PART III

DISCUSSION, CURRICULUM VITAE AND PUBLICATIONS
In this thesis we investigated the influence of blood pressure (BP) measures and angiotensin-converting enzyme (ACE) on brain structure (including vascular brain lesions and brain atrophy), and brain function (including self-rated health and cognition).

The major findings of this thesis are: 1) lower BP levels were associated with worse well-being, and more progression of subcortical brain atrophy in patients with arterial disease, whereas a decline in BP over time was associated with less progression of subcortical brain atrophy in patients with higher BP levels at baseline; 2) higher ACE levels were - particularly in the context of a high BP - associated with more vascular events and vascular brain lesions, but also with more CSF amyloid-β (indicating less plaque accumulation), less brain atrophy and less Alzheimer’s disease (AD). As such we found support for both the vascular hypothesis and the amyloid hypothesis of the association between ACE and AD. To appreciate these findings, in this chapter they are placed in a broader context, certain points of discussion are raised and clinical relevance and implications for future research are discussed.

Figure 1 shows a schematic overview of the relations between BP, ACE and Alzheimer’s disease. It does not attempt to be complete, but to explain the main relations described in this thesis.

**Blood pressure**

There is an on-going debate on the relation between BP and cardiovascular morbidity and mortality. Although it has been well established that high BP is a major risk factor for these outcomes,\(^1\) this association may be less straightforward than usually been thought. Increasing evidence shows that a low BP in certain populations may be harmful as well, particularly in older people.\(^2-4\) Yet, the evidence is inconsistent, where both increased and decreased mortality have been associated with higher BP levels.\(^1,5,6\) This could be explained by the large heterogeneity among (older) persons, such as differences in comorbidity and functioning. In some people, the biological age may be higher than their actual age, making them more vulnerable for low BP levels.\(^7,8\) In line with this, in specified patient groups, particularly patients with coronary artery disease or diabetes mellitus, a low BP is associated with higher morbidity and mortality.\(^9-11\) This might indicate that these patient groups are subject to early vascular aging. Their biological age may be higher than their actual age, making them at increased risk for low BP related adverse outcomes.
Figure 1. A detailed overview of the conflicting roles of Angiotensin Converting Enzyme (ACE) in the etiology of Alzheimer’s disease (AD)

Relations observed in this thesis (black) and known from literature (grey)
ACE: angiotensin-converting enzyme; Ach: acetylcholine; AD: Alzheimer’s disease; ANG: angiotensin; AP-A: aminopeptidase A; Aminopeptidase N; AT1R: angiotensin II type 1 receptor; BP: blood pressure; NEP: neprysilin.
Blood pressure and the brain

Similarly, a low BP may be harmful for the brain through vascular and neurodegenerative brain changes, causing cognitive decline and dementia including AD.\textsuperscript{12-14} Up to now, the literature shows that older people, but possibly also other subgroups including patients with coronary artery disease (CAD), may be at increased risk for these adverse brain outcomes when having low BP levels.\textsuperscript{12-14} This thesis contributes to the existing evidence, as we showed in Chapter 2 and 4 that in late-middle aged patients with manifest arterial disease, a low baseline BP was associated with reduced physical and mental functioning, and with increased progression of subcortical brain atrophy.\textsuperscript{15,16} Hence, our data suggest that the effect of BP on the brain in a high-risk population of middle-age is comparable to that of older persons in the community. Otherwise, our population may represent a vulnerable population with higher biological than actual age, in whom high BP levels are better.\textsuperscript{17,18} Several underlying mechanisms could be suggested. Possibly, a low BP (especially low diastolic BP) in this high-risk population is an indicator of arterial stiffness. The carotid arteries in our population with manifest arterial disease were relatively stiff compared to a general population at mid-life,\textsuperscript{19} although not as stiff as those of elderly in the community.\textsuperscript{20} Also, in Chapter 3 we showed a cross-sectional association between arterial stiffness and vascular brain lesions and brain atrophy in this population, although arterial stiffness was not associated with progression of these structural brain measures. Another suggested underlying mechanism is that low BP could be an indicator of decreased cardiac output.\textsuperscript{17} This is in line with our observation that the association between low DBP and progression of subcortical atrophy was particularly present in patients with CAD, who may often have a lower cardiac output. Finally, our population may be at increased risk of cerebral hypoperfusion through microvascular pathology and impaired cerebral autoregulation.\textsuperscript{18} This can make the brain more vulnerable to lower BP levels, whether resulting from antihypertensive treatment, arterial stiffness or reduced cardiac output.\textsuperscript{18} Another finding in Chapter 2 was that declining systolic and diastolic BP levels in our population were associated with less subcortical brain atrophy, particularly in patients with a higher baseline BP.\textsuperscript{16} These results suggest that lowering of BP is beneficial in patients with higher BP levels, but one should be cautious with further BP lowering in patients who already have a low BP.
Blood pressure and the brain: what is the role of ACE?

