APPENDIX A

Characteristics of oral cancer patients and controls and genomic concentration of samples

Sample No	Patient Code	Disease Statuse	Ethnicity	Gender	Smoking Statuse	Alcohol Drinking Status	Betel Quid Chewing Status	Age	Genetic Status	Genotype	gDNA Concentration ng/µl
1	01-0001-05	Cancer	Indian	Female	No	No	Yes	65	Wild	GG	823
2	01-0052-05	Cancer	Indian	Male	Yes	Yes	Yes	69	Polymorphism	GA	522
3	02-0004-05	Cancer	Indian	Female	No	Yes	Yes	70	Wild	GG	343
4	04-0007-05	Cancer	Indian	Female	No	Yes	Yes	69	Wild	GG	923
5	06-0002-05	Cancer	Indian	Female	No	No	Yes	65	Wild	GG	900
6	06-0006-05	Cancer	Indian	Female	No	No	Yes	72	Wild	GG	748
7	06-0018-05	Cancer	Indian	Female	No	No	Yes	75	Wild	GG	590
9	06-0043-05	Control	Indian	Male	No	Yes	No	26	Wild	GG	1318
10	06-0063-05	Control	Indian	Female	No	No	No	47	Wild	GG	830
12	01-0009-06	Cancer	Indian	Female	No	No	No	59	Wild	GG	743
13	01-0011-04	Cancer	Indian	Female	No	No	Yes	50	Wild	GG	778
14	01-0013-06	Control	Indian	Male	No	No	No	26	Wild	GG	395
15	04-0014-06	Cancer	Indian	Female	No	Yes	Yes	55	Wild	GG	823
16	06-0008-05	Control	Indian	Female	No	No	Yes	60	Wild	GG	430
17	06-0017-05	Control	Indian	Female	No	No	No	38	Wild	GG	493
18	06-0021-05	Control	Indian	Female	No	No	No	32	Wild	GG	728
19	06-0039-05	Cancer	Indian	Female	No	No	No	28	Polymorphism	GA	835
20	06-0045-06	Cancer	Indian	Female	No	Yes	Yes	67	Wild	GG	555
23	04-0015-05	Cancer	Indian	Female	No	No	Yes	72	Wild	GG	848
25	01-0008-05	Control	Indian	Female	No	No	No	57	Polymorphism	AA	818
26	01-0011-05	Cancer	Indian	Female	No	No	Yes	80	Wild	GG	470

28	01-0013-05	Control	Indian	Female	No	No	No	54	Wild	GG	528
29	07-0003-06	Cancer	Indian	Female	No	No	Yes	53	Wild	GG	502
32	01-0031-05	Control	Indian	Female	No	No	No	45	Wild	GG	810
33	06-0066-06	Cancer	Indian	Female	No	Yes	Yes	71	Wild	GG	378
35	01-0037-05	Cancer	Indian	Female	No	No	Yes	53	Wild	GG	618
36	06-0035-04	Cancer	Indian	Female	No	No	Yes	58	Wild	GG	898
37	04-0006-04	Cancer	Indian	Female	Yes	Yes	Yes	74	Wild	GG	1170
38	01-0004-04	Cancer	Indian	Female	No	No	Yes	57	Wild	GG	1538
39	06-0007-04	Cancer	Indian	Female	No	No	Yes	47	Wild	GG	1308
40	06-0029-06	Cancer	Indian	Female	No	No	Yes	59	Wild	GG	395
41	01-0022-05	Cancer	Indian	Female	No	No	Yes	59	Polymorphism	GA	542
42	01-0012-05	Control	Indian	Female	No	No	No	57	Wild	GG	875
43	06-0037-06	Cancer	Indian	Male	No	No	Yes	49	Wild	GG	733
44	06-0008-04	Cancer	Indian	Female	No	No	Yes	51	Wild	GG	808
45	06-0035-03	Cancer	Indian	Female	No	No	Yes	61	Wild	GG	1140
46	06-0021-04	Cancer	Indian	Female	No	No	Yes	66	Wild	GG	973
47	01-0010-04	Cancer	Indian	Female	No	No	Yes	65	Wild	GG	815
48	01-0008-04	Cancer	Indian	Female	No	Yes	Yes	58	Wild	GG	1018
49	03-0003-04	Cancer	Indian	Female	No	No	Yes	67	Wild	GG	.
50	06-0011-07	Cancer	Indian	Male	Yes	Yes	No	50	Wild	GG	253
51	04-0019-06	Cancer	Indian	Male	No	No	Yes	67	Wild	GG	260
52	07-0001-04	Cancer	Indian	Female	No	No	Yes	69	Polymorphism	GA	150
54	04-0017-06	Cancer	Indian	Female	No	No	Yes	68	Wild	GG	965
55	07-0013-06	Cancer	Indian	Female	No	No	Yes	59	Wild	GG	.
56	06-0022-06	Cancer	Indian	Female	No	Yes	Yes	72	Wild	GG	1188
58	06-0032-03	Cancer	Indian	Female	No	No	Yes	49	Wild	GG	1415
59	06-0028-04	Cancer	Indian	Female	No	No	Yes	68	Polymorphism	GA	925
60	06-0027-05	Cancer	Indian	Male	Yes	No	No	36	Wild	GG	905

