

**COMPARATIVE STUDY IN THE MANAGEMENT
OF BACTERIAL VAGINOSIS -
METRONIDAZOLE Vs LACTOBACILLUS**

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CERTIFICATE

Certified that this dissertation entitled “**Comparative study in the management of Bacterial vaginosis Metronidazole Vs lactobacillus**” is a bonafide work done by **Dr.S.Narmadha**, Post Graduate Student in **M.D. Dermatology, Venereology and Leprology**, Madras Medical College, Chennai-600 003, during the academic year 2008-2011.This work has not previously formed the basis for the award of any degree.

Prof.Dr.P.ELANGO VAN, M.D., D.V,
Additional Professor, & Director Incharge
Institute of Venereology,
Madras Medical College,
Chennai-600 003.

Prof. Dr. D.Prabhavathy, M.D.D.D.,
Professor and Head,
Department of Dermatology &
Leprology,
Madras Medical College,
Chennai-600 003.

Prof. Dr. J. MohanaSundaram, M.D., P.hd
Dean,
Madras Medical College,
Chennai-600 003.

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Introduction

INTRODUCTION

Bacterial vaginosis is due to imbalance in the normal bacterial flora of vagina. Bacterial vaginosis is a polymicrobial syndrome involving the replacement of the normal vaginal lactobacillus by variety of anaerobic bacteriae and mycoplasmas.

Bacterial vaginosis is the commonest cause of vaginal discharge (48%) in women with significant morbidity.¹ If untreated , patients are more prone for ascending endometrial infections, preterm delivery and increased risk of acquisition of Herpes genitalis, Gonorrhoea and HIV infections. Most of the cases are asymptomatic and recurrences are common even in treated cases. Recurrence of the disease , unknown incubation period , adverse outcomes on pregnancy and complications like pelvic inflammatory disease necessitated the need to find an alternative drug , in this emerging antibiotic resistance period.

In this background we planned to conduct this study to compare the efficacy of Metronidazole , the commonly used drug and Lactobacillus , a Probiotic alternative drug.

*Review of
Literature*

REVIEW OF LITERATURE

DEFINITION

Bacterial vaginosis is the most common form of vaginal infection in women of the reproductive age group which represents a disturbance of vaginal microbial ecosystem rather than a true tissue or epithelial infection. It is a polymicrobial syndrome involving replacement of the normal vaginal lactobacilli by a variety of anaerobic bacteria and mycoplasmas.²

SYNONYMS

Non specific vaginitis, Hemophilus vaginitis, Corynebacterium vaginitis, Gardnerella vaginitis, nonspecific vaginosis, anaerobic vaginosis.

EPIDEMIOLOGY

- It accounts for 48% of patients with abnormal vaginal discharge.
- The prevalence of bacterial vaginosis among adolescents varies from 13%-31.9%³

- Among post pubertal females , the prevalence of BV was lower with those who had no sexual experience than those with sexual experience.⁴
- BV is more prevalent in patients with tubal infertility (31.5%) as compared with non tubal infertility. (19.7%)⁵
- Among lesbians the prevalence is 29%.⁶
- High prevalence rates are seen in HIV positive commercial sex workers.⁷

VAGINAL ECOFLORA

The vagina is a dynamic ecosystem that is sterile at birth. Under the influence of maternal oestrogen the glycogen content of vaginal epithelial cells get elevated and the infant vagina is colonised by lactobacilli.

As the level of oestrogen diminishes, the glycogen content and the lactobacilli diminishes, which contribute to interspersed with coagulase negative staphylococci, streptococci and E.coli colonisation.⁸

In the adult female once again due to increased glycogen under the influence of oestrogen, the lactobacilli concentration rises to about 10^5 - 10^8 cfu/ml of vaginal secretion. Other organisms are present at concentrations as below

Gardnerella vaginalis 10^7

Mycoplasma hominis $10^{5.2}$

Anaerobic gram negative rods 10^6

Privately biviadisians $10^{5.5}$

LACTOBACILLUS

Various subspecies of lactobacillus are present in vagina like *Lactobacillus jensensinii*, *L.crispatus*, *L.gasseri*, *L.fermentum*, *L.oris*, *L.reuteri* and *L.vaginalis*.⁹

1. They produce Hydrogen peroxide which has inhibitory effect on bacterial growth, particularly catalase negative bacteria.
2. Hydrogen peroxide also interacts with halide ions in the presence of cervical peroxidase and forms H_2O_2 -halide-peroxidase antibacterial system which inhibit bacteria as well as viruses including HIV.

3. Lactobacillus also produces bacterial interference by colonisation.

VAGINAL pH

Vaginal pH in premenarchal female is near neutral (around 7).

At the time of puberty, the vaginal epithelium increases to 25 cell thickness, glycogen level increases, there is predominance of Lactobacillus and the production of lactic acid decreases vaginal pH to less than 4.5. This pH is maintained until menopause.

PATHOGENESIS OF BACTERIAL VAGINOSIS

In BV, this symbiotic relationship is broken up and overgrowth of pathogenic anaerobic bacteria produces amines by decarboxylation of fatty acids and amino acids which elevates the vaginal pH.

The effects of altered pattern of organic acids is uncertain. Succinic acid produced by vaginal anaerobes inhibit the chemotactic response of WBC.¹⁰

The vaginal fluid of women with BV has increased levels of endotoxin, sialidase, and mucinase. An increased host response is documented by increased levels of IL 1 α and prostaglandins in the cervical mucus.¹¹

BIOCHEMICAL CHANGES

There is increased microbial surface virulence factors including lipopolysaccharidases, sialidases, mucinases. In addition some strains of *Gardnerella vaginalis* cause cleavage of secretory IgA.¹²

Anaerobic bacteria produce enzymes, aminopeptidases that degrade protein. Decarboxylase convert amino acids and other compounds to amines such as putrescine, cadaverine and trimethylamine at alkaline pH. These amines contribute to increased vaginal pH and produce odoriferous discharge. *Mobiluncus* is known to produce trimethylamines.¹³

The vaginal fluid of women with BV has increased levels of endotoxin, sialidase and mucinase.

An increased host response is documented by increased levels of IL1 α and prostaglandins in the cervical mucus. The redox potential (eh) of vaginal epithelial surface is lower in women with BV.

RISK FACTORS

1. History of bacterial STDs
2. Increased number of lifetime sexual partners.¹⁴
3. Lower age of first intercourse¹⁵
4. Lesbians¹⁶
5. Smoking
6. High risk sexual behaviour
7. Intrauterine contraceptive devices¹⁷, vaginal douching, peccary use.
8. Gynaecological cancers¹⁸

CLINICAL FEATURES

Symptoms:

Malodorous vaginal discharge, more after sexual intercourse and during menstruation.

