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Brief Communication

Modified SADI-S in non-obese diabetic rats: Description of the surgical technique

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ABSTRACT

Background: Single anastomosis duodenoileal bypass with sleeve gastrectomy (SADI-S) is a predominantly malabsorptive technique that has shown excellent results in morbid obese patients. The aim of this study is to establish a rodent model modifying the SADI-S technique by performing a proximal duodenojejunal anastomosis. This model can be useful for the study of glucose metabolism without malabsorption observed after the SADI-S technique.

Methods: Goto-Kakizaki rats, a genetic model of non-obese and non-hypertensive type 2 diabetes mellitus, that develop hyperglycemia at an early age was used. Surgery consisted in a sleeve gastrectomy, duodenojejunal anastomosis and duodenal exclusion using three different techniques: duodenal transection (DT), duodenal ligation with hem-o-lock (DLH), and duodenal ligation with suture (DLS). Surgery time, weight loss, morbidity and mortality were recorded.

Results: A total of 16 animals were subjected to surgical intervention and overall mortality was 25 %, with the DT group showing the highest mortality rate (42.9 %). No differences were observed among groups in terms of weight loss.

Conclusion: The surgical technique described in this work is feasible and reproducible. Weight loss is comparable regardless of the technique used for duodenal exclusion.

Introduction

Gastrointestinal surgery provides the most substantial and sustainable weight loss in patients with obesity and diabetes mellitus (DM) [1]. Moreover, the benefits of bariatric surgery may extend beyond simply losing body weight, since glycemic control improves even before substantial weight loss after surgery [2]. Several studies, both in animal models and in humans, have shown the effectiveness of metabolic surgery [3,4]. The single anastomosis duodenoileal bypass with sleeve gastrectomy (SADI-S) is a predominantly malabsorptive technique that has shown excellent results in morbidly obese patients [2]. The advantages of SADI-S include the single-anastomosis procedure, no mesenteric opening and pyloric preservation and the physiological advantages of the short circuit for nutrient delivery from the stomach to the ileum [5]; however, by performing a proximal anastomosis in the duodenojejunal bypass with sleeve gastrectomy (SADJB-SG) malabsorption could be avoided.

New experimental models may help to dissociate some metabolic effects from weight loss. For this reason, the main goal of this study is to establish a new model modifying the SADI-S technique and performing a proximal anastomosis in an animal model of non-obese type 2 DM (T2DM) for further studies of the effect of surgery on glucose metabolism.

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Materials and methods

Animals

12-week-old male Goto-Kakizaki (GK) rats were used for this study. GK rats were purchased from Universidad Miguel Hernández (Alicante, Spain) and housed individually in environmentally controlled conditions (22 ± 1 °C, 60 % of humidity and 12 h light/dark cycle), and had access to water and food *ad libitum*. All experimental procedures were approved by the Ethical Committee for Animal Welfare of the Universidad Rey Juan Carlos and Comunidad Autónoma de Madrid, Spain.

Perioperative care

All animals were subjected to an exhaustive pre- and post-operative control and care. An important aspect of this care is the habituation of the animals to the management, as well as the adaptation to a soft diet to avoid malnutrition and dehydration. One week prior to surgery, rats were exposed to palatable gelatine tablets prepared with unflavored gelatin (Royal, Spain) in a mixture of water and liquid diet for diabetic patients (Diben; Fresenius Kabi, Spain), in a 1:1 ratio. One tablet/day is placed in each cage for rat habituation (Fig. 1A).

After the surgical procedure was completed, prewarmed saline were administered s.c. to prevent dehydration and during the first 72 h after surgery, analgesia and saline were administered every 12 h (Fig. 1B). From the second to the seventh postoperative day, palatable gelatin tablets were also administered. Finally, from the third postoperative day, a solid diet was gradually introduced until the seventh day, when free access to food was allowed.