61	04-0022-06	Cancer	Indian	Male	Yes	No	Yes	73	Wild	GG	180
62	01-0001-06	Cancer	Indigenous	Male	No	Yes	No	76	Polymorphism	GA	.
63	06-0033-04	Cancer	Indian	Male	No	Yes	Yes	70	Wild	GG	2005
64	04-0010-06	Cancer	Indian	Male	Yes	No	No	50	Wild	GG	465
65	01-0050-05	Cancer	Indian	Female	No	No	Yes	73	Wild	GG	.
66	07-0004-06	Cancer	Indian	Male	Yes	Yes	No	79	Wild	GG	1128
67	01-0032-05	Control	Indian	Male	No	No	No	50	Wild	GG	733
68	01-0006-04	Cancer	Indian	Female	No	No	Yes	80	Wild	GG	860
69	06-0022-05	Control	Indian	Male	No	No	No	43	Wild	GG	950
70	06-0028-05	Cancer	Indian	Female	No	No	Yes	67	Wild	GG	888
71	04-0012-04	Cancer	Indian	Female	Yes	No	No	99	Wild	GG	415
72	07-0001-05	Cancer	Indian	Male	No	No	Yes	52	Wild	GG	560
74	06-0044-05	Control	Indian	Male	No	No	No	76	Polymorphism	GA	773
76	07-0002-05	Cancer	Indian	Female	No	No	Yes	59	Wild	GG	695
77	01-0034-05	Control	Indian	Male	No	Yes	No	57	Wild	GG	1000
78	07-0017-05	Control	Indian	Male	No	No	No	50	Wild	GG	.
79	01-0015-05	Control	Indian	Female	No	No	No	41	Polymorphism	GA	900
80	01-0030-05	Control	Indian	Female	No	No	No	51	Polymorphism	GA	838
81	04-0022-05	Cancer	Indian	Female	No	No	Yes	93	Wild	GG	608
82	06-0052-05	Control	Indian	Male	No	No	No	36	Wild	GG	670
85	06-0011-05	Cancer	Indian	Male	Yes	Yes	Yes	56	Wild	GG	898
86	06-0038-04	Cancer	Indian	Female	No	No	Yes	82	Wild	GG	762
87	07-0012-05	Cancer	Indian	Female	No	No	Yes	71	Wild	GG	883
88	06-0051-05	Cancer	Indian	Male	Yes	Yes	No	65	Wild	GG	1053
89	01-0006-08	Control	Indian	Female	No	No	No	28	Wild	GG	872
90	01-0043-07	Control	Indian	Male	Yes	No	No	22	Wild	GG	590
91	01-0008-06	Cancer	Indian	Female	No	No	Yes	87	Polymorphism	GA	405
92	04-0006-06	Cancer	Indian	Female	No	No	Yes	71	Wild	GG	488

