Original Article

MOLECULAR METABOLISM

TRPV1 neurons regulate β-cell function in a sex-dependent manner

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ABSTRACT

There is emerging evidence to support an important role for the transient receptor potential vanilloid type 1 (TRPV1) sensory innervation in glucose homeostasis. However, it remains unknown whether the glucoregulatory action of these afferent neurons is sex-biased and whether it is pancreatic β-cell-mediated.

Objective: We investigated in male and female mice whether denervation of whole-body or pancreas-projecting TRPV1 sensory neurons regulates adult functional β-cell mass and alters systemic glucose homeostasis.

Methods: We used a combination of pharmacological and surgical approaches to ablate whole-body or pancreatic TRPV1 sensory neurons and assessed islet β-cell function and mass, aspects of glucose and insulin homeostasis, and energy expenditure.

Results: Capsaicin-induced chemodenervation of whole-body TRPV1 sensory neurons improved glucose clearance and enhanced glucose-stimulated insulin secretion without alterations in β-cell proliferation and mass, systemic insulin sensitivity, body composition, and energy expenditure. Similarly, denervation of intrapancreatic TRPV1 afferents by pancreas intraductal injection of capsaicin or surgical removal of the dorsal root ganglia projecting into the pancreas lowered post-absorptive glucose levels and increased insulin release upon glucose stimulation. The beneficial effects of TRPV1 sensory denervation on glucose tolerance and β-cell function were observed in male but not female mice.

Conclusion: Collectively, these findings suggest that TRPV1 neurons regulate glucose homeostasis, at least partly, through direct modulation of glucose-induced insulin secretion and that this regulation operates in a sex-dependent manner.

Keywords TRPV1 sensory innervation; Glucose homeostasis; β-cell function; Sex difference

1. INTRODUCTION

All forms of diabetes are commonly characterized by a scarcity in the number of functional pancreatic insulin-secreting β cells [1]. This deficit results in chronic hyperglycemia that, if not controlled, leads to multi-organ complications, impairs life quality and ultimately shortens lifespan [2]. Understanding the underlying biological processes enabling survival, growth, and function of islet β cells is of utmost importance in diabetes research as the identification of cells and molecules capable of enhancing functional islet β-cell mass would reverse diabetes and halt its complications. New insights from the field of integrative physiology highlight the relevance of metabolic organ cross-talk in pancreatic β-cell regeneration and function [3]. Studies focused on inter-organ communication between the pancreas and extra-pancreatic systems have yielded several molecular targets to enhance growth and/or activity of islet β cells [4–7]. In addition to the peripheral tissues, the autonomic nervous system plays key roles in pancreatic β cells [8,9]. The pancreas receives a large supply of parasympathetic, sympathetic and sensory neurons [8,10–12]. Parasympathetic innervation enhances insulin secretion during the meal-induced cephalic insulin response [13] and regulates β-cell proliferation and function in normal and insulin resistance settings [14,15]. Conversely, the sympathetic branch inhibits insulin release during hypoglycemia-induced metabolic stress [16]. While there is ample literature documenting the effect of efferent neurons and their respective neurotransmitters in pancreatic islet cells, the role of sensory neurons is largely unexplored despite the dense innervation of the endocrine pancreas [8,12,17,18]. Visceral afferent innervation mediates the adaptive response to a variety of internal and external stimuli and the disruption of sensory inputs is associated with the initiation and progression of various chronic diseases [19]. Recently, experimental evidence in rodents has linked dysfunction of sensory neurons to the development of type 1 and type 2 diabetes [20–23]. Interestingly, the development of diabetes (either type 1 or type 2) and the perception of sensory modalities (e.g., physical pain) are sex-biased. Men are inherently more prone to develop diabetes [24] and exhibit higher tolerance to pain perception [25], thus suggesting a relationship between glucose homeostasis and sensory systems. Moreover, the

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prevalence of diabetes in individuals with spinal cord injury is 3-fold higher compared to non-injured individuals [26,27]. These observations strengthen the idea that visceral sensory innervation plays a critical role in glucose homeostasis. Nevertheless, it remains unknown whether sensory regulation of glucose homeostasis operates similarly in males and females and whether pancreatic β cells are major targets of the visceral sensory inputs.

