

## Supplementary Material

### Azole-Based Compounds That Are Active against Candida Biofilm: In Vitro, In Vivo and In Silico Studies

Simone Carradori <sup>1,2</sup>, Alessandra Ammazalorso <sup>1,\*</sup>, Barbara De Filippis <sup>1</sup>, Ahmet Fatih Şahin <sup>3</sup>,  
Atilla Akdemir <sup>2,3</sup>, Anastasia Orekhova <sup>4</sup>, Graziana Bonincontro <sup>5</sup> and Giovanna Simonetti <sup>5</sup>

<sup>1</sup>Department of Pharmacy, “G. d’Annunzio” University of Chieti-Pescara, Via dei Vestini 31,  
66100 Chieti, Italy

<sup>2</sup>Computer-Aided Drug Discovery Laboratory, Department of Pharmacology, Faculty of Pharmacy,  
Bezmialem Vakif University, 34093 Istanbul, Turkey

<sup>3</sup>Department of Drug Discovery and Development, Institute of Health Sciences, Bezmialem Vakif Uni-versity, 34093  
Istanbul, Turkey

<sup>4</sup>Department of Public Health and Infectious Diseases, “Sapienza” University of Rome, 00185 Rome, Italy

<sup>5</sup>Department of Environmental Biology, “Sapienza”, University of Rome, 00185 Rome, Italy

\*Correspondence: alessandra.ammazzalorso@unich.it

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**Figure S1:** <sup>1</sup>H spectrum of compound **29**

29

Sample Name:  
AA816B  
Data Collected on:  
m300-mercury300  
Archive directory:  
/home/amazza/vnmrsys/data  
Sample directory:  
AA801B\_20170328\_01  
FidFile: AA816B

Pulse Sequence: PROTON (s2pul)  
Solvent: cdcl3  
Data collected on: Jun 12 2017

Temp. 20.0 C / 293.1 K  
Operator: amazza

Relax. delay 1.000 sec  
Pulse 45.0 degrees  
Acq. time 1.706 sec  
Width 4803.1 Hz  
16 repetitions  
OBSERVE H1, 300.1976543 MHz  
DATA PROCESSING  
PT size 16384  
Total time 0 min 45 sec

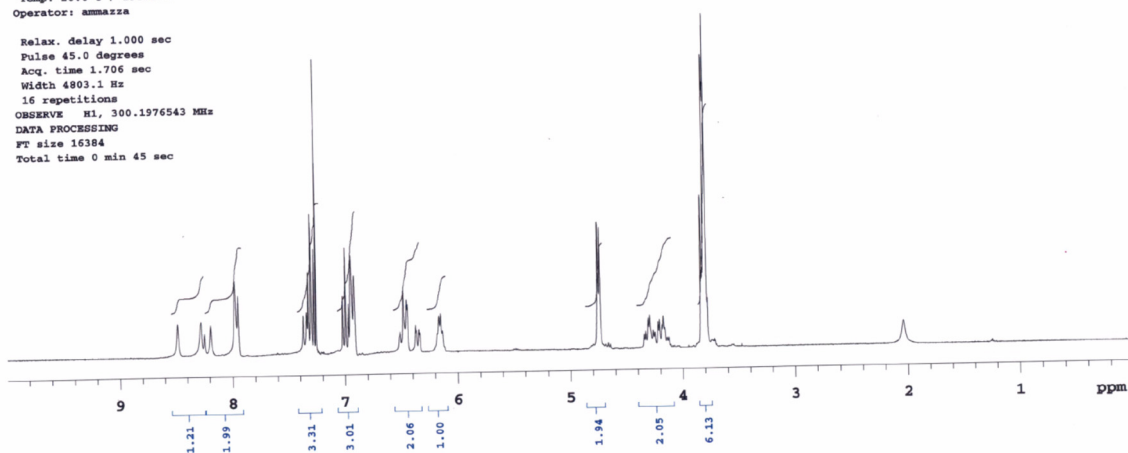
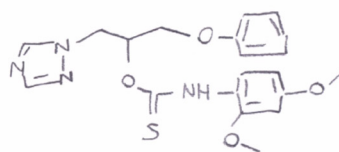


Figure S2:  $^{13}\text{C}$  spectrum of compound 29

29

Sample Name:  
AA816B  
Data Collected on:  
m300-mercury300  
Archive directory:  
/home/amazza/vnmrsys/data  
Sample directory:  
AA801B\_20170328\_01  
FidFile: AA816B\_c13