Surgical procedure

Animals were fasted for 6 h before surgery and allowed *ad libitum* access to water. Then, rats were anesthetized with isoflurane (4 % induction, 2 % maintenance) (Karizoo Laboratories, Spain) and a mixture of 0.05 mg/kg buprenorphine (Buprecare; Ecuphar NV, Belgium) and 1 mg/kg meloxicam (Meloxidolor; Dechra Veterinary Products, Spain) was administered subcutaneously (s.c.). Besides, Ciprofloxacin (0.1 mg/kg; Fresenius Kabi, Spain) was intraperitoneally (i.p.) administered. Ophthalmic gel (Lipolasic; Bausch & Lomb, Spain) was then applied and the animal was placed on a heating pad in supine position. Body temperature was continuously monitored by a rectal probe (Fig. 1A).

After shaving the abdomen and disinfecting the skin, the xiphoid was identified and a 3-cm midline abdominal incision was made. The ligament of Treitz was located and 15 cm distally a side-to-side duodenojejunal anastomosis was performed 3–5 mm distal to the pylorus using 7–0 polydioxanone suture (Ethicon, Cat No. Z149H). Subsequently, the first portion of the duodenum was excluded, just distal to the anastomosis. Three methods were used for duodenal exclusion, therefore, 3 experimental groups were established:

- DT: Transection and closing duodenal stumps with double ligation with transfixive 000 silk suture (Ethicon, Cat No. C013D) (Fig. 2A).
- DLH: Ligation with two hem-o-locks (Weck® Hem-o-lok®) (Fig. 2B).
- DLS: Ligation with suture using double 000 silk suture (Ethicon, Cat No. C013D) (Fig. 2C).

Once duodenal exclusion was completed, the stomach was exteriorized and the entire greater curvature was devascularized by ligating the gastrosplenic vessels with small ligaclips (Ethicon, Cat No. MCS20). Tubular gastrectomy of approximately 70 % of the stomach was performed by sectioning it with scissors and sutured with polydioxanone 5–0 (Ethicon, Cat No. Z503G) in a single layer with invaginating stitches. The laparotomy was closed using Polydioxanone 5–0 for the muscle plane and Vycril Rapide 3–0 with hidden stitches for the skin. Surgical time and intraoperative complications were recorded.

