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Quantitative basal CBF and CBF fMRI of rhesus monkeys using three-coil continuous arterial spin labeling

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Abstract

A three-coil continuous arterial-spin-labeling technique with a separate neck labeling coil was implemented on a Siemens 3T Trio for quantitative cerebral blood flow (CBF) and CBF fMRI measurements in non-human primates (rhesus monkeys). The optimal labeling power was 2 W, labeling efficiency was 92±2%, and optimal post-labeling delay was 0.8 s. Gray matter (GM) and white matter (WM) were segmented based on T_1 maps. Quantitative CBF were obtained in 3 min with 1.5-mm isotropic resolution. Whole-brain average $\Delta S/S$ was 1.0–1.5%. GM CBF was 104±3 ml/100 g/min ($n=6$, SD) and WM CBF was 45±6 ml/100 g/min in isoflurane-anesthetized rhesus monkeys, with the CBF GM/WM ratio of 2.3±0.2. Combined CBF and BOLD (blood-oxygenation-level-dependent) fMRI associated with hypercapnia and hyperoxia were made with 8-s temporal resolution. CBF fMRI responses to 5% CO₂ were 59±10% (GM) and 37±4% (WM); BOLD fMRI responses were 2.0±0.4% (GM) and 1.2±0.4% (WM). CBF fMRI responses to 100% O₂ were −9.4±2% (GM) and −3.9±2.6% (WM); BOLD responses were 2.4±0.7% (GM) and 0.8±0.2% (WM). The use of a separate neck coil for spin labeling significantly increased CBF signal-to-noise ratio and the use of small receive-only surface coil significantly increased signal-to-noise ratio and spatial resolution. This study sets the stage for quantitative perfusion imaging and CBF fMRI for neurological diseases in anesthetized and awake monkeys.

Keywords

Cerebral blood flow; BOLD; Two-coil ASL; Hemodynamic coupling

Introduction

Non-invasive cerebral blood flow (CBF) measurement using MRI is widely used to study normal physiology and pathophysiology. Quantitative CBF can be obtained at high temporal and spatial resolution. Functional MRI based on CBF change is spatially more specific to the site of increased neural activity, capable of resolving cortical columns (Duong et al., 2001), is

easier to interpret than the blood-oxygenation-level-dependent (BOLD) fMRI signals, has less susceptibility to pathologic perturbations, and less inter-subject and cross-day variability (Barbier et al., 2001). Combined cerebral blood flow and BOLD fMRI measurements offer the means to estimate the stimulus-evoked changes in cerebral metabolic rate of oxygen in a totally non-invasive manner (Kim and Ugurbil, 1997; Davis et al., 1998; Hoge et al., 1999). The main drawbacks of quantitative CBF and CBF fMRI measurements are relatively low temporal resolution, low signal-to-noise ratio (SNR) per unit time, and greater susceptibility to motion artifacts (Calamante et al., 1999; Barbier et al., 2001).

CBF can be measured by using an exogenous intravascular contrast agent or by magnetically labeling the endogenous water in blood (Calamante et al., 1999; Barbier et al., 2001). The former is efficient but it is incompatible with dynamic CBF fMRI because the long half life of the contrast agent allows only one CBF measurement per bolus injection. Arterial spin labeling (ASL) techniques, on the other hand, are totally non-invasive, and the labeled water has a short half-life ($\sim$ blood T_1) making it possible to perform repeated measurements which can be used to augment spatial resolution and/or signal-to-noise ratio. ASL is compatible with dynamic CBF fMRI studies.

ASL can be performed using pulsed labeling (Detre et al., 1994; Wong et al., 1998; Wang et al., 2005) or continuous labeling (Silva et al., 1995; Zaharchuk et al., 1999; Talagala et al., 2004); both are capable of multislice and whole-brain imaging. Continuous ASL (cASL) can be achieved with the same radiofrequency (RF) coil used for imaging or a separate neck coil. cASL with a separate neck coil is generally more sensitive relative to the single-coil technique (Kim, 1995; Wong et al., 1998; Wang et al., 2005), particularly in small animals such as rodents which have short arterial transit time (Silva et al., 1999; Duong et al., 2000b). With the separate neck coil, magnetization-transfer effect is eliminated if the coils are properly decoupled, resulting in a larger signal difference between labeled and non-labeled images, and thus improved CBF SNR. RF power deposition is localized to the neck area and unlabeled images can be acquired without labeling RF, reducing specific absorption rate (SAR) (Zhang et al., 1995). While cASL technique for measuring quantitative basal CBF and CBF-based MRI is more readily available on animal scanners for rodent imaging (Silva et al., 1999; Duong et al., 2000b), similar studies on humans and large non-human primates are sparse because clinical scanners generally lack the necessary hardware and software. cASL using a separate neck coil has been reported on General Electric scanners for human studies (Zaharchuk et al., 1999; Talagala et al., 2004), although its advantage over the single-coil arterial spin labeling technique remains to be demonstrated because of the long arterial transit time in humans. The typical spatial resolution of basal ASL CBF measurements on human scanners was $\sim 70 \text{ mm}^3$ (Zaharchuk et al., 1999; Talagala et al., 2004). CBF-based fMRI using cASL with a separate neck coil on human scanners remains to be demonstrated.