The pancreas is innervated by sensory neurons originating from the vagus and the spinal nerves with cell bodies located in the nodose ganglia and dorsal root ganglia (DRG), respectively [28]. DRG afferents perceive a variety of sensory modalities including thermal, chemical, mechanical and inflammatory stimuli [29]. A large proportion of these sensory neurons express the transient receptor potential vanilloid 1 (TRPV1), a non-selective cation channel that is sensitive to capsaicin — the pungent component of hot chili peppers, involved in detecting noxious thermal and chemical stimuli [30,31]. TRPV1-expressing neurons have dual sensory-efferent functions; that is, in addition to their ability to convey information arising from the tissues they innervate to the central nervous system, DRG neurons are also able to release substances locally from the same terminal that is excited by visceral cues [32]. TRPV1 neurons are primarily peptidergic and secrete calcitonin gene-related protein (CGRP) and substance P (SP); two circulating peptides elevated in insulin resistance and obesity [33]. Genetic and pharmacological ablation of TRPV1-expressing neurons in models of diabetes and obesity normalizes CGRP levels [34], enhances insulin release from the β cells [35] and improves insulin sensitivity in peripheral tissues [34]. It is unclear how desensitization of TRPV1 sensory neurons generates anti-diabetic effects. Capsaicin-sensitive neurons affect a variety of peripheral and central biological processes, including food intake, energy expenditure, body weight regulation, systemic inflammation, peripheral insulin resistance, GLP-1 release, adiponectin and leptin signaling, and β-cell mass and function [33]. A major obstacle that hinders progress in understanding the proportional contribution of individual peripheral tissues in the sensory control of glucose homeostasis is the long-standing lack of methods to manipulate neural activity in a tissue-specific manner.

In this study, we used a combination of chemical and surgical models to address whether TRPV1 sensory neurons directly modulate functional β-cell mass and consequently alter glucose homeostasis in male and female mice. We demonstrated that both whole-body and pancreas-selective chemodenervation of TRPV1 afferents improved glucose tolerance and glucose-stimulated insulin release without altering insulin sensitivity. Interestingly, chemical ablation of pancreatic TRPV1 neurons, via pancreas intraductal injection of capsaicin, enhanced glucose-stimulated insulin secretion and glucose excursion with no apparent change in pancreatic β-cell proliferation and mass. Mice lacking lower thoracic DRGs that contain pancreas-projecting sensory neurons also exhibited enhanced glucose clearance and glucose-induced insulin secretion. Finally, we demonstrated that the beneficial effect of TRPV1 sensory denervation on glucose homeostasis was restricted to male mice. Together, these data suggest that TRPV1 sensory neurons modulate, at least in part, directly pancreatic β-cell function in a sex-dependent manner.

2. METHODS

2.1. Animals

All mice studied were five-week-old wild-type males and females on the C57BL/6J background (stock #000664, The Jackson Laboratory) unless indicated otherwise. Mice were housed in pathogen-free facilities and maintained on a 12-hour light/dark cycle in the Animal Care Facility at Child Health Institute of New Jersey. All studies and protocols were approved by the Rutgers University Animal Care and Use Committee and were in accordance with National Institutes of Health guidelines. Blood glucose was monitored using an automated glucose monitor (Bayer) and plasma insulin was detected by ELISA (Crystal Chem).

2.2. Pancreas intraductal injection

Mice were anesthetized with ketamine (100 mg/kg BW) and xylazine (10 mg/kg BW) and received a subcutaneous injection of buprenorphine (0.05 mg/kg BW). A midline incision of 2-to-3 cm was made along the abdomen. The duodenum was exposed and the common bile duct was clamped caudal to the liver. The intraductal injection was performed using a 28G1/2 needle. The syringe was entered into the pancreatic duct from the duodenum through the ampulla of Vater and carefully advanced in a retrograde manner. The needle was secured by a micro clamp and 100 μl of vehicle or capsaicin (50 μg/100 μl) were slowly administered over one minute. After clamp removal, the wound on the ampulla was sealed with 3M Vetbond Tissue Adhesive (3M Animal Care). Abdominal muscle layer was closed with simple interrupted stitch with Polyamide Monofilament (B. Braun) and the overlying skin was sutured using 9 mm autoclips (Braintree Scientific).

2.3. Dorsal root ganglionectomy

Mice were anesthetized with ketamine (100 mg/kg BW) and xylazine (10 mg/kg BW) and received a subcutaneous injection of buprenorphine (0.05 mg/kg BW). The skin and muscles were separated exposing unilaterally the region of 11th, 12th and 13th thoracic vertebrae and the dorsal region transverse processes. To expose the DRGs, transverse processes were trimmed with a micro drill (Roboz Surgical Instrument). Whole DRGs were carefully cut with tipped Dumont #5 forceps and Vannas spring scissors (Fine Science Tools) without disrupting the spinal cord. Dorsal muscle layer was then closed with interrupted suture technique with Polyamide Monofilament (B. Braun). The overlying skin was sutured using 9 mm autoclips (Braintree Scientific).

2.4. Immunostaining

Pancreases were analyzed by immunolabeling using anti-BrdU (Dako) and anti-insulin (Cell Signaling Technology) antibodies. Quantification of β-cell mass was performed as described previously [4,6]. TRPV1 expression in the pancreas was analyzed by immunostaining using a rabbit anti-TRPV1 antiserum as described in [36]. DRGs were stained with antibodies for TRPV1 (Alomone Labs) and CGRP (Sigma).

