



OPEN

Comparison between an SGLT2 inhibitor and insulin in tumor-to-tissue contrasts in ^{18}F -FDG PET imaging of diabetic mice

Liang Meidai^{1,2,5}, Du Yujing^{1,3,5}, Liu Zhaoyu², Li Shanshi⁴, Zhao Guangyu¹, Fan Yan¹, Yang Xiuying²✉ & Zhang Jianhua¹✉

^{18}F -fluorodeoxyglucose positron emission tomography (^{18}F -FDG PET) has been widely utilized for tumor diagnosis. Hyperglycemia affects the ^{18}F -FDG uptake and reduces tumor-to-tissue contrasts, however, ideal hypoglycemic drugs are lacking. This study compared the role of insulin with the novel widely used hypoglycemic drug, sodium-glucose cotransporter 2 (SGLT2) inhibitor, on ^{18}F -FDG PET imaging in diabetic conditions. The streptozotocin (STZ)-induced diabetic C57BL/6N mice were inoculated with B16 (mouse melanoma) cells to establish the xenograft tumor model. After the mice had been administrated with dapagliflozin (30 mg/kg, IG) or insulin (0.75 U/kg, IP) for one hour, 9.25 MBq/10 g ^{18}F -FDG was injected. Biodistributions were detected by gamma counting and microPET imaging. The results showed dapagliflozin did not significantly affect the ^{18}F -FDG uptake in tumors but reduced uptake in reference tissues, resulting in a significant increase in the tumor-to-skeletal muscle ratio. Conversely, insulin increased ^{18}F -FDG uptake in tumors without significant reduction in uptake in reference tissues; Although there was an observable improvement in tumor imaging, it did not reach significantly statistical differences. This study suggests that insulin and SGLT2 inhibitor yield comparable effects on the quality of ^{18}F -FDG PET imaging in diabetic patients. Nevertheless, SGLT2 inhibitors would be more suitable when skeletal muscle is used as reference tissue.

Abbreviations

CT	Computed tomography
^{18}F -FDG	^{18}F -fluorodeoxyglucose
GLUT	Glucose transporters
IG	Intragastric administration
IP	Intraperitoneal administration
MRI	Magnetic resonance imaging
PET	Positron emission tomography
SGLT2	Sodium-glucose co-transporter 2
STZ	Streptozotocin
T1D	Type 1 diabetes

^{18}F -Fluorodeoxyglucose positron emission tomography (^{18}F -FDG PET), in combination with high-resolution anatomical structure detection such as computed tomography (CT) or magnetic resonance imaging (MRI), has

¹Department of Nuclear Medicine, Peking University First Hospital, Beijing 100034, People's Republic of China. ²Beijing Key Laboratory of Drug Target and Screening Research, Institute of Materia Medica of Peking, Union Medical College, Beijing 100050, People's Republic of China. ³Department of Nuclear Medicine, Beijing Chaoyang Hospital, Capital Medical University, Beijing 100020, People's Republic of China. ⁴Department of Radiation Oncology, Peking University First Hospital, Beijing 100034, People's Republic of China. ⁵These authors contributed equally: Liang Meidai and Du Yujing. ✉email: lucia@imm.ac.cn; zjh@bjmu.edu.cn

been widely used in clinical diagnostics^{1–3}. It entails the comparison of ¹⁸F-FDG uptake in tumors with that in reference tissues to ascertain the presence of tumor cells². However, elevated blood glucose levels can increase ¹⁸F-FDG uptake in tissues^{4,5}. In diabetic patients, hyperglycemia adversely influences the PET detection of multiple tumors by reducing the glucose uptake ratio between tumors and contrast tissues⁶, and consequently diminishes diagnostic accuracy⁷.

Currently, managing fasting blood glucose levels for diabetic patients who undergo ¹⁸F-FDG PET/CT examination remains a challenge^{1,5,8}. Collaborative nuclear medicine groups such as the European Nuclear Medicine Association (EANM) and the American Academy of Radiology (ACR) have proposed protocol guidelines for the management of diabetic patients undergoing ¹⁸F-FDG PET/CT examination¹. The current accepted guidance for diabetic patients is if their blood glucose level exceeds 11 mmol/L (approximately 200 mg/dL), the patient must either defer the examination, use rapid-acting insulin, or wait until their blood glucose meets the requirements^{1,5}. Since insulin can significantly increase tissue glucose uptake, the interval between insulin and ¹⁸F-FDG injection should exceed four hours¹. However, insulin notably influences glucose transporter 4 (GLUT-4), leading to excessive glucose/FDG uptake by skeletal muscle¹. In addition to the risk of hypoglycemia, insulin can also result in false-negative tumor detection^{1,8,9}. Additionally, 9.5% of diabetic patients still had blood glucose levels above 10 mmol/L⁸. Although limited research has been conducted, other oral hypoglycemic drugs, such as sulfonylureas, glinides, dipeptidyl peptidase IV inhibitors, glucagon-like peptide-1 receptor agonists, and thiazolidinediones, which can either directly or indirectly promote insulin secretion or enhance insulin's effects, potentially leading to nonspecific glucose/FDG absorption by tissues¹⁰. Metformin, one of the first-line hypoglycemic drugs for type 2 diabetic patients¹¹, can increase glucose/FDG uptake in tissues, especially in the intestine, necessitating discontinuation 48 h before the examination¹². Fasting alone can also enhance the sensitivity of ¹⁸F-FDG PET examination, but prolonged fasting time inconveniences patients^{7,13,14}.

