Potensi senyawa(1)-*N*-(3,4-dimetoksi benzoil)-1,10-fenantrolin-bromida dan senyawa (1)-*N*-(1-metoksibenzoil)-1,10-fenantrolin-bromida sebagai antijamur: kajian aktivitas dan toksisitas *in vitro* dan kemungkinan mekanisme aksinya

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Intisari

Latar Belakang. *Candida* sp adalah flora normal dijumpai dalam organ manusia. Namun *Candida* sp dapat bersifat patogen karena kemampuannya menginfeksi sel inang dan mengakibatkan penyakit apabila kondisi sistem imun inang lemah**.** Senyawa turunan 1.10 fenantrolin merupakan kelator logam yang memiliki efek antijamur. Pada studi pendahuluan senyawa (1)-*N*-2 metoksibenzil-1.10 fenantrolin bromide dan senyawa (1)-*N*-(1-metoksibenzoil)-1,10-fenantrolin bromida menghambat pertumbuhan jamur *C. albicans.*

Tujuan. Mengkaji aktivitas antijamur *in vitro* senyawa 1, senyawa 2 dan flukonazol terhadap *C. albicans* . Mengkaji toksisitas *in vitro* nya pada kultur sel normal selain itu juga mengkaji pengaruhnya terhadap penghambatan pembentukan dan pengurangan pembentukan biofilm *C. albicans* dan mengkaji integritas membran sel *C. albicans*.

Hasil Penelitian. Penelitian KHM senyawa 1 dan senyawa 2 terhadap *C.albicans* ATCC 10231, isolat klinik *Candida* 2, *Candida* 20 antara 0,78-3.13 µg/mL. KHM flukonazol antara 0,25-0,50 µg/mL. Penelitian KBM senyawa 1, 2 dan flukonazol terhadap *C. albicans* ATCC 10231 berturut-turut adalah 100 µg/mL, 100 µg/mL dan >64 µg/mL. Senyawa 1, 2 dan flukonazol terhadap *Candida* 2 adalah >100 µg/mL >100 µg/mL dan >64 µg/mL. Senyawa 1, 2 dan flukonazol terhadap *Candida* 20 adalah >64 µg/mL, >64 µg/mL dan >64 µg/mL. Penelitian Inhibitory Concentration $_{50}$ (IC 50) senyawa 1,2 pada *Candida* 2 adalah 56,17 ± 21,59 µg/mL; 84,93 ± 21,09 µg/mL dan flukonazol > 1000 µg/mL.

IC_{50} senyawa 1 dan 2 pada *Candida* 20 adalah 56,17 ± 21,59 µg/mL ; 84,93 ± 21,09 µg/mL dan flukonazol > 1000 µg/mL. IC $_{50}$ senyawa 1, 2 pada C. *albicans* adalah 84,93 ± 21,09 µg/mL 56,17 ± 21,59 µg/mL dan flukonazol > 1000 µg/mL. Indek sitotoksik (IS) senyawa 1, 2 dan flukonazol pada *Candida* 2 berturut turut 17,9 ; 27,1 dan > 4000. IS senyawa 1, 2 dan flukonazol pada

Candida 20 dan flukonazol berturut turut 54,44 ; 36,0 dan > 2000. IS senyawa 1, 2 dan flukonazol pada *C. albicans* berturut turut 54,44 ; 36,0 dan > 2000.

Penelitian Kadar Hambat Minimal (KHBM) Nilai $KHBM_{50}$ *C. albicans, Candida* 2 dan *Candida* 20 adalah 0,41;0,72 dan 0,48 µg/mL. Nilai $KHBM_{80}$ senyawa 1 pada *C. albicans,* Candida 2 dan *Candida* 20 adalah 0,21 ; < 0,04 dan < 0,04 µg/mL. Nilai $KHBM_{50}$ senyawa 2 pada *C. albicans, Candida 2 dan Candida 20* adalah 1,53 ; 1,21 dan 0,82 µg/mL. Nilai $KHBM_{80}$ senyawa 2 pada *C. albicans, Candida* 2 dan *Candida* 20 adalah < 0,80 0,29; 0,39 µg/mL. Nilai $KHBM_{50}$ flukonazol pada *C. albicans, Candida 2* dan *Candida 20* antara 0,53 ; 1,46 dan 0,83 µg/mL. Nilai $KHBM_{80}$ flukonazol pada *C. albicans, Candida* 2 dan *Candida* 20 adalah 0,18 ; 0,40 dan 0,30 µg/mL.

