A Retrospective cohort study on incidence of Trichomoniasis in high risk group patients with Leucorrhoea at RIMS General Hospital, Srikakulam.

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Background: Trichomoniasis becomes an epidemic in nearly all countries of the world during the past four decades. It has been noted that it has major impact on the maternal and infant morbidity and on human reproduction and infertility. Leucorrhoea is one of the common complaints encountered in Obstetric & Gynaecology and STD departments. The pathogens, which were frequently blamed for this type of abnormal vaginal discharge are T. vaginalis, candida species and Gardnerella vaginalis. Out of these three causes, T. vaginalis vaginitis is probably the most common cause among women afflicted with STD. The true incidence of trichomoniasis in our population is unknown. Hence we conducted a retrospective cohort study on incidence of Trichomoniasis in high risk group patients with Leucorrhoea attending OPD at RIMS General Hospital, Srikakulam.

Objective: To evaluate the true incidence of Trichomoniasis in high risk groups attending the OPD with Leucorrhoea.

Patients and methods: A retrospective analysis of 300 cases of vaginal discharge who attended themselves to the OPD and those who had not received any specific anti-trichomonal drug. T. vaginalis was isolated by a wet mount examination and by culture.

Results: We got 17.67% of incidence in our patient population. We observed a high incidence among the following, Between 15 and 29 years of age, with a low socio economic status and, In pregnant females. Most of the patients were the victims of sexually promiscuous husbands.

Conclusion: In our study, T. vaginalis was isolated in 53 cases of vaginal discharge by a wet mount examination and by culture. But we had poor culture results with Kupferberg’s trichomonas broth base medium and the poor culture results made us to depend completely on the wet mount examination. So, we prefer wet mount examination to culture in identification of T. vaginalis.

Introduceni:-
In India, ‘Leucorrhoea’ is one of the common complaints presented by the patients attending the Sexually Transmitted Diseases and Gynaecology out-patient departments for treatment. “Leucorrhoea” means ‘a running of white substance’. It describes any vaginal discharge which is not frankly purulent, but is best reserved for non-pathological discharges, caused by an excess of normal secretions and exudates from the genital tract. Thus this term should be restricted to mean an excessive amount of the normal discharge. Such ‘physiological’ discharge is often more common in women during the pre ovulatory phase of the menstrual cycle, sexual stimulation, premenstrual period, pregnancy, and in women taking oral contraceptives or with an I.U.D. (1)
The normal vaginal discharge consists of several components including water, electrolytes, microorganisms, epithelial cells and organic compounds, such as fatty acids, proteins and carbohydrates. The components combine to produce a vaginal discharge with a pH of less than 4.5. The largest volume of vaginal fluid arises from the egress of a serum transudate through local capillary walls and through the vaginal wall to the vaginal lumen. Smaller amounts of fluid are derived from Bartholin's glands, the cervix, endometrium and fallopian tubes. Cellular elements are sloughed from both cervical columner and vaginal squamous epithelium. The normal vaginal flora consists of microorganisms that colonize the vaginal fluid and the epithelial cell. White blood cells (W.B.C) though normally present in the secretory phase of the menstrual cycle, are usually found in only small numbers. The discharge although white or cream when fresh, it dries to leave a brownish yellow stain on clothing. The patients with leucorrhoea therefore become worried about, especially when they consider themselves at risk of infection.

Leucorrhoea is a nuisance in that it stains clothing and if the patient fails to take bath and change frequently causes excoriation and soreness of the vulva. But it never causes pruritus and is never offensive. Out of the three most common causes i.e., bacterial vaginosis candidasis and trichomoniasis, the last one is usually sexually transmitted.

Being a sexually transmitted disease, trichomoniasis is a common condition giving rise to vaginal discharge for which complaint female patients visit S.T.D and Gynaecology O.P departments frequently. Despite a seemingly limited number of causes for vaginal discharge, the woman often finds that she is plagued by a lack of symptomatic relief following therapy. There are multiple reasons for the failure to cure a vaginal discharge, which include most commonly diagnostic errors, therapeutic errors, and sexual reacquisition, but also non-sexual recurrence, drug resistance and depressed cellular immunity. Most patients incorrectly assume that the complaint is easily remediable. Failure to effectively treat the vaginal discharge often causes both patient and physician frustration.

