

## Anatomic peculiarities of cervix uteri ligaments in pre- and postnatal human ontogenesis

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### Abstract

**Background:** Uterus and uterine cervix ligament concept is a decisive factor for female pelvis surgery and for fundamental mechanisms of urogenital dysfunction ascertainment. According to publications, there are contradictory ideas of morphologic organization of female pelvis connective tissue. From this point of view necessity arises for system investigation of connective tissue around uterine and vagina in different age periods. Target: to ascertain peculiarities of uterine cervix ligaments macro- and micro-structure at 1<sup>st</sup>, 2<sup>nd</sup> adult, elderly and senile periods of ontogenesis.

**Material and methods:** Investigations have been performed in 35 anatomical specimens. The following investigational methods have been used: macroscopy, microscopy of consecutive histological sections series, conventional and thin preparations. Statistical data processing was performed with licensed program "Statistica" use. Received indices confidence was determined with Student's t-test use. Values with  $T < 0,05$  have been taken statistically significant.

**Results:** to ascertain peculiarities of uterine cervix ligaments their macro- and micro-structure in human ontogenesis using the morphologic and statistics methods.

**Conclusions:** The ligaments of uterine cervix have specific morphologic peculiarities of their structure and topography, and they contain different constituent parts in different age periods.

**Key words:** cervix uteri, human ontogenesis, ligaments, anatomy.

### Introduction

"Uterus and uterine cervix ligament" concept is a decisive factor for female pelvis surgery and for fundamental mechanisms of urogenital dysfunction ascertainment [7, 9]. According to publications [10], there are two contradictory ideas of morphologic organization of female pelvis connective tissue. On the one hand, it is known, that pelvis connective tissue consists of anteroposterior and transversal systems. Anteroposterior system is formed by sagittal plate of dense connective tissue, expanding from pubic bones to sacral bone. Dorsal part of this system is called uterosacral or rectouterine ligament, and it has been described in detail by Campell [4]. Transversal system consists of the broad ligament, which caudal part is a so-called Mackenrodt's transverse ligament of the uterine cervix (UC) or Kocks's cardinal ligament. Other authors deny existence of ligaments, placed around uterine and vagina, at all [1]. From this point of view necessity arises for system investigation of connective tissue around uterine and vagina in different age periods.

**Target:** to ascertain peculiarities of uterine cervix ligaments macro- and micro-structure at 1<sup>st</sup>, 2<sup>nd</sup> adult, elderly and senile periods of ontogenesis.

### Material and methods

Investigations have been performed in 35 anatomical specimens. The following investigational methods have been used: macroscopy, microscopy of consecutive histological sections series, conventional and thin preparations. Statistical data processing was performed with licensed program "Statistica" use. Received indices confidence was determined with Student's t-test use. Values with  $T < 0,05$

have been taken statistically significant. Investigation has been performed according to method recommendations of "Ethic and legislative regulations and requirements compliance at scientific morphologic investigation performance" and complying to basic provisions of the World Medical Association Declaration of Helsinki on Ethical Principles for Medical Research Involving Human Subjects (1964-2000), and MoH of Ukraine Order of 23/09/2009 No 690.

### Results and discussion

In embrionic period (9-10-weeks) pelvis cavity parts are filled with undifferentiated mesenchyme. Standard methods use does not permit to differentiate genital tube and adjoining tissue. The rectum of 9 week old embryos is covered with condensed mesenchyma layer, which is later differentiated to dense connective tissue. The urinary bladder, ventrally covered with condensed mesenchyma layer, has the appearance of the semicircle with a hole, located from back-side. Lateral dilatations of the semicircle diverge from the urinary bladder wall in posterolateral direction. Nerves and vessels, entering the muscle layer of the urinary bladder, are located dorso-laterally, where the organ is covered by loose mesenchyme only. At 11<sup>th</sup> and 12<sup>th</sup> weeks of pre-fetus development mesenchyme in the pelvis region is replaced with loose connective tissue containing a large number of thin collagenic fibers. In 4-month fetuses the condensed mesenchyme layer is already presented by the dense connective tissue. At the urinary bladder cervix level it is directly linked with pubovesical ligament ventrally. Therefore, it is medially linked with parietal pelvic fascia. In 5-6-month fetuses takes place condensation of the pelvis connective tissue fibers. In 6-month fetuses the apex of the pelvis parietal fatty

