

Supplementary Materials

Pooled Gene Screen - Variant Validation and Thresholds for Inclusion

A total of 27 variants identified in the pooled gene screen were assessed for validation to identify thresholds to classify variants as real or artefact, as well as to determine the likely number of alternate alleles present in the pool. The results of this, alongside manual inspection of the reads mapping to a variant in the SureCall Triage View (similar to IGV), was used to predict whether unassessed variants were likely to be real or sequencing artefacts. Evidence that a variant was an artefact included: the variant was present in multiple pools (three or more) despite having a MAF < 0.001 or no dbSNP ID; the variant was observed at an unexpectedly high frequency of reads in a single pool (ie. >40%); the variant was only present in a single population of amplicons; the variant was called in a region with low coverage (ie <20 reads); the variant was present in reads with multiple spurious variants; or the variant had previously been sequenced by direct sequencing in another DNA pool and was identified as an artefact. If, after collating the evidence, it was still not clear if the variant was likely to be real or not, the variant was validated by direct sequencing. Furthermore, based on the selected validation experiment, variants deemed to be real with a DNA pool frequency of 3.7% –10.6% were predicted to have a single alternate allele, whereas two alternate alleles were predicted for variants with a frequency between 10.7% and 15.5%.

To compare variant calls between the WES and the pooled gene screen data, the variants that fulfilled the inclusion criteria for the filtering strategy 1 (rare potentially pathogenic variants) were assessed in the 55 cases that were sequenced by both methods. Fourteen rare potentially pathogenic variants were identified in the WES data including two small deletions and 12 single nucleotide variants (SNVs). Of these, 12 were called in the DNA pool containing the matching DNA sample, one SNV and one small deletion were absent in the pooled gene screen data. The positions of these variants were manually assessed in the expected DNA pools using Triage View in SureCall, but no alternate alleles were observed. In contrast, a variant was identified in an individual that was

included in both sequencing strategies through the targeted gene screen that was not called in the WES dataset. Upon manual investigation of the WES data, the variant was in fact called, but had a depth of only five reads and was thus not included in the filtered data. This variant was therefore included in the chi squared test which included the pooled gene screen data, but was excluded in the SKAT analysis which only used the WES data.

Supplementary Table S1 – Custom probe panel designed to amplify *ZNF469* as part of the HaloPlex Target Enrichment System (Agilent Technologies). The probe identification code (Probe ID), the region captured and the target strand are reported.

Probe ID	Region	Strand
AM_614228_004711	chr16:88493634-88493932	+
AM_614228_004712	chr16:88493801-88494048	+
AM_614228_004713	chr16:88493804-88494052	-
AM_614228_004714	chr16:88493834-88493964	+
AM_614228_004715	chr16:88493846-88493999	+
AM_614228_004716	chr16:88493876-88494036	+
AM_614228_004717	chr16:88493876-88494036	-
AM_614228_004718	chr16:88493932-88494016	-
AM_614228_004719	chr16:88493960-88494053	-
AM_614228_004720	chr16:88493964-88494057	+
AM_614228_004721	chr16:88493999-88494055	+
AM_614228_004722	chr16:88494048-88494110	+
AM_614228_004723	chr16:88494052-88494113	-
AM_614228_004724	chr16:88494055-88494107	+
AM_614228_004725	chr16:88494071-88494184	+
AM_614228_004726	chr16:88494075-88494184	-
AM_614228_004727	chr16:88494094-88494255	-
AM_614228_004728	chr16:88494094-88494255	+
AM_614228_004729	chr16:88494096-88494344	+
AM_614228_004730	chr16:88494110-88494321	+
AM_614228_004731	chr16:88494122-88494342	+
AM_614228_004732	chr16:88494122-88494342	-
AM_614228_004733	chr16:88494184-88494377	+
AM_614228_004734	chr16:88494315-88494378	-
AM_614228_004735	chr16:88494316-88494379	+
AM_614228_004736	chr16:88494354-88494503	-

