The therapeutic activity of the epidermal growth factor receptor (EGFR)-directed monoclonal antibody cetuximab in gastric cancer is currently being investigated in clinical studies. Reliable biomarkers for the identification of patients who are likely to benefit from this treatment are not available. In this study, we assessed the activity of cetuximab in five gastric cancer cell lines (AGS, AZ521, Hs746T, LMSU and MKN1). The viability of two of these cell lines, AZ521 and MKN1, was significantly reduced by cetuximab treatment. High expression and secretion levels of the EGFR-binding ligand, amphiregulin (AREG), were associated with cetuximab responsiveness. MET activation and mutations in Kirsten-Ras gene (KRAS) were associated with cetuximab resistance. By introducing a hierarchy between these markers, we established a model that facilitated the correct classification of all five gastric cancer cell lines as cetuximab responsive or non-responsive. The highest priority was allocated to activating KRAS mutations, followed by MET activation and finally by the levels of secreted AREG. In order to validate these results, we used three additional human gastric cancer cell lines (KATOIII, MKN28 and MKN45). In conclusion, we propose that our model allows the response of gastric cancer cell lines to cetuximab treatment to be predicted.