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Blood–brain barrier permeable β-blockers linked to lower risk of Alzheimer’s disease in hypertension

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Alzheimer’s disease is a neurodegenerative disorder in which the pathological accumulation of amyloid-β and tau begins years before symptom onset. Emerging evidence suggests that β-blockers (β-adrenergic antagonists) increase brain clearance of these metabolites by enhancing CSF flow. Our objective was to determine whether β-blocker treatments that easily cross the blood–brain barrier reduce the risk of Alzheimer’s disease compared to less permeable β-blockers.

Data from the Danish national registers were used to identify a retrospective cohort of individuals with hypertension, and those treated with β-blockers were included in the analysis. People with indications for β-blocker use other than hypertension (e.g. heart failure) were only retained in a sensitivity analysis. β-blockers were divided into three permeability groups: low, moderate and high. We used multivariable cause-specific Cox regression to model the effect of β-blocker blood–brain barrier permeability on time to dementia outcomes, adjusting for baseline comorbidities, demographics and socioeconomic variables. Death was modelled as a competing risk. The 10-year standardized absolute risk was estimated as the averaged person-specific risks per treatment.

In a cohort of 69081 (median age = 64.4 years, 64.8% female) people treated with β-blockers for hypertension, highly blood–brain barrier-permeable β-blockers were associated with reduced risk of Alzheimer’s disease versus low permeability β-blockers (−0.45%, P < 0.036). This effect was specific to Alzheimer’s diagnoses and did not extend to dementia in general. Propensity score analysis matching high and low blood–brain barrier-permeable patients also detected a decreased Alzheimer’s risk (−0.92%, P < 0.001) in the high permeability group compared to the low, as did a 1-year landmark analysis (−0.57%, P < 0.029) in which events within the first year of follow-up were ignored as likely unrelated to treatment. Our results suggest that amongst people taking β-blockers for hypertension, treatment with highly blood–brain barrier permeable β-blockers reduces the risk of Alzheimer’s disease compared to low permeability drugs. Our findings support the hypothesis that highly permeable β-blockers protect against Alzheimer’s disease by promoting waste brain metabolite clearance.
Introduction

Alzheimer’s disease is a progressive neurodegenerative condition, characterized by an accumulation of amyloid-β and tau proteins that begins decades before symptoms occur. While the precise mechanisms are unclear, amyloid-β oligomers contribute to cell damage, and levels of pathogenic tau are closely linked to symptom severity. Alzheimer’s is the most common form of dementia, with similar prevalence in Denmark as other European countries. It affects more than 46 million people worldwide and is projected to nearly triple in prevalence by 2050.

CSF bulk flow propelled by cardiac-arterial pulsations may remove amyloid-β and tau via efflux along perivenous spaces and cranial nerves, a mechanism which is notably impaired in murine Alzheimer’s models. The importance of this CSF-dependent clearance is supported by human imaging, histopathology and genetic studies.

Norepinephrine binds to α- and β-adrenergic receptors (α and βARs) expressed throughout the CNS. A key neuromodulator of sleep and arousal, norepinephrine also regulates CSF-dependent clearance: reducing norepinephrine signalling improves CNS metabolite clearance, likely by diminishing glial cell volume, increasing interstitial space and lowering resistance to parenchymal flow. β-Blockers (βBs, i.e. β-adrenergic antagonists), typically used to treat cardiovascular conditions like hypertension by inhibiting cardiac βARs, may therefore also promote CSF-dependent clearance if they reach the CNS (Fig. 1).

Blood–brain barrier (BBB) permeability can be determined using a variety of methods, including biodistribution and autoradiography in animals, and invasive sampling, PET imaging or post-mortem investigations in humans. Such studies have shown consistent results that allow grouping of βBs by their ability to cross the BBB (Table 1).

Hypertension remains a main indication for βB use, although they are no longer recommended as one of the starting treatment options. It affects an estimated 1.13 billion people globally, is treated with interchangeable βBs of varying BBB permeability, but similar ability to control blood pressure, and often warrants treatment decades before typical Alzheimer’s onset. These features make hypertension an ideal backdrop for investigating the impact of βB use and BBB permeability on Alzheimer’s risk.

Here we used an epidemiological approach, interrogating data from the Danish national registry to test the hypothesis that treatment with highly BBB-permeable βBs reduces the risk of Alzheimer’s disease compared to βBs with low BBB permeability.