There are two major types of glucose carrier proteins within the body, namely facilitative glucose transporters (GLUTs) and active sodium-glucose cotransporters (SGLTs). Currently, sodium-glucose cotransporter 2 (SGLT2) inhibitors, such as dapagliflozin, are widely used to lower blood glucose levels in diabetic patients, particularly for elderly patients^{11,15}, who may have difficulty tolerating extended fasting preparation. This suggests that SGLT2 inhibitors could serve as a potential preparation intervention for ¹⁸F-FDG PET/CT examination. Unlike the aforementioned hypoglycemic drugs, the hypoglycemic effect of SGLT2 inhibitors is closely linked to the glomerular filtration rate, leading to the elimination of glucose from the body through the urinary system¹⁶, a pattern that aligns with the excretion of ¹⁸F-FDG¹¹. Studies have demonstrated that SGLT2 inhibitors effectively reduce the blood glucose levels in diabetic patients without significantly impacting the glucose uptake of tissues, and they have no significant effect on tissue insulin sensitivity^{11,15}. Malignant tumor cells predominantly utilize overexpressed glucose transporters, especially GLUT1, GLUT3, and SGLT2, for glucose uptake^{17–19}. However, the uptake of ¹⁸F-FDG is nearly unaffected by SGLT2 inhibitors because ¹⁸F-FDG is primarily a substrate of GLUTs and exhibits lower affinity for SGLTs^{11,15,20}.

It has been demonstrated that B16 cells (mouse melanoma cells) can be used to accurately estimate tumor volume in ¹⁸F-FDG PET imaging²¹. Melanoma is known to express GLUT1 and GLUT3 transporters²². Informatic analysis (<http://gepia.cancer-pku.cn/detail.php>) of SGLT2 gene expression in melanoma revealed that, in line with findings in most malignancies, there are no significant alterations in the expression.

Given the prevailing challenges in clinical ¹⁸F-FDG-PET imaging for patients with diabetes, this study aimed to compare the effects of SGLT2 inhibitors and insulin, and to provide clues for diabetic patients to choose appropriate hypoglycemic drugs in ¹⁸F-FDG PET imaging diagnosis.

Materials and methods

Animals

In this study, male C57BL/6J mice (20–22 g) were used. The animals were purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd. (Beijing, China). Mice were housed in environmentally controlled conditions with a 12-h light/dark cycle at temperature of 22 ± 3 °C and humidity of 55 ± 5%. Mice were given free access to food and water for seven days before the experiment. This study and all procedures using animals were in accordance with ARRIVE guidelines²³ and approved by the Animal Care and Use Committee of the Institute of Materia Medica, Chinese Academy of Medical Sciences. All the methods were performed in accordance with the relevant Ethical guidelines and regulations (Ethical committee approval No.00009471).

Cell culture and viability detection

B16 cell line (mouse melanoma cells) was purchased from FuHeng Biology Co., Ltd. (Shanghai, China). The cells were cultured in high glucose (4500 mg/L) Dulbecco's modified Eagle's medium (DMEM) (Thermo) supplemented with 10% FBS at 37 °C in 5% CO₂ atmosphere.

STZ-induced diabetic mouse modeling

After fasting for 16 h, diabetes was induced by intraperitoneal injections of streptozotocin (STZ, InnoChem Science & Technology Co., Ltd., Beijing, China) at a dose of 120 mg/kg. Control mice received an equal volume of the vehicle only (citrate buffer, pH 4.5). Seven days after the administration of STZ, 4-h fasting blood glucose levels were measured with an ACCU-CHEK® Active glucometer (Roche, Hoffmann, Germany). The mice whose fasting blood-glucose levels exceeded 11 mmol/L were chosen as the diabetic mouse models.

Inoculation for tumor xenografts in mice

The xenograft tumor model in mice was established following the procedures reported previously²⁴. In brief, after the tumor cells were digested with trypsin and harvested, the cells were suspended in phosphate buffer

solution (PBS) (pH 7.2) and kept on ice. Normal or diabetic modeling mice, which were 12-week-old male C57BL/6 mice, were routinely disinfected with 75% ethanol on the right axillary skin, and 0.1 mL of tumor cell suspensions (containing 1.5×10^6 cells) were injected under the axilla, while the control mice were injected with the equal volume of PBS. The body weights of mice and the sizes of tumor grafts were recorded regularly. The tumors were measured using calipers, and the volumes were calculated as $[0.52 \times (\text{length} \times \text{width}^2)]$. When the tumor grew up to 1000 mm³ in size, the animals were subjected to microPET scan and ¹⁸F-FDG biodistribution.

Animal groups and drug treatments

After the modeling of tumors combined with diabetes in mice, 4-h fasting blood glucose levels were tested. The tumor combined with diabetes mice were divided into three equal groups (n = 10) according to blood-glucose levels and body weights, including the diabetic tumor control group, dapagliflozin intragastric administration group (30 mg/kg, IG), and insulin intraperitoneal administration group (0.75 U/kg, IP). Euglycemic mice inoculated with tumor xenografts were used as the tumor control mice. Dapagliflozin was purchased from Shanghai Yuanye Bio-Technology Co., Ltd. (Cas 461432–26-8, Shanghai, China), and insulin for injection was purchased from Wanbang Biopharmaceuticals Co., Ltd. (Xuzhou, Jiangsu, China).

¹⁸F-FDG biodistribution detection in mice

Mice were fasted for 2 h, and mice in the administration groups were given drugs by the corresponding routes. One hour later (fasting for a total of three hours), 2-[¹⁸F]-FDG (radiochemical purity > 95%, atom Hi-Tech Co., Ltd., Beijing, China,) was injected into the tail vein at a dose of 9.25 MBq/10 g body weight (0.25 mCi/10 g). After ¹⁸F-FDG was distributed in conscious mice for one hour, the mice were anesthetized with isoflurane, and the blood, tumor, and tissues were collected. Gamma counting was performed following tissue weighing. After attenuation correction, the percentage of radiation uptake per gram of tissue (%ID/g) was calculated for each tissue and tumor.

