CLINICAL AND MICROBIOLOGICAL OBSERVATIONS ON NON-GONOCOCCAL INFECTIONS OF THE MALE AND FEMALE GENITO-URINARY TRACT*

BY

K. LEJMAN AND J. BOGDASZEWSKA-CZABANOWSKA

From the Dermatological Clinic, First Clinic of Gynaecology and Obstetrics, Medical Academy, Cracow

The non-gonococcal inflammatory conditions of the mucosa of the lower genito-urinary tract in male and female patients constitute a serious diagnostic and therapeutic problem in the gynaecological and venereological out-patient practice in Poland. Early preliminary diagnosis is important to permit immediate treatment or as an indication for further laboratory examinations.

Objects of the Present Study

- (1) To classify the vaginitides according to the macroscopic and microscopic character of the discharge;
- (2) To assess the relative proportions of bacterial, monilial, and trichomonal infections in male and female patients;
- (3) To observe the morphology of *C. albicans* and *T. vaginalis* in direct preparations;
- (4) To note the accompanying cytological phenomena.

Clinical Material

The study comprised 862 patients classified in three groups:

- (A) 600 women with non-gonococcal discharge who were examined by us before being treated in the gynaecological out-patients department;
- (B) 162 men suffering from non-gonococcal urethritis;
- (C) 100 pregnant women, clinically healthy.

Methods

Dark-field and phase-contrast techniques were used in all cases to examine the discharge. All the preparations were also stained by Gram's method, the usual and prolonged Giemsa method, and Löffler's methylene blue stain. Cultures from 120

female and 28 male patients were inoculated on several bacterial media, as well as on Sabouraud's maltose agar; the yeast-like fungi were identified according to routine mycological procedures. The diagnosis of *Trichomonas vaginalis* infestation was made in all cases by the dark-field and phase-contrast methods and by stained smears. Cultures for *T. vaginalis* were taken only in selected cases.

Results

(A) A preliminary analysis of the 600 female patients suffering from non-gonococcal discharge (Group A) is shown in Table I.

TABLE I
FINDINGS IN GROUP A

General Microbiological Features	Number of Patients	Per cent.
(I) Bacterial discharge	198	33
(II) Monilial discharge	108*	18
(III) Trichomoniasis	288	48
(IV) Spirochaetal discharge	3	0.5
(V) Abacterial discharge	3	0.5
Total	600	100

* In 18 cases together with *T. vaginalis*.

(I) *Bacterial Discharge*.—198 of the 600 female patients (Group A) with non-gonococcal discharge, showed only bacterial flora in the smears. Repeated observation made it possible to distinguish the several types of discharge shown in Table II (opposite).

To complete the data given in Table II it should be added that the cases of Type (a) were young women and girls at puberty. The discharge of Type (b) usually showed different strains of non-gonococcal *Neisseriae*, *Corynebacterium pseudodiphtheriae*, *Haemophilus vaginalis*, and various

* Paper read at the European Symposium of the I.U.V.D.T. at Cracow, Poland, in September, 1960.

TABLE II

TYPES OF BACTERIAL DISCHARGE IN THE 198 FEMALE PATIENTS IN GROUP I

Type of Discharge	Number of Patients	Percentage of whole Bacterial Group	Percentage of whole Group (A)	pH*	Clinical Symptoms							Subjective symptoms: vulvar pruritus, burning, tenesmus
					Vulvo-vaginitis	Cervicitis	Urethritis	Adnexitis	Bartholinitis	Erosion of the cervix	Intertrigo	
(a) White, epithelial, with normal flora† ...	14	7.1	2.3	4.5 to 4.8	—	—	—	—	—	—	—	—
(b) White, epithelial, with single or mixed flora ...	67	33.8	11.2	4.8 to 5.5	12	3	2	—	—	2	—	9
(c) Greyish-white, greyish, or greyish-yellow profuse epithelio-purulent discharge with single or mixed flora ...	85	42.9	14.2	5.2 to 7	24	6	7	3	—	4	3	34
(d) Purulent and sero-purulent profuse discharge with cellular detritus and mixed flora ...	32	16.2	5.3	5.2 to 7.5	32‡	5	6	4	1	5	3	32§
Total ...	198	100	33		68	14	15	7	1	11	6	75

* Tested by colorimetric method. † A small number of leucocytes frequently present. ‡ Twenty showed colpitis granulosa.