Penelitian Kadar Reduksi Biofilm Minimal (KRBM). Nilai $KRBM_{50}$ senyawa 1 pada *C. albicans, Candida* 2 dan *Candida* 20 berkisar antara 1,59 ; 3,67 dan 2,48 µg/mL. $KRBM_{80}$ senyawa 1 pada *C. albicans, Candida* 2 dan *Candida* 20 adalah 0,71 ; 1,79 dan 1,93 µg/mL.

$KRBM_{50}$ senyawa 2 pada *C. albicans, Candida* 2 dan *Candida* 20 adalah 1,08 – 3,89 dan 1,24 µg/mL. $KRBM_{80}$ senyawa 2 pada *C. albicans, Candida* 2 dan *Candida* 20 adalah 0,47 ; 2,31 dan 0,69 µg/mL.

$KRBM_{50}$ flukonazol pada *C. albicans, Candida* 2 *Candida* 20 adalah 0,60 ; 2,58 dan 1,09 µg/mL. $KHBM_{80}$ flukonazol pada ketiga isolat *C. albicans* antara 0,25 ; 0,74 dan 1,09 µg/mL.

Penelitian integritas membran sel *Candida* 2, peningkatan dosis senyawa 1 dan 2 yaitu 2 x KHM, 4 x KHM dan 8 x KHM sebagai berikut: senyawa 1 dengan nilai berturut-turut 46,50 ± 1,57 µg/mL ; 50,00 ± 3,01 µg/mL ; 69,62 ± 1,12 µg/mL. Sementara senyawa 2 memiliki nilai berturut-turut adalah 58,71 ± 0,71 µg/mL; 67,77 ± 0,35 µg/mL; 63,34 ± 9,43 µg/mL. Pada flukonazol dengan konsentrasi 8 x Kadar Hambat Minimal (KHM) adalah 51,47 ± 1,29 µg/mL.

Pada *Candida* 20, peningkatan dosis senyawa 1 dan 2 (2 x Kadar Hambat Minimal (KHM), 4 x Kadar Hambat Minimal (KHM) dan 8 x Kadar Hambat Minimal (KHM) sebagai berikut senyawa 1 dengan nilai berturut-turut 9,77 ± 0,27 µg/mL; 19,91 ± 0,21 µg/mL; dan 21,60 ± 0,65 µg/mL.

Sementara senyawa 2 memiliki nilai berturut-turut 20,95 ± 0,45 µg/mL; 32,39 ± 0,66 µg/mL dan 27,55 ± 0,25 µg/mL. Pada flukonazol dengan konsentrasi 8x Kadar Hambat Minimal (KHM) adalah 27,36 ± 0,78 µg/mL.

Pada *Candida albicans* ATCC 10231 peningkatan dosis senyawa 1 dan 2 (2 x Kadar Hambat Minimal (KHM), 4 x Kadar Hambat Minimal (KHM) dan 8 x Kadar Hambat Minimal (KHM): senyawa 1 dengan nilai berturut-turut 13,08 ± 0,62 µg/mL; 42,03 ± 0,91 µg/mL dan 31,16 ± 0,36 µg/mL. Sementara senyawa 2 memiliki nilai berturut-turut 26,91 ± 0,44 µg/mL; 20,63 ± 0,15 µg/mL dan 65,94

± 1,20 µg/mL. Pada flukonazol dengan konsentrasi 8 x Kadar Hambat Minimal (KHM) adalah 28,02 ± 0,30 µg/mL. Kelipatan dosis senyawa 1 dan 2 tidak selalu berefek meningkatkan kerusakan membran sel *C. albicans* ATCC 10231, isolat klinik *Candida* 2 *dan Candida* 20.

Senyawa 1, senyawa 2 dan flukonazol menghambat dan membunuh *C. albicans* ATCC 10231 isolat klinik *Candida* 2 dan *Candida* 20.

Senyawa 1, senyawa 2 dan flukonazol menghambat pertumbuhan sel Vero sampai 50 % terhadap *C. albicans* ATCC 10231, isolat klinik *Candida* 2 dan *Candida* 20.

