SUPPLEMENTARY INFORMATION

Supplementary Table 1 can be found near the end of this document on p.8-p.9.

Supplementary Table 2 can be found at the end of this document on p.10

Supplementary Figure 1. Formalin-induced paw licking behavior and paw edema in female and male C57BL/6J and C57BL/6N mice. The paw licking response after injection of 2.5% formalin concentration into the right paw of C57BL/6J versus C57BL/6N mice. **(A)** female and **(C)** male mice. Changes in paw edema, as measured by the difference in the ipsilateral paw diameter between before versus after injection (ΔPD), in C57BL/6J and C57BL/6N **(B)** female and **(D)** male mice 1 h after injection of formalin. Data are expressed as the mean ± S.E.M. of 6 animals per group. *$p<0.05$ significantly different from B6J mice.

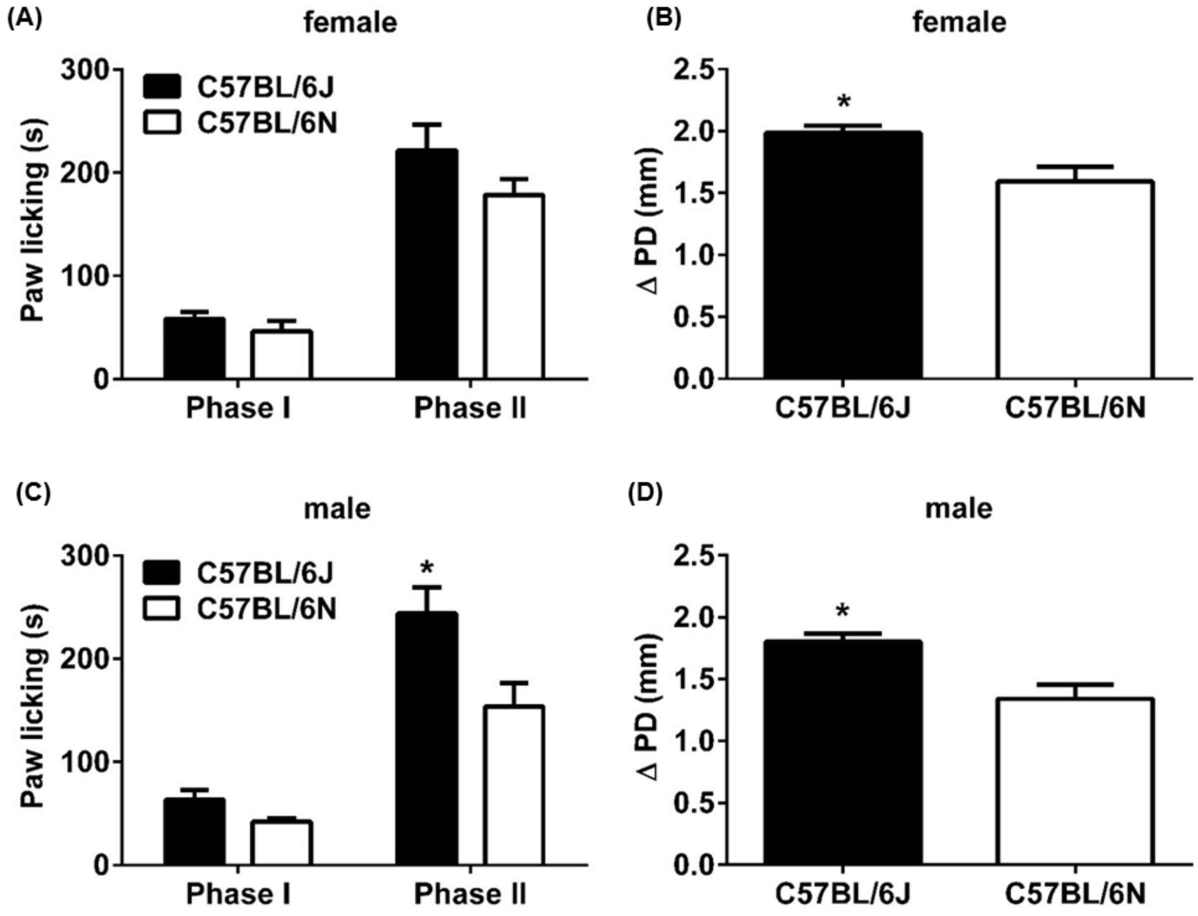
Supplementary Figure 2. CFA-induced mechanical & thermal hypersensitivity and edema in female and male C57BL/6J & C57BL/6N mice. Differences in mechanical paw withdrawal thresholds in C57BL/6J and C57BL/6N **(A)** female and **(C)** male mice at different days after injection of complete Freund's adjuvant (CFA, 10 % solution/20 µl). Differences in paw withdrawal latencies (Δ PWL=contralateral–ipsilateral hindpaw latencies) in C57BL/6J and C57BL/6N **(B)** female and **(D)** male mice. Data are expressed as the mean ± S.E.M. of 6 mice per group. *$p<0.05$ significantly different from baseline (BL).

