

EFFECT OF SOME PLANT OILS ON MYCELIAL GROWTH OF *AGARICUS BISPORUS* (LANGE) SING

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Abstract

The levels of 10µl, 30µl and 50µl concentration of plant oils of apricot (*Prunus armeniaca* L.), melon (*Cucumis melo* L.), parsley (*Petroselinum crispum* (Miller) A.W. Hill), pine stone (*Pinus pinea* L.), plum (*Prunus domestica* L.) and watermelon (*Citrullus lanatus* (Thunb) Matsum et Nakai) which was grown in Turkey were used for mycelial growth of *Agaricus bisporus* on the agar medium. After nine days of incubation, the 10µl and 30µl concentrations of apricot oil were activated to varying degrees in stimulative the mycelial growth compared with the control group. In addition, all concentrations of parsley completely showed inhibitory effect against mycelial growth. This study suggests that the data can be used as natural stimulating agents on the mycelial growth of mushroom of apricot and plum kernel oils.

Introduction

Cultivated mushroom is very important in the ecosystem because it biodegrade the substrate and therefore the waste of agriculture products can be used for cultivated mushroom. Many species of the edible mushrooms occur in forests and are harvested either commercially or as an outdoor recreation activity (Hosford *et al.*, 1995). The retinal pigment epithelium (RPE) plays a major role in the development of the anomalous retinal scarring response termed proliferative vitreoretinopathy. The present study was undertaken to investigate whether lectin obtained from *Agaricus bisporus* inhibited human RPE proliferation *In vitro*. Lectin is bounded to RPE cells so that inhibited by preincubation of lectin with asialomucin. Lectin obtained from *Agaricus bisporus* caused a dose-dependent inhibition of RPE proliferation (one-way ANOVA, F = 94.470, p<0.001) that was partially reversible on removal of the lectin (Kent *et al.*, 2003). The stimulation of the germination of *Agaricus bisporus* spores and mycelium of the same species has been shown to be due to a volatile metabolite, diffused into the culture medium and atmosphere. A wide range of other fungi has been found to effect *A. bisporus* spores in a similar manner. There was no evidence that the stimulus was carbon dioxide (Lösel, 1964).

Mushrooms have also been reported as therapeutic foods, useful in preventing diseases such as hypertension, hypercholesterolemia and cancer (Bobek & Galbavý, 1999; Bobek *et al.*, 1995). These functional properties are mainly due to their chemical composition such as fatty acids, dietary fibre, in particular, chitin (Szymczak, 1972; Hiroi & Tsuyuki, 1988; Manzi *et al.*, 2001) and beta glucans (Manzi & Pizzoferrato, 2000). Limited studies concerning with effect of edible oil on the mycelial growth of *A. bisporus* were carried out. The aim of this study was to determine the effect of plant oils on the mycelial growth of mushroom, *A. bisporus*, in culture media. In results, the rapid growth in a short time of mushroom spores will be profitable for mushroom cultivation.

There are studies about the effects of other materials on mycelia growth. Güler *et al.*, (2003) studied the effect of some Turkish propolis on the product quantity of *A.*

bisporus. As a result of this study, propolis showed stimulatory effects on the developmental stages and some parameters of the yield and an early yield of mushrooms, rapid growing and increased of total weight of harvested.

Material and Method

Agaricus bisporus mycelial used in this experiment was obtained from culture Mycology Research Laboratory of Department of Biology, Selçuk University in Konya in Turkey. *A. bisporus* was extremely mature and it's inside cover had not been opened. Apricot (*Prunus armeniaca*), melon (*Cucumis melo*), parsley (*Petroselinum crispum*), pine stone (*Pinus pinea*), plum (*Prunus domestica*) and watermelon (*Citrullus lanatus*) were used to obtain plant oils. Different parts of above mentioned plants (Table 1) were finely powdered. The oils were extracted with diethylether (50°C) in a Soxhlet apparatus. Extract was evaporated in vacuum. The lipid extract was collected in a flask. The extracted lipid was weighed to determine the oil content and stored under nitrogen at 4°C for further analyses (Doğan & Başoğlu, 1985).