The goal of this study was to implement a three-coil arterial spin-labeling technique on a Siemens 3T Trio clinical scanner for non-human primate (rhesus monkey) studies. Due to the difficulty and safety concerns in re-configuring the Siemens hardware, we instead constructed a stand-alone hardware unit for cASL using a separate neck coil. Hardware components which included an external RF amplifier, control electronics, optical-electrical relays, active decoupling circuits and radiofrequency probes were built, interfaced and tested on a Siemens 3T Trio. This approach was demonstrated by obtaining: (i) high-resolution (1.5-mm isotropic resolution or 3.3 mm^3 resolution) quantitative basal CBF in 3 min, and (ii) high-resolution combined CBF and BOLD fMRI with 8-s temporal resolution associated with hypercapnic and hyperoxic challenges. Technical issues associated with performing CBF measurements on rhesus monkeys are detailed.

Methods

Animal preparations

Rhesus monkeys ($n=6$, 5.6–8.3 kg) were initially anesthetized with ketamine (10 mg/kg, i.m.) and intubated. The animals breathed on their own under 0.9–1.1% isoflurane delivered to a non-rebreathing circuit. Gas flow to the animals was delivered at a rate of 2–3 ml/min. Animal was positioned on the stomach with the eyes facing along the magnet bore, stabilized in an animal holder with ear bars and mouth bar. Body temperature was maintained by a feedback-regulated circulating warm-water blanket. End-tidal CO_2 , O_2 saturation, heart rate, respiration rate, and rectal temperature were monitored continuously and maintained within normal physiological ranges throughout the entire studies unless otherwise perturbed.

Hypercapnic and hyperoxic challenges

Hypercapnic challenge used a premixed gas of 5% CO_2 with 30% O_2 and balance N_2 . Hyperoxic challenge used 100% O_2 . The baseline was 30% O_2 with balance N_2 which was used to ensure proper oxygenation under anesthesia. Each trial consisted of 3 min of data acquired during baseline and 3 min during hypercapnia or hyperoxia, and 4 min during baseline. At least a 10-min break was given between trials. Two to three repeated measurements were typically made. During hypercapnia, end-tidal CO_2 and respiration rate increased slightly as expected. All other monitored physiological parameters were not statistically different from normal physiologic ranges. Gases were manually switched at the multiple-inlet gas mixer. The time for the new gas to reach the animal was ~10 s and for complete exchange was ~50 s, as detected by capnometer reading at the intubation tube, accounting for the length of the capnometer line (BOLD signals could take up to 120 s to plateau).

Hardware implementation

We used the Siemens whole-body RF transmit coil for excitation, a home-made receive-only surface coil (single-loop, id=7 cm) with built-in pre-amplifier for brain imaging, and a home-made butterfly neck coil (id=2.9 cm, each loop) for arterial spin labeling. All RF coils were actively decoupled. The decoupling of the body RF transmit coil was done *via* Siemens built-in function. The decoupling of the custom-made brain and neck coils used a custom-built “decoupling box”. Although the receive-only surface coil could yield a non-uniform sensitivity profile, this was negligible because the coil size was sufficiently large for the monkey brain and uniform body-coil excitation was used.

