Review: Diphenylphosphane Derivatives of Ketoconazole and Oteseconazole/VT-1161, Promising New Azole Compounds in the Treatment of *Candida albicans* Infections

Muhammad Rosyid As Sabiq[1,2] and Peni Indrayudha[1(✉)]

[1] Faculty of Pharmacy, Universitas Muhammadiyah Surakarta (UMS), Jl. A. Yani Tromol Pos I, Pabelan, Kartasura, Sukoharjo, Indonesia
peni.indrayudha@ums.ac.id

[2] Master of Pharmacy, Graduate School of Universitas Muhammadiyah Surakarta, Jl. A. Yani Tromol Pos I, Pabelan, Kartasura, Sukoharjo, Indonesia

Abstract. In some treatments of diseases, there is a risk of possible fungal infection of Candida albicans. The risk of infection can be exacerbated by the occurrence of resistance to anti-fungal drugs, including resistance to azole compounds. The purpose of this research is to conduct a review of new azole compounds in the treatment of Candida albicans infections. This study uses a literature review method. The source of the data for this research comes from journals that have been published and indexed in the PubMed search with several inclusion and exclusion criteria. The results showed that all new drug candidates of azoles showed excellent antifungal activity against Candida albicans in vitro. In general, all azoles have a similar mechanism of action of inhibiting the enzyme 14α-demethylase (CYP51) through competitive inhibition. Inhibition of CYP51 causes fungal cells to lose ergosterol, implicated in cell death. The new azole compounds showed stronger CYP51 inhibitory activity and can suppress the growth of fluconazole-resistant strains. The new azole compounds showed better CYP51 inhibitory activity against Candida albicans resistance

Keywords: "Diphenylphosphane derivatives of ketoconazole" · oteseconazole · VT-1161 · "Candida albicans"

1 Introduction

Advances in modern medicine, such as cancer chemotherapy, the consumption of large amounts of antibiotics, and immunosuppression evidently can increase the risk of opportunistic fungal infections such as *Candida* albicans [1]. Every year, as many as 150 million people worldwide experience fungal infections. A total of 750,000 cases were due to invasive Candida infections. Azoles has been used as the first line therapy in fungal infections [2].

The azole antifungal drugs are known to have a mechanism of inhibition of the enzyme 14α-demethylase (CYP51) in *Candida albicans* [2]. CYP51 belongs to the

A. Sri Wahyuni et al. (Eds.): ICB-Pharma 2022, AHCPS 3, pp. 318–328, 2023.
https://doi.org/10.2991/978-94-6463-050-3_27

cytochrome P450 monooxygenase (CYP) superfamily and acts as an enzyme in the synthesis of ergosterol. Ergosterol is a fungus-specific sterol [3]. As a group of enzymes classified as a "superfamily", cytochrome P450 plays an important role as a catalyst in primary and secondary metabolic pathways [4]. Ergosterol determines the fluidity, permeability and activity of membrane-associated proteins [5].

The presence of azole drugs that bind to CYP51 causes disruption of the ergosterol synthesis cycle. The reduced amount of ergosterol in fungi will damage the cell membrane by weakening the structure and making it hollow [6].

Azole through its mechanism of inhibition against CYP51 has been shown to be able to kill fungal cells. Regarding their use, azole antifungals are relatively safe, have a high therapeutic index, and are easy to administer (often by oral). Triazoles (one of the azole groups) are the standard first-line therapy to fight fungal infections. However, long-term azole treatment and prophylactic use increase the risk of resistant strains [7]. Cases of antifungal drug resistance are also becoming more frequent, therefore the new drug development and better understanding of target receptors are needed [1].

The case of *Candida albicans* resistance to azoles encourages research on efforts to overcome this problem. In this review, we summarized 2 new azole compounds that are promising in the treatment of Candida albicans infections, namely diphenylphosphane derivatives of ketoconazole and oteseconazole/VT-1161.

2 Method

This study uses a literature review method. The source of the data for this research comes from journals that have been published and indexed in the PubMed search with several inclusion and exclusion criteria. Table 1 shows the inclusion and exclusion criteria of the publications used.

The selected publications were analyzed. Extraction of the data obtained will be presented in the results section and used to draw conclusions.

