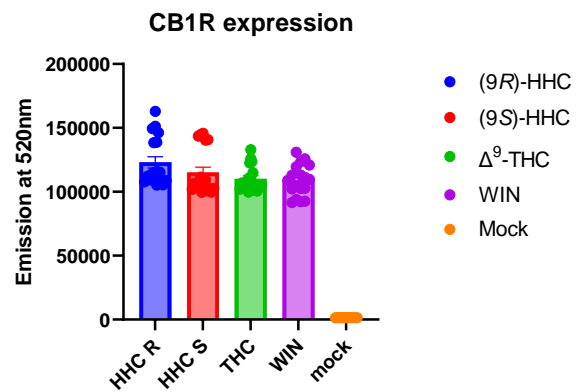


Hexahydrocannabinol (HHC) and Δ^9 -tetrahydrocannabinol (Δ^9 -THC) driven activation of cannabinoid receptor 1 results in biased intracellular signaling

Oleh Durydivka, Petr Palivec, Matej Gazdarica, Ken Mackie, Jaroslav Blahos & Martin Kuchar

Supplementary Information



Supplementary Figure 1. Expression levels of SNAP-CB1R in transiently transfected HEK293 cells used in the receptor internalization assay (related to Fig. 4). Emission of SNAPLumi4-Tb-tagged CB1R-SNAP was measured at 520 ± 10 nm upon excitation at 340 ± 26 nm on Mithras LB 940 microplate reader.

Supplementary Table 1. Curve-fitting analysis of the G₁₁ activation data, presented in Figure 2. LogEC50 and EC50 represent ligand potency, and Top represents ligand efficacy.

Compound	(9S)-HHC	(9R)-HHC	Δ ⁹ -THC	WIN
Best-fit values				
Bottom	-0.008036	-0.02847	-0.02623	-0.01680
Top	-0.06988	-0.1214	-0.1137	-0.1664
LogEC50	-6.597	-7.650	-7.876	-6.818
EC50	2.532e-007	2.238e-008	1.330e-008	1.522e-007
Span	-0.06184	-0.09290	-0.08749	-0.1496
95% CI (profile likelihood)				
Bottom	-0.01980 to 0.003972	-0.05146 to -0.004468	-0.04467 to -0.007311	-0.03316 to -0.0002765
Top	-0.08842 to -0.05251	-0.1484 to -0.09620	-0.1339 to -0.09491	-0.1921 to -0.1434
LogEC50	-7.327 to -5.928	-8.599 to -6.712	-8.648 to -6.966	-7.336 to -6.229
EC50	4.705e-008 to 1.179e-006	2.515e-009 to 1.943e-007	2.248e-009 to 1.081e-007	4.612e-008 to 5.905e-007
Goodness of Fit				
Degrees of Freedom	24	24	24	24
R squared	0.6062	0.5710	0.6702	0.8489
Sum of Squares	0.01192	0.03509	0.02047	0.01982
Sy.x	0.02229	0.03824	0.02921	0.02874

Supplementary Table 2. Curve-fitting analysis of the G_{oA} activation data, presented in Figure 2. LogEC50 and EC50 represent ligand potency, and Top represents ligand efficacy.

Compound	(9S)-HHC	(9R)-HHC	Δ ⁹ -THC	WIN
Best-fit values				
Bottom	-0.0007050	-0.009940	-0.01025	-0.01486
Top	-0.1031	-0.1330	-0.1255	-0.1498
LogEC50	-6.633	-7.623	-8.069	-7.223
EC50	2.329e-007	2.381e-008	8.529e-009	5.986e-008
Span	-0.1024	-0.1230	-0.1153	-0.1349
95% CI (profile likelihood)				
Bottom	-0.008361 to 0.007019	-0.01821 to -0.001553	-0.01768 to -0.002737	-0.02691 to -0.002565
Top	-0.1148 to -0.09178	-0.1423 to -0.1238	-0.1329 to -0.1182	-0.1654 to -0.1345
LogEC50	-6.911 to -6.357	-7.884 to -7.357	-8.298 to -7.846	-7.611 to -6.872
EC50	1.228e-007 to 4.393e-007	1.307e-008 to 4.397e-008	5.036e-009 to 1.425e-008	2.447e-008 to 1.341e-007
Goodness of Fit				
Degrees of Freedom	24	24	24	24
R squared	0.9117	0.9496	0.9581	0.9015
Sum of Squares	0.004908	0.004340	0.003158	0.01042
Sy.x	0.01430	0.01345	0.01147	0.02084

