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☐ Initial submission ☐ Revised version ☒ Final submission

Life Sciences Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form is intended for publication with all accepted life science papers and provides structure for consistency and transparency in reporting. Every life science submission will use this form; some list items might not apply to an individual manuscript, but all fields must be completed for clarity.

For further information on the points included in this form, see Reporting Life Sciences Research. For further information on Nature Research policies, including our data availability policy, see Authors & Referees and the Editorial Policy Checklist.

Experimental design

1. Sample size

Describe how sample size was determined.

The sample sizes of each set of animals were determined according to previous studies performed by our group and other scholars and were fixed in a prospective manner.

2. Data exclusions

Describe any data exclusions.

One mouse in Alox12-HTG/Rep 6h group died before the sample collection and thus were excluded for statistical analysis

3. Replication

Describe whether the experimental findings were reliably reproduced.

All in vitro experiments were performed in triplicate unless specified in the figure legends. The detailed replication of each experiments has been provided in Figure Legend. All attempts at replication were successful for all experiments.

4. Randomization

Describe how samples/organisms/participants were allocated into experimental groups.

Animals with the same genotype and similar baseline values were randomly assigned to the sham and surgery groups or the vehicle and ML355 groups.

5. Blinding

Describe whether the investigators were blinded to group allocation during data collection and/or analysis.

The experimenters were blinded to the animal genotype and grouping information.

Note: all studies involving animals and/or human research participants must disclose whether blinding and randomization were used.

6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)
☐	☒ A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ A statement indicating how many times each experiment was replicated
☐	☒ The statistical test(s) used and whether they are one- or two-sided (note: only common tests should be described solely by name; more complex techniques should be described in the Methods section)
☐	☒ A description of any assumptions or corrections, such as an adjustment for multiple comparisons
☐	☒ The test results (e.g. *P* values) given as exact values whenever possible and with confidence intervals noted
☐	☒ A clear description of statistics including central tendency (e.g. median, mean) and variation (e.g. standard deviation, interquartile range)
☐	☒ Clearly defined error bars

See the web collection on statistics for biologists for further resources and guidance.

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Software

Policy information about availability of computer code

7. Software

Describe the software used to analyze the data in this study.

Agilent MassHunter software (version B.08.00) was used for the quantitative analysis of UPLC-MS.
Philips IntelliSpace Portal workstation (version 06.0.3.12200) was used to analyze the perfusion CT data.
All clean reads were mapped to reference sequences using SOAP2 (version 2.21) in DGE analysis.
Proteome Discoverer software (version 1.4) was used to search against the Uniport mouse protein sequences in proteomics analysis.
MeV (version 4.9) was used for K-means clustering analysis.
GSEA was implemented in the java GSEA (version 2.2.4).
A circular layout showing the correlation coefficients and P values of the pathways was generated using Circos (version 0.69).
Clean reads were aligned by HISAT2 (version 2.1.0) in RNA-seq analysis.
SAMtools (version 1.4.1) was used to produce BAM files in RNA-seq analysis.
Cufflinks (version 2.2.1) was applied to estimate the gene expression in RNA-seq analysis.
The gene co-expression network was constructed using WGCNA (version 1.51).
All statistical analyses used in this study were performed using SPSS (Version 21.0).

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). *Nature Methods* guidance for providing algorithms and software for publication provides further information on this topic.

Materials and reagents

Policy information about availability of materials

8. Materials availability

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a for-profit company.

Agilent MassHunter software was used for the quantitative analysis.The data sets supporting the conclusions of this article are included within the article and its additional files. The DGE and transcriptomics data have been submitted to NCBI Sequence Read Archive with the database identifier of SPR117594, SPR117665 and SPR117667. The mass spectrometry proteomics data have been deposited to the ProteomeXchange Consortium via the PRIDE partner repository with the dataset identifier PXD007827. Plasmids are available upon reasonable request. A Life Sciences Reporting Summary for this paper is available.

9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).

