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Reporting Summary

Nature Portfolio 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 Portfolio policies, see our Editorial Policies 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	RADIUS All 2.2 (Build 21230) ; Q-Exactive HF X （Thermo Fisher Scientific， San Jose， CA）
Data analysis	All statistical analyses and graphics production was performed using Graph Pad Prism v.8.0 (GraphPad, La Jolla, CA, USA). All flow cytometry data were analyzed on FlowJo-V10 software. Raw data of DDA were processed and analyzed by MaxQuant (version 1.5.3.30). DIA were analyzed by Spectronaut (12.0.20491.14.21367). DEPs were assessed by MSstats R package (version: 3.2.1).

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 and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio 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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

The primary and processed proteomics data reported in this paper have been deposited in the OMIX, China National Center for Bioinformation/Beijing Institute of Genomics, Chinese Academy of Sciences (https://ngdc.cncb.ac.cn/omix: accession no. OMIX001255).

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 sample size calculation was performed. For the cohort, the maximum number of patients available were included for the study. For animal and cell culture experiments, sample size was approximated based on prior experiments and available data, which provide sufficient sample numbers to statistically detect differences between multiple experimental groups.
Data exclusions	No data was excluded from the analyses in our study.
Replication	The in vitro and in vivo experiments were performed by at least five replicates respectively, and all attempts at replication were successful. The details were described in the Figure Legends and Method sections.
Randomization	The samples/organisms/participants were allocated into experimental groups by random assignment.
Blinding	The investigators were blinded to group allocation during data collection and analyses.

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 and archaeology
☐	☒ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

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

Antibodies

Antibodies used

Flow Cytometry: FITC anti-human CD41a (BD, 555466), PE anti-human CD45 (ebioscience, 1:200, 12-9459-42), rabbit anti-mouse thrombocyte polyclonal antibody (Mybiosource, MBS524066), PE-Cy7 anti-mouse CD41 (Thermo Fisher, 25-0411-82), APC anti-human CD66b antibodies (BioLegend, 305118), PE anti-mouse Ly6G (BioLegend, 127607), Pyroptosis/Caspase-1 Assay (Green) kit (Immuno Chemistry, #9146), FAM-FLICA Caspase-3/7 Assay Kit (Immuno Chemistry, #94), FLICA 660 Caspase-3/7 Assay Kit (Immuno Chemistry, #9125), LC3B antibody Alexa Fluor 405 (NOVUSBIO, NB100-2220AF405) and Alexa Fluor 594-conjugated anti-rabbit IgG secondary antibody(Abcam, ab150084).

Immunofluorescence: goat polyclonal anti-NLRP3 (Abcam, ab4207), rabbit polyclonal anti-ASC (Novusbio, NBP1-78978), rabbit monoclonal anti-CD41 (Abcam, ab134131), Alexa Fluor 488 Anti-mouse CD41 (BioLegend, 133908), Hoechst (Thermo, H3570), mouse monoclonal anti-myeloperoxidase (Genetx, gtx75318), rabbit monoclonal anti-myeloperoxidase (Abcam, ab208670) and rabbit polyclonal anti-Histone H3 (citrulline R2 + R8 + R17, Abcam, ab5103), Alexa Fluor 488-conjugated anti-goat IgG antibody (BioLegend, 405508), Alexa Fluor 647-conjugated anti-rabbit IgG secondary antibody (Abcam, ab150079), Alexa Fluor 594-conjugated anti-rabbit IgG secondary antibody (Abcam, ab150084), Alexa Fluor 594-conjugated anti-mouse IgG secondary antibody (Abcam, ab150116), Alexa Fluor 488-conjugated anti-goat IgG secondary antibody (Abcam, ab150113), Alexa Fluor 488-conjugated anti-rabbit IgG secondary antibody (Abcam, ab150077) and Alexa Fluor 488-conjugated anti-mouse IgG secondary antibody (Abcam, ab150113).

ELISA: anti-MPO polyclonal (Biotin Conjugated) primary antibody (Bioss, bs-4943R-Biotin).

WB: polyclonal rabbit anti-GSDMD (Cell Signaling Technology, 1:1000, #96458S), anti-NLRP3 (Cell Signaling Technology, #15101S), anti-TMS1/ASC (Abcam, ab151700), anti-pro Caspase 1 + p10 + p12 (Abcam, ab179515), α-Actinin (Abcam, ab68194) and β-Actin (Abcam,ab6276).

