# The human gut microbiota contributes to Type-2 Diabetes non-resolution 5-years after Roux-en-Y gastric bypass

Jean Debédat, Tiphaine Le Roy, Lise Voland , Eugeni Belda, Rohia Alili, Solia Adriouch, Pierre Bel Lassen, Kazuyuki Kasahara, Evan Hutchison, Laurent Genser, Licia Torres, Camille Gamblin, Christine Rouault, Jean-Daniel Zucker, Nathalie Kapel, Christine Poitou, Geneviève Marcelin, Federico E. Rey, Judith Aron-Wisnewsky, and Karine Clément.

## Introduction

Despite persistent public health efforts, the prevalance of obesity, and associated co-morbidities like type 2 diabetes mellitus (T2DM), have remained on the rise globally[1]. Bariatric surgery refers to a collection of surgical procedures performed to manage obesity[2]. For most cases of severe obesity, it is currently the only intervention that can cause major and prolonged weight loss, while improving all obesity related co-morbidities[3]. However, only 50-60% of bariatric surgery patients undergo T2DM remission (DR) following bariatric surgery[4]. A few parameters have been proposed to predict the likelihood of DR like T2DM severity, and diabetes duration. However, these are not reliably accurate[5]. Some studies have shown that the gut microbiome might have some association with DR rates[6].

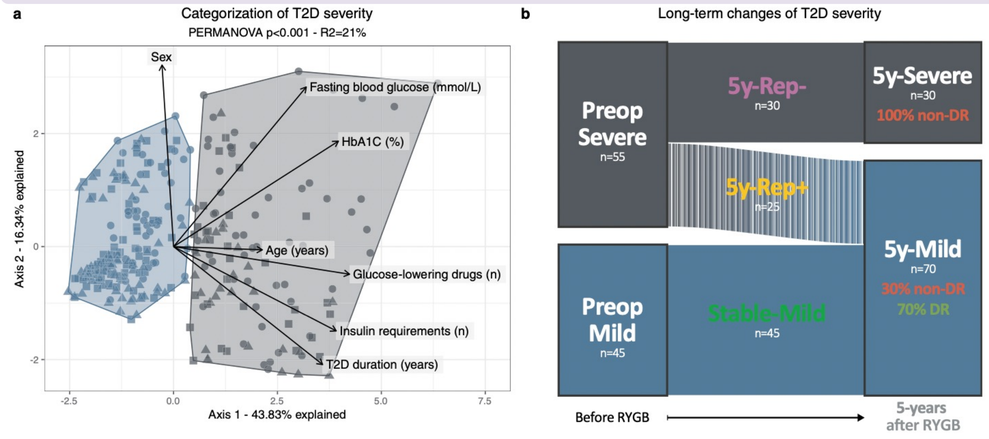
The aim of this study is to support the hypothesis that the gut microbiome can cause metabolic changes in patients that have undergone Roux-en-Y gastric bypass (RYGB), a type of bariatric surgery.

## Methods

Classification of T2D severity in patients pre- and post- RYGB → Extraction and GM sequencing of fecal DNA from patient stool samples → Antibiotic-induced gut microbiota depletion and fecal matter transplant (FMT) into recipient mice → Biochemical tests to quantify metabolic changes in mice