¹⁸F-FDG microPET scan in mice

Mice were imaged using a Mosaic HP Small Animal PET imager (Philips Medical Systems, Inc., Cleveland, Ohio, USA) for ¹⁸F-FDG PET imaging. The administration of drugs and 2-[¹⁸F]-FDG in mice was the same as for the biodistribution. ¹⁸F-FDG was distributed in the mice for 1 h while awake. To prevent movement contamination and confounding the result with ¹⁸F-FDG uptake in muscles, the anesthetized animals used here for biodistribution and imaging detection²⁵. A mixture of oxygen (95%) and isoflurane (5% for induction and $\leq 2\%$ for maintenance) was used to anesthetize the mice. Then, the mice were gently fixed with the prone position on an imaging bed and subjected to six-minute static PET scanning. The images were scanned, corrected and reconstructed using the ordered subset expectation maximization reconstruction algorithm (ESEM2D) of the MOSAIC HP PET system. Data analysis was performed according to the region of interest (ROI) drawn on the tumor and tissue. uWS-MI software (United Imaging Healthcare Co., Ltd, Shanghai, China) was used for image display and volume of interest (VOI) analysis. The displayed values were the pixel values. For tumors, the mean% and max% (the single largest pixel value in the ROI) were reported to avoid bias in the ROI by including any necrotic tumor areas.

Statistical analysis

The differences between the groups were tested using unpaired 2-tailed Student's *t* tests or ANOVAs with Tukey's post hoc analysis, as appropriate. Data were analyzed using GraphPad Prism (GraphPad Software, USA). Data were presented as the means \pm SEM unless otherwise stated. The significance level was set at $P \leq 0.05$.

Ethical approval

This study and all procedures using animals were approved by the Animal Care and Use Committee of the Institute of Materia Medica, Chinese Academy of Medical Sciences (No. 00009471).

Results

Both the dapagliflozin and insulin can effectively reduce blood glucose in T1D mice

To clarify the time effect of dapagliflozin and insulin on blood glucose in diabetic animals, dapagliflozin (30 mg/kg, IG) and rapid-acting insulin (0.75 U/kg, IP) were administered to diabetic animals. The results showed that within 2 h after administration, the glucose-lowering effects of both drugs were comparable. However, after three hours, dapagliflozin continued to reduce blood glucose, which was significantly stronger than that of insulin. The optimal hypoglycemic effects were achieved when drugs were administered after 1 h (blood-glucose level below 10 mmol/L) (Fig. 1a). Other studies have found that one hour after the rats⁹ or diabetic patients^{26,27} were given insulin, ¹⁸F-FDG injection imaging shows a good contrast between tumors and tissues. In present study, dapagliflozin and insulin were administered one hours before ¹⁸F-FDG injection, and biodistribution were detected another one hour later via gamma counting and microPET imaging. The experimental process is shown in Fig. 1b. Both drugs were effective in lowering blood glucose to comparable levels before ¹⁸F-FDG injection (Fig. 1c).

Effects of drugs on ¹⁸F-FDG uptake in tumor and reference tissues of diabetic mice

This study revealed that diabetes significantly decreased ¹⁸F-FDG uptake in tumor tissue compared to euglycemic state (Fig. 2a). Diabetic animals exhibited elevated levels of ¹⁸F-FDG in reference tissues when compared to non-diabetic animals, including blood (Fig. 2b), skeletal muscle (Fig. 2c), liver (Fig. 2d), and kidney (Fig. 2e). Dapagliflozin did not significantly affect ¹⁸F-FDG uptake in diabetic tumor tissues (Fig. 2a), but prominently

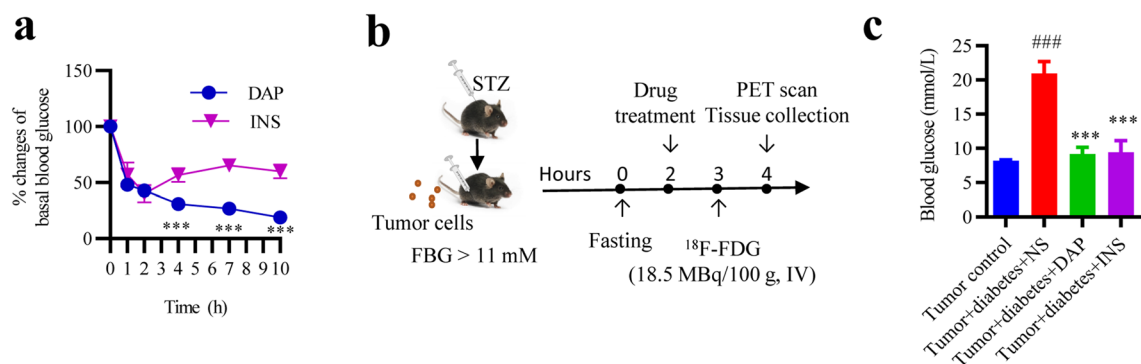


Figure 1. Effects of dapagliflozin and insulin on blood glucose levels in diabetic mice. **(a)** Comparison of the hypoglycemic effects between dapagliflozin and insulin. **(b)** Strategy for studying of dapagliflozin on ^{18}F -FDG PET imaging in diabetic mice with tumors. **(c)** Blood glucose levels of each group. Data represent the means \pm SEMs, $n = 10$; biological replicates. $###P < 0.001$ compared with tumor control, $***P < 0.001$ compared with tumor diabetic control or indicated group. Statistical analysis was performed by unpaired 2-tailed Student's *t* test **(a)** or one-way ANOVA followed by Fisher's LSD test **(c)**.