Supplementary Figure 3. CCI-induced mechanical and thermal hypersensitivity in female and male C57BL/6J and C57BL/6N mice. Differences in mechanical paw withdrawal thresholds in C57BL/6J and C57BL/6N **(A)** female and **(B)** male mice at different times (days) after chronic constrictive nerve injury (CCI) operation. Data are expressed as the mean ± S.E.M. of 6 animals per group. *$p<0.05$ significantly different from the value of baseline (BL).

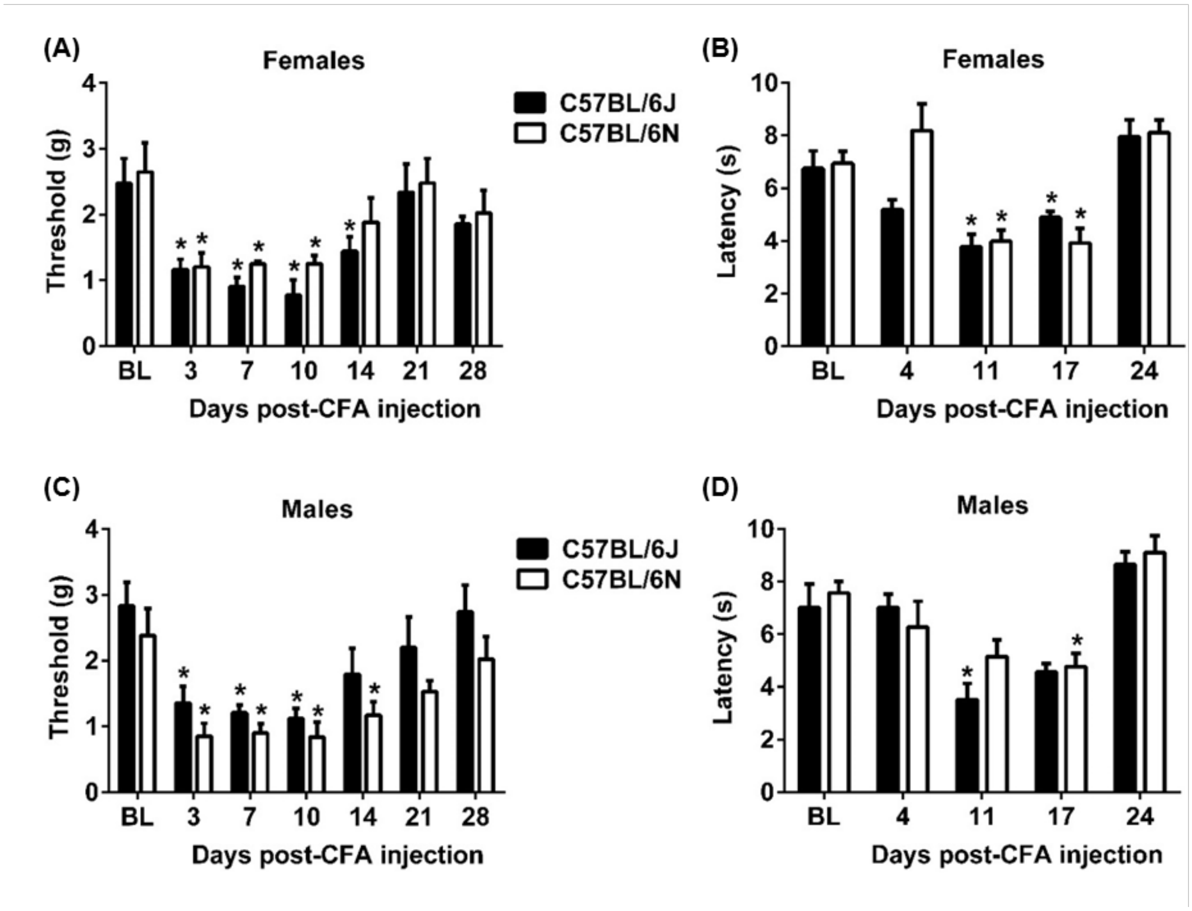
Supplementary Figure 4: B6 parental substrain differences, QTL plots and effect plots for baseline hot plate latency in female versus male B6J x B6NJ-F2 mice. (A): Female and male B6J mice (n=12 females, 12 male) showed lower hot plate latencies than their B6NJ counterparts (n=11females, 7 males). **(B):** When considering female F2 mice alone (n = 78), no genome-wide significant QTL was detected for hot plate sensitivity **(C):** When considering F2 males alone (n = 86), a genome-wide significant QTL was detected on chromosome 7. **(D):** Chromosome 7 LOD plot is shown for F2 females. **(E):** Chromosome 7 LOD plot is shown for F2 males. LOD = 5.67; peak = 15.30 cM (26.64 Mb); Bayes: 8.30-20.30 cM (14.78 Mb – 34.61 Mb); **(F):** Effect plot at peak associated marker (rs3148686; 31 Mb) for females (F, red bars) and males (M, blue bars). Geno = genotype at rs3148686. BB = homozygous for B6J; BN = heterozygous; NN = homozygous for B6NJ. Data for panels A and E are expressed as the mean ± S.E.M. Horizontal lines for panels B through E indicate the significance threshold (1000x permutations).

