

# Synergic Effect of Phthalide Lactones and Fluconazole and Its New Analogues as a Factor Limiting the Use of Azole Drugs against Candidiasis

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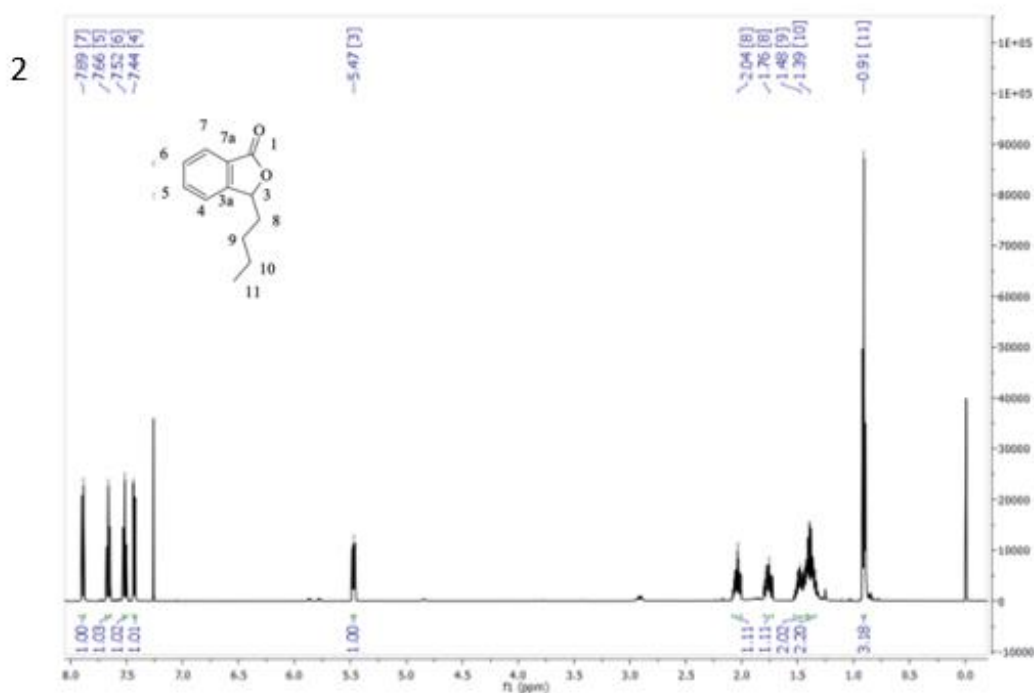


Figure S-1a. <sup>1</sup>H NMR of 3-*n*-butylphthalide (2).

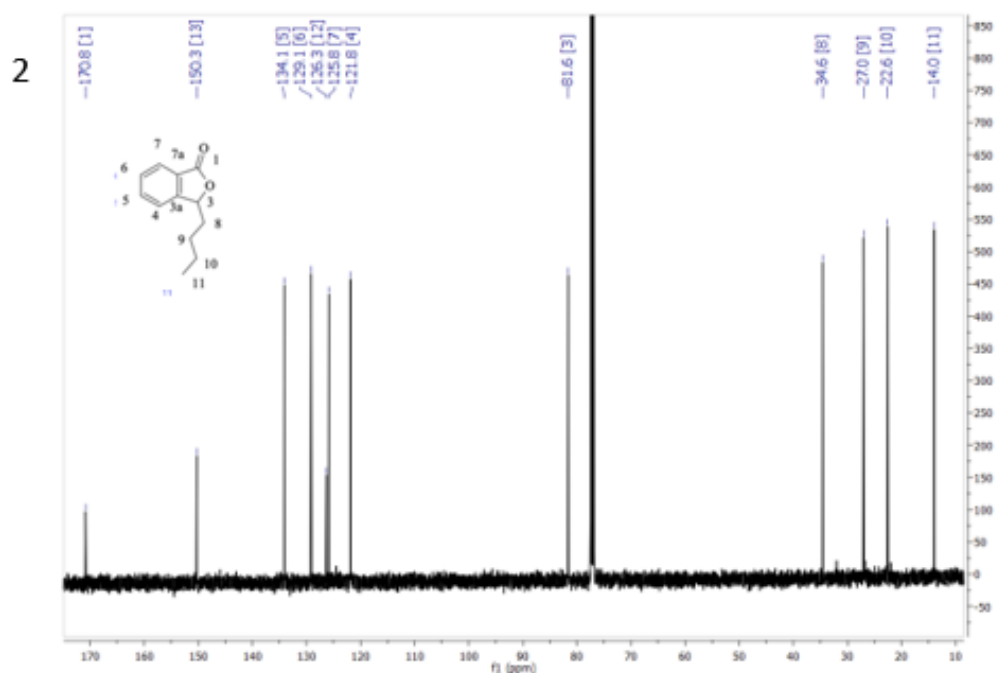


Figure S-1b. <sup>13</sup>C NMR of 3-*n*-butylphthalide (2).

3

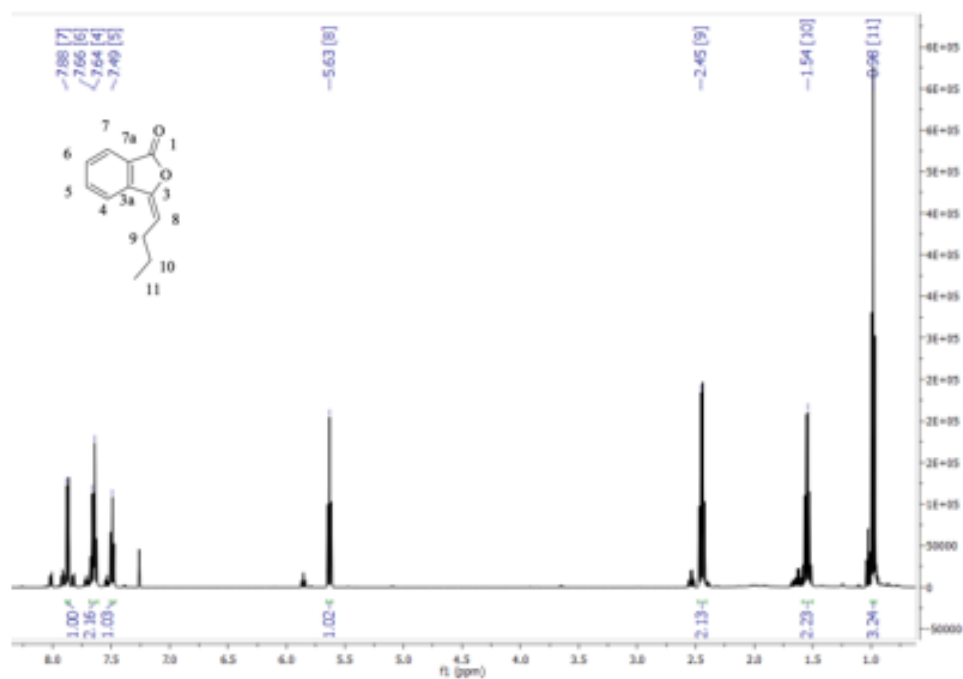


Figure S-2a. <sup>1</sup>H NMR of 3-butyldenephthalide (3).

3

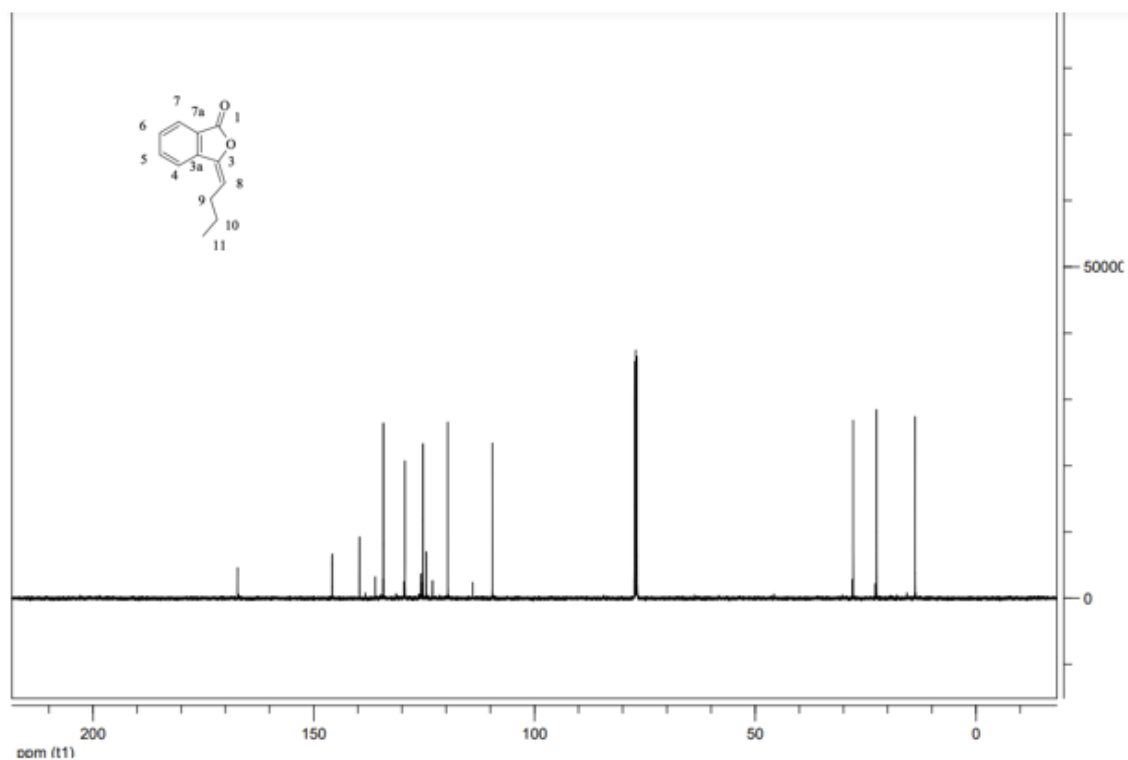
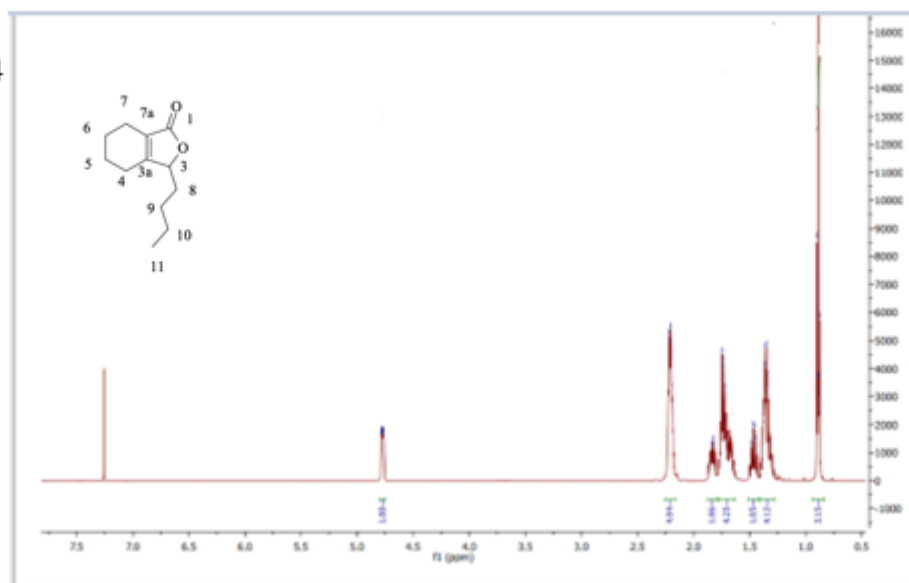


