

Epicardial Adipose Tissue in Hyperglucagonemia mice: an experimental model for GLP-1 triple agonists

Gianluca Iacobellis

giacobellis@med.miami.edu

University of Miami, Miller School of Medicine <https://orcid.org/0000-0001-7888-9530>

Lorena Mazuecos

Amanda Ferreira-Neves

University of Miami

Ernesto Bernal-Mizrachi

University of Miami

Brief Communication

Keywords: Epicardial adipose tissue, hyperglucagonemia, glucagon-like peptide-1 receptors (GLP-1Rs), glucose-dependent insulinotropic polypeptide (GIPR) and glucagon receptors (GCGRs)

Posted Date: August 26th, 2026

DOI: <https://doi.org/10.21203/rs.3.rs-10616455/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.
[Read Full License](#)

Additional Declarations: There is **NO** conflict of interest to disclose

Abstract

Human epicardial adipose tissue (EAT) plays a key role in coronary artery disease, atrial fibrillation and heart failure. However, experimental mouse models of EAT are virtually absent or at least scarce. Epicardial adipose tissue (EAT), a metabolically active visceral fat depot closely associated with coronary arteries and the myocardium, expresses the GLP-1 receptor and appears to respond directly to GLP-1 receptor activation. Given the established role of glucagon signaling in regulating lipid metabolism, energy expenditure, and adipose tissue biology, glucagon receptor activation may exert effects on EAT that are distinct from or complementary to those mediated by GLP-1 receptor signaling. However, the impact of glucagon receptor agonism on EAT remains poorly understood. In this study we analyzed EAT in the model of hyperglucagonemia in mice with activation of mTORC1 signaling by overexpression of Rheb (α RhebTg) and controls. We found that Wistar rats, α RhebTg and control mice have EAT and it is distributed in the same anatomical locations of the human heart. EAT was predominantly localized in the AV groove, atria, coronary arteries, but also on the ventricle in α RhebTg mice. EAT genetic and anatomical regionality plays a key role in the development of major cardiovascular diseases. While the effects of hyperglucagonemia on EAT are unknown, the activation of EAT glucagon-like peptide-1 receptors (GLP-1Rs), glucose-dependent insulinotropic polypeptide (GIPR) and glucagon receptors (GCGRs) is associated with genes encoding for adipose tissue and lipids metabolism. As EAT is a therapeutic target for GLP-1R analogs, α RhebTg mice could be used as an experimental model.

Introduction

Adipose tissue is no longer seen as fat storage and silent bystander (1). The role of epicardial adipose tissue (EAT) in cardiovascular diseases has recently emerged (2). EAT is involved in the pathogenesis and development of coronary artery disease (3), atrial fibrillation (4) and heart failure (5). EAT has unique anatomy, transcriptome and proteasome (6). EAT is situated between the myocardium and the visceral layer of the epicardium. Studies in humans showed that there is no muscle fascia to divide EAT and the underlying myocardium (7). This feature is unique to EAT as no other visceral fat depot has such unobstructed contact with the proximal organ. This anatomical peculiarity allows paracrine and vasocrine crosstalk between EAT and the myocardium and coronary arteries. Macroscopically, EAT is found within the atrio-ventricular groove, over the surface of the ventricles, around the left atrium and coronary arteries of adult human hearts (7). However, while many studies have shown the presence, function, and clinical measurability of EAT in humans, experimental mouse models of EAT are scarce (8). The fast metabolism of the mouse's heart and the small size of these tractable animals were thought to be associated with the scarcity of the EAT. This was considered a major limitation in fully understanding and modulating the physiology and pathology of EAT. Hence, in this study we sought to analyze the macroscopic presence and histological features of EAT in an experimental model of both rats and mice.

Methods

Experimental models

3-month-old male Wistar rats, from the National Paraplegic Hospital (Toledo, Spain) were maintained at 20–22°C temperature, 55% humidity and 12h light/dark cycle. Mice used in this study were housed in a pathogen-free environment and maintained on a 12 h light/dark cycle at University of Miami Facility. Animals were fed *ad libitum* with a standard chow diet and provided access to water. 12-week-old The Glucagon-Cre mice (generous gift from Dr. G. Gittes, University of Pittsburg) (9), expressing *Cre* recombinase driven by the glucagon promoter were crossed with Rheb (Ras homolog enriched in brain 1) transgenics (Rheb^{tg}) to conditionally activate Rheb expression in alpha cells (α Rheb^{Tg}). The Rheb transgene was generated as described previously (REF Diabetes paper). In this Tet-off model, expression of the tTA is turned upon *Cre*-mediated recombination, and removal of the neo gene cassette induces Rheb expression in the absence of doxycycline (Dox). Animal studies were performed in accordance with protocol approval by the University of Miami Institutional Animal Care and Use Committee (IACUC) and the Institutional Committee for Ethical Animal Care CEEA/UCLM/JCCM conducted with animal care regulations.

Hematoxiline-eosine (H&E) staining of heart sections

For macroscopical study and histological analysis, hearts were removed from the thorax after approved euthanasia by excision at the aortic arch and photographs were taken with a magnifying lens. Each cardiac tissue sample was bisected, and both halves were embedded separately with the cut surface-oriented face down to ensure optimal representation of the internal architecture. The samples were fixed in a 4% formaldehyde solution and were embedded in paraffin, cut into 4–5 μ m sections and stained routinely with hematoxylin and eosin for morphological examination. From each half, four tissue sections were obtained at different levels throughout the block to ensure representative sampling of basal, mid, and apical regions. Sections were spaced approximately 200 μ m apart. Serial sections were cut at a thickness of 4–5 μ m.

Attention was given during grossing and sectioning to include regions of the right ventricular free wall as well as the atrioventricular and interventricular grooves, where EAT is typically present. Sections were stained with hematoxylin, differentiated, blued, and counterstained with eosin. After dehydration and clearing, slides were covered using a permanent mounting medium. All stained sections were examined under light microscopy for histo-morphological evaluation.

