

Aortic distensibility alterations in adults with m.3243A>G MELAS gene mutation

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Summary

Principles: MELAS, or mitochondrial myopathy, encephalopathy, lactic acidosis and stroke-like episodes is a new distinctive clinical entity. The current study was designed to assess ascending aortic elasticity in adult patients with MELAS syndrome and in gene carriers, and to compare the results with age- and gender-matched healthy controls.

Methods: The study comprised eight patients with MELAS syndrome and four asymptomatic gene carriers. All subjects underwent complete 2-dimensional transthoracic echocardiography, and systolic and diastolic ascending aortic diameters (SD and DD respectively) were recorded in M-mode 3 cm above the aortic valve from a parasternal long-axis view. Aortic elastic properties were calculated using aortic data and forearm blood pressure values.

Results: SD and DD of MELAS patients and gene carriers were enlarged compared with controls. Aortic stiffness index was increased (16.4 ± 3.7 vs 3.6 ± 1.1 , $p = 0.00001$), while aortic strain ($0.035 \pm 0.012\%$ vs $0.146 \pm 0.050\%$, $p = 0.00002$) and aortic distensibility (1.03 ± 0.30 cm²/dynes 10⁻⁶ vs 4.70 ± 1.69 cm²/dynes 10⁻⁶, $p = 0.0002$) were decreased in MELAS patients compared with controls. Aortic elastic properties of gene carriers were between MELAS patients and controls.

Conclusions: Increased ascending aortic stiffness and enlarged aortic dimensions suggesting vascular remodelling were found in MELAS patients as compared with controls.

Key words: aortic; vascular; distensibility; stiffness; elasticity; MELAS

Introduction

MELAS, or mitochondrial myopathy, encephalopathy, lactic acidosis and recurrent stroke-like episodes, was first described by Pavlakis et al. in 1984 as a new distinctive clinical entity [1]. MELAS syndrome is one of the well-described syndromes caused by mutations in mitochondrial deoxyribonucleic acid (DNA), and is the most common form of maternally inherited mitochondrial encephalopathy. The first gene mutation causing MELAS syndrome was found in 1990 as a mitochondrial adenine-to-guanine transition at nucleotide pair 3243 (m.3243A>G) [2]. Later, other less common mitochondrial gene mutations associated with MELAS syndrome were also de-

scribed (m.3271T>C and MTND5 gene mutations) [3, 4]. In MELAS patients preferential vascular involvement is believed to be responsible for the episodic nature of its clinical manifestations, including transient cerebral ischaemia causing stroke-like episodes and progressive mental deterioration [5, 6]. It is known that transthoracic echocardiographic M-mode measurement of cyclic ascending aortic diameter changes can be used to evaluate aortic elasticity using forearm blood pressure values [7, 8]. However, vascular distensibility data in this patient population was missing. The current study was therefore designed to assess whether ascending aortic elastic-

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ity in a unique group of adults with MELAS syndrome and carriers of the m.3243A>G mutation is affected, and to compare their results with age-,

gender- and risk factor-matched genetically healthy controls.

Patients and methods

Study population

The study comprised 12 adult patients, 8 patients with MELAS syndrome and 4 asymptomatic gene carriers (table 1). Patients were diagnosed on the basis of histological, biochemical and DNA studies as previously described [9]. Quadriceps muscle biopsy samples were morphologically investigated for signs of mitochondrial disease. Ragged red fibres (deposits of mitochondrial material beneath the sarcolemma), visualised by modified Gomori trichrome staining, or succinate dehydrogenase enhanced stained fibers were noted when seen in >1% of all fibres. Gene carriers were genetically, but not phenotypically, identical with MELAS syndrome patients with m.3243A>G gene mutation. The results of the ultrasound studies were compared with 16 age- and gender-matched healthy controls. Informed consent was obtained from each patient and the study was approved by the institutional review board at Erasmus Medical Centre, Rotterdam, Netherlands.

