

1 **A greater burden of atrial fibrillation is associated with worse endothelial**
2 **dysfunction in hypertension**

3 Short title: Endothelial function in AF and hypertension

4 Ahsan A Khan, MRCP¹

5 Rehan T Junejo, PhD^{2,3}

6 Reem Alsharari, MSc⁴

7 Graham N Thomas, PhD¹ *

8 James P Fisher, PhD⁵ *

9 Gregory Y.H. Lip, MD^{3,6} *

- 10 1. Institute of Applied Health Research, University of Birmingham, United Kingdom
11 2. School of Sport, Exercise and Rehabilitation Sciences, College of Life and
12 Environmental Sciences, University of Birmingham, United Kingdom
13 3. Liverpool Centre for Cardiovascular Science, Institute of Ageing and Chronic Disease,
14 University of Liverpool, United Kingdom
15 4. Institute of Cardiovascular Sciences, University of Birmingham, United Kingdom
16 5. Department of Physiology, Faculty of Medical and Health Sciences, University of
17 Auckland, New Zealand
18 6. Aalborg Thrombosis Research Unit, Department of Clinical Medicine, Faculty of
19 Health, Aalborg University, Denmark

20 *Joint senior authors

21 **Correspondence to:**

22 Professor Gregory Lip gregory.lip@liverpool.ac.uk

23 Full mailing address University of Liverpool, William Henry Duncan Building, 6 West
24 Derby Street, Liverpool, L7 8TX

25 Telephone number 0151 794 9020

26 Word count: 4224 (not including abstract, figures, tables and references)

27

28 **Abstract**

29

30 Atrial fibrillation (AF) and hypertension often co-exist and both are associated with
31 endothelial dysfunction. We hypothesised that AF would further worsen endothelium-
32 dependent flow-mediated dilatation (FMD) in hypertension patients compared to those
33 without AF. In a cross-sectional comparison, we measured brachial artery diameter at rest
34 and during reactive hyperaemia following 5 minutes of arterial occlusion in two patient
35 groups: AF (and hypertension) (n = 61) and hypertension control groups (n = 33). The AF
36 (and hypertension) subgroups: permanent AF (n = 30) and paroxysmal AF (n = 31) were also
37 assessed. The permanent AF patients received heart rate and blood pressure (BP) control
38 optimisation and were then followed up after eight weeks for repeat FMD testing. There
39 was no significant difference in FMD between AF (and hypertension) group and
40 hypertension control group (4.6%, 95% CI [2.6 – 5.9%] vs 2.6%, 95% CI [1.9 – 5.3%]; p=0.25).
41 There was a significant difference in FMD between permanent AF and paroxysmal AF groups
42 (3.1%, 95% CI [2.3 – 4.8%] vs 5.9%, 95% CI [4.0 – 8.1%]; p=0.02). Endothelium-dependent
43 FMD response showed a non-significant improvement trend following eight weeks of heart
44 rate and BP optimisation (3.1%, 95% CI [2.3 – 4.8%] (baseline) vs 5.2%, 95% CI [3.9 – 6.5%]
45 (follow up), p=0.09). Presence of AF generally does not incrementally worsen endothelial
46 dysfunction in hypertension patients, although the duration and frequency of AF
47 (paroxysmal AF to permanent AF) does lead to worsening endothelial function. Eight weeks
48 of BP optimisation did not significantly improve endothelial dysfunction as measured by
49 FMD.

50

51 Introduction

52 Atrial fibrillation (AF) is associated with increased morbidity including stroke, heart failure,
53 thromboembolic complications and high mortality.¹ Hypertension accounts for more cases
54 of AF than other risk factors, increasing the risk of AF two-fold.² In the Framingham study,
55 for example, hypertension heralded an excess risk of AF by 50% in males and 40% in
56 females.³ Among individuals with a confirmed diagnosis of AF, hypertension is present in
57 about 60% to 80% of these patients.⁴ These 2 conditions often co-exist in the same patient,
58 and their prevalence is increasing globally. It is widely perceived that the combination of
59 these conditions confers a worse prognosis than either alone.⁵

60 Beat to beat variation in blood flow dynamics during AF has been related to presence of
61 endothelial dysfunction.⁶ It is well established that the endothelium plays a fundamental
62 role in the regulation of vascular tone by releasing a variety of vasodilatory substances,
63 particularly nitric oxide (NO). NO modulates vascular smooth muscle tone by exerting its
64 effects at a cellular level. A key consequence of normal endothelial function in vivo is the
65 ability to release NO in response to physiological stimuli, such as increased flow, reflecting
66 endothelial flow-mediated dilatation (FMD).⁷

67 Impaired FMD is associated with cardiovascular risk factors and provides important
68 prognostic information. FMD measurement using high-resolution ultrasound has become a
69 reliable and reproducible technique for assessment of endothelial dysfunction.⁸ When blood
70 flow through a vessel increases, the resultant increase in shear stress on the vascular
71 endothelium causes endothelium-dependent vasodilation. The magnitude of this
72 vasodilatory response can be used as an index of endothelial function.

73 Several studies have previously shown impaired FMD as a marker of endothelial dysfunction
74 in patients with various atherosclerotic risk factors, including advanced age, hypertension,
75 hypercholesterolaemia, diabetes mellitus, tobacco use and postmenopausal status.⁹⁻¹² FMD
76 is also found to be impaired in patients with AF.¹³⁻¹⁶ Since AF and hypertension, commonly
77 co-exist, we hypothesised that endothelium-dependent FMD will be reduced in patients
78 with AF (and hypertension) compared to hypertensive controls and this may partly explain
79 the poor prognosis in such patients.

80 We therefore aimed to assess whether presence of AF leads to worsening of endothelial
81 dysfunction in hypertensive patients through assessment by FMD, to assess whether there
82 are any differences in FMD between permanent AF and paroxysmal AF, and lastly whether
83 improvement in blood pressure (BP) control can lead to improvement in FMD.

84 **Methods**

85 Participants were provided with detailed information sheets, and written informed consent
86 was obtained from all participants, in accordance with the Declaration of Helsinki (2013).
87 Eligible participants underwent screening against inclusion and exclusion criteria before
88 being invited to take part in the study (see **supplementary material**). The study was
89 approved by the Health Research Authority (HRA) and National Research and Ethics Service
90 (NREC) Committee London – Camden & Kings Cross (18/LO/1064). Anonymized data and
91 materials have been made publicly available at the Harvard Dataverse and can be accessed
92 at <https://doi.org/10.7910/DVN/QKG7DL>.

93 A total of 94 participants were recruited from the atrial fibrillation and hypertension
94 services at Sandwell and West Birmingham Hospitals NHS Trust between October 2018 –

95 March 2019. We recruited 2 groups of patients: AF (and hypertension) (n = 61) and
96 hypertension control (n = 33). Patients with AF were stable on rate control and
97 antithrombotic medication. The AF (and hypertension) group was further subdivided into
98 permanent AF (n = 30) and paroxysmal AF (n = 31). Permanent AF was defined as an episode
99 of AF in which efforts to restore normal sinus rhythm had either failed or been abandoned.
100 Paroxysmal AF was defined as an episode of AF that terminates spontaneously or with
101 intervention in less than seven days. The hypertension control group included patients with
102 hypertension (defined as previous diagnosis of hypertension or clinic BP of $\geq 140/90$ mmHg)
103 but not AF. These patients had additional cardiovascular risk factors similar to the other two
104 AF groups and acted as the control group.

