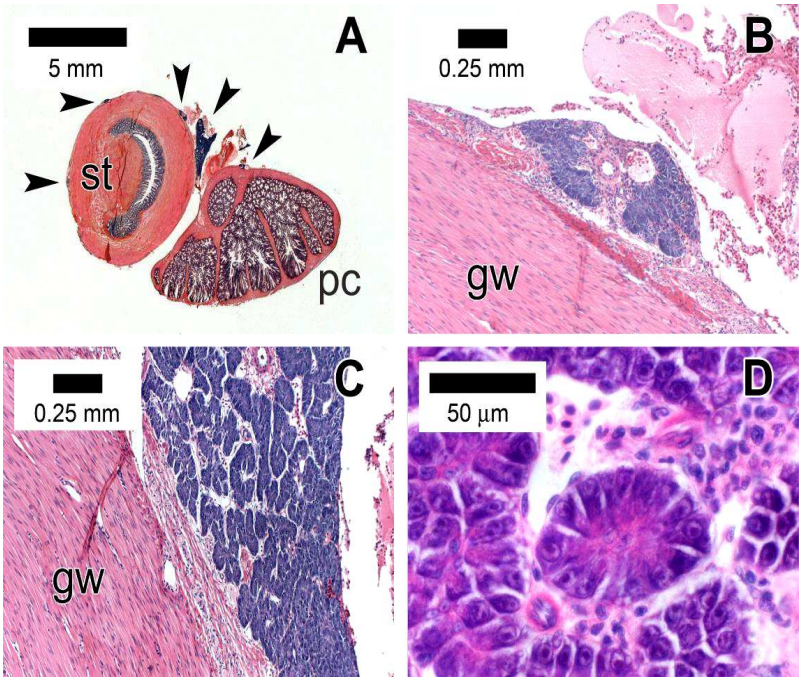

Fig. 7. Haematoxylin-eosin staining, transversal section. Photograph A: gastric pyloric sphincter (st), and pyloric caecum (pc) with attached pancreatic tissues (black arrowheads). Photographs B and C: details of pancreatic tissues attached to the stomach wall (gw). At this level it is not possible to see the islets of Langerhans. Photograph D: details of the exocrine acinar cells of pancreatic parenchyma in the gastric region, basophilic staining of the nuclei and cytoplasm, eosinophilic zymogen granules are apparent in the luminal apex.

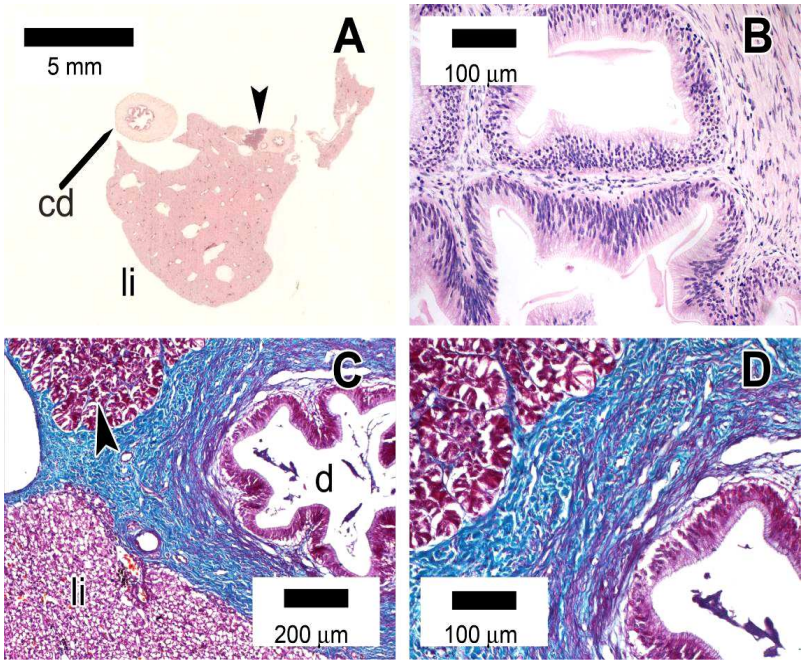

Fig. 8. Photographs A and B: stained with haematoxylin-eosin. Photographs C and D: stained with Crossmon tri-chromic. Photograph A: liver (li) section at the common bile duct origin point (cd). It is possible to see a small branch of the pancreas (black arrowhead) and another duct on the right. Photograph B: details of the common bile duct. Photograph C: details of the pancreatic tissue (black arrowhead) and the duct (d) near the liver tissue (li). Photograph D: details of the duct wall in photograph C.