Transthoracic echocardiography

Echocardiographic studies were performed by the Sonos 7500 ultrasound system (Philips Medical Systems, Best, Netherlands) using a 3–5 MHz phased-array transducer. All echocardiographic studies were digitally stored and evaluated by experts who were blinded to the clinical data. All echocardiographic measurements were averaged from three beats. M-mode echocardiography was used to measure left ventricular (LV) internal dimensions. American Society of Echocardiography convention as the most

accepted border definition criteria was used to measure the leading edge of each layer [8]. Fractional shortening was calculated by the standard formula. Left ventricular mass (LVM) was calculated according to the Penn convention: $LVM (Penn) = [(EDD + IVS + PW)^3 - EDD^3] \times 1.04 - 13.6$ g, where EDD is the LV end-diastolic diameter, IVS is the interventricular septum and PW is the LV posterior wall [10].

Measurement of aortic stiffness

In all patients blood pressure was measured in the supine position with a mercury sphygmomanometer. None of the patients or control subjects received therapeutic drugs, used caffeinated beverages within one hour before blood pressure measurements or was a smoker. Systolic and diastolic ascending aortic diameters (SD and DD respectively) were recorded during M-mode echocardiography 3 cm above the aortic valve from a parasternal long-axis view, according to a method described previously in more detail [8] (fig. 1). SD and DD were measured at the time of maximum aortic anterior motion and at the peak of the QRS complex respectively. The following aortic elastic indices were calculated:

- Aortic strain = $(SD - DD) / DD$.
- Aortic stiffness index (β) = $\ln(SBP / DBP) / [(SD - DD) / DD]$, where SBP and DBP are the systolic and diastolic blood pressures, and “ln” is the natural logarithm.
- Aortic distensibility = $2 \times (SD - DD) / [(SBP - DBP) \times DD]$.

Table 1

Clinical, echocardiographic and blood pressure data in MELAS patients, gene carriers and controls.

	MELAS patients	Gene carriers	Controls
n	8	4	16
Age (years)	36.0 ± 12.1	31.5 ± 14.5	32.8 ± 9.3
Males (%)	4 (50)	1 (25)	6 (38)
Diabetes (%)	3 (38)	1 (25)	4 (25)
Hypertension (%)	2 (25)	1 (25)	4 (25)
SBP (mm Hg)	124.4 ± 24.3	124.5 ± 9.2	124.8 ± 13.0
DBP (mm Hg)	72.4 ± 16.4	75.0 ± 7.6	77.1 ± 8.7
Aortic PP (mm Hg)	52.0 ± 9.9	49.5 ± 8.2	47.7 ± 8.2
LV-EDD (mm)	49.4 ± 7.0	46.3 ± 4.6	46.8 ± 3.0
LV-ESD (mm)	34.8 ± 5.7	28.3 ± 4.7	29.7 ± 3.0 ‡
LV-FS (%)	29.6 ± 4.2	39.0 ± 6.6 †	36.5 ± 5.4 *
LV mass index (g/m ²)	118.4 ± 33.9	79.3 ± 3.5	78.4 ± 4.3 *
Aortic DS (mm)	28.8 ± 2.4	27.4 ± 3.5	25.9 ± 3.3
Aortic DD (mm)	27.9 ± 2.3	25.1 ± 1.9	22.6 ± 3.1 #
DS-DD (mm)	0.98 ± 0.33	2.25 ± 1.76	3.25 ± 1.04 *

Continuous variables are given as mean ± standard deviation

‡ p = 0.02 vs MELAS patients

* p = 0.0001 vs MELAS patients

p = 0.003 vs MELAS patients

† p = 0.03 vs MELAS patients

Abbreviations: SBP: systolic blood pressure, DBP: diastolic blood pressure, PP: aortic pulse pressure, DS: systolic diameter, DD: diastolic diameter, FS: fractional shortening

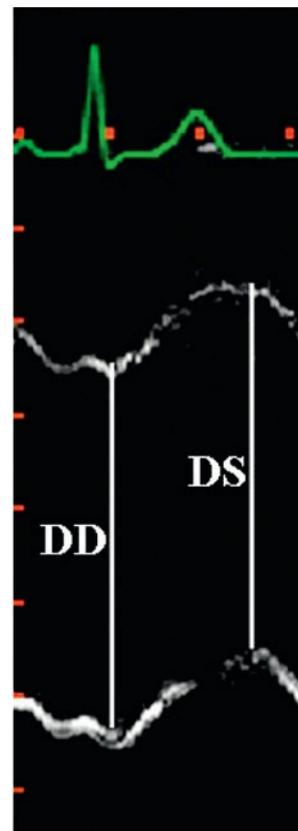


Figure 1

Measurements of systolic (SD) and diastolic (DD) diameters of the ascending aorta are shown on the M-mode tracing obtained 3 cm above the aortic valve.