Pruritus (very rarely)

Pain during coitus

Lower abdominal pain

Mental stress due to the symptoms

Signs:

Gross vaginitis and vulvitis are not characteristic of this condition.

1. Excessive ,homogenous uniformly adherent vaginal discharge, in contrast to the floccular vaginal discharge of normal individuals.
2. Elevated vaginal pH >4.5 . pH determination is assessed by narrow range pH paper applied to the withdrawn speculum or directly over the vagina.If it is more than 4.5 then it is abnormal.¹⁹

Also vaginal pH can be assessed by self test pH glove. In this, One finger of a medical examination glove has an attached pH indicator which the patient can introduce into her vagina and then read the measured result.²⁰

3. Sniff / Whiff test:

The odour of vaginal secretions is tested by smelling the withdrawn speculum. Rotten or dead fishy odour is smelt due to the presence of amines like putrescine, cadaverine and trimethylamine.

This can also be tested by adding a few drops of 10% KOH to a few drops of vaginal secretion. The test will give the characteristic odour.

The KOH test was reported to be the most powerful single predictor for the diagnosis of BV. This test may be the least sensitive of the clinical tests for diagnosis of BV.

Electronic sensor assay (electronic nose) The idea of using sensor arrays coupled with interpretation of the resulting signals is based on the assumption that the signal pattern detected might be an electronic counterpart to the human sensation of smell and has attracted some attention.

4. Saline wet mount and gram staining of vaginal secretion should be done to look for clue cells.

Clue cells are squamous epithelial cells with large number of BV organisms densely attached in clusters to their surface. The cell borders are not clearly discernible, with fuzzy cytoplasm, like staining with black pencil.²¹

The presence of > 20 % of clue cells is diagnostic. Clue cells represent the best single diagnostic criteria of BV.

5. Absent or few lactobacilli in Gram staining.

AMSEL'S CRITERIA:

1. Excessive, homogenous, uniformly adherent vaginal discharge
2. Elevated vaginal pH > 4.5
3. Positive amine test (Whiff test)
4. Clue cells constituting 20% or more of total vaginal epithelial cells.

The above four diagnostic features are the features of Amsel's criteria. The patients should fulfil at least 3 out of 4 criteria for bacterial vaginosis.²²

NUGENT'S SCORING SYSTEM

Vaginal Gram stain had comparable sensitivity but a much greater specificity and predictive value than vaginal culture for *G.vaginalis*. Culture does not constitute test of cure, since many women without clinical signs of BV have positive culture for this organism following treatment.²³

This system is based on the bacterial morphology in Gram staining.

Bacterial morphology type	Score: none	0	1+	2+	3+	4+
Lactobacilli (large, elongated, gram Positive bacteria)	>30	6-30	1-5	<1	0	
Gardnerella vaginalis (Small, Gram variable Coccobacilli)	0	<1	1-5	6-30	>30	
Mobiluncus (Curved, gram negative Bacilli)	0	<1	1-5	6-30	>30	
SCORE	0-3	Normal				
	4-6	Intermediate (test to be repeated later)				
	7-10	Bacterial vaginosis				

SCHMIDTS SCORING SYSTEM

This system of wet smear examination of vaginal secretion resembles Nugent's scoring system. The ranking of lactobacilli and cocci is similar but the demarcation of the intervals differ.²⁴

This system does not recognise mobiluncus.

This method has been validated for the diagnosis of BV in primary care population.

HAYS/ ISON SYSTEM

This is another scoring system based on the observation of gram stain to estimate the ratios of the observed morph type rather than the exact number of bacteria.

Originally the observations are divided into three categories - normal, intermediate or BV. Later, in order to obtain a more precise classification, two additional categories were added as compared to Nugent's scoring. The new groups are those that contain no bacteria at all (Group 0) and those that contain large amounts of gram positive cocci such as streptococcus or staphylococcus morphotypes.²⁵

DIAGNOSIS

1. History, Clinical examination
2. Satisfying Amsel's criteria, Nugent's scoring
3. Gas liquid chromatography for succinate and lactate ratio

Many anaerobic Gram negative rods produce succinate, as a metabolic product and lactobacilli produce lactate, as a metabolic product. The ratio of succinate to lactate has been found to be increased in vaginal fluid of patients with BV. This is done by gas liquid chromatographic analysis.²⁶

4. Amine estimation;

Diamines and polyamines in the vaginal fluid show 86% sensitivity and 92% specificity by Femexam method. They can also be estimated by colorimetric detection of diamines.²⁷

5. Serodiagnosis to determine the presence of antibodies to the microbes.

6. Nucleic acid probe

7. DNA probe²⁸

8. Culture

Gardnerella Vaginalis: Semi selective human blood in Tween 80 bilayered agar + 5% Carbon di oxide will give tiny colonies with a zone of haemolysis in a period of 3-5 days. *G.vaginalis* can be recovered from all women with BV but also from upto 58% of those without BV.

Mobiluncus: Human blood Colombia agar or semi selective foetal calf serum and rabbit's blood bilayered agar with 5% Carbon di oxide yields tiny translucent colonies in 3-5 days.

Mycoplasma hominis: Hominis agar or H-H broth with arginine will give a fried egg colony.

9. Detection of metabolic products:

Proline amino peptidase test – Bacteria associated with BV produce proline aminopeptidase , whereas lactobacilli do not produce this enzyme.

After inoculation of vaginal fluid with enzyme substrate in a micro titre plate for 4 hours at 35.5° C, rapid garnet green is added to produce a colour reaction. Red or pink colour indicates a positive test whereas an orange or yellow colour indicates a

negative test. It was reported to have 81% sensitivity and 96% specificity in comparison to clinical criteria for diagnosis of BV.²⁹

10. BV blue test:

It is a test for detection of sialidases activity. A vaginal swab is to be placed in the BV Blue vial containing the chromogenic substance of bacterial sialidase and a laboratory timer is to be started. Two drops of BV Blue developer solution is to be added at 10 minutes. Blue or green colour was recorded as a positive result, whereas yellow colour represents negative result. The test is performed at room temperature.