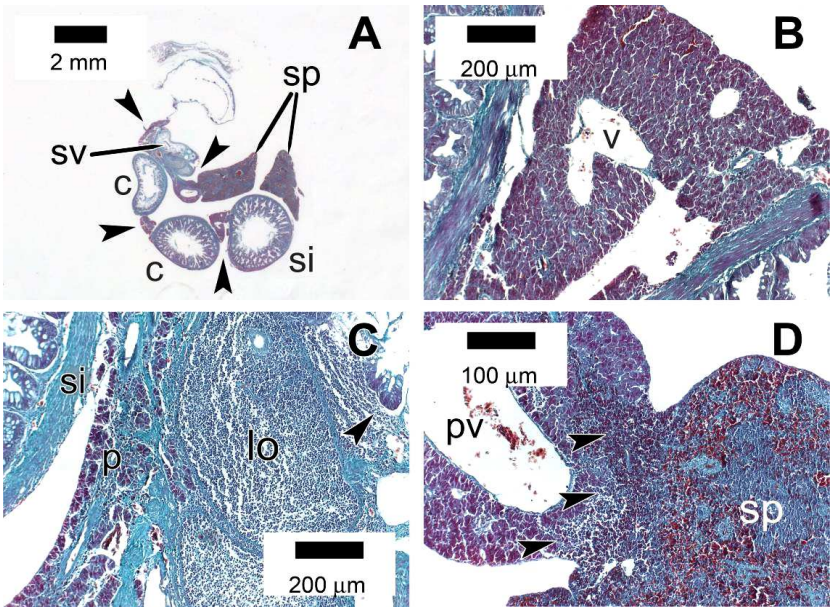

Fig. 9. Transverse sections, stained with Crossmon tri-chromic, passing through the S curve of the small intestine and the point of origin of the spiral valve. Photograph A: macro view of the organs, with the pancreas and the spiral valve (sv) and pancreatic tissues inside the ligaments (black arrowheads). The tracts of the S curve (c), the small intestine (si), and the spleen (sp). Photograph B: details of the pancreas inside the intestinal ligaments. It is possible to see the vein inside the pancreas (v). Photograph C: details of the point of origin of the spiral valve with lymphoid organ (lo) and spiral valve mucosa (black arrowhead), pancreas tissue (p) and the last tract of the small intestine (si). Photograph D: junction between the spleen body (sp) and the pancreas. Lymphocytes (black arrowheads) separate the two different tissues, into the pancreas parenchyma and spleen. It is possible to see the large pancreatic vein (pv).

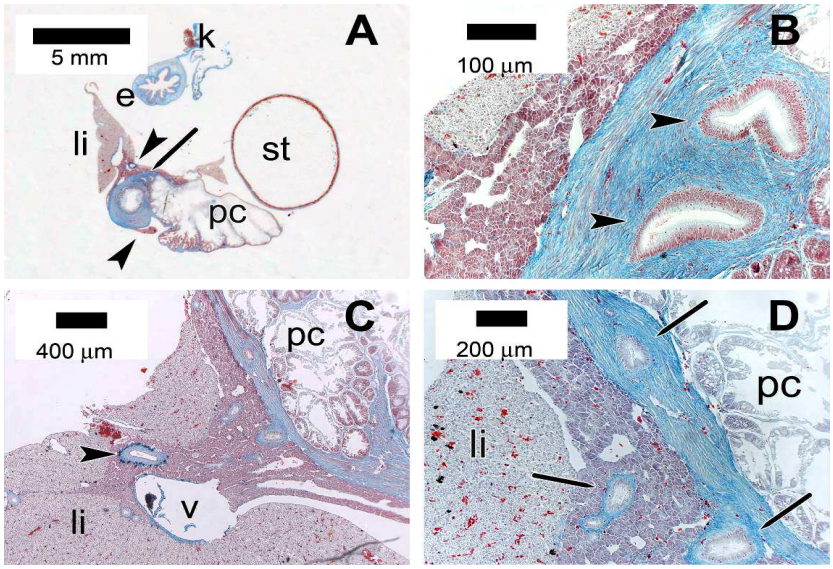

Fig. 10. Transversal sections (stained with Mallory AZAN tri-chromic). Photograph A: intestinal package just behind the opening of the pyloric caecum. The kidneys (k) are visible with the ligament that attaches the first part of the stomach (e) to the vertebral column, right lobe of the liver with the centre-pancreatic vein extending into the liver (li). The pancreatic tissues (black arrowheads) are darker than the liver, is visible the crossing point of the common bile duct and pancreatic duct (black rod) through the intestinal wall. The stomach between the large gastric curve (st) and pyloric caecum (pc) is visible. Photograph B: details of the ducts in photograph A (black arrowheads). Photograph C: section before photograph A with the pyloric caecum (pc), liver (li), centre pancreatic vein (v), and artery vessels with melano-macrophages centre (black arrowhead). At this level, the ducts cross the pyloric caecum wall. Photograph D: details of photograph C with the pyloric caecum (pc) and liver (li). At this level, the ducts (black rods) start to cross the intestinal wall.

