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Histology of Umbilical Cord in Mammals

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Abstract

The histology is one of the most important disciplines for studying of organic tissues such as the placenta, umbilical cord and related structures which has been poorly studied in mammals, mainly in domestic animals. The umbilical cord, connection between fetus and mother, is one of the most important anatomical structures related to the development of the animal during their stage of formation due to its participation as responsible of the exchange of nutrients and protection of the structural vessels. There are several researches which have described anatomic, histological, immunohistochemical characteristics of the main constituents of the umbilical cord in wild and domestic animals due to its importance in the maintaining of the fetus during their development. The present chapter recompiles and describes the majority of researches about histological characteristics of the constituents of the umbilical cord included images and graphics that improve the understanding about the histological main differences found among mammals and also, this review aims to be a great helpful for the pathologist, surgeons and staff dedicated to reproduction and research about stem cells, this last considered one of the fields of special interest since some years ago in veterinary medicine.

Keywords: allantoic duct, histology, umbilical artery, umbilical cord, umbilical vein, mammals

1. Introduction

When an animal is born, it is necessary that several changes in the fetal circulation appear such as fast closure of some anatomical communication channels by coordinated mechanisms which arrows to autonomic life. The umbilical orifice permits the passage of the umbilical cord, being this characteristic a natural defect [1].

The majority of mammals have an umbilical cord and eventually an umbilicus, but sea shellfish, the whole animals, looks like an umbilicus [1].

The umbilical cord was originated from the embryonic stem which connects the bladder of both yolk sac and amniotic. This is discarded after birth in all species of mammals [2]. The umbilical cord forms a connection between placenta and the fetus. This structure is responsible for exchange of nutrients during the gestation. The main characteristic that umbilical cord shows is a specific gross morphology of vein and arteries surrounded of mucous connective tissue. It is known that fetus-mother nutrients exchange is very delicate and difficult to maintain; however, many morphological and functional alterations may produce changes in this mechanism of exchange easily.

In relation to the hematopoietic cells, these have been widely studied functionally, molecularly and structurally, but there are few studies about ultrastructural characterization [3]. The use of stem cells obtained from umbilical cord have originated a lot of expectative for use it in cell therapy and regeneration of organs [4]. Furthermore, it is known that the umbilical cord cells have been studied only in preclinical approaches [5].

The scarcity of bibliographic information about studies of optical microscopy of the umbilical cord in mammals and lack researches about these topics in mammalian species have motivated to execution of this type of reviews that reveal main characteristics in relation to the histology of umbilical cord and comparative aspects between different species of mammals of interest.

2. Umbilical cord

2.1. Structure

The umbilical cord is a structure discarded after the birth and the transplant of their cells may present less risks of causing reactions, resulting immune reactions, resulting in minimum for the recipient of its cells [4]. This structure has amniotic and allantoic segments. The amniotic segment of the umbilical cord contains two arteries and a vein that arborize into the amnion. These segments continue as multiple vessels in the allantoic segment of the cord with mainly two arteries and two veins with branches (**Figure 1**) [6]. The urachus courses within the cord from the fetus and empties into the allantoic cavity [6].

The umbilical cord is a unique mammalian fetal attachment and was attached to the center of the placental disk [7, 8]. This structure plays an important role in the transport of maternal nutrients for developing of fetus during gestation [1]. The umbilical cord shows distinct types of composition with respect to the number of blood vessels. There are many morphological changes that appear at the birth into local anatomical structures around the umbilical cord form a complex device to help the organism to severe relation with the placenta. The relation between anatomical structures that form the umbilical cord arrows the morphological support as basis of contraction to eliminate remnants that was inside the abdominal cavity [1, 6].

As has already been mentioned, this structure has a main function of making the connection between the fetus and placenta, ensuring its viability mainly during the later stages of pregnancy [9]. In relation to the umbilical vessels, these are not supplied by *vasa vasorum* and thus

