

Minireview Article

Vaginal or Fecal Contamination Which Contributes to Parasite Existence in Urine Sediment

ABSTRACT

Aims: intended to deepen our knowledge to date regarding parasites that might found in urine sediment as consequences of vaginal or fecal contamination during voiding

Discussion: Urine in the bladder is a sterile waste product composed of water soluble nitrogen products. But during voiding, the passage of the urine through lower/outer portion of the urinary tract might come across normal microbiota which function in maintaining the urothelial integrity and preventing urinary tract infection (UTI), as well as promoting local immune function. Parasites and parasitic ova may be seen in urinary sediments as a result of fecal or vaginal contamination. Identification of parasites in centrifuged deposits of urine sediment is a relatively rare occurrence in clinical practice but still must be considered carefully, whether as a consequence of contamination or definite causes of infection. Two of the common parasite found in urine sediment are *Trichomonas vaginalis* and *Enterobius vermicularis*. Urine collection procedure must be explained briefly to the patient along with appropriate informed consent prior obtaining the sample.

Conclusion: The possibility of parasites being found in urine sediment remain to happen. An in-depth analysis needs to be carried out to determine whether this occurs due to vaginal or rectal contamination or whether it is due to a definitive infection occurring in the urinary tract.

Keywords: *Trichomonas vaginalis*, *Enterobius vermicularis*, centrifugation, contact, urinalysis

1. INTRODUCTION

Parasitic diseases with its characteristics of tissue tropism [1] may develop slow and causing chronic ailment that facilitate other infection to happen [2,3]. In most cases, it exhibits unclear, non-specific symptoms [4] along with non-pathognomonic physical signs [5] and make it difficult to withdraw and conclude a parasite based diagnosis [6]. Through brief medical history and physical examination, doctor can choose what lab or radiology test appropriate for the patient's subjective complaint [7]. The definite diagnosis is primarily based on the result of laboratory tests result or radiographic findings [8,9]. The commonly applied method for making correct diagnosis of a parasitic diseases include anamnesis, physical examination and laboratory/supporting examination [10], including urinalysis [11,12], that confirmed diagnosis.

Urinalysis is the examination of urine for certain pattern of physical properties, solutes, cells, casts, crystals, organisms, metabolomes or certain particulate matter [13-15]. Because the procedure and steps of urinalysis is easy to conduct, economically affordable, and productive in terms of the result, it is recommended as part of the initial examination of all adult hospitalized patients [16] and should always be recommended to be reiterated as clinically needed [17]. The rationale and technique of urinalysis are always straightforward [18]. Nevertheless, various circumstances, whether patient based (excessive exercise. Excessive food consumption etc.) or transportation based (time from collection to analysis or handling the specimen), may alter the final information obtained after analysis conducted

[19]. Sometime, urinalysis can reveal parasite during microscopic examination [11,12,16]. This mini review intended to deepen our knowledge to date regarding parasites that might found in urine sediment as consequences of vaginal or fecal contamination during voiding.

2. CHARACTERISTIC OF URINE

As study conducted by Rotter et al [20] revealed the instability of urine's composition based on time (e.g., delay in processing) [20,21], temperature [20,22] and on the addition of preservatives [22,23]. Urine contrasts substantially in ion and other solute constituent from plasma and destined to accommodates harmful and filthy substances [24] that must be initially stored for some time prior to voiding- the bladder then acts as the storage site for this waste product until higher-order centers within the central nervous system initiate the voiding process, which then in order to do so is quickly removed the urine into the urethra, located on the inferior aspect of the bladder [25]- an activity conducted when it is socially suitable to do so.

Alteration of its composition occurs as soon as it is expelled from the body through voiding [20-25]. Appropriate protocol of collection, storage, and handling are pivotal to maintaining the sample's integrity and consequently the result of analysis [26]. For the purpose of analysis, urine samples collected from the first void or "morning urine" are considered the best representative for compared to spot urine, e.g. to asses' microalbuminuria [27] due to the reason that urine accumulated overnight in the bladder is more concentrated [28], thus, provides an insight into the kidneys' concentrating capacities [29,30] and allows for the detection of trace amounts of substances in concentrated rather than diluted samples [31]. Urinary elements concentrations have any systematic relationships with gender and/or diet [31].