Public RNA-seq Dataset Analysis

Bulk RNA-sequencing data from human adipose tissue depots were obtained from the publicly available dataset GSE125856 (10). Raw gene-level count matrices and corresponding sample annotations were downloaded from the Gene Expression Omnibus (GEO) database. Gene expression abundance was quantified as counts per million (CPM) using the edgeR package. CPM values were calculated by

normalizing raw counts to the total sequencing depth of each sample. Expression of glucagon-like peptide-1 (GLP-1R), glucose-dependent insulinotropic polypeptide (GIPR) and glucagon (GCGR) receptors was extracted from the normalized expression matrix and transformed as $\log_{10}(\text{CPM} + 1)$ for visualization.

Results

Macroscopical examination of EAT

Macroscopical examination of dissected hearts revealed the presence of EAT deposits in C57BL/6J mice, NOD mice and Wistar rats, confirming that its presence across different rodent strains and species under basal dietary conditions (Fig. 1A, B, C). In C57BL/6J mice as well as in NOD mice (Figs. 1A and 1B), EAT was observed not only mostly in atrial regions (AV groove) but also surrounding coronary arteries (A2, A3, A4, B2 depicted by black arrowheads). In Wistar rats, EAT is only distributed, at least at macroscopical level, along the atrial and periaortic regions (Fig. 1C).

H&E staining of EAT in a hyperglucagonemia mice model ($\alpha\text{Rheb}^{\text{Tg}}$)

The $\alpha\text{Rheb}^{\text{Tg}}$ mouse is an inducible model, reversible by doxycycline (TetOff), where Rheb1 (Ras homolog enriched in brain 1) is overexpressed selectively in pancreatic α cells, activating mTORC1 signaling and increasing pS6 in glucagon-positive cells as previously reported (11). mTORC1 activation in this model leads to α -cell hypertrophy and hyperplasia with chronic endogenous hyperglucagonemia, while β -cell size and insulin secretion remain largely unchanged.

To further evaluate EAT distribution in the model of hyperglucagonemia, we analyzed the histological architecture of different cardiac regions by hematoxylin and eosin (H&E) staining of control and $\alpha\text{Rheb}^{\text{Tg}}$ mice. To provide a clearer assessment of the regional distribution of adipose tissue depots, the images were segmented into atrial/periaortic (Fig. 2) and atrioventricular regions (Fig. 3). Both control and $\alpha\text{Rheb}^{\text{Tg}}$ mouse heart sections exhibited the presence of EAT surrounding the atrial and periaortic regions (Fig. 2, sections 2A-2E). EAT was predominantly localized in the atrial region and within the atrioventricular (AV) valve mesenchyme (Fig. 2, sections 2A, 2B, 2D, and 2E), as well as encircling the major arteries (sections 2B and 2E). In addition, scattered adipocytes were observed along the inferior wall of the ventricle (section 2C) in control mice, indicating a limited but detectable distribution of adipose cells beyond the primary epicardial depots.

Regarding the atrioventricular groove region, control mice displayed a clear presence of EAT along the AV groove (Fig. 3, sections 3A-3C). Notably, this feature was more pronounced in $\alpha\text{Rheb}^{\text{Tg}}$ mice, in which the number of adipocytes constituting the EAT was visibly increased (Fig. 3, sections 3D-3F).

Furthermore, the structural organization of EAT differed between hyperglucagonemic $\alpha\text{Rheb}^{\text{Tg}}$ mice and

controls. In α Rheb^{Tg} hearts, EAT within the AV groove appeared more compact and spatially restricted, with adipocytes clustered within a confined area, resulting in a higher local adipocyte density. In contrast, control mice exhibited a more diffuse and evenly distributed pattern of adipocytes along the myocardial wall.

The microscopic appearance of EAT in the α RhebTg mice has several anatomical similarities with the human EAT, as shown in Fig. 4. In particular, we found EAT in the atrial region of the α RhebTg mice heart within the subepicardial myocardium as it was observed in human hearts (Fig. 5).

Bulk RNA-sequencing data from human adipose tissue depots

The expression of GLP-1, GIP, GCG receptors in EAT in comparison to other fat depots, such mediastinal and subcutaneous fat are shown in Fig. 6 (panel A) and stratified by lean/obese groups as shown in the panel B.

In this study, we show that α RhebTg mice have EAT and it is distributed in the same anatomical locations of the human heart. Rodents are generally considered to not have much EAT, but rather paracardial fat, an anatomically and functionally different fat depot not in contiguity with the myocardium. EAT was previously described in the AV groove in mice and thought to derive by mesenchymal transformation of the epicardium (12). We found the macroscopic presence of EAT in both rats and mice. EAT was observed not only in the AV groove but also surrounding the coronary arteries and the atria. Unlike previous reports (7), we found the presence of EAT also on the ventricle wall.

In the α RhebTg mice, our histological analysis clearly showed the regional distribution of EAT that was predominantly localized in the AV groove, atrial region, the coronary arteries, and in smaller quantities in the left ventricle. It was previously noticed that the presence of EAT in the mouse AV groove starts around 2 weeks after birth and represents epicardium differentiation that requires peroxisome proliferator-activated receptor gamma (PPAR- γ) expression to adopt an adipocyte fate (8). Interestingly, reactivation of the epicardium is thought to lead to myocardial fatty infiltration in atrial fibrillation (13).

Yamaguchi et al concluded that fat depot in mouse AV groove is EAT based on subepicardial location, the perfusion by coronary vasculature and the brown adipose tissue characteristics (8). These are the same unique features of the human EAT that is supplied by branches of coronary arteries and has also brown fat-like and beige fat-like features (14).

We believe that our results are of great interest, as EAT distribution and microscopic appearance in the α RhebTg mice is like the one observed in human hearts (2) (Fig. 4). EAT is not equally distributed with the human heart, and its genetic and anatomical regionality plays a key role in the development of major cardiovascular diseases (15). Peri-coronary, left atrial and myocardial EAT are different and can now be detected with common imaging techniques (16–17). Peri-coronary EAT affects the proximal coronary arteries and peri-atrial causes atrial myopathy contributing to the development of atrial fibrillation (18–

19). It is very interesting that we found EAT in the atrial region of the α RhebTg mice heart within the subepicardial myocardium.