2.5. Transmission electron microscopy

Pancreas tissue samples were minced and fixed in 2.5% glutaraldehyde in 0.1M phosphate buffer and processed for routine electron microscopy as previously described [37]. Images were obtained using Philips 301 Transmission Electron Microscope. Three cytoplasmic areas (4 × 4 micrometer) were randomly selected from each cell for the quantification of the dense core insulin-containing vesicles. Total of seven cells from three vehicle-treated mice and sixteen cells from four capsaicin-treated mice were quantified and represented as the average number of dense core vesicles per um^2.

2.6. Statistical analysis

All data are presented as mean ± SEM. Data were analyzed using Student's t-test and a "p" value < 0.05 was considered statistically significant.

3. RESULTS

3.1. Whole-body chemical ablation of TRPV1 sensory neurons improves β-cell function and glucose tolerance in a sex-dependent manner

To determine whether TRPV1 sensory neurons regulate glucose homeostasis in a sex-dependent manner, we used the capsaicin-induced denervation paradigm [38]. Briefly, vehicle or capsaicin (50 mg/kg) was subcutaneously injected in five-week-old male and female C57BL/6J mice on three consecutive days into the scruff of the neck. Mice were then subjected to a series of metabolic phenotyping assays (Figure 1A). Ten days post-treatment, capsaicin-treated mice displayed improved glucose tolerance (Figure 1B) without alteration in insulin sensitivity (Figure 1C). The effect of TRPV1 sensory denervation on post-absorptive glucose levels sustained up to 30 days after capsaicin treatment (data not shown) despite identical body weight, random-fed glucose and insulin levels in vehicle- and capsaicin-treated male mice (Figure 1D–F). To investigate whether the beneficial effect of capsaicin on glucose clearance was due to changes in body composition, we used dual-energy X-ray absorptiometry (DEXA) to assess body fat and lean tissue contents in control and sensory denervated mice. DEXA analysis revealed unaltered body composition in capsaicin-injected mice when compared to their age-matched vehicle-injected littermates. Lean mass and fat content from visceral, subcutaneous and interscapular regions were identical in the experimental groups (Figure 1G,H). The lack of differences in body composition was confirmed by weighing the tissues post mortem (Figure S1A). In contrast to male mice, no significant differences in glucose excursion were observed in capsaicin-injected female mice (Figure 1I). Body weight, blood glucose and insulin levels, insulin sensitivity and body composition were similar between chemodenervated female mice and vehicle-treated littermates (Figure 1J–O). To further characterize the metabolic profile of TRPV1 sensory chemodenervated male mice, we used the Oxymax/comprehensive laboratory analysis monitoring system (CLAMS) system. Animals were single-housed in metabolic cages over a period of five days. To circumvent variations introduced by adaptation to a new environment, data from only the last three-day period were considered. CLAMS analysis did not reveal major changes in total activity, energy expenditure (as assessed by heat production), oxygen consumption (VO2), carbon dioxide production (VCO2) and RER (respiratory exchange ratio = VO2/VCO2) (Figure 2A–E). A slight, but significant, increase in accumulated food intake was noted in capsaicin-treated mice (Figure 2F and S1B). To determine whether the stimulatory action of glucose on insulin release was affected by chemodenervation, vehicle- and capsaicin-treated mice were subjected to glucose-stimulated insulin secretion (GSIS) tests. Our data revealed that while fasting levels of insulin are similar between the experimental groups, chemodenervated male – but not female – mice displayed higher insulin release upon acute glucose challenge when compared to their control littermates (Figure S1C and S1D). Ultrastructural and morphometric analyses of pancreases derived from vehicle- and capsaicin-treated mice revealed no alterations in the number of dense-core vesicles in islets, thus excluding changes in the formation of hormone-containing vesicles as a significant factor modifying β-cell function upon capsaicin treatment (Figure S2). Collectively, these data provide