Materials and methods

Registry data

Data were retrieved from the Danish Civil Registration System, the Danish National Patient Registry, National Prescription Registry, the Danish national registers are centrally maintained and contain information on nearly all residents of Denmark. Any prescription drugs dispensed are legally required to be recorded. Hospital reporting to the Danish National Patient Registry is mandatory for public institutions, and psychiatric and outpatient clinic visits have been included since 1995. We had access to data for more than 7 million current and former residents of Denmark.

Study population

We identified people with hypertension based on their use of two different antihypertensive drug classes in two consecutive quarters. Individuals were included at the end date of the second quarter and retained if either of the drug classes was βB. We considered the most common βB drugs available from 1995–2017: metoprolol, atenolol, bisoprolol, propranolol, carvedilol and sotalol (Table 1). We established a relatively homogenous and comparable primary cohort by excluding those with evidence of competing indications for βB treatment. Individuals were followed from inclusion until an event (death or diagnosis of dementia), emigration or the end of the study period on 31 December 2017 (Supplementary Fig. 1). Diagnosis and medication codes are listed in Supplementary Table 1.

Exclusion criteria

People with competing indications for βB treatment (heart failure, arrhythmia or ischaemic heart disease) were excluded, along with those diagnosed with hypertension as a complication of a more serious underlying condition (secondary hypertension) or with concomitant intake of >2 antihypertensive drug classes. Persons treated with βBs for liver disease (almost always using propranolol) were excluded to avoid violating model assumptions.

We retained individuals between 50 and 99 years of age at inclusion, so as to capture the onset of most Alzheimer’s disease cases.

Those with prior diagnoses of Alzheimer’s or other types of dementia were removed from the analysis. Individuals were required to have a minimum of 5 years intact data prior to inclusion. Therefore, the earliest possible inclusion date for our study was 1 January 2000. Persons included before 2000, and those who lived outside Denmark within 5 years prior to inclusion, were removed due to incomplete data.

Keywords: Alzheimer’s disease; antihypertensive drugs; adrenergic antagonists; glymphatic clearance; blood–brain barrier

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Materials and methods

Registry data

Data were retrieved from the Danish Civil Registration System, the Danish National Patient Registry, National Prescription Registry, the Death Register, the National Migration Register and the Family Income Register. Diagnoses since 1994 are recorded using the Danish edition of the ICD-10 (International Classification of Diseases, 10th Revision), and prescriptions are registered by their ATC (Anatomical Therapeutic Chemical) code. Data were anonymized without possibility for identification of individuals. Ethical approval is not required for registry-based studies in Denmark.

The Danish national registers are centrally maintained and contain information on nearly all residents of Denmark. Any prescription drugs dispensed are legally required to be recorded. Hospital reporting to the Danish National Patient Registry is mandatory for public institutions, and psychiatric and outpatient clinic visits have been included since 1995. We had access to data for more than 7 million current and former residents of Denmark.
Outcomes

Our main outcome was diagnosis of Alzheimer’s disease, and for comparison, a diagnosis of any dementia (including Alzheimer’s, unspecified dementia, and other causes). Both registry diagnoses have previously been validated, with positive predictive values of 81.0% and 86.8%, respectively.\textsuperscript{41}

Measures

Individuals with hypertension were divided into groups based on which β-blocker they were taking at baseline. Atenolol,\textsuperscript{23–27} bisoprolol\textsuperscript{28} and sotalol\textsuperscript{28} were classified as low BBB permeability; metoprolol\textsuperscript{23–26} as moderate BBB permeability; and carvedilol\textsuperscript{30,31} and propranolol\textsuperscript{23–26,28,29} as high BBB permeability (Table 1). Our analyses defined exposure by prescriptions at inclusion, regardless of discontinuation. This framework avoided bias due to non-random discontinuation, including probable confounding by unobservable features. Unlike a traditional prospective intention-to-treat framework, however, participants were not randomized to a treatment group.

Covariates

Each individual’s medical history, socioeconomic group and living situation were determined from the years prior to inclusion. Covariates relating to prescriptions pertained to medications received in the 6 months preceding inclusion (Supplementary Fig. 1 and Supplementary Table 1).

