Can erythrocyte proteins be used as dementia biomarkers?

Dianne Marquez Lopez*, Paul Khoo, Elizabeta Mukaetova-Ladinska
Institute of Neuroscience, Campus of Ageing and Vitality, Newcastle University
* d.m.lopez@newcastle.ac.uk

Aims
To explore and identify erythrocyte proteins that can be used as blood biomarkers for dementia, by measuring changes in erythrocyte membrane protein levels in demented and control subjects.

Introduction
- Alzheimer’s Disease (AD), Vascular dementia (VaD) and Dementia with Lewy bodies (DLB) are the leading causes of dementia in the older people. Improving current diagnostic methods to facilitate early dementia treatments remains an optimal goal. Recent research primarily focuses on plasma proteins as dementia peripheral (blood) biomarkers, whereas blood cellular components, including erythrocytes (ERC), are poorly investigated.
- ERC’s shape and morphology relies on the internal structures of cytoskeletal membrane. ERC proteins such as Ubiquitin and IgG play an important role in membrane integrity and stability (Figure 1).
- Ubiquitin is important for protein-degradation by interacting with ERC proteasomes. Failure to activate ubiquitination of cytoskeletal proteins may result to pathological changes in ERC morphology that results in AD (2).

Materials and Methods
- Blood samples (n=72) came from: 28 controls, 17 AD, 20 VaD and 7 DLB subjects.
- All participants had an extensive clinical assessment for their cognitive and behavioural functioning.
- Blood samples were processed to obtain plasma, platelets and ERC fractions (Figure 2).
- Novel ELISA immunoassays were developed and used to measure Ubiquitin and IgG using commercially available immuno probes.

Results
- Mean protein concentration for Ubiquitin and IgG for each subject.

Discussion
- This is the first study to explore the clinical utility of ERC protein content for diagnosis of dementia.
- ERC ubiquitin levels were highest in AD subjects. Control and dementia subjects had similar ERC IgG showing no significant difference (Figure 3).
- The ubiquitin ERC measures had high accuracy to distinguish dementia from cognitively intact subjects (74.8%) (Figure 4).

Conclusion
Further work is needed to validate the ERC ubiquitin assay in a larger clinical sample with various extent of cognitive impairment and aetiologies so that it can be used in routine clinical setting for dementia diagnosis.

Figure 1: ERC proteins and dementia pathology. Western blot refers to a control ERC sample at pH 5.7. 1, IgG, 2, ubiquitin. (*) indicates location of target protein size.

Figure 2: ERC extraction (pH5.7) and use in Western blot and ELISAs.

Figure 3: Ubiquitin (a) and IgG (b) ERC content in control and dementia subjects. Y-axes values represent Relative Values (%). ** p<0.01; ***, p=0.0001.

Figure 4: Area under the curve (AUC) indicating sensitivity and specificity of Ubiquitin and IgG as ERC dementia biomarkers.

Reference
1. Mohrnty et al. Proteome Sci 2010, 8; 1