Chapter 6

Aortic disease and recurrence of congenital heart disease in first and second-degree relatives of patients with PDA

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Abstract

Aims: Persistent ductus arteriosus (PDA) and familial thoracic aneurysm and dissection (TAAD) are genetically heterogeneous. Data on the prevalence of aortic disease including TAAD, the familial recurrence of PDA, and other congenital heart disease (CHD) provide essential information for the investigation of genetic factors of these diseases.

Methods and results: Retrospective patient-based study using a questionnaire assessing the prevalence of cardiovascular disease in family members of children with PDA. A thorough family history provided information on three generations. The data were analyzed by a binominal test. The prevalence in the general population was derived from a Dutch population-based study.
Over the last 30 years 320 patients older than 3 months with isolated PDA have been treated in our institution. Data on175 patients and their 2336 family members were available. The prevalence of aortic disease was higher in the study group compared to the general Dutch population. Aortic disease was reported in 4/590 = 0.7% of first grade relatives and 29/1339 = 2% of second grade relatives. In second grade relatives the prevalence of TAAD was increased to 0.7% (9/1339) compared to 0.016% in the general population ($p<0.001$). The recurrence rate of CHD was higher (2.7%) in first-degree relatives than in second-degree relatives (0.79%).

Conclusion: These data provide evidence for a genetic association of PDA and aortic disease. The prevalence of PDA and other CHD is increased in first grade relatives of the index patient, suggestive of common genetic factors in the determination of CHD and aortic disease.

Introduction

Isolated persistent ductus arteriosus (PDA) is a common minor congenital cardiac anomaly. The prevalence varies between about 0.8 and 10.2 per 1000 live births.[1,2] Symptoms and therapeutic consequences of the persisting fetal connection between aorta and pulmonary artery vary with patient's age and the size of the PDA. The large variability of available data is explained by the difficulty to differentiate clinically between the delayed closing ductus arteriosus (DA) and the structurally abnormal PDA in young infants, mild or absent symptoms in patients with small PDA, and the increasing survival of premature babies. The present study focuses on the families of patients treated for an isolated PDA above the age of 3 months, in whom we considered PDA as a primary congenital malformation. Nowadays, it is possible to treat most of the affected patients by catheter techniques.[3] Surgery, considered the golden standard for PDA treatment in the past, is restricted to patients who are not suitable for catheter interventional therapy. Clinical problems, caused by the left-to-right shunt between aorta and the pulmonary system, disappear after closure. Therefore, patients with isolated PDA are in general not followed after successful treatment has been performed and documented.[4]

Isolated PDA beyond the neonatal age is considered a sporadic and multifactorial disease. Examples for environmental factors include maternal exposure to the rubella virus[5], and several drugs that increase the incidence of PDA.[6-8] Bokenkamp et al.[9] reviewed a correlation of PDA with some monogenic defects, such as Char syndrome and specific genetic defects including trisomy 21, 18 and CHARGE syndrome. In addition, populations with increased parental consanguinity have been reported with autosomal recessive inheritance with variable penetrance.[10] Although familial cases of PDA are rare the recurrence rate of 5% in sibs[11] suggests a genetic component also in less consanguineous populations.

The association of thoracic aorta aneurysm and dissection (TAAD) with PDA has been documented in two families carrying mutations in genes encoding smooth muscle cell (SMC)- specific beta-myosin (*MYH11*)[12] or aortic smooth muscle alpha-actin (*ACTA2*)[13]. This suggests that unimpaired SMC contractility is a prerequisite for proper DA remodeling in addition to its critical role in maintaining the structural integrity of the aorta.[14] Recently it has been documented that *ACTA2* mutations predispose patients to a variety of diffuse and diverse vascular diseases, including TAAD, premature coronary artery disease, ischemic strokes, and Moyamoya disease.[15] Whether other cases of familial TAAD and PDA,[16] can be attributed to

anomalies in contractile proteins is unclear. Considering the embryonic processes responsible for the formation of vessel walls in general and the DA in particular, all mutations impairing endothelial (EC) and SMC function as well as elastogenesis might be responsible for PDA and TAAD. DA closure and normal aortic distensibility both depend on proper interaction between EC and SMC and involve turnover and repair of extra cellular matrix substances.[17]
Different from earlier studies on the familial segregation of vascular diseases in families with TAAD mutations[12,13] we investigated whether aortic pathology, specifically TAAD, occurs more frequently in families of children with isolated PDA of unknown origin. In addition to the determination of the prevalence of aortic disease in these families we aimed to study the familial recurrence of PDA and other types of CHD in first and second-degree relatives of patients with PDA.

