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Supplemental Material to:

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Delineating the functional map of the interaction between nimotuzumab and the epidermal growth factor receptor

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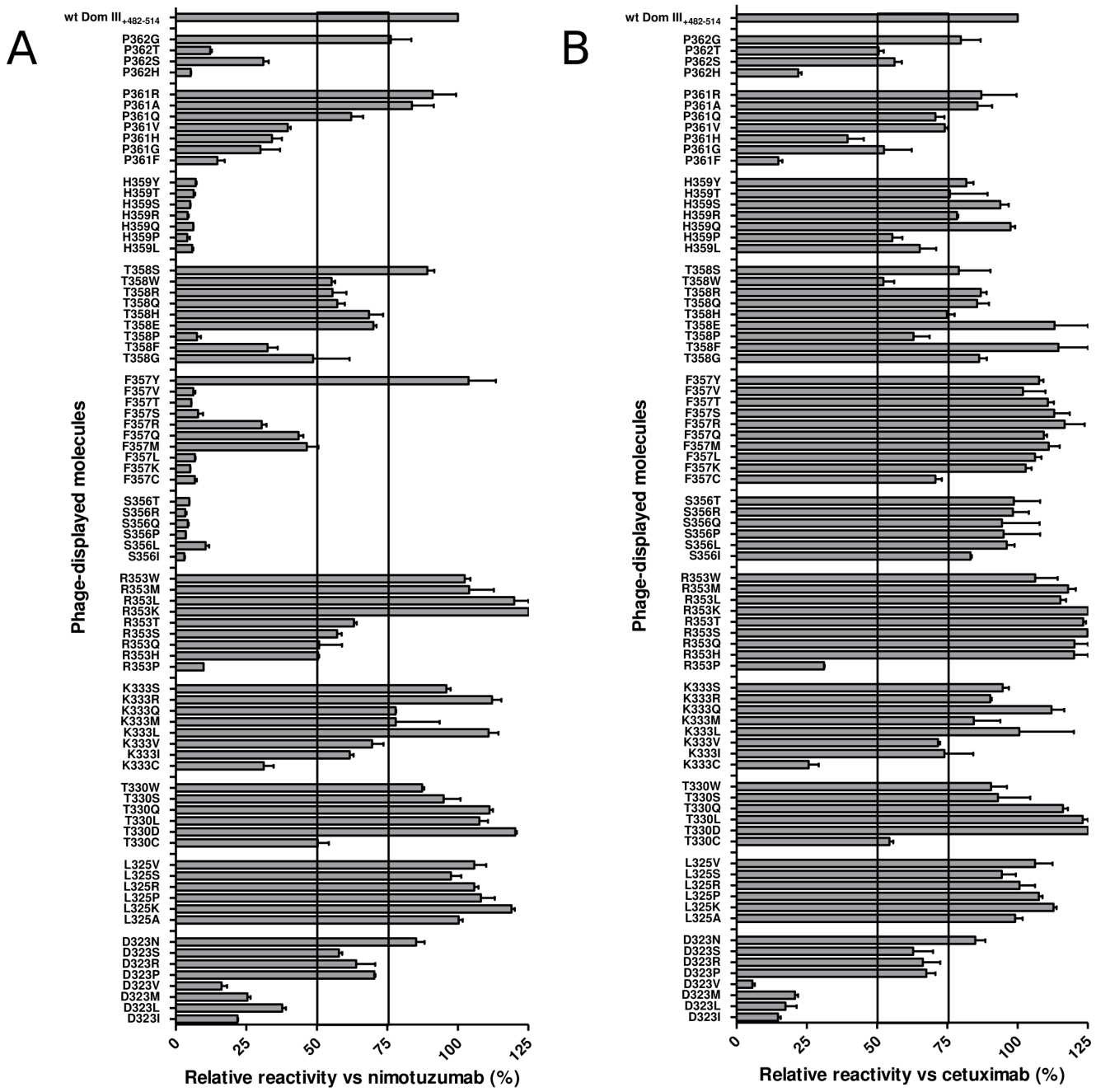

Figure S1: **Recognition of phage-displayed EGFR domain III variants with mutations in the antigenic region recognized by nimotuzumab**. Phages displaying EGFR Dom $III_{+482-514}$ mutated variants with replacements within the solvent-exposed area surrounding the critical residue H359 were produced at a 50 ml scale. Phage-displayed wt Dom $III_{+482-514}$ was included as a control. Purified phages (10^{12} viral particles/ml) were incubated on microtiter plates coated with either anti-EGFR mAbs (nimotuzumab (A) and cetuximab (B)) or the anti-*c-myc* tag 9E10 mAb. Bound phages were detected with an anti-M13 mAb conjugated to horseradish peroxidase. Normalized reactivity for each variant was estimated by dividing the signal obtained with each mAb by the reference signal (measured with the anti-tag mAb). Relative reactivity (%) was calculated as the ratio between normalized reactivity of each variant and that of wt domain III. Lines indicate 50% and 75% or relative reactivity.