Supplementary Table 3. Curve-fitting analysis of the GRK3-CB1R interaction data, presented in Figure 3. LogEC50 and EC50 represent ligand potency, and Top represents ligand efficacy.

Compound	(9S)-HHC	(9R)-HHC	Δ^9 -THC	WIN
Best-fit values				
Bottom	-0.002394	0.001923	-0.0004729	0.005882
Top	0.02599	0.03543	0.01409	0.06538
LogEC50	-4.983	-6.172	-6.250	-6.712
EC50	1.041e-005	6.731e-007	5.624e-007	1.940e-007
Span	0.02838	0.03351	0.01456	0.05950
95% CI (profile likelihood)				
Bottom	-0.006911 to 0.001333	-0.001247 to 0.005034	-0.003035 to 0.001880	1.282e-005 to 0.01166
Top	0.01168 to 0.5234	0.02976 to 0.04153	0.009846 to 0.01960	0.05691 to 0.07450
LogEC50	-7.057 to -2.711	-6.690 to -5.700	-7.324 to -5.146	-7.163 to -6.245
EC50	8.780e-008 to 0.001943	2.044e-007 to 1.997e-006	4.737e-008 to 7.138e-006	6.872e-008 to 5.690e-007
Goodness of Fit				
Degrees of Freedom	24	24	24	24
R squared	0.5735	0.8513	0.6729	0.8692
Sum of Squares	0.001474	0.0008500	0.0004568	0.002604
Sy.x	0.007837	0.005951	0.004363	0.01042

Supplementary Table 4. Curve-fitting analysis of the β -arrestin2-CB1R interaction data, presented in Figure 3. LogEC50 and EC50 represent ligand potency, and Top represents ligand efficacy.

Compound	(9S)-HHC	(9R)-HHC	Δ^9 -THC	WIN
Best-fit values				
Bottom	-0.002803	-0.005400	0.0005880	0.0008429
Top	0.01618	0.01657	0.004772	0.03307
LogEC50	-4.165	-5.284	-6.765	-6.344
EC50	6.846e-005	5.200e-006	1.717e-007	4.526e-007
Span	0.01899	0.02197	0.004184	0.03223
95% CI (profile likelihood)				
Bottom	-0.004790 to -0.0008480	-0.007917 to -0.002964	-0.001292 to 0.002402	-0.001783 to 0.003441
Top	0.004019 to ND	0.009883 to 0.02542	0.002288 to 0.009830	0.02875 to 0.03762
LogEC50	-5.513 to ND	-6.082 to -4.612	-10.35 to -3.970	-6.687 to -6.013
EC50	3.070e-006 to ND	8.281e-007 to 2.444e-005	4.417e-011 to 0.0001070	2.056e-007 to 9.701e-007
Goodness of Fit				
Degrees of Freedom	24	24	24	24
R squared	0.4298	0.7050	0.2390	0.8877
Sum of Squares	0.0004427	0.0006098	0.0002752	0.0005947
Sy.x	0.004295	0.005041	0.003386	0.004978

ND, non-determinable

Supplementary Table 5. Statistical analysis of the data presented in Fig. 4 by ANOVA

