The development of muscular layers in the wall of fetal urinary bladder during the fetal period

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Özet

Fetal dönem boyunca fetal mesane duvarındaki kas tabakalarının gelişimi

Amaç: Çalışmamızda fetal dönem boyunca mesane duvarının kas tabakalarının gelişimi incelenmiştir. Gereç ve Yöntem: Bu çalışma, gestasyonel yaşları 9-40. haftalar arasında değişen 130 insan fetusu üzerinde yapıldı. İlk olarak mesane, pelvis boşluğundaki yerinden çıkarıldı. Daha sonra, mesane corpusunun ön duvarından örnekler alındı. Rutin histolojik doku takip yöntemlerinden sonra, alınan örnekler parafine gömüldü, parafin bloklardan 5 ìm'lik seri kesitler alındı ve kesitler hematoksilen-eozin ile boyandı. Işık mikroskobunda total duvar kalınlığı, longitudinal ve sirküler kas tabakalarının kalınlıkları ölçüldü. Bulgular: Elde edilen veriler değerlendirildiğinde, parametrelerde cinsler arasında fark olmadığı gözlendi ($p>0.05$). Tüm parametrelerin gestasyonel yaşla birlikte arttığı tespit edildi ($p<0.001$). 1. trimesterde vakaların %33'ü iki tabakalı kas yapısına sahip iken, %67'sinde üç tabakalı kas yapısı mevcuttu. İkinci trimesterde vakaların %86'sında, 3. trimesterde %97'sinde ve full term'de ise tamamında üç tabakalı kas yapısı gözlendi. İki tabakalı kas yapısına ait olguların yüzde dağılımı azalırken, üç tabakalı kas yapısına sahip olguların yüzde dağılımının full terme doğru arttığı belirlendi. Tartışma: Mesane duvarının kas tabakalarında fetal dönemde meydana gelen değişikliklerin bilinmesinin farklı patoloji ve anomalilerin tanımlanmasına katkıda bulunacağı düşünülmektedir.

Anahtar kelimeler: mesane, fetal gelişim, kas tabakası, insan fetusu

Abstract

Objective: The aim of our study is to investigate the development of muscular layers in the wall of urinary bladder during the fetal period. Material and Methods: We performed this study on 130 human fetuses aged between 9-40 weeks of gestation obtained with families' consent from Isparta Maternity and Children's Hospital between 1997 and 2002. We did the study at the department of anatomy in medicine faculty of Suleyman Demirel University 2004. At first, we removed the bladder from the pelvic cavity. Then, we obtained the specimens from anterior wall of body of bladder. After the routine histological procedure, we embedded the specimens in parraffine. We took serial 5 µm sections from the paraffine blocks and we stained the sections with hematoxylin-eosin. We measured the thickness of total wall, longitudinal and circular muscle layers and evaluated the development of them in the light microscope during the fetal period. Findings: There was no significant difference between sexes for the parameters ($p>0.05$). All parameters were increasing with gestational age ($p<0.001$). In the first trimester, 33% of the cases had two layered muscular structure and 67% of the cases had three layered muscle. 86% of the cases in the second trimester and 97% of the cases in the third trimester had three layered muscular structure. All cases had three layers in the full term. Conclusion: Having the knowledge of the changes in smooth muscle of urinary bladder during the fetal period will contribute to define the different pathology and anomalies.

Key words: urinary bladder, fetal development, muscular layer, human fetus

Introduction

Urinary bladder is an organ made by muscles and membranes which stores and discharge the urine filtered by kidneys for a while before it leaves the body (1).

For understanding the malformations and anomalies

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of bladder, it is necessary that embryonic myogenesis should be known well. Taking shape of urinay bladder muscular layer begins by condensation of mesenchymal tissue and first muscular layer appears at the beginning of the week 7 or at week 8 (2). Matsuno et al. (3), stated that producing and flowing of urine and mechanical extension of bladder may stimulate the myogenesis. Wu et al. (4) reported an interactive relationship between the differentiation of smooth muscle and development of epithel in rat bladder. The increase of bladder wall thickness by stress incontinans and detrusor overactivity was also shown in some previous studies and it's thougt that such kind of changes in urinary bladder may reflect different pathophysiological mechanisms (5). We couldn't find enough study related to maturation of bladder musculature during the fetal period. Therefore, we aimed to study the normal muscle development in bladder wall during the fetal period.