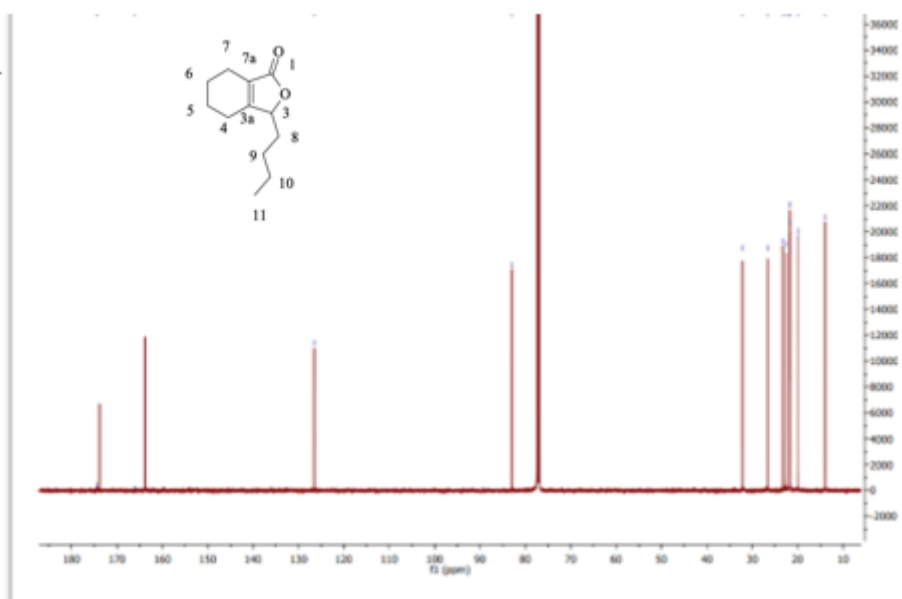
Figure S-2b. <sup>13</sup>C NMR of 3-butyldenephthalide (3).

4



**Figure S-3a.**  $^1\text{H}$  NMR of 3-butyl-4,5,6,7-tetrahydrophthalide (4).

4



**Figure S-3b.**  $^{13}\text{C}$  NMR of 3-Butyl-4,5,6,7-tetrahydrophthalide (4)

5

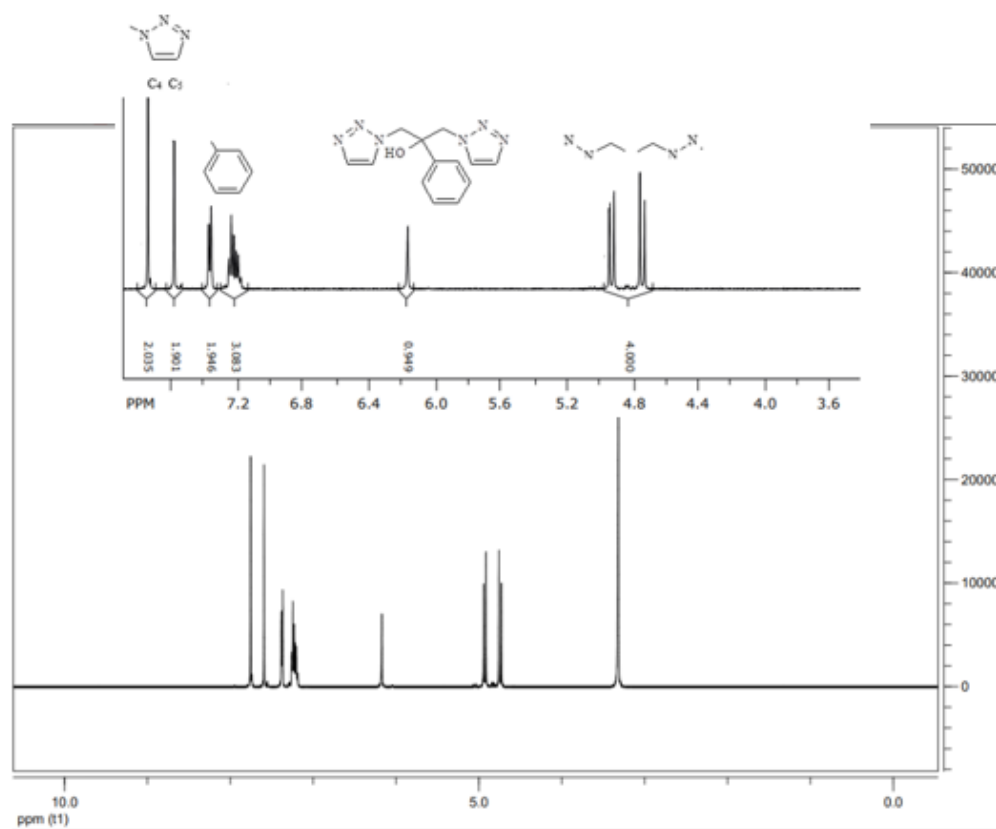


Figure S-4a. <sup>1</sup>H NMR of 1,3-bis(1H-1,2,3-triazol-1-yl)-2-phenylpropan-2-ol (5).

5

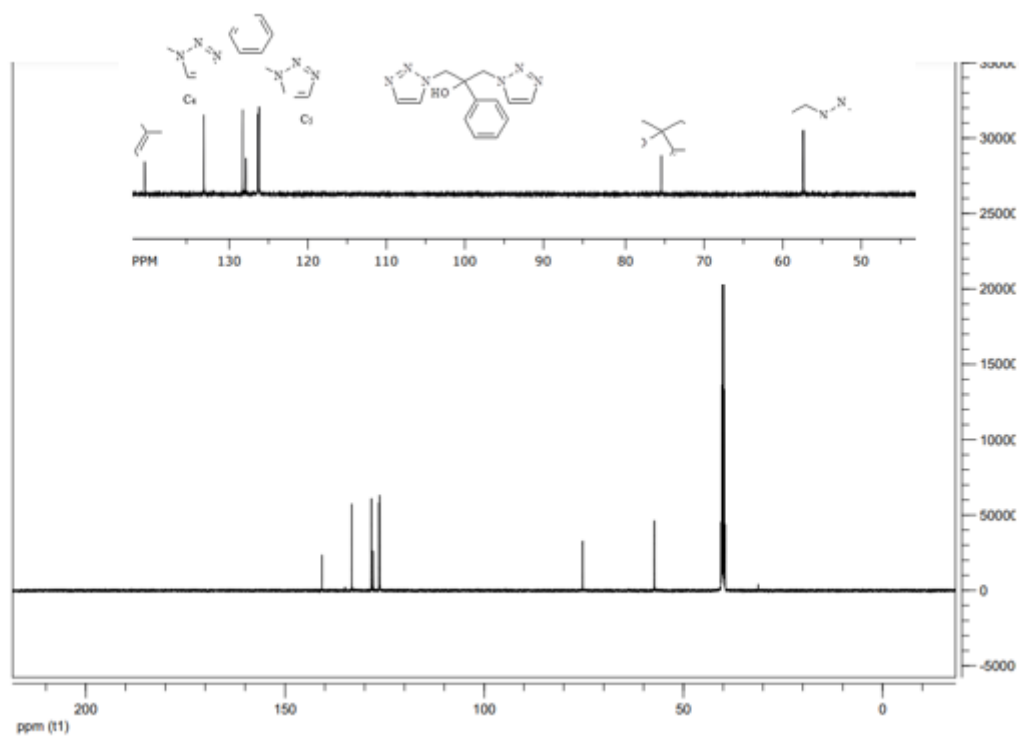


Figure S-4b. <sup>13</sup>C NMR of 1,3-bis(1H-1,2,3-triazol-1-yl)-2-phenylpropan-2-ol (5).