105 Initially, a cross-sectional age and clinical characteristics-matched comparison of the two
106 main groups, AF (and hypertension) versus hypertension control was carried out. This was
107 followed by the two subgroups of AF (and hypertension) group. Lastly, the patient group
108 with permanent AF (and hypertension) (n = 30) were studied longitudinally with a single
109 follow-up interval of 8 weeks duration following optimisation of their heart rate (HR) and BP
110 medication. The medication optimisation was carried out by a single clinician with
111 experience in managing these conditions and involved either increasing the dosage of
112 existing cardiovascular medication or addition of a new medication (for which the
113 prescription was provided) according to participants' needs, allergy status, known
114 contraindications and clinical indication. These patients underwent the same measurements
115 as at their first visit.

116

117 Experimental protocol

118 Participants were expected to fast from food, water, caffeine and withhold their
119 cardiovascular medications, except anticoagulation, for at least 12 hours prior to their
120 appointment. They were advised to refrain from smoking for at least 4 hours, physical
121 exercise for 12 hours and drinking alcohol for at least 24 hours prior to their appointment.
122 At the experimental appointment, a detailed medical history was taken from the
123 participants including medications history and a physical examination carried out. This
124 included anthropometric measurements such as height and weight to determine BMI
125 (weight/height²; kg/m²). An ECG was performed on all participants to determine rhythm.

126 Baseline blood samples to test for full blood count, renal, liver and thyroid function, fasting
127 glucose, lipid, and clotting profile, were taken from participants from their left antecubital
128 fossa if they have not had these tests taken within 6 months of their study appointment. A
129 full transthoracic echocardiogram study was performed if a participant did not have a recent
130 echocardiogram. Subsequent measurements were performed in a temperature-controlled
131 room under uniform conditions with participants resting quietly in the supine position on a
132 medical examination couch.

133 Measurements

134 Three serial BP readings were taken non-invasively from the left brachial artery using an
135 automated sphygmomanometer over 5 minutes to determine an average. Vascular function
136 was assessed by measuring brachial artery blood flow velocity and diameter. The
137 measurements were obtained from the right arm positioned at heart level by Doppler
138 ultrasound (CX50 CompactXtreme; Philips, Amsterdam, Netherlands) by a single

139 experimenter, using a 10-MHz multi-frequency linear-array transducer. B-mode imaging was
140 used to measure arterial diameter, and peak blood velocity was simultaneously measured
141 using the pulse-wave mode. Measurements were made in accordance with recent technical
142 recommendations.¹⁷ The ultrasound machine was connected via a HDMI AV.io (Epiphan
143 Video Systems Inc, California, USA) video grabber to a laptop with a dedicated FMD
144 software, QUIPU Cardiovascular Suite (Quipu srl, Pisa, Italy) with edge-detection capability
145 and real-time processing and recording of B-mode ultrasound image sequence, removing
146 the need for ECG gating.¹⁸ This software utilises image based automated edge detection and
147 wall tracking algorithms working independently of investigator influence. This system has
148 been used and validated in other studies involving human participants.^{18, 19}

149 Participants lay supine on the couch with their right arm extended out and had a narrow
150 inflatable cuff (5-cm width; Hokanson, Bellevue, WA) placed 5 – 7 cm distal to the medial
151 epicondyle. The arm was positioned in a comfortable position. The brachial artery was
152 imaged 10-15 cm proximal to the medial epicondyle at 60° insonation angle in the
153 longitudinal plane. Duplex imaging was used to obtain a B-mode image of vessel diameter
154 and pulse-wave mode for peak blood velocity. Ultrasound measurements were made in
155 accordance with technical recommendations.¹⁷ Following 1 minute of baseline diameter
156 recording, the arterial occlusion cuff was inflated to 50 mmHg above systolic BP for 5
157 minutes. Following this, the cuff was rapidly deflated and arterial image recording continued
158 for further 2 minutes. Recordings were screen captured and stored as video files and off-line
159 analysis carried out with automated edge detection and wall tracking software
160 (Cardiovascular Suite version 3.4.1; FMD Studio, Pisa, Italy).

161

162 Data analysis

163 Patients were matched for age and clinical characteristics to reduce chances of
164 confounders. Body mass index (BMI) was expressed as the ratio of the participants' weight
165 and their height squared. Digitally recorded data were extracted in an anonymized manner.
166 Mean arterial pressure (MAP) was the mean blood pressure over each cardiac cycle.
167 Brachial artery FMD was taken as the maximal change in brachial artery diameter following
168 cuff deflation. The time to peak diameter was obtained between the cuff deflation and the
169 maximal artery dilation, and the time to peak blood flow (reactive hyperaemia) was
170 obtained between cuff deflation and maximal flow velocity. Shear rate (positive shear rate
171 area to peak) was calculated as an integral between the cuff deflation and the maximal
172 artery dilation. FMD was expressed as absolute (mm) and relative change (%) in diameter.
173 Based on recent guidelines, covariate-corrected FMD was presented, adjusting for
174 differences in baseline diameter between the two groups using analysis of covariance
175 (ANCOVA).²⁰

176 Statistical analysis

177 Descriptive statistics are presented as mean \pm standard deviation (SD) or median with
178 interquartile range, as appropriate for continuous variables. Categorical variables are
179 expressed as numbers and percentages. Statistical analysis was performed using SPSS
180 software (version 26.0; SPSS Inc., Chicago, Illinois). Continuous variables were tested for
181 normality using the Shapiro-Wilk test. If passed, data was analysed using independent
182 Student's t-test between the two groups. Data found to be not normally distributed were
183 analysed with Mann-Whitney U test. For longitudinal comparison, continuous variables

184 were tested for normality using the Shapiro-Wilk test. If passed, data was analysed using
185 Student's paired t-test. Data found to be not normally distributed were analysed with
186 Wilcoxon Signed Rank test. A p value of < 0.05 was considered statistically significant.
187 Associations between FMD and co-variables were assessed before and after adjustment for
188 potential confounders (age, sex, BMI) using linear regression analysis.

189 To test specific hypothesis 1 ("Patients with AF and hypertension will have worse
190 parameters of vascular function compared to hypertension control group"), we recruited 94
191 patients in total, split between 2 groups (a) AF and hypertension (b) hypertension control.
192 This part of the study was powered based on independent t-test, comparing the flow-
193 mediated dilatation values across the two groups. Skalidis *et al* reported a mean FMD of 8.1
194 (standard deviation (SD) = 3.6) in a pre-treatment (i.e. cardioversion) AF group.⁶ Assuming
195 our SD is similar, the minimum sample size was computed as 18 patients per group at 90%
196 power, 5% alpha and effect size of 1.14.

197 To test specific hypothesis 2 ("Patients with permanent AF and hypertension will have
198 worse parameters of vascular function compared to patients with paroxysmal AF and
199 hypertension"), we recruited 61 patients in total, split between the 2 groups. This part of
200 the study was powered based on an independent t-test, assessing the difference in FMD
201 between permanent AF and paroxysmal AF. Mazaris *et al* reported a mean FMD of 4.09 (SD
202 = 1.67) in permanent AF group compared to mean FMD of 6.83 (SD = 1.38) in paroxysmal AF
203 group.¹⁶ Assuming our SD is similar, the minimum sample size was computed as 8 patients
204 per group at 90% power, 5% alpha and effect size of 1.79.

205 To test specific hypothesis 3 (“Eight weeks of intensive anti-hypertensive and
206 anticoagulation therapy will improve vascular function in patients with permanent AF and
207 hypertension”) we recruited 30 patients and tested them before and after intensification of
208 their antihypertensive and anticoagulation treatment. This part of the study was powered
209 based on a paired t-test, assessing the change in flow mediated dilation from pre- to post-
210 treatment. It was assumed that the mean pre- intervention flow mediated dilation would be
211 8.1 (SD=3.6), as per Skolidis et al, and that the effect size would be 1.06.⁶ If this is the case,
212 then the minimum number of patients required is 12 at 90% power and 5% alpha.

