Effects of Anesthetic Agents and Fasting Duration on ^{18}F-FDG Biodistribution and Insulin Levels in Tumor-Bearing Mice

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Small-animal PET has opened the way for imaging ^{18}F-FDG uptake in murine tumor models, but the need for anesthesia raises concern over its potential influence on ^{18}F-FDG kinetics. We thus investigated such effects on cultured cells and on tumor-bearing mice after short- and long-term fasting. **Methods:** Lewis lung carcinoma (LLC) cells and cardiomyoblasts were treated for 2 h with a 100 μmol/L concentration of xylazine, ketamine, xylazine plus ketamine (Xy/Ke), or pentobarbital and were measured for ^{18}F-FDG uptake. LLC tumor–bearing C57BL6 mice that had been kept fasting for either 4 or 20 h were injected with Xy/Ke, pentobarbital, or saline and were administered 1.8 MBq of ^{18}F-FDG 15 min later. Biodistribution studies and plasma glucose and insulin assays were performed 45 min after injection. Separate anesthetized and control mice underwent ^{18}F-FDG PET. **Results:** ^{18}F-FDG uptake in LLC cells was unaffected by anesthetic agents, whereas xylazine and ketamine caused a small increase of uptake in cardiomyoblasts. In mice kept fasting 4 h, Xy/Ke induced a marked elevation of ^{18}F-FDG activity (percentage injected dose [%ID]) in blood (6.8 ± 0.9 %ID/g vs. 1.1 ± 0.6 %ID/g) and kidneys while decreasing myocardial uptake (2.3 ± 1.3 %ID/g vs. 4.7 ± 1.8 %ID/g). Target-to-blood ratios were significantly reduced. Pentobarbital caused a moderate increase in blood activity (2.5 ± 0.8 %ID/g), decreased myocardial uptake (2.8 ± 0.5 %ID/g), and reduced target-to-blood ratios. PET images of mice kept fasting 4 h were consistent with the biodistribution data. Insulin levels were lower with Xy/Ke and higher with pentobarbital. In mice kept fasting 20 h, Xy/Ke and pentobarbital increased blood ^{18}F-FDG activity (5.5 ± 2.2 and 4.9 ± 0.9 %ID/g vs. 2.4 ± 0.3 %ID/g) and reduced target-to-blood ratios, but these changes were substantially attenuated, compared with those in mice kept fasting 4 h. In addition, insulin levels were low and unaffected by anesthesia. **Conclusion:** Xy/Ke anesthesia markedly elevates blood ^{18}F-FDG activity and reduces tumor uptake ratios through inhibition of insulin release in mice kept fasting 4 h, whereas pentobarbital induces a similar but less severe response through insulin resistance. These metabolic effects, however, are substantially attenuated after 20 h of fasting. Hence both the choice of anesthetic and the duration of fasting have important effects on ^{18}F-FDG kinetics and PET images of tumor-bearing mice and should be considered when such studies are performed.

Key Words: anesthesia; ^{18}F-FDG, tumor; glucose; insulin

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An augmented demand for glucose use is a fundamental metabolic feature of tumor cells that is exploited by oncologic ^{18}F-FDG PET studies. The use of ^{18}F-FDG PET in cancer patients has now gained widespread acceptance for initial staging of disease, management of recurrent disease, and monitoring of therapeutic response (*1*). More recently, molecular imaging in small animals has emerged as a powerful technique for investigating disease processes at the molecular and cellular levels. As such, small-animal PET with ^{18}F-FDG provides a unique opportunity to advance our understanding of glucose metabolism in tumors of living mouse models (*2–4*). Imaging small animals requires anesthesia to prevent motion artifacts. This presents a potential problem for ^{18}F-FDG PET, because various anesthetic agents have been found to affect glucose turnover rates and alter its use in such tissues as the brain (*5–10*). These findings imply that anesthetic agents may also influence ^{18}F-FDG distribution and PET image results in animal tumor models, but this issue has not yet been elucidated. Moreover, important questions such as relative magnitudes of effect by different anesthetic agents, their potential mechanisms of effect, and the role of fasting duration have not been addressed. In this study, we thus investigated the effects of the widely used anesthetic agents xylazine plus ketamine (Xy/Ke) and pentobarbital on ^{18}F-FDG uptake in cultured cells and on ^{18}F-FDG biodistribution in tumor-bearing mice after a short- or long-term fast.

