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Dose-dependent, Saturable Occupancy of the Metabotropic Glutamate Subtype 5 Receptor by Fenobam as Measured with [¹¹C]ABP688 PET Imaging

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Abstract

Fenobam is a negative allosteric modulator of the metabotropic glutamate receptor subtype 5 (mGluR5) with inverse agonist activity and is expected to contribute to the treatment of neuropsychiatric disorders involving dysfunction of mGluR5 including Fragile × syndrome. This study examined whether [¹¹C]ABP688, an antagonist PET radioligand, competes with fenobam for the same binding site in the non-human primate brain and would allow examination of occupancy-plasma concentration relationships in the evaluation of the drug for target disorders in the human brain. Four paired PET studies with [¹¹C]ABP688 were performed in baboons at a baseline condition and after intravenous treatment with fenobam at different dose levels (0.3 - 1.33 mg/kg). Total distribution volume (V_T) and binding potential (BP_{ND}) using the cerebellum as a reference region were obtained by the plasma reference graphical method. Then it was examined whether occupancy follows a dose-dependent, saturating pattern that was predicted by a modified first-order Hill equation in individual regions. Baseline regional V_T and BP_{ND} values agreed with previously published data. Occupancy showed dose-dependent and saturating patterns in individual regions, reaching >90% occupancy at 1.33 mg/kg dose of fenobam in the majority of regions. To our knowledge, this is the first use of PET to characterize the mGluR5 therapeutic drug fenobam. This study demonstrates a proof of principle for determining the in vivo occupancy of fenobam in primates. The results indicate that [¹¹C]ABP688 and PET may be useful for examination of occupancy of mGluR5 by fenobam, which should prove to be useful for designing future studies and treatment of human disease states.

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Conflict of Interest. Dr. Fabrizio Gasparini is an employee of Novartis Pharma AG. The authors declare that they have no other conflict of interest.

Keywords

[¹¹C]ABP688; fenobam; glutamate; mGluR5; occupancy; PET

INTRODUCTION

Glutamate is considered to be the major excitatory neurotransmitter in the central nervous system, and balanced glutamatergic transmission is required for normal brain function (Pin et al., 1995). Glutamate receptors are divided into two classes, ionotropic and metabotropic. The metabotropic glutamate receptors (mGluRs) are a family of eight G-protein-coupled receptors and are divided into three groups based on their structure and function (Niswender et al., 2010). Group I mGluRs include the subtypes mGluR1 and mGluR5. These receptors are mainly expressed at postsynaptic neurons (Shigemoto et al., 1997) as well as glia cells (D'Ascenzo et al., 2007). Their role is to modulate excitatory transmission by stimulating the release of Ca²⁺ from intracellular stores and activation of *N*-methyl-D-aspartic acid (NMDA) receptors (Spooren et al., 2003; Pisani et al., 2001).

Much attention has been devoted to mGluR5 due to its possible involvement in a variety of disorders such as anxiety (Ballard et al., 2005), depression (Deschwenden et al., 2011), schizophrenia (Krystal et al., 2010), pain (Jacob et al., 2009), addiction (Olive et al., 2009), Parkinson's disease (Morin et al., 2010), and amyotrophic lateral sclerosis (Anneser et al., 2004). Recent findings have also shown a connection between mGluR5 and the most common inherited form of mental retardation and autism, Fragile × syndrome (Yan et al., 2005; Dölen et al., 2007). Several mGluR5 antagonists are currently in clinical trials for the treatment of Fragile × syndrome (Berry-Kravis et al., 2009; Levenga et al., 2011). One of these drugs, fenobam, was identified using rodent model for anxiety and previously investigated as an anxiolytic in a number of phase II studies in the early 1980s (Itil et al., 1978; Friedman et al., 1980; Pecknold et al., 1982). The molecular mechanism of action of fenobam was discovered 25 years later by Porter et al. (2005) and shown to be a negative allosteric modulator of mGluR5. A recent study showed that combined fenobam and amantadine treatment produced robust antidyskinetic effects in a primate model of Parkinson's disease (Ko et al., 2014).