Various studies were carried out on this disease by various workers. But still the knowledge on this disease is not sufficient to clear the doubts. A trial was therefore conducted to know the incidence of vaginal discharge due to T.vaginalis vaginitis and its epidemiological features in our own patient population. At the same time, we want to compare a single dose treatment with 2 gms. Metronidazole with a 7 day treatment i.e., 200 mg three times a day.

The present study was carried out on the high risk group patients with leucorrhoea attending the out-patient departments of S.T.D &Gynaecology, Government General Hospital, RIMS, Srikakulam, Andhra Pradesh with the following aims: To know the incidence of trichomoniasis in the high risk population of our area. To know the epidemiological features of trichomoniasis in our own patient population. To find out the efficacy of a single dose treatment with 2 gm. metronidazole and to compare with the results of 5 days therapy. To compare our findings with the findings of the previous reports.

**Method of Examination:**

**In Female Patients:**
The patients who were positive for Trichomonas vaginalis were studied for the efficacy of metronidazole. All the patients were thoroughly examined both clinically and parasitologically, after taking a detailed history.

**History taking:**
Every patient was made to sit down comfortably and made to feel relaxed as far as possible. A friendly approach was employed to reassure the patient.

Detailed history was elicited in each case regarding the present complaints and duration. Salient features were noted according to the protocol.

**Clinical examination:**
Complete and detailed local examination in each case was performed and documented. Genital examination was performed with the patient in the lithotomy position in strict privacy for the patient in a good source of light. First, the inguinal region was inspected and then palpated for evidence of lymphadenopathy. At the same time, the pubic area and surrounding exposed skin and the inner aspects of thighs were inspected for evidence of excoriations, pediculosis, folliculitis or other skin lesions.

Later, attention focused on the external genitalia and introitus. The discharge on that area was wiped off with a wet cotton swab. The labia majora were gently separated and then the labia minora to exclude any labial erythema,
oedema, fissuring or vulval lesions. At the same time, palpated to assess any enlargement of Bartholin’s glands and inspected their ductal openings for evidence of discharge. The buttocks were then separated for inspection of the perianal region for any evidence of excoriation.

Next, a sterile cusco’s bi-valve speculum or a sim’s speculum was introduced. After introducing the speculum, the cervix was examined and then a specimen for the appropriate test was taken. The appearance of the cervical epithelium, any abnormality and the presence or absence of any cervical exudate noted. Vaginal secretion from the posterior fornix was collected onto a sterile swab for culture and then a little onto a sterile platinum loop for wet-mount preparation. Then the speculum was carefully withdrawn while inspecting the walls of the vagina for any lesions. After removing the speculum, the urethra was examined and milked for any discharge.

Finally, a bimanual examination was done, both to assess the size of the uterus and to detect the presence of any abnormalities in the adnexae. Later, systemic examination was done methodically.

**Parasitological examination:-**

Vaginal discharge was collected from the posterior fornix of the vagina, with a sterile cotton swab for culturing T.vaginalis and with a sterile loop for wet-mount preparation.

A small amount of discharge was mixed with a drop of normal saline over a clean glass slide. It was examined under a microscope after placing a cover slip on that drop. Immediate diagnosis could be made by identifying motile trichomonads, if the patient was suffering from trichomonas vaginitis.

- **Culture:-**
  The medium which we used in our study was the "Kupferberg Trichomonas Broth Base Medium", supplied by the hi-media laboratories pvt ltd, bombay in a powdery form, within a plastic container.

The prepared medium was dispensed in small screw capped bottles. After inoculation of the specimen into this medium, it was incubated at 37 degrees C for up to 7 days. The organisms thus grown up on culture were examined again microscopically by getting a drop from the medium with a sterile loop.

Examination in male patients: The male partners were thoroughly examined both clinically and parasitological. After thorough examination, the same drug with the same dosage was given to them.

