GENCEFE

Synthetic DNA Libraries

The synthetic DNA library is a combination of a large number of DNA mutant sequences constructed to screen mutant proteins with specific functions. It has been widely used in research fields such as protein directed evolution and antibody screening. GenCefe Biotech's experienced R&D and production team relies on the advanced AI bioinformatics platform and gene synthesis technology platform to provide customized synthetic DNA library services. We can mutate the amino acid or nucleotide sequences specified by customers, or design and synthesize mutation libraries according to customer requirements, such as site-directed mutagenesis libraries, random mutant libraries, degenerate mutation libraries, controlled libraries, sgRNA libraries and other types of library construction services.

Our Services

- Site-directed mutagenesis libraries
- Random mutant library
- Degenerate mutation libraries
- Controlled libraries
- sgRNA libraries

Service Features

- **Advanced technology platform:** Experienced R&D and production teams, leading bioinformatics and gene synthesis technology platforms, provide you with high-quality library design and synthesis services;
- **Comprehensive library types:** site-directed mutagenesis libraries, random mutant libraries, degenerate mutation libraries, controlled libraries, sgRNA libraries and other types of library construction services;
- **Professional technical support:** provide considerate pre-sales technical consultation and after-sales service, and timely update project progress.

Site-Directed Mutagenesis Libraries

Site-directed mutagenesis libraries refer to a group of gene sequences with mutated amino acids formed by replacing one or more amino acids encoding proteins or peptides with specified amino acids. Each gene sequence in the library generally carries only one mutation, and different gene sequences with different mutations can be delivered as separate clones or in the form of a mixture.

Site-directed mutagenesis libraries are representative of alanine/glycine scanning libraries, which means that each amino acid in the coding protein or peptide sequence is replaced with alanine or glycine one by one, and only one position of each mutant gene is replaced. If it is a protein containing 200 amino acids, 200 mutant genes will be generated, and each mutant gene will be replaced by alanine or glycine at positions from 1 to 200. The library will be delivered as 200 individual clones or as a pool of 200 genes upon request.

Alanine Scanning Library

Recommended Applications

- Research on protein structure and function
- Study of the enzyme active sites
- Study on the antibody binding domains
- Protein property modifications, such as changing thermal stability, substrate binding specificity, etc.

Service Specifications

Delivery Format	Advantage	Turnaround Time
Single Clone	▪ The sequence of each clone is clear ▪ No loss in library replication Suitable for mutations of a few amino acid positions or replacement of multiple amino acid positions with a single amino acid	10-15 business days
Mixed Library	▪ Applicable for larger library capacity ▪ Cost effective ▪ Reduce the screening efforts	10-15 business days

Deliverables

- For single clone: 5μg plasmid containing the mutated gene for each clone
- For mixed mutation library: >100μg plasmid or plasmids with more than 100 times the library capacity
- Sequence verification information
- Statistical analysis of base distribution of mutation sites

Random Mutant Library

The random mutation library refers to the random introduction of nucleotide mutations in the gene sequence by a specific method, thereby generating a mutant gene mixture containing random nucleotide mutations. Each gene molecule in this mixture contains a certain number of random mutation points in a designated region. To make the results analyzable, usually each gene contains an average of <20/kb mutation points. Depending on the length of the gene and the average number of mutations, the theoretical library capacity is also different. The random mutation library can not only randomly mutate the full length of the gene, but also randomly mutate a certain region of the full-length gene. The purpose of random mutagenesis in a region is to focus on this region and can greatly reduce the theoretical library capacity and thus reduce subsequent screening efforts.

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AATGGTAGATAGCGGGCTGGCTGAAAATTAGATCCCAGAGATGCGCTCGAGATAGGGCTCGCTGGGATATTTACACATGCCCGATAGTAGATAGAGGG
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Random Mutant Library

Recommended Applications

- Research on protein structural functional domains;
- Antibody humanization;
- Protein property modifications, such as the determination of the catalytic domain of the enzyme, the improvement of catalytic properties, etc.

