**Supplementary Information**

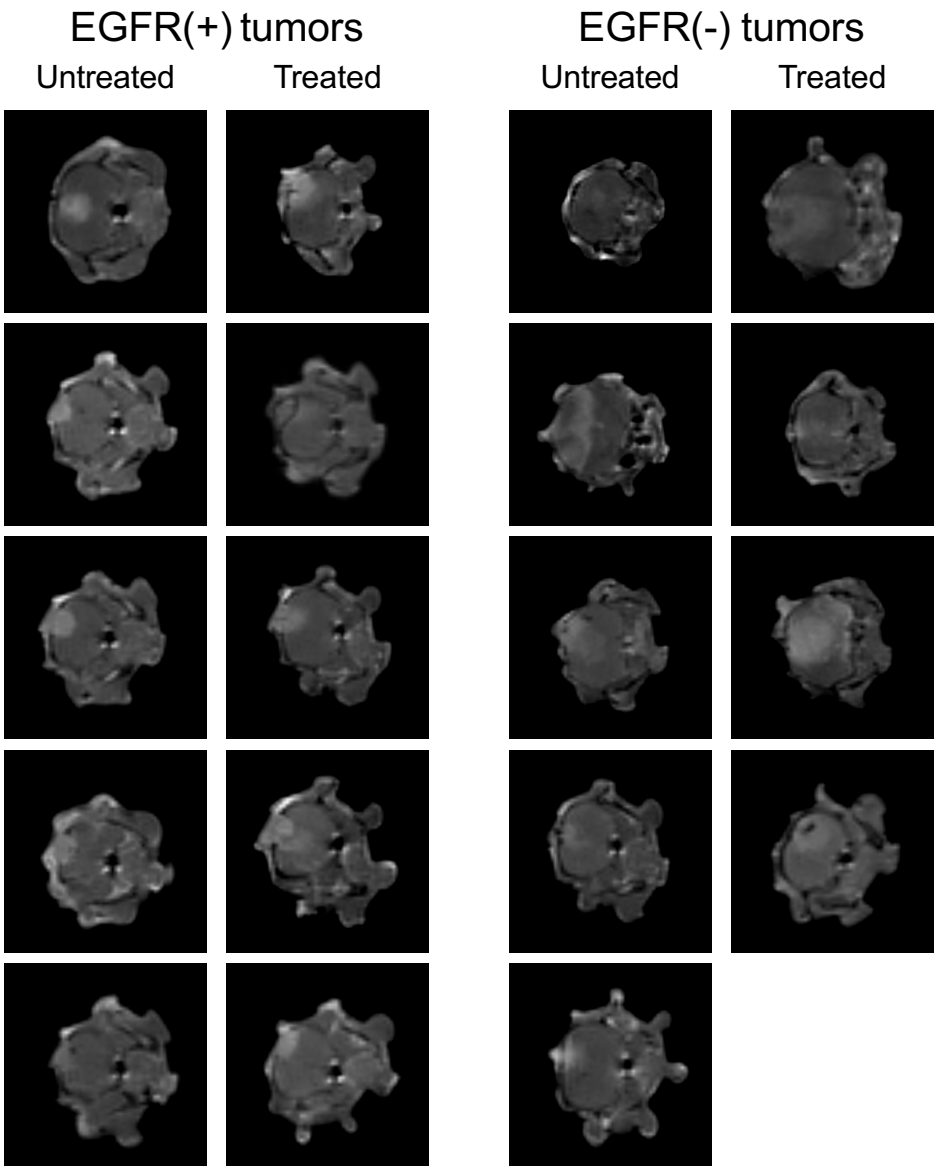


**Figure S1**. Head scans of gadolinium-enhanced T1-weighted MR images for each animal included in the study (one slice per mouse shown). Indentations in the tissue show the locations of the optical fibers.

