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Corresponding author(s): Srikanth Singamaneni

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Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see Authors & Referees and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☒	☐	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Image Studio 4.0, Horiba datastation, UVProbe 2.43, LAS V4.7, guavaSoft 3.1.1, Olympus FV1000 confocal-microscope software.
Data analysis	ImageJ 1.6.0_24, NanoScope Analysis 1.5, Origin 2016, DAS6 v6.1, Image Studio Lite 5.2, GraphPad Prism 8.0.1, FV10-ASW 3.0 Viewer, FlowJo 10.5.0

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors/reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

The main data supporting the results in this study are available within the paper and its Supplementary Information. Raw imaging data (collected and analysed via the software indicated above) are available from figshare with the identifier https://doi.org/10.6084/m9.figshare.11888055.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	No statistical method was employed to determine the sample size, and no sample size was necessary for this study. We provided indicatations that the pFLISA detects NGAL quickly in the urine of normal individuals and in the urine of patients known to have elevated urinary NGAL.
Data exclusions	No data were excluded from the analysis.
Replication	We have replicated each experiment at least three times independently, to support all of the claims made in this manuscript.
Randomization	The samples were chosen randomly.
Blinding	The investigator was not blinded to group allocation during the experiment. Blinding was not relevant because the main purpose of the study was to test the performance of the plasmonic-fluor-enhanced fluoroimmunoassay (that is, the accuracy, required assay time and sensitivity were compared to the state-of-the-art).

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology
☐	☒ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used	human IL-6 capture antibody (R&D, catalog number DY 206, lot number P173353, PART# 840113, 120-fold dilution), human IL-6 detection antibody (R&D, catalog number DY 206, lot number P173353, PART# 840114, 60-fold dilution), mouse TNF-alpha and IL-6 capture antibody which were precoated on the microbeads (R&D, catalog number LXSAMSM-03, lot number L126064, PART# 894724), mouse TNF-alpha and IL-6 detection antibody cocktail (R&D, catalog number LXSAMSM-03, lot number L126064, PART# 894666, 11-fold dilution), human kidney biomarker antibodies precoated on the NC membrane (R&D, catalog number ARY019, lot number 1311110, PART# 893967), human kidney biomarker detection antibody cocktail (R&D, catalog number ARY019, lot number 1311110, PART# 893966), anti-human HER-2/biotin (eBioscience, BMS120BT, Clone 2G11, 186281000), anti-Mo CD80/biotin (Invitrogen, 13-0801-82, Clone 16-10A1, 1934784, 5000-fold dilution), human NGAL capture antibody (R&D, catalog# DY1757, lot# P195696, PART# 844864, 120-fold dilution), human NGAL detection antibody (R&D, catalog# DY1757, lot# P195696, PART# 844865, 60-fold dilution), mouse IL-6 capture antibody (R&D, catalog# DY406-05, lot# P195781, PART# 840171, 120-fold dilution), mouse IL-6 detection antibody (R&D, catalog# DY406-05, lot# P195781, PART# 840172, 60-fold dilution)
Validation	The antibodies purchased from R&D systems have been tested for proper performance and function under their established Quality Control Testing criteria according to the certificate of analysis from the manufacturer. The anti-human HER-2/biotin and anti-Mo CD80/biotin were recommended by their provider companies for cell staining. They have been tested by Quality Control and passed internal specifications. According to the vendor, anti-human HER-2/biotin has been validated for human and anti-Mo CD80/biotin has been validated for dog, mouse, and pig. Anti-Mo CD80/biotin has been tested by flow-cytometric analysis of activated mouse splenocytes, according to the provider.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Human epithelial breast cancer cells (SK-BR-3) were purchased from ATCC.
Authentication	The SK-BR-3 cell line was authenticated via STR analysis (performed by ATCC).
Mycoplasma contamination	The cell line tested negative for mycoplasma contamination (test performed by ATCC).
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	Female mice C57BL/6 (H-2b), 5-to-6 weeks of age. Female mice C57BL/6J, 9-to-10 weeks of age.
Wild animals	The study did not involve wild animals.
Field-collected samples	The study did not involve samples collected from the field.
Ethics oversight	IACUC from Washington University in St. Louis.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Human research participants

Policy information about studies involving human research participants

Population characteristics	The characteristics of the human participants are provided in the Supplementary Information.
Recruitment	Patients with imaged renal masses were recruited under IRB 201601082 for the study, and urine samples were obtained from these patients. Consent was obtained for the use of their samples for other studies. Healthy volunteers were recruited under IRB 201202051, and urine samples were obtained from these patients. There was no perceived bias in the recruitment process.
Ethics oversight	Washington University IRB 201601082 “Nanotech Biomarkers For Renal Cancer Intervention: Clinical Validation and Utility”; Washington University IRB 201202051 "Urine Proteome of Surgical Patients and Healthy Volunteers"

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	For the isolation of BMDCs, female C57BL/6 (H-2b) mice that were 5-to-6 weeks of age were purchased from Jackson Labs (Bar Harbor, ME, USA). The mice were maintained under pathogen-free conditions. All experiments employing mice were performed in accordance with laboratory animal protocol approved by the School of Medicine Animal Studies Committee of Washington University in St. Louis. Mouse were euthanized using CO2 asphyxiation and cervical dislocation. The euthanized mouse was kept in 70% (v/v) ethanol for 1 min. Both the femurs and tibiae were isolated, and the muscle attachments were carefully removed using gauze pads. Both ends of the bones were cut with scissors and the marrow was centrifuged in an adapted centrifuge tube (0.6 ml tube with a hole inserted in 1.5 ml tube) at 1000 rpm for 10 seconds. The pellet was resuspended by vigorous pipetting in RPMI 1640 media. The cells were passed through a 70 µm cell strainer to prepare a single cell suspension. After one washing (1200 rpm, 5 min), red blood cells were depleted with RBC lysis buffer (Sigma-Aldrich). The bone marrow cells were collected and cultured in 100-mm Petri dishes containing 10 mL RPMI medium supplemented with 10% heat-inactivated FBS, 50 IU mL-1 penicillin, 50 µg mL-1 streptomycin, and 20 ng mL-1 mouse recombinant granulocyte- macrophage colony-stimulating factor (GM-CSF, R&D Systems, MN, USA). Around one million BMDCs were cultured in 6 well plates and were stimulated by adding 1 ml of different concentrations of LPS (0.5 µg/ml, 0.2 µg/ml, 0.1 µg/ml, 0.05 µg/ml, 0.01 µg/ml, and 0 µg/ml) for 24 hours. Cells were harvested using a cell scraper for further staining and flow cytometry analysis.

Instrument	Guava easyCyte
Software	Collection: guavaSoft 3.1.1; Analysis: FlowJo 10.5.0.
Cell population abundance	No sorting was performed.
Gating strategy	The cells were gated by preliminary FSC/SSC gates to exclude any dead cells or cell debris. A specific example of the gating strategy is provided in the Supplementary Information.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.