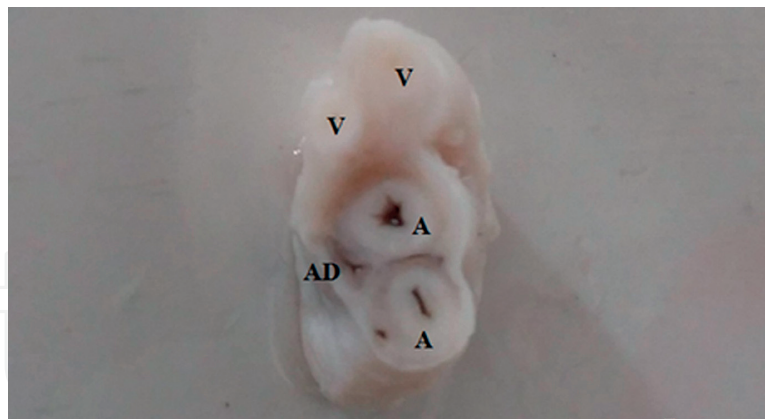


Figure 1. Photograph of umbilical cord of alpaca (*Vicugna pacos*) showing two umbilical veins (V) and umbilical arteries (a). Next to the umbilical arteries is located allantoic duct (AD). (adapted from barrios-Arpi et al. 2017. Histological characterization of umbilical cord in alpaca (*Vicugna pacos*).

depend on their oxygen supply making them more vulnerable to changes originated by hemodynamic disorders and similar conditions [10].

This structure is coated by amniotic epithelium (simple squamous epithelium) and the conjunctive layer is adhered closely to the fundamental substance of the umbilical cord majority known as fetal mesenchyme, which is constituted of mesenchymal connective tissue with stellate cells and amorphous ground substance which contains abundant glycogen. This gelatinous composition also called Wharton's jelly or mucous connective tissue has been of great interest and potential impact due to research about tissue repair and differentiation [10]. However, many of the investigations have not been able to continue due to the lack of an animal model that can be used in the preclinical studies [11]. This mucous connective tissue (Wharton's jelly) is an active metabolically tissue involved in fluid exchange between umbilical vessels and amniotic fluid [12].

In the majority of domestic species, two arteries and two veins wound spirally being immersed in a mucous connective tissue appears [13, 14]; however, it is known that in some species, appear the formation of anastomosis of arteries in middle third and proximal to the maternal part of the placenta appears [15].

There are important changes evidenced in the structure of umbilical cord in different animal species. It is indicated that from the beginning, vessels of umbilical cord are represented by two umbilical veins and arteries in species such as bovines and small ruminants [16], zebu-crossed bovines [17], buffaloes [18–20], pigs [21], African lions and gazelles [22], and Bactrian camels and dromedaries [23, 24] (**Figure 2**); however, it is known that in some species the disintegration of right umbilical vein appears without have a strong explanation about this feature [25] (**Figure 3**). Among these species are carnivores, horses, guinea pigs, nutrias, chinchillas, cavies and rock cavies [26–32].

Finally, as the stages of gestation, the tunica adventitia is becoming thicker in relationship to the tunica intima in both arteries and veins [23].

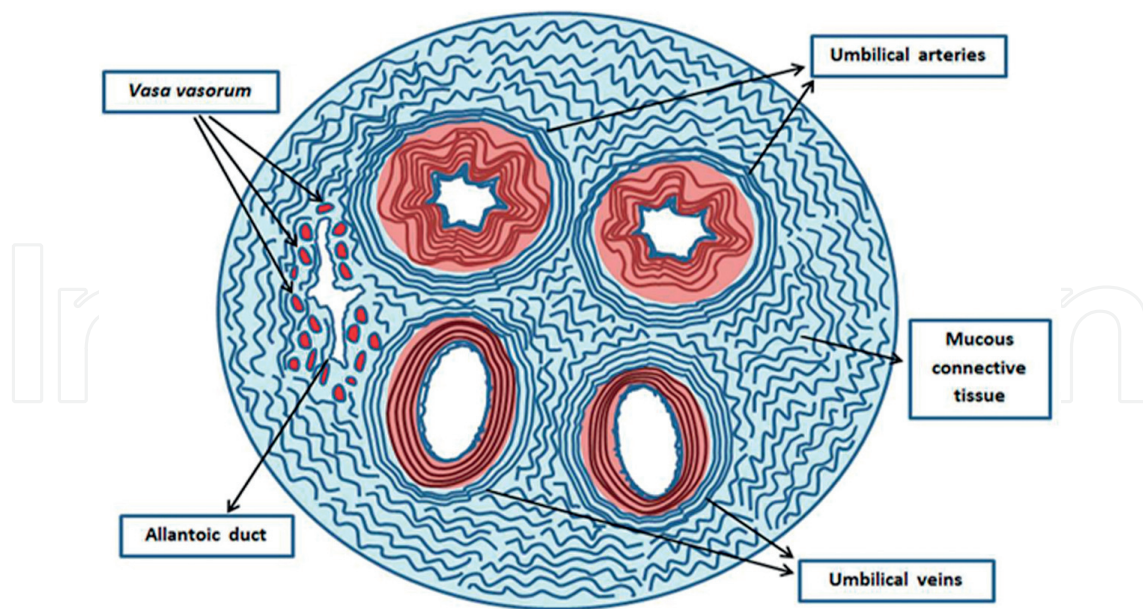


Figure 2. Schematic representation of structure of umbilical cord in cattle, sheep, goat, buffalo, alpaca, canine and feline showing two arteries and two veins. Furthermore, this figure shows the localization of allantoic duct and their relation with blood vessels.