Pulse Sequence: CARBON (s2pul)  
Solvent: cdcl3  
Data collected on: Jun 12 2017

Temp. 20.0 C / 293.1 K  
Operator: amazza

Relax. delay 1.000 sec  
Pulse 45.0 degrees  
Acq. time 0.868 sec  
Width 18867.9 Hz  
832 repetitions  
OBSERVE C13, 75.4847602 MHz  
DECOUPLE H1, 300.1991980 MHz  
Power 38 dB  
continuously on  
WALTZ-16 modulated  
DATA PROCESSING  
Line broadening 0.5 Hz  
PT size 32768  
Total time 1 hr, 36 min

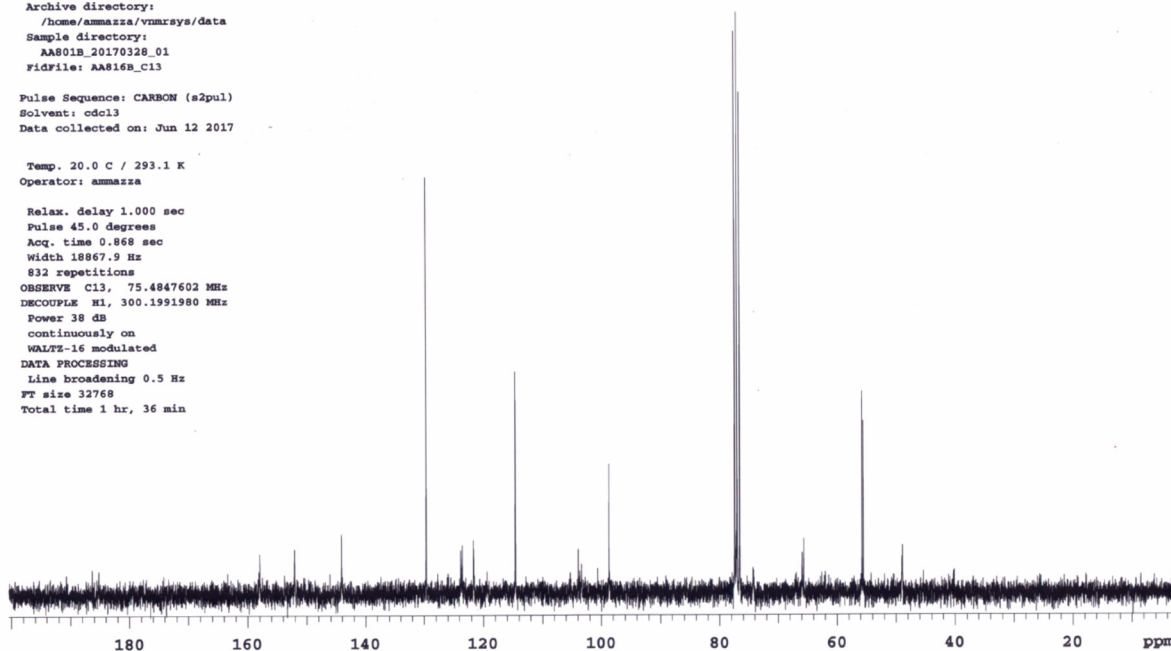


Figure S3:  $^1\text{H}$  spectrum of compound 30

Sample Name:  
AA832A  
Data Collected on:  
m300-mercury300  
Archive directory:  
/home/ammazza/vmrays/data  
Sample directory:  
AA801B\_20170328\_01  
FidFile: AA832A

Pulse Sequence: PROTON (s2pul)  
Solvent: cdcl3  
Data collected on: Sep 28 2017

Operator: ammazza

Relax. delay 1.000 sec  
Pulse 45.0 degrees  
Acq. time 1.706 sec  
Width 4803.1 Hz  
8 repetitions  
OBSERVE H1, 300.1976543 MHz  
DATA PROCESSING  
FT size 16384  
Total time 0 min 23 sec