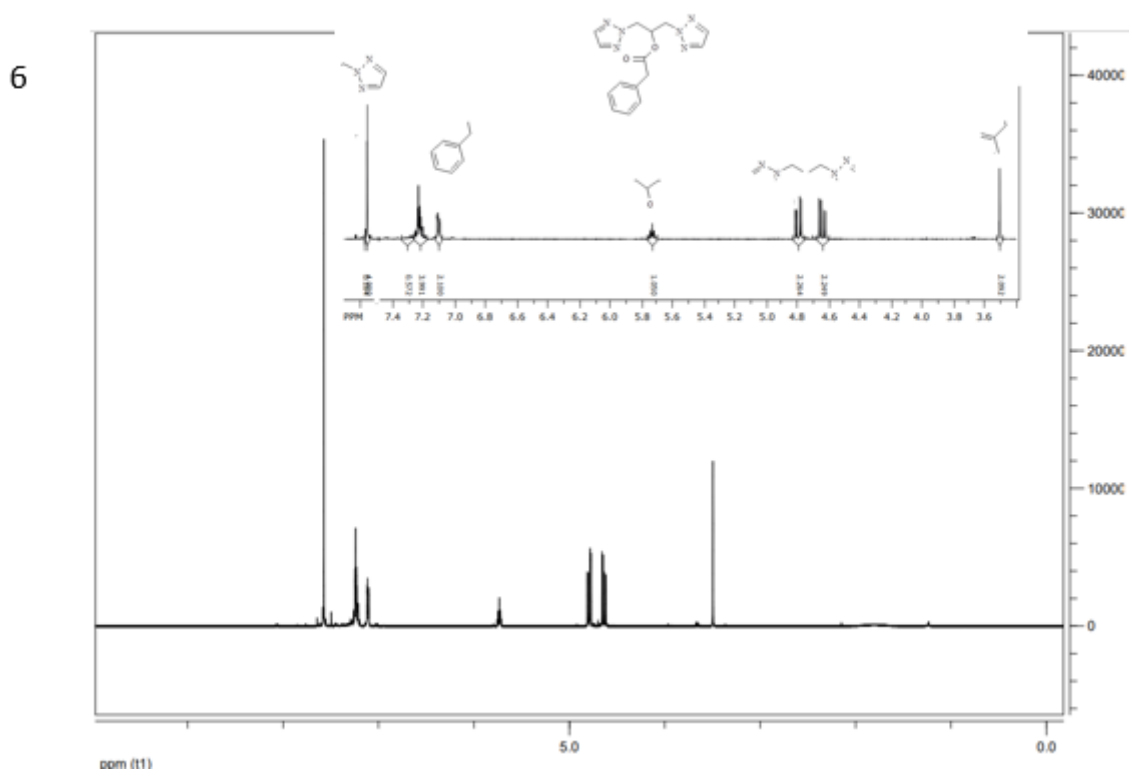


Figure S-5a.  $^1\text{H}$  NMR of 1,3-di(1,2,3-triazol-2-yl)propan-2-yl phenylacetate (6).

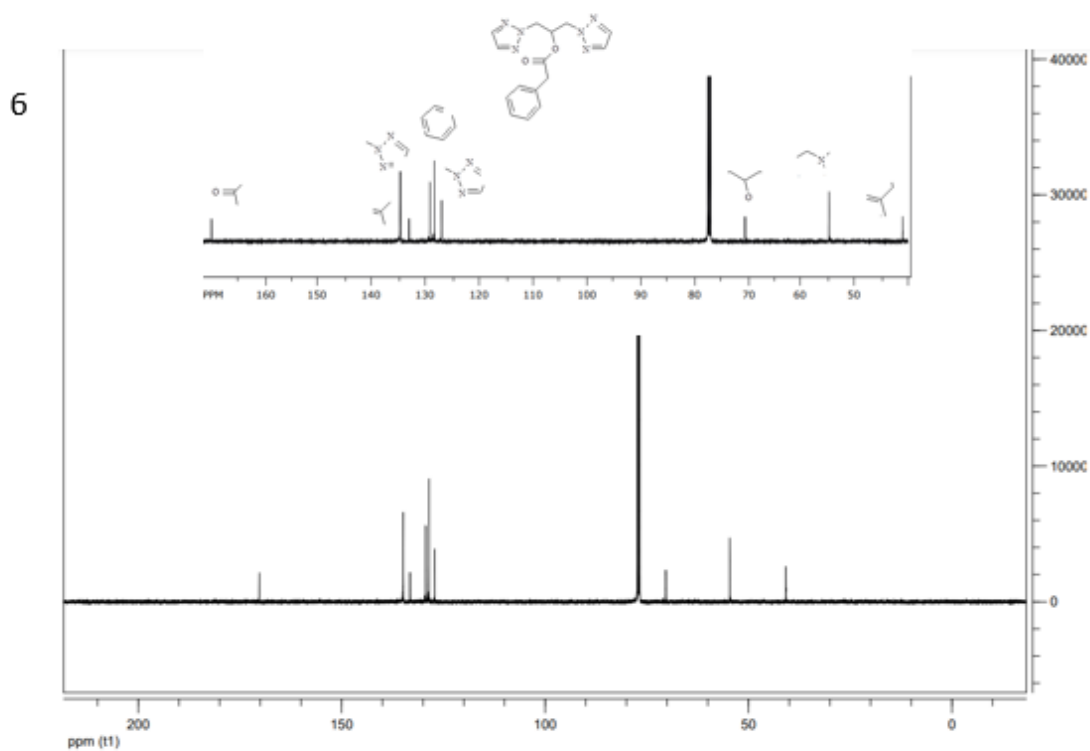


Figure S-5b.  $^{13}\text{C}$  NMR of 1,3-di(1,2,3-triazol-2-yl)propan-2-yl phenylacetate (6).

1



8

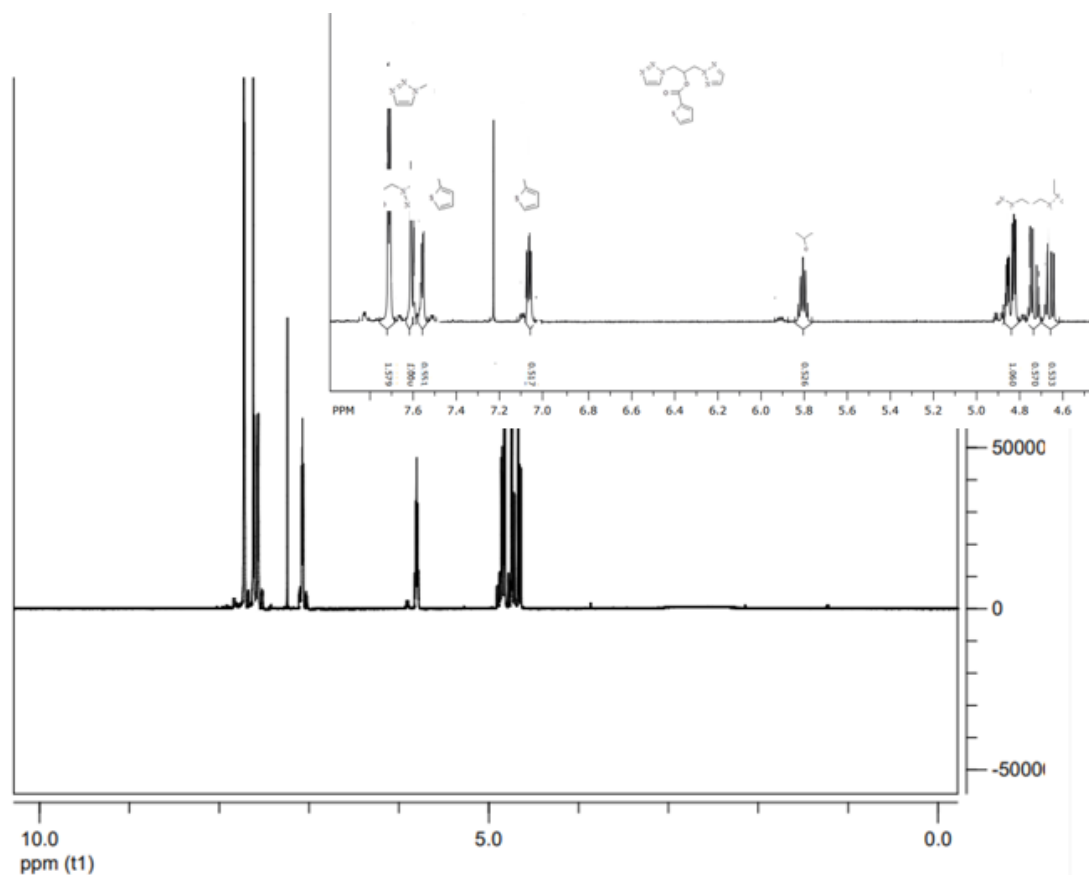


Figure S-7a. <sup>1</sup>H NMR of 1-(1,2,3-triazol-1-yl)-3-(1,2,3-triazol-2-yl)propan-2-yl 2-phenylacetate (8).