Weight record

Animal weight was recorded weekly from 2 weeks before surgery

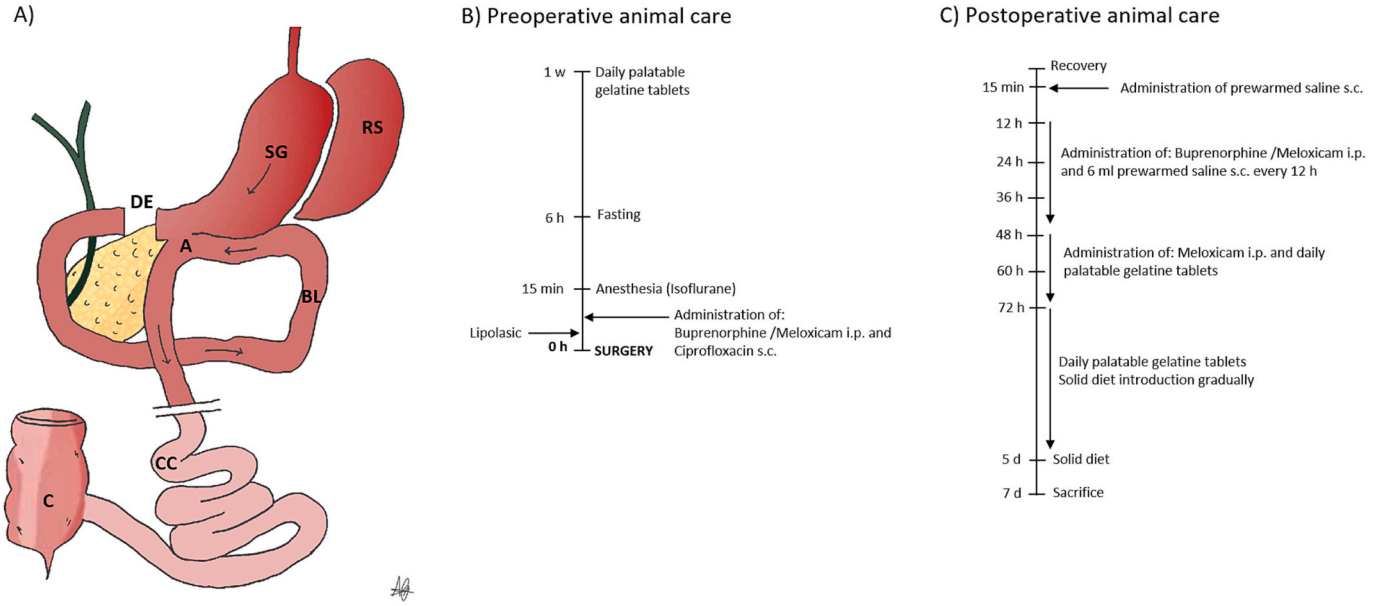

Fig. 1. Graphical diagram of the surgical technique and timeline of pre-operative and post-operative care. A) Schematic illustration of the technical modification and anatomy of single anastomosis duodenojejunal bypass plus sleeve gastrectomy. DE: duodenal exclusion, A: loop duodenojejunal anastomosis, SG: sleeve gastrectomy, RS: resected stomach, BL: biliopancreatic limb, CC: common channel, C: colon. B) During the preoperative phase, the animals were habituated to the palatable gelatins made with liquid diet to ensure good acceptance during the postoperative phase. Just before the procedure, they were administered analgesia and antibiotics, and their eyes were protected with ophthalmic gel. C) The animals were subjected to an exhaustive postoperative control. During the first week after surgery, analgesia was administered for pain management. Both weight and food intake were monitored daily and the animals were gradually switched from gelatine to solid diet according to their tolerance.

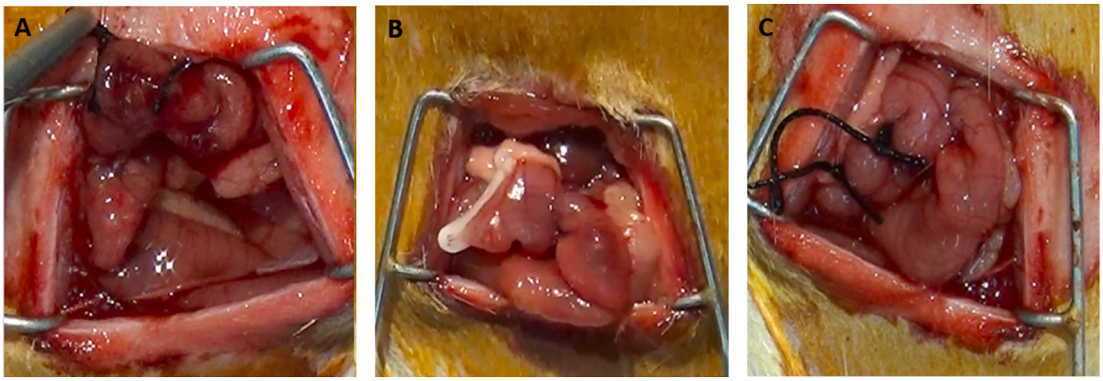

Fig. 2. Representative photographs of the three methods used for duodenal exclusion. A) DT and closing duodenal stumps with double ligation with transfixive 000 silk suture. B) Duodenal ligation with two hem-o-locks. C) Duodenal ligation with suture.

and daily during the first postoperative week. Data were analyzed with STATA 17.0. Mixed model effects with unstructured covariance matrix was adjusted to estimate trend in weight measure over time. A p value $<$ 0.05 was considered as statistically significant.

Results

A total of 16 animals were used, of which 7 underwent DT, 4 underwent DLH and 5 underwent DLS. There was no intraoperative mortality. Bleeding occurred in two cases that were controlled by suture ligation and clips. Despite the mastery of microsurgery, the most important technical challenge was the performing the duodenojejunal anastomosis. Besides, despite the 6 h of fasting, the stomach of the rats was full in all cases, which required the stomach to be emptied and washed out before sleeve gastrectomy. The mean operative time was 98 min, with some differences depending on the duodenal exclusion technique, being 110, 90 and 95 min in DT, DLH and DLS respectively.

