

Cellecta Next-Gen Sequencing Report

Sequencing of Samples from Genetic Screen (screening done with Cellecta Library)

Customer: Joe Smith
 Institution: Customer University
 Cat.#: CANA-SQD + CANA-DNA
 Order#: 90000
 Quote#: SMI160208A-1
 Date: 3/18/16

Library Used: DECIPHER Human Module 1, Pathway Targets (plasmid)
 Vector: pRSI12-U6-sh-UbiC-TagRFP-2A-Puro
 Lot#: 11070805
 Sample Type: Cell Pellets

Sample description

#	Sample	Description	Cell Type	Cell #	DNA
1	C01	Mock, Collected, Day 4	HEK 293	5.5 x 10 ⁷	500 µg
2	D01	DRUG #1 treatment (7 days)	HEK 293	5.5 x 10 ⁷	500 µg
3	D02	DRUG #2 treatment (7 days)	HEK 293	5.5 x 10 ⁷	500 µg

Comments

1. Genomic DNA was isolated from the cell pellets provided. Yields are shown in the table above.
2. Amplification and purification of shRNA barcodes was successful.
3. The NGS data passed our quality standards.

Sequencing Data:

Please see the file [NGS-Data-CustomerU-90000-160318.xlsx](#) for sequencing results. Description of columns are provided below:

Column	Header	Description
A:	label	shRNA label
B:	sequence	Sequence of shRNA/barcode insert
C:	order	Order of oligo in library (arbitrary)
D:	shRNA#	shRNA# for the particular gene (arbitrary)
E:	gene#	Gene# in library (arbitrary)
F:	replicate#	Barcode# for particular shRNA (e.g. controls have 3 barcodes/shRNA)
G:	refseq#	RefSeq#

See Page 3 for a screen shot of the Sample NGS Data file.



H:	symbol	HUGO Gene Symbol
I:	plasmid	shRNA reads in plasmid library
J:	plasmid_20M	shRNA reads in plasmid library, adjusted to 20M reads
K:	C	shRNA reads in sample "C"
L:	C_20M	shRNA reads in sample "C", adjusted to 20M reads
M:	D1	shRNA reads in sample "D1"
N:	D1_20M	shRNA reads in sample "D1", adjusted to 20M reads
O:	D2	shRNA reads in sample "D2"
P:	D2_20M	shRNA reads in sample "D2", adjusted to 20M reads

Description of Controls (applicable to Human DECIPHER Libraries only):

The Human DECIPHER 27K modules are made of two shRNA sets: shRNAs 1 - 27,290 (body of the library) and shRNAs 27,291 - 27,500 (control block).

The 1st block is made up of 27,290 shRNAs targeting about 5,000 genes (column E), with 5-6 shRNAs/gene, (column D). Each shRNA sequence is represented once, with its own unique barcode (column F).

The 2nd block is made of 70 shRNAs targeting 10 control genes (7 shRNA/gene). Each shRNA sequence is represented 3 times, with 3 different barcodes (see column E and column F):

$$10 \text{ genes} \times 7 \text{ shRNA sequences} \times 3 \text{ barcodes} = 210 \text{ barcodes}$$

The control block is intended for QC (the three shRNA replicates should behave similar to each other, because they are the same shRNA sequence (only the non-transcribed barcode is different). **Merging this block with the body of the library is not recommended**, as many of the same shRNA sequences are already present in the body of the library. If you want to merge, you should first remove all of the shRNA replicates within the control block (2 out of 3 shRNAs for each shRNA sequence), and then also check for duplicates in the body of the library.

If you have any questions, please contact us at tech@cellecta.com.