The spore print was obtained under sterile conditions. Spores were diluted with 2 ml sterile distilled water and inoculated into prepared potato dextrose agar. Secondary mycelia were obtained from spores germinated on the medium. Essential oils at concentration 10, 30 and 50µl were added into medium autoclaved at 121°C, 1.5 atm at 20 min. Vegetative mycelium pieces of 1cm² size were transferred in the centre of plate containing essential oils and culture medium. All petri dishes were incubated at 23-25°C. The colony diameter was measured and the percentage mycelial inhibition calculated as follows (Deans & Svoboda, 1990). Three replicates of each treatment were run and averages were calculated. Control sets were simultaneously run without using the plant oils.

$$I = \frac{C - T}{C} \times 100$$

I = Inhibition (%)

C = Colony diameter of control (mm)

T = Colony diameter of tested mycelium (mm)

Table 1. Plants used in experiments.

English name	Botanic name	Family	Used parts
Apricot	<i>Prunus armeniaca</i> Lam.	Rosaceae	Kernel
Melon	<i>Cucumis melo</i> L.	Cucurbitaceae	Seed
Parsley	<i>Petroselinum crispum</i> (Miller) A.W. Hill	Apiaceae	Fruit
Pine	<i>Pinus pinea</i> L.	Pinaceae	Seed
Plum	<i>Prunus domestica</i> L.	Rosaceae	Kernel
Watermelon	<i>Citrullus lanatus</i> (Thunb) Matsum et Nakai	Cucurbitaceae	Seed

Mycelial parts of *A. bisporus* showed growth samples inoculated into the PDA medium without using the oils, and were observed for a week period. Mycelial growth in this period exposed the fungicidal and fungistatic activities of the oils, respectively.

Statistical analyses: The SPSS program was used for data analyses. Control group and between groups MANOVA analysis was performed for evaluation. The "Tamhane Test" results in Tables 2 & 3 is shown different groups as a statically. The mean difference is significant at the 0.05 level.

Table 2. Multiple comparison table.

	(I) Groups	(J) Groups	Mean difference (I-J)	Std. Error	Sig.	95% Confidence interval	
						Lower bound	Upper bound
Tamhane	Control	pine	38,7492*	4,89179	,000	22,4704	55,0280
		parsley	41,6437*	4,87993	,000	25,3890	57,8983
		apricot	6,7994	6,70920	1,000	-14,5769	28,1757
		plum	4,7916	6,97591	1,000	-17,4194	27,0027
		watermelon	11,6399	5,96207	,705	-7,3904	30,6702
		melon	13,4364	5,99845	,468	-5,7252	32,5979
Pine	Pine	control	-38,7492*	4,89179	,000	-55,0280	-22,4704
		parsley	2,8945*	,34035	,000	1,7608	4,0282
		apricot	-31,9498*	4,61687	,000	-47,5634	-16,3362
		plum	-33,9576*	4,99655	,000	-50,7811	-17,1340
		watermelon	-27,1093*	3,44215	,000	-38,5531	-15,6655
		melon	-25,3128*	3,50478	,000	-37,1012	-13,5244
Parsley	Parsley	control	-41,6437*	4,87993	,000	-57,8983	-25,3890
		pine	-2,8945*	,34035	,000	-4,0282	-1,7608
		apricot	-34,8443*	4,60430	,000	-50,4349	-19,2537
		plum	-36,8521*	4,98494	,000	-53,6537	-20,0504
		watermelon	-30,0038*	3,42528	,000	-41,4131	-18,5945
		melon	-28,2073*	3,48822	,000	-39,9643	-16,4503
Apricot	Apricot	control	-6,7994	6,70920	1,000	-28,1757	14,5769
		pine	31,9498*	4,61687	,000	16,3362	47,5634
		parsley	34,8443*	4,60430	,000	19,2537	50,4349
		plum	-2,0078	6,78596	1,000	-23,7000	19,6845
		watermelon	4,8405	5,73866	1,000	-13,5685	23,2495
		melon	6,6370	5,77644	,998	-11,9100	25,1839
Plum	Plum	control	-4,7916	6,97591	1,000	-27,0027	17,4194
		pine	33,9576*	4,99655	,000	17,1340	50,7811
		parsley	36,8521*	4,98494	,000	20,0504	53,6537
		apricot	2,0078	6,78596	1,000	-19,6845	23,7000
		watermelon	6,8483	6,04832	,998	-12,5713	26,2678
		melon	8,6447	6,08419	,976	-10,9012	28,1907
Watermelon	Watermelon	control	-11,6399	5,96207	,705	-30,6702	7,3904
		pine	27,1093*	3,44215	,000	15,6655	38,5531
		parsley	30,0038*	3,42528	,000	18,5945	41,4131
		apricot	-4,8405	5,73866	1,000	-23,2495	13,5685
		plum	-6,8483	6,04832	,998	-26,2678	12,5713
		melon	1,7965	4,88879	1,000	-13,7689	17,3618
Melon	Melon	control	-13,4364	5,99845	,468	-32,5979	5,7252
		pine	25,3128*	3,50478	,000	13,5244	37,1012
		parsley	28,2073*	3,48822	,000	16,4503	39,9643
		apricot	-6,6370	5,77644	,998	-25,1839	11,9100
		plum	-8,6447	6,08419	,976	-28,1907	10,9012
		melon	-1,7965	4,88879	1,000	-17,3618	13,7689

