

Renal FMD may not confer a familial hypertensive risk nor is it caused by *ACTA2* mutations

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Abstract Renal fibromuscular dysplasia (FMD) can cause hypertension, and previous reports suggest that FMD is familial. We hypothesized that, in families containing an individual with proven FMD, relatives of index cases would have an increased risk of hypertension. *ACTA2* mutations cause a spectrum of extra-renal arteriopathy, leading to our second hypothesis that mutations are implicated in FMD. The blood pressure of first-degree relatives was measured using standard devices and, when indicated, with 24-h ambulatory monitoring. Leucocyte DNA was obtained from FMD index cases and *ACTA2* sequenced. Thirteen unrelated index cases, aged 2–32 (median 15) years, were recruited. Blood pressure was

assessed in 40 first-degree relatives, comprising 22 parents aged 28–58 (median 44) years and 18 siblings aged 3–30 (median 13) years. Hypertension was evident in six (27%) parents but in none of the eight adult siblings. Of the ten screened siblings aged less than 18 years, one teenager was pre-hypertensive (90th–95th centile), the remainder being normotensive. No *ACTA2* mutations were found in 13 index cases. Hypertension was evident in 20% of all assessed adult first-degree relatives and is therefore not increased relative to 25% of the adult population. Although hypertensive parents did not undergo angiography to assign FMD status, this observation, together with the lack of hypertension in 18 siblings, indicates that FMD is unlikely to confer an excess hypertension risk in first-degree relatives up to middle-age. Furthermore, in our cohort, FMD was not caused by *ACTA2* mutations.

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Introduction

Although primary hypertension can occur in children, raised blood pressure before adulthood is often secondary to endocrine (e.g. adrenal tumors), renal parenchymal or renovascular disease [1]. In the latter category, some children with renovascular hypertension (RVH) have defined multi-organ, genetic disorders, such as neurofibromatosis type 1 and Williams–Beuren syndrome [2, 3]. The commonest cause of RVH in children is, however, renal artery stenosis (RAS) caused by fibromuscular dysplasia (FMD), a defect of the arterial wall sometimes also found in extra-renal vessels such as internal carotid arteries and the aorta [4, 5].

Renal FMD itself is likely to be a heterogeneous condition, but the most common lesion, medial fibroplasia, is characterized by a “string of beads” appearance on angiography [6]. FMD lesions are generally not inflammatory, nor are they atheromatous [6], so FMD could be envisaged as a primary disorder of arterial differentiation and/or maturation. The pathogenesis of FMD is unknown, but one adult study found that smoking in index cases was a risk factor [7]. However, this environmental factor seems unlikely to play an important role in children with FMD. Furthermore, given that FMD is sometimes diagnosed in very young children, [5] some of the initial lesions may be congenital, and therefore developmental defects.

A unique family has been described with four of nine siblings affected by hypertension and FMD, associated with congenital digital and cardiac anomalies [8]. Although the gene (or genes) causing this syndrome has yet to be reported, the publication does raise the prospect that non-syndromic forms of FMD may be familial and have genetic bases. In a report from the USA, Rushton described 20 families, each with an index case with FMD [9]. It was noted that 12 of the families “contained between one and 11 other relatives who appeared to have FMD”, and it was concluded that the inheritance pattern was most consistent with an autosomal dominant trait with variable penetrance.

Recently, it has been shown that mutations in *ACTA2*, which codes for smooth muscle cell α -actin, a major structural protein in vascular muscle, cause a wide spectrum of diseases, including sporadic and familial thoracic aneurysms, as well as premature coronary artery disease and ischemic strokes [10]. This protein is also expressed in the developing renal arterial tree [11].

Therefore, renal arterial FMD can cause hypertension, and FMD may run in families. We hypothesized that, in kindreds containing an individual with proven FMD, relatives of index cases would have an increased risk of hypertension. Furthermore, because *ACTA2* mutations cause a spectrum of extra-renal arteriopathy, we hypothesized that mutations are also implicated in FMD.

Methods

The family blood pressure and the genetic arms of the study were approved by the Local Ethics Committee, and individuals were recruited and investigated after appropriate consent/assent. RVH index cases were identified through searches in both the Paediatric Nephrology and the Renovascular databases held at Great Ormond Street Hospital for Children NHS Trust. Clinical data were obtained and reviewed in patients, including those who had been transitioned to the care of adult nephrologists and/or primary care physicians. Of 28 children confirmed to have

RVH on angiography, seven were excluded from this study of non-syndromic FMD because they had neurofibromatosis type 1 ($n=5$) or Williams–Beuren syndrome ($n=2$). A further eight index cases declined entry to the study or could not be contacted. Therefore, 13 index cases and their families were recruited to this study; each index case came from a separate family.