The recent availability of mGluR5 PET radioligands has allowed in vivo investigations of the distribution and function of this receptor subtype (Hamill et al., 2005; Ametamy et al., 2006; Siméon et al., 2007). The highest expression of mGluR5 in the brain is in regions such as caudate/putamen, cortex, and hippocampus (Romano et al., 1995). Brain structures expressing this receptor are known to play important roles in emotion, motivation and motor control, leading to the hypothesis that mGluR5 may play a critical role in affective disorders, including anxiety, depression, and neurodegenerative diseases involving motor dysfunction (Spooren et al., 2003). It has been demonstrated that exaggerated signaling through mGluR5 can account for multiple cognitive and syndromic features of Fragile × syndrome (Dölen et al., 2008).

This finding combined with the potential availability of the phase II validated drug fenobam, has led to preliminary investigation of the efficacy of fenobam treatment for Fragile ×

syndrome (Berry-Kravis et al., 2009). Fenobam dosing in the aforementioned study ranged from 50 to 150 mg per adult subject and were selected based on doses in previous studies where CNS side effects were observed. One tool that would prove valuable for dosing determination is measurement of occupancy by PET. Therefore, we sought to determine the PET occupancy of mGluR5 by fenobam at various doses using [^{11}C]ABP688 for target engagement. A similar study using 3-((2-methyl-1,3-thiazol-4-yl)ethynyl)pyridine (MTEP) as a blocker was recently reported for one fixed dose by DeLorenzo et al. (2011a). A more recent study was conducted in humans using AZD2066 as a blocker (Kågedal et al., 2013). The aim of this study is to demonstrate that fenobam occupies the same receptor as [^{11}C]ABP688 in the non-human primate brain in a dose-dependent and saturable manner. Such a finding would indicate that [^{11}C]ABP688 and PET could be a powerful tool for examining occupancy-drug plasma concentration relationships in the human brain, and to understand occupancy levels by fenobam associated with effective dose levels in a number of neuropsychiatric disorders.

MATERIALS AND METHODS

Synthesis of [^{11}C]ABP688

[^{11}C]ABP688 was synthesized using a procedure previously described (Ametamey et al., 2007) with slight modification. Briefly, 1 mg of desmethyl-ABP688 in 200 μL of dimethylformamide was added to 0.5 mg of sodium hydride and reacted with [^{11}C]methyl iodide at 80°C for 5 min. The product was purified by semi-preparative high-performance liquid chromatography using a Phenomenex C18(2) Luna column (250 $\times$ 10 mm, 5 μm) and eluting with 60:40 acetonitrile:water (0.1M ammonium formate) at a flow rate of 10 mL/min. The collected fraction (t_R = 5.3 min) was diluted with 50 mL of water and loaded onto a Waters C18 Sep-Pak plus. The Sep-Pak was washed with 10 mL of normal saline and eluted with 1 mL of ethanol. The ethanol solution was diluted with 10 mL of saline and filtered through a 0.22 μm Millex-FG sterile filter. The average specific activity at end-of-synthesis was 500 ± 259 GBq/ μmol (13.5 ± 7.0 Ci/ μmol).

Preparation of animals and PET imaging

The experimental protocol was approved by the Animal Care and Use Committee of the Johns Hopkins Medical Institutions. PET experiments were performed on four male baboons (*Papio anubis*) on a High Resolution Research Tomograph with an axial resolution 2 mm (HRRT, CPS Innovations, Inc., Knoxville, TN). Each animal underwent one baseline scan and one or two blocking scans following pretreatment with fenobam at various dose levels on separate dates (Table 1). The animals were fasted for 12 hours prior to each PET study. Anesthesia was induced with intramuscular Ketamine (5-10 mg/kg) and maintained with a continuous intravenous infusion of Propofol at 0.4-0.6 mg/kg/min throughout the PET experiment. One venous catheter was inserted for the radioligand injection, and one arterial catheter was inserted to obtain arterial blood samples for radioactivity determination in plasma. The head of the animal was immobilized with a thermoplastic mask to reduce head motion inside the PET scanner. A transmission scan was acquired using a rotating Cs-137 source for attenuation correction. Then a dynamic PET acquisition was performed in a three-dimensional list mode for 90 min following an intravenous bolus injection over 30 s of

[¹¹C]ABP688 (Radioactivity range: 492-777 MBq (13.3-21.0 mCi); Cold mass range: 0.21-1.45 µg). For the blocking scans, fenobam was dissolved in 0.5 mL of ethanol made acidic with 20 µL of acetic acid and diluted with 4.5 mL of polyethylene glycol and 5.0 mL of normal saline. After sterile filtration (0.22 µm), the 10 mL of fenobam solution was given intravenously 5 min before the start of the second emission PET scan. In all scans, arterial blood samples were collected at very short intervals (< 5 s) initially and gradually prolonging intervals throughout the 90 min PET study for plasma radioactivity determination.

