

**Rekap Korespondensi dengan Journal of The Egyptian Women's Dermatologic Society (JEWDS)**

<b>No</b>	<b>Tanggal</b>	<b>Uraian</b>	<b>Halaman</b>	<b>Ket</b>
1	10 December 2020	Submit Paper to JEWDS	<b>Page 1</b>	
2	24 December 2020	Submitted paper has been assigned the following manuscript number: JEWDS-D-20-00061	<b>Page 2</b>	
3	13 February 2021	The suggestions of the reviewers (to revision submitted paper)	<b>Page 3-5</b>	
4	11 April 2021	Revised Manuscript #JEWDS-D-20-00061 <b>R1</b> has been submitted to JEWDS	<b>Page 6</b>	
5	25 May 2021	The suggestions of the reviewers (2) : substantial revisions	<b>Page 7-9</b>	
6	4 July 2021	Revised Manuscript #JEWDS-D-20-00061 <b>R2</b> has been submitted to JEWDS	<b>Page 10</b>	
7	16 August 2021	Minor revision about point-by-point response to the comments of the reviewers (3)	<b>Page 11-12</b>	
8	20 August 2021	Revised Manuscript #JEWDS-D-20-00061 <b>R3</b> has been submitted to JEWDS	<b>Page 13</b>	
9	21 August 2021	Minor revision about point-by-point response to the comments of the reviewers (4)	<b>Page 14-15</b>	
10	23 August 2021	Paper has been accepted	<b>Page 16</b>	
11	2 January 2022	Paper published online (JEWDS Vol 19 Issue 1, January-April 2022)	<b>Page 17-23</b>	

# JEWDS Submission Confirmation for Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Kamis, 10 Desember 2020 18.12 WIB

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Dec 10, 2020

Dear dr. Murtiastutik,

Your submission entitled "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients" has been received by the journal editorial office.

You will be able to check on the progress of your paper by logging on to Editorial Manager as an author.

<https://www.editorialmanager.com/jewds/>

Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=25300&l=PVCPPBPJ>

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Thank you for submitting your work to this journal.

Kind Regards,

Journal of Egyptian Women's Dermatologic Society

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**PAGE 1**

## A manuscript number has been assigned to your JEWDS submission

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Kamis, 24 Desember 2020 03.32 WIB

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TODAYS\_DATE%

Dear dr. Murtiastutik,

Your submission entitled "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients" has been assigned the following manuscript number: JEWDS-D-20-00061.

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Thank you for submitting your work to Journal of Egyptian Women's Dermatologic Society.

Kind Regards,

Nayera Mofteh, MD

Editor-in-Chief

Journal of Egyptian Women's Dermatologic Society

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## Your Submission to Journal of Egyptian Women's Dermatologic Society

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Sabtu, 13 Februari 2021 12.58 WIB

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Feb 13, 2021

RE: JEWDS-D-20-00061, entitled "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients"

Dear dr. Murtiastutik,

The editorial and peer review of the above-referenced manuscript have been completed. While the reviewers believe that your studies are of potential interest to our readers, substantial revisions are necessary before the paper can once again be considered for publication in the Journal of Egyptian Women's Dermatologic Society.

Should you choose to revise the manuscript, please be sure to take into careful consideration the suggestions of the reviewers, provided below. I will send the revised paper to the original reviewers for further appraisal upon receipt.

Please include with your revised submission an itemized, point-by-point response to the reviewers which details the changes made. The revised manuscript should be submitted by May 14, 2021 to avoid being considered as a new submission.

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Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=25763&l=DRO3LPY8>

Please feel free to contact the Journal Office with any questions or concerns. We look forward to receiving the revised manuscript.

With Kind Regards,

Dr Nermeen Abdel Fattah  
Associate Editor  
Journal of Egyptian Women's Dermatologic Society

Reviewer Comments:

Reviewer #1: Thanks for giving me the opportunity to revise manuscript subjected for publication in JEWDS.

Some points have to be addressed.

- Proper rephrasing of the results in the abstract. Data written is not clear.
- Abbreviation of "C" has to be written as candida at first before the abbreviation to be clear.
- type of the study, methods of patients sampling and sample size have to be added in the methodology.
- did the patients have other immunosuppressive diseases or on immunosuppressive therapy or prolonged use of antibiotics? they have to be added in the exclusion criteria.
- What is the importance of taking the patient education level & occupation written in the results ? I think these data are out of the point of the aim of the study

- the chief complaint was white patches in 100 patients. what about the remaining 14 patients ? Data from all patients is better to be written in the results, not just in the tables.
- 20 isolates out of 30 patients is about 53%. Authors wrote it as 44%. Revision is needed or proper rephrasing to avoid this confusion.
- The remaining of the isolates were candida albicans or no growth ? Details have to be written in the results of each group of the three groups of CD4 counts.
- Results written in the discussion are better to be written in the results section not in the discussion only to be clear to the reader and researcher.

