

## TNT-cloning System

### Disclosure Number

201403357

### Technology Summary

Our technology describe a novel synthetic biology tool, called TNT-cloning, that in vitro assembles DNA parts in a simple, fast, efficient, flexible and, if desired, automated manner. Our system combines all cloning elements into one single universal library allowing a pre-determined assembly without the need for linkers/adaptors, which ultimately creates a scar-free product. Also, there are no sequence homology requirements and up to 3 fragments can be cloned at once in as little as one hour using our optimized TNT-buffer. The binary plasmids support secondary and tertiary assembling, which require minimal re-cloning of fragments and makes the final construct reusable as well as ready for plant transformation. Because this technique is compatible with isothermal (Gibson) assembly, virtually any fragment can be used as an element in the library and circularized without the need to carry the binary-backbone, expanding the technology use to other systems. This novel cloning platform will greatly facilitate the engineering of different pathways as well as provide different fields with a malleable and trustworthy cloning system.

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