Analysis of radioactive metabolites in plasma

Selected samples taken at 0, 5, 10, 20, 30, 60, and 90 min were analyzed by high-performance liquid chromatography for the presence of [¹¹C]ABP688 and its radioactive metabolites using a column-switching technique (Hilton et al., 2000). Briefly, 3 mL of plasma in 8M urea acidified with 50 mg of citric acid was passed through a capture column (19 × 4.6 mm Strata-X, Phenomenex, Torrance, CA) at 2 mL/min followed by 1% acetonitrile in water to wash plasma proteins from the column. The effluent from the capture column, containing only highly polar components, passed quickly through a dual bismuth germanium oxide detector (Bioscan, Washington, DC). The solvent was then switched to 65% acetonitrile in 10 mM sodium phosphate buffer pH 5.5 (2 mL/min) for elution of the radiolabeled components bound to the capture column onto the analytical column (Synergi Polar-RP, 10 micron, 250 × 4.6mm, Phenomenex, Torrance, CA).

Reconstruction of PET images

Emission PET scans were reconstructed using the iterative ordered-subset expectation-maximization (OSEM) algorithm correcting for attenuation, scatter, and dead time. The radioactivity was corrected for physical decay to the injection time and re-binned to 35 dynamic PET frames of 256 (left-to-right) by 256 (nasion-to-inion) by 207 (neck-to-cranium) voxels. The frame schedules were, four 15-s, four 30-s, three 1-min, two 2-min, five 4-min, and twelve 5-min frames.

PET data analysis

Volumes of interest (VOIs)—VOIs that were previously prepared on individual animals' T1-weighted MRIs as described in our previous publication (Kuwabara et al., 2012) were used for this study and edited manually. VOIs were transferred to the baseline PET according to co-registration parameters given by SPM5 using the mutual information theory (West et al., 1997; Maes et al., 1997), and adjusted for radioactivity distribution of the baseline scan for each animal. The customized VOI template was transferred to the PET spaces of blocking scans using PET-to-PET co-registration parameters given by the co-registration module of SPM5 (Gitelman et al., 2003). Time activity curves (TACs) of regions were obtained by applying the VOIs onto the PET frames.

Derivation of PET outcome variables—Plasma reference graphical analysis (PRGA) (Logan et al., 1990) was used to obtain regional distribution volume (V_T), after confirming in a preliminary study an excellent agreement against the unconstrained two tissue compartmental model (TTCM-UC: =x) which was shown to be optimal for the baboon brain

($y = 1 \cdot x + 0.2$; $R^2 = 0.977$) (DeLorenzo et al. 2011a). In PRGA, PET frames up to 60 min was used with t^* (the start of asymptote) set at 10 min because a preliminary analysis showed stable estimates of V_T after 40 min ($V_{T40} = 0.96 \cdot V_{T60} - 0.23$; $R^2 = 0.995$, where subscripts 40 and 60 indicate the circulation times). Regional binding potentials (BP_{ND}) were obtained using cerebellum (Cb) as the reference region (i.e., region-to-Cb V_T ratio less 1) as previously recommended (DeLorenzo et al., 2011a; Ametamey et al., 2007), as well as after confirming negligible changes in Cb V_T between baseline and post-fenobam scans in this study.

Calculation of occupancy and ED₅₀—Occupancy of mGluR5 by fenobam was calculated using the percent change in BP_{ND} (baseline minus post-fenobam) as follows:

$$O_R = \Delta BP_{ND} / (\text{baseline } BP_{ND} - 1) \quad \dots (1)$$

A plot of occupancy versus fenobam dose in mg/kg, pooled for all baseline-blocking scan pairs was fitted by the following modified first-order Hill equation in each region:

$$O_R = D \cdot O_{\max} / (D + ED_{50}) \quad \dots (2)$$

where ED_{50} stands for the inhibition dose (ED_{50}) that caused 50% of the maximal occupancy ($O_{\max}$; fixed at 100%) and D for doses to individual animals

The pharmacokinetic plasma levels of fenobam were determined for one animal, B15070 at 0.90 mg/kg by analysis of the plasma sample by LC/MS/MS using atmospheric pressure chemical ionization.

