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laboratory established in 1924 although it seemed to be the only way in which to raise the standard of testing in this country; it was then almost literally true that there were as many ways of conducting the Wassermann test as there were laboratories practising it, the attitude of the average pathologist being that his method was as good as any and better than most. To extend this control would require a great increase in staff but if the Society expressed the view that there should be a strong control, it might help his successor to continue the good work.

Major Lane had mentioned the modification of tests. He agreed that serologists would modify a given technique; of course they would and it would be wrong if they were hidebound but he would ask above all that everyone should first learn a method which had stood up to strict examination, and then, if he wished to modify it, he should try the modification in parallel with the original test on at least a few hundred sera. It would be a good thing, as was suggested before the last war, to circulate problem sera.

He was grateful to Lt.-Col. Bensted for reminding him about typhus. Another point that had been underlined was that by Lt.-Col. King and others that the positive reaction was simply an indication for further investigation.

TRICHOMONAS VAGINALIS INFESTATION IN WOMEN*

By SQUADRON-LEADER R.A.F.V.R. JEAN H. MORTON, M.D., D.P.H.

To those of us who are constantly employed in the investigation of venereal disease in the female, and of gynaecological complaints, the question of Trichomonas vaginalis infestation and its treatment is becoming more insistent. I think that all will agree that in the past we have regarded this condition only as a minor gynaecological complaint, which is equally annoying both to the patient and to the doctor whose prestige is likely to suffer because he frequently fails to achieve a permanent cure. Therefore we should regard this condition in women with as much gravity as we do a gonococcal infection and bear in mind that any acute infection of the vagina and cervix may ascend to the Fallopian tubes with subsequent complications. Hence I would claim that this is no mean gynaecological entity.

Though Trichomonas vaginalis infestation has become a well recognized condition only during the last ten years, it was first described by Donné as a cause of leucorrhoea in women as far back as 1836.

A great deal of literature has been written on the subject much of which is contradictory, and I have based this paper on the more logical findings and my own observations and conclusions.

Morphology

The *Trichomonas vaginalis* is a lozenge-shaped protozoon from ten to thirty μ in length with a large macro-nucleus at the anterior end; anterior to this is a smaller nucleus, the blepharoplast, which is connected to the larger nucleus by a fine filament, the rhizoplast. Extending from the blepharoplast are fine hair-like flagella capable of rapid movement. The number of the flagella varies according to the species of trichomonas. Each flagellum has a central fibre, the axoneme, covered by a fine layer of bodily substance.

One axoneme is more closely bound to the body of the protozoon often throughout the whole length, by a layer of bodily substance from the blepharoplast backwards, which is very extensile, communicating to this membrane a wave-like motion, hence the name of the undulating membrane. In some protozoa the axoneme extends beyond the undulating membrane forming a posterior flagellum, but the *Trichomonas vaginalis* has no posterior flagellum. The axostyle, which extends from the macro-nucleus throughout the body, can be protruded and retracted and can fix itself by means of a sticky substance discharged from its point. The flagella cause currents and waft particles of food towards the organism. Movement of the organism is effected by means of the flagella and the undulating membrane; amoeboid movement may also be seen microscopically amongst pus cells. Penetration of the epithelium of a mucous membrane has been

* An address read to the Medical Society for the Study of Venereal Diseases, May, 1943.
THE BRITISH JOURNAL OF VENEREAL DISEASES
demonstrated in a section of the bowel (Wenyon, 1926). The raised papillae
seen in the vagina in infected women may be due to this penetration. The
protozoon feeds like an amoeba and multiplication is by simple fission. Various
species of trichomonads can be distinguished from one another by the number of
their flagella which may amount to three, four or five.

The two species with four flagella found in human beings—Trichomonas
buccalis in the mouth and Trichomonas vaginalis in the vagina—may be distin-
guished from one another by the fact that Trichomonas vaginalis has an
undulating membrane extending along two-thirds of its body differing from
Trichomonas buccalis which has an undulating membrane along the whole of its
length, neither having a posterior flagellum.

