TWIST BIOPHARMA

Combinatorial Assembly Libraries

For comprehensive screening of user defined combinations of custom gene parts, such as domains from TCRs, CARs, or antibody VHH/scFv fragments.

Twist's state-of-the-art DNA synthesis platform streamlines the development of advanced cell therapies by enabling the creation of large-scale, highly diverse T-cell Receptor (TCR), chimeric antigen receptor (CAR), and multispecific VHH libraries. Through combinatorial assembly, Twist generates highly uniform screening libraries with precise, user-defined combinations of gene fragments, enabling efficient and comprehensive screening of the desired combinations of custom gene parts, such as domains from TCRs, CARs, or antibody VHH/scFv fragments.

SPECIFICATIONS

- Product format: NGS-verified DNA cloned into the vector of your choice
- Delivery and yield: 100–250 ug of endotoxin-free plasmid DNA
- Price: Project dependent
- Turnaround time: Starting at 8 weeks after custom vector onboarding

KEY BENEFITS

High Diversity

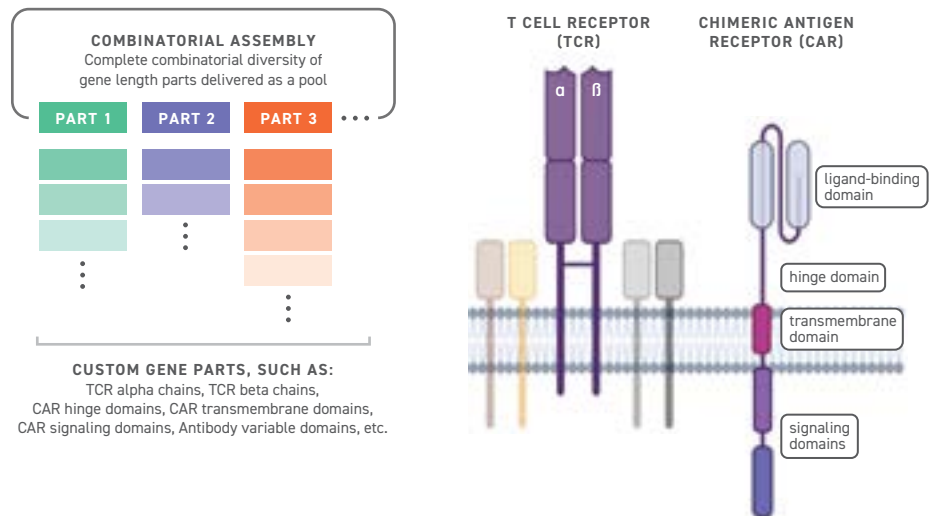- Up to 10,000 gene fragment combinations

High Quality

- NGS-verified libraries with > 90% of possible variants present within 10x of the mean

High Flexibility

- Insert sequences up to 1.5 kb in length

High Throughput

- Libraries designed at scale with diversity across multiple elements of the sequence
- TCR: combinatorial variants across alpha and beta chains
- CAR: combinatorial variants across hinge, transmembrane, and signaling domains
- CAR ligand-binding domains: access to leading-edge in vivo and in vitro technologies to rapidly identify the best scFv and/or VHH candidates against your target of interest

COMBINATORIAL ASSEMBLY
Complete combinatorial diversity of gene length parts delivered as a pool

PART 1   PART 2   PART 3   ...

CUSTOM GENE PARTS, SUCH AS:
TCR alpha chains, TCR beta chains,
CAR hinge domains, CAR transmembrane domains,
CAR signaling domains, Antibody variable domains, etc.

T CELL RECEPTOR (TCR)

α   ß

CHIMERIC ANTIGEN RECEPTOR (CAR)