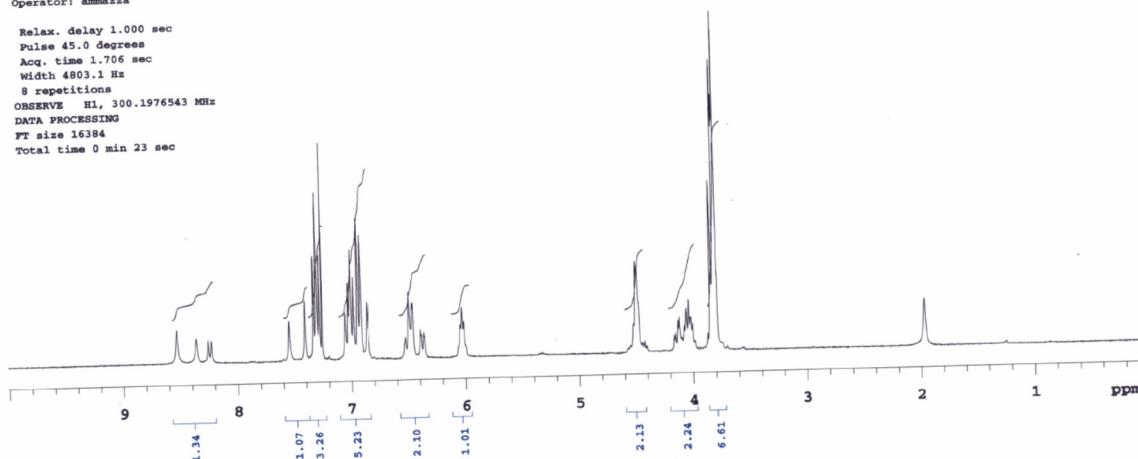
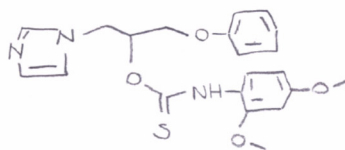


Figure S4:  $^{13}\text{C}$  spectrum of compound 30

Sample Name:  
AA832A  
Data Collected on:  
m300-mercury300  
Archive directory:  
/home/ammazza/vmrays/data  
Sample directory:  
AA801B\_20170328\_01  
FidFile: AA832A\_C13

Pulse Sequence: CARBON (s2pul)  
Solvent: cdcl3  
Data collected on: Sep 28 2017

Operator: ammazza

Relax. delay 1.000 sec  
Pulse 45.0 degrees  
Acq. time 0.868 sec  
Width 18867.9 Hz  
1024 repetitions  
OBSERVE C13, 75.4847602 MHz  
DECOUPLE H1, 300.1991980 MHz  
Power 38 dB  
continuously on  
WALTZ-16 modulated  
DATA PROCESSING  
Line broadening 0.5 Hz  
FT size 32768  
Total time 1 hr, 36 min

