

## THE TREATMENT OF TRICHOMONAS VAGINALIS VAGINITIS WITH THE LACTOBACILLUS\*

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It is with some hesitation that we present another method of treating vaginitis due to trichomonads. Time and further clinical studies have shown, in the case of so many of the methods already advocated, that the claims made for them were unwarranted.

Some one has truly said that a trichomonas infection is an ideal condition to treat in transients. By this it is meant that when an out-of-town patient consults a physician because of an irritating discharge caused by trichomonads he can usually give prompt temporary relief and acquire her gratitude. The trouble is that so often the symptoms return with the next or subsequent menstrual periods.

The return of symptoms with menstruation is due probably to the decreased acidity of the vagina which occurs at this time. In many instances a few trichomonads survive the prescribed treatment and continue to live in the upper vagina, and with the decrease in acidity they increase in number, set up an inflammation, and cause a return of the vaginal discharge.

The importance of these recurring exacerbations of trichomonas vaginalis vaginitis cannot be overemphasized, for in women during menstrual life the trichomonas vaginalis is probably the commonest cause of leukorrhoea. There is, in our minds, not the slightest doubt that this statement is true when one is referring to the type of women seen in private practice and, we believe, it is also true for dispensary patients, although gonorrhoeal endocervicitis in this latter type of patient runs the trichomonas a close second as a cause of leukorrhoea. Moreover, the trichomonads must always be thought of and looked for when a woman complains of pruritus and dyspareunia.

Trichomonads cause a vaginitis and little, if any, endocervicitis. The characteristic lesion is most often seen in the upper posterior part of the vagina just behind the cervix. One not infrequently sees in this area minute red spots, giving the upper posterior vaginal wall a strawberry-like appearance.

Some of those who have written about trichomonas infections have stated that the discharge often contains bubbles and is apt to have a slight greenish tint but personally we have seldom observed these peculiarities in the discharge. In our experience there is nothing that is characteristic in the appearance of the discharge. It may be thick or thin, yellow or white. It is, however, apt to cause much more irritation than does the discharge caused by endocervicitis.

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The trichomonads occasionally invade the urethra, Skene's and Bartholin's glands. Cases of salpingitis, due to the trichomonas vaginalis, have been reported but, although we have seen many hundreds of patients with a leukorrheal discharge due to these organisms, we have never seen a case of salpingitis for which we thought they were responsible.

The trichomonas is a protozoa with actively moving flagella at one extremity and an undulating membrane. The latter cannot always be seen as easily as the flagella. These organisms are larger than an ordinary pus cell and yet smaller than the epithelial cells which line the vagina and which can always be seen in vaginal smears. The trichomonads are very motile. Under the microscope their flagella can be seen moving rapidly. When the protozoa happen to be caught under a mass of epithelial cells they often make the whole clump of cells move.

The diagnosis is made by demonstrating the organisms with the microscope. No special apparatus is needed. All that is required is a little normal salt solution and an ordinary microscopic slide. In some articles you will read that hollow ground slides should be used in studying the trichomonas but they are not necessary. The technic for demonstrating these organisms is as follows: The gloved finger is inserted high in the vagina behind the cervix and a drop of the secretion taken on the gloved finger. This is mixed with a little normal salt solution and the preparation examined at once. The organisms can be seen under low or high grade power. It is not necessary to stain them.

In some women, smears examined in the intermenstrual period may fail to show the organisms, while those taken immediately before or after a period will reveal them. In fact, in some instances the organisms can more easily be demonstrated if the patient is examined when she is actually menstruating.

The diagnosis is often missed for two reasons. The first is the same reason that explains why so many conditions remain undiagnosed. It is that they are not thought of. The second reason that explains why physicians sometimes fail to make the diagnosis is that while they think of trichomonas vaginalis as a possible cause of leukorrhoea they think of it too minutes too late. That is what often happens. A woman comes to a doctor complaining of a leukorrheal discharge. He thinks of endocervicitis, polyps, carcinoma and many other gynecologic conditions as being the possible cause of the discharge. He at once puts green-soap or some other lubricant on his fingers and makes a pelvic examination. After his examination has failed to demonstrate any of the conditions mentioned above he then thinks of the possibility of a vaginitis due to the trichomonas vaginalis, takes smears, looks at the material under the microscope and fails to see the organisms. This may be due to the fact that the green-soap on his fingers killed the trichomonas which were in the superficial tissues and by which the diagnosis could have been made. We feel that it is advisable to take smears for trichomonas vaginalis as the first step in the study of all patients complaining of leukorrhoea, pruritus, and dyspareunia.

