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Are Inflammatory Biomarkers the New Frontier in Alzheimer's Disease Research?

JACOBO MINTZER, MD, MBA

PSYCHIATRIST, RALPH. H. JOHNSON VAMC

DIRECTOR, SOUTH CAROLINA INSTITUTE FOR BRAIN HEALTH

LOWCOUNTRY CENTER FOR VETERANS RESEARCH

RALPH H. JOHNSON VA MEDICAL CENTER AFFILIATED 501C3

PROFESSOR, COLLEGE OF HEALTH PROFESSIONS

MEDICAL UNIVERSITY OF SOUTH CAROLINA

Disclosures

Consulting

- AARP – Global Council on Brain Health
- AARP – Staying Sharp
- ACADIA
- Alzheimer's Association, South Carolina Chapter
- Alzheimer's Clinical Trials Consortium
- Alzheimer's Disease Cooperative Study
- Biopharma Connex
- Corium
- Elder Court
- Exciva
- Genentech (affiliate of F. Hoffman-La Roche Ltd.)
- International Psychogeriatric Association
- Ironshore Pharmaceuticals
- ICG Pharmaceuticals
- Lundbeck
- ***NeuroQuest***
- Praxis Bioresearch
- Recruitment Partners
- Sumitomo
- Sygnature Discovery
- Technology Accelerator Company

Research

- Alzheimer's Association
- Cerevel Therapeutics
- Eisai Inc.
- National Endowment for the Arts
- National Institute on Aging
- National Institute of Health

NeuroQuest

Identifying Alzheimer's Disease

- Alzheimer's disease (AD) is a neurodegenerative disorder characterized by cognitive impairment, functional deterioration, and dementia.
- Pathologically, AD is defined by the presence of amyloid plaques and neurofibrillary tangles due to the accumulation of 42-amino-acid amyloid-β peptide ($A\beta_{42}$) and phosphorylated tau protein.[1]
- The diagnosis of AD now relies on clinical assessment and information from imaging and biofluid markers aimed to detect the presence of $A\beta_{42}$ and phosphorylated tau protein by using[2]:
  - Positron emission tomography (PET)
  - Neuroimaging
  - Cerebrospinal fluid (CSF) measurements.
- Evidence has shown that amyloid lesions may be present in the brain up to 20 years before the onset of symptoms.

1. Scheltens P, Blennow K, Breteler MMB, et al. Alzheimer's disease. The Lancet. 2016;388:505-517.
2. Jack CR Jr, Bennett DA, Blennow K, et al. A/T/N: an unbiased descriptive classification scheme for Alzheimer disease biomarkers. Neurology. 2016;87:539-547.

The Future of Alzheimer's Disease Treatment and Clinical Research

- There are currently two drugs that have been approved by the FDA with the ability the reduce amyloid load in the brain:
  - Aducanumab
  - Lecanemab
- These drugs appear to be more effective the earlier in the disease process treatment is started.
- The challenge is to find an easy-to-administer blood-based biomarker that will identify asymptomatic patients presenting with very low loads of amyloid in the brain.

Mechanism of Action of New Treatments

- Aducanumab and lecanemab are monoclonal antibodies that block the amyloid formation chain at different stages.
- Specifically, lecanemab is a humanized immunoglobulin gamma 1 (IgG1) monoclonal antibody.
  - It is directed against aggregated soluble and insoluble forms of $A\beta_{42}$ for the treatment of AD.
  - It prevents the attachment of the fibrils at the membrane level of brain cells.

Limitations of Current Available Biomarkers

- Current available biomarkers rely on:
  - The presence of amyloid in plasma and/or its metabolites.
  - The presence of tau and/or its metabolites.
- Although the specificity of these biomarkers is very high, the sensitivity is unknown.
- It is safe to assume there must be a minimal threshold of brain amyloid load for these tests to become positive. However, the required level of the amyloid load remains unknown.