The data search strategy in PubMed uses 4 groups of keywords that are aligned with the inclusion criteria and combined with Boolean operators as follows:

Table 1. Inclusion and exclusion criteria

Inclusion	Exclusion
Indexed in searches using PubMed	Not Indexed in searches using PubMed
Year of publication in the range 2011–2021	Year of publication < 2011
Publication in English	Publication in a language other than English
There is discussion regarding trials of new azole compounds or azole resistance to *Candida albicans*	The discussion is not related to trials of new azole compounds or azole resistance to *Candida albicans*
Research article, full text	Only in the form of manuscript fragments, or only research abstracts

(new azole* OR azole* OR azoles) AND (inhibit* OR inhibits* OR inhibition* OR inhibitory) AND (14α-demethylase* OR demethylase* OR cyp51* OR 14alpha-demethylase*) AND (c.albicans* OR "candida albicans" OR albicans*).

3 Results

The search results obtained as many as 218 publications. Based on the inclusion criteria, there are more than 100 journals that meet the criteria. However, the authors chose to focus on discussing the diphenylphospane derivatives of ketoconazole and oteseconazole/VT-1161.

3.1 Diphenylphosphane Derivatives of Ketoconazole

Diphenylphosphane derivatives of ketoconazole are promising antifungal agents [8]. There were 4 derivatives studied, namely:

a. Ketoconazole Diphenylphosphane (KeP),
b. Ketoconazole Diphenylphosphane Oxide (KeOP)
c. Ketoconazole Diphenylphosphane Sulphide (KeSP) and
d. Ketoconazole Diphenylphosphane Selenide (KeSeP).

Metal complexes are modification of drugs that gain interest nowadays. They are characterized to have abundances of easily modifiable geometries and accessible redox states of metal centers [9].

The synthesis process of these compounds can be seen in Fig. 1. The principle of this group of compounds is to form a complex of azole compounds with metal ions to increase the anti-fungal ability. The azole used was ketoconazole, then reacted with aminomethylphospane.

Ketoconazole Diphenylphosphane (KeP) was synthesized using deacylated ketoconazole in a modified Mannich condensation reaction using hydroxy methylphosphanes with amines. Ketoconazole Diphenylphosphane Oxide (KeOP) was synthesized using KeP with a stoichiometric amount of H2O2. In the synthesis of Diphenylphosphane Sulphide (KeSP), in addition to H2O2 S8 (Sulfur) was added, while in the synthesis of Diphenylphosphane Selenide (KeSeP) Se (Selenium) was added.

The four compounds were then tested for anti-fungal activity using multiple strains of Candida albicans. Based on Table 2 there are 6 sources of strains used, namely: CAF2–1; DSY150; B3; B4; Gu4 and Gu5. Each of the 2 strains was a pair before and after mutation. DSY150 is a mutation of CAF2–1. Strain B4 is a mutation of B3, and Gu5 is a mutation of Gu4. Information on the characteristics of the strains is listed in full in Table 2. The antifungal test method used was fungistatic MIC_{50} in vitro compared with fluconazole. Complete data are presented in Table 3.

Based on the data in Table 3, ketoconazole was 16 times more active than fluconazole against CAF2–1 strain. In the same strain, KeP, KeSP and KeSeP were 2 times more active than fluconazole, while KeOP was 8 times more active. The MIC50 value in the

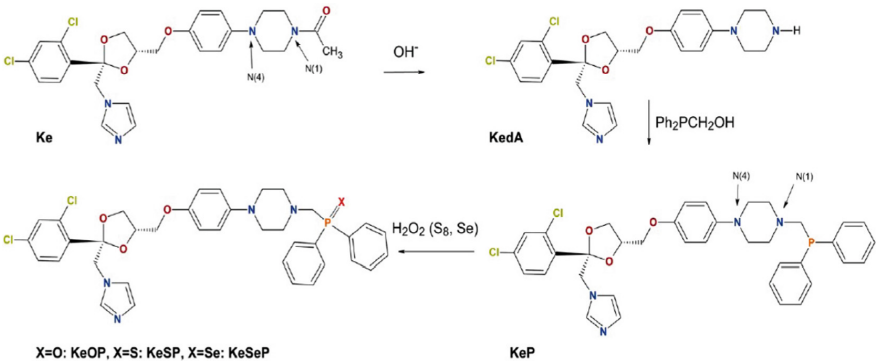

Fig. 1. The synthesis process of KeP, KeOP, KeSP and KeSeP (Adapted from "New diphenylphosphane derivatives of ketoconazole are promising antifungal agents")