Dobell (1934) undertook experiments which showed that a single species of
trichomonads can live in different hosts, for example monkeys and men, also in
different situations such as the intestines and vagina and that their presence
may not cause any symptoms. The trichomonads infecting man are, however,
selective both in their site and in the medium in which they grow, the normal
medium of the Trichomonas vaginalis being the vagina.

Incidence
In approximately 400 of my consecutive patients the Trichomonas vaginalis
was present, in sixty per cent of those with a vaginal discharge. Also forty-two
per cent of the gonorrhoea cases were infested with the Trichomonas vaginalis.

Karnaky (1940) investigated 500 consecutive patients in a gynaecological
out-patient clinic. He found thirty-seven per cent harboured T. vaginalis of
which forty-six per cent were negro women and twenty-four per cent white women.
Their ages ranged from eleven to fifty-two years, the majority being between
nineteen and thirty-three years.

Sites of infection with Trichomonas vaginalis in humans

Females clinically normal.—I have found T. vaginalis in the vagina of normal
healthy women who have had no complaints or any treatment, and also in the
normal vagina of women who have been previously treated for trichomonas
and who were reporting some months after cessation of the treatment. And I
am sure that if more extensive examinations could be made on normal healthy
vaginæ, it would be found that a fairly large percentage of women harbour
small numbers of T. vaginalis.

Females clinically infected.—I have recovered T. vaginalis from the following
sites: vagina, cervix, urethra, Bartholin ducts and in the urine; also from the
rectum in three cases with a very heavy infection of the vagina, the rectum
showing infection of the mucous membrane and large blobs of pus. Under the
microscope the Trichomonas vaginalis recovered from this pus was identical
with that recovered from the vagina of the same patient. Nitschke (1936) reported
that the parasite can travel from the urethra to the bladder causing cystitis, also
Lewis and Carroll (1928) reported extension of infection from the urinary tract.

Males.—Liston and Lees (1940) state that males may become infected from
females suffering from Trichomonas vaginitis and may subsequently infect or
re-infect females. In males the trichomonas is generally found beneath the
prepuce or in the anterior urethra, but less commonly in the posterior urethra,
from whence it may pass along the urinary tract to the bladder and kidneys
causing pyelitis (Rosenthal, 1931) or by the genital tract to the prostate, seminal
vesicles and testis (Liston and Lees).

A stricture may result from a persistent posterior urethral infection. Owing
to its dryness, the male urethra is not supposed to be a kindly habitat for the
parasite, but when infected the symptoms of urethritis are only slight.

Incidence in males.—Liston and Lees examined 400 men, attending a V.D.
oclinic, not suffering from syphilis. Only sixteen of this number were found to
have T. vaginalis in some part of the urogenital tract and with one exception,
a case of gonorrhoea, the T. vaginalis was found to be the sole cause of the
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urethritis. The result of this investigation disposes of the surmise that T. vaginalis infestation is a venereal disease.

Comparison of conditions in the normal and infested vagina

There are three outstanding differences between the contents of the normal adult vagina and the infested vagina in the inter-menstrual period.

1. The epithelial lining.—The adult female vagina is lined with squamous epithelial cells and in a film of the vaginal contents stained with Best’s Carmine or Iodine the epithelial cells are found to be full of glycogen. At birth the vaginal epithelial cells are also found to contain glycogen; but all these are shed within the first month of life and are replaced by glycogen free cells. With the onset of puberty glycogen is again found and persists until some years after the menopause.

The presence of glycogen in these cells has been attributed by some authors (Liston, 1940) to oestrogenic substances, obtained in the case of the baby from the maternal circulation, hence lost soon after birth and again intrinsically coming into action at puberty and lasting until after the menopause. The important point to note is that T. vaginalis infestation is found only in adult women, but not in children and rarely after the menopause. I, myself have never seen a child infected, though one case of a child of five weeks (Brady and Reid, 1942) and another of three years (Cornell, Goodwin and Matthies, 1931) have been reported. It would appear, therefore, that the presence of glycogen in the vaginal epithelial cells is necessary for the development of the trichomonas.