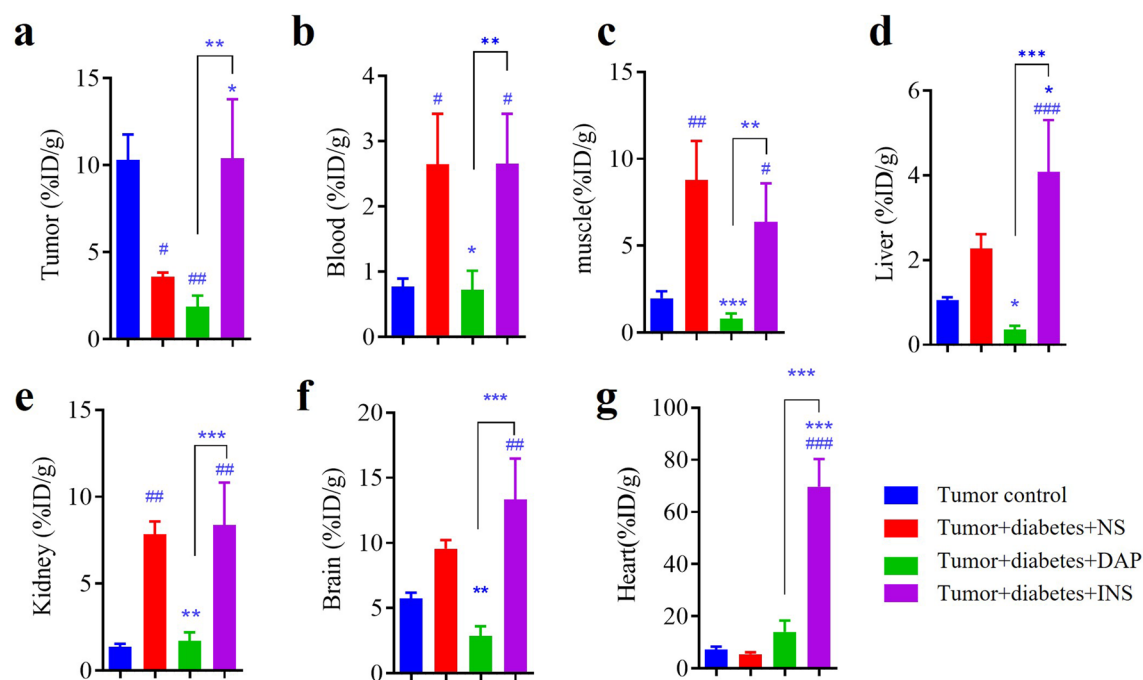


Figure 2. Effects of drugs on ^{18}F -FDG uptakes in diabetic mice based on gamma counting. **(a)** Tumor tissue. **(b)** Blood. **(c)** Skeletal muscle. **(d)** Liver. **(e)** Kidney. **(f)** Brain. **(g)** Heart. Data represent the means \pm SEMs, $n = 10$; biological replicates. $^{\#}P < 0.05$, $^{\#\#}P < 0.01$, $^{\#\#\#}P < 0.001$ compared with tumor control, $*P < 0.05$, $**P < 0.01$, $***P < 0.001$ compared with tumor diabetes control or the indicated group. Statistical analysis was performed by one-way ANOVA followed by Fisher's LSD test.

reduced ^{18}F -FDG levels in blood (Fig. 2b), skeletal muscle (Fig. 2c), liver (Fig. 2d), kidney (Fig. 2e), as well as brain (Fig. 2f). Insulin administration significantly increased ^{18}F -FDG uptake in tumor tissue (Fig. 2a), liver (Fig. 2d), and heart (Fig. 2g) in diabetic animals but did not have a significant impact on ^{18}F -FDG uptake in blood (Fig. 2b), skeletal muscle (Fig. 2c), kidneys (Fig. 2e), and brain (Fig. 2f). Surprisingly, insulin increased the uptakes of ^{18}F -FDG in tumors in diabetic animals, which was a result markedly different from the effects of dapagliflozin administration (Fig. 2a).

Biodistribution detection results of the ^{18}F -FDG uptake ratios between tumor and reference tissues

This study demonstrated the ^{18}F -FDG tumor-to-tissue uptake ratios were ranked in descending order as blood, liver, skeletal muscle, kidney, brain, and heart under euglycemic condition (Fig. 3a–g). In diabetic conditions, ^{18}F -FDG tumor-to-tissue uptake ratios decreased, meanwhile the order remained similar to that of euglycemic

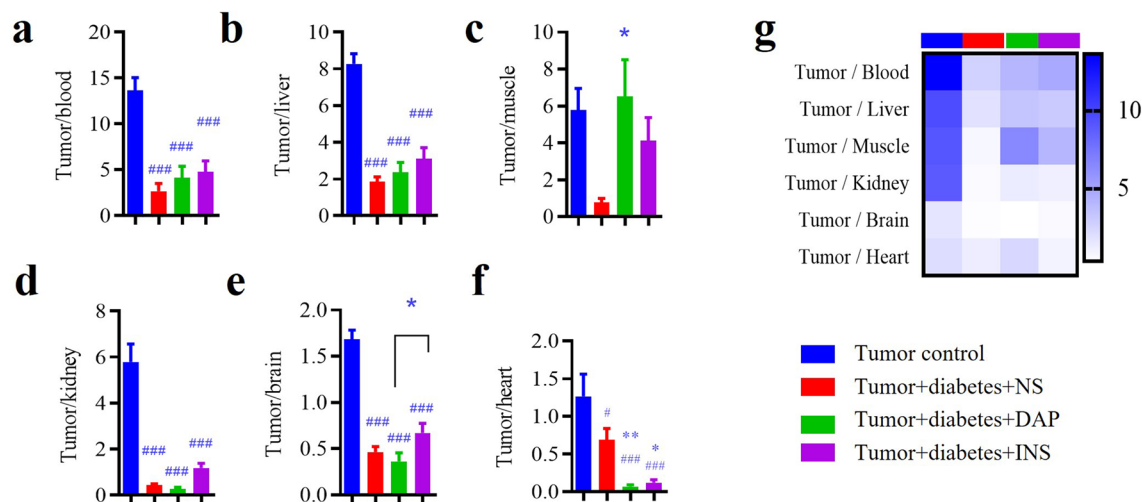


Figure 3. Effect of drugs on tumor-to tissue ratios of ^{18}F -FDG uptake by gamma counting. (a) Tumor to blood. (b) Tumor to liver. (c) Tumor to skeletal muscle. (d) Tumor to kidney. (e) Tumor to brain. (f) Tumor to heart. (g) Heatmap of the ^{18}F -FDG uptake ratios among groups. Data represent the means \pm SEMs, $n = 10$; biological replicates. $###P < 0.001$ compared with tumor control, $*P < 0.05$, $**P < 0.01$ compared with tumor diabetes control or the indicated group. Statistics analyzed by one-way ANOVA followed by Fisher's LSD test.