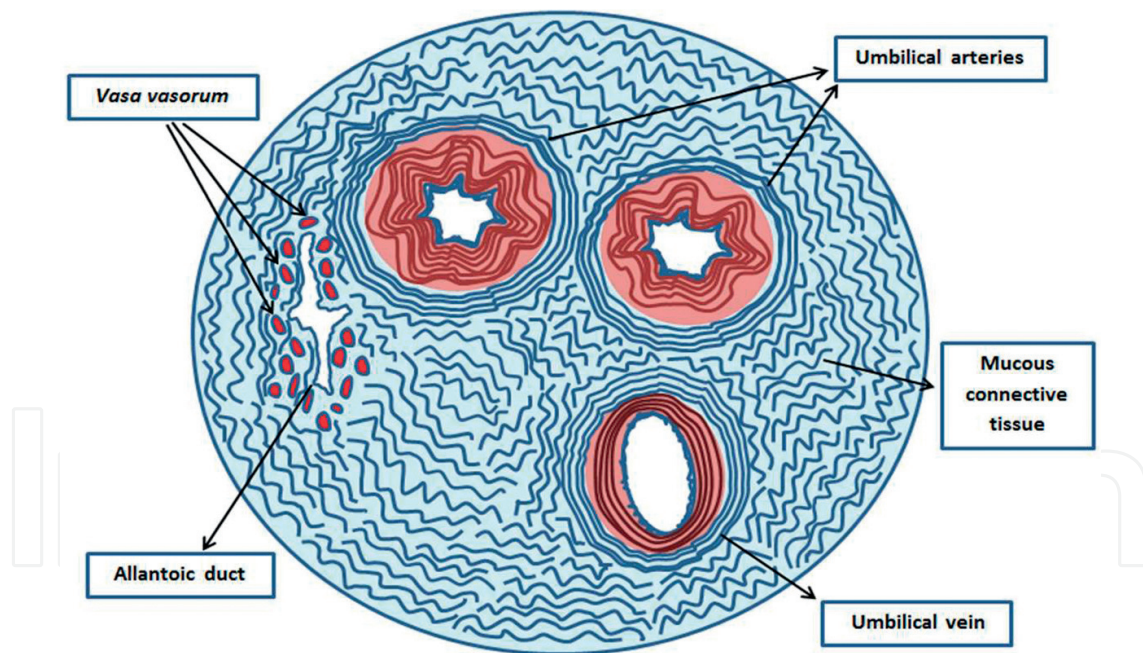


Figure 3. Schematic representation of structure of umbilical cord in horses, pigs and humans showing two arteries and only one vein. Furthermore, this figure shows the localization of allantoic duct and their relation with blood vessels.

2.2. Umbilical artery

At parturition the umbilical arteries retract into the abdomen and close by smooth muscle contraction. This process appears in response to the increased partial pressure of oxygen in the blood [33].

There are some characteristics that differ between lumens of umbilical arteries in different mammalian species. In the majority of mammals, umbilical arteries show a very large lumen and irregular shape. In some species such as buffaloes, bovines and zebu cows, the umbilical arteries show a lumen of star-shaped lumen [17, 18, 34–36]; however, in species as South American Camelids, the animals present a lumen of slightly star-shaped [37]. In mammals, the umbilical artery is constituted by tunics intima, media and external/adventitia (**Figure 4**).