§ Two complained of abdominal pain.

unidentified Gram-negative and Gram-positive bacilli. Type (c) was characterized by the presence of *Staphylococcus albus* and *aureus*, enterococci, haemolytic and non-haemolytic streptococci, and different strains of non-gonococcal *Neisseriae*. *Escherichia coli*, *Proteus vulgaris*, *B. pyocyaneum*, *Corynebacterium pseudodiphtheriae*, *Gaffkyia tetragena*, and various Gram-positive bacilli were also observed in particular cases in different combinations. The discharge of Type (d) showed almost the same bacterial flora, but the enterococci and streptococci were more frequent.

(II) *Monilial Discharge*.—Fungal elements were found in 108 (18 per cent.) women of Group (A). *Candida albicans* was identified in 98 cases and other *Candida* in six cases. In four cases we failed to identify the fungi observed.

Almost all cases infected with *C. albicans* showed the clinical symptoms of vulvo-vaginitis with profuse, dense, caseous discharge, often forming greyish-white colonies on swollen vaginal mucosa of bluish hue. The pH of the discharge ranged from 6 to 8. The microscopical picture of the wet preparations or of the stained smears corresponded with the clinical observations. Fungal elements were present in all preparations, but abundant pseudomycelium or mycelial threads growing into the

vaginal epithelium (Fig. 1, overleaf) were seen, particularly in acute cases. However, in other acute cases, blastospores were prevalent.

Acute monilial vaginitis was microscopically characterized by profuse desquamation of the vaginal epithelium and the presence of leucocytes. *C. albicans* was found in the urethral specimens of thirty patients (28 per cent. of all monilial infections) and in the cervical discharge of 28 (26 per cent.).

The accompanying bacterial flora in cases of monilial vaginitis was scanty, mostly Gram-negative and Gram-positive bacilli and cocci. Eighteen patients (16.6 per cent.) had acute infection with both trichomonads and *C. albicans*, with abundant mixed bacterial flora. All these patients showed the clinical symptoms of vulvo-vaginitis with profuse, purulent discharge.

Thirty women (27.7 per cent.) of the cases with monilial discharge were pregnant, compared with only 2 per cent. of the remaining 492 patients of Group (A). It is evident that infection with *C. albicans* is the commonest aetiological agent of vaginal inflammation and monilial discharge in pregnant women.

(III) *Trichomoniasis*.—Trichomonads were found in the vaginal specimens of 288 (48 per cent.)

FIG. 1.—Mycelium of *Candida albicans* growing into islets of desquamated vaginal epithelium. Phase-contrast, $\times 1,500$ approx.

women suffering from discharges. In 214 of these patients acute and subacute infection was recognized (Table III).

TABLE III

INCIDENCE OF *T. VAGINALIS* INFESTATION

Trichomoniasis	Number of Patients	Percentage of Group (A)	<i>T. vaginalis</i> present in		
			Vagina	Cervix	Urethra
Acute and Subacute ...	214	35.7	214	26	55
Chronic ...	74	12.3	74	8	22
Total ...	288	48	288	34	77

The acute and subacute trichomonal vaginitis was characterized by profuse, often foamy, purulent, foul-smelling discharge, greyish-yellow or greyish-white in colour. Almost all patients also showed vulvitis of varying intensity, sometimes with severe pruritus. The pH of the discharges was between 6 and 8. The number of flagellates in one field in stained smears ranged from one to ten. In cases of chronic infestation, the number of trichomonads was smaller; and they seemed to play a minor role

compared with the usually very abundant, mixed bacterial flora.