Methods

Study population

For this study 322 patients treated for PDA after 3 months of age have been identified from the patient databases of the Centre of Anomalies of the Heart Amsterdam/Leiden (CAHAL) covering a time-span of 30 years.
The 322 patients, or in case of patients younger than 16 years, their parents or guardians, were sent a questionnaire appropriate to the patient's age and familial situation. (Supplement questionnaire) 175 questionnaires were returned resulting in a response rate 54%. These index patients provided information spanning 2336 family members with a mean of 14.3 family members per patient. Data from 634 first-degree relatives (parent, sibling or offspring) and 1390 second degree relatives (grandparent, uncle, aunt, nephew, niece, half-sibling) were included in our study.

Recorded data included age, sex, cardiovascular morbidity and mortality and general mortality in the families. All cardiovascular interventions and a positive family history for connective tissue disorders were noted. Aortic disease was concluded from answers that were suggestive for any type of aortic disease including aortic aneurysms, acute or chronic dissections, aortic root or valve disease. A more detailed description of the aortic disease including "ruptured aorta" or "dilated aorta" was necessary to conclude that a relative had TAAD. The recurrence rate of PDA and CHD in general was analyzed in the cohort of first-

degree relatives. The recurrence rate was defined as percentage of patients affected related to the number of index cases. The family history was part of a more extended study on the elastic properties of the aorta in PDA patients and had been approved by the medical ethical board of the LUMC.

Statistics

SPSS 19.0 was used for analysis of the family history data. The prevalence of TAA was based on data describing a prevalence of 0.0016 in the Dutch population.[18,19] A Wald test was used to compare the proportions of patients and family members with aortic disease to the prevalence in the Dutch population. To take into account a possible dependence within families, robust standard errors were computed by using the generalized estimating equations module in SPSS and assuming an independence working correlation. A p-value <0.05 was considered as statistically significant. Cardiovascular morbidity and mortality data were compared to data published for the Dutch population in the same study period.[20]

Results

Baseline characteristics of the patients are summarized in (Table 6.1). Our study cohort consisting of 175 patients who responded to the questionnaire showed a male preponderance (male to female ratio of 1.76). The majority of the patients (142, 81.1%) were treated by catheter techniques. Treatment for PDA took place at a mean age of 2.8 (0.3-46.5) years.

Table 6.1 Baseline characteristic of the study population.

Characteristics	Total population	PDA patients
Number of patients	2511	175
Male, n (%)	1020 (40.6)	111 (63.1)
Female, n (%)	1052 (41.9)	63 (35.8)
Unknown, n (%)	439 (17.5)	1 (1.1)
mean age at inclusion (in years)	42.8 (0-110)	12.0 (1-50
mean age at PDA treatment (in years)	n.a.	2.8 (0.3-46.5)
PDA surgery, n (%)	n.a.	29 (16.6)
PDA catheter treatment, n (%)	n.a.	142 (81.1)
PDA treatment unknown,n (%)	n.a.	4 (2.3)

n.a. = not applicable

The distribution of aortic disease among the PDA patients and relatives is depicted in Table 6.2A and 6.2B. Thirty-six relatives of PDA patients fulfilled our definition of

aortic disease (Table 6.2A). In 11 out of these 36 relatives we concluded that the aortic disease was TAAD. According to the binominal test for proportions TAAD was significantly more frequent in second- degree relatives of PDA patients than in the Dutch general population. Results are provided in Table 6.2B.

Table 6.2 Distribution of aortic disease in the PDA patient group and their relatives.

A

Degree of relationship	Aortic disease	n	Observed Prop.	Test Prop.	p
PDA patients	no	113	0.974	0.998	0.002
	yes	3	0.026		
	total	116	1.0		
First-degree relatives	no	586	0.993	0.998	0.045
	yes	4	0.007		
	total	590	1.0		
Second-degree relatives	no	1309	0.978	0.998	<0.001
	yes	30	0.002		
	total	1339	1.0		

B

Degree of relationship	TAAD	n	Observed Prop.	Test Prop.	p
PDA patients	no	116	1.0	0.998	0.793
	yes	0	0		
	total	116	1.0		
First-degree relatives	no	588	0.997	0.998	0.454
	yes	2	0.003		
	total	590	1.0		
Second-degree relatives	no	1330	0.993	0.998	<0.001
	yes	9	0.007		
	total	1339	1.0		