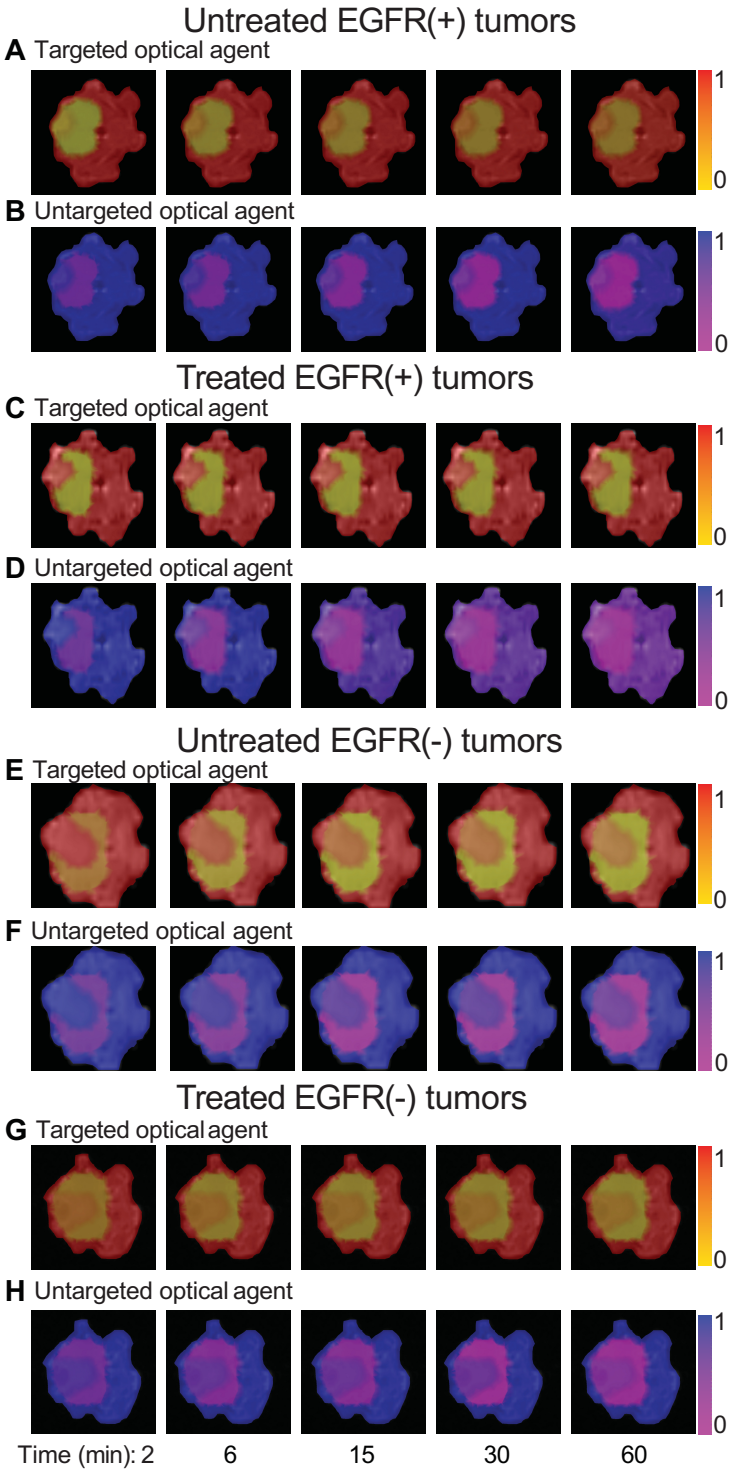


**Figure S2**. Recovered values of fluorescence yield (targeted and untargeted channels) overlaid on MRI scans for four animals (one from each group). All reconstructions were done using the hard-priors technique in 3-D and a slice extracted for visualization. Selected frames over the sequence are shown. Values from the tumor were extracted and used in the paired-agent models to determine RA.

***Testing data distribution normality:***

To confirm the RA data follow a normal distribution, we performed a Shapiro-Wilk test as well as a Q-Q (quantile-quantile) plot. Shapiro-Wilk test statistics and p-values are shown in Table S1, and indicate that all cohorts show p-values above the alpha level ($\alpha = 0.05$). Thus, the test was unable to reject the null hypothesis that the data populations are normally distributed. The Q-Q plot is provided in Figure S1 and shows strong similarity between experimental distributions and theoretical Gaussian distributions for all cohorts (Figure S1).