In this study, by using mice with inducible induction of Rheb1 in α cells (11), we produced hyperglucagonemia through increased glucagon secretion. Epicardial adipocytes architecture and accumulation was different between hyperglucagonemic mice and controls. Interestingly, epicardial adipocytes in α RhebTg mice were clustered within a confined area. This appearance highly resembles the histopathologic feature of the high density of crown-like structures (CLS), marker of adipose tissue inflammation and apoptotic changes, that was observed in EAT from subjects with coronary artery disease (20).

Depending on the exposure time, hyperglucagonemia can cause different cardio-metabolic effects (21), although this was outside the scope of our observational study. We demonstrated that the α RhebTg mouse is a suitable model to study reversible hyperglucagonemia in physiology and disease states (11). Glucagon action plays a critical role in glucose metabolism, insulin sensitivity and liver fat accumulation, all related to EAT in humans (22–23). One of the appealing features of human EAT is the expression and presence of glucagon-like peptide-1 (GLP-1R), glucose-dependent insulinotropic polypeptide (GIPR) and glucagon (GCGR) receptors, as discovered by our group (24–25). The immunohistochemical analysis indicated that the receptors were mainly located in the CLS of EAT, microscopic, and inflammatory formation where macrophages gather in a ring or "crown" around apoptotic fat cells (24). In this study, we expanded and confirmed the presence of GLP-1R, GIPR and GCGR in EAT by performing RNA-sequencing data from human adipose tissue depots obtained from publicly available dataset. While the effects of hyperglucagonemia on EAT are unknown, the activation of EAT GLP-1R, GIPR and GCGR is associated with genes encoding for adipose tissue and lipids metabolism (24). EAT GLP-1R, GIPR and GCGR are related to genes modulating free fatty acids metabolism, promoting adipogenesis, such as the PPAR- γ , and white-to-brown adipocyte differentiation such as uncoupling protein 1 (UCP-1) (24). Clinical studies showed that EAT is a therapeutic target for GLP-1R and dual GLP1R-GIPR analogs, such as liraglutide, semaglutide and tirzepatide, respectively (26–28). The recent development of dual and triple agonists that combine GLP-1 receptor activation with GIP and GCG receptor agonism has generated considerable interest because these agents produce greater weight loss and metabolic improvements than traditional GLP-1 receptor agonists. Interestingly, the expression of EAT receptors was present in both lean and obese subjects, potentially supporting the use and cardiovascular beneficial effects of the GLP-1R/ GIPR analogs in subjects who are not obese. We showed that EAT thickness, marker of visceral fat, could help to better stratify GLP-1 and GIP receptors expression and identify the best candidate to receive this treatment (25). As next generation of glucagon receptor, GLP1 and GIP dual and triple agonist therapies enter clinical practice, understanding the direct effects of glucagon receptor signaling on EAT will become particularly important, as EAT remodeling may represent a previously unrecognized mechanism contributing to their enhanced cardiometabolic benefits. However, the impact of glucagon receptor agonism on EAT remains poorly understood. Defining the relative contributions of GLP-1, GIP, and glucagon receptor signaling to EAT remodeling may provide important mechanistic insights into the

cardiovascular benefits observed with these emerging therapies. α RhebTg mice could be used as experimental model to test the effects of these drugs on EAT.

In conclusion, our study shows the presence of EAT in an experimental mouse model and its distribution and architecture resembling that are observed in the human heart. The hyperglucagonemia in our mice model could modulate EAT responsiveness to GLP-1R and GCGR analogs.

However, this was an observational study and therefore no correlation with blood or genetic markers was performed. The presence of EAT GCGR found in our previous study in humans (24) could not be confirmed in α RhebTg mice, as staining the mouse GCGR requires highly specific antibodies. Adipocytes could not be quantified or characterized in these animals. Further studies to evaluate the reproducibility and suitability of this experimental model seem to be warranted.

Declarations

Acknowledgements/Funding resources

Mazuecos L. was supported by “José Castillejo” international grant from Ministerio de Ciencia e Innovación (CAS23/00295, Spain).

We would like to acknowledge Camila Rossetti for technical support and Valentina Pita-Grisanti for their generosity in permitting the macroscopic analysis of their mice.

References

1. Oikonomou, E. K. & Antoniades, C. The role of adipose tissue in cardiovascular health and disease. *Nat. Rev. Cardiol.* 16, 83–99 (2019).
2. Iacobellis G. Epicardial adipose tissue in contemporary cardiology. *Nat Rev Cardiol.* 9, 593–606. (2022)
3. Mazurek T, et al. Human epicardial adipose tissue is a source of inflammatory mediators. *Circulation* 108, 2460–6 (2003)
4. McCauley MD, et al. Targeting the Substrate for Atrial Fibrillation: JACC Review Topic of the Week. *J Am Coll Cardiol.* 83, 2015–2027 (2024)
5. Packer M. Causal role of adipose tissue-derived adipokines in the pathogenesis of heart failure with preserved ejection fraction: experimental demonstration as the basis for clinical targeting. *Eur Heart J.* 47, 2286–2289 (2026)
6. Iacobellis G. Local and Systemic effects of the multifaceted Epicardial Adipose Tissue Depot. *Nat Rev Endocrinology*; 11, 363–371 (2015)
7. Iacobellis G, Corradi D, Sharma AM. Epicardial adipose tissue: anatomic, biomolecular and clinical relationships with the heart. *Nat Clin Pract Cardiovasc Med* 2, 536–543 (2005)

