DUNEDIN STUDY CONCEPT PAPER FORM

Provisional Paper Title: Associations between retinal neuronal and microvascular parameters and brain structural integrity: Is the retina a biomarker of brain health?

Proposing Author: Ashleigh Barrett-Young

Author's Email: Ashleigh.barrett-young@otago.ac.nz

P.I. Sponsor: Richie Poulton & Graham Wilson
(if the proposing author is a student or colleague of an original PI)

Today's Date: 6 December 2021

Please describe your proposal in 2-3 pages with sufficient detail for helpful review.

Objective of the study:

The retina has potential as a biomarker of brain health, including Alzheimer's disease (AD), because it is the only part of the central nervous system which can be easily imaged, and has advantages over brain imaging technologies of being non-invasive, cheap, and widely available (London et al., 2013). Existing treatments for AD may be most effective in the earliest stages of the disease (Musiek & Morris, 2021). Thus, screening tools to identify people at high risk of developing AD or with early AD are imperative to initiating treatments at the optimal stage, hopefully improving patients' quality of life.

There is growing evidence that retinal thinning is evident in preclinical AD. Retinal thinning has been associated with indicators of preclinical Alzheimer's, including amyloid burden (Ko et al., 2018; Santos et al., 2018), family history and genetic risk (Santos et al., 2018), and cognitive performance (Barrett-Young et al., in submission; Ko et al., 2018). Few have directly compared structural measurements of the brain with retinal measurements, and these pilot studies have involved elderly participants or very small sample sizes (Donix et al., 2021; Mejia-Vergara et al., 2021; Méndez-Gómez et al., 2018; Ong et al., 2015; Uchida et al., 2020). A recent study from the UK Biobank suggests that retinal measures are associated with smaller cortical and hippocampal volume in a middle-aged sample (Chua et al., 2020). However, the question of

whether retinal thinning is associated with brain structure in the decades before an Alzheimer's diagnosis remains.
Thus, the objective of this study is to investigate whether the retinal neuronal and microvascular structures are associated with structural brain integrity, determined using neurostructural measurements acquired from MRI.
We hypothesise that thinner retinal nerve fibre layer (RNFL), ganglion cell layer (GCL), and lower vessel density (VD) will be associated with higher brainAGE, indicating more advanced brain ageing. In addition, we hypothesise that thinner RNFL, and GCL, and lower VD will be associated with smaller cortical surface area, lower cortical thickness, and lower grey matter volumes, particularly in areas known to be affected in AD (i.e. the hippocampus).
Note that RNFL and GCL are measures of neuronal thickness, and VD is a measure of the amount of microvasculature in the retina.

Data analysis methods:

As OCT and MRI data were collected at age 45 only, this will be a primarily cross-sectional study. The first stage of analysis will involve using regression to test for associations between OCT variables and brainAGE, a composite measure of structural brain integrity. Posthoc analyses based on these initial findings will be conducted to investigate any associations between cortical thickness, surface area, grey matter volume, and white matter hyperintensities with OCT variables, to determine if specific structural features may have a larger contribution than others towards any overall association.
All models will be adjusted for sex and other physiological covariates, e.g. ocular (intraocular pressure, axial length, optic disc size, signal strength), cardiovascular (BMI, mean arterial blood pressure, smoking), and potential MRI confounders (total brain volume, intracranial volume, and/or cerebrospinal fluid volume). A false discovery rate procedure will be used to correct for multiple comparisons.

Variables needed at which ages:

- Retinal variables (obtained via optical coherence tomography [OCT] and optical coherence tomography angiography [OCTA] at phase 45):
    - Retinal nerve fibre layer (RNFL)—average, all quadrants.
    - Ganglion cell layer (GCL)—average, all segments.
    - Vessel density (VD)—full mean, all segments.
    - Ocular covariates:
        - Intraocular pressure
        - Axial length
        - Optic disc size
        - OCT/OCTA scan signal strength
- Structural brain variables (obtained via magnetic resonance imaging [MRI] at phase 45):
    - Cortical thickness—whole brain and regional

    - Surface area—whole brain and regional
    - Subcortical volume—all areas, incl. hippocampus
    - White matter hyperintensity volume
    - brainAge
    - MRI covariates:
        - Total brain volume
        - Intracranial volume
- Potential covariates:
    - Sex
    - Cognitive decline (ages 7, 9, 11 to age 45; residualised IQ change)
    - IQ variables from ages 7, 9, 11, and 45—FSIQ and indices.
    - BMI (age 45)
    - Mean arterial blood pressure (age 45)

Significance of the Study (for theory, research methods or clinical practice):

Retinal imaging technologies have a huge potential as a biomarker of preclinical AD, and as a potential predictive tool for conversion from mild cognitive impairment to AD (Ng et al., 2021). However, while artificial intelligence and machine learning research using retinal imaging is progressing at a rapid pace, there remain several questions about the relationship between the retina and the AD-affected brain.