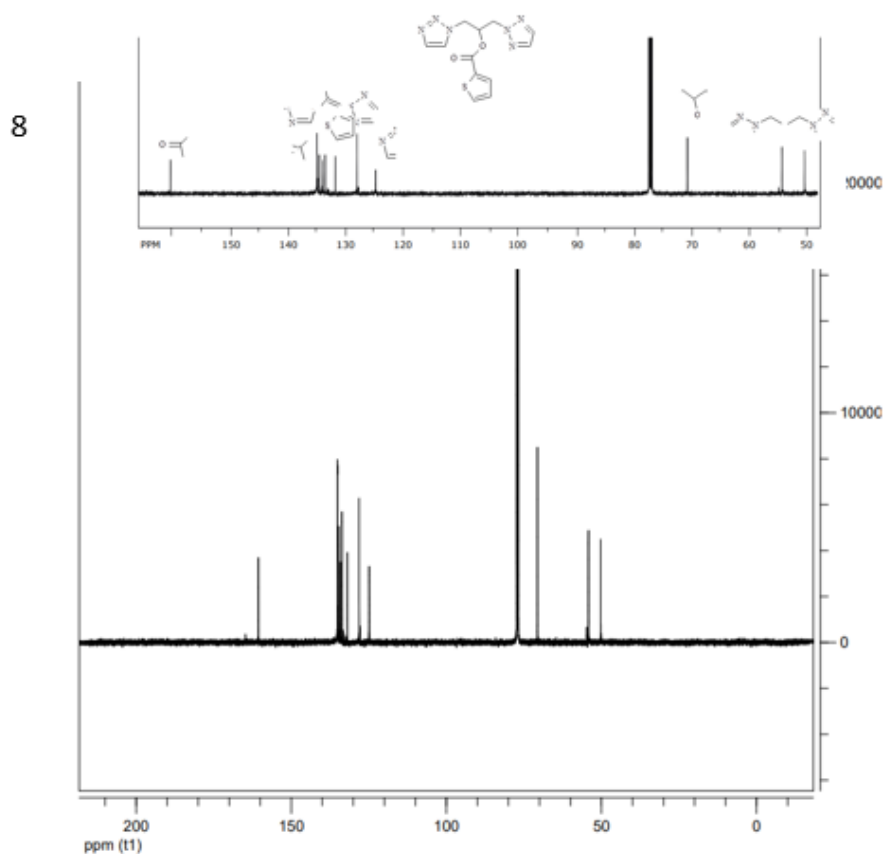
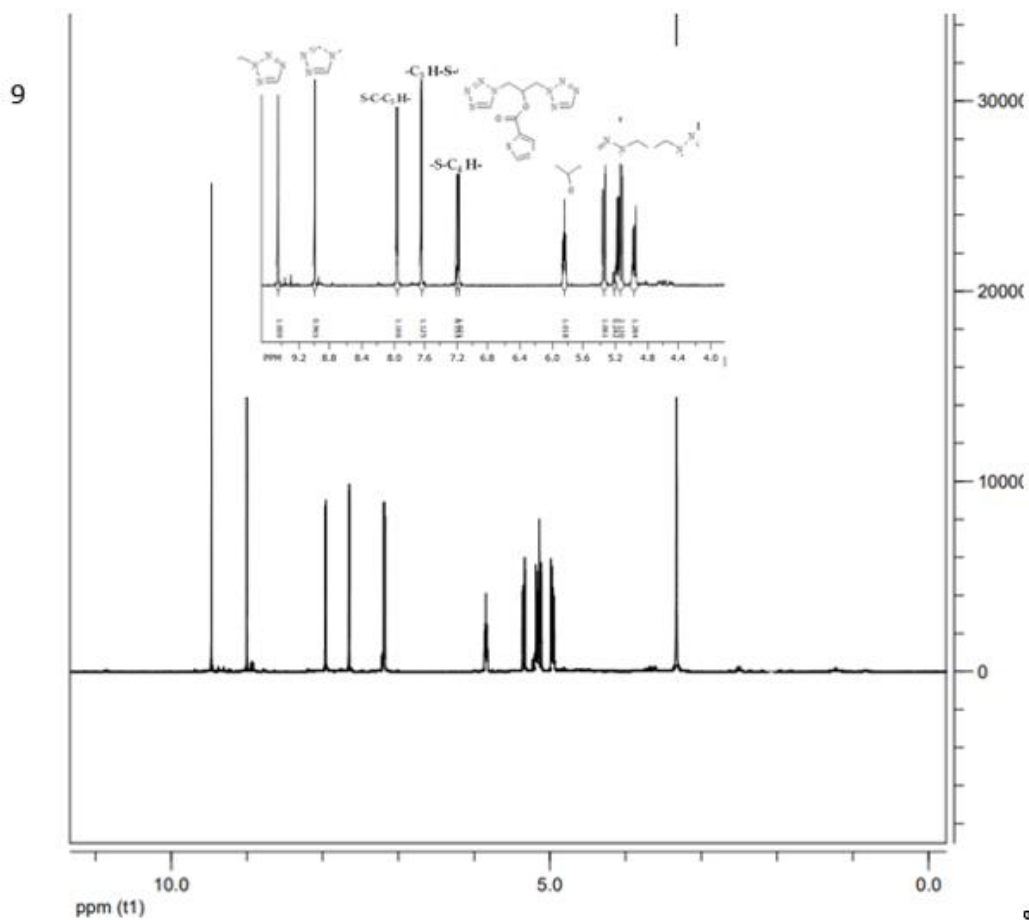


Figure S-7b. <sup>13</sup>C NMR of 1-(1,2,3-triazol-1-yl)-3-(1,2,3-triazol-2-yl)propan-2-yl 2-phenylacetate (8).



Figure

S-8a.  $^1\text{H}$  NMR of 1-(tetrazol-1-yl)-3-(tetrazol-2-yl)propan-2-yl 2-tiophenylacetate (9).

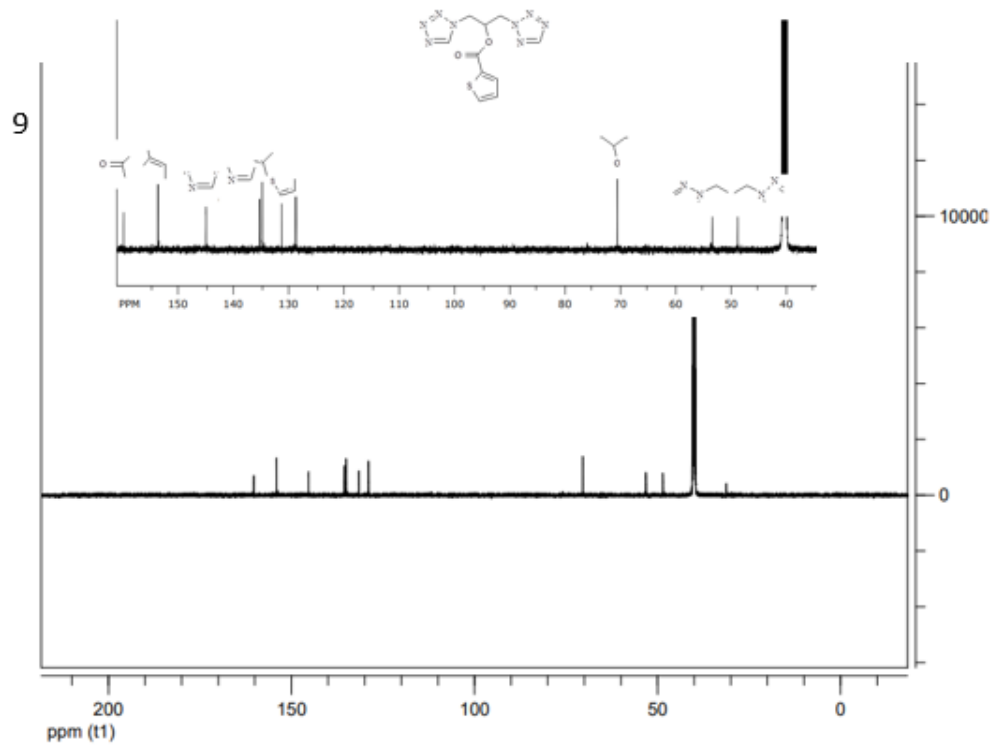


Figure S-8b.  $^{13}\text{C}$  NMR of 1-(tetrazol-1-yl)-3-(tetrazol-2-yl)propan-2-yl 2-tiophenylacetate (9).



10

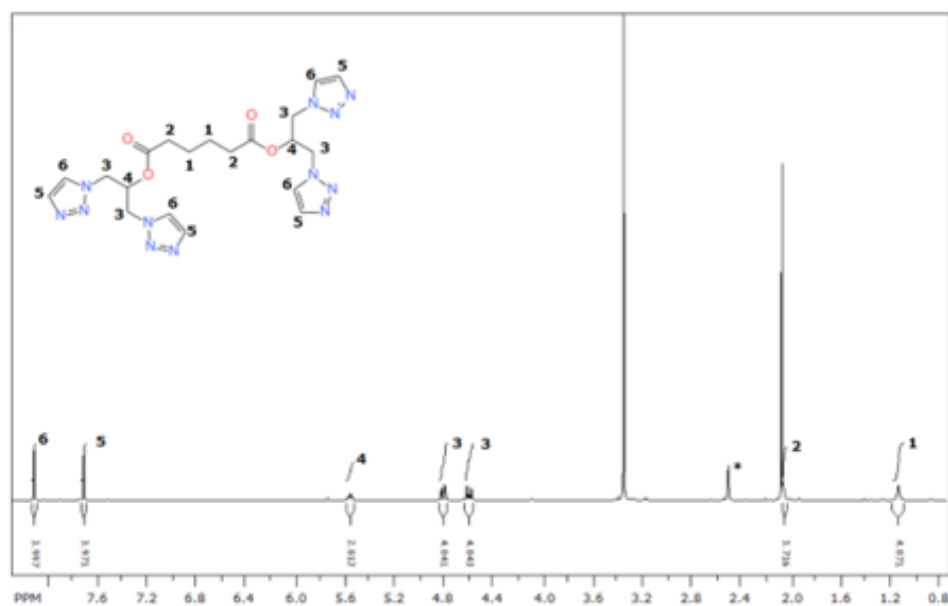


Figure S-9a. <sup>1</sup>H NMR of 1,6-bis[1-(1,2,3-triazol-1-yl)-3-(1,2,3-triazol-1-yl)propan-2-yl] adipate (10).