Senyawa 1, senyawa 2 dan flukonazol aman terhadap *C. albicans* ATCC 10231, isolat klinik *Candida* 2 dan *Candida* 20, terlihat pada nilai indek sitotoksiknya.

Senyawa 1, senyawa 2 dan flukonazol menghambat dan mengurangi pertumbuhan *C. albicans* ATCC 10231, isolat klinik *Candida* 2 dan *Candida* 20.

Kenaikan dosis senyawa 1 dan senyawa 2 tidak selalu meningkatkan kerusakan dinding sel *C. albicans* ATCC 10231, isolat klinik *Candida* 2 dan *Candida* 20.

Kata kunci: senyawa (1)-*N*-(3,4-dimetoksibenzoil)-1,10-fenantrolin-bromida dan senyawa (1)-*N*-(1-metoksibenzoil)-1,10-fenantrolin-bromida, flukonazol, KHM, KBM, sitotoksisitas, KHBM, KRBM, integritas membran.

Potential action of (1)-N-(3,4-dimethoxy benzyl -1.10 phenantroline bromide (1)-N-(1-methoxy–3 methoxy benzyl-1.10 phenantroline bromide compounds as antifungal agent: Studies on activity and *in vitro* toxicity and the possibility of its mechanism of action

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ABSTRACT

Background. The 1.10 phenantroline derivative compound is a metal-based drugs possessing effects as an anti plasmodium, anti-cancer and anti-fungal. In a preliminary study of the compound (1)-N-2-methoxy-benzyl 1.10 phenantroline bromide proven to inhibit the growth of fungi Candida spp.

Aim. Reviewing invitro activity of the anti-fungal, the in vitro toxicity in normal cell culture, anti-fungal activity through its influence on the formation of biofilm and membrane integrity of the (1)-N-(3,4-dimethoxy benzyl -1.10fenantrolin bromide [1st compound], (1)-N-(1-methoxy –3methoxy benzyl -1.10 fenantrolin bromide [2nd compound] and fluconazole against Candida albicans ATCC 10231, clinical isolates of Candida 2 and 20.

Results. MIC values of the 1st and 2nd compounds towards Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20 were between 0,78-3.13 µg/mL, while the MIC values of fluconazole were between 0,25-0,50 µg/mL. The research on MBC values of 1st and 2nd compounds and fluconazole towards Candida albicans ATCC 10231 were 100 µg/mL, 100 µg/mL and >64 µg/mL, respectively. As the MBC values of 1st and 2nd compounds and fluconazole towards Candida 2 were >100 µg/mL >100 µg/mL and >64 µg/mL, consecutively; while the MBC values of 1st and 2nd compounds and fluconazole towards Candida 20 were >64 µg/mL, >64 µg/mL and >64 µg/mL, respectively . The research on Inhibitory Concentration$_{50}$ (IC_{50}) values of 1st and 2nd compounds for Candida 2 were 56,17 ± 21,59 µg/mL; 84,93 ± 21,09 µg/mL as

fluconazole was >1000 µg/mL. IC_{50} values on 1st and 2nd compounds for Candida 20 were 56,17 ± 21,59 µg/mL; 84,93 ± 21,09 µg/mL as fluconazole were >1000 µg/mL, while the IC_{50} values for Candida albicans were 84,93 ± 21,09 µg/mL; 56,17 ± 21,59 µg/mL and fluconazole was >1000 µg/mL.

Index Cytotoxic (IC) of the 1st and 2nd compounds, and fluconazole towards Candida 2 consecutively were 17,9; 27,1 and > 4000. The IC values towards Candida 20, respectively were 54,44; 36,0 and > 2000, while IC values towards Candida albicans were 54,44; 36,0 and > 2000.