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MATERIALS AND METHODS

Cell Culture and ^{18}F-FDG Uptake Experiments

Lewis lung carcinoma (LLC) cells (established from lung cancer of a C57BL6 mouse) and H9C2 embryonic rat cardiomyoblasts were purchased from the American Type Culture Collection and maintained in Dulbecco's modified Eagle medium (Gibco BRL) supplemented with 10% fetal bovine serum, 2 mmol of glutamine per liter, 100 U of penicillin per milliliter, and 100 mg of streptomycin per liter at 37°C in 5% CO_2. Cardiomyoblasts were differentiated by being cultured in medium with 2% fetal bovine serum for 3 d, followed by incubation in 0.5% fetal bovine serum starting the day before the experiments. The experiments were performed on 70%–80% confluent cells in 12-well plates. The cells were treated with a 100 μmol/L concentration of xylazine, ketamine, Xy/Ke, or pentobarbital added to the culture medium for 2 h, followed by incubation with 1.8 MBq of ^{18}F-FDG at 37°C and 5% CO_2 for 45 min, then rapidly washed twice with phosphate-buffered saline and measured for radioactivity on a high-energy γ-counter (Wallac).

Animal Tumor Model Preparation

All animal procedures were in accordance with the National Institutes of Health *Guide for the Care and Use of Laboratory Animals* (*11*) and were approved by the Institutional Committee on Animal Care and Use. Tumors were grown in male C57BL6 mice by subcutaneous injection of 2×10^7 LLC tumor cells into the right flank. Biodistribution experiments were performed when tumor diameter reached approximately 2 cm.

Anesthesia

All experiments began in the late morning; food had been withheld from the animals since early morning (4 h of fasting) or the evening before (20 h of fasting). Water was allowed ad libitum. On the day of the experiment, tumor-bearing mice were given an intraperitoneal injection of Xy/Ke (9 and 110 mg/kg) or pentobarbital (50 mg/kg). Fifteen minutes later, the animals were checked for depth of anesthesia by a toe pinch to verify no response and then were injected with ^{18}F-FDG. Control animals were intraperitoneally injected with saline, injected with ^{18}F-FDG by immobilization in a restrainer, and placed in a quiet and dark cage until they were sacrificed.

^{18}F-FDG Biodistribution and Glucose and Insulin Measurements

A 1.8-MBq injection of ^{18}F-FDG mixed with isotonic saline to a total volume of 150 μL was administered through a tail vein. After the 45-min ^{18}F-FDG uptake period, the animals were sacrificed by cervical dislocation, and major organs were promptly extracted, washed, weighed, and measured for radioactivity on a γ-counter. Organ and tumor uptake levels were expressed as mean ± SD percentage injected dose (%ID) per gram of tissue. The plasma of each animal was assayed for glucose concentration using a dedicated glucose analyzer (2300 STAT Plus; YSI Life Sciences) and for insulin concentration using an immunoradiometric assay kit (INS-IRMA kit; BioSource).

^{18}F-FDG PET Imaging of Tumor-Bearing Mice

LLC tumor–bearing mice were intraperitoneally injected with Xy/Ke, pentobarbital, or saline as above. Fifteen minutes later, a 9.2-MBq injection of ^{18}F-FDG was administered through a tail vein with the mouse under anesthesia or, in the case of saline-injected controls, immobilized in a restrainer. Image acquisition started 45 min after injection and was performed on an Advance PET scanner (GE Healthcare) without attenuation correction. The control animals were sacrificed by cervical dislocation immediately before imaging to prevent motion.

Statistical Analysis

The significance of differences in ^{18}F-FDG uptake levels, organ-to-blood uptake ratios, and blood insulin levels between multiple groups was analyzed by 1-way ANOVA, with P values < 0.05 considered significant. The Bonferroni multiple-comparison test was performed as a post hoc test to determine the significance of the difference between any 2 groups.

RESULTS

^{18}F-FDG Uptake in Cultured Cells Treated with Anesthetic Agents

In cultured LLC tumor cells, none of the anesthetic agents tested affected ^{18}F-FDG uptake levels. In the cardiomyoblasts, minor increases of ^{18}F-FDG uptake were induced by xylazine, ketamine, and Xy/Ke (to 123% ± 1%, 117% ± 4%, and 128% ± 4%, respectively, of controls) but not by pentobarbital (Fig. 1).