Service Specifications

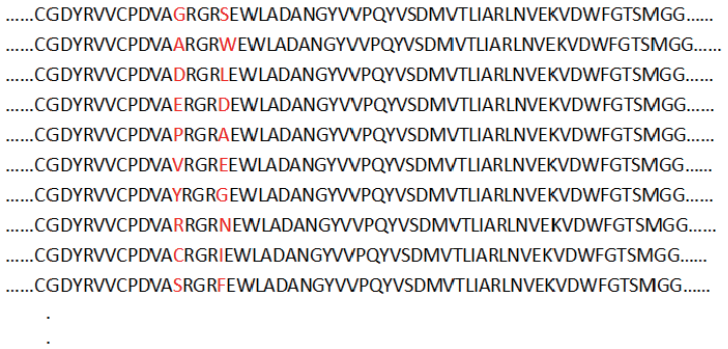
Services	Description	Turnaround Time
Gene synthesis	Refer to gene synthesis service specifications	5-8 business days
Vector construction	Clone the synthetic gene into the specified vector	2-3 business days
Random mutation	The mutation region length is 200-1,500 bp, and we provide three different types of mutation specifications. ▪ 1-4 mutations/kb ▪ 5-10 mutations/kb ▪ 11-20 mutations/kb	2-3 business days
Cloning	Delivered as plasmid:10^4-10^7 library capacity Delivered as PCR products: up tp10^{11} library capacity	2-20 business days

Deliverables

- 20-100 μg of plasmid containing the mutated gene
- More than 80% positive rate, and the mutation frequency meets the specified requirements
- The library capacity meets the customer's specified requirements
- At least 20 sequencing results, and the results meet the design specifications

Degenerate Mutation Libraries

A degenerate mutation library, also known as a site-directed saturation library, refers to the introduction of NNK or NNN at single or multiple positions in a gene sequence. After these gene sequences are translated into proteins, mutations containing any of the 20 amino acids are formed at the corresponding positions. Considering all the sequences as a whole, these positions are a gene library containing 20 different amino acids instead of a fixed one among the 20. This kind of gene library is called a degenerate library. The library is often formed by using a mixture of four single nucleotides at the corresponding codon positions during gene synthesis to replace a specific single nucleotide in conventional gene synthesis.



Degenerate Mutation Libraries

Recommended Applications

- Research on protein structural functional domains;
- Directed evolution of proteins, enzymes, antibodies;
- Determination and modification of key amino acids in enzyme active sites;
- Antibody humanization, fine-tuning of antibody affinity and binding properties.

Service Specifications

Services	Description	Turnaround Time
Gene synthesis	Refer to gene synthesis service specifications	5-8 business days
Vector construction	Clone the synthetic gene into the specified vector	2-3 business days
Degenerate mutation	There is no limit to the number of degenerate amino acids, within 45 bp is counted as one. It is usually recommended to have less than 10 degenerate regions. Too many degenerate amino acids will cause the library to not contain all theoretical mutant genes.	2-3 business days
Cloning	Delivered as plasmid:10^4 -10^7 library capacity Delivered as PCR products: up tp10^{11} library capacity	2-20 business days

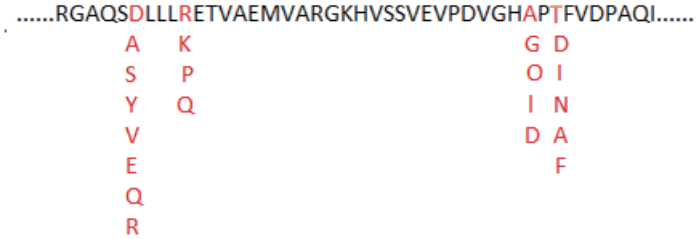
Deliverables

- 20-100 μg of plasmid containing the mutated gene
- More than 80% positive rate, and the mutation position and frequency meet the specified requirements
- The library capacity meets the customer's specified requirements
- At least 20 sequencing results, and the results meet the design specifications