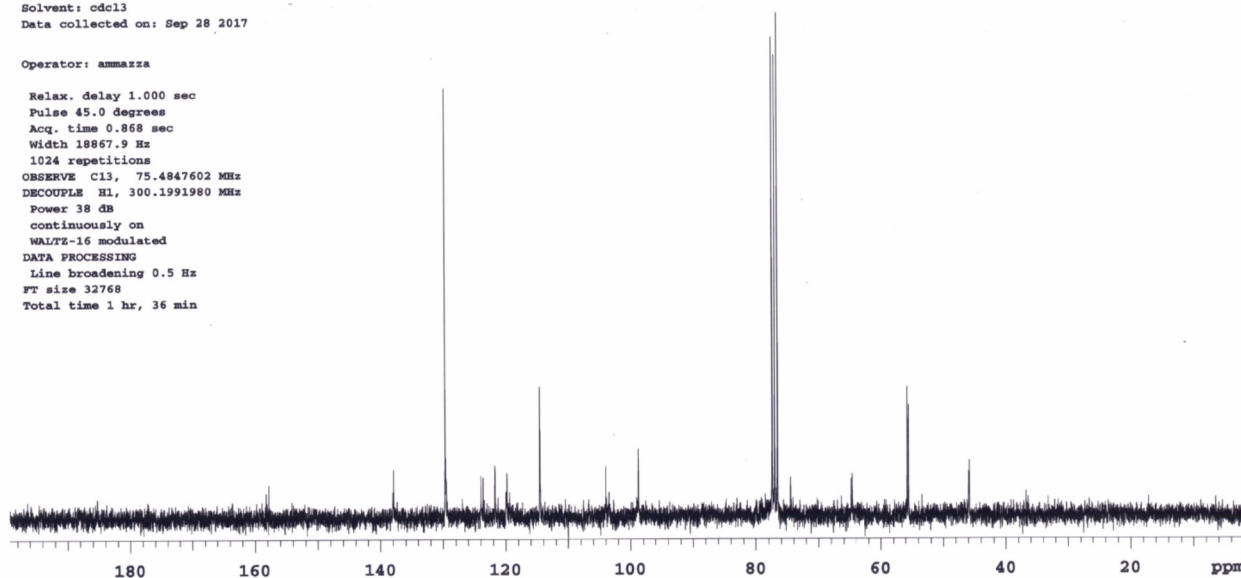
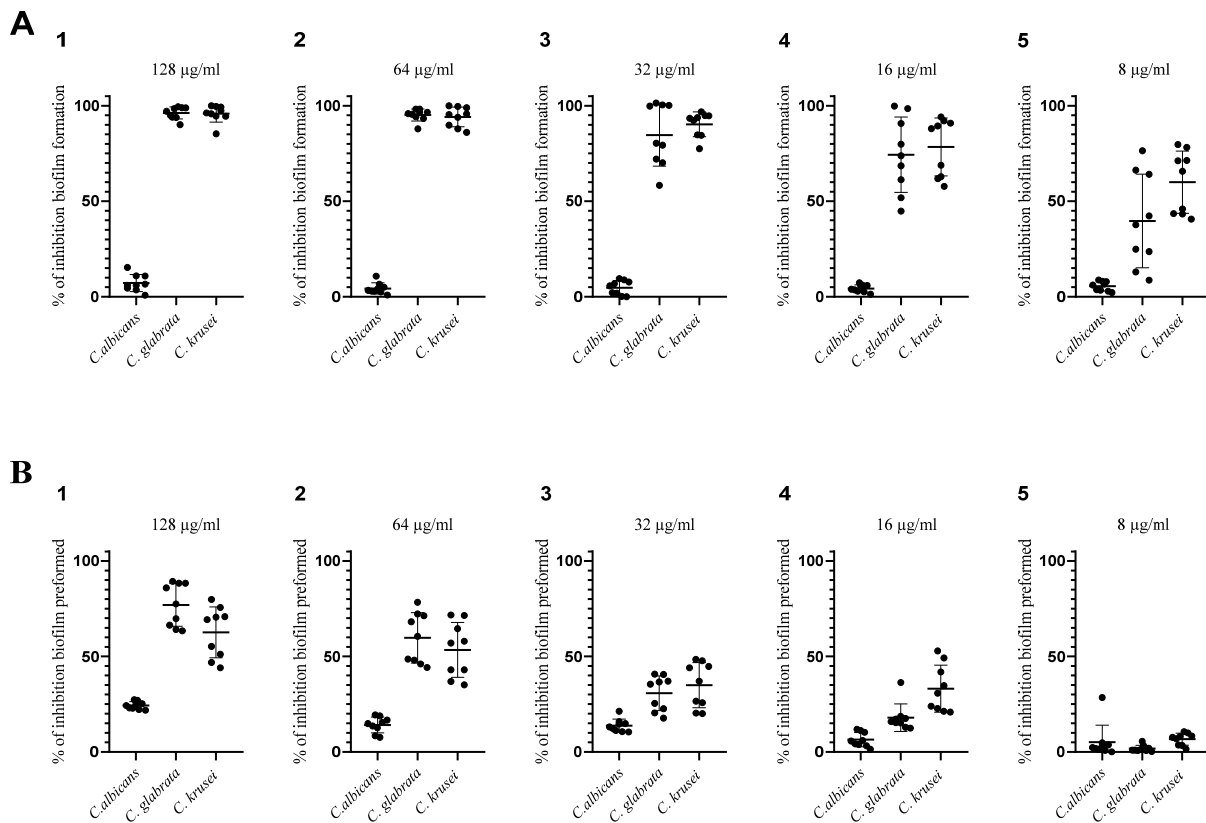


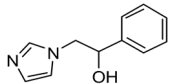
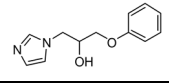
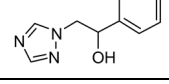
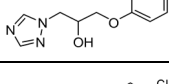
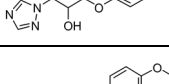
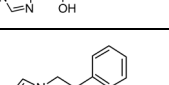
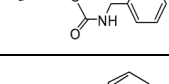
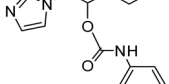
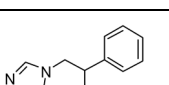
Figure S5. Data distribution by Kolmogorov-Smirnov test

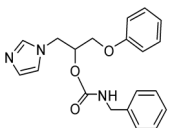
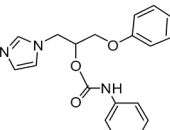
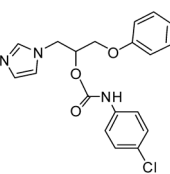
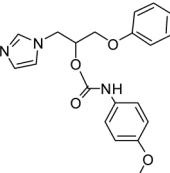
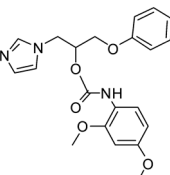
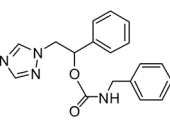
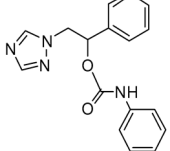
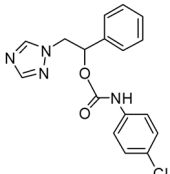
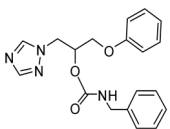
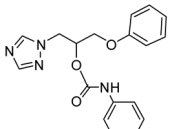