Based on observed means

*= The mean difference is significant at the ,05 level

Table 3. According to measurements, the multiple comparison table.

Dependent variable		(I) Groups	(J) Groups	Mean difference (I-J)	Std. Error	Sig.	95% confidence interval			
							Lower bound	Upper bound		
Ten	Tamhane	control	pine	33,4028*	4,99539	,000	16,8932	49,9123		
			parsley	42,0862*	4,88993	,000	25,7983	58,3741		
		pine	control	-33,4028*	4,99539	,000	-49,9123	-16,8932		
			parsley	8,6834*	1,02105	,000	5,2824	12,0845		
			apricot	-29,5166*	5,24792	,000	-47,1374	-11,8957		
			plum	-26,8092*	5,05333	,000	-43,6945	-9,9240		
			watermelon	-16,2128*	3,03380	,000	-26,1226	-6,3029		
			melon	-17,8035*	3,18229	,000	-28,3126	-7,2944		
		parsley	control	-42,0862*	4,88993	,000	-58,3741	-25,7983		
			pine	-8,6834*	1,02105	,000	-12,0845	-5,2824		
			apricot	-38,2000*	5,14763	,000	-55,6303	-20,7697		
			plum	-35,4927*	4,94910	,000	-52,1736	-18,8118		
			watermelon	-24,8962*	2,85681	,000	-34,4120	-15,3804		
			melon	-26,4869*	3,01404	,000	-36,6457	-16,3282		
		apricot	pine	29,5166*	5,24792	,000	11,8957	47,1374		
			parsley	38,2000*	5,14763	,000	20,7697	55,6303		
		plum	pine	26,8092*	5,05333	,000	9,9240	43,6945		
			parsley	35,4927*	4,94910	,000	18,8118	52,1735		
		watermelon	pine	16,2128*	3,03380	,000	6,3029	26,1225		
			parsley	24,8962*	2,85681	,000	15,3804	34,4120		
		melon	pine	17,8035*	3,18229	,000	7,2944	28,3126		
			parsley	26,4869*	3,01404	,000	16,3282	36,6457		
		Thirty	Tamhane	control	pine	41,6724*	4,91957	,000	25,2857	58,0591
					parsley	41,6724*	4,91957	,000	25,2857	58,0591
pine	control			-41,6724*	4,91957	,000	-58,0591	-25,2857		
	apricot			-38,0868*	4,62747	,000	-53,7558	-22,4178		
	plum			-38,3454*	4,96555	,000	-55,0817	-21,6091		
	watermelon			-37,5628*	4,36849	,000	-52,1138	-23,0117		
	melon			-37,4546*	5,14879	,000	-54,8085	-20,1007		
	parsley			control	-41,6724*	4,91957	,000	-58,0591	-25,2857	
apricot				-38,0868*	4,62747	,000	-53,7558	-22,4178		
plum				-38,3454*	4,96555	,000	-55,0817	-21,6091		
watermelon				-37,5628*	4,36849	,000	-52,1138	-23,0117		
melon				-37,4546*	5,14879	,000	-54,8085	-20,1007		
apricot				pine	38,0868*	4,62747	,000	22,4178	53,7558	
	parsley			38,0868*	4,62747	,000	22,4178	53,7558		
plum	pine			38,3454*	4,96555	,000	21,6091	55,0817		
	parsley			38,3454*	4,96555	,000	21,6091	55,0817		
watermelon	pine			37,5628*	4,36849	,000	23,0117	52,1138		
	parsley			37,5628*	4,36849	,000	23,0117	52,1138		
melon	pine			37,4546*	5,14879	,000	20,1007	54,8085		
	parsley			37,4546*	5,14879	,000	20,1007	54,8085		