All data are reported as mean \pm standard deviation. Variables were compared by the Student *t* test. A value of $p < 0.05$ was considered to be statistically significant.

Statistical analysis

SPSS 12.0 software was used for statistical calculations (SPSS Inc., Chicago, Illinois, USA). Interobserver reproducibility for measurement of systolic and diastolic ascending aortic diameters in our institution is 84% and 88% respectively [8].

Results

Clinical and demographic data of MELAS patients and gene carriers and controls are presented in table 1. Systolic and diastolic aortic diameters

of MELAS patients and gene carriers were enlarged compared with controls. Aortic stiffness index was increased (16.4 ± 3.7 vs 3.6 ± 1.1 , $p = 0.00001$), while aortic strain ($0.035 \pm 0.012\%$ vs $0.146 \pm 0.050\%$, $p = 0.00002$) and aortic distensibility (1.03 ± 0.30 cm²/dynes 10^{-6} vs 4.70 ± 1.69 cm²/dynes 10^{-6} , $p = 0.0002$) were decreased in MELAS patients compared with controls. Aortic elastic properties of gene carriers were between MELAS patients and controls. Aortic strain was decreased ($0.087 \pm 0.061\%$ vs $0.146 \pm 0.050\%$, $p = 0.05$), while aortic stiffness index was increased (8.5 ± 5.8 vs 3.6 ± 1.1 , $p = 0.002$) in gene carriers compared with controls. Individual values for aortic strain, aortic distensibility and aortic stiffness index in patients and controls are presented in figures 2, 3 and 4 (respectively).

Figure 2

Individual aortic strain values of MELAS patients, gene carriers and control subjects.

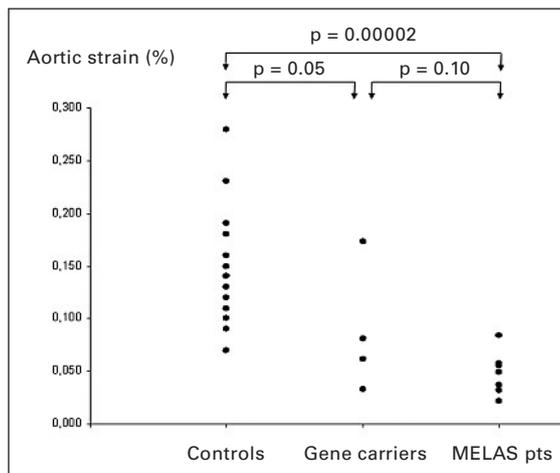


Figure 3

Individual aortic distensibility values of MELAS patients, gene carriers and control subjects.

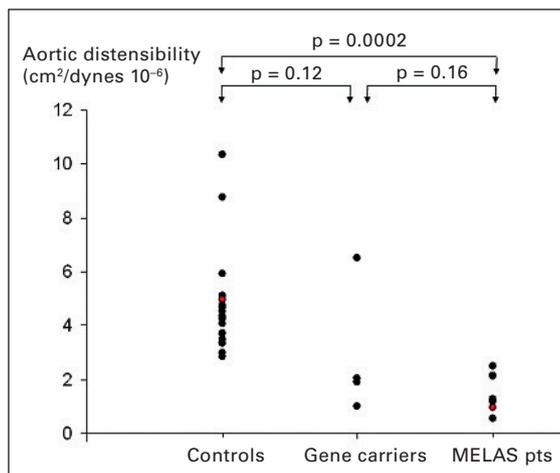
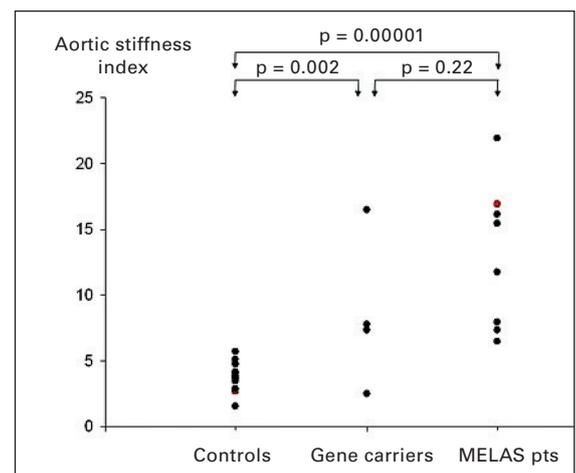