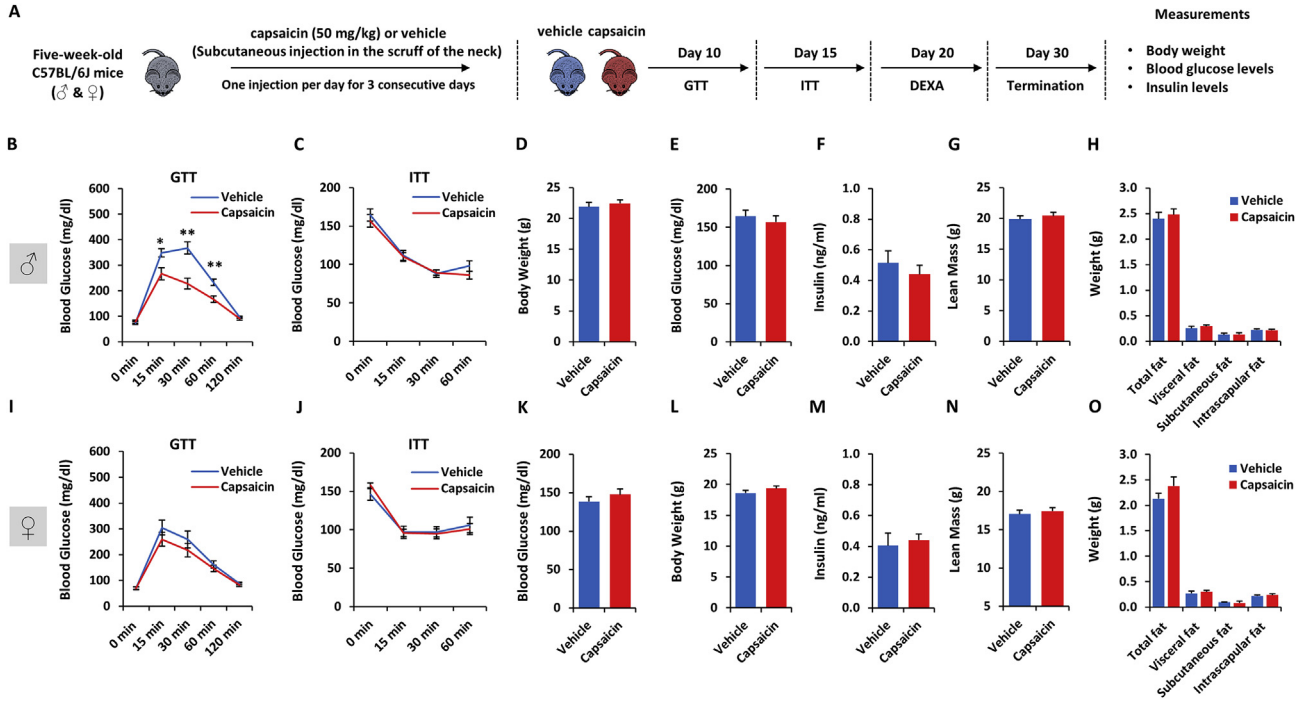

Figure 1: Whole-body chemical ablation of TRPV1 sensory neurons improves glucose tolerance in a sex-dependent manner: Five-week-old male and female C57BL/6J mice were subcutaneously injected into the scruff of the neck with capsaicin (50 mg/kg) or vehicle once per day for three consecutive days. Ten days post-treatment, mice were subjected to metabolic phenotyping tests. **A.** Schematic of the experimental design. **B.** Glucose tolerance test (males). **C.** Insulin tolerance test (males). **D.** Body weight (males). **E.** Random-fed blood glucose (males). **F.** Random-fed insulin levels (males). **G.** Lean mass evaluated by DEXA analysis (males). **H.** Quantification of fat mass by DEXA analysis (males). **I.** Glucose tolerance test (females). **J.** Insulin tolerance test (females). **K.** Body weight (females). **L.** Random-fed blood glucose (females). **M.** Random-fed insulin levels (females). **N.** Lean mass evaluated by DEXA analysis (females). **O.** Quantification of fat mass by DEXA analysis (females). Data represent mean ± SEM. $^{*}p \leq 0.05$, $^{**}p \leq 0.01$ (n = 6–7 per group).

