

1 **Human cerebral collateral arteriole function in normal cognition, mild cognitive**  
2 **impairment and dementia subjects**

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19 **Keywords:** cerebrovascular disease, Alzheimer's disease, vascular dementia, endothelial

20 function, disease model

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22 **New and Noteworthy:** We present novel findings that brain collateral arteriole function did not  
23 differ among cognitively normal, mild cognitive impairment and dementia (Alzheimer's disease,  
24 vascular dementia) subjects. Although arteriole function was impaired by vascular stressors ( $\beta$ -  
25 amyloid, palmitic acid and medin), responses did not differ between those with or without  
26 dementia. The cognitive dysfunction in dementia disorders is not attributable to differences in

## Collateral circulation function and dementia disorders

27 baseline brain collateral circulation function but may be influenced by vascular exposure to  
28 metabolic stressors.

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**Abstract**

Clinical and preclinical studies suggest a link between cardiovascular disease and dementia disorders but the role of collateral brain circulation in cognitive dysfunction remains unknown. We aim to test the hypothesis that leptomeningeal arteriole (LMA) function and response to metabolic stressors differ among dementia, mild cognitive impairment (MCI) and normal cognition (CN) subjects. Following rapid autopsy, LMA were isolated from CN (N=10), MCI (N=12) or dementia subjects (N=42, Alzheimer's disease [AD], vascular dementia [VaD], or Other Dementia) and endothelial and smooth muscle-dependent function were measured at baseline and following exposure to  $\beta$ -amyloid (A $\beta$ 42 2  $\mu$ M), palmitic acid (PA 150  $\mu$ M) or medin (5 $\mu$ M) and compared. There was no difference among the groups in baseline endothelial function (maximum dilation to acetylcholine: CN 74.1 $\pm$ 9.7, MCI 67.1 $\pm$ 4.8, AD 74.7 $\pm$ 2.8, VaD 72.0 $\pm$ 5.3, Other Dementia 68.0 $\pm$ 8.0%) and smooth muscle-dependent function (CN 93.4 $\pm$ 3.0, MCI 83.3 $\pm$ 4.1, AD 91.8 $\pm$ 1.7, VaD 91.7 $\pm$ 2.4, Other Dementia 87.9 $\pm$ 4.9%). There was no correlation between last cognitive function score and baseline endothelial or smooth muscle-dependent function. LMA endothelial function and to a lesser extent smooth muscle-dependent function were impaired post-treatment with A $\beta$ 42, PA and medin. Post-treatment LMA responses were not different between CN/MCI versus dementia subjects. Baseline responses and impaired vasoreactivity following treatment with metabolic stressors did not differ among CN, MCI and dementia subjects. The results suggest that the cognitive dysfunction in dementia disorders is not attributable to differences in baseline brain collateral circulation function but may be influenced by exposure of the vasculature to metabolic stressors.

52 Clinical and preclinical data show that vascular disease and cardiovascular risk factors  
53 are associated with Alzheimer's disease (AD) and AD-related dementia disorders(2, 7).  
54 Impaired cerebrovascular hemodynamics found in early AD patients(5) and pathologic vascular  
55 changes suggesting altered arteriolar vasoregulation(11) and hemodynamic disturbances(13)  
56 point to the critical role of vascular dysfunction in disease pathogenesis. Leptomeningeal  
57 collateral arterioles (LMA) link the 3 major arterial territories over the brain surface. Although it  
58 is known that LMAs play a significant role in cerebrovascular autoregulation, represent an  
59 important compensatory mechanism in cerebrovascular disease and affect prognosis following  
60 stroke(3, 16), the role of LMA function in the pathophysiology of dementia disorders remains  
61 poorly understood despite pathologic observations that LMA are widely affected in AD(23).  
62 Additionally, vascular dysfunction may result from exposure to metabolic stressors such as fatty  
63 acids or amyloidogenic proteins such as  $\beta$ -amyloid ( $A\beta$ ) or medin (aging-associated vascular  
64 amyloid protein)(15, 17, 21, 25, 26) but the differential susceptibility of brain collateral circulation  
65 between patients with dementia disorders and those without remains unknown. Using a unique  
66 and novel model using ex-vivo human LMA isolated following rapid autopsy of brain donors(25),  
67 the study aims to test the hypothesis that LMA endothelium-dependent function (EFn) and  
68 smooth muscle-dependent function (SMFn) are impaired in patients with dementia (AD,  
69 vascular dementia [VaD] or other causes) when compared to patients with normal cognition  
70 (CN) or mild cognitive impairment (MCI). We also aim to test the hypothesis that LMA from  
71 patients with dementia have worse response when exposed to vascular metabolic stressors  
72 ( $A\beta$ , palmitic acid [PA] or medin) compared to LMA from CN/MCI patients.