213 **Results**

214 Matched AF (and hypertension) group vs matched hypertension control group

215 Participants from AF (and hypertension) group and hypertension control group were
216 matched for age and clinical characteristics (see table 1). Participants’ medication history is
217 displayed in figure 1. There were no significant differences in age, sex, height, weight and
218 BMI. Past medical history of all participants between the groups was similar except that
219 participants in hypertension control group had significantly more patients with a
220 background of chronic kidney disease (CKD) ($p = 0.01$). The CHA₂DS₂-VASc score and HAS-
221 BLED score were similar between the two groups. The mean heart rate was significantly
222 lower in the hypertension control group ($p = 0.02$). There were no significant differences in
223 mean blood pressure (systolic and diastolic) between the two groups, baseline glycaemia
224 control (HBA1c), kidney function (creatinine clearance) and left ventricular ejection fraction
225 (EF (%)).

226 Baseline diameter of brachial artery was significantly smaller in the AF (and hypertension)
227 group compared to hypertension control group (4.6 mm, 95% confidence interval (CI) [4.4 –
228 4.9 mm] vs 5.2 mm, 95% CI [4.8 – 5.6 mm]; $p = 0.02$) (see table 2). Following 5 minutes of
229 forearm ischaemia, there was no significant difference in absolute FMD between AF (and
230 hypertension) group and hypertension control group (0.2 mm, 95% CI [0.1 – 0.3 mm] vs 0.2
231 mm, 95% CI [0.1 – 0.3 mm]; $p = 0.61$) or FMD percentage (4.6%, 95% CI [2.6 – 5.9%] vs 2.6%,
232 95% CI [1.9 – 5.3%]; $p = 0.25$) respectively. The FMD (%) means were adjusted for baseline
233 diameter and showed no significant difference between the two groups (4.9%, 95% CI [3.8 –
234 6.0%] (AF (and hypertension) group) vs 4.3%, 95% CI [2.8 – 5.9%] (hypertension control
235 group), $p = 0.56$).

236 The peak diameter was significantly different between the two groups (4.9 mm, 95% CI [4.6
237 – 5.2 mm] (AF (and hypertension) group) vs 5.4 mm, 95% CI [5.0 – 5.8 mm] (hypertension
238 control group); $p = 0.03$). There were no significant differences in time to peak diameter and
239 shear rate between the two groups ($p = 0.07$ and $p = 0.41$ respectively). No variables were
240 identified on univariate and stepwise multivariate analysis as independent predictors of
241 reduced FMD.

242 Permanent AF (and hypertension) vs PAF (and hypertension) groups

243 Participants in the two AF subgroups (permanent AF vs paroxysmal AF) were well matched
244 for age, sex, clinical characteristics including height, weight, BMI, mean blood pressure,
245 HBA1c, creatinine clearance and left ventricular EF (%) (see table 3). Participants'
246 medication history is displayed in figure 1. There was a significantly higher incidence of

247 ischaemic heart disease in paroxysmal AF group ($p < 0.001$) and mean heart rate was found
248 to be significantly slower in participants in paroxysmal AF group ($p = 0.003$).

249 On FMD measurement, there were no significant difference in baseline diameter between
250 the two groups (permanent AF (4.5 mm, 95% CI [4.2 – 5.0 mm]) vs paroxysmal AF (4.8 mm,
251 95% CI [4.6 – 5.1 mm]) $p = 0.67$) (see table 4). Following 5 minutes of forearm ischaemia,
252 there was a significant difference in absolute FMD change between permanent AF and
253 paroxysmal AF (0.1 mm, 95% CI [0.1 – 0.2 mm] vs 0.3 mm, 95% CI [0.2 – 0.4 mm]; $p = 0.01$
254 respectively). There was also a significant difference in FMD percentage between the two
255 groups (3.1%, 95% CI [2.3 – 4.8%] (permanent AF) vs 5.9%, 95% CI [4.0 – 8.1%] (paroxysmal
256 AF); $p = 0.02$). This difference persisted with correction for baseline diameter (3.9%, 95% CI
257 [2.8 – 5.0%] (permanent AF) vs 5.9%, 95% CI [4.8 – 7.0%] (paroxysmal AF); $p = 0.01$).

258 There was no significant difference in peak diameter ($p = 0.49$), time to peak diameter ($p =$
259 0.23) and shear rate ($p = 0.40$) between the two groups. Presence of permanent AF
260 (Spearman's rho 0.295; $p = 0.02$) and ischaemic heart disease (Spearman's rho 0.280; $p =$
261 0.03) were identified as independent predictors of reduced FMD on univariate analysis ($p =$
262 0.03) but only permanent AF was identified as an independent predictor of reduced FMD on
263 stepwise multivariate analysis (R^2 0.090; F 5.855; $p = 0.02$).

264 Permanent AF (and hypertension) group – longitudinal comparison

265 Following optimisation of HR and BP medication, patients with permanent AF (and
266 hypertension) were followed up after eight weeks and FMD repeated (see table 5). There
267 was significant improvement in mean heart rate (77 beats per minute (bpm) \pm 18 (baseline)
268 vs 72 bpm \pm 17 (follow up), $p = 0.01$), systolic BP (140 mmHg [128 – 148] (baseline) vs 131

269 mmHg [122 – 146] (follow up), $p = 0.03$), diastolic BP (81 mmHg \pm 13 (baseline) vs 77 mmHg
270 \pm 12 (follow up), $p = 0.02$) and mean arterial pressure (MAP) (100 mmHg \pm 9 (baseline) vs 97
271 mmHg \pm 13 (follow up), $p = 0.01$).

272 Both groups had a similar baseline brachial artery diameter ($p = 0.34$). Endothelium-
273 dependent FMD response was better following eight weeks of HR and BP optimisation but
274 this 68% relative improvement did not reach statistical significance (3.1%, 95% CI [2.3 –
275 4.8%] (baseline) vs 5.2%, 95% CI [3.9 – 6.5%] (follow up), $p = 0.09$). The FMD (%) means
276 were adjusted for baseline diameter and showed no significant difference between the two
277 groups (4.0%, 95% CI [3.0 – 4.9%] (baseline) vs 5.1%, 95% CI [4.2 – 6.1%] (follow up), $p =$
278 0.09). The difference was also not significant in absolute change in diameter (0.14 mm, 95%
279 CI [0.11 – 0.25 mm] (baseline) vs 0.20 mm, 95% CI [0.17 – 0.28 mm] (follow up), $p = 0.15$).
280 The time to peak diameter, peak diameter and shear rate stimulus were similar between the
281 two groups (table 5). No variables were identified on univariate or stepwise multivariate
282 analysis as independent predictors of reduced FMD.

283 Discussion

284 This is the first study investigating whether the presence of AF worsens the endothelial
285 dysfunction seen in patients with hypertension. The results are consistent with other studies
286 looking at FMD in hypertension and AF individually and confirms that endothelial
287 dysfunction is present.^{9, 13-16, 21} Our findings extend previous work by demonstrating that the
288 presence of AF generally does not incrementally worsen endothelial dysfunction, nor was AF
289 an independent predictor of endothelial dysfunction on multivariate analysis. However,
290 permanent AF compared to paroxysmal AF does have significantly worse FMD parameters

291 with permanent AF being an independent predictor on multivariate analysis. Lastly, we did
292 not find any significant improvement in FMD following 8 weeks of HR and BP optimisation in
293 permanent AF and hypertension patients.

294 There are potentially several reasons that may explain the lack of differences seen between
295 AF (and hypertension) and hypertension control group in our study. These can be broadly
296 categorised into oxidative stress, inflammation and the role of endothelial nitric oxide
297 synthase (eNOS). Increase in systemic oxidative stress is thought to play a part in endothelial
298 dysfunction seen in patients with hypertension, whereas a reduction has been shown to
299 reverse endothelial dysfunction.²² Risk factors for AF are similar to those of atherosclerosis
300 and hypertension, diseases known to be perpetuated by oxidative stress. This can explain
301 why the addition of AF does not significantly worsen endothelial dysfunction seen in
302 patients with hypertension.