The development of mixed bacterial flora with the consecutive rapid disappearance of lactobacilli, even in the presence of small numbers of flagellates in the discharge, should be regarded as a characteristic feature of trichomoniasis in women. 42 per cent. of the patients suffering from trichomonal infestation also showed streptococci, which were noted in only 19 per cent. of the whole group with bacterial discharge (I).

(IV) *Spirochaetal Discharge*.—This was observed in three patients (0.5 per cent.). In two cases the spirochaetes were associated with fusiform bacilli, but with no necrotizing lesions.

(V) *Abacterial Discharge*.—This was noted in three patients (0.5 per cent.).

(B) An analysis of the 162 cases of non-gonococcal urethritis in male patients (Group B) is shown in Table IV (opposite).

In the cases of bacterial urethritis the flora chiefly comprised staphylococci, diplococci, different strains of non-gonococcal *Neisseriae*, and only

TABLE IV
FINDINGS IN GROUP B

Type of Urethritis	Number of Patients	Per-centage	Days of Incuba-tion	No. of Patients with Clinical Complications
Bacterial ...	73	45.1	4-17	11
<i>C. albicans</i> * ...	2	1.2	Undeter-mined	2
<i>T. vaginalis</i> ...	20	12.3	7-14	2
Spirochaetal†	2	1.2	Undeter-mined	0
Abacterial ...	65	40.2	10-24	4
Total ...	162	100		19

* Together with monilial balanoposthitis. The patients had been infected by their pregnant wives.

† Caused by *Spirochaeta balanitidis* and co-existent with endo-urethral condylomata.

rarely streptococci. *Corynebacterium pseudodiphtheriae* was relatively common. Subacute epididymitis was noted in four (in two cases co-existent with phlebitis dorsi penis). Five patients suffered from chronic prostatitis and one from cowperitis.

In the group with *T. vaginalis* infestation two cases showed a great number of flagellates in the para-urethral ducts. Abacterial urethritis was complicated by subacute epididymitis in four cases, and enlargement of inguinal lymph nodes was relatively common among all cases of non-gonococcal urethritis.

Biomorphology of *Trichomonas vaginalis*

A search for cyst formation and the multidivision phenomena of *T. vaginalis* gave negative results. Only bi-division of these flagellates was noted. Occasionally, relatively large forms of *T. vaginalis* were observed in wet preparations among living parasites (Fig. 2) and in stained smears (Fig. 3), resembling to some degree Lapierre's drawings of cysts (Lapierre, 1957). In our opinion, however, these represent initial stages before bi-division.

In four cases of acute or subacute male urethritis, big groups of *T. vaginalis* were seen to be packed closely together (Fig. 4, overleaf). One patient showed these groups in the para-urethral duct.

This phenomenon may be regarded as a result of multiplication of the parasites in the urethral diverticula not rinsed off by the urinary stream. However, spontaneous grouping of these flagellates cannot be excluded in such cases.

Phagocytosis of *T. vaginalis* in the vaginal and urethral discharge in women was rarely observed. However, it was seen more frequently in male patients and in some cases to a significant degree.

FIG. 2.—Large round living form of *T. vaginalis* from vaginal discharge. Phase-contrast $\times 1,500$ approx.

FIG. 3.—Large round form of *T. vaginalis* from vaginal discharge, corresponding to that seen in Fig. 2. Löffler's methylene blue. $\times 1,500$ approx.

In one, the number of protozoa engulfed by leucocytes amounted to 83 per cent. of the total parasites observed in the smears.

While the different stages of phagocytosis can be well observed in stained smears the best method is by the phase-contrast technique (Figs 5 and 6, overleaf).

In stained preparations the structure of the engulfed parasites at the beginning of the process of phagocytosis shows little or no damage. In the latter stages, however, the protozoa show different involuntary changes, e.g. homogenization and partial resorption.

FIG. 4.—Group of 38 trichomonads from specimen of male urethral discharge. Prolonged Giemsa staining. $\times 900$ approx.