The neck transmit-only coil was driven by a stand-alone unit of custom-made hardware which included an RF synthesizer (PTS, Model 250 S4N1G, Littleton, MA), variable attenuators, active RF switches (Mini-Circuits, Brooklyn, NY), a Watt meter (Bird Electronic Corporation, Model 43, Cleveland, OH), and RF power amplifier (Dressler HF-Technik GmbH, Model LPPA 1305, Stolberg-Vicht, Germany). To alleviate electrical safety concerns, interfaces with the Siemens console was done *via* optical signals. Optical-to-electrical relays were built to convert the optical signal from the Siemens console to electrical signal: (i) to drive the external second RF channel used for arterial spin labeling, (ii) to trigger the “decoupling box” for tuning/detuning of the two custom-made RF coils, and (iii) to synchronize the RF frequency synthesizer to the 10 MHz clock from the Siemens console. The schematic layout of the implementation is shown in Fig. 1.

MRI parameters

To verify proper position of the neck coil with respect to the carotid and vertebral arteries, cross-sectional images of the neck were acquired using neck coil to transmit and receive MRI signals. To verify proper decoupling, sagittal images were obtained without active decoupling

and with active decoupling. If the coils were not properly decoupled, the tissue water under the brain coil would be excited although no direct RF was applied to the brain coil. This could result in unwanted magnetization transfer effects.

CBF images were acquired with 1.5-mm isotropic resolution using single-shot, gradient-echo planar imaging (EPI) with TR=3.9 s, labeling duration=2.0 s, sixteen consecutive 1.5-mm slices with 0.5-mm gap between slices, TE=31 ms, matrix=64×64, FOV= 9.6×9.6 cm, and labeling gradient of 0.3 G/cm to achieve flow-related adiabatic inversion. The non-labeled images were acquired without RF excitation to the neck coil. Labeling power levels were explored from 0 to 4 W measured at the neck coil (excluding the resistive loss of the long RF cable) to determine the optimal labeling power at post-labeling delay of 0 s. Post-labeling delays were also evaluated from 0.4 to 1.4 s (quoted for the first slice acquired) to visually optimize tissue CBF contrasts at the optimal labeling power. The acquisition time between neighboring imaging slices was 68.7 ms.

T_1 maps were obtained using inversion-recovery EPI on the same imaging slices, FOV and matrix size, with TR=8000 ms, TE=31 ms, 13 inversion delays (TI=0.1, 0.2, 0.3, 0.5, 0.7, 1.0, 1.5, 2.0, 2.5, 3.5, 4.5, 5.5, and 6.5 s), and two averages. The 3D time-of-flight (TOF) angiography was acquired to identify large vessels using TR=39 ms, flip angle=15°, TE=5.74 ms, slice thickness=1 mm, FOV=96×96 mm, matrix=448×448, 36 slices, and single average.

Imaging data analysis

Data analyses were performed using Matlab (MathWorks, Natick, MA) and Stimulate software (University of Minnesota). Labeling efficiency was calculated from the arrayed labeling power data set using the formula, $(S_{\text{non-labeled}} - S_{\text{labeled}})/S_{\text{non-labeled}}$, where $S_{\text{non-labeled}}$ and S_{labeled} are signal intensities in the arteries of the non-labeled and labeled images, respectively. The locations of these vessels were confirmed by MRI angiography.

CBF in ml/100 g/min was calculated using the formula of Ye et al. (1997) for the case where magnetization-transfer effect is absent (valid if the labeling neck coil and brain imaging coils are properly decoupled),

$$\text{CBF} = \frac{\lambda}{2\alpha T_1} \frac{(S_{\text{unlabeled}} - S_{\text{labeled}})}{M_0} \times \frac{1}{e^{-\text{ATT}/T_{1A}} e^{-(\text{PLD}-\text{ATT}/T_1)} (1 - e^{-\text{LD}/T_1})}, \quad (1)$$

where λ is the water brain–blood partition coefficient, T_1 is the spin-lattice relaxation time of tissue, α is the arterial spin-labeling efficiency, $S_{\text{non-labeled}}$ and S_{labeled} are signal intensities of the non-labeled and labeled images, respectively, M_0 is the equilibrium magnetization (spin density), ATT is the arterial transit time, T_{1A} is T_1 of arterial blood, PLD is the post-labeling delay and LD is the labeling duration. T_1 maps were obtained on the same slices and M_0 maps were obtained *via* T_1 fitting. Group-average α of 0.92 (see Results), λ of 0.98 ml/g for gray matter and 0.82 ml/g for white matter (Herscovitch and Raichle, 1985), ATT of 0.8 s instead of the 1 s commonly used in humans (Butman et al., 2002), and T_{1A} of 1.66 s at 3 T (Lu et al., 2004) were used. PLD for different imaging slices were taken into account in the CBF calculation based on known acquisition time per slice.