mice (Fig. 3g). Dapagliflozin administration apparently increased the tumor-to-skeletal muscle ratio (Fig. 3c), which was comparable to that of normal diabetic animals. The ratios in dapagliflozin-administered group were ranked from high to low as skeletal muscle, blood, and liver (Fig. 3g), while the order of ratios was blood, skeletal muscle, and liver in insulin treated group (Fig. 3g). Insulin administration did not significantly increase ^{18}F -FDG uptake ratios of tumor to reference tissues in diabetic mice (Fig. 3a–f).

Effects of the two drugs on ^{18}F -FDG PET imaging in diabetic mice

The results of PET semiquantitative analysis manifested diabetic condition played an important role in increasing ^{18}F -FDG imaging pixels in various reference tissues (Fig. 4a). The semiquantitative analysis results of PET showed similar ranking of tumor-to-tissue ratios among different groups as observed in biodistribution detection (Fig. 4b). Dapagliflozin increased the ^{18}F -FDG uptake imaging contrast of tumor to skeletal muscle of diabetic animals (Fig. 4c). Considering the possibility of central necrosis in tumor that could affect the results, the ratios of tumor mean% pixel and max% pixel to reference tissues were calculated (Fig. 4b, 4c). Comparatively, the results of max% pixels aligned more closely with the findings of biodistribution assay.

Discussion

This study is the first to discover the SGLT2 inhibitor can effectively reduce ^{18}F -FDG uptake in skeletal muscle and significantly increase the tumor-to-skeletal muscle ratio in diabetic mice compared with insulin.

The present work found that the FDG uptake in tumor tissues of diabetic mice decreased, but in reference tissues increased, which is consistent with the results previously reported⁹. In addition, we found that the SGLT inhibitor had no obvious effect on ^{18}F -FDG level in tumor tissue, which is in agreement with a previous report that SGLT1/2 has little influence on the transfer of ^{18}F -FDG to tumor cells¹¹.

Compared to diabetes controls, we also discovered that insulin promoted ^{18}F -FDG uptake in B16 tumor tissue. Although insulin can increase glucose uptake in various tissues¹, the effect of insulin on ^{18}F -FDG uptake in tumor tissue under diabetic conditions remains to be elucidated. An in vitro study showed there was no significant change of ^{18}F -FDG uptake in human adenocarcinoma cells with chronic hyperglycemia (300 mg/dL), while acute hyperglycemia markedly reduced ^{18}F -FDG uptake²⁸. Studies have found in the mammary carcinoma of STZ-induced diabetic rats, after 1 h of insulin administration, insulin didn't affect the FDG uptake of tumor, the uptake ratio of tumor to liver and blood increased, but to skeletal muscle decreased⁹. One study reported that insulin reduced FDG uptake in allogenic hepatoma cells (KDH-8) in euglycemic rats²⁹. Clinical studies show that insulin administration one hour before ^{18}F -FDG injection neither improves imaging quality²⁷ nor affect ^{18}F -FDG uptake in lung cancer tissue of diabetic patients²⁶. In present study, we found that the imaging of ^{18}F -FDG PET showed an improvement trend with insulin treatment, but without statistically significant differences. Insulin stimulates the up-regulation of GLUT4, resulting in the transfer of FDG distribution to peripheral muscle, soft tissue and fat, with a low ratio of tumor targeting to reference tissues uptake³⁰. Although insulin has no obvious effect on the expression of transcription factors other than GLUT4, it can induce the transfer of GLUT1³¹, GLUT3³² to cell membranes. Melanoma has also been reported to express GLUT4³³. Thus, we speculate that the increased uptake of insulin-promoted FDG in B16 melanoma may be related to the above factors, which requires further verification.

Generally, GLUTs are the main transporters for 2-FDG entry into tumor cells, yet, 2-FDG is also a substrate of SGLTs²⁰. Studies have reported that glucose uptake in some types of tumor cells is dependent on SGLTs³⁴. SGLT2 is expressed in the early development of lung cancer, specifically in precancerous lesions and well-differentiated