- **Treatment:-**
  20 patients were given 2 gm of metronidazole as 5 tablets of 400 mg each. The entire dose was given at one time under direct supervision. The remaining 18 patients were given 400 mg of metronidazole, three times daily for 5 days. The consorts were treated simultaneously with the same drug and dosage in a majority of cases. No other treatment either local or systemic was given. Patients were advised to avoid intercourse during the period of study and advised not to consume alcohol.

- **Duration of Follow up:-**
  Follow-up examinations were essentially the same as described in the method of examination. They were arranged for 4th, 7th, 14th, 21st, 28th post treatment days. Only the results of the 28th post-treatment day were used for the evaluation of treatment. It is very difficult to arrive at the optimum time for the follow up after treatment. There are two problems: (a) temporary reduction in the number of parasites, leading to falsely high cure rates and (b) re-infection in endemic areas which is likely to give falsely low cure rates.

In case of true failures, the number of parasites gradually increases during the post-treatment period, and by the 4th week the vaginal discharge becomes positive for T.vaginalis. Therefore a follow-up examination earlier than 4 weeks is likely to give falsely high cure rates.

If there was any re-infection, it would take a similar period, for the discharge to become positive because the effect of anti-trichomonal drug will decrease over 1-2 weeks, and another week or two would be required for the growth of the parasites. Thus, evaluation on the 28th day seems to be optimum.
Criterion of cure:-
The criterion of cure was absence of Trichomonas vaginalis both in the post -treatment vaginal smear and culture showing complete or marked relief of symptoms and signs.

6. Treatment failures: Patients with presence of T.vaginalis both in post - treatment vaginal smear and culture with or without relief of symptoms were considered treatment failures.

7. Number of patients: A total of 300 patients were screened. Out of them, 53 patients were enrolled and given the trial treatment.

Results:-
Incidence of Trichomonas vaginalis vaginitis:

| Age Group      | Patients with vaginal discharge | Patients with T.vaginitis |
|----------------|---------------------------------|---------------------------|
| 15-19          | 63                              | 13                        |
| 20-24          | 117                             | 19                        |
| 25-29          | 47                              | 14                        |
| 30 & Above     | 73                              | 7                         |
| Total          | 300                             | 53                        |

Out of 300 cases taken into study, 53 cases were positive for T.vaginalis vaginitis. Thus the present study shows 17.67% incidence of T.vaginitis. The youngest was 15 years old and the oldest 62. maximum: 20-24 i.e., 35.85%.

Socio - Economic status:-
The main bulk of patients was from the low income group. More than 90% women had never entered school and a few had received only elementary education.

Marital Status:

| Patients with Vaginal Discharge | Patients positive for T.vaginalis |
|---------------------------------|----------------------------------|
| 1. Married                      | 264                              | 45                        |
| 2. Unmarried                    | 36                               | 8                         |
| 3. Separated                    | 4. Widows                        |                           |
| 5. Pregnant                     | 27                               | 6                         |

Table 2 shows out of 300 patients with leucorrhoea, 264 patients were married and the remaining were either unmarried or separated or widows.

Out of 264 married patients, 45 patients are positive for T.vaginalis and in the remaining 36 unmarried, separated and widows patients, 8 patients are positive for T.vaginalis.

Table 2 also shows out of 300 patients with leucorrhoea, 27 patients were pregnant. Out of 27 pregnant ladies, 6 patients were positive for T.vaginalis.

Out of them, 5 patients were in the 5th month of gestation and the other patient was 10 weeks pregnant.

6. Source of infection:
Careful history taking revealed the source of infection. It was, marital in 34 cases, professional in 6 cases, regular or casual partners in 9 cases and boy-friends in another 4 cases.