The Unmet Need

- New clinical trials will focus on disease prevention in asymptomatic individuals.
- Researchers will need to identify not only the individuals who will accumulate amyloid in the brain, but also those who are likely to develop symptoms of dementia when amyloid accumulates in their brain.
- Approximately 10-20% of individuals showing high amyloid loads in their brain do not show, and will never show, symptoms of dementia, even when 100 years of age or more.
- The question is – are there other specific characteristics that will determine the likelihood of the individuals who will develop symptoms of dementia in the presence of amyloid in their serum and/or brain?

Potential Immune-Based Theory to Understand Alzheimer's Disease

- Schwartz and colleagues[1] demonstrated the presence of an active interchange of immune cells between the central nervous system (CNS) and the periphery in the choroid plexus in animal models of AD.
- Interferon (IFN)-γ-dependent recruitment of monocyte-derived macrophages to the brain resulted in the clearance of cerebral Aβ plaques and improved cognitive performance in a mouse AD model.[2]
- These findings suggest the presence of immune deficits that "permit" the accumulation of amyloid.[2]
- The identification of these "immune deficits", especially in individuals who present with amyloid in their serum and/or brain, will provide a more accurate predictor of an individual's risk to develop symptoms of dementia.

1. Kunis G, Baruch K, Rosenzweig N, et al. IFN-gamma-dependent activation of the brain's choroid plexus for CNS immune surveillance and repair. Brain. 2013;136:3427-3440.
2. Baruch K, Deczkowska A, Rosenzweig N, et al. PD-1 immune checkpoint blockade reduces pathology and improves memory in mouse models of Alzheimer's disease. Nat Med. 2016;22:135-137.
3. Huang, X., Li, Y., Fowler, C., Doecke, J. D., Lim, Y. Y., Drysdale, C., Zhang, V., Park, K., Trounson, B., Pertile, K., Rumble, R., Pickering, J. W., Rissman, R. A., Sarsoza, F., Abdel-Latif, S., Lin, Y., Doré, V., Villemagne, V., Rowe, C. C., Fripp, J., … Gu, B. J. (2022). Leukocyte surface biomarkers implicate deficits of innate immunity in sporadic Alzheimer's disease. Alzheimer's & dementia : the journal of the Alzheimer's Association, 10.1002/alz.12813. Advance online publication.

Identifying Immune-Based Biomarkers in Serum for Alzheimer's Disease

- The goal of the experiments we will describe was to identify potential immune-based biomarkers that would predict an individual's risk to develop symptoms of cognitive impairment.
- We hypothesized that individuals who progressed to symptomatic AD would have, in addition to amyloid deposition in the brain, an additional risk that would allow the amyloid deposition to result in brain damage.
- We suggested that the risk was an immune deficit that interferes with the regulation of the inflammatory response the brain generates in the presence of amyloid.
- In order to identify those deficits, we had to demonstrate the presence of an immune deficit in all stages of AD.