Family histories were obtained of all first-degree relatives of index cases. Parents and siblings had their blood pressure recorded in their homes by a research nurse (AG) trained in manual blood pressure measurements. Three readings were obtained with a suitable-sized cuff after 15 min of rest. For relatives under 18 years of age, hypertension was defined as blood pressure >95th centile appropriate for age, sex and height centile [12]. For older individuals, hypertension was defined as blood pressure measurements >140/90 mmHg (National Institute of Health and Clinical Excellence clinical guideline endorsed by the British Hypertension Society; <http://www.nice.org.uk/CG034>). A diagnosis of hypertension was also accepted if a relative had already been diagnosed as having this condition and was being treated with anti-hypertensive medications. Where relatives had borderline high blood pressures or noted to be hypertensive, 24-h ambulatory blood pressure monitoring was undertaken prior to referral back to the relative’s general practitioner.

The *ACTA2* gene (locus NM_001613) has two variants; these differ only in the 5’ untranslated region (UTR) and encode the same protein. *ACTA2* has an 1134-bp coding sequence consisting of nine exons, of which eight are coding, resulting in a 377-amino acid protein. In this study, all exons including splice sites were sequenced in all 13 index FMD patients. Genomic DNA extracted from leucocytes were subjected to PCR amplification using exon-specific primers (available upon request). PCR products were analysed using standard procedures, as previously described [13].

Results

Figure 1 depicts the 13 unrelated index cases, of whom eight (62%) were female, with their respective family trees (RASFMD1–13). All families were Caucasian, with the exception of two Asian families where there was consanguinity with the parents being first cousins (RASFMD7 and -10). The index cases were aged 0.03–14 (median 7) years and 2–32 (median 15) years at the time of clinical presentation and study entry, respectively. All were hypertensive at presentation, and the other clinical details are outlined in Table 1. Around one-half of the cohort had central nervous system manifestations, with cardiac failure being the second most common mode of presentation. Eight

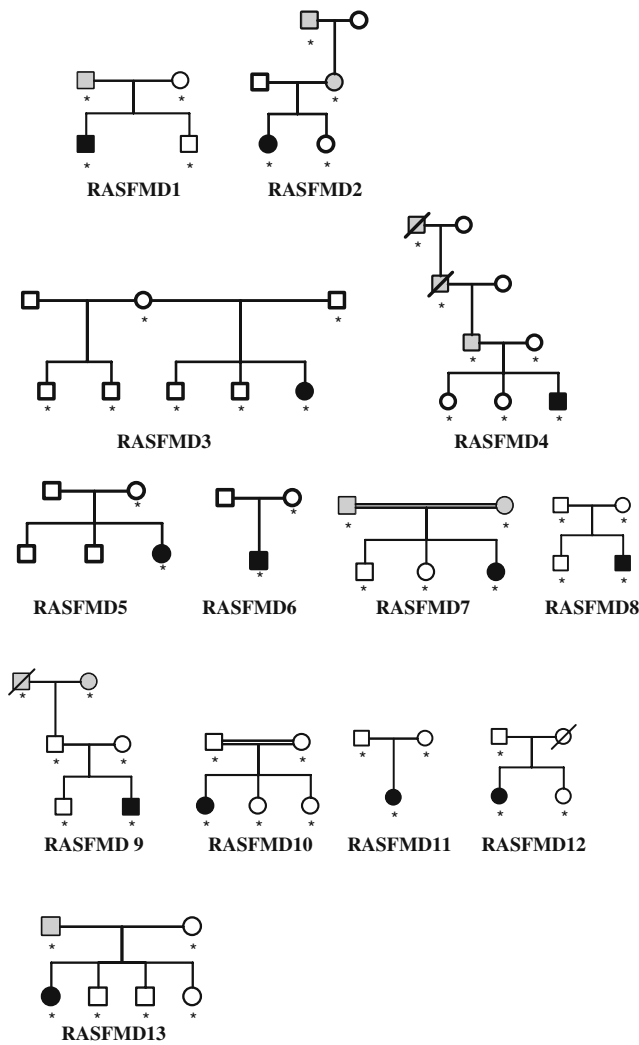


Fig. 1 Family trees of the 13 index cases with fibromuscular dysplasia. *Black-shaded icons* Index cases, *grey-shaded icons* individuals with hypertension. *Asterisk below each icon* indicates that either the blood pressure was assessed in this study or that there was a pre-existing history of hypertension. Note that the step-father in RASFMD3 was not a formal part of this study

individuals (62%) of the cohort had bilateral RAS/FMD, and three (23%) had proven carotid artery or intracranial arterial involvement. As indicated in Table 1, the 13 index cases had undergone various medical, interventional radiological and surgical therapies for hypertension and FMD, and these aspects of our patient cohort have been reported previously [14, 15].