10

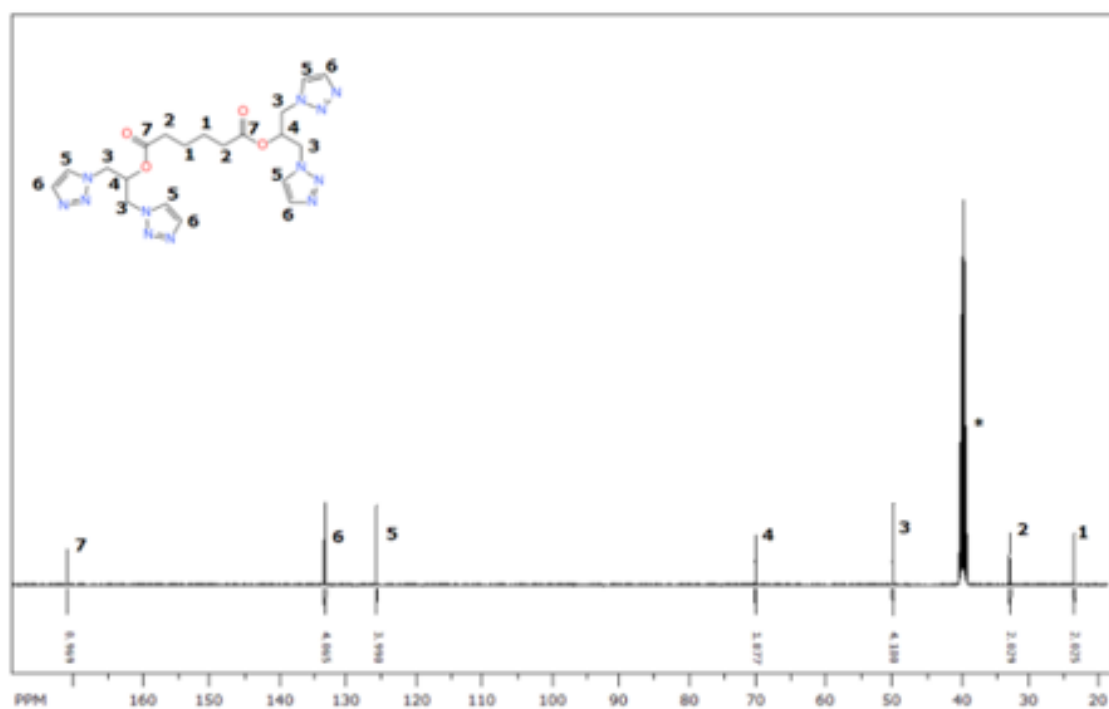


Figure S-9b. <sup>13</sup>C NMR of 1,6-bis[1-(1,2,3-triazol-1-yl)-3-(1,2,3-triazol-1-yl)propan-2-yl] adipate (10).

11

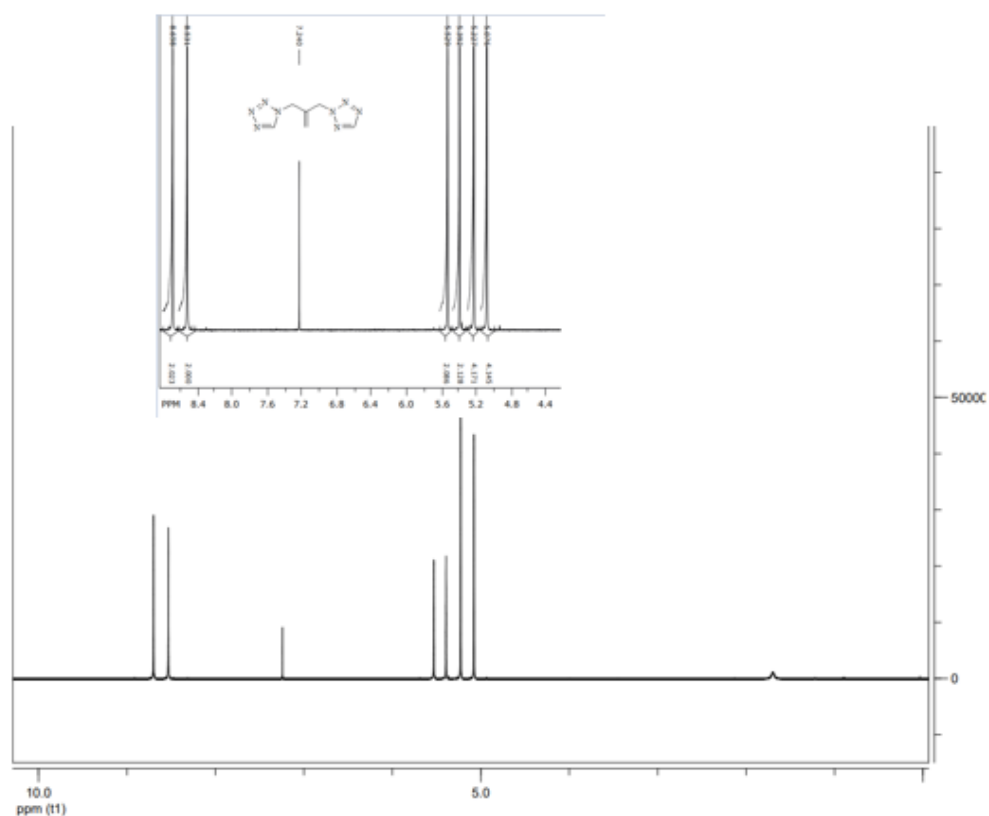


Figure S-10a.  $^1\text{H}$  NMR of 1-[2-(2H-tetrazol-2-ylmethyl)prop-2-en-1-yl]-1H-tetrazole (11).

11

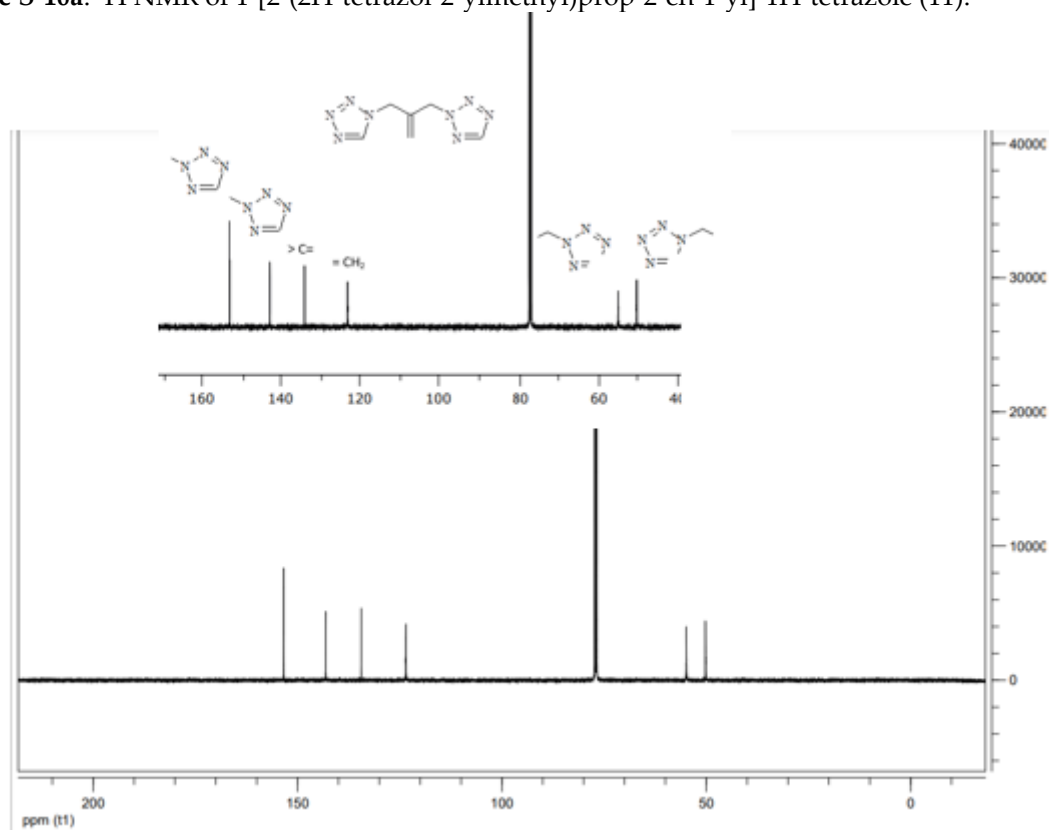


Figure S-10b.  $^{13}\text{C}$  NMR of 1-[2-(2H-tetrazol-2-ylmethyl)prop-2-en-1-yl]-1H-tetrazole (11).

12

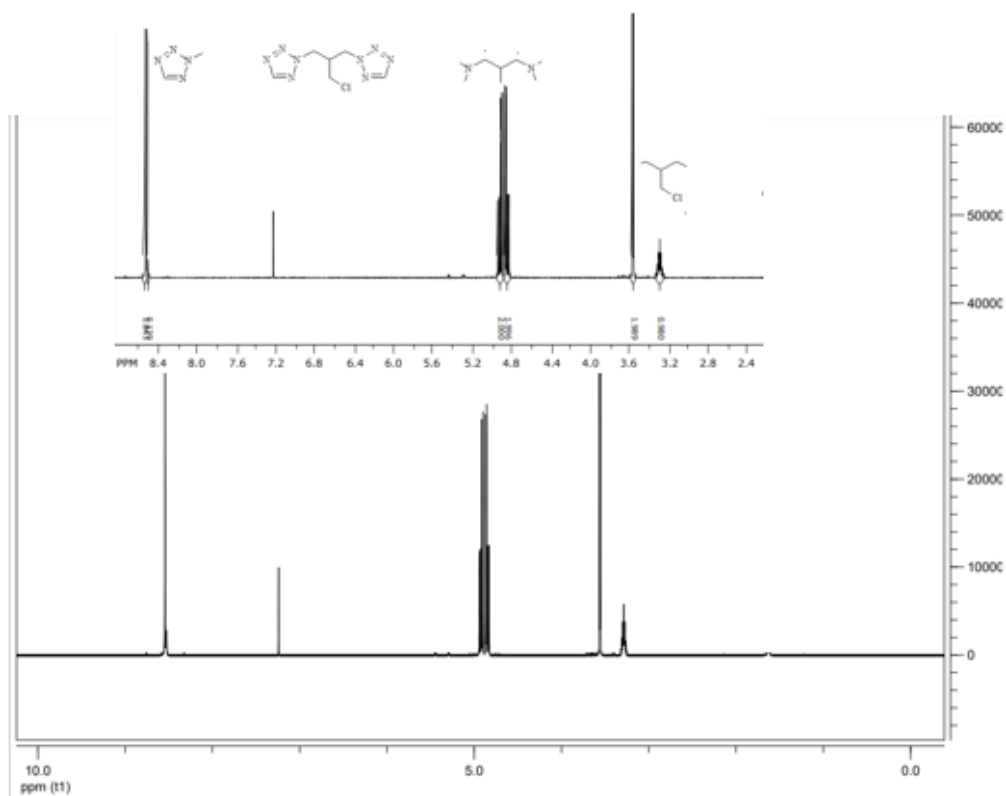


Figure S-11a.  $^1\text{H}$  NMR of 1,3-di(tetrazol-2-yl)-2-chloromethylpropane (12).