Effect of Xy/Ke on ^{18}F-FDG Biodistribution in Mice Kept Fasting 4 h

The biodistribution of ^{18}F-FDG in LLC tumor–bearing C57BL6 mice is shown in Table 1. In mice kept fasting 4 h, the effect of Xy/Ke anesthesia was distinguished by a marked, 5.9-fold, increase of blood ^{18}F-FDG activity over control levels. Another prominent finding was a 7.9-fold increase of activity in the kidneys. Liver uptake was also

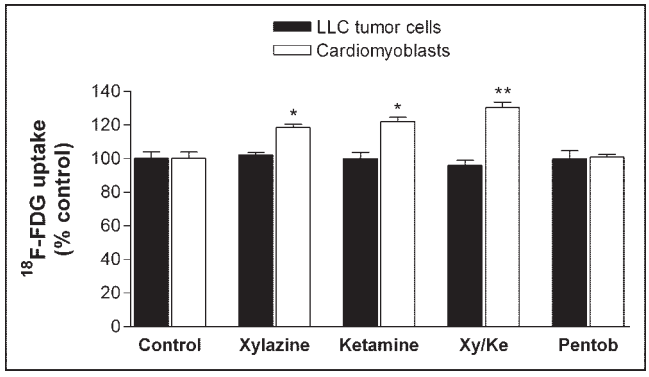

FIGURE 1. Effect of anesthetic agents on ^{18}F-FDG uptake in cultured LLC cells and H9C2 cardiomyoblasts. Uptake was measured after cells had been incubated for 45 min with ^{18}F-FDG and after a 2-h pretreatment with a 100 μmol/L concentration of xylazine, ketamine, Xy/Ke, or pentobarbital (Pentob). Data are mean ± SD of percentage uptake relative to control cells, obtained from triplicate samples of a single experiment representative of 2 separate experiments. *$P < 0.01$. **$P < 0.001$.

TABLE 1
^{18}F-FDG Biodistribution, Plasma Glucose, and Insulin Levels Obtained With or Without Anesthesia and After 4 or 20 Hours of Fasting in LLC-Bearing Mice

	4 h of Fasting			20 h of Fasting		
Tissue	Control (n = 7)	Xy/Ke (n = 7)	Pentobarbital (n = 6)	Control (n = 4)	Xy/Ke (n = 4)	Pentobarbital (n = 4)
Biodistribution						
Blood	1.15 ± 0.62	6.78 ± 0.91†	2.50 ± 0.84†	2.45 ± 0.32	5.51 ± 2.18†	4.95 ± 0.85†
Heart	4.71 ± 1.77	2.31 ± 1.27‡	2.76 ± 0.54†	2.94 ± 0.72	3.06 ± 0.66	2.41 ± 0.28
Lung	4.22 ± 1.83	4.65 ± 1.73	3.65 ± 0.67	8.17 ± 1.11	9.32 ± 0.61	8.62 ± 1.33
Liver	2.56 ± 3.05	6.61 ± 1.73‡	2.83 ± 1.10	2.93 ± 0.21	5.73 ± 2.24†	5.34 ± 1.05
Spleen	2.93 ± 0.85	4.33 ± 2.00	3.52 ± 0.77	7.89 ± 0.67	8.89 ± 1.33	8.59 ± 1.07
Kidney	1.54 ± 0.65	12.17 ± 4.96†	3.54 ± 1.58	3.04 ± 0.40	8.27 ± 3.29†	7.17 ± 1.84
Muscle	4.49 ± 2.40	1.08 ± 0.43‡	0.83 ± 0.15†	3.95 ± 4.04	1.19 ± 0.40	1.00 ± 0.24
Tumor	6.69 ± 2.91	5.63 ± 2.56	6.61 ± 1.28	23.61 ± 2.24	30.38 ± 2.57†	26.47 ± 3.32
Plasma glucose (g/mL)	230.1 ± 31.2	418.8 ± 113.5‡	195.9 ± 44.6	75.7 ± 8.0	75.3 ± 10.6	69.3 ± 12.3
Plasma insulin (μIU/ml)	29.6 ± 18.9	10.1 ± 0.9†	49.6 ± 29.5	8.1 ± 0.5	8.1 ± 0.5	6.7 ± 1.6

*P < 0.001 compared with controls.
†P < 0.05 compared with controls.
‡P < 0.01 compared with controls.
Biodistribution values are given as mean ± SD of %ID/g of tissue. P values are significance of difference compared with respective controls.

increased 3.8-fold, whereas myocardial uptake was reduced to less than half of control levels. Skeletal muscle uptake was higher in control animals, probably because they were able to move about after ^{18}F-FDG injection. Although Xy/Ke did not significantly affect absolute tumor uptake levels, high blood activity led to a substantial reduction in tumor-to-blood ratios (0.8 ± 0.3 vs. 6.7 ± 3.6) and myocardium-to-blood ratios (0.3 ± 0.2 vs. 4.3 ± 1.3; Fig. 2A).