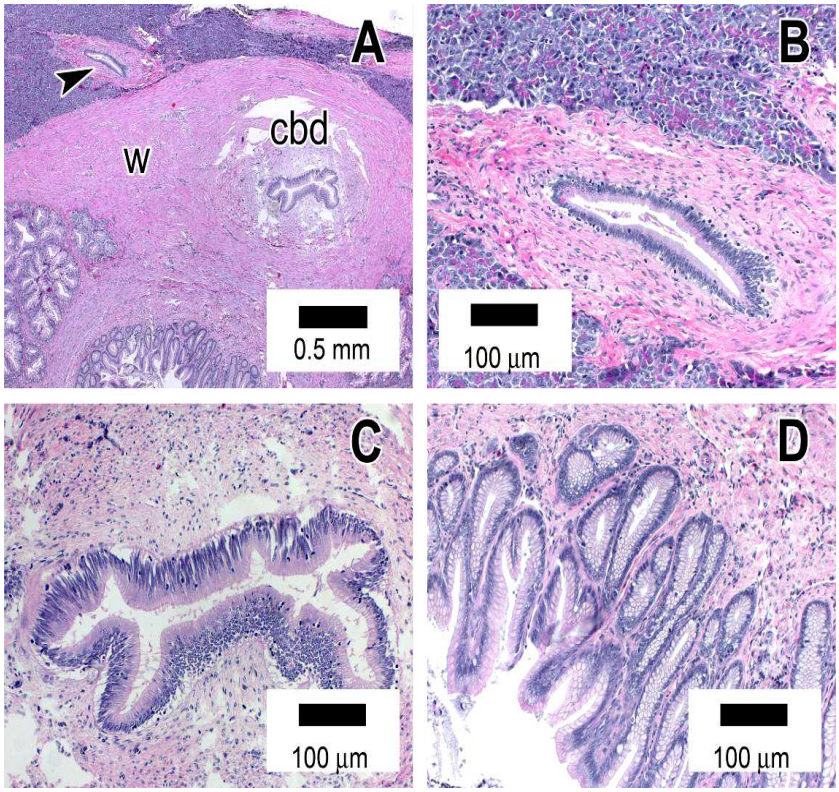

Fig. 11. Transverse sections of the pyloric caecum stained with haematoxylin eosin. Photograph A: common bile duct (cbd) that crosses the pyloric caecum wall (w). Pancreatic duct in the pancreas tissue (black arrowhead). Photograph B: details of the pancreatic duct. Photograph C: details of the common bile duct. Photograph D: details of the pyloric caecum mucosa.

The central pancreatic veins collect the intestinal venous blood, each pancreatic lobes have a large vein inside the parenchyma. All pancreatic veins and the right lobe of the pancreas merge, in the cranial tract of the small intestine, into the long lobe of the pancreas and the veins vessel terminate in the liver (Fig. 10C).

The common bile duct and pancreatic ducts pass over the intestinal wall in dorsal position (Fig. 10A), merge in a structure that resembles the major duodenal papilla of mammals, and opens in to the pyloric caecum. Two ducts (Fig. 6) or only one (Fig. 11) characterizes the papilla.

Macroscopic observation of the sturgeon gut mucosa shows changes in its structure along different tracts and we use those as parameter for dividing and naming the different tracts. Where the pharynges becomes the oesophagus (Fig. 12A), it is possible to see that the large pyramidal papillae are reduced and become small longitudinal folds. The gastric mucosa has high longitudinal folds next to the large gastric curve (Fig. 12B). The small intestine starts from the pyloric sphincter of the stomach: the lumen of the pyloric caecum and the small intestine mucosa present a villous and velvet-like aspect, respectively (Fig. 12C). The small intestine mucosa display a complex architecture from the end of the pyloric caecum region till the S curve, has a characteristic net shape (Fig. 12D). The intestinal S curve mucosa has villi with papillose aspect (Fig. 12E). The spiral valve has a villous aspect of the mucosa and it is possible to distinguish nodular structures inside its axis that correspond to lymphoid organs (Fig. 12F). The dimensions of these nodes become smaller proceeding in the caudal direction.

Histological sections indicate that the oesophageal mucosa has a pseudo-stratified epithelium, rich in goblet cells and that the sub-mucosa has a connective tissue structure with wide spread fatty cell clusters. After the junction with the pneumatic duct, acinar glands begin to appear and, at this point, the oesophagus becomes the stomach (Fig. 13D). Histological sections allowed observing the distribution of the nervous structures in the oesophagus wall. In the oesophagus cranial part, we saw strips of myelinated nerve fibres between the serosa and the muscular layer; whereas in the caudal direction (few millimetres) the nerves can be seen inside the muscular layer and in the sub-mucosa, resembling the vagus nerve system of mammals.