layer penetrates to dense connective tissue layer plate of the urinary bladder ventrally and separates it to more thick medial and thinner lateral parts. In 5-7 fetuses differentiation of the connective tissue takes place, and it becomes possible to distinguish dense and loose connective tissue. Inside the pelvis cavity particles of fatty tissue are already formed, but differentiation and expansion of the fatty tissue is still in the process, therefore it is possible to detect clearly the organization of the pelvis connective tissue. Sacral bone does not have flexure yet, but coccyx flexure takes place in parallel to the rectum wall. Uterine body and UC in this development period are still placed in the same plane, anteflexio still exists. But due to the formed angle between vagina and UC one may say about anteversio. There is also insignificant flexure between cranial and caudal parts of vagina. Dense connective tissue is developed around uterine and vagina, and it is closely connected with subperitoneal connective tissue of rectouterine and vesicouterine recesses. At the level of these recesses there is a uterine body with wide ligaments on both sides located. Both peritoneal recesses are circularly covered with dense subperitoneal connective tissue. Thereby, ventral and dorsal subperitoneal layers of wide ligaments also consist of the dense connective tissue. Between these layers areolar connective tissue is detected, located along nerves and vessels of the uterine. Both, anterior and posterior uterine walls are covered tangentially with dense connective tissue. Subperitoneal connective tissue of the rectouterine recess consists of collagen fibers, whereas neither elastic fibers, nor smooth muscles cells have been detected. At the level of the caudal end of the rectouterine recess UC is dorsally embraced with dense connective tissue. Ventrally, loose connective tissue is located between the urinary bladder wall and UC wall. Connective tissue fibers are located tangentially along the UC posterior wall, and separate fibers are directed to its lateral wall. These fibers are also integrated into circular system and do not immediately fix the uterine to the pelvis lateral wall. Vagina posterior wall is loosely connected with rectal adventitia, its ventral wall conjugates tightly to urinary bladder and ureter walls with fibrous connective tissue. There were no definite uterine ligations detected. Adjoining to uterine and vagina connective tissue is an integral part of the neighboring structures. Rectouterine ligament (RUL) in 8-month fetuses is macroscopically visualized as a peritoneal fold only if fundus of uterus pulled off in anterior direction and fixed. Each fold connects UC with the sacral bone, being mildly fixed to the rectum medially. In histological sections RUL consisted of fatty and connective tissues, in which numerous vessels and nerve fibers have been detected. Moreover, clusters of sympathetic fibers were available in all cases, whereas there were no due ligament structures – histological signs of formed connective tissue were absent, small number of fibroblasts, fatty inclusions were available. In a newborn, fatty particles start expanding between dense connective tissue fibers of rectal adventitia. This layer is covered by the dense connective tissue plate, which may be considered a rectal fascia. It is clearly

seen on pelvis parietal fascia at the level of the pelvic floor. Rectal vessels, nerves and lymph nodes are located inside rectal adventitia.

RUL is considered an important supporting structure together with vesicouterine, round ligaments, and paracervix [8]. Autonomous nerves of small pelvis organs, responsible for viscerosensory innervation of uterus, UC, vagina, ureter, urinary bladder, and rectum, are usually located within RUL. Thereby, in our judgment, such structure, as “ligament”, is a complex, containing connective tissue, vessels, and nerves, integrated into visceral pelvic fascia (VPF).

At 1<sup>st</sup>, 2<sup>nd</sup> adult, elderly and senile ages RUL consists of two symmetric peritoneal folds along posterior uterine surface, forms curve around rectum and reaches pelvis surface of the sacral bone. RUL starts with fan-shaped fibers nearby sacral bone at the level S1-S3, sometimes at S4, proximally narrowing to UC. Two VPF folds, covering RUL behind uterine and laterally from the rectum, have been cut medially to ureters. There were lower inferior hypogastric plexus, hypogastric nerves and pelvis nerves detected between them. Hypogastric nerve fibers were located about 12,0-21,0 mm under each ureter. Hypogastric nerve is available in each side under ureter, goes posterior-anterior, top-down, and rounds RUL from the outer side. Pelvis nerves have been identified as derivatives of the third and the fourth anterior branches of sacral plexus, changing their direction down to RUL. They join, forming lower inferior hypogastric plexus in the lateral part of RUL and in the posterior layer of the broad ligament. Ureter is of great topographic significance, because it crosses lower inferior hypogastric plexus top-down, outside-inside. Distance from UC head to RUL – is 10.0-12.0 cm. RUL composition includes fatty inclusions, vessels, nerve fibers and clusters of sympathetic fibers. But there were larger amounts of fibroblasts and fatty tissue inclusions observed, compared to pre-natal ontogenesis period.

