Synfluenza (Synthetic Influenza) Clone Set, Recombinant in *Escherichia coli*, Plate 7 (Hemagglutinin)

Catalog No. NR-45096

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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 HA gene from NR-45096 was manufactured from seven individually-designed, double-stranded DNA construct cassettes produced by assembly of eight chemically-synthesized oligonucleotides using the Gibson Assembly™ process.[2-6] The seven 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-45096 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 7 (Hemagglutinin), NR-45096."

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.

Support Provided by NIAID

Table 1: Synfluenza Clone Set, Plate 7, (NR-45096)[1]

Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
A01	A/Wisconsin/32/2007 (H3N2)	HUMAN_H3N2_HA_M000882:1135630362036	EU516207.1	168828232	3745	5'-3'
A02	A/Qingdao/1046/2009 (H3N2)	HUMAN_H3N2_HA_M000920:1135630361046	CY050138.1	301638051	3744	5'-3'
A03	A/New York/UR06-0515/2007 (H3N2)	HUMAN_H3N2_HA_M000945:1135630360470	CY025485.1	157281904	3744	5'-3'
A04	A/Georgia/02/2008 (H3N2)	HUMAN_H3N2_HA_M000995:1135630352456	EU716476.1	188076400	3746	3'-5'
A05	A/Denmark/77/2006 (H3N2)	HUMAN_H3N2_HA_M001021:1135630362317	EU103655.1	156691519	3744	3'-5'
A06	A/Hawaii/06/2006 (H3N2)	HUMAN_H3N2_HA_M001043:1135630351391	EU100695.1	156123357	3745	5'-3'
A07	A/swine/NC/00573/2005 (H1N1)	PORCINE_H1N1_HA_M000010:1135630360742	FJ638306.1	221326987	3758	5'-3'
A08	A/swine/Hong Kong/NS29/2009 (H1N1)	PORCINE_H1N1_HA_M000063:1135630361478	GQ229301.1	239618845	3762	3'-5'
A09	A/swine/Hong Kong/1562/2005 (H1N2)	PORCINE_H1N2_HA_M000036:1135630360378	GQ229333.1	239618853	3761	5'-3'
A10	A/swine/Italy/116114/2010 (H1N2)	PORCINE_H1N2_HA_M000051:1135630364201	CY067662.1	312839912	3762	3'-5'
A11	A/swine/Missouri/03035/2010 (H1N2)	PORCINE_H1N2_HA_M000065:1135630363204	HM754219.1	300676661	3759	5'-3'
A12	A/swine/Shandong/133/2007 (H3N2)	PORCINE_H3N2_HA_M000019:1135630360426	GU086113.1	261889679	3744	3'-5'
B01	A/Hawaii/10/2009 (H3N2)	HUMAN_H3N2_HA_M000893:1135630362285	GQ385854.1	254564323	3744	3'-5'
B02	A/Qingdao/1045/2009 (H3N2)	HUMAN_H3N2_HA_M000923:1135630361058	CY050136.1	301638047	3746	5'-3'
B03	A/Arizona/11/2009 (H3N2)	HUMAN_H3N2_HA_M000950:1135630346703	GQ385909.1	254564367	3746	5'-3'

Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
B04	A/Iowa/02/2008 (H3N2)	HUMAN_H3N2_HA_M001004:1 135630349643	EU852009.1	193084882	3744	5'-3'
B05	A/Hawaii/47/2008 (H3N2)	HUMAN_H3N2_HA_M001027:1 135630362407	FJ686944.1	222433775	3746	5'-3'
B06	A/Nepal/921/2006 X PR8 X-165 (H3N2)	HUMAN_H3N2_HA_M001046:1 135630351345	EU100702.1	156123371	3745	3'-5'
B07	A/swine/Minnesota/07002083/2007 (H1N1)	PORCINE_H1N1_HA_M00001 2:1135630360766	FJ611898.1	220172404	3758	5'-3'
B08	A/swine/Germany/SIV04/2008 (H1N1)	PORCINE_H1N1_HA_M00007 0:1135630362742	FN429078.1	283481678	3762	5'-3'
B09	A/swine/Guangxi/17/2005 (H1N2)	PORCINE_H1N2_HA_M00003 7:1135630360389	EF556201.1	146706487	3762	5'-3'
B10	A/swine/Thailand/CU-CHK4/2009 (H1N2)	PORCINE_H1N2_HA_M00005 2:1135630364191	GU454849.1	284181771	3761	3'-5'
B11	A/swine/Minnesota/44837-3/2009 (H1N2)	PORCINE_H1N2_HA_M00006 6:1135630363277	HQ424891.1	310693821	3754	5'-3'
B12	A/swine/Sichuan/01/2006 (H3N2)	PORCINE_H3N2_HA_M00002 6:1135630364159	EU655692.1	194338967	3745	3'-5'
C01	A/California/VRDL338/2009 (H3N2)	HUMAN_H3N2_HA_M000897:1 135630362216	CY068625.1	302183946	3745	3'-5'
C02	A/California/VRDL176/2009 (H3N2)	HUMAN_H3N2_HA_M000930:1 135630361352	CY067921.1	302136505	3745	3'-5'
C03	A/Wellington/8/2005 (H3N2)	HUMAN_H3N2_HA_M000952:1 135630346655	CY014095.1	113171404	3744	3'-5'
C04	A/Tennessee/04/2008 (H3N2)	HUMAN_H3N2_HA_M001006:1 135630349717	EU885518.1	194293982	3745	3'-5'
C05	A/Colorado/05/2008 (H3N2)	HUMAN_H3N2_HA_M001030:1 135630362612	FJ686919.1	222433725	3744	3'-5'
C06	A/Colorado/03/2007 (H3N2)	HUMAN_H3N2_HA_M001048:1 135630362996	EU199257.1	158187915	3745	5'-3'
C07	A/swine/Iowa/03032/2010 (H1N1)	PORCINE_H1N1_HA_M00001 3:1135630360761	HM585495.1	299766147	3768	3'-5'
C08	A/swine/Ratchaburi/NIAH101942/2008 (H1N1)	PORCINE_H1N1_HA_M00007 6:1135630362696	AB514936.1	291461521	3762	3'-5'
C09	A/swine/Korea/JL04/2005 (H1N2)	PORCINE_H1N2_HA_M00003 9:1135630360419	EU798783.1	190403708	3761	5'-3'
C10	A/swine/Tochigi/1/2008 (H1N2)	PORCINE_H1N2_HA_M00005 3:1135630364119	AB514929.1	291461507	3761	5'-3'
C11	A/swine/South Dakota/152B/2009 (H1N2)	PORCINE_H1N2_HA_M00006 7:1135630363241	HQ840338.1	317016306	3759	5'-3'
C12	A/swine/Ratchaburi/NIAH874/2005 (H3N2)	PORCINE_H3N2_HA_M00002 7:1135630364145	EU296617.1	163676487	3744	3'-5'
D01	A/Sao Paulo/16363/2010 (H3N2)	HUMAN_H3N2_HA_M000906:1 135630361400	HM628692.1	300488895	3744	5'-3'
D02	A/California/VRDL360/2009 (H3N2)	HUMAN_H3N2_HA_M000935:1 135630361117	CY068065.1	302138095	3745	3'-5'
D03	A/Iraq/WRAIR1157P/2009 (H3N2)	HUMAN_H3N2_HA_M000954:1 135630346618	CY069477.1	302424683	3745	5'-3'
D04	A/New York/462/2005 (H3N2)	HUMAN_H3N2_HA_M001014:1 135630350482	CY006291.1	82494224	3746	3'-5'
D05	A/Western Australia/65/2005 (H3N2)	HUMAN_H3N2_HA_M001031:1 135630362662	CY015988.1	115290931	3744	5'-3'
D06	A/California/VRDL339/2009 (H3N2)	HUMAN_H3N2_HA_M001057:1 135630363516	CY067969.1	302136619	3746	3'-5'
D07	A/swine/Iowa/03031/2010 (H1N1)	PORCINE_H1N1_HA_M00001 4:1135630360809	HM585494.1	299766145	3761	5'-3'
D08	A/swine/Chachoengsao/NIAH587/2005 (H1N1)	PORCINE_H1N1_HA_M00008 8:1135630362521	EU296601.1	163676471	3767	5'-3'
D09	A/swine/Korea/PZ14/2006 (H1N2)	PORCINE_H1N2_HA_M00004 3:1135630363761	EU798787.1	190403716	3761	3'-5'

Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
D10	A/swine/Hainan/1/2005 (H1N2)	PORCINE_H1N2_HA_M000055:1135630364103	EF556203.1	146706513	3762	5'-3'
D11	A/swine/Nakhon pathom/NIAH586-1/2005 (H3N2)	PORCINE_H3N2_HA_M000001:1135630360722	AB434360.1	216409375	3744	3'-5'
D12	A/swine/Heilongjiang/10/2007 (H3N2)	PORCINE_H3N2_HA_M000029:1135630364182	HM765432.1	301752275	3744	3'-5'
E01	A/California/VRDL313/2009 (H3N2)	HUMAN_H3N2_HA_M000908:1135630348074	CY068497.1	302183267	3745	3'-5'
E02	A/New Jersey/14/2007 (H3N2)	HUMAN_H3N2_HA_M000937:1135630347240	EU516033.1	168828082	3744	3'-5'
E03	A/Cheongju/H471/2008 (H3N2)	HUMAN_H3N2_HA_M000957:1135630360532	FJ009476.1	196481144	3745	3'-5'
E04	A/Hong Kong/HKU42/2005 (H3N2)	HUMAN_H3N2_HA_M001017:1135630350562	CY038679.1	225908067	3745	3'-5'
E05	A/Waikato/1/2005 (H3N2)	HUMAN_H3N2_HA_M001032:1135630362623	CY014015.1	113170900	3744	5'-3'
E06	A/swine/Fujian/0325/2008 (H1N1)	PORCINE_H1N1_HA_M000002:1135630360934	GU646016.1	289900516	3758	5'-3'
E07	A/swine/Iowa/46519-2/2008 (H1N1)	PORCINE_H1N1_HA_M000016:1135630360568	HQ378732.1	308737072	3762	3'-5'
E08	A/swine/Saraburi/NIAH13021/2005 (H1N2)	PORCINE_H1N2_HA_M000002:1135630361201	EU296607.1	163676477	3761	3'-5'
E09	A/swine/Korea/S11/2005 (H1N2)	PORCINE_H1N2_HA_M000047:1135630363802	DQ666933.1	109501337	3761	5'-3'
E10	A/swine/Hong Kong/NS1889/2009 (H1N2)	PORCINE_H1N2_HA_M000056:1135630364045	CY061837.1	295424734	3760	3'-5'
E11	A/swine/Jilin/5/2007 (H3N2)	PORCINE_H3N2_HA_M000005:1135630360648	GU215018.1	269930132	3745	3'-5'
E12	A/swine/Korea/CAN04/2005 (H3N2)	PORCINE_H3N2_HA_M000032:1135630364019	EU798790.1	190403722	3745	3'-5'
F01	A/New Mexico/02/2008 (H3N2)	HUMAN_H3N2_HA_M000915:1135630361644	FJ179356.1	198241607	3745	5'-3'
F02	A/Acre/15093/2010 (H3N2)	HUMAN_H3N2_HA_M000938:1135630361082	HM628693.1	300488897	3745	3'-5'
F03	A/Boston/1/2008 (H3N2)	HUMAN_H3N2_HA_M000960:1135630353028	CY044429.1	256386087	3739	3'-5'
F04	A/New York/382/2005 (H3N2)	HUMAN_H3N2_HA_M001018:1135630362121	CY002032.1	71564841	3744	3'-5'
F05	A/Ontario/1252/2007 (H3N2)	HUMAN_H3N2_HA_M001034:1135630362568	EU399751.1	165880677	3746	5'-3'
F06	A/swine/Chonburi/05CB1/2005 (H1N1)	PORCINE_H1N1_HA_M000005:1135630360922	EU296603.1	163676473	3766	5'-3'
F07	A/swine/Oregon/10-004060/2009 (H1N1)	PORCINE_H1N1_HA_M000023:1135630360449	GU984384.1	290873670	3762	3'-5'
F08	A/swine/Miyazaki/1/2006 (H1N2)	PORCINE_H1N2_HA_M000003:1135630361238	AB441170.1	192806776	3761	5'-3'
F09	A/swine/Hong Kong/1110/2006 (H1N2)	PORCINE_H1N2_HA_M000048:1135630363974	GQ229373.1	239618863	3762	5'-3'
F10	A/swine/Groitzsch/IDT6016-2/2007 (H1N2)	PORCINE_H1N2_HA_M000060:1135630363325	GQ161143.1	238057038	3762	3'-5'
F11	A/swine/Minnesota/7931/2007 (H3N2)	PORCINE_H3N2_HA_M000007:1135630360600	FJ519977.1	217384849	3745	5'-3'
F12	A/swine/Guangdong/211/2006 (H3N2)	PORCINE_H3N2_HA_M000035:1135630363794	GU086121.1	261889681	3740	5'-3'
G01	A/Qingdao/1157/2009 (H3N2)	HUMAN_H3N2_HA_M000916:1135630348909	CY050108.1	301637991	3744	5'-3'
G02	A/California/VRDL259/2009 (H3N2)	HUMAN_H3N2_HA_M000939:1135630361188	CY068321.1	302182835	3745	5'-3'
G03	A/Pennsylvania/PIT39/2008 (H3N2)	HUMAN_H3N2_HA_M000967:1135630353092	CY038495.1	225907649	3746	3'-5'