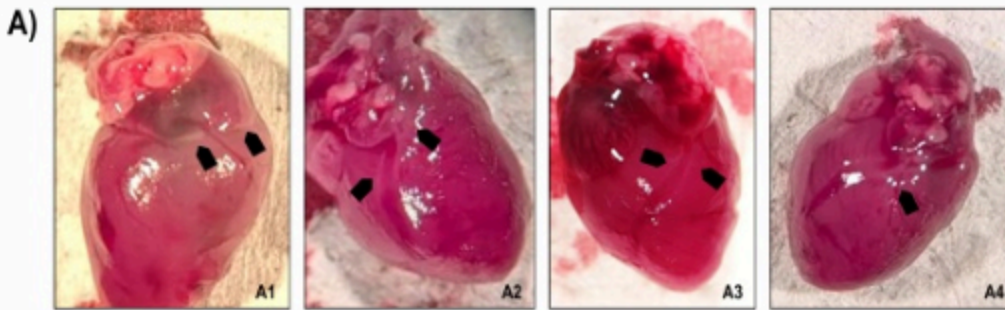
8. Yamaguchi Y, et al. Adipogenesis and epicardial adipose tissue: a novel fate of the epicardium induced by mesenchymal transformation and PPAR γ activation. *Proc Natl Acad Sci U S A*. 112, 2070–2075 (2015)
9. Shiota C, et al. α -Cells are dispensable in postnatal morphogenesis and maturation of mouse pancreatic islets. *Am J Physiol Endocrinol Metab*. 305, E1030-40 (2013)
10. Chechi K, et al. UCP1 expression-associated gene signatures of human epicardial adipose tissue. *JCI Insight*. 4:e123618 (2019)
11. Lubaczeuski C, et al. Time-dependent effects of endogenous hyperglucagonemia on glucose homeostasis and hepatic glucagon action. *JCI Insight*. 8, e16225 (2023)
12. Liu Q, et al. Epicardium-to-fat transition in injured heart. *Cell Res* 24, 1367–1369 (2014)
13. Suffee N, et al. Reactivation of the epicardium at the origin of myocardial fibro-fatty infiltration during the atrial cardiomyopathy. *Circ. Res*. 126:1330–1342 (2020)
14. Sacks HS, et al. Adult epicardial fat exhibits beige features. *J Clin Endocrinol Metab*. 98, E1448-55 (2013)
15. Gaborit B, et al. Human epicardial adipose tissue has a specific transcriptomic signature depending on its anatomical peri-atrial, peri-ventricular, or peri-coronary location. *Cardiovasc. Res*. 108, 62–73 (2015)
16. Iacobellis, G. et al. Epicardial fat from echocardiography: a new method for visceral adipose tissue prediction. *Obes. Res*. 11, 304–310 (2003).
17. West HW, et al. Deep-Learning for Epicardial Adipose Tissue Assessment With Computed Tomography: Implications for Cardiovascular Risk Prediction. *JACC Cardiovasc Imaging*. 16, 800–816 (2023)
18. de Vos, A. M. et al. Peri-coronary epicardial adipose tissue is related to cardiovascular risk factors and coronary artery calcification in post-menopausal women. *Eur. Heart J*. 29, 777–783 (2008).
19. Batal, O. et al. Left atrial epicardial adiposity and atrial fibrillation. *Circ. Arrhythm. Electrophysiol*. 3, 230–236 (2010)
20. Malavazos AE, et al. The density of crown-like structures in epicardial adipose tissue could play a role in cardiovascular diseases. *Eat Weight Disord*. 27, 2905–2910 (2022)
21. Riahi Y, et al. Hyperglucagonaemia in diabetes: altered amino acid metabolism triggers mTORC1 activation, which drives glucagon production. *Diabetologia*. 66, 1925–1942 (2023)
22. Iacobellis G, Leonetti F. Epicardial adipose tissue and insulin resistance in obese subjects. *J Clin Endocrinol Metab*. 90:6300–2 (2005)
23. Perdomo CM, et al. Transient elastography and serum markers of liver fibrosis associate with epicardial adipose tissue and coronary artery calcium in NAFLD. *Sci Rep*. 12, 6564 (2022)
24. Iacobellis G, et al. Human Epicardial Fat Expresses Glucagon-Like Peptide 1 and 2 Receptors Genes. *Horm Metab Res*. 49:625–630 (2017)

25. Malavazos AE, et al. Human epicardial adipose tissue expresses glucose-dependent insulinotropic polypeptide, glucagon, and glucagon-like peptide-1 receptors as potential targets of pleiotropic therapies. *Eur J Prev Cardiol.* 30, 680–693 (2023)
26. Goldberger JJ, et al. The Liraglutide Effects in Atrial Fibrillation (LEAF) Study. *JACC Clin Electrophysiol.* S2405-500X(26)00270–7 (2026)
27. Manubolu VS, et al. Effect of Semaglutide on Epicardial Adipose Tissue in Type 2 Diabetes: Insights From the STOP (Semaglutide Treatment effect On coronary atherosclerosis Progression) Randomized Trial. *J Am Coll Cardiol.* 84, 865–867 (2024)
28. Packer M, et al; SUMMIT Trial Study Group. Influence of Type 2 Diabetes on the Effects of Tirzepatide in Patients With Heart Failure and a Preserved Ejection Fraction With Obesity: A Prespecified Stratification-Based Analysis. *J Am Coll Cardiol.* 86:696–707 (2025)