Reviewer #2: Dear Author,

Thank you for the article entitled "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients".

- \* The exclusion/inclusion criteria are not clear enough. Were patients under antibiotic therapy included? Females with contraceptive use? Diabetics or prediabetics? Patients with dental caries and/or plaque?
- \* It would be beneficial to divide the patients into those had ARV and those who do not take, to compare the study parameters in patients who took ARV versus those who did not take.
- \* Statistical analysis methodology is deficient and lacks p value specifications.
- \* The authors excluded patients with no growth of fungal colonies, however, there was no clear information about no growth in study population.
- \* In page 6 the details of isolated Candida non albicans are mentioned in correlation with lymphocyte count but no details about isolated Candida albicans.
- \* The number of isolated colonies are confusing and not arranged in ascending or descending manner. In addition, they are not correlated with lymphocyte count.
- \* CD4+ count is not correlated with the extent/number of lesions.
- \* I assumed that since Candida non albicans were isolated in 45 specimens, Candida albicans were isolated in 114-45, thus, 69. However, the total number of C. albicans isolates are 104. Does this mean that in some samples both Candida albicans and non albicans were isolated? It worth study this population and highlight possible underlying factors if any.
- \* It is well-known that CD4+ lymphocyte count of 200 cell/ $\mu$ l is considered a cut off value for opportunistic infections, so what is the purpose of comparing those with CD4+ lymphocyte count of >100 cell/ $\mu$ l and between 101-200 cell/ $\mu$ l?
- \* The types of isolates were not correlated with the clinical symptoms.
- \* In page 9, it was mentioned that "Candida non-albicans began to be identified. This study found C. glabrata in 12 (8.1%), C. tropicalis in six (4.05%), C. krusei in 22 (14.85%), C. dubliniensis in two (1.3%), C. lypolitica in one (0.7%), and C. parapsilosis in two (1.3%) of isolates". This information is not supported in the results section.
- \* Table 2, Location section is confusing, what is "combination"? what is "tongue combination"? What is the difference between "Tongue, mucosa, combination" and "Tongue / mucosa"?
- \* The quality of the photos needs improvement.
- \* Figure 1, the identified colonies particular species are not mentioned neither in figure nor in legend.
- \* References need to be updated. Suggested references to be added are:

Arch Oral Biol . 2020 Jul;115:104741.

doi: 10.1016/j.archoralbio.2020.104741. Epub 2020 May 7.

Dynamic study of oral Candida infection and immune status in HIV infected patients during HAART

Xian Du 1, Hui Xiong 2, Yao Yang 3, Jihui Yan 4, Shuqiong Zhu 4, Fangchun Chen 5

Pan Afr Med J. 2020 May 19;36:23.

doi: 10.11604/pamj.2020.36.23.18202. eCollection 2020.

The prevalence, risk factors and antifungal sensitivity pattern of oral candidiasis in HIV/AIDS patients in Kumba District Hospital, South West Region, Cameroon

Ngwa Fabrice Ambe 1, Njunda Anna Longdoh 1, Patience Tebid 1, Tanyi Pride Bobga 1, Claude Ngwayu Nkfusai 2 3, Sangwe Bertrand Ngwa 1, Frankline Sanyuy Nsai 2 3, Samuel Nambile Cumber 4 5 6

\* After addressing all the above-mentioned points, discussion section needs to be revised.

Much obliged

Complete Revision Instructions:

PRELIMINARY STEPS:

**PAGE 4**

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Revised Manuscript # JEWDS-D-20-00061R1 has been submitted to JEWDS

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Minggu, 11 April 2021 01.20 WIB

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Apr 10, 2021

Dear DR. dr. Murtiastutik,

The Journal of Egyptian Women's Dermatologic Society has received your revised submission, JEWDS-D-20-00061R1, entitled, "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients."

You may check the status of your manuscript at any time by logging in to Editorial Manager as an author.

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Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=26529&l=RUCSVLTN>

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Kind Regards,

Journal of Egyptian Women's Dermatologic Society

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## Your Submission to Journal of Egyptian Women's Dermatologic Society

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Selasa, 25 Mei 2021 04.20 WIB

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May 24, 2021

RE: JEWDS-D-20-00061R1, entitled "Association between etiologic species of oral candidiasis and CD4 T lymphocyte count in HIV / AIDS patients"

Dear DR. dr. Murtiastutik,

The editorial and peer review of the above-referenced manuscript have been completed. While the reviewers believe that your studies are of potential interest to our readers, substantial revisions are necessary before the paper can once again be considered for publication in the Journal of Egyptian Women's Dermatologic Society.

Should you choose to revise the manuscript, please be sure to take into careful consideration the suggestions of the reviewers, provided below. I will send the revised paper to the original reviewers for further appraisal upon receipt.

Please include with your revised submission an itemized, point-by-point response to the reviewers which details the changes made. The revised manuscript should be submitted by Aug 22, 2021 to avoid being considered as a new submission.