Well	Strain	Clone Name	Locus (CDS)	Gene ID[3]	Vector Total Size	Insert Orientation
G04	A/New York/373/2005 (H3N2)	HUMAN_H3N2_HA_M001019:1135630362147	CY002456.1	73665921	3745	3'-5'
G05	A/New York/238/2005 (H3N2)	HUMAN_H3N2_HA_M001038:1135630351167	CY006139.1	80973916	3745	5'-3'
G06	A/swine/Okinawa/2/2005 (H1N1)	PORCINE_H1N1_HA_M000008:1135630360887	AB573800.1	302190116	3761	3'-5'
G07	A/swine/Shandong/128/2008 (H1N1)	PORCINE_H1N1_HA_M000051:1135630361723	FJ536781.1	218138355	3761	3'-5'
G08	A/swine/Iowa/02998/2010 (H1N2)	PORCINE_H1N2_HA_M000008:1135630361288	HM193850.1	296044860	3759	5'-3'
G09	A/swine/Guangdong/1222/2006 (H1N2)	PORCINE_H1N2_HA_M000049:1135630363891	GU086081.1	261889671	3762	5'-3'
G10	A/swine/Doetlingen/IDT4735/2005 (H1N2)	PORCINE_H1N2_HA_M000061:1135630363308	EU053133.1	153957928	3758	5'-3'
G11	A/swine/Minnesota/03008/2010 (H3N2)	PORCINE_H3N2_HA_M000010:1135630360291	HM217202.1	296238981	3745	3'-5'
G12	A/swine/Korea/CY07/2007 (H3N2)	PORCINE_H3N2_HA_M000039:1135630363828	EU798795.1	190403732	3746	3'-5'
H01	A/Mississippi/UR06-0197/2007 (H3N2)	HUMAN_H3N2_HA_M000918:1135630348860	CY035882.1	211593612	3745	3'-5'
H02	A/Nanjing/1/2009 (H3N2)	HUMAN_H3N2_HA_M000940:1135630360499	GU907114.1	289900184	3746	5'-3'
H03	A/Oregon/01/2008 (H3N2)	HUMAN_H3N2_HA_M000974:1135630353716	EU885532.1	194294010	3746	3'-5'
H04	A/Hong Kong/HKU62/2005 (H3N2)	HUMAN_H3N2_HA_M001020:1135630350681	CY038695.1	225908105	3745	5'-3'
H05	A/Colorado/04/2007 (H3N2)	HUMAN_H3N2_HA_M001042:1135630362853	EU199258.1	158187917	3746	3'-5'
H06	A/swine/Minnesota/03000/2010 (H1N1)	PORCINE_H1N1_HA_M000009:1135630360865	HM193852.1	296044864	3757	3'-5'
H07	A/swine/Beijing/21/2008 (H1N1)	PORCINE_H1N1_HA_M000058:1135630361807	FJ536762.1	218138409	3761	3'-5'
H08	A/swine/Argentina/CIP051-StaFeN2/2010 (H1N2)	PORCINE_H1N2_HA_M000030:1135630360320	CY075864.1	309378110	3759	3'-5'
H09	A/swine/Hong Kong/1479/2009 (H1N2)	PORCINE_H1N2_HA_M000050:1135630364218	CY061661.1	295424327	3760	5'-3'
H10	A/swine/Illinois/03036/2010 (H1N2)	PORCINE_H1N2_HA_M000063:1135630363387	HM754220.1	300676663	3759	5'-3'
H11	A/swine/Guangdong/02/2005 (H3N2)	PORCINE_H3N2_HA_M000015:1135630360348	EU620723.1	197344285	3745	5'-3'
H12	A/swine/Korea/CY10/2007 (H3N2)	PORCINE_H3N2_HA_M000040:1135630363646	EU798797.1	190403736	3743	5'-3'

[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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