Probe ID	Region	Strand
AM_614228_004737	chr16:88494356-88494409	-
AM_614228_004738	chr16:88494356-88494409	+
AM_614228_004739	chr16:88494358-88494507	+
AM_614228_004740	chr16:88494377-88494718	+
AM_614228_004741	chr16:88494381-88494718	-
AM_614228_004742	chr16:88494382-88494450	-
AM_614228_004743	chr16:88494383-88494451	+
AM_614228_004744	chr16:88494387-88494597	-
AM_614228_004745	chr16:88494447-88494603	-
AM_614228_004746	chr16:88494505-88494563	-
AM_614228_004747	chr16:88494520-88494620	-
AM_614228_004748	chr16:88494524-88494616	+
AM_614228_004749	chr16:88494569-88494805	-
AM_614228_004750	chr16:88494583-88494667	+
AM_614228_004751	chr16:88494594-88494828	+
AM_614228_004752	chr16:88494597-88494832	-
AM_614228_004753	chr16:88494603-88494956	-
AM_614228_004754	chr16:88494603-88494956	-
AM_614228_004755	chr16:88494603-88494956	+
AM_614228_004756	chr16:88494603-88494956	+
AM_614228_004757	chr16:88494620-88494758	-
AM_614228_004758	chr16:88494667-88494725	-
AM_614228_004759	chr16:88494725-88494778	-
AM_614228_004760	chr16:88494758-88494832	-
AM_614228_004761	chr16:88494762-88494836	+
AM_614228_004762	chr16:88494804-88495037	+
AM_614228_004763	chr16:88494805-88495038	-
AM_614228_004764	chr16:88494816-88494882	-
AM_614228_004765	chr16:88494817-88494883	+
AM_614228_004766	chr16:88494832-88494932	-
AM_614228_004767	chr16:88494838-88494936	-
AM_614228_004768	chr16:88494840-88494897	+
AM_614228_004769	chr16:88494883-88494938	+
AM_614228_004770	chr16:88494909-88494968	+
AM_614228_004771	chr16:88494909-88494968	-
AM_614228_004772	chr16:88494950-88495055	+
AM_614228_004773	chr16:88494956-88495074	+
AM_614228_004774	chr16:88494956-88495074	-
AM_614228_004775	chr16:88494968-88495066	-
AM_614228_004776	chr16:88494968-88495066	-
AM_614228_004777	chr16:88494968-88495066	+
AM_614228_004778	chr16:88494968-88495066	+
AM_614228_004779	chr16:88495064-88495169	-

Probe ID	Region	Strand
AM_614228_004780	chr16:88495066-88495141	+
AM_614228_004781	chr16:88495066-88495141	-
AM_614228_004782	chr16:88495068-88495173	+
AM_614228_004783	chr16:88495074-88495156	-
AM_614228_004784	chr16:88495074-88495156	+
AM_614228_004785	chr16:88495080-88495152	+
AM_614228_004786	chr16:88495151-88495238	-
AM_614228_004787	chr16:88495152-88495239	+
AM_614228_004788	chr16:88495171-88495239	+
AM_614228_004789	chr16:88495173-88495241	+
AM_614228_004790	chr16:88495195-88495477	-
AM_614228_004791	chr16:88495195-88495477	+
AM_614228_004792	chr16:88495238-88495312	-
AM_614228_004793	chr16:88495241-88495318	+
AM_614228_004794	chr16:88495312-88495613	-
AM_614228_004795	chr16:88495313-88495614	+
AM_614228_004796	chr16:88495325-88495429	+
AM_614228_004797	chr16:88495402-88495554	-
AM_614228_004798	chr16:88495402-88495554	-
AM_614228_004799	chr16:88495433-88495879	-
AM_614228_004800	chr16:88495441-88495548	-
AM_614228_004801	chr16:88495477-88495734	+
AM_614228_004802	chr16:88495477-88495734	-
AM_614228_004803	chr16:88495548-88495766	-
AM_614228_004804	chr16:88495594-88495735	+
AM_614228_004805	chr16:88495598-88495738	-
AM_614228_004806	chr16:88495645-88495732	+
AM_614228_004807	chr16:88495645-88495732	+
AM_614228_004808	chr16:88495655-88495892	+
AM_614228_004809	chr16:88495655-88495893	-
AM_614228_004810	chr16:88495679-88495758	+
AM_614228_004811	chr16:88495770-88495836	+
AM_614228_004812	chr16:88495775-88495880	+
AM_614228_004813	chr16:88495832-88495957	-
AM_614228_004814	chr16:88495836-88495961	+
AM_614228_004815	chr16:88495855-88495905	-
AM_614228_004816	chr16:88495856-88495906	+
AM_614228_004817	chr16:88495875-88495984	+
AM_614228_004818	chr16:88495879-88495984	-
AM_614228_004819	chr16:88495880-88495959	-
AM_614228_004820	chr16:88495892-88496156	+
AM_614228_004821	chr16:88495893-88496157	-
AM_614228_004822	chr16:88495957-88496192	-