Another factor that sometimes interferes with the diagnosis of trichomonas

infections and also of gonorrhoea is that many women through a natural sense of cleanliness will take a douche immediately before coming to a doctor's office and thus prevent the physician from making the diagnosis. It is well to ask every woman who comes to you complaining of leukorrhoea when she took her last douche.

Until we began working on the method of treatment which we are now presenting, we had, as most of us have, tried one after another of the usual procedures recommended for trichomonas vaginalis vaginitis. All of them were helpful in some cases. However, as has been the experience of so many gynecologists there were numerous cases in which a recurrence of symptoms occurred following the menstrual periods with a reappearance of the protozoa. Moreover, there were a few patients to whom the treatments did not give even temporary relief.

Although there have been many treatments recommended for trichomonas vaginalis vaginitis, they may be divided into two groups based on what it is hoped the treatment will accomplish. In the first, an effort is made to destroy all the protozoa by the use of antiseptics. Silver picrate, aldarsone and carbarsone are among those that have been recommended. Sometimes mechanical cleansing with green-soap or other solutions is added to the use of antiseptics.

In the second group of treatments less attention is paid to antiseptics than to measures which it is hoped will restore the normal defenses of the vagina. They are based on the general idea that it is possible for the protozoa to exist only when the normal mechanism which protects the vagina against infection has been disturbed.

In general, this defense mechanism of the vagina is believed to depend primarily on three conditions. First, that the vaginal secretion remains at its normal low  $p_H$ ; second, that the Döderlein or vaginal Lactobacilli are present in sufficient numbers to form lactic acid and finally that there is sufficient carbohydrate, perhaps in the form of glycogen, in the vaginal epithelial cells or spaces between the cells to afford adequate nourishment for the continued growth and activity of the Döderlein bacilli.

With the idea that in vaginitis acid douches might restore the bactericidal power of the vagina, lactic acid and vinegar douches have been prescribed in trichomonas vaginalis. They do give the patient considerable relief but apparently are not sufficient, in themselves, to completely correct the condition. Few if any cures have been brought about by simply using acid douches.

Adair and Hesseltine believe that, in the treatment of trichomonas vaginalis vaginitis, the most important therapeutic measure is to furnish sufficient nourishment to maintain a normal bacterial flora and, hence, they recommended, in 1936, that vaginal tablets containing 95 per cent lactose and 5 per cent citric acid be used. Karnaky with the same idea became enthusiastic over the use of vaginal suppositories, containing lactose and dextrose. The preparation which Karnaky recommended, and which was placed on the market under the trade name of floraquin, does also contain an antiseptic

known as diodoquin but the antiseptic is thought to have less therapeutic value than the sugars.

We have used floroquin in a large number of cases, with some satisfactory results. However, there were in the series of patients quite a few who continued to harbor the parasites in the vagina and to have recurrences of leukorrhoea and pruritus with the menstrual periods. Being rather discouraged by the results obtained from the numerous preparations that had been recommended for the treatment of vaginitis due to the trichomonas vaginalis we decided to attempt to build up the vaginal defense by simultaneously carrying out several measures. Not only was nourishment suitable for the growth of Döderlein's bacillus to be introduced into the vagina but also viable Lactobacilli.

Such a procedure if feasible seemed worth trying. To quote from Hesseltine on an article dealing with trichomonas infestations: "One thing is well established and that is with the restoration of the vagina to a normal cellular and bacteriologic flora the disease entity vanishes."