12

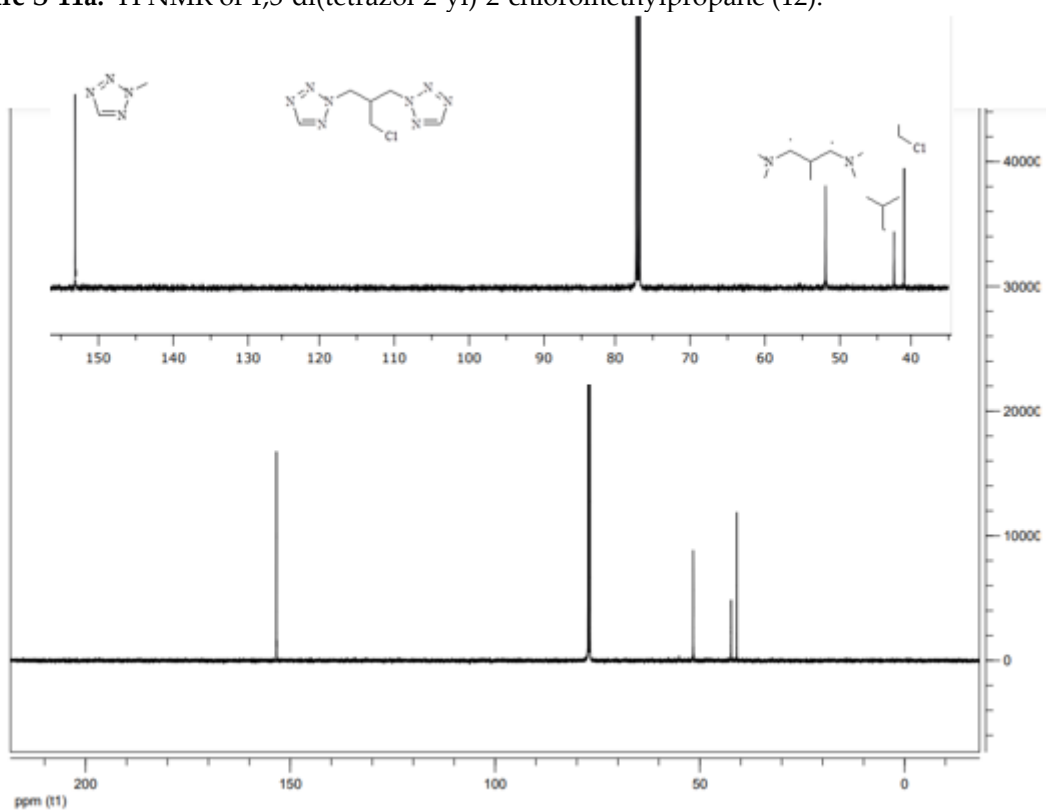


Figure S-11b.  $^{13}\text{C}$  NMR of 1,3-di(tetrazol-2-yl)-2-chloromethylpropane (12).

13

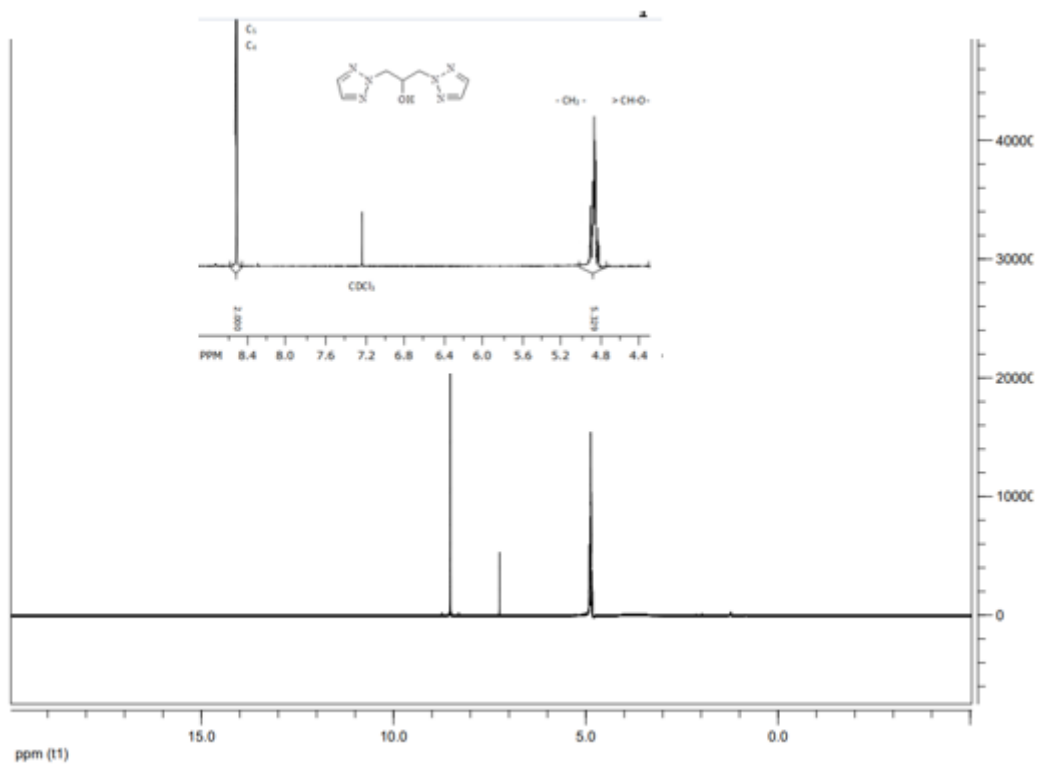


Figure S-12a. <sup>1</sup>H NMR of 1,3-di(1,2,3-triazol-2-yl)propan-2-ol (13).

13

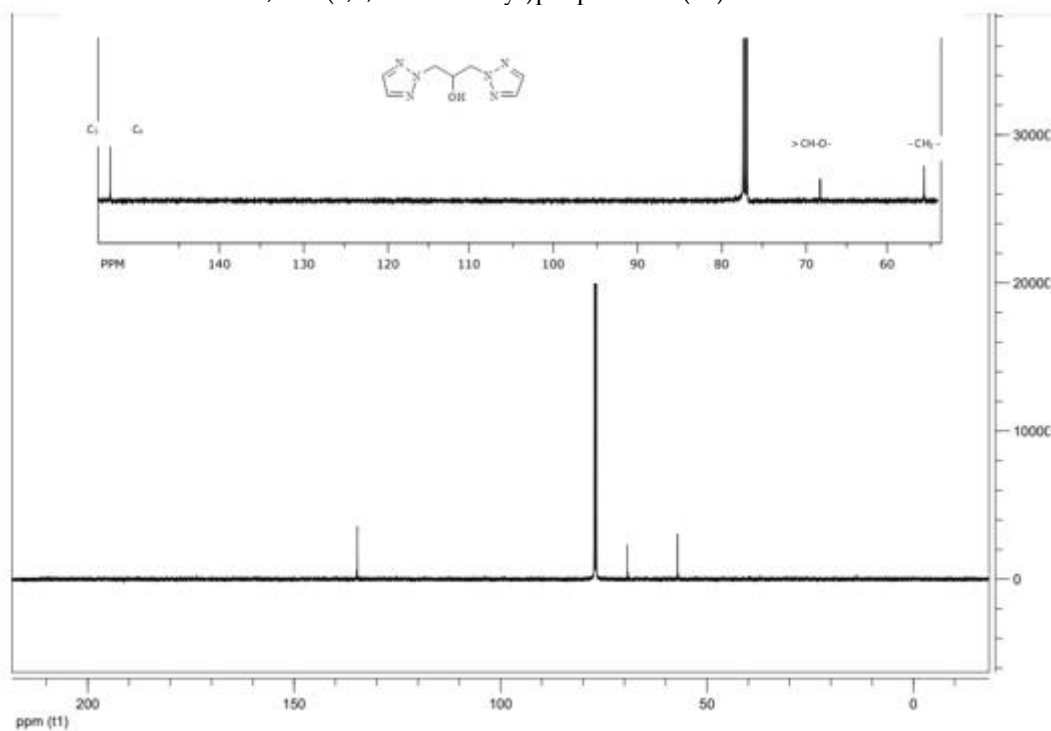


Figure S-12b. <sup>13</sup>C NMR of 1,3-di(1,2,3-triazol-2-yl)propan-2-ol (13).