Material and Methods

We carried out this study on human fetuses aged between 9 and 40 weeks of gestation obtained with families' consent from Isparta Maternity and Children's Hospital between 1997 and 2002 gestation. We performed the study at the department of anatomy in medicine faculty of Suleyman Demirel University in 2004. We carried out our measurements on 130 fetuses (72 boys, 58 girls) with no external pathology or anomaly. An approval from the Ethics Board of Suleyman Demirel University Faculty of Medicine was obtained prior to the commencement of the study. We determined the gestational ages of the fetuses by using Crown-Rump Length (CRL) until week 12, and biparietal diameter, head circumference and foot length between 13 and 40 weeks (1). We assigned fetuses to one of 4 groups according to their gestational ages: Group I (1st trimester), Group 2 (2nd trimester), Group 3 (3rd trimester) and Full term (Term). We dissected the abdomens of all fetal materials by anatomical dissection method. We exposed the bladder and neighbouring structures. We didn't include the cases which had an anomaly and malformation to the study.

At first, we removed the bladder from its place in pelvic cavity. Then, we had our specimens from the anterior wall of the body of the bladder. Following the routine histological methods, we embadded the specimens into paraffine, we took serial sections using systematical random sampling and stained the sections by hematoxylin-eosin. We measured the total wall thickness, thicknesses of longitudinal and circular muscle layers and evaluated the characteristics of muscular layers during the fetal period (Figures 1-4).

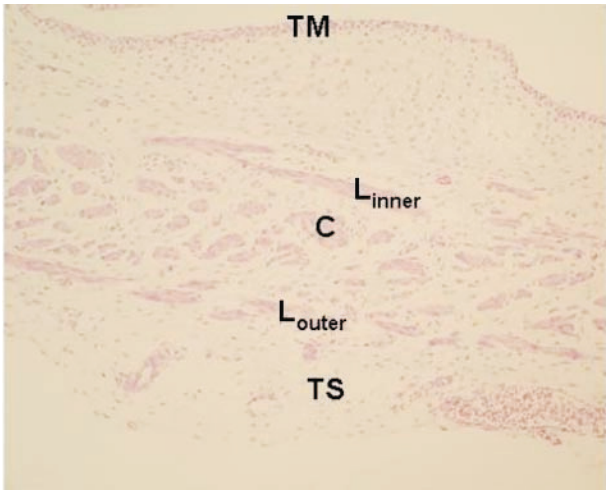

Figure 1: Muscle layer of a 12 week-old male fetus (HE, X48), TM: tunica muscularis, L_{inner}: inner longitudinal muscle layer, C: circular muscle layer, L_{outer}: outer longitudinal muscle, TS: tunica serosa

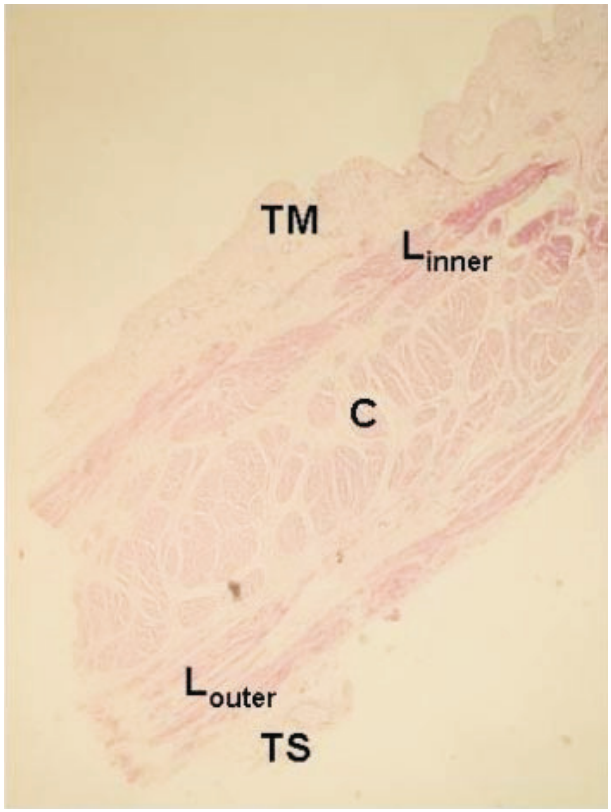

Figure 2: Muscle layer of a 22 week-old male fetus (HE, X48) TM: tunica muscularis, L_{inner}: inner longitudinal muscle layer, C: circular muscle layer, L_{outer}: outer longitudinal muscle, TS: tunica serosa

Figure 3: Muscle layer of a 33 week-old male fetus (HE, X48) TM: tunica muscularis, L_{inner}: inner longitudinal muscle layer, C: circular muscle layer, L_{outer}: outer longitudinal muscle, TS: tunica serosa

