Title: Repurposing of ribavirin as an adjunct therapy against invasive *Candida* strains: *In vitro* study

Running title: *In vitro* ribavirin activity against *Candida* species.

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Abstract

The use of antifungal agents in clinical settings is limited by the appearance of drug resistance and adverse side effects. There is, therefore, an urgent need to develop new drugs to strengthen the treatment of invasive fungal diseases. The aim of this study is to describe the potential repurposing of ribavirin as an adjunct therapy against *Candida* spp.

Primary screening of Prestwick chemical library against *Candida albicans* ATCC 90028 and fluconazole-resistant *Candida albicans* was performed. Subsequently, we evaluated the response of 100 *Candida* spp strains to ribavirin, an antiviral agent, using the broth microdilution method as recommended by CLSI. We checked the involvement of efflux pump activity in the development of ribavirin-resistance. We studied time-kill curves and performed a checkerboard assay for ribavirin-antifungals combinations study.

Twenty-one nonstandard antifungal compounds were identified, including ribavirin. Ribavirin had, *in vitro,* an antifungal activity against 63 *Candida* strains including *C. albicans, C. parapsilosis* and *C. tropicalis*, with a minimum inhibitory concentrations (MICs) ranging from 0.37 to 3.02 µg/ml, while MICs for *C. krusei*, *C. glabrata, C. lusitaniae* and some *C. albicans* remain high (≥ 24.16 µg/ml). No relation was observed between efflux pump activity and ribavirin-resistance. Ribavirin exhibited a fungistatic activity against multidrug-resistant (MDR) *C. albicans* and a fungicidal activity against *C. parapsilosis* strain. In addition, ribavirin acted synergistically with azoles against *Candida* strains for which ribavirin MICs were < 24.4 µg/ml.

This study highlights the potential clinical application of ribavirin, alone or in association with other antifungal agents, as an adjunct anti-*Candida* drug.

Introduction

Fungi are considered to be one of the main causes of human infections, particularly among immunocompromised individuals and hospitalized patients with serious immunosuppressive conditions such as HIV and organ transplantation (1). Infections due to *Candida* spp are one of the most common invasive fungal diseases (2). In a recent study, *Candida* spp were identified as the most frequent cause of bloodstream infection in hospitalized patients (3). Only three classes of antifungal agents are currently used to treat invasive *Candida* species infections, namely azoles, polyenes and echinocandins (4). Although these classes of antifungal agents are usually active against many fungal infections, their applicability and efficacy could be compromised due to their high toxicity, narrow spectrum of activity and the development of drug resistance (5). Hence, there is a constant need for other compounds that possess antifungal properties. An interesting thought in this field has been addressed via the drug repurposing, where FDA-approved drugs could be tested and used in another therapeutic class (6). Applying this approach by screening 1,920 compounds belonging to three FDA-libraries drugs against *Candida albicans* strains, Tournu et *al.,* recently identified, ribavirin, a purine nucleoside analog, as a potential *C. albicans* vacuole disrupting agent (7). Ribavirin is a guanosine analog that produces broad-spectrum activity against many RNA and DNA viruses. However, it is mainly used to treat hepatitis C virus (HCV) in combination with interferon-α (8). Here, we investigated the *in vitro* activity of ribavirin against different clinical *Candida* species, including emerging fluconazole-resistant *Candida albicans*. We also evaluated the eventual synergy between ribavirin and common antifungal agents.

62 **Results and discussion**

63 By testing the FDA-approved library of 1,280 drugs against *C. albicans* ATCC 90028
64 (quality control) and *C. albicans* (Q181103513) that have high resistance to fluconazole and
65 all echinocandin antifungal agents, 21 non-standard antifungal hits showed fungal growth
66 inhibition greater than 90%. These primary compounds include 7 antibacterial drugs, 3
67 antihelmintic agents and 11 compounds belonging to different therapeutic classes (Table 1).

