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

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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.
\boxtimes	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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
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
	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), indicating how they were calculated
	Our was collection an statistics for biologists contains articles on many of the points above

Software and code

Policy information about <u>availability of computer code</u>

Data collection

RNA-Seq data were sequenced with PE150 mode on Illumina NovaSeq6000 platform.
Full-length cDNA data were processed with the Iso-Seq3 pipeline (v3.4.0; https://github.com/PacificBiosciences/IsoSeq).
qPCR analysis were using QuantStudio 3.

Dual luciferase reporter analysis were using Infinite F50.

Data analysis

The raw subreads were processed with the Iso-Seq3 pipeline (v3.4.0). CCS reads generator (--min-rq 0.9 --min-passes 3 -j 6 --min-length 200). cDNA primers and poly(A) tails were removed with Lima (v2.1.0) and IsoSeq3 (v3.4.0). The FLNC reads from the same transcript were clustered with SMRTLink software (v10.1). The high-quality consensus isoforms were aligned with minimap2 (v2.20-r1061). RNA-seq data were used to extend the data of reference genome with StringTie software (v2.1.6, -eB -G). cDNA_Cupcake (v28.0.0) was used to filter out reads and to merge reads. SQANTI3 (v5.1.1) filter script and aggregate the reads. BUSCO (v3.0.2) was used to assess the integrity of the transcriptome. Protein-coding potential was predicted with SQANTI3 (v5.1.1). Clean reads obtained by RNA-Seq were mapped using HISAT2 (HISAT2 2.0.4, --dta -p 6 --max-intronlen 5000000). The uniquely mapped reads were assembled using StringTie software (v2.2.1, --merge -F 0.1 -T 0.1). DEGs were identified using DESeq2 1.30.1 (default: test="Wald", fitType="parametric"). The expression level of gene/transcript was provided by StringTie. Gene clustering was performed with Mfuzz (http://mfuzz.sysbiolab.eu). Scatter map was constructed with the OmicShare tools (https://www.omicshare.com/tools). Heatmap and venn diagram were plotted with online tools (http:// www.bioinformatics.com.cn/). PPI analysis was performed with string (https://cn.string-db.org/, v12.0) and were visualized with cytoscape (v3.9.1). Astalavista (v3.2) was used to analyze the basic AS types. The AS events were compared by using rMATS (v4.0.2). The rMATS (v4.0.2) statistical is used to measure the p-value of the differences by likelihood-ratio test. In AS analysis, the default threshold for rMATS screening is $|\Delta\psi| > c$ (c=0.0001). APA sites were processed with TAPIS pipeline (v1.1.3) (https://bitbucket.org/comp_bio/tapis/overview). WGCNA was performed with ImageGP (http://www.ehbio.com/ImageGP/). The differences of qPCR and Dual luciferase reporter analysis were compared with unpaired t-test by using GraphPad Prism 9.0.

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 Portfolio 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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

The datasets presented in this study have been deposited to the National Genomics Data Center (NGDC, https://ngdc.cncb.ac.cn/) with the dataset accession number CRA013591 (https://bigd.big.ac.cn/gsa/browse/CRA013591)

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation),

Research involving human participants, their data, or biological material

and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender

Reporting on race, ethnicity, or other socially relevant groupings.

No reporting on race, ethnicity, or other socially relevant groupings.

Population characteristics

Population characteristics

Population characteristics are not relevant for this study.

Recruitment

Recruitment is not relevant for this study.

Ethics oversight

Ethics oversight is not relevant for this study.

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

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.

	15 Min pigs, a Chinese local pig breed, were both male used here. The longissimus dorsi muscles were collected from pigs at 7-, 30-, 60-, 90 and 210-d-old, each with three individuals.	
Data exclusions	No exclusion	
Replication	All attempts at replication were successful	

D	Alla sationa			
Randomization	Allocation was i	location was random		
Blinding	Blinding is not r	relevant for this study		
Reporting	g for sr	pecific materials, systems and methods		
We require informatio	on from authors	about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.		
Materials & exp	perimental s	vstems Methods		
n/a Involved in the		n/a Involved in the study		
Antibodies		ChiP-seq		
Eukaryotic o	cell lines	Flow cytometry		
	ogy and archaeol			
Animals and Clinical data	d other organism	IS Control of the con		
	a search of concer	n		
Plants				
,				
Eukaryotic ce	ell lines			
Policy information a	about <u>cell lines</u>	and Sex and Gender in Research		
Cell line source(s)		PK-15 cells (RRID: CVCL_2160) : from Cellosaurus.		
Authentication		NO		
Mycoplasma contamination		The cell lines were tested negative for mycoplasma contamination.		
Commonly misidentified lines		NO		
(See <u>ICLAC</u> register)				
Animals and	other res	earch organisms		
		nvolving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in		
Laboratory anima		Min pigs, a Chinese local pig breed. The pigs were obtained from the Institute of Animal Husbandry, Heilongjiang Academy of Agricultural Sciences, Harbin, China. The ages were 7-, 30-, 60-, 90- and 210-d-old.		
Wild animals	No wile	No wild animals		
Reporting on sex	Male	Male		
Field-collected sar	mples The stu	S The study did not involve samples collected from the field		
Ethics oversight	Labora	Laboratory Animal Welfare and Ethics Committee of Northeast Agricultural University		
Note that full information on the approval of the study protocol must also be provided in the manuscript.				
Plants				
Seed stocks	No pla	No plants		
Novol plant				
Novel plant genotypes No plants				
Authentication	No aut	No authentication		