73

## 74 **METHODS**

75 Consecutive brain donor LMA were isolated from 64 cadavers (88.7 $\pm$ 1.2 years, 32  
76 females) following rapid autopsy (post-mortem interval, PMI, or the time elapsed from official  
77 declaration of death to brain removal, 2.8 $\pm$ 0.1 hour) who prior to expiration provided informed

78 consent for brain donation (program information at [www.brainandbodydonationprogram.org](http://www.brainandbodydonationprogram.org))(1).  
79 Program operations were approved by the Banner-Sun Health Research Institute (BSHRI)  
80 Institutional Review Board (IRB) and the study was approved by the Phoenix VA IRB.  
81 Each participant in the BSHRI Brain and Body Donation Program undergoes general  
82 neurological examinations and neurological functional battery tests annually and clinical  
83 diagnostic classification (CN, MCI, specific dementia or neurologic disorders) is performed after  
84 each annual assessment at a consensus conference attended by neurologists, psychiatrists and  
85 neuropsychologists(1). Final clinicopathologic diagnoses are assigned after death following  
86 review of all standardized clinical data, the most recent private medical records and  
87 neuropathological examination findings. All CN (N=10) and MCI (N=12) subjects did not meet  
88 criteria for dementia at the time of death and are without a major neuropathological diagnosis.  
89 AD (N=34) is defined as at least intermediate or high NIA-Reagan criteria(12). VaD cases  
90 (N=13) are classified using modified NINDS-AIREN criteria(24). All subjects had final  
91 pathologic diagnoses. In 10 donors, AD and VaD were both present. Donors with dementia  
92 whose final pathologic diagnoses were not AD or VaD were grouped under Other Dementia  
93 (N=5, 2 Parkinson's disease, 2 Dementia with Lewy bodies, 1 Pick's disease). Donors were  
94 also classified as to whether cerebral amyloid angiopathy (CAA) was present (N=31) or not  
95 (N=14). Clinical and pathologic data are summarized in Table 1.

96 Methodologic details on vasoreactivity procedures were reported previously(17, 18). In  
97 brief, LMA were cannulated, pressurized in sequence to 30 mm Hg and then to physiologic 60  
98 mm Hg pressure (30 minutes stabilization) and precontracted to ~60% of maximum diameter  
99 with endothelin-1 ( $2-50 \times 10^{-9} \text{M}$ ). Baseline dilator responses to acetylcholine ( $10^{-9}$ - $10^{-4} \text{M}$  to  
100 measure EFn) and papaverine or diethylenetriamine NONOate ( $10^{-4} \text{M}$  to measure SMFn) were  
101 measured by videomicroscopy, and LMA arteriole responses averaged for each donor. EFn  
102 (maximum acetylcholine dilation and acetylcholine half-maximal effective concentration  
103 [EC50])(17, 18) and SMFn were compared among 1) CN, MCI, AD and Other Dementia

104 subjects, and 2) CN, MCI, VaD and Other Dementia subjects (separate analyses were  
105 performed due to overlap in subjects having both AD and VaD). LMA function were also  
106 compared between donors with cerebral amyloid angiopathy (CAA) versus those without CAA.

107       Following washout, some LMA were exposed for 1 hour to one of the following: A $\beta$ 42 (2  
108  $\mu$ M, Anaspec, Fremont CA), PA (150  $\mu$ M, the most common saturated fatty acid in the Western  
109 diet) or medin (5  $\mu$ M, recombinantly produced as detailed previously)(17) and a second  
110 measurement of dilator responses were obtained; absolute differences with baseline LMA  
111 responses were computed and compared between CN/MCI subjects versus subjects with  
112 AD/VaD. The doses selected represent physiologically relevant concentrations observed in-vivo  
113 in humans(15, 17, 19).

