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A Rapid 1-Day Cloning to Protein Expression Workflow

Molecular biology tools enable the custom generation of proteins with complete control of sequence, purification tags, secretion signals, and other performance characteristics. While the breadth of tools allows researchers to create their desired protein, this process often involves a low-throughput and time-consuming, multiday workflow using live cells. To overcome these limitations, we have demonstrated a completely in vitro workflow that combines Golden Gate DNA Assembly, rolling circle amplification (RCA), and cell-free protein expression (CFPE) to rapidly screen the impact of multiple protein designs simultaneously (Figure 1). This workflow enables researchers to generate an array of protein variants in as little as a single day using a basic set of custom DNA vectors or insertion fragments. It also provides a means to assess the engineering constraints that are unique to each protein of interest and allows for the rapid identification of soluble protein.

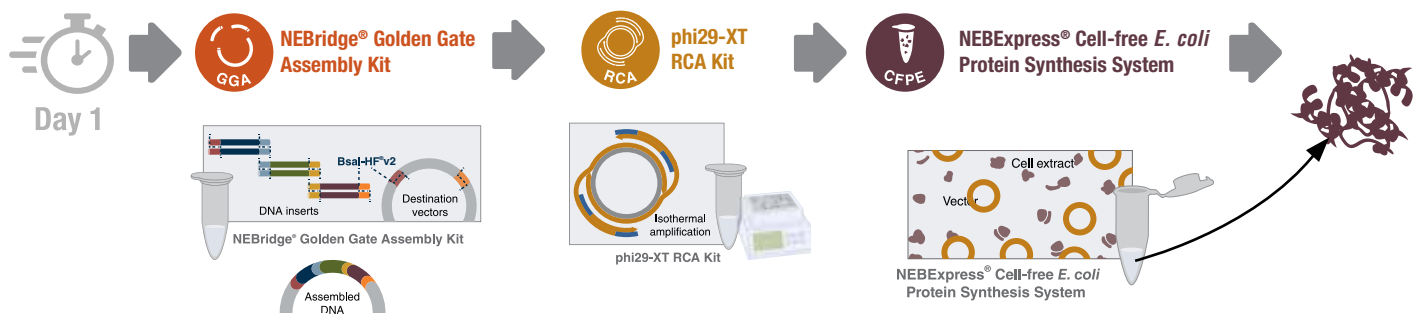
APPLICATION NOTES:

Accelerating DNA Construction to Protein Expression A Rapid 1-Day Workflow Using NEBridge Golden Gate Assembly



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Figure 1: **Rapid expression of proteins using NEBridge® Golden Gate Assembly and NEBExpress Cell-free E. coli Protein Synthesis System**



	NEB#	LIST PRICE	PROMOTIONAL PRICE
NEBridge® Ligase Master Mix	M1100S	\$135	\$94.50
	M1100L	\$539	\$377.30
Bsal-HF® v2	R3733S	\$107	\$74.90
	R3733L	\$444	\$310.80
phi29-XT RCA Kit	E1603S	\$479	\$335.30
	E1603L	\$2,030	\$1,421.00
NEBExpress® Cell-free <i>E. coli</i> Protein Synthesis System	E5360S	\$328	\$229.60
	E5360L	\$2,654	\$1,857.80
NEBExpress® Ni-NTA Magnetic Beads	S1423S	\$284	\$198.80
	S1423L	\$1,250	\$875.00
SNAP-Capture Magnetic Beads	S9145S	\$361	\$252.70
Monarch® DNA Gel Extraction Kit	T1020S	\$139	\$97.30
	T1020L	\$633	\$443.10



Learn more about NEBridge Golden Gate Assembly at international.neb.com/goldengate. Try our NEBridge Ligase Fidelity Tools for Golden Gate Assembly at ligasefidelity.neb.com



For more information about the phi29-XT RCA Kit, please visit www.neb.ca/E1603



Learn more about protein expression at NEB at international.neb.com/proteinexpression

For more information visit:
www.neb.ca/neb-rapid-1-day-workflow-nebridge-24



Howard Cukier, Ph.D.
Regional Manager,
Ontario and Manitoba
Cell: (416) 527-2352
hcukier@neb.com

Nicholas Prevedel, MSc.
Associate Field Sales Representative,
Ontario and Manitoba
Cell: (437) 431-8820
nprevedel@neb.com

Alexandre Rochette, Ph.D.
Territory Manager,
Eastern Canada
Cell: (514) 233-4557
arochette@neb.com

Martin Chenal, Ph.D.
Associate Field Sales
Representative,
Eastern Canada
Cell: (438) 466-2884
mchenal@neb.com

Julie Wong, Ph.D.
Territory Manager,
Western Canada
Cell: (604) 767-5078
jwong@neb.com



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