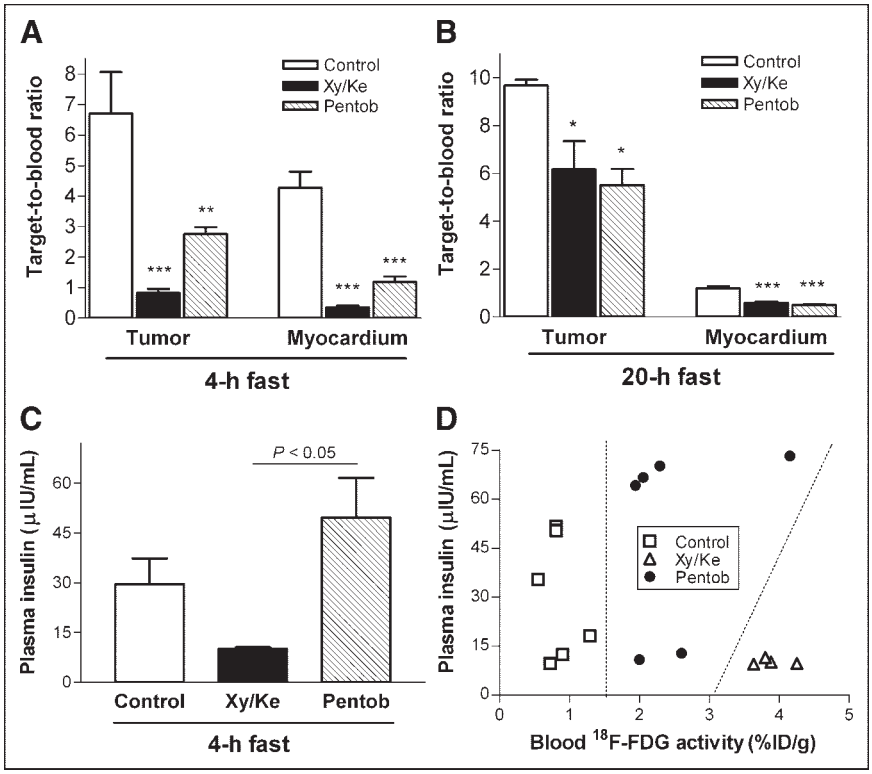

FIGURE 2. Effect of anesthetic agents on organ-to-blood ^{18}F-FDG ratio, insulin level, and blood ^{18}F-FDG activity. (A and B) Tumor- and myocardium-to-blood ratios of ^{18}F-FDG uptake in mice that had been kept fasting for 4 h (A) or 20 h (B). (C and D) Plasma insulin levels (μIU/mL) (C) and relationship to blood ^{18}F-FDG activity (D) in mice that had been kept fasting for 4 h (dotted lines were arbitrarily drawn to separate the 3 groups). All bar graphs are represented as mean ± SE. Pentob = pentobarbital. *P < 0.05. **P < 0.01. ***P < 0.001 compared with controls.

Effect of Pentobarbital on ^{18}F-FDG Biodistribution in Mice Kept Fasting 4 h

Pentobarbital anesthesia also induced a significant, 2.2-fold, elevation of blood ^{18}F-FDG activity over control levels, although the magnitude was not as pronounced as with Xy/Ke (Table 1). Myocardial uptake was significantly decreased. High blood activity in this group also led to a significant reduction of tumor-to-blood ratios (2.8 ± 0.6) and myocardium-to-blood ratios (1.2 ± 0.4; Fig. 2A).

^{18}F-FDG PET Imaging Results in Mice Kept Fasting 4 h

PET studies of mice kept fasting 4 h revealed images consistent with the biodistribution results. In the nonanesthetized mouse, the subcutaneous tumor and the myocardium were clearly visualized, with high target-to-background contrast. The Xy/Ke- and pentobarbital-anesthetized mice demonstrated significantly inferior tumor contrast, and visualization of the myocardium was poor (Fig. 3).