93	04-0004-05	Cancer	Indian	Female	No	Yes	Yes	70	Wild	GG	568
94	04-0004-04	Cancer	Indigenous	Male	Yes	Yes	No	66	Wild	GG	.
95	06-0019-06	Cancer	Indian	Male	Yes	Yes	Yes	76	Wild	GG	.
96	06-0015-06	Cancer	Indian	Female	No	No	Yes	54	Wild	GG	253
97	06-0002-06	Cancer	Indian	Female	No	No	Yes	52	Wild	GG	710
98	06-0001-06	Cancer	Indian	Female	No	Yes	Yes	59	Wild	GG	690
101	06-0075-09	Control	Indian	Female	No	No	Yes	57	Wild	GG	.
102	11-0019-05	Control	Indigenous	Female	No	No	No	40	Wild	GG	355
103	06-0023-05	Control	Indian	Male	Yes	Yes	No	44	Wild	GG	.
104	06-0058-09	Control	Indian	Male	No	Yes	No	63	Polymorphism	GA	.
105	06-0086-09	Control	Indian	Female	No	No	No	48	Wild	GG	.
106	06-0070-09	Control	Indian	Male	No	Yes	Yes	67	Wild	GG	.
107	11-0029-05	Control	Indigenous	Female	No	No	No	70	Wild	GG	780
108	06-0060-09	Control	Indian	Male	Yes	Yes	Yes	60	Wild	GG	.
109	01-0009-05	Control	Indian	Female	No	No	No	54	Wild	GG	868
110	06-0071-09	Control	Indian	Male	No	No	Yes	64	Wild	GG	.
111	06-007609	Control	Indian	Male	No	No	No	42	Wild	GG	.
112	01-0014-05	Control	Indian	Female	No	No	No	49	Wild	GG	565
113	06-0061-09	Control	Indian	Male	Yes	No	No	58	Wild	GG	.
114	11-0023-05	Control	Indigenous	Female	No	Yes	No	40	Wild	GG	553
115	06-0067-09	Control	Indian	Female	No	No	Yes	78	Wild	GG	.
116	10-0016-05	Control	Indigenous	Male	No	No	No	47	Wild	GG	925
117	06-0079-09	Control	Indian	Male	No	Yes	Yes	78	Wild	GG	.
118	06-0073-09	Control	Indian	Male	Yes	Yes	No	75	Wild	GG	.
119	06-0082-09	Control	Indian	Female	No	No	Yes	53	Wild	GG	.
120	06-0077-09	Control	Indian	Female	No	No	Yes	60	Wild	GG	.
121	06-0072-09	Control	Indian	Male	No	No	No	63	Wild	GG	.
122	06-0062-09	Control	Indian	Female	No	No	Yes	55	Wild	GG	.

123	06-0083-09	Control	Indian	Female	No	No	Yes	48	Wild	GG	.
124	06-0059-09	Control	Indian	Female	No	No	Yes	57	Wild	GG	.
125	11-0026-05	Control	Indigenous	Female	No	No	No	54	Wild	GG	780
126	06-0046-05	Control	Indian	Female	No	No	No	31	Wild	GG	908
127	10-0008-04	Control	Indigenous	Female	No	No	No	71	Wild	GG	522
128	10-0008-05	Control	Indigenous	Male	Yes	Yes	No	41	Wild	GG	998
129	10-0007-05	Control	Indigenous	Female	No	No	No	34	Wild	GG	735
130	11-0020-05	Control	Indigenous	Female	No	No	No	36	Wild	GG	423
131	06-0074-09	Control	Indian	Male	Yes	Yes	No	58	Wild	GG	.
132	11-0017-05	Control	Indigenous	Female	No	No	No	47	Wild	GG	978
133	11-0015-05	Control	Indigenous	Female	No	No	No	50	Wild	GG	900
135	06-0081-09	Control	Indian	Male	Yes	Yes	No	46	Wild	GG	.
136	10-0013-05	Control	Indigenous	Female	No	No	No	48	Wild	GG	.
137	06-0064-09	Control	Indian	Male	Yes	Yes	No	42	Wild	GG	.
138	06-0053-05	Control	Indian	Female	No	No	No	29	Wild	GG	525
139	11-0028-05	Control	Indigenous	Male	Yes	Yes	No	58	Wild	GG	260
140	11-0022-05	Control	Indigenous	Male	No	No	Yes	104	Wild	GG	520
141	11-0016-05	Control	Indigenous	Female	No	No	No	48	Wild	GG	958
142	11-0024-05	Control	Indigenous	Male	No	No	No	40	Wild	GG	900
143	10-0009-05	Control	Indigenous	Female	No	No	No	54	Wild	GG	975
145	06-0063-09	Control	Indian	Female	No	No	No	40	Wild	GG	.
146	11-0025-05	Control	Indigenous	Female	No	No	No	47	Wild	GG	372
147	06-0085-09	Control	Indian	Female	No	No	Yes	68	Wild	GG	.
148	06-0078-09	Control	Indian	Male	Yes	Yes	No	40	Wild	GG	.
149	06-0044-05	Control	Indian	Male	No	No	No	27	Polymorphism	GA	880
150	06-0057-09	Control	Indian	Female	No	No	Yes	62	Wild	GG	.
151	06-0056-09	Control	Indian	Male	No	No	No	49	Polymorphism	GA	.
153	06-0040-05	Cancer	Indian	Female	No	No	Yes	75	Wild	GG	212

