Abstracts

P08.35 CLINICAL OUTCOMES FOR GBM PATIENTS WITH SOLITARY, MULTIFOCAL AND MULTICENTRIC DISEASE
Bea ton oncology centre, Glasgow, United Kingdom.

INTRODUCTION: Based on imaging features, glioblastoma (GBM) can be classified as solitary, multifocal or multicentric. The incidence of mono-
centric/multicentric GBM has been reported to range from 0.5 to 35% and some studies have reported poorer survival in patients with multiple lesions. Two potential reasons have been proposed: (1) intrinsic differences in tumour biology; (2) failure to encompass the entire tumour within the radiotherapy planning volume. To address this question we investigated clinical imaging and genetic features in a cohort of GBM patients.

METHODS: Imaging, clinical, treatment, MGMT methylation and outcome data were collected retrospectively for all consecutive GBM patients treated in a single cancer centre between January 2011 and June 2012. Tumours were categorised as solitary, multifocal or multicentric by a consultant neuroradiologist. Patients with MGMT promoter status was unmethylated or methylated in 37% and unknown in 15%. Preoperative imaging was CT in 70% and MRI in 30% of patients. Overall, the proportion of patients with solitary, multifocal and multicentric tumours was 78%, 15.5% and 6.5% respectively, but in patients undergoing MR imaging these proportions were 60%, 26.5% and 13.5%. Gross total resection was achieved in 65% of solitary compared with 42% of multifocal and 23% of multicentric cases, and radical chemoradiation was delivered to 50% of solitary, 38% of multifocal and 11% of multicentric cases. Overall, median survival was increased in solitary compared with multifocal/ multicentric patients (9.9 vs. 6.7 months, p < 0.046). In patients receiving radical chemoradiation, however, there was no difference in overall survival between solitary and multifocal/multicentric patients (18.5 vs. 16.8 months, p = 0.57).

RESULTS: 122 patients with GBM were identified. Median age was 60 and male:female ratio was 2:1.1. MGMT promoter status was unmethylated in 48% of tumours, methylated in 37% and unknown in 15%. Preoperative imaging was CT in 70% and MRI in 30% of patients. Overall, the proportion of patients with solitary, multifocal and multicentric tumours was 78%, 15.5% and 6.5% respectively. In patients undergoing MRI imaging, these proportions were 60%, 26.5% and 13.5%. Gross total resection was performed in 65% of solitary compared with 42% of multifocal and 23% of multicentric cases, and radical chemoradiation was delivered to 50% of solitary, 38% of multifocal and 11% of multicentric cases. Overall, median survival was increased in solitary compared with multifocal/multicentric patients (9.9 vs. 6.7 months, p < 0.046). In patients receiving radical chemoradiation, however, there was no difference in overall survival between solitary and multifocal/multicentric patients (18.5 vs. 16.8 months, p = 0.57). MGMT promoter methylation was associated with increased survival in patients with solitary tumours (14.6 vs. 8.5 months, p = 0.014) but not multifocal/multicentric tumours (3.8 vs. 6.9 months, p = 0.3).

CONCLUSION: In this retrospective study the incidence of multifocal/multicentric GBM was in line with previous studies. Pre-operative CT imaging may underestimate the incidence of multifocal/multicentric disease. Our main finding was that multifocality or multicentricity did not affect survival in patients to whom radical chemoradiotherapy could be delivered.

P08.26 RADIORESISTANCE OF GBM STEM-LIKE CELLS IS ASSOCIATED WITH DNA REPLICATION STRESS, WHICH IS A PROMISING THERAPEUTIC TARGET
S. U. Ahmed1, R. Carruthers2, D. Bisso1, N. Gomez-Roman2, L. Gilmour3, K. Strathdee3, A. Hedley1, G. Kalina1, E. Hammonds1, A. J. Chalmers2
1University of Sunderland, Sunderland, United Kingdom, 2University of Glasgow, Glasgow, United Kingdom, 3University of Oxford, Oxford, United Kingdom, 4CR-UK Beatson Institute, Glasgow, United Kingdom.

