ORIGINAL ARTICLE

UTILITY OF URINE KOH IN DETECTING CANDIDURIA IN INFANTS

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The authors declare that the data presented are original material and has not been previously published, accepted or considered for publication elsewhere; that the manuscript has been approved by all authors, and all authors have met the requirements for authorship.

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ABSTRACT

Background: Candida species are common cause of urinary tract infection in infants requiring medical care. Candida fungal elements may be demonstrated in urine using microscopic examination with potassium hydroxide (KOH). However, detection of these elements does not always correlate with candiduria.

Objectives: To establish the utility of urine KOH in identifying candiduria and to determine the risk factors, as well as urinalysis and CBC parameters associated with candiduria.

Methods: This prospective cross-sectional study included admitted infants 1 year and below with urine culture and with any risk factor/s for candiduria. Additional urine KOH testing was done using clean catch or catheter method. Urine culture was used as the gold standard.

Results: Among the 90 study participants with both urine culture and urine KOH, 13 (14%) had candiduria. The use of indwelling catheter, presence of urinary tract anomalies, positive leukocyte esterase in urinalysis, and increased monocyte counts in CBC are all associated with candiduria. Urine KOH has sensitivity of 100%, (CI 75.2-100%), specificity 59.7%, (CI 47.9-70.7%), PPV 29.5%, (CI 17.7-45.2%), and NPV 100%, (CI 92.2-100%) in detecting candiduria.

Conclusions: Negative urine KOH has excellent negative predictive value, while positive urine KOH result may warrant further investigation. Urine KOH results should be interpreted with caution depending on patient's risk factors, clinical status, and other laboratory results prior to initiation of empiric antifungal therapy. Positive urine KOH may not always require treatment.

KEYWORDS: *urine KOH, candiduria, Candida*

INTRODUCTION

Candida species are one of the common causes of urinary tract infection (UTI) in neonates and infants requiring medical care.[1] Fungal elements (e.g. yeast cells and pseudohyphae) may be seen in *Candida* infected body fluid specimens such as urine with microscopic examination using 10-20% potassium hydroxide suspension (KOH).[2] However, detection of these fungal elements in urine does not always correlate with candiduria or UTI. Urine KOH results are frequently used to diagnose candiduria in many clinical settings because it is affordable, readily available, and yields immediate results compared to urine culture, the gold standard for detecting candiduria. However, the role of KOH in urine for detection of candiduria has not been well studied. There was no available data or study conducted both locally and internationally that compared urine KOH to urine culture, hence this study was undertaken. This study will help and guide clinicians regarding the value of urine KOH in the diagnosis of candiduria in infants. If highly sensitive or specific, it may be a valuable screening tool for candiduria. However, if not sensitive, we will be able to prevent unnecessary urine KOH testing, and thus unnecessary expenses. And if shown that it is not specific, this will prevent unwarranted exposure of patients to antifungal therapy.

The main objective of this study is to establish the utility of urine KOH in identifying candiduria in infants. Specifically, to determine the sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of urine KOH in detecting candiduria in infants compared with urine culture, to identify the risk factors significantly associated with candiduria in infants, and to detect association of urinalysis findings and CBC parameters with candiduria in infants.

Candiduria is defined by presence of $>10^3$ CFU/mL in urine culture (by suprapubic aspiration), $>10^4$ CFU/mL (by urethral catheterization), or $>10^5$ (by clean catch method), and a positive Urine KOH is the presence of fungal elements (yeast or hyphae) in urine specimen.

METHODOLOGY

This prospective cross-sectional study determined the utility of urine KOH in detecting candiduria in infants conducted from September 2017 – June 2018. The study commenced upon the approval of the Institutional Review Board and Ethics Committee of our institution. This study did not have any financial sponsors. No conflicts of interest are hereby declared.

Subject and Sample Size Computation

Inclusion Criteria: Admitted infants 1 year old and below, with urine culture request, and with any of the following risk factors: low birth weight (<2500g), prematurity (<37 weeks AOG), on prolonged steroids (>14 days), with congenital urinary tract anomalies, on broad-spectrum antibiotics (e.g. third and 4th generation cephalosporins, piperacillin tazobactam, vancomycin, carbapenems), on parenteral nutrition, admitted at ICU, on endotracheal intubation, with indwelling urinary catheter or on clean intermittent catheterization, those who underwent recent (≤ 1 month) abdominal, pelvic or urologic surgery, with hematologic malignancies, or those on immunosuppressive drugs (e.g. on chemotherapy). Patients on antifungal prophylaxis or previously given antifungal were included as long as they were able to fulfill the inclusion criteria.