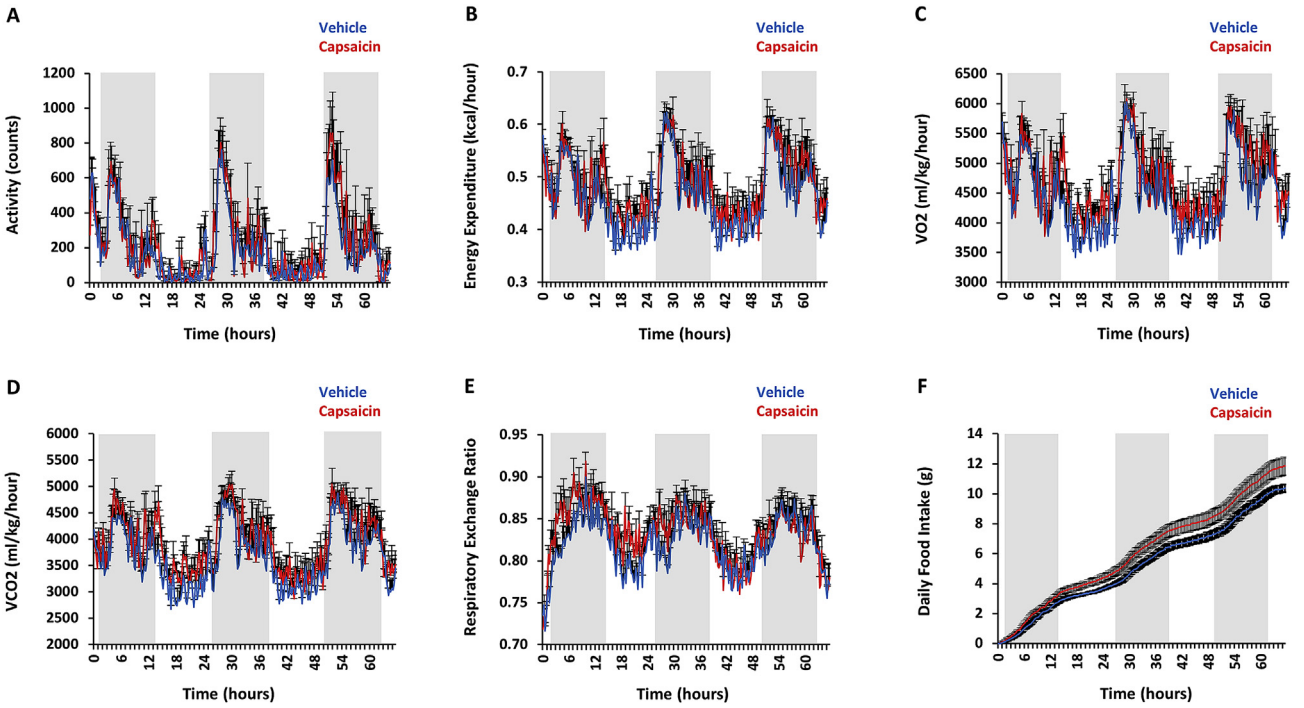

Figure 2: Whole-body TRPV1 sensory denervation does not affect energy expenditure in male mice: Five-week-old male C57BL/6J mice were subcutaneously injected into the scruff of the neck with capsaicin (50 mg/kg) or vehicle once per day for three consecutive days. Twenty-five days post-treatment, indirect calorimetric assays were performed using the Comprehensive Laboratory Animal Monitoring System (CLAMS). **A.** Locomotor activity (counts). **B.** Energy expenditure (kcal/hour) **C.** Oxygen consumption (ml/kg/hour). **D.** Carbon dioxide release (ml/kg/hour). **E.** Respiratory Exchange Ratio. **F.** Diurnal profiles of food intake (g).

evidence for a sexually dimorphic role of TRPV1 sensory neurons in glucose homeostasis. This sensory-mediated glucoregulation occurs independently of changes in insulin sensitivity, body composition and energy expenditure, and implicates the pancreatic β cell as an important target of TRPV1-expressing neurons.

3.2. Denervation of pancreas-projecting TRPV1 afferents enhances β-cell function and glucose tolerance in a sex-dependent manner

To determine whether TRPV1 neurons regulate glucose homeostasis through direct modulation of islet β cells, we performed a single injection of vehicle or capsaicin (50 μg/mouse) into the pancreatic duct of five-week-old male and female C57BL/6J mice (Figure 3A). To confirm capsaicin-induced ablation of pancreas-projecting TRPV1 afferents, we harvested lower thoracic DRGs located at vertebral levels T10 through T13 and conducted TRPV1 and CGRP staining. Morphometric analyses demonstrated that the number of TRPV1-labeled neurons decreased by nearly 35% in capsaicin-treated mice when compared to vehicle-treated littermates (Figure S3A and S3B). CGRP neurons were unaffected by capsaicin treatment indicating a specific action of capsaicin toward TRPV1 afferents (Figure S3A and S3C). The number of TRPV1/CGRP double-positive neurons was reduced in mice that received pancreas intraductal injection of capsaicin suggesting elimination of TRPV1 peptidergic neurons (Figure S3A and S3D). Similar to whole-body sensory desensitization, pancreas chemodenervation of capsaicin-sensitive afferents resulted in enhanced glucose tolerance and unchanged insulin sensitivity (Figure 3B and C) in male mice. Further, glucose-induced insulin secretion was higher in capsaicin-injected male mice when compared to male littermates injected with vehicle (Figure 3D). Co-immunostaining of pancreatic sections did not reveal expression of TRPV1 in islets β cells (Figure S4A–D), hence ruling out the β cell as a direct target of capsaicin in the pancreas. As a positive control, dorsal root ganglia revealed multiple TRPV1-positive neuronal cell bodies (Figure S4E). To explore whether enhanced GSIS upon chemodenervation in male mice is secondary to increased β-cell mass, we intraperitoneally injected the experimental groups with bromodeoxyuridine (BrdU; 100 mg/kg body weight) three times, once every other two days. The mice were then sacrificed and pancreases were harvested for quantitation of BrdU incorporation in β cells and morphometric analysis of the pancreatic islets. Our data revealed no difference in β-cell proliferation and mass between capsaicin-treated mice and their vehicle-treated littermates (Figure 3E,F). Female mice that received an intraductal injection of capsaicin in the pancreas exhibited no differences in glucose clearance, insulin sensitivity, glucose-stimulated insulin release, and β-cell proliferation and mass (Figure 3G–K). To exclude potential confounding effects generated by vagal TRPV1-expressing sensory neurons, we used five-week-old male and female C57BL/6J mice to surgically ablate lower thoracic DRGs T11 through T13 (Figure S4F and S4G) known to contain pancreas-projecting TRPV1 afferents [39] and assessed glucose clearance, insulin sensitivity, GSIS and β-cell proliferation and mass (Figure 4A). Ablation of DRG afferents projecting into the pancreas resulted in improved glucose tolerance, unaltered insulin sensitivity and enhanced glucose-induced insulin release in absence of noticeable changes in β-cell proliferation and mass in male mice (Figure 4B–F). Female mice that underwent a similar procedure exhibited a normal metabolic phenotype when compared to age-matched mice subjected to sham surgery (Figure 4G–K). Body weight and random-fed blood glucose and insulin levels were normal in mice that underwent pancreas deafferentation and dorsal root ganglionectomy procedures (Figure S5). Taken together, these data