2.2.1. Intima

The intima layer consists of elongated endothelium to the long axis of the blood vessel. The endothelium of tunica intima corresponds to the thinnest layer comprised of simple squamous

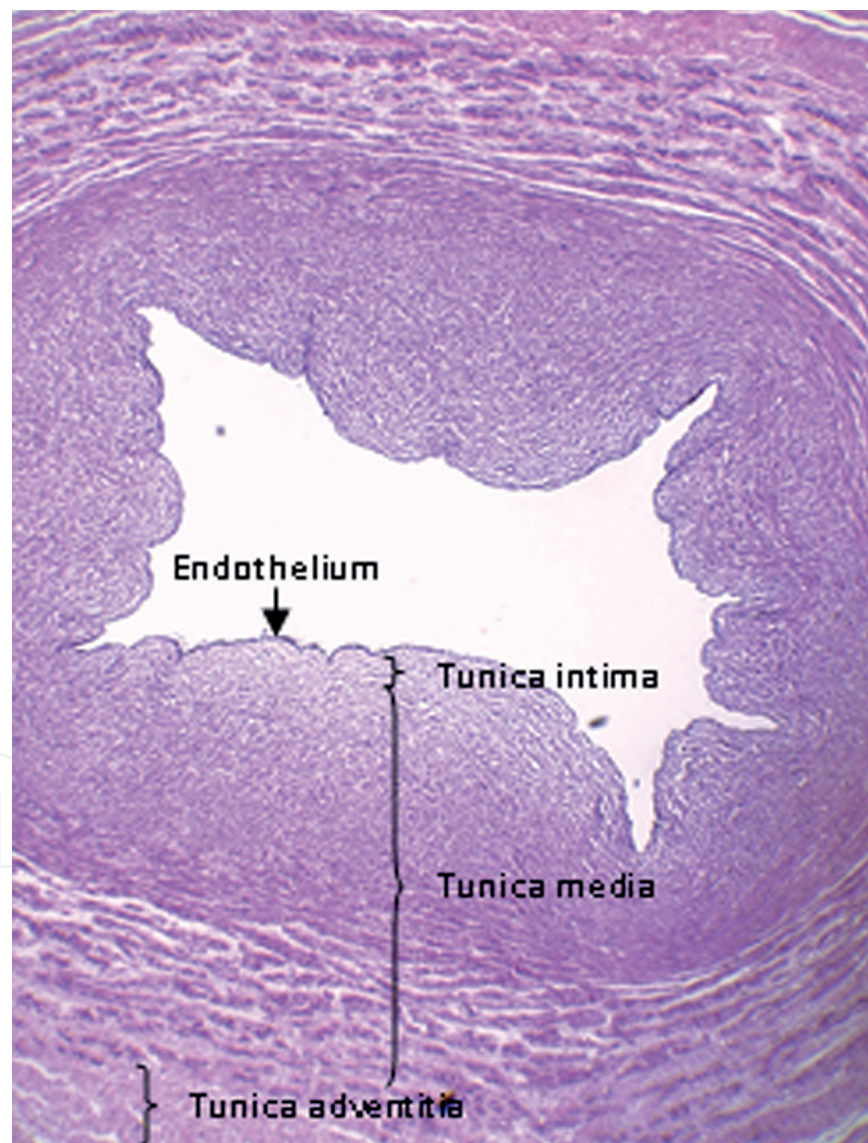


Figure 4. Umbilical artery, light microscopy, H-E stain. The tunica intima is constituted by endothelium and a small endothelial space. The tunica media is formed mainly by smooth muscle and has double size of tunica adventitia. The abundant collagen and elastic fibers of the tunica adventitia contrasts with muscular tissue and collagen fibers of the tunica media. 40 x.

epithelium. Most animals, the internal elastic lamina is discontinuous and thin [17, 18, 32–38] with exception of humans and camels which the umbilical arteries possess no internal elastic lamina [10, 23]. In some species as alpacas is possible to observe a very small sub endothelial space which consisted of muscular fibers (non-differentiated muscular cells) cross and cut diagonally (circular disposition), and connective tissue fibers [37] (**Figure 5**).

2.2.2. Media

The media layer is located below to the sub endothelial space and the thickness of this layer is double the size of the external/adventitia. The tunica media is constituted by a double layered muscular of smooth muscle bundles, characterized for inner circular muscular layer (collagen and reticular fibers) and outer longitudinal layer (**Figure 6**). Another characteristic of the media layer is absence of elastic lamina, a presence of reticular fibers and both collagen and elastic fibers and presence of capillaries [10, 17, 32–38].

2.2.3. Adventitia

The tunica adventitia is the most outer layer that forms the umbilical cord wall in all mammals. The tunica adventitia consists of collagen, smooth muscle and elastic fibers. This layer is constituted by smooth muscular fibers which invade part of tunica media and muscle fibers cross-sectional [10, 17, 18, 32, 34–36, 38].