The stomach muscular layer thickens increase progressively from the beginning till the pyloric sphincter where the maximum is reached (Fig. 7A). The gastric glands have acinar structure with small lumen and cells with dense cytoplasm in the cranial part of the stomach (Fig. 13C,D) and in the next tract have the typical structure of the oxynticopeptic glands of the stomach (Fig. 13B). Histology shows that the mucosal architecture

is particularly complicated in the small intestine (Fig. 6D) and in the pyloric caecum (Fig. 7A) and it is cavernous with elaborate networks of villi.

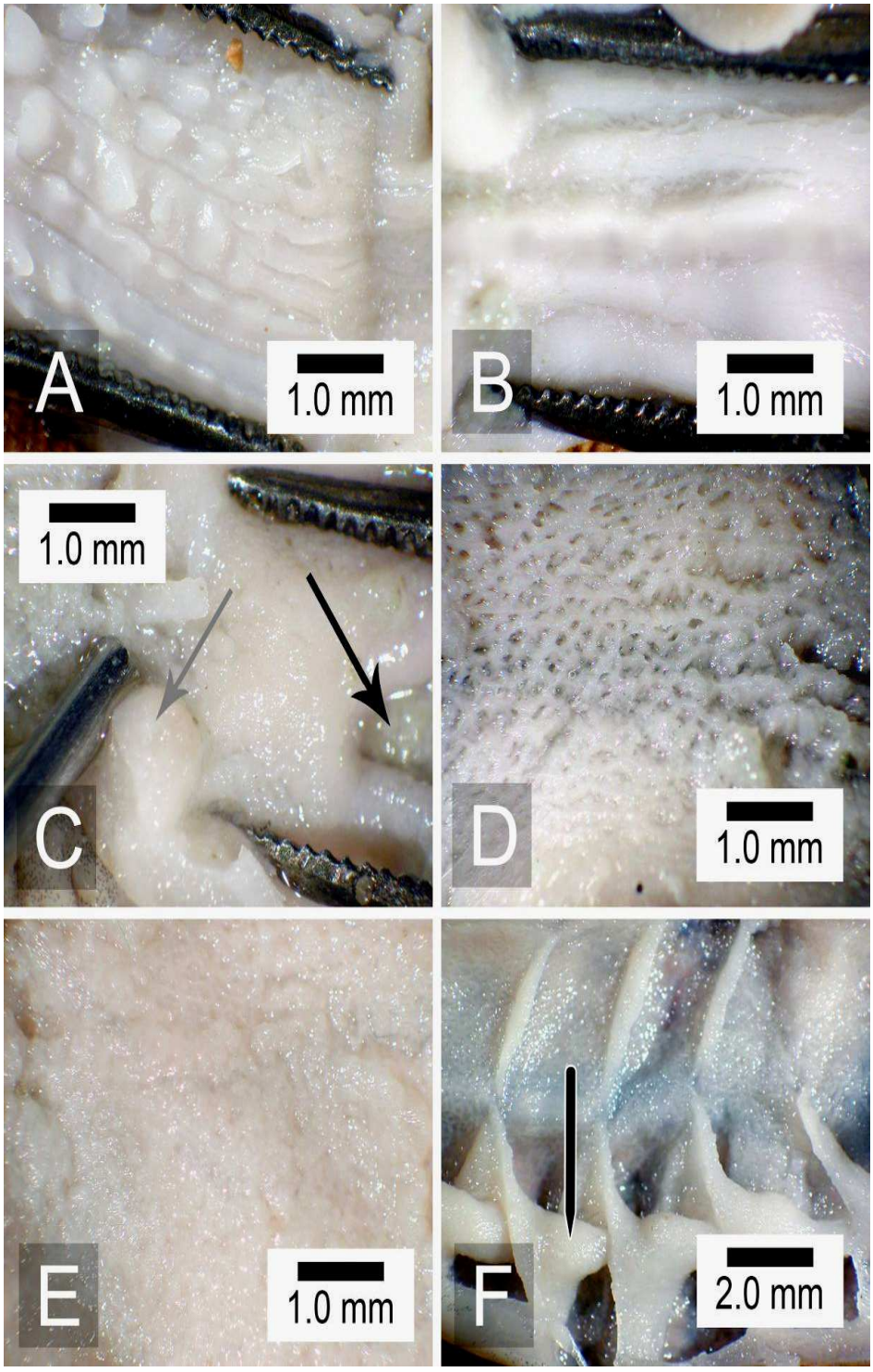

Fig. 12. Macroscopic views of different tracts of sturgeon gut after partial formalin fixation: the cranial side of the intestine is on the left side in all photographs. Photograph A: zone where the pharynx becomes the oesophagus. Photograph B: gastric mucosa with its longitudinal folds next to the large curve. Photograph C: pyloric sphincter of the stomach on the left (grey arrow) and the pyloric caecum opens onto the small intestine on the right (black arrow). Photograph D: small intestine mucosa and its characteristic net shape. Photograph E: mucosa of the intestinal S curve with a papillary aspect. Photograph F: mucosa of the spiral valve with a villous aspect and it is possible to distinguish node structures in its axis, which correspond to the lymphoid organs (black rod indicates one knot).