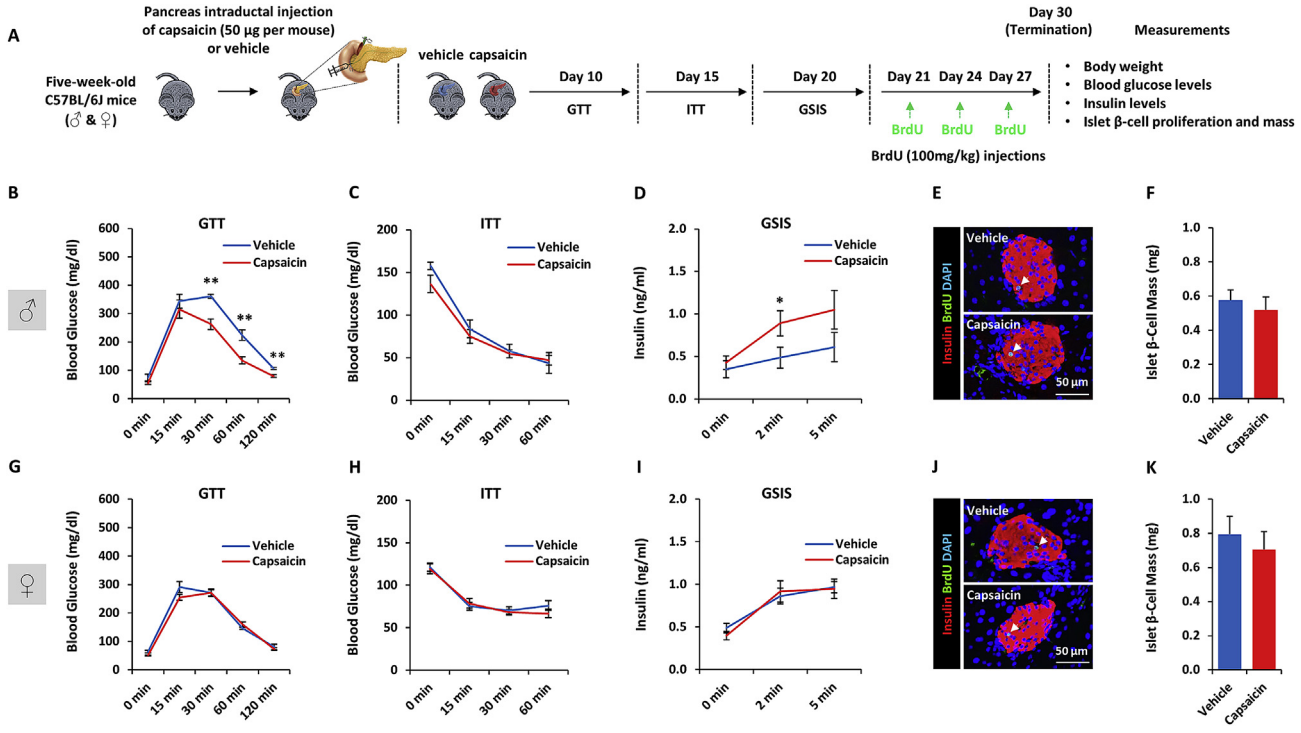

Figure 3: Chemodenervation of pancreas-projecting TRPV1 afferents enhances β-cell function and glucose tolerance in a sex-dependent manner: Five-week-old male and female C57BL/6J mice received a pancreatic intraductal injection of 50 µg of capsaicin per 100 µl of vehicle or vehicle alone. Ten days post-treatment, metabolic phenotyping was performed. **A.** Schematic of the experimental design. **B.** Glucose tolerance test (males). **C.** Insulin tolerance test (males). **D.** Glucose-stimulated insulin secretion (males). **E.** Representative fluorescence images of pancreases co-stained for BrdU (green), insulin (red) and DAPI (blue). Pancreases were harvested from male mice injected with vehicle (upper panel) or capsaicin (lower panel). **F.** Quantification of β-cell mass (males). **G.** Glucose tolerance test (females). **H.** Insulin tolerance test (females). **I.** Glucose-stimulated insulin secretion (females). **J.** Representative fluorescence images of pancreases co-stained for BrdU (green), insulin (red) and DAPI (blue). Pancreases were harvested from female mice injected with vehicle (upper panel) or capsaicin (lower panel). **K.** Quantification of β-cell mass (females). Data represent mean ± SEM. $^{*}p \leq 0.05$ and $^{**}p \leq 0.01$ (n = 4–6 per group).

suggest the existence of a physiological crosstalk between spinal DRG neurons and pancreatic islet β cells, whereby pancreas-projecting spinal sensory neurons modulate insulin release in a sex-dependent fashion.

