Multiplex barcoded Paired-End ditag (mbPED) library construction for ultra high throughput sequencing.

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摘要

Multiplex barcoded Paired-End Ditag (mbPED) library construction for ultra high throughput sequencing is disclosed. The mbPED library comprises multiple types of barcoded Paired-End Ditag (bPED) nucleic acid fragment constructs, each of which comprises a unique barcoded adaptor, a first tag, and a second tag linked to the first tag via the barcoded adaptor. The two tags are the 5'- and 3'-ends of a nucleic acid molecule from which they originate. The barcoded adaptor comprises a barcode, a first polynucleotide sequence comprising a first restriction enzyme (RE) recognition site, and a second polynucleotide sequence comprising a second RE recognition site and covalently linked to the first polynucleotide sequence via the barcode. The two REs lead to cleavage of a nucleic acid at a defined distance from their recognition sites. The length of the adaptor is set so that the bPED nucleic acid fragment fits one-step sequencing.

智財權狀態

美國US 8.481.699 B2已獲證、美國臨時案已申請

技術優勢

This is a sweet, creative design for making paired-end diTag library with barcode inserted in the adaptor position in the fragment library. The clone of "barcoded paired-end ditags (bPETs)" is constructed prior to libribary construction, thus the cloning process is quite flexible. Using the barcode as recognition signature, multiplex paired-end diTag (mPET) library could be created by mixing multiple bPET library together. In the end, ultra high throughput sequencing can be achieved with reduced sequencing running time and cost. Compared to current sequencing technology, the advantages of this novel sequencing approach include: to simplify experimental procedure for PEmediated investigations to reduce sequencing running cost to reduce sequencing time to increase sequencing throughtput

應用範圍

Worldwide DNA sequencing and proteomics markets are expected to rise to \$17.5 billion in 2009. The novel sequencing approach is advantagous in the aspect of ultra high throughput sequencing, therefore it would have great market potentials.

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