Anova results					
Two-way RM ANOVA	Matching: Stacked				
Assume sphericity?	No				
Alpha	0.05				
Source of Variation	% of total variation	P value	P value summary	Significant?	Geisser-Greenhouse's epsilon
Time x agonist	5.127	<0,0001	****	Yes	
Time	71.2	<0,0001	****	Yes	0.2536
agonist	14.35	<0,0001	****	Yes	
Subject	8.563	<0,0001	****	Yes	
ANOVA table	SS	DF	MS	F (DFn, DFd)	P value
Time x agonist	2162881	18	120160	F (18, 192) = 72,51	P<0,0001
Time	30036521	6	5006087	F (1,521, 48,68) = 3021	P<0,0001
agonist	6054030	3	2018010	F (3, 32) = 17,88	P<0,0001
Subject	3612152	32	112880	F (32, 192) = 68,12	P<0,0001
Residual	318174	192	1657		
Data summary					
Number of columns (agonist)	4				
Number of rows (Time)	7				
Number of subjects (Subject)	36				

Number of missing values 0

Multiple comparisons

Number of families 7

Number of comparisons per family 6

Alpha 0.05

Tukey's multiple comparisons test	Mean Diff,	95,00% CI of diff,	Below threshold?	Summary	Adjusted P Value
------------------------------------------	-------------------	---------------------------	-------------------------	----------------	-------------------------

Row 1

HHC R vs. HHC S	23.95	-118,4 to 166,3	No	ns	0.9619
------------------------	-------	-----------------	----	----	--------

HHC R vs. THC	66.53	-49,63 to 182,7	No	ns	0.3789
----------------------	-------	-----------------	----	----	--------

HHC R vs. WIN	98.79	-12,22 to 209,8	No	ns	0.0878
----------------------	-------	-----------------	----	----	--------

HHC S vs. THC	42.58	-85,73 to 170,9	No	ns	0.768
----------------------	-------	-----------------	----	----	-------

HHC S vs. WIN	74.84	-49,31 to 199,0	No	ns	0.3226
----------------------	-------	-----------------	----	----	--------

THC vs. WIN	32.26	-52,93 to 117,5	No	ns	0.7013
--------------------	-------	-----------------	----	----	--------

Row 2

HHC R vs. HHC S	94.79	-58,51 to 248,1	No	ns	0.321
------------------------	-------	-----------------	----	----	-------

HHC R vs. THC	141.2	1,899 to 280,4	Yes	*	0.0466
----------------------	-------	----------------	-----	---	--------

HHC R vs. WIN	-65.93	-203,0 to 71,17	No	ns	0.505
HHC S vs. THC	46.38	-72,86 to 165,6	No	ns	0.6782
HHC S vs. WIN	-160.7	-277,0 to -44,47	Yes	**	0.0064
THC vs. WIN	-207.1	-295,8 to -118,4	Yes	****	<0,0001
Row 3					
HHC R vs. HHC S	131.7	-53,52 to 317,0	No	ns	0.2159
HHC R vs. THC	203.9	35,64 to 372,1	Yes	*	0.0161
HHC R vs. WIN	-215.3	-387,5 to -43,04	Yes	*	0.0128
HHC S vs. THC	72.16	-77,46 to 221,8	No	ns	0.5212
HHC S vs. WIN	-347	-501,6 to -192,4	Yes	****	<0,0001
THC vs. WIN	-419.2	-547,9 to -290,4	Yes	****	<0,0001
Row 4					
HHC R vs. HHC S	135.5	-64,82 to 335,8	No	ns	0.2528
HHC R vs. THC	241.6	63,21 to 420,1	Yes	**	0.0072
HHC R vs. WIN	-269.6	-467,9 to -71,25	Yes	**	0.0065
HHC S vs. THC	106.1	-64,86 to 277,1	No	ns	0.3141
HHC S vs. WIN	-405.1	-597,4 to -212,8	Yes	****	<0,0001
THC vs. WIN	-511.2	-679,5 to -342,9	Yes	****	<0,0001

Row 5

HHC R vs. HHC S	113.4	-93,51 to 320,4	No	ns	0.423
HHC R vs. THC	242.8	63,43 to 422,1	Yes	**	0.0071
HHC R vs. WIN	-316.4	-516,1 to -116,7	Yes	**	0.0017
HHC S vs. THC	129.3	-52,46 to 311,1	No	ns	0.2116
HHC S vs. WIN	-429.8	-631,6 to -228,1	Yes	****	<0,0001
THC vs. WIN	-559.2	-731,6 to -386,7	Yes	****	<0,0001