155	06-0026-05	Cancer	Indian	Female	No	No	Yes	71	Wild	GG	868
156	10-0083-05	Cancer	Indigenous	Male	No	No	No	100	Wild	GG	618
157	10-0080-05	Cancer	Indigenous	Female	No	No	Yes	33	Wild	GG	753
158	11-0014-06	Cancer	Indigenous	Female	No	No	No	59	Wild	GG	878
159	10-0014-05	Control	Indigenous	Female	No	No	No	43	Wild	GG	350
160	11-0003-06	Cancer	Indigenous	Male	Yes	Yes	Yes	65	Wild	GG	202
161	02-0007-05	Cancer	Indigenous	Male	Yes	Yes	No	67	Wild	GG	1020
163	04-0017-06	Cancer	Indian	Female	No	No	Yes	65	Wild	GG	965
164	10-0003-04	Cancer	Indigenous	Female	No	No	No	53	Wild	GG	372
165	11-0005-04	Cancer	Indigenous	Male	Yes	Yes	Yes	57	Wild	GG	628
166	10-0018-05	Cancer	Indigenous	Female	No	Yes	Yes	70	Wild	GG	858
167	11-0017-06	Cancer	Indigenous	Female	No	Yes	Yes	58	Wild	GG	780
168	10-0082-05	Cancer	Indigenous	Female	No	No	No	69	Wild	GG	415
169	10-0001-05	Cancer	Indigenous	Female	No	Yes	Yes	51	Wild	GG	542
170	11-0043-05	Cancer	Indigenous	Female	No	No	Yes	105	Wild	GG	720
171	04-0005-04	Cancer	Indian	Female	No	Yes	Yes	70	Wild	GG	682
172	04-0020-05	Cancer	Indian	Male	Yes	Yes	No	64	Wild	GG	718
173	10-0004-04	Cancer	Indigenous	Female	No	No	Yes	64	Wild	GG	312
174	11-0009-05	Cancer	Indigenous	Male	No	No	No	71	Wild	GG	700
175	10-0019-05	Cancer	Indigenous	Male	Yes	No	No	68	Wild	GG	815
176	11-0039-05	Cancer	Indian	Female	No	No	Yes	52	Wild	GG	888
177	10-0002-04	Cancer	Indigenous	Male	Yes	Yes	No	61	Polymorphism	GA	482
178	10-0016-04	Cancer	Indigenous	Male	Yes	Yes	No	65	Wild	GG	910
179	10-0005-04	Cancer	Indigenous	Male	Yes	Yes	Yes	76	Wild	GG	610
181	10-0015-04	Cancer	Indigenous	Male	Yes	No	No	37	Wild	GG	725
182	11-0016-06	Cancer	Indigenous	Female	No	Yes	Yes	72	Wild	GG	325
183	11-0006-06	Cancer	Indigenous	Female	No	No	Yes	71	Wild	GG	912
184	10-0015-05	Cancer	Indigenous	Female	No	No	Yes	62	Wild	GG	630