Probe ID	Region	Strand
AM_614228_004823	chr16:88495957-88496192	-
AM_614228_004824	chr16:88495959-88496111	-
AM_614228_004825	chr16:88495959-88496111	-
AM_614228_004826	chr16:88495959-88496111	+
AM_614228_004827	chr16:88495959-88496111	+
AM_614228_004828	chr16:88495984-88496076	-
AM_614228_004829	chr16:88496033-88496249	+
AM_614228_004830	chr16:88496042-88496209	+
AM_614228_004831	chr16:88496056-88496276	+
AM_614228_004832	chr16:88496184-88496291	-
AM_614228_004833	chr16:88496184-88496291	+
AM_614228_004834	chr16:88496208-88496286	-
AM_614228_004835	chr16:88496229-88496545	-
AM_614228_004836	chr16:88496229-88496545	+
AM_614228_004837	chr16:88496249-88496312	+
AM_614228_004838	chr16:88496252-88496315	-
AM_614228_004839	chr16:88496276-88496395	-
AM_614228_004840	chr16:88496276-88496395	+
AM_614228_004841	chr16:88496306-88496373	-
AM_614228_004842	chr16:88496307-88496374	+
AM_614228_004843	chr16:88496339-88496502	-
AM_614228_004844	chr16:88496339-88496502	+
AM_614228_004845	chr16:88496395-88496466	+
AM_614228_004846	chr16:88496413-88496481	-
AM_614228_004847	chr16:88496466-88496517	-
AM_614228_004848	chr16:88496466-88496517	+
AM_614228_004849	chr16:88496500-88496632	-
AM_614228_004850	chr16:88496501-88496633	+
AM_614228_004851	chr16:88496517-88496567	+
AM_614228_004852	chr16:88496517-88496567	-
AM_614228_004853	chr16:88496532-88496647	+
AM_614228_004854	chr16:88496534-88496647	-
AM_614228_004855	chr16:88496545-88496747	+
AM_614228_004856	chr16:88496545-88496747	-
AM_614228_004857	chr16:88496567-88496734	-
AM_614228_004858	chr16:88496672-88496808	+
AM_614228_004859	chr16:88496672-88496808	-
AM_614228_004860	chr16:88496708-88496763	-
AM_614228_004861	chr16:88496709-88496764	+
AM_614228_004862	chr16:88496734-88496949	+
AM_614228_004863	chr16:88496734-88496949	-
AM_614228_004864	chr16:88496747-88497176	+
AM_614228_004865	chr16:88496747-88497176	-

Probe ID	Region	Strand
AM_614228_004866	chr16:88496810-88497129	+
AM_614228_004867	chr16:88496879-88497115	-
AM_614228_004868	chr16:88496879-88497115	+
AM_614228_004869	chr16:88496949-88497025	-
AM_614228_004870	chr16:88496949-88497025	+
AM_614228_004871	chr16:88496984-88497104	-
AM_614228_004872	chr16:88497023-88497199	-
AM_614228_004873	chr16:88497023-88497199	-
AM_614228_004874	chr16:88497027-88497203	+
AM_614228_004875	chr16:88497027-88497203	+
AM_614228_004876	chr16:88497043-88497474	+
AM_614228_004877	chr16:88497043-88497478	-
AM_614228_004878	chr16:88497121-88497201	-
AM_614228_004879	chr16:88497121-88497201	+
AM_614228_004880	chr16:88497128-88497253	+
AM_614228_004881	chr16:88497128-88497253	-
AM_614228_004882	chr16:88497129-88497194	+
AM_614228_004883	chr16:88497194-88497384	+
AM_614228_004884	chr16:88497198-88497387	-
AM_614228_004885	chr16:88497203-88497331	+
AM_614228_004886	chr16:88497225-88497369	-
AM_614228_004887	chr16:88497336-88497392	-
AM_614228_004888	chr16:88497337-88497393	+
AM_614228_004889	chr16:88497383-88497596	+
AM_614228_004890	chr16:88497391-88497463	+
AM_614228_004891	chr16:88497391-88497463	+
AM_614228_004892	chr16:88497395-88497466	-
AM_614228_004893	chr16:88497395-88497466	-
AM_614228_004894	chr16:88497470-88497551	+
AM_614228_004895	chr16:88497470-88497551	-
AM_614228_004896	chr16:88497487-88497552	+
AM_614228_004897	chr16:88497511-88497745	+
AM_614228_004898	chr16:88497514-88497749	-
AM_614228_004899	chr16:88497541-88497598	+
AM_614228_004900	chr16:88497551-88497806	-
AM_614228_004901	chr16:88497551-88497806	+
AM_614228_004902	chr16:88497556-88497970	-
AM_614228_004903	chr16:88497600-88497796	-
AM_614228_004904	chr16:88497736-88497888	-
AM_614228_004905	chr16:88497745-88497867	+
AM_614228_004906	chr16:88497753-88498113	+
AM_614228_004907	chr16:88497753-88498113	-
AM_614228_004908	chr16:88497806-88497954	+