The majority of research in this area has investigated people with established AD, largely in cross-sectional case-control studies. The clinical utility of retinal imaging, however, is in the preclinical stages, before symptoms progress to the level where daily living is affected and irreversible neurological damage has occurred. Comparisons between OCT/A and MRI measures have been lacking due to the practicalities and costs of these technologies, so here we have a unique opportunity to investigate the associations between the retina and structural brain data in a well-characterised cohort, where we can also control for various other risk factors if required.

The overarching goal of this project is to provide evidence for the potential of OCT/A as a clinically-useful screening tool for identifying those at risk of developing AD and for monitoring disease progression. These findings will hopefully inform applications to improve screening of AD risk at an early stage of the disease and to ensure equitable access to such screening through the use of existing retinal imaging technology. OCT/A technology is already widely available in regional clinics, as well as urban hospital settings, and increasingly available in retail optometrists. Work is progressing on using OCT/A images in artificial intelligence/machine learning applications for the diagnosis of AD, and manufacturers of commercial ophthalmology imaging tools are likely to implement models into their devices when evidence on a model's efficacy is clear. Potential incorporation of machine learning into OCT/A technology would widen the availability of retinal AD screening to regional populations, as well as other marginalised groups in Aotearoa, such as Māori and Pasifika.

References:

Barrett-Young, A., Ambler, A., Cheyne, K., Guiney, H., Kokaua, J., Steptoe, B., Tham, Y. C., Wilson, G. A., Wong, T. Y., & Poulton, R. (n.d.). Associations between retinal nerve fibre layer and ganglion cell layer and cognition from childhood to middle age. *Manuscript Submitted for Publication*.

Chua, J., Hu, Q., Ke, M., Tan, B., Hong, J., Yao, X., Hilal, S., Venketasubramanian, N., Garhöfer, G., Cheung, C. Y., Wong, T. Y., Chen, C. L.-H., & Schmetterer, L. (2020). Retinal microvasculature dysfunction is associated with Alzheimer's disease and mild cognitive impairment. *Alzheimer's Research & Therapy*, *12*(1), 161. https://doi.org/10.1186/s13195-020-00724-0

Donix, M., Wittig, D., Hermann, W., Haussmann, R., Dittmer, M., Bienert, F., Buthut, M., Jacobi, L., Werner, A., Linn, J., Ziemssen, T., & Brandt, M. D. (2021). Relation of retinal and hippocampal thickness in patients with amnestic mild cognitive impairment and healthy controls. *Brain and Behavior*, e02035. https://doi.org/10.1002/brb3.2035

Ko, F., Muthy, Z. A., Gallacher, J., Sudlow, C., Rees, G., Yang, Q., Keane, P. A., Petzold, A., Khaw, P. T., Reisman, C., Strouthidis, N. G., Foster, P. J., Patel, P. J., & for the UK Biobank Eye & Vision Consortium. (2018). Association of retinal nerve fiber layer thinning with current and future cognitive decline: A study using optical coherence tomography. *JAMA Neurology*, *75*(10), 1198–1205. https://doi.org/10.1001/jamaneurol.2018.1578

London, A., Benhar, I., & Schwartz, M. (2013). The retina as a window to the brain—From eye research to CNS disorders. *Nature Reviews Neurology*, *9*(1), 44–53. https://doi.org/10.1038/nrneurol.2012.227

Mejia-Vergara, A. J., Karanjia, R., & Sadun, A. A. (2021). OCT parameters of the optic nerve head and the retina as surrogate markers of brain volume in a normal population, a pilot study. *Journal of the Neurological Sciences*, *420*, 117213. https://doi.org/10.1016/j.jns.2020.117213

Méndez-Gómez, J. L., Pelletier, A., Rougier, M.-B., Korobelnik, J.-F., Schweitzer, C., Delyfer, M.-N., Catheline, G., Monfermé, S., Dartigues, J.-F., Delcourt, C., & Helmer, C. (2018). Association of Retinal Nerve Fiber Layer Thickness With Brain Alterations in the Visual and Limbic Networks in Elderly Adults Without Dementia. *JAMA Network Open*, *1*(7), e184406. https://doi.org/10.1001/jamanetworkopen.2018.4406

Musiek, E. S., & Morris, J. C. (2021). Possible Consequences of the Approval of a Disease-Modifying Therapy for Alzheimer Disease. *JAMA Neurology*, *78*(2), 141. https://doi.org/10.1001/jamaneurol.2020.4478

Ng, W. Y., Cheung, C. Y., Milea, D., & Ting, D. S. W. (2021). Artificial intelligence and machine learning for Alzheimer's disease: Let's not forget about the retina. *British Journal of Ophthalmology*, *105*(5), 593–594. https://doi.org/10.1136/bjophthalmol-2020-318407

Ong, Y.-T., Hilal, S., Cheung, C. Y., Venketasubramanian, N., Niessen, W. J., Vrooman, H., Anuar, A. R., Chew, M., Chen, C., Wong, T. Y., & Ikram, M. K. (2015). Retinal neurodegeneration on optical coherence tomography and cerebral atrophy.

