

Enzyme Engineering: Old and New Approaches

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Editorial

Enzymes, as biocatalyzers, are proteins that allow that a great number of biological reactions take place at rate much quicker than in its absence. These molecules show high specificity for the recognition of the substances to be transformed and they have evolved during thousands of years to make possible a wide variety of transformations related to all forms of life. In this line, a first and primitive wave of enzyme engineering was developed as a way to use enzymes in industrial processes. One of the first fields was food industry. Microbial and plant enzymes, such hydrolases or pectinases were used in food preparation, such as bread, beer, milk derivatives or juice preparation. Other industries, as detergents, waste treatment in bioremediation or pharmaceutical synthesis have also developed in parallel with the use of enzymes as much specific, efficient and stable as possible [1].

In order to purify enzymes for the improvements of all processes, the techniques for enzyme extraction and purification from natural sources have been designed in very careful ways to get better preparations adapted to the processes, and this in a branch of the biotechnology well developed during the last two decades of the XX century. Other strategies toward the design of improved enzymes and enzymes with new activities using novel scaffolding and computational models to design or improve performance of multi-enzyme systems are actively developed [2,3].

On the other hand, during the last XXI century, enzyme engineering is opened to a new concept of biotechnology within the context of genome engineering and expansion of the natural genetic code. This is one of the areas of knowledge from which great expectations are constantly arising in order to complement or even to avoid chemical methods for protein modification. The addition of new chemical and biological properties to enzymes is recently pursued through the incorporation of non-canonical amino acids analogs into proteins during natural protein synthesis in cell-free systems. These new methods are promising economical and convenient tools for the screening of a high number of amino acid analogs in a variety of protein targets for the study of functional improvements of enzymes in comparison to the currently employed natural ones [4]. Alternatively, sense codon reassignment is a very promising tools, using the pyrrolysine system. Unlike most aminoacyl-tRNA synthetases, the synthetase for this rare 22nd proteinogenic amino acid shows low

selectivity towards its substrate and low selectivity towards the anticodon of tRNA. This allows the pyrrolysine machinery to be easily used for the genetic incorporation of non-natural amino acids or even α -hydroxy acids into proteins at the amber stop codon [5]. This codon was also target for an engineered system for specific cotranslational phosphoserine incorporation into any desired position in a protein by an *Escherichia coli* strain. This system could be very promising in molecular biology and disease research as phosphoserine is the most abundant amino acid in the eukaryotic phosphoproteome after natural post-translational modification [6].

Finally, in this context of genetic code expansion, a different approach is the sense codon emancipation using some low-usage codons, as the isoleucine AUA, for liberation from its natural decoding function [7].

In sum, these and other similar approaches might allow us hopefully the design of new enzymes with a number of new designers amino acids to optimize the catalytic properties and stability of a great variety of enzymes to improve the efficiency of industrial and biosynthetic processes, open new frontiers to classical enzyme engineering.

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