Effect of Anesthesia on Insulin Levels in Mice Kept Fasting 4 h

When we evaluated plasma insulin levels at the time of the biodistribution studies, mice kept fasting 4 h and anesthetized with Xy/Ke had substantially lower insulin levels than did pentobarbital-anesthetized mice (Table 1; Fig. 2C). On a scattergram display, the control group had low blood ^{18}F-FDG activity with variable plasma insulin levels, the Xy/Ke group had markedly increased blood ^{18}F-FDG activity with very low plasma insulin levels, and the pentobarbital group had moderately increased blood ^{18}F-FDG activity with plasma insulin levels that were high in 4 mice and low in 2 mice (Fig. 2D). Those 2 mice showed normal-range plasma glucose levels despite high blood ^{18}F-FDG activity, suggesting that plasma glucose and insulin levels had returned to normal before blood ^{18}F-FDG activity had.

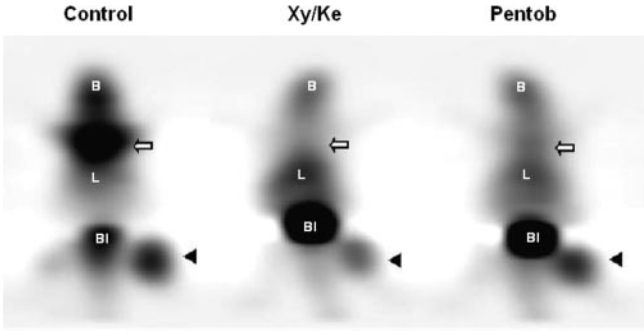

FIGURE 3. Coronal whole-body ^{18}F-FDG PET images of mice bearing subcutaneous Lewis lung tumors (arrowheads) in right flank and kept fasting 4 h. Images were obtained on clinical whole-body PET scanner rather than dedicated small-animal PET scanner. Arrow = heart; B = brain; Bl = urinary bladder; control = nonanesthetized mouse; L = liver; pentob = pentobarbital-anesthetized mouse; Xy/Ke = Xy/Ke-anesthetized mouse.

Effect of Anesthesia on ^{18}F-FDG Distribution and Insulin Levels in Mice Kept Fasting 20 h

In mice kept fasting 20 h, Xy/Ke elevated blood ^{18}F-FDG activity 2.2-fold over control levels and increased uptake in the liver and kidneys (Table 1). Interestingly, a mild increase in tumor ^{18}F-FDG uptake was seen in these mice. Pentobarbital injection elevated blood ^{18}F-FDG activity 2.0-fold over control levels but had no effect on tumor uptake (Table 1). Tumor- and myocardium-to-blood ratios were significantly reduced with both anesthetic agents, but less so than in mice kept fasting 4 h (Fig. 2B). Plasma insulin levels during distribution studies were similarly low in all mice kept fasting 20 h, regardless of whether they were administered Xy/Ke, pentobarbital, or saline (Table 1).

DISCUSSION

Xy/Ke and pentobarbital are the most widely used injectable agents to anesthetize rodents for imaging studies. The results of this investigation demonstrated that, in mice kept fasting 4 h, both Xy/Ke and pentobarbital anesthesia induced a substantial increase of blood ^{18}F-FDG activity. The manifest effects of anesthesia on blood glucose or ^{18}F-FDG levels are the result of a complex interaction of multiple mechanisms that include changes in endocrine secretion, hepatic and renal handling, and uptake by peripheral tissues. In our study, altered ^{18}F-FDG biodistribution by Xy/Ke and pentobarbital was accompanied by opposite directions of change in plasma insulin concentrations, indicating different mechanisms of effect.

Xylazine (2-(2,6-xylidino)-5,6-dihydro-4H-1,3-thiazine hydrochloride) is a clonidine analog with sedative, anesthetic, analgesic, and muscle relaxation effects. When ketamine is used, xylazine is routinely added to provide stable sedation and avoid muscle rigidity (*12*). Our results showed that Xy/Ke markedly increased blood ^{18}F-FDG activity and substantially reduced plasma insulin levels. These findings are consistent with the well-known property of xylazine of stimulating pancreatic α_2-adrenergic receptors to block insulin secretion and induce a strong hyperglycemic effect (*13,14*). Such an effect has been reported in various animal species including dogs (*15*), rats (*16*), and recently C57BL6 mice (*17*). Ketamine is a phencyclidine derivative used for its anesthetic effect of dissociating thalamic and limbic activity from neocortical function. Although increased blood glucose has been associated with the use of ketamine (*18*), whether it commonly induces hyperglycemia is less well established. Thus, the high blood ^{18}F-FDG activity and glucose concentration for Xy/Ke-injected animals in our study was most likely caused by the hypoinsulinemic effect of xylazine. Because elevated plasma glucose levels can decrease ^{18}F-FDG uptake by competition for transport and metabolism, it is likely that such a decrease contributed to the altered ^{18}F-FDG biodistribution that was observed.