Since the time when Metchnikoff proposed the ingestion of Lactobacillus bulgaricus as an "elixir of life" to the more recent work of Rettger and many others, aciduric bacteria have enjoyed periodic popularity as agents for the relief of certain human ailments. Within the last two decades, "acidophilus therapy" has soared and fallen. It was used principally for the relief of gastrointestinal symptoms and depended upon implantation of the organisms in the intestinal tract with subsequent elimination of other, less desirable, types of bacteria. The literature from 1910 to 1930 contained a great deal of evidence that such therapy was justified. On the other hand, its popularity as a panacea has been rightfully questioned. When indicated, massive doses have doubtless given desirable results. It is not our purpose to take issue with either view in the discussion which arose in this respect. There can be no doubt that many commercial preparations during the heyday of acidophilus therapy were of little or of no value due to the lack of viable Lactobacilli.

Perhaps, however, some of the unfavorable reports, such as those by Bass and James, on Lactobacillus preparations were in part due to media difficulties for, as will be brought out shortly, we had in the beginning of this work considerable trouble in growing Lactobacilli on the usual solid medias. Moreover, James, in a recent personal communication, has expressed the opinion that conditions in the intestinal tract and in the vagina are so different it is not surprising that we have been successful in implanting Lactobacilli in the vagina.

Our first attempts to treat trichomonas vaginalis vaginitis with lactic acid-forming bacilli plus sufficient carbohydrate for the nourishment of the bacilli were rather crude. The junior author, who at the time this work was started, was the Bacteriologist to the Gynecological Department of the Johns Hopkins Hospital, had a culture of Lactobacillus which had been obtained from the United States Department of Agriculture. The culture secured from Washington is known as Hansen's strain, and has been used throughout this investi-

gation. Two lumps of ordinary sugar were coated with this culture and placed in the vagina. Quite naturally the heat of the tissues soon melted the sugar, converting it into syrup which ran out of the vagina. Tampons made of ordinary cotton were of no help as they quickly absorbed the syrup and prevented its coming in contact with the vaginal epithelium. Tampons made of nonabsorbent cotton were then used, with more success. In fact, many women obtained prompt relief by this crude treatment.

Encouraged by these results we then endeavored to work out a simpler method of carrying out this treatment. Tablets were then prepared for us under the careful supervision of Dr. John Brewer, Bacteriologist for Hynson, Westcott and Dunning. The tablets are prepared in the following manner:

Sterile skim milk is inoculated with a culture of *Lactobacillus bulgaricus*, U.S.D.A. Hansen's strain. After 48 hours' incubation, 1,000 cc. of this culture is mixed with 325 grams each of XXXX sugar and milk sugar. This is thoroughly dissolved in the milk and the mixture is spread in thin layers on enamel pans and dried in a vacuum oven at 37° C.

The mixture is scraped from the pans, broken into bits, and replaced in a drier to complete desiccation. When completely dried six cubic centimeters of white mineral oil and 38 Gm. of starch are added. The mixture is then pressed into tablets. The finished tablets weigh 1.3 Gm., and represent 1.25 cc. of original culture.

We were, of course, interested in determining the number of live *Lactobacilli* to the tablet. Tomato juice agar, using the formula of Valley and Herter, and also dehydrated tomato juice agar (Difco) was not found to be satisfactory for this purpose. On the other hand, it was possible to prove the presence of viable organisms in milk and tablets when dilutions were made in tubes of litmus-milk and later in thioglycolate medium. By this method we were able to determine the number of living organisms in a tablet or in one cubic centimeter of the milk culture. Dilutions were made by placing one tablet into 10 cc. sterile water. One cubic centimeter of this, when dissolved, was transferred to a tube containing nine cubic centimeters of sterile water, one cubic centimeter of this dilution transferred to a second tube containing nine cubic centimeters, and so on. One cubic centimeter of each dilution was transferred to 10 cc. of thioglycolate medium or litmus-milk, making the final dilution from the first tube, one tablet per 100; second tube, one tablet per 1,000, etc. Growth of the *Lactobacilli* was determined by coagulation of the milk or clouding of the thioglycolate medium, and Gram's stain of these media after 24 hours' incubation showing presence of typical gram-positive bacilli.

By this procedure we have determined the approximate number of viable *Lactobacilli* in tablets kept for as long as 12 months. Numerous samples taken from different lots of tablets all showed between 10,000 and 100,000 viable *Lactobacilli* in each tablet after they had been stored in a refrigerator for 12 months.