Neuroscience Letters, *584*, 12–16. https://doi.org/10.1016/j.neulet.2014.10.010
Santos, C. Y., Johnson, L. N., Sinoff, S. E., Festa, E. K., Heindel, W. C., & Snyder, P. J. (2018). Change in retinal structural anatomy during the preclinical stage of Alzheimer's disease. *Alzheimer's & Dementia: Diagnosis, Assessment & Disease Monitoring*, *10*(1), 196–209. https://doi.org/10.1016/j.dadm.2018.01.003
Uchida, A., Pillai, J. A., Bermel, R., Jones, S. E., Fernandez, H., Leverenz, J. B., Srivastava, S. K., & Ehlers, J. P. (2020). Correlation between brain volume and retinal photoreceptor outer segment volume in normal aging and neurodegenerative diseases. *PLOS ONE*, *15*(9), e0237078. https://doi.org/10.1371/journal.pone.0237078

DATA SECURITY AGREEMENT

Provisional Paper Title	Associations between retinal neuronal and microvascular parameters and brain structural integrity: Is the retina a biomarker of brain health?
Proposing Author	Ashleigh Barrett-Young
Today's Date	6/12/2021

Please keep one copy for your records and return one to the PI Sponsor

Please initial your agreement: (customize as necessary)

✓	I am current on Human Subjects Training [CITI www.citigrogram.org] or equivalent.
✓	My project is covered by the Dunedin Study's ethics approval OR I have /will obtain ethical approval from my home institution (please specify).
✓	I will treat all data as "restricted" and store in a secure fashion. My computer or laptop is: • encrypted (recommended programs are FileVault2 for Macs, and Bitlocker for Windows machines) • password-protected • configured to lock-out after 15 minutes of inactivity AND • has an antivirus client installed as well as being patched regularly.
✓	I will not "sync" the data to a mobile device.
✓	In the event that my laptop with data on it is lost, stolen or hacked, I will immediately contact my PI Sponsor or Study Director, Richie Poulton (richie.poulton@otago.ac.nz).
✓	I will not share the data with anyone, including my students or other collaborators not specifically listed on this concept paper.
✓	I will not post data online or submit the data file to a journal for them to post. *Some journals are now requesting the data file as part of the manuscript submission process. The Dunedin Study Members have not given informed consent for unrestricted open access, so we have a managed-access process. Speak to your PI Sponsor or Richie Poulton for strategies for achieving compliance with data-sharing policies of journals.*
✓	I will delete all data files from my computer after the project is complete. Collaborators and trainees may not take a data file away from the office. The data remains the property of the Study and cannot be used for further analyses without an approved concept paper for new analyses.

Signature: _Ashleigh Barrett-Young_____

CONCEPT PAPER RESPONSE FORM

A To be completed by the proposing author:

Provisional Paper Title	Associations between retinal neuronal and microvascular parameters and brain structural integrity: Is the retina a biomarker of brain health?
Proposing Author	Ashleigh Barrett-Young
Other Contributors	Richie Poulton, Graham Wilson, Jesse Gale, Terrie Moffitt, Avshalom Caspi, Ahmad Hariri, Annchen Knodt, Ross Keenan, Cliff Abraham, Tracy Melzer, Tien Yin Wong, Yih Chung Tham, David Ireland, Sandhya Ramrakha, Sean Hogan
Potential Journals	Molecular Psychiatry
Today's Date	6 December 2021
Intended Submission Date	September 2022

Please keep one copy for your records and return one to the proposing author

B. To be completed by potential co-authors:

☐ Approved ☐ Not Approved ☐ Let's discuss, I have concerns

Comments: _____

_____

_____

_____

_____

Please check your contribution(s) for authorship:

	Conceptualizing and designing the longitudinal study
	Conceptualizing data collection protocols and creating variables
	Data collection
	Conceptualizing and designing this specific paper project
	Statistical analyses and interpretation (or reproducibility check)
	Writing
	Reviewing manuscript drafts
	Final approval before submission for publication

	Agreement to be accountable for the work
	Acknowledgment only, I will not be a co-author

Signature: _____

Name: _____