Row 6

HHC R vs. HHC S	44.44	-203,6 to 292,5	No	ns	0.9548
HHC R vs. THC	233.6	26,44 to 440,7	Yes	*	0.0252
HHC R vs. WIN	-376.5	-612,2 to -140,8	Yes	**	0.0016
HHC S vs. THC	189.1	-25,76 to 404,0	No	ns	0.0928
HHC S vs. WIN	-420.9	-662,7 to -179,1	Yes	****	0.0007
THC vs. WIN	-610.1	-808,3 to -411,8	Yes	****	<0,0001

Row 7

HHC R vs. HHC S	-78.04	-319,9 to 163,9	No	ns	0.7901
HHC R vs. THC	232.9	42,50 to 423,3	Yes	*	0.0144
HHC R vs. WIN	-481.6	-702,0 to -261,3	Yes	****	<0,0001
HHC S vs. THC	311	83,12 to 538,8	Yes	**	0.007

HHC S vs. WIN	-403.6	-653,8 to -153,3	Yes	**	0.0015
THC vs. WIN	-714.5	-917,7 to -511,4	Yes	****	<0,0001

Test details	Mean 1	Mean 2	Mean Diff,	SE of diff,	N1	N2	q	DF
Row 1								
HHC R vs. HHC S	2021	1997	23.95	49.67	9	9	0.6 82	15. 74
HHC R vs. THC	2021	1954	66.53	40.09	9	9	2.3 47	14. 37
HHC R vs. WIN	2021	1922	98.79	37.68	9	9	3.7 08	12. 66
HHC S vs. THC	1997	1954	42.58	43.87	9	9	1.3 73	13. 34
HHC S vs. WIN	1997	1922	74.84	41.68	9	9	2.5 4	11. 74
THC vs. WIN	1954	1922	32.26	29.63	9	9	1.5 4	15. 3

Row 2								
HHC R vs. HHC S	2183	2088	94.79	53.36	9	9	2.5 12	15. 43
HHC R vs. THC	2183	2042	141.2	47.24	9	9	4.2 26	12. 59
HHC R vs. WIN	2183	2249	-65.93	46.04	9	9	2.0 25	11. 76
HHC S vs. THC	2088	2042	46.38	41.07	9	9	1.5 97	14. 11
HHC S vs. WIN	2088	2249	-160.7	39.68	9	9	5.7 28	13. 18
THC vs. WIN	2042	2249	-207.1	30.96	9	9	9.4 61	15. 79

Row 3								
--------------	--	--	--	--	--	--	--	--

HHC R vs. HHC S	2410	2279	131.7	64.58	9	9	2.8	15.84	64
HHC R vs. THC	2410	2207	203.9	57.48	9	9	5.0	13.16	27
HHC R vs. WIN	2410	2626	-215.3	59.33	9	9	5.1	14.32	12
HHC S vs. THC	2279	2207	72.16	51.69	9	9	1.9	14.74	48
HHC S vs. WIN	2279	2626	-347	53.74	9	9	9.1	15.32	23
THC vs. WIN	2207	2626	-419.2	44.95	9	9	13.	15.19	82

Row 4

HHC R vs. HHC S	2573	2438	135.5	69.99	9	9	2.7	15.38	95
HHC R vs. THC	2573	2332	241.6	61.41	9	9	5.5	14.65	04
HHC R vs. WIN	2573	2843	-269.6	69.28	9	9	5.5	15.03	9
HHC S vs. THC	2438	2332	106.1	59.08	9	9	2.5	14.4	49
HHC S vs. WIN	2438	2843	-405.1	67.22	9	9	8.5	15.23	99
THC vs. WIN	2332	2843	-511.2	58.23	9	9	12.	14.42	65

Row 5

HHC R vs. HHC S	2667	2553	113.4	72.33	9	9	2.2	15.18	99
HHC R vs. THC	2667	2424	242.8	61.88	9	9	5.5	14.48	33
HHC R vs. WIN	2667	2983	-316.4	69.79	9	9	6.4	15.12	95
HHC S vs. THC	2553	2424	129.3	62.65	9	9	2.9	14.19	19