It is of interest, and possibly important, to note that in no case were contaminating organisms found in or on any tablets tested, in spite of the fact

that no precautions were taken, during storage, to maintain sterility. This, of course, may be explained on the basis of the acidity of the original milk cultures used in making the tablets.

These tablets have been used in several different ways, and it may be that the best way has not yet been worked out. However, the method which we are employing at present is the following: As soon as the diagnosis is made a bivalve speculum is introduced into the vagina, the cervix inspected for complicating endocervicitis and Skene's and Bartholin's glands inspected for possible involvement. The vagina is then dried with cotton and two Lactobacillus tablets inserted high in the vagina in the posterior fornix behind the cervix. The vaginal orifice is then plugged with a tampon of nonabsorbent cotton. When the patient returns on the next day the tampon is removed, material taken from the vagina for microscopic study and the treatment carried out on the preceding day repeated. On this second visit practically every patient will report that the itching has been much less, and it is very unusual to be able to demonstrate organisms at this time.

Such office treatments are repeated daily for from four to six days. The patient is then told to insert one Lactobacillus tablet high in the vagina each night. She is told to take douches only if she becomes uncomfortable from unabsorbed particles of the tablets coming out of the vagina and causing irritation. A white vinegar douche (5 per cent acetic acid) is recommended in a strength of from two to four tablespoonfuls to two quarts of water. Two douches a week are usually sufficient.

This home treatment is continued from three to six weeks and longer if necessary. However, if the organisms promptly disappear and show no immediate tendency to recur, the tablets need be used only every other night. It is especially important that they be used while the patient is menstruating, as that is, of course, the time when the vaginal defenses against the trichomonads are weakest.

If there is an endocervicitis which in itself is producing some of the leukorrhea it is treated by a cauterization. All patients are given the usual instructions regarding toilet technic, just as should be given no matter what method of treatment is being carried out. This consists of instructing the patient to clean herself, after defecation, from in front backwards, that is from the vagina towards the rectum, rather than in the reverse manner as is usual in most women. This lessens the likelihood of the condition being kept up by constant reinfection from the anus. When it is possible to clear up the infection quickly it is better to advise against sexual relations. When the treatment must be continued for a longer period of time the patient is instructed to take a plain water douche before coitus.

Unfortunately, some dispensary patients cannot come for daily office treatments during the first week after the diagnosis has been made, and in these cases we have to rely largely on the home treatments carried out by the patients themselves. We have not given patients tampons of nonabsorbent

cotton with which they themselves might plug the vaginal orifice, although this might be advisable.

These tablets have now been used on 50 consecutive, unselected cases. Thirty-one were white women, 19 colored. There were two children in the series, age six and seven, respectively, and two women beyond the menopause. The remaining 46 were in the reproductive age.

Just a few days ago a five-weeks-old child was brought into the dispensary with a profuse leukorrheal discharge which was thought to be gonorrhoeal in origin. However, smears for gonococci were negative while examinations for trichomonads showed a large number of the protozoa. Because the vagina was so small we have had difficulty in getting the Lactobacilli into it. However, we have been fairly successful with some long, very narrow tablets. Perhaps suppositories would be more effective, but their preparation has not yet been completed. After two treatments the discharge almost entirely disappeared, although there are a few organisms still present. This is, I believe, one of the youngest cases of vaginitis due to the trichomonas vaginalis which has been recorded. It is of interest that the mother also has a vaginitis due to the trichomonas vaginalis, indicating that the child probably acquired the disease from the mother.

All 50 of the women in our series complained of a vaginal discharge, 40 of pruritus vulvae and ten of dyspareunia. Fifteen had received treatment for the trichomonas infestation before coming under our care. Thirty-seven had had symptoms for over six months before we saw them, and 25 for a year or more. In our series there were only four patients whose symptoms were only a few weeks' duration.