The role of ACE in relation to AD received increasing attention the last decades. Yet, the two main hypotheses on this association are conflicting. The vascular hypothesis suggests that higher ACE-activity leads to higher BP levels, thereby increasing the risk of vascular brain lesions and AD. Conversely, the amyloid hypothesis suggests that higher ACE activity leads to increased amyloid-β (Aβ) degradation, and hence to less plaque accumulation, decreased Aβ mediated neuronal damage and brain atrophy, and reduced risk of AD. Figure 1 shows the two hypotheses, in more detail than before, including the most relevant newly discovered components of the RAS.

In this thesis, we found support for both hypotheses. In Chapter 5 we observed that in patients with manifest arterial disease, higher serum ACE levels were related to more recurrent ischemic stroke and coronary heart disease, particularly in the presence of hypertension. Also, higher serum ACE levels were associated with more progression of WML volume (Chapter 6). Further, higher CSF and to a lesser extent serum ACE levels were borderline significantly related to severe WMLs and presence of microbleeds, in AD patients with hypertension (Chapter 8). This is in line with the current evidence supporting the vascular hypothesis, which mainly comes from secondary outcome measures of clinical trials showing that ACE-inhibitors reduced the risk of stroke, CSVD, cognitive decline, and dementia.

Interestingly, high BP was not merely an intermediate in these associations, but an effect modifier. We observed that the associations of ACE with vascular outcomes were independent of BP, yet there was an apparent synergistic effect of ACE and hypertension on these outcomes. It is however unclear how ACE independently of BP influences vascular pathology. Possibly the enzymatic actions of ACE, through formation of ANGII, promotes endothelial dysfunction, blood-brain barrier permeability, inflammation and atherosclerosis eventually leading to ischemic heart disease, stroke, WMLs and microbleeds. Since all these conditions are more severe in hypertensive patients, this could explain the modifying effect of hypertension.

We also found evidence for the amyloid hypothesis, as we observed that higher ACE levels, particularly CSF ACE protein levels, were associated with higher CSF Aβ levels, indicating less Aβ accumulation in the brain (Chapter 7). Also, higher serum ACE protein levels were associated with less progression of cortical brain atrophy in patients with manifest arterial disease (Chapter 5), and higher CSF ACE activity levels were associated with less global cortical atrophy in a
memory clinic cohort (Chapter 8). This suggests that ACE indeed may degrade Aβ in living humans, thereby preventing accumulation of Aβ in the brain, brain atrophy and possibly the development or rate of progression of AD. Thereby, this study is in line with the numerous laboratory-based findings (in vitro and in animal models) showing that ACE degrades Aβ.

A remaining question is whether ACE is causally related to brain Aβ pathology or whether the down regulation of ACE is a general phenomenon associated with neurodegeneration. Our data may be consistent with the first possibility since the associations were also present in the control group without cognitive impairment. It is therefore reasonable to assume that the down-regulation of ACE is not pathology-driven, and that lower ACE levels might precede the pathology. This however, remains to be further investigated in a larger, longitudinal study.

Of interest, in this thesis we showed that ACE was associated with vascular and neurodegenerative brain measures in patients with high vascular risk, as well as patients (at risk for) AD. Whereas the evidence on the vascular hypothesis was mainly based on ACE inhibitor (ACEi) use in high vascular risk populations, and the evidence on the amyloid hypothesis (in humans) particularly on ACE genotype in patients with AD, the differences in populations could have partly explained the conflicting results. Yet, results presented in this thesis were comparable in both populations, indicating that the functions of ACE (induced effects) are largely independent of the study populations.

We did not observe an association between serum ACE protein levels and cognitive decline within the SMART-MR study, possibly because of the relatively intact cognition in this population (Chapter 5). However, we found that CSF and serum ACE levels were lower in patients with AD compared to controls in a memory clinic cohort (Chapter 7). In general, previous work showed lower ACE protein levels in AD as well, whereas studies with ACE activity were inconsistent.