Of the 40 first-degree relatives assessed in the study, 22 were parents (age range 28–58, median 44 years) and 18 were siblings, including two half-siblings (RASFMD3), (age range 3–30, median 13 years). Hypertension was noted in six (27%) of the 22 parents assessed. Of these, two were in the same kindred (RASFMD7). Five of the hypertensive parents had already been diagnosed and were on anti-hypertensive medication. Two fathers (RASFMD1 and -13)

were diagnosed during routine workplace health screening examinations when aged 40–49 years and two fathers (RASFMD4 and -7) were investigated when aged 28–35 years due to family history of hypertension. One mother (RASFMD7) was diagnosed with hypertension during her first pregnancy, and another mother (RASFMD2) was discovered to be hypertensive during this study, initially based on manual blood pressures and subsequently confirmed with 24-h ambulatory blood pressure monitoring. There was no family history of myocardial infarction or cerebrovascular accidents in the parents of the index cases, or of their parents’ siblings or parents. None of the parents of the index cases have yet undergone specific investigations to identify a cause for their hypertension (such as digital subtraction renal angiography) as their blood pressures are well controlled.

All eight adult siblings who were assessed were normotensive. Moreover, of the ten screened siblings under 18 years of age, none were hypertensive, with 60% (6 siblings) and 30% (3 siblings) with blood pressures <50th centile and between the 50th and 90th centiles for age, sex and height centile, respectively. Only one (brother in RASFMD1) sibling was noted to be pre-hypertensive, with manual blood pressure readings between the 90th and 95th centile for age, sex and height centiles. In three families (RASFMD2, -4 and -9), more distant relatives were known to have been diagnosed with hypertension and, for completeness, these are indicated in Fig. 1.

Finally, none of the 13 FMD index cases showed any sequence variations in *ACTA2*. We were unable to detect pathological or other sequence variants in our cohorts or controls.

Discussion

Hypertension occurs in 20–25% of all adults [16, 17], and so the incidence of hypertension in adult first-degree relatives appears to be unremarkable [27% in assessed parents, 0% in assessed adult siblings and 20% in all assessed adult first-degree relatives (parents and adult siblings)]. In addition, none of the siblings assessed in their childhood years were hypertensive. From these observations, we conclude that FMD diagnosed in a child is unlikely to confer a marked excess hypertension risk in first-degree relatives, at least up to middle-age.

Hypertension was noted in 27% parents [95% confidence interval (CI) 13–48%], which is comparable to 20–27% prevalence found in the general population (95% CI 13–34%). Although our parental population was young (age range 28–58 years), the majority of our parental hypertensive patients were aged 50–58 years. Our cohort was compared to the age-standardized and age-specific prevalence data of adult hypertension in the USA from the

Table 1 Presentation features, treatment and follow-up of index cases

Sex	Presenting age (years)	Presentation features	Maximum BP (mmHg)	Uni- or bilateral disease	Extra-renal arterial disease	Treatment	Follow-up (years)
M	0.7	Headaches	240/165	Bilateral	No	A (6), R (1)	10.5
F	6	Headaches	190/110	Bilateral	No	A (6), R (4), S (2)	7.5
M	0.03	Cardiac failure	140/90	Bilateral	No	A (3)	18
F	14	Headaches, dizzy spells	140/90	Unilateral	No	A (2), R (1)	3.5
M	9	Incidental (head injury)	190/135	Unilateral	No	A (3), R (1)	14
F	13	Encephalopathy	180/110	Unilateral	No	A (3), R (1), S (1)	20
M	9	Intracranial haemorrhage	170/100	Bilateral	Berry aneurysm	A (3), R (2)	6
F	7	Incidental (operation)	160/100	Bilateral	Carotid artery	A (4), R (2)	3
F	4	Headaches	150/90	Unilateral	No	A (5), R (2)	7
F	11	Incidental (appendicitis)	150/120	Bilateral	No	A (5), R (2)	17
F	0.3	Cardiac failure	140/90	Unilateral	No	A (4), R (1), S (1)	2.5
M	4	Facial palsy	240/120	Bilateral	Carotid artery	A (5), R (1)	0.5
F	3	Cardiac failure	150/100	Bilateral	No	A (4), R (1)	14