114       Group comparisons were performed using one-way analysis of variance (ANOVA) with  
115 pairwise Holm-Sidak method for normally distributed data and ANOVA on ranks with pairwise  
116 Kruskal-Wallis for non-normally distributed data. Baseline and post-treatment dilator responses  
117 were compared using paired t-test. Post-treatment responses (change in dilator response) were  
118 compared between CN/MCI versus AD/VaD subjects and between donors with CAA versus  
119 those without CAA using unpaired t-test. Comparisons between 2 groups with non-normally  
120 distributed data were done using Mann Whitney rank sum test. Correlation analyses were done  
121 using Pearson's correlation. Chi-square analysis or Fisher exact test was used for categorical  
122 variables. Data are expressed as means $\pm$ standard error of means. In case of missing data, no  
123 imputation was performed. Significant p-value was set at p<0.05.

124

## 125 **RESULTS:**

126       The donor pool consisted of elderly subjects, most whom had cardiovascular risk factors  
127 or disease (Table 1). Donors with Other Dementia (N=5) were significantly younger than CN,  
128 MCI, AD or VaD donors. There were no significant differences in gender composition, PMI,  
129 presence of cardiovascular co-morbidities (coronary or peripheral arterial disease, hypertension,

130 diabetes, hyperlipidemia) among the groups (Table 1). As expected, there were significant  
131 differences in cognitive function (last MMSE score) among the groups.

132         There were no significant differences in post-mortem brain weight (Table 1). Total senile  
133 amyloid plaque density was significantly higher in AD donors compared to Other Dementia  
134 donors, but did not differ when compared with CN or MCI donors. Although CAA score (density  
135 of amyloidotic vessels) did not differ among the groups, the proportion of donors with CAA was  
136 higher in AD group versus Other Dementia group, but not with CN or MCI groups.

137         There were no significant differences in unpressurized vessel diameters among the  
138 groups, although Other Dementia subjects showed a trend towards larger baseline values  
139 (Table). Average arteriole diameters increased from 0 to 30 mm Hg intraluminal pressure.  
140 There were no significant differences in vessel diameters among CN, MCI, AD and VaD  
141 subjects; however, vessel diameter was significantly higher in Other Dementia subjects at 30,  
142 60 mm Hg and maximum dilation versus other groups, although the changes in diameters  
143 following pressurization did not differ among the groups. The average maximum vessel  
144 diameter is  $177\pm 5\ \mu\text{m}$ .

145         There was no significant difference in EFn or SMFn among CN, MCI, AD and Other  
146 dementia subjects, and among CN, MCI, VaD and Other dementia subjects (Figure 1). In  
147 separate analyses, donors with both AD+VaD were compared to CN, MCI, AD without VaD and  
148 Other Dementia. AD+VaD maximum endothelium-dependent dilation to acetylcholine  
149 ( $72.1\pm 6.2\%$ ), acetylcholine logM EC50 ( $-6.0\pm 0.4$ ) and smooth muscle-dependent dilation  
150 ( $91.1\pm 3.1\%$ ) did not significantly differ when compared to the other groups.

151         There was no significant correlation between last mini-mental state examination (MMSE)  
152 score and endothelial function or smooth muscle function (Figure 2).

153         There was no difference between subjects with CAA (N=31) and those without CAA  
154 (N=14) in terms of EFn (maximum acetylcholine dilation  $71.0\pm 3.2$  versus  $81.1\pm 3.8\%$  and

155 acetylcholine log M EC50  $-5.8 \pm 0.2$  versus  $-6.2 \pm 0.2$ , respectively, both  $p=NS$ ) and SMFn  
156 ( $91.5 \pm 1.7$  versus  $93.9 \pm 1.7\%$ , respectively,  $p=NS$ ).

157 Exposure of LMAs to physiologically relevant doses of A $\beta$ 42, PA and medin caused  
158 profound impairment in EFn ( $-31.4 \pm 5.9$ ,  $-32.8 \pm 8.9$ ,  $-39.6 \pm 7.2\%$  versus baseline control  
159 response, respectively) while also causing impairment in SMFn ( $-11.3 \pm 5.2$ ,  $-29.8 \pm 8.7$ , -  
160  $16.6 \pm 6.1\%$  versus baseline control response, respectively) (Fig. 3). There was no significant  
161 difference in response to A $\beta$ 42, PA or medin between subjects with CN/MCI versus those with  
162 AD/VaD.