RESULTS

Radioactive metabolites and plasma TACs

[¹¹C]ABP688 was rapidly metabolized after injection with total radioactive metabolites reaching 40% of total radioactivity at 3 min, 64% at 5 min, and 94% at 90 min. The time profiles of total radioactive metabolites did show differences between baseline and post-fenobam scans and appeared to have a dose-dependent relationship between fenobam dose and the rate of metabolism (Figure 1, Panel A).

Plasma time-activity curves, expressed in standard uptake value ($SUV = \text{radioactivity (mCi/L)} / (\text{injected radioactivity dose (mCi)} / \text{body weight (kg)}) \times 100\%$) are shown in Figure 1, panel B, as mean of four baseline scans. Total and metabolite corrected radioactivity peaked within one minute of the bolus injection, decreased sharply by 2 min, and decreased slowly thereafter, while total radioactive metabolite TACs decreased uneventfully after the peak.

Concentrations of fenobam in plasma following a bolus injection are shown in Table 2 as a function of time after the radioligand injection. The time-course remained relatively stable between 30 and 90 min post-radioligand injection.

Radioactivity images and tissue TACs

Figure 2 shows coronal and sagittal images of averaged PET frames for baseline and post-fenobam scans of B15070 who received 1.33 mg/kg of fenobam. On visual inspection, cingulate cortex, especially anterior cingulate cortex, caudate nucleus, putamen, and insula cortex showed the highest accumulation of radioactivity, while cerebellum showed the lowest accumulation across baseline scans. Tissue TACs for selected brain regions are shown in Figure 3. In baseline scans, the caudate nucleus showed the highest radioactivity, followed by putamen, cortical regions, and hippocampus, while the cerebellum showed the lowest radioactivity. Treatment with 1.33 mg/kg of fenobam inhibited the uptake of [^{11}C]ABP688 in all brain regions including some reduction in the cerebellum.

Regional V_T and BP_{ND}

Baseline regional V_T and BP_{ND} values of PRGA are shown in Figure 4. Anterior temporal region, anterior cingulate cortex, insula, putamen, and caudate nucleus showed high V_T and BP_{ND} values exceeding 1, as predicted by averaged radioactivity images, followed by posterior cingulate cortex and hippocampus. Other cortical regions and thalamus showed low V_T and BP_{ND} values while Cb showed the lowest V_T among examined regions and in the brain by visual inspection of radioactivity images. Although post-fenobam scans showed slightly lower V_T in Cb (3.7 ± 2.2 mL/mL, mean $\pm$ SD) than baseline scans (baseline: 4.6 ± 1.5 mL/mL), no statistical differences were noted ($t = 1.80$; $df = 3$; $p > 0.1$, paired t-test).

Occupancy of mGluR5 by fenobam

Plots of occupancy versus dose followed Equation 2, as exemplified for anterior cingulate cortex and putamen in Figure 5. AIC values ranged from 16.9 in anterior cingulate cortex (the best fit) to 24.1 in thalamus (the poorest fit among 12 brain regions examined). AIC values were lower for the saturation model (Equation 2) than a no-saturation model (occupancy = $\alpha \cdot \text{dose}$) ($t = -5.45$, $df = 11$; $p < 0.001$, paired t-test), suggesting that the saturation model fitted the data better than a no-saturation model. ED_{50} estimates ranged from 0.3 mg/kg (thalamus) to 1.1 mg/kg (parietal cortex).

DISCUSSION

Although other PET tracers such as [^{18}F]FPEB now exist, we used [^{11}C]ABP688 because of its availability and lack of uptake in the cerebellum. Regional V_T patterns and magnitudes for [^{11}C]ABP688 agreed with published data (DeLorenzo et al., 2011a; Ametamey et al., 2007), with caudate nucleus and putamen showing high V_T . Some methodology-oriented studies indicated that TTCM-UC yielded more robust estimates of V_T than TTCM-C for [^{11}C]ABP688 (Ametamey et al., 2007; Treyer et al., 2007). However, all methodology-intensive studies on [^{11}C]ABP688 to our knowledge claimed that BP_{ND} could be obtained as the (region-to-Cb V_T ratio) - 1 (DeLorenzo et al., 2011a; Burger et al., 2010) alone for [^{11}C]ABP688 due to unstable BP_{ND} estimates given by the k_3 - k_4 ratio as defined by Innis et al. (2007). Therefore, in order to claim that [^{11}C]ABP688 yields BP_{ND} , it is critical to confirm negligible specific binding in Cb. In support of this requirement, Elmenhorst et al. (2010) reported B_{max} values of 80 and 2300 fmol/mg tissue (about 30-fold differences) for Cb and putamen-caudate nucleus, respective in the rat brain using [^3H]ABP688. In addition,

