

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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☐ | ☒ | 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
<i>Give P values as exact values whenever suitable.</i> |
| ☒ | ☐ | 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 Measurements of metabolites were conducted on Agilent QTOF6545 or 6520 using Agilent's LC/MS Data Acquisition software (Version B.08.00).

Data analysis Metabolomics data were analyzed using Agilent Qualitative Analysis (Version 10.0 or 10.1). Other data were analyzed on Graphpad Prism (Version 10.0.3).

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](#)

All supporting data can be found within the paper or in the supplementary files.

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender	Sex and gender were clearly described in the Methods section.
Reporting on race, ethnicity, or other socially relevant groupings	Race/ethnicity, by self-identification, was reported in the Methods section for the human study involved.
Population characteristics	10 young white male subjects were in the human meal study. More information is in Voldstedlund, C. T. et al. Exercise-induced increase in muscle insulin sensitivity in men is amplified when assessed using a meal test. Diabetologia 67, 1386–1398 (2024).
Recruitment	Individuals were given written informed consent to participate in the study. More information can be found in the original human study Voldstedlund, C. T. et al. Exercise-induced increase in muscle insulin sensitivity in men is amplified when assessed using a meal test. Diabetologia 67, 1386–1398 (2024).
Ethics oversight	The study was approved by the Research Ethics Committee of Copenhagen (H-18006850) and complied with the Declaration of Helsinki.

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](https://www.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	The minimal sample size was pre-determined by the nature of the experiments. For biochemical measurements, at least 3-4 different mice or pythons per group were used. For behavioral measurements, 7-9 different mice per group were included. For histology studies, the same experiment was repeated in at least 3 different mice or pythons. For electrophysiological studies, at least 6 different neurons from 3 different mice were included. Sample sizes were based on previously published work and literature in the field that have conducted similar experimental designs. Biochemical measurements and mouse feeding studies were based on Li et al., Nature, 2022 (https://doi.org/10.1038/s41586-022-04828-5). Neuronal recordings were based on Liu et al., J. Neuroscience, 2024 (DOI: 10.1523/JNEUROSCI.2192-23.2024). TRAP or cFos studies were based on DeNardo et al., Nat. Neurosci., 2019 (doi: 10.1038/s41593-018-0318-7).
Data exclusions	One mouse per group was excluded from RER and EE analyses due to malfunctioning of two chambers in the TSE system. However, food intake data for these mice were unaffected and therefore included.
Replication	Each experiment has at least 3 biological replicates (with the exception of Burmese python 1DPF plasma samples, N = 2 as in Figure 1e) The number of replicates is specified in each of the figure legends.
Randomization	Animals of the same genotype was randomly assigned into different experiment groups or endpoints. For in vitro experiments, treatments were randomly assigned to each well.
Blinding	The investigators were not blinded due to the quantitative nature of the measurements. All samples were processed in the exact same way. All experiments were conducted in the exact same way.

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
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

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

Antibodies

Antibodies used	1:1000 dilution mouse anti-FLAG antibody (Sigma, F1804); 1:1000 dilution rabbit anti-beta tubulin antibody (Origene, TA301569). 1:1000 dilution goat anti-rabbit IRDye 800RD (LI-COR, 926-32211); 1:1000 dilution goat anti-mouse IRDye 680RD (LI-COR, 925-68070). 1:500 dilution rabbit anti-cFos antibody (Synaptic System, 226008).
Validation	All antibodies used in this study are commercially available and validated by the manufacturers.

Eukaryotic cell lines

Policy information about [cell lines and Sex and Gender in Research](#)

Cell line source(s)	HEK293T (CRL-3216) from American Type Culture Collection (ATCC).
Authentication	The HEK293T cell line was authenticated by ATCC.
Mycoplasma contamination	Cells were tested negative for mycoplasma.
Commonly misidentified lines (See ICLAC register)	Not applicable.

Animals and other research organisms

Policy information about [studies involving animals](#); [ARRIVE guidelines](#) recommended for reporting animal research, and [Sex and Gender in Research](#)

Laboratory animals	Mouse strains used in this study include: 1) WT male C57BL/6J mice from The Jackson Laboratory (000664) at 8-16 weeks old. 2) WT male diet-induced obese C57BL6/J mice from The Jackson Laboratory (380050) at 14-16 weeks old; 3) TRAP2/Ai14 male mice were crossed from TRAP2 (JAX, 030323) and Ai14 (JAX, 007914) and were used at 8-14 weeks old. 4) TRAP2 male mice from The Jackson Laboratory (007914) at 8-14 weeks old. Pythons were purchased from Bob Clark Reptiles. Burmese pythons were approximately 2 years old and weighed 1.5–2.5kg at the start of the study. Ball pythons were approximately 1 year old at the start of the experiments and weighed 400–500g.
Wild animals	Wild animals were not used in this study.
Reporting on sex	Only male mice were used in this study, sex was not considered in study design. Both male and female pythons were used and were randomized into different groups.
Field-collected samples	Field-collected samples were not used in this study.
Ethics oversight	Animal protocols and procedures involving pythons were approved by the Institutional Care and Use Committee (IACUC) of the University of Colorado Boulder. All mouse experiments were performed according to procedures approved by the IACUC of Stanford University and Baylor College of Medicine.

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

Plants

Seed stocks	Not applicable.
Novel plant genotypes	Not applicable.
Authentication	Not applicable.