Supplementary Figure 5. QTLs and effect plots for hot plate latency in B6J x B6NJ-F2 mice with a prior history of repeated SAL versus OXY injections. (A,B): Qualitatively similar, trending but non-significant QTL plots were observed, regardless of whether mice had a prior history of SAL injections (**A;** n = 83) or OXY injections (**B**; n = 81; see Methods for details on treatment history). **(C):** Corresponding to the similarly trending QTL plots, qualitatively similar decreases in hot plate latency were observed in mice with a prior history of SAL injections (black bars) versus mice with a prior history of OXY injections (white bars) injections. Mice with two copies of the B6J allele (BB) showed a lower hot plate latency (s) than mice with two copies of the B6NJ allele (NN). Heterozygous mice (BN) were intermediate. Data are expressed as the mean ± S.E.M.

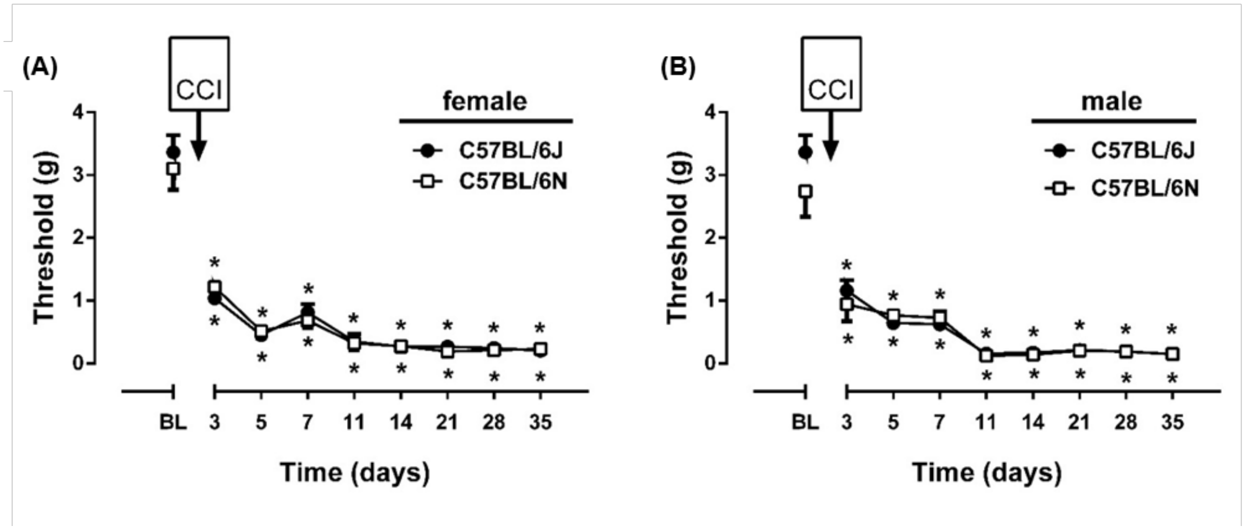
SUPPLEMENTARY FIGURE 1



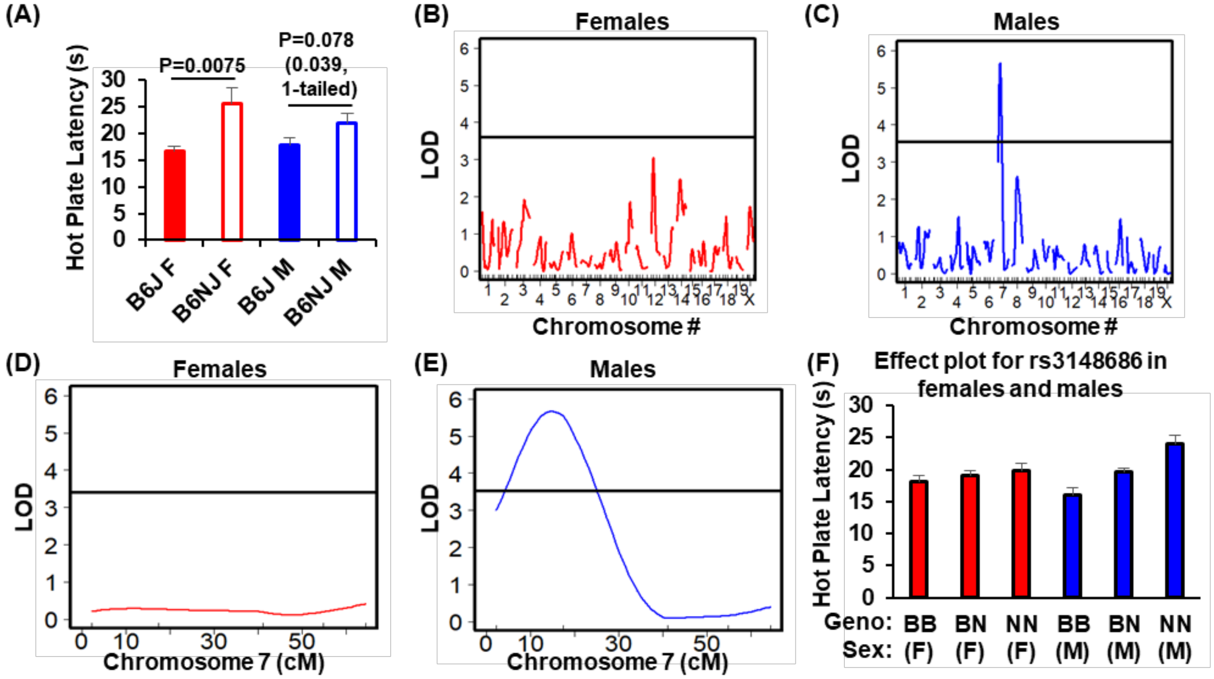
SUPPLEMENTARY FIGURE 2



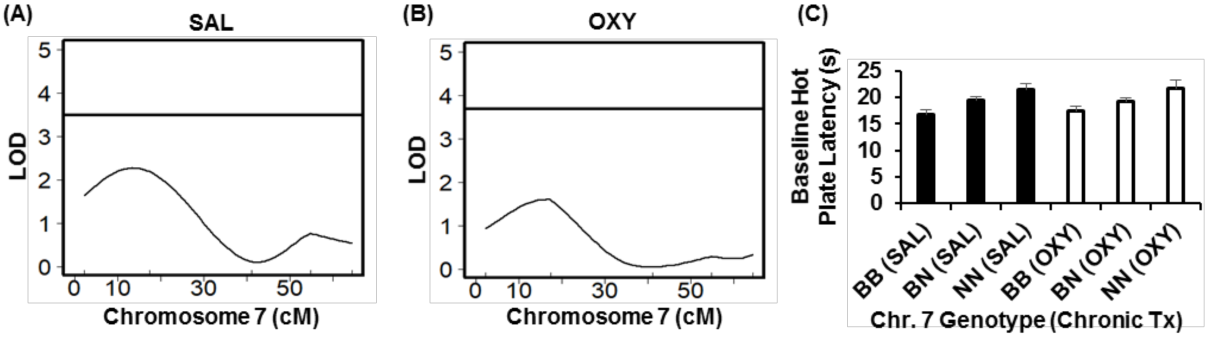
SUPPLEMENTARY FIGURE 3



SUPPLEMENTARY FIGURE 4