Anesthesia with pentobarbital in mice kept fasting 4 h also led to a significant elevation of blood ^{18}F-FDG activity.

However, unlike the insulin-lowering effect of Xy/Ke, pentobarbital caused a prominent increase in plasma insulin levels. The effect of pentobarbital anesthesia on glucose use and glucose tolerance has previously been evaluated in rats by several investigators (*19–23*). Pénicaud et al. observed in pentobarbital-injected rats a transient elevation of blood glucose at 3 min that returned to basal levels by 40 min (*20*). Clark et al. (*21*) and Vera et al. (*22*) also found mild elevations of plasma glucose in rats injected with pentobarbital, whereas Johansen et al. did not (*23*). It may be that the observed levels of blood glucose after pentobarbital injection vary with the time of sampling because its induction of hyperglycemia is relatively mild and transient. In our study, increases in blood ^{18}F-FDG activity were not as marked with pentobarbital as with Xy/Ke. Moreover, glucose levels in the pentobarbital group were not elevated despite the significantly higher ^{18}F-FDG activity in the same blood samples. A possible reason may be that ^{18}F-FDG kinetics are not identical to glucose kinetics. Also contributing may be the fact that ^{18}F-FDG was administered 15 min after pentobarbital and had that much less time to return to normal levels than did glucose. The effect of pentobarbital on plasma insulin levels has also previously been studied in rats. Whereas Pénicaud et al. did not observe a significant effect (*20*), Vera et al. (*22*) and Johansen et al. (*23*) found a significant elevation of plasma insulin levels after pentobarbital anesthesia. The hyperinsulinemia induced by pentobarbital implies that its hyperglycemic effect is associated with an insulin-resistant state. Speculative mechanisms for this phenomenon include stimulated secretion of growth hormone, which can induce insulin resistance (*24*), and a direct inhibitory effect on glucose transporters (*25,26*).

In our study, the reduced tumor ^{18}F-FDG uptake ratios and PET image contrast caused by both Xy/Ke and pentobarbital anesthesia were due to high blood activity rather than to differences in absolute tumor uptake levels. The results of our in vitro uptake experiment also indicated no direct effect on ^{18}F-FDG uptake by tumor cells, although an effect of anesthetic agents on ^{18}F-FDG metabolism might have caused a difference in the relative contribution of unmetabolized ^{18}F-FDG to net ^{18}F-FDG uptake. On the other hand, Xy/Ke and pentobarbital both significantly decreased absolute myocardial ^{18}F-FDG uptake and caused poor visualization of the heart on PET images. A similar effect has previously been observed on ^{18}F-FDG small-animal PET images of Xy/Ke-anesthetized normal BALB/c mice, although the authors of that study did not obtain biodistribution data (*9*). No evidence suggests that xylazine or pentobarbital may directly interfere with glucose uptake by cardiomyocytes. This lack of interference is also supported by the results of our cultured cardiomyoblast experiments, in which no reduction of ^{18}F-FDG uptake by the anesthetic agents was observed. However, both Xy/Ke anesthesia (*27–30*) and pentobarbital anesthesia (*29,30*) are known to markedly depress heart rate and left ventricular ejection fraction in mice. Hence, the reduction of ^{18}F-FDG uptake in the myocardium of mice anesthetized by Xy/Ke or pentobarbital is most likely an indirect effect of depressed cardiac contractility.

For the Xy/Ke group, another prominent feature was a marked increase of ^{18}F-FDG activity in the kidneys, likely related to the high urinary bladder activity seen on the PET images. In rats, xylazine anesthesia is known to enhance urinary flow rate and urinary sodium excretion (*31*). This diuretic and natriuretic response is thought to result from the inhibitory effect of xylazine on vasopressin secretion and the direct vasopressin antagonistic action of xylazine on the distal nephron (*32,33*). Hence, the high renal ^{18}F-FDG activity caused by Xy/Ke anesthesia appears to be due to a combination of blood activity–elevating effect and diuretic effect. This high renal activity may be a disadvantage of Xy/Ke anesthesia for ^{18}F-FDG PET of small animals when the region of interest is near the kidneys or bladder.