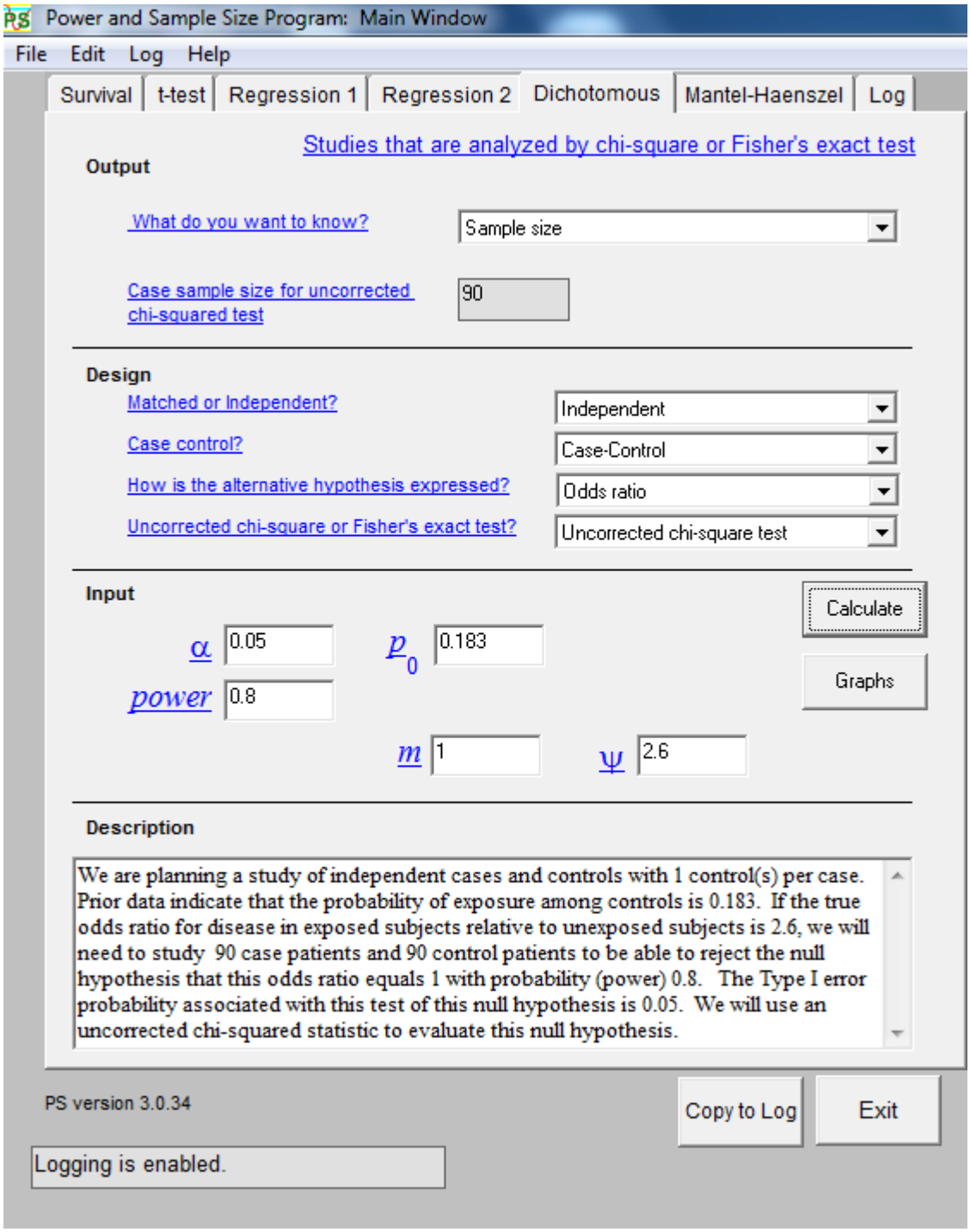
185	04-0019-05	Cancer	Indian	Female	No	No	Yes	55	Wild	GG	608
186	10-0013-04	Cancer	Indigenous	Female	No	No	No	45	Wild	GG	825
187	10-0020-05	Cancer	Indigenous	Female	No	No	Yes	51	Wild	GG	1055
190	11-0008-04	Cancer	Indigenous	Female	No	No	Yes	54	Wild	GG	475
191	10-0017-05	Cancer	Indigenous	Female	No	Yes	Yes	67	Wild	GG	390
192	11-0013-06	Cancer	Indigenous	Male	Yes	Yes	No	43	Wild	GG	735
194	11-0012-06	Cancer	Indigenous	Male	Yes	Yes	Yes	79	Wild	GG	710
195	10-0002-05	Cancer	Indigenous	Female	No	No	Yes	62	Wild	GG	1128
196	10-0079-05	Cancer	Indigenous	Female	No	Yes	Yes	69	Wild	GG	580
198	06-0060-06	Cancer	Indian	Female	No	No	Yes	58	Wild	GG	1100
199	04-0017-06	Cancer	Indian	Female	No	No	Yes	68	Wild	GG	965
200	01-0017-04	Cancer	Indian	Female	No	No	Yes	74	Wild	GG	963
202	06-0026-06	Control	Indian	Female	No	No	No	37	Wild	GG	920
203	06-0058-06	Cancer	Indian	Male	Yes	Yes	No	38	Wild	GG	845
204	06-0045-04	Cancer	Indian	Female	No	Yes	Yes	48	Polymorphism	GA	.
205	07-0003-07	Cancer	Indian	Female	Yes	No	Yes	72	Polymorphism	GA	.
206	01-0063-07	Cancer	Indian	Female	No	No	Yes	50	Wild	GG	.
208	11-0004-04	Cancer	Indigenous	Female	Yes	No	No	59	Polymorphism	GA	698
210	11-0033-05	Cancer	Indigenous	Male	Yes	Yes	No	36	Wild	GG	918
211	11-0040-05	Cancer	Indigenous	Female	Yes	Yes	Yes	63	Wild	GG	1055
212	10-0009-04	Cancer	Indigenous	Male	Yes	Yes	No	47	Wild	GG	548
213	11-0044-05	Cancer	Indigenous	Male	Yes	Yes	No	64	Wild	GG	1088
214	06-0009-06	Cancer	Indigenous	Female	No	No	Yes	61	Wild	GG	725
215	11-0007-06	Cancer	Indigenous	Female	No	No	No	55	Wild	GG	1203
216	06-0031-03	Cancer	Indigenous	Male	Yes	Yes	Yes	66	Wild	GG	1453
217	01-0022-07	Cancer	Indian	Male	Yes	No	No	49	Wild	GG	.
218	01-0017-07	Cancer	Indian	Female	No	No	Yes	61	Wild	GG	.
219	04-0023-06	Cancer	Indian	Female	No	No	No	83	Wild	GG	383