For fMRI analysis, non-labeled images were taken as BOLD images. Cross-correlation analysis (Bandettini et al., 1993) was performed on the CBF and BOLD time-series images to obtain hypercapnia- and hyperoxia-induced CBF and BOLD percent-change maps. Similar conclusions were reached with fMRI analysis of the ΔS and $\Delta S/S$ data from the CBF measurements but they are not shown to avoid redundancy. Percent-change maps were overlaid on echo-planar images.

White matter (WM), gray matter (GM) and cerebrospinal fluid (CSF) were segmented based on T_1 maps (with typical T_1 ranges of 0.60–0.85 s for WM, 0.95–1.15 s for GM, and 1.6–3.0 s for CSF). Quantitative CBF and physiologically induced BOLD and CBF percent-changes were tabulated for the segmented WM and GM. All values are reported as mean $\pm$ SD.

Results

Optimizing ASL parameters

Locations of the brain and neck RF coils and the imaging slabs are shown in Fig. 2A. This MR image was acquired using a “volume coil” which were not used in our subsequent CBF measurements. Fig. 2B shows the head-holder setup, brain and neck RF coils. Cross-sectional images of the neck using the neck coil were acquired to ensure sufficient coverage and, thus, proper labeling of the carotid and vertebral arteries (Fig. 2C). To verify proper coil-to-coil electromagnetic decoupling, images were acquired without active decoupling (Fig. 2D) and compared with those with decoupling (Fig. 2E). The optimal labeling RF power was 2.0 W (Fig. 3). The labeling efficiency was $92\pm 2\%$ ($n=5$, Table 1). No visible burn marks or skin discoloration in the neck area were observed in any studies.

Fig. 4 shows the 16 consecutive $\Delta S/S$ images as a function of PLD for proximal-to-distal and distal-to-proximal slice acquisitions. Based on visual inspection of the $\Delta S/S$ images, a PLD of 0.8 s (quoted for the first slice acquired) was determined to be optimal for *tissue* CBF contrasts in which majority of the large vessels were no longer evident while minimizing CBF signal loss due to T_1 recovery. The typical $\Delta S/S$ in percentage ranged from 0.8% (last slice acquired) to 3% (first slice acquired) with an average whole-brain $\Delta S/S$ of 1.0–1.5% for 0.8-s PLD. All subsequent studies employed the optimal parameters of 2 W labeling power for 2.0 s labeling duration, and 0.8 s PLD.

Quantitative CBF

Quantitative CBF images with 1.5-mm isotropic resolution obtained in 3 min showed excellent blood flow contrasts among WM, GM and CSF (Fig. 5). WM, GM and CSF were segmented based on T_1 maps for CBF quantification (Fig. 6). Basal GM CBF was 104 ± 3 ml/100 g/min ($n=6$, SD) and basal WM CBF was 45 ± 6 ml/100 g/min under $\sim 1\%$ isoflurane (Table 1). The CBF GM/WM ratio was 2.3 ± 0.2 .

Combined BOLD and CBF fMRI

We targeted a 8-s temporal resolution (one pair of images) for combined BOLD and CBF fMRI measurements. Fig. 7A shows the hypercapnia-induced CBF and BOLD fMRI percent-change maps and the corresponding time courses from one animal. Large and heterogeneous CBF and BOLD changes were observed across the entire brain as expected. Similarly, hyperoxia also markedly and heterogeneously increased BOLD signals but decreased CBF signals (Fig. 7B, one animal). Group-average percent-changes during hypercapnia and hyperoxia are summarized in Table 2.

Discussion

A three-coil continuous arterial-spin-labeling technique using a separate neck coil was implemented on a Siemens 3T Trio for quantitative CBF MRI and CBF-based fMRI measurements. Multislice and quantitative basal CBF images were obtained in 3 min at 1.5-mm isotropic resolution. The optimal labeling RF power, labeling efficiency and post-labeling delay were determined. Quantitative GM and WM CBF were obtained. Combined BOLD and CBF fMRI measurements were made with an 8-s temporal resolution at 1.5-mm isotropic resolution. Hypercapnia increased both CBF and BOLD signals. Hyperoxia increased BOLD

but slightly decreased CBF signals. This approach can be used to study neurological diseases in non-human primates. This stand-alone ASL hardware unit can also be readily implemented on other MRI scanners manufactured by different vendors.

