Spontaneous Closure of Isolated Ventricular Septal Defect in the Pika (*Ochotona rufescens rufescens*)

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(Received 14 August 1985/Accepted 22 October 1985)

The incidence of spontaneously occurring ventricular septal defects (VSD) in PIKA neonates and its fate during development are investigated. A total of 160 PIKAs were used in the present study. They consisted of three groups; first, 56 live neonates, second, 37 3-week-old live animals and third, 68 animals which had died during the first two weeks after birth from unknown causes. As high as 8.9% (5 cases out of 56) of the live neonates revealed isolated ventricular septal defects of membranous type. Decreased incidence (2.7 %) was seen in 3-week-old PIKAs. Its implication and significance are evaluated in the discussion.

Puget [13] introduced Pika (*Ochotona rufescens rufescens*) for the first time as a new experimental animal. Nomura, Director of the Central Institute for Experimental Animals noticed this and obtained several pairs of this animal through the courtesy of Puget in 1974. Following the successful breeding of this animal by Matsuzaki [7], Nomura pushed forward the joint study on teratogenicity of several drugs which was conducted under the collaboration of 23 toxicological research institutes. During the progress of this study, it was revealed that in many institutes, the term fetuses of the vehicle treated control animals showed a high incidence (near 8%) of spontaneous ventricular septal defect (VSD) [8].

The purpose of the present study is to confirm whether or not such a high incidence is shown in the non-treated fetuses of this animal species and, as the supplement of the study, to examine the early postnatal history of this defect.

Materials and Methods

During the breeding of Pika in the Central

Institute for Experimental Animals of Japan as mentioned by Matsuzaki et al. [7], 3 groups of animals, that is, live neonates, 3-week-old live animals and the animals which had died during the first two weeks after birth, 160 cases in total, were singled out non-selectively from the colony and used for the present study.

The live animals were chloroformed and sacrificed. The body cavities were opened and the entire animals were fixed in 70% ethylalcohol to be examined for any defect under the binocular dissecting microscope.

For cardiovascular examination, we adopted the modification of Barrow's method as is described by Nishimura [9] since this method seemed to offer the more close view of the ventricular septum than the Wilson's method [15]. After removing the anterior chest wall, thymus, lungs and diaphragm were observed. Then, the thymus was carefully removed and attention was paid to the bifurcations of the great vessels and external view of the heart. Next, the right and left auricular appendages were cut by the ophthalmologist's scissors and the atrial septum was closely checked from

both sides. The heart was then taken out and cut horizontally by the razor blade into two pieces at the widest level of the ventricle. The left ventricular wall of the lower part was cut and the muscular septum was viewed from the left ventricular cavity. The left ventricular wall and the arotic wall of the upper part were also cut and the membranous septum was observed. To detect the presence of VSD, a human hair was inserted gently into the opening in order to confirm whether of not the opening reaches the right ventricular cavity.

Results

A. Pathology: All of VSDs found in the present study belong to isolated and membranous type. In the neonates, most of their openings were round and a little larger than

A : Aorta, P: Pulmonary trunk, V: Ventricle
Fig. 1. Isolated ventricular septal defect detected in the live neonate of Pika. Relatively large defect is situated in the middle of membranous triangle of ventricular septum (arrow). (X11).

the orifice of the coronary artery. A few cases had slit-like openings. Such defects were located just below the semilunar valve of the aorta and between the right coronary cusp and the non-coronary cusp of the valve. One case had a relatively large defect in the middle of the membranous trinagle (Fig. 1).

No significant difference in sex preponderance and body weight was noted between the neonates with VSD and those without VSD. B. Natural history of isolated VSD: Incidence of the VSD at various postnatal ages is shown in Table 1 and Fig. 2.

- Main findings shown in Table 1 are:
- 1) Incidence of VSD in live neonates was almost the same with that in the term fetuses of the vehicle-treated animals [8].
 - 2) There was no significant difference in the incidence of VSD between the live and dead neonates. Therefore, death of the newborn seems not to be associated with occurrence of VSD.
 - 3) Decreased incidence of VSD is shown in live 3-week-old Pikas compared with that of the neonates.