Observed Prop. = observed proportion of absence (no) or presence (yes) of aortic pathology or of TAAD in the study group, Test Prop. = test proportion absence of aortic pathology or absence of TAAD in the Dutch population, p = p-value of the binominal test

We observed an increased prevalence of CHD in first and second-degree relatives of patients with PDA (2.73% resp. 0.79%) as compared to the Dutch population exhibiting 0. 43% CHD (confidence interval 0. 35 and 0.54). The frequency and the type of congenital heart disease (CHD) among relatives of PDA patients are summarized (Table 6.3). CHD was present in 17 (2.73%) first-degree relatives, including isolated PDA in 9 and the combination of PDA with atrial septal defect (ASD), ventricular septal defect (VSD) or arrhythmia in 5. In addition, 3 other first-degree relatives had non-specified valve anomalies (2) and non-specified septal defect (1). Eleven (0.79%) second-degree relatives were diagnosed with CHD, among those were VSD in 2, valve anomalies in 7 and transposition of the great arteries in 2 cases.

Table 6.3 Frequency and the type of congenital heart disease (CHD) among relatives of PDA patients.

Degree of relationship	Diagnosis	n	Percentage (%)
First-degree relatives	PDA, isolated	9	1.4
	PDA + ASD	2	0.3
	PDA + VSD	2	0.3
	ASD of VSD	1	0.2
	PDA + arrhythmia	1	0.2
	valve anomaly	2	0.3
	PDA total	14	2.2
	all anomalies	17	2.7
Second-degree relatives	VSD	1	0.1
	TGA	2	0.1
	valve anomaly	7	0.5
	all anomalies	10	0.7

ASD = atrial septal defect, VSD = ventricular septal defect, TGA = transposition of the great arteries

The familial recurrence of CHD was further analyzed in the cohort of first-degree relatives. The recurrence rate of CHD in general was 9.7%. Isolated PDA recurred in 9/17 first-degree relatives conform to a recurrence rate of 5%. As all 14 cases with PDA, regardless whether the PDA was isolated or associated with other lesions, the recurrence rate of PDA was 8%. Compared to the general population exhibiting a frequency of PDA of 0.05%.[21] PDA was more frequent in first degree-relatives of PDA patients (1.4% isolated PDA and 2.2% all types of PDA) as extrapolated from our data.

Even higher than the direct concordance of 52% (9/17) (i.e. another family member with an isolated PDA) is the partial concordance for CHD (i.e. another family member with a CHD) of 82% (14/17) in first degree-relatives. In the group of second-degree relatives no PDA was reported. 2/10 individuals with various CHD were born with a transposition of the great arteries, which is more than the expected birth prevalence of 0.2/1000.[22] One index patient was exposed to maternal rubella infection. Apart from one case of trisomy 21 no other genetic syndromes were diagnosed in index patients. Out of the 18 deaths in the group of 626 first-degree relatives 2 were reported as a consequence of CHD in infancy. Among the 283 deaths of second-degree relatives just one was related to CHD in an infant.

We analyzed the data on the frequency of and mortality due to CVDs to get insight in the total prevalence of cardiovascular diseases (CVDs) according to the definition of the WHO. This includes coronary, cerebrovascular, peripherial arterial,

rheumatic, congenital disease and deep vein thrombosis and pulmonary embolism in families of PDA patients. Among the first degree-relatives 42/553 (6.6%) had CVDs. The mortality due to CVDs was 0.2%. As expected among second-degree relatives the prevalence 172/1174 (12.4%) and the mortality due to CVDs (2%) was higher, most probably due to the increased age of these individuals at inclusion. With respect to coronary disease, we did not find a higher prevalence in relatives of PDA patients than in the general Dutch population. Table 6.4 provides an overview of the distribution of coronary disease in comparison to the prevalence in the population as provided by the Netherlands Heart Foundation.[20]

Table 6.4 Overview over the frequency of coronary disease in the study population compared to the prevalence in the Dutch population.