68 The principal aim of our study is to identify new molecules whose antifungal action is not
69 known and/or well investigated and which can be used in clinical practices without any
70 constraints for patients. Among these 21 hits obtained, 14 of them have already been well
71 identified and studied in previous works (Table 1), confirming the validity and the
72 reproducibility of our results and our screening technique. However, to our knowledge, the
73 fungal growth inhibition induced by the remaining 7 compounds including pinaverium
74 bromide, avermectin B1, triclabendazole, tetraethylenepentamine pentahydrochloride,
75 thioguanine, and anthralin, has never been previously reported neither well studied as well as
76 the ribavirin compound (Table 1).

77 Among these unknown antifungal hits, we focused our study on ribavirin drug for several
78 purposes; (i) it presents a percentage of fungal growth reduction equal to 96%. (ii) Ribavirin
79 has been shown to disrupt vacuolar function in the pathogen *C. albicans* (7), however, no
80 report describes directly its antifungal efficacy (i.e. MICs, fungicidal or fungistatic activity,
81 synergy with antifungals …). (iii) Most of these 7 hits have been designed for a specific non-
82 infectious indication (antispastic, antilipemic, antineoplastic, antipsoriatic) (Table 1) whereas
83 ribavirin was approved to treat liver infectious disease (9), so its repurposing as an antifungal
84 agent could be admissible. (iv) Reversible hemolytic anemia is the unique adverse effect
85 observed with ribavirin treatment, unlike the cytotoxic effect that of other molecules, such as

thioguanine and avermectin B1, may have on human cells due to their non-selective activities (10, 11). (v) Ribavirin can be administered orally (capsule or tablet), intravenously and through inhalation (9), while the topical application is only possible for some molecules such as anthralin (12).

That is why, in this study, ribavirin susceptibility testing against a large panel of 100 *Candida* spp strains was performed, and ribavirin MICs were determined. Consequently, we report here the efficacy of this compound against a collection of 60 *Candida* spp strains (total No = 100 strains), including 6 *C. parapsilosis*, 5 *C. tropicalis,* and 49 *C. albicans*, with MIC ranging from 1.56 to 12.5 µM (i.e. 0.37 to 3.02 µg/ml). Among these 60 clinical strains tested, *C. parapsilosis* and *C. tropicalis* showed the greatest susceptibility to ribavirin with MICs mainly ≤ 3.02 µM (i.e. ≤ 0.75 µg/ml). Interestingly, ribavirin was effective against the clinical strain that was resistant to both fluconazole and echinocandin agents (*C. albicans* Q181103513), with a low MIC equal to 6.25 µM (i.e. 1.51 µg/ml). On the other hand, the ribavirin MICs obtained against *C. krusei*, *C. lusitaniae, C. auris,* and *C. glabrata* were mainly ≥ 100 µM (i.e. ≥ 24.16 µg/ml) (Fig 1).

In order to study fungicidal and fungistatic activities of ribavirin compound, time-kill curves were performed with *C. albicans* and *C. parapsilosis*. Ribavirin exhibited fungistatic activity against MDR *C. albicans* and *C. albicans* ATCC 90028 strains tested. At ribavirin concentrations equal to 0.5 X MIC, no inhibitory effect was observed and the curve was nearly identical to those for the control. At concentrations ≥ 1 X MIC, a concentration-independent fungistatic effect was observed. In contrast, ribavirin fungicidal activity against *C. parapsilosis* was noted with Minimal Fungicidal Concentration (MFC) ≥ to two-fold its MIC value (MFC = 3.12 µM; i.e. 1.5 µg/ml) (Fig 2).

