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Reporting Summary

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Statistical parameters

When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. 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
☐	☒	An indication of 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 statistics 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
☐	☒	Clearly defined error bars *State explicitly what error bars represent (e.g. SD, SE, CI)*

Our web collection on statistics for biologists may be useful.

Software and code

Policy information about availability of computer code

Data collection	Zeiss Zen Lite Imaging Software 2012, ANSYS CFX R17.2 software, SlideBook version 5.5, BioTek Gen5 Microplate Reader and Imager Software, QuantaSoft ddPCR software, and LabVIEW 2016.
Data analysis	ImageJ, Excel 2016, QuantaSoft ddPCR software, Leica Application Suite X (LAS X), R, and Origin 2016.

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 upon request. 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 authors declare that all data supporting the findings of this study are available within the paper and its Supplementary Information. The raw and analysed datasets generated during the study are available for research purposes from the corresponding author on reasonable request.

Field-specific reporting

Please select 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/authors/policies/ReportingSummary-flat.pdf

Life sciences study design

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

Sample size	As a proof-of-concept, we examined plasma samples collected from OvCa patients (n = 20) and age-matched non-cancer controls (n = 10) (Supplementary Table S2). This sample size is sufficiently large to evaluate diagnostic accuracy with reasonable statistical errors (Table S3).
Data exclusions	No data were excluded from the analyses.
Replication	All the measurements were repeated for at least three times. Negative and positive controls were included to correct the measurement when relevant.
Randomization	Human plasma samples were randomly pulled out from the KU Cancer Center's Biospecimen Repository Core Facility (BRCF) and patients were randomly selected from an age range of 50-80 years old (Table S1).
Blinding	The study was not blinded.

Reporting for specific materials, systems and methods

Materials & experimental systems

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

Methods

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

Antibodies

Antibodies used	All the antibodies used are provided in Table S1.
Validation	The antibodies have all been validated by the manufacturers, and their specificity and cross-reactivity for the protein targets of interest have been also validated using either microplate ELISA, Western Blot, or on-chip ELISA.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	COLO-1 and MCF-7 cells were cultured and EVs were purified from the culture media by HansaBioMed, Inc. SKOV3 and OVCAR3 cell lines were obtained from ATCC and cultured in the lab of Dr. Godwin (co-author) at KUMC.
Authentication	COLO-1 and MCF-7 cells were authenticated with Short Tandem Repeat (STR) profiling by the provider biobank (Interlab Cell Line Collection, Italy) for HansaBioMed, Inc. All the cell lines cultured in Dr Godwin's lab were authenticated by STR allele profiling and sequencing by an independent source (University of Arizona Genetics Core, Tucson, AZ –Cell line Authentication Core). All cell lines in Dr Godwin's lab are screened for mycoplasma contamination every 6 months.
Mycoplasma contamination	Cell lines tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

Human research participants

Policy information about studies involving human research participants

Population characteristics	Human plasma samples were obtained from the University of Kansas Cancer Center's Biospecimen Repository Core Facility under the protocol (IRB #5929) approved by the internal Human Subjects Committee. Samples were collected from 50–80 years old females diagnosed with no cancer or with stage I–IV ovarian cancer of various histological subtypes. The detailed population information is provided in Table S2.
Recruitment	No recruitment was necessary.