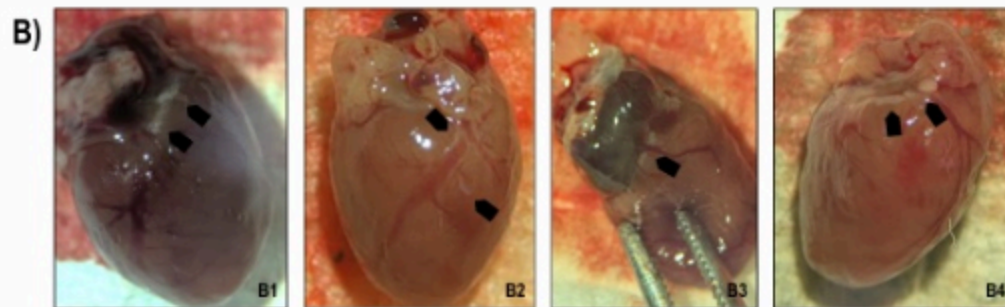
Figures

EAT observed in rodents

C57BL/6J mice



NOD mice



Wistar rat

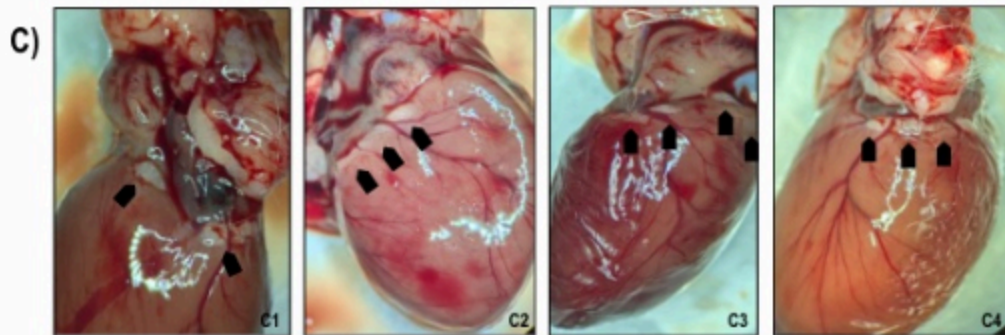


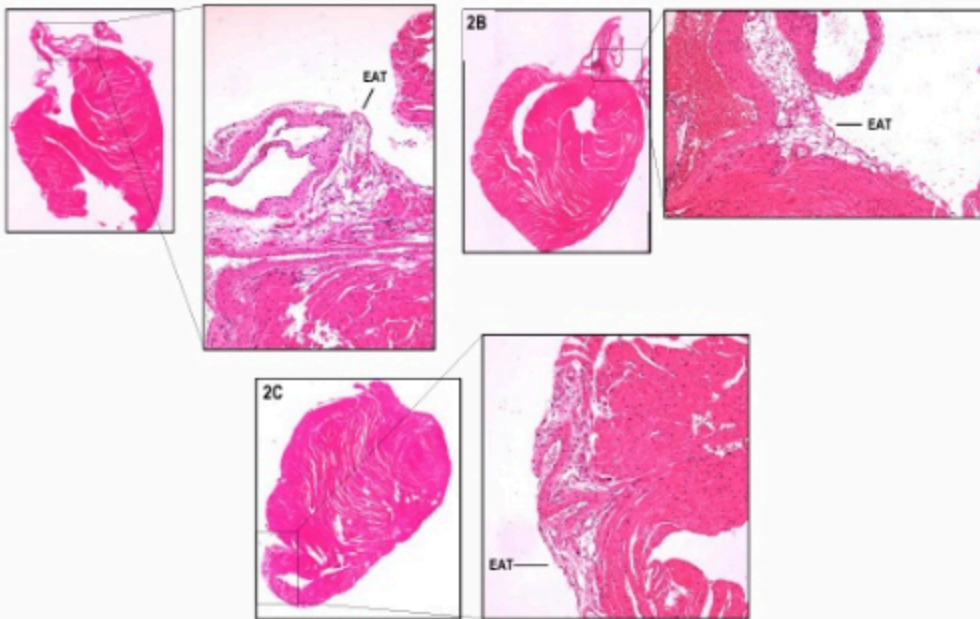
Figure 1

Location of EAT in rodents

Macroscopical images of epicardial adipose tissue (EAT) observed in dissected hearts of A) C57BL/6J mice (n=4), B) NOD mice (n=4) and C) Wistar rats (n=4) under chow diet. Biological replicates are indicated by an uppercase letter and a subscript number in the lower right corner.

H&E α Rheb^{Tg} mice: atrial and periaortic regions

Control mice



α Rheb^{Tg} mice

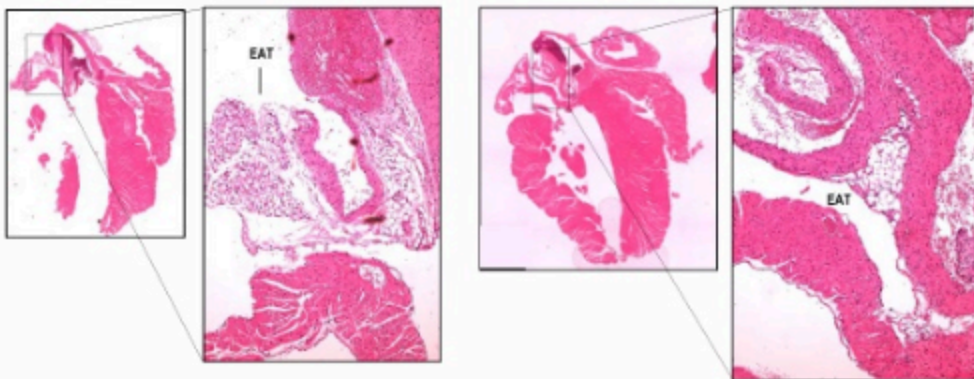


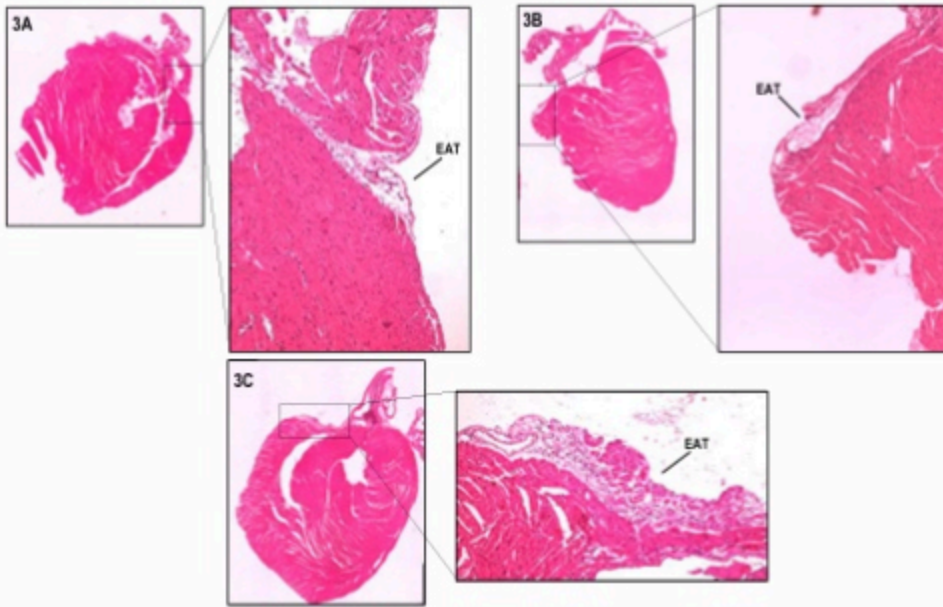
Figure 2

Histological appearance of EAT in atrial and periaortic regions by H&E staining.