Unlike other mammals, in alpacas, the inner layer is not well defined [37]. In alpacas, the concentration of collagen fibers is increased towards longitudinally oriented muscular fibers and towards periphery. The elastic fibers are abundant between the tunicas media and

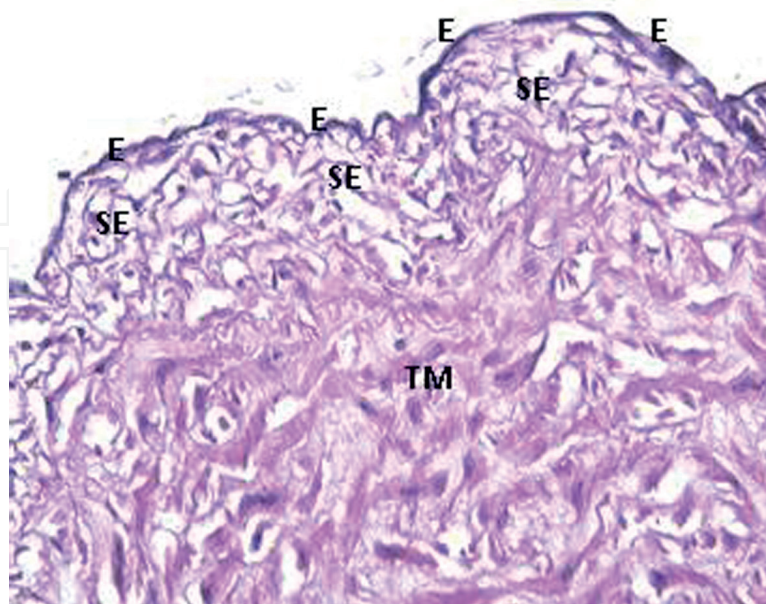


Figure 5. Umbilical artery, light microscopy, H-E stain. The tunica intima is comprised of a simple squamous epithelium (E) and a small endothelial space (SE) characterized by elastic and reticular fibers. Note that the internal elastic lamina is not well-defined. TM: Tunica muscular. 400 x.

adventitia. Finally, the tunica adventitia is constituted by small blood vessels denominated *Vasa vasorum*, non-myelinated nerves cross-sectional and clearly separated from mucous connective tissue (**Figure 7**) [10, 17, 18, 32, 34–38].

2.3. Umbilical vein

At parturition the umbilical vein and urachus remain outside the abdomen. In relation to the vein, this structure closes soon by smooth muscle contraction and the urachus shrinks and dries within a day [1].

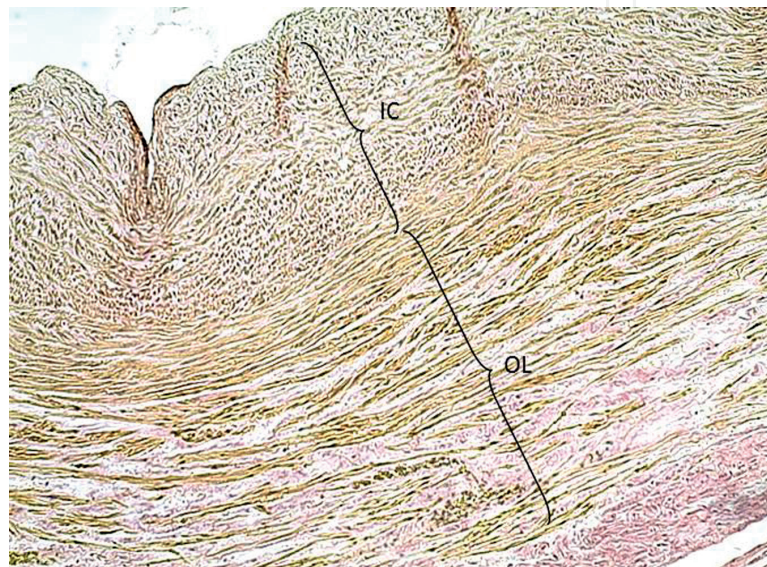


Figure 6. Umbilical artery, light microscopy, van Gieson stain. The tunica media is constituted by a double layered muscular of smooth muscle bundles, characterized for inner circular (IC) and outer longitudinal muscular (OL) layers. 100 x.