11. Pulse field gel electrophoresis rib typing

12. 16S rDNA restriction fragment length polymorphism

13. Multiplex polymerase chain reaction

14. Triplet arbitrary primed PCR

DIFFERENTIAL DIAGNOSIS

PHYSIOLOGICAL

1. At the time of menarche
2. At the time of menopause
3. After sexual intercourse
4. At the time of ovulation
5. Before and after menstruation
6. Pregnancy

PATHOLOGICAL

Vaginal discharge is considered abnormal if

1. Hypervaginal secretion not associated with menstruation
2. Offensive or malodorous discharge
3. Yellowish discharge

INFECTIVE

Vaginitis - Bacterial vaginosis

Vaginal candidiasis

Vaginal trichomoniasis

Cervicitis - Neisseria gonorrhoeae

Chlamydia trachomatis

NON INFECTIVE

Foreign bodies - IUCD

Tampons and other materials

Chemical irritants - Antiseptics

Deo spray

Bath additives

Detergent, spermicidal agents

Douches

Perfumed soaps

Gynaecological conditions – Endo cervical polyps

Fistulae

Post operative

Radiation effects

Tumours

Medication and nutrition³⁰

COMPLICATIONS

Gynaecological complications:

1. Plasma cell endometritis
2. Silent endometritis - Spontaneous PID that occurs without instrumentation may also be related to BV. A small amount of intermenstrual bleeding or increased bleeding with menses is a frequent occurrence in patients with endometritis and salpingitis.³¹
3. Vaginal cuff cellulitis
4. BV and HIV - There is a twofold increase in the prevalence of HIV in women with BV. Women with BV are more likely to seroconvert to HIV than those without BV.

A low vaginal pH inhibit CD₄ lymphocytes activation and reduces the target for HIV in the vagina. In BV, elevated vaginal pH makes the vagina more conducive to HIV survival and adhesion. The level of acidity of vagina may affect CD₄ lymphocytes activation. The more alkaline the pH, more CD₄ cells are activated and is a more suitable target for HIV.

BV also increases IL -10, which increases the susceptibility to HIV.

Main degrading enzymes in BV will make it easier for HIV to infect, by breaking down the cervico vaginal mucus.³²

BV and other STIs

BV is present most frequently as a co infection with other STIs like Gonorrhoea. The leading hypothesis to explain this is that the absence of protective Lactobacilli in BV facilitates the acquisition of other STIs.³³

An inhibitory effect of the bacterial amines putrescine and cadaverine on the cell division and germ tube formation of candida albicans has recently been reported.³⁴

Obstetric complications

1. Intra amniotic fluid infection - BV associated organisms ascend to cause infection of the decidua , placenta or amniotic fluid . Those patients , have elevated levels of endotoxin on activation of the prostaglandin system and could provoke preterm labour. The concentration of PGE₂ and PG F₂ are significantly higher. Even in the absence of upper genital tract infection, BV associated pathophysiology might lead to preterm labour.³⁵

2. Chorioamnionitis
3. Premature rupture of membrane
4. Preterm delivery
5. Low birth weight
6. Post partum endometritis
7. Post abortion endometritis - Invasion of anaerobes like *Gardnerella vaginalis* and *Mycoplasma hominis* increases the risk of postpartum endometritis and vaginitis if delivered vaginally or by LSCS.

TREATMENT

The cure or improvement was usually defined as resolution of two or more of Amsels criteria for diagnosis.³⁶

TREATMENT GUIDELINES

WHO (2003)

Recommended regimen

- T. Metronidazole 400 mg or 500 mg orally, twice daily X 7 days.

Alternative regimen

- T.Metronidazole 2 gm orally as a single dose or
- Clindamycin 2 % vaginal cream, 5 gm intra vaginally at bedtime for 7 days or
- Metronidazole 0.75% gel, 5 gm intravaginally , twice daily for 5 days or
- T.Clindamycin 300 mg orally twice daily for 7 days.

CDC guidelines (2006):

Recommended regimen:

- T.Metronidazole 500 mg orally twice a day for 7 days or
- Metronidazole gel 0.75 % one full applicator (5 gm) intravaginally once a day for 5 days or
- Clindamycin cream 2 % one full applicator (5 gm) intravaginally at bedtime for 7 days.

Alternative regimen:

- T. Clindamycin 300 mg orally twice a day for 7 days or
- Clindamycin ovules 100 mg intravaginally once at bedtime for 3 days

NACO guidelines (2007):

Recommended guidelines for vaginitis (TV, BV & VVC):

T. Secnidazole 2 gm orally single dose

Or

T. Tinidazole 2 gm orally twice daily for 5 days.

T. Metaclopramide to be taken 30 minutes before T. Secnidazole to prevent gastric intolerance.

Along with T. Fluconazole 150 mg orally single dose (or)
Local Clotrimazole 500mg vaginal pessary once.³⁹

Gardnerella vaginalis is more susceptible to the hydroxyl metabolite of metronidazole. *Mycoplasma curtissii* is not susceptible to metronidazole. The long term recurrence rate following Clindamycin and Metronidazole approach 80% in the year after treatment.³⁸

Alternative treatments for BV includes restoration of normal vaginal ecology ;

1. Chlorhexidine pessaries 150 mg x 7 days
2. Povidone iodine pessary bid x 14 days

3. The use of vaginal acidifiers in the form of gels, suppositories, and acid soaked tampons has varied widely from 18-80%

They suppress but do not kill vaginal anaerobes.

Lactic acid suppository:

Lactate pH 3.5, 5 gm gel daily x 7 days

Lactate pH 3.8, 5 gm gel intermittent use x 6 weeks

Acetate gel:

5% Acetic acid tampon bd x 7 days

Yogurt;

PH < 4.5 douche 10-15 ml bd x 14 days

Lactobacillus suppository bid x 6 days

Hydrogen peroxide producing lactobacillus acidophilus in combination with 0.03 mg of estriol.

Unresolved issues in the patient management

1. How common is late recurrence?

Up to 70 % women develop recurrent BV within 3 months after therapy.⁴⁰ Other studies show long term recurrence rates of approximately 50%.⁴¹

2. Why do recurrences happen?
 - a. Reinfection by a male partner colonised with BV associated micro organisms.
 - b. Recurrence due to the persistence of BV associated organisms.
 - c. Failure to establish the normal and perhaps protective lactobacillus rich flora following therapy.
 - d. Persistence of another yet unidentified host factor in the patient.
 - e. Microbiological studies established that half of women lack vaginal lactobacilli that produce H_2O_2 following therapy with clindamycin or Metronidazole.⁴²

3. Should women with signs of BV be treated if asymptomatic?

Women with BV should be informed of their diagnosis and treatment should be offered if requested.

4. Does BV contribute to PID among women undergoing IUCD insertion ?

IUCD users are at increased risk of BV. It also increases the risk of endometritis and salpingitis following IUCD insertion.

5. Should HIV infected women with asymptomatic BV receive treatment ?

HIV infection may be particularly common in BV associated PID. It may be prudent to recommend treatment of asymptomatic BV in HIV infected women.