Discussion

VSD has been found in the term fetuses or newborns of many animal species, and it has been known to constitute an appreciable number of the cases with congenital heart disease [1]. As for their types, the highly located defects are common. According to the present study, Pika also demonstrated such pathology in accord with many other animals. The pathology and incidences of VSD in the

Table 1. Incidence of ventricular septal defects in PIKA

age (days)	No. of PIKAs	No. of dams	with VSD	incidence (%)
1— 7	90	25		
	live : 56 (32m+24f)		5 (2m+3f)	8.9
	dead : 34 (17m+17f)		3 (2m+1f)	8.8
8—14	33	unknown		
	live : 0			
	dead : 33 (18m+15f)		2 (2m)	6.1
15—21	37	6		
	live : 37 (19m+18f)		1 (1f)	2.7

VSD: ventricular septal defect m : male, f : female

CRL: Crown-Rump Length
Fig. 2. VSD (Ventricular Septal Defect) detected in PIKAs

term fetuses of various strains of rats were reviewed by Okamoto [11]. He noted that the prevalence of VSD in rats were 2.6%–7.5%, all but the Long-Evans (Olson-Goss) strain (17.3%) which was the result of selective breeding. Igarashi et al. [4] reported a paper (unpublished) that 3.2% of VSD found at term in untreated Wistar-Imamichi rats dropped to 1.5% in another untreated control group at 7 days of age. The incidence of VSD in the mouse (C57BL/c) was reported to be less than 1% [10] and that in rabbit (NZW) was 0.01% [12]. It can be concluded that compared with all the above species except the Long-Evans (Olson-Goss) rat, spontaneous occurrence of VSD in the neonates of Pika may have a prospect for production of a good animal model allowing the study on spontaneous closure of VSD during early postnatal stages.

Next, how do we explain the decrease of VSD in live 3-week-old Pikas? Two possibilities can be considered: the first is that the live neonates with VSD may die selectively during the suckling period, resulting in the sufficient elimination of VSD cases: the second is that a high number of VSD cases heal spontaneously after the neonatal stage. The first possibility is unlikely since no difference in incidence of VSD was found between live

and dead neonates in the present study. Also, the fact that in some other species such as the dog and the human, spontaneous closure of VSD in the early postnatal stage has been occasionally found, strengthening the second possibility. Examples of the spontaneous closure are as follows: Breznock [2] described two cases of spontaneous closure among 20 dogs which occurred within 15 months after successful surgical occlusion of PDA (patent ductus arteriosus) and estimated that the process of the closure could be hypertrophy of the muscle of interventricular septum. In humans, spontaneous closure of isolated VSD is now well documented [3, 14]. The authors presumed that the closure occurred by self-adherence of the tricuspid valve or by ingrowth of fibrous tissues. According to Li et al. [6], the frequency of complete postnatal closure of the ventricular septum in humans was 25%; 57% of such cases took place before 3 years of age and 89% under 8 years of age. Such examples seem to suggest that in many animal species, spontaneous closure of VSD in early postnatal stage may be found if careful examination is conducted. It can be concluded that the close consecutive postnatal observation of the heart is important in teratological study, especially when testing for reproductive hazards.

Acknowledgement

The authors would like to acknowledge Mr. K. Arishima and Mis. C. Uwabe for their technical assistance and Mr. T. Matsuzaki for supplying of Pikas respectively. The authors are also very grateful to Dr. T. Nomura of the Central Institute for Experimental Animals and Prof. O. Tanaka, Department of Anatomy, Shimane Medical University for their promotion of this investigation.

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新生仔期のナキウサギに認められた心室中隔欠損の 自然閉鎖について

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1974年に実験動物中央研究所に導入された新実験動物であるナキウサギを用いて実験奇形学的研究を進めるにあたり、先づその自然発現奇形の背景値を得る試みがなされた(1983, 西村ら)。その中で特に注目されたのは、ナキウサギにおいては心室中隔欠損が高頻度に観察されたことである。しかしながら、この値については観察を行なった研究機関によって数%から35%といった数値のばらつきがあったが、その観察法が Wilson 法によっている事もその原因の1つと考えられた。われわれは合計160匹のナキウサギを用いて、胸部内臓のより詳細な観察の可能な、1974年西村(大日本製薬)による Barrow の変法によって心臓観察を行ない、併せてその心奇形の

生後発育に伴う変化についても検討した。その結果、新生仔期には、生存例56例中5例(8.9%), 死亡例34例中3例(8.8%)に膜性心室中隔欠損が認められた。3週齢では、生存例37例中1例のみ(2.7%)に膜性心室中隔欠損が認められた。そこで、生後1週から3週までに何らかの原因によって死亡した33例を調べたところ、その2例(6.1%)に膜性中隔欠損が認められたのみであった。このことから、膜性心室中隔欠損例が特に選択的に死亡しているというより、むしろ、新生仔期に認められた高頻度の膜性心室中隔欠損の大部分が生後発育の初期の段階に自然閉鎖するのではないかと推察された。