Table 3. (Cont'd.).

Dependent variable		(I) Groups	(J) Groups	Mean difference (I-J)	Std. Error	Sig.	95% confidence interval	
							Lower bound	Upper bound
Fifty	Tamhane	control	pine	41,1724*	4,83606	,000	25,0639	57,2809
			parsley	41,1724*	4,83606	,000	25,0639	57,2809
			melon	20,4920*	5,36458	,010	3,1355	37,8485
		pine	control	-41,1724*	4,83606	,000	-57,2809	-25,0639
			apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363
			plum	-36,7181*	5,04632	,000	-53,7266	-19,7095
			watermelon	-27,5524*	3,06611	,000	-37,7654	-17,3394
			melon	-20,6804*	2,32191	,000	-28,5063	-12,8544
			parsley	control	-41,1724*	4,83606	,000	-57,2809
		apricot	apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363
			plum	-36,7181*	5,04632	,000	-53,7266	-19,7095
			watermelon	-27,5524*	3,06611	,000	-37,7654	-17,3394
		plum	control	-41,1724*	4,83606	,000	-57,2809	-25,0639
			apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363
			plum	-36,7181*	5,04632	,000	-53,7266	-19,7095
		watermelon	control	-41,1724*	4,83606	,000	-57,2809	-25,0639
			apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363
			plum	-36,7181*	5,04632	,000	-53,7266	-19,7095
		melon	control	-20,4920*	5,36458	,010	-37,8485	-3,1355
			pine	20,6804*	2,32191	,000	12,8544	28,5063
parsley	20,6804*		2,32191	,000	12,8544	28,5063		
pine	28,2460*		4,07837	,000	14,4363	42,0557		
parsley	28,2460*		4,07837	,000	14,4363	42,0557		
plum	36,7181*		5,04632	,000	19,7095	53,7266		
pine	control	-41,1724*	4,83606	,000	-57,2809	-25,0639		
	apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363		
	plum	-36,7181*	5,04632	,000	-53,7266	-19,7095		
parsley	control	-41,1724*	4,83606	,000	-57,2809	-25,0639		
	apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363		
	plum	-36,7181*	5,04632	,000	-53,7266	-19,7095		
watermelon	control	-41,1724*	4,83606	,000	-57,2809	-25,0639		
	apricot	-28,2460*	4,07837	,000	-42,0557	-14,4363		
	plum	-36,7181*	5,04632	,000	-53,7266	-19,7095		
melon	control	-20,4920*	5,36458	,010	-37,8485	-3,1355		
	pine	20,6804*	2,32191	,000	12,8544	28,5063		
	parsley	20,6804*	2,32191	,000	12,8544	28,5063		

