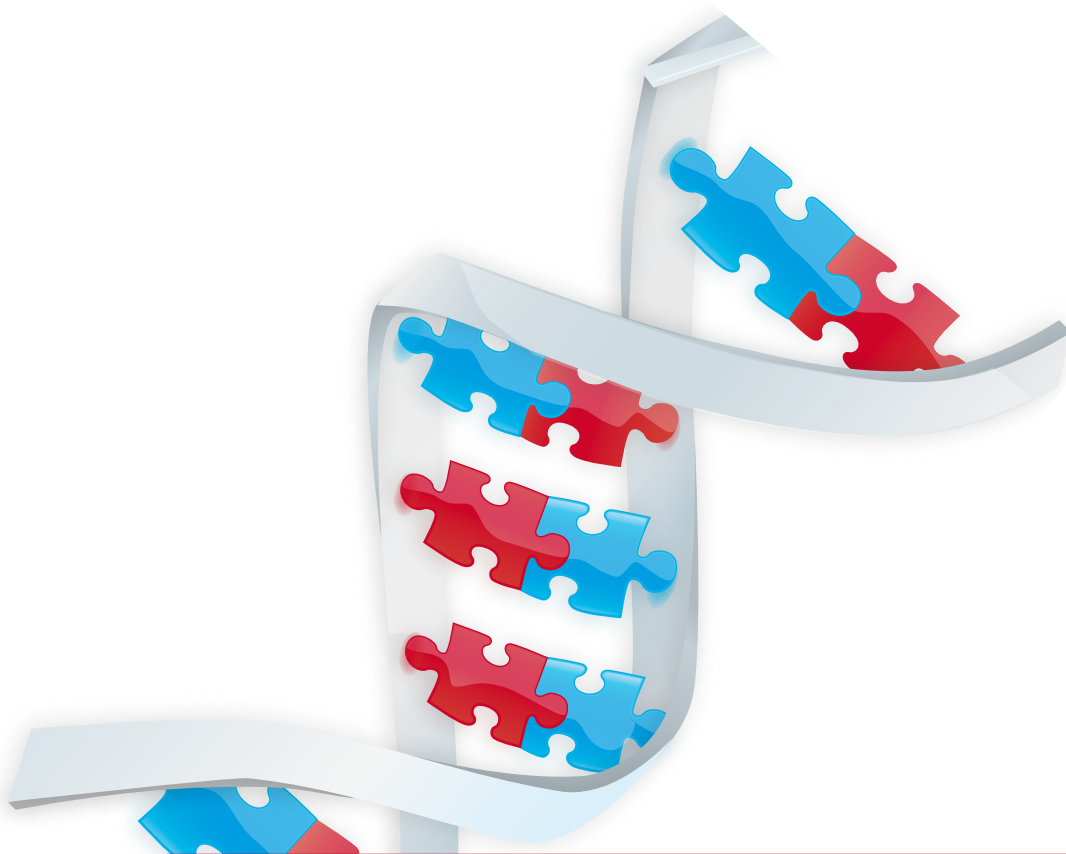


# cDNA library construction



## We offer you

- Customized cDNA library construction services
- EasyClone cDNA library construction kit
- Our cDNA library collection

# High complexity cDNA library construction

## High complexity cDNA library construction

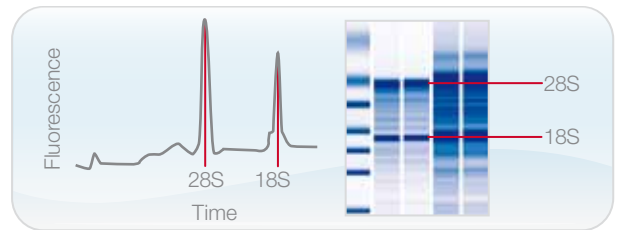
- ▶ High representation of 5' ends
- ▶ Guaranteed coverage of tissue complexity
- ▶ Large average insert sizes (>1.5 kb)\*
- ▶ Constructed in a vector of your choice



## RNA quality control by Experion chip and *in silico* gel electrophoresis

The quality of a cDNA library depends on the purity and preservation of the starting material. We examine the purified total RNA by Experion chip analysis and *in silico* gel electrophoresis. The 28S and 18S RNA are clearly visible as peaks and strong bands, respectively.

