Limitation of DCE-MRI in Xenograft Model Study

Septian Hartono[1], Tong San Koh[2], Quan Sing Ng[2], Richard Ong[2], Hung The Huynh[2], Sidney Yu[3], Sotirios Bisdas[4], Laurent Martarello[5], and Choon Hua Thng[2]
[1]Nanyang Technological University, Singapore, Singapore, [2]National Cancer Centre, Singapore, Singapore, [3]Singapore General Hospital, Singapore, [4]Eberhard Karls University, Tübingen, Germany, [5]F. Hoffmann-La Roche, Switzerland

Introduction: DCE-MRI has been used extensively in preclinical and clinical trials as a biomarker of drug effect. It is commonly assumed that a decrease in blood volume and blood flow is related to drug effect. This study examines the pitfall of such an assumption and illustrates decrease in perfused blood volume in untreated tumor as derived by DCE-MRI.

Materials and Methods: Mice: Male BALB/c mice (6 weeks old, 30±2g) were maintained according to the Guide for the Care & Use of Laboratory Animals (NIH). They were implanted with human-derived hepatocellular carcinoma xenograft line. The experiment design was as follow:

Group	Number of days after implantation							IHC
	13 (BL)	15 (D0)	16 (D1)	20 (D5)	23 (D8)	27	34	
Control A (n = 6)	MRI	Xolair	MRI	MRI	MRI	MRI	MRI	No
Control B (n = 6)	MRI	Xolair	MRI	MRI	MRI+IHC			Yes
Treated (n = 6)	MRI	Bevacizumab	MRI	MRI	MRI+IHC			Yes

The control groups were given an IP human anti-IgE antibody Xolair at 10mg/kg while the treated group was given with antiVEGF antibody Bevacizumab (Avastin) at 10mg/kg.

DCE-MRI: MRI was performed on a 7T scanner (Bruker ClinScan, Bruker BioSpin MRI GmbH, Germany). A 3D VIBE sequence was used with following parameters: TR = 3.04ms, TE = 1.23ms, FOV = 36 × 36 mm, 128 × 128 matrix, 8 slices with thickness of 1 mm, & temporal resolution 2 s. Five sets of baseline images were acquired with α = 6° & 14°. It was followed by a dynamic sequence of 130 sets of images (α = 14°). A dose of 100 µL of Gd-DOTA (Dotarem, Guerbet SA, France) at 1 mmol/kg was injected through the tail vein after the first set of dynamic images.

Data Processing: Region of interests corresponding to the xenograft and aorta were manually outlined. Microcirculatory parameters such as blood volume (v_1) were derived from the two-compartment model of Brix.

Results: For control group A, blood volume was 5.15 ± 1.49 % at baseline increasing to 6.91 ± 2.82 % at Day 20 before dropping to 1.18 ± 0.41 % at Day 23. The low level of blood volume was sustained until the terminal time point at the level of 1.23 ± 0.39%. An example of such drop between Day 20 and Day 23 was shown in Fig 1 and the overall trend plot was displayed in Fig 2.

For control group B, baseline blood volume was 8.17 ± 2.98 %, increasing to 10.46 ± 2.03 % and subsequently dropping to 4.47 ± 3.03%. For the Bevacizumab group, baseline blood volume was 8.80 ± 2.95% which dropped to 4.51 ± 1.98% one day after dosing and subsequently remained as low as the other control groups. The individual plots of blood volume trends for control group B and bevacizumab group can be seen in Fig 3.

Although the measured blood volume was similar for all groups at end of timeline, CD34 staining for vessels, as shown in Fig 4, showed that the control group has more stained vessels than the treated group. The low DCE MRI reading for blood volume was related to vessels shut down in the treated group. We postulate that the low blood volume measured by DCE MRI in the control group was related to increased proportions of non-perfused vessels possibly due to increase in interstitial fluid pressure.