Covariates were based on established risk factors for Alzheimer’s disease.\textsuperscript{42,43} We adjusted for the following: age; sex;
acting agents, potassium-sparing diuretics, loop diuretics, and thiazide and other diuretics. This allowed for appropriate adjustment of the BBB permeability of the β2-adrenergic receptor at higher doses. Carvedilol, a third generation β-blocker, is additionally active at the α1-adrenergic receptor. The number of people treated is reported before and after applying exclusion criteria. Classification of β-blockers as low, moderate or high BBB permeability was based on available literature reporting distribution in human or animal tissue. Sotalol is used primarily as an anti-arrhythmic; persons with this prescription were excluded from the primary cohort since arrhythmia is a competing indication for β-blocker use.

whether the subject was living alone or with others; socioeconomic quartile based on equivalized household income; area of residence as a proxy for provider density, environmental exposures and regional affluence; diabetes; stroke; head trauma; hyperlipidaemia; atherosclerosis; chronic obstructive pulmonary disease; and depression and anxiety.

Diagnostic criteria and treatment of both hypertension and dementia, as well as β-blocker usage in general, changed over the timeframe of our study. In particular, prescriptions declined after hypertension treatment guidelines were revised in 2009. We corrected for the relative time of inclusion to accommodate this potential confounder.

Lastly, we corrected for the second antihypertensive used alongside β-blocker at inclusion, grouped as: calcium channel blockers, angiotensin-II receptor blockers, other renin-angiotensin system-acting agents, potassium-sparing diuretics, loop diuretics, and thiazides and other diuretics. This allowed for appropriate adjustment given possible unrelated protective effects of calcium channel blockers, angiotensin-II receptor blockers and potassium-sparing diuretics that have been described previously.

**Statistical analysis**

Baseline characteristics for each group were compared using the Chi-squared or Kruskal–Wallis H-tests as appropriate. Unless otherwise specified, numbers are presented as the median and interquartile range (IQR).

Non-standardized absolute risk was calculated using the Aalen Johansen estimator. When adjusting for covariates, cause-specific Cox regression was used to model the treatment effect and covariates on time to outcome. Death was considered as a competing risk, such that in the case of two events (e.g. dementia, then death) the first to occur was considered the outcome for that individual, and death without a dementia diagnosis was not equivalent to survival without dementia. The standardized absolute risk of an outcome was estimated as the averaged person-specific absolute risk over each cohort under a given treatment modality. Risks were calculated at 10 years from the date of inclusion and are shown as a percentage alongside 95% confidence intervals (CIs).

Because age and relative time of inclusion did not meet the linearity assumption of Cox models, their effects were modelled using B-splines (basis regression splines).

Standardized absolute risk was calculated every 6 months up to 10 years in each group. The differences in outcome risk were then computed between low and high BBB permeability using the average treatment effect. Adjustment for multiple comparisons over time was performed using the quantile of the confidence bands.

Data were processed using SAS (version 9.4, SAS Institute, Cary, NC, USA) and R statistical software (version 4.0.3, R development core team), including the R package riskRegression (version 2020.12.08) to compute the standardized absolute risks and confidence bands.

**Sensitivity analyses**

In order to investigate our hypothesis in a less homogenous but more representative sample, we repeated the analysis in a secondary cohort where competing diagnoses for β-blocker use (arrhythmia, ischaemic heart disease and heart failure) were corrected for instead of excluded.

To compare our populations with previous investigations, we evaluated the use of any β-blocker together with a second antihypertensive drug class versus treatment with two non-β-blocker antihypertensive drug classes. Exclusion criteria were otherwise the same as in the main analysis.

Defining exposure by baseline prescriptions does not factor in possible discontinuation of the drug assigned at the start of the study or switching between groups. We estimated the effect of the intention to treat rather than the treatment effect; this definition avoided bias due to non-random discontinuation likely related to features unavailable in the registers. Therefore, to check how well the designated group represented actual treatment, we calculated coverage by the BBB permeability group assigned at inclusion (on-target) and by non-assigned groups (off-target) using dispensation records during the follow-up period. We also performed a sensitivity analysis in which subjects were censored upon switching groups or discontinuing β-blocker treatment, forcing perfect compliance.