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Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=26961&l=WBILTEXB>

Please feel free to contact the Journal Office with any questions or concerns. We look forward to receiving the revised manuscript.

With Kind Regards,

Dr Nermeen Abdel Fattah  
Associate Editor  
Journal of Egyptian Women's Dermatologic Society

Reviewer Comments:

Reviewer #1: Thanks for giving me the opportunity to revise manuscript subjected to publication in JEWDS. Most of the previous comments were corrected but there are some other comments that have to be considered.

Authors mentioned in the patients and method section "Patients receiving oral antifungals within the past 3 months were included. Then they wrote patients receiving oral antifungals within two weeks from the study were excluded. Proper rephrasing is needed.

Authors included patients received or not antiretroviral drugs for HIV. Better results will be provided if patients who received these drugs were excluded. to overcome this, authors have to divide results into two groups accordingly.



what other the possible suggestions for isolation of. C. albicans only, C non albicans only and both types in different groups of patients with different CD4 counts?

Revision is needed regarding the number of patients with pseudomembranous candidiasis "103" and those written in table 6 "104".

Limitations of the study have to be added at the end of the discussion.

Reviewer #2: Dear Author,

Although you addressed most of the comments, I still have the following comments to consider:

- \* Please divide the patients into those had ARV and those who did not take, to compare the study parameters in patients who took ARV versus those who did not take, since ARV is not an exclusion criterion.
  - \* Please correlate CD4 lymphocyte count with the number of isolated colonies as well as the extent/number of lesions.
  - \* Please compare the patients with isolated only C. albicans to those with isolated only C. non albicans and those with isolated both C. albicans and non-albicans and highlight details and possible underlying factors for isolation of both types.
  - \* In page 7: "In this study, all subjects grew fungal colonies in culture and one study subject may grow more than one Candida species". How come only one subject grew more than one species? It has been mentioned that 30 patients grew more than one type of colony. "In CD4 T lymphocyte count of 1-100, C. albicans and C. non-albicans grew in 38 (25.5%) and 20 (13.4%) isolates respectively". I assumed that since the total number of patients in this group is 38, that 28 patients of them grew only C. albicans, while 20 patients grew both C. albicans and non albicans. However, it was mentioned that 15 subjects with CD4 lymphocyte count of 1-100, grew both colonies. Numbers needs revision.
  - \* Please add study limitations and recommendations at the end of discussion section.
  - \* The numbers mentioned in Table 6 for the clinical types are confusing and not matching with the study population or the number of isolates, either, with total of 167?!
  - \* Figure 2 A is not showing obvious pseudomembranous candidiasis. It seems coated tongue, recommended to be changed.
- Thank you

Complete Revision Instructions:

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11. If you find problems with the manuscript, please click "Edit Submission" from the Action menu. Make the appropriate changes, beginning again at step 3.
12. If you find no problems with the manuscript, please click "Approve Submission" from the Action menu. Your re-submission is now complete!

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Revised Manuscript # JEWDS-D-20-00061R2 has been submitted to JEWDS

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Minggu, 4 Juli 2021 15.57 WIB

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Jul 04, 2021

Dear DR. dr. Murtiastutik,

The Journal of Egyptian Women's Dermatologic Society has received your revised submission, JEWDS-D-20-00061R2, entitled, "Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV / AIDS patients."

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<https://www.editorialmanager.com/jewds/l.asp?i=27335&l=HB2YCRSG>

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Kind Regards,

Journal of Egyptian Women's Dermatologic Society

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# Journal of The Egyptian Women's Dermatologic Society Decision

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Senin, 16 Agustus 2021 06.22 WIB

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Aug 15, 2021

RE: JEWDS-D-20-00061R2, entitled "Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV / AIDS patients"

Dear DR. dr. Murtiastutik,

I am pleased to inform you that your paper has been found acceptable for publication pending minor revision. I anticipate that you will easily be able to answer the criticisms of the reviewers in a satisfactory manner. I will verify that this has been done upon receipt of the revised manuscript. Please find the comments of the reviewers listed below.

Please include with your revised submission an itemized, point-by-point response to the comments of the reviewers. The revisions should be completed by Nov 13, 2021 to avoid being considered as a new submission.

To submit a revision, go to <https://www.editorialmanager.com/jewds/> and log in as an Author. You will see a menu item called "Submission Needing Revision." Please click on this item to obtain your submission record and begin the revision process.

Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=27512&l=2WE86TF4>

With Kind Regards,

Dr Nermeen Abdel Fattah  
Associate Editor  
Journal of Egyptian Women's Dermatologic Society

Reviewer Comments:

Reviewer #1: Thanks for revising the manuscript, correcting the mistakes and did the corrections point by point.

Few corrections are needed.

Conclusion in the abstract:

authors wrote "Candida species was significantly associated with CD4 count"

This needs correction and proper rephrasing to conclude that candida albicans were detected more in patients with higher CD4 count compared to non candida species detected in patients with lower CD4 count, and then to write the clinical features of OC between candida, non candida species or mixed types to fulfill the aim of your study as the sentence wrote in the discussion page 15, lines 176-180.