A switching point can be appreciated in the blood volume time course graph beyond which there is a significant and sharp drop. The biological processes responsible for this drop require further study. As decrease in DCE-MRI measured blood volume can occur in control groups, control groups should not be omitted in DCE MRI experimental designs.

Conclusion: Our study show that as control tumor grows, it will reach a switching point where there is a significant and sharp drop in blood volume presumably related to increase in interstitial fluid pressure and decrease in perfused vessels. Thus, characterization study must be performed for every study to determine the switching point for the particular xenograft. DCE MRI studies are best performed before the switching point to avoid this confounding effect. Interpretation of DCE-MRI results in these studies must be done very carefully as well, especially if no characterization study has been done before hand or if no sham controls were included.

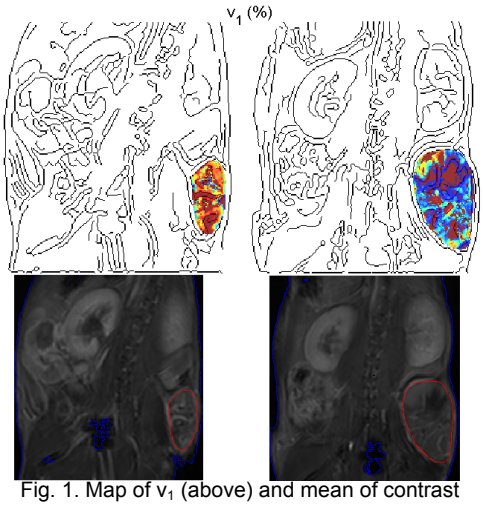

Fig. 1. Map of v_1 (above) and mean of contrast enhancement (below) of mouse #2 in Control A before (left) and after (right) the switching point

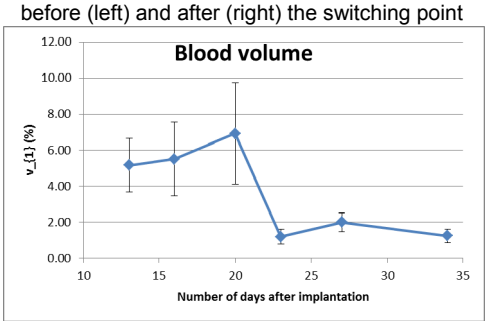

Fig 2. Plot of blood volume for control group A

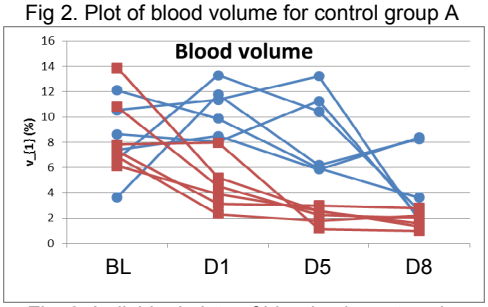

Fig. 3. Individual plots of blood volume trends for control group B (blue) & treated group (red)

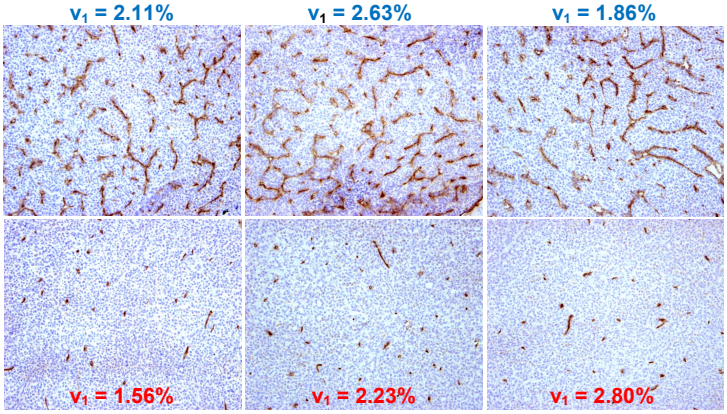

Fig. 4. CD34 staining for control group B (above) and treated group (bottom) and their respective measured blood volume by DCE-MRI