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山手3号館2階共通セミナー室

第4回 岡崎発動分子科学セミナー

Accurate high-throughput screening based on digital protein synthesis in a massively parallel femtoliter droplet array



国立研究開発法人海洋研究開発機構
超先鋭研究開発部門研究員

張 翼 博士

This presentation reports a general strategy based on Poisson statistics that enables a rapid and efficient acquisition of enzyme mutants from just a few clones within a day. We prepared a high-density femtoliter droplet array (FemDA) consisting of one million uniform droplets per 1 cm² to carry out high-throughput protein synthesis and screening. Single DNA molecules from a mutagenesis library were randomly distributed into each droplet following a Poisson process to initiate the protein synthesis, and then can be recovered by a microcapillary. The protein yield in each droplet was proportional to the number of DNA molecules, meaning that droplets with apparent intensities higher than the Poisson distribution-predicted maximum can be readily identified as the exact hits exhibiting the desired increased activity, rather than a co-encapsulation of multiple DNA molecules. This unique strategy combining the miniaturized device technology and the mathematical modeling enabled cost-effective screening of several kinds of novel alkaline phosphatases with remarkably improved activities from just a few droplets using less than 10 nL reagents.

連絡先：飯野亮太（内線5230） 協賛：新学術領域研究「発動分子科学」
<http://www.molecular-engine.bio.titech.ac.jp/>



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