109 Ribavirin is part of the WHO Model List of Essential Medicines, as an effective and safe drug
110 used in a health system. Ribavirin is administered orally at a dose of 1,200 mg (600 mg twice
111 a day) during 4 weeks to achieve a stable plasmatic concentration of 2.2 µg/ml (9). A
112 retrospective study performed by J.F. Jen *et al,* on patients infected with the HCV, showed
113 that 49% of the sustained virological response was achieved at week 4 via ribavirin plasmatic
114 concentrations that ranged from 3.5 to 4 µg/ml. In addition, the authors preconized the
115 achieving of higher ribavirin plasmatic concentrations in the treatment of a patient infected
116 with HCV genotype 1 (13). Moreover, a prospective open-label study in patients infected with
117 HCV genotype 1, demonstrated that the increase in the ribavirin treatment dose (maximum
118 daily dose of 3,600 mg) resulted in ribavirin plasmatic concentrations of 5.37 µg/ml. In this
119 case, undetectable HCV RNA on 9/10 patients during the fourth week was obtained but
120 associated with more frequent side effects, such as anemia (14). Thus, plasma concentrations
121 equal to the MICs obtained from most of the *Candida* spp strains tested in this study can be
122 easily achieved. The main side effect of ribavirin treatment is reversible hemolytic anemia
123 that may be observed in patients with chronic hepatitis C (15). The use of this drug for a short
124 time therapy, along with the use of hematopoietic growth factors, can manage the risk of
125 severe anemia in maintaining an effective concentration.

126 Nonetheless, some *Candida* strains were resistant to ribavirin, including some *C. albicans*
127 strains and all the tested *C. glabrata*, *C. krusei* and *C. lusiatnia* strains. Among the many drug
128 resistance mechanisms that have been described in yeast, one specific or non-specific
129 mechanism is the overexpression of membrane-associated carriers acting as drug efflux
130 pumps (16). Thus, we supposed that reduced permeability of the membrane to ribavirin drug
131 and the potential role of efflux pumps in the extrusion of ribavirin molecule can be the
132 mechanism which explains the ribavirin-resistance. That is why we checked the efflux-pumps
133 activity using CCCP (carbonyl cyanide 3-chloro-phenylhydrazone) nonspecific efflux-pumps

inhibitor (EPI) and the specific calcium channel-blocker; verapamil against ribavirin-resistant strains. The activity of ribavirin did not change with the addition of CCCP or verapamil. No changes were reported in the ribavirin MICs of all strains tested after EPIs treatment, which could indicate the potential normal transport of ribavirin in the isolates tested, although we did not quantify the intracellular level of ribavirin. This result leads us to the possibility of the existence of a specific pathway that could induce the development of ribavirin resistance in yeasts.

Mutation frequencies of *C. albicans* ATCC on RPMI medium containing 1.52, 3.04, 6.08 µg/ml of ribavirin were $1.5x10^{-4}$, $1.1x10^{-4}$, $9.2x10^{-4}$ respectively. Mutation frequencies of *C. albicans* (Q181103513) on RPMI-agar plates with 3.04, 6.08 and 12.16 µg/ml of ribavirin were $1.6x10^{-4}$, $1.1x10^{-4}$, $9.8x10^{-4}$ respectively. The mutation frequency was approximatively about 10^{-4} which is relatively higher than that reported for classical antifungals (17). This result indicates that it is preferable to use ribavirin for a short time or in association with other antimicrobial drugs. Therefore, in addition to the discovery of new agents effective against fungi, a pragmatic alternative would be to improve the activity of old antifungal agents by using a combination of drugs. Thus, testing the *in vitro* ribavirin-antifungals combinations could be very useful to increase the antimicrobial spectrum of the usual antifungals used and to minimize the development of drug resistance and side effects by reducing the drug concentration.