In this thesis, we showed that ACE protein levels in CSF and serum correlated strongly, suggesting the synthesis of the enzyme appears to be well correlated in different physiological compartments. Yet, levels of ACE activity did not correlate within serum and CSF and similar apparent disconnected correlations were found between ACE activity in CSF and brain tissue post mortem. This suggests that the eventual resultant catalytic activity of the enzyme could be modified by different ‘environmental’ conditions within these compartments. Therefore, more clarifications of the modifications that may occur are
needed to examine whether ACE protein and activity levels could be potential biomarkers for AD.

The knowledge on the RAS system has increasingly expanded the last years and many new angiotensins and functions have been identified.\textsuperscript{36} ANGII has been shown to not only increase BP, but also increase inflammation, and inhibit acetylcholine release, further increasing the risk of AD (Figure 1).\textsuperscript{21,36} In relation to AD, the most important new angiotensins are probably ANG1-7 and ANGIV. ANG1-7, among others formed as byproduct of ANGII, counterbalances the (negative) effect of ANGII on the vasculature, and therefore its role in the vascular hypothesis. Further, ANGII is converted to ANGIII and ANGIV, of which the latter may act to increase acetylcholine release, thereby leading to cerebroprotective effects, and enhanced memory.\textsuperscript{21} It would be of interest to also examine these angiotensins in relation to vascular brain lesions and brain atrophy, to get a better understanding of all components of the RAS potentially involved in AD development.

**Clinical relevance and future directions**

Since AD is one of the major health problems of the current century, there is an urgent need for disease-modifying treatment. Currently, no therapeutic options are available to target the neurodegenerative component of dementia, but the vascular component might offer opportunities for treatment and prevention strategies.\textsuperscript{37} Clinical trials investigating whether antihypertensive treatment decreased the incidence of AD are, however, inconclusive.\textsuperscript{38-40} This could be explained by selective loss-to-follow-up, the use of different, and mainly secondary endpoints, different antihypertensive drugs, and different study populations at baseline.\textsuperscript{38} In this thesis, we observed that a low baseline BP was associated with more progression of subcortical brain atrophy and worse functioning in patients with arterial disease, and a decline in BP over time was associated with less progression of brain atrophy only in patients with higher BP levels at baseline. This suggests that patients with manifest arterial disease are vulnerable to early vascular aging, and have a higher biological than actual age. It might be interesting to re-analyze observational and intervention studies on BP and adverse brain outcome measures (including AD) on the modifying presence of vulnerability or vascular aging, for example walking speed, self-perceived health, perceived age by the physician, or measures of arterial stiffness.\textsuperscript{7,41-43}
Further, it would be of interest to perform anti-hypertensive treatment trials where vulnerable participants are randomized to achieve more and less stringent BP targets to see if the brain is affected to different degrees when the BP is higher or lower. Yet, in the meanwhile, to avoid unintended harm when pushing the BP down, we should try to identify persons with increased vulnerability and higher biological than actual age, and take that into account when considering (stringent) antihypertensive treatment.

Further, we observed that higher ACE levels were associated with both detrimental and beneficial effects on the brain. Whereas the detrimental effects are particularly due to the ANGII effects via the ANGII type 1 receptor (AT1R), the beneficial effects are due to the ACE enzyme itself. Therefore, we suggest that angiotensin II type 1 receptor blockers (ARBs) instead of ACE-inhibitors (ACEi) might be a better antihypertensive therapy for patients at risk for dementia (e.g. with a positive family history, the APOE-e4 allele or subjective complaints). This is supported by some observational data, showing that ACEi contributed to higher AD risk and mortality than ARBs, and two small-scaled RCTs suggesting that ARBs might be better than ACEi in preserving cerebral hemodynamics and cognitive function. This however warrants further investigation in larger scaled RCTs, therefore, we suggest to compare the use of ARBs or ACEi (particularly those that can cross the blood-brain barrier) in a RCT, with brain MRI, cognitive measures and incidence of AD included as outcome measures. Further, it is currently not known whether ACEi use in humans influences Aβ accumulation. So far only one small-scaled RCT examined the association between ACEi use and CSF Aβ and found no association. We suggest it would be of interest to include CSF AD biomarkers and PIB-PET as outcome measures in the abovementioned RCTs, to examine whether ACEi or ARBs differently influence Aβ accumulation.
References