Abbreviation of ARV not mentioned in the abstract.

Material & methods:

No need to write the equation used for sample size.

What is the importance of writing the education level of the patients?

Table 3 is much better with clear data.

Complete Revision Instructions:

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RESUBMISSION STEPS:

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11. If you find problems with the manuscript, please click "Edit Submission" from the Action menu. Make the appropriate changes, beginning again at step 3.
12. If you find no problems with the manuscript, please click "Approve Submission" from the Action menu. Your re-submission is now complete!

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Revised Manuscript # JEWDS-D-20-00061R3 has been submitted to JEWDS

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Jumat, 20 Agustus 2021 03.05 WIB

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Aug 19, 2021

Dear DR. dr. Murtiastutik,

The Journal of Egyptian Women's Dermatologic Society has received your revised submission, JEWDS-D-20-00061R3, entitled, "Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV / AIDS patients."

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You will notified of the editorial decision regarding your revised manuscript once it has been received.

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# Journal of The Egyptian Women's Dermatologic Society Decision

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Dari: Journal of The Egyptian Women's Dermatologic Society (em@editorialmanager.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Sabtu, 21 Agustus 2021 04.57 WIB

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Aug 20, 2021

RE: JEWDS-D-20-00061R3, entitled "Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV / AIDS patients"

Dear DR. dr. Murtiastutik,

I am pleased to inform you that your paper has been found acceptable for publication pending minor revision. I anticipate that you will easily be able to answer the criticisms of the reviewers in a satisfactory manner. I will verify that this has been done upon receipt of the revised manuscript. Please find the comments of the reviewers listed below.

Please include with your revised submission an itemized, point-by-point response to the comments of the reviewers. The revisions should be completed by Nov 18, 2021 to avoid being considered as a new submission.

To submit a revision, go to <https://www.editorialmanager.com/jewds/> and log in as an Author. You will see a menu item called "Submission Needing Revision." Please click on this item to obtain your submission record and begin the revision process.

Your username is: dwimurtiastutik

<https://www.editorialmanager.com/jewds/l.asp?i=27578&l=VGEH282S>

With Kind Regards,

Dr Nermeen Abdel Fattah  
Associate Editor  
Journal of Egyptian Women's Dermatologic Society

Reviewer Comments:

Minor comments are in the attached file which you can get from action links

Complete Revision Instructions:

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Dari: Journal of the Egyptian Women's Dermatologic Society (editor.jewds@journalonweb.com)

Kepada: dwimurtiastutik@yahoo.co.id

Tanggal: Senin, 23 Agustus 2021 17.04 WIB

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Dear Dr Murtiastutik,

The Editorial team of Journal of The Egyptian Women's Dermatologic Society is pleased to inform you that your manuscript jewd\_44\_21 entitled "Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV / AIDS patients", is accepted for publication in the journal.

We will be sending you page proofs of the manuscript in about 2-3 weeks for final checking and corrections if any., through the manuscript management site before the publication of the manuscript.

Please note that the journal reserves the right to make changes in the language, grammar, presentation, etc. to suit the journal's requirements.

We thank you for submitting your valuable research work to Journal of The Egyptian Women's Dermatologic Society.

With warm personal regards,

Yours sincerely,

The Editorial Team

Journal of The Egyptian Women's Dermatologic Society

# Association between etiologic species with CD4 count and clinical features of oral candidiasis among HIV/AIDS patients

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## Background

Oral candidiasis (OC) is an oral mucosal disorder due to *Candida* genus. Its predisposing factor among patients with HIV/AIDS is mainly decreasing CD4 count. OC is commonly caused by *Candida albicans*. As CD4 decreases, the shift to *C. non-albicans* has been observed.

## Objective

To evaluate the association of *Candida* species with CD4 count and clinical features in HIV/AIDS patients with OC.

## Patients and methods

This is a cross-sectional study. A total of 114 oral rinse solution samples from HIV/AIDS patients with OC were collected. *Candida* species identification was done by culture in Chromagar followed by VITEK 2. The association of *Candida* species with CD4 count and clinical features was analyzed using Pearson's  $\chi^2$  and Kruskal-Wallis tests.

## Results

There was growth of 149 isolates in culture from 114 patients. *C. albicans* was found in 104 (69.7%) isolates. *Candida non-albicans* were found in 45 (30.3%) isolates, namely *Candida krusei* in 22 (14.85%), *Candida glabrata* in 12 (8.1%), *Candida tropicalis* in six (4.05%), *Candida dubliniensis* in two (1.3%), *Candida parapsilosis* in two (1.3%), and *Candida lipolytica* in one (0.7%) isolate. *Candida* species was significantly associated with clinical types, episode types, pain on swallowing, CD4 count, and antiretroviral (ARV) use among all patients.