Relation to the indexpt.	Mean age (y)	Coronary disease (n)	Coronary disease (%)	Prevalence
Maternal grandmother	67.0	10	5.7	86.97/1000
Maternal grandfather	66.4	24	13.7	192.06/1000
Paternal grandmother	69.9	4	2.3	126.06/1000
Paternal grandfather	69.4	16	9.1	192.6/1000
Mother	41.5	0	0	4.45/1000
Father	44.9	2	1.1	27.29/1000
Sister	16.0	0	0	0/1000
Brother	13.7	0	0	0.3/1000
Maternal aunt	39.9	0	0	4.54/1000
Maternal uncle	37.9	2	1.2	3.97/1000
Paternal aunt	40.7	0	0	4.54/1000
Paternal uncle	42.8	0	0	8.1/1000
Child	5.3	0	0	0/1000

Indexpt. = indexpatient, y=years, prevalence = prevalence in the Dutch population after.[20]

Discussion

The most important result of our study is that aortic disease and especially TAAD occurs more frequently in relatives of patients with PDA. As expected from data on familial patterns of TAAD[23] significantly more cases than in the general population were found in the cohort of second-degree relatives, which included the generation of the grandparents whose mean age ranged between 66.4 and 69.9 years. The asoociation of PDA and TAAD indicates a common molecular mechanism that might be responsible for both diseases. This might put PDA patients, usually treated in childhood, and their families at a higher risk to develop aortic disease later in life.

As DA closure and normal aortic distensibility both depend on proper interaction between EC and SMC involving turnover and repair of extra cellular matrix

substances[17], all mutations impairing EC and SMC function and elastogenesis are candidate culprits. Only a small number of mutations affecting contractile proteins have been documented in studies on the familial segregation of vascular diseases in families with TAAD mutations.[13,24] To our knowledge studies taking the PDA-patient as starting-point to study the incidence of aortic disease and the underlying genetic background have not been published yet.

Various mutations under which *MYH 11* and *ACTA 2* mutations might link PDA with TAAD or aortic disease in our study. Defects in the transcription factor myocardin diminish the contractile function of vascular SMC and associate PDA with aortic aneurysms in animal models.[25,26] Mutations in *TFAP2B* impair the differentiation of SMC during fetal development[27] and are responsible for PDA in Char syndrome,[28] although aortic aneurysms have not been described. The lack of aortic disease in Char syndrome could be explained by a specific spatial overexpression of *Tfap2B i*n the DA as documented in rats (data submitted).

Aortic aneurysms and dissections are the leading clinical problem in genetic syndromes impairing elastogenesis such as Marfan and Loeys-Dietz syndrome (LDS). As the loss of intimate spatial association between elastin deposits and vascular smooth muscle cells is the histological hallmark of LDS it is plausible that patients with LDS show an increased incidence of PDA.[29] In the Brown-Norway rat small PDA and aortic fragility including aneurysms is related to an abnormal distribution of elastic fibers and diminished elastin content.[30-32] In our study population only one family member, but no index patient, was diagnosed with Marfan syndrome and Char syndrome has not been reported. In view of the increased incidence of aortic disease and TAAD in families with PDA we advocate a thorough family history in all PDA patients. In selected cases genetic testing will be needed to define the underlying genetic abnormality in a family.

The high prevalence of CHD (2.7% among first-degree relatives of patients with PDA) corroborates larger population–based studies[33-35] and is comparable to the recurrence rate of other CHD in first degree-relatives detected by detailed fetal echocardiography.[36] In 5% of our index patients a first-degree relative was diagnosed with an isolated PDA. This recurrence rate is in agreement with other studies[11,37] and supports the idea of a genetic component in the pathogenesis of isolated non-syndromic PDA.[10] Our study documents a relatively high complete (52%) and partial (82%) concordance for recurrent PDA in first-degree relatives. The degree of concordance for various types of CHD varies between the different studies. So is the concordance for PDA higher than for other CHD based on fetal echoscreening[38] or at the same level as described by others.[36,39] Despite the

relatively high concordance for recurrent PDA in our study, a familial association of PDA with more complex CHD was also present. In this point our data support the international guidelines that recommend a fetal echocardiographic study in all first degree-relatives of patients with PDA. When counseling relatives about prenatal diagnostic tests it must be realized that the DA is a normal fetal structure. An isolated PDA cannot be detected on a fetal echocardiogram. The recurrence rate of 5% and the relatively high complete and partial concordance compared to other lesions justifies a postnatal clinical and echocardiographic investigation in addition to the fetal examination. In second-degree relatives we found an increased frequency of valve anomalies. This observation was made in the older cohort of second –degree relatives and has two potentially interconnected reasons. In the first place the higher frequency might reflect the usually late clinical manifestation and diagnosis of minor structural aortic valve anomalies such as bicuspid aortic valves. Secondly, the valve anomaly might be related to the dilatation of the ascending aorta in patients with aortic disease, which is also more frequent at an older age. We are aware of the limitations inherent to survey research i.e. low response rate and the lack of details in the diagnosis data.[40] The response rate in our study was 54%, which is more than expected from a mailed questionnaire.[40] According to the answers we categorized the diseases into CHD and other CVDs. While aortic disease, PDA and CHD were overrepresented in our sample, coronary disease was not more frequent and did not show an earlier onset in our study population as compared to the general population. As assessed by the frequency of coronary disease the study population was a representative sample of the general Dutch population. The premature onset of coronary disease, which might occur with *ACTA2* mutations,[15] was not observed.