Table 2. Candida albicans strains used

No	Strain	Description
1	CAF2–1	Normal *Candida albicans*
2	DSY150	Mutations of CAF2–1 lacking the CDR1, CDR2 and MDR1 genes
3	B3	*Candida albicans* from patients before taking fluconazole
4	B4	*Candida albicans* from patients after taking fluconazole with mutations that increase MDR1 expression
5	Gu4	*Candida albicans* from patients before taking fluconazole
6	Gu5	*Candida albicans* from patients after taking fluconazole with mutations that increase CDR1 and CDR2 expressions

Table 3. MIC_{50} (μM) values of fluconazole, ketoconazole and ketoconazole derivatives

Strain	Flc	Ke	KeP	KeOP	KeSP	KeSeP
CAF2–1	6.25	0.05	3.13	0.78	3.13	3.13
DSY150	0.78	0.02	0.78	0.2	0.39	0.78
B3	6.25	0.05	3.13	1.56	1.56	3.13
B4	50	0.39	3.13	3.13	3.13	6.25
Gu4	6.25	25	3.13	12.5	50	12.5
Gu5	> 200	50	50	25	> 200	> 200

DSY150 strain was smaller than the value in the CAF2–1 strain. This is because the mutation that occurs does not give rise to the CDR1, CDR2 and MDR1 genes.

The difference in the MIC_{50} value of fluconazole was much adrift between B3 and B4 due to a mutation in the increased expression of the MDR1 gene that was resistant

to fluconazole and voriconazole. Ketoconazole and its derivatives increased the MIC_{50} value, but not significantly. In Gu5 strain, all test azoles had a marked increase in MIC_{50}. There was an increase in the expression of CDR1 and CDR2 genes which were widely resistant to antifungals [8].

The research was continued by synergizing KeP and KeOP with fluconazole to overcome resistance from Gu5 strain. The results are presented in the form of a graph can be seen in Fig. 2.

Based on the data in Table 4, the use of a combination of azole compounds has been shown to suppress the viability of Candida albicans. The addition and increase in the number of KeP and KeOP in the combination of fluconazole + ketoconazole derivatives was shown to decrease the MIC50 value of fluconazole compared to controls.

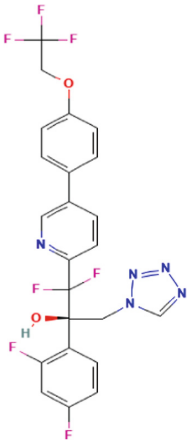

Fig. 2. Chemical structure of oteseconazole/VT-1161 (Adapted from https://pubchem.ncbi.nlm.nih.gov)

Table 4. *Candida albicans* Viability (%) against control (fluconazole), fluconazole + KeP and fluconazole + KeOP. **(*0.01 < P < 0.05; **0.001 < P < 0.01; ***P < 0.0001). ± SD, n = 3.)**

Compound		Viability (%)				
Flc (μM)		0	0.78	1.56	3.13	6.25
	Without KeOP or KeP	100 ± 4.48	86.94 ± 7.84	71.35 ± 13.94	73.85 ± 4.35	37.99 ± 8.94
KeOp (μM)	0.1	102.18 ± 2.22	84.62 ± 7.26	86.97 ± 12.74	61.89 ± 8.16	16.38 ± 3.65
	0.2	93.93 ± 7.22	62.2 ± 18.77	56.29 ± 13.73	55.87 ± 18.43	13.09 ± 6.22*
	0.39	91.2 ± 10.66	70.97 ± 12.08	17.89 ± 13.6*	0 ± 12.12**	0 ± 7.32*
KeP (μM)	0.39	114.94 ± 9.06	82.35 ± 4.89	70.35 ± 3.98	55.69 ± 4.27*	33.18 ± 8.69
	0.78	0.78	101.89 ± 1.98	71.98 ± 5.97	55.32 ± 10.76*	59.7 ± 3.19*
	1.56	1.56	96.76 ± 3.21	49.28 ± 14.1*	21.39 ± 13.95*	0 ± 8.36***