Figure 4: Muscle layer of a 40 week-old female fetus (HE, X48) TM: tunica muscularis, L_{inner}: inner longitudinal muscle layer, C: circular muscle layer, L_{outer}: outer longitudinal muscle, TS: tunica serosa

We computed the means of each parameter with respect to gestational weeks and groups using SPSS 6.0 statistical package (SPSS Inc, Chicago, Illinois, USA). Level of significance was set at 0.05. We tested the relationships between parameters using Pearson's correlation. Data with respect to weeks were expressed as means and data with respect to groups were expressed as mean ± standard deviation. We also used non-parametric tests for comparing groups due to insufficient number of cases. Significant groups according to Kruskal Wallis analysis of variance were compared pair-wise using Mann-Whitney U test. Levels of significance were assessed with Bonferroni correction. With regard to non-parametric data, chi-square test was used for comparisons of percent distributions among groups and p and ki-square values were presented in Results section and in relevant tables.

Findings

We determined the mean values of all parameters of each fetus with respect to gestational weeks. There were no differences in any of the parameters between genders ($p>0.05$). The parameters of total wall thickness, thicknesses of outer longitudinal, circular and inner longitudinal muscular layers were observed to be increased with gestational ages ($p<0.001$, Table 1).

When we assessed these parameters with respect to groups, we determined a significant difference in all parameters ($p<0.05$, Table 2).

When we evaluated the development of muscular layers with respect to groups, we observed that there was circular muscle layer in all trimesters. We couldn't observe the inner longitudinal muscle layer in 3 of 9 cases, and outer longitudinal muscle in only 1 of 9 cases in first trimester. We couldn't see both inner and outer longitudinal muscle layers in 5 of 71 cases in second trimester. In third trimester, only inner longitudinal muscle layer couldn't observed in 2 of 38 cases and in full term, all of 12 cases had both inner and outer longitudinal muscle layers (Table 3).

In our study, we were also assessed the characteristics of muscular layers. With respect to this; 33% of the cases had two layered (longitudinal+circular) muscular structure while there was three layered (inner longitudinal+circular+outer longitudinal) muscular structure in 67% of them in first trimester (Table 4). In 86% of the cases in second trimester, in 97% of the cases in third trimester and in all of the cases in full term, three layered muscular structure was observed (Table 4). Longitudinal or circular musle layers couldn't observed. In addition, the percentage distribution of the two layered cases was determined to be decreased while three layered cases was determined to be increased towards full term.

Table 1: Means of muscular layers during the fetal period with respect to gestational weeks (mm)

	Total Wall		L_{outer}		C		L_{inner}	
Age (weeks)	N	Thickness	N	Thickness	N	Thickness	N	Thickness
9	2	260,0	2	10,0	2	50,0	1	10,0
10	2	280,0	2	50,0	2	80,0	1	30,0
11	1	330,0	1	50,0	1	85,0	1	43,3
12	4	445,0	3	53,3	4	111,2	3	45,7
13	5	680,0	5	78,6	5	160,0	3	50,0
14	13	778,8	11	100,0	13	175,3	13	60,0
15	4	965,0	4	111,2	4	205,0	4	62,5
16	3	1066,6	3	115,9	3	205,8	2	77,5
17	5	1170,0	4	130,0	5	253,3	5	105,0
18	6	1181,2	5	133,7	6	256,2	6	105,0
19	5	1275,0	5	157,0	5	285,0	4	114,0
20	5	1300,0	5	158,8	5	305,0	4	137,5
21	5	1387,5	5	161,0	5	362,5	5	145,7
22	9	1415,0	8	177,5	9	381,4	9	148,7
23	5	1437,5	5	185,0	5	460,0	5	165,0
24	4	1663,7	4	199,0	4	475,0	4	166,6
25	2	1665,0	2	200,0	2	483,2	2	175,0
26	3	1711,5	3	213,3	3	486,6	3	191,6
27	4	1756,2	4	237,5	4	504,9	3	193,7
28	5	1812,5	5	241,2	5	575,0	5	212,5
29	2	1833,3	2	242,0	2	625,0	2	213,3
30	3	1916,6	3	250,0	3	641,6	3	236,6
31	5	2025,0	5	250,0	5	659,8	5	237,4
32	4	2101,8	4	266,6	4	700,0	4	252,7
33	3	2219,5	3	278,9	3	702,0	3	256,6
34	2	2266,3	2	291,6	2	749,9	2	268,2
35	2	2637,5	2	316,6	2	874,8	2	275,0
36	4	2744,4	4	316,6	4	949,6	3	300,0
37	1	3082,5	1	325,0	1	962,2	1	416,5
38	3	3095,0	3	352,7	3	979,8	3	424,9
39	2	3610,0	2	485,5	2	1041,3	2	438,7
40	7	4354,3	7	641,5	7	1228,8	7	490,2

