Main structural targets for engineering lipase substrate specificity

ABSTRACT

Microbial lipases represent one of the most important groups of biotechnological biocatalysts. However, the high-level production of lipases requires an understanding of the molecular mechanisms of gene expression, folding, and secretion processes. Stable, selective, and productive lipase is essential for modern chemical industries, as most lipases cannot work in different process conditions. However, the screening and isolation of a new lipase with desired and specific properties would be time consuming, and costly, so researchers typically modify an available lipase with a certain potential for minimizing cost. Improving enzyme properties is associated with altering the enzymatic structure by changing one or several amino acids in the protein sequence. This review detailed the main sources, classification, structural properties, and mutagenic approaches, such as rational design (site direct mutagenesis, iterative saturation mutagenesis) and direct evolution (error prone PCR, DNA shuffling), for achieving modification goals. Here, both techniques were reviewed, with different results for lipase engineering, with a particular focus on improving or changing lipase specificity. Changing the amino acid sequences of the binding pocket or lid region of the lipase led to remarkable enzyme substrate specificity and enantioselectivity improvement. Site-directed mutagenesis is one of the appropriate methods to alter the enzyme sequence, as compared to random mutagenesis, such as error-prone PCR. This contribution has summarized and evaluated several experimental studies on modifying the substrate specificity of lipases.

Keyword: Lipase; Specificity; Binding pocket; Lid; Oxyanion hole; Protein engineering; Chemoselectivity; Regioselectivity; Stereoselectivity