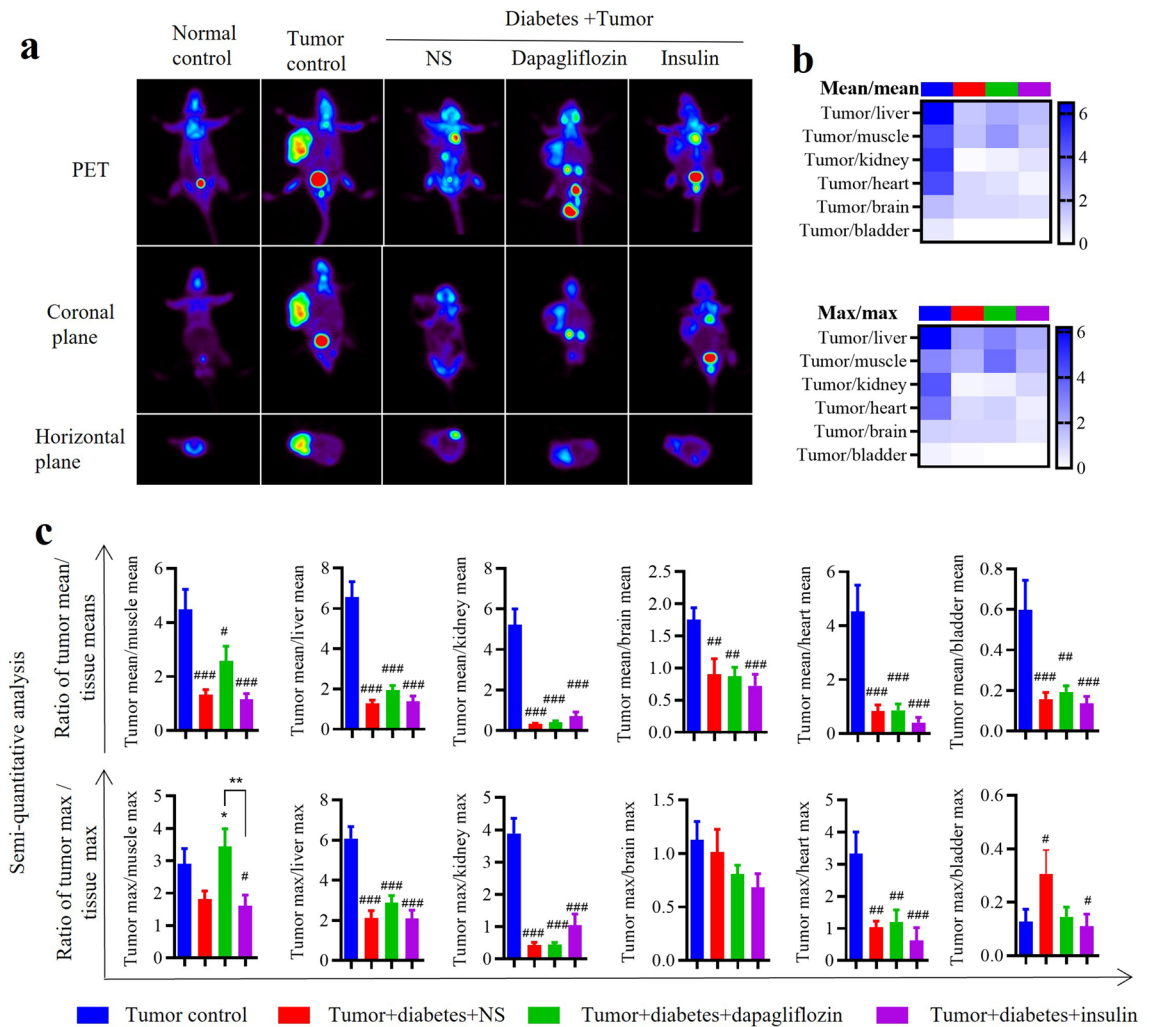


Figure 4. Effect of dapagliflozin and insulin on ¹⁸F-FDG PET imaging in diabetic mice. (a) Representative photos of ¹⁸F-FDG PET examination. (b) Heatmap of the ¹⁸F-FDG PET semiquantitative analysis results of tumor mean% to reference tissue mean% (upper), and tumor max% to reference tissue max% (lower). (c) Ratio of tumor mean% to reference tissue mean% and tumor max% to reference tissue max%. Data represent the means ± SEMs, n = 7–10; biological replicates. #P < 0.05, ##P < 0.01, ###P < 0.001 compared with tumor control, *P < 0.05 compared with tumor diabetes control or the indicated group. Statistical analysis was performed by one-way ANOVA followed by Fisher’s LSD test.

adenocarcinoma¹⁸. In cases of SGLT2 overexpression and lower GLUT1 expression, false-negative outcomes may occur in certain tumor metastases³⁵. Considering that SGLT2 gene expression levels are lower than GLUTs in most solid tumors³⁶, and ¹⁸F-2-FDG is not a specific substrate of SGLT2, we speculate the use of hypoglycemic SGLT2 inhibitors would be more beneficial in ¹⁸F-FDG PET diagnosis than insulin for most patients with hyperglycemia combined with tumors.

Importantly, our study found that when the SGLT2 inhibitor was used, the blood ¹⁸F-FDG levels significantly decreased compared with both the diabetic control and insulin groups, which has important implications for clinical application. Although SGLTs have far lower affinity for FDG than glucose³⁷, approximately 10% of FDG is reabsorbed by the proximal tubule of the kidney^{38,39}. In diabetic state, SGLT2 expression is elevated in the proximal tubule⁴⁰. Therefore, we infer the reasons for the lower ¹⁸F-FDG levels in reference tissues, including blood and tissues, may be due to the inhibition of SGLT2, thereby promoting its excretion, whereas insulin has no effect in this regard.

Excitingly, this study uncovered a novel effect, unlike the effect of insulin, SGLT2 inhibitors can dramatically reduce ¹⁸F-FDG uptake in diabetic skeletal muscle. Decreasing the ¹⁸F-FDG uptake of reference tissues under hyperglycemia is a crucial way to enhance the imaging contrast of tumor tissue for ¹⁸F-FDG PET detection. Previous studies have reported that SGLT2 inhibitors could effectively reduce the blood-glucose level of diabetic patients without significantly increasing the glucose uptake of tissues^{11,15}, yet our study showed that SGLT2 inhibitors decreased the glucose/FDG uptake in reference tissues, especially in skeletal muscle. It is noteworthy that the similar uptake in tumor tissues and reduced uptake in nontarget tissues also suggests that

SGLT2 inhibitors may be have beneficial for patient by reducing the radiation load to organs, and decreasing the harmful radiation toxicity.

The work concurrently has several limitations. One issue is that a study has shown diabetic state increases the expression of renal tubular SGLT2⁴⁰. Therefore, the impact of diabetes on the expression of SGLTs and GLUTs in tumors and reference tissues may affect ¹⁸F-FDG imaging and need to be clarified when SGLT2 inhibitors administered. Additionally, the influence of SGLT2 inhibitors on the half-life of ¹⁸F-FDG in the blood is also a consideration. Dapagliflozin reaches its peak within two hours after entering the bloodstream and has a half-life of 12.9 hours⁴¹. Different SGLT2 inhibitors also have different half-lives and hypoglycemic effects, so researches on the time and dose effect of SGLT2 inhibitors on ¹⁸F-FDG tissue uptake and elimination will further guide clinical applications. Furthermore, more clinical studies are needed to observe and validate whether the results of this study can be applied to humans.