In case of high quality total RNA, the ratio of the intensities of the 28S and 18S bands is 1.5-2.5 : 1.



## Construction process

- Isolation of total or polyA+ RNA
- Chip-based quality control
- 1<sup>st</sup> cDNA generation and size fractionation
- 2<sup>nd</sup> strand cDNA generation on two independent size pools
- Directional ligation into appropriate vector
- Amplification of primary library
- Isolation of plasmid DNA

## Specifications and deliverables

- Special 5' adaptor annealing procedure preserves full-length cDNAs
- Average insert sizes > 1.5 kb
- High complexities ( $2 \times 10^6 - 5 \times 10^7$ )
- Cloned into DUALmembrane, DUALhunter and DUALhybrid library vectors, or a vector of your choice
- Average turnaround time 2 months
- You receive primary glycerol stocks and at least 500 µg of purified plasmid DNA

▶ Order number: S03002

\* Average insert sizes depend on the organism and RNA quality

# EasyClone cDNA library construction kit

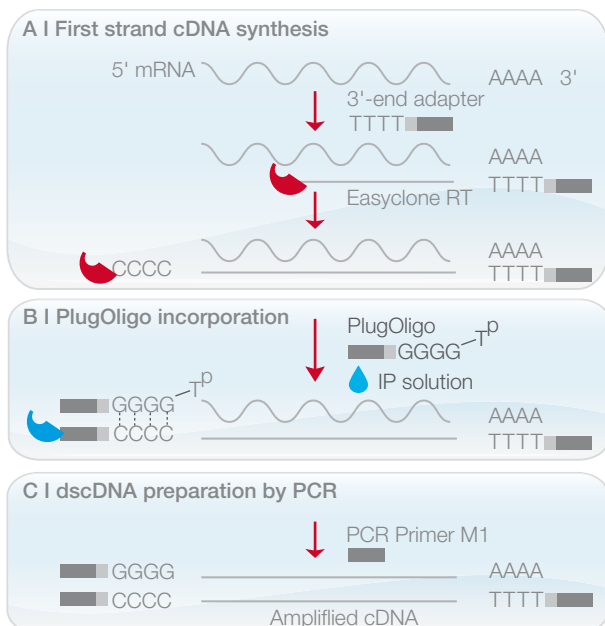
- Fast and easy synthesis of highly complex, representational cDNAs
- High representation of full length transcripts
- Directional cloning of cDNAs into DUALmembrane and DUALhybrid vectors
- Only small starting amounts of RNA required

➤ **Order number: P01010**

**No extensive know-how in library construction is needed – the kit supplies all reagents for library construction and the detailed manual guides you through all steps of construction**

## ■ Schematic outline of the cDNA synthesis workflow

During first strand cDNA synthesis the EasyClone reverse transcriptase (RT) starts synthesis at the Oligo(dT) adapter and ends by adding several non-template nucleotides to the 3' end of the newly synthesized strand (A). The PlugOligo (second adapter) base-pairs with the newly synthesized oligo(dC) stretch, leading to the incorporation of the PlugOligo sequence at the 3' end of the cDNA strand by RT (B). In the final step, EasyClone polymerase and special PCR primers are used to synthesize full length cDNA flanked by PlugOligo and 3' end adapter sequences (C).