Representative hematoxylin and eosin (H&E)–stained sections of atrial regions of the heart from control (n=2) (upper panel) and α Rheb^{Tg} (n=2) mice (lower panel). The boxed regions are shown at higher magnification. EAT: epicardial adipose tissue.

H&E α Rheb^{Tg} mice: atrioventricular groove

Control mice



α Rheb^{Tg} mice

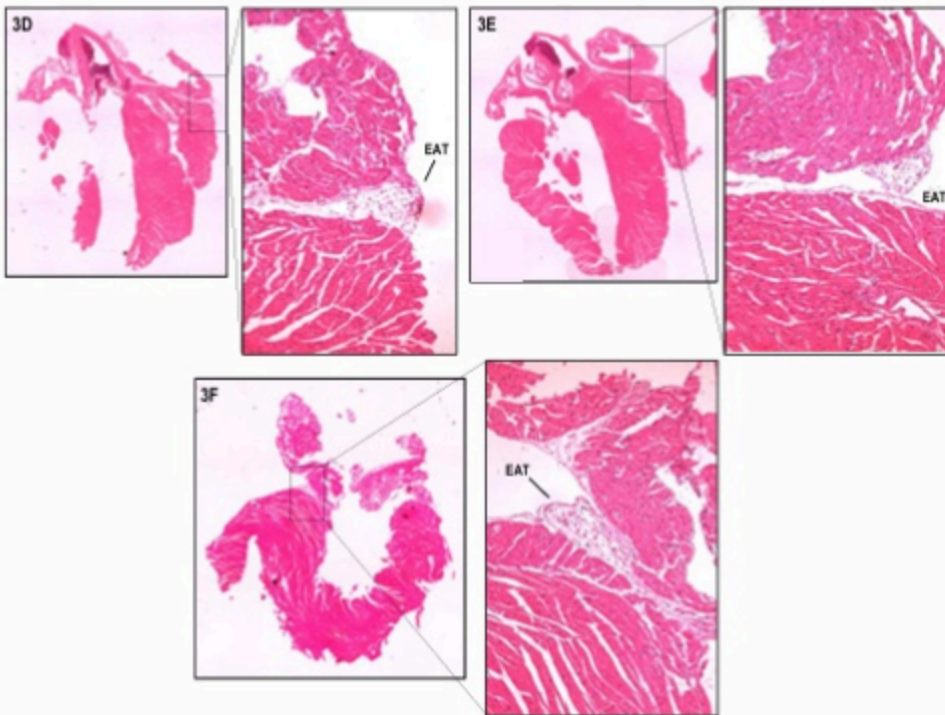


Figure 3

Histological appearance of EAT in atrio-ventricular groove by H&E staining.

Representative hematoxylin and eosin (H&E)–stained sections of the atrioventricular groove of the heart from control (n=2) (upper panels) and α Rheb^{Tg} (n=2) (lower panels) mice. The boxed regions are shown at higher magnification. EAT: epicardial adipose tissue.

