## Antibody-functionalized aluminum oxide-coated particles targeting neutrophil receptors in a multifrequency microfluidic impedance cytometer

Brandon K Ashley Department of Biomedical Engineering, Rutgers, the State University of New Jersey, Piscataway, NJ, 08854

Jianye Sui Department of Electrical Engineering, the State University of New Jersey, Piscataway, NJ, 08854

Mehdi Javanmard Department of Electrical Engineering & Department of Biomedical Engineering Rutgers, the State University of New Jersey, Piscataway, NJ, 08854

Umer Hassan\* Department of Electrical Engineering, Department of Biomedical Engineering, and Global Health Institute Rutgers, the State University of New Jersey, Piscataway, NJ, 08854

\*Corresponding Author. Email: <u>umer.hassan@rutgers.edu</u>

**Electronic Supplemental Information (ESI)** 



ESI Fig. 1: Flow chart of A) signal acquisition and B) digital signal processing to obtain electrical signatures from multifrequency microfluidic impedance cytometer.



PMT2: PE-CD11b fluorescence intensity (a. u.)

ESI Fig. 2: Two-stain and gated flow cytometry results for CD11b expression (stained with phycoerythrin or PE) and CD66b expression (stained with fluorescein isothiocyanate or FITC) as reference receptor expression to compare with multifrequency impedance cytometry results.

ESI Table 1: Percentage of neutrophils with significant receptor expression of CD11b, CD66b, or both measured from flow cytometry

No expression (Q1)	CD11b+ only (Q2)	CD66b+ only (Q3)	CD11b+ & CD66b+ (Q4)
35.3%	27.7%	4.3%	32.7%



ESI Fig. 3: Additional scatter plot comparisons between different frequencies for bipolar pulse data collected. (red: isolated neutrophils alone, green: neutrophils combined with 10 nm aluminum oxide coated Janus microparticles (MOJPs) functionalized with anti-CD11b antibodies, blue: neutrophils combined with 30 nm MOJPs functionalized with anti-CD66b antibodies).

Machine Learning Model	Accuracy	ROC AUC	Sensitivity	Selectivity
Fine Tree	73.30%	82.00%	67.7%	79.0%
Medium Tree	73.6%	82.0%	68.8%	78.5%
Coarse Tree	72.1%	78.0%	70.1%	74.0%
Gaussian Naïve Bayes	68.1%	80.0%	45.1%	91.1%
Kernel Naïve Bayes	73.5%	82.0%	65.5%	81.5%
Linear Support Vector Machine (SVM)	73.4%	80.0%	61.7%	88.4%
Quadratic SVM	73.7%	82.0%	62.8%	84.7%
Cubic SVM	63.1%	72.0%	84.1%	42.1%
Fine Gaussian SVM	74.8%	82.0%	73.7%	75.9%
Medium Gaussian SVM	75.4%	83.0%	67.5%	83.3%
Coarse Gaussian SVM	73.2%	80.0%	61.6%	84.9%
Fine K-Nearest Neighbors (KNN)	68.0%	68.0%	66.9%	69.0%
Medium KNN	73.7%	82.0%	71.0%	76.3%

ESI Table 2: Cells alone vs. Cells/10nmMOJP/anti-CD11b all Machine Learning Results

Coarse KNN	74.0%	83.0%	64.5%	83.5%
Cosine KNN	73.9%	81.0%	73.4%	74.4%
Cubic KNN	73.4%	81.0%	70.3%	76.5%
Weighted KNN	73.7%	81.0%	68.4%	78.9%
Boosted Trees	75.1%	84.0%	72.1%	79.0%
Bagged Trees	74.9%	80.0%	75.7%	74.2%
Subspace KNN	70.2%	77.0%	69.2%	71.1%
Narrow Neural Network	75.8%	85.0%	74.0%	78.4%
Medium Neural Network	74.6%	84.0%	72.3%	76.9%
Wide Neural Network	71.3%	80.0%	71.1%	71.4%
Bilayered Neural Network	75.2%	84.0%	73.6%	76.8%
Trilayered Neural Network	74.6%	84.0%	71.4%	77.8%

