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Comparison between modern rapid immunochromatographic strips and traditional methods for detecting *Candida albicans, Trichomonas vaginalis, a*nd *Neisseria gonorrhea in* Kerbala and Babylon cities

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Abstract

About thirty various viruses, bacteria, and parasites **are** the causes of sexually transmitted diseases (STDs). These diseases are spread predominantly by sexual contact, including vaginal, anal, and oral sex. This study aims to evaluate the efficiency of modern immunochromatographic strips in diagnosing *Candida albicans*, *Trichomonas vaginalis*, *a*nd *Neisseria gonorrhea* in K*erbala* and Babylon cities and compare them with traditional diagnostic methods. The study started on 1 November 2022 and ended on 30 June 2023. This is a cross-sectional study constituted 300 patients (150 male and 150 female) seeking treatment for abnormal vaginal discharge, itching, dysuria, dyspareunia, urethritis, and prostatitis at private clinics in Kerbala and Babylon cities. The entire patient database was enclosed in a single questionnaire. Immunochromatography strips were used to detect *Candida*, *Trichomonas*, and *Neisseria*. The investigations achieved 144 positive cases of STDs distributed as 81, 43, and 20 positive samples for *Candida albicans*, *Trichomonas vaginalis*, and *Neisseria gonorrhea* respectively. Whilst, the remaining 156 negative samples may be referred to other causative agents. The highest infection among marital state was in married persons than unmarried. The study approved significantly increased STDs in patients who lived in urban areas than in rural areas. **In conclusion**, this study revealed that the new immunochromatography strip test was rapid and more sensitive and specific in diagnosis of *T. vaginalis*, *N. gonorrhea*, and *C. albicans* than routine conventional diagnostic tests.

Keywords: *Trichomonas vaginalis*, *Neisseria gonorrhea*, *Candida albicans*, immunochromatography

Introduction

According to previous publications, more than 30 different viruses, bacteria, and parasites are transmitted through sexual contact, including vaginal and anal intercourse. Some STDs can also be transmitted from mother to child during pregnancy, childbirth, and

breastfeeding. There are eight pathogens linked to the highest incidence of STDs. Four of those infections can be cured: trichomoniasis, gonorrhea, syphilis, and chlamydia. All of them threaten growing challenges in providing adequate services for STD prevention and control (1).

Immunochromatographic capillary flow strip detects *Candida albicans, Trichomonas vaginalis*, and *Neisseria gonorrhea* membrane proteins in 10-15 minutes. This rapid diagnostic strips that identify *Candida albicans, Trichomonas vaginalis, and Neisseria gonorrhea* antigens or nucleic acids have an advantage over microscopy and culture because they are not limited by timely transportation and specimen processing. The Rapid test has a sensitivity from 77 to 98 % with a specificity of 99-100% when performed on vaginal secretions or swabs; however, it should not be utilized in asymptomatic females or males. False positives are possible, particularly in low-prevalence groups (2, 3). A study was conducted at the Institute of Microbiology and Immunology (IMI), Faculty of Medicine, University of Ljubljana, compared the efficacy of three techniques for detecting *Candida albicans, Trichomonas vaginalis,* and *Neisseria gonorrhea* in urogenital swabs: wet mount microscopy, culture, and real-time PCR. Specimens were obtained from 75 male and 80 female patients who had STD symptoms, sexual risk behavior, or a sex partner with definite STDs, where the results of the study showed that wet mount microscopy revealed that all 155 specimens (100%) were negative. After culture, 154 (99.4%) vaginal swabs were negative, and one (0.6%) was positive; however, six specimens tested positive using real-time PCR (five vaginal swabs from five female patients with an average age of 31.4 years and one urethral swab from a 31-year-old male patient) (4). A literature review revealed scarce publications regarding the effectiveness of immunochromatographic strips in diagnosing some urogenital diseases (*Candida albicans, Trichomonas vaginalis* and *Neisseria gonorrhea*). Therefore, this study aimed to investigate the efficacy of immunochromatographic strips for diagnosis some urogenital diseases (*Candida albicans, Trichomonas vaginalis* and *Neisseria gonorrhea*) and compare it with traditional methods.

Materials and Methods

The investigation began from November 1, 2022, to June 30, 2023; a cross-sectional study was conducted on 300 patients (150 male and 150 female), who sought care at private clinics in Kerbala and Babylon cities. Each patient, whose ages varied from 20 and above, had their complete data entered into a unique questionnaire. High vaginal swabs, cervical swabs in females, and urethral swabs for males were collected from each patient using sterile swabs after a thorough clinical examination. Swabs were taken without the use of any antiseptic creams. Phosphate buffer saline was used immediately to soak each swab. All sample collection is transported in an ice pack box until processing for rapid immunochromatography assay and traditional methods such as direct microscopy and culture.