163

#### 164 **DISCUSSION:**

165 Cerebrovascular disease leading to brain hypoperfusion is believed to contribute to the  
166 pathophysiology of VaD, sporadic AD and aging-related dementia(10). Collateral perfusion  
167 through LMA is a critical component of cerebrovascular autoregulation; it is emerging as a key  
168 determinant of clinical outcome following ischemic stroke(16). A preclinical investigation  
169 showed impaired LMA vasoreactivity in a hypertensive rat model that may contribute to  
170 perfusion deficits(4). Leptomeningeal atherosclerosis was associated with AD  
171 neuropathological lesions suggesting that atherosclerosis-induced hypoperfusion contributes to  
172 AD pathology(22). Our observations represent the first measurement and comparison of human  
173 LMA vascular function among CN, MCI, AD, VaD and Other Dementia patients that we know of.  
174 Contrary to our hypotheses, EFn and SMFn (within the limits of our experimental conditions) did  
175 not differ among CN, MCI and dementia subjects suggesting that AD or VaD does not confer  
176 fixed phenotypic alteration in baseline LMA function. Consistent with this finding, there was also  
177 a lack of correlation between LMA vasoreactivity and cognitive function score. Although one  
178 could be tempted to interpret the results as being not supportive of the etiologic contribution of  
179 LMA dysfunction to cognitive dysfunction, our results following LMA exposure to stressors (A $\beta$ ,  
180 PA and medin) suggest an alternative interpretation. The idealized, plasma-free, ex-vivo

181 conditions in our baseline vasoreactivity experiments likely do not reflect the metabolic milieu  
182 present in-vivo in terms of presence of metabolic abnormalities that could impair microvascular  
183 function (such as hyperlipidemia, exposure to circulating amyloid proteins, proinflammatory  
184 state), and in the absence of vascular metabolic stressors as represented by our baseline  
185 conditions, no significant differences in microvascular response were noted. However, the in-  
186 vivo metabolic milieu may be different among the groups tested. Adverse metabolic milieu was  
187 simulated in our ex-vivo model following exposure to A $\beta$ , PA or medin. Post-treatment arteriole  
188 vasoreactivity showed consistent impaired responses to a malignant metabolic milieu,  
189 conditions that are known to exist in vivo. Because the doses of vascular stressors used in this  
190 study are within levels reported in humans(15, 17, 19), living conditions/in-vivo states where  
191 these conditions exist could recapitulate our post-treatment experimental conditions and cause  
192 microvascular dysfunction, which especially in the setting of periodic and chronic exposure,  
193 could potentially lead to brain hypoperfusion. Future studies focusing on differences in the  
194 magnitude and duration of exposure *in-vivo* to vascular metabolic stressors in subjects with or at  
195 risk for AD or VaD versus CN subjects may yield insights on how microvascular dysfunction  
196 contributes to cognitive dysfunction that might also serve as therapeutic targets for modulation.

197         The study is the first that we know of showing impaired brain collateral circulation  
198 function following exposure to PA, a well-established dietary cardiovascular risk factor. Earlier  
199 efforts by our group showed that A $\beta$ 42(25, 26) and medin(17) caused LMA endothelial  
200 dysfunction. The current study vastly expanded on these datasets, and in doing so, we not only  
201 validated prior observations but the larger sample size also allowed comparisons of differential  
202 responses between dementia and non-demented subjects, a novel aspect of this study.  
203 Interestingly, we also showed the lack of differences in LMA EF<sub>n</sub> and SMF<sub>n</sub> between donors  
204 with and without CAA suggesting that soluble species, and not the insoluble amyloid forms,  
205 confer vascular toxicity. Finally, the ability to measure arteriole function following rapid autopsy  
206 in a large series of brain donors points to the potential of this new human tissue model to study

207 vascular determinants of neurodegenerative diseases that could bridge the translational gap  
208 between preclinical transgenic mouse models and human clinical trials.

209           In terms of basal myogenic tone, it is interesting to note that the gradual increase in  
210 LMA diameters from 0 to 30 mm Hg was similar to response observed in rat posterior cerebral  
211 artery(20) but rat posterior cerebral arteries, unlike human LMA, showed constriction response  
212 (signifying development of myogenic tone) when exposed to 60 mm Hg intraluminal pressure.  
213 Future studies should investigate whether human collateral LMA responses differ from  
214 intraparenchymal or penetrating arteriole responses as they may have important implications in  
215 vasoregulation.