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**Table 1 β-blocker classification**

<table>
<thead>
<tr>
<th>Drug name</th>
<th>Codes</th>
<th>Number of people</th>
<th>BBB permeability</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before exclusion criteria</td>
<td>After exclusion criteria</td>
<td></td>
</tr>
<tr>
<td><strong>β1 selective antagonists</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metoprolol</td>
<td>C07AB02</td>
<td>298'032</td>
<td>43'055</td>
</tr>
<tr>
<td></td>
<td>C07BB02</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>C07FB02</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Atenolol</td>
<td>C07AB03</td>
<td>57'483</td>
<td>12'883</td>
</tr>
<tr>
<td></td>
<td>C07BC03</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bisoprolol</td>
<td>C07AB07</td>
<td>26'516</td>
<td>36'47</td>
</tr>
<tr>
<td><strong>Non-selective β antagonist</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Propranolol</td>
<td>C07AA05</td>
<td>31'867</td>
<td>6'974</td>
</tr>
<tr>
<td></td>
<td>C07AA07</td>
<td>40'66</td>
<td></td>
</tr>
<tr>
<td>Sotalol</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carvedilol</td>
<td>C07AG02</td>
<td>36'548</td>
<td>2'520</td>
</tr>
</tbody>
</table>

Name, activity, anatomical therapeutic chemical system (ATC) code, and number of people treated for each β-blocker considered in the study. First generation β-blockers (propranolol, sotalol) target both the β1-adrenergic receptor and the β2-adrenergic receptor, whereas second generation β-blockers (metoprolol, atenolol, bisoprolol) only bind the β2-adrenergic receptor at higher doses. Carvedilol, a third generation β-blocker, is additionally active at the α1-adrenergic receptor. The number of people treated is reported before and after applying exclusion criteria. Classification of β-blockers as low, moderate or high BBB permeability was based on available literature reporting distribution in human or animal tissue. Sotalol is used primarily as an anti-arrhythmic; persons with this prescription were excluded from the primary cohort since arrhythmia is a competing indication for β-blocker use.
To reduce any overt selection bias, we compared a subset of highly BBB-permeable β-blockers and low BBB permeability users with similar likelihoods of taking highly BBB-permeable β-blockers. This was achieved by using logistic regression models based on the same covariates described below to calculate each subject’s propensity for taking highly BBB-permeable β-blockers. For each high BBB permeability individual, we then selected (without replacement) the closest propensity low BBB permeability individual.

Finally, to reduce the impact of latency between dementia onset and diagnosis, we ran landmark analyses in which follow-up began at 1, 3 or 5 years after inclusion and any person who had started β-blockers, stopped β-blockers, had an event (death or dementia) or been censored (e.g. emigrated) prior to that point was excluded.

Data availability
Access to anonymized Danish National Registry data for research purposes is controlled by Statistics Denmark.

Results
We labelled 1275458 individuals taking one or more antihypertensive drug from two different classes as having hypertension, 454508 (35.6%) of whom were prescribed β-blockers. After exclusion criteria (Fig. 2), we retained a primary cohort of 69081 people with a total follow-up of more than 522065 person-years, a median follow-up time of 9.8 (IQR: 5.3 to 10.0) years and a median age of 64.4 years (IQR: 57.7 to 72.3 years; 64.8% female). The cohort was divided into low, moderate and high BBB permeability β-blocker groups (see the ‘Materials and methods’ section) as shown in Fig. 2. Baseline demographics are summarized in Supplementary Table 2.

Highly BBB-permeable β-blockers are associated with reduced risk of Alzheimer’s disease

At the end of follow-up, there were 837 Alzheimer’s events, 13505 deaths and 2075 cases of any type of dementia (including Alzheimer’s, unspecified and other dementia). Our conclusions were supported by a landmark analysis, where any event (death or dementia) was included as an event. As with the primary cohort, Alzheimer’s risk decreased numerically along the gradient from low to moderate to high BBB permeability, without reaching statistical significance. Absolute risk of death was slightly higher in both moderate [+0.71% (CI: 0.21% to 1.21%), P < 0.012] and high [+1.86% (CI: 1.18% to 2.53%), P < 0.001] BBB permeability groups compared to the low BBB permeability group.

Comparing β-blocker use as a group against alternative hypertensive drug classes in the wider hypertension population, we did not find any significant difference in terms of either Alzheimer’s risk or risk of any dementia (Supplementary Table 4). Risk of death was increased by 21.1% for non-β-blockers to 22.4% in β-blocker users [+1.36% (CI: 1.02% to 1.70%), P < 0.001, n = 368892]. When using a propensity score approach, Alzheimer’s risk was decreased by 0.92% (CI: −1.42% to −0.42%; P < 0.011; n = 16166) for highly BBB-permeable β-blockers as compared to propensity-matched low BBB permeability users, and risk of death was increased by 1.82% (CI: 0.69% to 2.94%; P < 0.002; n = 16166) (Table 3).

