Synfluenza (Synthetic Influenza) Clone Set, Recombinant in *Escherichia coli*, Plate 14 (Neuraminidase)

Catalog No. NR-45832

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Contributor and Manufacturer:

Pathogen Functional Genomics Resource Center at the J. Craig Venter Institute

Product Description:

The Synfluenza clone set is part of a National Institute of Allergy and Infectious Diseases (NIAID) initiative to create 1000 influenza gene segment clones from 12 host subtypes that span the protein sequence diversity of influenza viruses between 2005 and 2010. Each clone is designed from GenBank sequences with consensus untranslated regions. The purpose of the project is to develop the ability to create and stockpile synthetic DNA encoding influenza gene segments. These segments can then be used to generate virus seed stocks and a library of clones for vaccine, diagnostic and basic research.[1]

The NIAID Genome Sequencing Center at the J. Craig Venter Institute constructed synthetic influenza neuraminidase (NA) and hemagglutinin (HA) genes using automated DNA synthesis and assembly. There are nine synthetic NA influenza clone plates (BEI numbers NR-45827 through NR-45833, NR-45090 and NR-45091) and six synthetic HA influenza clone plates (BEI numbers NR-45092 through NR-45097) in the set.

Each synthetic NA gene from NR-45832 was manufactured from five individually-designed, double-stranded DNA construct cassettes produced by assembly of eight chemically-synthesized oligonucleotides using the Gibson Assembly™ process.[2-6] The five cassettes were combined into the pSMART®-LCKan vector (Lucigen®) to establish gene segment clones in One Shot® TOP10 competent (Invitrogen™) *Escherichia coli* (*E. coli*) cells. Detailed information for each clone on the plate is shown in Table 1.

Material Provided:

Each well of the 96-well plate contains approximately 200 µL of *E. coli* culture in Yeast Extract Tryptone media containing 25 µg/mL kanamycin supplemented with 10% glycerol.

Note: Production in the 96-well format has increased risk of cross-contamination between adjacent wells. Individual clones should be purified (e.g. single colony isolation and purification using good microbiological practices) and sequence-verified prior to use.

Packaging/Storage:

NR-45832 was packaged aseptically in a 96-well plate. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:
Yeast Extract Tryptone broth or agar containing 25 µg/mL kanamycin
Incubation:
Temperature: 37°C
Atmosphere: Aerobic
Propagation:
1. Scrape top of frozen well with a pipette tip and streak onto agar plate.
2. Incubate the plate at 37°C for 18 to 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Synfluenza (Synthetic Influenza) Clone Set, Recombinant in *Escherichia coli*, Plate 14 (Neuraminidase), NR-45832."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

1. D. Wentworth, Personal Communication.
2. Gibson, D. G. et al. "Creation of a Bacterial Cell Controlled by a Chemically Synthesized Genome." Science 329 (2010): 52-56. PubMed: 20488990.
3. Gibson, D. G. et al. "Enzymatic Assembly of DNA Molecules up to Several Hundred Kilobases." Nat. Methods 6 (2009): 343-345. PubMed: 19363495.
4. Gibson, D. G. et al. "Chemical Synthesis of the Mouse Mitochondrial Genome." Nat. Methods 7 (2010): 901-903. PubMed: 20935651.
5. Gibson, D. G. et al. "Complete Chemical Synthesis, Assembly, and Cloning of a *Mycoplasma genitalium* Genome." Science 319 (2008): 1215-1220. PubMed: 18218864.
6. Dormitzer, P. R. et. al. "Synthetic Generation of Influenza Vaccine for Rapid Response to Pandemics." Sci Transl Med. 185 (2013): 1-12. PubMed: 23677594.