ligand-binding domain
hinge domain
transmembrane domain
signaling domains

Expedited Cell Therapy Applications

Developing novel TCR and CAR-T cell-based therapies is a particularly challenging task. Antigen recognition for both technologies is dictated by a constellation of amino acids spread across multiple protein domains. Additionally, the downstream effects of antigen-binding is dependent on the composition of each protein's cytoplasmic tail. To hone in on receptors with potential therapeutic applications, researchers need the ability to test a wide range of sequences containing unique combinations of variants in alpha and beta chains, scFV domains, and more. Twist's Combinatorial Assembly Libraries produce large-scale, highly uniform variant libraries in which each custom gene part contains a unique assemblage of gene fragments that are selected by the researcher. This allows researchers to focus their screening on exactly the variants they designed without wasting resources on irrelevant or non-functional variants. Such a library streamlines the process of TCR-T and CAR-T cell development.
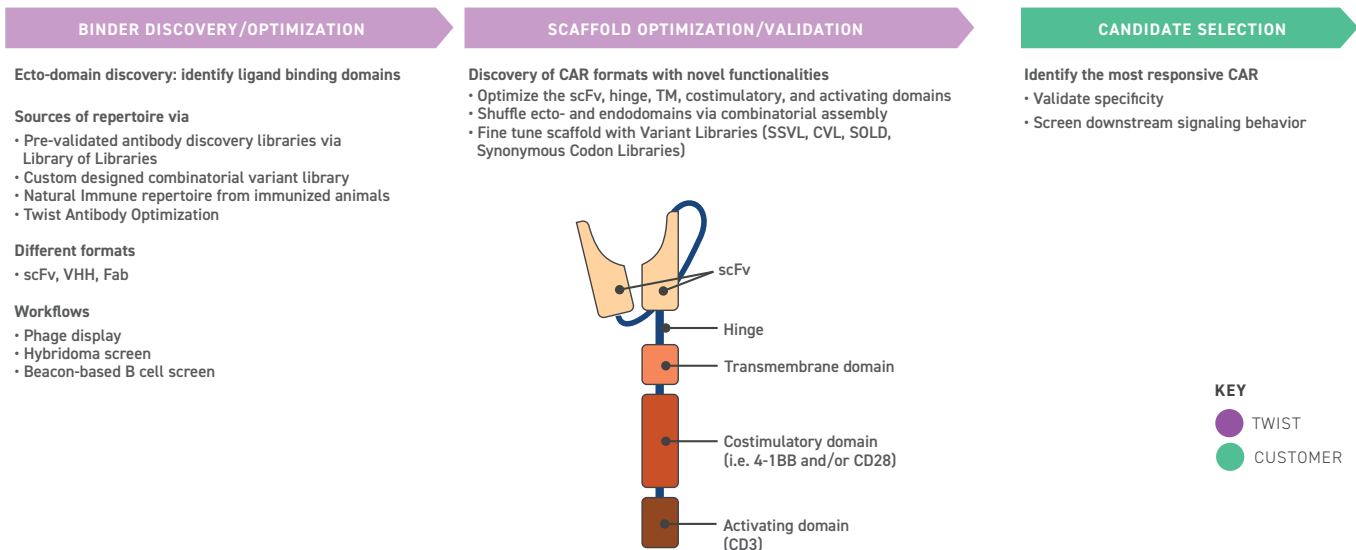
Custom, Rapidly Generated Libraries for High-Throughput Screening & Characterization

Combinatorial Assembly Libraries utilize seamless assembly of multiple variant domains to rapidly create libraries that enable high-throughput screening and characterization of novel and known variants for therapeutic discovery.

TCR Discovery

------- *WHAT TWIST CAN DO FOR YOU* -------

ANALYZE & IDENTIFY | DESIGN / BUILD | TEST

Antigen-presenting cell — T cell
HLA — TCR
Tumor-associated peptides
or peptides derived from infectious disease

- ✓ Rebuild the natural hit repertoire in arrayed and pooled formats
- ✓ Explore additional CDR3 diversity
- ✓ Create stabilized frameworks
- ✓ Optimize frameworks

✓ Hit Validation

Antigen-presenting cell — T cell
HLA — TCR
Tumor-associated peptides
or peptides derived from infectious disease

- ✓ Explore potential pHLA targets
- ✓ Perform TCR binder validation

✓ Hit Validation/Lead selection

Tumor cell — BITE antibody — T cell
Cytokine release

- ✓ Identify novel combinations of target and T cell binding moieties

✓ Scaffold optimization

CAR Discovery and Optimization

BINDER DISCOVERY/OPTIMIZATION

Ecto-domain discovery: identify ligand binding domains

Sources of repertoire via
- Pre-validated antibody discovery libraries via Library of Libraries
- Custom designed combinatorial variant library
- Natural Immune repertoire from immunized animals
- Twist Antibody Optimization

Different formats
- scFv, VHH, Fab

Workflows
- Phage display
- Hybridoma screen
- Beacon-based B cell screen

SCAFFOLD OPTIMIZATION/VALIDATION

Discovery of CAR formats with novel functionalities
- Optimize the scFv, hinge, TM, costimulatory, and activating domains
- Shuffle ecto- and endodomains via combinatorial assembly
- Fine tune scaffold with Variant Libraries (SSVL, CVL, SOLD, Synonymous Codon Libraries)

scFv
Hinge
Transmembrane domain
Costimulatory domain
(i.e. 4-1BB and/or CD28)
Activating domain
(CD3)

CANDIDATE SELECTION

Identify the most responsive CAR
- Validate specificity
- Screen downstream signaling behavior

KEY
- ● TWIST
- ● CUSTOMER

PARTNER WITH US. Get in touch at **biopharma@twistbioscience.com** or learn more at **twistbiopharma.com**