Anatomic investigation of RUL in any section reveals the availability of nerve fibers net in the connective tissue, which probably correspond to lower inferior hypogastric plexus, and it grounds the use of the term “ligamentous complex” [4, 6]. These ligaments are fixed to posterolateral UC wall at the level of the internal os. Near UC RUL have the appearance of clearly defined stripes, covered with peritoneum, but as they form in posterior direction upper border of the rectouterine pouch, they become rarer with less apparent folds of adjacent peritoneum [5]. Posterior one third of the ligament has fan-shaped form and consists of thinner fibers, fixed to presacral fascia, which is an integral part of the pelvis parietal fascia, located between fascial compartment of the rectum, upper fascia of pelvic diaphragm, and sacral bone, opposite from the lower part of the sacroiliac joint. RUL dimensions are  $6.1 \pm 0.8$  cm,  $3.1 \pm 0.4$  cm and  $2.5 \pm 0.3$  cm in areas of sacral, intermediate, and cervix parts correspondingly. In the cervix end of ligaments there is a large number of vessels, which are branches of uterine arteries and veins. In perineal one third of the

RUL the moderate number of vessels is present, and in the sacral one – still lesser number. In posterior two thirds they consist of branch anastomoses of uterine and hemorrhoidal systems. It is often difficult to identify nerve components of these ligaments macroscopically. Parasympathetic nerve fibers of 2, 3, and 4 sacral segments may be detected in intermediate and anterior one thirds of RUL. They provide parasympathetic autonomous innervation of small pelvis organs. In the location of the RUL cervix fixing, there are sympathetic fibers detected, reaching UC, which accompany uterine artery, and just they provide uterine sympathetic vegetative innervation. In the process of VPF ectomy, nerve fibers have been detected, leading from UC to the sacral bone. Anatomic and histologic peculiarities in further periods do not differ considerably; there are nerve fibers, sympathetic fibers clusters, and arterial, venous and lymph vessels inside connective tissue available too. Histologically RUL may be divided into 3 parts. Anterior or cervix department contains smooth muscles, which amount is dominating among all listed components, fibroelastic connective tissue, blood and lymph vessels, and nerves. In intermediate RUL one third dominating tissue component is connective tissue. There is a dense connective tissue layer available, located lower than peritoneum, which may be considered “a fibrous layer”. Intermediate one third consists of the connective tissue, concentrated in subserous fibrous layer, under which there is a less dense net of chaotically expanded fibers of connective tissue. Nerves elements are numerous, and blood vessels are available in moderate number. In this region smooth muscle fibers and lymph vessels are detected here and there. Posterior or sacral part is almost completely composed of loose connective tissue and fatty inclusions. Insignificant number of blood and lymph vessels is present here.

It was established that transverse cervical ligament (TCL) in the 1<sup>st</sup>, 2<sup>nd</sup> adult, elderly and senile ages is a mesenteric-like structure, anteriorly and posteriorly covered with VPF, and it is an extension of the perivascular cover of internal and external iliac arteries and veins [3]. Pelvis organs in their natural position, and without pathologies, do not have expressed structures of ligament nature in the area of TCL. TCL total length is 8.0-10.0 cm on average, it may be conditionally divided into such departments: distal (cervix) department 2.0±0.2 cm thick and 2.1±0.3 cm long; intermediate department 3.4±0.2 cm long and 1.8±0.2 cm wide; proximal (pelvis) department has triangular form in transverse incision, its length is 4.6±0.3 cm and the largest width is 2.1±0.2 cm. In the distal department TCL is fixed to the UC lateral surface. In the rear side it is connected with RUL fixing place. TCL is caudally connected with the upper fascia of the pelvis diaphragm (nearby levator ani). In the intermediate department there were observed noticeable ventral (vessel) and dorsal (nerve) areas, ureter was visualized, which along surface was crossed by uterine artery and vein; located in depth branches of uterine veins often separated