6. Should male partners be treated?

No study has convincingly shown that treatment of male partners decrease the risk of recurrence.⁴³

PREVENTION

1. Abstinence
2. Promotion of condom use
3. Recolonization of vagina with H₂O₂ lactobacilli
4. Genital hygiene practice in the male partner.⁴⁴

METRONIDAZOLE

History

Isolation of the antibiotic azomycin (2-nitro-imidazole) from a streptomycete by Maeda in 1953. 1- β -hydroxyethyl-2 methyl-5-
itroimidazoles has especially high activity in vivo and in vitro
against the anaerobic protozoa *T.vaginalis* and *E.histolytica*.⁵³

PHARMACOLOGY

Metronidazole is an amoebicidal, trichomonocidal and bactericidal drug. A chemically reactive reduced form of metronidazole is thought to be responsible for the drug's activity. The reduced substrate affects anoxic or hypoxic cells causing loss of the helical structure of DNA strand and impairment of cellular function.

SPECTRUM OF ACTIVITY

1. Anaerobic gram negative bacilli including most bacterioides species, fusobacterium and veillonella.
2. Anaerobic gram positive cocci including clostridium, eubacterium, peptococcus and peptostreptobacillus.

3. Helicobacter pylori, Gardnerella vaginalis and the protozoa Endamoeba histolytica, Trichomonas vaginalis and Giardia lamblia.
4. Not against fungi, viruses and most other aerobic or facultative anaerobic bacteriae like Actinomyces, Lactobacilli and Propionio bacterium acnes.⁵⁴

MECHANISM OF ACTION

1. It is a prodrug. It requires reductive activation of the nitro group by susceptible organisms. The anaerobic organisms contain electron transport components such as ferredoxins, small Fe –s proteins that have a sufficiently negative redox potential to donate electrons generated by energy metabolism. The single electron transfer forms a highly reactive nitro radical anion that kills the susceptible organisms by targeting DNA and other vital bio molecules.

The anaerobic organisms derive energy from the oxidative fermentation of keto acids such as pyruvate. Pyruvate decarboxylation catalysed by pyruvate : ferredoxin oxido reductase (PFOR) produces electrons that reduce ferredoxins which in turn catalytically

donates its electrons to biological electron acceptors or to metronidazole.⁵⁵

2. Suppression of cell mediated immunity.
3. Sensitization of hypoxic cells to radiation, carcinogenicity and mutagenicity in experimental animals.

CLINICAL RESISTANCE

1. When there is impaired oxygen scavenging capabilities, there will be increased local concentrations of oxygen. This causes decreased activation of Metronidazole.⁵⁶
2. Lower levels of PFOR and ferredoxin owing to reduced transcription of the ferredoxin gene.⁵⁷
3. Increased expression of superoxide dismutase and peroxiredoxin also cause Metronidazole resistance.

PHARMACOKINETICS

Following oral administration, Metronidazole is well absorbed from the gastrointestinal tract.

Peak serum levels following an oral dose occurs in 1 to 2 hours. Mean effective concentrations in plasma of 8-13 micro

gm/ml is achieved within 0.25 to 4 hours after a single dose of 500 mg.

Repeated doses every 6 to 8 hours result in some accumulation of the drug. The plasma half life is 6 to 8 hours.

It reaches cell, tissues and fluids with CSF concentrations reaching approximately 43% of serum concentration.

The drug crosses the placenta and is secreted in breast milk. Metronidazole is metabolized in the liver. It is excreted primarily in the urine as metabolites with 20% of the dose excreted as unchanged drug.

The half life of Metronidazole in adults ranges between 6 and 12 hours. Accumulation may occur in patients with severely impaired hepatic function. Dosage reduction may be indicated. But dosage adjustment is unnecessary in patients with decreased renal function.⁵⁸

DRUG INTERACTIONS

1. Alcohol should be avoided because metronidazole and alcohol together can cause severe nausea, vomiting, cramps, flushing and head ache.

2. Disulfiram like reaction may result in confusion and psychotic reactions.
3. Cimetidine increases blood levels of metronidazole.
4. Cholestyramine reduces blood levels of metronidazole by reducing its absorption.
5. Metronidazole should not be combined with Amprenavir for treating HIV because Amprenavir contains propylene glycol. Metronidazole blocks the breakdown of propylene glycol in the liver leading to accumulation of propylene glycol in blood. This would cause seizures, increased heart rate and kidney failure.
6. Barbiturates cause increased metabolism of Metronidazole, thereby the concentration of Metronidazole is decreased.

INDICATIONS

1. Bacterial infection:
 - 1 a. Serious infection caused by susceptible anaerobic bacteria such as Bacterioides fragilis ,Clostridium, Fuso bacterium, Peptostreptococcus and peptococcus species.
 - 1 b. Mixed aerobic and anaerobic infections

2. Antibiotic induced diarrhoea and colitis including pseudo membranous colitis caused by *Clostridium difficile*.
3. In multi drug regimens for the treatment of *Helicobacter pylori* associated peptic ulcer disease.
4. Bacterial vaginosis –In 1995 Canadian STD guidelines recommended metronidazole for the treatment of this condition.
5. Periodontal diseases like acute necrotizing ulcerative gingivitis caused by spirochetes, *Fusobacteria* and *bacterioides* species.
6. Protozoal diseases like Trichomoniasis in men and women, hepatic and intestinal amoebiasis, Giardiasis
7. Acne rosacea

CONTRA INDICATIONS

- Hyper sensitivity to metronidazole
- Patients with active neurological disorders or history of blood dyscrasias
- First trimester of pregnancy. In addition, it is advisable that administration be avoided during the second and third

trimester. Its use requires that the potential benefits be weighed against the possible risks.

- Lactation-unnecessary exposure to metronidazole should be avoided. If a nursing mother is treated with metronidazole the breast milk should be expressed and discarded during treatment. Breast feeding can be resumed 24-48 hours after treatment.

ADVERSE EFFECTS

CVS: Palpitations and chest pain

CNS: Peripheral neuropathy, Seizures, transient ataxia, dizziness, drowsiness, confusion, insomnia, headache, incoordination, ataxia and encephalopathy.

DERMATOLOGICAL: Urticaria, flushing, rash, pruritus, furry tongue, glossitis, stomatitis and acute exacerbation of candidiasis. Rarely can cause Steven Johnson syndrome⁵⁹, toxic epidermal necolysis. But recent reports suggest concomitant administration with Mebendazole increases the risk of SJS and TEN

GIT: Diarrhoea, nausea, vomiting, unpleasant metallic taste, anorexia, epigastric distress, constipation, dry mouth, glossitis, stomatitis, and candidiasis.