Probe ID	Region	Strand
AM_614228_004909	chr16:88497858-88498171	-
AM_614228_004910	chr16:88497871-88497999	-
AM_614228_004911	chr16:88497889-88498005	+
AM_614228_004912	chr16:88497966-88498072	+
AM_614228_004913	chr16:88497970-88498072	-
AM_614228_004914	chr16:88497994-88498106	-
AM_614228_004915	chr16:88497996-88498084	+
AM_614228_004916	chr16:88498018-88498350	-
AM_614228_004917	chr16:88498019-88498351	+
AM_614228_004918	chr16:88498072-88498170	-
AM_614228_004919	chr16:88498084-88498188	+
AM_614228_004920	chr16:88498087-88498191	-
AM_614228_004921	chr16:88498162-88498600	+
AM_614228_004922	chr16:88498162-88498600	-
AM_614228_004923	chr16:88498171-88498445	-
AM_614228_004924	chr16:88498175-88498449	+
AM_614228_004925	chr16:88498191-88498451	-
AM_614228_004926	chr16:88498228-88498603	-
AM_614228_004927	chr16:88498301-88498505	-
AM_614228_004928	chr16:88498367-88498476	-
AM_614228_004929	chr16:88498384-88498546	+
AM_614228_004930	chr16:88498451-88498653	-
AM_614228_004931	chr16:88498476-88498722	-
AM_614228_004932	chr16:88498477-88498723	+
AM_614228_004933	chr16:88498546-88498705	+
AM_614228_004934	chr16:88498628-88498718	+
AM_614228_004935	chr16:88498643-88498781	-
AM_614228_004936	chr16:88498650-88498815	+
AM_614228_004937	chr16:88498661-88498879	-
AM_614228_004938	chr16:88498728-88498903	+
AM_614228_004939	chr16:88498751-88499074	+
AM_614228_004940	chr16:88498751-88499074	-
AM_614228_004941	chr16:88498815-88499046	+
AM_614228_004942	chr16:88498852-88499023	+
AM_614228_004943	chr16:88498878-88499159	+
AM_614228_004944	chr16:88498993-88499131	+
AM_614228_004945	chr16:88499048-88499213	-
AM_614228_004946	chr16:88499049-88499130	-
AM_614228_004947	chr16:88499052-88499217	+
AM_614228_004948	chr16:88499100-88499384	+
AM_614228_004949	chr16:88499100-88499384	-
AM_614228_004950	chr16:88499127-88499412	+
AM_614228_004951	chr16:88499130-88499188	-

Probe ID	Region	Strand
AM_614228_004952	chr16:88499141-88499246	-
AM_614228_004953	chr16:88499148-88499261	+
AM_614228_004954	chr16:88499233-88499333	-
AM_614228_004955	chr16:88499261-88499567	-
AM_614228_004956	chr16:88499289-88499621	+
AM_614228_004957	chr16:88499289-88499621	+
AM_614228_004958	chr16:88499290-88499622	-
AM_614228_004959	chr16:88499290-88499622	-
AM_614228_004960	chr16:88499341-88499425	-
AM_614228_004961	chr16:88499384-88499450	-
AM_614228_004962	chr16:88499386-88499709	-
AM_614228_004963	chr16:88499399-88499572	-
AM_614228_004964	chr16:88499403-88499576	+
AM_614228_004965	chr16:88499450-88499644	-
AM_614228_004966	chr16:88499544-88499682	+
AM_614228_004967	chr16:88499572-88499668	-
AM_614228_004968	chr16:88499596-88499650	-
AM_614228_004969	chr16:88499616-88499689	+
AM_614228_004970	chr16:88499621-88499810	+
AM_614228_004971	chr16:88499622-88499811	-
AM_614228_004972	chr16:88499650-88499800	+
AM_614228_004973	chr16:88499692-88499929	-
AM_614228_004974	chr16:88499711-88499803	+
AM_614228_004975	chr16:88499750-88499903	+
AM_614228_004976	chr16:88499783-88499860	+
AM_614228_004977	chr16:88499800-88499878	-
AM_614228_004978	chr16:88499802-88499934	-
AM_614228_004979	chr16:88499859-88499961	-
AM_614228_004980	chr16:88499859-88499961	+
AM_614228_004981	chr16:88499878-88500325	+
AM_614228_004982	chr16:88499878-88500325	-
AM_614228_004983	chr16:88499926-88500114	+
AM_614228_004984	chr16:88499929-88500117	-
AM_614228_004985	chr16:88499961-88500032	+
AM_614228_004986	chr16:88499994-88500252	-
AM_614228_004987	chr16:88499994-88500252	+
AM_614228_004988	chr16:88500022-88500109	+
AM_614228_004989	chr16:88500039-88500264	+
AM_614228_004990	chr16:88500114-88500444	+
AM_614228_004991	chr16:88500117-88500447	-
AM_614228_004992	chr16:88500125-88500201	-
AM_614228_004993	chr16:88500126-88500202	+
AM_614228_004994	chr16:88500231-88500297	-