Other types of urine specimens may be ordered according to specific purposes, whether random collection, 2-hours postprandial or 24-hour collection where each of these three has specific purpose. It can screen and monitor drugs metabolites [32] and diagnose metabolic disorders [33] and urinary tract infections [34] and by doing so will help doctor to interpret and determine the cause of bloody urine, abdominal pains, burning sensations or whatever complaints associated with micturition [35].

Urine should be ideally examined as soon as possible [34,35], or at least within the first hour after the collection (unless preservative agent/condition added) [36] due to the instability of some urinary components (cells, casts, and crystals) [31]. If not possible, the sample should be refrigerated at 4^o C for up to 24 hours [37], which will slow down the decomposition process without the application of additional preservatives [38]. Any specimen older than 24 hours without any treatment at all should not be used for urinalysis [35-38].

There are two methods to obtain a urine specimen: non-invasive (for example, urine bags, pads, or clean catch) [43] and invasive (for example, catheter or SPA) techniques [44]. Spontaneous voiding is the main non-invasive technique, such as commonly used in cases of dysfunctional voiding [45] although other strategies may be used in children who cannot yet control their voiding, such as the use of bag urine [46]. In contrast, urethral catheterization [47] and suprapubic bladder puncture [48] are the two invasive procedures commonly applied nowadays. Urine collection preference order are as follows: cystocentesis [39], followed by sterile urinary catheterization [40], followed by mid-stream void [41] and last but no least contaminated first void [42].

The fundamental principle of either approach is to obtain the best specimen without any external contamination.

3. PARASITOLOGY URINALYSIS

Urine tests can surely make a definite diagnosis of parasitic infections [11-13]. In the era of automated machine-based analysis [13-16], these parasitic organisms are easily overlooked [14] because it is not available in the system or possible overlapping between parasite morphology [15], and though doing things manually becomes inconvenient, subjectively and clinically [16], but applying microscopic examination of urinary sediments is very important for the diagnosis of these parasitic organisms [11]. Several actively motile organisms that can be seen morphologically in urine are *Trichomonas vaginalis* with its distinctive pyriform appearance, undulating membrane and actively moving flagellae [49], Microfilaria of *Wuchereria* spp or *Brugia* spp [50] which usually found in urinary sample with specific appearance of achylous hematuria [51], *Schistosoma haematobium* eggs in urine which usually increased number of eggs is shed in the urine around midday, so an optimum urine specimen for diagnosis should be collected at noon [52], and other rare organisms such as *Balantidium coli* [53] and even *Paramecium* [54].

In the context of urinary parasites, the incidence is clinically rare compared to GI tract parasites [10-12,55,56]. The rarity perhaps due to mild or transient or even non-specific symptom which sometime make it unnoticed/neglected [57] even overlooked [58] by clinician or lab tech. Incidental finding in routine urine examination is seldom reported [12,59,60] ; even though from the laboratory work perspective, urine is consistently appraised as a flawless material for making correct diagnostic out of clinical sample due to its simpleness to collect, directly applicable and conveniences to the patient because of its non-invasiveness [11].

Parasites that may be found in urinary sediments include *Trichomonas vaginalis* [11,61], *Enterobius vermicularis* [11,62], and *Schistosoma haematobium* (Chronically infected adults pass few eggs in the urine, which are often missed when current diagnostic methods are used) [11,63]. Except for *S. haematobium*, parasites and parasitic ova are usually present in urine sediment as a result of vaginal or fecal contamination [64,65]. The next section will briefly focus on the parasitic contamination which can be detected in urinalysis.