Conclusion

This study is the first to investigate the impact of SGLT2 inhibitor on ¹⁸F-FDG PET imaging and provides a comparative analysis with the effects of insulin. In this study, we have unveiled that SGLT2 inhibitors exhibit the capacity to notably reduce ¹⁸F-FDG uptake in skeletal muscle compared with that of insulin under hyperglycemic condition, and enhance the tumor-to-skeletal muscle contrast. This study implies that SGLT2 inhibitors may be beneficial for ¹⁸F-FDG PET diagnostic in diabetic patients with skeletal muscle as the reference tissue. However, further clinical validation is still needed.

Data availability

The datasets used or analyzed during the current study are available from the corresponding author on reasonable request.

Received: 17 August 2023; Accepted: 16 October 2023

Published online: 26 October 2023

References

- Nakamoto, R. *et al.* Diffusely decreased liver uptake on FDG PET and cancer-associated cachexia with reduced survival. *Clin. Nucl. Med.* **44**(8), 634–642 (2019).
- Zaucha, J. M., Chauvie, S., Zaucha, R., Biggii, A. & Gallamini, A. The role of PET/CT in the modern treatment of Hodgkin lymphoma. *Cancer Treat. Rev.* **77**, 44–56 (2019).
- Hughes, D. J. *et al.* (18)F FDG PET/CT and novel molecular imaging for directing immunotherapy in cancer. *Radiology* **304**(2), 246–264 (2022).
- Boellaard, R. *et al.* FDG PET/CT: EANM procedure guidelines for tumour imaging: Version 2.0. *Eur. J. Nucl. Med. Mol. Imaging* **42**(2), 328–354 (2015).
- Eskian, M. *et al.* Effect of blood glucose level on standardized uptake value (SUV) in (18)F-FDG PET-scan: A systematic review and meta-analysis of 20,807 individual SUV measurements. *Eur. J. Nucl. Med. Mol. Imaging* **46**(1), 224–237 (2019).
- Diederichs, C. G., Staib, L., Glattig, G., Beger, H. G. & Reske, S. N. FDG PET: Elevated plasma glucose reduces both uptake and detection rate of pancreatic malignancies. *J. Nucl. Med.* **39**(6), 1030–1033 (1998).
- Cremerius, U. *et al.* Fasting improves discrimination of grade 1 and atypical or malignant meningioma in FDG-PET. *J. Nucl. Med.* **38**(1), 26–30 (1997).
- Roy, F. N., Beaulieu, S., Boucher, L., Bourdeau, I. & Cohade, C. Impact of intravenous insulin on 18F-FDG PET in diabetic cancer patients. *J. Nucl. Med.* **50**, 178–183. <https://doi.org/10.2967/jnumed.108.056283> (2009).
- Torizuka, T., Fisher, S. J., Brown, R. S. & Wahl, R. L. Effect of insulin on uptake of FDG by experimental mammary carcinoma in diabetic rats. *Radiology* **208**, 499–504. <https://doi.org/10.1148/radiology.208.2.9680582> (1998).
- Dahlén, A. D. *et al.* Trends in antidiabetic drug discovery: FDA approved drugs, new drugs in clinical trials and global sales. *Front. Pharmacol.* **12**, 807548 (2021).
- Wright, E. M. SGLT2 and cancer. *Pflugers Arch.* **472**(9), 1407–1414 (2020).
- Zhang, X. *et al.* Effect of metformin on (18)F-fluorodeoxyglucose uptake and positron emission tomographic imaging. *Br. J. Radiol.* **95**(1130), 20200810 (2022).
- Surasi, D. S., Bhambhani, P., Baldwin, J. A., Almodovar, S. E. & O'Malley, J. P. ¹⁸F-FDG PET and PET/CT patient preparation: A review of the literature. *J. Nucl. Med. Technol.* **42**(1), 5–13 (2014).
- Belohlavek, O. & Jaruskova, M. [¹⁸F]FDG-PET scan in patients with fasting hyperglycemia. *Q. J. Nucl. Med. Mol. Imaging* **60**(4), 404–412 (2016).
- Latva-Rasku, A. *et al.* The SGLT2 inhibitor dapagliflozin reduces liver fat but does not affect tissue insulin sensitivity: A randomized, double-blind, placebo-controlled study with 8-week treatment in type 2 diabetes patients. *Diabetes Care* **42**, 931–937. <https://doi.org/10.2337/dc18-1569> (2019).
- Rasul, S. *et al.* Response evaluation of SGLT2 inhibitor therapy in patients with type 2 diabetes mellitus using (18)F-FDG PET/MRI. *BMJ Open Diabetes Res. Care.* **8**(1), e001135 (2020).
- Macheda, M. L., Rogers, S. & Best, J. D. Molecular and cellular regulation of glucose transporter (GLUT) proteins in cancer. *J. Cell Physiol.* **202**(3), 654–662 (2005).
- Scafoglio, C. R. *et al.* Sodium-glucose transporter 2 is a diagnostic and therapeutic target for early-stage lung adenocarcinoma. *Sci. Transl. Med.* **10**(467), eaat5933 (2018).
- Pliszka, M. & Szablewski, L. Glucose transporters as a target for anticancer therapy. *Cancers* **13**(16), 4184 (2021).
- Barrio, J. R. *et al.* Does 2-FDG PET accurately reflect quantitative in vivo glucose utilization. *J. Nucl. Med.* **61**(6), 931–937 (2020).
- Puaux, A. L. *et al.* A comparison of imaging techniques to monitor tumor growth and cancer progression in living animals. *Int. J. Mol. Imaging* **2011**, 321538. <https://doi.org/10.1155/2011/321538> (2011).
- Parente, P. *et al.* Immunohistochemical expression of the glucose transporters Glut-1 and Glut-3 in human malignant melanomas and benign melanocytic lesions. *J. Exp. Clin. Cancer Res.* **27**, 34. <https://doi.org/10.1186/1756-9966-27-34> (2008).
- Percie du Sert, N. *et al.* Reporting animal research: Explanation and elaboration for the ARRIVE guidelines 2.0. *PLoS Biol.* **18**, e3000411. <https://doi.org/10.1371/journal.pbio.3000411> (2020).
- Kumar, M. *et al.* Longitudinal molecular imaging of progesterone receptor reveals early differential response to endocrine therapy in breast cancer with an activating ESR1 mutation. *J. Nucl. Med.* **62**(4), 500–506 (2021).