Histologically, the pancreas is composed of exocrine parenchyma organized in acini. The acinar cells of parenchyma have basophilic cytoplasm and the zymogen granules are eosinophilic sited in the luminal apex of the cells (Fig. 7D). Endocrine cells are grouped in structures similar to Islets of Langerhans in the long lobe of the pancreas (Figs. 14A,B; 15). These islets are not visible in neither main portion of pancreas or in the small branches attached to the intestinal wall (Fig. 14D). In Siberian sturgeon islets of Langerhans are cord structures with histological organization similar to those of mammals. A morphometric evaluation of the histological sections of the islets showed that their mean area is 8.32 $mm^2 \cdot 10^{-3}$, with a range from 2.31 to 22.99 $mm^2 \cdot 10^{-3}$ (Fig. 16) in our specimens. The mean length is 68 µm but they can attain a maximum of 161 µm for fish between 50 and 300 g of body weight.

The serous membrane envelops the entire pancreas and follows blood vessels that enter and exit from the gut organs and pancreatic tissue (Figs. 7; 10; 14). Where the long pancreas lobe touches the right liver lobe, there is a thin layer of connective tissue that dividing the pancreas from the liver. The same organization was found for the other pancreatic branches attached to intestinal or gastric walls.

Arteries, veins, and ducts are apparent in the pancreatic parenchyma, veins and duct, increase in diameter as they proceed in cranial direction (Fig. 5). Histologically, the pancreatic small arteries and small pancreatic ducts are similar except for the epithelial lining, indeed a simple layer of endothelial cells characterizes arteries and veins whereas the small pancreatic ducts have a simple prismatic epithelium.

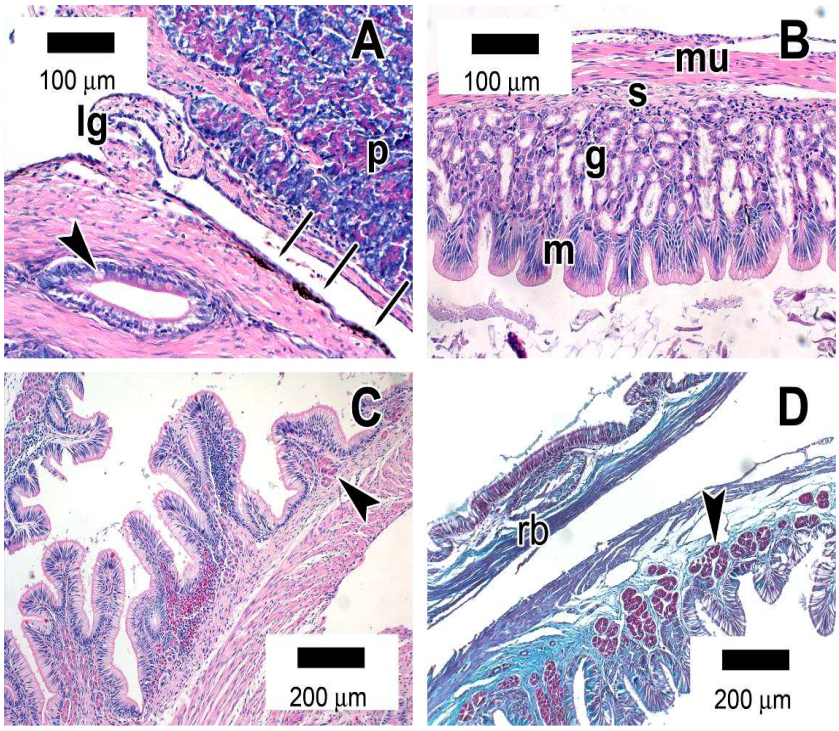

Fig. 13. Sections A, B and C stained with haematoxylin-eosin. Photograph A: details of the ligament that join the intestinal organs and the pancreas. It is possible to see bile or a pancreatic duct (black arrowhead), the structure one of the ligament (lg), and pancreas tissue (p). The melano-macrophages centre in the serous membrane of the gut (black rods) can be clearly seen; they provide the characteristic macular pigmentation of the gut organs. Photograph B: details of the thickness of the gastric wall next to the large curve. The stomach is replete and the folds of the mucosa are stretch; the muscular (mu) and sub-mucosa (s) layers are thin. The glands (g) and gastric mucosa (m) layers appear flat. Photograph C: zone where the oesophagus becomes the stomach. The sub mucosa glands (black arrowhead) have a different structure from the next stomach tract shown in photograph B, and are not distributed throughout the sub mucosa. Photograph D: section of the first tract of the stomach. The sub mucosa glands (black arrowhead) have the same structure as the glands in photograph C but are evenly distributed throughout the sub mucosa. The red body of the swim bladder (rb) closer to the stomach (Crossmon stain).