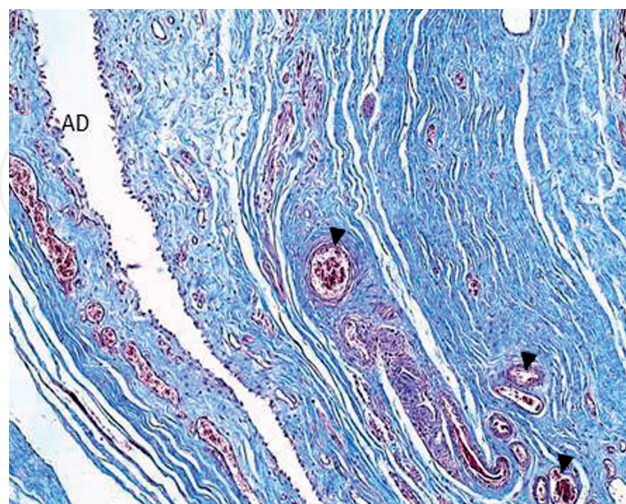


Figure 7. Umbilical artery, light microscopy, Masson's trichrome stain. The tunica adventitia/external is the most external layer of umbilical cord and is constituted by small blood vessels denominated *Vasa vasorum* (short arrows). AD: Allantoic duct. 100 x.

Similar to the umbilical artery, there are some characteristics that differ between lumen of umbilical arteries in different mammalian species. In some mammals such as buffaloes, carnivores, horses, guinea pigs, nutrias, chinchillas, cavies and rock cavies, umbilical vein shows a lumen in elliptic shape with wall thinner than umbilical artery [26–32]; however, in species as South American Camelids and horses, the umbilical vein displays a lumen obliterated into star-shaped [29, 37, 38]. In mammals, umbilical vein is constituted of tunics intima, media and external/adventitia (**Figure 8**).

2.3.1. Intima

The tunica intima consists of endothelium but lack of external elastic lamina with less organization line compared to the umbilical artery. However, in general, endothelium showed similar characteristics as observed in the umbilical artery.

There are several studies that confirm this conformation of the tunica intima which is thin and lack internal elastic lamina [10, 17, 23, 34–38]. Similar to alpacas, is possible to observe a small



Figure 8. Umbilical vein, light microscopy, H-E stain. The tunica intima is constituted by endothelium and a very small endothelial space. The tunica media is formed mainly by smooth muscle and has a thickness similar to the tunica adventitia. The abundant collagen and elastic fibers of the tunica adventitia is slightly invaded by muscular tissue and collagen fibers of the tunica media. 40 x.

sub endothelial space which consist of muscular fibers (non-differentiated muscular cells) cross and cut diagonally (circular disposition), and connective tissue fibers [37] (**Figure 9**).

2.3.2. *Media*

The media layer is located below to the sub endothelial space and the thickness of this layer is variable in relation to the external/adventitia in different mammalian species. Most animals display a tunica media which is smaller than tunica adventitia including species such as buffaloes, carnivores, horses, guinea pigs, nutrias, chinchillas, cavies and rock cavies [26–32]. Additionally, the South American camelids present a tunica media which is larger in dimension than the tunica adventitia [37], however, the cause of this morphologic evidence is not known.

This tunica is constituted by a double layered muscular of smooth muscle bundles, characterized for inner circular muscular layer (collagen and reticular fibers) and outer longitudinal layer (**Figure 10**). The tunica media comprises the inner circular and outer longitudinal muscular layers, with an absence of outer elastic lamina and presence of reticular fibers. Also, it presented a small amount of collagen and elastic fibers [10, 17, 23, 34–38]. Furthermore, in alpacas is known that smooth, cross-sectional muscle fibers invade part of the tunic adventitia consisting in muscular fibers cross-transverse, abundant collagen fibers, collagen and muscular fibers arranged longitudinally [37].

