Epidermal Growth Factor Receptor Targeted Fluorescein for Glioblastoma Boundary Delineation: A Novel Fluorophore with Superior Specificity

Ranjith Babu MS; Chunhui Di; Roger McLendon MD; Allan H. Friedman MD; D. Cory Adamson MD PhD MPH MHSc
Duke University Medical Center, Durham, NC, USA; Durham VA Medical Center, Durham, NC, USA

Introduction

Fluorescence-guided resection has emerged as a potential tool for defining the often obscure tumor-brain interface during glioblastoma tumor surgery [1]. However while agents such as fluorescein and aminolevulinic acid have been shown to increase the extent of resection, these fluorophores may result in the fluorescence of the surrounding edematous brain. In this study, we have evaluated the specificity of epidermal growth factor conjugated fluorescein (EGF-FL) and have compared it to conventional fluorescein in vitro and in vivo.

Methods

In the in vitro study, U87 (low-EGFR expression) and U87-MG-EGFR (high-EGFR expression) cells were incubated with various concentrations of fluorescein and EGF-FL. In the in vivo study, athymic nude mice were intracranially implanted with U87-MG or U87-MG-EGFR cells. Mice were intravenously administered fluorescein sodium or EGF-FL and then sacrificed.

Results

Cellular fluorescence in U87 cells was highest following treatment with 5ug/mL of EGF-FL, with this fluorescence being significantly greater than that achieved by 15ug/mL of fluorescein ($p=0.0014$). Also treatment of U87-MG-EGFR cells with 0.5ug/mL of EGF-FL resulted in more cellular fluorescence than 15ug/mL of fluorescein at both 1 and 3 hours ($p<0.0001$). Evaluation of fluorescein tumor specificity revealed tumor tissue to account for only 68.4% and 59.1% of total fluorescence in low and high EGFR expressing tumors, respectively (Figure 1). Use of EGF-FL in U87 tumors revealed similar tumor specificity as fluorescein (70.9% vs. 68.4%, $p=0.71$). However, the use of EGF-FL in U87-MG-EGFR tumors revealed a tumor specificity of 93.2% which was significantly greater than that seen for fluorescein ($p<0.0001$) (Figures 2 and 3).

Figure 1. Fluorescence and histological images of tumor boundaries following treatment with fluorescein demonstrating extratumoral fluorescence in U87-MGEGFR tumors

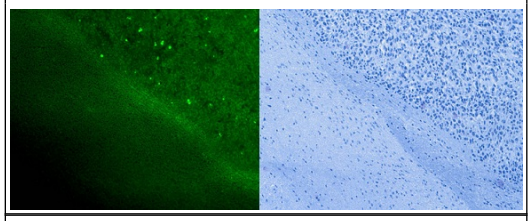


Figure 2: Fluorescence and histological images of tumor boundaries following treatment of U87-MGEGFR tumors with EGF-FL demonstrating no fluorescence in the surrounding normal brain

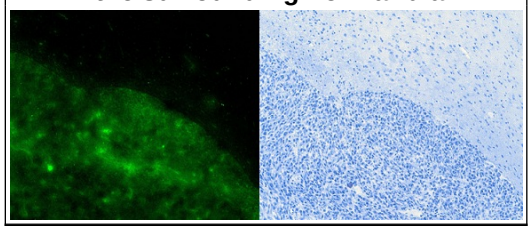
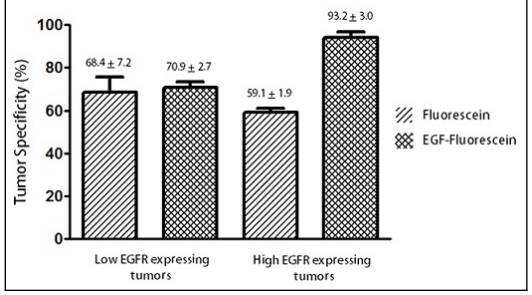


Figure 3: Bar graphs showing high EGFR-expressing tumors to demonstrate increased tumor specificity for targeted EGF-FL compared to nonspecific fluorescein ($p < 0.0001$)



Conclusions

EGF-FL is a highly specific fluorophore for the delineation of EGFR-expressing tumors and has comparable efficacy to conventional fluorescein in tumors with low EGFR expression. Due to the established safety profile of fluorescein and the ease of administration, EGF-fluorescein has direct clinical applicability for use in fluorescence-guided resections and requires further study.

Learning Objectives

By the conclusion of this session, participants should be able to: 1) Describe the utility of tumor-specific intraoperative fluorescent agents, 2) Discuss, in small groups, the efficacy and limitations of currently used fluorophores, 3) Understand the future implications of the current study.

References

1. Babu R, Adamson C: Fluorescence-Guided Malignant Glioma Resections. Curr Drug Discov Technol. 2012;9(4):256-67