Figure 4

Individual β indices of MELAS patients, gene carriers and control subjects.



Discussion

The present study is an extension of a previous one in which cardiac involvement was examined in adults with m.3242A>G gene mutation [8]. It was postulated that vascular involvement is also present in these patients. To the best of the authors' knowledge this is the first time that alterations in ascending aortic elasticity in MELAS patients and gene carriers with m.3242A>G mitochondrial DNA mutation have been demonstrated. Increased ascending aortic stiffness and enlarged aortic dimensions suggesting vascular remodelling were found in MELAS patients as

compared with age- and gender-matched healthy controls. Moreover, progressively increased aortic stiffness was found in gene carriers and patients with MELAS phenotype.

Direct measurement of arterial stiffness requires invasive techniques unsuitable for routine clinical practice. At present there exist non-invasive methods of characterising aortic distensibility, including measurement of pulse-wave velocity or other indices / moduli based on cyclic aortic diameter changes and blood pressure values. Stefanadis et al. have demonstrated that the non-

invasively evaluated β index as a determinant of aortic stiffness is comparable with invasive methods with a high degree of accuracy [7].

In a recent study by our group major cardiac abnormalities including abnormal systolic and diastolic LV function and hypertrophy were seen in half of patients with MELAS syndrome, but in none of the gene carriers [9]. The other patients with MELAS syndrome had isolated abnormalities on systolic and diastolic tissue Doppler imaging. All these cardiac abnormalities were confined to patients with MELAS syndrome with ragged red fibres in skeletal muscle biopsy samples. Others have described cardiovascular manifestations in adults with MELAS syndrome including preexcitation, atrioventricular heart block, dilated and hypertrophic cardiomyopathy and hypertension [11].

Although the underlying mechanisms of stroke-like episodes in MELAS are not completely understood, mitochondrial angiopathy has been demonstrated in pathological studies with accumulation of abnormal mitochondria in the endothelial cells and smooth muscle cells of the pial arterioles and small cerebral arteries [6]. Takahashi et al. showed that a MELAS patient may have not only pathological but also functional vascular involvement [12]. Tay et al. described familial large vessel involvement with rupture of the aortic vessel in a family with MELAS [13]. They found a relatively higher mutation load in the aortic vessel wall compared with other tissues, and decreased immunostaining of the mitochondrial encoded cytochrome *c* oxidase I subunit of the vas vasorum of the aortic vessel wall and of the smooth muscle cells of the aorta. This suggests that the underlying mechanism of large vessel involvement is an impaired oxidative phosphorylation system causing a failure of energy production. Interestingly, there are only a

limited number of studies describing functional vascular alterations in MELAS syndrome. Koga et al. demonstrated that MELAS patients have decreased vasodilation capacity in small arteries [5]. They reported that L-arginine therapy, a precursor of nitric oxide, rapidly reduced the severity of stroke-like symptoms in MELAS, enhanced the dynamics of the microcirculation, reduced tissue injury due to ischaemia, and lowered the frequency and severity of stroke-like episodes [6]. Our study further supports the concept of vascular involvement in MELAS patients.

Limitations

This was a single-centre experience limited by the relatively small number of patients. The study would have been statistically stronger if a larger number of patients had been evaluated. No other methods were used to evaluate other vascular functional alterations (e.g., coronary flow reserve, flow-mediated vasodilation etc.). It is also known that classic cardiovascular risk factors including male gender, age, diabetes and hypertension could also affect aortic distensibility, thus influencing our results. Patient populations, however, were matched for these parameters. Medication used was not considered during evaluations. However, further studies are warranted to compare our findings with other functional vascular assessments.

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