## nature research

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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 our <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

### **Statistics**

| For | all st | atistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.                                                                                                                           |
|-----|--------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| n/a | Cor    | nfirmed                                                                                                                                                                                                                                                       |
|     | ×      | The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement                                                                                                                                           |
| X   |        | 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<br>Only common tests should be described solely by name; describe more complex techniques in the Methods section.                                                                         |
| X   |        | 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)<br>AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals) |
|     | ×      | For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i> ) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted Give <i>P</i> values as exact values whenever suitable.                           |
| X   |        | For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings                                                                                                                                                              |
| X   |        | For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes                                                                                                                                        |
| X   |        | 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.                                                                                                                                                                |
| So  | ftw    | vare and code                                                                                                                                                                                                                                                 |

# Policy information about <u>availability of computer code</u> Data collection Data analysis Graphpad Prism 6, SPSS software (iBM SPSS Statistics version 2.2.0)

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 and 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 <u>data availability statement</u>. 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

Source data are provided with this paper

## Life sciences study design

| An studies must disclose on these points even when the disclosure is negative. |                                                                                                                                                           |  |  |  |
|--------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------|--|--|--|
| Sample size                                                                    | Sample size for animal experiments was determined using www.gpower.hhu.de to detect an effect with power = 0.8                                            |  |  |  |
| Data exclusions                                                                | No data were excluded from the analyses of this work                                                                                                      |  |  |  |
| Replication                                                                    | All experiments in teh main figures have been successfully repeated at least three times.                                                                 |  |  |  |
| Randomization                                                                  | Animals involved in the same experiments were all syngenic and of the same age, so they have been randomly allocated in the different experimental groups |  |  |  |
| Blinding                                                                       | All the processes of this work, from group allocation to data collection and analysis, have been performed in blind.                                      |  |  |  |

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

## 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

#### Methods

| n/a | Involved in the study                |  |
|-----|--------------------------------------|--|
|     | X Antibodies                         |  |
|     | Eukaryotic cell lines                |  |
| ×   | Palaeontology and archaeology        |  |
|     | × Animals and other organisms        |  |
|     | <b>X</b> Human research participants |  |
| ×   | Clinical data                        |  |
| ×   | Dual use research of concern         |  |

| n/a | Involved in the study |
|-----|-----------------------|
| ×   | ChIP-seq              |
| ×   | Flow cytometry        |
|     |                       |

▼ | MRI-based neuroimaging

## Antibodies

| Antibodies used | - anti-BMP1.3 monoclonal antibody (clone No. 6B5A3) from Promab (Richmond, CA). in vitro neutralization assay: 5µg; in vivo                                                                                                 |
|-----------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Antibodies used | administration: 50 $\mu$ g/kg; in vitro administration: 5 $\mu$ g/ml                                                                                                                                                        |
|                 | - anti-BMP2 antibody (abcam ab6295) clone number: 65529.111. IHC: 1:200                                                                                                                                                     |
|                 | - anti-BMP5 antibody (Thermo Fisher Scientific, PA5-78878). IHC: 1:1000                                                                                                                                                     |
|                 | - anti-TGFB1 antibody (Abcam, ab92486). IHC: 1:200                                                                                                                                                                          |
|                 | - anti-TGFβ1 antibody (BioXCell, BE0057). In vivo administration: 2 mg/kg                                                                                                                                                   |
|                 | - anti-LOX2L (R&D Systems, MAB2639 clone number: 262418). IHC: 1:100                                                                                                                                                        |
|                 | - anti-Sarcomeric Alpha Actinin antibody (Abcam, ab9465): IHC 1:3000 IF 1:200                                                                                                                                               |
|                 | - anti-SMAD2/3 (Cell Signalling Rabbit mAb #8685): WB: 1:1000                                                                                                                                                               |
|                 | - anti-pSMAD2/3 antibodies (Cell Signaling Rabbit mAb #8828): WB: 1:1000                                                                                                                                                    |
|                 | - anti-pSMAD2 (kind donation from Peter ten Dijke). IHC 1:100                                                                                                                                                               |
|                 | - anti-cleaved Caspase 3 (Cell Signalling Rabbit pAb #9661). IF:1:100                                                                                                                                                       |
|                 | - anti Donkey anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488 (Invitrogen A-21202). IF 1:500                                                                                                 |
|                 | - Mouse and Rabbit Specific HRP/AEC IHC Detection Kit - Micro-polymer; Abcam: ab236467                                                                                                                                      |
|                 | - goat anti-rabbit HRP-conjugated antibody (DAKO, ref p0448): WB: 1:2000                                                                                                                                                    |
|                 |                                                                                                                                                                                                                             |
| Validation      | - anti-BMP1.3 monoclonal antibody (clone No. 6B5A3) from Promab (Richmond, CA). Monoclonal BMP1.3 antibody (internal code                                                                                                   |
|                 | 84303#1; clone number 6B5A3) was produced by Promab Biotechnologies (Richmond, CA)) from specific peptide (sequence is                                                                                                      |
|                 | included in the manuscript) upon order from the Laboratory of Mineralized Tissues, University of Zagreb School of Medicine, Zagreb,                                                                                         |
|                 | Croatia, and is not commercially available. Monoclonal BMP1.3 antibody was produced for laboratory use only.                                                                                                                |
|                 | - anti-BMP2 antibody (abcam ab6295) clone number: 65529.111. no validation statement available. datasheet at: https:// www.abcam.com/bmp2-antibody-65529111-ab6285.html                                                     |
|                 | - anti-BMP5 antibody (Thermo Fisher Scientific, PA5-78878). "no validation statement available". Testing data available at: https://                                                                                        |
|                 | www.thermofisher.com/antibody/product/BMP5-Antibody-Polyclonal/PA5-78878                                                                                                                                                    |
|                 | - anti-TGFβ1 antibody (Abcam, ab92486). "Full length, inactive 44 kD TGFB1 is cleaved into mature TGFB1 (13 kD). TGFB1 also                                                                                                 |
|                 | homodimerizes and heterodimerizes with TGFB2, so there is potential for multiple different band sizes in WB.ab92486 detects the                                                                                             |
|                 | mature secreted form of the protein at 13 kDa. Our internal testing data suggests that this does not detect the full length protein at                                                                                      |
|                 | 44kDa. As cell lysates are unsuitable for detection of the mature form, tissue lysates should be used when using this antibody."                                                                                            |
|                 | Testing data at <:https://www.abcam.com/tgf-beta-1-antibody-ab92486.html                                                                                                                                                    |
|                 | anti-TGFβ1 antibody (BioXCell, BE0057). "Western Blot data confirm that this clone binds to its target antigen. For lot specific bindin                                                                                     |
|                 | validation data, email technical service". Testing data at: https://bxcell.com/product/m-h-tgf-beta/<br>- anti-LOX2L (R&D Systems, MAB2639 clone number: 262418). no validation statement available. Datasheet at: https:// |
|                 | - anti-LOAZE (R&D systems, WAD2059 clone number: 202416). No Validation statement available. Datasneet at: https://                                                                                                         |