## Conclusion

Among HIV/AIDS patients with OC, growth of *C. albicans* only was more common in higher CD4 count, while mixed growth of *C. albicans* and *C. non-albicans* was more common in lower CD4 count. Clinical features associated with growth of *C. albicans* only were pseudomembranous type, recurrent OC, absence of pain on swallowing, and patients on ARV, whereas those associated with mixed growth of *C. albicans* and *C. non-albicans* were cheilitis type, first-episode OC, presence of pain on swallowing, and ARV-naïve patients.

## Keywords:

AIDS, *Candida albicans*, candidiasis, CD4 counts, HIV

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## Introduction

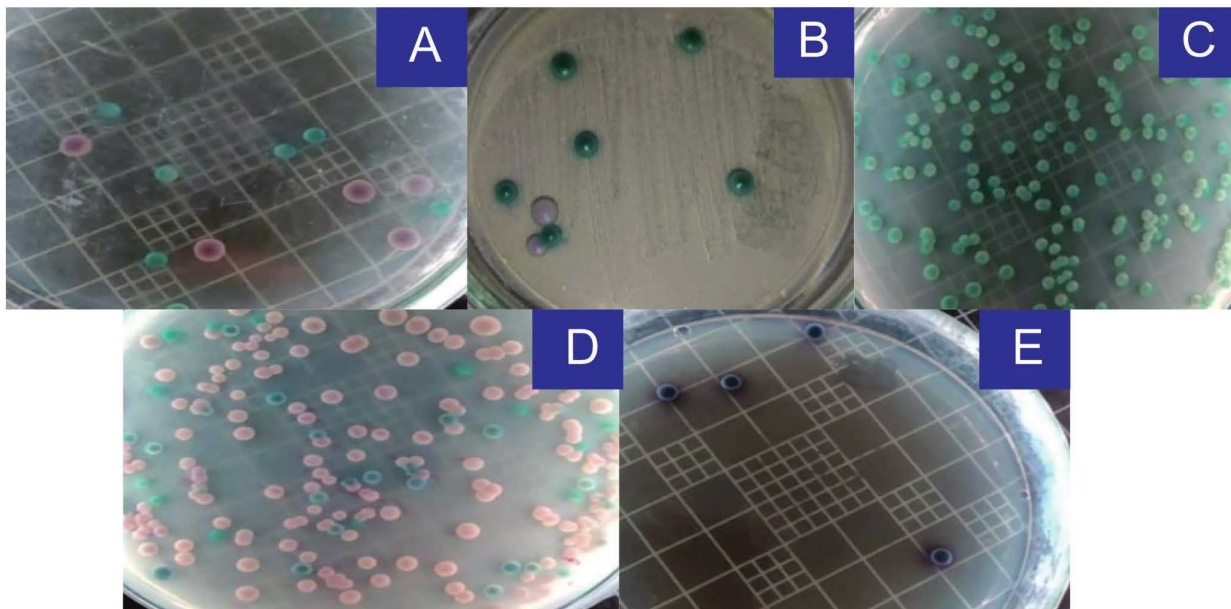
Oral candidiasis (OC) is an oral mucosal disorder due to *Candida* genus [1], and an independent predictor of immunodeficiency in AIDS patients [2]. The predisposing factor for OC among HIV/AIDS patients is mainly decreasing CD4 count [1]. OC is commonly caused by *Candida albicans*. As CD4 decreases, the shift to *C. non-albicans* like *Candida tropicalis*, *Candida glabrata*, and *Candida krusei* has been observed recently [2]. *Candida* species has been associated also with other features such as antiretroviral (ARV) use among HIV/AIDS patients with OC [3,4]. This study evaluates the association of *Candida* species with CD4 count and clinical features among HIV/AIDS patients with OC.

## Patients and methods

This was an observational analytical cross-sectional study. The participants in this study were HIV/AIDS patients who visited our institution and had OC opportunistic infections. Diagnosis of HIV/AIDS was done by rapid test/HIV three methods using Fokus (PT Fokus Diagnostic Indonesia, Jakarta Selatan, Indonesia), InTec (InTec Product Inc., Xiamen, China), and VIKIA (Biomérieux, SA, Marcy l'Etoile, France). Diagnosis of OC was based on

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Figure 1



*Candida* colonies in Chromagar: (a) *Candida albicans* (green) and *Candida glabrata* (baby purple), (b) *C. albicans* (green) and *Candida parapsilosis* (white), (c) *C. albicans* (green), (d) *C. albicans* (green) and *Candida krusei* (pink), (E) *Candida tropicalis* (prussian blue).

clinical examination and 10–20% KOH examinations, followed by culture in Chromagar media (Becton Dickinson and Company, Franklin Lakes, New Jersey, USA) with subsequent culture in VITEK 2 (Biomérieux). CD4 T-lymphocyte count was evaluated with BD FACSCalibur device (Becton, Dickinson and Company, Franklin Lakes, New Jersey, USA).