SUPPLEMENTARY FIGURE 5



Supplementary Table 1. SNP markers for QTL mapping.

Chr	Position (bp)	cM	B6J	B6NJ	Type	dbSNP	Target
1	29,549,273	11.33	G	A	SNP	rs263412184	Intergenic
1	59,847,167	30.44	G	A	SNP	rs227085647	*Bmpr2*
1	88,806,692	44.98	C	T	SNP	rs242608911	Intergenic
1	116,037,566	51.85	A	C	SNP	rs32180662	*Cntnap5a*
1	163,132,699	72.42	A	T	SNP	rs6341208	Intergenic
1	181,318,003	84.58	A	C	SNP	rs51237371	Intergenic
2	24,187,191	16.24	A	G	SNP	rs254201911	*Il1f9*
2	38,946,612	24.43	A	G	SNP	rs33064547	Intergenic
2	60,051,388	34.36	A	T	SNP	rs256008253	*Baz2b*
2	78,799,176	47.11	C	T	SNP	rs33162749	Intergenic
2	138,480,020	68.27	T	C	SNP	rs13476801	Intergenic
2	152,781,403	75.36	A	G	SNP	rs217443774	*Bcl2l1*
3	5,370,727	1.85	C	T	SNP	rs13476956	Intergenic
3	23,824,920	9.23	T	A	SNP	rs13477019	Intergenic
3	54,914,906	25.91	A	G	SNP	rs30751697	*6030405A18Rik*
3	95,734,876	41.34	A	G	SNP	rs13474735	*Ecm1*
3	119,108,318	52.04	A	G	SNP	rs50526143	*Dpyd*
3	159,811,101	82.64	G	T	SNP	rs260058764	Intergenic
4	21,873,684	9.22	C	G	SNP	rs13460207	*Sfrs18*
4	40,805,010	20.58	A	G	SNP	rs46850777	*B4galt1*