3.1 *Trichomonas vaginalis* and how it contaminated Urine

Trichomoniasis is the commonest non-viral sexually transmitted disease [66], and it is caused exclusively by the protozoan flagellate *T. vaginalis* [49,61,66]. Although highly prevalent in specific sub population namely reproductive age of sexually active women [67], but unfortunately it was often leave unnoticed in other sub groups of potentially infected population such as male [68]. These findings raise concerns about a surge in transmission to and between sexual partners, especially if those concerned engage in promiscuous practices [69]. Trichomoniasis increases both transmission and acquisition of HIV among women [71]. Anderson et al [72] studied African women suffer from co-infection of trichomoniasis and HIV and they revealed that successful treatment for *T. vaginalis* can decrease the potency of HIV genital shedding. Trichomoniasis also reported to give rise to horrifying adverse birth outcomes of pregnancy [73], which has raised wide spread concern regarding *T. vaginalis* detection [74] and increased the need for highly sensitive diagnostic tests [75,76].

As an obligate extracellular parasite [77], this flagellate invaded and then occupied right on the facet of the epithelium of the urogenital tract, especially the cervical epithelium [78]. They acquire its energy via anaerobic pathways where they obtain glucose or maltose and used it as their own primary carbon and energy commencement [79]; its existence in this specific milieu maintained via contact dependent cytolysis of epithelial cell [80].

Trichomoniasis is a gender-related parasitic infection mainly exert influence on women with roughly calculated to have a global ratio prevalence of 8.1% among women compared to only one percent among men [81]. Surprisingly, most men are capable of briskly remove the infection, because infection only lasts transiently and self-limiting [82] a condition which made male with trichomoniasis often referred as Trichomonads asymptomatic carrier [83]. This discrepancy perhaps facilitated by the differences in the specific urogenital micro-environments which directly affecting trichomonad ability to exists and conduct its pathogenesis [77]. *T. vaginalis* benefited from an iron-rich environment in the vagina, especially during menstruation because iron, an essential nutritional and metabolic element for *T. vaginalis* parasitism available in higher concentrations during menses and seems to be entangled in the resistance to complement lysis and contributes to the immune evasion [82]. On the other hand, this parasite confronted with a belligerent and dangerous milieu of zinc-rich surrounding in the prostatic glands [84]. Zinc is a known antioxidant, anti-inflammatory and also antimicrobial chemical substance which act as first line of defense in humans [85]. A preliminary report conducted by Krieger and rein [86] revealed that men with lower amount of zinc in their prostatic secretions (<1.6 mM) are more tolerate to *T. vaginalis*, a condition which facilitate them to agonize chronic trichomonads prostatitis.

Trichomonas vaginalis replicates by binary longitudinal fission, with mitotic division of the nucleus [87]; even though study conducted by Yusof and Kumar [88] confirms that multiple modes of nuclear division do exist in *T. vaginalis* and are a precursor to progeny formation. The life cycle of this parasite possesses a trophozoite form without a cystic stage. However, the presence of non-proliferative and non-motile, yet viable and reversible spherical forms with internalized flagella, denominated pseudocysts, has been commonly observed for this parasite [89]. There is no evidence regarding existence of this parasite in the external environment outside its host; it seems that the parasite's survivability outside the host is questionable [90].

This parasite stays in the female lower genital tract [67,68,71,72] while for the male patient, it occupies the epithelial urethra and prostate [68,76]. It is responsible of causing trichomoniasis in women that is usually characterized by vaginitis with a thin purulent discharge accompanied by vulvar and cervical lesions, abdominal pain, or dysuria [91,92]. The incubation period is 5–28 days. In men, the infection can be asymptomatic or have characteristics of urethritis, epididymitis, and prostatitis [68,76,92]. Trichomoniasis is usually diagnosed by direct wet mount microscopy analysis of vaginal swab sample [93,94], which has an average sensitivity of 60 to 80 percent for detection of the motile *T. vaginalis* organisms.

In urine specimens which actually come from female patient, it is believed that the presence of *T. vaginalis* represents vaginal contamination [92], specifically is contaminated by organisms from the vaginal discharge [64,65]. Despite the fact that the midstream clean-catch technique is commonly used for urine collection [42,64], contaminated urine cultures are commonly happening with distressing regularity [64,65]. Practically in the hospital/clinic, the midstream clean-catch procedure is tedious to describe and because of that it is commonly not performed correctly by patients, costly for supplies, often embarrassing for patients, staff and or lab technician, and sometime also of unproven benefit [95]. However, all these risks must be minimized by the hospital good clinical practice [96] so that the urine collected is free of contamination and the results of the urinalysis examination are not biased. Sufficient explanation in a brief informed consent is a mandatory prior to specimen collection [97].

