REGIONAL CENTRE FOR BIOTECHNOLOGY

Seminar series

Gene Targeting by ZFNs and TALENs in zebrafish

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Seminar Room

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Abstract

Zinc Finger Nucleases (ZFNs) and Transcription Activator-Like Effector Nucleases (TALENs) provide powerful platforms for genome editing in plants and animals. Both ZFNs and TALENs are composed of the nuclease domain from the FokI endonuclease fused to a tandem array of DNA-binding domains that can be engineered to recognize and cleave at desired sequences in the genome. ZFNs can be constructed from zinc finger modules with predefined specificities but the lack of reliable modules results in low success rate of ZFNs. We employed bacterial-one-hybrid based selections to obtain zinc finger modules that improved the DNA binding specificities of ZFNs resulting in a higher success rate of ZFNs. Employing these modules, ~95% protein-coding genes can be targeted by ZFNs. However, the newly described TALEN platform for gene editing surpasses ZFNs in not only being more successful but also being able to target virtually any DNA sequence. Employing TALENs and ZFNs, we created two double stranded breaks on the same chromosome simultaneously using two pairs of nuclesases allowing deletion of large genomic regions of upto 5.5 Mb in zebrafish. The ability to efficiently delete genomic segments in a vertebrate developmental system will facilitate the study of functional non-coding elements on an organismic level.