3.2 Oteseconazole/VT-1161

Oteseconazole/VT-1161 is a candidate for a new azole drug that belongs to the class of organic compounds known as phenylpyridines. It is a polycyclic aromatic compound containing a benzene ring linked to a pyridine ring via CC or CN bonds (go.drugbank.com). The structure of oteseconazole is shown in Fig. 2.

Oteseconazole/VT-1161 is a tetrazole designed to be effective against a range of fungal pathogens [10]. In-vitro test MIC_{50} of oteseconazole/VT-1161 was compared with 4 azole drugs already on the market. The four drugs are: fluconazole, itrazonazole, voriconazole and posaconazole. Candida strains were obtained from patient isolates [11] Complete data are presented in Table 5.

As many as 5 of 20 isolates (25%) of Candida albicans showed MIC_{50} of fluconazole that exceeded the CLSI susceptible limit. A total of 14 from the 31 candida isolates (45%) showed MIC_{50} of fluconazole 4 mg/L. Different results were shown by oteseconazole/VT-1161 where only 1 isolate showed MIC_{50} value 4 mg/L. Thereby, oteseconazole/VT-1161 had in vitro activity against candida by 97% (30 of 31 isolates).

There is a study comparing the potency of otoseconazole/VT-1161 to other azoles by comparing its inhibitory ability against CYP51 [1]. The enzyme/inhibitor/substrate molar ratio was 1:2:50, with a P450 concentration at 0.5 M (37 °C, 60 min reaction). The experiment was carried out in triplicate, and the results are presented as mean ± SD, shown in Fig. 3.

Fluconazole has an inhibitory value of 54%, the lowest compared to other azole drugs. Clotrimazole and miconazole had similar inhibitory abilities, namely 78% and 79%. Sequentially voriconazole, ketoconazole and itraconazole had inhibitory abilities of 84%, 85% and 91%. Posaconazole and the new drug candidate oteseconazole/VT-1161 had the greatest inhibition value of 98%.

In an in-vivo assay, administration of oteseconazole/VT-1161 in mice resulted in significant accumulation of mucosal drug and elimination of infections caused by fluconazole-susceptible and resistant Candida strains [12].

Oteseconazole has experienced a faster research development compared to Diphenylphosphane derivatives of ketoconazole. Oteseconazole/VT-1161 has been studied in treating Vulvovaginal candidiasis (VVC) and Recurrent VVC (VVC) and is potentially more efficacious than fluconazole [12]. In Table 6, it can be seen that fluconazole-resistant strains were sensitive to oteseconazole/vt-1161.

In 2021, the publication of a randomized phase 2 study of oteseconazle/VT-1161 for the treatment of acute vulvovaginal candidiasis showed that all treatment groups achieved cure on day 28 [13].

Table 5. MIC_{50} (mg/L) of Oteseconazole/VT-1161 compared with other azole drugs