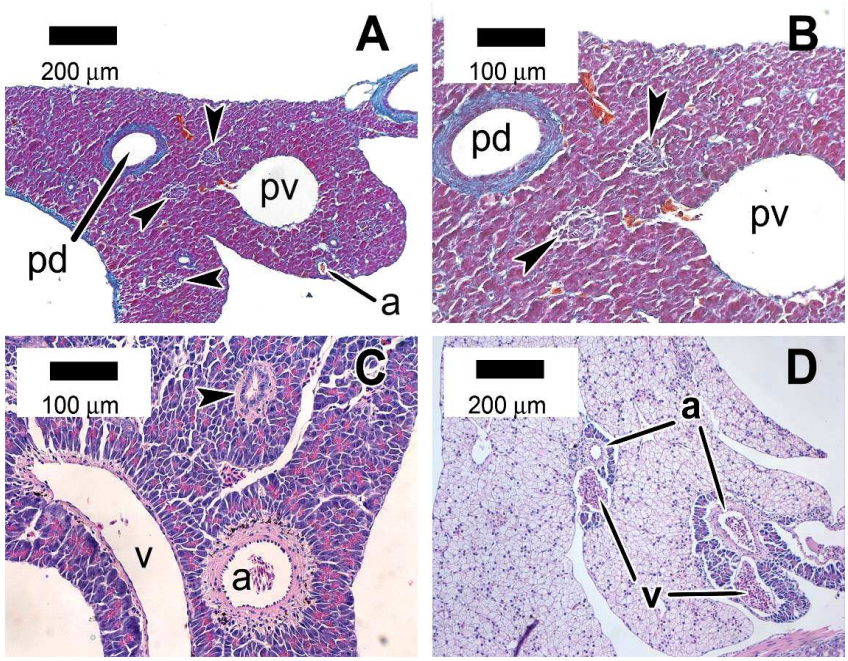

Fig. 14. Transverse sections, stained with tri-chromic Crossmon. Photograph A: the islets of Langerhans (black arrowheads) can be easily seen and it is possible to distinguish the arterial vessel (a) with red blood cells inside the pancreatic vein (pv) in the point where the small vein joins the pancreatic duct (pd). Photograph B: details of the mid zone of photograph A. Photographs C and D stained with haematoxylin-eosin. Photograph C: transverse section of the long lobe of the pancreas where it is possible to observe the arterial (a) and venous (v) vessels and a small pancreatic duct (black arrowhead). Photograph D: a branch of the liver where it is possible to distinguish the pancreatic tissue around the blood vessels, arteries (a) and veins (v).

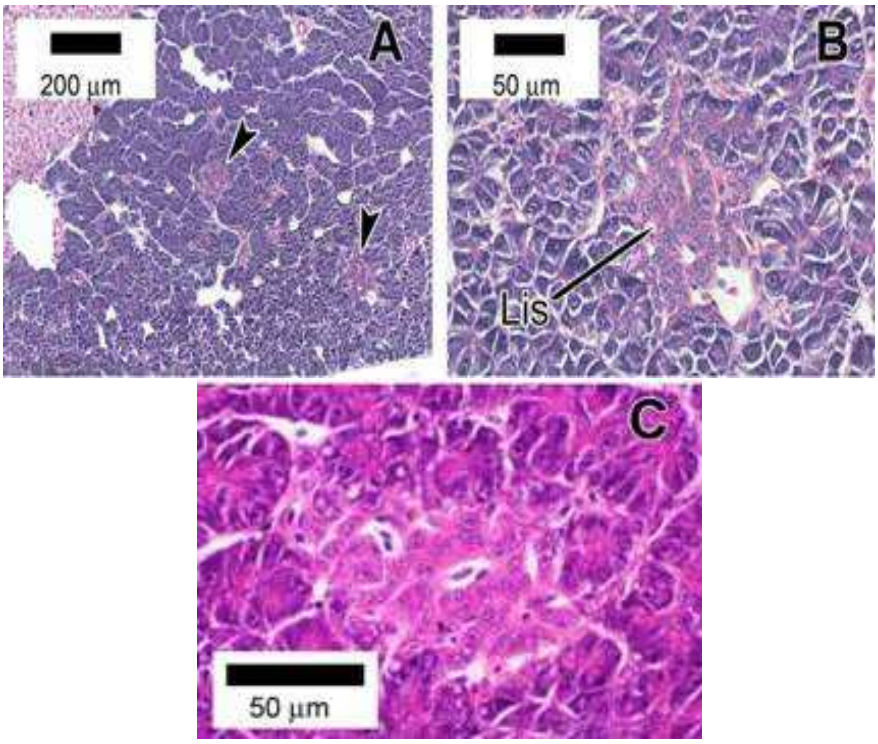

Fig. 15. Pancreas sections stained with haematoxylin-eosin. Photograph A: zone where the long lobe touches the right liver lobe. The islets of Langerhans (black arrowheads) are visible. Photograph B: details of the islet (Lis). Photograph C: detail of Islet of Langerhans and erythrocytes of its blood vessel.