216           Our study is limited by the lack of detailed morphological assessment of the LMA  
217 vessels which should be comprehensively studied in the future to gain insights on morphology-  
218 function relationships. We also did not study the role of penetrating and intracerebral arterioles  
219 and capillaries which play critical roles in cerebral perfusion. We lack data on differential  
220 responses to shear stress as well as in-vivo status of collateral brain circulation from imaging  
221 data that could have provided additional context to our findings. An additional study limitation is  
222 the lack of therapeutic intervention to restore microvascular function following exposure to  
223 vascular stressors that would inform mechanisms of injury and may uncover differential  
224 responses among CN, MCI and dementia subjects, if they exist. Despite this limitation, our  
225 previous work on a more limited number of donor leptomeningeal arterioles did point to specific  
226 signaling mechanisms underlying the vascular injury. We showed that A $\beta$ 42(26) and medin(6,  
227 17) induced endothelial cell oxidative and nitrative stress and reduced nitric oxide bioavailability  
228 that were reversed by agents that have antioxidant effects. Additionally we showed that  
229 inhibition of receptor for advanced glycation end product (RAGE) also restored LMA function  
230 following exposure to medin(17), suggesting that RAGE may be a common pathway by which  
231 A $\beta$ (8, 9) and medin affect the microvasculature. Similar to the LMA response to PA shown in  
232 this study, human adipose arterioles using similar experimental setup also showed profound

233 endothelial dysfunction when exposed to fatty acid-rich very low density lipoprotein lipolysis  
234 products(15) and PA, like A $\beta$  and medin, is known to impair endothelial cell nitric oxide  
235 production(14). Our findings support the need to systematically study the acute and chronic  
236 effects of PA and fatty acids, enriched in Western diet, on cerebrovascular function.

237         The impaired treatment responses to A $\beta$ 42, PA and medin are likely not due to impaired  
238 tissue viability following baseline response. In our prior work, co-treatment of A $\beta$ 42 with  
239 phosphatidic acid-containing nanoliposomes(26) and co-treatment of medin with antioxidant  
240 polyethylene glycol superoxide dismutase or RAGE inhibitor FPS-ZM1(17) fully restored  
241 microvascular function, suggesting that vessels continue to be viable in these experimental  
242 conditions. It is important to note that in assessing smooth muscle-dependent dilator response,  
243 we only used one high concentration ( $10^{-4}$ M) dose and did not perform full dose-response  
244 studies which might reveal differences not seen with the high dose.

245         In summary, our results show the novel finding that LMA endothelial and smooth  
246 muscle-dependent function did not differ among CN, MCI and dementia subjects while impaired  
247 vasoreactivity was seen in, yet did not differ, among CN, MCI and dementia subjects following  
248 exposure to A $\beta$ , PA and medin. Unlike their emerging critical role in stroke pathophysiology, the  
249 role of collateral cerebral circulation in the pathophysiology of cognitive dysfunction in AD and  
250 related dementia disorders remains to be fully explored and defined.

251

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261 **Declaration of Interest-None**