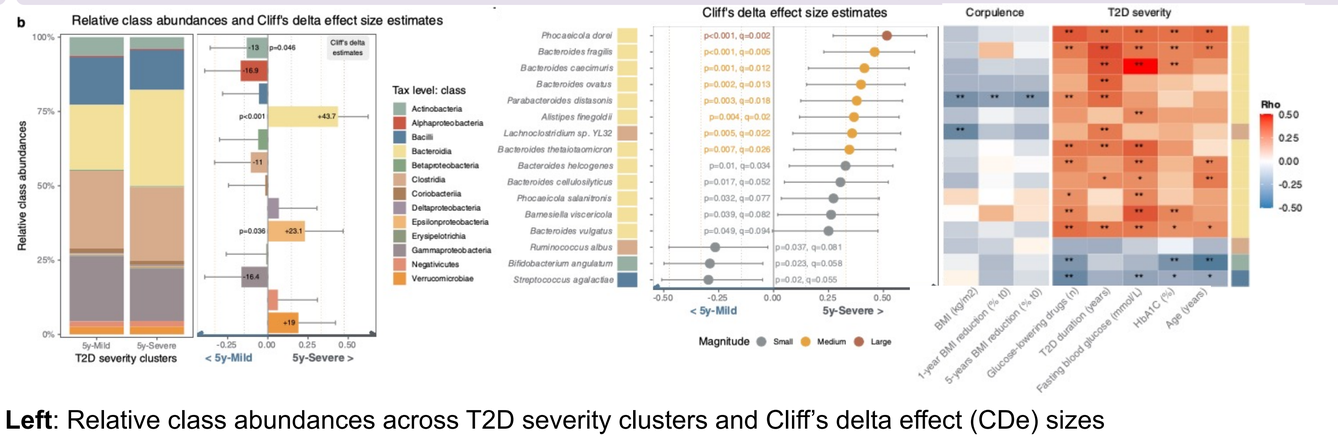
## Results

**Fig 1:** Categorization and long-term evolution of T2D severity after RYGB




**Left**: Patients' principal component analysis (PCA) projection according to their T2D severity (mild – blue, severe – grey) at all time points (● initial, ▲1y, ■ 5y). Arrows show association between clinical variables and T2D severity.
**Right**: Long-term trajectories of T2D severity and association with long-term DR. Rep+, good responders; Rep-, poor responders

**Fig 2:** T2D severity after RYGB is associated with specific GM signatures in patient stool samples

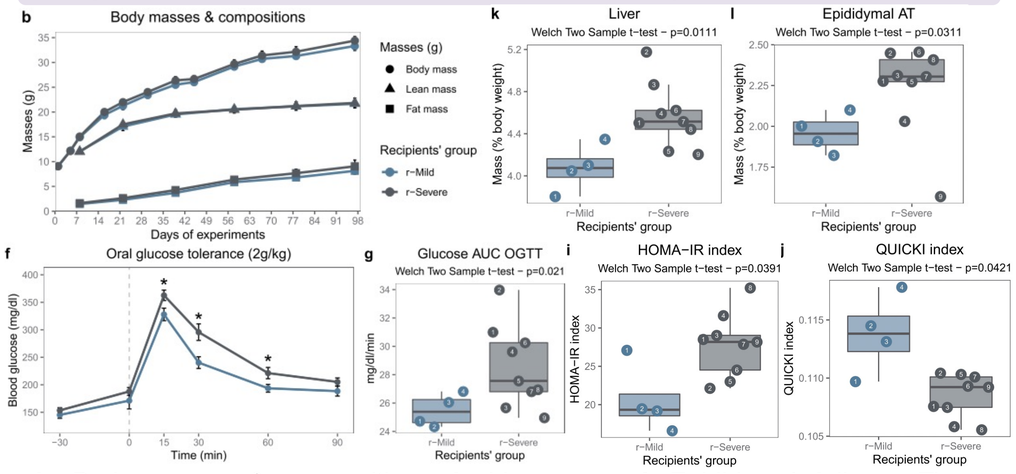



**Left**: Relative class abundances across T2D severity clusters and Cliff's delta effect (CDe) sizes

**Middle**: CDe size estimates, showing 13 bacterial genera were significantly more prevalent in 5y-Severe patients (most from Bacteroides class), 3 species were more abundant in 5y-Mild patients

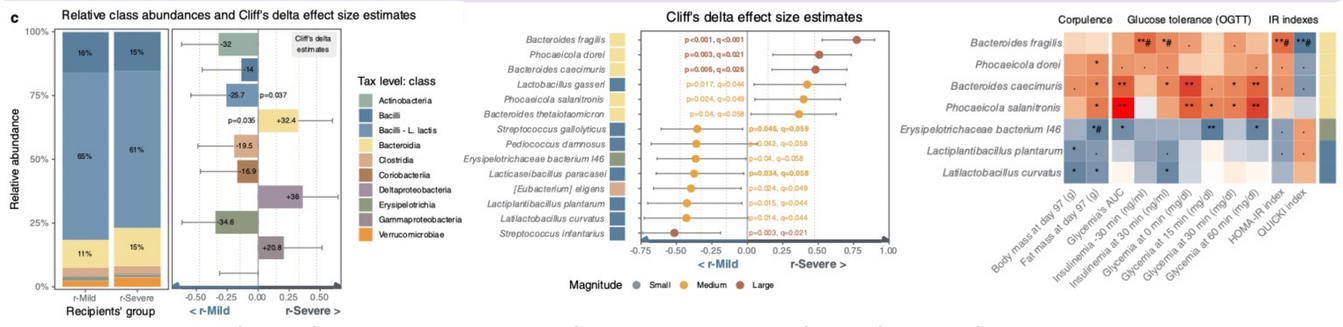
**Right**: Heatmap of Spearman's rank correlations between the 16 species and 5-year clinical variables (*: p<0.05; **:q<0.1), showing association between bacterial species and altered metabolism, related to corpulence and T2D severity