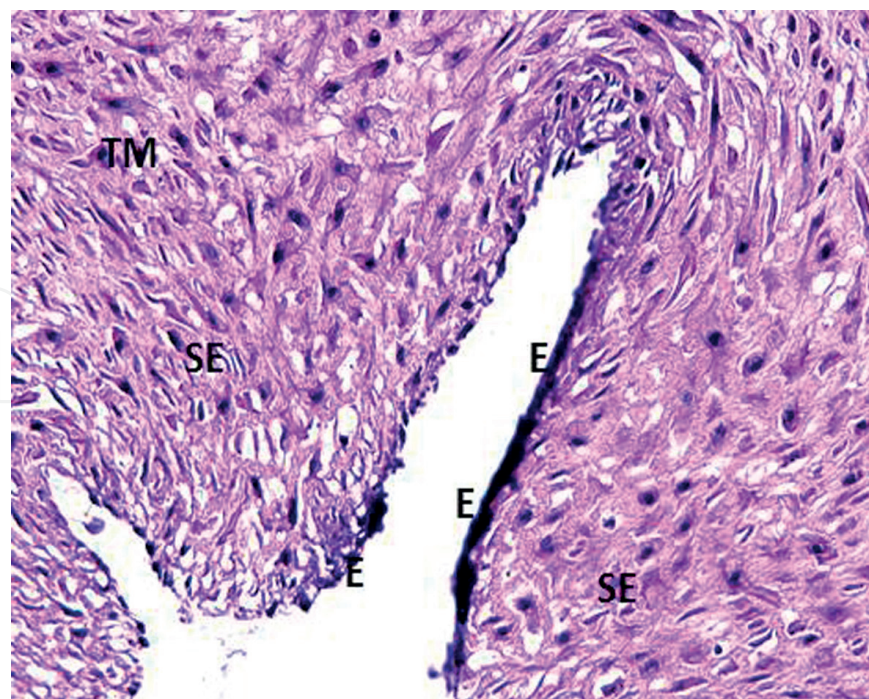


Figure 9. Umbilical vein, light microscopy, H-E stain. The tunica intima is comprised of a simple squamous epithelium (E) and a small endothelial space (SE). TM: Tunica muscular. 400 x.

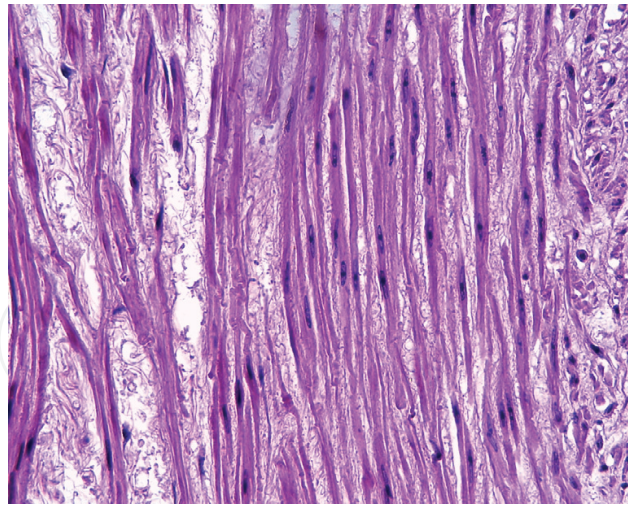


Figure 10. Umbilical vein, light microscopy, H-E stain. The tunica media is constituted by abundant muscular fibers separated by elastic and fibrous tissue. 400 x.

2.3.3. Adventitia

In the majority of mammalian species, the tunica adventitia is larger than the tunica media [26–32] and this layer consists of collagen and elastic fibers, and small blood vessels called *Vasa vasorum* (**Figure 11**). Additionally, the tunica adventitia is slightly smaller than the tunica media in alpacas [37]. Generally, this tunica not is clearly separated of the mucous connective tissue also called Wharton's Jelly, however, in species as alpacas, it is possible observe a clear separation among both structures [37]. This tunica displays variations in their thickness where most mammalian species present a great number of transversal nerves in all thickness fibers [10, 17, 23, 34–38].

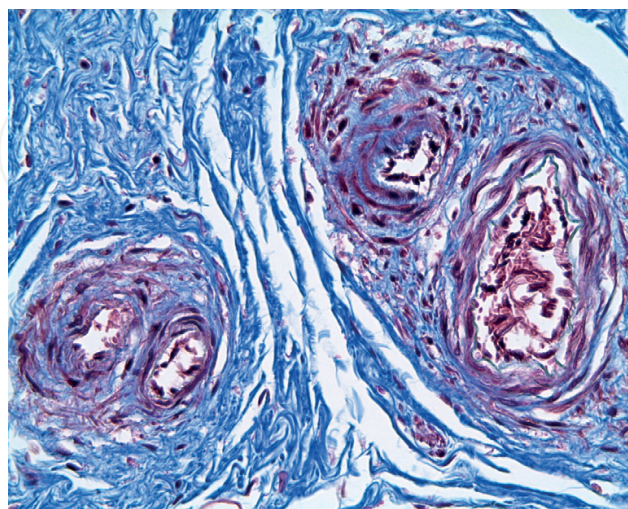


Figure 11. Umbilical vein, light microscopy, Masson's trichrome stain. The tunica adventitia/external is constituted by small blood vessels denominated *Vasa vasorum*. Note the presence abundant collagen and elastic fibers. 400 x.