INTRODUCTION: The inevitability of tumour recurrence in glioblas-
toma (GBM) patients despite multi-modality treatment consisting of surgery, radiotherapy and chemotherapy, is reflected by a median survival of only 14 months. Tumour recurrence is thought to be driven by a small population of glioblastoma stem-like cells (GSCs) that are resistant to conventional therapies. DNA damage response (DDR) pathways have been shown to be up-regulated in GSCs and implicated in radioresistance and treatment failure. However the precise cause of enhanced DDR signalling in GSCs and the extent to which these signalling networks contribute to therapy resistance remains elusive. The objectives of this study were to investigate the underly-
ing cause of DDR upregulation and treatment resistance in GSCs with a view to identifying novel and promising therapeutic targets.

MATERIALS AND METHODS: A panel of primary patient derived GBM cell lines cultured under conditions to enrich for or deplete the tumour stem cell population (GSC vs bulk respectively) were utilised in order to investigate enhanced GSC DDR under basal conditions and in response to ionising radiation. Confirmatory studies were also performed in cells sorted for the putative GSC marker CD133. The effects of a panel of small molecule DDR inhibitor agents on cell survival in GSC and bulk cells were quantified.

RESULTS: GSCs exhibited higher levels of total and activated DDR targets ATR, CHK1, ATM and PARP1 under basal conditions and were also more resistant compared to paired bulk populations. This was not due to increased levels of reactive oxygen species (ROS). Instead, we show that RPA is significantly higher in replicating GSCs and confirm by DNA fibre assays that GSCs and CD133+ cells have increased numbers of stalled replication forks, fewer new origins and slower DNA replication compared to bulk or CD133− populations, demonstrating for the first time that replication stress (RS) is a hallmark of GSCs. We identify increased expression of long neural genes as a likely mechanism for RS and DNA double strand breaks (DSBs) in GSCs and show that their radioresistance is reversed by dual inhibition of key RS and DDR proteins ATR and PARP.

CONCLUSIONS: This study demonstrates the novel finding that replica-
tion stress is a hallmark of GSCs and resonates with recently published studies showing neuronal progenitors cells showing that RS preferentially induces DNA DSB in long neural genes. Taken together, we implicate RS as a driver of enhanced DDR in GSCs and identify novel therapeutics with potential to improve clinical outcomes by overcoming the radioresistance of GBM.

P08.37 TUMOR ASSOCIATED M2 MACROPHAGE INFILTRATION IN GlioBLASTOMA
Kobe University Graduate School of Medicine, Kobe, Japan.

INTRODUCTION: Anti-inflammatory phenotype (M2) macrophage is known to secrete various cytokines and promote tumor-growth. However, the role of M2 macrophage in glioblastoma is not clear yet.

METHODS: We evaluated the specimens resected from thirty-three patients with glioblastoma, who underwent surgery at Kobe University Hos-
pital from November 2006 to December 2013. We investigated the infil-
tration of tumor-associated M2 macrophages by means of immunostaining for CD163, which is M2 cell marker. Also, we examined the association between infiltration levels of M2 macrophages and CSF cytokines (IL-6, IL-10, IL-4). Spearman’s correlation coefficient by rank test. RESULTS: The mean infiltration rate of CD163-positive M2 cells in all glioblastoma specimens was 23.9% (3.7–64.7%). CD163-positive M2 macrophages tended to infiltrate in perivascular regions. Mean CSF concentration of IL-6, IL-10, IL-4 in all glioblastoma patients were 30.5 pg/ml (IL-6), 2 pg/ml (IL-10), and <50 pg/ml (IL-4), respectively. The mean concentration of IL-6 was 40 pg/ml. In relationship between infiltration levels of M2 macrophages and CSF cytokines, IL-6 had significant shorter PFS in comparison with those with lower levels of IL-6 (p=0.046).

CONCLUSIONS: We observed high infiltration of CD163-positive M2 macrophages around the perivascular regions in glioblastoma. In addition, the infiltration levels of M2 macrophages correlated with IL-6 levels in CSF. The infiltration level of CD163-positive M2 macrophages may be associated with the prognosis of patients with glioblastoma.

P08.38 IRRADIATION OF SUBVENTRICULAR ZONE IN GliOBLASTOMA: ITS IMPACT ON TUMOR PROGRESSION AND SURVIVAL
E. Sert, S. Hoca, S. Kam, Y. Aruşık
Ege University, Izmir, Turkey.

Irradiation of Subventricular Zone in Glioblastoma: Its Impact on Tumor Progression and Survival