www.rndsystems.com/products/human-lysyl-oxidase-homolog-2-loxl2-antibody-262418\_mab2639

- anti-Sarcomeric Alpha Actinin antibody (Abcam, ab9465): tested in WB: Mouse and rat skeletal muscle and rat heart tissue lysates. IHC-P: Human skeletal muscle tissue. Testing data at: https://www.abcam.com/sarcomeric-alpha-actinin-antibody-ea-53ab9465.html

-anti-SMAD2/3 (Cell Signalling Rabbit mAb #8685):"This Ab has been validated using SimpleChiP Enzymatic Chromatin IP Kits". Testing data at: https://www.cellsignal.com/products/primary-antibodies/smad2-3-d7g7-xp-rabbit-mab/8685

-anti pSMAD (Cell Signaling Rabbit mAb # 8828): no validation statement available. Datasheet at: https://www.cellsignal.com/ products/primary-antibodies/phospho-smad2-ser465-467-smad3-ser423-425-d27f4-rabbit-mab/8828

- anti-cleaved Caspase 3 (Cell Signalling Rabbit pAb : no validation statement available. Datasheet at: https://www.cellsignal.com/ products/primary-antibodies/cleaved-caspase-3-asp175-antibody/9661

- anti Donkey anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488 (Invitrogen A-21202). no validation statement available. Testing data at: https://www.thermofisher.com/antibody/product/Donkey-anti-Mouse-IgG-H-L-Highly-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21202

- Mouse and Rabbit Specific HRP/AEC IHC Detection Kit - Micro-polymer; Abcam: ab236467no validation statement available. Testing data at: https://www.abcam.com/mouse-and-rabbit-specific-hrpaec-ihc-detection-kit-micro-polymer-ab236467.html - goat anti-rabbit HRP-conjugated antibody (DAKO, ref p0448): no validation statement available. datasheet at https://

www.citeab.com/antibodies/3288347-p0448-goat-anti-rabbit-immunoglobulins-hrp-affinity

## Eukaryotic cell lines

| Policy information about <u>cell lines</u>                  |                                                      |  |  |  |
|-------------------------------------------------------------|------------------------------------------------------|--|--|--|
| Cell line source(s)                                         | HEK293T, Chinese Ovary Hamster CHO,                  |  |  |  |
| Authentication                                              | Cell lines used in this study were not authenticated |  |  |  |
| Mycoplasma contamination                                    | Not tested                                           |  |  |  |
| Commonly misidentified lines<br>(See <u>ICLAC</u> register) | None                                                 |  |  |  |

## Animals and other organisms

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

| Laboratory animals      | Sprague-Dawley rats (age 2 days, both genders; 2 months , male); C57BL6 mice (age 2 months, male); aSMA-RFP/COLL-EGFP mice (age 2 months, male).                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                      |
|-------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Wild animals            | This study did not involve the use of wild animals                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |
| Field-collected samples | Thi study did not involve the use of sample collected by field                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
| Ethics oversight        | All animal experiments were conducted in accordance with guidelines from the Directive 2010/63/EU of the European Parliament on animal experimentation in compliance with European guidelines and International Laws and Policies (EC Council Directive 86/609, OJL 34, 12 December 1987) and were approved by the ICGEB Animal Welfare Board, the Ethical Committee and the Italian Ministry of Health (authorization number 1303/2015-PR) and by Institutional Animal Care Committee of Medical Faculty, University of Zagreb and Directorate for Veterinary and Food Safety, Ministry of Agriculture (Republic of Croatia), authorization number 525-10/0255-15-6. |

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

## Human research participants

| Policy information about <u>studies involving human research participants</u> |                                                                                                                                                                                                                                                                                                                |  |  |  |
|-------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--|--|--|
| Population characteristics                                                    | Patients of both genders, who suffered an acute myocardial infarction (AMI), and aged/gender matched healthy donors without comorbidities were recruited in this study. Among AMI patients, 6 had hyperlipidemia and others did not have significant comorbidities. All participants were older than 40 years. |  |  |  |
| Recruitment                                                                   | Patients were consecutively recruited according to their admittance and controls were aged/gender matched. We do not foresee any major selection bias.                                                                                                                                                         |  |  |  |
| Ethics oversight                                                              | Study was approved by Ethical Committee of School of Medicine, University of Zagreb (EC KBC 5367001).                                                                                                                                                                                                          |  |  |  |

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