**Table S1**. Shapiro-Wilk normality test of RA distributions for each experiment cohorts

| **Shapiro-Wilk test** | **EGFR(+) Treated** | **EGFR(+) Untreated** | **EGFR(-) Treated** | **EGFR(-) Untreated** |
|---|---|---|---|---|
| **W** | 0.8737 | 0.9039 | 0.8311 | 0.9620 |
| **P value** | 0.2816 | 0.4320 | 0.1706 | 0.8220 |
| **Passed normality test (alpha=0.05)?** | Yes | Yes | Yes | Yes |

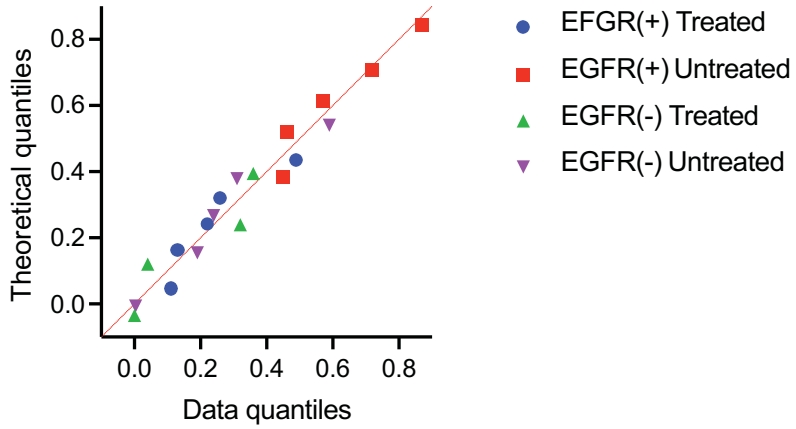


**Figure S3**. Normal Q-Q plot of RA distributions.

***Further analysis of Confidence Intervals:***

To correct for familywise error rate in multiple comparison procedure (MCP), a Tukey post hoc correction was applied for one-way ANOVA analysis. Test result indicates statistically significant in one-way ANOVA test. Assuming normal distribution, 95% Confidence Interval analysis with Tukey correction of differences in the means further confirms the test results (Figure S2)

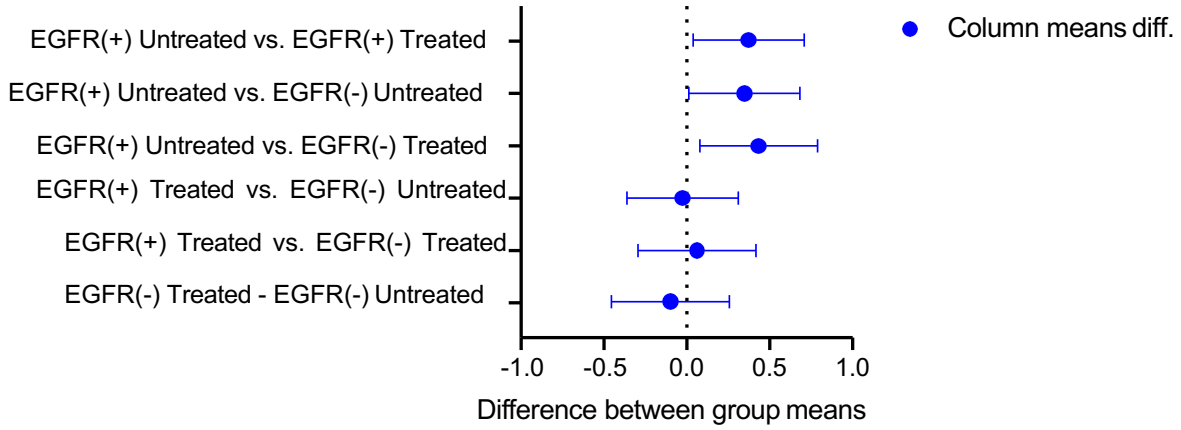


**Figure S4**. 95% Confidence Interval plot showing the difference between group means for multiple comparisons.