303 Inflammation has also been implicated in the pathophysiology of hypertension as well as
304 initiation and perpetuation of AF and AF-related adverse effects.^{23, 24} Endothelial
305 dysfunction seen in hypertension relates to local vascular inflammation and systemic
306 inflammation.²⁵ Also, inflammation contributes to the pathophysiology of AF, both directly
307 and through AF-promoting cardiovascular conditions that have an inflammatory aetiology.²⁶
308 FMD has been shown to be inversely associated with serum C-reactive protein (CRP) levels
309 in chronic AF patients, implying disruption by inflammation.²⁷ Since inflammation plays an
310 important role in causing endothelial dysfunction in both conditions, it is perhaps
311 unsurprising that we did not see a significant difference in the FMD response between the
312 groups, suggesting that endothelial perturbation seen in AF may reflect underlying
313 comorbidities rather than AF per se. Interestingly, endothelial dysfunction itself enhances

314 oxidative stress and leads to increase in recruitment of proinflammatory agents promoting a
315 vicious cycle.²⁸ The complex interplay involving oxidative stress and inflammation seen in
316 both conditions is summarised in figure 2.

317 eNOS, a key regulator of vascular tone is found to be reduced or dysfunctional in both
318 hypertension and AF.^{29, 30} eNOS produces NO to mediate relaxation of blood vessels and
319 preservation of vascular function. When eNOS is deprived of its critical cofactor
320 tetrahydrobiopterin or its substrate L-arginine, it results in synthesis of large volumes of
321 reactive oxygen species such as peroxynitrite (superoxide) instead of NO, leading to nitric
322 oxide synthase (NOS) uncoupling. Superoxide production by uncoupled eNOS further
323 sustains oxidative stress in the vasculature, resulting in endothelial dysfunction, impaired
324 endothelium-dependent vasorelaxation and elevated BP.²⁹ This inadvertently leads to tissue
325 damage that promotes pathological remodelling of the myocardium contributing to
326 initiation and propagation of AF.³¹ Since the aetiology and pathophysiology of endothelial
327 dysfunction are similar in both hypertension and AF, this supports our finding that AF and
328 hypertension had similar effect on the FMD with no significant difference seen between the
329 two groups. Our study also suggests that AF, as opposed to hypertension, is perhaps the
330 dominant condition responsible for endothelial dysfunction in these patients as permanent
331 AF group showed a worse FMD compared to paroxysmal AF group and 8 weeks of intensive
332 hypertensive therapy revealed a non-significant improvement trend in FMD.

333 Interestingly, we were able to see a significant difference in FMD between permanent AF
334 and paroxysmal AF groups with more impaired FMD noted in permanent AF group. This
335 suggests that frequency and duration of AF episode or type of AF may be important in
336 progression of endothelial dysfunction. Our findings are similar to other studies showing

337 that patients with permanent AF have worse FMD compared to patients with paroxysmal
338 AF.^{15, 16, 32} However, unlike previous studies, our study included patients with AF and
339 hypertension, which has not been looked at before.

340 Although, our study did show that improvement in HR and BP can lead to improvement in
341 FMD in hypertensive patients despite the presence of AF, however this 68% improvement
342 did not reach statistical significance ($p = 0.09$). These results are similar to the study
343 performed by Modena and colleagues who looked at hypertensive patients (without AF) and
344 showed that 6 month of BP optimisation led to improvement in FMD and was associated
345 with a more favourable prognosis.³³ Thus, longer-term improvement in FMD may have a
346 prognostic implication.³⁴ Furthermore, it supports previous work showing modulation of
347 endothelial function is possible and that endothelial dysfunction is a reversible condition.²⁵

348 Our study has several important clinical implications. We have been able to show that
349 endothelial dysfunction is present in patients with AF and hypertension. This may explain
350 the increased risk of stroke and heart attack in these patients as endothelial function may
351 be involved in the pathophysiology of these conditions, in addition to the prothrombotic
352 state seen in AF. We have been able to show that increased frequency and duration of AF
353 leads to worsening of endothelial function and thus these patients may benefit from closer
354 monitoring and perhaps consideration for AF ablation. We have also shown that
355 improvement in HR and BP leads to improvement in FMD, although it was not significant in
356 our study. Nevertheless, it does suggest that endothelial function may be a reversible
357 condition if risk factors such as blood pressure are controlled and optimised.

358

359 *Strengths and limitations*

360 We did not use nitrate to assess for endothelium-independent vasodilation as this has been
361 studied previously in both AF and hypertension.^{9, 13, 15, 21} Furthermore, use of intra-arterial
362 acetylcholine would have been advantageous to investigate brachial artery endothelial
363 function but FMD is a well established surrogate.¹⁷ Given the widespread prevalence of AF
364 in hypertensive patients, the inclusion of separate hypertension groups with and without
365 AF, is a strength of our study. There have been limited studies looking at vascular function in
366 patients with AF arrhythmia and therefore this makes our study unique. Participants in our
367 group were well-matched for age, sex composition, comorbidities, CHA₂DS₂-VASc score,
368 HAS-BLED score, BMI, BP, glycaemic control and LV systolic function. Nonetheless, the
369 hypertension control group did have a significantly higher number of patients with CKD
370 which may have been a source of bias. We accommodated for this and other potential
371 confounders by utilisation of linear regression analysis. The longitudinal comparison of
372 permanent AF (and hypertension) group in assessing FMD response to intervention has not
373 been looked at before. The utilisation of edge detection software, assessment of shear rate
374 and correcting for differences in group baseline diameters shows robustness of our
375 methodological approach.

376 In contrast, our study has some limitations. Endothelial function was examined using the
377 well-established brachial artery flow mediated dilatation technique in accordance with
378 recent technical recommendations, however we acknowledge that this may not provide an
379 optimal assessment of endothelial dysfunction.¹⁷ Second, it would have been useful to
380 compare our findings with a healthy control group and/or a group with AF but no
381 hypertension as the relation between hypertension and AF is bi-univocal. Third, the use of

382 anti-hypertensives and other concomitant medications may have influenced endothelial
383 function long term which cannot be excluded. Additionally, whilst we were able to show
384 reduction in HR and BP in our longitudinal study, the short duration of 8 weeks may not be
385 enough to reveal significant improvement in endothelial function. Fourth, we did not
386 measure other potential causes for endothelial dysfunction such as changes in free fatty
387 acids, inflammatory cytokines, inflammatory markers such as c-reactive protein (CRP), nitric
388 oxide synthase expression and endothelin. However, this real world cohort has ecological
389 validity and makes our observations more representative of the clinic. Future studies should
390 look at whether and how endothelial function progresses in patients with AF over time and
391 compare it to patients with hypertension to assess if there are any differences.

392 **Conclusions**

393 The presence of AF generally does not incrementally worsen endothelial dysfunction in
394 hypertension, nor was AF an independent predictor of endothelial dysfunction on
395 multivariate analysis. However, duration and frequency of AF leads to worsening endothelial
396 function as demonstrated in our study. Eight weeks of BP optimisation did not give a
397 significant improvement in endothelial dysfunction as measured by FMD.

398

399 Summary Table

What is known about topic?
<ul style="list-style-type: none">• Atrial fibrillation and hypertension commonly co-exist and the combination of these two conditions confers a worse prognosis than either alone.• Endothelial dysfunction is present in both atrial fibrillation and hypertension.• Flow-mediated dilatation is a reliable tool to assess endothelial function.
What this study adds?
<ul style="list-style-type: none">• Presence of AF generally does not incrementally worsen endothelial dysfunction in hypertension patients• The duration and frequency of AF (paroxysmal AF to permanent AF) does lead to worsening endothelial function.• There is potential for endothelial dysfunction to improve following optimisation of BP suggesting modulation of endothelial function is possible in patients with permanent AF and hypertension.