Based on observed means

*= The mean difference is significant at the, 05 level

Results and Discussion

The effects of three different concentrations of plant oils (pine stone, parsley, and apricot, plum, and watermelon and melon seed and kernel oils) on the mycelial growth of mushroom are presented in Table 4. The inhibitory effects of oils against mycelial growth of *A. bisporus* changed depending on differences of oils. After 9 days of incubation, both concentrations (10 and 30µl) of apricot oil were activated to varying degrees in stimulative mycelial growth of mushroom and compared with the control test (Table 4). But, all concentrations of parsley completely exhibited inhibitory effect. The mycelial growth of all other samples (except for plum) treated showed slow growth according to control group. Also effects of oils on mycelial growth were different comparing with the control group. In addition, after 19 and 23 days of incubation, 30 and 10 levels of plum oils were activated mycelial growth of mushroom during incubation period. It was observed a slow growth with watermelon and melon, while all concentrations of parsley and, 30 and 50 levels of pine showed weak effect on mycelial growth (Table 5). Also, all concentrations of apricot and plum oils had showed higher a stimulating effect than those of other oils during incubation period. But the effects on mycelial growth of all concentrations of oils were similar upto 5 days. Percentage stimulative activities of 30µl levels of both apricot and plum oils were

found higher according to other concentrations (Table 5). After 21 days of incubation, 30 µl level of melon oil exhibited stimulative effect on mycelial growth of mushroom compared with the control group.

With 1% sesame oil, highest growth was established and less growth period was determined as 27.4 days. All of the samples with sesame oil showed better growth according to control, being the stimulative effect of sesame oil higher than that of bitter almond oil (Kalyoncu *et al.*, 1999). The results suggest that high concentrations may enhance potency of antifungal activity of parsley oil. But all plant oils showed partly stimulatory effects on secondary mycelial growth of mushroom. The variations of inhibitory and stimulatory activities of tested plant oils may be probably due to their fatty acid contents and position in triglycerides. Also, the data can support the use of apricot and plum kernel oils as natural stimulating agents on mycelial growth of mushroom.

The exo-biopolymer production and mycelial growth were substantially increased (almost tripled) by supplementation of certain vegetable oils into the medium. In particular, sunflower oil at the level of 2% led to a significant increase in exo-biopolymer concentration from 2.3 to 7.5 g/l, while the addition of 4% olive oil dramatically increased mycelial biomass of *Cordyceps militaris* from 5.8 to 19.0 g/l (Park *et al.*, 2002).

Table 4. The effects of three different concentrations of six plant oils on the mycelial growth of *A. bisporus*.