A potential limitation of our PET experiment is that the control mouse had to be sacrificed immediately before PET acquisition to prevent motion whereas the anesthetized animals were imaged while alive. However, because the PET acquisition lasted only 5 min, the image result is not likely to have significantly been affected.

For ^{18}F-FDG PET studies of mice, food is generally withheld starting the morning of the experiment, usually resulting in fasting durations of approximately 3–4 h. However, to further reveal the impact of fasting duration on the effects of anesthetic agents on ^{18}F-FDG biodistribution, we extended our investigation to mice kept fasting 20 h. In these mice, we found that the elevating effects of both Xy/Ke and pentobarbital on blood ^{18}F-FDG activity were substantially attenuated, compared to the effects in mice kept fasting 4 h. Furthermore, the anesthetic agents did not affect the plasma insulin levels of mice kept fasting 20 h.

CONCLUSION

In conclusion, the present study demonstrated that in tumor-bearing mice kept fasting short-term, both Xy/Ke anesthesia and pentobarbital anesthesia resulted in a significant elevation of blood ^{18}F-FDG activity. The Xy/Ke-induced effects were greater, probably through the mediation of inhibited insulin secretion, and were accompanied by markedly increased renal uptake. Pentobarbital-induced effects were less severe than Xy/Ke-induced effects and appeared to be mediated through an insulin resistance–like state. The metabolic effects of these anesthetic agents, however, were minimized when mice had been kept fasting longer. Hence, both the choice of anesthetic and the duration of fasting are important factors that can affect biodistribution and image results in ^{18}F-FDG PET of tumor-bearing mice and should be considered in such studies.

