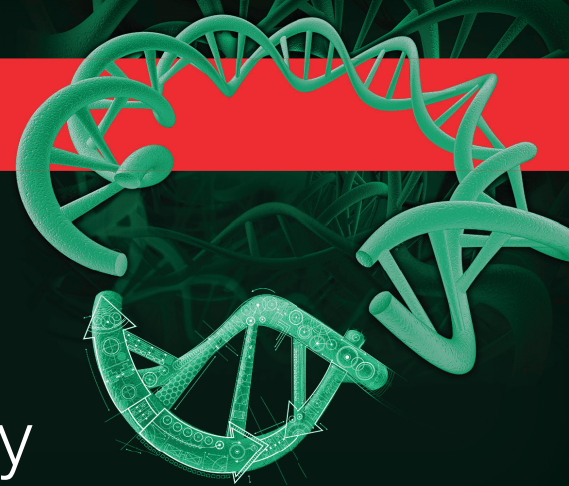


GeneArt Elements Vector Construction and Combinatorial Parts Assembly



The **Invitrogen™ GeneArt™ Elements™ Vector Construction** parts collection comprises a growing subset of biologically well-characterized parts with defined sequences and functionalities, such as promoters, terminators, enhancers, operators, open reading frames, and more. The GeneArt Elements parts provided can be combined with custom parts to design individualized vectors.

- **Flexible**—GeneArt Elements parts can be seamlessly assembled with minimal design restrictions
- **Reliable**—all offered parts are 100% sequence-verified
- **Easy to use**—an intuitive CAD-like software within the Invitrogen™ GeneArt™ portal will assist you with your vector design

The Invitrogen™ GeneArt™ Elements™ template breaks down five commonly used vectors (e.g., pcDNA3.1) into their functional parts (Figure 1). Simply build your vector of choice by adding, deleting, or substituting DNA parts.

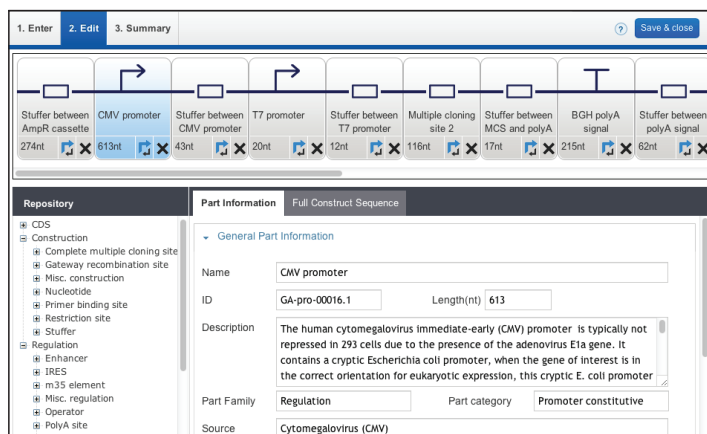


Figure 1. Screen shot of the GeneArt Elements Vector Construction template.

To demonstrate the flexibility of the offering, two vectors were made that contain green fluorescence protein (GFP) with different secretion leaders: IgK and tPA. HEK 293 cells were transiently transfected with the indicated constructs, and the conditioned media from the transfected cells were subjected to western analysis with an anti-GFP antibody (Figure 2). This shows that the GeneArt Elements Vector Constructions were functional, generating positive controls for protein secretion.

How to order GeneArt Elements Vector Construction

GeneArt Elements Vector Construction can be ordered online via the GeneArt portal. Conveniently drag and drop GeneArt Elements parts from the repository and add custom parts as needed to create the vector of your choice.

Ordering is also available via an Excel™ order form at thermofisher.com/geneartelementsvc

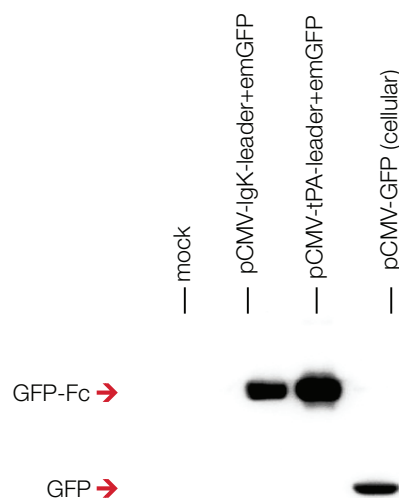


Figure 2. Western blot showing protein expressed from pCMV-GFP vectors constructed using GeneArt Elements Vector Construction. The proteins in conditioned media were probed with an anti-GFP antibody.