400

401 **Acknowledgements**

402 The time and effort expended by all the participants is greatly appreciated.

403 **Conflict of Interest**

404 Authors declare no conflict of interests for this article.

405 GYHL has served as a consultant for Bayer/Janssen, BMS/Pfizer, Biotronik, Medtronic,
406 Boehringer Ingelheim, Microlife and Daiichi-Sankyo; and a speaker for Bayer, BMS/Pfizer,
407 Medtronic, Boehringer Ingelheim, Microlife, Roche and Daiichi-Sankyo. No personal fees
408 received.

409 **Sources of Funding**

410 None

References

- 411
412
- 413 1. Kannel WB, Wolf PA, Benjamin EJ and Levy D. Prevalence, incidence, prognosis, and
414 predisposing conditions for atrial fibrillation: population-based estimates. *Am J Cardiol.*
415 1998; 82: 2n-9n.
 - 416 2. Lau YF, Yiu KH, Siu CW and Tse HF. Hypertension and atrial fibrillation: epidemiology,
417 pathophysiology and therapeutic implications. *J Hum Hypertens.* 2012; 26: 563-9.
 - 418 3. Benjamin EJ, Levy D, Vaziri SM, D'Agostino RB, Belanger AJ and Wolf PA. Independent risk
419 factors for atrial fibrillation in a population-based cohort. The Framingham Heart Study.
420 *JAMA.* 1994; 271: 840-4.
 - 421 4. Nabauer M, Gerth A, Limbourg T, Schneider S, Oeff M, Kirchhof P, et al. The Registry of
422 the German Competence NETwork on Atrial Fibrillation: patient characteristics and initial
423 management. *Europace.* 2009; 11: 423-34.
 - 424 5. Dzeshka MS, Shantsila A, Shantsila E and Lip GYH. Atrial Fibrillation and Hypertension.
425 *Hypertension.* 2017; 70: 854-861.
 - 426 6. Skalidis EI, Zacharis EA, Tsetis DK, Pagonidis K, Chlouverakis G, Yarmenitis S, et al.
427 Endothelial Cell Function During Atrial Fibrillation and After Restoration of Sinus Rhythm.
428 *Am J Cardiol.* 2007; 99: 1258-1262.
 - 429 7. Joannides R, Haefeli WE, Linder L, Richard V, Bakkali EH, Thuiliez C, et al. Nitric oxide is
430 responsible for flow-dependent dilatation of human peripheral conduit arteries in vivo.
431 *Circulation.* 1995; 91: 1314-9.
 - 432 8. Corretti MC, Anderson TJ, Benjamin EJ, Celermajer D, Charbonneau F, Creager MA, et al.
433 Guidelines for the ultrasound assessment of endothelial-dependent flow-mediated
434 vasodilation of the brachial artery. A report of the International Brachial Artery Reactivity
435 Task Force. 2002; 39: 257-265.

- 436 9. Iiyama K, Nagano M, Yo Y, Nagano N, Kamide K, Higaki J, et al. Impaired endothelial
437 function with essential hypertension assessed by ultrasonography. *Am Heart J.* 1996; 132:
438 779-782.
- 439 10. Simons LA, Sullivan D, Simons J and Celermajer DS. Effects of atorvastatin monotherapy
440 and simvastatin plus cholestyramine on arterial endothelial function in patients with severe
441 primary hypercholesterolaemia. *Atherosclerosis.* 1998; 137: 197-203.
- 442 11. Lekakis J, Papamichael C, Anastasiou H, Alevizaki M, Desses N, Souvatzoglou A, et al.
443 Endothelial dysfunction of conduit arteries in insulin-dependent diabetes mellitus without
444 microalbuminuria. *Cardiovasc Res.* 1997; 34: 164-168.
- 445 12. Thomas GN, Chook P, Yip TW, Kwong SK, Chan TY, Qiao M, et al. Smoking without
446 exception adversely affects vascular structure and function in apparently healthy Chinese:
447 implications in global atherosclerosis prevention. *Int J Cardiol.* 2008; 128: 172-7.
- 448 13. Freestone B, Chong AY, Nuttall S and Lip GY. Impaired flow mediated dilatation as
449 evidence of endothelial dysfunction in chronic atrial fibrillation: relationship to plasma von
450 Willebrand factor and soluble E-selectin levels. *Thromb Res.* 2008; 122: 85-90.
- 451 14. Borschel CS, Rubsamen N, Ojeda FM, Wild PS, Hoffmann BA, Prochaska JH, et al.
452 Noninvasive peripheral vascular function and atrial fibrillation in the general population. *J*
453 *Hypertens.* 2019; 37: 928-934.
- 454 15. Komatsu T, Kunugita F, Ozawa M, Satoh Y, Yoshizawa R, Owada S, et al. Relationship
455 between Impairment of the Vascular Endothelial Function and the CHA2DS2-VASc Score in
456 Patients with Sinus Rhythm and Non-valvular Atrial Fibrillation. *Intern Med.* 2018; 57: 2131-
457 2139.

- 458 16. Mazaris S. TD, Siasos G., Zisimos K., Oikonomou E., Kokkou E., Konsola T., Lazaros G.,
459 Chrysohoou C., Stefanadis C. The role of endothelial function on paroxysmal and chronic
460 atrial fibrillation. *Circ J.* 2014; 130.
- 461 17. Thijssen DHJ, Bruno RM, van Mil ACCM, Holder SM, Fata F, Greyling A, et al. Expert
462 consensus and evidence-based recommendations for the assessment of flow-mediated
463 dilation in humans. *Eur Heart J.* 2019; 40: 2534-2547.
- 464 18. Gemignani V, Bianchini E, Fata F, Giannarelli C, Plantinga Y, Ghiadoni L, et al. Ultrasound
465 measurement of the brachial artery flow-mediated dilation without ECG gating. *Ultrasound
466 Med Biol.* 2008; 34: 385-91.
- 467 19. Junejo RT, May S, Alsalahi S, Alali M, Ogoh S and Fisher JP. Cerebrovascular carbon
468 dioxide reactivity and flow-mediated dilation in young healthy South Asian and Caucasian
469 European men. *American Journal of Physiology-Heart and Circulatory Physiology.* 2020; 318:
470 H756-H763.
- 471 20. Atkinson G and Batterham AM. Allometric scaling of diameter change in the original
472 flow-mediated dilation protocol. *Atherosclerosis.* 2013; 226: 425-427.
- 473 21. Felmeden DC, Spencer CGC, Chung NAY, Belgore FM, Blann AD, Beevers DG, et al.
474 Relation of thrombogenesis in systemic hypertension to angiogenesis and endothelial
475 damage/dysfunction (a Substudy of the Anglo-Scandinavian Cardiac Outcomes Trial
476 [ASCOT]). *The American Journal of Cardiology.* 2003; 92: 400-405.
- 477 22. Kizhakekuttu TJ and Widlansky ME. Natural antioxidants and hypertension: promise and
478 challenges. *Cardiovasc Ther.* 2010; 28: e20-32.
- 479 23. Schiffrin EL. The immune system: role in hypertension. *Can J Cardiol.* 2013; 29: 543-8.
- 480 24. Guo Y, Lip GY and Apostolakis S. Inflammation in atrial fibrillation. *J Am Coll Cardiol.*
481 2012; 60: 2263-70.