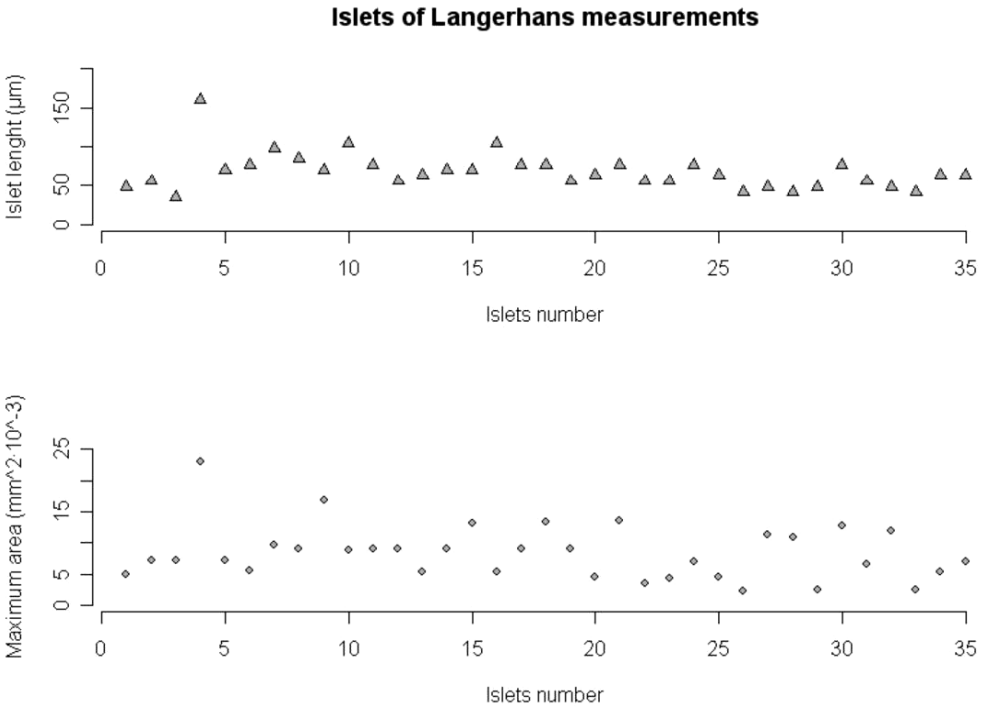

Fig. 16. Islets of Langerhans measurements: On the X-axis, there is the number of the sampled islets. The graphic A has on the Y-axis the islets length (▲) in µm. The graphic B have on the Y-axis the maximum area of the islets (●) in $mm^2 \cdot 10^{-3}$.

Moreover, some arteries have melano-macrophages cells in the adventitia (Fig. 10C), and they are not present in pancreatic ducts adventitia. Small pancreatic ducts have a simple cubic epithelium while in the larger ducts, the epithelium is pseudo-stratified (Figs. 10B,D; 13A; 14). Melano-macrophage centres are present, especially in the liver, around arterial vessels inside the organs, and between the serous membrane and muscular layer (Figs. 10C; 13A).

The observation on unfed sturgeon did not highlight anatomical differences compared with fed sturgeon, except for the pigmentation of the abdomen and the gut due to melano-macrophage centres. When feed was withheld, the gut organs appeared relatively pale (Fig. 1, lower), in contrast to the highly pigmented (Fig. 2B) appearance generally observed fed fish.

Discussion

The pancreas of the Siberian sturgeon is an organ with a principal part "pancreatic body" situated on the dorsal side of the pyloric caecum, between the small gastric curve and the liver. Three lobes originate from the pancreatic body in the visceral cavity. The long lobe ends in the spiral valve dorsal vein ligament. Two smaller lobes, left lobe and right lobe, are located on both sides of the small intestine. Nicolas (1904) observed, in *Acipenser sturio* larvae, that the dorsal embryonic process ends before the spiral valve, running dorsal of the visceral cavity. This is partially in agreement with our observations. The structure, named "dorsal embryonic process" by Nicolas, appears to correspond to the long lobe of the pancreas that we observed in the Siberian sturgeon examined by us. The long lobe shows a cranio-caudal and ventro-dorsal pathway that is quite different compared to the dorsal embryonic process that appears as cranio-caudal in the dorsal position. Moreover, the long lobe ends between the first and the second coil of the spiral valve upper side, which is in contrast with observations made by Nicolas in the larvae.