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
1	label	sequence	order	shRNA#	gene#	replicate#	refseq#	symbol	plasmid	plasmid_20M	C01	C01_20M	D01	D01_20M	D02	D02_20M
2	4501844 NM_001088.1 38903	TAGAAGACGCACCGGGCCTTTGAGATTGAGT	1	1	0	0	NM_001088	AANAT	966	443	3376	811	2464	705	3415	879
3	4501844 NM_001088.1 38904	TAGAAGACGCACCGGTGAAGCCTTTATCTCTC	2	2	0	0	NM_001088	AANAT	1693	777	7019	1686	1556	445	6059	1560
4	4501844 NM_001088.1 38905	TAGAAGACGCACCGGCTTCTATGAGAGGTTT	3	3	0	0	NM_001088	AANAT	2819	1293	3930	944	6963	1993	2877	741
5	4501844 NM_001088.1 38906	TAGAAGACGCACCGGCCAGTGAGTTTCGTTG	4	4	0	0	NM_001088	AANAT	1154	529	3093	743	342	98	2584	665
6	4501844 NM_001088.1 38907	TAGAAGACGCACCGGCCTCACCTTTATGGAG	5	5	0	0	NM_001088	AANAT	1483	680	1586	381	3880	1111	1480	381
7	4501844 NM_001088.1 54352	TAGAAGACGCACCGGGCTCAGCTGAGTATAG	6	6	0	0	NM_001088	AANAT	2801	1285	828	199	5150	1474	771	199
8	4501848 NM_001089.1 46287	TAGAAGACGCACCGGGCTGAAGATTCAGTT	7	1	1	0	NM_001089	ABCA3	920	422	2246	540	2262	647	2575	663
9	4501848 NM_001089.1 46288	TAGAAGACGCACCGGGCCAGCTTATTGGGA	8	2	1	0	NM_001089	ABCA3	823	378	6699	1610	1025	293	6378	1642
10	4501848 NM_001089.1 46289	TAGAAGACGCACCGGGCTCCATACACCTTTG	9	3	1	0	NM_001089	ABCA3	1140	523	1050	252	1410	404	876	226
11	4501848 NM_001089.1 46290	TAGAAGACGCACCGGGCATATATCAGTGGGT	10	4	1	0	NM_001089	ABCA3	1107	508	1154	277	128	37	907	234
12	4501848 NM_001089.1 46291	TAGAAGACGCACCGGGTTCAGTTACACATGG	11	5	1	0	NM_001089	ABCA3	1628	747	5877	1412	5977	1711	5112	1316
13	4501848 NM_001089.1 54353	TAGAAGACGCACCGGCGCATCTCCATGTTTG	12	6	1	0	NM_001089	ABCA3	5104	2341	1765	424	8165	2337	2023	521
14	4501870 NM_003501.1 41765	TAGAAGACGCACCGGGCAGCAGACAAGCAA	13	1	2	0	NM_003501	ACOX3	1916	879	1771	426	1572	450	1095	282
15	4501870 NM_003501.1 41766	TAGAAGACGCACCGGCCTGTCATAGGAAGTT	14	2	2	0	NM_003501	ACOX3	525	241	3834	921	709	203	3570	919
16	4501870 NM_003501.1 41767	TAGAAGACGCACCGGCGGCAGTAATACTAAC	15	3	2	0	NM_003501	ACOX3	1869	857	3005	722	3694	1057	2262	582
17	4501870 NM_003501.1 41768	TAGAAGACGCACCGGCCATTTCTCTAAGTCG	16	4	2	0	NM_003501	ACOX3	1097	503	976	235	2550	730	566	146
18	4501870 NM_003501.1 41769	TAGAAGACGCACCGGGACCTGTCTTTGGAGA	17	5	2	0	NM_003501	ACOX3	2129	977	4144	996	2390	684	2234	575
19	4501870 NM_003501.1 54354	TAGAAGACGCACCGGCGGAGATTGTTGTTGC	18	6	2	0	NM_003501	ACOX3	861	395	3204	770	1738	497	3588	924
20	4501882 NM_001613.1 49980	TAGAAGACGCACCGGCCGATAGAATATGGC	19	1	3	0	NM_001613	ACTA2	2328	1068	3395	816	907	260	2752	709
21	4501882 NM_001613.1 49981	TAGAAGACGCACCGGGCAAGTGATTACCATT	20	2	3	0	NM_001613	ACTA2	1639	752	3226	775	2922	836	3072	791
22	4501882 NM_001613.1 49982	TAGAAGACGCACCGGCGATAGAACATGGCAT	21	3	3	0	NM_001613	ACTA2	1141	523	3531	848	959	275	3547	913