14

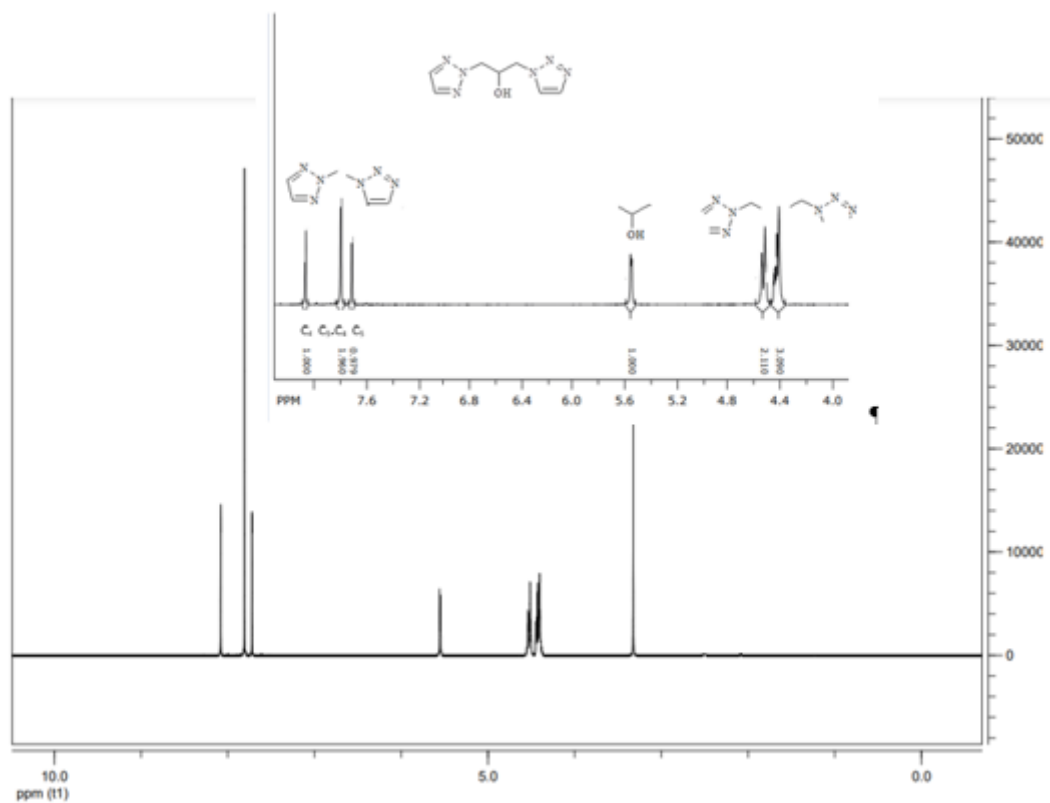


Figure S-13a.  $^1\text{H}$  NMR of 1-(1,2,3 triazol-1-yl)-3-(1,2,3-triazol-2-yl)propan-2-ol (14).

14

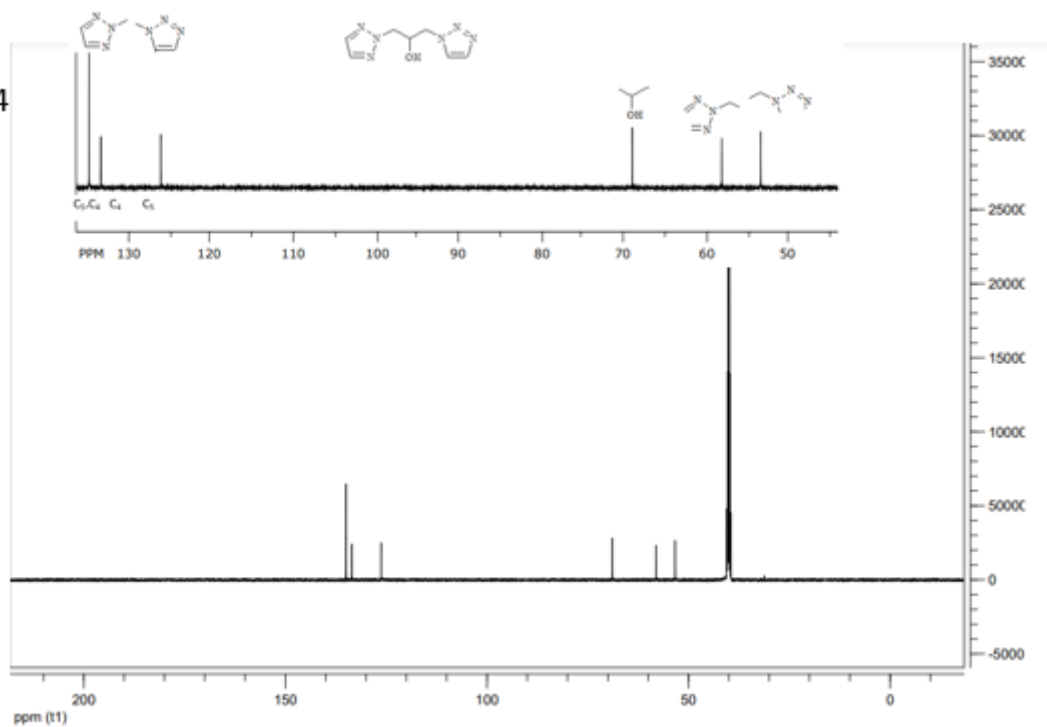
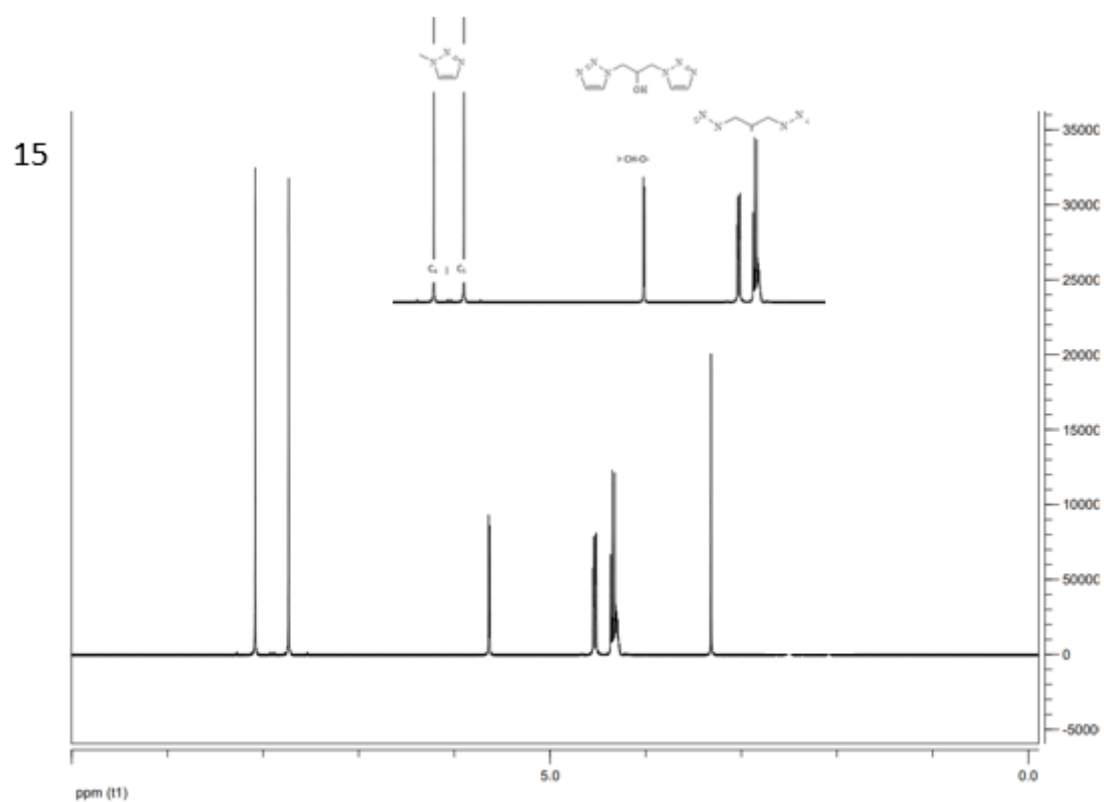
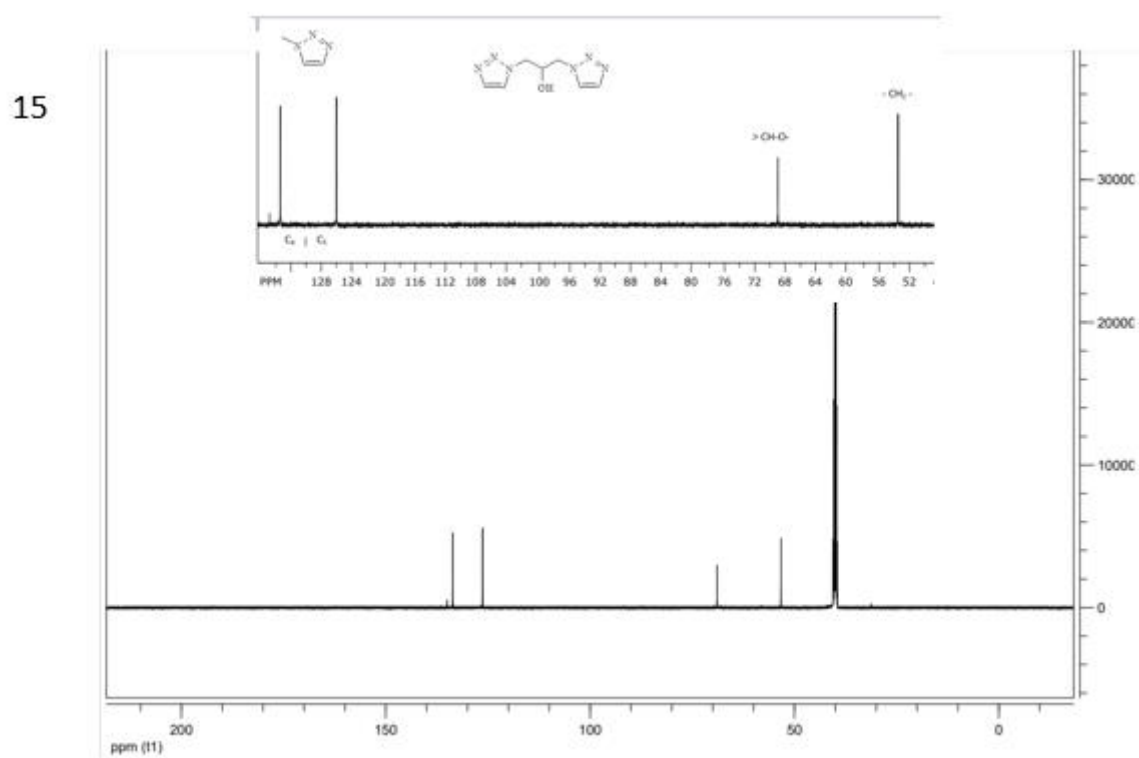


Figure S-13b.  $^{13}\text{C}$  NMR of 1-(1,2,3 triazol-1-yl)-3-(1,2,3-triazol-2-yl)propan-2-ol (14).