4. DISCUSSION

Neuromodulation of end-organ systems is an attractive strategy to treat common conditions [40]. With the advent of new neurotechnologies and the reconstruction of high-resolution neuroanatomical maps of the pancreas [17], investigating the neural control of the endocrine β cells holds considerable promise for neuromodulation-based therapies for diabetes [41–43]. Here, we demonstrated that TRPV1 sensory neurons regulate post-absorptive glucose levels through direct modulation of pancreatic β-cell function in a sex-dependent manner.

The major finding that TRPV1 sensory innervation controls glucose homeostasis in a sex-dependent manner is highly relevant as it provides new insights into the role of sensory innervation in the well-known sexual dimorphic pattern of glucose homeostasis. It also offers a valuable clue that can be harnessed to predict the response to neuromodulation-based therapies in men and women [24,44]. One possible explanation is that high levels of estrogen in female mice provides neuroprotection that counters capsaicin-induced chemodenervation of TRPV1 afferents [45]. In support of this hypothesis, a recent study has demonstrated that prolonged exposure of dorsal root ganglia neurons to estradiol reduces TRPV1 response to capsaicin [46]. Because the protective effect of capsaicin on glucose excursion was observed in male mice, an alternative explanation is that male steroid hormones influence TRPV1-expressing neurons and/or functionally associated neural networks. In this context, a recent study has revealed that testosterone binds and activates the transient receptor potential melastatin 8 (TRPM8) known to exhibit functional interactions with TRPV1 [47]. Interestingly, mice lacking *trpm8* gene exhibit decreased glucose-stimulated insulin secretion [48]. It is possible that the sexually dimorphic pattern observed in sensory-mediated control of glucose homeostasis is a consequence, among others, of a testosterone-regulated reciprocal functional interaction between TRPV1- and TRPM8-expressing sensory neurons. Further investigations are needed to address whether the sex-biased action of TRPV1 sensory neurons in insulin secretion is mediated by gonadal hormones and/or driven by intrinsic molecular targets, encoded by sex chromosomes, expressed in islet β cells [49] and/or DRG sensory neurons [50].

Our data indicate that whole-body chemodenervation of TRPV1 sensory afferents in male mice enhances glucose-induced insulin output and improves glucose clearance without alterations in body weight, insulin sensitivity, lean and fat mass, and energy expenditure. These results contradict the idea that deafferentation of capsaicin-sensitive neurons increases glucose clearance via enhancement of insulin sensitivity [51] and strongly pinpoint to the pancreatic β cell as a major target of TRPV1 neurons [21,52]. Indeed, pharmacological and surgical ablation