GeneArt Elements Combinatorial Parts Assembly (CPA) is a way to combine predefined DNA parts (promoters, ribosomal binding sites, open reading frames, terminators, etc.) in order to build a diverse set of larger constructs (Figure 3). Users simply provide individual part sequences (custom or predefined) and progression of the parts within the order form. The final construct is synthesized, with sequence junctions and reading frame all controlled seamlessly by the assembly.

All conceivable part combinations can be created to build and test new metabolic pathways or a variety of expression cassettes to identify the most valuable combination for your research needs.

- **Tested for accuracy**—all CPAs that are delivered as separate constructs are sequenced as part of our ISO 9001:2008–certified quality management system; we only ship constructs that are 100% verified
- **Cost-effective**—DNA parts have the potential to be used multiple times in the final constructs
- **Comprehensive**—all permutations of the available genetic elements are possible

To demonstrate this concept, we generated a combinatorial library composed of a small set of yeast promoters and corresponding terminators from the GeneArt Elements part collection to analyze relative luciferase expression levels. In this experiment, the highest firefly luciferase expression was shown using a TEF1 promoter/TEF1 terminator combination (Figure 4). This demonstrates the proof of concept and can be extrapolated to test other functional elements, such as in metabolic pathways.

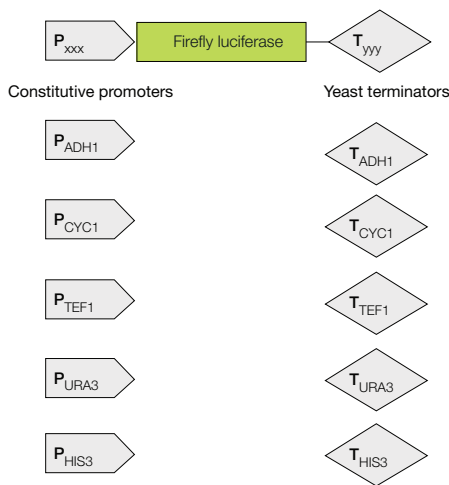


Figure 3. Yeast constitutive promoters and terminators from the GeneArt Elements CPA collection used for creation of an example combinatorial promoter/terminator library.

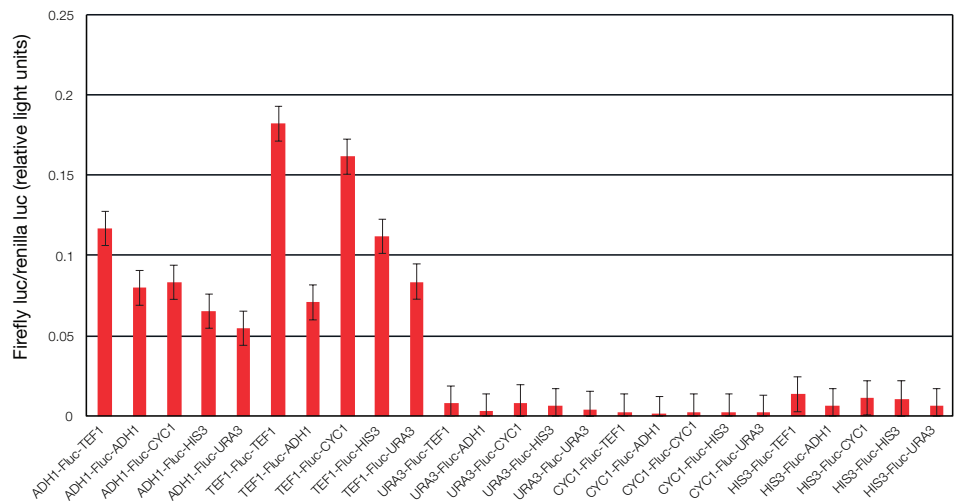


Figure 4. Twenty-five different promoter/terminator combinations were created to control firefly luciferase reporter expression. Reporter expression levels were measured and normalized against cotransformed renilla luciferase driven by a standard promoter.

How to order GeneArt Elements Combinatorial Parts Assembly

Fill out the Excel order form available at thermofisher.com/genartelemntscpa with relevant DNA part information, order, and sequence, and send it to geneartsupport@thermofisher.com

Find out more at thermofisher.com/genartelemntsvc or thermofisher.com/genartelemntscpa

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