Study Design and Methods

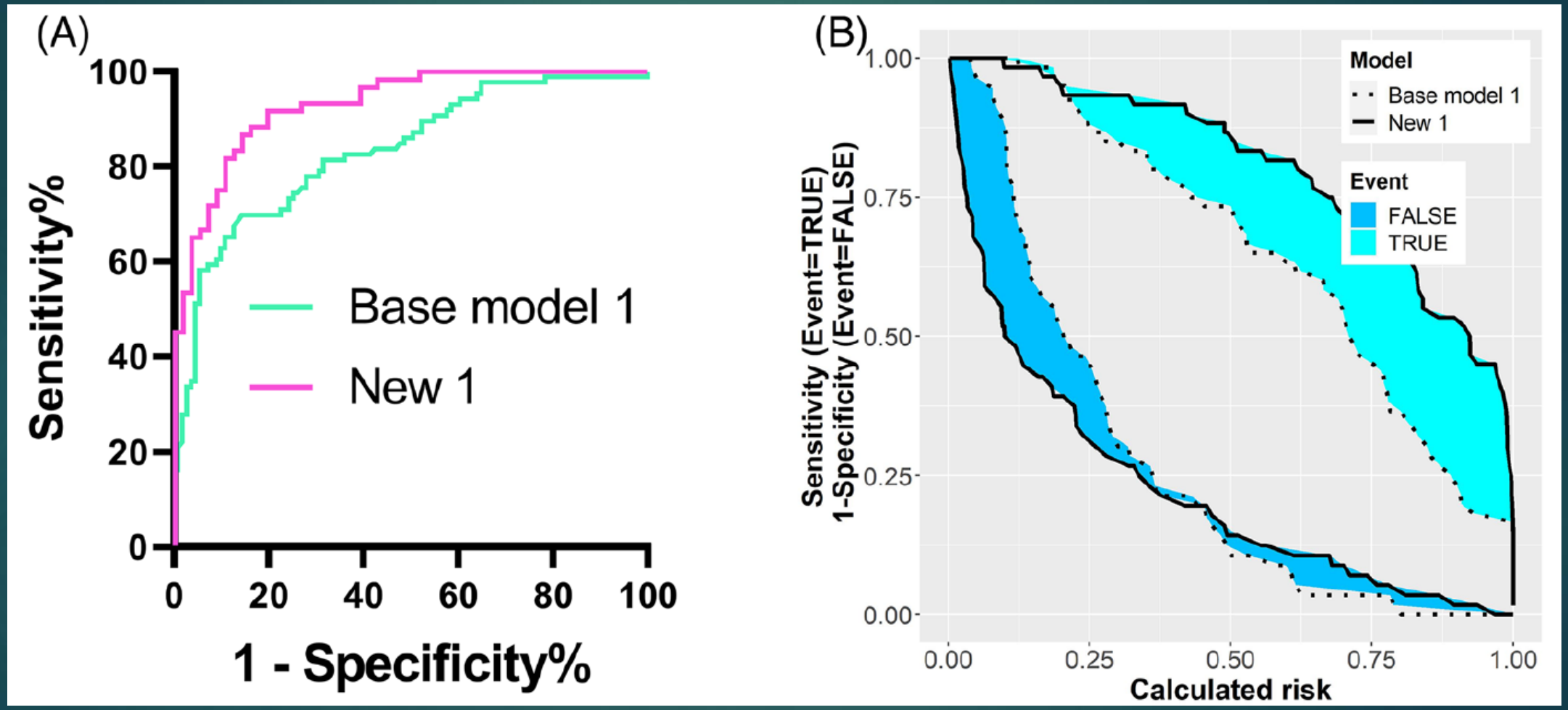
- A cross-sectional study comprised four stages was conducted.
  - Stage I was exploratory research to investigate major leukocyte markers.
    - 76 Subjects
    - A total of 34 leukocyte antigens were examined by flow cytometry immunophenotyping. 13 were differentially expressed in patients with clinically defined MCI/AD-dementia.
  - Stage II was an update for Stage I
    - 142 Subjects
    - Stage II confirmed the decreased expression of CD11c in the MCI/AD-dementia compared to CN.
  - Stage III was designed to prospectively confirm the findings from previous stages.
    - 200 Subjects
    - 11 leukocyte markers were differentially expressed, including five upregulated markers and six downregulated markers.
  - Stage IV was designed to confirm the findings from Stage III.
    - 112 Subjects