Days	Cont	Pine			Parsley			Apricot			Plum			Watermelon			Melon		
		10	30	50	10	30	50	10	30	50	10	30	50	10	30	50	10	30	50
1	1.17	0.25	0.00	0.00	0.00	0.00	0.00	1.83	3.17	1.67	1.83	3.00	2.00	1.00	0.50	0.50	2.67	1.33	1.50
2	3.50	1.33	0.00	0.00	0.00	0.00	0.00	4.00	6.00	2.33	3.67	5.00	5.00	2.33	1.33	1.83	4.50	3.33	3.08
3	5.67	1.33	0.00	0.00	0.00	0.00	0.00	6.67	8.33	4.67	5.00	6.67	5.50	4.67	4.33	5.00	5.83	4.83	4.92
4	7.67	1.67	0.00	0.00	0.00	0.00	0.00	8.50	10.33	6.33	6.00	8.83	7.33	6.33	6.00	6.17	7.33	6.00	5.92
5	10.83	2.25	0.00	0.00	0.00	0.00	0.00	10.67	12.33	7.50	7.00	10.50	8.83	7.67	8.33	7.67	9.17	7.17	6.92
6	13.00	3.08	0.00	0.00	0.00	0.00	0.00	12.67	14.67	9.00	8.00	12.00	10.17	9.00	12.00	9.33	11.00	8.33	8.17
7	15.33	3.08	0.00	0.00	0.00	0.00	0.00	16.00	17.00	12.33	12.00	13.33	12.67	10.83	15.33	11.67	13.83	11.67	9.83
8	18.67	3.67	0.00	0.00	0.00	0.00	0.00	18.33	20.33	13.67	13.33	16.33	15.00	11.33	16.67	13.67	16.33	17.67	12.17
9	20.50	4.58	0.00	0.00	0.00	0.00	0.00	20.50	23.67	15.67	17.33	18.67	18.33	12.50	19.67	14.67	18.17	22.00	13.67
10	22.67	5.58	0.00	0.00	0.00	0.00	0.00	23.00	27.67	17.67	20.33	24.33	21.67	13.83	23.67	15.67	20.00	25.67	15.08
11	27.33	6.50	0.00	0.00	0.00	0.00	0.00	28.33	31.67	20.00	24.33	28.00	25.00	16.33	27.00	18.00	21.33	28.33	16.67
12	32.00	7.50	0.00	0.00	0.00	0.00	0.00	32.00	35.67	22.33	28.00	31.00	28.00	18.33	30.00	20.67	23.17	30.83	18.42
13	36.00	7.75	0.00	0.00	0.00	0.00	0.00	36.17	39.33	25.33	31.33	34.33	30.83	20.67	33.33	24.00	25.00	33.33	20.33
14	39.00	7.75	0.00	0.00	0.00	0.00	0.00	40.00	40.33	26.00	36.00	39.00	34.33	21.67	36.67	25.67	26.33	35.33	21.00
15	42.33	8.33	0.00	0.00	0.00	0.00	0.00	43.67	44.33	28.33	39.00	43.00	39.00	24.00	39.33	29.00	28.67	39.83	23.58
16	45.00	9.00	0.00	0.00	0.00	0.00	0.00	47.33	47.67	31.33	42.67	48.00	43.67	26.67	42.33	32.67	30.67	44.00	25.67
17	47.67	9.33	0.00	0.00	0.00	0.00	0.00	50.33	50.67	35.33	46.33	50.67	47.00	28.33	44.33	34.33	32.33	46.67	27.17
18	51.00	10.00	0.00	0.00	0.00	0.00	0.00	57.00	53.67	38.67	50.33	53.67	50.67	30.33	46.00	35.67	34.17	50.33	28.42
19	55.00	10.67	0.00	0.00	0.00	0.00	0.00	60.00	55.33	42.67	52.67	56.67	54.67	31.83	48.50	37.50	35.83	53.67	29.75
20	59.00	11.33	0.00	0.00	0.00	0.00	0.00	64.00	57.67	46.33	56.33	59.33	59.00	33.67	51.67	39.33	37.33	57.33	30.83
21	61.67	12.00	0.00	0.00	0.00	0.00	0.00	68.00	62.33	50.33	60.33	63.33	63.00	35.67	54.00	40.67	40.33	63.33	32.17
22	64.67	12.67	0.00	0.00	0.00	0.00	0.00	72.00	66.33	55.33	64.67	67.33	67.33	38.00	56.00	42.00	43.17	69.00	33.50
23	68.00	13.17	0.00	0.00	0.00	0.00	0.00	76.00	71.67	59.33	69.00	71.33	71.00	40.00	58.00	43.00	46.33	75.50	35.00
24	71.00	14.00	0.00	0.00	0.00	0.00	0.00	78.00	75.33	64.00	72.67	75.33	76.00	42.00	62.00	45.00	49.33	78.67	36.67
25	74.33	14.67	0.00	0.00	0.00	0.00	0.00	80.00	76.67	70.00	76.33	78.00	78.67	44.00	65.00	47.00	51.67	79.67	38.00
26	76.67	15.50	0.00	0.00	0.00	0.00	0.00	80.00	78.33	75.33	78.33	79.33	80.00	45.67	67.67	48.33	54.17	80.00	39.25
27	78.33	16.33	0.00	0.00	0.00	0.00	0.00	80.00	79.67	78.67	79.67	80.00	80.00	47.33	71.00	46.67	56.67	80.00	40.83
28	79.67	18.33	0.00	0.00	0.00	0.00	0.00	80.00	80.00	80.00	80.00	80.00	80.00	48.67	73.33	51.33	58.33	80.00	43.67
29	80.00	20.17	0.00	0.00	0.00	0.00	0.00	80.00	80.00	80.00	80.00	80.00	80.00	49.33	75.33	52.00	59.33	80.00	44.17
Standard deviation	26.28	5.50	0.00	0.00	0.00	0.00	0.00	25.74	25.39	24.81	26.92	26.09	25.73	15.38	23.53	16.51	17.52	26.25	13.14