Immunochromatography assay procedure

Five drops of collection solution were added to the extraction tube, the sample swab was placed inside, and the swap was forcefully rotated on the tube's side at least 10 times to mix. Mixing the sample with the solution yields the best results, and after saturating in the extraction buffer for at least one minute, the swab was squeezed as much liquid as possible by pinching the side of the flexible extraction tube. Capillary migration requires half the specimen buffer solution in the tube. The extracted tube was tipped. The device identified the patient or control. Three drops of extracted buffer on the test cassette. After 15 minutes, the result was read.

Ethics approval

The Helsinki Declaration and later amendments for human research was applied in this study. Each patient was informed the complete information about the nature and aims of this study. The patients then signed and informed consent form. The protocol was approved by the Ethics Committee, Applied medical science college, University of Kerbala, Iraq (Ref. # 2022)**.** In order to collect data on the patients' socioeconomic status, clinical history, and sexual behavior, a questionnaire was distributed to them. Each patient provided a sample, while working with medical staff at the institution's designated time.

Exclusion criteria: The study excluded the following: no active sexual partner, less than 18 years and above 59 years, urine samples for both sexes, patient taking antibiotics, and sample drying or delayed.

Statistical analysis: All data were arranged in a Microsoft Excel (2010) sheet. Variable data were calculated using the student Chi-square (χ 2) test.

Results and Discussions

The current study was comprising 300 samples, with an equal distribution of 150 samples for each gender, the results revealed that 81 samples (27%) tested positive for *Candida albicans*, 43 samples (14.33%) tested positive for *Trichomonas vaginalis*, and 20 samples (6.67%) tested positive for *Neisseria gonorrhea*. The remaining 156 samples (52%) were negative and may be attributed to other causative agents. The distribution of the infection in the clinical samples was analyzed based on gender. The results indicated that *Candida albicans* were present in 19.67% of females, while *Trichomonas vaginalis* and *Neisseria gonorrhea* were found in 11.33% and 2% of females, respectively. Additionally, the samples above-exhibited prevalence rates of 7.33%, 3%, and 4.67% in males for *Candida albicans, Trichomonas vaginalis*, and *Neisseria gonorrhea*, respectively. The current investigation observed a distinct and statistically significant disparity (X2= 16.74, P= 0.0002) in the prevalence of certain sexually transmitted diseases among individuals of different genders. Specifically, *Trichomonas vaginalis* exhibited a higher infection rate among females than males, whereas *Neisseria gonorrhea* demonstrated a higher infection rate among males than females (Table.1).

Table.1: The Percentages of sexual transmitted microorganisms among gender.

Types of microorganisms	**Female No.**	**%**	**Male No.**	**%**	**Total**	**%**
Positive *Candida*	59	19.67	22	7.33	81	27
Positive *Trichomonas*	34	11.33	9	3	43	14.33
Positive *Neisseria*	6	2	14	4.67	20	6.67
Negative sample	51	17	105	35	156	52
Total	150		150		300	100
Statistical analysis	X^2= 16.74, DF= 2. P=0.0002 ****The colors in each tables represented the highest values**					

The table (2) shows that there is a non-significant relationship between marital status and the type of microbial infection in females. It was found that the married patients had an insignificance increased incidence compared to the unmarried, with percentages of 56.8%, 55.8% and 65% for *Candida, Trichomonas, and Neisseria* respectively. These outcomes

agree with some local studies, which found that married participants 157 (80.92%) out of 194 more prevalence of *Trichomonas vaginitis* than unmarried 13 (40.62%) out of 30 cases among females in some regions of Maysan city (5). The current study's results agree with those of other studies, which explain that the prevalence of STDs among married females increased significantly from 1.6% in 2006 to 2.5% in 2016. Likewise, STDs increased significantly among married males, from 0.5% in 2006 to 1.1% in 2016 (6). Nonetheless, the result of the current study disagrees with the previously another reported factors that significantly related to sexually transmitted disease intercourse with unmarried patients in Duhok City (7). However, the current results disagreed with the findings of the previous reported studies that revealed unmarried male and female had a much higher risk of having a positive test for *gonorrhea* and *chlamydia* in comparison to married males and female (8). The limitation of this investigation related to the fact that sexual behaviors are a part of a person's private life. Not all examined female participants felt confident enough to respond honestly, even when promised confidentially. The results agree with other research, which shows that the diagnosis of trichomoniasis was more likely to occur in married individuals (9). The results of the current investigation are compatible with another previously published study that found married males had more prevalent sexually transmitted diseases than other marital statuses. They recorded that 44.4% out of 200 patients have STDs in Nigeria (10). Moreover, the percentage of married adults in the US has decreased, whereas the percentage of single and divorced adults was increased. The shifting makeup of married Americans may influence sexual behavior and risk factors for contracting STDs (11).