The inclusion criteria were male or female patients and above 18 years of age. Patients who have or have not taken ARV were included in this study. The exclusion criteria were patients with no growth of fungal colony in the culture, history of other immunosuppressive diseases, consumption of antifungal medications within 3 months before the study, immunosuppressive medications, prolonged antibiotics or contraceptives, and dental caries. Patients who agreed to take part in the study received explanation about the study and gave informed consents. Ethical clearance has been given by the Health Research Ethics Committee of our institution with number of 1129/KEPK/IV/2019, dated April 20, 2019.

The patients were grouped based on CD4 T-lymphocyte level into three groups: patients with CD4 T-lymphocyte level of 1–100 cells/ $\mu$ l, CD4 T-lymphocyte level of 101–200 cells/ $\mu$ l, and CD4 T-lymphocyte level more than 200 cells/ $\mu$ l. Each group had 38 patients and the total sample was 114 patients.

Samples were taken by consecutive sampling from patients fulfilling the inclusion criteria, until the required number of samples for each group was fulfilled.

The study commenced on May 2019 until 114 patients were obtained. Samples were taken from oral rinse solutions collected by asking patients to gargle using 25 ml of sterile aquadest, which were then stored in a sterile container and sealed, labeled with the identity of the patient, date and time of collection, and sent to the laboratory. Fungal culture is done in Chromagar media (Fig. 1). In order to identify the species of *Candida*, subsequent culture was done in VITEK 2.

#### Statistical analysis

The results were analyzed with SPSS, version 17 (IBM, Armonk, New York, USA). Pearson's  $\chi^2$  test was used to analyze the association between *Candida* species and sex, clinical types, episode types, pain on swallowing, CD4 count, and ARV use. Kruskal–Wallis test were used to analyze the association between *Candida* species and age. A *P* value of <0.05 was considered statistically significant.

#### Results

This study described 114 patients with OC and HIV/AIDS who fulfilled the inclusion criteria as study participants. The age characteristic of the study

Figure 2



Clinical types of OC: (a) pseudomembranous, (b) cheilitis, (c) acute atrophic.

**Table 1 Clinical features of the study patients**

Characteristics	n (%)
<b>Sex</b>	
Male	83 (72.8)
Female	31 (27.2)
<b>Age (years)</b>	
Range	18–59
Mean±SD	36.4±9.8
<b>Chief complaint</b>	
Whitish patches in oral cavity	100 (87.7)
Reddish patches in oral cavity, patches and sores on corners of lips	7 (6.2)
Whitish and reddish patches in oral cavity	4 (3.5)
Whitish patches in oral cavity, patches and sores on corners of lips	3 (2.6)
<b>Location</b>	
Tongue	54 (47.3)
Tongue and mucosa	49 (43.0)
Tongue, mucosa and lips	10 (8.8)
Mucosa	1 (0.9)
<b>Clinical types</b>	
Acute pseudomembranous	103 (90.3)
Cheilitis	10 (8.8)
Acute atrophic	1 (0.9)
Chronic hyperplastic	0
<b>Episode types</b>	
First time	33 (28.9)
Recurrent	81 (71.1)
<b>Pain on swallowing</b>	
Yes	50 (43.9)
No	64 (56.1)
Total	114 (100.0)

patients was mean±SD of 36.4±9.8 years old with range from 18 to 59 years old. There were more male (72.8%) than female patients (27.2%). This study showed chief complaint of whitish patches in

the oral cavity in 100 (87.7%) patients, reddish patches in the oral cavity with patches and sores on the corners of the lips in seven (6.2%) patients, whitish and reddish patches in the oral cavity in four (3.5%) patients, and whitish patches in the oral cavity with patches and sores on the corners of the lips in three (2.6%) patients. The most common location was on the tongue in 54 (47.4%) patients, tongue and buccal and palatal mucosa in 49 (43.0%) patients, tongue, buccal, and palatal mucosa and lips in 10 (8.8%) patients, and buccal and palatal mucosa only in one (0.9%) patient. The clinical types of OC were mostly pseudomembranous type (Fig. 2a), followed by cheilitis type (Fig. 2b) and acute atrophic type (Fig. 2c), in 103 (90.4%) patients, 10 (8.8%) patients, and one (0.9%) patient, respectively. Pain on swallowing was absent in 64 (56.1%) patients and present in 50 (43.9%) patients. Most patients had recurrent OC (88 patients, 71.1%), while others had first episode of OC (33 patients, 28.9%) (Table 1).

