NetraAI Reveals Subpopulations of Bulbar and Limb ALS Using Transcriptomic Analysis

Joseph Geraci[1,2,3,4], Ravi Bhargava[5,6], Bessi Qorri[1], Paul Leonchyk[1], Douglas Cook[1,7], Moses Cook[8], Fanny Sie[6], Luca Pani[1,9,10,11]

[1]NetraMark Corp, [2]Department of Pathology and Molecular Medicine, Queen's University, [3]Centre for Biotechnology and Genomic Medicine, Medical College of Georgia, Augusta University, [4]Arthur C. Clarke Center for Human Imagination, School of Physical Sciences, University of California San Diego, [5]Department of Biomedical and Molecular Sciences, Queen's University, [6]Science and Research, Roche Integrated Informatics, F. Hoffmann La-Roche, [7]Department of Surgery, Queen's University, [8]Department of Medical Biophysics, University of Toronto, [9]Department of Psychiatry and Behavioral Sciences, Leonard M. Miller School of Medicine, University of Miami, [10]Department of Biomedical, Metabolic, and Neural Sciences, University of Modena and Reggio Emilia, [11]VeraSci

BACKGROUND

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative and neuromuscular disease that can present with either bulbar or limb onset.

CHALLENGES:

- Bulbar onset is associated with rapid disease progression and poor prognosis, requiring immediate and aggressive intervention.
- Limb onset progresses more slowly and allows for more time to explore treatment options.

NEED:

- Understanding of the molecular mechanisms underlying bulbar and limb onset of ALS to identify targeted therapies.

OBJECTIVE:

Use a machine learning (ML) systems-level approach to analyze ALS patient data to identify covariates and clusters of factors that provide a classification of disease onset and potential therapeutic targets specific to each subtype.

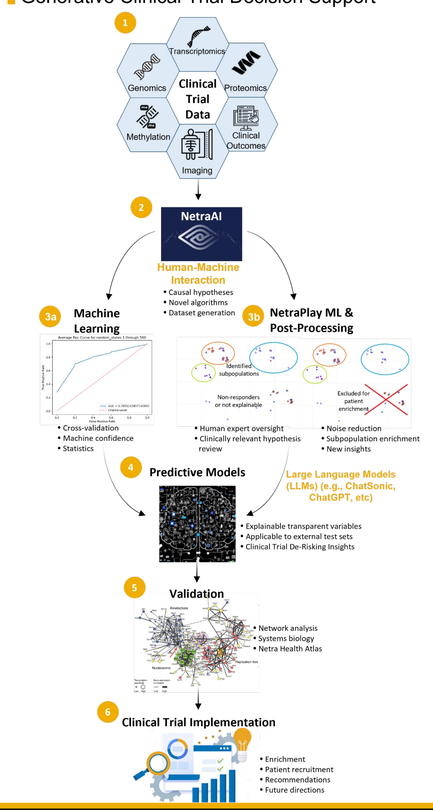
METHODOLOGY & WORKFLOW

DATASET:

- Publicly available dataset provided by patient advocacy groups AnswerALS and EverythingALS.
- 31 bulbar onset and 85 limb onset ALS patients from 8 neuromuscular clinics in the US.
- iPSC lines generated from patients' peripheral blood mononuclear cells.
- Consortium generated multi-omics data, focusing on transcriptomics.

PERSPECTIVE ANALYTICS AND HYPOTHESIS-GENERATION USING NETRAAI:

Using NetraAI to discover unknown subpopulations that are defined by multiple genes and provide explanations for underlying driving mechanisms behind each subpopulation.



RESULTS

NetraAI identified genetic drivers of a subpopulation of limb onset ALS patients.