Overall postoperative mortality was 25 %, the analysis of mortality rates by group indicated that it was 42.9 %, 22.2 % and 0 % in the DT, DLS and DLH respectively (Table 1). All deaths occurred within 24 h postoperatively due to duodenal or anastomotic loop ischemia. Necropsies also revealed an abdominal abscess related to the sleeve gastrectomy. Regarding body weight, there was a gradual and statistically significant weight loss in the three experimental groups (Table 1). This weight loss was slightly higher in the DLS group (Linear trend: -0.37, CI 95 %: -0.42; -0.33).

Discussion

Based on the clinical experience of laparoscopic sleeve gastrectomy with duodenojejunal bypass in diabetic non-obese patients and the single anastomosis approach [2,6], we have developed an animal model of single anastomosis duodenojejunal bypass with sleeve gastrectomy in GK rats in order to further investigate the long-term metabolic effects. In 2018, Montana and colleagues first developed SADI-S technique in Wistar rats [5]. Recently, only one experience has been published using SADJB-SG but they focus on the metabolic and inflammatory effects but not on the surgical details and difficulties [7].

A very important issue when dealing with surgery in rodents is the perioperative care in order to reduce mortality, pain and ensure animal welfare. Among the preoperative care, although most studies use liquid diets [8,9], we habituated the rats for 1 week using gelatins prepared with liquid diet. In our experience, the gelatin was well tolerated by rats and allowed us to reduce the frequency of s.c. saline administration during the postoperative period, thus reducing the stress of the animals. On the other hand, continuous intraoperative temperature monitoring is crucial to prevent the death of the animal due to hypothermia. We used equipment for temperature control and a heating pad, which maintained a constant body temperature of 37 °C. Moreover, the abdominal cavity was frequently irrigated with prewarmed saline.

Regarding the surgical time, other authors have reported that the mean surgical time for SADI-S was 45 min and it did not differ according to the duodenal exclusion technique used [5]. The mean operative time in our series was 98 ± 5 min, depending on the duodenal exclusion technique used. We performed a hand sewn sleeve gastrectomy that leads to prolonged operative time in comparison with Montana that uses surgical staplers [5]. Although these small differences have not had an impact on intraoperative mortality, gastrointestinal surgery is associated with high morbidity and mortality in rats [10]. Despite the exhaustive care of the animals, postoperative mortality was high in the DT group, consistent with the mortality described by other authors [10]. Necropsies revealed the presence of signs of duodenal ischemia mainly related to the anastomosis in both DT and DLS, so the duodenal exclusion technique employed could explain the differences in mortality observed. Another important consideration is the possibility of duodenal repermeabilization when the DT is not performed for its exclusion. In our experience, for long term and follow up studies we recommend DT technique for exclusion, even assuming a higher mortality but not risking duodenal repermeabilization seen more frequently in duodenal ligation techniques.

Conclusion

SADJB-SG technique in non-obese diabetic GK rats is a feasible and reproducible experimental model. The description of this model provides the rationale for future studies of the metabolic effects of the surgery.

Table 1
Mixed model results. EM: estimated mean; SE: standard error; CI: confidence interval.

	BASELINE		7 DAYS MEASUREMENT		LINEAR TREND					
GROUP	EM	SE	EM	SE	LINEAR SLOPE	CI 95 %		LINEAR SLOPE TEST	INTERACTION EFFECT TEST	
DT	266.42	9.91	207.35	10.53	−0.35	−0.40	−0.30	<0.001	0.142	0.541
DLS	273.44	10.78	210.89	11.18	−0.37	−0.42	−0.33	<0.001	0.036	ref
DLH	287.38	12.11	237.38	12.70	−0.30	−0.35	−0.24	<0.001	ref	

Abbreviations

DLH	duodenal ligation with two hem-o-locks
DLS	duodenal ligation with suture
DM	diabetes mellitus
DT	duodenal transection
SADI-S	single-anastomosis duodenoileal bypass with sleeve gastrectomy
SADJB-SG	single anastomosis duodenojejunal bypass with sleeve gastrectomy
T2DM	type 2 diabetes mellitus

CRediT authorship contribution statement

Sirio Melone: Data curation, Funding acquisition, Investigation, Methodology, Writing – original draft, Writing – review & editing, Formal analysis. **José Mª Fernández-Cebrián:** Investigation, Supervision, Validation. **Mario Amores:** Data curation, Investigation, Methodology. **Elia Pérez-Fernández:** Data curation, Formal analysis, Methodology. **Carlos Guijarro:** Methodology, Supervision, Validation. **Sagrario Martínez:** Investigation, Supervision. **Maria Ruth Pazos:** Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Supervision, Writing – review & editing, Writing – original draft.