In this study, all patients grew fungal colonies in culture. There were 149 isolates of *Candida* species from 114 patients. The predominant species in this study was *C. albicans* in 104 (69.7%) isolates. *Candida* non-*albicans* were found in 45 (30.3%) isolates, namely *C. krusei* in 22 (14.85%), *C. glabrata* in 12 (8.1%), *C. tropicalis* in six (4.05%), *Candida dubliniensis* in two (1.3%), *Candida parapsilosis* in two (1.3%), and *Candida lipolytica* in one (0.7%) of the isolates. Growth of *C. albicans* only was found in 69 patients and *C. non-albicans* only were found in 10 patients. Mixed growth of *C. albicans* and *C. non-albicans* was found in 35 patients. *C. krusei* was found growing alone in nine patients and mixed with *C. albicans* in 13

patients. *C. tropicalis* was found growing alone in one patient and mixed with *C. albicans* in five patients. Other *C. non-albicans* species were found in mixed growth with *C. albicans*, namely *C. glabrata* in 12 patients, *C. dubliniensis* in two patients, *C. parapsilosis* in two patients, and *C. lipolytica* in one patient.

*Candida* species was significantly associated with clinical types ( $P=0.011$ ), episode types ( $P=0.003$ ), pain on swallowing ( $P=0.002$ ), CD4 count ( $P=0.000$ ), and ARV use ( $P=0.004$ ), but was not significantly associated with sex ( $P=0.747$ ) and age ( $P=0.188$ ) among all patients. *Candida* species was associated with pain on swallowing and CD4 count, regardless of ARV use. *Candida* species was also significantly associated with clinical types among patients on ARV, and with sex among ARV-naïve patients (Table 2). In general, growth of *C. albicans* only was more common in patients with higher CD4 count, pseudomembranous type and recurrent OC, without pain on swallowing, and patients on ARV, while mixed growth of *C. albicans* and *C. non-albicans* was more common in patients with lower CD4 count, cheilitis type and first episode of OC, presence of pain on swallowing, and ARV-naïve patients (Table 2).

## Discussion

OC is the most common opportunistic fungal infection among individuals infected with HIV/AIDS [2]. OC can affect various age groups and sex, from infants to the elderly. Marak and Dhanashree [5] showed that the most affected age group is 51–60-year-old group, which may be due to the low immunity and immunosuppressive diseases. In this study, there were more male than female patients. This is concordant with data from the Health Ministry of the Republic of Indonesia in 2017, which showed that HIV patients were more common in males than females [6]. Ambe *et al.* [7] showed that the prevalence of OC was not significantly associated with sex. In general, there is no difference of the prevalence of OC based on sex, because OC is different from vulvovaginal candidiasis that is influenced by hormonal factor [8,9]. The age of the patients had mean±SD of 36.41±9.825 years old with range of 17–59 years old. This result may be due to that adults are productive and sexually active, thus engaging in many unsafe sexual practices risky for HIV transmission [10].

OC may be the first sign or symptom of HIV/AIDS disease, and may sometimes be the

presenting complaint for patients [11]. *Candida* can spread extensively, directly from the oral cavity to other organs such as the digestive tract and many more [12]. The most common symptom was whitish patches in the oral cavity and the most common locations were tongue and also mucosa, which correspond to the most common clinical type of OC, pseudomembranous OC [2,9]. There was no associated pain on swallowing in the majority of patients. Pain on swallowing is a clinical sign that the lesion affects the pharynx and esophagus [9].

In this study, *C. albicans* was the predominant species growing in 104 isolates (69.7%), while *C. non-albicans* were found in 45 (30.3%) isolates, namely *C. krusei*, *C. glabrata*, *C. tropicalis*, *C. dubliniensis*, *C. parapsilosis*, and *C. lipolytica*. This proved that *C. albicans* was still the leading cause of OC, but *Candida* non-*albicans* began to be identified. Ambe *et al.* [7] showed similar result in which there were more *C. albicans* isolates (60.2%) than *C. non-albicans* (39.8%), and there were similar isolated *Candida* non-*albicans* species such as *C. glabrata*, *C. krusei*, *C. tropicalis*, and *C. parapsilosis* from HIV patients with OC in Cameroon. A meta-analysis study in sub-Saharan Africa from 2005 to 2015 also showed a prevalence of 33.5% for *C. non-albicans*, the most common among which was *C. glabrata*, followed by *C. krusei* and *C. tropicalis* [13]. Nelwan *et al.* [14] found *C. albicans* in 56.2% and *C. glabrata* in 15.3% of OC in Indonesian HIV/AIDS patients.

Predominance of *C. albicans* in OC among HIV/AIDS patients may be related to the pathogenicity of this species by production of hydrolytic enzymes, presence of multiple-adhesion factor, and ability to convert from yeast form to hyphal form that is crucial for tissue invasion [7]. *C. albicans* also synergistically aided colonization and infection by *C. glabrata*, while it was suppressed by *C. krusei*, which may explain the mixed growth of *C. glabrata* with *C. albicans*, and the growth of *C. krusei* only in some patients of this study [15]. *Candida* non-*albicans* are considered less virulent than *C. albicans* because of their lower adherence ability to mucosal and endothelial surface, less production of proteinase enzymes, and lack or lower ability to form hyphae [16]. However, increasing prevalence of *Candida* non-*albicans* is of importance because these species are inherently resistant to widely available fluconazole, resulting in a public health problem among HIV/AIDS population in sub-Saharan Africa [13,17].

