

DNAWorks (v3.2.4)

Automatic oligonucleotide design for PCR-based gene synthesis

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The availability of sequences of entire genomes has dramatically increased the number of protein targets, many of which will need to be overexpressed in cells other than the original source of DNA. Gene synthesis often provides a fast and economically efficient approach. The synthetic gene can be optimized for expression and constructed for easy mutational manipulation without regard to the parent genome. Yet design and construction of synthetic genes, especially those coding for large proteins, can be a slow, difficult and confusing process. DNAWorks automates the design of oligonucleotides for gene synthesis by PCR-based methods.

A description of the original DNAWorks can be found in our publication ([Hoover, D. and Lubkowski, J., 2002](#)).

For further improvements in the performance of DNAWorks, feedback on the success or failure of gene synthesis using DNAWorks is very important. [Please let us know about the outcome of your synthesis experiments.](#)



DNAWorks is open-source, and can be downloaded from <https://github.com/davidhoover/DNAWorks>. If you can find a Fortran compiler, you can run DNAWorks via commandline.

Job Name: You must enter a name for your job. Acceptable characters are [A-Za-z0-9_].

Job name:

E-mail Address (optional):

Entering your e-mail address is entirely voluntary. The results will be made available online at the end of the program run, but if an address is entered, the results will also be sent to that address. **Online results will be deleted after two days.**

E-mail Address:

Mutant run (optional): All parameters from original logfile will override those from this page, except for job name, email and sequences.

Enable mutant run: ☐

Codon Frequency Table:

GCG CodonFrequency format only. For more organisms, check out the [Codon Usage Database](#)

Choice 1 - Choose a standard organism: ☒

E. coli class II
H. sapiens
C. elegans
D. melanogaster
M. musculus

Choice 2 - Enter custom codon frequency table manually: ☐

Parameters:

The total number of trials is capped at 64

Annealing Temperature:	62	°C	to		°C	
Oligo Length:	60	nt	to		nt	Random: ☐
Codon Frequency Threshold:	10	%	Random:	☐	Strict:	☐
			Scored:	☐		
Oligonucleotide Concentration:	1E-7	M				
Na+/K+ Concentration:	0.05	M				
Mg2+ Concentration:	0.002	M				
Number of Solutions:	1					
TBIO Mode:	☐					
No gaps in assembly:	☐					

Restriction Site Screen (optional):

Sites selected will be excluded from the protein coding region of the synthetic gene

Non-degenerate sites:	☐ AatII ☐ Acc65I ☐ AclI ☐ AcuI ☐ AfeI ☐ AflII ☐ AgeI ☐ AlwI ☐ ApaI ☐ ApaLI ☐ AscI ☐ AseI	Degenerate sites:	☐ AccI ☐ AflIII ☐ AhdI ☐ AleI ☐ AlwNI ☐ Apol ☐ Aval ☐ BaeI ☐ BanI ☐ BanII ☐ BcgI ☐ BglI
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Custom Site Screen (optional):

Input your own site to be excluded from the protein coding region(s) of the synthetic gene

Enter name and sequence:	
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Weights (optional):

Enable Weights: ☐	TWT:	CWT:	RWT:	MWT:	GWT:	AWT:	LWT:	PWT:	FWT:
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Sequence(s) (*formats*):

Enter up to 99 sequence elements.

protein

nucleotide

reverse sequence

☐

fix sequence in gap

☐

Choice 1 - Upload sequence file:

☐

Choose File

No file chosen

Choice 2 - Enter sequence manually:

☒

Add Sequence Field

Design Oligos

Reset

[Help](#)

Remove Sequence Field

Example:

> lysozyme, 129 aa.

```
KVFGRCELAAAMKRHGLDNYRGYSLGNWVCAAKFESNFNTQATNRNTDGSTDYGILQINS  
RWWCNDGRTPGSRNLCNIPCSALLSSDITASVNCAKKIVSDGNGMNAWVAWRNRCKGTDV  
QAWIRGCRL
```

> pUC18 fragment

```
cgtagcgctct cctgtttccga ccctgccgct taccggatac ctgtccgcct  
ttctcccttc gggaagcgtg gcgctttctc aaagctcacg ctgtaggtat  
ctcagttcgg tgtaggtcgt tcgctccaag ctgggctgtg tgcacgaacc  
cccgtttcag ccgaccgct gcgccttata cggttaactat cgtcttgagt  
ccaacccggt aagacacgac ttatcgccac tggcagcagc cactggtaac  
aggattagca gagcgaggta ttagggcggg gctacagagt tcttgaagtg  
gtggcctaac tacggctaca ctagaagaac agtatttggt atctgcgctc
```

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