A clean catch urine sample or specimen [46] is one of the least invasive procedures for a urine culture or urinalysis. The clean catch method aims to intercept any microorganisms

from the skin of the penis or vagina and also from vaginal discharge from contaminating the urine specimen [98]. It is mandatory for the patient to follow the clean catch process to have accurate results from an uncontaminated sample.

Some clinics provide a clean catch kit consisting of a plastic container with a lid, a label for the patient to write their name on, and an individually clean and wrapped moist towel. Others ask that the patient to use soapy water to clean their perineal and genital region instead of providing a moist towel. Note that for best urinalysis result, it is important to collect a urine sample midstream [34]. This means that the patient should start urinating, then stop the flow, followed by put the collection container underneath their genital area and then release the urine flow again and collect it in sufficient amount (not too much or too little). The patient can usually see similar instructions to those below listed on a laminated instruction sheet posted in the clinic bathroom.

Female patients should use a packaged, moist towel to clean the vulva and perianal areas starting from front to back. Repeat with a second moist towel. On the other hand, male patients should retract the foreskin from the penis if necessary and use the packaged towel to clean the penis from the tip to the base. Repeat with second towel.

Female patients should then spread their labia with one hand and start urinating into the toilet. With the other free hand, put the urine container under the genital area to catch the stream of urine without touching any skin. Male patients should retract the foreskin if necessary with one hand and start urinating into the toilet. Then, position the urine container with the other hand to catch the stream without touching any skin.

Patients must be instructed not to fill urine to the top of the sterile container. No more than half a container is necessary. Place the lid on the container and set it on the sink or someplace stable while the patient finish urinating into the toilet. Screw the lid securely on the container and wipe it off. The patient must wash their hands and drop off the container to the laboratory as instructed.

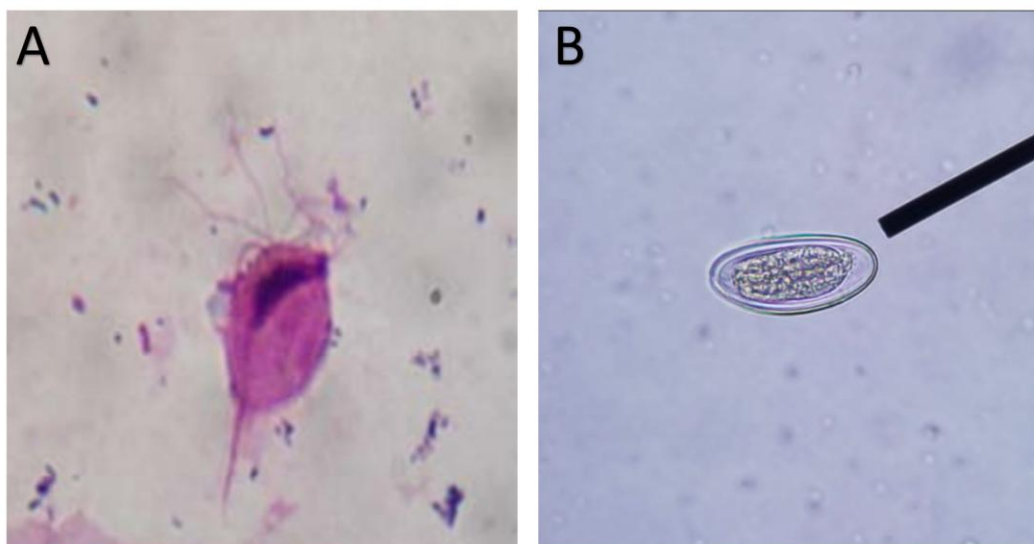


Fig. 1 A. *Trichomonas vaginalis*, 1 B. the egg of *Enterobius vermicularis* (courtesy dept. of Parasitology, Faculty of Medicine, Universitas Kristen Indonesia, Jakarta-Indonesia)