GENITO URINARY: Dysuria,proliferation of candida albicans in the vagina,vaginal dryness and burning .

Dysuria, cystitis and darkening of urine due to the metabolite of Metronidazole.

HAEMATOLOGICAL: Transient eosinophilia or leucopenia

HYPERSENSITIVITY: Erythematous rash, urticaria, serum sickness like reaction.

ENDOCRINOLOGICAL: Gynaecomastia

DOSAGE

Available as injection, oral tablets, vaginal gel, vaginal cream, vaginal inserts topical cream and topical gel.

Adults : oral 500 mg every 8 hours (7.5 mg/kg every 6-8 hours) to a maximum of 4 g/24 hours.

IV -500mg by IV infusion every 8 hours to a maximum of 4 gm /24 hours. IV infusion at the rate of 5 ml/minute.

Children : 30 mg/kg /dayIV in 3 divided doses or 15-30 mg/kg/day orally in 3-4 divided doses.

Bacterial vaginosis: Adult dose of 500 mg orally twice a day
x7 days Or

Metronidazole gel 0.75 % one applicator full intra vaginally
twice daily x 5 days

Or

Metronidazole 2 gm orally in a single dose.

Routine treatment of male sexual partners is not necessary.

ORAL PROBIOTICS -CAPSULE LACTOBACILLUS

DEFINITION

Probiotics are a mixture of putatively beneficial lyophilized bacteria given orally.⁴⁵

HISTORY

Probiotic means 'for life'. Metchinkoff in 20th century suggested the possibility of modification of gut flora by replacing the harmful microbes by useful microbes.

Henry Jissie isolated Bifidobacterium for managing diarrhoea. Alfred Misside in 1970 isolated E.coli strain which was followed

by isolation of Metchinkoffs bulgarius bacillus later called as Lactobacillus bulgarius.

In 1985 Lactobacillus acidophilus was isolated. The term probiotics was introduced in 1983. Later Lactobacillus species.

Lactobacillus rhamnosus and reuteri were found to have beneficial effects on urogenital tract infections.

LACTOBACILLUS

Lactobacilli are rod shaped gram positive, fermentive, organotropes. They are usually straight although they can form spiral or coccobacillary forms under certain conditions. They are found in pairs or chains of varying length. They derive energy from the conversion of glucose to lactic acid during fermentation. They generate ATP by nonoxidative substrate level phosphorylation.

They have a generation time ranging from 25 minutes to several hundred minutes. They grow optimally between the temperature of 30 and 40 degree celsius.

MECHANISM OF ACTION

1. The lactobacillus strain, colonises over a sufficient period to confer health benefits to the host. Adhesion and colonization of the vaginal epithelium by the lactobacillus provides⁴⁶ an effective barrier against pathogenic vaginal organisms.
2. The dried lactobacilli used in vaginal suppositories appear to be capable of hydrating from the capsule and interferes with pathogenic organisms.
3. Most urogenital micro flora originate from the gut and ascend via the rectum.⁴⁷
4. DNA extracted from probiotic organism could mediate anti inflammatory activity through toll like receptor 9 signalling pathway.
5. Modulation of immunity by production of bio surfactants and collagen binding proteins that inhibit pathogen adhesion.^{48,49}
6. L.rhamnosus and L. reuteri adhere to cells , persist ,multiply and prevent adhesion of other pathogenic organisms.
7. They produce lactic acid, hydrogen peroxide , reutericin and bacterioicin that are antagonistic to pathogen growth.⁵⁰

8. They resist vaginal microbicides including spermicides.
9. Competition for nutrients which are essential for the survival of pathogens.
10. Stimulate the secretory local IgA for combating the infection.
11. Natural vaginal strain *L.jensenii* being engineered to deliver the potent HIV inhibitor cyanovirin is under study.

EFFICACY AND SAFETY

Effective in bacterial vaginosis, prevent the development of yeast vaginitis , inhibits the growth of *Candidia albicans*.

COMPOSITION

Each capsule contains

Equal proportion of 1 billion C.F.U of *Lactobacillus rhamnosus* GR-1 and *Lactobacillus reuteri* RC - 14^{50,51}

The unique capsule technology protects the strains in the canister against gastric acid by using a spherical polysaccharide matrix. The matrix releases the live protective bacteria in the small intestine. In Indian women *Lactobacillus rhamnosus* GR-1 and

Lactobacillus reuteri RC- 14 are the more common strain. L. rhamnosus are particularly effective in inhibiting growth , adhesion and biofilm formation against gram negative pathogens and Candida albicans.

While L.reuteri RC-14 is effective against Gram positive pathogens such as Staphylococci, enterococci and Streptococci. A combination of such type of lactobacilli can be more useful as a probiotic. So we had taken these strains in our study.

INDICATION

1. Bacterial vaginosis, Yeast vaginitis, urinary tract infection and its recurrence.
2. Bacterial vaginosis leading to urethritis, cystitis, urethral syndrome,post catheterization syndrome.⁵²
3. Bacterial vaginosis increasing the risk of PID , cervicitis,endometritis,pre mature rupture of membrane, abortion and preterm labour.
4. Treatment of Bacterial vaginosis prior to in vitro fertilization procedures.

DOSAGE AND DURATION

1 Capsule bid for the first 30 days followed by

1 Capsule o.d for the next 30 days.

Preferably to be taken along with meals with a glass of water.

The total duration of therapy is 60 days.

ADVERSE EFFECTS

Rare cases of lactobacillemia have been reported in severely immunosuppressed individuals. On the other hand , in HIV affected persons reduce shedding of the virus.

In cardiac valve replaced patients, can produce valve sepsis. Some cases of liver abscess have been reported.

STORAGE

Capsules are to be stored in a cool , dry place and to be Protected from light and heat.

The lid to be replaced immediately after taking the capsule.

PRESENTATION

Canister containing 10 capsules and 30 capsules.

Aim and Objectives

AIM & OBJECTIVES

1. To analyse the epidemiological pattern of the disease among the study population.
2. To compare the efficacy of metronidazole and lactobacillus in the management of bacterial vaginosis.
3. To evaluate safety, adverse effects, compliance of metronidazole and lactobacillus.
4. To monitor the recurrence of Bacterial vaginosis following each therapeutic modality.

Materials and Methods

MATERIALS AND METHODS

TRIAL DESIGN

Prospective, open labelled, comparative clinical trial with the approval of ethical committee.