4	65,944,235	34.50	T	C	SNP	rs13477746	Intergenic
4	109,874,495	51.36	A	G	SNP	rs3680956	Intergenic
4	124,654,566	57.85	T	G	SNP	rs32865425	*Pou3f1*
4	138,221,673	70.22	C	T	SNP	rs234702424	*Hp1bp3*
4	150,685,581	81.46	T	C	SNP	rs32407463	Intergenic
5	18,710,388	8.37	A	G	SNP	rs33367397	Intergenic
5	40,761,789	21.97	C	T	SNP	rs33508711	Intergenic
5	59,833,642	31.78	T	C	SNP	rs33209545	Intergenic
5	70,931,531	37.56	T	C	SNP	rs29547790	Intergenic
5	131,541,036	70.96	A	G	SNP	rs3719791	*Auts2*
5	149,026,872	89.13	T	C	SNP	rs33404968	Intergenic
6	39,400,456	18.21	T	A	SNP	rs30899669	*Mkrn1*
6	60,591,379	29.06	A	G	SNP	rs13478783	Intergenic
6	90,159,627	40.07	A	C	SNP	rs245539697	Intergenic
6	117,470,880	55.04	C	G	SNP	rs13478995	Intergenic
6	145,246,710	77.35	A	G	SNP	rs30119689	*Kras*
7	4,006,930	2.30	A	G	SNP	rs31995355	Intergenic
7	30,306,475	17.41	A	G	SNP	rs3148686	*Clip3*
7	71,816,909	41.11	C	G	SNP	rs32060039	Intergenic
7	102,973,309	54.80	C	T	SNP	rs243575509	Intergenic
7	120,135,179	64.46	G	A	SNP	rs235606231	*Zp2*
8	14,127,388	6.54	C	T	SNP	rs236928053	*Dlgap2*
8	29,097,075	16.52	T	C	SNP	rs13479672	Intergenic

