

# Northeast Structural Genomics Consortium Primer Prim'er – PCR Primer Design Software

[http://www.nesg.org/primer\\_primer](http://www.nesg.org/primer_primer)

Primer Prim'er (Everett et al., 2004) is a PCR primer design tool that completely automates the primer design process. Primer Prim'er generates vector specific PCR primer sets designed to amplify and insert DNA targets into laboratory vectors. Both restriction endonuclease and viral recombination cloning strategies are supported. It is implemented under Adobe's flash player

Primer Prim'er possesses a rich interactive interface designed to be both a teaching tool as well as a powerful tool for structural genomic efforts (Figure 1). The software introduces endonuclease restriction and viral recombination sites into calculated primer sets and adds additional nucleotides in order to preserve frame with vector based fusions. Primer Prim'er is very customizable. Users can readily define and use any set of vectors, select from a variety of annealing temperature formulas, design mutagenic primer sets and automatically create series of primer sets designed to vary the boundaries of targeted regions of DNA. Calculated primer sets are reported in both a graphical format and several text based formats for ease of ordering. Calculated primer sets can be sorted upon a variety of criteria such as expected PCR product length and agarose gels can be simulated to demonstrate how the expected PCR products would appear in the laboratory.

The software has been used by the NESG protein sample production pipeline (Acton et al., 2005) to design PCR primers for more than 20,000 expression vectors.

Everett, J.K.; Acton, T.B.; Montelione, G.T. *J. Struct. Funct. Genomics* 2004, 5: 13-21. Primer Prim'r: A web based server for automated primer design.

Acton, T.B.; Gunsalus, K.C.; Xiao, R.; Ma, L-C.; Aramini, J.M.; Baran, M.C.; Chiang, Y-W.; Climent, T.; Cooper, B.; Denissova, N.; Douglas, S.M; Everett, J.K.; Ho, C.K.; Macapagal, D.; Paranjji, R.K.; Shastry, R.; Shih, L-Y.; Swapna, G.V.T.; Wilson, M.; Wu, M.; Gerstein, M.; Inouye, M.; Hunt, J.F.; Montelione, G.T. *Meth. Enzymology* 2005, 394:210-243. Robotic cloning and protein production platform of the Northeast Structural Genomics Consortium.

The screenshot displays the Primer Prim'er web interface. On the left is a navigation menu with options: Home, Start, Vectors, Options, Help, and Target control. The Target control section includes buttons for 'previous target', 'next target', and 'automate'. The main area shows a target named 'WR2' with target number '2 / 3'. Below this, four vector options are listed: pET 14-15A, pET 14-15B (selected), pET 14-15C, and pET 14-15d. The selected vector, pET 14-15B, is shown with a graphical representation of the DNA sequence. The 5' site is EcoRI and the 3' site is HindIII. The N nmr sequence is S H M A N S and the C nmr sequence is -. The interface also shows restriction sites (NdeI, EcoRI, BamHI, SacI, Sall, HindIII, XhoI) and a legend for 5' and 3' restriction site markers.

**Figure 1.** Primer Prim'er possess a rich interactive interface. The non-native amino and carboxy terminal that will be expressed dynamically update as the user selects different cloning sites by clicking and dragging the 3' and 5' site markers.