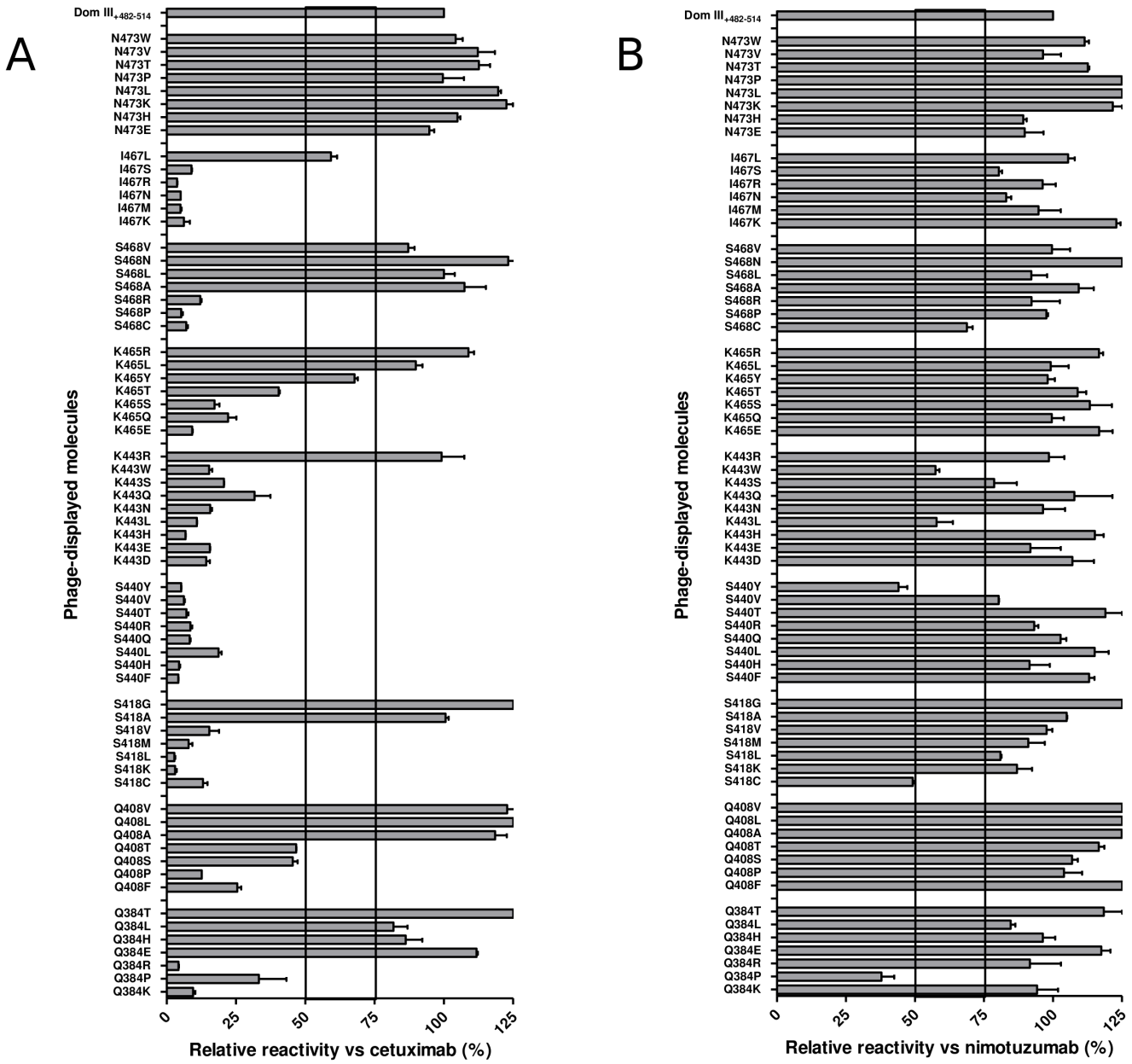

Figure S2: **Recognition of phage-displayed EGFR domain III variants with mutations within the cetuximab structural epitope**. Phages displaying EGFR Dom III$_{+482-514}$ mutated variants with replacements within the cetuximab structural epitope were produced at a 50 ml scale. Phage-displayed wt Dom III$_{+482-514}$ was included as a control. Purified phages (10^{12} viral particles/ml) were incubated on microtiter plates coated with either anti-EGFR mAbs (cetuximab (A) and nimotuzumab (B)) or the anti-*c-myc* tag 9E10 mAb. Bound phages were detected with an anti-M13 mAb conjugated to horseradish peroxidase. Normalized reactivity for each variant was estimated by dividing the signal obtained with each mAb by the reference signal (measured with the anti-tag mAb). Relative reactivity (%) was calculated as the ratio between normalized reactivity of each variant and that of wt domain III. Lines indicate 50% and 75% or relative reactivity.