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Support Provided by NIAID

Table 1: Synfluenza Clone Set, Plate 14 (NR-45832)[1]

Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
A01	A/Xian/009/2009 (H1N1)	HUMAN_H1N1PDM_NA_M002843:1135661846222	HM006725.1	291195537	3442	5'-3'
A02	A/Maryland/01/2009 (H1N1)	HUMAN_H1N1_NA_M000437:1135661845596	GQ476027.1	255960786	3446	3'-5'
A03	A/Hong Kong/CUHK5300/2005 (H3N2)	HUMAN_H3N2_NA_M000024:1135630356219	EU857328.1	194269538	3449	3'-5'
A04	A/Managua/1181.03/2010 (H3N2)	HUMAN_H3N2_NA_M000145:1135630356827	CY070953.1	303303795	3448	3'-5'
A05	A/Yamagata/K16/2006 (H3N2)	HUMAN_H3N2_NA_M000220:1135661846272	AB271786.1	126364472	3450	3'-5'
A06	A/Hong Kong/CUHK53907/2006 (H3N2)	HUMAN_H3N2_NA_M000338:1135661845289	EU857338.1	194269558	3450	5'-3'
A07	A/Kisumu/7604/2008 (H3N2)	HUMAN_H3N2_NA_M000447:1135630357342	HQ214398.1	306494533	3451	5'-3'
A08	A/Thailand/322/2008 (H3N2)	HUMAN_H3N2_NA_M000562:1135630356594	AB501493.1	261399535	3449	3'-5'
A09	A/Babol/37/2005 (H3N2)	HUMAN_H3N2_NA_M000744:1135661844396	FJ769868.1	224482738	3451	5'-3'
A10	A/Illinois/UR06-0600/2007 (H3N2)	HUMAN_H3N2_NA_M000803:1135661846458	CY030199.1	168481171	3451	3'-5'
A11	A/Thailand/CU-B657/2009 (H3N2)	HUMAN_H3N2_NA_M000959:1135661845536	GQ902827.1	258578569	3450	5'-3'
A12	A/Managua/17/2007 (H3N2)	HUMAN_H3N2_NA_M001002:1135661843431	CY034086.1	194352261	3450	5'-3'
B01	A/Qingdao/1215/2009 (H1N1)	HUMAN_H1N1PDM_NA_M002844:1135661846193	CY050114.1	301638003	3442	5'-3'
B02	A/Denmark/116/2005 (H1N1)	HUMAN_H1N1_NA_M000480:1135661847095	EU097719.1	158830160	3445	5'-3'
B03	A/Yamagata/K55/2006 (H3N2)	HUMAN_H3N2_NA_M000035:1135661843642	AB271792.1	126364484	3450	5'-3'

