

HISTOMORPHOMETRIC STUDY OF DUCTUS ARTERIOSUS IN FETUSES OF DIFFERENT AGE GROUP

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ABSTRACT

Ductus arteriosus (DA) from six fetuses of different gestational period (20-30 weeks) were studied microscopically to observe the gradual sequential changes that occur in the wall during the process of closure. DA was found to originate either from left pulmonary artery or from bifurcation of pulmonary trunk. Gradual increases in length, outer and inner diameter of DA were observed. Microscopically tunica media and tunica intima showed changes in the arrangement of muscle fibers, collagen and elastic fibrils. The appearance of intimal cushion due to the proliferation and migration of muscle fibers marked the occlusion process of lumen of DA.

KEY WORDS: Ductus arteriosus, tunica intima, media, adventitia, intimal cushions

INTRODUCTION

Ductus arteriosus (DA) is a shunt connecting the pulmonary artery to the arch of aorta in fetal life. It plays an essential role in protecting the non-functional lungs from overloading and allows the left ventricle to strengthen. Functional closure of DA occurs immediately after birth mediated through bradykinin released from lungs during initial inflation, a fall in prostaglandin levels and increased blood oxygen tension due to the initiation of pulmonary ventilation¹. Anatomical obliteration of DA is completed by proliferation of intimal cushion in a period of 1-3 months.

Early closure of DA resulted in right ventricular hypertrophy and congestive heart failure, while prolonged patency of DA caused left to right shunt with cardiopulmonary distress. Various factors like genetic predisposition² maternal rubella/viral infections during pregnancy³ may lead to the persistence of ductus arteriosus (PDA).

Cauqui et al. classified the DA as a muscular artery⁴. However, Holmes reported the presence of

concentrically arranged elastic laminae and fibrils in the tunica media of DA⁵. Sy Ho et.al reported gradual proliferation of tunica media and intimal mounds, mucoid filled spaces and fragmented elastic lamina⁶. The formation of intimal cushion is due to the migration of muscle cells from tunica media to sub endothelial layer of tunica intima^{7, 8}. The closure of DA begins from pulmonary artery to aorta. The thickening of intimal cushions plays a major role in the closure of ductus arteriosus⁹.

The present work is carried on to study the process of appearance of intimal mounds produced by the migration of muscle cells in the gestation period 20-40 weeks along with morphometric data like diameter, thickness of tunics and other constituents of DA layers.

MATERIALS AND METHODS

Six aborted fetuses of age group 20-40 weeks were received from the department of Obstetrics & Gynaecology after obtaining consent from the parents/relatives and clearance from Institutional ethical committee. The heart was removed along with great blood vessels and preserved in formalin. Following observation was recorded:

- Crown rump (CR) length of the fetuses
- Position of DA from pulmonary artery/trunk
- Length and patency of DA

The DA along with the connecting part of aorta and pulmonary trunk was removed and processed for routine histological study. Six micron thick sections were cut and stained with H&E and Van Giessen's stain. Using ocular scale the diameter and thickness of the tunica intima, media and adventitia were measured.

OSERVATIONS

In the Ductus arteriosus of 20 weeks male fetus, the tunica intima (TI) showed molded cushions bulging towards the lumen. The cushions were structured with

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multilayered elastic fibers and smooth muscles. Tunica media and adventitia were comparatively thin. (Figure.1) In 34 weeks fetus, tunica adventitia (TA), tunica media (TM) and intima appeared thicker. The intimal cushions were prominent and the sub endothelial tissue showed circular and longitudinally oriented muscle fibers mingled with elastic and collagen fibers (Figure.2 insert). Another added feature was the presence of intercellular spaces. Tunica media appeared thicker on one side and tunica adventitia showed vasa vasorum and nervosum (Figure.2).