220	01-0059-07	Cancer	Indian	Female	No	No	Yes	55	Wild	GG	.
221	11-0022-06	Cancer	Indigenous	Female	Yes	Yes	Yes	64	Wild	GG	.
223	06-0029-03	Cancer	Indian	Female	No	No	Yes	63	Wild	GG	588
225	01-0051-07	Cancer	Indian	Female	No	No	Yes	72	Polymorphism	AA	.
226	11-0021-06	Cancer	Indigenous	Female	No	No	Yes	50	Wild	GG	673
227	01-0023-07	Cancer	Indian	Female	No	No	Yes	83	Polymorphism	AA	.
228	04-0020-06	Cancer	Indian	Female	No	No	Yes	54	Wild	GG	895
229	06-0033-08	Cancer	Indian	Female	No	No	Yes	70	Wild	GG	.
230	06-0023-09	Cancer	Indian	Female	No	No	Yes	55	Polymorphism	GA	.
231	01-0003-05	Cancer	Indian	Male	No	No	Yes	50	Wild	GG	773
232	06-0001-08	Cancer	Indian	Female	No	No	Yes	61	Wild	GG	.
233	01-0040-07	Cancer	Indian	Male	Yes	Yes	No	73	Wild	GG	.
234	01-0002-04	Cancer	Indian	Female	No	Yes	Yes	80	Polymorphism	GA	1535
235	06-0006-08	Cancer	Indian	Female	No	Yes	Yes	64	Wild	GG	.
236	07-0004-07	Cancer	Indian	Female	No	No	Yes	59	Wild	GG	.
237	10-0004-05	Cancer	Indigenous	Female	No	No	Yes	52	Wild	GG	840
238	06-0023-08	Cancer	Indian	Male	Yes	Yes	Yes	65	Wild	GG	.
239	01-0027-05	Cancer	Indian	Female	No	No	Yes	54	Wild	GG	792
240	11-0015-06	Cancer	Indigenous	Female	No	No	Yes	73	Wild	GG	933
241	07-0007-06	Cancer	Indigenous	Female	Yes	No	No	57	Wild	GG	843
243	06-0013-08	Cancer	Indian	Female	No	Yes	Yes	100	Polymorphism	GA	.
244	07-0001-07	Cancer	Indian	Male	Yes	Yes	Yes	80	Wild	GG	.
245	01-0029-06	Cancer	Indian	Female	No	No	Yes	66	Wild	GG	350
246	01-0030-07	Control	Indigenous	Female	No	No	No	24	Polymorphism	GA	845
247	01-0074-07	Control	Indian	Male	No	No	No	63	Wild	GG	.
248	01-0092-07	Control	Indian	Female	No	No	No	100	Wild	GG	.
250	01-0075-07	Control	Indigenous	Female	No	No	No	88	Wild	GG	.
251	01-0049-08	Control	Indigenous	Male	No	Yes	No	34	Polymorphism	GA	.

252	06-0087-09	Control	Indian	Female	No	No	Yes	49	Wild	GG	.
253	04-0017-05	Cancer	Indian	Female	No	No	No	66	Wild	GG	.
255	01-0001-06	Cancer	Indian	Female	No	No	Yes	71	Wild	GG	372
256	01-0084-07	Control	Indian	Female	No	Yes	No	24	Wild	GG	.
257	07-0014-05	Control	Indian	Female	No	No	No	32	Polymorphism	GA	.

APPENDIX B

Power and Sample Size Program version 3.0.17



APPENDIX C

Agarose gel electrophoresis

Preparation of 50X TAE buffer

Tris base	242 g
Glacial Acetic Acid	57.1 ml
0.5 M EDTA (pH 8.0)	100 ml

1X concentration was diluted with distilled water before use.