Investigations for potential *in vitro* synergy between ribavirin and antifungal agents were tested against *Candida* strains which are resistant to any of azoles (fluconazole (No = 6 strains), itraconazole (No = 9 strains), posaconazole (No = 19 strains) and/or flucytosine (No = 11 strains) (Table 2). The combination of ribavirin and fluconazole was evaluated in 6 fluconazole-resistant *Candida*. spp strains (5 *C. glabrata* and one MDR *C. albicans*) (Table 2). Interestingly, a significant synergism (Fractional Inhibitory Concentration Index (FICI) =

159 0.12) was demonstrated against the MDR *C. albicans* strain (Q181103513), since this
160 combination has lowered the MICs of fluconazole and ribavirin from 256 µg/ml and 1.5
161 µg/ml to 1 µg/ml and 0.18 µg/ml, respectively. However, this combination (i.e. ribavirin with
162 fluconazole) was only synergistic for one *C. glabrata* strain and was indifferent to the
163 remaining 4 strains. It is worthy to note that these latter strains have initially high ribavirin
164 MICs (MICs ≥24.4 µg/ml) (Table 2). When ribavirin was combined with itraconazole and
165 tested against 9 itraconazole-resistant strains, synergistic activities were observed for the two
166 *C. albicans* strains tested, while indifferent activities were noted for most *C. glabrata* strains
167 (5/7) analyzed in this study (Table 2). Furthermore, in ribavirin-posaconazole combinations,
168 the highest rates of synergy were observed for *C. albicans* strains (7/9; 78%) and for *C.*
169 *tropicalis* (2/2; 100%) (Table 2), but not for *C. glabrata* strains (1/7) and for *C. krusei* (0/1). It
170 is worth mentioning again that ribavirin-azole association was mainly synergistic against
171 *Candida* strains that have ribavirin MICs < 24.4 µg/ml. Synergy was not seen with ribavirin
172 in combination with flucytosine against any of the *Candida* spp strains tested in this study (11
173 strains; Table 2). This could be explained by the fact that both molecules (i.e. ribavirin and
174 flucytosine) belong to the same therapeutic class of nucleoside analogues which can disturb
175 the DNA synthesis of a given microorganism. Finally, none of the tested combinations was
176 found antagonism against *Candida* strains.

177 Mechanisms of action of ribavirin on the HCV have been correctly elucidated, where four
178 pathways were described; (i) inhibition of HCV replication (RNA polymerase), (ii) inhibition
179 of IMPDH enzyme, (iii) induction of RNA mutagenesis and (iv) adaptation of immune
180 response to the HCV(9). In contrast, the mechanism of ribavirin action on *Candida* species
181 remains unclear. Thus, the most important perspective of this study will be the
182 characterization of the mode of action of ribavirin against *Candida* species.

Our findings demonstrate that ribavirin exhibit antifungal activities against *Candida* species especially against *C. albicans*, *C. parapsilosis* and *C. tropicalis*. Its efficacy against MDR *C. albicans* could be a promising alternative strategy for the treatment of invasive fungal infections. Nonetheless, the ribavirin MICs of some *Candida* strains (especially those belonging to *C. glabrata*, *C. krusei* and *C. lusitaniae*) remain higher than the achievable plasmatic concentration in human. This latter phenomenon of high MICs is not associated with the overexpression of efflux pump's activity. Finally, ribavirin has synergistic activities with either fluconazole, itraconazole or posaconazole, but do not have with flucytosine, against particularly *Candida* strains whose ribavirin MICs are < 24.4 µg/ml. Consequently, *in vivo* activities of ribavirin need further investigation in order to introduce ribavirin as an antifungal agent, alone or in association with other antifungals, in clinical mycology practice.

Materials and methods

Screening of 1280 FDA-approved drugs against C. albicans

Using the same concept of drugs-repurposing, an initial FDA-approved library of 1,280 drugs (Prestwick, Illkirch graffenstaden, France) (18) screen was performed against *Candida albicans* ATCC 90028 (quality control) and clinical *Candida albicans* strain, which is resistant to fluconazole and all echinocandins agents at a fixed concentration of 10 µM. This concentration was chosen according to previous screening studies in order to select "hits" which present antifungal activities at low concentrations (i.e. avoiding the toxicity and adverse effect of tested drugs at high concentrations). The fungal inoculum was prepared using RPMI-1640 medium (Sigma Aldrich, St Louis, France) according to the Clinical and Laboratory Standards Institute (CLSI M27-A2, Vol 22, NO.15) protocol. Sixteen of 96-well plates were used. Each plate contains 80 compounds, the first column served for the positive control with untreated fungi and the last one contains the medium as a negative control. The

plates were incubated 24 hours at 37°C. Then, the optical density (OD) was obtained using a spectrophotometer and the percentages of fungal growth inhibition were calculated in relation to the untreated wells.