In conclusion our data provide evidence for an association of isolated PDA and aortic disease in members of the same family. This links a minor cardiac anomaly usually treated in childhood to serious aortic disease that will manifest later in life. In addition we showed that the prevalence of PDA and other more complex CHD is increased in first-grade relatives of the index patient. This suggests common genetic factors in the determination of CHD and aortic disease in these families. Moreover, searching specifically for mutations in these families could help to identify new genes that may contribute to PDA, other CHD and cause aortic disease.

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Appendix

Supplements

Supplement 1

Questionnaire for patients of 16 years and older:

1. Are your grandparents alive?
2. What is the age of your grandparents? / At what age have your grandparents died?
3. Has or had one or more of your grandparents a heart or vessel disease?
4. If yes, who has/had what kind of heart or vessel disease?
5. Has or had one or more of your parents been operated on his/her heart or vessels?
6. If yes, who had which kind of operation?
7. Has or had one or more of your grandparents a disease of the aorta?
8. Is your father alive?
9. How old is your father or how old was your father when he died?
10. Has or had your father heart or vessel disease?
11. If yes, which kind of heart or vessel disease?
12. Has your father been operated on his heart or vessels?
13. Has or had your father a disease of the aorta?
14. Is your mother alive?
15. How old is your mother or how old was your mother when she died?
16. Has or had your mother heart or vessel disease?
17. If yes, which kind of heart or vessel disease?
18. Has your mother been operated on her heart or vessels?
19. Has or had your mother a disease of the aorta?
20. Do you have brothers or sisters?
21. How many brothers and sisters?
22. How old are your brothers and sisters?
23. Are all your brothers and sisters alive?
24. What was the cause of death?
25. At what age have they died?
26. Has of had one of your brothers or sisters a heart or vessel disease?
27. What kind of heart or vessel disease?
28. Has one of your brothers or sisters been operated on his/her heart or vessels?
29. Has one of your brothers or sisters a disease of the aorta?
30. Do you have children?
31. If yes, how many children do you have? (How many boys how many girls?)
32. How old are your children?
33. Are all of them alive?
34. If not, what was the cause of death?
35. At what age have they died?
36. Has or had one of your children a heart or vessel disease?
37. If yes, which kind of heart and vessel disease?
38. Has or had one of your children a disease of the aorta?
39. Has or had one of your family members a connective tissue disease such as Marfan, Ehlers Danlos syndrome or does one have the syndrome of Char?
40. Does one of your family members sleepwalk?

Supplement 2

1. Questionnaire for mother/father of an index-patient:
2. Are your parents alive?
3. What is the age of your parents? / At what age have your parents died?
4. Has of had one of your parents a heart or vessel disease?
5. If yes, what kind of heart or vessel disease?
6. Has one of your parents been operated on his/her heart or vessels?
7. Has one of your parents a disease of the aorta?
8. Do you have brothers or sisters?
9. How many brothers and sisters?
10. How old are your brothers and sisters?
11. Are all your brothers and sisters alive?
12. What was the cause of death?
13. At what age have they died?
14. Has of had one of your brothers or sisters a heart or vessel disease?
15. What kind of heart or vessel disease?
16. Has one of your brothers or sisters been operated on his/her heart or vessels?
17. Has one of your brothers or sisters a disease of the aorta?
18. Do you have a heart or vessel disease?
19. What kind of heart or vessel disease?
20. How old are you?
21. Have you been operated on your heart or vessels?
22. If yes, what kind of operation?
23. Do you have a disease of the aorta?
24. How many children do you have? How many boys? How many girls?
25. What is the age of your children?
26. Are all your children alive?
27. If not, what was the cause of death?
28. How old was the child that has died?
29. Has one or have more of your children a heart of vessel disease?
30. If yes, what kind of heart or vessel disease?
31. Has one or have more of your children a disease of the aorta?
32. Has or had one of your family members a connective tissue disease such as Marfan, Ehlers Danlos syndrome or does one have the syndrome of Char?
33. Does one of your family members sleepwalk?