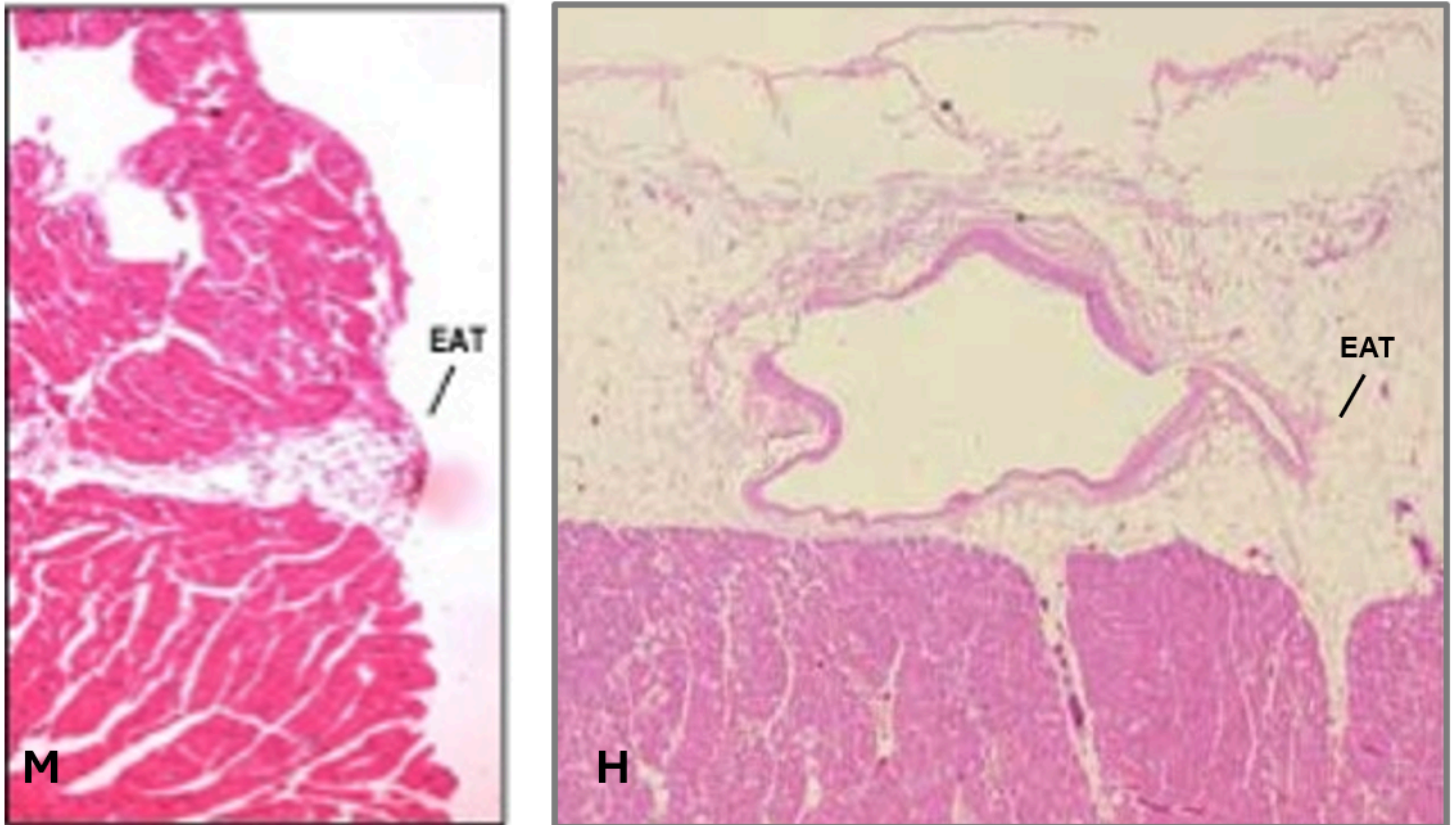


Figure 4

Microscopic appearance of EAT in mice and humans

Histological appearance of epicardial adipose tissue (EAT) in the atrio-ventricular groove in α RhebTg mice (M panel on the left) and in human (H right panel on the right), collected from a patient who underwent cardiac surgery. Both the mouse and human EAT show no fascial structure separating the fat depot from the underlying myocardium. It can be also appreciated that both mice and human EAT present with a dense number of adipocytes. Representative Hematoxylin & Eosin (H&E) stained sections.

Picture of human EAT from Iacobellis G. Nat Rev Endocrinol. 2015; 11:363-71

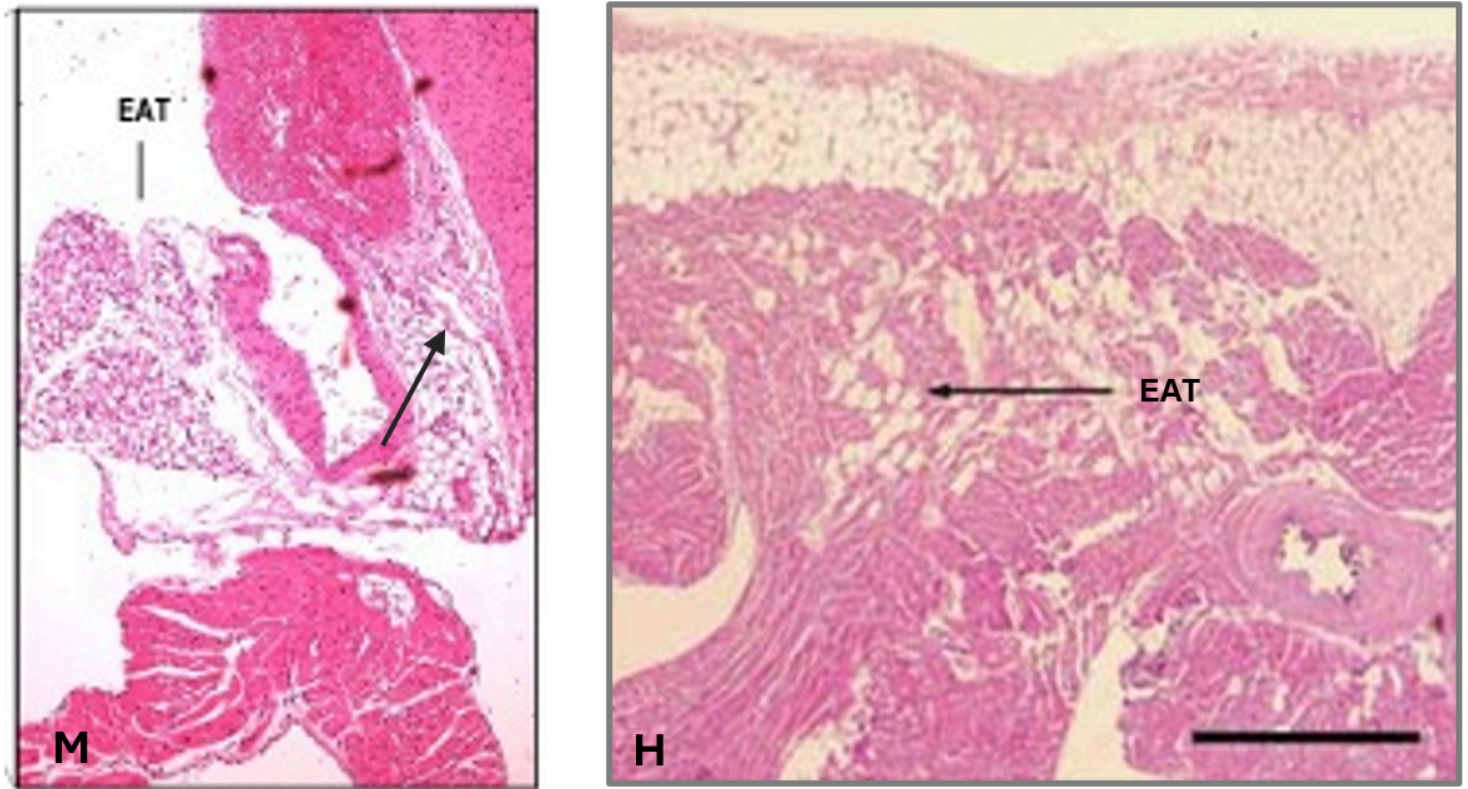
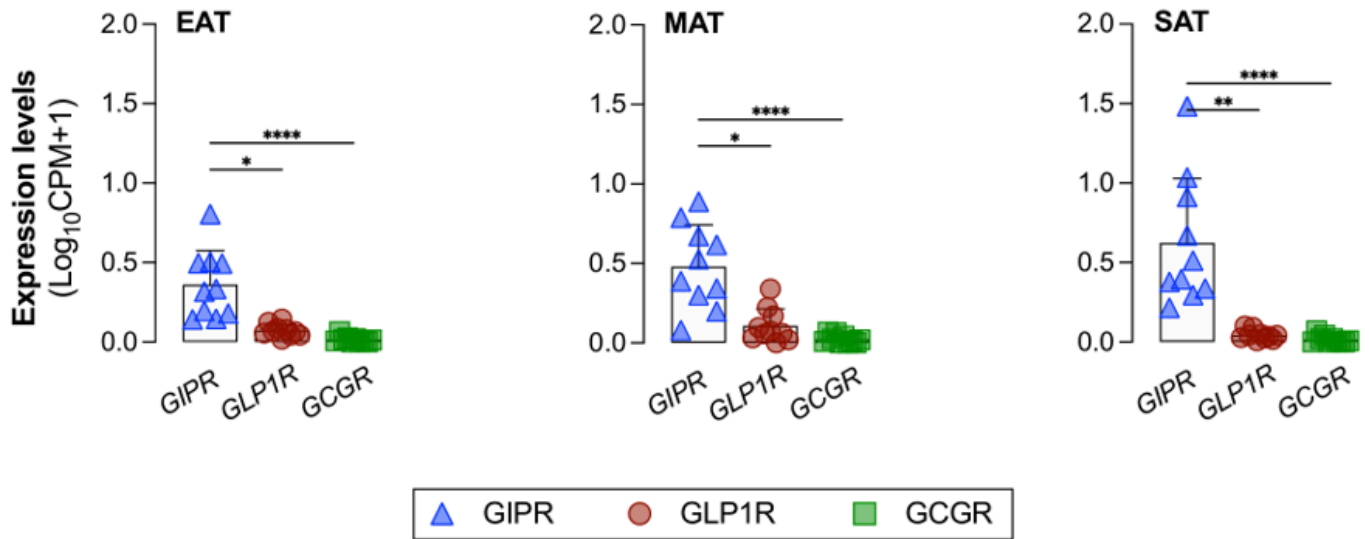