Machine Learning Model	Accuracy	ROC AUC	Sensitivity	Selectivity
Fine Tree	72.1%	78.0%	76.4%	82.5%
Medium Tree	68.1%	80.0%	69.6%	85.5%
Coarse Tree	73.5%	82.0%	73.0%	75.9%
Gaussian Naïve Bayes	73.4%	80.0%	65.7%	86.8%
Kernel Naïve Bayes	73.7%	82.0%	69.0%	83.5%
Linear SVM	63.1%	72.0%	69.1%	85.3%
Quadratic SVM	74.8%	82.0%	80.0%	85.7%
Cubic SVM	75.4%	83.0%	86.9%	69.4%
Fine Gaussian SVM	73.2%	80.0%	80.9%	79.1%
Medium Gaussian SVM	82.3%	91.0%	78.5%	86.1%
Coarse Gaussian SVM	78.6%	85.0%	68.5%	88.6%
Fine KNN	75.6%	76.0%	76.4%	75.0%
Medium KNN	80.0%	89.0%	81.6%	78.4%
Coarse KNN	80.8%	90.0%	91.7%	84.1%

ESI Table 3: Cells alone vs. Cells/30nmMOJP/anti-CD66b all Machine Learning Results

Cosine KNN	80.0%	88.0%	78.3%	81.8%
Cubic KNN	80.4%	89.0%	81.8%	79.0%
Weighted KNN	80.0%	89.0%	79.5%	80.5%
Boosted Trees	81.5%	90.0%	78.4%	84.5%
Bagged Trees	81.3%	90.0%	80.6%	82.0%
Subspace KNN	80.1%	89.0%	79.1%	81.1%
Narrow Neural Network	82.8%	92.0%	81.6%	84.0%
Medium Neural Network	82.3%	91.0%	81.8%	82.8%
Wide Neural Network	78.3%	88.0%	78.4%	78.1%
Bilayered Neural Network	82.7%	91.0%	82.5%	83.1%
Trilayered Neural Network	82.0%	91.0%	81.9%	82.1%

ESI Table 4: Cells/10nmMOJP/anti-CD11b vs. Cells/30nmMOJP/anti-CD66b all Machine Learning Results

Machine Learning Model	Accuracy	ROC AUC	Sensitivity	Selectivity
Fine Tree	75.7%	81.0%	80.5%	70.9%
Medium Tree	76.0%	82.0%	83.5%	68.4%
Coarse Tree	73.2%	76.0%	91.0%	55.4%
Gaussian Naïve Bayes	61.4%	76.0%	90.8%	31.9%
Kernel Naïve Bayes	72.9%	80.0%	76.6%	69.2%
Linear SVM	64.9%	70.0%	68.5%	61.2%
Quadratic SVM	79.5%	87.0%	89.3%	69.2%
Cubic SVM	77.9%	84.0%	83.9%	72.0%
Fine Gaussian SVM	75.9%	82.0%	75.2%	76.5%
Medium Gaussian SVM	78.8%	87.0%	88.0%	69.6%
Coarse Gaussian SVM	69.9%	77.0%	74.4%	65.4%
Fine KNN	69.5%	69.0%	71.7%	67.2%
Medium KNN	75.1%	83.0%	86.2%	64.0%

Coarse KNN	76.1%	85.0%	89.2%	63.0%
Cosine KNN	75.6%	83.0%	81.7%	69.5%
Cubic KNN	74.8%	82.0%	85.8%	63.7%
Weighted KNN	75.8%	83.0%	83.6%	67.6%
Boosted Trees	77.5%	85.0%	84.9%	70.1%
Bagged Trees	78.0%	85.0%	83.3%	72.7%
Subspace KNN	75.3%	82.0%	81.1%	69.5%
Narrow Neural Network	79.5%	87.0%	84.8%	74.2%
Medium Neural Network	78.4%	86.0%	83.3%	73.4%
Wide Neural Network	73.3%	81.0%	74.4%	72.2%
Bilayered Neural Network	78.4%	86.0%	83.2%	73.7%
Trilayered Neural Network	78.1%	86.0%	82.9%	73.3%