2.4. Mucous connective tissue (Wharton jelly)

The mucous connective tissue surrounding the umbilical artery is almost the same in thickness and share histological features with that of umbilical vein region except that smaller blood vessels and blood capillaries are more numerous towards periphery (**Figure 12**). Moreover, small nerve bundles cut in different profiles (and some structures resembling to ganglion) have been also observed towards periphery of the mucous connective tissue [10, 17, 18, 32, 34–38].

In alpacas has been observed some larger cells that had triangular or star-shaped with less basophilic nuclei and strongly eosinophilic cytoplasm, probably mesenchymal stem cells. A few round cells with differently stained nuclei were also observed which showed similar features to lymphoid cells [37].

2.5. Allantoic duct

The allantoic duct present irregular lumen and is comprised by simple cuboidal to columnar epithelium [10, 17, 18, 32, 34–38]. In alpacas has been recognized some characteristics such as the less basophilic nuclei of varying shapes that are oriented in mid portion of the epithelium, and eosinophilic dense and finely granular cytoplasm [37].

Most animals display the outer layer consisting of band of smooth muscle bundles arranged in different directions; oblique, circular and longitudinal [10, 17, 18, 32, 34–38].

In alpacas, this structure presents abundant fine blood vessels between arterioles, venules and capillaries which have been reduced to adjacent portion of the mucous connective tissue (**Figure 13**) [37].

Umbilical cord is covered by simple squamous epithelium in all mammalian species [10, 17, 18, 32, 34–38].

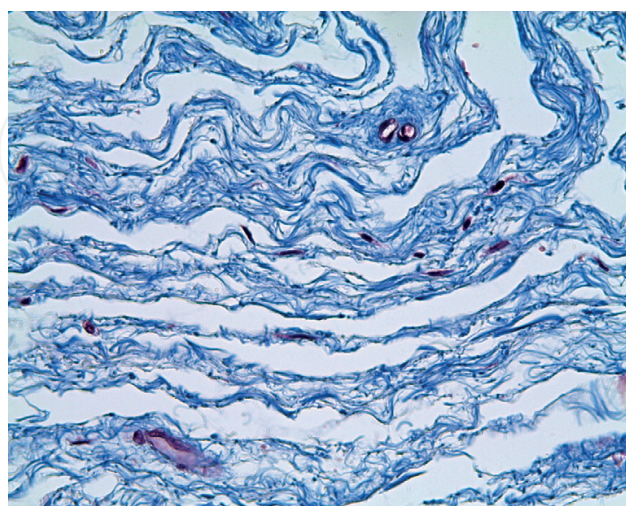


Figure 12. Mucous connective tissue, light microscopy, Masson's trichrome stain. In the most periphery part is observed great number of blood vessels that alternate with abundant reticular, elastic and collagen fibers. 400 x.

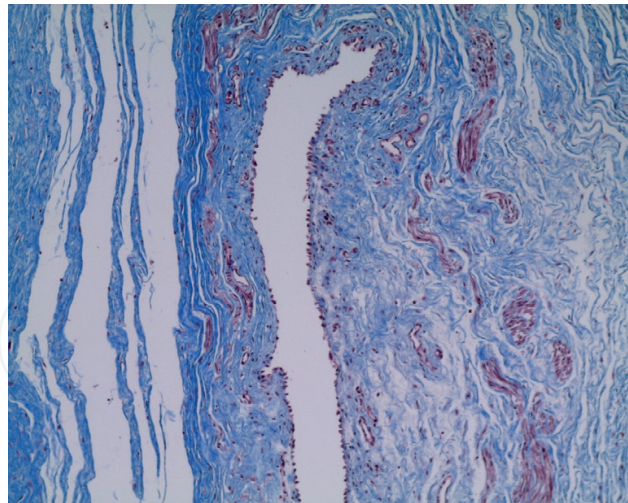


Figure 13. Allantoic duct, light microscopy, Masson's trichrome stain. Note the irregular shape lumen with plane epithelium and presence of abundant small blood vessels located near to this structure immersed in abundant connective tissue. 100 x.

Conclusions

Histology plays an important role in the characterization of umbilical cord in mammals. There are many structures which enable to be recognized through of this method and furthermore, allow compares and differentiate among different animal species. According with the presence of certain structures and characteristics that comprise the wall of arteries and veins is possible a well-characterized description of the umbilical cord in mammals.

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