There are two possible explanations of why the Trichomonas vaginalis requires glycogen. Staining methods have shown that the trichomonas feeds on glycogen. Also glycogen is required to maintain a certain degree of acidity in the vagina which is favourable for the development of the organism. In adults this acidity is dependent on the production of lactic acid from glycogen by the action of Döderlein’s bacillus.

In children and women after the menopause the neutral reaction of the vagina is due to the absence of glycogen, hence the absence of the trichomonas in such age groups.

2. The vaginal flora.—This has been found to fall into three types according to the acidity of the vagina.

Type (1) This flora is found in normal healthy adult women with the highest acidity, namely, a pH of 3·8 to 4. A film of the vaginal contents shows large numbers of Gram positive bacilli—Döderlein’s bacilli. Very few other organisms and trichomonas are supposed to live in this acidity.

Type (2) This flora in a less acid vagina consists chiefly of Gram positive diphtheroid and Gram negative bacilli. Thrush fungus and trichomonas will live in this acidity.

Type (3) As the medium is still less acid the flora consists of a much greater variety of organisms, but mainly small Gram negative cocci and bacilli. This is supposed to be the most favourable medium for the growth of T. vaginalis.

3. The hydrogen-ion concentration of the vagina.—The pH of the normal vagina in the adult female varies between 3·8 and 4 with its accompanying flora rising with a fall in acidity till it reaches the neutral point of 7 which is the pH found in the vagina of children and women after the menopause. Liston in an analysis of 100 cases of trichomonas infestation found that sixty of them gave a pH of between 5 and 6, the other forty having variations from the normal. I would stress this question of the pH, the normal and abnormal, as I think the whole question of infestation and its treatment depend upon it.

Signs and symptoms

Trichomonas vaginitis may be acute, subacute, or chronic. The complaint common to all is a vaginal discharge of varying amount, from very slight to very profuse, and is frequently accompanied by a sensation of itching of the vulva. Frequency of micturition is also present in a number of cases. Examination of the patient in the lithotomy position shows the usual signs of inflammation of the genital tract, varying according to the degree and chronicity of the infection, but
exhibiting certain characteristic signs. A well-marked case presents injection of the membranes of the introitus and a urethritis, but the most outstanding feature of all is the vaginitis, which this condition essentially is. The vagina has a characteristic appearance, sometimes described as "strawberry," due to the raised papillae, most marked in the upper third, and rough to palpation. The discharge varies from a purulent to a muco-purulent consistency and is frequently frothy. This frothiness has been attributed to the *Vibrio alcaligenes*. The outer margin of the cervix also takes part and exhibits small punctate eroded areas, presenting a granular appearance. The presence of an extensive cervical erosion may be found and probably this has preceded the infection.

Complications such as cystitis and pyelitis have already been mentioned, and a proctitis in three cases. Certain conditions coincident with but not caused by *T. vaginalis* infestation are worthy of note.

*Salpingitis.*—If the history of infestation is carefully investigated a certain number of patients will complain of lower abdominal pain and menorrhagia dating from the onset of the discharge. Though pelvic examination is sometimes negative in such cases, I would suggest that a mild salpingitis has occurred and this is borne out by the fact that a small number of my own patients while in hospital have had a definite salpingitis, one of them developing pelvic peritonitis needing subsequent operative treatment.

*Bartholinitis.*—This also, has been present in a certain number of cases.

*Condylomata Accuminata.*—During the past year I have had four cases of trichomonas infestation associated with a massive growth of condylomata accuminata of the vulva. The patients noted that the growth developed subsequently to the discharge.

**Isolation of Trichomonas vaginalis**

The most suitable method of keeping samples of the discharge will depend upon the time and microscopic facilities available. If immediate examination is possible take a loopful of discharge from the posterior or lateral fornix. Place the secretion on a slide and mix if necessary with normal saline, apply a coverslip, and preferably examine by dark ground illumination which is easier than the ordinary illumination. The edges of the coverslip may be sealed with paraffin wax and the slide can then be set aside for some hours before examination.