Exclusion Criteria: infants with urine culture without any risk factor for candiduria, infants with cutaneous candidiasis on the pelvic/perineal area (i.e. satellite pustules with erythematous base,

and marginal scaling), and infants with diaper dermatitis.

Using Epi Info Version 7, the minimum sample size requirement is 90 based on the specificity of KOH smear in evaluation of fungal foot infection (62%)[16], with a margin of error 10% and confidence interval of 95%.

Study Procedure

Admitted infants (≤12 months old) with urine culture request were identified from the laboratory logbook daily. Once identified, the risk factors for candiduria were determined if present in these infants, which was done through history and physical examination of the patient and chart review by the principal investigator. If a risk factor was present, and the infant had no clinical signs of diaper dermatitis or cutaneous candidiasis on the pelvic or perineal area, an informed consent was obtained from the parents/guardian of the infant for inclusion in the study. Thereafter, urine collection for KOH testing was obtained for those infants without prior urine KOH test. Infants with recent KOH test (past 24 hours) were included in the study but no additional KOH testing was done. Urine specimen for KOH testing was collected either via clean catch method, or from catheter (in catheterized patients), within 24 hours of urine culture collection.

Data Processing and Analysis

Data analysis was performed in Stata SE version 13. Quantitative variables were summarized as mean and standard deviation, while qualitative variables were tabulated as frequency and percent. Accuracy of urine KOH in predicting candiduria were computed in terms of its sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV). Factors associated with candiduria was analyzed using logistic regression. The level of significance was set at 5%.

RESULTS

In our study, 69% (62) were males and 67% (60) were infants more than 1 month old. The most common risk factors identified for candiduria in the study participants were the use of broad-spectrum antibiotics in 72 of the 90 cases (80%), followed by admission to an Intensive Care Unit 59 (66%), and having endotracheal intubation 32 (36%) (Table 1).

Table 1. Risk factors Identified for Candiduria

Risk factors	**n = 90**	**%**
Low birth weight (<2500g)	24	27%
Prematurity (<37 weeks AOG)	18	20%
Prolonged steroids (≥14 days) use	1	1%
Congenital urinary tract anomalies	14	16%
Broad-spectrum antibiotics (e.g. 3rd and 4th generation cephalosporins, piperacillin, vancomycin, carbapenems) use	72	80%
Parenteral nutrition	5	6%
Admitted at Intensive Care Unit (ICU), Neonatal ICU (NICU)	59	66%
Endotracheal intubation	32	36%
Urinary catheter use	20	22%
Central vascular catheters (central lines)	17	19%
Recent (≤ 1 month) abdominal, pelvic or urologic surgery	11	12%
Hematologic malignancies	3	3%
Immunosuppressive drugs (e.g. on chemotherapy) use	1	1%
Others: Candidemia	3	3%

In our study, 56 (62%) did not have any growth in urine culture, 21 (23%) had bacterial growth, and 13 (14%) had candiduria. Among these, 10 (77%) were non-albicans *Candida* spp. and 3 (23%) were *Candida albicans.*

Of the 13 infants with candiduria, 8 (62%) were males, and 8 (62%) were more than 1 month old. The most common identified predisposing risk factor in those with candiduria was the use of broad spectrum antibiotics (100%), followed by admission to an Intensive Care Unit (69%), and having a urinary catheter (61%) (Table 2).

The presence of congenital urinary tract anomalies, or those with urinary catheter are the significant risk factors identified for candiduria in this study. The odds of developing candiduria is 4.72 if a patient has congenital urinary tract anomaly, and 8.67 if with urinary catheter (Table 2).

Table 2. Association of Patients' Characteristics and Risk factors with Candiduria

Characteristics Risk factors	***With*** candiduria n = 13	***Without*** candiduria n = 77	**Odds ratio**	**P value**	**C.I**
Sex (male)	8 (62%)	54 (70%)	0.68	0.538	0.20 – 2.30
Low birth weight	0	24 (31%)	-	-	-
Prematurity	0	18 (23%)	-	-	-
Prolonged steroids use	1 (8%)	0	-	-	-
Congenital urinary tract anomalies	5 (38%)	9 (12%)	4.72	0.021	1.27 – 17.60
Broad-spectrum antibiotics use	13 (100%)	59 (77%)	-	-	-
Parenteral nutrition	0	5 (6%)	-	-	-
Admitted at ICU/NICU	9 (69%)	50 (65%)	1.2	0.763	034 – 4.31
Endotracheal intubation	6 (46%)	26 (34%)	1.68	0.391	0.51 – 5.51
Urinary catheter	8 (62%)	12 (16%)	8.67	0.001	2.41 – 31.04
Central vascular catheters	2 (15%)	15 (19%)	0.75	0.728	0.15 – 3.75
Recent (≤ 1 mo) abdomen, urologic/pelvic surgery	1 (8%)	10 (13%)	0.56	0.594	0.06 – 4.77
Hematologic malignancies	0	3 (4%)	-	-	-
Immunosuppressive drugs use	0	1 (1%)	-	-	-

Of the 13 infants with candiduria, 1 (8%) showed positive nitrite results, and 9 (69%) showed positive for leukocyte esterase. The mean values for urine WBC (63/hpf) and RBC (62/hpf), as well as CBC parameters were also noted (Table 3).