this study showed no statistical differences in V_T of Cb between baseline and post-fenobam scans, albeit a $< 20\%$ decrease in mean value. In a study using another specific mGluR5 antagonist MTEP at a fixed dose of 1 mg/kg, Cb (gray matter only as in this study) showed a 7% (SD: 30.0%) decrease in V_T between baseline and blocking scans while other brain regions showed close to or more than 50% decreases in V_T (DeLorenzo et al., 2011a). Although no statistics were provided for the aforementioned study, it is not likely to have a statistically different decrease in V_T for Cb because of the relatively large SD. Lastly, mRNA for mGluR5 is reported to be negligible in Cb (Berthele et al., 1999). Therefore, it appeared to be appropriate to claim negligible displacements of [^{11}C]ABP688 binding in Cb for fenobam and MTEP blocking experiments, and that BP_{ND} is measurable with [^{11}C]ABP688 and PET. Regional BP_{ND} patterns were very similar to regional V_T patterns using the BP_{ND} calculation formula. In addition to caudate nucleus and putamen, we noted high BP_{ND} in anterior cingulate cortex, anterior temporal cortex, and insula as previously reported for baboons (DeLorenzo et al., 2011a) and humans (Ametamey et al., 2007; Treyer et al., 2008; DeLorenzo et al., 2011b) with [^{11}C]ABP688 and PET.

This study demonstrated that in vivo binding of [^{11}C]ABP688 was blocked by pre-treatment with fenobam in a dose-dependent, saturable manner, approaching close to full occupancy ($> 90\%$) at a dose of 1.33 mg/kg. Although the plasma concentration of fenobam was only measured for one study (0.90 mg/kg), the measured levels at later time points compared favorably with those observed during oral dosing in humans (Itil et al., 1978). It has been demonstrated that fenobam inhibits mGluR5 receptor activity via its actions as an inverse agonist (Porter et al., 2005). Therefore, the agreement of the observed occupancy-dose curve with the first-order (i.e., Hill coefficient = 1) Hill equation (Equation 2) suggested that [^{11}C]ABP688 and fenobam compete for one single site of mGluR5. MTEP, another mGluR5 antagonist exhibited relatively homogenous blockade of [^{11}C]ABP688 binding (72% - 88%) with a fixed dose (1 mg/kg) in the baboon brain (DeLorenzo et al., 2011a). In rodents, a dose-dependent, saturable blocking of [^{11}C]ABP688 binding that followed the same saturation equation was reported by pre-treatment with MPEP, a mGluR5 antagonist (Elmenhorst et al., 2010). Lastly, a dose-dependent, saturable blockade of [^3H]ABP688 was reported with CTEP, yet another highly specific mGluR5 antagonist in the mouse brain (Lindemann et al., 2011). Taken altogether, it is very likely that [^{11}C]ABP688 may be appropriate for drug occupancy studies of above mentioned drugs and their analogues targeting mGluR5. However, it should be noted that applications of [^{11}C]ABP688 on humans remain uncertain because a human test-retest study (DeLorenzo et al., 2011b) reported significant increases ($>15\%$) in BP_{ND} in retest scans in a same-day scanning design. However, [^{18}F]FPEB showed stable estimates when repeated a few days apart (Wong et al., 2013). At present, it is not clear whether the phenomenon of 'unstable V_T ' is specific to [^{11}C]ABP688. On the other hand, no specific bias toward test or retest scans were found in the baboon brain (DeLorenzo et al., 2011a). Thus, it is likely that the use of [^{11}C]ABP688 is justified for drug occupancy studies on baboons.