When immediate examination is not possible, pipette some discharge into a test tube containing a small quantity of normal saline and incubate until time for examination is available. If desired, films can be stained by Gram's method or with Leishman's stain. Repeated examination is sometimes necessary before the parasite is discovered.

The following culture media is used by Liston:

\[ R \]

\[
\text{Sod. Chloride} \quad ... \quad 6 \text{ gms.} \\
\text{Pot. Chloride} \quad ... \quad 0-1 \text{ "} \\
\text{Calc. Chloride} \quad ... \quad 0-1 \text{ "} \\
\text{Sodi. Bicarb.} \quad ... \quad 0-1 \text{ "} \\
\text{Distilled water} \quad ... \quad \text{to 1 litre.}
\]

The solution is sterilized and cooled and 2-5 grammes of Loeffler's dried blood serum and twenty cubic centimetres of fresh human serum are added. The solution is passed through a sterile bacterial Steitz filter. Subculture is advised every sixth hour as the plates tend to become overgrown with bacteria.

**Type of patient**

On looking back over a number of years and on consulting my present records, certain facts stand out. The majority of women infected with *T. vaginalis* are under the age of thirty years. Very few virgins have come to me for treatment of this condition; one frequently gets the history that the vaginal discharge started after sexual intercourse, which suggests a traumatic factor in the causation. This condition is more prevalent amongst women with a low standard of personal hygiene, and whose habits are somewhat promiscuous.
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I have found that *T. vaginalis* was present in forty-two per cent of the gonorrheal cases and I believe that this figure would have been higher if I had continued the search. Therefore it is well to bear in mind that a certain number of cases treated for trichomonas infestation will reveal on completion of treatment, a gonococcal infection which had previously been sought for without success.

Mode of infection

This has been attributed to bath water, towels and clothing of infected persons, but I think this mode of spread must be rare, as the parasite requires a certain degree of moisture and heat to keep it alive. In civilian life I was struck by the fact that women of low economic standards were more frequently affected and I thought that vitamin deficiency might play a part, but this cannot be said of women in the Armed Services, who today are having a mixed and well balanced diet which is far superior to that of a large percentage of the population in pre-war days.

One author maintains that *T. vaginalis* appears in the vagina merely as a secondary invader, in conditions caused by other organisms, e.g. *B. coli* and the gonococcus.

Hibbert and Falls (1938) thought that an independent causative agent other than *T. vaginalis* might produce the clinical syndrome and that a symbiotic relationship between the trichomonas and a streptococcus may be the causative factor. They also suggested the failure of normal carriers of the parasite to show clinical signs may be due to the immunity established against these two factors.

Conclusions

1. I would suggest that a large number of women are carriers of *T. vaginalis* which, when the vaginal pH is normal, are of no significance.
2. In this group of women, *T. vaginalis* becomes pathogenic, only when the pH of the vagina has been primarily altered by other invading organisms, for example, gonococci, *B. coli*, streptococci and others. Trauma, uncleanliness, the presence of cervical erosions and general debility after an illness are adjuvant factors.
3. An external invasion of *T. vaginalis*, the source being difficult to state, may take place in any unhealthy vagina with an abnormal pH.
4. The condition which is widespread amongst the female community, both in civilian and service life and which is of medical and economic importance (for many hours are spent in hospitals and treatment centres) is well worthy of further investigation.

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Ante-natal serological tests for syphilis

The desire in many medical circles for the application of routine tests for syphilis to all pregnant women has recently received the approbation of the executive committee of the National Association of Maternity and Child Welfare Centres.

The opinion of the executive committee of this Association was expressed in the resolution adopted at its meeting on 24th May, which considered that the physical and social disadvantages to a child with congenital syphilis are such that everything practicable should be done to diagnose the condition at the earliest possible time, including the routine application of syphilis blood tests to the mother during pregnancy in order that effective treatment may be given. A resolution was also adopted that the Ministry of Health should give practical effect to this by propaganda and by stimulating welfare authorities, voluntary hospitals and clinics.
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*Br J Vener Dis* 1943 19: 121-125
doi: 10.1136/sti.19.3.121

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