25. Karunanithi, S. *et al.* Spectrum of physiologic and pathologic skeletal muscle (18)F-FDG uptake on PET/CT. *AJR Am. J. Roentgenol.* **205**, W141–149. <https://doi.org/10.2214/AJR.14.13457> (2015).
26. Turcotte, E., Leblanc, M., Carpentier, A. & Bénard, F. Optimization of whole-body positron emission tomography imaging by using delayed 2-deoxy-2-[F-18]fluoro-D-glucose Injection following I.V. Insulin in diabetic patients. *Mol. Imaging Biol.* **8**, 348–354. <https://doi.org/10.1007/s11307-006-0064-1> (2006).
27. Song, H. S. *et al.* Ultrashort-acting insulin may improve on 18F-FDG PET/CT image quality in patients with uncontrolled diabetic mellitus. *Nucl. Med. Commun.* **34**, 527–532. <https://doi.org/10.1097/MNM.0b013e32836066a2> (2013).
28. Torizuka, T., Clavo, A. C. & Wahl, R. L. Effect of hyperglycemia on in vitro tumor uptake of tritiated FDG, thymidine, L-methionine and L-leucine. *J. Nucl. Med.* **38**, 382–386 (1997).
29. Zhao, S. *et al.* Effects of insulin and glucose loading on FDG uptake in experimental malignant tumours and inflammatory lesions. *Eur. J. Nucl. Med.* **28**, 730–735. <https://doi.org/10.1007/s002590100517> (2001).
30. Viglianti, B. L. *et al.* Effect of hyperglycemia on brain and liver (18)F-FDG standardized uptake value (FDG SUV) measured by quantitative positron emission tomography (PET) imaging. *Biomed. Pharmacother.* **88**, 1038–1045. <https://doi.org/10.1016/j.biopha.2017.01.166> (2017).
31. Cifuentes, M. *et al.* Insulin regulates GLUT1-mediated glucose transport in MG-63 human osteosarcoma cells. *J. Cell Physiol.* **226**(6), 1425–1432 (2011).
32. Simpson, I. A. *et al.* The facilitative glucose transporter GLUT3: 20 years of distinction. *Am. J. Physiol. Endocrinol. Metab.* **295**(2), E242–E253 (2008).
33. Zeng, Q. *et al.* Identification of TDP-43 as an oncogene in melanoma and its function during melanoma pathogenesis. *Cancer Biol Ther.* **18**, 8–15. <https://doi.org/10.1080/15384047.2016.1250984> (2017).
34. Scafoglio, C. *et al.* Functional expression of sodium-glucose transporters in cancer. *Proc. Natl. Acad. Sci. U S A* **112**(30), E4111–4119 (2015).
35. Taira, N. *et al.* Comparison of GLUT-1, SGLT-1, and SGLT-2 expression in false-negative and true-positive lymph nodes during the (18)F-FDG PET/CT mediastinal nodal staging of non-small cell lung cancer. *Lung Cancer* **123**, 30–35 (2018).
36. Madunić, I. V., Madunić, J., Breljak, D., Karaica, D. & Sabolić, I. Sodium-glucose cotransporters: New targets of cancer therapy. *Arh. Hig. Rada. Toksikol.* **69**(4), 278–285 (2018).
37. Qiao, H., Bai, J., Chen, Y. & Tian, J. Kidney modelling for FDG excretion with PET. *Int. J. Biomed. Imaging* **2007**, 63234 (2007).
38. Kobayashi, M. *et al.* Comparison of the transcellular transport of FDG and D-glucose by the kidney epithelial cell line, LLC-PK1. *Nucl. Med. Commun.* **31**(2), 141–146 (2010).
39. Rasul, S. *et al.* Response evaluation of SGLT2 inhibitor therapy in patients with type 2 diabetes mellitus using 18F-FDG PET/MRI. *BMJ Open Diabetes Res. Care* **8**(1), e001135 (2020).
40. Rahmoune, H. *et al.* Glucose transporters in human renal proximal tubular cells isolated from the urine of patients with non-insulin-dependent diabetes. *Diabetes* **54**(12), 3427–3434 (2005).
41. Dhillon, S. Dapagliflozin: A review in type 2 diabetes. *Drugs* **79**(10), 1135–1146 (2019).

Author contributions

Z.J. and Y.X. conceived of the study and participated in its design. L.M. and D.Y. participated in the experiments and drafted the manuscript and collected PET/CT images. L.M., D.Y. and L.Z. conducted the statistical analysis. L.S., Z.G., F.Y., Y.X., and Z.J. carried the methodology, review and editing the manuscript. All authors read and approved the final manuscript. All authors of the current manuscript meet the specified criteria for authorship and agreed to publish this manuscript.

Funding

Xiuying Yang has received grants from the CAMS Innovation Fund (2021-1-I2M-029), Beijing Municipal Natural Science Foundation (7222119). Jianhua Zhang has received grants from the Beijing TCM Science and Technology Foundation (JJ-2020-04), and Interdisciplinary clinical research project of Peking University First Hospital (2021CR32).

Competing interests

The authors declare no competing interests.

Additional information

Correspondence and requests for materials should be addressed to Y.X. or Z.J.

Reprints and permissions information is available at www.nature.com/reprints.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

© The Author(s) 2023