One limitation of this study is that it included only 4 pairs of baseline and post-fenobam scans. However, this number appeared appropriate for the purpose of demonstrating proof of principle for determining the in vivo occupancy of a mGluR5 therapeutic drug fenobam since the distribution of doses was relatively equally spaced, and near full blockade was

observed at the highest dose (1.33 mg/kg). Second, plasma concentrations of fenobam were measured in one study alone. Despite that occupancy-plasma drug concentration plots may be preferable as shown for other drug-receptor system combinations (Sparshatt et al., 2009; Bishara et al., 2013), our study showed acceptably good fit of the plot to the occupancy equation (Equation 2) compared to occupancy-dose plots of these meta-analysis articles. Thus, it is likely that occupancy-dose plots were sufficient for the purpose of demonstrating proof of principle with [^{11}C]ABP688. It was assumed that fenobam occupies mGluR5 in a stable manner throughout the duration of PET studies as commonly assumed in preclinical drug occupancy evaluations (e.g., Kuwabara et al., 2012). Lastly, $\text{Cb } V_T$ was used as the estimate of V_{ND} in this study while DeLorenzo et al. (2011a) proposed the use of the transposed Lassen plot (i.e., scatter plots V_T (= baseline - post-fenobam) versus baseline V_T of regions to yield V_{ND} as the x-intercept of the regression line (Lassen et al., 1995; Cunningham et al., 2010) to obtain a 'theoretically correct' V_{ND} . Although excellent correlations were observed at high dose experiments, uncertainties arose on estimates of V_{ND} (i.e., substantially different from $\text{Cb } V_T$) in low dose experiments because of the relatively flat arrangement of the plot (i.e., small V_T). For this reason, together with aforementioned line of evidence supporting negligible mGluR5 in Cb (gray matter) in the brain of non-human primate, the Lassen plot was not employed in this study.

CONCLUSIONS

This study indicates that [^{11}C]ABP688 may be appropriate for examination of the in vivo occupancy of the mGluR5 therapeutic drug fenobam. To our knowledge, this is the first use of PET to characterize fenobam, albeit preliminary. Existing literature has indicated that [^{11}C]ABP688 could be applicable to various candidate drugs targeting mGluR5. To be fully applicable to humans, the test-retest issues in the literature raise the question whether one [^{11}C]ABP688 scan influences subsequent [^{11}C]ABP688 scans in the human brain. Nevertheless, these baboon PET studies suggest that [^{11}C]ABP688 could be a promising PET radioligand for human occupancy studies.

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Fig. 1.

Time profiles of total radioactive metabolites in plasma (mean and SD bars) given by HPLC analysis, separately for baseline and post-Fenobam scans (panel A). Total and metabolite-corrected time-activity curves (TACs) in plasma, mean of 4 baseline scans (panel B). The inset shows TACs for early time points.

Fig. 2. PET images, averages of 10-90 min frames of [^{11}C]ABP688 scans, baseline (upper row) and post-fenobam (lower row; dose: 1.33 mg/kg) scans. This dose of fenobam showed almost complete occupancy of mGluR5s. Coronal view images (left column), showing high radioactivity accumulations in putamen (Pu), caudate nucleus (CN), insula (In), and cingulate cortex (Cg), while sagittal images (right column) about 4 mm from the mid-plane show high accumulations in anterior cingulate cortex (aCg).

Fig. 3. Time-activity curves (TACs) of selected brain regions following a bolus injection of $[^{11}\text{C}]\text{ABP688}$, average of 4 baseline scans (panel A) and one post-fenobam scan with 1.33 mg/kg dose (B), expressed in standardized uptake value (SUV) in percentage.

Fig. 4.

Histograms showing regional values of V_T given by an unconstrained two-tissue compartmental model (TTCM-UM) and the plasma reference graphical analysis (PRGA) (panel A), and regional values of BP_{ND} given by PRGA and the reference tissue graphical analysis (RTGA) (panel B). Regions are frontal (Fr), temporal (Tp), parietal (Pa), and occipital (Oc) lobes, anterior (aCg) and posterior (pCg) cingulate cortices, insula (In), hippocampus (Hp), putamen (Pu), caudate nucleus (CN), thalamus (Th), and cerebellum (Cb).

Fig. 5. Scatter plots of occupancy versus dose for the anterior cingulate cortex (panel A) and putamen (B). Dotted lines indicate model-predicted occupancy-dose curves using Equation 2.

Table 1

Fenobam doses for blocking scans

Baboon #	Fenobam dose (mg/kg)
17436	0.20
17436	0.67
15070	0.90
C18	1.33

Table 2

Plasma levels of Fenobam (0.90 mg/kg)

Time (min)	Amount (ng/mL)	Amount (nM)
5	1,116	4,184
15	772	2,896
30	654	2,453
60	479	1,798
90	345	1,292