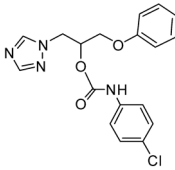
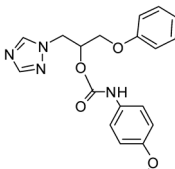
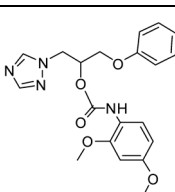
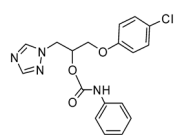
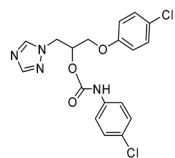
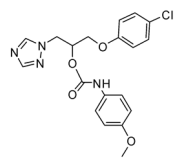
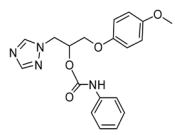
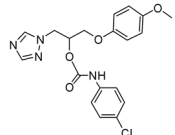
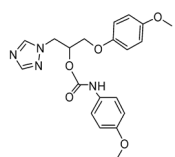
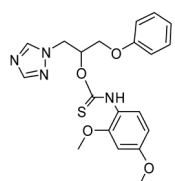
The activity of compound **12** against *C. albicans* ATCC 10231, *C. glabrata* PMC 0849, *C. krusei* PMC 0603 biofilm formation (**A**) and mature biofilm (**B**). Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.22 (**A 1**); Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.44 (**A 2**); Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.33 (**A 3**); Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.44 (**A 4**); Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.22 (**A 5**); Biofilm formation Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 0.88; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.55 (**A 6**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.44 (**B 1**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 0.4; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.44 (**B 2**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 0.88; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 0.88; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.44 (**B 3**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 1.0; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.88 (**B 4**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 1.0; Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. krusei* PMC 0603 0.88; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.88 (**B 5**). Mature biofilm Kolmogorov-Smirnov D between *C. albicans* ATCC 10231, *C. glabrata* PMC 0849 0.44; Kolmogorov-Smirnov D between *C.*

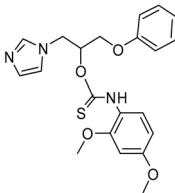
*albicans* ATCC 10231, *C. krusei* PMC 0603 0.55; Kolmogorov-Smirnov D between *C. glabrata* PMC 0849, *C. krusei* PMC 0603 0.66 (**B 6**). The value is expressed as a media of at least three independent biological replicates.

**Table S1.** *In vitro* activity of studied compounds against *A. fumigatus* and dermatophytes.

cpd	structure	<i>Aspergillus fumigatus</i> DSM 790	<i>Microsporum gypseum</i> DSM 3824	<i>Trichophyton rubrum</i> PMC 6604	<i>Trichophyton mentagrophytes</i> DSM 4870
		MIC <sub>50</sub> µg/mL median value			
1		>128	>128	>128	>128
2		>128	>128	>128	>128
3		>128	>128	>128	>128
4		>128	>128	>128	>128
5		>128	>128	>128	>128
6		>128	>128	>128	>128
7		>128	>128	>128	>128
8		>128	>128	>128	>128
9		>128	>128	>128	>128

10		>128	>128	>128	>128
11		>128	>128	>128	>128
12		>128	90.51	>128	>128
13		>128	>128	>128	>128
14		>128	>128	>128	>128
15		>128	>128	>128	>128
16		>128	>128	>128	>128
17		>128	>128	>128	>128
18		>128	>128	>128	>128
19		>128	>128	>128	>128

20		>128	>128	>128	>128
21		>128	>128	>128	>128
22		>128	>128	>128	>128
23		>128	>128	>128	>128
24		>128	>128	>128	>128
25		>128	>128	>128	>128
26		>128	>128	>128	>128
27		>128	>128	>128	>128
28		>128	>128	>128	>128
29		>128	>128	>128	>128

<b>30</b>		>128	90.58	>128	>128
<b>FL C</b>		>128	16	32	16

MIC<sub>50</sub>: minimal inhibitory concentration resulting in 50% reduction in growth compared to control.

FLC: fluconazole.