A well-developed tunica intima with cushions and discontinuous internal elastic lamina was observed in 40 weeks fetus (Figure. 4). The sub endothelial connective tissue showed smooth muscles and collagen fibers. The tunica media showed muscle layer with intercellular spaces and no structural change discernable in tunica adventitia. (Figure.3)

The outer and inner diameter, and the thickness of all layers of DA were measured using ocular scale (Table 1, 2). The length was found to increase with age (2.5 – 10.33 mm). The maximum outer diameter and minimum outer diameter ranged from 1.423 – 3.385mm and 0.648 – 1.912 mm respectively depicting a correlation with age. The maximum and minimum inner diameter varied from 1.107- 2.591 mm and 9.34 -1.076 mm respectively. The thickness of TI, TM and TA ranged from 0.01 – 0.076 mm, 0.112 – 0.453 mm, 0.0275 – 0.148 mm respectively. All these measurements showed a gradual increase with fetal age.

DISCUSSION

Ductus arteriosus (DA) also called as Ductus Botalli is a shunt connecting pulmonary trunk to arch of aorta in fetal life. DA allows most of the blood from right ventricle to bypass nonfunctional fetal lung preventing it from overloading. The gradual obliteration of DA was observed to begin during gestational period of 20 weeks marked with structural changes in the walls of DA. The functional closure occurs immediately after birth under the influence of bradykinin, prostaglandin levels, oxygen tension^{10,1}. The process of closure of DA starts before birth with structural modification in the walls with formation of intimal cushion, migration of muscle layer and arrangements of collagen and elastic fibers.



Figure.1. Photomicrograph shows appearance of DA from pulmonary artery and prominent intimal cushion. The cushion mounds show elastic and muscular fibers. H&E x100

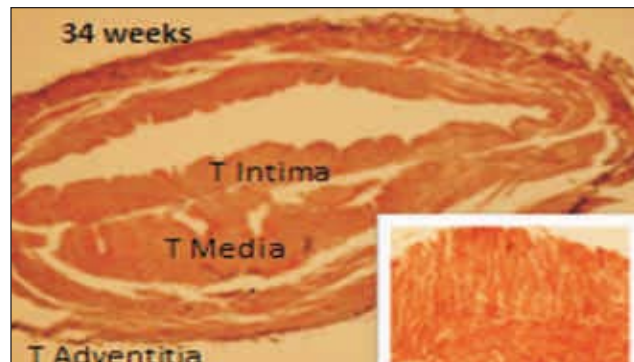


Figure.2. Photomicrograph shows DA wall thicker on one side and the longitudinal orientation of fibers with migrated muscle fibers (insert with asterisk) from tunica media. H&E x100

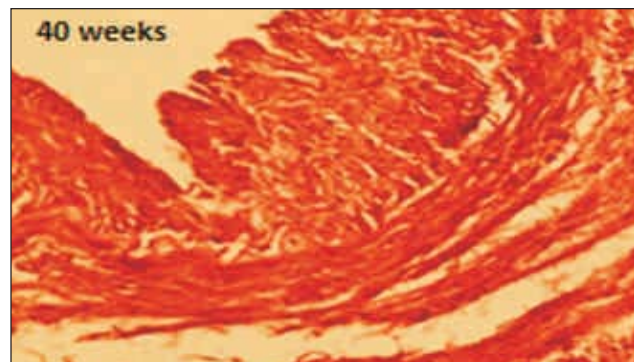


Figure.3. Photomicrograph shows sub endothelial tissue proliferation with prominent intercellular spaces. H&E x400

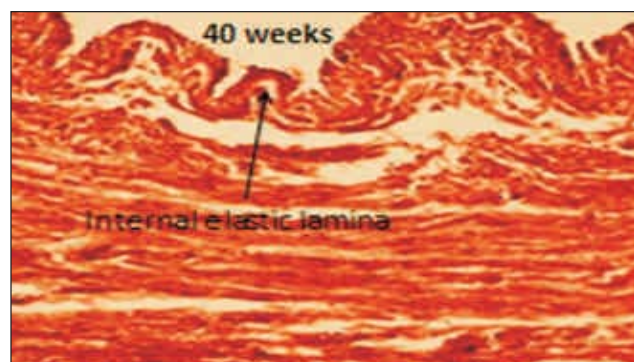


Figure. 4. Photomicrograph of DA shows an interrupted internal elastic lamina (arrow) and appearance of wide intercellular spaces. H&E x400

In the present study, six fetuses of age group 20- 40 weeks were studied to analyze the structure of DA macroscopically and microscopically. The length of DA ranged from 2.5 – 10.3 mm (Table .1) and the length increased with advancement of gestation. M.Szpinda in his study of fetuses in age group of 15 – 34 weeks observed the length ranged from 3.95 -12.2 mm. This variation in length could be regarded as regional variation¹¹.