HHC S vs. WIN	2553	2983	-429.8	70.47	9	9	8.6	15.
							26	92
THC vs. WIN	2424	2983	-559.2	59.7	9	9	13.	14.
							24	74
Row 6								
HHC R vs.	2785	2740	44.44	86.67	9	9	0.7	15.
HHC S							252	97
HHC R vs. THC	2785	2551	233.6	70.98	9	9	4.6	13.
							54	58
HHC R vs. WIN	2785	3161	-376.5	82.35	9	9	6.4	15.
							65	95
HHC S vs. THC	2740	2551	189.1	73.37	9	9	3.6	13.
							45	21
HHC S vs. WIN	2740	3161	-420.9	84.42	9	9	7.0	15.
							51	83
THC vs. WIN	2551	3161	-610.1	68.22	9	9	12.	14.
							65	03
Row 7								
HHC R vs.	2958	3036	-78.04	84.02	9	9	1.3	15.
HHC S							14	13
HHC R vs. THC	2958	2725	232.9	66.19	9	9	4.9	15.
							77	23
HHC R vs. WIN	2958	3440	-481.6	76.94	9	9	8.8	15.
							52	86
HHC S vs. THC	3036	2725	311	77.94	9	9	5.6	13.
							42	4
HHC S vs. WIN	3036	3440	-403.6	87.26	9	9	6.5	15.
							41	65
THC vs. WIN	2725	3440	-714.5	70.26	9	9	14.	14.
							38	58

Supplementary Table 6. Statistical analysis of the data presented in Fig. 5 by ANOVA

Anova results

Two-way RM ANOVA	Matching: Stacked
Assume sphericity?	No
Alpha	0.05

Source of Variation	% of total variation	P value	P value summary	Significant ?	Geisser-Greenhouse's epsilon
Time x agonist	4.528	0.0002	***	Yes	
Time	6.802	<0,0001	****	Yes	0.7975
agonist	5.781	0.4345	ns	No	
Subject	65.85	<0,0001	****	Yes	

ANOVA table	SS	DF	MS	F (DFn, DFd)	P value
Time x agonist	156142	18	8675	F (18, 192) = 2,835	P=0,0002
Time	234564	6	39094	F (4,785, 153,1) = 12,78	P<0,0001
agonist	199339	3	66446	F (3, 32) = 0,9364	P=0,4345
Subject	2270733	32	70960	F (32, 192) = 23,19	P<0,0001
Residual	587435	192	3060		

Data summary

Number of columns (agonist)	4
Number of rows (Time)	7
Number of subjects (Subject)	36

Number of missing values 0

Multiple comparisons

Number of families 7

Number of comparisons per family 6

Alpha 0.05

Tukey's multiple comparisons test	Mean Diff,	95,00% CI of diff,	Below threshold ?	Summary	Adjusted P Value
------------------------------------------	-------------------	---------------------------	--------------------------	----------------	-------------------------

Row 1

R-HHC vs. S-HHC	38.63	-126,7 to 203,9	No	ns	0.9074
------------------------	-------	-----------------	----	----	--------

R-HHC vs. THC	8.796	-144,0 to 161,6	No	ns	0.9983
----------------------	-------	-----------------	----	----	--------

R-HHC vs. WIN	11.8	-132,9 to 156,5	No	ns	0.9951
----------------------	------	-----------------	----	----	--------

S-HHC vs. THC	-29.83	-181,2 to 121,6	No	ns	0.9407
----------------------	--------	-----------------	----	----	--------

S-HHC vs. WIN	-26.82	-170,0 to 116,4	No	ns	0.9469
----------------------	--------	-----------------	----	----	--------

THC vs. WIN	3.008	-123,1 to 129,1	No	ns	0.9999
--------------------	-------	-----------------	----	----	--------

Row 2

R-HHC vs. S-HHC	-11.08	-122,7 to 100,6	No	ns	0.9916
------------------------	--------	-----------------	----	----	--------