A, Anti-hypertensive agents (number); BP, blood pressure; F, female; M, male; R, renal angioplasties (number); S, surgical revascularizations (number)

National Health and Nutrition Examination Survey (NHANES of 1999–2004; <http://www.cdc.gov/nchs/nhanes.htm>) where hypertensive rates for 30- to 39-, 40- to 49- and 50- to 59-year-old Caucasian males and females were 12.5, 23.9 and 36.5%, and 5.4, 19.9 and 39.8%, respectively.

On the face of the evidence in our study, one could reason that familial FMD is indeed a rather rare entity, although we are unable to exclude polygenic factors or autosomal dominant inheritance with variable penetrance (partial penetrance has not been excluded). Previously reported data has suggested an autosomal recessive inheritance, and this cannot be refuted from our current cohort study. The fact that none of the eight adult siblings were hypertensive could have arisen out of chance, and 10% of siblings being pre-hypertensive could be expected of a normal healthy teenage population at the present time.

The strength of our study is that all 13 index cases presented with marked hypertension, often also having striking cardiovascular symptoms and signs, and had FMD proven by digital subtraction angiography. An additional strength was the fact that blood pressures were measured in the great majority of first-degree relatives by a trained nurse. However, we are aware that we ascertained only 87% of all possible first-degree relatives. In this respect, there were two siblings who we were unable to assess (brothers in RASFMD5; Fig. 1). We were unable to assess three fathers (RASFMD2, 5 and 6) who were alive at the time of the study and one mother had died (RASFMD12) from cancer. As far as we could establish, none of these four family relatives had a history of hypertension.

However, another caveat to our study is that none of the hypertensive parents underwent angiography to assign a definite FMD status nor are they likely to do so in the near future because their blood pressures are well controlled on current medications. Therefore, because of the lack of angiographic data, a case could be made that some of our families may have other members with hypertension and FMD.

As reasoned by Plouin et al., probably less than 1% of the total population will experience hypertension caused by renal arterial FMD [18]. However, to complicate matters, angiographic findings suggestive of FMD have been noted in up to 4% of otherwise healthy potential kidney donors [19]. Such individuals (who have not donated an affected kidney) are at excess risk compared to the general population of developing hypertension over the next decade [19]. Consistent with this observation is the fact that the radiographic severity of RAS associated with FMD can progress over time [6]. These observations may be relevant to the non-hypertensive first-degree relatives in our study because it remains possible that they have structural renal artery FMD but are “pre-hypertensive”. It would seem clinically inappropriate for them to currently undergo angiography given that they are well, and screening angiography could not be justified in a research study such as ours. On the other hand, we suggest that such individuals should have their blood pressures checked on an annual basis as an increased life-long risk of hypertension compared to the general population cannot be excluded.

Interestingly, the diagnosis of an “affected relative” in Rushton’s study, cited in the [Introduction](#), in which the possible familial nature of FMD was first raised, was

overwhelmingly based on the presence of hypertension or a cardiovascular event (such as myocardial infarction) rather than by a formal angiographic diagnosis of FMD [9]. Therefore, one could question the conclusion of that study, namely that FMD is not uncommonly inherited. In another study of 104 unrelated hypertensive patients with FMD [20], 11 index cases, all female with multifocal lesions, had at least one sibling identified as having FMD on angiography. However, this study from France did not systematically screen siblings by this technique. Index FMD cases can have asymptomatic carotid artery lesions and, in one study, non-invasive screening to seek such lesions demonstrated an increased prevalence in relatives of index cases in a pattern consistent with autosomal dominant inheritance [4].

If non-syndromic FMD has genetic causes, what might they be? In our cohort, FMD was not caused by *ACTA2* mutations, at least as assessed by the sequencing of all exons in all 13 index cases. A study of sibling pairs affected by FMD in whom a polymorphism in the gene encoding elastin (mutated in Williams–Beuren syndrome) was analysed excluded this locus. Other studies have provided preliminary evidence that polymorphisms in HLA [7] and the gene encoding angiotensin converting enzyme [21] may be associated with FMD, but these observations await replication in other cohorts.

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