23	4501882 NM_001613.1 49983	TAGAAGACGCACCGGGCGTGAGATTGTTCCG	22	4	3	0	NM_001613	ACTA2	1452	666	1182	284	2359	675	956	246
24	4501882 NM_001613.1 49984	TAGAAGACGCACCGGCCTTGAGAAGAGTTAT	23	5	3	0	NM_001613	ACTA2	2301	1055	4739	1139	3784	1083	5601	1442
25	4501882 NM_001613.1 54355	TAGAAGACGCACCGGCTCTAGCACACAATTG	24	6	3	0	NM_001613	ACTA2	4624	2121	1688	406	10360	2966	1881	484
26	4501892 NM_001103.1 44335	TAGAAGACGCACCGGCGCTTTGCTATTTAGG	25	1	4	0	NM_001103	ACTN2	1434	658	10729	2578	2654	760	11512	2964
27	4501892 NM_001103.1 44336	TAGAAGACGCACCGGGCCTGATGGATTATGA	26	2	4	0	NM_001103	ACTN2	1384	635	2982	716	4053	1160	3354	864
28	4501892 NM_001103.1 44337	TAGAAGACGCACCGGGAATGGAGAAATTGC	27	3	4	0	NM_001103	ACTN2	2880	1321	5223	1255	6457	1848	6731	1733
29	4501892 NM_001103.1 44338	TAGAAGACGCACCGGGAGACAACATCATCA	28	4	4	0	NM_001103	ACTN2	1255	576	3633	873	711	204	3652	940
30	4501892 NM_001103.1 44339	TAGAAGACGCACCGGCGACGAGGGTGAGTA	29	5	4	0	NM_001103	ACTN2	1761	808	3403	818	2910	833	2191	564
31	4501892 NM_001103.1 54356	TAGAAGACGCACCGGCCTGGGATTGGATAGT	30	6	4	0	NM_001103	ACTN2	2040	936	2461	591	5158	1476	2502	644
32	4501900 NM_000666.1 43565	TAGAAGACGCACCGGGCACAAGTTGTAAAC	31	1	5	0	NM_000666	ACY1	1357	622	532	128	2127	609	633	163
33	4501900 NM_000666.1 43566	TAGAAGACGCACCGGCACCTTAGAGTTTGCT	32	2	5	0	NM_000666	ACY1	684	314	5981	1437	2266	649	5548	1429
34	4501900 NM_000666.1 43567	TAGAAGACGCACCGGTGCCACTGATAACCGT	33	3	5	0	NM_000666	ACY1	1095	502	1914	460	3906	1118	1966	506
35	4501900 NM_000666.1 43568	TAGAAGACGCACCGGCACATGATCTTTGTGC	34	4	5	0	NM_000666	ACY1	850	390	6973	1675	2098	601	7989	2057
36	4501900 NM_000666.1 53551	TAGAAGACGCACCGGTGCCACTGATAACCGT	35	5	5	0	NM_000666	ACY1	1589	729	1451	349	1803	516	683	176
37	4501900 NM_000666.1 54357	TAGAAGACGCACCGGCCTGGAACCTCTAAAT	36	6	5	0	NM_000666	ACY1	2349	1077	2794	671	5787	1657	2789	718
38	4501944 NM_001124.1 49019	TAGAAGACGCACCGGCCTCACTATTACTTGA	37	1	6	0	NM_001124	ADM	1905	874	2620	630	8348	2390	2739	705
39	4501944 NM_001124.1 49020	TAGAAGACGCACCGGCCGCCAGAGTATGAAC	38	2	6	0	NM_001124	ADM	811	372	2481	596	2287	655	1298	334
40	4501944 NM_001124.1 49021	TAGAAGACGCACCGGGTTTCGAAAGAAGTG	39	3	6	0	NM_001124	ADM	2096	961	5966	1433	2008	575	6047	1557
41	4501944 NM_001124.1 53448	TAGAAGACGCACCGGGTACATACAGAGGTA	40	4	6	0	NM_001124	ADM	1340	615	3167	761	669	191	2834	730
42	4501944 NM_001124.1 53449	TAGAAGACGCACCGGCCCACTTTCTTTGGGA	41	5	6	0	NM_001124	ADM	1285	589	6635	1594	1471	421	5111	1316
43	4501944 NM_001124.1 54358	TAGAAGACGCACCGGCCAGGTTAAGGAGAC	42	6	6	0	NM_001124	ADM	431	198	2021	486	1255	359	1772	456
44	4501988 NM_001134.1 44635	TAGAAGACGCACCGGCCCTTGAATGCTAA	43	1	7	0	NM_001134	AFP	1549	711	2122	510	2978	852	1861	479
45	4501988 NM_001134.1 44636	TAGAAGACGCACCGGCCTGTCTTCTGGAAG	44	2	7	0	NM_001134	AFP	2029	931	2990	718	4596	1316	1919	494