- 482 25. Dharmashankar K and Widlansky ME. Vascular endothelial function and hypertension:
483 insights and directions. *Curr Hypertens Rep.* 2010; 12: 448-455.
- 484 26. Harada M, Van Wagoner DR and Nattel S. Role of inflammation in atrial fibrillation
485 pathophysiology and management. *Circulation journal : official journal of the Japanese*
486 *Circulation Society.* 2015; 79: 495-502.
- 487 27. Tousoulis D, Zisimos K, Antoniades C, Stefanadi E, Siasos G, Tsioufis C, et al. Oxidative
488 stress and inflammatory process in patients with atrial fibrillation: the role of left atrium
489 distension. *Int J Cardiol.* 2009; 136: 258-62.
- 490 28. Guazzi M, Casali M, Berti F, Rossoni G, Colonna VD and Guazzi MD. Endothelium-
491 mediated modulation of ergoreflex and improvement in exercise ventilation by acute
492 sildenafil in heart failure patients. *Clin Pharmacol Ther.* 2008; 83: 336-41.
- 493 29. Li Q, Youn J-Y and Cai H. Mechanisms and consequences of endothelial nitric oxide
494 synthase dysfunction in hypertension. *J Hypertens.* 2015; 33: 1128-1136.
- 495 30. Cai H, Li Z, Goette A, Mera F, Honeycutt C, Feterik K, et al. Downregulation of
496 endocardial nitric oxide synthase expression and nitric oxide production in atrial fibrillation:
497 potential mechanisms for atrial thrombosis and stroke. *Circulation.* 2002; 106: 2854-8.
- 498 31. Fares F, Smith Y, Azzam N, Zafrir B, Lewis BS and Amir O. The 894G Allele of the
499 Endothelial Nitric Oxide Synthase 3 (eNOS) is Associated with Atrial Fibrillation in Chronic
500 Systolic Heart Failure. *J Atr Fibrillation.* 2012; 5: 757-757.
- 501 32. Siasos G, Mazaris S, Zisimos K, Oikonomou E, Kokkou E, Konsola T, et al. THE IMPACT OF
502 ATRIAL FIBRILLATION ON ENDOTHELIAL DYSFUNCTION. *J Am Coll Cardiol.* 2015; 65: A477.
- 503 33. Modena MG, Bonetti L, Coppi F, Bursi F and Rossi R. Prognostic role of reversible
504 endothelial dysfunction in hypertensive postmenopausal women. *J Am Coll Cardiol.* 2002;
505 40: 505-10.

506 34. Ghiadoni L, Taddei S and Virdis A. Hypertension and endothelial dysfunction: therapeutic
507 approach. *Curr Vasc Pharmacol.* 2012; 10: 42-60.

508 **Legends**

509 **Figure 1**

510 Medication use by class of drugs

511 ACE inhibitor = Angiotensin converting enzyme; ARB = Angiotensin receptor blocker

512 **Figure 2**

513 Complex interplay between hypertension, AF, oxidative stress, inflammation and
514 endothelial dysfunction

515 **Table 1**

516 Descriptive data are presented as numbers (with percentages). Normally distributed data
517 are expressed as mean \pm standard deviation. Non-normally distributed data are displayed as
518 median with interquartile ranges. Statistical differences were tested for matched groups
519 using an independent t-test for normally distributed data and Mann-Whitney U test for non-
520 normally distributed data. Categorical data was compared using Chi-square test. Where Chi-
521 square test was not valid, Fisher's Exact Test was used. Significance $p \leq 0.05$. - = unable to
522 calculate p value as sample size too small/statistical test not valid

523 AF = atrial fibrillation; TIA = Transient Ischaemic Attack; COPD = Chronic Obstructive
524 Pulmonary Disease; BMI = Body Mass Index; bpm = beats per minute; BP = blood pressure;
525 HbA1c = Haemoglobin A1C; CrCl = Creatine Clearance (Cockcroft-Gault method); TSH =
526 Thyroid Stimulating Hormone; INR = International Normalised Ratio

527

528 **Table 2**

529

530 Normally distributed data are expressed as mean [95% confidence intervals (CI)]. Identified
531 by superscript a. Non-normally distributed data are displayed as median [95% CI]. Identified
532 by superscript b. Statistical differences were tested for matched groups using independent
533 t-test (for parametric data) or Mann-Whitney U test (for non-parametric data). Significance
534 $p \leq 0.05$.

535 AF = atrial fibrillation; FMD = flow-mediated dilatation; FMDc = FMD % mean [95% CI]
536 adjusted for baseline diameter

537 **Table 3**

538 Descriptive data are presented as numbers (with percentages). Normally distributed data
539 are expressed as mean \pm standard deviation. Non-normally distributed data are displayed as
540 median with interquartile ranges. Statistical differences were tested using an independent t-
541 test for normally distributed data and Mann-Whitney U test for non-normally distributed
542 data. Categorical data was compared using Chi-square test. Where Chi-square test was not
543 valid, Fisher's Exact Test was used. Significance $p \leq 0.05$. - = unable to calculate p value as
544 sample size too small/statistical test not valid

545 AF = atrial fibrillation; TIA = Transient Ischaemic Attack; COPD = Chronic Obstructive
546 Pulmonary Disease; BMI = Body Mass Index; bpm = beats per minute; BP = blood pressure;
547 HbA1c = Haemoglobin A1C; CrCl = Creatine Clearance (Cockcroft-Gault method); TSH =
548 Thyroid Stimulating Hormone; INR = International Normalised Ratio

549

550 **Table 4**

551 Normally distributed data are expressed as mean [95% confidence intervals (CI)]. Identified
552 by superscript a. Non-normally distributed data are displayed as median [95% CI]. Identified
553 by superscript b. Statistical differences were tested using independent t-test (for parametric
554 data) or Mann-Whitney U test (for non-parametric data). Significance $p \leq 0.05$.

555 AF = atrial fibrillation; FMD = flow-mediated dilatation

556

557 **Table 5**

558 Normally distributed data are expressed as mean \pm standard deviation for descriptive data
559 and mean [95% confidence interval (CI)] otherwise. Identified by superscript a. Non-
560 normally distributed data are displayed as median with interquartile ranges for descriptive
561 data and median [95% CI] otherwise. Identified by superscript b. Normality test was
562 performed using Shapiro-Wilk test. Statistical differences were tested using paired t-test (if
563 passed) or Wilcoxon signed rank test (if failed). Significance $p \leq 0.05$. AF = atrial fibrillation;
564 bpm = beats per minute; BP = blood pressure; FMD = flow mediated dilatation