Isolate	Species	MIC_{50} (mg/L)				
		Oteseconazole	Fluconazole	Itraconazole	Voriconazole	Posaconazole
Y31	*C.albicans*	0.12	32	0.5	0.5	0.5
Y37	*C.albicans*	0.03	64	0.12	0.12	0.06
Y42	*C.albicans*	0.03	<=0.12	0.03	<=0.008	<=0.008
Y43	*C.albicans*	0.03	0.25	0.06	<=0.008	0.015
Y46	*C.glabrata*	0.06	8	0.5	0.25	1
Y47	*C.albicans*	0.03	0,.25	<=0.015	<=0.008	<=0.008
Y48	*C.albicans*	0.03	0.5	0.06	<=0.008	0.015
Y49	*C.glabrata*	2	128	>16	2	>8
Y51	*C.albicans*	0.03	4	0.12	0.06	0.12
Y52	*C.glabrata*	1	256	>16	2	>8
Y54	*C.glabrata*	0.5	128	16	2	8
Y55	*C.albicans*	0.5	16	0.12	0.25	0.12
Y57	*C.albicans*	>16	4	0.06	0.015	0.06
Y72	*C.albicans*	0.03	0.5	0.06	0.015	0.03
Y73	*C.nivariensis*	0.06	16	0.5	0.25	1
Y75	*C.albicans*	0.03	1	0.06	<=0.008	0.06
Y79	*C.albicans*	0.03	0.5	0.06	0.015	0.03
Y82	*C.utilis*	0.06	4	0.25	0.12	0.25
Y83	*C.albicans*	0.03	2	0.12	0.06	0.06
Y84	*C.albicans*	0.03	0.5	0.06	0.015	0.015
Y87	*C.glabrata*	0.03	8	0.25	0.25	0.5
Y88	*C.albicans*	0.03	0.5	0.03	<=0.008	0.015
Y92	*C.dubliniensis*	0.03	0.25	0.06	<=0.008	0.06
Y93	*C.albicans*	0.03	0.5	0.06	0.008	0.03
Y107	*C.albicans*	0.06	0.5	0.06	0.015	0.015
Y111	*C.albicans*	0.03	0.5	0.03	0.008	0.015
Y121	*C.glabrata*	0.12	16	0.5	0.5	1
Y125	*C.krusei*	0.12	16	0.5	0.5	1
Y126	*C.parapsilosis*	0.03	0.25	0.06	<=0.008	0.03
Y152	*C.albicans*	0.03	<= 0.12	<= 0.015	<=0.008	<=0.008
Y160	*C.albicans*	0.03	1	0.03	0.03	0.015

Besides *C. albicans*, vt-1161 has also been shown to be effective against *Coccidioides* sp [14]; *Rhizopus arrhizus* var. *Arrhizus* [15]; *Candida glabrata* [16] [17]; *Candida krusei* [17]; *Cryptococcus* spp [18]; *T. rubrum* [19] [20]; *T. mentagrophytes* [20] and *Epidermophyton floccosum* [20].

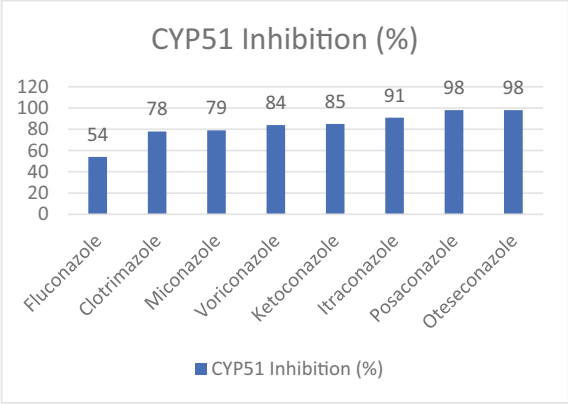

Fig. 3. Comparison of the inhibitory ability of azoles against CYP51

Table 6. MIC values of oteseconazole/VT-1161 and fluconazole MIC against clinical isolates from VVC/RVVC patient

Strain	MIC (μg/ml)	
	Oteseconazole	Fluconazole
3153A (lab strain)	≤0.015	0.25
MR700–13	≤0.015	0.25
SM692–08	≤0.015	2
CC330–10	≤0.015	4
BR160–09	≤0.015	8
JJ330–05	0.12	8
AF313–10	≤0.015	32
AR466–06	≤0.015	64
LP1158–07	2	64

4 Conclusions

Diphenylphosphane derivatives of ketoconazole and oteseconazole/VT-1161 showed excellent antifungal activity against *Candida albicans*. As drug candidates, both have the potential to be developed and researched further. Furthermore, oteseconazole is already in the clinical research phase. The availability of new, better azole compounds is one of the efforts to combat the incidence of *Candida albicans* resistance to existing azole compounds.

Acknowledgments. We grateful to The Faculty of Pharmacy at the University of Muhammadiyah Surakarta (UMS) for the research opportunity it has been given.

Authors' Contributions. Muhammad Rosyid As Sabiq, did data collection; data analysis and interpretation; drafting the article.

Peni Indrayudha did critical revision of the article; final approval of the version to be published.