This paper is essentially a report of the results obtained by the use of tablets containing Lactobacilli and not a detailed clinical study of vaginitis due to the trichomonas vaginalis. However, we wish to state that on the basis of our observations we are in complete accord with those investigators who in the past few years have brought out the fact that trichomonads not infrequently cause an inflammation of the Bartholin glands. This is a point of sufficient importance to deserve being stressed. Until a few years ago many gynecologists taught that when an examination showed bilateral thickening of the Bartholin glands one could be almost certain that at some time the woman had had gonorrhoea. This is incorrect. Shelanski and Savitz, in 1939, reported that they had demonstrated trichomonads in Bartholin's and Skene's glands, and proved conclusively that an inflammation of these glands may be due to trichomonads. In our series there were several cases of bartholinitis, in all probability due to the trichomonas vaginalis. In one patient, an unmarried woman with a hymen which would not admit the tip of the finger, the glands reached the size of olives. One case in our series interested us especially. Trichomonads were persistently present in the voided specimen of urine, while smears taken from the walls of the vagina showed no protozoa. After several examinations the trichomonads were found to be coming from an infected Skene's duct.

We realize that a series of 50 cases is not large enough to draw any very definite conclusions from regarding the efficacy of a method of treatment. Nevertheless, we do feel that our results warrant this preliminary report of the work we are doing.

Every patient treated with the *Lactobacillus* tablets showed immediate improvement and in many instances the organisms disappeared at once. Nevertheless treatment was continued from three to six weeks and when necessary longer. There have been recurrences in six patients whom we had thought were cured, perhaps in some instances these were reinfestations. Usually the supposed recurrence cleared up more rapidly than did the first infestation.

Forty-eight out of the series now have negative smears, and are asymptomatic. Both of the patients with positive smears are women in whom recurrences or reinfestations recently developed. They are now being treated.

All the patients whom we have considered cured have been instructed to report for examination immediately at the termination of each menstrual period, and without taking a douche before coming for examination. Thirty-five patients have remained symptom-free and have shown persistently negative smears for three months. We have 20 patients who are apparently well after six months.

We are enthusiastic about the prompt results which we have obtained with the *Lactobacillus* tablets, and we have had numerous patients go without treatment for from three to six months in order to prove that they have really been cured of the infestation. Nevertheless, we feel that every woman who has had a trichomonas infection should continue for considerable time to take either acetic acid douches or to use the *Lactobacillus* tablets for a few days each month around the time of the menstrual period. In fact, no matter what treatment is carried out for vaginitis due to the trichomonas vaginalis it should, in our opinion, be repeated at each menstrual period for from six months to a year.

There is still a great deal to learn about the trichomonads. Perhaps what we need is not a more effective way of treating this condition but to learn the necessity of treating it for a longer period of time than we have in the past. It is possible that the organisms, when attacked by various antiseptics, develop a few very resistant forms with the power to live much longer than the ordinary trichomonads.

There are of course certain questions that at once present themselves when one considers the advisability of using lactic acid-forming bacilli in vaginitis and some of these we hope to answer in the near future. For instance while we have clinical results to report which indicate the effectiveness of the method which we have used, and are also able to prove that the tablets themselves contained great numbers of viable lactic acid-forming bacilli, even when refrigerated 12 months, we have not shown just how rapidly these tablets restore the vaginal flora to its normal state.

We have analyzed the bacteriologic flora of a series of women, and



selected six from whose vagina it was impossible to grow any Lactobacilli. These six were instructed to insert one Lactobacillus tablet in the vagina every night for six nights and then to report for examination 48 hours later. Five of these six were then shown by culture to have large numbers of viable Lactobacilli in the vagina.

#### SUMMARY

A method of treating vaginitis due to the trichomonas vaginalis is presented. It differs from other methods in that viable lactic acid-forming bacilli are introduced into the vagina. There have been a few recurrences or reinfestations, but in all cases the method has yielded almost immediate relief of symptoms and in the great majority of instances the vaginas have remained free of trichomonads. Many of these patients have now been followed for six months.

We realize that much additional clinical and laboratory work must be carried out to establish, definitely, the value of Lactobacillus therapy in the treatment of vaginitis due to the trichomonas vaginalis. In spite of the relatively good results which we have obtained we do not claim that this method of treatment will completely eliminate recurrences in all patients. However, we feel our results justify this preliminary report.

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