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Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
B04	A/Cheongju/H393/2007 (H3N2)	HUMAN_H3N2_NA_M000160:1135630357326	FJ009480.1	196481136	3447	5'-3'
B05	A/Miyagi/S125/2006 (H3N2)	HUMAN_H3N2_NA_M000249:1135661844253	AB271721.1	126364424	3451	3'-5'
B06	A/Hong Kong/CUHK40210/2005 (H3N2)	HUMAN_H3N2_NA_M000339:1135661845053	EU857280.1	194269442	3451	5'-3'
B07	A/Tennessee/04/2008 (H3N2)	HUMAN_H3N2_NA_M000456:1135630357029	FJ686924.1	222433735	3449	3'-5'
B08	A/Isiolo/7514/2008 (H3N2)	HUMAN_H3N2_NA_M000586:1135630358496	HQ214371.1	306494479	3450	3'-5'
B09	A/Nagasaki/N113/2006 (H3N2)	HUMAN_H3N2_NA_M000751:1135661843918	AB271005.1	124484549	3449	3'-5'
B10	A/Hawaii/12/2007 (H3N2)	HUMAN_H3N2_NA_M000814:1135630359562	EU516171.1	168825313	3449	5'-3'
B11	A/California/VRDL166/2009 (H3N2)	HUMAN_H3N2_NA_M000963:1135661845275	CY068179.1	302182498	3451	3'-5'
B12	A/Managua/3616.01/2007 (H3N2)	HUMAN_H3N2_NA_M001003:1135661843416	CY038513.1	225907692	3451	5'-3'
C01	A/Singapore/ON2416/2009 (H1N1)	HUMAN_H1N1PDM_NA_M002849:1135661845974	CY063824.1	297206577	3442	5'-3'
C02	A/Rheinland-Pfalz/1/2006 (H1N1)	HUMAN_H1N1_NA_M000484:1135661847423	FJ231836.1	208972451	3445	5'-3'
C03	A/Australia/19/2009 (H3N2)	HUMAN_H3N2_NA_M000043:1135630356605	CY061900.1	295191782	3450	5'-3'
C04	A/Bethesda/NIH12-D6/2008 (H3N2)	HUMAN_H3N2_NA_M000199:1135630358298	GU294118.1	281335531	3439	3'-5'
C05	A/Hong Kong/CUHK40022/2005 (H3N2)	HUMAN_H3N2_NA_M000266:1135661842510	EU857277.1	194269436	3451	3'-5'
C06	A/Denmark/22/2006 (H3N2)	HUMAN_H3N2_NA_M000344:1135630356977	EU103915.1	156692029	3450	3'-5'
C07	A/Thailand/381/2007 (H3N2)	HUMAN_H3N2_NA_M000466:1135630357977	AB501504.1	261399557	3449	5'-3'
C08	A/New York/461/2005 (H3N2)	HUMAN_H3N2_NA_M000592:1135630358735	CY006078.1	78706391	3449	3'-5'
C09	A/Nagasaki/N76/2006 (H3N2)	HUMAN_H3N2_NA_M000752:1135661843933	AB271713.1	126364408	3451	5'-3'
C10	A/Thailand/173/2007 (H3N2)	HUMAN_H3N2_NA_M000815:1135661846867	AB501496.1	261399541	3451	3'-5'
C11	A/Hanoi/TN403/2005 (H3N2)	HUMAN_H3N2_NA_M000968:1135661845066	AB281257.1	146197732	3451	3'-5'
C12	A/Managua/28/2007 (H3N2)	HUMAN_H3N2_NA_M001007:1135661843356	CY032535.1	189231073	3451	3'-5'
D01	A/Iran/16743/2009 (H1N1)	HUMAN_H1N1PDM_NA_M002860:1135661845511	HM581910.1	299152143	3441	3'-5'
D02	A/Niedersachsen/192/2005 (H1N1)	HUMAN_H1N1_NA_M000489:1135661847456	FJ231819.1	208972417	3445	3'-5'
D03	A/Hong Kong/CUHK6898/2005 (H3N2)	HUMAN_H3N2_NA_M000095:1135630358543	EU857360.1	194269602	3451	3'-5'
D04	A/Hanoi/TN405/2005 (H3N2)	HUMAN_H3N2_NA_M000202:1135630358184	AB281260.1	146197739	3449	3'-5'
D05	A/Fukuoka/F52/2006 (H3N2)	HUMAN_H3N2_NA_M000269:1135661842462	AB271803.1	126364506	3451	5'-3'
D06	A/California/VRDL207/2009 (H3N2)	HUMAN_H3N2_NA_M000349:1135630356874	CY068784.1	302371742	3450	5'-3'
D07	A/Hong Kong/CUHK6422/2005 (H3N2)	HUMAN_H3N2_NA_M000470:1135661845401	EU857352.1	194269586	3450	5'-3'
D08	A/California/VRDL332/2009 (H3N2)	HUMAN_H3N2_NA_M000664:1135630354640	CY068587.1	302183856	3449	3'-5'
D09	A/California/09/2008 (H3N2)	HUMAN_H3N2_NA_M000754:1135661843896	FJ532079.1	216960165	3451	3'-5'