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362

Table 1. Demographics, Pathology and Leptomeningeal Arteriole Tone

	Cognitively Normal (N=10)	Mild Cognitive Impairment (N=12)	Alzheimer's Disease (N=34) <sup>a</sup>	Vascular Dementia (N=13) <sup>a</sup>	Other Dementia (N=5)	p-value (CN/MCI/AD, Other)	p-value (CN/MCI/VaD, Other)
<b>A. DEMOGRAPHICS</b>							
<b>Age (years)</b>	<b>90.6±2.2</b>	<b>93.3±2.1</b>	<b>88.4±1.7</b>	<b>94.1±1.5</b>	<b>75.8±2.7</b>	<b>0.04<sup>b</sup></b>	<b>&lt;0.001<sup>b</sup></b>
Female (N/%)	3 (30%)	5 (42%)	19 (56%)	5 (62%)	3 (60%)	NS	NS
PMI (hours)	3.1±0.2	2.9±0.2	2.7±0.1	2.8±0.1	3.4±0.1	NS	NS
<b>Last MMSE</b>	<b>28.4±0.4</b>	<b>25.2±0.9</b>	<b>16.5±1.4</b>	<b>21.1±1.4</b>	<b>14.2±5.6</b>	<b>&lt;0.001<sup>c</sup></b>	<b>&lt;0.001<sup>c</sup></b>
CAD/PAD (N/%)	5 (56%)	9 (75%)	12 (39%)	7 (70%)	2 (50%)	NS	NS
Hypertension (N/%)	6 (67%)	11 (92%)	19 (63%)	6 (60%)	1 (25%)	NS	NS
Diabetes mellitus (N/%)	3 (33%)	5 (42%)	4 (13%)	2 (20%)	1 (25%)	NS	NS
Hyperlipidemia (N/%)	7 (78%)	9 (75%)	22 (71%)	6 (60%)	1 (25%)	NS	NS
<b>B. PATHOLOGY</b>							
Brain weight (g)	1097±32	1041±95	1125±26	1062±34	1101±72	NS	NS
<b>Brain Total Plaque<sup>d</sup></b>	<b>8.4±1.8</b>	<b>7.8±1.7</b>	<b>11.7±0.7</b>	<b>9.9±1.7</b>	<b>1.3±0.6</b>	<b>0.001<sup>e</sup></b>	<b>NS</b>
<b>CAA present (N/%)</b>	<b>6 (67%)</b>	<b>4 (80%)</b>	<b>21 (84%)</b>	<b>6 (67%)</b>	<b>0 (0%)</b>	<b>0.01<sup>f</sup></b>	<b>NS</b>
CAA score <sup>g</sup>	2.6±0.9	2.8±1.2	3.7±0.7	2.9±1.2	0±0	NS	NS
<b>C. LMA TONE</b>							
Diameter, no pressure (µm)	129±7	149±13	139±6	138±12	183±20	NS	NS
<b>Diameter, 30 mm Hg (µm)</b>	<b>147±13</b>	<b>167±12</b>	<b>149±6</b>	<b>144±10</b>	<b>210±12</b>	<b>0.03<sup>h</sup></b>	<b>0.04<sup>h</sup></b>
<b>Diameter, 60 mm Hg (µm)</b>	<b>142±11</b>	<b>168±11</b>	<b>152±6</b>	<b>155±11</b>	<b>211±9</b>	<b>0.01<sup>i</sup></b>	<b>0.02<sup>i</sup></b>
<b>Maximum diameter (µm)</b>	<b>163±14</b>	<b>199±13</b>	<b>166±7</b>	<b>166±12</b>	<b>224±10</b>	<b>0.005<sup>j</sup></b>	<b>0.02<sup>j</sup></b>
Endothelin-1 dose used (x10 <sup>-9</sup> M)	9.7±2.5	7.0±2.3	8.5±21.1	7.4±1.3	12.5±9.5	NS	NS

364

365 Legend: CN-cognitively normal, MCI-mild cognitive impairment, VaD-vascular dementia, PMI-post-mortem interval, MMSE-mini mental  
366 state examination score, CAD-coronary artery disease, PAD-peripheral arterial disease, CAA-cerebral amyloid angiopathy, NS-not  
367 significant, LMA-leptomeningeal arteriole; <sup>a</sup>N=10 were diagnosed with both AD and VaD; <sup>b</sup>Other Dementia age is significantly less than  
368 CN, MCI, AD or VaD by pairwise analyses; <sup>c</sup>CN significantly higher than AD, VaD or Other Dementia by pairwise analyses; <sup>d</sup>brain total  
369 plaque: senile amyloid plaque density scored according to CERAD templates, with highest possible total being 15<sup>7</sup>; <sup>e</sup>AD is significantly  
370 higher than Other by pairwise analysis; <sup>f</sup>proportion of CAA present in AD significantly higher than Other on Fisher's exact test; <sup>g</sup>density  
371 of amyloidotic blood vessels (scored as 0=none, 1=sparse, 2=moderate, 3=frequent) summed from frontal, temporal, parietal and  
372 occipital regions<sup>7</sup>, <sup>h</sup>Other Dementia diameter is significantly larger than AD or VaD on pairwise analyses; <sup>i</sup>Other Dementia diameter is

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373 significantly larger than CN or AD on pairwise analyses, <sup>i</sup>Other Dementia diameter is significantly larger than CN, AD or VaD on  
374 pairwise analyses; no imputation of missing data was performed.  
375