Probe ID	Region	Strand
AM_614228_004995	chr16:88500232-88500298	+
AM_614228_004996	chr16:88500297-88500362	-
AM_614228_004997	chr16:88500300-88500393	-
AM_614228_004998	chr16:88500304-88500397	+
AM_614228_004999	chr16:88500345-88500499	-
AM_614228_005000	chr16:88500345-88500499	-
AM_614228_005001	chr16:88500377-88500562	-
AM_614228_005002	chr16:88500377-88500562	+
AM_614228_005003	chr16:88500397-88500630	+
AM_614228_005004	chr16:88500438-88500522	-
AM_614228_005005	chr16:88500495-88500591	+
AM_614228_005006	chr16:88500499-88500591	-
AM_614228_005007	chr16:88500562-88500626	+
AM_614228_005008	chr16:88500562-88500628	-
AM_614228_005009	chr16:88500591-88500950	-
AM_614228_005010	chr16:88500598-88500945	-
AM_614228_005011	chr16:88500599-88500941	+
AM_614228_005012	chr16:88500634-88500813	-
AM_614228_005013	chr16:88500674-88500777	+
AM_614228_005014	chr16:88500674-88500777	-
AM_614228_005015	chr16:88500738-88500793	+
AM_614228_005016	chr16:88500738-88500793	-
AM_614228_005017	chr16:88500756-88500867	-
AM_614228_005018	chr16:88500777-88500953	+
AM_614228_005019	chr16:88500817-88500924	+
AM_614228_005020	chr16:88500817-88500924	+
AM_614228_005021	chr16:88500917-88501158	-
AM_614228_005022	chr16:88500917-88501158	+
AM_614228_005023	chr16:88500927-88501026	+
AM_614228_005024	chr16:88500941-88501002	-
AM_614228_005025	chr16:88500941-88501248	+
AM_614228_005026	chr16:88500945-88501252	-
AM_614228_005027	chr16:88500980-88501133	-
AM_614228_005028	chr16:88501029-88501226	-
AM_614228_005029	chr16:88501029-88501226	-
AM_614228_005030	chr16:88501158-88501277	+
AM_614228_005031	chr16:88501197-88501260	-
AM_614228_005032	chr16:88501216-88501293	-
AM_614228_005033	chr16:88501217-88501294	+
AM_614228_005034	chr16:88501222-88501337	+
AM_614228_005035	chr16:88501226-88501340	-
AM_614228_005036	chr16:88501244-88501597	+
AM_614228_005037	chr16:88501244-88501597	-

Probe ID	Region	Strand
AM_614228_005038	chr16:88501323-88501430	-
AM_614228_005039	chr16:88501324-88501431	+
AM_614228_005040	chr16:88501403-88501643	+
AM_614228_005041	chr16:88501403-88501643	-
AM_614228_005042	chr16:88501413-88501562	+
AM_614228_005043	chr16:88501416-88501565	-
AM_614228_005044	chr16:88501427-88501546	-
AM_614228_005045	chr16:88501548-88501610	-
AM_614228_005046	chr16:88501549-88501611	+
AM_614228_005047	chr16:88501565-88501617	-
AM_614228_005048	chr16:88501566-88501730	-
AM_614228_005049	chr16:88501570-88501734	+
AM_614228_005050	chr16:88501597-88501684	+
AM_614228_005051	chr16:88501617-88501741	-
AM_614228_005052	chr16:88501653-88501709	-
AM_614228_005053	chr16:88501684-88501877	+
AM_614228_005054	chr16:88501684-88501877	-
AM_614228_005055	chr16:88501713-88501883	-
AM_614228_005056	chr16:88501713-88501883	+
AM_614228_005057	chr16:88501730-88502037	-
AM_614228_005058	chr16:88501734-88502041	+
AM_614228_005059	chr16:88501793-88501935	-
AM_614228_005060	chr16:88501803-88501905	-
AM_614228_005061	chr16:88501804-88501906	+
AM_614228_005062	chr16:88501877-88501994	+
AM_614228_005063	chr16:88501877-88501994	-
AM_614228_005064	chr16:88501964-88502039	+
AM_614228_005065	chr16:88501964-88502039	+
AM_614228_005066	chr16:88501964-88502039	-
AM_614228_005067	chr16:88501964-88502039	-
AM_614228_005068	chr16:88501978-88502074	+
AM_614228_005069	chr16:88501979-88502075	-
AM_614228_005070	chr16:88502004-88502069	-
AM_614228_005071	chr16:88502069-88502136	-
AM_614228_005072	chr16:88502070-88502137	+
AM_614228_005073	chr16:88502136-88502196	-
AM_614228_005074	chr16:88502137-88502197	+
AM_614228_005075	chr16:88502178-88502265	+
AM_614228_005076	chr16:88502178-88502269	-
AM_614228_005077	chr16:88502192-88502376	-
AM_614228_005078	chr16:88502192-88502376	+
AM_614228_005079	chr16:88502265-88502415	+
AM_614228_005080	chr16:88502269-88502534	-