ureter from nerve structures of the dorsal part. In the proximal department fixing place of TCL to the lateral pelvis wall had triangular form, which top is the 1<sup>st</sup> diverticulum of the internal iliac artery, and basis is an upper fascia of the pelvis diaphragm. Loose connective tissue, surrounding blood vessels and pelvis nerve plexus, appears nearby hypogastric artery and occupies large anterior and medial area, reaches lateral UC surface; it contains less fat and is more compact, than the rest of the retroperitoneal loose connective tissue. This condensing is mostly expressed at lateral surfaces of UC and vagina, and expands down to the level of the pelvic floor. It cannot be separated from thinner parietal pelvis fascia, but this structure does not expand around vagina and UC. Laterally to the uterine artery vessels number grows, loose connective tissue becomes less compact and it is loosely connected with upper fascia of pelvis diaphragm by several thin fibers. While approaching the pelvis wall these tissues fan-shapely expand and continuously connect to the retroperitoneal connective tissue. With uterine pulled off to the opposite side, more differentiated tissue structure becomes evident, which may be easily distinguished from the areolar tissue, located in front of it, and from pelvis parietal fascia, located behind, around the rectum. Vessels are located in the area between two thick stripes, which pass from the lateral edges of UC and vagina to the pelvis lateral wall nearby the top of the hypogastric artery. The most part of TCL contains blood vessels (mostly veins), nerves, originating in the pelvis plexus, lymph vessels, and adjacent loose connective tissue. Paracervix may be identified as cardinal ligament due to its strength, and it surrounds so-called venous “root”, it is formed from one or more transversally oriented venous vessels, overlying one another and joining to paravisceral venous plexus into the subperitoneal vein [2]. Microscopic investigation established, that TCL main mass includes blood vessels (mostly veins), nerves of the lower inferior hypogastric plexus, lymph vessels, and areolar connective tissue, surrounding these structures. There are joints of collagen thin fibers with upper fascia of the pelvis diaphragm available. Loose connective tissue is the densest in the area, where blood vessels penetrate the fascia. Plain muscle fibers are present in blood vessels walls and adventitia composition only. There was large amount of cell elements, especially fibroblasts observed. Several isolated elastic fibers outside vessel walls have been detected. Thereby, TCL consists mostly of vessels, loose connective tissue and separate nerve fibers. It may be divided into vessel ((cranial, parametrium)) and nerve (tail, paracervix) parts. Vessel area is a prolongation of the perivascular cover of internal iliac vessels, leading to genital roots, whereas nerve part is a prolongation of the inferior hypogastric plexus.

Pubocervical ligament (anterior ligament) consists of vesicouterine fold of peritoneum, which is projected onto the urinary bladder from the anterior part of uterine, at the border of UC and body, and changes during human ontogenesis are almost absent.

### Conclusions

1. In 9-10-week embryos pelvis cavity departments are filled with undifferentiated mesenchyme. In 11-12-week pre-fetuses mesenchyme in the pelvis area is replaced by areolar connective tissue, containing large number of thin collagen fibers. In 4-month fetuses the condensed mesenchyme layer is already presented by the dense connective tissue.

2. In 5-7 month fetuses differentiation of connective tissue occurs, areolar and dense connective tissues are distinguished. Dense connective tissue develops around uterine and vagina and is tightly tied with subperitoneal connective tissue of rectouterine and vesicouterine pouches.

3. At 1<sup>st</sup>, 2<sup>nd</sup> adult, elderly and senile ages the uterine cervix transverse ligament is a mesenteric-like structure 8,0-10,0 cm long, anteriorly and posteriorly covered with visceral pelvis fascia, and contains vessels, loose connective tissue and separate nerve fibers, and has cervix, intermediate and distal departments.

4. In the cervix end of the rectouterine ligament there is a large number of vessels available, which are branches of uterine arteries and veins, it contains smooth muscles, dense connective tissue, blood and lymph vessels, and nerves; in the intermediate one third vessels are present in moderate number, main tissue component is connective tissue; and in the sacral one – even less, it consists of loose connective tissue and fatty inclusions.

5. Rectouterine ligament dimensions are  $6.1\pm 0.8$ cm,  $3.1\pm 0.4$ cm i  $2.5\pm 0.3$ cm in areas of sacral, intermediate and cervix departments correspondingly.

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