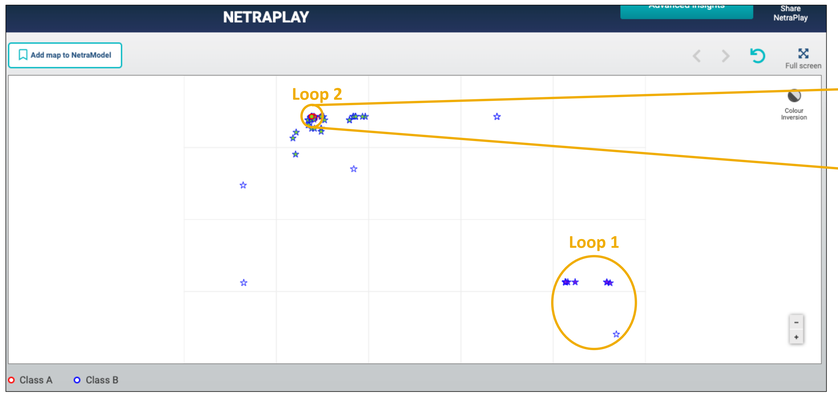


Figure 1. NetraPlay map of limb and bulbar onset ALS patients. Loop 1 corresponds to a subpopulation of limb onset ALS patients. Loop 2 corresponds to a subpopulation of bulbar onset ALS patients.

A subpopulation of limb onset ALS patients was identified (Loop 1) that were characterized by higher expression of:

- *IL20RA*
- *LRRC23*

The remaining limb onset patients, accounting for the majority of patients in the dataset, were characterized by the opposite expression of the bulbar subpopulation (Loop 2):

- Higher expression of: *TBC1D20, ALG3P1, CROCC2, AC109439.1, FAM151A,* and *NKX21-1-AS1*
- Lower expression of *TMEM14A*

NetraAI identified a subpopulation of bulbar onset ALS patients.

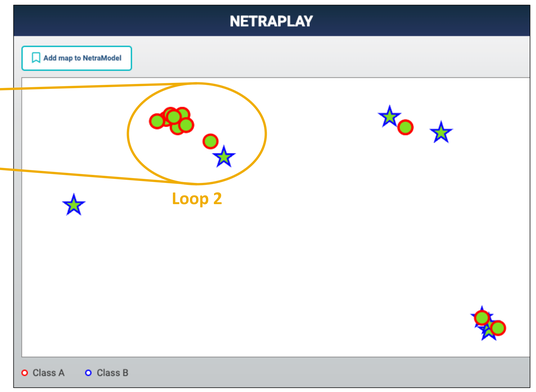


Figure 2. Zoomed NetraPlay map highlighting a subpopulation of bulbar onset ALS patients.

Using the interactive and "zoom" ability of NetraPlay, we identified a pure subpopulation of bulbar onset ALS patients characterized by:

- Lower expression of: *TBC1D20, ALG3P1, CROCC2, AC109439.1, FAM151A,* and *NKX21-1-AS1*
- Higher expression of *TMEM14A*

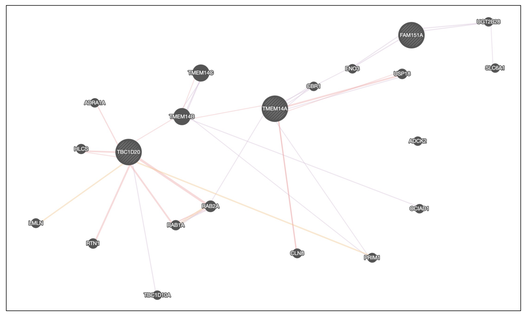


Figure 3. Gene interaction network with drivers of interest.

A gene interaction network was constructed, consisting of *TMEM14A* and *FAM151A.* A link to *RAB1* and *RAB2,* was identified, suggesting dysfunction in ER-Golgi transport is characteristic in bulbar onset ALS, with mutant SOD-1.

CONCLUSIONS & SIGNIFICANCE

SIGNIFICANCE OF METHODOLOGY AND FINDINGS

Identification of limb and bulbar onset ALS patient subpopulations within a heterogeneous ALS patient population dataset.

- In an Open Science challenge using this dataset, we were the only group to discern this subpopulation stratification.
- The high expression of *TMEM14A* and *FAM151A* linked to *RAB1* and *RAB2* present as unique findings linking intracellular transport dysfunction to mutant *SOD1* that is characteristic of ALS patients and warrants further investigation in the progression and treatment of bulbar onset ALS patients.
- Identification of molecular subpopulations may inform targeted therapeutic approaches.

Clinical Trial Significance

Empowering Biostatistics Through Clear Hypotheses & Causal Variables

Having a system capable of generating hypotheses that are dually scrutinized for:

- Statistical significance testing
- Biological plausibility

results in a better understanding of patient populations.

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