Incidence of Trichomoniasis in male partners of patients with T.vaginitis:

| TOT NO OF CASES | patients positive for T.vaginalis | percentage |
|-----------------|-----------------------------------|------------|
| 42              | 2                                 | 4.76       |

Trichomonads were found by immediate microscopy in the urethral discharges of 2, out of 42 contacts of the patients.
Incidence of Symptoms:

| Predominant symptom       | no of patients | percentage |
|---------------------------|----------------|------------|
| No symptoms               | 16             | 30.19      |
| vaginal discharge         | 29             | 54.72      |
| pruritus                  | 8              | 15.09      |
| Dysuria                   | 6              | 11.32      |
| Symptoms attributable to other STD | 4              | 7.55       |

16 out of 53 cases were symptomless. In the remaining, only 14 patients had symptoms like discharge, dysuria and pruritus attributable to trichomoniasis. The characteristic greenish, yellow or white, frothy vaginal discharge with unpleasant fishy odour smell of musty hay was present in 29 cases. Only in 4 cases, there were symptoms attributable to other STD.

Laboratory findings:

| tot no of cases | T.vaginalis positive in wet mount examination |
|-----------------|-----------------------------------------------|
| no of cases     | percentage                                    |
| 300             | 53                                            |
|                 | 17.67                                         |

A total number of 300 cases were screened for Trichomonas vaginitis by wet mount examination with normal saline and by culture. In wet mount examination with normal saline, 53 cases were positive for T.vaginalis organisms and the remaining 247 cases negative.

Both in culture for T.vaginalis on kupferberg's trichomonas broth base medium, the findings were not satisfactory and even not encouraging as the results were very poor even in the well established cases of T.vaginitis proved by wet mount examination.

Association with other STD:

| Disease         | no of cases | percentage |
|-----------------|-------------|------------|
| candidiasis     | 16          | 30.19      |
| Gonorrhoea      | 5           | 9.43       |
| venereal warts  | 4           | 7.55       |
| Latent syphilis | 2           | 3.77       |
| Early syphilis  | 1           | 1.89       |

Gonorrhoea was found in 5 of the 53 cases, candidiasis in 16 cases, venereal warts in 4 cases, Latent syphilis in 2 cases.

Defaults:

Out of 300 cases, 53 cases got T.vaginalis smear positive but, 15 of them defaulted after the first visit leaving 38 patients for assessment. All 38 patients completed the trial according to the plan of study.

Out of 38 patients who completed the trial, 20 patients were given 2gm stat Metronidazole. The remaining 18 patients were given 400 mg of Metronidazole 3 times a day for 5 days.

Response with Metronidazole:

(a) With 2 gm stat. regime:
Table 8:

| last seen | no of cases followed | successful | failure rate(%) | admitted for further infection |
|-----------|---------------------|------------|----------------|-------------------------------|
| 0 day     | 53                  | -          | -              | -                             |
| 4th day   | 20                  | 20         | -              | -                             |
| 7th day   | 20                  | 20         | -              | -                             |
| 14th day  | 20                  | 20         | -              | -                             |
| 21st day  | 20                  | 20         | -              | -                             |
| 28th day  | 20                  | 20         | -              | -                             |

All the 20 patients completed the trial according to the plan of study. The response of the patients to treatment was shown in the table. There was no failure in the treatment with Metronidazole 2 gms.

(b) With 400 mg of metronidazole, 3 times a day for 5 days.

Table 9:

| last seen | no of cases followed | successful | failure rate(%) | admitted for further infection |
|-----------|---------------------|------------|----------------|-------------------------------|
| 0 day     | 53                  | -          | -              | -                             |
| 4th day   | 18                  | 18         | -              | -                             |
| 7th day   | 18                  | 18         | -              | -                             |
| 14th day  | 18                  | 18         | -              | -                             |
| 21st day  | 18                  | 18         | -              | -                             |
| 28th day  | 18                  | 18         | -              | -                             |

All the 18 patients in this 5 day treatment schedule, completed the trial according to the plan of study, the response was good both in the immediate follow-up period and after 4 weeks. There was no treatment failure in the schedule also as observed in the single dose regimen.

Parasitological and clinical response to treatment:

Table 10:

| Response | Metronidazole 2gm single dose/400mg tid for 5 days |
|----------|---------------------------------------------------|
| n        | percentage                                       |
| (a) Parasitological: |                     |
| cure ------------------------ | 38 | 100 |
| failure---------------------- | 0  | 0   |
| (b) clinical:               |                     |
| cure or marked improvement | 38 | 100 |
| in symptoms and signs       | 0   | 0   |
| slight or no improvement    | 0   | 0   |

Results were analysed both parasitologically and clinically all patients showed a uniformly good parasitological response. Examination on the 4th and 7th post treatment days showed that the flagellates had disappeared from the vaginal secretions.