Validation	Antibodies used for flow cytometry, Immunofluorescence, and Western blotting were previously validated for the respective application by the distributor. Anti-MPO polyclonal (Biotin Conjugated) primary antibody (Bioss, bs-4943R-Biotin) used for ELISA was validated for this application by the investigators in preceding study (Middleton, E.A., et al., Blood, 136, 1169-1179, 2020).

Animals and other organisms

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

Laboratory animals	The platelet-specific Gsdmd KO (Gsdmd^fl/fl PF4-Cre) mice (by crossing Gsdmd^fl/fl mice with PF4-Cre mice) and global S100a9 deficient mice (using CRISPR/Cas9) were designed and generated (Shanghai Model Organisms Center, China). The global Tlr4 deficient (mice, mT/mG: PF4-Cre mice and wild type (WT) mice were obtained from the Jackson Laboratory. The C57BL/6J mice were obtained from the Jackson Laboratory and backcrossed to C57BL/6 mice (Bar Harbor, USA). All mice (five-week-old, male) were housed three per cage in controlled environment with a constant temperature of 18 °C–22 °C and a humidity of 55%–60% on a 12:12-h light/dark cycle. After acclimation for seven days, mice were supplied with food and water freely every day.
Wild animals	No wild animals were used in the study.
Field-collected samples	No field collected samples were used in the study.
Ethics oversight	Procedures were approved by the Institutional Animal Care and Use Committee of Guangzhou Medical University (No. SYXK2018-266).

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	All participants were enrolled from Guangzhou Women and Children's Medical Center, Guangzhou, China (Table 1). According to international guidelines (Pediatr Crit Care Med 6, 2-8 (2005)), pediatric sepsis is defined by a suspected or proven infection caused by any pathogen or a systemic inflammatory response syndrome associated with a high probability of infection. The age specific vital signs and laboratory variables of pediatric sepsis were divided into six distinct categories in Supplementary Table 3 (Curr Opin Pediatr 28, 380-387 (2016)). According to clinical spectrum of severity, it encompasses sepsis (systemic inflammatory response syndrome in the presence of infection), severe sepsis (sepsis in the presence of cardiovascular dysfunction, acute respiratory distress syndrome, or dysfunction of ≥ 2 organ systems) and septic shock (sepsis with cardiovascular dysfunction persisting after at least 40 ml/kg of fluid resuscitation in one hour). The ages of patients with sepsis were 0 to 18 years old, which are divided into 0~1 year, 1~5 years, 5-12 years and 12-18 years. All the patients with sepsis were treated with antibiotics including cephalosporin, vancomycin and broad-spectrum penicillin after admission.
Recruitment	93 pediatric sepsis patients (0-18 ages) and 75 age-matched HS, who were from Guangzhou Women and Children's Medical Center, were recruited in this study. Our medical center is a major tertiary transfer medical center in China, and thus the patients transferred to our center are more severe than that in other hospitals. The mortality of sepsis, severe sepsis, and septic shock was 0%, 14%, and 50% respectively (Table 1). However, the mortality of sepsis (including sepsis, severe sepsis, and septic shock) (22%) in our cohort is consistent with that of the previous international study (25%).
Ethics oversight	The study was approved by the Institutional Review Board of Guangzhou Women and Children's Medical Center and informed consent was obtained from each subject (Human Investigation Committee No. 2019-44102-1). Implementations followed the International Ethical Guidelines for Research Involving Human Subjects as stated in the Helsinki Declaration.The legal guardians of all participants signed the consent forms.

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	Venous blood samples were drawn from septic patients when they were enrolled first day. Venous blood samples (3 ml) were drawn from HS and severe sepsis patients with blood collection tubes containing 3.8% trisodium citrate (w/v). In mice, blood samples (approximately 0.8 ml) were directly obtained from the right cardiac ventricle into 3.8% trisodium citrate. Platelet-rich plasma (PRP) was centrifuged at 250 g at 25°C for 15 minutes. PRP were then treated with 100 nM prostaglandin E1 (PGE1, Sigma, 745-65-3) and centrifuged at 1000 g for 5 minutes. After discarding the supernatant, the platelet pellet was washed and resuspended with 3 ml Hank's Balanced Salt Solution (HBSS, Gibco, 14025092). Platelet suspensions (10^8

	platelets/ml) were incubated with antibodies at 37°C for 1 hour and analyzed by FACS. Each sample was analyzed using FACS. Data was collected from 20,000 platelets.
Instrument	BD, FACScanto, V657338000204.
Software	FlowJo-V10 software.
Cell population abundance	No cell sorting was used.
Gating strategy	Gating strategy was specificed in the relevant figures. This is included in Extended Data Fig.1a of the revised Supplemental material.

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