References

1. Hargrove T.Y., Friggeri L., Wawrzak Z., Qi A., Hoekstra W.J., Schotzinger R.J., York J.D., Peter Guengerich F. and Lepesheva G.I., 2017, Structural analyses of Candida albicans sterol 14α-demethylase complexed with azole drugs address the molecular basis of azole-mediated inhibition of fungal sterol biosynthesis, *Journal of Biological Chemistry*, 292 (16), 6728–6743. DOI: https://doi.org/10.1074/2Fjbc.M117.778308
2. Binjubair F.A., Parker J.E., Warrilow A.G., Puri K., Braidley P.J., Tatar E., Kelly S.L., Kelly D.E. and Simons C., 2020, Small-Molecule Inhibitors Targeting Sterol 14α-Demethylase (CYP51): Synthesis, Molecular Modelling and Evaluation Against Candida albicans, *ChemMedChem*, 15 (14), 1294–1309. DOI: https://doi.org/10.1002/cmdc.202000250
3. Zhang J., Li L., Lv Q., Yan L., Wang Y. and Jiang Y., 2019, The fungal CYP51s: Their functions, structures, related drug resistance, and inhibitors, *Frontiers in Microbiology*, 10 (April). DOI: https://doi.org/10.3389/fmicb.2019.00691
4. Lamb D.C., Waterman M.R., Kelly S.L. and Guengerich F.P., 2007, Cytochromes P450 and drug discovery, *Current Opinion in Biotechnology*, 18 (6), 504–512.
5. Jordá T, Puig S. Regulation of Ergosterol Biosynthesis in Saccharomyces cerevisiae. Genes (Basel). 2020 Jul 15;11(7):795. Doi: https://doi.org/10.3390/genes11070795
6. Hargrove T.Y., Friggeri L., Wawrzak Z., Sivakumaran S., Yazlovitskaya E.M., Hiebert S.W., Guengerich F.P., Waterman M.R. and Lepesheva G.I., 2016, Human sterol 14α-demethylase as a target for anticancer chemotherapy: Towards structure-aided drug design, *Journal of LipiResearch*, 57 (8), 1552–1563. DOI: https://doi.org/10.1194/jlr.M069229
7. Warrilow A.G., Nishimoto A.T., Parker J.E., Price C.L., Flowers S.A., Kelly D.E., David Rogers P. and Kelly S.L., 2019, The evolution of Azole resistance in Candida albicans Sterol 14-demethylase (CYP51) through incremental amino acid substitutions, *Antimicrobial Agents and Chemotherapy*, 63 (5), 1–16. DOI: https://doi.org/10.1128/AAC.02586-18
8. de Almeida R.F.M., Santos F.C., Marycz K., Alicka M., Krasowska A., Suchodolski J., Panek J.J., Jezierska A. and Starosta R., 2019, New diphenylphosphane derivatives of ketoconazole are promising antifungal agents, *Scientific Reports*, 9 (1), 1–14. DOI: https://doi.org/10.1038/s41598-019-52525-7
9. Starosta R , de Almeida RFM , Puchalska M , Białońska A , Panek JJ , Jezierska A , Szmigiel I , Suchodolski J , Krasowska A . New anticandidal Cu(i) complexes with neocuproine and ketoconazole derived diphenyl(aminomethyl)phosphane: luminescence properties for detection in fungal cells. *Dalton Trans*. 2020 Jul 7;49(25):8528-8539. DOI: https://doi.org/10.1039/D0DT01162B
10. Monk BC, Keniya MV, Sabherwal M, Wilson RK, Graham DO, Hassan HF, Chen D, Tyndall JDA. 2019. Azole resistance reduces susceptibility to the tetrazole antifungal VT-1161. *Antimicrob Agents Chemother* 63:e02114-18. https://doi.org/10.1128/AAC.02114-18.
11. Break T.J., Desai J. V., Natarajan M., Ferre E.M.N., Henderson C., Zelazny A.M., Siebenlist U., Hoekstra W.J., Schotzinger R.J., Garvey E.P. and Lionakis M.S., 2018, VT-1161 protects mice against oropharyngeal candidiasis caused by fluconazole-susceptible and -resistant Candida albicans, *Journal of Antimicrobial Chemotherapy*, 73 (1), 151–155. DOI: https://doi.org/10.1093/jac/dkx352