**Table 2** *Candida* species and associated clinical and laboratory features

Clinical and laboratory features	All patients				Patients on ARV				ARV-naïve patients			
	<i>Candida albicans</i> only	<i>Candida non-albicans</i> only	Mixed <sup>#</sup>	P value	<i>Candida albicans</i> only	<i>Candida non-albicans</i> only	Mixed <sup>#</sup>	P value	<i>Candida albicans</i> only	<i>Candida non-albicans</i> only	Mixed <sup>#</sup>	P value
<b>Sex</b>												
Male	52	7	24	0.747	38	6	13	0.951	14	1	11	0.038 <sup>*</sup>
Female	17	3	11		17	3	5		0	0	6	
Age (mean±SD)	35.3±8.7	40.9±9.3	37.5 ±11.3	0.188	35.9±9.4	39.8±9.1	38.8 ±10.1	0.339	33.1±5.1	51	36.2 ±12.6	0.353
<b>Clinical types</b>												
Pseudomembranous	66	10	27	0.011 <sup>*</sup>	53	9	14	0.021 <sup>*</sup>	13	1	13	0.278
Cheilitis	2	0	8		2	0	4		0	0	4	
Acute atrophic	1	0	0		0	0	0		1	0	0	
<b>Episode types</b>												
First episode	16	0	17	0.003 <sup>*</sup>	8	0	4	0.305	8	0	13	0.198
Recurrent	53	10	18		47	9	14		6	1	4	
<b>Pain on swallowing</b>												
Yes	23	3	24	0.002 <sup>*</sup>	20	2	12	0.036 <sup>*</sup>	3	1	12	0.015 <sup>*</sup>
No	46	7	11		35	7	6		11	0	5	
<b>CD4 (cells/μl)</b>												
1–100	18	0	20	0.000 <sup>*</sup>	18	0	7	0.002 <sup>*</sup>	0	0	13	0.000 <sup>*</sup>
101–200	18	6	14		11	5	10		7	1	4	
>200	33	4	1		26	4	1		7	0	0	
<b>ARV use</b>												
Yes	55	9	18	0.004 <sup>*</sup>	55	9	18	-	0	0	0	-
No	14	1	17		0	0	0		14	1	17	
<b>Total</b>	<b>69</b>	<b>10</b>	<b>35</b>		<b>55</b>	<b>9</b>	<b>18</b>		<b>14</b>	<b>1</b>	<b>17</b>	

ARV, antiretroviral. <sup>#</sup>Growth of *Candida albicans* and *Candida non-albicans* in one patient. \*P-value <0.05 is considered statistically significant.

In this study, growth of *C. albicans* only was more common in patients with higher CD4 count, pseudomembranous type and recurrent OC, without pain on swallowing, and patients on ARV, while mixed growth of *C. albicans* and *C. non-albicans* was more common in patients with lower CD4 count, cheilitis type and first episode of OC, presence of pain on swallowing, and ARV-naïve patients. Mushi *et al.* [3] found that *C. non-albicans* was increased in patients with low CD4. Substantial immunosuppression in HIV as reflected by low CD4 count may promote the growth of the less-pathogenic *C. non-albicans* and cause OC [3,13]. Lam-Ubol *et al.* [4] showed that *C. non-albicans* was more common in highly active ARV therapy (HAART)-naïve patients, and use of HAART was associated with the decrease of these species. HAART may act directly by inhibition of secretory aspartyl proteinase enzymes needed for growth of Candidal hyphae, and indirectly by improving CD4 levels to reduce *Candida* colonization [18,19]. Nair and Shetti [20] showed the association between pseudomembranous OC and *C. albicans*. Ribeiro *et al.* [21] found that *C. albicans* was more common in recurrent OC, while *C. non-albicans* was found in primary OC. Although *C. albicans* was the most common cause of esophageal candidiasis [9], Redding *et al.* [22] found that the presence of *C. non-albicans* produced more severe pain on swallowing, as also seen in this study. Previous antifungal treatment increased the prevalence of *C. non-albicans* by exerting positive-selection pressure to these species that were less sensitive to these drugs [15]. Antibiotics use has also been associated with mixed infection by *C. non-albicans* and *C. albicans* [17]. However, intake of those medications has been excluded in this study.

This study has some limitations such as the relatively smaller number of ARV-naïve patients and lack of details of ARV therapy. Future studies with larger samples of both patients on ARV and ARV-naïve patients, and analysis of the association of duration and types of ARV to *Candida* species, are recommended.

In conclusion, *Candida* species was significantly associated with CD4 count, ARV use, clinical and episode types, and pain on swallowing among HIV/AIDS patients with OC. *C. albicans* was the predominant species, but growth of *C. non-albicans* alone or mixed with *C. albicans* was identified. Mixed growth was more common in lower CD4 count, cheilitis type, first-episode OC, pain on swallowing, and ARV-naïve patients.

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#### Conflicts of Interest

There are no conflicts of interest.

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