Activation induced BOLD and CBF responses upon acetazolamide administration: implications for inferring neurovascular and metabolic coupling in patients with impaired cerebrovascular reactivity

Jeroen C.W. Siero[1], Nolan Hartkamp[1], Anita A. Harteveld[1], Annette Compter[1], Bart van der Worp[1], Manus J. Donahue[2], Esben T. Petersen[1], and Jeroen Hendrikse[1]
[1]*University Medical Center Utrecht, Utrecht, Utrecht, Netherlands,* [2]*Radiology and Radiological Sciences, Vanderbilt University, TN, United States*

Introduction. Blood-oxygenation-level-dependent (BOLD) MRI is widely used for inferring neuronal activation and is becoming increasingly popular for assessing cerebrovascular reactivity (CVR) in combination with a vasoactive stimulus [1,2,3]. The rationale for using the BOLD signal is that it reflects underlying changes in cerebral blood flow (CBF) that carry information regarding neurovascular coupling and CVR. The BOLD signal, however, does not only reflect purely CBF changes, but is also modulated by changes in CBV and CMRO2. In patients with cerebrovascular disease it has been demonstrated that uncoupling between changes in CBF, CBV and CMRO2 can lead to discrepant BOLD responses [4]. This can affect correct interpretation on functional activation when solely using BOLD MRI. However, in the above study only neuronal activation was evaluated, and it remains unclear whether vascular stimuli, either in isolation or in conjunction with neuronal stimuli, may influence neurovascular and metabolic coupling. Here, we investigated BOLD and CBF responses in patients with vertebral basilar artery stenosis. Specifically, we evoked neuronal BOLD and CBF responses in visual cortex before and after a vasodilatory challenge (acetazolamide). Acetazolamide (ACZ) is a carbonic anhydrase inhibitor that increases the baseline CBF due to vasodilation - the degree of which gives information regarding CVR [5,6]. We observe that additional measurements of CBF contain crucial information in this population, and therefore that solely BOLD MRI cannot correctly assess the activation and condition of the vasculature and surrounding tissue.

Materials and Methods. *Data acquisition:* Patients (N=8, age=61±5) that were included were diagnosed with vertebral basilar (VBA) stenosis or occlusion. *ASL/BOLD fMRI*; PCASL data were acquired on a Philips 3T system (8-channel head coil) before and after (15min) ACZ administration (14mg/kg) using GE-EPI with background suppression: TR/TE=4000/20 ms, flip angle=90°, SENSEfactor=2.5, voxel size=3mm, FOV=240×240×105 mm^3, 15 slices, 45 control/label pairs, scan-time=6.2 min. An additional M0 scan was acquired for CBF quantification. During data acquisition, the participants were presented with 40s periods of visual stimulation (8Hz checkerboard) interleaved with 40s showing a uniform gray screen. *Analysis:* Local subtraction and addition of control and label images yielded respectively CBF and BOLD weighted images. Region-of-interest (ROI) analysis was performed on the combined CBF and BOLD activation map (|z-stat| ≥1.6, p<0.05, using FSL FEAT [7]) confined to the visual cortex. Visual-activation induced BOLD and CBF responses were computed as well as maps of baseline CBF and CVR (percentage CBF increase upon ACZ administration).

Results Fig. 1 shows for three patient cases maps of CVR, and respectively pre- and post-ACZ baseline CBF, ΔCBF and ΔBOLD. Group results for pre- and post-ACZ baseline CBF, ΔCBF and ΔBOLD are shown in **Fig. 2**. We observed an overall increase in basal CBF upon ACZ administration (**Fig. 2A**), and a reduced ΔBOLD upon visual stimulation while absolute ΔCBF values showed no significant differences (**Fig. 1A**, and **Fig. 2B,C**). Interestingly, in 3 out of 8 patients substantial negative ΔBOLD changes were found only post-ACZ (area 30% - 70% of visual-activation ROI (two patients shown in **Fig. 1B** and **1C**). **Fig. 3A** shows the visual-activation induced post-ACZ ΔBOLD and ΔCBF response from the negative BOLD area for patient 3. Results show that the negative post-ACZ ΔBOLD findings are not consistent with large response delays or vascular steal (i.e., CBF reduction below baseline); the post-ACZ ΔCBF response is positive for these regions and has similar temporal evolution. Interestingly, no evidence was found for the appearance of negative ΔBOLD being related to differences in CVR between the negative and positive ΔBOLD areas (**Fig. 3B**). **Fig. 3C,D** shows ΔBOLD versus ΔCBF for all patients; the pre-ACZ ΔBOLD changes show a positive linear relationship with ΔCBF changes (**Fig. 3C**). Post-ACZ, however, this apparent one-to-one relationship is absent (**Fig. 3C**), consistent with the occurrence of large areas with negative ΔBOLD (blue shaded area).