Figure 1

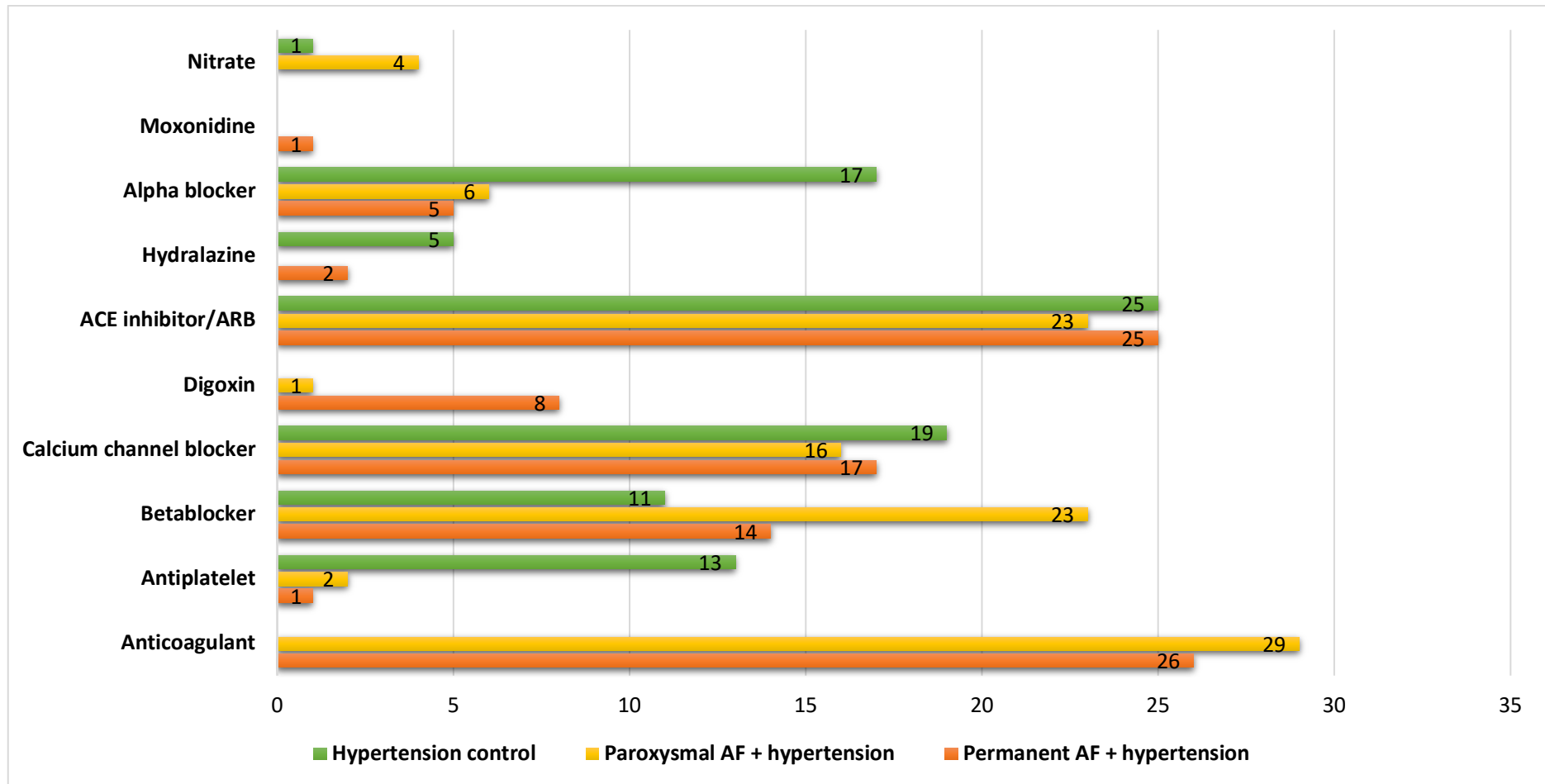


Figure 2

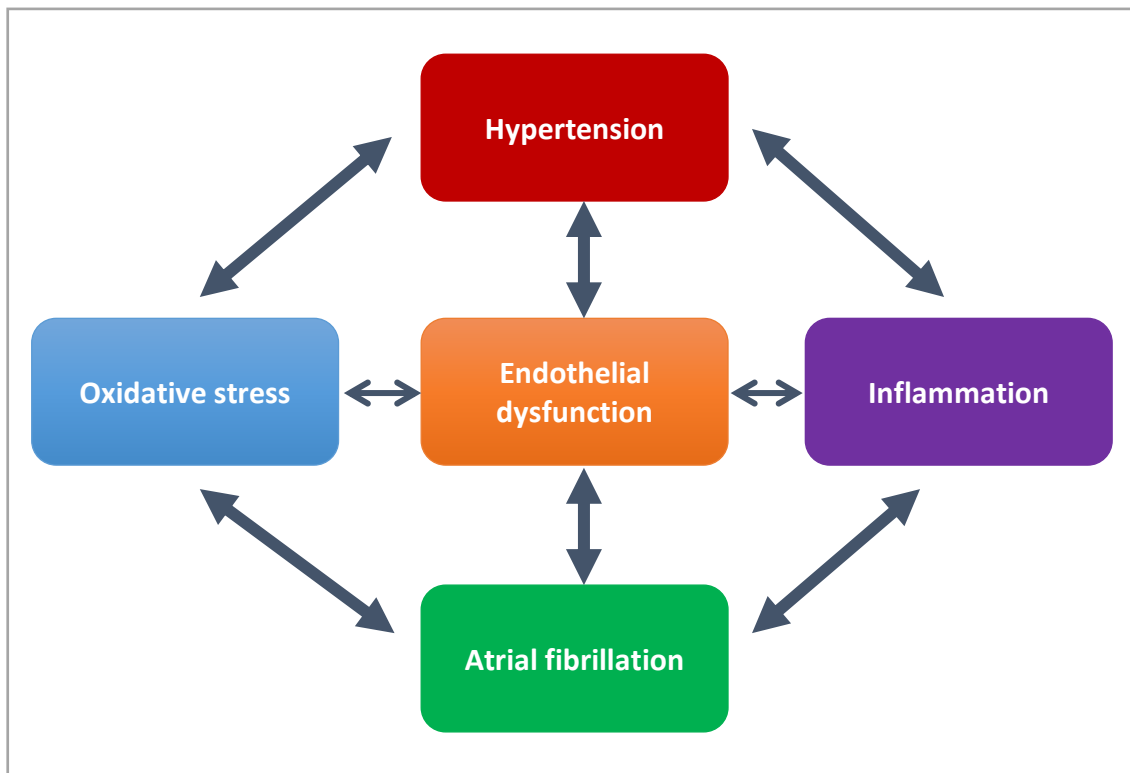


Table 1 – Demographics and clinical characteristics of matched AF (and hypertension) group and hypertension control group

	AF + hypertension group (n = 40)	Hypertension control group (n = 20)	Matched groups p
Demographics			
Age, years	66 ± 7	65 ± 7	0.71
Sex			
Male	29	15	0.84
Female	11	5	
Ethnicity			
Caucasians, n (%)	34 (85%)	10 (50%)	-
Blacks, n (%)	3 (7.5%)	6 (30%)	
Asians, n (%)	3 (7.5%)	3 (15%)	
Mixed, n (%)	0 (0%)	1 (5%)	
Clinical characteristics			
Heart failure, n (%)	2 (5%)	0 (0%)	0.55
IHD, n (%)	5 (12.5%)	5 (25%)	0.28
Diabetes Mellitus, n (%)	10 (25%)	8 (40%)	0.23
Previous stroke/TIA, n (%)	5 (12.5%)	5 (25%)	0.28
Asthma/COPD, n (%)	5 (12.5%)	2 (10%)	0.57
Chronic liver disease, n (%)	0 (0%)	0 (0%)	-
Chronic kidney disease, n (%)	1 (2.5%)	5 (25%)	0.01
Anaemia, n (%)	0 (0%)	2 (10%)	0.11
Thyroid disorder, n (%)	3 (7.5%)	4 (20%)	0.21
Hypercholesterolaemia, n (%)	19 (47.5%)	11 (55%)	0.58
Arthritis, n (%)	24 (60%)	8 (40%)	0.14
CHA ₂ DS ₂ -VASc score	2 [2 – 4]	3 [1 – 4]	0.74
HAS-BLED score	1 [1 – 1]	2 [1 – 2]	0.06
Smoking status			
Never smoked, n (%)	19 (47.5%)	13 (65%)	-
Ex-smoker, n (%)	18 (45%)	7 (35%)	
Current, n (%)	3 (7.5%)	0 (0%)	
Alcohol			
None, n (%)	9 (22.5%)	5 (25%)	0.54
Recommended, n (%)	31 (77.5%)	15 (75%)	
Height (cm)	170.1 ± 8.9	169.4 ± 11.1	0.80
Weight (kg)	95.5 ± 18.4	92.3 ± 14.7	0.50
BMI (kg/m ²)	32.9 ± 5.2	32.1 ± 4.2	0.58
Heart rate (bpm)	70 [60 – 82]	63 [58 – 67]	0.02
Systolic BP (mm/Hg)	142 [133 – 152]	148 [135 – 175]	0.12
Diastolic BP (mm/Hg)	83 ± 14	85 ± 13	0.53
Mean Arterial Pressure (MAP) (mm/Hg)	103 ± 15	109 ± 16	0.23
HbA1c (mmol/mol)	41 [39 – 48]	45 [38 – 56]	0.32
CrCl (mL/min)	98.8 ± 29.6	85 ± 28.1	0.09
Ejection fraction (%)	58 ± 11	62 ± 7	0.14