R-HHC vs. THC	-20.63	-157,9 to 116,7	No	ns	0.9707
----------------------	--------	-----------------	----	----	--------

R-HHC vs. WIN	-52.16	-209,8 to 105,4	No	ns	0.7636
S-HHC vs. THC	-9.551	-152,4 to 133,3	No	ns	0.9973
S-HHC vs. WIN	-41.09	-203,0 to 120,8	No	ns	0.8789
THC vs. WIN	-31.53	-207,8 to 144,7	No	ns	0.9547
Row 3					
R-HHC vs. S-HHC	17.42	-111,2 to 146,0	No	ns	0.9795
R-HHC vs. THC	-11.64	-164,8 to 141,5	No	ns	0.9961
R-HHC vs. WIN	-177.2	-360,6 to 6,082	No	ns	0.0595
S-HHC vs. THC	-29.06	-177,4 to 119,2	No	ns	0.9404
S-HHC vs. WIN	-194.7	-374,6 to -14,68	Yes	*	0.0326
THC vs. WIN	-165.6	-359,9 to 28,65	No	ns	0.1086
Row 4					
R-HHC vs. S-HHC	35.11	-86,56 to 156,8	No	ns	0.8398
R-HHC vs. THC	8.921	-111,2 to 129,1	No	ns	0.9964
R-HHC vs. WIN	-79.09	-247,3 to 89,17	No	ns	0.5401
S-HHC vs. THC	-26.19	-132,6 to 80,23	No	ns	0.894
S-HHC vs. WIN	-114.2	-275,8 to 47,38	No	ns	0.2106
THC vs. WIN	-88.01	-248,7 to 72,71	No	ns	0.4036

Row 5

R-HHC vs. S-HHC	-8.598	-166,3 to 149,1	No	ns	0.9985
R-HHC vs. THC	4.088	-133,0 to 141,1	No	ns	0.9998
R-HHC vs. WIN	-64.75	-226,0 to 96,55	No	ns	0.6577
S-HHC vs. THC	12.69	-151,2 to 176,6	No	ns	0.9959
S-HHC vs. WIN	-56.15	-238,4 to 126,1	No	ns	0.8142
THC vs. WIN	-68.84	-236,1 to 98,40	No	ns	0.6449

Row 6

R-HHC vs. S-HHC	20.84	-117,5 to 159,2	No	ns	0.9723
R-HHC vs. THC	-17.95	-226,7 to 190,8	No	ns	0.9939
R-HHC vs. WIN	-17.85	-164,9 to 129,2	No	ns	0.985
S-HHC vs. THC	-38.78	-248,0 to 170,4	No	ns	0.9457
S-HHC vs. WIN	-38.69	-186,8 to 109,4	No	ns	0.8759
THC vs. WIN	0.09302	-213,2 to 213,4	No	ns	>0,9999

Row 7

R-HHC vs. S-HHC	14.28	-87,46 to 116,0	No	ns	0.977
R-HHC vs. THC	-4.98	-114,5 to 104,5	No	ns	0.9992
R-HHC vs. WIN	-35.03	-184,1 to 114,1	No	ns	0.9008
S-HHC vs. THC	-19.26	-119,5 to 80,93	No	ns	0.9447

S-HHC vs. WIN	-49.3	-193,6 to 94,98	No	ns	0.7446
THC vs. WIN	-30.05	-178,4 to 118,3	No	ns	0.9327

Test details	Mean 1	Mean 2	Mean Diff,	SE of diff,	N1	N2	q	DF
--------------	--------	--------	------------	-------------	----	----	---	----

Row 1								
R-HHC vs. S-HHC	605	566.4	38.63	57.77	9	9	0.9	16
R-HHC vs. THC	605	596.2	8.796	53.17	9	9	0.2	15.
R-HHC vs. WIN	605	593.2	11.8	49.86	9	9	0.3	14.
S-HHC vs. THC	566.4	596.2	-29.83	52.73	9	9	0.8	15.
S-HHC vs. WIN	566.4	593.2	-26.82	49.39	9	9	0.7	14.
THC vs. WIN	596.2	593.2	3.008	43.92	9	9	0.0	15.
								456
								339
								348
								002
								681
								968
								7