Table. 2: The percentages of sexual transmitted microorganisms presented according to marital status

Types of microorganisms	Number	Unmarried No. (%)	Married No. (%)
Positive *Candida*	81	35 (43.2)	46 (56.8)
Positive *Trichomonas*	43	19 (44.2)	24 (55.8)
Positive *Neisseria*	20	7 (35)	13 (65)
Statistical analysis	X^2= 0.526,DF= 2, P=0.769		

The current results found a significantly higher prevalence of STD microorganisms in patients who lived in urban areas in Kerbala and Babylon cities compared to rural areas, with 43 (53.1%) of 81 cases in urban areas and 38 (46.9%) of 81 cases in rural areas (Table.3). The results of the current study are consistent with those of previous study in Kerbala city, which found that the prevalence of Trichomonas vaginalis in urban areas was (10%) 11/111 more than in rural areas, which was (7.43%) 9/121 cases (5). Some authors found that the infection rate of trichomoniasis was much more significant for rural residents than for those who lived in urban areas in Najaf city, which disagrees with the current study's results (12). The current investigations found all cases of Neisseria in the rural area of Babylon City and no case in the urban area. This result was consistent with another study, which found multi resistant antibiotics in patients with Neisseria (13). The prevalence of Trichomonas in Vanuatu females is significantly higher than in developed countries. Females in rural settings are less likely to have access to prevention and treatment programs for STDs (14).

Table. 3: The Percentage of sexually transmitted microorganisms according to residence

Types of microorganisms	Number	Kerbala / Babylon	
		Urban No. (%)	Rural No. (%)
Positive *Candida*	81	43 (53.1)	38 (46.9)
Positive *Trichomonas*	43	31 (72.09)	12 (27.91)
Positive *Neisseria*	20	15 (75)	5 (25)
Statistical analysis	$X^2 = 6.011$, DF = 2, P = 0.049		

Table (4) introduces the relationship of microbial infections with the level of education. The results found that patients with a low level of education are more infected at a non-significant level than patients with a primary, intermediate, or higher level of education. Furthermore, these results are compatible with a previously published study (12), which revealed that females with low education have the highest percentage of infection (18.7%). In addition, another study in Baghdad and Al-Najaf cities showed that uneducated female illiterates were more associated with the disease than other age classes (12).

Table 4: The Percentage of sexual transmitted microorganisms according to education levels

microorganisms	Number	No Education	Primary Education	Secondary Education	Higher Education
Positive *Candida*	81	27 (33.34)	20 (24.69)	18 (22.22)	16 (19.75)
Positive *Trichomonas*	43	14 (32.55)	11 (25.59)	10 (23.26)	8 (18.6)
Positive *Neisseria*	20	9 (45)	7 (35)	2 (10)	2 (10)
Total	144	50	38	30	26
Statistical analysis	$X^2 = 3.551$, DF=6, P= 0.737				

This outcome was consistent with other local research, which found that low education was associated with *Trichomonas vaginalis* more than other educational levels. It was recorded 87 (77.67%) out of 112 participants in Maysan City / Iraq (5). The current results disagree with another previously published study that showed a higher prevalence of *Trichomonas vaginalis* in secondary education than other stages. The study recorded 6 (14.63%) cases out of 41 cases in Kerbala city (5). The current investigations are compatible with some national studies that explained candidiasis's greater prevalence in low education than in other education levels in Tikrit City, which recorded 68 (59%) from 115 (15). Although education and counseling are the main strategies for the prevention and control of STDs, as well as the development of structured educational programs that must include the generation of adequate knowledge in order to change risk attitudes and practices, some STDs have been recorded in patients who have a higher education level. The WHO reports showed that half of the new cases occur in adolescents and young adults (16). The present results disagree with other studies on females with vaginal delivery recorded 155 (65%) out of 239 with secondary education, while 76 (31.7%) were low education level, and 8 (3.3%) out of 239 cases with a university degree infected with *Neisseria gonorrhea* (17). The incidence of trichomoniasis has declined sharply in developed countries in recent years, probably due to early diagnosis, the use of better diagnostic techniques, proper

management, and emphasis on behavioral change. In contrast, the infection is widespread in undeveloped countries and disadvantaged groups (18).

Conclusions

The results of the current study approved that the new immunochromatographic strip was rapid, sensitive, specific and accurate method for diagnosis STDs that caused by *T. vaginalis*, N. gonorrhea and *C. albicans* compare to routine diagnostic technique such as microscopic examination and culture tests.

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Competing interests statement

No conflict of interest related with publishing of this article.

Ethics statement

All authors approved that this research follows the journal's ethical guidelines as appeared on the journal's author guidelines page.

Author contributions

MHJ: provide the concept of the research and collected samples, worked the laboratory investigation tests, wrote and arranged the draft of the manuscript; **HRH** Contributed in writing and arranging the research; **HAM:** Contributed with the laboratory investigation **; ZAM**: Contributed in samples collection and revising of the manuscript**; SSH**: Contributed to collecting references and writing of the manuscript.

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