Our consistent observation of zymogen granules in the exocrine pancreas confirms prior studies with the larvae of green sturgeon, *Acipenser sturio* larvae (Nicolas 1904) and *Acipenser medirostris* (Gisbert and Doroshov 2003).

We used gross anatomy, stereomicroscopy and light microscopy to identify the pancreas and the pancreatic ducts. Einarsson and Davies (1997) described the pancreatic ducts in Atlantic salmon observed in haematoxylin-eosin stained sections. They noted that the histological structure of ducts is a monolayer of flat epithelium. Our observations on small pancreatic ducts are in agreement with the previous cited observation but, in contrast; we observed in Siberian sturgeon that big pancreatic ducts have a pseudo-stratified epithelium, and displayed a

underlying muscular layer. Overall, the anatomical organisation of sturgeon pancreatic ducts was similar to those of mammals.

A structure resembling the major duodenal papilla of mammals was apparent in the pyloric caecum. However, it was not possible to discriminate the common bile duct from the pancreatic duct because they have a similar histological appearance. Nicolas (1904) stated that during embryonic development pancreatic ducts merge in the wall of the common bile duct, near the caudal end of the liver. They proceed along the dorsal wall of the small intestine and end up close to the junction point between the small intestine and the pyloric caecum. Nicolas also reported that the last tract of the common bile duct is surmounted by the dorsal pancreatic bud that, at this point, merges with the two ventral pancreatic buds, closer to the liver. In our observations on Siberian sturgeon, we did not find the same anatomical interaction. The pancreatic lobes of Siberian sturgeon merge in the body of the pancreas, at the level of the pyloric caecum dorsal face. The common bile duct was completely embedded in the pancreatic tissue for two/thirds of its length, and we found that the body of the pancreas had many small ramifications, in the serosa layer, that follow blood vessels entering and exiting from the gut organs.

Two types of Islets of Langerhans were described in *Silurus asotus* (*Siluridae*) and *Siniperca scherzeri* (*Centropomidae*) (Lee et al. 2001). They differ in size and location according to the different endocrine activities and functions. In the present study on Siberian sturgeon, the main location of the Islets of Langerhans was in the long lobe and left lobe caudal portion of the pancreas. Different sized islets were present; no such structure was found in any of the pancreatic small branches to the gut organs. The variability in size of the Islets of Langerhans in fish, which was studied by Massari (1898) and Diamare (1899), is affected by feeding status and during periods of starvation the atrophy of this gland occurs. In the present study pancreatic atrophy or Islet of Langerhans modification were not apparent in sturgeon held without feed for two weeks, we suppose that sturgeon do not modify pancreatic structure in response to food deprivation period in contrast with other fish pecies.

Melano-macrophage centre in Siberian sturgeon is related to the pigmentation, and its activity is linked to digestive processes and body size, but unfortunately no data are available in literature about the function the relation of these particular structure of sturgeon, further studies are needed to clarify them function.

Gisbert et al. (1999) provided prior observations of sturgeon intestine tracts, liver and pancreatic tissue reactivity, treated with different histological stains. However, their work was limited to the ontogenesis of these organs, but no anatomical description or deal with the relationship between the pancreas and the other gut organs in successive body developmental stages are available. Cataldi et al. (2002) reported the gut ontogenesis description of the Acipenser naccarii but they did not describe pancreatic tissue. The histological description of the alimentary tract of white sturgeon (Domeneghini et al. 2002) provided a framework for comparison with these studies in Siberian sturgeon. Andoh and co-workers (Andoh et al. 2000) have similarly provided useful observations about molecular forms of insulin and glucagons in *Huso dauricus* whole homogenised gut but no mention were done concerning anatomical description of the pancreas or the possibility to sampling only this specific tissue.

In conclusion, different intestinal tracts were defined on the base of the mucosa morphological aspect. The sturgeon pancreas is histologically similar to those of mammals, and it is anatomically quite different from that of other fish. The pancreas of Siberian sturgeon is grossly visible during dissection and is independent of the liver and other abdominal organs, even if in many portion the serous membrane joint it to gut wall and liver. Three large lobes start from the pancreas body and they run in a caudal direction. To our knowledge this is the first work providing an anatomical description and disposition of sturgeon gut organs in post larval stage with detailed photographic tables enclosed, moreover it seems that photographs of historical works on larval stage (from 1898 to 1929) are not available. Our findings further define the gastrointestinal anatomy of Siberian sturgeon and provide a viable baseline for comparative studies of sturgeon anatomy and organ health when assessing dietary formulations.

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