NRF2 knockdown as a mean to sensitize Glioblastoma Neurospheres to UVA Radiation

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INTRODUCTION

Glioblastoma (GBM) is the most fatal and frequent primary brain tumor, with an incidence rate of 3.22 per 100,000 population per year [1]. Glioma stem cells (GSC) in GBM are radio- and chemoresistant and have been pointed as the main cause of GBM’s frequent relapse [2]. The inhibition of NRF2, the master regulator of antioxidant defense, is becoming an interesting approach to increase GSC vulnerability towards oxidative stress induced by radiation exposure [3] (Fig 1). We previously showed a reduction of stem-like properties in GSCs with reduced NRF2 expression (N18) compared to wildtype GSCs (U87MG) after treatment with ionizing radiation [4]. Nevertheless, detailed molecular data regarding this transfected cell line is still missing.

METHODS

PART 1. U87MG vs N18 SENSITIVITY TO UVA

RESULTS

500 N18 and U87MG cells in 20µL SFM* followed by UVA irradiation (0, 5, 10, 15 and 20 min) Neurorsphere Counting 3 days in 100µL SFM

PART 2. ML385 TOXICITY ASSAYS

RESULTS

500 U87MG in 200µL SFM* with ML385 6 days 37°C and 5% CO2

PART 3. CRISPR-CAS9 KEEAP1 KNOCKOUT

RESULTS

4 days 24h 37°C and 5% CO2

Protein extraction and quantification for Western Blot analysis

References