Declaration of competing interest

The authors declare that they have no conflict of interest.

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Ethical approval

All experimental procedures were approved by the Ethical Committee for Animal Welfare of the Universidad Rey Juan Carlos and Comunidad Autónoma de Madrid, Spain (PROEX 281/19). The experimental protocol met European and Spanish (2010/63/EEC and RD 53/2013) guidelines for the protection of experimental animals.

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References

[1] de Campos Martins MVD, Peixoto AA, Schanaider A, Esposito CC, Aratanha CBA. Glucose tolerance in the proximal versus the distal small bowel in Wistar rats. Obes Surg 2009;19(2):202–6. https://doi.org/10.1007/s11695-008-9651-3.

[2] Sánchez-Pernaute A, Rubio MA, Cabrerizo L, Ramos-Levi A, Pérez-Aguirre E, Torres A. Single-anastomosis duodenoileal bypass with sleeve gastrectomy (SADI-S) for obese diabetic patients. Surg Obes Relat Dis 2015;11(5):1092–8. https://doi.org/10.1016/j.soard.2015.01.024.

[3] Rubino F, Marescaux J. Effect of duodenal–jejunal exclusion in a non-obese animal model of Type 2 diabetes: a new perspective for an old disease. Ann Surg 2004;239(1):1–11. https://doi.org/10.1097/01.sla.0000102989.54824.fc.

[4] Higa K, Ho T, Tercero F, Yunus T, Boone KB. Laparoscopic Roux-en-Y gastric bypass: 10-year follow-up. Surg Obes Relat Dis 2011;7(4):516–25. https://doi.org/10.1016/j.soard.2010.10.019.

[5] Montana L, Lamon M, Barrat C, Hansel B, Magnan C, Arapis K. Establishing a reproducible murine animal model of Single Anastomosis Duodenoileal Bypass with Sleeve Gastrectomy (SADl-S). Obes Surg 2018;28(7):2122–5. https://doi.org/10.1007/s11695-018-3254-4.

[6] Navarrete SA, Leyba JL, Llopis SN. Laparoscopic sleeve gastrectomy with Duodenojejunal bypass for the treatment of Type 2 diabetes in non-obese patients: technique and preliminary results. Obes Surg 2011;21(5):663–7. https://doi.org/10.1007/s11695-011-0371-8.

[7] Yu H, Song Z, Zhang H, Zheng K, Zhan J, Luo Q, et al. Duodenojejunal bypass plus sleeve gastrectomy reduces infiltration of macrophages and secretion of TNF-α in the visceral white adipose tissue of Goto-Kakizaki rats. Obes Surg 2019;29(6):1742–50. https://doi.org/10.1007/s11695-019-03755-1.

[8] Zubiaga L, Abad R, Ruiz-Tovar J, Enriquez P, Vílchez JA, Calzada M, et al. The effects of one-anastomosis gastric bypass on glucose metabolism in Goto-Kakizaki rats. Obes Surg 2016;26(11):2622–8. https://doi.org/10.1007/s11695-016-2138-8.

[9] Bruinsma BG, Uygun K, Yarmush ML, Saeidi N. Surgical models of Roux-en-Y gastric bypass surgery and sleeve gastrectomy in rats and mice. Nat Protoc 2015;10(3):495–507. https://doi.org/10.1038/nprot.2015.027.

[10] Inabnet WB, Milone L, Korner J, Durak E, Ahmed L, Pomrantz J, et al. A rodent model of metabolic surgery for study of type 2 diabetes and positron emission tomography scanning of beta cell mass. Surg Obes Relat Dis 2009;5(2):212–7. https://doi.org/10.1016/j.soard.2008.09.007.