Fungal strains

A collection of 96 *Candida* spp strains was tested, including 72 *C. albicans,* 5 *Candida parapsilosis,* 8 *Candida glabrata,* 5 *Candida tropicalis,* 3 *Candida krusei* and 3 *Candida lusitaniae* strains with different antifungal susceptibilities (Table S1). The species of all isolates were identified using Matrix-assisted laser desorption/ionization mass spectrometry (MALDI-TOF MS) (19). All strains were recovered from La Timone University Hospital in Marseille and were isolated from different clinical samples, mainly from blood culture, urine and vaginal swabs (Supplementary Table 1). *C. albicans* ATCC 90028, *C. parapsilosis* ATCC 22019, *C. auris* DSMZ 21092 and *C. krusei* ATCC 6258 were used as quality control (Table S1).

Antifungals susceptibility testing

Antifungal susceptibility testing was performed using commercial broth microdilution plates; Sensititre®YeastOne® (Thermo Fisher Scientific, Schwerte, Germany) containing 9 antifungals belonging to the 4 therapeutic classes, namely polyene (amphotericin B), azoles (fluconazole, posaconazole, voriconazole and itraconazole), echinocandin (anidulafungin, caspofungin and micafungin) and 5-flucytosine. The minimum inhibitory concentrations (MICs) obtained for *Candida* spp strains tested (Table S1) were compared to the breakpoints provided by the manufacturers or, to the Clinical and Laboratory Standards Institute (CLSI) cut-offs (CLSI M27-A2, Vol 22, NO.15), to assess the susceptibility of each strain to the different antifungal agents tested.

Ribavirin susceptibility testing

Using the broth microdilution method as outlined by the CLSI, ribavirin MICs were performed. First, ribavirin powder (Sigma Aldrich, St Louis, France) was dissolved in water to obtain a stock solution at a concentration of 10 mg/ml. Then, the ribavirin stock solution was diluted in RPMI-1640 broth medium (Sigma Aldrich, St Louis, France) to obtain a concentration of 200 µM (i.e. 48.32 µg/ml); twofold more concentrated than the starting concentration. Fungal suspensions at 0.5 MacFarland were prepared and working yeast suspensions were obtained by a 1:100 dilution followed by a 1:20 dilution of the initial suspension in RPMI broth medium. Further, serial ribavirin dilutions (twofold dilutions) ranging from 100 to 0.2 µM (i.e. 24.16 to 0.047 µg/ml) were tested by inoculating 100 µl of ribavirin-containing well at the appropriate concentration, with 100 µl of final fungal suspension. Ribavirin concentration which is achievable in human plasma without toxic effects (≈ 6 µg/ml) was considered to establish the concentration ranges used in the two-fold dilution testing. The MIC of ribavirin was defined as the concentration that resulted in complete growth inhibition compared to untreated control wells. This later was determined visually after 24 hours of incubations at 37°C and using OD by spectrophotometric measurement.

Time-kill assay

In order to assess the fungicidal or fungistatic activity of ribavirin compound against three fungal strains (2 *C. albicans* and 1 *C. parapsilosis*), ribavirin time-kill analysis was performed as previously described (20); (*C. albicans* ATCC 90028 was used as quality control). The ribavirin action was investigated at 4 different concentrations (0.5xMIC, 1xMIC, 2xMIC, 4xMIC) and the fungal growth was followed-up by the CFU/ml number after 2, 6, 12, 24, 36, 48 hours of incubation at 37°C. The results were analyzed using the GraphPad Prism 5.3 software (GraphPad Inc., San Diego, CA, USA).