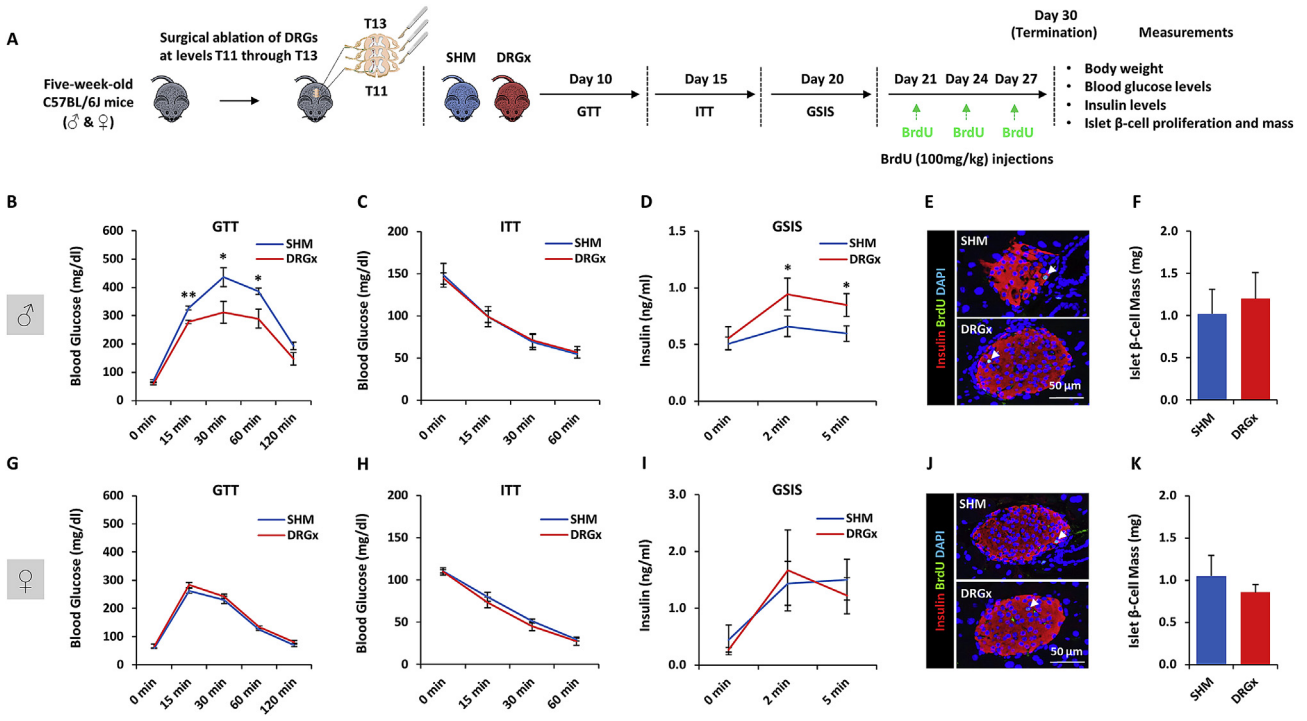

Figure 4: Dorsal root ganglionectomy enhances β-cell function and glucose tolerance in a sex-dependent manner: Five-week-old male and female C57BL/6J mice were subjected to unilateral ganglionectomy of DRGs located at levels T11, T12 and T13. **A.** Schematic of the experimental design. **B.** Glucose tolerance test (males). **C.** Insulin tolerance test (males). **D.** Glucose-stimulated insulin secretion (males). **E.** Representative fluorescence images of pancreases co-stained for BrdU (green), insulin (red) and DAPI (blue). Pancreases were harvested from male mice injected with vehicle (upper panel) or capsaicin (lower panel). **F.** Quantification of β-cell mass (males). **G.** Glucose tolerance test (females). **H.** Insulin tolerance test (females). **I.** Glucose-stimulated insulin secretion (females). **J.** Representative fluorescence images of pancreases co-stained for BrdU (green), insulin (red) and DAPI (blue). Pancreases were harvested from female mice injected with vehicle (upper panel) or capsaicin (lower panel). **K.** Quantification of β-cell mass (females). Data represent mean ± SEM. $^{*}p \leq 0.05$, $^{**}p \leq 0.01$ (n = 4–6 per group).

of pancreas-projecting sensory neurons in male mice enhanced glucose-induced insulin release and improved glucose tolerance in absence of changes in insulin sensitivity. We postulate that TRPV1 neurons exert a physiological repressive activity on insulin release as part of a negative feedback loop mechanism, whereby insulin-induced neuropeptide release represses insulin secretion. This hypothesis is substantiated by studies suggesting that insulin receptors are expressed in pancreas-projecting TRPV1 neurons [39] and that CGRP inhibits insulin release [53]. A potential caveat in this study is that whole-body chemodenervation can generate confounding extra-pancreatic effects that may indirectly affect islet β-cell response. However, the enhanced glucose-induced insulin release observed upon ablation of pancreas-projecting TRPV1 afferents via pancreas intraductal injection of capsaicin *in situ* and after surgical removal of the DRGs projecting in the pancreas support the DRG-islet crosstalk model. Additional studies are warranted to identify the molecular signature and the precise function of TRPV1 neurons projecting exclusively in the endocrine pancreas.

It is relevant to highlight that systemic chemodenervation of TRPV1 sensory neurons increased food intake. This observation is consistent with a recent study which demonstrated that proopiomelanocortin (POMC) neurons in the arcuate nucleus of the hypothalamus also express TRPV1 molecules and which, when activated, reduce food intake [54]. In agreement with the respective roles of central and peripheral TRPV1-expressing neurons in feeding behavior and glucose clearance, another study reported that male Zucker Diabetic Fatty (ZDF) rats treated with high doses of capsaicin displayed lower hyperglycemia, enhanced glucose tolerance and glucose-stimulated insulin secretion, and increased food intake and body weight over time [55]. Overall, these observations highlight the dual roles of TRPV1 neurons in energy balance via central and peripheral mechanisms and further underscore the necessity of developing targeted approaches to manipulate the activity of TRPV1 neurons in a tissue-specific manner.

Taken together, we demonstrated that TRPV1 neurons regulate glucose-stimulated insulin release and glucose homeostasis in a sex-dependent manner. This regulation occurs independently of changes in insulin sensitivity and implicates the pancreatic islet β cell as a major target of sensory neurons. The identification of the genetic makeup of TRPV1 neurons projecting in the endocrine pancreas will lay strong neurobiological foundations to develop selective neuromodulation-based approaches to enhance β-cell activity in individuals with diabetes.

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CONFLICT OF INTEREST

None declared.

APPENDIX A. SUPPLEMENTARY DATA

Supplementary data to this article can be found online at https://doi.org/10.1016/j.molmet.2018.10.002.

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