Standardized absolute risk of Alzheimer’s disease was also reduced in the moderate and high compared to low BBB permeability group in a 1-year landmark analysis (moderate: −0.46% (CI: −0.81% to −0.12%), P < 0.020; high: −0.57% (CI: −1.02% to −0.12%), P < 0.029, n = 54769). Here, any events (i.e. death or dementia) within the first year were excluded to offset the effect of lag between dementia onset and diagnosis (Table 4). Absolute risk was not significantly different between high and low groups for death [+1.07% (CI: 0.02% to 2.12%), P = 0.108] or any dementia [−0.27% (CI: −0.85% to 0.31%), P = 0.623]. The direction of these results was similar in 3- and 5-year landmark analyses, but differences were not significant (data not shown).

Discussion
In this nation-wide retrospective cohort study of Danish residents with hypertension, we demonstrated that treatment with β-blockers that readily cross the BBB is associated with a reduced Alzheimer’s risk (−0.47%; risk ratio: 75.9%) compared to treatment with less BBB-permeable β-blockers. The reduction in Alzheimer’s risk for the highly BBB-permeable β-blockers was significant after 1.5 years (Fig. 3A), a delay which is consistent with the gradual progression of Alzheimer’s disease. Our conclusions were supported by a landmark analysis,
Figure 2. Flow chart describing cohort selection. A total of 1,275,458 individuals took at least two different antihypertensive drug classes between 1995 and 2017. For 454,508 (35.6%) of them, βBs were one of the drug classes. Following exclusion criteria, we included 69,081 people in the primary cohort, from which people with indications for βBs other than hypertension were excluded. Individuals were further divided into groups based on the BBB permeability of the βB prescribed at baseline, with atenolol and bisoprolol considered low; metoprolol moderate; and carvedilol and propranolol high BBB permeability. Altogether, 23.9% of people were grouped as taking low BBB permeability βBs, 62.3% as moderate, and 13.7% as high.

Table 2 Ten-year outcomes

<table>
<thead>
<tr>
<th></th>
<th>Events, n (% of population)</th>
<th>Non-standardized absolute risk, % (95% CI)</th>
<th>Standardized absolute risk, % (95% CI)</th>
<th>Standardized absolute risk difference, % (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alzheimer’s disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>231 (1.4%)</td>
<td>1.54 (1.34 to 1.74)</td>
<td>1.96 (1.65 to 2.30)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>494 (1.1%)</td>
<td>1.44 (1.32 to 1.57)</td>
<td>1.63 (1.43 to 1.85)</td>
<td>−0.33 (−0.63 to −0.03)</td>
<td>0.074</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>112 (1.2%)</td>
<td>1.54 (1.25 to 1.83)</td>
<td>1.49 (1.20 to 1.82)</td>
<td>−0.47 (−0.85 to −0.10)</td>
<td>&lt;0.036</td>
</tr>
<tr>
<td>Death (as competing risk with Alzheimer’s disease)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>3304 (20.0%)</td>
<td>21.9 (21.2 to 22.5)</td>
<td>22.9 (22.2 to 23.5)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>8123 (18.9%)</td>
<td>23.1 (22.6 to 23.5)</td>
<td>23.8 (23.3 to 24.3)</td>
<td>0.91 (0.24 to 1.58)</td>
<td>&lt;0.022</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>2078 (21.9%)</td>
<td>27.2 (26.2 to 28.3)</td>
<td>24.4 (23.6 to 25.2)</td>
<td>1.55 (0.63 to 2.47)</td>
<td>&lt;0.003</td>
</tr>
<tr>
<td>Any dementia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>530 (3.2%)</td>
<td>3.52 (3.22 to 3.81)</td>
<td>3.71 (3.38 to 4.06)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>1226 (2.8%)</td>
<td>3.53 (3.34 to 3.73)</td>
<td>3.54 (3.31 to 3.78)</td>
<td>−0.17 (−0.54 to 0.20)</td>
<td>0.641</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>319 (3.4%)</td>
<td>4.28 (3.81 to 4.74)</td>
<td>3.66 (3.27 to 4.08)</td>
<td>−0.05 (−0.55 to 0.44)</td>
<td>0.973</td>
</tr>
</tbody>
</table>

Number of Alzheimer’s disease, death and any dementia (including Alzheimer’s, unspecified dementia, and other causes) outcomes in the primary cohort, and risk at 10 years. Death was treated as a competing risk (see the ‘Materials and methods’ section). Non-standardized risk was uncorrected, while standardized absolute risk was modelled using the covariates listed below. P-values refer to standardized risk differences. There were 16,530 people taking low, 43,056 taking moderate and 9,495 taking high BBB-permeable βBs. People taking βBs with high BBB permeability had decreased risk of Alzheimer’s disease compared to low permeability βB users.

