BOLD fMRI of Anesthetized Baboons

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INTRODUCTION Non-human primates (NHPs) are important animal models because of their overall similarities to humans, resulting in better recapitulation of many human diseases compared to the more commonly used rodent models. Only a few BOLD fMRI studies of awake (*1-6*) and anesthetized NHP (*7, 8*) have been reported. Most of these studies were in rhesus and smaller primates such as squirrel monkeys. In this study, we present an anesthetized baboon model for BOLD fMRI studies of visual and somatosensory/motor stimulations with the long-term goals of applying this model to investigate stroke and epilepsy. The advantages of baboons are their larger brain size compared to rhesus and they are more commonly used in genetic, infectious disease and obesity studies. Toward optimizing the BOLD fMRI responses, we compared BOLD fMRI data obtained under two commonly used anesthetics (isoflurane and ketamine) with and without paralytics.

METHODS Four repeated BOLD fMRI sessions were performed on three normal female baboons (10-20kg). Animals were first studied under 0.8~1.0% isoflurane followed by ketamine (6-8mg/kg/hr) drips in the same fMRI session with and without paralytics (vecuronium 0.1mg/kg first dose, 0.02 mg/kg supplement dose as needed). Animal was positioned supine in an animal holder and mechanically ventilated. End-tidal CO2, O2 saturation, heart rate, respiration rate, and rectal temperature were monitored continuously and maintained within normal physiological ranges throughout the entire studies. At the end of the MRI experiments, neostigmine (0.5-2mg) was administered to reverse paralytic effects. Somatosensory/motor stimulation via a pneumatic stimulator was applied to the animal's right hand. Achromatic light flickering at 10 Hz delivered via fiber optics was applied to both eyes simultaneously. Stimulation paradigm used a block design of three 50s-on/off epochs.

MRI studies were performed on 3T Siemens TIM TRIO. BOLD fMRI was acquired using gradient echo-planar imaging with TR = 2500, TE = 30 ms, matrix = 64x64 or 100x100, field of view (FOV) = 12.8x12.8 or 15x15 cm, with a resolution of 2x2x5 or 1.5x1.5x4mm, and 10 slices. Typically, 2-4 repeated fMRI stimulation trials were measured in each session. Data were processed using FMRIB Software Library (FSL). Activation maps were threshold to Z>2.3 ($p<0.01$) and registered to a high-resolution anatomical template. BOLD percent changes were tabulated for region-of-interests the primary (S1), secondary (S2) somatosensory cortex, motor (M) and supplementary motor cortex (SMA), thalamus (Th), primary visual (V1), and lateral geniculate nucleus (LGN).

RESULTS & DISCUSSION All physiological parameters were maintained at normal physiological ranges. Isoflurane at > 1.2% abolished essentially all activations in all animals studied. With 0.8-1.0% isoflurane without paralytic, the animals remained adequately anesthetized as determined by the absence of pinching responses and normal heart rates. Activations in the expected brain regions were often detected but are often contaminated by signal drifts and large temporal fluctuations likely due to motion-related physiological noises. Consequently, activations were inconsistent in repeated fMRI trials within the same sessions as well as between animals. Similar findings were reported with ketamine at a minimal dose that adequately anesthetized the animals without paralytics.

In contrast, with vecuronium paralytics, time courses were very stable and BOLD activations were robust and consistent across multiple repeated fMRI trials in the same sessions and between animals. Vecuronium was effectively reversed paralysis and all animals were able to breathe unassisted within 5-10 mins after administration of neostigmine. **Figure 1A** shows the activation maps of simultaneous somatosensory/motor and visual stimulations under 0.8-1.0% isoflurane and paralytic. **Figure 1B** shows representative V1 and LGN BOLD time courses under isoflurane and ketamine with paralytic for the ROI shown in Figure 1A. Activations in S1, S2, M, SMA, Th, V1, and LGN were robustly detected. Frequencies of detectable activation, BOLD percent changes and Z-scores are summarized in **Table 1**. Isoflurane and ketamine yielded reliable fMRI responses with comparable sensitivity. Although BOLD percent changes were statistically different in SMA and Th, the average Z scores were not statistically different between two anesthetics in the ROI's analyzed.

CONCLUSIONS This study establishes a robust anesthetized baboon model for BOLD fMRI studies on a 3T human scanner. At the appropriate dosages, isoflurane or ketamine with paralytics yields reliable fMRI responses with comparable sensitivity. Future study will utilizes this model to map functional changes associated with stroke and epilepsy studies.

Table 1. Frequency of detection, averaged BOLD changes and averaged Z scores by ROI (mean ± SEM, n=9 for Iso, and n=6 for Ket). * or # $p< 0.05$

	Frequency of detection		BOLD % changes		Z scores	
	Iso	Ket	Iso	Ket	Iso	Ket
S1/S2	100%	100%	0.57±0.05	0.56±0.06	4.5±0.3	4.5±0.3
M	89%	83%	0.62±0.04	0.60±0.12	4.5±0.3	4.2±0.5
SMA	33%	50%	0.45±0.01 *	0.82±0.32 *	4.8±0.6	4.3±0.6
Th	33%	50%	0.89±0.31	0.67±0.38	4.8±0.7	4.2±0.6
V1	100%	83%	0.72±0.08#	0.94±0.08#	4.2±0.3	4.1±0.4
LGN	67%	67%	0.77±0.07	0.90±0.05	4.9±0.4	5.1±0.6

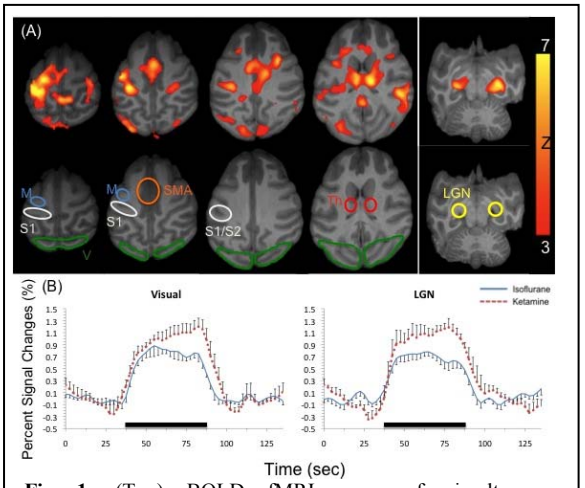

Fig 1. (Top) BOLD fMRI maps of simultaneous somatosensory/ motor and visual stimulations under 0.8-1.0% isoflurane and vecuronium from a single animal. (Bottom) Group-averaged time courses from two representative ROI's of the V1 and LGN. Error bars are SEM (n=9 for isoflurane, and n=6 for ketamine).

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