Biological relevance and changes of potential biomarkers in AD

GO class (direct)	Potential biomarkers	Biological relevance to AD	Changes in AD
Amyloid-beta binding and microglial cell activation	CD11b	Integrin subunit alpha M; complement component receptor 3 alpha; phagocytosis; integrins; C3b binding.	Stage I, III Nil; Stage II ↓
Integrin binding	CD11c	Integrin subunit alpha X; complement component receptor 4 alpha; regulation of actin cytoskeleton.	Stage I, II, III ↓
Amyloid-beta binding and microglial cell activation	CD18/ITGB2	Integrin subunit beta 2; combines with different alpha chains, for example, CD11b and CD11c, to form different integrin heterodimers also referred to as CR3 and CR4.	Stage III ↓
Protein binding	CD33	GWAS associated gene for AD[17]; myeloid cell surface antigen; immunoglobulin superfamily cell adhesion molecule.	Stage II ↓
Complement component C3b/4b binding	CD35/CR1	GWAS associated gene for AD[18]; phagocytosis; integrins; complement receptor 1.	Stage III ↑
Amyloid-beta binding and antigen processing and presentation	CD36	Scavenger receptor class B member 1; phagocytosis; receptor for oxidized low-density lipoprotein (LDL)[19]; receptor for Aβ.[20]	Stage III ↓
Protein binding	CD59	Complement inhibitory protein; prevent formation of the complement membrane attack complex (MAC).[21]	Stage III ↑
Amyloid-beta binding and scavenger receptor activity	CD91	Genetically associated wi AD[22–24]; low-density lipoprotein receptor-related protein 1; mediates the endocytosis and degradation of secreted amyloid precursor protein.[25]	Stage III ↑
Scavenger receptor activity	CD163	Scavenger receptor.	Stage III ↓
Protein binding and protein phosphorylation	MerTK	Receptor protein-tyrosine kinase; mediates phagocytosis in microglial cells[26] and retinal pigment epithelium.[27]	Stage III ↓
Extracellularly ATP-gated cation channel activity	P2X7	Innate phagocytosis.[2]	Stage I, III ↓; Stage II Nil
Amyloid-beta binding and microglial cell activation	RAGE/AGER	Mediates Aβ transport across the blood-brain barrier and accumulation in brain[28,29].	Stage III ↑
Amyloid-beta binding and scavenger receptor activity	SCARA-1/MSR1	Macrophage scavenger receptor 1; phagocytosis; mediates uptake of fibrillar amyloid.[30]	Stage III ↑

GO: Gene ontology.
↑up-regulation.
↓down-regulation.
Nil: No change.