Testing of efflux-pumps activity

The implication of efflux-pumps in ribavirin resistance mechanism in some ribavirin-resistant *Candida* spp strains (2 *C. glabrata*, 1 *C. krusei*, 1 *C. lusiataniae*, 1 *C. albicans*) was verified using CCCP (carbonyl cyanide 3-chloro-phenylhydrazone) and verapamil inhibitors, which decrease the MICs when resistance was correlated to this mechanism (21). The CCCP and verapamil inhibitors were tested at two final concentrations (0.5 µg/ml, 10 µg/ml). To ensure that these concentrations do not affect yeast growth, control growth wells containing efflux-pumps inhibitors (EPIs) alone in RPMI medium were added. We determined ribavirin MICs for all strains before and after adding EPIs to the RPMI medium.

Mutant frequency

The frequencies of spontaneous mutations were determined on *C. albicans* ATCC 90028 and multidrug-resistant (MDR)- *C. albicans* (Q181103513) by plating 100 µl of growing yeast with appropriate dilution (10^3 CFU) on ribavirin-free plates and without dilution (10^5 CFU) on ribavirin-containing plates (17). RPMI-1640 agar plates with different ribavirin concentrations (1xMIC, 2xMIC, 4xMIC) were prepared as previously described (22). Spontaneous mutation frequencies were calculated, after 24 hours of incubation at 37°C, by dividing the number of resistant colonies grown on a given plate by the initial inoculum pipetted. Mutant resistance phenotypes were confirmed by determining the ribavirin MICs of mutant colonies.

***In vitro* interaction studies of ribavirin combination with antifungal drugs**

In order to exploit the potential for improved efficacy, reduced toxicity and reduced risk of drug resistance development (23), ribavirin-antifungal agent combinations were tested against 29 *Candida* spp strains, including 12 *C. albicans* strains, 8 *C. glabrata* strains, 4 *C. tropicalis* strains, 1 *C. lusitaniae* strain and 4 *C. krusei* strains. Based on the checkerboard association

assay, combinations of ribavirin with each of the following antifungals: fluconazole, itraconazole, posaconazole, and flucytosine (Sigma Aldrich, St Louis, France) were tested. The effects of different combinations performed on different strains were analyzed using the Fractional Inhibitory Concentration Index (FICI) calculated as previously described (23) and interpreted as follows : if the FICI is ≤0.5, then there is synergy between the tested antimicrobials; if it is >0.5 but ≤4, there is indifference or additivity between the tested antimicrobials, and if the FICI is >4, that means that there is an antagonism between the tested antimicrobials.

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289

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384

385 **Figures legends**

386 **Fig 1**. Distribution of ribavirin MICs for all clinical *Candida* spp strains tested in this study.
387 *C.parapsilosis* (brown), *C. glabrata* (green), *C. auris* (yellow), *C. albicans* (red), *C.*
388 *krusei* (blue), *C. lusitaniae* (black), *C. tropicalis* (grey), MICs – Minimum Inhibitory
389 Concentrations.

390 **Fig 2**. Values for Log_{10} of number of CFU/mL versus time for *Candida* strains tested against
391 ribavirin; *C. albicans* Q181103513 (A), *C. albicans* ATCC 90028 (B) and *C. parapsilosis*
392 Q181208447 (C) at the following concentrations: control (■ filled square), 0.5XMIC (▲
393 triangle), 1XMIC (● circle), 2XMIC (♦ lozenge), 4XMIC (─ line).