376 **Figure Legends**

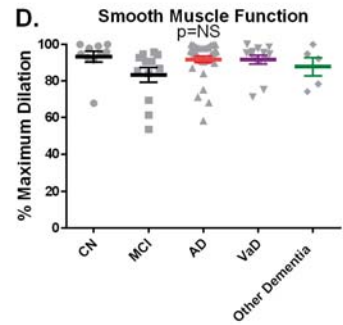
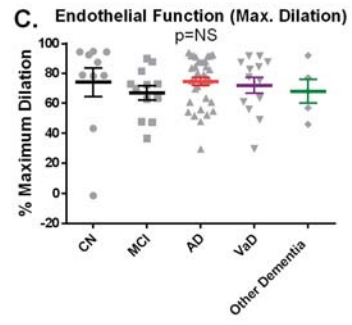
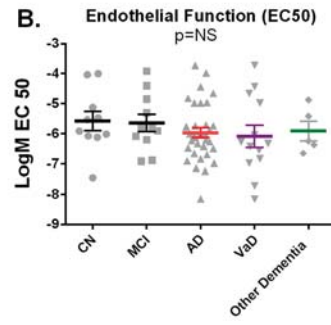
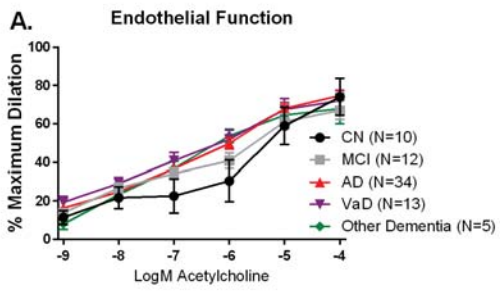
377 **Figure 1. Leptomeningeal Arteriole Vasoreactivity.** There was no difference in baseline  
378 endothelial function (A-C) when comparing subjects with CN, MCI, AD and Other Dementia and  
379 when comparing subjects with CN, MCI, VaD and Other Dementia. Dilator response to a single  
380 dose of papaverine or DETA-NONOate to reflect smooth-muscle dependent function (D) also  
381 showed no significant difference among the groups.

382

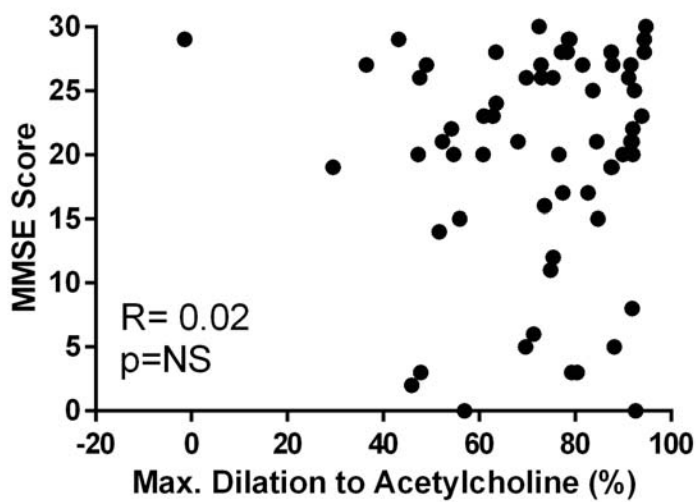
383 **Figure 2. Leptomeningeal Arteriole Function and Cognitive Function.** There was no  
384 correlation between last cognitive function score (MMSE score) and measures of LMA  
385 endothelial (A) or smooth muscle-dependent function (B).

386

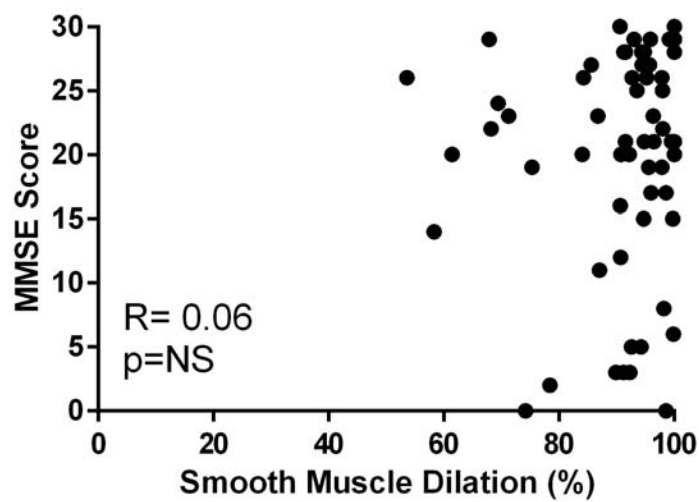
387 **Figure 3. Vasoreactivity Response to Vascular Stressors.** There was significant impairment  
388 in endothelial function when LMAs were exposed to A $\beta$ 42 (2  $\mu$ M), PA (150  $\mu$ M) and medin (5  
389  $\mu$ M) compared to baseline control response (A-C). The impairment in response was not different  
390 between LMAs from CN/MCI versus AD/VaD subjects (D-F). There was impaired smooth  
391 muscle-dependent function in LMAs exposed to A $\beta$ 42, PA and medin (G-I). Post-treatment  
392 smooth muscle-dependent vasoreactivity responses were not different between LMAs from  
393 N/MCI versus AD/VaD subjects.