Table 5. Percentage inhibitions of some selected plant oils on mycelial growth of *Agaricus bisporus*.

Days	Pine			Parsley			Apricot			Plum			Watermelon			Melon		
	10	30	50	10	30	50	10	30	50	10	30	50	10	30	50	10	30	50
1	-78.63	-100.00	-100.00	-100.00	-100.00	-100.00	56.41	170.94	42.74	56.41	156.41	70.94	-14.53	-57.26	-57.26	128.21	13.68	28.21
2	-62.00	-100.00	-100.00	-100.00	-100.00	-100.00	14.29	71.43	-33.43	4.86	42.86	42.86	-33.43	-62.00	-47.71	28.57	-4.86	-12.00
3	-76.54	-100.00	-100.00	-100.00	-100.00	-100.00	17.64	46.91	-17.64	-11.82	17.64	-3.00	-17.64	-23.63	-11.82	2.82	-14.81	-13.23
4	-78.23	-100.00	-100.00	-100.00	-100.00	-100.00	10.82	34.68	-17.47	-21.77	15.12	-4.43	-17.47	-21.77	-19.56	-4.43	-21.77	-22.82
5	-79.22	-100.00	-100.00	-100.00	-100.00	-100.00	-1.48	13.85	-30.75	-35.36	-3.05	-18.47	-29.18	-23.08	-29.18	-15.33	-33.80	-36.10
6	-76.31	-100.00	-100.00	-100.00	-100.00	-100.00	-2.54	12.85	-30.77	-38.46	-7.69	-21.77	-30.77	-7.69	-28.23	-15.38	-35.92	-37.15
7	-79.91	-100.00	-100.00	-100.00	-100.00	-100.00	4.37	10.89	-19.57	-21.72	-13.05	-17.35	-29.35	0.00	-23.87	-9.78	-23.87	-35.88
8	-80.34	-100.00	-100.00	-100.00	-100.00	-100.00	-1.82	8.89	-26.78	-28.60	-12.53	-19.66	-39.31	-10.71	-26.78	-12.53	-5.36	-34.82
9	-77.66	-100.00	-100.00	-100.00	-100.00	-100.00	0.00	15.46	-23.56	-15.46	-8.93	-10.59	-39.02	-4.05	-28.44	-11.37	7.32	-33.32
10	-75.39	-100.00	-100.00	-100.00	-100.00	-100.00	**1.46	22.06	-22.06	-10.32	7.32	-4.41	-38.99	4.41	-30.88	-11.78	13.23	-33.48
11	-76.22	-100.00	-100.00	-100.00	-100.00	-100.00	3.66	15.88	-26.82	-10.98	2.45	-8.53	-40.25	-1.21	-34.14	-21.95	3.66	-39.00
12	-76.56	-100.00	-100.00	-100.00	-100.00	-100.00	0.00	11.47	-30.22	-12.50	-3.13	-12.50	-42.72	-6.25	-35.41	-27.59	-3.66	-42.44
13	-78.47	-100.00	-100.00	-100.00	-100.00	-100.00	0.47	9.25	-29.64	-12.97	-4.64	-14.36	-42.58	-7.42	-33.33	-30.56	-7.42	-43.53
14	-80.13	-100.00	-100.00	-100.00	-100.00	-100.00	2.56	3.41	-33.33	-7.69	0.00	-11.97	-44.44	-5.97	-34.18	-32.49	-9.41	-46.15