From the results of the study, the significant laboratory parameters associated with candiduria are presence of leukocyte esterase on urinalysis and elevated monocyte counts on CBC. The presence of leukocyte esterase in urinalysis increases the odds of having candiduria as well as increase in monocyte counts in CBC (Table 3).

Table 3. Association of Patients' Laboratory Parameters with Candiduria

Laboratory Parameters	***With*** candiduria Mean ± SD n = 13	***Without*** candiduria Mean ± SD n = 77	Odds ratio	P value	C.I
Urinalysis					
Nitrite (+)	1 (8%)	5 (6%)	1.20	0.873	0.12 – 11.18
Leukocyte esterase (+)	9 (69%)	17 (22%)	7.94	0.002	2.17 – 28.99
WBC	62.9 ± 122.0	23.3 ± 67.0	1.00	0.122	0.99 – 1.01
RBC	62.1 ± 158.0	112.4 ± 534.9	1.00	0.740	0.99 – 1.00
CBC					
Hemoglobin	11.7 ± 2.0	11.6 ± 2.6	1.02	0.697	0.81 – 1.28
Hematocrit	35.3 ± 5.9	34.5 ± 7.5	1.02	0.939	0.93 – 1.09
WBC	14.9 ± 7.0	21.0 ± 24.1	0.97	0.366	0.91 – 1.03
Neutrophil	55.6 ± 21.3	55.2 ± 22.6	1.00	0.957	0.97 – 1.02
Lymphocyte	30.3 ± 17.9	36.6 ± 22.9	0.98	0.350	0.95 – 1.01
Eosinophil	3.3 ± 5.01	1.9 ± 2.6	1.12	0.159	0.95 – 1.30
Monocyte	9.2 ± 4.2	5.1 ± 4.0	1.27	0.004	1.08 – 1.49
Platelet ($x10^3$)	398.0 ± 259.7	244.5 ± 164.9	1.00	0.010	1.00 – 1.00

As seen in table 4, of the 90 study participants, 44 patients (49%) had positive urine KOH while 46 patients (51%) had negative urine KOH. Of the 44 patients who tested positive for urine KOH, 13 infants (30%) had positive urine culture result. Thirty-one (31) patients (70%) who tested positive for urine KOH was negative for urine culture. Of the 46 patients who tested negative for urine KOH, all 46 patients (100%) were also negative for urine culture.

Table 4. Urine KOH and Urine Culture Results

	Urine CS (+)	Urine CS (-)	Total
Urine KOH (+)	13	31	**44**
Urine KOH (-)	0	46	**46**
	13	***77***	90

The sensitivity of urine KOH in detecting candiduria is 100%, which means that all patients with candiduria tested positive for urine KOH. The specificity of urine KOH is 59.74%, which means that 59.74% of patients without candiduria tested negative for urine KOH. The positive predictive value (PPV) of urine KOH is 29.55%, while the negative predictive value (NPV) of urine KOH is 100% (Table 5).

Table 5. Accuracy of Urine KOH in Detecting Candiduria

	Overall % (C.I)
Sensitivity	**100% (75.2-100%)**
Specificity	**59.7% (47.9-70.7%)**
PPV	**29.5% (16.7-45.2%)**
NPV	**100% (92.2-100%)**

DISCUSSION

Candida spp. are commensal organisms commonly found in the gastrointestinal and genitourinary tracts of healthy individuals.[3] *Candida* is the most important cause of fungal infection in health care settings, including those of the urinary tract. In the majority of asymptomatic persons, the presence of yeast in the urine indicates contamination or colonization. However, in symptomatic patients or those with risk factors for candiduria, the presence of yeast may indicate true infection.