Potential errors in absolute CBF values

The main factors that could cause errors in quantitative CBF measurements by MRI are magnetization-transfer (McLaughlin et al., 1997; Golay et al., 1999), transit-time (Zhou and van Zijl, 1999; Yang et al., 2000) and water-exchange effect (Silva et al., 1997; St. Lawrence et al., 2000; Zhou et al., 2001) and they have been reviewed. Magnetization-transfer effect was not an issue for cASL here because a separate neck labeling coil was used and all RF coils were actively decoupled. Of note, the proximity of the neck coil and the cerebellum/occipital lobe could cause direct saturation. In our initial attempt, this effect was not negligible because the monkey neck is relatively short as indicated by the position dependent effect on the non-labeled signals (data not shown). The solution to avoid saturation effect was to place the neck coil as far away from the brain coil as possible and to make the neck coil as small as possible while the blood flowing through the vertebral and carotid arteries were still effectively labeled. The neck coil size was adjusted based on the coverage of the carotid and vertebral arteries in the cross-sectional neck images.

In almost all ASL measurements to date including ours, a single arterial transit time was used because of the difficulty in obtaining it in an image form and CBF is weakly dependent on arterial transit times within the physiologic ranges. ATT of 0.8 s was used instead of the 1 s commonly used in humans (Butman et al., 2002). This is justified because of the smaller size of the monkey, which is further supported by the shorter optimal PLD compared to humans. Moreover, we performed simulations using experimental parameters to investigate errors with respect to ATT. A 20% decrease in ATT (i.e., from 1.0 to 0.8 s) was estimated to result in 8% increase in CBF value. The arterial transit-time effect can be and was minimized by using adequate post-labeling delay (Alsop and Detre, 1996).

The water-exchange effect on CBF quantification is complex, depending on the precise difference between blood T_1 and tissue T_1 , blood flow values, the extent of water exchange, magnetic field strength, CBF measurement techniques, and CBF calculation models, etc. (St. Lawrence et al., 2000; Parkes and Tofts, 2002). Some of these effects counterbalance each other. Water-exchange effect is generally more significant in white matter than in gray matter because of its larger T_1 difference from blood T_1 . In this study, we employed a widely used single-compartment CBF quantification model which ignores water exchange. Comparison of different CBF quantification models, including those accounting for water exchange across the blood–brain barrier, indicates that CBF values obtained using different models were overall similar although there are some distinct differences (Steger et al., 2005). Improving absolute CBF quantification remains an active area of research. This is particularly important for low flow conditions such as in ischemic brain injury where water exchange and transit time effects on quantitative CBF are amplified and may not be negligible.

It is evident that the optimal PLD of 0.8 s quoted was not a universal optimal value across all slices. 3D scan and faster parallel imaging can be used to obtain the universal optimal PLD for CBF measurements.

SAR and optimal ASL parameters

Talagala et al. used a tissue mesh model and estimated a SAR value of 3.8 W/kg in the human neck for cASL using a separate neck coil, 1.7 W optimal labeling power, 3 s labeling duration, fast spin-echo acquisition (9 echoes) and background suppression (Talagala et al., 2004). Wang et al. used a homogeneous sphere model of a human head and estimated a SAR value of 1 W/

kg for single-coil ASL (Wang et al., 2005). These SAR values are considerably below the FDA recommendation of 8 W/kg in the human head for 15 min (FDA, 1998). Our three-coil setup is similar to Talagala's two-coil setup. We however expect that our SAR to be lower because, in our case, "control" images were acquired without labeling RF and the labeling duration (2.0 s) was shorter albeit at slightly higher labeling power. Our setup yielded an optimal power of 2 W delivered to the labeling coil and a labeling efficiency of 92%. Labeling efficiency had been reported to be 75% using similar separate neck-coil cASL (Talagala et al., 2004), and 74% (Wang et al., 2005) and 71% (Ye et al., 2000a) using single-coil ASL. The average whole-brain $\Delta S/S$ was 1.0–1.5% for 0.8 s PLD, significantly larger than those reported previously in humans (0.69% in GM and 0.26% in WM (Talagala et al., 2004), and 0.7% (Wang et al., 2005)), consistent with the higher labeling efficiency reported here. The optimal post-labeling delay was 0.8 s, comparable to slightly shorter than those in humans (1.0–1.5 s (Ye et al., 2000a; Talagala et al., 2004; Wang et al., 2005)), consistent with size difference of the subjects studied.