STUDY POPULATION

Hundred patients with bacterial vaginosis diagnosed on clinical grounds, Amsel's criteria, Nugent's scoring system, attending the female STD out patient clinic of Institute of Venereology, Chennai for 2 years from 21.8.2008 to 20.8.2010.

INCLUSION CRITERIA

1. Patients aged above 18 years.
2. Those who fulfilled Amsels and Nugent's criteria for bacterial vaginosis.
3. Those who were willing to come for the follow up.

EXCLUSION CRITERIA

1. Patients aged less than 18 years
2. Pregnant individuals
3. Lactating mothers
4. Treatment with any antibiotics 6 weeks prior to study period.
5. Patients with other causes of vaginal discharge

METRONIDAZOLE GROUP

1. Known hypersensitivity to metronidazole.
2. Underlying neurological disorder
3. Alcohol consumption

LACTOBACILLUS GROUP

1. History of any drug hypersensitivity
2. Cardiac valve replacement surgery
3. On immunosuppressive drugs
4. Severe immunosuppression

WITH DRAWALS AND DROPOUTS

Subjects were informed that they were free to dropout from the study at any time, without stating any reason and records were maintained.

TREATMENT PROTOCOL AND METHODOLOGY

Hundred patients were randomly allotted to the two groups(50 each) to receive either tablet metronidazole 400 mg bd for 7 days orally or capsule Lactobacillus 1 bd for one month followed by 1 od for the next month orally.

The study was approved by the Institutional Ethical Committee, Government General Hospital & Madras Medical College, Chennai . All patients signed a written informed consent document.

EVALUATION AT THE FIRST VISIT

During the screening period all the patients were evaluated as Follows:

History

General examination

Systemic examination

Genital examination, Sniff / whiff test, vaginal pH

Saline mount, KOH examination,

Gram staining of vaginal smear.

Gram staining of endocervical smear

Endocervical swab for gonococci culture

Urine examination for albumin, sugar, cast

VDRL, VCTC tests

MONITORING AT FOLLOW UP VISITS:

The patients were followed up at one week, one month, and 2 months 6 months after starting treatment.

At each visit vaginal pH, Sniff test, Whiff test, saline mount, KoH test and gram staining were repeated.

USG was done at the end of 2 months of initiation of treatment.

PROCEDURES

COLLECTION OF SPECIMEN

The patient was asked to lie in the lithotomy position. Cusco's self retaining speculum was placed in position. Vaginal smear was collected from the lateral or posterior fornix of vagina with a sterile cotton swab. An endocervical swab was taken by inserting the swab 1-2 cm in to the endocervical canal and by rotating the swab for 10-20 seconds. It was subjected to Gram stain and culture for Gonococci.

TESTS OF SPECIMEN

1. Vaginal pH test:

The vaginal swabs were touched on the pH paper reading ranging from 3.5 to 6.0. Otherwise, the pH paper was touched to the tip of the vaginal speculum after removing it from the vagina or pH paper directly touched to the wall of the vagina. The pH paper was not allowed to contact with cervical secretion.

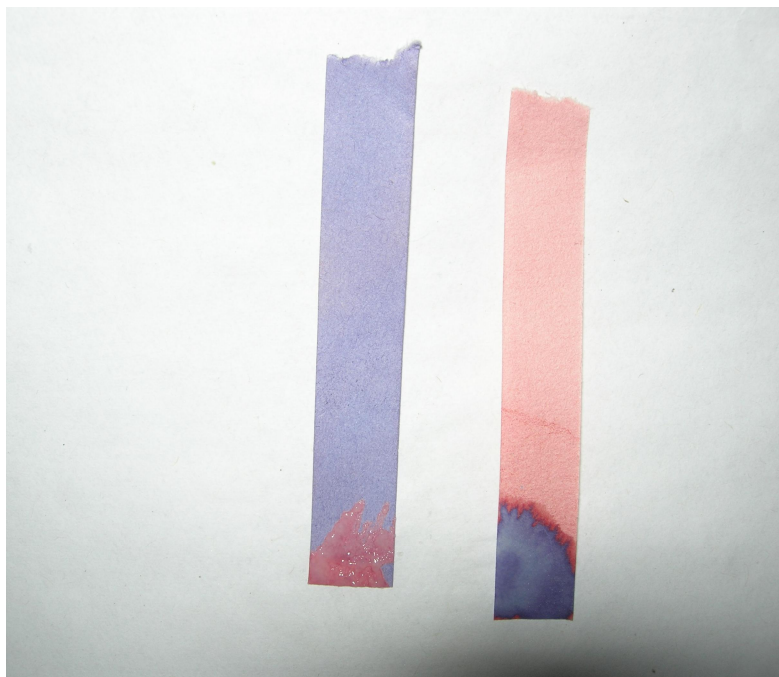
CUSCO'S BIVALVED, SELF RETAINING SPECULUM EXAMINATION



CAPSULE LACTOBACILLUS



pH PAPER TEST



READING

Normal adult vagina has an acid pH of 1- 4.5. In bacterial vaginosis the pH is raised above 4.5

Presence of menstrual blood, cervical mucus, semen and Trichomonas vaginalis infection may also rise the vaginal pH.

2. AMINE TEST

A drop of vaginal fluid was put on a clean microscopic slide.

To this one drop of 10 % KOH solution was added. The slide was brought close to the nose to smell the amine odor. An intense, putrid fishy odor denoted a positive reaction.

3. MICROSCOPY

Wet mount test:

A drop of normal saline was put over a clean , grease free microscopic slide. To this a drop of vaginal fluid was added and mixed well. A cover slip was put over the mixture ,so that there was a uniform spread without air bubble. The slide was observed under 40 x magnification of the objective.

READING

Presence of clue cells suggested the diagnosis. (A clue cell is a squamous epithelial cell with many coccobacillary organisms attached to its surface giving it a granular surface. The cells do not have a well defined edge because of the presence of bacteria and disintegration of the cell).

Gram staining of the smear:

A vaginal smear was prepared on clean, grease free microscopic slide by rolling the swab on the slide.

The smear was air dried and heat fixed. Crystal violet solution was poured over the slide and left for one minute and washed with distilled water.

Gram's iodine solution was added on the slide and kept for one minute and washed again with water.

The slide was held between the thumb and forefinger and flooded with a few drops of the acetone - alcohol decolourizer until the violet colour washed off.

The slide was washed with running water. Carbol fuchsin solution was poured over the slide and allowed to act for one minute.

The slide was washed with water and blotted by drying in between two blotting papers.

A drop of liquid paraffin was put over the slide and observed under oil immersion.

Reading:

Gardnerella vaginalis - Gram variable to Gram negative rods small (1.5-2.5 micron x 0.5 micron) shows variation in shape.