In the study by Gholamipour et al, the highest frequency of candiduria was seen in patients who had received more than 2 or 3 antibiotics during their hospitalization (37% and 24%, respectively).[4] Other risk factors identified in their study include admission in ICU (24.5%) and NICU (12%), those with cardiovascular disorder (18%), with urinary catheter (12%), respiratory diseases (10%), anomaly of the urinary tract (10%), gastrointestinal and liver diseases (9%) and neurologic disorders (8.5%). In relation to this, this study has identified the use of broad-spectrum antibiotics in all (100%) patients who developed candiduria. Furthermore, other common risk factors for candiduria that were identified in this study include the following: admission to intensive care unit (69%), use of indwelling urinary catheter (61%), on endotracheal intubation (46%), and presence of congenital urinary tract anomaly (38%).

In the study of Paul et al, prior antimicrobial use was documented in 92% with candiduria (OR 9.1; 95%; CI 2.1-31.9) [5], while in this study as mentioned above, prior antimicrobial use was documented in 100% of patients with candiduria. Furthermore, Alfouzan et al. reported that aside from long term urinary catheterization, prior antibiotic use is the next most significant risk factor for candiduria.[6] In this study, the most frequently used antibiotic in patients with candiduria were cephalosporins (3rd and 4th generations).

The use of broad-spectrum antibiotics leads to alteration of the normal bacterial flora, that results to a more conducive environment for the growth of yeasts. The higher number of candiduria cases noted in ICU patients are probably secondary to others factors such as underlying diseases, relative immunodeficiency status, multiple manipulations by health care team and altered bacterial flora secondary to use of broad-spectrum antibiotics.[7]

In this study, the odds of developing candiduria increases in the presence of urinary tract anomaly, or use of indwelling urinary catheter. Urinary tract anomalies noted in patients with candiduria in this study included horseshoe kidneys, cloacal exstrophy, prune belly syndrome, and bladder exstrophy. Alfouzan et al. also reported that long-term urinary catheterization is considered to be the most significant risk factor for candiduria.[6]

The presence of pyuria, hematuria, or leukocyte esterase in urinalysis maybe useful in distinguishing infection from contamination or colonization.[3] In this study, urinalysis and CBC parameters were compared between those with candiduria and those without candiduria. Significantly, the presence of positive leukocyte esterase in urine specimen increased the odds of a patient having candiduria. Monocytes, along with neutrophils and macrophages are important antifungal effector cells. Residing phagocytes in infected organs are involved in the killing of invading Candida, whereas neutrophils and monocytes are recruited to the site of infection.[3] The mean percentage of monocyte in infants is 5%.[8] In our study, increase in monocyte counts was noted to be associated with increased odds of having candiduria. The presence of low platelet count has been associated to candidemia in several studies, especially in the neonates.[9,10] In our study, platelet counts has no significant association with the presence or development of candiduria, probably because our study population involved more infants than neonates.

Among the 13 *Candida* species isolated in our study, 10 were non-albicans *Candida* spp. (77%), and 3 were *Candida albicans* (23%). In the study of Malhotra et al, *C. albicans* were isolated in 37 of 333 cases (11.1%) and non-albicans *Candida* spp. were noted in 35 patients (10.5%). [11]

The accuracy of urine KOH in its ability to detect significant candiduria has not been well studied. There is scarcity in data regarding the use of urine KOH in predicting candiduria when compared to urine culture as the gold standard. In this study, the sensitivity of urine KOH was noted at 100%, implying that all patients with candiduria tested positive with urine KOH. On the other hand, the specificity of urine KOH was noted at 59.74%. The 100% sensitivity also means that urine KOH will detect virtually every infant who has candiduria but its low specificity means that it will be falsely positive for a number of infants who actually don't have candiduria. Comparison of urine KOH with the standard urine culture is important, since urine KOH is a cheaper, readily available especially in remote areas, and yields more rapid results.

From the results of the study, not all patients with positive urine KOH implies candiduria. Of the 44 patients with positive urine KOH, only 13 (30%) showed with positive urine culture results. Thus, it is prudent to not immediately treat patients with positive urine KOH result with antifungals such as fluconazole, unless correlated with urine cultures and clinical status of the patient. Correlation of the patient's clinical status is also important, as not all infections are detected by urine culture even though it is the gold standard in detecting candiduria. Furthermore, exposure of patients to unnecessary drugs or antimicrobials (antifungals included), has its drawbacks and disadvantages.