*Models were corrected for sex, age (B-splines), relative time of inclusion (B-splines), socioeconomic group, municipality, living alone, loop diuretic use, diabetes mellitus, hyperlipidaemia, depression, stroke, head trauma, atherosclerosis, chronic obstructive pulmonary disease and second antihypertensive drug class.
BBB-permeable βBs and Alzheimer’s risk

Figure 3 Standardized absolute risk over time. Standardized absolute risk over 10 years for the primary cohort, where subjects with indications other than hypertension for treatment with β-adrenergic antagonists were excluded. 95% CIs are shaded. Models were corrected and P-values adjusted for multiple comparisons as described in the ‘Materials and methods’ section. (A) Absolute risk of Alzheimer’s disease with death as a competing risk, showing significantly reduced risk for those taking high BBB permeability βBs (pink) compared to low BBB permeability βBs (blue) from 1.5 years onward. (B) Absolute risk of death with Alzheimer’s disease as a competing risk. (C) Absolute risk of any dementia (including Alzheimer’s, unspecified dementia and other causes) with death as a competing risk. n.s. = not significant.

where events within the first year were ignored as likely unrelated to β treatment (Table 4). Our cohort was carefully curated for increased rigor at the expense of generalizability; however, we had almost no selection or attrition bias thanks to the integrity of the registry data. Alzheimer’s risk was also numerically lower for the high BBB-permeability β group versus the low BBB-permeability β group in a larger cohort with fewer exclusions (Supplementary Table 3), suggesting a degree of generalizability.

We found no significant change in risk for any dementia (including Alzheimer’s disease, unspecified dementia and other causes) between BBB permeability groups, either in the main analysis (Fig. 3C) or any of the sensitivity measures. This alluded to an Alzheimer’s-specific process, tallying with the hypothesis of enhanced glymphatic clearance of amyloid-β and tau. This supported the comparability between groups, as this was an outcome we did not expect to be affected by β treatment.

βBs treat cardiovascular disease via inhibition of cardiac and renal adrenergic receptors\(^1\) and are about as effective as alternatives in treating hypertension.\(^2\) Relative to other βBs, Cochrane reports show little difference between the drugs of interest in terms of diastolic or systolic blood pressure control.\(^3,4\) However, those βBs that cross the BBB have an additional impact on central ARs. Within the CNS, norepinephrine and βARs are involved in a wide range of functions central to Alzheimer’s disease pathology, including memory consolidation, synaptic plasticity, glial function and sleep.\(^5\) Non-REM sleep is believed to be the main regulator of glymphatic clearance, where central adrenergic signalling and locus coeruleus activity play an important inhibitory role.\(^6,7\) Specifically, \(\alpha_1\)-antagonists and βBs inhibit norepinephrine release in a similar yet less potent manner than \(\alpha_2\)AR agonists\(^8\) (e.g. dexmedetomidine) to ultimately reduce arousal, promote sleep and improve glymphatic function.\(^\)\(^,7\) While BBB-permeable βBs in particular are associated with fatigue\(^9\) and subjective changes in sleep,\(^10\) these side effects cannot easily be separated from the impact of βBs on peripheral blood pressure,\(^11\) and objective effects of non-REM sleep in a hypertensive population have not yet been investigated. Therefore, further studies are needed to clarify the impact of βBB permeability on blood pressure control, sleep quality and Alzheimer’s risk.

The role of βARs in Alzheimer’s pathogenesis remains controversial.\(^12\) Several preclinical studies suggest that βBs can increase Alzheimer’s risk\(^13\) and that βAR activation can be protective against Alzheimer’s disease,\(^14,15\) effects that seem contradictory to our findings. However, other data show that βBs can decrease Aβ accumulation and reduce cognitive deficits in mice models\(^16\) and slow cognitive decline in people with Alzheimer’s disease.\(^17\) Genetic studies have found that βAR gene variants associated with decreased Alzheimer’s risk also make the βAR receptors less responsive to norepinephrine.\(^18\) Finally, a case control study of recently diagnosed Alzheimer’s patients showed that cases were significantly less likely to have been using βBs across a 3-year period.\(^19\) Taken together with our findings, these results suggest that central βARs may modulate Alzheimer’s disease risk and development.