15	-80.32	-100.00	-100.00	-100.00	-100.00	-100.00	3.17	4.72	-33.07	-7.87	1.58	-7.87	-43.30	-7.09	-31.49	-32.27	-5.91	-44.29
16	-80.00	-100.00	-100.00	-100.00	-100.00	-100.00	5.18	5.93	-30.38	-5.18	6.67	-2.96	-40.73	-5.93	-27.40	-31.84	-2.22	-42.96
17	-80.43	-100.00	-100.00	-100.00	-100.00	-100.00	5.58	6.29	-25.89	-2.81	6.29	-1.41	-40.57	-7.01	-27.98	-32.18	-2.10	-43.00
18	-80.39	-100.00	-100.00	-100.00	-100.00	-100.00	11.76	5.24	-24.18	-1.31	5.24	-0.65	-40.53	-9.80	-30.06	-33.00	-1.31	-44.27
19	-80.60	-100.00	-100.00	-100.00	-100.00	-100.00	9.09	0.60	-22.42	-4.24	3.04	-0.60	-42.13	-11.82	-31.82	-34.85	-2.42	-45.91
20	-80.80	-100.00	-100.00	-100.00	-100.00	-100.00	8.47	-2.25	-21.47	-4.53	0.56	0.00	-42.93	-12.42	-33.34	-36.73	-2.83	-47.75
21	-80.54	-100.00	-100.00	-100.00	-100.00	-100.00	10.26	1.07	-18.39	-2.17	2.69	2.16	-42.16	-12.44	-34.05	-34.60	2.69	-47.84
22	-80.41	-100.00	-100.00	-100.00	-100.00	-100.00	11.33	2.57	-14.44	0.00	4.11	4.11	-41.24	-13.41	-35.05	-33.25	6.70	-48.20
23	-80.63	-100.00	-100.00	-100.00	-100.00	-100.00	11.76	5.40	-12.75	1.47	4.90	4.41	-41.18	-14.71	-36.76	-31.87	11.03	-48.53
24	-80.28	-100.00	-100.00	-100.00	-100.00	-100.00	9.86	6.10	-9.86	2.35	6.10	7.04	-40.85	-12.68	-36.62	-30.52	10.80	-48.35
25	-80.26	-100.00	-100.00	-100.00	-100.00	-100.00	7.63	3.15	-5.83	2.69	4.94	5.84	-40.80	-12.55	-36.77	-30.49	7.18	-48.88
26	-79.78	-100.00	-100.00	-100.00	-100.00	-100.00		2.17	-1.75	2.17	3.47	4.34	-40.43	-11.74	-36.96	-29.35	4.34	-48.81
27	-79.15	-100.00	-100.00	-100.00	-100.00	-100.00		1.71	0.43	1.71	2.13		-39.58	-9.36	-40.42	-27.65		-47.87
28	-76.99	-100.00	-100.00	-100.00	-100.00	-100.00		0.41	0.41	0.41			-38.91	-7.96	-35.57	-26.79		-45.19
29	-74.79	-100.00	-100.00	-100.00	-100.00	-100.00							-38.34	-5.84	-35.00	-25.84		-44.79
Standard deviation	3.62	0.00	0.00	0.00	0.00	0.00	11.47	33.94	15.73	16.83	31.33	19.30	8.09	14.31	8.07	31.43	13.26	16.05

* = Inhibition
 ** = Stimulation

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