The CBF spatial resolution reported here (1.5 mm isotropic resolution or 3.3 mm³) is significantly higher than those reported previously in humans using MRI (3.75×3.75×5 mm or 70 mm³ (Zaharchuk et al., 1999; Talagala et al., 2004)) and PET (4.2-mm isotropic resolution or 74 mm³ (Enlund et al., 1997; Kudomi et al., 2005)). Such increase in spatial resolution is necessary and roughly compensates for the small brain volume of the rhesus monkey (89 cm³ adult male and 71 cm³ adult female (Franklin et al., 2000)) compared to adult human brain volume of ~1400 cm³ (Milner, 1990). Further improvement is expected with segmented EPI acquisition and is under investigation.

CBF comparison

CBF GM/WM ratio was 2.3 which is within the ranges reported previously of 1.7 (Talagala et al., 2004), 2.7 (Ye et al., 2000b) 3 (Pedersen et al., 2004) by MRI, and 2.0 by PET (Ye et al., 2000a). GM CBF was 104±3 ml/100 g/min and WM CBF was 45±6 ml/100 g/min in isoflurane-anesthetized monkeys. Surprisingly, we did not find any published quantitative CBF data in rhesus monkeys using MRI. Dynamic contrast-enhanced MRI only reported a CBF GM/WM ratio in anesthetized monkeys (Pedersen et al., 2004). PET reported quantitative CBF of 23–43 ml/100 g/min (whole-brain) in propofol-anesthetized monkeys (Kudomi et al., 2005), and GM CBF value of 56–68 ml/100 g/min in the cortices and a WM CBF value of 34 ml/100 g/min in ketamine-anesthetized monkeys (Enlund et al., 1997). Our CBF values in isoflurane-anesthetized monkeys were high compared to those in awake humans and anesthetized monkeys. The major cause of such difference is likely due to isoflurane anesthetic which is an established potent vasodilator. Similarly high CBF under isoflurane (Sicard et al., 2003; Sicard and Duong, 2005) relative to other anesthetics (Duong et al., 2000b) and awake conditions (Sicard et al., 2003) have been well documented in rodents (isoflurane/awake CBF ratio in rat was 1.48 (Duong et al., 2000b; Sicard et al., 2003) and the isoflurane/ α -chloralose CBF ratio was 1.84 (Sicard et al., 2003)). Other factors such as APT could also contribute to the high CBF in isoflurane-anesthetized monkeys as discussed above. Further studies are needed to rule out other technical contributing factors and parameters in CBF calculations as well as to determine the extent of high CBF under isoflurane by investigating other commonly used anesthetics.

Combined BOLD and CBF fMRI of hypercapnia and hyperoxia

BOLD and CBF fMRI studies of rhesus monkeys under physiologic or task-induced stimulations are sparse. Pfeuffer et al. reported CBF fMRI associated with visual stimulation on rhesus monkeys using the FAIR CBF technique (Pfeuffer et al., 2004). No quantitative CBF was reported in that study.

Hypercapnia-induced BOLD and CBF increases in isoflurane-anesthetized monkeys are in general agreement with those obtained in awake humans (Kety and Schmidt, 1948; Kim and Ugurbil, 1997; Davis et al., 1998; Hoge et al., 1999; Posse et al., 2001; Cohen et al., 2002) and in anesthetized rodents (Liu et al., 2004; Sicard and Duong, 2005). Hyperoxia increased BOLD as expected, consistent with those widely reported in the literature (Kim and Ugurbil, 1997; Davis et al., 1998; Hoge et al., 1999; Posse et al., 2001; Cohen et al., 2002; Liu et al., 2004; Sicard and Duong, 2005). Hyperoxia decreased CBF slightly (CBF: $-9.4 \pm 2\%$ in GM and $-3.9 \pm 2.6\%$ in WM), in good agreement with that reported in awake humans measured by PET (-13% CBF decrease) (Kety and Schmidt, 1948). MRI CBF measurement of hyperoxia is not widely available for comparison. Oxygen consumption and CBF remain relatively unchanged during brief hyperoxia and the hyperoxia-induced reduction in blood flow is a result of vasoconstriction due to hyperoxia (Kety and Schmidt, 1948). We did not quantitatively compare our data with human and rodent data because the difference in species, anesthetics and other experimental parameters will likely make these comparisons less meaningful.