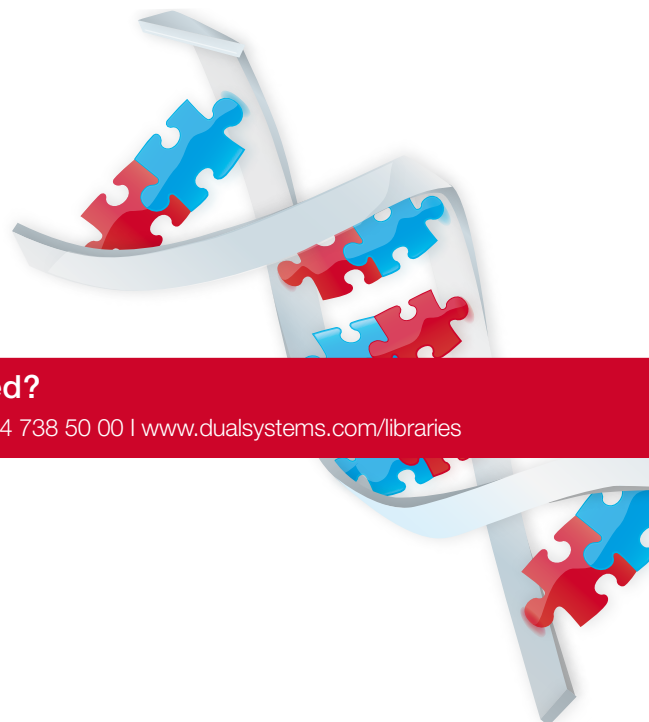
## ■ Kit contents

Box 1 Components	Amount
5X first strand buffer	80 µl
DTT (20 mM)	30 µl
10X dNTP mix (10 mM each)	25 µl
PlugOligo-1 adapter (15 µM)	25 µl
CDS-1 adapter (10 µM)	25 µl
PlugOligo-3M adapter (15 µM)	25 µl
CDS-3M adapter (10 µM)	25 µl
EasyClone reverse transcriptase	20 µl
IP-solution	130 µl
Control RNA template	15 µl
Sterile RNase free water	1.8 ml

Box 2 Components	Amount
50X EasyClone polymerase mix	50 µl
10X EasyClone buffer	300 µl
PCR Primer M1 (10 µM)	100 µl
50X dNTP mix (10 mM each)	80 µl
Sterile RNase free water	1.8 ml
Control amplified cDNA sample 1	25 µl
Control amplified cDNA sample 2	25 µl

**Interested?**

Phone +41 44 738 50 00 | [www.dualsystems.com/libraries](http://www.dualsystems.com/libraries)



# Our cDNA library collection

We supply a wide range of premade cDNA libraries for use with our yeast-based screening systems. All cDNA libraries are delivered as purified plasmid DNA, ready for screening.

## ■ DUALhybrid libraries

### Human libraries

P02102 | Human colon  
P02103 | HeLa cell line  
P02104 | LNCaP human prostate cancer cells  
P02106 | Human hepatocellular carcinoma cells

### Mouse libraries

P02101 | Mouse 11d total embryo  
P02107 | Mouse adult heart

### C.elegans libraries

P02105 | C. elegans whole adult

### Arabidopsis libraries

P02109 | Arabidopsis thaliana

### S. cerevisiae libraries

P02108 | Saccharomyces cerevisiae

## ■ DUALmembrane/hunter libraries

### Mouse libraries

P02201 | Mouse adult brain (NubG-x)  
P02206 | Mouse adult heart (x-NubG)  
P02207 | Mouse adult heart (NubG-x)  
P02224 | Mouse adult spleen (x-NubG)  
P02231 | Mouse adult kidney (NubG-x)  
P02234 | Mouse whole embryo, 11 days (NubG-x)

### Rat libraries

P02233 | rat neonatal cardiomyocyte (NubG-x)

### C.elegans libraries

P02218 | C. elegans whole adult (NubG-x)  
P02222 | C. elegans whole eggs (NubG-x)

### Drosophila libraries

P02223 | D. melanogaster embryo (NubG-x)  
P02229 | Drosophila melanogaster (NubG-x)

### Normalized libraries

P02301 | Normalized mouse embryo (x-NubG)  
P02302 | Normalized human spleen (NubG-x)

### Plant libraries

P02210 | Arabidopsis thaliana(NubG-x)  
P02235 | Medicago nodules (x-NubG)  
P02236 | Medicago nodules (NubG-x)

### Human libraries

P02205 | Jurkat T cells (NubG-x)  
P02208 | Human adult kidney (x-NubG)  
P02226 | Human adult kidney (NubG-x)  
P02212 | HeLa cell line (NubG-x)  
P02213 | Human embryonal brain (NubG-x)  
P02221 | Human adult brain (NubG-x)  
P02227 | Human adult brain (x-NubG)  
P02215 | Human adult colon (NubG-x)  
P02220 | Human adult liver (NubG-x)  
P02228 | LNCaP cell line (NubG-x)  
P02230 | Human adult lung (NubG-x)  
P02232 | Mammary epithelial cells (NubG-x)

### S. cerevisiae libraries

P02237 | Saccharomyces cerevisiae (NubG-x)

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