Mobiluncus species - Slender (0.3- 0.4 micron), slightly curved rods, either singly or in pairs with the appearance of gull wings.

M. curtisii is Gram intermediate

M. mulieris is Gram negative

Lactobacillus - Seen normally in vaginal secretion as Gram positive rods. These were markedly reduced in BV.

Clue cells - epithelial cells coated with bacteria.

Grams staining reagents

Solution A : Crystal violet powder 2 gram

Ethyl alcohol 20 ml

Solution B : Ammonium oxalate 0.8 gram

Distilled water 100 ml

Mix solutions A and B and filter.

2. Grams Iodine

Potassium iodide 2 grams

Iodine crystals 1 gram

Add 100 ml distilled water

3. Decolourizer

Acetone 50 ml

95 % Ethyl alcohol 50 ml

4. Counter stain

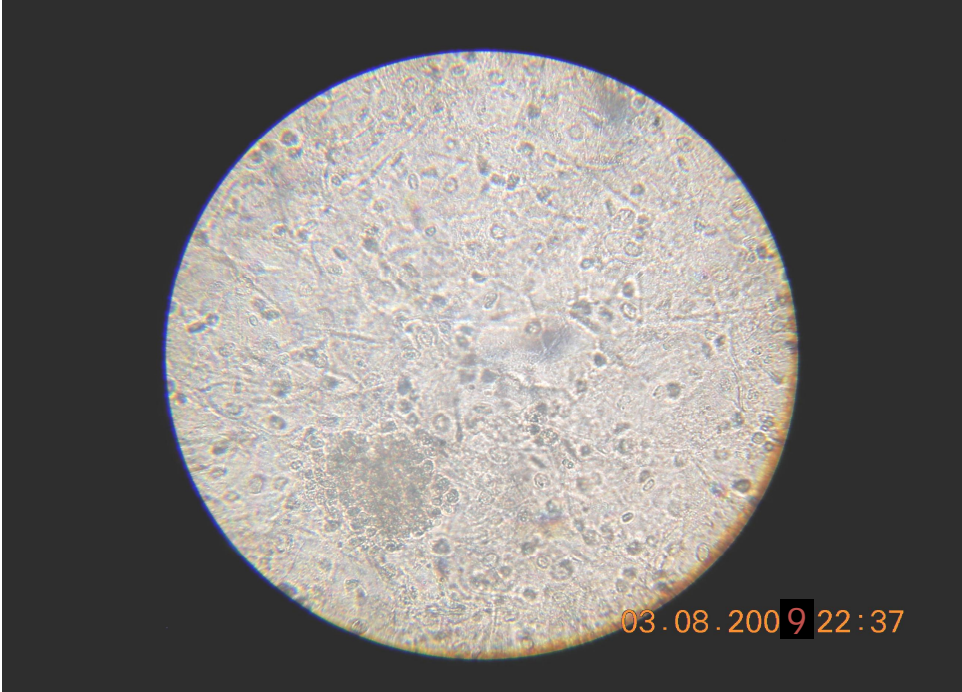
Stocking solution Safari or Carol fustian 2.5 gram