Preparation of 1X TAE buffer

$M_1\ V_1 = M_2\ V_2 =$ initial concentration

(50X) (V_1) = (1X) (1000ml) $V_1 =$ volume required

V_1 = 20 ml $M_2 =$ final concentration

$V_2 =$ final volume

Therefore, to prepare 1 liter of 1X TAE buffer, add 20 ml of 50X TAE buffer stock solution and top up to 1000 ml distilled water (diluent).

APPENDIX D

DNA Purification protocol

1. Add 5 volumes of buffer PB to 10 µl of PCR product which was taken from selected PCR sample.
2. Mix the solution well and pipe it into a QIAquick spin column which placed in a 2 ml collection tube.
3. Apply the sample to the QIAquick spin column and centrifuge for 30-60 seconds to bind DNA.
4. Discard flow-through. Place the QIAquick column back into the same tube.
5. Add 750 µl of buffer PE to the QIAquick column and centrifuge for 30-60 seconds for washing.
6. Discard flow-through. Place the QIAquick column back into the same tube. Centrifuge the QIAquick column for additional 1 minute.
7. Place the QIAquick column in a clean 1.5 ml micro centrifuge tube.
8. To elute DNA, add 50 µl of elution buffer (10mM Tris.Cl, pH 8.5) directly to the center of the column and centrifuge it instantly for 1 minute.
9. Alternatively, for increased DNA concentration, add 30µl of elution buffer to the centre of QIAquick membrane. After that, incubate it at room temperature for 1 minute and then centrifuge it.
10. The elution obtained is the purified product.

APPENDIX E

SPSS tables

INDIAN SMOKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	19.123	13397.657	.000	1	.999	20193434 2.539	.000	.
	Constant	-21.203	13397.656	.000	1	.999	.000		

a Variable(s) entered on step 1: Disease_Status.

INDIAN NON-SMOKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-.227	.505	.201	1	.654	.797	.296	2.146
	Constant	-1.609	.387	17.269	1	.000	.200		

a Variable(s) entered on step 1: Disease_Status.

INDIAN ALCOHOL DRINKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	.736	1.173	.393	1	.531	2.087	.209	20.811
	Constant	-2.485	1.041	5.700	1	.017	.083		

a Variable(s) entered on step 1: Disease_Status.

INDIAN NON-ALCOHOL DRINKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-.265	.545	.236	1	.627	.767	.264	2.233
	Constant	-1.665	.412	16.318	1	.000	.189		

a Variable(s) entered on step 1: Disease_Status.

INDIAN BETEL QUID CHEWERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	19.411	10377.780	.000	1	.999	269245744.344	.000	.
	Constant	-21.203	10377.780	.000	1	.998	.000		

a Variable(s) entered on step 1: Disease_Status.

INDIAN NON-BETEL QUID CHEWERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-1.118	1.110	1.015	1	.314	.327	.037	2.877
	Constant	-1.447	.393	13.558	1	.000	.235		

a Variable(s) entered on step 1: Disease_Status.

INDIGENOUS SMOKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	19.063	28420.724	.000	1	.999	190055878.212	.000	.
	Constant	-21.203	28420.724	.000	1	.999	.000		

a Variable(s) entered on step 1: Disease_Status.

INDIGENOUS NON-SMOKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-1.022	1.263	.654	1	.419	.360	.030	4.281
	Constant	-2.197	.745	8.690	1	.003	.111		

a Variable(s) entered on step 1: Disease_Status.

INDIGENOUS ALCOHOL DRINKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-1.204	1.372	.770	1	.380	.300	.020	4.418
	Constant	-1.099	1.155	.905	1	.341	.333		

a Variable(s) entered on step 1: Disease_Status.

INDIGENOUS NON-ALCOHOL DRINKERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Disease_Status(1)	-.258	1.451	.032	1	.859	.773	.045	13.268
	Constant	-2.833	1.029	7.581	1	.006	.059		

a Variable(s) entered on step 1: Disease_Status.

INDIGENOUS BETEL QUID CHEWERS

Warnings

No measures of association are computed for the cross tabulation of Genetic_Status * Disease_Status. At least one variable in each 2-way table upon which measures of association are computed is a constant.

Chi-Square Tests

	Value
Pearson Chi-Square	.(a)
N of Valid Cases	26

a No statistics are computed because Genetic_Status is a constant.