Figure 5

Microscopic appearance of EAT within the subepicardic myocardium in mice and humans

Histological appearance of EAT in atrial region in α RhebTg and in the the right ventricle in human. The arrow shows the isles of mature adipocytes. Adipocytes are seen (black arrows) within the subepicardic myocardium in both mice and human EAT. Scale bar = 1 mm 10. *Picture of human EAT from Iacobellis G. Nat Rev Endocrinol. 2015; 11:363-71*

A



B

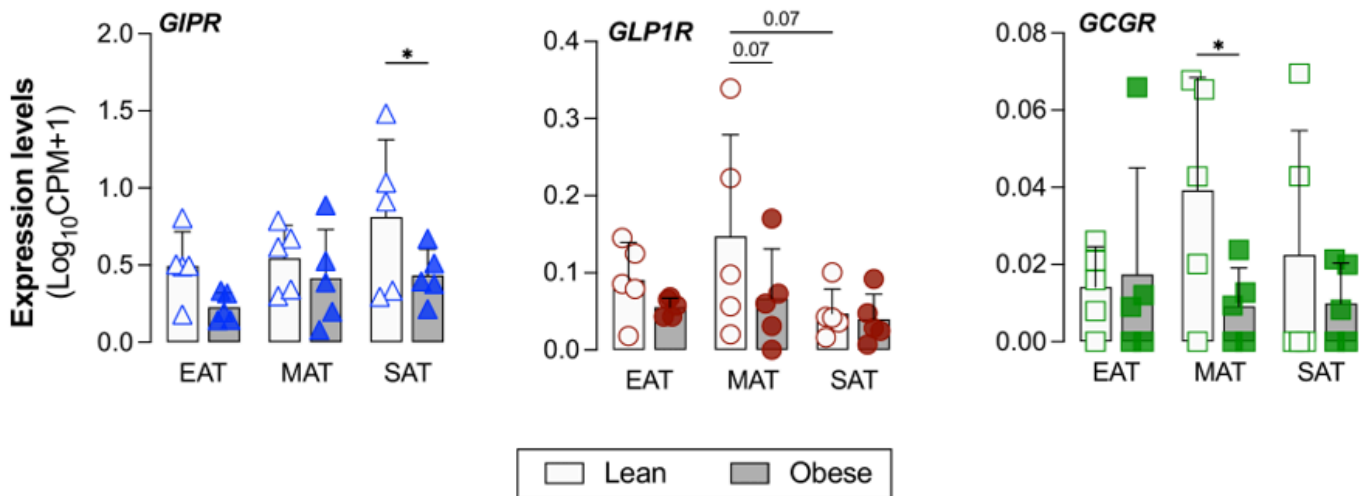


Figure 6

Expression of GIPR, GLP1R, and GCGR in human adipose tissue depots

Panel A

Transcript abundance was quantified from bulk RNA-sequencing data and expressed as log₁₀(CPM + 1). Expression levels of GIPR, GLP1R, and GCGR are shown in epicardial adipose tissue (EAT, n=10), mediastinal adipose tissue (MAT, n=10), and subcutaneous adipose tissue (SAT, n=10). Each symbol corresponds to an individual biological sample. Bars indicate mean ± SD. Kruskal-Wallis test followed by Dunn's multiple-comparison test. *P < 0.05; ****P < 0.001.

Panel B

Transcript abundance was quantified from bulk RNA-sequencing data and expressed as $\log_{10}(\text{CPM} + 1)$. Expression levels of GIPR, GLP1R, and GCGR were stratified by clinical group (lean n=5 or obese n=5) within each adipose depot, including epicardial adipose tissue (EAT), mediastinal adipose tissue (MAT), and subcutaneous adipose tissue (SAT). Each symbol corresponds to an individual biological sample. Bars indicate mean $\pm$ SD. Statistical comparisons were performed between lean and obese groups for each adipose depot within each receptor. Two-way ANOVA followed by Šídák's multiple-comparison test. *P < 0.05.