**Fig 3:** FMT-induced Altered Metabolic Phenotype in Recipient Mice




**b**: Body masses & compositions; **k**: Liver mass; **l**: Epididymal Adipose Tissue, showing that the body mass and fat mass of the r-Severe group significantly increased more than r-Mild group, indicating higher corpulence
**f**: Blood Glucose Levels during OGTT; **g**: Area under Curve of OGTT; **i**: HOMA-IR index; **j**: QUICKI index, showing that r-Severe group has significantly poorer glucose tolerance and is significantly more insulin resistant than r-Mild group

**Fig 4:** Bacterial species from class *Bacteroidia* associated with increased corpulence, glucose intolerance and insulin resistance in recipient mice




**r-Mild**: recipients of FMT from 5y-Mild cluster; **r-Severe**: recipients of FMT from 5y-Severe cluster

**Left**: Relative class abundances across recipients' clusters and CDe sizes

**Middle**: CDe size estimates, showing 8 bacterial species were more prevalent in r-Mild animals (CDe<0), and 6 species in r-Severe animals (CDe>0), of which 5 species belonged to the class Bacteroidia and were also more prevalent in humans from the 5y-Severe cluster

**Right**: Heatmap of Spearman's rank correlations between 6 species and animal's phenotypic variables (*: p < .05; **: q < 0.1), showing association between bacterial species and altered metabolism, related to corpulence, glucose intolerance (OGTT) and insulin resistance (IR indices)

## Key findings

- The *Bacteroidia* class of bacteria was associated with greater T2DM severity and alterations of glucose homeostasis markers.
- The gut microbiome (GM) of good metabolic responders was like that of Stable-Mild patients, suggesting that post RYGB metabolic improvements could be attributed to a partial restoration of a "Mild" GM profile.
- The hallmark GM changes associated with T2DM severity were partially transmittable in mice via FMT and resulted in a change in the recipient mice's metabolism.
- Following transplantation, there were strong similarities between donors and recipient mice's phenotypes, as well as bacterial abundances. This suggests that there is partial involvement of the GM in T2DM severity and persistence after RYGB.

Implications of findings

- Considering that the GM composition plays a role in post bariatric surgery DR, modulating the GM composition, on top of existing therapies, could improve outcomes for post-RYGB patients dramatically.
- However, more studies have to be done to validate the efficacy of GM modulation in reducing DR in post bariatric surgery patients.
- Therefore, a future research question could be whether modulating the GM composition in post bariatric surgery patients could reduce DR rates more significantly compared to conventional therapies.

## Conclusion

This study concludes that GM contributes to long-term T2D persistence and severity after RYGB.

## References

1. NCD Risk Factor Collaboration (NCD-RisC). Rising rural body-mass index is the main driver of the global obesity epidemic in adults. Nature. 2019May8;569(7755):260–4.
2. Lim RB. Bariatric surgery for management of obesity: Indications and preoperative preparation [Internet]. Jones D, Chen W, editors. UpToDate. [cited 2022Nov24]. Available from: https://www.uptodate.com/contents/bariatric-surgery-for-management-of-obesity-indications-and-preoperative-preparation
3. Sjöström L. Bariatric surgery and reduction in morbidity and mortality: Experiences from the SOS Study. International Journal of Obesity. 2009Jan12;32(S7).
4. Debédat J, Sokolovska N, Coupaye M, Panunzi S, Chakaroun R, Genser L, et al. Long-term relapse of type 2 diabetes after roux-en-Y gastric bypass: Prediction and clinical relevance. Diabetes Care. 2018;41(10):2086–95.
5. Aron-Wisnewsky J, Sokolovska N, Liu Y, Comaneshter DS, Vinker S, Pecht T, et al. The advanced-DIAREM score improves prediction of diabetes remission 1 year post-roux-en-y gastric bypass. Diabetologia. 2017;60(10):1892–902.
6. Aron-Wisnewsky J, Prifti E, Belda E, Ichou F, Kayser BD, Dao MC, et al. Major microbiota dysbiosis in severe obesity: Fate after bariatric surgery. Gut. 2018;68(1):70–82.