Highly BBB-permeable βBs are less selective for βARs (Table 1), which raises the possibility that the observed Alzheimer’s risk reduction is driven by increased βAR inhibition rather than higher BBB permeability. While both types of drug are comparably effective at controlling blood pressure,\(^20,21\) β-selective βBs mainly act by reducing heart rate and contractility to lower cardiac output, whereas non-selective βBs additionally induce slight peripheral
To account for differences between drugs, such as the sensitivity analyses with each drug removed and found similar vasocontraction. However, the decreasing trend in risk from low to moderate to high BBB permeability argues against β selectivity as the defining feature. Furthermore, β₁-selective βBs inhibit both β₁ and β₂ ARs at higher doses. To account for differences between drugs, such as the α₁AR antagonist activity of carvedilol, we ran sensitivity analyses with each drug removed and found similar results (data not shown), suggesting that our results cannot be attributed to any single compound.

We considered two statistical approaches to estimate risk; the cause-specific Cox model, which relies upon correct specification of the model for risk of Alzheimer’s disease and death, and propensity score, which relies upon correct specification of the model for treatment allocation. The propensity score analysis mirrored the main results, showing a 0.92% decrease in Alzheimer’s disease risk for the high BBB-permeability group compared to the low (Table 3). However, we also saw an elevated risk of death in the high (1.82%) and moderate (1.55%) BBB-permeability groups compared to the low (Table 2). The high permeability group risk increased was observed throughout the follow-up period, including at the earliest timepoints (Fig. 3B), and was not related to dementia (as follow-up ends at a dementia diagnosis). The effect is likely explained by an increased unmeasured comorbidity burden in the high BBB-permeability group, which should in principle also raise the Alzheimer’s risk, thereby reducing our effect size. However, when we removed events within the first year after inclusion as unrelated to recently initiated βB treatment, Alzheimer’s risk was still reduced for the high BBB permeability group compared to the low, but risk of death was no longer significantly increased (Table 4). A contributing factor could be the poor tolerability of BBB-permeable compounds relative to newer non-permeable options, and their resulting low popularity. Although those taking more than two antihypertensives at a time were excluded, individuals receiving high BBB-permeability βBs may still have been on their third or fourth treatment option for more severe or difficult to control hypertension, suggesting they were more ill and at risk for both death and Alzheimer’s disease. Moreover, the well-known CNS side effects of the high BBB-permeability drugs could have led prescribers to avoid these options for people with pre-existing cognitive problems (i.e. prodromal dementia), introducing a possible selection bias. Randomized prospective clinical trials are needed to establish a causal link between βB BBB permeability and Alzheimer’s disease risk.

Previous epidemiological investigations of the impact of βB treatment on dementia outcomes, which typically compared βBs against other antihypertensives or no treatment, have failed to show a consistent protective effect. However, such reports have generally considered βBs as a single group and found modest or insignificant effects on Alzheimer’s risk. When we combined βBs into a single group and compared it to alternative hypertension treatments without accounting for BBB permeability, we likewise did not detect any significant difference for risk of Alzheimer’s disease or any type of dementia (Supplementary Table 4). Our results thereby highlighted that the ability of βBs to access the CNS is relevant to Alzheimer’s risk. It is unfortunate that prior investigations have failed to account for this feature. Any central βAR-mediated impact on Alzheimer’s risk and pathogenesis requires that βBs pass through the BBB. Our study was specifically designed to address this important gap in the literature and investigate the role of BBB permeability on βB-mediated modulation of Alzheimer’s risk using real-world clinical data. The finding that higher BBB permeability is associated with reduced Alzheimer’s risk for those taking βBs favours the hypothesis that these medications confer a protective effect that may be independent of their capacity as antihypertensives, if they are able to cross the BBB and reach the site of Alzheimer’s disease pathogenesis. Further investigations are warranted to determine the underlying mechanism behind our findings.