First, unnecessary exposure of patients to antifungals may lead to emergence of resistant strains of *Candida* species such as *C. glabarata* and *C. krusei*. In the study of Prasad et al, they identified that patients older than 2 years, those with recent surgical procedure, and prior fluconazole use were independent risk factors for infection with *C. glabrata* and *C. krusei* in children.[12] Second, the general recommendation for treatment of candidemia is the use of Amphotericin B, which is usually nephrotoxic and may cause electrolyte imbalances (e.g. hypercalciuria, hypokalemia, hypomagnesemia), renal tubular acidosis, renal failure, acute hepatic failure, and hypotension.[8] In relation to this, patients who are not on prior azole use (e.g. fluconozaole) and not critically ill may use fluconazole for treatment of candidemia with susceptible isolates.[2] However, in the instance that a patient was previously treated with fluconazole because of other conditions (e.g. positive urine KOH), then we can no longer use fluconazole (a relatively safer agent compared to amphotericin B) to treat candidemia; amphotericin B will be given and continued for at least 14 days, thereby increasing the risk for possible detrimental side effects of this antifungal as previously mentioned.

Lastly, unnecessary use of antifungals such as fluconazole provides additional economic burden to patient's family. Locally, IV fluconazole approximately costs 1,500 pesos per vial of 2mg/ml (100ml), while oral fluconazole capsule costs 400 pesos per 200mg tablet.

Fluconazole is highly water soluble and is mainly excreted in the urine as an active drug (urinary concentrations are more than 10-fold compared to those in serum). With this, fluconazole is considered the drug of choice for both candida cystitis and pyelonephritis.[13] For asymptomatic candiduria, elimination of predisposing factors such as indwelling urinary catheters catheter is strongly recommended. Antifungal treatment is not recommended unless patients has high risk of candidemia (blood stream infection), such as neutropenia, very low birth weight, and patients who will undergo urologic manipulation.[2] In patients with indwelling catheter, removal of the device maybe adequate to resolve the candiduria without antifungal therapy.[3] It is recommended that asymptomatic catheter-associated bacteuria or

candiduria should not be treated while the catheter remains in place since this may lead to evolution of resistant flora.[14]

In the review of Lundstrom et al., management of candiduria depends on the clinical manifestations of patients. For those with asymptomatic candiduria, modification of risk factors such as catheter removal, or rational use of broad-spectrum antibiotics, is sufficient to address the condition. For those who are symptomatic with cystitis (dysuria, hematuria, frequency, urgency, and suprapubic tenderness), or those with pyelonephritis (fever, leukocytosis, costovertebral angle tenderness), treatment with fluconazole is given.[15] Thomas et al., supported this management concept for candiduria and indicated that antifungal therapy is only required in symptomatic or high-risk cases, because spontaneous resolution is common in patients with asymptomatic colonization.[16]

Based from the recommendations of other literatures and the results of this study, this study recommends the following approaches which may be done in patients with positive urine KOH:

1. For patients *without* risk factor for candiduria, who are *asymptomatic,* and with positive urine KOH, no treatment is necessary and observation or monitoring for clinical signs and symptoms suggestive of urinary tract infection maybe done.
2. On the other hand, for patients *with* risk factor/s for candiduria, who remain to be *asymptomatic*, and with positive urine KOH, a urine culture should be done to verify presence of candiduria; treatment is then directed once urine culture and sensitivities are available. Furthermore, for patients with risk factors such as presence of urinary tract abnormality or those with indwelling catheter (which were both found to increase the odds of developing candiduria in this study), an antifungal therapy maybe started pending urine culture. Treatment is then directed once urine culture results are available.
3. Lastly, for patients *with* risk factor/s for candiduria, who are *symptomatic* (e.g. febrile, frequency, dysuria) or critically ill (e.g. admitted at ICU, intubated), and with positive urine KOH, antifungal therapy with fluconazole maybe empirically started with urine collection for culture. Treatment is then continued, stopped, or directed once urine culture result and sensitivities are available, with correlation on the clinical status of the patient.

CONCLUSION AND RECOMMENDATIONS

The most common risk factors seen with candiduria are prior use of broad-spectrum antibiotics, admission to intensive care units, and use of indwelling urinary catheter. The use of indwelling catheter, presence of urinary tract anomalies, the positive leukocyte esterase in urinalysis, and elevated monocyte counts in CBC are all associated with increased odds of developing candiduria. When compared to urine culture, a negative urine KOH has excellent negative predictive value, while a positive urine KOH result will warrant further investigation with urine culture and correlation with patient's condition, prior to initiation of empiric antifungal therapy.

This study recommends that urine KOH results be approached individually with caution depending on patient's risk factors and clinical status to prevent emergence of resistant candida strains, promote rational use of antifungals, and avoid additional economic burden to the family with the use of unnecessary antifungals.

The study recommends to extend the scope of population to VLBW babies since candiduria is significant in these age group, as well as in older children (beyond infancy > 1 year old) so as to determine value of urine KOH in candiduria in this age group. Comparison of use of urine KOH for those without risk factors for candiduria against those with risk factors is also recommended for future studies.

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