12. Garvey E.P., Hoekstra W.J., Schotzinger R.J., Sobel J.D., Lilly E.A. and Fidel P.L., 2015, Efficacy of the clinical agent VT-1161 against fluconazole-sensitive and -resistant Candida albicans in a murine model of vaginal candidiasis, *Antimicrobial Agents and Chemotherapy*, 59 (9), 5567–5573. DOI: https://doi.org/10.1128/AAC.00185-15
13. Brand S.R., Sobel J.D., Nyirjesy P., Ghannoum M.A., Schotzinger R.J. and Degenhardt T.P., 2021, A Randomized Phase 2 Study of VT-1161 for the Treatment of Acute Vulvovaginal Candidiasis, *Clinical infectious diseases : an official publication of the Infectious Diseases Society of America*, 73 (7), e1518–e1524. DOI: https://doi.org/10.1093/cid/ciaa1204
14. Shubitz LF, Trinh HT, Galgiani JN, Lewis ML, Fothergill AW, Wiederhold NP, Barker BM, Lewis ERG, Doyle AL, Hoekstra WJ, Schotzinger RJ, Garvey EP. 2015. Evaluation of VT-1161 for treatment of coccidioidomycosis in murine infection models. *Antimicrob Agents Chemother* 59:7249 –7254. DOI: https://doi.org/10.1128/AAC.00593-15
15. Gebremariam T, Wiederhold NP, Fothergill AW, Garvey EP, Hoekstra WJ, Schotzinger RJ, Patterson TF, Filler SG, Ibrahim AS. 2015. VT-1161 protects immunosuppressed mice from *Rhizopus arrhizus* var. *arrhizus* infection. *Antimicrob Agents Chemother* 59:7815–7817. https://doi.org/10.1128/AAC.01437-15.
16. Nishimoto AT, Whaley SG, Wiederhold NP, Zhang Q, Yates CM, Hoekstra WJ, Schotzinger RJ, Garvey EP, Rogers PD. Impact of the Major Candida glabrata Triazole Resistance Determinants on the Activity of the Novel Investigational Tetrazoles VT-1598 and VT-1161. *Antimicrob Agents Chemother*. 2019 Sep 23;63(10):e01304-19. DOI: https://doi.org/10.1128/AAC.01304-19.
17. Schell WA, Jones AM, Garvey EP, Hoekstra WJ, Schotzinger RJ, Alexander BD. Fungal CYP51 Inhibitors VT-1161 and VT-1129 Exhibit Strong *In Vitro* Activity against Candida glabrata and C. krusei Isolates Clinically Resistant to Azole and Echinocandin Antifungal Compounds. *Antimicrob Agents Chemother.* 2017 Feb 23;61(3):e01817–16. DOI: https://doi.org/10.1128/AAC.01817-16
18. Wang L, Zhang M, Guo J, Guo W, Zhong N, Shen H, Cai J, Zhu Z, Wu W. In vitro activities of the tetrazole VT-1161 compared with itraconazole and fluconazole against Cryptococcus and non-albicans *Candida* species. *Mycologia.* 2021 Sep-Oct;113(5):918–925. DOI: https://doi.org/10.1080/00275514.2021.1913949.
19. Warrilow AGS, Parker JE, Price CL, Garvey EP, Hoekstra WJ, Schotzinger RJ, Wiederhold NP, Nes WD, Kelly DE, Kelly SL. The Tetrazole VT-1161 Is a Potent Inhibitor of Trichophyton rubrum through Its Inhibition of T. rubrum CYP51. *Antimicrob Agents Chemother*. 2017 Jun 27;61(7):e00333–17. DOI: https://doi.org/10.1128/AAC.00333-17.
20. Garvey EP, Hoekstra WJ, Moore WR, Schotzinger RJ, Long L, Ghannoum MA. VT-1161 dosed once daily or once weekly exhibits potent efficacy in treatment of dermatophytosis in a guinea pig model. *Antimicrob Agents Chemother*. 2015 Apr;59(4):1992-7. DOI: https://doi.org/10.1128/AAC.04902-14.