Controlled Library

Controlled libraries, also known as site-directed unsaturation libraries or Trimer libraries, are a class of libraries similar to degenerate libraries. The difference is that the amino acid mutation point of the controlled library is replaced by some of the other 20 amino acids, not all 20. And the ratio of several amino acids to be replaced can be average, or it can be artificially controlled according to a specific ratio. The main purpose of the controlled library is to reduce the theoretical library capacity and remove some high-probability nons ense mutations, thereby reducing the later screening effort. Focus on the sequence of the mutants that are most likely to be obtained, so as to facilitate the in-depth study of mutations of a few amino acids. For example, in-depth study of the effect of a few amino acids around the catalytic site of the enzyme on the catalytic properties of the enzyme.

Controlled libraries are often implemented with Trimer primers. Unlike ordinary primers where the single nucleotides are synthesized one by one, Trimer primers are three nucleotides that are pre-linked into a nucleotide triplet, called Trimer. When synthesizing primers, just connect the corresponding Trimer at the required position. If you need several amino acids to appear at this position, just mix the Trimers of these amino acids and connect them. The ratio of each amino acid at this position in the finished library is controlled by controlling the ratio of each Trimer.



Controlled Libraries

CRISPR sgRNA libraries

CRISPR sgRNA libraries serve as powerful tools for high-throughput genomic screening. By silencing or activating gene expression on a large scale, the link between genes and phenotypes can be established, and the genes responsible for specific phenotypes can be identified. The sgRNA library can replace the previous research method of a single or a small number of genes. By studying a large number of genes at one time, usually more than 10,000 genes, it greatly improves efficiency and saves time and cost. The design and synthesis of sgRNA is simple and convenient. An sgRNA library usually consists of thousands to tens of thousands of sgRNAs designed for different target genes. Multiple sgRNAs can be designed for each gene to improve the success rate and effectiveness of genome editing. CRISPRs sgRNA libraries have been widely used to study diseases or specific phenotypes, such as immunotherapy response, viral infection mechanisms, oncology, etc.

Recommended Applications

- Large-scale gene knockout, activation and suppression
- Identification and verification of new drug targets
- Identification of cancer treatment targets
- Applications in agricultural field, such as development of disease-resistant crops

Service Specifications

Services	Description	Turnaround Time
sgRNA Design	gRNA design targeting specific genes according to customer's requirements	1-2 weeks
sgRNA Library Construction	sgRNA and control synthesis Constructed the sgRNA to designated vector	5-7 weeks
Plasmid Preparation	Transfection grade plasmid preparation	1 week
QC	Extra 20 business days will be needed if QC by NGS is required	Get a Quote

GenCefe Biotech Gene Synthesis and Related Services

GenCefe's experienced R&D and production team has established an advanced gene synthesis technology platform, standardized operating procedures, and a strict quality control system. We provide high-quality customized services including gene synthesis, codon optimization, PCR cloning, subcloning, plasmid preparation, site-directed mutagenesis, and mutation library construction.

Just submit the gene sequence you need, and GenCefe will deliver the desired plasmid to you on time!

Service Features

- **Advanced technology platform:** our team has successfully synthesized various types of difficult sequences, such as repetitive sequences, GC/AT-rich sequences, etc., and delivered the plasmids according to customer-specific requirements
- **Professional technical support:** considerate pre-sales and after-sales services, free codon optimization and project design, timely project updates, and free technical consulting
- **On-time delivery:** experienced production and R&D teams ensure the on-time delivery rate of over 95%
- **Intellectual property protection:** the nucleic acid/amino acid sequence provided by the customer is kept strictly confidential and will not be distributed to third-party in any form

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