

TERATOGENIC EFFECT OF ADRIAMYCIN ON SEPTAL HEART DEVELOPMENT OF EARLY FETUSES

ERKEN DÖNEM FETUSLARIN KALPLERİNDE SEPTAL GELİŞİM ÜZERİNE ADRIAMYCİNİN TERATOJENİK ETKİSİ

Harun ÜLGER¹, Saim ÖZDAMAR², Arzu YAY²

¹Department of Anatomy, University of Erciyes, Kayseri, Turkey

²Department of Histology-Embryology, University of Erciyes, Kayseri, Turkey

Abstract:

Esophageal atresia is often associated with cardiovascular and other malformations. The present study tests the heart development which may be abnormal in the rat model of esophageal atresia.

Time-mated pregnant rats received 2 mg/kg adriamycin intraperitoneally on days 6-9 of gestation. The heart malformations were investigated on day 13 of gestation under the microscope in early term fetuses.

Adriamycin caused resorption of most embryos (or early fetuses) and only seven embryos obtained from adriamycin received dams. Control fetuses had no heart malformations, whereas five of the seven fetuses exposed to adriamycin had interatrial and interventricular septal defects besides esophageal atresia.

As a result, adriamycin administration to pregnant rats, results of esophageal atresia and cardiac malformations in embryos.

Key words: Adriamycin, heart, malformation, embryo, development

INTRODUCTION

The vascular system is the earliest system to begin development in a rapidly growing embryo. The heart rudiment develops rapidly in 6-somite embryos (day 8) and the circulatory system is established by the 11-12th day of gestation in the mouse (1). This period also a period of organogenesis and any distribution of embryo by teroto-

Özet

Özofageal atrezi sıklıkla kardiyovasküler ve diğer malformasyonlar ile ilişkilidir. Bu çalışmada kalp gelişiminin sıçanlarda oluşturulan özofageal atrezi modelinde anormal olabileceği sınandı.

Zaman kontrolü yapılarak çiftleştirilen gebe sıçanlara, gebeliğin 6-9. günlerinde 2 mg/kg adriamycin intraperitoneal olarak verildi. Kalp malformasyonları 13 günlük erken dönem fetuslarda mikroskopik olarak incelendi.

Adriamycine maruz kalmış anne sıçanların embriyolarının (erken fetuslarını) çoğunda adriamycin rezorpsiyona neden olmuştu ve sadece yedi embriyo elde edilebildi. Kontrol fetuslar hiçbir kalp malformasyonuna sahip değilken adriamycine maruz kalmış yedi fetusun beşinde, özofagus atrezisi yanında interatriyal ve interventriküler septal defektler görüldü.

Sonuç olarak, gebe sıçanlara adriamycin uygulaması embriyolarda özofageal atrezi ve kalp malformasyonlarının oluşmasına sebep olur.

Anahtar kelimeler: Adriamycin, kalp, kusurluluk, embriyo, gelişim

gens can cause congenital malformation. It has been reported that more than half of babies born with esophageal atresia (EA) had associated congenital malformations and one the most frequently encountered groups of anomalies are cardiovascular among these (2,3). The molecular basis of EA and associated congenital malformations

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Corresponding Author: Dr. Harun Ülger Department of Anatomy, University of Erciyes, Kayseri, 38039, TURKEY.

Tel: + 90 352 4374901/ 23229

Fax: 00 90 352 4375285

e-mail: ulger@erciyes.edu.tr

is still far from being clear. Adriamycin (doxorubicin), a member of the anthracycline family, continues to be one of the most effective anticancer antibiotics. However, patients receiving adriamycin treatment can develop cardiomyopathy and subsequent severe congestive heart failure (4). It is also teratogenic in rats, producing vertebral, anal, cardiac, tracheal, esophageal, renal, limb and notochord anomalies and gastrointestinal atresias (5,6). However, there are not many studies on cardiac malformations in the rat model caused by adriamycin. It is possible to produce EA in fetal rats by giving 2 mg/kg intraperitoneal adriamycin on early gestational days and it could be good model to study associated congenital malformations (6-9). In the present study, septal malformations of the early foetal heart were investigated in the adriamycin-exposed pregnant rats.

MATERIALS AND METHODS

This study was approved by the Experimental Animals Ethic Committee, Erciyes University, Turkey. Time-mated female Wistar rats (weighing 200-250 g) obtained from Laboratory Animal Unit of Experimental and Clinical Research Centre, Erciyes University. Ten rats received 2 mg/kg/d adriamycin dissolved in sterile saline (0.5 mg/mL) intraperitoneally each day (one dose) from gestational day six to day nine (i.e., four injections). The control rats (n=5) received the same volume of saline injection during the corresponding period of gestation. On day 13 of gestation, the fetuses were removed, sacrificed, immersed in 10% formalin, and maintained for over night at room temperature for fixation. After fixation, the fetuses were embedded in paraffin blocks. Serial 6µ sections were cut in the transversal plane from blocks and stained hematoxylin-eosin for histologic study of interatrial and interventricular septal defects of heart development. The serial sections from control and adriamycin treated fetuses were carefully examined and photographs were taken under light microscope.