Probe ID	Region	Strand
AM_614228_005081	chr16:88502271-88502337	+
AM_614228_005082	chr16:88502271-88502337	-
AM_614228_005083	chr16:88502273-88502538	+
AM_614228_005084	chr16:88502325-88502430	+
AM_614228_005085	chr16:88502347-88502409	-
AM_614228_005086	chr16:88502348-88502410	+
AM_614228_005087	chr16:88502376-88502612	+
AM_614228_005088	chr16:88502376-88502612	-
AM_614228_005089	chr16:88502385-88502589	-
AM_614228_005090	chr16:88502385-88502589	+
AM_614228_005091	chr16:88502415-88502789	+
AM_614228_005092	chr16:88502419-88502789	-
AM_614228_005093	chr16:88502534-88502587	-
AM_614228_005094	chr16:88502538-88502591	+
AM_614228_005095	chr16:88502559-88502625	-
AM_614228_005096	chr16:88502559-88502625	+
AM_614228_005097	chr16:88502589-88502741	+
AM_614228_005098	chr16:88502589-88502741	-
AM_614228_005099	chr16:88502591-88502743	+
AM_614228_005100	chr16:88502612-88502693	-
AM_614228_005101	chr16:88502612-88502693	+
AM_614228_005102	chr16:88502625-88502783	-
AM_614228_005103	chr16:88502625-88502783	+
AM_614228_005104	chr16:88502655-88502753	-
AM_614228_005105	chr16:88502693-88502894	+
AM_614228_005106	chr16:88502694-88502894	-
AM_614228_005107	chr16:88502750-88502841	+
AM_614228_005108	chr16:88502753-88502844	-
AM_614228_005109	chr16:88502762-88502827	-
AM_614228_005110	chr16:88502762-88502827	+
AM_614228_005111	chr16:88502768-88502926	-
AM_614228_005112	chr16:88502841-88503006	+
AM_614228_005113	chr16:88502845-88503099	+
AM_614228_005114	chr16:88502848-88503043	-
AM_614228_005115	chr16:88502888-88503093	-
AM_614228_005116	chr16:88502966-88503162	+
AM_614228_005117	chr16:88503010-88503106	+
AM_614228_005118	chr16:88503020-88503115	-
AM_614228_005119	chr16:88503024-88503119	+
AM_614228_005120	chr16:88503043-88503164	-
AM_614228_005121	chr16:88503124-88503177	-
AM_614228_005122	chr16:88503126-88503265	+
AM_614228_005123	chr16:88503126-88503265	-

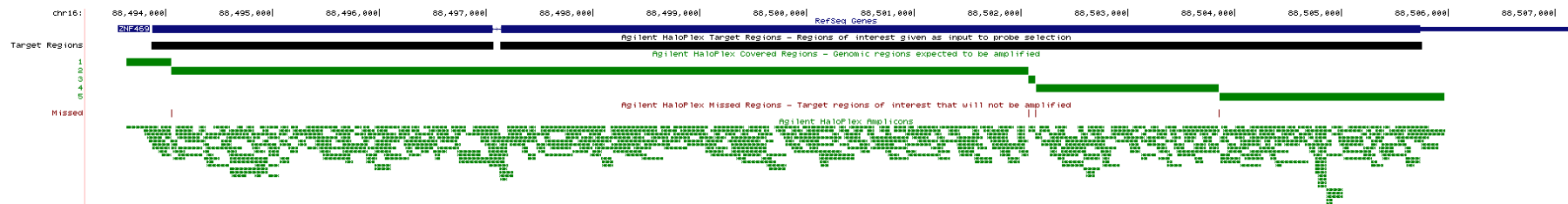
Probe ID	Region	Strand
AM_614228_005124	chr16:88503128-88503181	+
AM_614228_005125	chr16:88503162-88503365	+
AM_614228_005126	chr16:88503177-88503359	-
AM_614228_005127	chr16:88503181-88503363	+
AM_614228_005128	chr16:88503215-88503284	+
AM_614228_005129	chr16:88503274-88503367	-
AM_614228_005130	chr16:88503274-88503367	+
AM_614228_005131	chr16:88503284-88503392	-
AM_614228_005132	chr16:88503284-88503392	+
AM_614228_005133	chr16:88503349-88503459	-
AM_614228_005134	chr16:88503349-88503459	+
AM_614228_005135	chr16:88503365-88503417	-
AM_614228_005136	chr16:88503366-88503418	+
AM_614228_005137	chr16:88503367-88503507	+
AM_614228_005138	chr16:88503395-88503452	-
AM_614228_005139	chr16:88503399-88503456	+
AM_614228_005140	chr16:88503418-88503495	+
AM_614228_005141	chr16:88503431-88503528	+
AM_614228_005142	chr16:88503431-88503528	-
AM_614228_005143	chr16:88503459-88503857	+
AM_614228_005144	chr16:88503459-88503857	-
AM_614228_005145	chr16:88503468-88503632	+
AM_614228_005146	chr16:88503511-88503847	-
AM_614228_005147	chr16:88503525-88503645	-
AM_614228_005148	chr16:88503526-88503646	+
AM_614228_005149	chr16:88503528-88503620	+
AM_614228_005150	chr16:88503620-88503731	+
AM_614228_005151	chr16:88503620-88503731	-
AM_614228_005152	chr16:88503628-88503699	-
AM_614228_005153	chr16:88503632-88503703	+
AM_614228_005154	chr16:88503677-88503759	-
AM_614228_005155	chr16:88503699-88503784	-
AM_614228_005156	chr16:88503703-88503788	+
AM_614228_005157	chr16:88503759-88503850	-
AM_614228_005158	chr16:88503760-88503851	+
AM_614228_005159	chr16:88503788-88503840	+
AM_614228_005160	chr16:88503857-88503931	-
AM_614228_005161	chr16:88503860-88503950	-
AM_614228_005162	chr16:88503864-88503954	+
AM_614228_005163	chr16:88503869-88504057	+
AM_614228_005164	chr16:88503869-88504057	-
AM_614228_005165	chr16:88503873-88503934	-
AM_614228_005166	chr16:88503891-88504207	-