The detailed information of all antibodies used in our present study has been provided in the "Western blot analysis and antibodies" section in ONLINE METHODS. Antibodies against the following proteins were purchased from Cell Signaling Technology (Beverly, Massachusetts, USA): p-Ikba (#9246), Ikba (#4814), p-p65 (#3033), p65 (#4764), p-p38 (#4511), p38 (#9212), p-ERK1–ERK2 (#4370), ERK1–ERK2 (#4695), p-JNK1–JNK2 (#4668), JNK1–JNK2 (#9252), BCL2 (#3498), BAX (#2772), c-caspase3 (#9664), p-PKC (#2055), PKC (#2058), ALOX5 (#3289), CPLA2 (#2832) and GAPDH (#2118). Antibodies against ALOX12 were obtained from Santa Cruz (sc-365194; 1:200; Dallas, TX, USA) and Sigma (SAB2100109; St.Louis, MO, USA). ALOX15 (sc-133085) antibody was purchased from Santa Cruz Biotechnology. Antibodies against GPR31 (ab75579), GPR40 (ab211049), GPR75 (ab75581) and CYP4A (ab3573) were obtained from Abcam. GPR120 (sc-390752) was obtained from Santa Cruz Biotechnology. Secondary antibodies used in this study included Peroxidase AffiniPure goat anti-rabbit-IgG (H+L) (#111-035-003) and goat anti-mouse-IgG (H+L) (#115-035-003), which were purchased from the Jackson Laboratory (Bar Harbar, ME, USA). The Flag antibody was obtained from medical and biological laboratories (MBL, Nagoya, Japan). Unless specified, all primary antibodies were used at the dilution of 1:1000, and secondary antibodies were used at a 1:5,000 dilution.

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10. Eukaryotic cell lines

a. State the source of each eukaryotic cell line used.

The HEK293T and L02 cells were purchased from the Type Culture Collection of the Chinese Academy of Sciences (Shanghai, China).

b. Describe the method of cell line authentication used.

The HEK293T and L02 cell lines were verified by short tandem-repeat DNA profiling before the study.

c. Report whether the cell lines were tested for mycoplasma contamination.

No mycoplasma contamination was observed in the cells.

d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by ICLAC, provide a scientific rationale for their use.

No commonly misidentified cells were used in this study.

Animals and human research participants

Policy information about studies involving animals; when reporting animal research, follow the ARRIVE guidelines

11. Description of research animals

Provide details on animals and/or animal-derived materials used in the study.

The detailed information about animals and animal-derived materials used in this study has been provided in our ONLINE METHODS section.
Male mice aged 8-10 weeks (24-27 g) were subjected to a 70% warm hepatic IR injury. Mice were maintained in a standard SPF environment with ad libitum access to food and water. ML355 (HY-12341; MCE, Monmouth Junction, NJ, USA) or CDC (ab141560; Abcam, Cambridge, MA, USA) in the solution (DMSO : Solutol : PEG400 : water; 5/10/20/65 v/v/v/v) was injected into mice via the tail vein at 30 min before ischemia and at 6 h after reperfusion at a dose of 3 mg/kg. A blank solution was used as the vehicle control.
Primary hepatocytes were isolated from male mice aged 6-8 weeks using collagenase perfusion method.
All animal protocols used in this study were approved by the Animal Care and Use Committee of Renmin Hospital of Wuhan University and Institutional Animal Care and Use Committee of the Institute of Model Animal of Wuhan University. Animals received humane cares according to Guide for the Care and Use of Laboratory Animals published by the National Academy of Sciences and the National Institutes of Health. Abuse and maltreatment were avoided in our study.

Policy information about studies involving human research participants

12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.

The descriptions of characteristics of human samples have been provided in the "Human samples" section and in Supplementary Table 2.
The human serum and liver samples used in this study were obtained from patients who underwent liver resection surgery due to hepatocellular carcinoma or hepatic cyst. Liver samples were collected from normal liver tissues of individuals at 3 time points, including before ischemia (baseline group), after ischemia but before reperfusion (ischemia group), and after reperfusion (reperfusion group). The serum samples were obtained at the same time points as the liver samples. Informed consent forms were signed by all donors or their families. All these samples were used only to achieve experimental objective, and the studies were performed according to the principles outlined by the Declaration of Helsinki. All procedures involving human samples were approved by the Renmin Hospital of Wuhan University Review Board and Xijing hospital of The Fourth Military Medical University Review Board.

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