In the present study DA showed the features of muscular artery⁷ with multilayered elastic fibers in tunica media as reported by Holmes⁵ (Figure 4). Deslingers et al and Danesino et al in their study reported the appearance of intimal cushions in tunica intima not before 24 weeks^{12,13}. The presence of intimal cushions in 20 weeks fetus in the present study confirmed the observation of Gittenberger et al⁸ who observed intimal cushions in 18 – 21 weeks fetus and Toda et al.⁷ who reported its presence in 15 weeks fetus.

In the tunica media of 20 weeks fetus, DA showed closely packed muscle bundles; and in 34 – 40 weeks , the bundles were organized as inner and outer longitudinal with circular layer in between . The thickness of DA appeared more on one side due to the proliferation and migration of muscles fibers in tunica media. Similar findings were observed by Mirza et al.⁹ (Figure.2. Insert). The thickness of DA wall was a stage, marking the beginning of gradual occlusion of lumen of DA. It was well marked in 34 – 40 weeks fetus. In addition to this, more elastic, collagen fibers and intercellular spaces were observed. These findings are in accordance with Toda et al⁷ and Mirza et al⁹. These intercellular spaces were filled with mucoid materials which were lost during the processing¹².

Tunica adventitia showed a gradual increase in thickness with collagen and elastic fibers; vasa vasorum and nerve fibers were also observed.

Szpinda measured the outer diameter of DA as 1.34- 3.49 mm in 15- 34 weeks fetus¹¹. In the present study an increase in outer diameter of DA was observed ranging from 1.43 -3.58 mm and the inner diameter varied from 1.107 -2.591 mm in 20- 40 weeks fetus showing an increase in diameter with correlation of age (Table 2). T.Schuppert stated that if the diameter of DA is more

than 9mm, it may lead to PDA postnatally¹⁴.

The thickness of TI, TM and TA (Table 2) showed a gradual increase as age advanced due to increase of muscle fibers, elastic and collagen fibers and increase of mucoid filled spaces.

CONCLUSION

Ductus arteriosus (DA) from different age group (20- 40 weeks) showed sequential structural changes like reorganization of muscle layer and appearance of lamellate elastic fibers. The length and diameters of DA showed a direct impact on the gestational age of the fetus. Marked proliferation and migration of muscles in the tunica media towards tunica intima created intimal cushions that appeared in 20 weeks fetus. The thickening of wall of DA on one side and gradual formation of intercellular spaces and presence of elastic fibers and collagen fibers increased the size of cushions that narrowed the lumen and began the process of occlusion of lumen as observed in 34 -40 weeks fetus. The complete obliteration of DA occurred postnatally.

TABLE 1 : LENGTH AND DIAMETER OF DA (N= 2×3=6)

Age of fetus (wks)	Length (mm)	Outer diameter of DA (mm)		Inner diameter of DA (mm)		Thickness Of DA wall (mm)
		maximum	minimum	maximum	Minimum	
20(2) Type equation	2.5	1.423 ±0.35 79	0.648 ±0.6 83	1.107 ±0.22 9	0.34±0.06 91	0.2395
34(2)	8.32	3.292 ±712	1.571 ±0.4 2	2.256 ±0.49 6	0.654±0.1 47	0.493
40(2)	10.33	3.585 ±0.26 5	1.912 ±0.3 78	2.591 ±0.20 4	1.076±0.1 723	0.561

TABLE 2 : THICKNESS OF TUNICS OF DA

Age of fetus (weeks)	Tunica Intima (mm)	Tunica media (mm)	Tunica adventitia (mm)
20 (2)	0.01	0.112	0.0275
34 (2)	0.01	0.336±0.06	0.03±0.005
40 (2)	0.076±0.0122	0.453±0.136	0.148±0.0768

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