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REFERENCES

1. Kostakoglu L, Agress H Jr, Goldsmith SJ. Clinical role of FDG PET in evaluation of cancer patients. *Radiographics.* 2003;23:315–340.
2. Cherry SR, Gambhir SS. Use of positron emission tomography in animal research. *ILAR J.* 2001;42:219–232.
3. Phelps ME. Positron emission tomography provides molecular imaging of biological processes. *Proc Natl Acad Sci U S A.* 2000;97:9226–9233.
4. Tatsumi M, Nakamoto Y, Traughber B, Marshall LT, Geschwind JF, Wahl RL. Initial experience in small animal tumor imaging with a clinical positron emission tomography/computed tomography scanner using 2-[F-18]fluoro-2-deoxy-D-glucose. *Cancer Res.* 2003;63:6252–6157.
5. Cavazzuti M, Porro CA, Biral GP, Benassi C, Barbieri GC. Ketamine effects on local cerebral blood flow and metabolism in the rat. *J Cereb Blood Flow Metab.* 1987;7:806–811.
6. Eintrei C, Sokoloff L, Smith CB. Effects of diazepam and ketamine administered individually or in combination on regional rates of glucose utilization in rat brain. *Br J Anaesth.* 1999;82:596–602.
7. Nelson SR, Howard RB, Cross RS, Samson F. Ketamine-induced changes in regional glucose utilization in the rat brain. *Anesthesiology.* 1980;52:330–334.
8. Saija A, Princi P, Pasquale RD, Costa G. Modifications of the permeability of the blood-brain barrier and local cerebral metabolism in pentobarbital- and ketamine-anesthetized rats. *Neuropharmacology.* 1989;28:997–1002.
9. Toyama H, Ichise M, Liow JS, et al. Evaluation of anesthesia effects on [^{18}F]FDG uptake in mouse brain and heart using small animal PET. *Nucl Med Biol.* 2004;31:251–256.
10. Toyama H, Ichise M, Liow JS, et al. Absolute quantification of regional cerebral glucose utilization in mice by ^{18}F-FDG small animal PET scanning and 2-^{14}C-DG autoradiography. *J Nucl Med.* 2004;45:1398–1405.
11. *Guide for the Care and Use of Laboratory Animals*. Washington, DC: National Academy Press; 1996.
12. Green CJ, Knight J, Precious S, Simpkin S. Ketamine alone and combined with diazepam or xylazine in laboratory animals: a 10 year experience. *Lab Anim.* 1981;15:163–170.
13. el Motal SMA, Sharp GWG. Inhibition of glucose-induced insulin release by xylazine. *Endocrinology.* 1985;116:2337–2340.
14. Muggaberg J. Effect of adrenergic drugs on glucose and plasma glucagons and insulin responses to xylazine in sheep. *Res Vet Sci.* 1982;33:118–120.
15. Ambrisko TD, Hikasa Y. Neurohormonal and metabolic effects of medetomidine compared with xylazine in beagle dogs. *Can J Vet Res.* 2002;66:42–49.
16. Amouzadeh HR, Sangiah S. Effects of streptozotocin-induced diabetes on xylazine-ketamine anesthesia. *Vet Hum Toxicol.* 1990;32:19–22.
17. Pomplun D, Mohlig M, Spranger J, Pfeiffer AF, Ristow M. Elevation of blood glucose following anaesthetic treatment in C57BL/6 mice. *Horm Metab Res.* 2004;36:67–69.
18. Otoide VO, Omueme C, Ojobo S. Elevated serum glucose levels following ketamine intravenous anaesthesia: a report of 2 cases. *Int J Obstetric Anesthesia.* 2001;10:206–208.
19. Davidson MB. Studies on the mechanism of pentobarbital-induced glucose intolerance. *Horm Metab Res.* 1971;3:243–247.
20. Pénicaud L, Ferré P, Kande J, Leturque A, Issad T, Girard J. Effect of anesthesia on glucose production and utilization in rats. *Am J Physiol.* 1987;252:E365–E369.
21. Clark PW, Jenkins AB, Kraegen EW. Pentobarbital reduces basal liver glucose output and its insulin suppression in rats. *Am J Physiol.* 1990;258:E709–E707.
22. Vera ER, Battell ML, Bhanot S, McNeill JH. Effects of age and anesthetic on plasma glucose and insulin levels and insulin sensitivity in spontaneously hypertensive and Wistar rats. *Can J Physiol Pharmacol.* 2002;80:962–970.
23. Johansen O, Vaaler S, Jorde R, Reikeras O. Increased plasma glucose levels after Hypnorm anaesthesia, but not after pentobarbital anaesthesia in rats. *Lab Anim.* 1994;28:244–248.
24. Dominici FP, Turyn D. Growth hormone-induced alterations in the insulin-signaling system. *Exp Biol Med.* 2002;227:149–157.
25. Honkanen RA, McBath H, Kushmerick C, et al. Barbiturates inhibit hexose transport in cultured mammalian cells and human erythrocytes and interact directly with purified GLUT-1. *Biochemistry.* 1995;34:535–544.
26. Haspel HC, Stephenson KN, Davies-Hill T, et al. Effects of barbiturates on facilitative glucose transporters are pharmacologically specific and isoform selective. *J Membrane Biol.* 1999;169:45–52.
27. Roth DM, Swaney JS, Dalton ND, Gilpin EA, Ross J Jr. Impact of anesthesia on cardiac function during echocardiography in mice. *Am J Physiol Heart Circ Physiol.* 2002;282:H2134–H2140.
28. Hart CY, Burnett JC Jr, Redfield MM. Effects of Avertin versus xylazine-ketamine anesthesia on cardiac function in normal mice. *Am J Physiol Heart Circ Physiol.* 2001;281:H1938–H1945.
29. Yang XP, Liu YH, Rhaleb NE, Kurihara N, Kim HE, Carretero OA. Echocardiographic assessment of cardiac function in conscious and anesthetized mice. *Am J Physiol.* 1999;277:H1967–H1974.
30. Rottman JN, Ni G, Khoo M, et al. Temporal changes in ventricular function assessed echocardiographically in conscious and anesthetized mice. *J Am Soc Echocardiogr.* 2003;16:1150–1157.
31. Cabral AM, Varner KJ, Kapusta DR. Renal excretory responses produced by central administration of opioid agonists in ketamine and xylazine-anesthetized rats. *J Pharmacol Exp Ther.* 1997;282:609–616.
32. Cabral AD, Kapusta DR, Kenigs VA, Varner KJ. Central alpha2-receptor mechanisms contribute to enhanced renal responses during ketamine-xylazine anesthesia. *Am J Physiol.* 1998;275:R1867–R1874.
33. Menegaz RG, Kapusta DR, Cabral AM. Role of intrarenal alpha(2)-adrenoceptors in the renal responses to xylazine in rats. *Am J Physiol Regul Integr Comp Physiol.* 2000;278:R1074–R1081.