RESULTS

Results showed that each animal from control group has normal litters between 8 and 13, average 9. Embryonic development was normal in all control animals, and no malformations were seen. However, 7 of the 10 animals from adriamycin-exposed group have necrosed fetuses in the uterus. Other three animals have few survival fetuses, two animals have two and one has three litters, and other fetuses were also necrosed. The all survival fetuses from adriamycin-exposed groups had distinguishable embryonic growth

retardation. The retardation was seen especially in size of fetuses.

In contrast with control animals, which had consistently normal heart development, the adriamycin-exposed five of the seven fetuses had heart malformations with EA. The heart from control group has well developed atrioventricular septum, interatrial septum (both septum primum and septum secundum) and mostly developed interventricular septum. The hearts of the adriamycin group has interatrial and interventricular septal defects. The septum secundum was not seen and there was retardation in the interventricular septal development in adriamycin exposed embryo. Fig. 1. and Fig. 2. summarize the heart development in control and adriamycin-exposed fetuses.

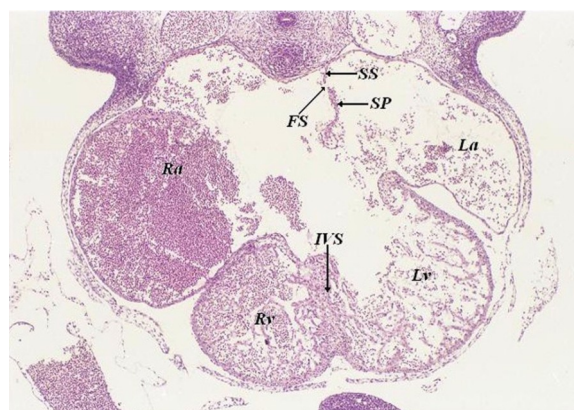


Fig. 1. Heart development in control embryo. Well-developed atrial septum secundum (SS) and septum primum (SP) with functional foramen secundum (arrowhead). Developed interventricular septum (IVS) (arrowhead). (La-left atrium, Ra- right atrium, Lv- left ventricle, Rv- right ventricle).

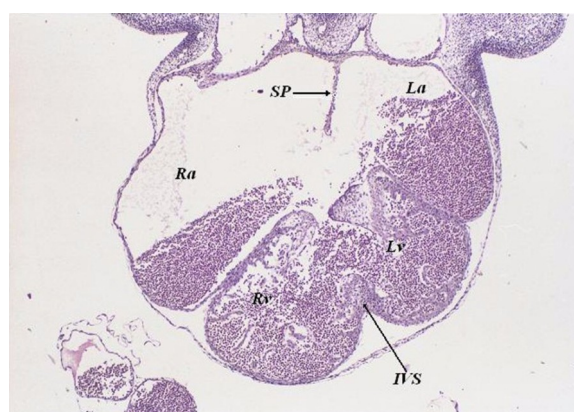


Fig. 2. Heart development in adriamycin-exposed embryo. Developed atrial septum primum (SP) and completely absent SS. Maldeveloped IVS (arrowhead). (La- left atrium, Ra- right atrium, Lv- left ventricle, Rv- right ventricle)

DISCUSSION

It has been reported that the first anomalies occurred at a dose of 1.25 mg/kg/d of adriamycin, and 100% were affected at 2.25 mg/kg/day. As the dose increased, the number of embryo resorptions increased. A dose of 2.5 mg/kg/d of adriamycin was lethal to all embryos (10). In this study, administration of 2 mg/kg/d of adriamycin (timed-pregnant rats on days 7-10) caused resorption of most embryos.

Drug-induced toxic changes in the myocardium have become an increasing problem and the effect of drugs on heart morphology may be acute or cumulative (11). Adriamycin is an anthracycline antibiotic that has been demonstrated to possess a broad spectrum of antitumorally solid tumors. Treatment with adriamycin resulted in significant augmentation of the cardiotoxic effects of this drug demonstrated by electrocardiographic measurements and myocardial histopathology (12). Its major toxicity is manifested by the depression of normal cell proliferation in the bone marrow and a delayed dose-dependent cardiac toxicity eventually resulting in congestive heart failure (13,14). A reported record of 53 patients treated with adriamycin, 17 of whom developed congestive heart failure, shows that it has risk factor for congestive heart failure (15). The deleterious effects of adriamycin in heart cells and cytoplasmic organelles were shown previously (16-18). Qi et al. (7) reported that administration of 2 mg/kg/d of adriamycin on days 6-9 caused 50% (6/12) atrial and/or ventricular septal defects in 21 days fetuses. There was 70.1% (5/7) interatrial and interventricular septal retardation in 13 days early fetuses in the present study. This differences between two studies could be restoration during the fetal development on days from 13 to 21.

Development of embryonic heart and related vessels are complex organogenetic processes and the neural crest is undoubtedly involved in these processes. Some observations suggest that the pattern of associated cardiovascular anomalies with EA may be related to an abnormal neural crest development (9,19). A recent review suggests that cardiac neural crest cells influence heart development directly and indirectly. They are essential in building the outflow septum and arch artery patterning (10).

Present and the previously available evidence suggest that adriamycin accounts for a cluster of malformations resulting from abnormal endodermal-mesenchymal interaction and disturbed somitic segmentation in fetal rats are caused by

abnormal neural crest development invite consideration of the molecular mechanisms related to EA with its associated malformations. The adriamycin rat model could be the most appropriate investigative tool for further research in this field.

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