394 **Table 1**. Hits obtained by the primary screening of 1,280 FDA-approved drugs of Prestwick chemical library at 10 μM against both *C. albicans*

395 ATCC 90028 and fluconazole-resistant *C. albicans* (strain Q181103513).

Drug	Therapeutic class	Fungal growth inhibition (%)	References
Auranofin	Analgesic	99	26
Disulfiram	Antabuse effect	98	27
Pinaverium bromide	Antispastic	98	Unknown
Avermectin B1	Antihelmintic	98	Unknown
Pyrvinium pamoate	Anthelmintic	97	28
Triclabendazole	Anthelmintic	96	Unknown
Pentamidine isethionate	Antiprotozoal	96	29
Tetra ethylenepentamine pentahydrochloride	Antilipemic	94	Unknown
Ribavirin	Antiviral	96	7
Pentetic acid	Chelating/Radioprotectant	98	29
Thioguanosine	Antineoplastic	96	Unknown
Anthralin	Antipsoriatic	96	Unknown
Thonzonium bromide	Antiseptic	98	30
Clioquinol	Antiamebic/Antiseptic	98	7
Clotrimazole	Antibacterial	98	31
Chloroxine	Antibacterial	98	32
Chlorhexidine	Antibacterial/Antiseptic	98	32
Dequalinium dichloride	Antibacterial/Antiseptic	98	33
Methyl benzethonium chloride	Antibacterial	98	34
Benzethonium chloride	Antibacterial/Antiseptic	98	34
Ciclopirox ethanolamine	Antibacterial	97	35

396

397 **Table 2**. Minimum inhibitory concentrations (MIC) and fractional inhibitory concentration indexes (FICI) for the combinations of ribavirin with

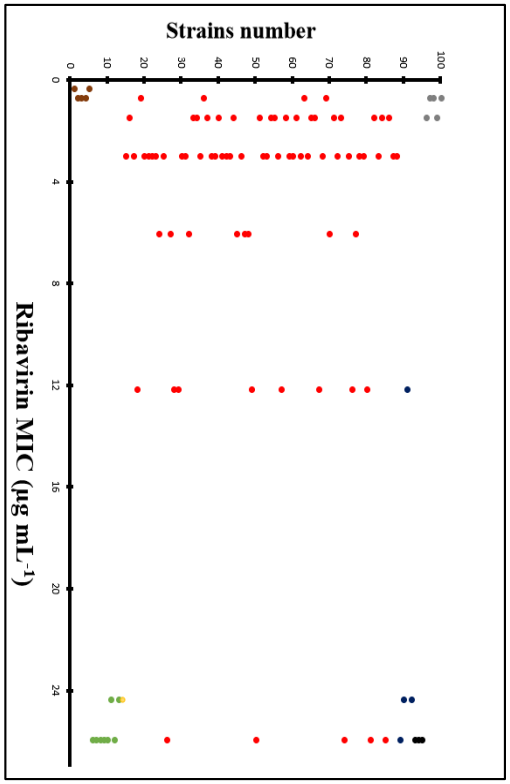
398 antifungals.