Table 2 – Differences in flow mediated dilatation (FMD) between matched AF (and hypertension) and hypertension control groups – cross sectional comparison

	AF + hypertension group (n = 40)	Hypertension control group (n = 20)	Matched groups p
Baseline diameter (mm)	4.6 [4.4 – 4.9] ^a	5.2 [4.8 – 5.6] ^a	0.02
Peak diameter (mm)	4.9 [4.6 – 5.2] ^a	5.4 [5.0 – 5.8] ^a	0.03
Absolute FMD change (mm)	0.2 [0.1 – 0.3] ^b	0.2 [0.1 – 0.3] ^b	0.61
FMD (%)	4.6 [2.6 – 5.9] ^b	2.6 [1.9 – 5.3] ^b	0.25
FMDc (%)	4.9 [3.8 – 6.0] ^a	4.4 [2.7 – 6.0] ^a	0.60
Time to peak diameter (sec)	58 [40 – 90] ^b	36 [21 – 65] ^b	0.07
Shear rate (Positive shear rate area to peak) [sec.-1]	4421 [2800 – 6077] ^b	3300 [1296 – 6887] ^b	0.41

Table 3 – Demographics and clinical characteristics of permanent AF (and hypertension) group and paroxysmal AF (and hypertension) group

	Permanent AF + hypertension group (n = 30)	Paroxysmal AF + hypertension group (n = 31)	p
Demographics			
Age, years	70 ± 8	72 ± 11	0.64
Sex			0.46
Males	22	20	
Females	8	11	
Ethnicity			-
Caucasians, n (%)	28 (93.3%)	25 (80.6%)	
Blacks, n (%)	1 (3.3%)	3 (9.7%)	
Asians, n (%)	1 (3.3%)	3 (9.7%)	
Mixed, n (%)	0 (0%)	0 (0%)	
Clinical characteristics			
Heart failure, n (%)	3 (10%)	0 (0%)	0.11
IHD, n (%)	0 (0%)	10 (32.3%)	<0.001
Diabetes Mellitus, n (%)	7 (23.3%)	7 (22.6%)	0.81
Previous stroke/TIA, n (%)	5 (16.7%)	2 (6.5%)	0.26
Asthma/COPD, n (%)	9 (30%)	4 (12.9%)	0.10
Chronic liver disease, n (%)	0 (0%)	0 (0%)	-
Chronic kidney disease, n (%)	0 (0%)	1 (3.2%)	1.00
Anaemia, n (%)	1 (3.3%)	1 (3.2%)	1.00
Thyroid disorder, n (%)	1 (3.3%)	4 (12.9%)	0.35
Hypercholesterolaemia, n (%)	14 (46.7%)	15 (48.4%)	0.89
Arthritis, n (%)	14 (46.7%)	16 (51.6%)	0.70
CHA ₂ DS ₂ -VASc score	3 [2 – 4]	3 [2 – 4]	0.56
HAS-BLED score	1 [1 – 1]	1 [1 – 1]	0.18
Smoking status			-
Never smoked, n (%)	13 (43.3%)	17 (54.8%)	
Ex-smoker, n (%)	15 (50%)	13 (42%)	
Current, n (%)	2 (6.7%)	1 (3.2%)	
Alcohol			0.85
None, n (%)	9 (30%)	10 (32.3%)	
Recommended, n (%)	21 (70%)	21 (67.7%)	
Height (cm)	169.3 ± 8.4	167.3 ± 10.1	0.40
Weight (kg)	89.6 ± 19.1	87.2 ± 21.7	0.66
BMI (kg/m ²)	31.1 ± 5.1	31.0 ± 6.3	0.95
Heart rate (bpm)	77 [68 – 86]	62 [58 – 70]	0.003
Systolic BP (mm/Hg)	140 [128 – 148]	144 [134 – 153]	0.24
Diastolic BP (mm/Hg)	81 ± 13	76 ± 15	0.16
Mean Arterial Pressure (MAP) (mm/Hg)	101 ± 12	101 ± 16	0.87
HbA1c (mmol/mol)	41 [38 – 46]	41 [40 – 51]	0.94
CrCl (mL/min)	86.2 ± 30.8	75.9 ± 38.1	0.72
Ejection fraction (%)	55 [55 – 62]	62 [55 – 68]	0.22

Table 4 – Differences in flow mediated dilatation (FMD) between permanent AF and paroxysmal AF groups – cross sectional comparison

	Permanent AF + hypertension group (n = 30)	Paroxysmal AF + hypertension group (n = 31)	P
Baseline diameter (mm)	4.5 [4.2 – 5.0] ^b	4.8 [4.6 – 5.1] ^b	0.67
Peak diameter (mm)	4.7 [4.4 – 5.2] ^b	5.2 [4.6 – 5.3] ^b	0.49
Absolute FMD change (mm)	0.1 [0.1 – 0.2] ^b	0.3 [0.2 – 0.4] ^b	0.01
FMD (%)	3.1 [2.3 – 4.8] ^b	5.9 [4.0 – 8.1] ^b	0.02
FMDc (%)	3.9 [2.8 – 5.0] ^a	5.9 [4.8 – 7.0] ^a	0.01
Time to peak diameter (sec)	50 [29 – 85] ^b	80 [36 – 93] ^b	0.23
Shear rate (Positive shear rate area to peak) [sec.-1]	4592 [2278 – 5734] ^b	4800 [2800 – 8102] ^b	0.40

Table 5 – Haemodynamic and FMD data for longitudinal comparison of Permanent AF (and hypertension) group

	Permanent AF + hypertension group (Baseline) [n = 30]	Permanent AF + hypertension group (Follow up) [n = 30]	p
Clinical characteristics	Mean ± SD / Median [IQR]	Mean ± SD / Median [IQR]	
Weight (kg)	89.6 ± 19.1	90.1 ± 19.4	0.13
BMI (kg/m ²)	31.1 ± 5.1	31.2 ± 5.2	0.11
Heart rate (bpm)	77 ± 18	72 ± 17	0.01
Systolic BP (mm/Hg)	140 [128 – 148]	131 [122 – 146]	0.03
Diastolic BP (mm/Hg)	81 ± 13	77 ± 12	0.02
Mean Arterial Pressure (MAP) (mm/Hg)	100 ± 9	97 ± 13	0.01
CHA ₂ DS ₂ -VASc score	3 [2 – 4]	3 [2 – 4]	1.00
HAS-BLED score	1 [1 – 1]	1 [1 – 1]	1.00
FMD measurements	Mean [95% CI] ^a / Median [95% CI] ^b	Mean [95% CI] ^a / Median [95% CI] ^b	
Baseline diameter (mm)	4.5 [4.2 – 5.0] ^b	4.4 [4.1 – 5.1] ^b	0.34
Peak diameter (mm)	4.9 [4.5 – 5.3] ^a	4.8 [4.5 – 5.2] ^a	0.69
Absolute FMD change (mm)	0.14 [0.11 – 0.25] ^b	0.20 [0.17 – 0.28] ^b	0.15
FMD (%)	3.1 [2.3 – 4.8] ^b	5.2 [3.9 – 6.5] ^b	0.09
FMDc (%)	4.0 [3.0 – 4.9] ^a	5.1 [4.2 – 6.1] ^a	0.09
Time to peak diameter (sec)	50 [29 – 85] ^b	72 [36 – 102] ^b	0.29
Shear rate stimulus (Positive shear rate area to peak) [sec.-1]	4592 [2278 – 5734] ^b	3961 [3526 – 8190] ^b	0.54