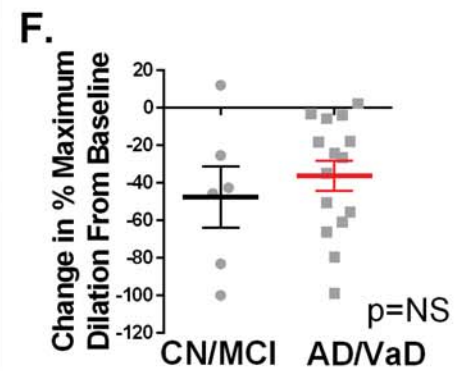
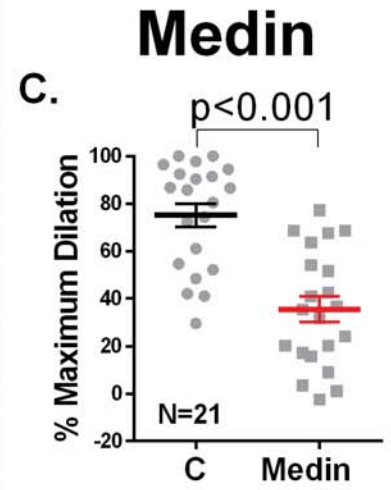
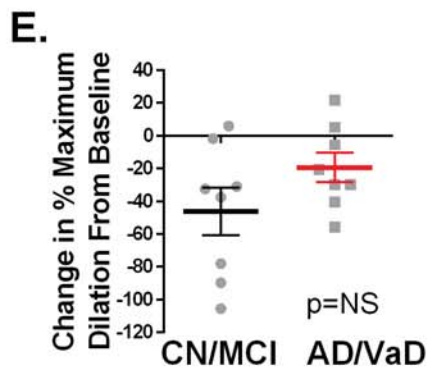
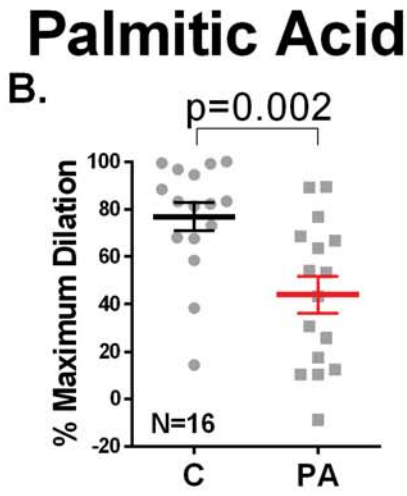
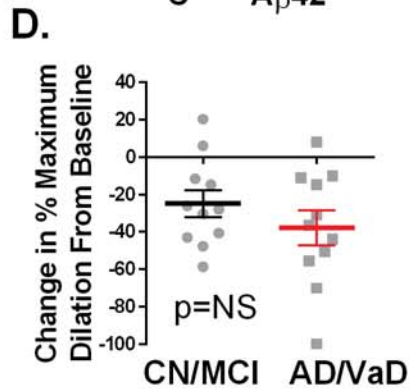
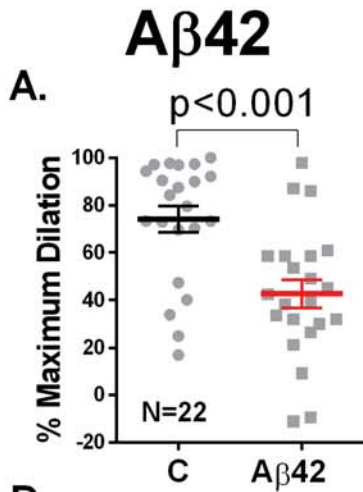
**A. Cognitive Function Score and LMA Endothelial Function**



**B. Cognitive Function Score and LMA Smooth Muscle Function**



# Endothelial Function



# Smooth Muscle Function