Row 2								
R-HHC vs. S-HHC	569.7	580.8	-11.08	38.92	9	9	0.4	15.
R-HHC vs. THC	569.7	590.3	-20.63	47.05	9	9	0.6	13.
R-HHC vs. WIN	569.7	621.8	-52.16	53.27	9	9	1.3	12.
S-HHC vs. THC	580.8	590.3	-9.551	49.5	9	9	0.2	14.
S-HHC vs. WIN	580.8	621.8	-41.09	55.45	9	9	1.0	13.
THC vs. WIN	590.3	621.8	-31.53	61.42	9	9	0.7	15.
								024
								2
								85
								729
								48
								26
								57

Row 3								
R-HHC vs. S-HHC	594.8	577.4	17.42	44.9	9	9	0.5	15.487
R-HHC vs. THC	594.8	606.5	-11.64	53.24	9	9	0.3	15.092
R-HHC vs. WIN	594.8	772.1	-177.2	62.74	9	9	3.9	13.95
S-HHC vs. THC	577.4	606.5	-29.06	51.28	9	9	0.8	14.014
S-HHC vs. WIN	577.4	772.1	-194.7	61.09	9	9	4.5	12.06
THC vs. WIN	606.5	772.1	-165.6	67.46	9	9	3.4	15.72
Row 4								
R-HHC vs. S-HHC	657.4	622.3	35.11	42.34	9	9	1.1	15.73
R-HHC vs. THC	657.4	648.5	8.921	41.74	9	9	0.3	15.022
R-HHC vs. WIN	657.4	736.5	-79.09	57.98	9	9	1.9	14.29
S-HHC vs. THC	622.3	648.5	-26.19	37.19	9	9	0.9	15.957
S-HHC vs. WIN	622.3	736.5	-114.2	54.79	9	9	2.9	12.48
THC vs. WIN	648.5	736.5	-88.01	54.33	9	9	2.2	12.91
Row 5								
R-HHC vs. S-HHC	643.1	651.7	-8.598	54.48	9	9	0.2	14.232
R-HHC vs. THC	643.1	639	4.088	47.82	9	9	0.1	15.209
R-HHC vs. WIN	643.1	707.9	-64.75	55.61	9	9	1.6	14.47
S-HHC vs. THC	651.7	639	12.69	57	9	9	0.3	15.147

S-HHC vs. WIN	651.7	707.9	-56.15	63.68	9	9	1.2	15.47	98
THC vs. WIN	639	707.9	-68.84	58.09	9	9	1.6	15.76	14

Row 6									
R-HHC vs. S-HHC	647.8	627	20.84	48.35	9	9	0.6	16.094	
R-HHC vs. THC	647.8	665.7	-17.95	70.64	9	9	0.3	12.593	4
R-HHC vs. WIN	647.8	665.6	-17.85	51.32	9	9	0.4	15.92	75
S-HHC vs. THC	627	665.7	-38.78	70.91	9	9	0.7	12.735	52
S-HHC vs. WIN	627	665.6	-38.69	51.69	9	9	1.0	15.59	81
THC vs. WIN	665.7	665.6	0.09302	72.96	9	9	0.0	13.018	38
								03	

Row 7									
R-HHC vs. S-HHC	598.4	584.1	14.28	35.41	9	9	0.5	15.702	41
R-HHC vs. THC	598.4	603.4	-4.98	38.27	9	9	0.1	15.84	99
R-HHC vs. WIN	598.4	633.4	-35.03	51.09	9	9	0.9	13.695	55
S-HHC vs. THC	584.1	603.4	-19.26	34.9	9	9	0.7	15.803	54
S-HHC vs. WIN	584.1	633.4	-49.3	48.62	9	9	1.4	12.34	04
THC vs. WIN	603.4	633.4	-30.05	50.74	9	9	0.8	13.374	36