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Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
D10	A/Alabama/UR06-0545/2007 (H3N2)	HUMAN_H3N2_NA_M000817:1135661846792	CY025861.1	157282783	3451	3'-5'
D11	A/Oklahoma/372/2005 (H3N2)	HUMAN_H3N2_NA_M000983:1135661843609	EF462548.1	126566796	3450	5'-3'
D12	A/Managua/29/2007 (H3N2)	HUMAN_H3N2_NA_M001008:1135661843338	CY032479.1	189230940	3450	5'-3'
E01	A/Hawaii/05/2008 (H1N1)	HUMAN_H1N1_NA_M000037:1135661846113	EU779644.1	189303419	3445	5'-3'
E02	A/Berlin/60/2005 (H1N1)	HUMAN_H1N1_NA_M000490:1135661847389	FJ231813.1	208972405	3446	5'-3'
E03	A/Tokyo/Ut-Sk-1/2007 (H3N2)	HUMAN_H3N2_NA_M000098:1135661845826	CY049750.1	285138740	3450	3'-5'
E04	A/Hong Kong/CUHK53798/2006 (H3N2)	HUMAN_H3N2_NA_M000205:1135630358194	EU857337.1	194269556	3452	5'-3'
E05	A/Queensland/57/2005 (H3N2)	HUMAN_H3N2_NA_M000285:1135630354707	CY017621.1	118318340	3450	5'-3'
E06	A/Denmark/98/2006 (H3N2)	HUMAN_H3N2_NA_M000365:1135630356338	EU103962.1	156692123	3449	5'-3'
E07	A/Denmark/35/2006 (H3N2)	HUMAN_H3N2_NA_M000487:1135630358571	EU103960.1	156692119	3450	3'-5'
E08	A/California/VRDL331/2009 (H3N2)	HUMAN_H3N2_NA_M000667:1135630354567	CY068579.1	302183837	3450	5'-3'
E09	A/Cheongju/H471/2008 (H3N2)	HUMAN_H3N2_NA_M000788:1135661843187	FJ009488.1	196481146	3447	5'-3'
E10	A/Sendai-H/401/2006 (H3N2)	HUMAN_H3N2_NA_M000826:1135661845579	AB441983.1	213385027	3449	5'-3'
E11	A/New York/352/2005 (H3N2)	HUMAN_H3N2_NA_M000986:1135661843625	CY002722.1	75180829	3450	3'-5'
E12	A/Managua/7/2007 (H3N2)	HUMAN_H3N2_NA_M001009:1135661843301	CY032543.1	189231092	3450	3'-5'
F01	A/England/593/2006 (H1N1)	HUMAN_H1N1_NA_M000274:1135661844223	FJ445058.1	212381569	3446	3'-5'
F02	A/Kentucky/UR06-0339/2007 (H1N1)	HUMAN_H1N1_NA_M000609:1135661842503	CY027781.1	159150069	3445	5'-3'
F03	A/Hong Kong/CUHK64082/2006 (H3N2)	HUMAN_H3N2_NA_M000100:1135661842784	EU857351.1	194269584	3450	5'-3'
F04	A/Miyagi/S709/2006 (H3N2)	HUMAN_H3N2_NA_M000212:1135661845788	AB271733.1	126364448	3450	5'-3'
F05	A/Hong Kong/HKU78/2005 (H3N2)	HUMAN_H3N2_NA_M000314:1135661846059	CY043770.1	255529094	3450	5'-3'
F06	A/Kisii/7566/2008 (H3N2)	HUMAN_H3N2_NA_M000377:1135630356568	HQ214385.1	306494507	3450	3'-5'
F07	A/Illinois/UR06-0436/2007 (H3N2)	HUMAN_H3N2_NA_M000510:1135630355612	CY025901.1	157282878	3450	5'-3'
F08	A/Qingdao/1118/2009 (H3N2)	HUMAN_H3N2_NA_M000677:1135661842314	CY050107.1	301637989	3451	3'-5'
F09	A/Memphis/15d/2008 (H3N2)	HUMAN_H3N2_NA_M000793:1135661842990	CY068889.1	302372435	3451	5'-3'
F10	A/Canterbury/232/2005 (H3N2)	HUMAN_H3N2_NA_M000845:1135661844926	CY013242.1	112787492	3451	5'-3'
F11	A/Niigata/F183/2006 (H3N2)	HUMAN_H3N2_NA_M000990:1135661843406	AB271704.1	126364390	3450	5'-3'
F12	A/Managua/1507.01/2007 (H3N2)	HUMAN_H3N2_NA_M001013:1135661844332	CY038553.1	225907787	3450	3'-5'
G01	A/Berlin/6/2006 (H1N1)	HUMAN_H1N1_NA_M000276:1135661844211	FJ231830.1	208972439	3445	5'-3'
G02	A/Illinois/UR06-0227/2007 (H1N1)	HUMAN_H1N1_NA_M000617:1135661843216	CY027789.1	159150088	3446	5'-3'
G03	A/New York/390/2005 (H3N2)	HUMAN_H3N2_NA_M000127:1135630354075	CY002482.1	73666598	3449	3'-5'