Finally, it should be noted that isoflurane, being a strong vasodilator, has a significant effect on vascular reactivity and, thus, the BOLD and CBF percent-changes. It has been shown previously that isoflurane causes sluggish BOLD responses as indicated by the delays of the BOLD time-to-peak associated with visual stimulation (Duong et al., 2000a). Moreover, the high basal CBF is expected to reduce the head room for hypercapnia-evoked CBF and/or BOLD increases (Sicard and Duong, 2005). It is thus interesting to compare isoflurane data with those obtained under other common anesthetics and awake conditions in monkeys.

Conclusions

This study reports quantitative CBF measurements with 3-min resolution and the combined CBF and BOLD fMRI with 8-s resolution in rhesus monkeys at 1.5-mm isotropic resolution. Further improvements in spatial and temporal resolution are expected. With its unique advantages, quantitative perfusion imaging and perfusion-based fMRI using cASL with a separate neck coil are expected to have increasing applications in both experimental research and clinical setting. These results set the stage for quantitative perfusion and CBF fMRI of awake monkeys, imaging stroke and other neurological diseases in non-human primates. The stand-alone ASL hardware unit can be readily implemented onto scanners manufactured by other vendors.

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Fig. 1.

Schematics of a stand-alone unit for the three-coil arterial-spin-labeling technique.

Fig. 2.

(A) MR image of a monkey head and neck using a “volume coil” for demonstration of the approximate positions of the coils only. (B) A picture of the head-holder apparatus with neck and brain coils. (C) Cross-sectional image of the neck showing the carotid and vertebral arteries. Neck coil was used as the transceiver. Sagittal images were obtained (D) without active decoupling and (E) with active decoupling. In the presence of electromagnetic coupling (i.e., the coils are not properly decoupled) the tissue water under the brain coil was excited which could result in unwanted magnetization transfer effects.

Fig. 3. Labeling efficiency. $\Delta S/S$ images at different labeling RF powers and the MRA images are shown. The ROIs of the arteries were used to quantify labeling efficiency. The optimal labeling power was 2 W which yielded 92% labeling efficiency, as indicated by the arrowhead.

Fig. 4.

Effects of post-labeling delays (PLD). $\Delta S/S$ images of 16 slices were acquired (A) proximal to distal and (B) distal to proximal. The signal drop off is due to T_1 recovery. The optimal PLD was 0.8 s. The typical $\Delta S/S$ in percentage ranged from 0.8% (last slice acquired) to 3% (first slice acquired) with an average whole-brain $\Delta S/S$ of 1.0–1.5% for 0.8-s PLD. The gray scale bar indicates 0 to 3% changes.

Fig. 5. Quantitative CBF images using the optimized parameters. Quantitative CBF images however did not show a drop off in signal across imaging slices compared to $\Delta S/S$ images in Fig. 4 because the effects of post-labeling delays were taken into account in the CBF calculation.

Fig. 6.
 T_1 maps and the segmented white matter, gray matter and CSF. Expanded views of the 7th slides are also shown.

Fig. 7. Single-animal BOLD and CBF fMRI of (A) hypercapnic and (B) hyperoxic challenge. Color bar indicates cross-correlation coefficients. BOLD and CBF changes are heterogeneous with larger changes in the gray matter and smaller changes in the white matter. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1

Labeling efficiency α , cerebral blood flow (CBF) values in gray mater (GM) and white mater (WM)

Subjects	α (%)	CBF (ml/100 g/min)		Ratio GM/WM
		GM	WM	
1	93.5	107.8	55.9	1.9
2	88.5	104.8	44.3	2.4
3	94.5	106.5	46.1	2.3
4	93.4	102.3	42.7	2.2
5	91.3	102.6	39.8	2.6
6	–	99.9	41.4	2.4
Average	92 $\pm$ 2	104 $\pm$ 3	45 $\pm$ 6	2.3 $\pm$ 0.2

GM and WM were segmented based on T_1 map.

Table 2

BOLD and CBF fMRI percent changes in gray and white matter associated with hypercapnia and hyperoxia

		Gray matter (%)	White matter (%)
Hypercapnia (<i>n</i> =4)	CBF	59±10	37±4
	BOLD	2.0±0.4	1.2±0.4
Hyperoxia (<i>n</i> =3)	CBF	-9.4±2	-3.9±2.6
	BOLD	2.4±0.7	0.8±0.2