Side effects:

Subjective side effects were reported by 3 patients, 2 females and one male. Vomiting occurred in 2 female patients while travelling in the bus. These patients who vomited after treatment had negative tests for Trichomonas vaginalis after the 4 weeks follow up. Vomiting occurred in a male patient in the clinic itself. The patient who vomited after treatment had negative test for the organism both before and after treatment.

Discussion:

Leucorrhoea is one of the common complaints encountered in Obstetrics and Gynaecology and STD departments. In the past, the vaginal discharge was assumed to be a part of female adulthood and motherhood. This is not true today and more woman are presenting themselves for the treatment of this problem. The pathogens, which were frequently blamed for this type of abnormal vaginal discharge, are T.vaginalis, candida species and Gardnerella vaginalis. The relative frequencies and symptoms of these three types vary markedly in different population. Out of these three causes, T.vaginalis vaginitis is probably the most common cause among women afflicted with STD. Trichomonas vaginitis without U.T.I is characterized by one or more of the following symptoms: Increased volume of discharge, Abnormal yellow or green colour of discharge, Vulval itching, irritation or burning, Introtital dyspareunia, Malodour.
T. vaginalis usually produces a profuse, yellow, purulent, homogeneous discharge which is often malodorous and may be frothy. The vaginal epithelium is inflamed and petechial lesions may be present on the cervix. Sexual transmission of T. vaginalis is well established. Routine sampling indicates that many women and most men with infection are asymptomatic. Among women with asymptomatic infection, however, the potential for development of symptoms is high.

The diagnosis in women is confirmed by demonstration of motile trichomonads in vaginal secretions mixed with normal saline and examined promptly under a low-power or high-power microscopic fields. The diagnosis of T. vaginalis in men is difficult and the organism can be demonstrated only in one-third of male sex partners of women with T. vaginitis. Hence culture of the organisms is required with the early morning urethral scrapings obtained before voiding, on the basis of these data and in view of the difficulty in demonstrating T. vaginalis in men, routine treatment of sex partners whether symptomatic or asymptomatic is advisable to reduce the reservoir of infection and the rise of transmission and to prevent the future development of symptoms.

Successive treatment of Trichomoniiasis requires that both the patient and her consort be treated simultaneously and adequately, which can be greatly facilitated by the availability of an effective single dose therapy.

Incidence of Trichomoniiasis:-
The true incidence in the population is unknown but it is estimated to be present in between 10-20% of women during the reproductive years \(^{(3)}\). In our study, we got 17.67% incidence of T. vaginitis. Age-distribution: In our study, 86.8% of women being in 15-29 years of age. The lowest age noted was 16 and the highest 55. This is an agreement with Willcox \(^{(4)}\) who noted a similar finding.

Socio economic status:-
High incidence of the disease in women during their years of greatest sexual activity and in those of low socio-economic status is consistent with the findings of similar studies by others \(^{(5,6)}\).

Marital status & source of infection:-
On careful history taking, it was known that 34 patients who got married had no extramarital contact. After thorough interrogation of their husbands, it came into light that they were sexually promiscuous individuals and they had multiple exposures with the prostitutes. This indicates that these 34 female patients might have acquired the infection through their husbands.

Relative incidence in pregnancy:-
27 women with vaginal discharge were pregnant, out of 300 practices. Cases screened. Out of 53 cases of trichomonas vaginitis, 6 patients were pregnant. Thus this is an agreement with Trussell \(^{(7)}\) who noted an increased incidence among the pregnant. Incidence of symptoms: 16 out of 53 cases were symptomless. In the remaining only 14 women, had symptoms like discharge, dysuria and pruritus attributable to Trichomoniiasis. Association with other STD: study of Trichomoniiasis associated with other STD is of epidemiological interest. The most frequent association noted in this study was candidiasis and next gonorrhoea. The frequency of association is very striking. The frequent association of candidiasis and gonorrhoea with trichomoniiasis demands the performance of full test for candidiasis and gonorrhoea, whenever a patient is treated for trichomoniiasis. While this procedure is adopted in all STD clinics it is not always so in other departments or in general.