Performance Assessment of the Top Four AD-Associated Leukocyte Markers



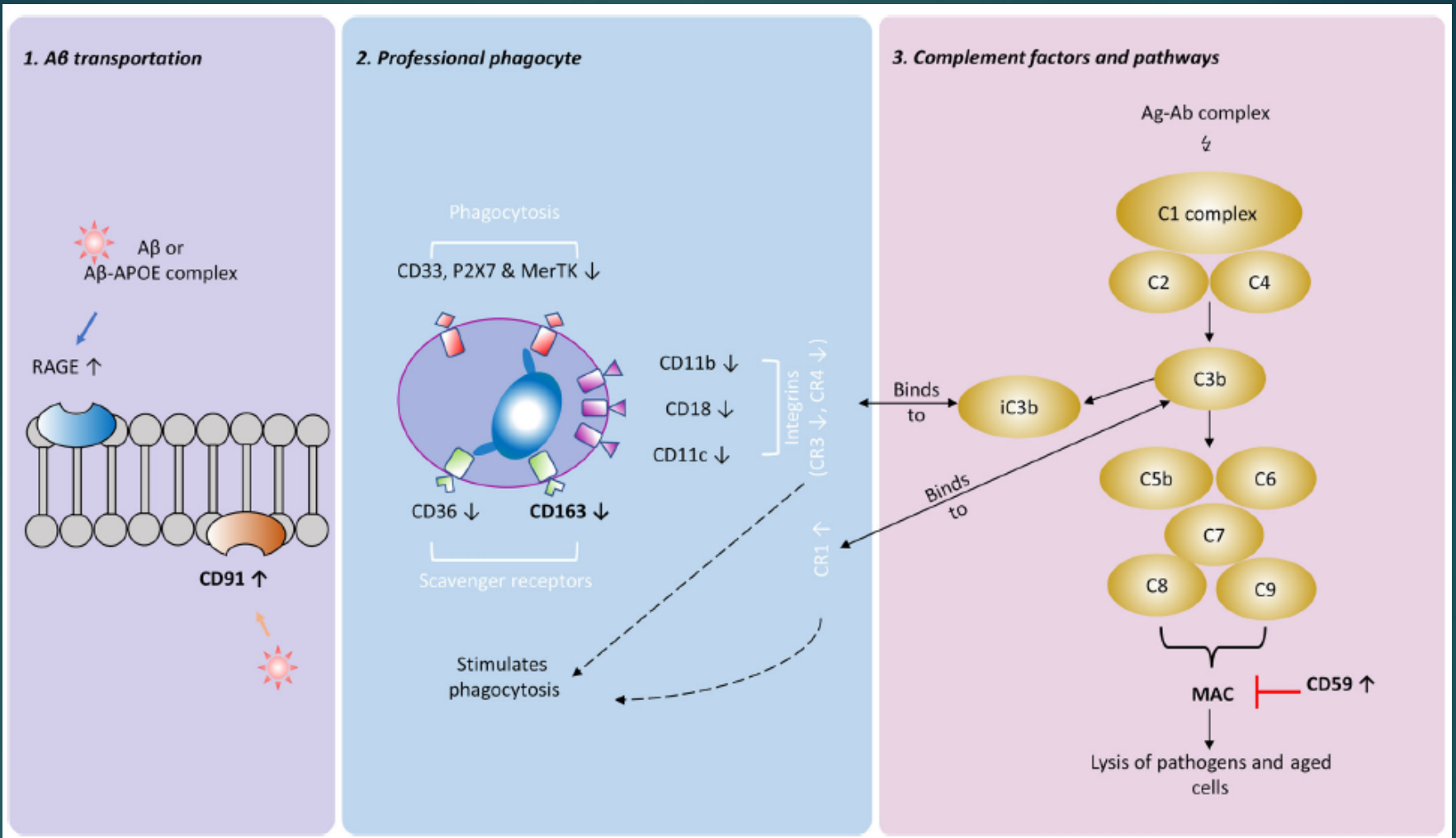


FIGURE 4 Scientific hypothesis and molecular pathways inhibiting phagocytosis and complement. Left. Aβ cross-membrane transportation is augmented given that both CD91 and RAGE increased in AD; Middle. A schematic phagocyte shows down-regulation in phagocytic receptors, MerTK and P2X7; scavenger receptors, CD36 and CD163; and cell adhesion integrins, CD11b, CD11c, and CD18, which can form CR3 and CR4; Right (adapted from KEGG map04610). Complement pathways are activated by antigen-antibody complex. It triggers cascade to form C3b. C3b binds to CR1 thatis upregulated in AD, and subsequentlystimulates phagocytosis. Whilst, iC3b binds to CR3 and CR4 thatare downregulated in AD, and subsequently inhibit phagocytosis. On the other hand, C5b, C6, C7, C8, and C9 form the membrane attack complex (MAC). CD59 prevents formation of MAC and lysis of target cells. Both inhibit opsonization, phagocytosis and clearance of pathogens which are involved in AD pathogenesis and development.

Developing a Biomarker Algorithm

- Leukocyte markers differentially expressed in the patients with AD were identified.
- Pathway analysis revealed a complex network involving upregulation of complement inhibition and downregulation of cargo receptor activity and Aβ clearance.
- A proposed panel including four leukocyte biomarkers – **CD11c, CD59, CD91, and CD163** – was found to predict patients' PET Aβ status with 97% sensitivity and 88% specificity.

Summary Results

- This blood-based test:
  - Presents with 97% sensitivity and 88% specificity.
  - Is independent of the amyloid load level in the individual's brain.
  - Could potentially predict the likelihood of the individual to develop dementia.
- The detection of the immune markers at the cell membrane level may be relevant to specific treatment approaches.
- These findings need to be confirmed in longitudinal studies.

Conclusion

- The discovery of changes in peripheral leukocyte surface antigens in AD implicate the deficit in innate immunity.
- Leukocyte-based biomarkers prove to be both sensitive and practical for AD screening, diagnosis, and prognosis.