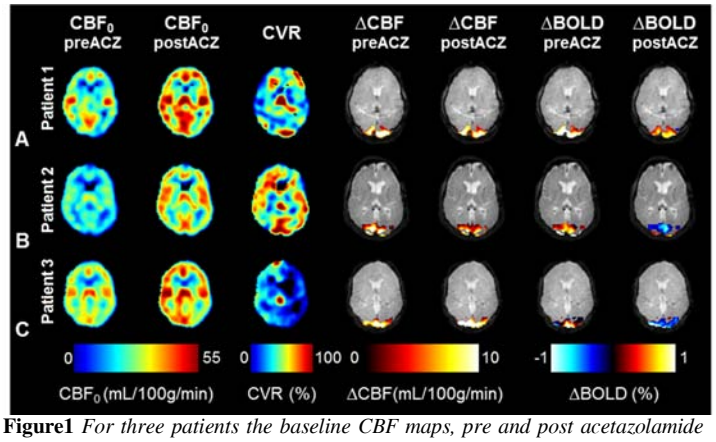

Figure1 *For three patients the baseline CBF maps, pre and post acetazolamide (ACZ) administration, the reactivity map (CVR), and the vascular responses (ΔCBF and ΔBOLD) upon visual activation, pre and post-ACZ administration.* ***A)*** *Results for patient1 shows similar CBF changes pre/post-ACZ, while the post-ACZ BOLD response is reduced, in line with previous findings in healthy subjects [8].* ***B,C)*** *for patients 2 and 3, however, we observe large regions with negative BOLD post-ACZ changes whilst the CBF changes remained similar.*

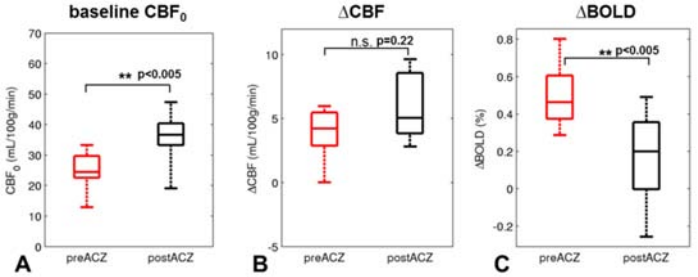

Figure2 ***A)*** *Baseline CBF,* ***B)*** *visual-activation induced CBF changes (ΔCBF), and* ***C)*** *BOLD changes (ΔBOLD), respectively for pre- and post-ACZ administration across all patients. Baseline CBF increases post-ACZ due to the vasodilatory challenge, causing ΔBOLD to reduce upon visual-activation while ΔCBF did not change significantly.*

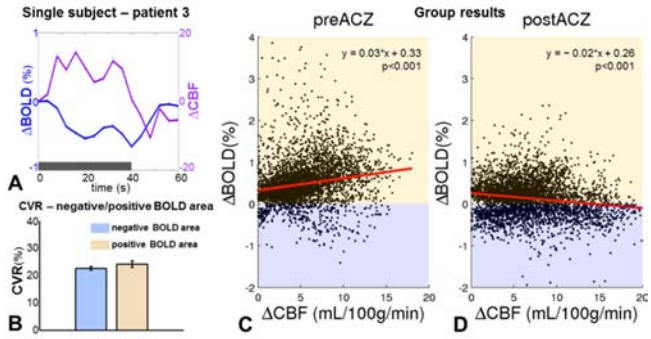

Figure3 ***A)*** *the post-ACZ ΔBOLD and ΔCBF response from the negative BOLD area for a representative subject (patient 3). The gray bar indicates the visual stimulus duration.* ***B)*** *CVR results for respectively the negative (blue) and positive BOLD area for the same patient. No clear CVR differences are observed.* ***C)*** *ΔBOLD versus ΔCBF for pre-ACZ, and* ***D)*** *post-ACZ showing data from all patients. A positive relationship between ΔBOLD and ΔCBF is found pre-ACZ, whereas post-ACZ this relationship is severely diminished and shows even a negative correlation due to a large regions of negative ΔBOLD (blue shaded area, and* ***Fig.1 A,B****).*

Discussion

We show that upon ACZ administration the neuronal-activation induced BOLD response is reduced for patients with impaired CVR while the CBF response remained relatively stable. At first glance, the group results are in line with previous findings in healthy subjects using BOLD fMRI and a vasodilatory challenge [8,9], and can largely be explained by the Davis' model for the BOLD signal (reduced available deoxyhemoglobin content post-ACZ) [10]. Interestingly, we observe substantial negative BOLD areas post-ACZ in 3 of 8 patients while CBF responses remained positive. An explanation for these results is more complex and likely gears towards an altered neurovascular and/or metabolic coupling occurrence. Different possibilities may lead to a negative BOLD and positive CBF response [11]: i) increased CMRO2, ii) excessive compliant vasculature (increased ΔCBV), or iii) strong autoregulation where ΔCBF and ΔCBV are disproportionally increased compared to CMRO2, and combinations of above. Our findings show that caution is warranted when inferring cortical activation and neurovascular coupling in these patients when solely using BOLD fMRI as the conventionally assumed one-to-one relationship between BOLD and CBF responses is responses is compromised (**Fig. 3B,C**).

References:[1] Lythgoe DJ et al. MRI 1999, [2] van der Zande FHR et al. NeuroRad2005, [3] Yezhuvath et al. NMRBiom. 2009, [4] Blicher JU et al. JCBFM2012, [5] Vorstrup et al. 1984, [6] Russel D et al. Stroke 1990, [7] Smith SM et al. NI2004, [8] Brown GG et al. JCBFM2003, [9] Bruhn et al. JCBFM1994, [10] Davis et al. PNAS1998, [11] Blicher et al JCBFM2012