L_{outer}: Outer longitudinal muscle layer, C: Circular muscle layer, L_{inner}: Inner longitudinal muscle layer

Table 2: Mean values and standart deviations of muscular layers with respect to groups (mm)

	Total Wall		L_{outer}		C		L_{inner}	
Group	N	Thickness	N	Thickness	N	Thickness	N	Thickness
(Trimester)								
1st trimester (9-12 wk)	9	360,0 ± 117,4	6	46,2 ± 17,6	9	91,6 ± 29,7	8	36,6 ± 15,0
2nd trimester (13-25 wk)	71	1256,0 ± 503,8	66	171,9 ± 118,8	71	308,3 ± 191,0	66	117,7 ± 73,7
3rdtrimester (26-37 wk)	38	2172,9 ± 852,9	36	251,6 ± 117,6	38	686,0 ± 419,3	38	244,8 ± 145,4
Term (38-40 wk)	12	3956,2 ± 1715,7	12	416,5± 244,8	12	1019,1 ± 469,6	12	460,9 ± 244,3
Total (9-40 wk)	130	1711,2 ± 1171,0	120	211,9 ± 155,6	130	469,3 ± 394,0	124	186,1 ± 164,6

L_{outer}: Outer longitudinal muscle layer, C: Circular muscle layer, L_{inner}: Inner longitudinal muscle layer
The difference between groups, $p<0,05$

Table 3: Case numbers and percentage distribution of the muscular layers observed in sections with respect to groups (n (%))

Gruop (Trimester)	Total Case Number (N)	L_{outer}	C	L_{inner}
1. trimester (9-12 wk)	9	8 (88,8)	9 (100)	6 (66,6)
2. trimester (13-25 wk)	71	66 (92,9)	71 (100)	66 (92,9)
3. trimester (26-37 wk)	38	38 (100)	38 (100)	36 (94,7)
Full term (38-40 wk)	12	12 (100)	12 (100)	12 (100)
Total (9-40 wk)	132	124 (93,9)	130 (98,4)	120 (90,9)

L_{outer}: Outer longitudinal muscle layer, C: Circular muscle layer, L_{inner}: Inner longitudinal muscle layer
Percent distribution is calculated along the rows.
$P>0.05$, x^2: 4.51 : No difference among groups

Table 4: Percentage distribution of case numbers of the layers with respect to groups (n (%))

Group (Trimester)	L_{outer}+C+L_{innerr}	L_{outer}	Total
1. trimester (9-12 wk)	6 (67)	3 (33)	9
2. trimester (13-25 wk)	61 (86)	10 (14)	71
3. trimester (26-37 wk)	37 (97)	1 (3)	38
Full term (38-40 wk)	12 (100)	-	12
Total (9-40 wk)	116	13	130

L_{outer}: Outer longitudinal muscle layer, C: Circular muscle layer, L_{inner}: Inner longitudinal muscle layer
$P < 0.001$, Ki-square: 61.661, the difference between groups

Discussion

In previous studies, it is shown smooth muscle of bladder at bladder dome by histological staining at the gestational week 7 (3,6,7). It is also stated that muscle cells at bladder dome are in the shape of muscle bundles and don't yet composed regular layers at gestational week 7 (3). Muscle layers are identified which distinguished from each other at the gestational week 17 in the bladder wall and the increase of volume of the circular muscle layer is found at week 21 (3).

In our study, distinguishable muscle layers were determined by week 9. When first trimester was assessed, 33% of the cases was found to be two layered and the rest of the cases (67%) was found to be three layered. In our study, formation of the smooth muscle of urinary bladder was determined beginning from first trimester. Therefore, there is no agreement between our data and Matsuno et al. (3) who observed the formation of smooth muscle layers beginning from week 17. When second and third trimesters were investigated in our study, two layered structure was observed to be changing into three layered structure quickly. Matsuno et al. (3) defined three layered structure at 21st week in the bladder wall and stated that the circular layer in the middle was well-developed. In our study, three layered muscular structure has been observed well developed by the middle of the second trimester in most of the cases. Besides in our study, circular layer was also determined well developed in this period. Therefore, our findings are similar with Matsuno et al. (3). We couldn't find any study related to full term period. All of our full term cases had three layered muscular structure. According to this data, it was determined that 86% of our cases completed its development in second trimester, and 97% of our cases completed its development in third trimester.

To conclude, we think that the changes in smooth muscle of urinary bladder during the fetal period will contribute to define the different pathology and anomalies.

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