### Strengths and limitations
This study benefitted from negligible loss to follow-up, near complete population coverage and comprehensive prescription drug information, as is characteristic of the Danish national registry system. The relative racial homogeneity of the Danish population limited interpretation for non-Scandinavian individuals but improved comparability between groups. However, relying on registry data meant that we could not take additional variables that may have influenced the outcomes of interest into account, e.g. body mass index, APOE genotype and tobacco use.

We corrected for the baseline demographics and diagnoses known to influence Alzheimer’s risk to accommodate for variability between groups. People taking highly BBB-permeable drugs have notably higher levels of diabetes mellitus, depression, anxiety, stroke and...
Table 4 Ten-year outcomes for 1-year landmark analysis

<table>
<thead>
<tr>
<th></th>
<th>Events, n (% of population)</th>
<th>Non-standardized absolute risk, % (95% CI)</th>
<th>Standardized absolute risk, % (95% CI)</th>
<th>Standardized absolute risk difference, % (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Alzheimer’s disease</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>211 (1.6%)</td>
<td>1.75 (1.51 to 1.98)</td>
<td>2.16 (1.81 to 2.56)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>412 (1.2%)</td>
<td>1.54 (1.39 to 1.69)</td>
<td>1.69 (1.47 to 1.95)</td>
<td>−0.46 (−0.81 to −0.12)</td>
<td>&lt;0.020</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>86 (1.3%)</td>
<td>1.68 (1.32 to 2.04)</td>
<td>1.59 (1.25 to 1.99)</td>
<td>−0.57 (−1.02 to −0.12)</td>
<td>&lt;0.029</td>
</tr>
<tr>
<td><strong>Death (as competing risk with Alzheimer’s disease)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>2782 (20.6%)</td>
<td>22.8 (22.1 to 23.6)</td>
<td>23.8 (23.1 to 24.6)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>6601 (19.1%)</td>
<td>23.8 (23.3 to 24.3)</td>
<td>24.3 (23.7 to 24.8)</td>
<td>0.45 (−0.29 to 1.20)</td>
<td>0.449</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>1549 (22.9%)</td>
<td>28.6 (27.4 to 29.9)</td>
<td>24.9 (24.0 to 25.8)</td>
<td>1.07 (0.02 to 2.12)</td>
<td>0.108</td>
</tr>
<tr>
<td><strong>Any dementia</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low BBB permeability</td>
<td>461 (3.4%)</td>
<td>3.79 (3.45 to 4.13)</td>
<td>3.99 (3.60 to 4.40)</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Moderate BBB permeability</td>
<td>977 (2.8%)</td>
<td>3.58 (3.35 to 3.80)</td>
<td>3.56 (3.31 to 3.82)</td>
<td>−0.43 (−0.85 to −0.01)</td>
<td>0.102</td>
</tr>
<tr>
<td>High BBB permeability</td>
<td>233 (3.4%)</td>
<td>4.40 (3.84 to 4.96)</td>
<td>3.72 (3.26 to 4.23)</td>
<td>−0.27 (−0.85 to 0.31)</td>
<td>0.623</td>
</tr>
</tbody>
</table>

Number of Alzheimer’s disease, death and any dementia (including Alzheimer’s, unspecified dementia and other causes) outcomes in the primary 1-year landmark cohort, and risk at 10 years. For the landmark analyses, any person with an event in the first year was excluded, and inclusion +1 year was used as the start point. Death was treated as a competing risk (see the ‘Materials and methods’ section). Non-standardized risk was uncorrected, while standardized absolute risk was modelled using the covariates listed below. P-values refer to standardized risk differences. There were 13500 people taking low, 34495 taking moderate and 6774 taking high BBB-permeable βs. People taking βs with high BBB permeability had reduced risk of Alzheimer’s disease. Risk of death was increased for both moderate and high BBB permeability βs users.

Models were corrected for the baseline covariates sex, age (B-splines), relative time of inclusion (B-splines), and second antihypertensive drug class, as well as 1-year values for competing risk (see the ‘Materials and methods’ section). Non-standardized risk was uncorrected, while standardized absolute risk was modelled using the covariates listed below. P-values refer to standardized risk differences. There were 13500 people taking low, 34495 taking moderate and 6774 taking high BBB-permeable βs. People taking βs with high BBB permeability had reduced risk of Alzheimer’s disease. Risk of death was increased for both moderate and high BBB permeability βs users.

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# Competing interests
S.C.H. is now a full-time employee of Roche Pharmaceuticals. All other authors report no disclosures.

# Supplementary material
Supplementary material is available at Brain online.

# References