Probe ID	Region	Strand
AM_614228_005167	chr16:88503892-88504208	+
AM_614228_005168	chr16:88503931-88504015	-
AM_614228_005169	chr16:88503932-88504064	+
AM_614228_005170	chr16:88503936-88504068	-
AM_614228_005171	chr16:88504004-88504091	-
AM_614228_005172	chr16:88504008-88504095	+
AM_614228_005173	chr16:88504057-88504243	+
AM_614228_005174	chr16:88504057-88504243	+
AM_614228_005175	chr16:88504057-88504243	-
AM_614228_005176	chr16:88504057-88504243	-
AM_614228_005177	chr16:88504066-88504137	+
AM_614228_005178	chr16:88504095-88504273	+
AM_614228_005179	chr16:88504139-88504471	-
AM_614228_005180	chr16:88504140-88504504	+
AM_614228_005181	chr16:88504140-88504504	-
AM_614228_005182	chr16:88504228-88504376	+
AM_614228_005183	chr16:88504232-88504379	-
AM_614228_005184	chr16:88504232-88504379	-
AM_614228_005185	chr16:88504282-88504332	-
AM_614228_005186	chr16:88504283-88504333	+
AM_614228_005187	chr16:88504331-88504384	-
AM_614228_005188	chr16:88504335-88504388	+
AM_614228_005189	chr16:88504376-88504682	+
AM_614228_005190	chr16:88504379-88504437	+
AM_614228_005191	chr16:88504379-88504686	-
AM_614228_005192	chr16:88504380-88504438	-
AM_614228_005193	chr16:88504388-88504600	+
AM_614228_005194	chr16:88504441-88504564	-
AM_614228_005195	chr16:88504465-88504609	-
AM_614228_005196	chr16:88504486-88504567	-
AM_614228_005197	chr16:88504556-88504658	+
AM_614228_005198	chr16:88504596-88504722	-
AM_614228_005199	chr16:88504600-88504726	+
AM_614228_005200	chr16:88504658-88504911	-
AM_614228_005201	chr16:88504658-88504911	+
AM_614228_005202	chr16:88504686-88504741	-
AM_614228_005203	chr16:88504701-88504754	+
AM_614228_005204	chr16:88504701-88504754	-
AM_614228_005205	chr16:88504736-88504815	-
AM_614228_005206	chr16:88504737-88504816	+
AM_614228_005207	chr16:88504754-88504858	-
AM_614228_005208	chr16:88504754-88504858	-
AM_614228_005209	chr16:88504754-88504858	-

