context dependence. Perhaps, in this convoluted sequence space, context-dependent positions are in some way related to intragenic suppressors.

Regardless of context, these experiments show high gain factors, whether we are grouping residues along one face of a helix or walking down a sequence in a blindly contiguous manner. Stemmer calculates that in using REM, "For a 285-residue protein, like ß-lactamase, the fraction of all possible amino-acid combinations present in the initial library is only 3 x 10^{-363}." He also calculates (independent of method used), that 10^{300} universe masses are required to synthesize all possible proteins of this length.[1] GA-based technologies are by their very nature sparse searches. Algorithmically, this is their power—to converge on a solution without searching the entire space. The important experimental point is that such sparse searches utilize physically realistic syntheses. In this regard, all GA-based technologies are very similar: REM, EEM, and GA-PCR "learn" from their initial sparse search and then generate interesting new proteins within a few iterations.

Which GA-based technology is best? That probably depends on the protein and the specific engineering goal. One advantage of REM is its synergistic interaction with the structure of the genetic code[4,5,8,16] having to do with hydropathy and molar volume, important determinants of protein structure and function. The "highly impractical,[1]" labor intensive aspects of EEM are lessened by new PCR methodology,[17] laboratory automation, and high throughput screening. However, given the fact that the field of combinatorial chemistry is still in its infancy, it is probably wise to consider all of the proven mutagenesis methods: GA-PCR, REM and its relatives, error-prone PCR, parsimonious mutagenesis,[18] and structurally guided or sequential random mutagenesis.[19]

All of these methods are currently being compared in my laboratory, using the green fluorescent protein as a model, with the goal of finding derivatives with novel spectroscopic properties.[17]

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Comments and opinions on editorials, articles, and research in Bio/Technology *are welcome. Letters to the editor may be addressed to: 345 Park Ave. South New York, NY 10010 fax: 212 696 9635 (e-mail: m.ginsberg@natureny.com).*

Errata:

In last month's issue, the incorrect picture appeared in the sidebar on p. 659 of "Protein Modeling by E-mail." The correct picture and caption are shown below.

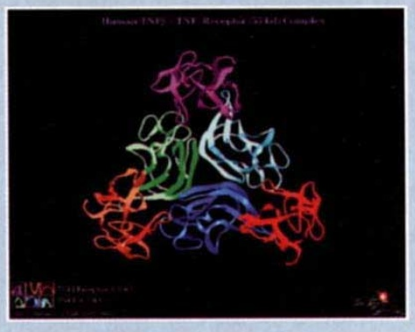

FIGURE 1. Sample Swiss-3DImage: The human tumor necrosis factor (TNF)-receptor complex.