Risk Estimate

	Value
Odds Ratio for Genetic_Status (Wild / .)	.(a)

a No statistics are computed because Genetic_Status is a constant.

Warnings

The dependent variable has less than two non-missing values. For logistic regression, the dependent value must assume exactly two values on the cases being processed.

This command is not executed.

Case Processing Summary

Unweighted Cases(a)		N	Percent
Selected Cases	Included in Analysis	26	16.8
	Missing Cases	129	83.2
	Total	155	100.0
Unselected Cases		0	.0
Total		155	100.0

a If weight is in effect, see classification table for the total number of cases.

INDIGENOUS NON-BETEL QUID CHEWERS

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
		Lower	Upper	Lower	Upper	Lower	Upper	Lower	Upper
Step 1(a)	Genotype(1)	-.517	.972	.283	1	.595	.596	.089	4.008
	Constant	.111	.334	.111	1	.739	1.118		

a Variable(s) entered on step 1: Genotype.

APPENDIX F

a) Calculation of Chi-square (x^2) test to determine Hardy-Weinberg Equilibrium for controls among Malaysian Indians

Genotypes	Observed #	Expected #	x^2 $\Sigma \frac{(O-E)^2}{E}$	p value
GG (Homozygote reference)	49	48.4		
GA (Heterozygote)	7	8.3	1.38	0.24
AA (Homozygote variant)	1	0.4		

1. If P < 0.05 - not consistent with HWE.
2. Not accurate if <5 individuals in any genotype group.

b) Calculation of Chi-square (x^2) test to determine Hardy-Weinberg Equilibrium for controls among Malaysian Indigenous

Genotypes	Observed #	Expected #	x^2 $\Sigma \frac{(O-E)^2}{E}$	p value
GG (Homozygote reference)	20	20		
GA (Heterozygote)	2	1.9	0.05	0.82
AA (Homozygote variant)	0	0		

3. If P < 0.05 - not consistent with HWE.
4. Not accurate if <5 individuals in any genotype group.

APPENDIX G

DNA sequencing blast result: Wild type of the *TNF-α* gene (G allele)

ref> |**NG_007462**| 9763bp DNA linear

Homo sapiens tumor necrosis factor (TNF), RefSeqGene on chromosome 6

Sequences producing significant alignments:

Accession	Description	Max score	Total score	Query coverage	E value	Max ident	Links
8275		193	193	72%	1e-54	99%	

```
>lcl|8275
Length=124

 Score =  193 bits (104),  Expect = 1e-54
 Identities = 106/107 (99%), Gaps = 0/107 (0%)
 Strand=Plus/Minus

Query  2    AGGCAATAGGTTTTGAGGGGCATGGGGACGGGGTTCAGCCTCCAGGGTCCTACACACAAA  61
            |||||||||||||||||| ||||||||||||||||||||||||||||||||||||||||||
Sbjct  121  AGGCAATAGGTTTTGAGGGCCATGGGGACGGGGTTCAGCCTCCAGGGTCCTACACACAAA  62

Query  62   TCAGTCAGTGGCCCAGAAGACCCCCCTCGGAATCGGAGCAGGGAGGA  108
            |||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  61   TCAGTCAGTGGCCCAGAAGACCCCCCTCGGAATCGGAGCAGGGAGGA  15
```

DNA sequencing blast result: variant types of *TNF-α* codon −308 (A allele)

ref> |**NG_007462**| 9763bp DNA linear

Homo sapiens tumor necrosis factor (TNF), RefSeqGene on chromosome 6

Sequences producing significant alignments:

Accession	Description	Max score	Total score	Query coverage	E value	Max ident	Links
45455		189	189	73%	2e-53	98%	

```
>lcl|45455
Length=136

 Score =  189 bits (102),  Expect = 2e-53
 Identities = 106/108 (98%), Gaps = 0/108 (0%)
 Strand=Plus/Minus

Query  1    GAGGCAATAGGTTTTGAGGGGCATGGGGACGGGGTTCAGCCTCCAGGGTCCTACACACAA  60
            |||||||||||||||||||| |||| ||||||||||||||||||||||||||||||||||
Sbjct  124  GAGGCAATAGGTTTTGAGGGCCATGAGGACGGGGTTCAGCCTCCAGGGTCCTACACACAA  65

Query  61   ATCAGTCAGTGGCCCAGAAGACCCCCCTCGGAATCGGAGCAGGGAGGA  108
            ||||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  64   ATCAGTCAGTGGCCCAGAAGACCCCCCTCGGAATCGGAGCAGGGAGGA  17
```