Comparisons of the Efficacy of the Jak1/2 Inhibitor AZD1480 with the VEGF signaling inhibitor cediranib (AZD2171) and Sham Treatments in Mouse Tumors Using DCE-MRI, DW-MRI, and Histology

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INTRODUCTION

Jak1/2 inhibition suppresses Stat3 phosphorylation that is characteristic of many tumor types. Activated Stat3 promotes the transcription of factors that enhance tumor growth, survival, and angiogenesis[1]. AZD1480 is a novel small molecule inhibitor of Jak 1/2, which have been shown to be key mediators of Stat3 activation[2]. Both DW-MRI and DCE-MRI have been used to monitor cancer treatment[3,4]. This work seeks to assess the utility of DW-MRI and DCE-MRI in determining AZD1480 efficacy compared to the potent anti-angiogenic drug cediranib at early treatment time points.

MATERIALS and METHODS

Thirty mice were injected with Calu-6 lung cancer cells in the hind limb; once the tumors reached approximately 200 mm^3, the mice were randomized into the following treatment groups: AZD1480 (50 mg/kg, *p.o. q.d.*), cediranib (6 mg/kg *p.o. q.d.*), and vehicle control. All animals were imaged at 9.4T at baseline, day 3 and day 5 post-treatment time points. DCE-MRI. Pre-contrast T_1 maps were obtained using an IR FLASH gradient echo sequence with eight inversion times with *TR**TE* \α= 12100 ms\ 3.44 ms\15° and NEX = 4, FOV = 35 mm^2, and matrix = 128^2 for fifteen 1 mm slices. The dynamic acquisition employed a SPGR sequence with *TR*\TE*\α* = 100 ms\2.83 ms\10°, and NEX = 4. A bolus of 0.1 mmol/kg Gd-DTPA was delivered *via* a jugular catheter using an automated syringe pump. A population derived VIF (C_p) was used to fit the tissue signal data (C_t) in the central slice for each mouse at each time point using a standard model[5]. DW-MRI. A gated and navigated PGSE sequence was used with the following parameters: TR\TE\α = 2000 ms\42 s\15°, acquisition matrix = 128^2, FOV = (35 mm)^2, and NEX = 2 with Δ = 35.00 ms and δ =5.00 ms for b-values of 150.88, 500.2, and 800.22 mm^2/s. The diffusion weighted signal at each b (S(b)) was fit to the following equation to extract the apparent diffusion coefficient (ADC)[6]

$$S(b) = S_0 e^{-ADC \cdot b}.$$

The percent change from baseline measurements for K^{trans}, *ADC*, and v_e were calculated for each time point. The Wilcoxon rank test identified significant changes in imaging parameters while H&E, CD31, cParp, and Ki-67 histology data validated the results.

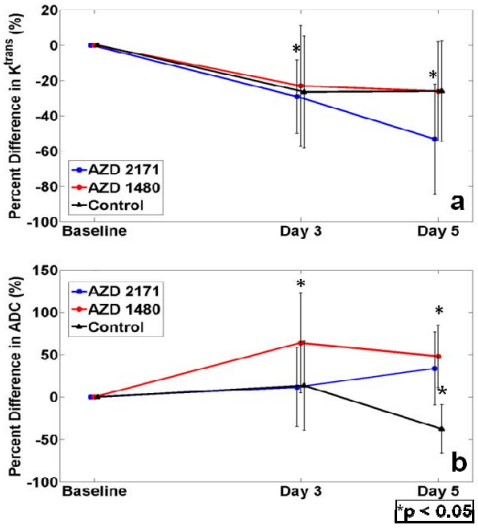

Fig 1. Percent change in K^{trans} (**a**) and *ADC* (**b**) for each treatment group.

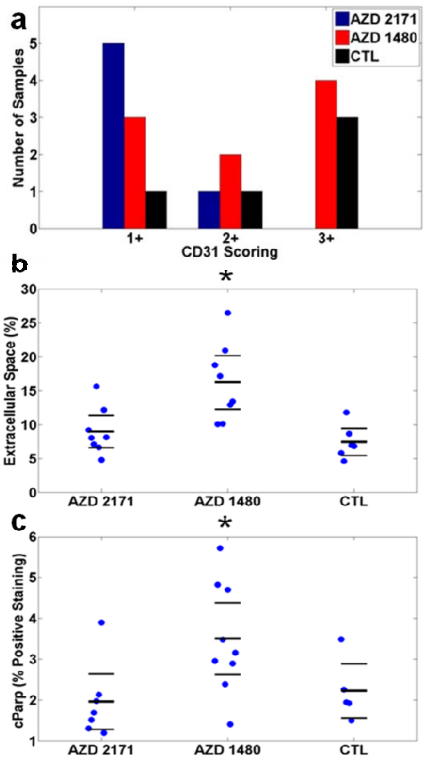

Fig 2. Plots demonstrating the number of samples in each CD31 scoring level (**a**), *EC* (%) for each treatment group (**b**), and cParp (%) staining for each group (**c**).

RESULTS

Fig. 1 presents the resulting K^{trans} (**1a**) and *ADC* (**1b**) changes for each treatment group. A significant decrease (indicated by '*') in K^{trans} was found at both day 3 and day 5 post-tx time points for the cediranib group. Interestingly, no significant changes in K^{trans} occurred for the AZD1480 group compared to controls, but a significant increase in *ADC* was demonstrated at day 3 and day 5. Control data showed a significant decrease in *ADC* at day 5. No significant changes in v_e were demonstrated in any treatment group except a decrease in the cediranib group at day 5. Histology acquired after day 5 (**Fig. 2**) indicated attenuated vasculature in the cediranib treatment group (1+ = reduced vascularity while 3+ = high vascularity) while no significant changes were shown in the AZD1480 or control groups (**2a**). Significant apoptotic activity (positive cParp staining in **2c**) and significantly increased extracellular space (**2c**) was present in the AZD1480 group with no changes in the cediranib/control groups (**2c**).

DISCUSSION

The lack of significant group changes in K^{trans} demonstrate that day 3 and day 5 of a 50 mg/kg *q.d.* dose of AZD1480 may be too early to see anti-angiogenic effects, compared to the VEGF signaling inhibitor cediranib. The more sensitive measure for AZD 1480 treatment seemed to be *ADC*. Significant increases in *ADC* were indicated while histology indicated a significant increase in apoptosis and extracellular space in this treatment group. Thus, tumor cell death was evident in AZD1480-treated xenografts in the absence of anti-angiogenic activity while anti-angiogenic activity (decreased K^{trans}) was detectable in xenografts treated with cediranib.

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