

Perfusion and vascular response as early markers in a bigenic mouse model of Alzheimer's Disease

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Target Audience

This abstract is relevant to researchers interested in the functional link between brain perfusion abnormalities and neurodegeneration, as well as to those interested in the implementation of arterial spin labeling in mice.

Purpose

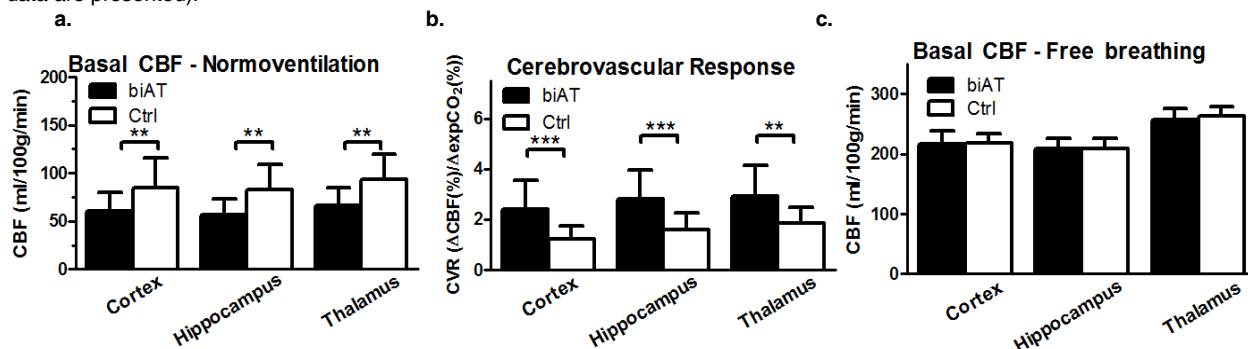
An accumulating body of evidence suggests a causative link between cerebrovascular dysfunction and Alzheimer's disease (AD) (1,2). Over recent years, arterial spin labeling (ASL) MRI has, like other perfusion imaging modalities, demonstrated brain hypoperfusion a long time before the onset of dementia (3,4). Although several transgenic AD models are available, only a limited amount of research has been dedicated to brain perfusion (5,6). Presymptomatic biomarkers are, however, essential not only to confirm that rodent models reflect human disease pathogenesis, but also as non-invasive readouts of AD, useful for the evaluation of novel pharmacological treatments targeting the early stages of the disease. Recent studies suggest that regional cerebrovascular response (CVR) to changes in arterial or expired CO₂ may be a more sensitive biomarker than basal cerebral perfusion (7). In aging rats, the variation in cerebral blood flow (CBF) and blood oxygen level dependent (BOLD) response to changes in pCO₂ allowed the prediction of mild cognitive impairment (8). A limited number of patient studies has also investigated the CVR using BOLD fMRI contrast (7) or transcranial Doppler (9), finding a reduced CVR to CO₂ breathing in AD subjects vs. healthy controls. In addition, abnormal vascular response has been reported in aging mice overexpressing amyloid precursor protein (APP) in a protocol combining mechanical ventilation and acetazolamide injection (10,11). In the current study, we implement ASL to evaluate both basal CBF and CVR, evaluating hypoventilation-induced hypercapnic CBF increases as an early biomarker in a double transgenic AD mouse model that expresses mutant human tau and APP (12,13).

Methods

We analyzed double transgenic APP.V717I \times Tau.P301L mice, denoted as biAT, which develop combined amyloid and tau pathology (12,13). All measurements were performed on two cohorts of young biAT mice (age 3 months; cohort 1, n=10; cohort 2, n=8) and of age-matched FVB wild-type control mice (cohort 1, n=12; cohort 2, n=5) and later on pooled for analysis. Two consecutive scanning sessions were performed within 1 week. During the first session, ASL MRI was performed under free breathing conditions and isoflurane anesthesia (1-1.6%) in pure oxygen. In the second session, the animals were anesthetized using a mixture of 150 mg/kg ketamine (Anesketin, Eurovet, Bladel, NL), 3.8 mg/kg midazolam (Dormicum, Roche, Brussels, BE) and 0.5 mg/kg atropine (Atropine sulfate, Sterop, Brussels, BE) intraperitoneally (i.p.), divided over two to three injections over 20 minutes. Animals were subsequently intubated and mechanically ventilated with pure oxygen as described previously (3), followed by an i.p. injection of 8 mg/kg rocuronium bromide (Esmeron, Organon, Oss, NL) for respiratory muscle paralysis. Expired CO₂ values were continuously monitored (Vaisala Carbocap Carbon dioxide transmitters series, Bonn, GE). The hypoventilation challenge was achieved by reducing the respiratory rate and tidal volume by 25% and 20%, respectively. MR measurements were performed on a 9.4T Biospin small animal MR system (20cm horizontal bore, Bruker Biospin, Ettlingen, GE), using a 7 cm linearly polarized resonator for transmission and an actively-decoupled mouse brain surface coil for receiving (Rapid Biomedical, Rimpf, GE). ASL data were acquired using a FAIR approach (14) and a RARE readout with the following specific parameters: TR 18s, TE 5.2 ms, rare factor 72, FOV 2.5x2.5 cm, matrix 128x128 with partial FT acceleration to 128x72, fourteen inversion times from 300-4000ms, using an inversion hyperbolic secant of 14ms, (Paravision 5.1, Bruker). CBF values were calculated using the T₁ difference method and assuming an arterial T₁ of 2400ms. CVR values were calculated by taking the ratio of the relative increase in cerebral blood flow (CBF) over the relative increase in expired CO₂. Statistical analysis was performed using one-way ANOVA with Bonferroni's correction for multiple testing. Data in Figures show average with standard deviation (SD).

Results

We report significantly reduced CBF values in the hippocampus, cortex and thalamus of young biAT animals compared to age- and sex-matched wild-type mice using the ventilation protocol (fig. 1a, p<0.01 in all regions). This hypoperfusion was not observed in the scan sessions of free breathing animals under isoflurane anesthesia (fig. 1c). Furthermore, we report significantly increased CVR in response to hypoventilation in young biAT transgenic mice (age 3 months) (fig. 1b, p<0.001 in cortex and hippocampus, p<0.01 in thalamus). This was observed in both cohorts (pooled data are presented).



Discussion

An early reduction in basal perfusion corresponds with previous reports in other AD models (5,6) and agrees with what has been reported in patients (1-4). The absence of perfusion differences under the free breathing conditions may be masked by isoflurane's strong vasodilatory effect or by ventilatory compensation mechanisms. We also report a strongly significant increase in CVR in all regions at 3 months of age. This seems to contradict previous reports on decreased CVR in AD to CO₂ or acetazolamide challenges (7), although they differ in the exact type of challenge and were made at later stage of the disease. At the young age studied here, the counterintuitive increased CVR is possibly the result of impaired autoregulatory mechanisms, eliciting a disproportionately strong vasodilatory response to the hypoventilation-induced hypercapnic challenge.

Conclusion

Three month old biAT animals display significantly increased cerebrovascular reactivity to hypoventilation-induced hypercapnia. To our knowledge, CVR has not yet been evaluated in transgenic mouse models for AD at this young age. Although the precise neurophysiological basis of this effect remains to be investigated (7), this observation is made prior to the development of amyloid plaque or tau tangle pathology (13), suggesting the use of CVR as an early, non-invasive imaging biomarker for AD to complement basal perfusion information.

References

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