Incidence in male partners:-
Trichomonads were found by immediate microscopy in the urethral discharges of 2, out of 42 contacts of the female patients. This observation agrees with the previous reports by others stating that the diagnosis of T. vaginalis infection in men is difficult and can be demonstrated only in one-third of male sex partners of women with T. vaginalis.

Response with Metronidazole:-
After treatment with metronidazole, the symptoms and signs improved rapidly in all 38 patients satisfied the criterion of cure and 100% efficacy was observed. With single dose treatment, tolerance was good. But in 7 days treatment 3 patients i.e., 2 females and 1 male had vomitings, after taking this drug. In other studies clinical trial reports on metronidazole indicate that a single oral dose of 2 gm cured trichomoniiasis in over 95% of cases when sexual partners are simultaneously treated \(^{(8,9,10,11)}\). Statistical analysis, however, showed no significant difference.
between the results of the two schedules of treatment i.e., 400 mg three times a day for 5 days or 2 gm as a single dose both in the immediate follow up period and upto 4 weeks after treatment. It is our opinion that metronidazole seems to be a highly efficacious drug in Trichomoniases in a single dose of 2 gm. The practical significance of single dose is obvious since it facilities complete treatment of patients as well as consorts right in the physician’s presence and thus removing the necessity of relying on the patient to complete a longer course. Since the results of treatment with 2 gm of metronidazole was good and the incidence of side effects no different from the other trials, metronidazole in our opinion, fills the need for an effective well tolerated single dose anti-trichomonal drug.

Laboratory findings:-
We got 53 cases positive for T. vaginalis, out of 300 cases of vaginal discharge screened, by wet mount examination. To culture T. vaginalis, we used Kupferberg's trichomonas broth base medium. But the findings were not satisfactory and the poor results by culture method made us to depend completely on the wet mount examination. The poor culture results were probably due to loss during transit from the O.P.D to the laboratory in the sunny climate, as the organisms being sensitive to temperature change.

Our observations regarding culture favours the Woodcock's statement\(^{(12,13)}\) that the culture methods were not substantially more reliable or more sensitive than simple microscopy, an opinion shared by Campbell\(^{(14)}\) and McCann\(^{(15)}\). But this observation is against the findings of Whittington\(^{(16)}\) who reported that the culture technique was more sensitive and thin et al \(^{(17)}\) who found that culture and microscopy gave similar results.

Summary and conclusions:-
The present study is an analysis of 300 cases of vaginal discharge. These cases were taken into study from the out patients departments of STD & Gynaecology. The patients were selected on the criteria of female patients who are attending themselves with the complaint of vaginal discharge and those who had not received any specific anti-trichomonal drug. In our study, T. vaginalis was isolated in 53 cases of vaginal discharge by a wet mount examination and by culture. But we had poor cultures results with Kupferberg's trichomonas broth base medium and the poor culture results made us to depend completely on the wet mount examination. So, we prefer wet mount examination to culture in identification of T. vaginalis.

We got 17.67% of incidence in our patient population. We observed a high incidence among the following Between 15 and 29 years of age, with a low socio economic status and In pregnant females. Most of the patients were the victims of sexually promiscuous husbands. Nearly about one-third of the patients were symptomless, about one-fourth of the patients had symptoms typical of trichomonas vaginitis and only complaint of vaginal discharge without dysuria and pruritus was present in half of the cases. In only 4 cases, patients had symptoms attributable to other STD's. The incidence was very low among the male partners and we could identify the T. vaginalis only in 2 male partners of 42 cases examined With 2 gm Metronodizole as a single dose, we did not get any treatment failure in our study. Our patients tolerated the dose very well. So, we recommend Metronodizole 2 gm stat. dose for the patients with Trichomoniases.

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