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Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
G04	A/Morioka/18/2006 (H3N2)	HUMAN_H3N2_NA_M000217:1135630358516	AB433823.1	183396546	3450	3'-5'
G05	A/Nagasaki/N01/2005 (H3N2)	HUMAN_H3N2_NA_M000329:1135661844709	AB271003.1	124484545	3448	5'-3'
G06	A/California/VRDL138/2009 (H3N2)	HUMAN_H3N2_NA_M000379:1135630356587	CY064825.1	298347697	3449	5'-3'
G07	A/Thailand/CU-1102/2008 (H3N2)	HUMAN_H3N2_NA_M000539:1135630354426	EU625367.1	193888331	3450	3'-5'
G08	A/California/VRDL315/2009 (H3N2)	HUMAN_H3N2_NA_M000698:1135661842806	CY068507.1	302183291	3451	3'-5'
G09	A/Boston/47/2008 (H3N2)	HUMAN_H3N2_NA_M000796:1135661842959	CY044646.1	256386681	3450	3'-5'
G10	A/Denmark/83/2005 (H3N2)	HUMAN_H3N2_NA_M000900:1135661847366	EU103914.1	156692027	3451	3'-5'
G11	A/Niigata/F272/2006 (H3N2)	HUMAN_H3N2_NA_M000992:1135661843784	AB271502.1	124484585	3451	3'-5'
G12	A/Texas/12/2007 (H3N2)	HUMAN_H3N2_NA_M001025:1135661843880	FJ445772.1	212383381	3455	3'-5'
H01	A/Peru/WRAIR1298P/2007 (H1N1)	HUMAN_H1N1_NA_M000434:1135661845750	CY070074.1	302634319	3445	3'-5'
H02	A/Kentucky/UR06-0028/2007 (H1N1)	HUMAN_H1N1_NA_M000808:1135661844664	CY026221.1	157368131	3445	3'-5'
H03	A/Yamagata/K32/2006 (H3N2)	HUMAN_H3N2_NA_M000136:1135661842590	AB271788.1	126364476	3451	3'-5'
H04	A/Miyagi/S704/2006 (H3N2)	HUMAN_H3N2_NA_M000219:1135661846099	AB271731.1	126364444	3451	5'-3'
H05	A/Wisconsin/43/2006 (H3N2)	HUMAN_H3N2_NA_M000332:1135630357960	EU100654.1	156123541	3450	3'-5'
H06	A/Thailand/536/2008 (H3N2)	HUMAN_H3N2_NA_M000380:1135630356477	AB501498.1	261399545	3450	5'-3'
H07	A/Kentucky/01/2008 (H3N2)	HUMAN_H3N2_NA_M000542:1135630357443	FJ532067.1	216960082	3449	3'-5'
H08	A/Thailand/CU-B106/2009 (H3N2)	HUMAN_H3N2_NA_M000740:1135661844419	GQ983550.1	260100716	3450	3'-5'
H09	A/New York/UR06-0510/2007 (H3N2)	HUMAN_H3N2_NA_M000797:1135661842887	CY025845.1	157282745	3450	3'-5'
H10	A/USA/AF1087/2007 (H3N2)	HUMAN_H3N2_NA_M000912:1135661847072	CY024911.1	155965957	3451	5'-3'
H11	A/New York/928/2006 (H3N2)	HUMAN_H3N2_NA_M000999:1135661843707	CY020087.1	131055740	3451	3'-5'
H12	A/Qingdao/2199/2009 (H3N2)	HUMAN_H3N2_NA_M001027:1135661843823	CY050096.1	301637967	3450	3'-5'

[1]All information in this table was provided by J. Craig Venter Institute at the time of deposition.
[2]All clones contain full length inserts, HA inserts are 1716 to 1803 base pairs, NA inserts are 1453 to 1557 base pairs.
[3]Genbank gene ID

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