8	59,125,561	30.78	T	C	SNP	rs245638874	Intergenic
8	85,365,179	41.58	A	T	SNP	rs245783224	*Mylk3*
8	119,945,761	68.67	C	T	SNP	rs230879074	*Gm20388*
9	3,994,759	0.39	C	T	SNP	rs226723272	Intergenic
9	25,130,622	10.16	C	G	SNP	rs217366063	*Herpud2*
9	58,886,107	31.82	G	A	SNP	rs238448868	*Neo1*
9	86,220,923	46.49	A	C	SNP	rs30522453	Intergenic
9	120,016,907	71.41	T	G	SNP	rs49019729	*Xirp1*
10	29,612,666	16.69	C	A	SNP	rs29330320	Intergenic
10	67,238,174	34.92	T	C	SNP	rs13480628	*Jmjd1c*
10	88,091,833	43.81	T	C	SNP	rs246274290	*4930547N16Rik*
11	30,675,691	18.15	A	C	SNP	rs48169870	Intergenic
11	46,222,615	27.63	G	A	SNP	rs240617401	*Cyfip2*
11	90,480,671	55.09	C	T	SNP	rs238893157	*Stxbp4*
11	120,306,788	84.05	T	C	SNP	rs49027247	Intergenic
12	28,409,763	10.93	G	T	SNP	rs29165094	Intergenic
12	55,758,373	24.10	G	T	SNP	rs6385807	Intergenic
12	91,595,674	44.60	A	C	SNP	rs235238684	Intergenic
12	117,167,625	62.92	C	T	SNP	rs229717662	*Ptprn2*
13	27,037,150	12.24	A	G	SNP	rs13481734	Intergenic
13	52,758,052	27.35	T	C	SNP	rs29248623	Intergenic
13	88,426,920	45.27	G	A	SNP	rs29622109	Intergenic
13	119,143,521	67.25	C	T	SNP	rs29235721	Intergenic
14	9,937,385	5.80	G	A	SNP	rs31187642	Intergenic
14	74,415,721	39.25	T	A	SNP	rs30264676	Intergenic
14	90,847,095	45.14	G	T	SNP	rs262164631	Intergenic
14	116,603,597	60.23	A	C	SNP	rs236603300	Intergenic
15	11,336,383	5.58	G	T	SNP	rs246033409	*Adamts12*
15	37,462,413	14.99	A	T	SNP	rs31810918	Intergenic
15	71,632,551	32.31	T	C	SNP	rs31858887	Intergenic
16	12,261,912	7.23	T	C	SNP	rs4162529	*Shisa9*
16	35,291,544	25.15	G	A	SNP	rs258334795	*Adcy5*
16	60,954,616	36.06	C	G	SNP	rs4193066	Intergenic
16	90,181,009	51.59	C	G	SNP	rs4217166	Intergenic
17	5,332,903	3.15	T	C	SNP	rs4137196	*Arid1b*
17	26,854,246	13.56	A	T	SNP	rs46703123	Intergenic
17	67,752,883	38.88	G	A	SNP	rs33387924	*Lama1*
18	9,450,891	4.87	C	G	SNP	rs29560146	Intergenic
18	54,614,841	29.22	A	C	SNP	rs13483369	Intergenic
18	90,448,757	59.64	A	G	SNP	rs263687961	Intergenic
19	9,027,005	6.04	G	C	SNP	rs31112038	*Ahnak*
19	16,602,218	11.66	T	C	SNP	rs30709918	Intergenic
19	52,359,370	46.77	A	G	SNP	rs30608930	Intergenic
X	64,312,439	34.62	C	T	SNP	rs246222509	Intergenic
X	90,719,435	40.39	A	C	SNP	rs221876655	Intergenic
X	120,419,803	49.63	T	A	SNP	rs226119295	*Pcdh11x*
X	152,774,935	70.34	A	G	SNP	rs226776950	intergenic