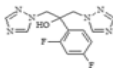



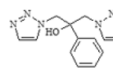
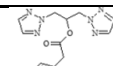
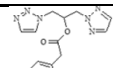
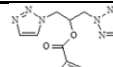
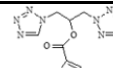
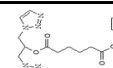
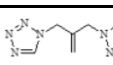
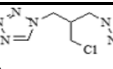
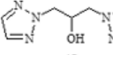
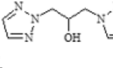
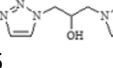


**Figure S-14a.**  $^1\text{H}$  NMR of 1,3-di(1,2,3-triazol-1-yl)propan-2-ol (15).



**Figure S-14b.**  $^{13}\text{C}$  NMR of 1,3-di(1,2,3-triazol-1-yl)propan-2-ol (15).

**Table S-1.** Experimentally determined fungistatic activity of fluconazole and lactones **2-4** and fluconazole analogues **5-15** derivative against *Candida albicans* ATCC 10231, *C. albicans* ATCC 2091, *C. zeylanoides* KKP 3528, and *C. guilliermondii* KKP 3390. MIC values are in µg/mL

		Microorganism				
		ATCC 10231	ATCC 2091	AM 38/20	KKP 3390	KKP 3528
 <b>1</b>	MIC <sub>50</sub>	1.56	2.19	7.22	8.41	6.25
	MIC <sub>90</sub>	6.25	5.57	≥18.0	≥18.0	18.75
 <b>2</b>	MIC <sub>50</sub>	45.55	30.79	78.52	55.0	46.29
	MIC <sub>90</sub>	137.72	92.34	150.00	80.00	144.64
 <b>3</b>	MIC <sub>50</sub>	38.90	17.61	85.04	36.96	62.24
	MIC <sub>90</sub>	150.12	110.0	157.76	58.95	151.13
 <b>4</b>	MIC <sub>50</sub>	62.72	20.10	49.49	46.15	58.56
	MIC <sub>90</sub>	150.00	120.10	142.51	80.34	131.29
 <b>5</b>	MIC <sub>50</sub>	179.69	70.42	163.98	146.28	154.47
	MIC <sub>90</sub>	300.0	280.95	330.65	258.96	278.42
 <b>6</b>	MIC <sub>50</sub>	225.0	69.18	118.89	95.90	174.27
	MIC <sub>90</sub>	326.16	304.47	207.78	302.15	276.84
 <b>7</b>	MIC <sub>50</sub>	194.05	82.90	203.95	146.29	164.73
	MIC <sub>90</sub>	296.0	145.40	303.95	260.57	293.34
 <b>8</b>	MIC <sub>50</sub>	201.41	127.11	225.0	144.84	119.27
	MIC <sub>90</sub>	292.32	265.04	300.0	269.84	301.09
 <b>9</b>	MIC <sub>50</sub>	197.50	122.71	226.45	148.29	180.24
	MIC <sub>90</sub>	308.61	265.57	408.27	265.94	322.59
 <b>10</b>	MIC <sub>50</sub>	133.49	107.75	166.64	68.98	38.45
	MIC <sub>90</sub>	225.44	274.42	348.45	132.48	212.36
 <b>11</b>	MIC <sub>50</sub>	249.16	173.34	260.81	118.60	-
	MIC <sub>90</sub>	378.19	345.02	414.65	185.26	300.00
 <b>12</b>	MIC <sub>50</sub>	162.67	139.41	224.32	127.48	192.37
	MIC <sub>90</sub>	329.33	277.34	321.88	203.82	297.63
 <b>13</b>	MIC <sub>50</sub>	122.29	170.59	150.0	-	-
	MIC <sub>90</sub>	219.85	295.59	304.74	56.54	167.54
 <b>14</b>	MIC <sub>50</sub>	149.78	128.43	208.80	77.89	-
	MIC <sub>90</sub>	297.93	262.21	325.42	107.52	120.45
 <b>15</b>	MIC <sub>50</sub>	138.23	154.11	140.03	45.48	75.0
	MIC <sub>90</sub>	221.56	302.26	300.0	94.26	300.0

ATCC 10231-*Candida albicans* ATCC 10231,

ATCC 2091-*C. albicans* ATCC 2091,

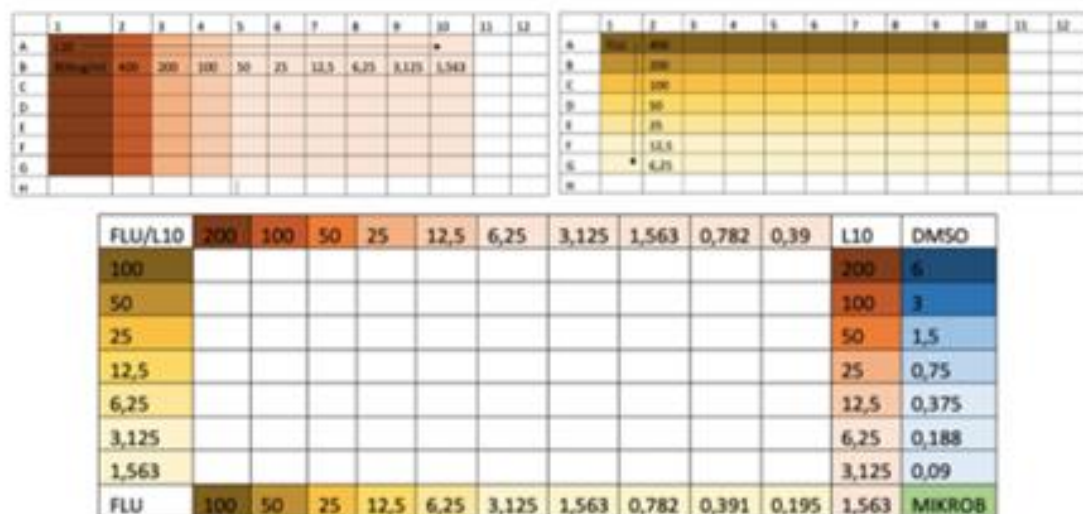
AM 38/20- *C. albicans* AM 38/20.

KKP 3528-*C. zeylanoides* KKP 3528,

KKP 3390-*C. guilliermondii* KKP 3390.

MIC<sub>50</sub> (the amount of fungistatic substance (in µg/mL) inhibiting yeast growth by 50%)

MIC<sub>90</sub> (the amount of fungistatic substance (in µg/mL) inhibiting yeast growth by 90%)



**Figure S-15a.** Graphical display of microplate used in checkboard method. Using double dilutions, a series systems consisting of phthalide lactone (L10) and chemical compound (FLU) were made. The plate designed in this way was inoculated with the microorganism under study (Private archive).

	1	2	3	4	5	6	7	8	9	10	11	12
A	0,109	0,081	0,093	0,102	0,13	0,124	0,134	0,103	0,136	0,093	0,116	0,352
B	0,094	0,096	0,124	0,258	0,295	0,317	0,376	0,402	0,412	0,468	0,158	0,538
C	0,094	0,094	0,132	0,366	0,465	0,677	0,744	0,77	0,696	0,868	0,429	0,634
D	0,101	0,104	0,24	0,441	0,551	0,704	0,751	0,572	0,69	0,766	0,538	0,614
E	0,107	0,112	0,347	0,554	0,693	0,913	0,834	0,811	0,853	1,115	0,767	0,771
F	0,094	0,234	0,234	0,604	0,59	0,835	0,718	0,92	1,036	0,92	0,685	0,61
G	0,081	0,111	0,241	0,647	0,705	0,612	0,916	0,846	0,899	0,806	0,87	0,818
H	0,102	0,23	0,422	0,703	0,769	0,696	0,556	0,825	0,797	0,7	0,72	0,537
	1	2	3	4	5	6	7	8	9	10	11	12
A	0,108	0,081	0,092	0,103	0,133	0,127	0,138	0,104	0,141	0,094	0,114	0,376
B	0,094	0,096	0,125	0,264	0,306	0,32	0,379	0,404	0,416	0,476	0,148	0,539
C	0,094	0,094	0,132	0,368	0,466	0,679	0,75	0,783	0,706	0,889	0,44	0,626
D	0,101	0,105	0,242	0,444	0,555	0,708	0,759	0,577	0,696	0,775	0,542	0,591
E	0,106	0,109	0,345	0,548	0,695	0,918	0,838	0,816	0,859	1,122	0,77	0,762
F	0,094	0,237	0,221	0,602	0,583	0,833	0,708	0,915	1,033	0,916	0,659	0,582
G	0,081	0,11	0,234	0,632	0,696	0,576	0,9	0,827	0,882	0,788	0,851	0,797
H	0,102	0,23	0,4	0,676	0,746	0,671	0,526	0,805	0,779	0,671	0,702	0,527

**Figure S-15b.** Results of crude OD values for *C. albicans* ATCC 109231 in repetition for the FIC assay between compounds 2 and 8 from Gen5 program (Private archive).