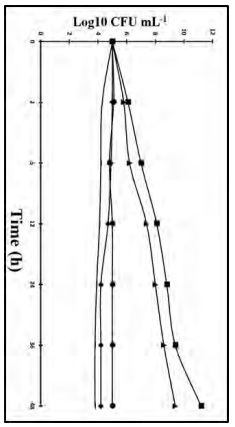
Candida spp	Strains	FLU MIC	ITRA MIC	POSA MIC	5-FC MIC	RBV MIC	RBV-FLU FICI	RBV-ITRA FICI	RBV-POSA FICI	RBV-5-FC FICI
C. glabrata	Q181201614	8	8	1	<0.06	>24.4	ND	0.7 (I)	ND	ND
C. glabrata	Q181198565	64	1	8	<0.06	>24.4	0.7 (I)	1 (I)	0.7 (I)	ND
C. glabrata	Q181203672	128	1	>8	<0.06	>24.4	0.6 (I)	0.6 (I)	0.4 (S)	ND
C. glabrata	Q181202903	16	4	8	0.06	>24.4	ND	0.7 (I)	1 (I)	ND
C. glabrata	Q181255715	64	16	4	<0.06	>24.4	0.6 (I)	0.6 (I)	0.8 (I)	ND
C. glabrata	Q181280604	4	0.25	0.5	<0.06	24.4	ND	ND	0.7 (I)	ND
C. glabrata	8070855959	128	16	8	<0.06	>24.4	0.6 (I)	0.1 (S)	2 (I)	ND
C. glabrata	Q181208658	128	16	8	<0.06	24.4	0.5 (S)	0.5 (S)	1 (I)	ND
C. albicans	Q181103513	256	0.25	0.25	<0.06	1.52	0.12 (S)	ND	0.1 (S)	ND
C. albicans	Q181198103	4	0.25	1	<0.06	3.05	ND	ND	0.2 (S)	ND
C. albicans	Q181201380	0.5	0.06	0.03	>64	1.52	ND	ND	ND	1.5 (I)
C. albicans	Q181203339	1	0.06	0.12	<0.06	>24.4	ND	ND	0.7 (I)	ND
C. albicans	490890568	1	0.5	0.25	0.5	3.05	ND	ND	0.4 (S)	ND
C. albicans	Q181264539	0.25	0.12	0.06	>64	0.76	ND	ND	ND	1.2 (I)
C. albicans	Q181252599	1	0.12	0.12	0.12	6.1	ND	ND	0.3 (S)	ND
C. albicans	Q181253570	<0.012	0.03	0.15	0.12	>24.4	ND	ND	0.5 (S)	ND
C. albicans	Q181211559	2	1	0.5	1	12.2	ND	0.4 (S)	0.6 (I)	ND
C. albicans	Q181226987	0.25	0.06	0.03	64	6.1	ND	ND	ND	2 (I)
C. albicans	Q181260978	2	1	0.5	0.5	12.2	ND	0.5 (S)	0.5 (S)	ND
C. albicans	Q181108884	16	0.5	0.5	1	1.52	ND	ND	0.3 (S)	ND
C. krusei	ATCC 6258	32	0.12	0.25	8	>24.4	ND	ND	0.6 (I)	ND
C. krusei	Q181262141	32	0.12	0.12	4	24.4	ND	ND	ND	0.8 (I)
C. krusei	Q181263662	16	0.12	0.12	2	12.2	ND	ND	ND	0.6 (I)
C. krusei	Q181208438	32	0.25	0.25	8	24.4	ND	ND	ND	0.6 (I)

C. lusitaniae	Q181206420	0.5	0.12	0.03	32	>24.4	ND	ND	ND	0.7 (I)
C. tropicalis	Q181257439	1	0.12	0.12	64	0.76	ND	ND	0.4 (S)	0.7 (I)
C. tropicalis	Q181250041	1	0.12	0.06	64	1.52	ND	ND	ND	1 (I)
C. tropicalis	8070845333	2	0.12	0.12	>64	1.52	ND	ND	0.4 (S)	1.1 (I)
C. tropicalis	Q181203338	1	0.12	0.06	64	0.76	ND	ND	ND	1 (I)

399 Abbreviations: FLU, fluconazole; ITRA, itraconazole; POSA, posaconazole; 5-FC, flucytosine; RBV, ribavirin; S, synergy; I, indifference; ND,

400 not determined.





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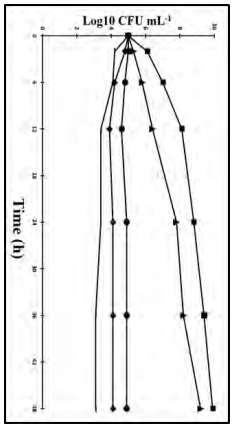

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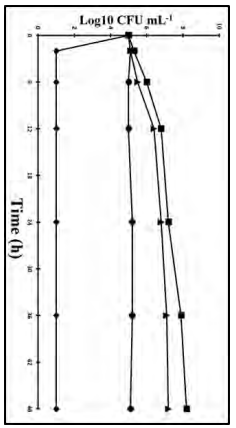

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