3.2 *Enterobius vermicularis* and how it contaminated Urine

Enterobius vermicularis, also called pinworm, is one of the most common nematode infections in the world. Originally, *E. vermicularis* was named *Oxyuris vermicularis* [99]. Humans are the only known natural host for this pinworm; as the result of study conducted by Paknazhad et al [100] in Iran regarding paleoparasitological evidence active enterobiasis in a female adolescent residing in ancient Tehran over 7000 years ago. Transmission highly facilitated in area where individuals living in overcrowded urban slum areas [101] and spread more easily within families where closed contact is imaginable [102].

Enterobiasis or Oxyuriasis commonly occurs in children [103]. Transmission is via fecal-oral route, either directly by hand, e.g., in children with finger sucking habit [104] (this route of infection named auto infestation) or indirectly through contaminated toys, clothing, toilette seat, bedding, eating utensil, food, or other articles, or even during sexual contact, especially oral sex with an infected person, including men having sex with men and women having sex with women [105]. Most infections are asymptomatic [99,103,105]. The cure rate is actually high (up to >90%) [106], but unfortunately recurrences are quotidian [107].

Asymptomatic Enterobiasis or Oxyuriasis occurred, approximately 30 to 40% of infested patients do not show any clinical symptoms of the disease [108]. For symptomatic patients, the most common presenting symptom is nocturnal pruritus ani and with addition of possible perineal pruritus [109]. The worms emerge from the anal canal at night till dawn in order to lay down their eggs in the perianal region; thus, pruritus is worst at those times. It is precisely under these conditions that contamination of urine can occur. For example, children who have previously experienced enterobiasis, then for other reasons (for example, suspected urinary tract infections) need to have their urine checked; If urine sampling is not done carefully and does not follow good clinical practice and correct sampling protocols, urine being collected can pass through the perineal area and become contaminated with *Enterobius* eggs.

Because urinary tract infection is particularly common in young girls and *Enterobius vermicularis* (pinworm) is one of the most prevalent worms found in children worldwide encourages Ok et al [111] to search and explore a possible relationship between urinary tract infection among young girls, with or without urinary tract infection, with Enterobiasis, they reported that urinary tract infections may be related to pinworms. When a urinary tract infection is diagnosed in young girls, cellulose tape should also be applied to both the perianal and the perineal regions on at least three consecutive occasions in order to find the primary source. The diagnosis of enterobiasis is best established by using the cellophane tape test [103,106]. The sensitivity of one single test is around 50%; however, the sensitivity increases to approximately 90% with tests performed on three different consecutive mornings [112]. Whenever a pinworm is envisaged in the perianal area or the stool, a morpho-parasiological analysis of the worm will relinquish a definitive diagnosis of enterobiasis. Because mature pinworms and eggs are not normally excreted in the stool, copro-parasitological examination is not suggested.

The urinary tract is actually rarely affected by this worm and only few cases have been scarcely reported. Sammour et al [113] report a case of bladder infestation by mature female worms of *E. vermicularis* in a woman presenting with irritative voiding symptoms. While Zahariou et al [114] reported an extremely rare manifestation of enterobius vermicularis infection since an intestinal-breeding worm is rarely found in the male genital tract of a patient complaint with mild voiding difficulties such as urgency, frequency, nocturia, dysuria, mild low back pain or perineal discomfort. The patient's prostatic secretions showed a large number of inflammatory cells and several eggs. These reports showed us that, urinary tract involvement in enterobiasis can also be an active ectopic infection, rather than

contamination and this is become the main reason to look carefully and determined whether it is caused by an active ectopic infection or just contamination.

4. CONCLUSION

The possibility of parasites being found in urine sediment remain to happen. An in-depth and careful analysis needs to be accomplish to determine whether this occurs due to vaginal or rectal contamination or whether it is due to a definitive infection occurring in the urinary tract.

CONSENT (WHERE EVER APPLICABLE)

Not needed

ETHICAL APPROVAL (WHERE EVER APPLICABLE)

Not needed

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