FIGS 5 and 6.—Reverse process of phagocytosis of *T. vaginalis* by a neutrophilic leucocyte, observed by the phase-contrast microscope in a moist-slide preparation of vaginal discharge. The engulfed parasite (Fig. 5.) still waving vigorously with its flagella and undulating membrane, releases itself from the surrounding protoplasm of the leucocyte, after 20 minutes' observation at room temperature (Fig. 6). $\times 1,500$ approx.

(C) The group of 100 healthy pregnant women had no symptoms of vaginitis and were seen for routine antenatal examination. The vaginal secretion was composed of desquamated epithelium, sometimes with small numbers of leucocytes. An analysis of the results of the examination of the stained smears is shown in Table V.

Yeast-like fungi in direct moist slide preparations and in the stained smears were noted in 22 cases. The specimens of vaginal secretion cultured on Sabouraud's media gave positive results in thirty cases (19—*C. albicans*; 8—*C. tropicalis*; 2—*C. krusei*; 1—*C. stellatoidea*).

TABLE V
FINDINGS IN GROUP C

General Microbiological Features	Number of Patients
Normal Flora (lactobacilli exclusively) ...	53
Scarce Mixed Flora	17
<i>Candida albicans</i> * + Mixed Flora ...	19
<i>T. vaginalis</i> + Mixed Flora	8
<i>T. vaginalis</i> + <i>C. albicans</i> + Mixed Flora	3
Total	100

*and other species of this group.

Discussion

The increased incidence of clinical complications in the group of women with purulent discharge (Table II), characterized by the development of profuse mixed bacterial flora, suggests the possibility of an aetiological role of these micro-organisms. On the other hand, the flora may only develop secondarily to primary immunological disturbance of the mucosa. However, even the secondary flora can exert a pathogenic effect on the mucosa in many cases. The results of the examination of healthy pregnant women confirm the opinion of Gardner, Dampeer, and Dukes (1957) that *C. albicans* may be regarded as a normal inhabitant of the vagina and that the development of clinical moniliasis depends on environmental conditions. Thus positive culture results may arise without clinical candidiasis, as the colony of these fungi may develop from even a solitary blastospore. The diagnosis of clinical candidiasis should always be confirmed by examining direct (moist-slide or stained) preparations of the discharge, searching for the characteristic behaviour of fungal elements, indicating their active relation to the vaginal epithelium.

The correlation between *T. vaginalis* infestation and the extensive development of a mixed vaginal flora seems to be proved. The phagocytosis of *T. vaginalis* by leucocytes, observed in particular cases of male urethritis, requires further study.

Summary

The non-gonococcal vaginitides are classified according to the macroscopic and microscopic

appearance of the discharge. The value of examining direct (moist-slide or stained) preparations is emphasized. The relative value of smears or moist-slide examinations and of culture examinations in cases of *C. albicans* infection is discussed. Several phenomena observed in the course of trichomonal infestation in the male and female, such as large round forms of *T. vaginalis*, large groups of *T. vaginalis*, and phagocytosis of *T. vaginalis* by leucocytes, are described.

REFERENCES

- Gardner, H. L., Dampeer, T. K., and Dukes, C. D. (1957). *Amer. J. Obstet. Gynec.*, 73, 1,080.
Lapierre, J. (1957). "Introduction à l'étude des formes de résistance du *T. vaginalis* Donne", in "Les infestations à trichomonas", 1er Symposium Européen, Reims, 1957, p. 271. Masson, Paris.

Observations cliniques et microbiologiques des infections non-gonococciques des voies génito-urinaires de l'homme et de la femme

Résumé

Les vaginites non-gonococciques sont classifiées d'après l'apparence macroscopique et microscopique de la perte. L'importance de l'examen de préparations directes (lames humides ou colorées) est soulignée. L'importance relative des examens des frottis ou des plaques humides et des examens des cultures en cas d'infection par *C. albicans* est discutée. Plusieurs phénomènes observés au cours de l'infestation par le *Trichomonas* de l'homme et de la femme sont décrits, parmi lesquels il faut citer les grandes formes rondes de *T. vaginalis*, les grands groupes de *T. vaginalis*, et la phagocytose de *T. vaginalis* par les leucocytes.