The microvascularization of the human rectal continence organ: Scanning Electron Microscopy & Light Microscopy

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The fine continence organ of the anus consists of several functional units of the rectal wall: the Corpus cavernosum recti, external and internal anal sphincters (IAS), the M. canalis ani and M. puborectalis, the transitional epithelium and the anoderm [1].
Especially the Corpus cavernosum recti, a cavernous body in the submucosa, and the IAS play an important role in maintaining the continence. The IAS is in a stage of permanent tonic contraction, guaranteeing the closure of the anus and assisting in the swelling of the Corpus cavernosum recti by decreasing its venous outflow [2]. The cavernous body consists of large dilated veins, which form the anal cushions, bulging as mucosal folds into the lumen of the anal canal and thus closing the anus completely. Though it is generally assumed that the dilated veins fill through arterio-venous anastomoses [3], there is no proofed evidence of their existence yet. It is more likely that sphincter structures and abrupt caliber changes in the dilated veins block venous outflow [4] what could explain their rapid filling after defecation with a big amount of blood.
The aim of the present study is to investigate the vascular supply of the tissue layers of the rectal continence organ with special emphasis on the submucosal layer, to gain insight into the cavernous body blood flow regulatory mechanisms, and the role these structures could play in the development of haemorrhoidal disease.

The vascular bed of excised human recta was cast post mortem. Briefly, the resin Mercox-Cl-2B® (Ladd Res. Inc., Burlington, VA, USA) was injected with manual pressure via a canula inserted into the superior rectal artery. After hardening injected specimens were macerated in 7.5% potassium hydroxide (40°C), rinsed in running tap water, cleaned in 5% formic acid and frozen in distilled water. While frozen in ice specimens were cut longitudinally and/or transversally by a bandsaw. Samples were freeze-dried (LYOVAC GT2, Leybold-Heraeus) and mounted by the “conductive bridge-method” [5]. After evaporating with carbon and gold vascular casts were analysed in the scanning electron microscopes (Cambridge Stereoscan 250 and Philips ESEM XL30) at 10 kV accelerating voltage. For further details on SEM and vascular corrosion casting see [6]. For light microscopy specimens of the whole anal canal (with part of the rectum) were fixed in 10% buffered formaline, dehydrated in a graded series of ethanol and embedded in paraplast. Longitudinal sections of 7 or 10µm thickness were cut and stained with AZAN. To visualize vascular smooth muscle cells sections were stained with anti-smooth muscle α-actin antibodies conjugated to DAB. For documentation stained sections were digitally photographed with an Olympus microscope (BX-51, Olympus, Vienna) using the Cell-A software (Olympus).

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All specimens revealed a distinct network of venous vessels in the submucosa of the anal cushions as well as in the subcutaneous layer of the anoderm at the height of the pecten analis. In both layers venous vessels showed many conspicuous dilatations of varying sizes, shapes and interconnections (Fig. 1). In the anal cushions the dilated venules formed an extensive plexus immediately beneath the subepithelial vascular layer (Figs. 1,2). Dilatations were interconnected by very small to large caliber vessels and showed a diameter of 300 - 600µm. Dilated venules locally exhibited venous valves, prominent narrowings indicating sites of sphincters, and "holes" representing sites of intussusceptive microvascular growth (IMG) (Fig. 2). IMG and its facets were clearly seen to act in shaping the wide dilated veins. Arterio-venous-anastomoses could not be found. In some specimens the submucosa also revealed undilated but very tortuous veins with diameters up to 1mm. Venules of the subepithelial vascular bed were found to drain into nearby wide submucosal veins, whereas in the subcutaneous layer of the anoderm prominent pearl-string-like venules were present. If and how these veins are interconnected with the dilated veins still remains open for further investigations.

Histology revealed that dilated venules localized throughout the whole anal canal in the submucosal and subcutaneous layers. The biggest dilatations appeared at the region where the mucosa changed into the squamous epithelium of the anoderm. In this region the IAS achieved its maximum thickness. Within the wall of dilated venules smooth muscle cells were accumulated locally forming sphincters (Fig. 3). Again, AV-anastomoses could not be found.

We conclude that in the cavernous body of the rectal continence organ sphincter structures together with venous valves of the wide dilated veins act as flow regulatory mechanisms.

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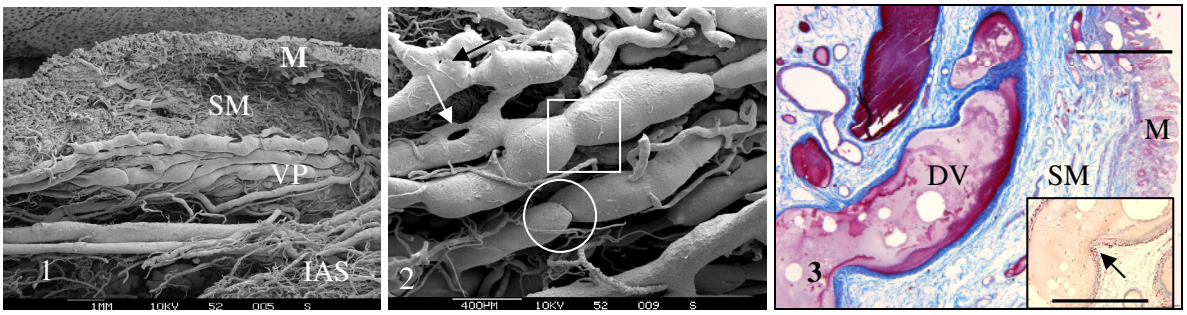

Figure 1. Human anal cushion. Microvasculature. Vascular corrosion cast (VCC). Longitudinal section. Scanning electron micrograph. IAS internal anal sphincter muscle, LU lumen, M mucosa, SM submucosa, VP venous plexus. **Figure 2.** Venous plexus in the submucosa. Detail view. Enboxed area indicates a sphincter site, encircled area outlines a venous valve. Arrows point to sites of IMG. **Figure 3.** Histomorphology of the human anal cushion. M Mucosa, SM submucosa containing a wide dilated vein (DV). AZAN staining. Inset: Accumulation of smooth muscle cells within the wall of the dilated vein forming a sphincter (arrow). Immunostaining for alpha-smooth muscle actin. Bars = 1mm.

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