Supplementary Table 2. Genes possessing *cis*-eQTLs within the chromosome 7 hot plate QTL that also possess genetic variants.

Chr	Position	Gene	dbSNP	Ref	C57BL_6NJ	Csq
7	12920539	Rps5	rs52344512	G	T	upstream_gene_variant
7	12925890	Rps5	-	T	t/g*	intron_variant upstream_gene_variant downstream_gene_variant
7	25104248	Pou2f2	-	C	c/t	intron_variant
7	25109388	Pou2f2	-	A	a/c	intron_variant
7	25137484	Pou2f2	-	CCTCTGCC	C*	intron_variant upstream_gene_variant _
7	25139545	Pou2f2	rs233121482;rs242658957	CTCTCCTT	ccttctcctt/cctcctt*	intron_variant _
7	25139566	Pou2f2	-	T	TC*	intron_variant _
7	25156931	Pou2f2	-	T	t/g*	intron_variant downstream_gene_variant _
7	27315143	Ltbp4	rs238888133	T	TC*	intron_variant upstream_gene_variant
7	27337648	Ltbp4	-	A	a/c*	5'UTR
7	28772685	Sirt2	-	T	t/g*	intron_variant upstream_gene_variant _
7	28777599	Sirt2	rs235180046	T	TGATA*	intron_variant _
7	28933274	Actn4	-	C	c/a	intron_variant
7	28933277	Actn4	rs587198568	C	c/a	intron_variant
7	28953469	Actn4	-	T	t/g*	intron_variant upstream_gene_variant
7	29034448	Ryr1	rs232379481	CAT	C	intron_variant
7	29034450	Ryr1	-	T	C	intron_variant
7	29034466	Ryr1	-	C	c/g	intron_variant
7	29034467	Ryr1	-	A	a/c	intron_variant
7	29034468	Ryr1	-	C	c/a	intron_variant
7	29044042	Ryr1	-	T	t/g	intron_variant
7	29045754	Ryr1	-	C	c/a	intron_variant
7	29046466	Ryr1	rs243592502	G	g/c	intron_variant
7	29055959	Ryr1	-	G	g/a	intron_variant
7	30305589	Clip3	-	T	t/g*	intron_variant upstream_gene_variant _
7	30306475	Clip3	rs3148686	A	G*	intron_variant upstream_gene_variant _
7	30625151	Cox6b1	-	A	a/c	intron_variant