The results of $MBIC_{50}$ values 1st compound for Candida albicans, Candida 2 and Candida 20 were 0,41;0,72 and 0,48 µg/mL. The $MBIC_{80}$ values of 1st compound for Candida albicans, Candida 2 and Candida 20 were 0,21; < 0,04 and < 0,04 µg/mL. The $MBIC_{50}$ of 2nd compound for Candida albicans, Candida *2* and Candida 20 were 1,53; 1,21 and 0,82 µg/mL. The $MBIC_{80}$ 2nd compound for Candida albicans, Candida 2 and Candida 20 were < 0,80; 0,29; and 0,39 µg/mL. The $MBIC_{50}$ fluconazole for Candida albicans, Candida 2 and Candida 20 were between 0,53; 1,46 and 0,83 µg/mL. $MBIC_{80}$ values for fluconazole for Candida albicans, Candida 2 and Candida 20 were 0,18; 0,40 and 0,30 µg/mL.

The $MBRC_{50}$ values of 1st compound for Candida albicans, Candida 2 and Candida 20 ranged between 1,59; 3,67 and 2,48 µg/mL. $MBRC_{80}$ values of 1st compound for Candida albicans, Candida 2 and Candida 20 were 0,71; 1,79 and 1,93 µg/mL.

$MBRC_{50}$ of the 2nd compound for Candida albicans, Candida 2 and Candida 20 were 1,08 – 3,89 and 1,24 µg/mL. $MBRC_{80}$ of the 2nd compound for Candida albicans, Candida 2 and Candida 20 were 0,47; 2,31 and 0,69 µg/mL.

$MBRC_{50}$ of the fluconazole for Candida albicans, Candida 2 and Candida 20 were 0,60; 2,58 and 1,09 µg/mL. $MBRC_{80}$ fluconazolefor the three isolates Candida albicans ranged between 0,25; 0,74 and 1,09 µg/mL.

The research on Candida 2 membrane cells integrity; the dose multiplication for 1st and 2nd compound were 2 x MIC, 4 x MIC and 8 x MIC are as follows: 1st compound are 46,50 ± 1,57 µg/mL ; 50,00 ± 3,01 µg/mL ; 69,62 ± 1,12 µg/mL, respectively. As for the 2nd compound were 58,71 ± 0,71 µg/mL; 67,77 ± 0,35 µg/mL; 63,34 ± 9,43 µg/mL. For fluconazole with 8 x MIC were 51,47 ± 1,29 µg/mL.

On Candida 20, the dose multiplication for 1st and 2nd compound were 2 x MIC, 4 x MIC and 8 x MIC as follows: 9,77 ± 0,27 µg/mL; 19,91 ± 0,21 µg/mL; dan 21,60 ± 0,65 µg/mL.

As for the 2nd compound has consecutive values as follows 20,95 ± 0,45 µg/mL; 32,39 ± 0,66 µg/mL dan 27,55 ± 0,25 µg/mL. For fluconazole with 8 x MIC were 27,36 ± 0,78 µg/mL.

On Candida albicans ATCC 10231 For fluconazole with 8 x MIC were as follows: 1st compound with consecutive values 13,08 ± 0,62 µg/mL; 42,03 ± 0,91 µg/mL and 31,16 ± 0,36 µg/mL. As for 2nd compound were 26,91 ± 0,44 µ; 20,63 ± 0,15 µg/mL and 65,94 ± 1,20 µg/mL, respectively. For fluconazole with 8 x MIC were 28,02 ± 0,30 µg/mL. The multiplication dose of the 1st and the 2nd compounds does not constantly increasing the damaging effect on Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20 membrane cells integrity.

The 1st and the 2nd compounds, as well as fluconazole inhibits and killed Candida albicans ATCC 10231, clinical isolates Candida 2 and Candida 20.

The 1st and the 2nd compounds, as well as fluconazole inhibits the growth of vero cell until 50 % towards Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20.

The 1st and the 2nd compounds, as well as fluconazole were considered safe towards Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20 as seen on the Cytotoxic Index.

The increasing doses of the 1st and the 2nd compounds, as well as fluconazole inhibits the growth of vero cell until 50% towards Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20.

The multiplication dose of the 1st and the 2nd compounds does not constantly increasing the damaging effect on Candida albicans ATCC 10231, clinical isolates of Candida 2 and Candida 20 membrane cells integrity.

Keywords: (1)-*N*-(3,4-dimetoksibenzoil)-1,10-phenantroline-bromide, (1)-*N*-(1-metoksibenzoil)-1,10-phenantroline-bromide, fluconazole, MIC, MBC, cytotoxicity, MBIC**,** MBRC**,** membrane integrity