Probe ID	Region	Strand
AM_614228_005210	chr16:88504754-88504858	-
AM_614228_005211	chr16:88504754-88504858	+
AM_614228_005212	chr16:88504754-88504858	+
AM_614228_005213	chr16:88504754-88504858	+
AM_614228_005214	chr16:88504754-88504858	+
AM_614228_005215	chr16:88504776-88504856	-
AM_614228_005216	chr16:88504776-88504856	-
AM_614228_005217	chr16:88504776-88504856	-
AM_614228_005218	chr16:88504776-88504856	-
AM_614228_005219	chr16:88504780-88504860	+
AM_614228_005220	chr16:88504780-88504860	+
AM_614228_005221	chr16:88504856-88504909	-
AM_614228_005222	chr16:88504858-88505012	+
AM_614228_005223	chr16:88504858-88505012	+
AM_614228_005224	chr16:88504858-88505012	-
AM_614228_005225	chr16:88504858-88505012	-
AM_614228_005226	chr16:88504860-88504913	+
AM_614228_005227	chr16:88504860-88504913	+
AM_614228_005228	chr16:88504911-88505008	+
AM_614228_005229	chr16:88504911-88505008	-
AM_614228_005230	chr16:88504967-88505197	-
AM_614228_005231	chr16:88504971-88505201	+
AM_614228_005232	chr16:88504973-88505204	+
AM_614228_005233	chr16:88504977-88505204	-
AM_614228_005234	chr16:88505008-88505070	-
AM_614228_005235	chr16:88505014-88505281	+
AM_614228_005236	chr16:88505018-88505285	-
AM_614228_005237	chr16:88505064-88505254	-
AM_614228_005238	chr16:88505184-88505241	-
AM_614228_005239	chr16:88505204-88505318	-
AM_614228_005240	chr16:88505205-88505319	+
AM_614228_005241	chr16:88505226-88505276	-
AM_614228_005242	chr16:88505230-88505280	+
AM_614228_005243	chr16:88505264-88505376	-
AM_614228_005244	chr16:88505264-88505376	+
AM_614228_005245	chr16:88505276-88505385	-
AM_614228_005246	chr16:88505278-88505337	+
AM_614228_005247	chr16:88505278-88505337	-
AM_614228_005248	chr16:88505280-88505389	+
AM_614228_005249	chr16:88505318-88505400	-
AM_614228_005250	chr16:88505319-88505401	+
AM_614228_005251	chr16:88505350-88505491	+
AM_614228_005252	chr16:88505350-88505491	-

Probe ID	Region	Strand
AM_614228_005253	chr16:88505376-88505606	+
AM_614228_005254	chr16:88505376-88505606	-
AM_614228_005255	chr16:88505385-88505447	-
AM_614228_005256	chr16:88505389-88505451	+
AM_614228_005257	chr16:88505392-88505582	+
AM_614228_005258	chr16:88505447-88505599	-
AM_614228_005259	chr16:88505506-88505814	+
AM_614228_005260	chr16:88505507-88505815	-
AM_614228_005261	chr16:88505528-88505588	-
AM_614228_005262	chr16:88505588-88505646	-
AM_614228_005263	chr16:88505589-88505647	+
AM_614228_005264	chr16:88505603-88505738	+
AM_614228_005265	chr16:88505603-88505738	+
AM_614228_005266	chr16:88505606-88505680	-
AM_614228_005267	chr16:88505614-88505967	-
AM_614228_005268	chr16:88505615-88505963	+
AM_614228_005269	chr16:88505680-88505769	-
AM_614228_005270	chr16:88505742-88505903	-
AM_614228_005271	chr16:88505742-88505903	-

Supplementary Figure S1 – A schematic of the gene screen design (HaloPlex, Agilent Technologies) across *ZNF469* visible as custom tracks on the UCSC genome browser (hg19). The first track in blue shows the RefSeq Gene schematic for *ZNF469*. The black track indicates the target regions for the HaloPlex design. The third track indicates the five covered regions in the HaloPlex design (numbered 1 to 5) in green, which correspond to regions in which at least one probe is designed to amplify. The fourth track indicates in red the regions for which probes could not be designed to cover and the last track shows the 561 amplicons in green, where the direction of the arrows indicates which strand is amplified.



Supplementary Table S2 – A summary of the coverage metrics for the pooled gene screen dataset as reported by Agilent Technologies, averaged across all DNA pools. Agilent reports coverage metrics for each covered region in the assay design. For each covered region (numbered 1-5 as in Supplementary Figure S1), the genomic position, the mean minimum and maximum depths, the mean of the mean depths and the mean number of bases with a depth below 10 for each region are presented. The ranges for these values are also reported. In 34 of the 44 DNA pools a 70bp region in covered region 2, corresponding to the distal end of exon 1, did not reach the minimum depth threshold for inclusion (chr16:88496809-88496879). This region was excluded from analysis. Of the remaining DNA pools, six DNA pools had no bases below threshold, three DNA pools had insufficient coverage for a 2bp region (chr16:88496809-88496810), and one had a 140bp region containing bases below threshold (chr16:88496809-88496949).

Covered region	Genomic position (hg19)	Mean minimum depth (range)	Mean maximum depth (range)	Mean depth (range)	Mean number of bases below threshold (range)
1	chr16:88493639-88494052	282 (113 – 452)	5058 (2249 – 7176)	1676 (713 – 2255)	0
2	chr16:88494053-88502070	10 (0 – 32)	8090 (3791 – 12249)	2559 (1149 – 3623)	57 (0 – 140)
3	chr16:88502074-88502132	1054 (472 – 1865)	1890 (871 – 2822)	1082 (485 – 1897)	0
4	chr16:88502141-88503852	120 (45 – 197)	5705 (2519 – 8305)	2627 (1168 – 3749)	0
5	chr16:88503862-88505962	132 (46 – 227)	12993 (6381 – 19874)	3219 (1475 – 4570)	0