95% Ethyl alcohol 100 ml

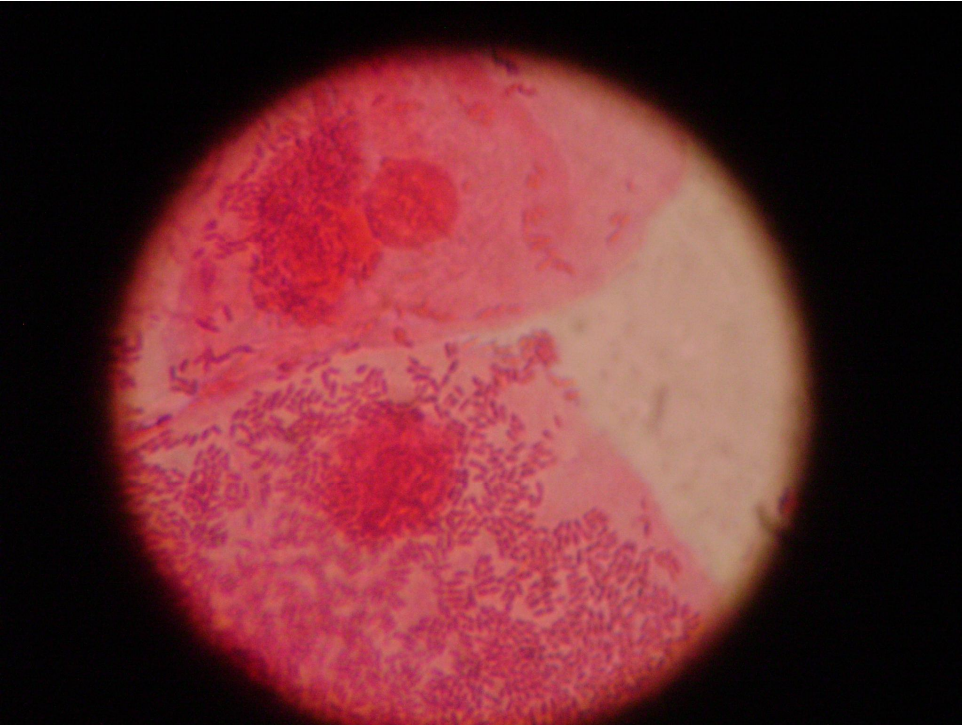
Working solution - Stock solution 10 ml

Distilled water 100 ml

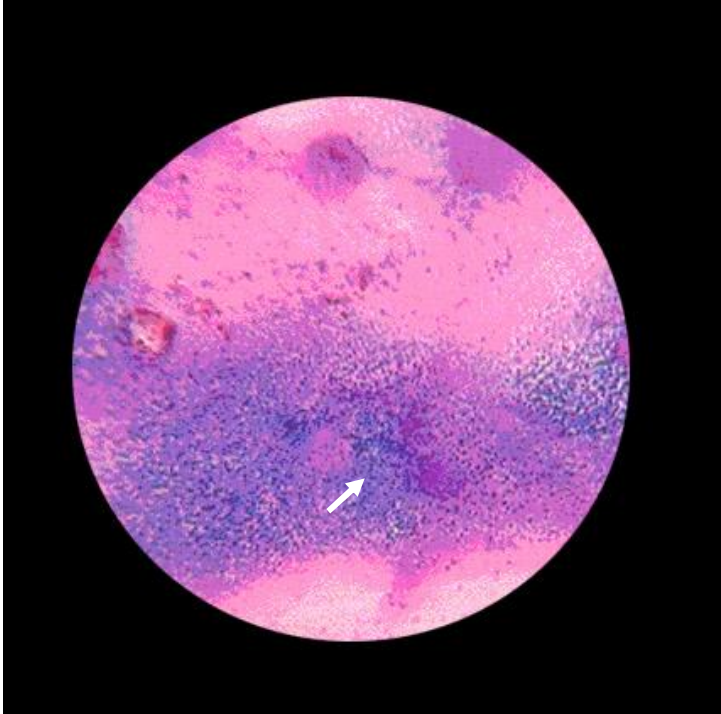
CLUE CELLS IN SALINE MOUNT



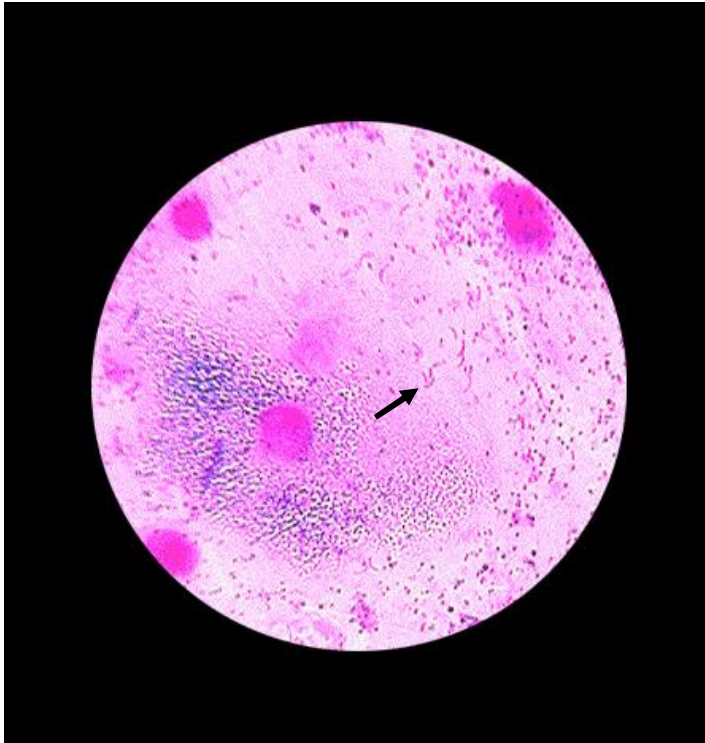
CLUE CELL IN GRAM'S STAIN



GRAM POSITIVE COCCOBACILLI IN GRAM'S STAIN



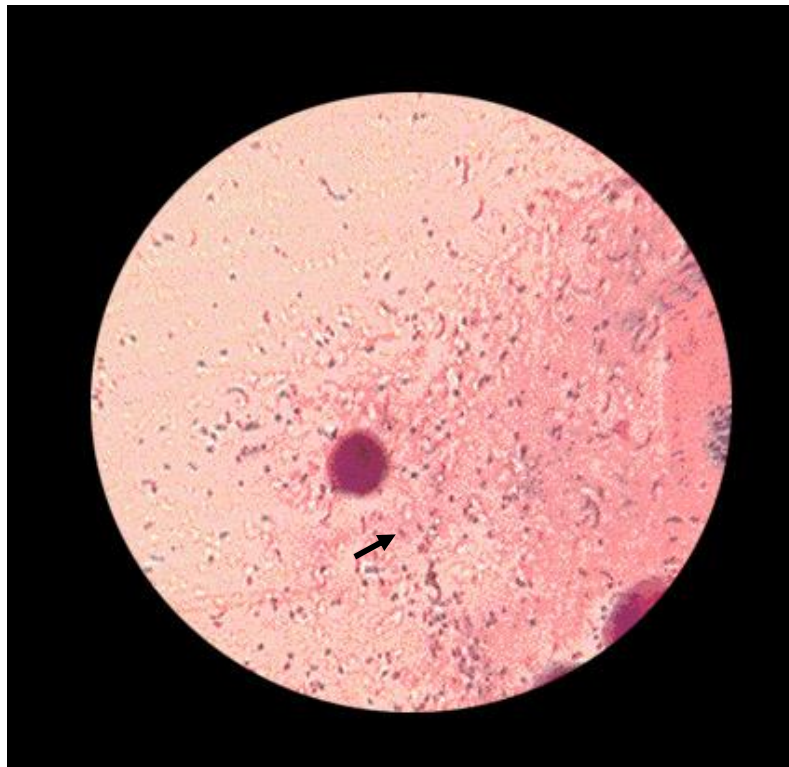
MOBILUNCUS IN GRAM'S STAIN



LACTOBACILLUS IN GRAM'S STAIN



GARDNERELLA VAGINALIS IN GRAM'S STAIN



Observation

OBSERVATION

DEMOGRAPHIC CHARACTERS

	Group 1	Group2	p value
Age	32.3 ₋ + 8.9	31.4 ₋ + 8	0.25
Duration	2(0.06-120)	2(0.3- 120)	0.22
PVD	21	17	0.57
Obstetric history	43	42	0.25
Partner assessment	11	11	0.78
Menstrual history	42	44	0.59

In this trial the mean age of patients was 32.4₋+ 8.4 years with the P=0.25 in the matched groups. The highest age was 51 years and the lowest age was 20 years.

There was no significant difference in the mean age between group 1 and group 2 (P=0.25).

DURATION OF SYMPTOMS

There was no significant difference in the median duration between group 1 and group 2 (p=0.22).

The average period of duration of the disease was about 2 months which was comparable in between the two groups.

MENSTRUAL HISTORY

Menstrual period was regular in 86 individuals out of 100 patients, irregular menstrual cycle observed in 10 out of 100 patients and 4 patients attained menopause.

The proportions of menstrual history were comparable in both the groups. (P value = 0.59).

COMPLAINTS

COMPLAINT	GROUP 1	GROUP 2	
GENITAL DISCHARGE	34	33	
GENITAL ITCHING	4	6	P=0.77 (N.S)
GENITAL MAL ODOR	5	2	

*Chi-square test was used to calculate the P-Value

INFERENCE

- There was no significant difference in the distribution of complaints between group 1 and group 2 (P=0.77). Out of 100 individuals, 67 patients came with genital discharge, 10 Individuals with genital itching & 7 individuals with both genital odors while 16 were asymptomatic.

MARITAL HISTORY

Marital history	Group 1	Group 2	P-value*
	N0 (%)	N0 (%)	
Married	46 (92.0)	46 (92.0)	1.00 (N.S.)
Unmarried	4 (8.0)	4 (8.0)	

*Fishers Exact Test (2-tailed) was used to calculate the P-Value

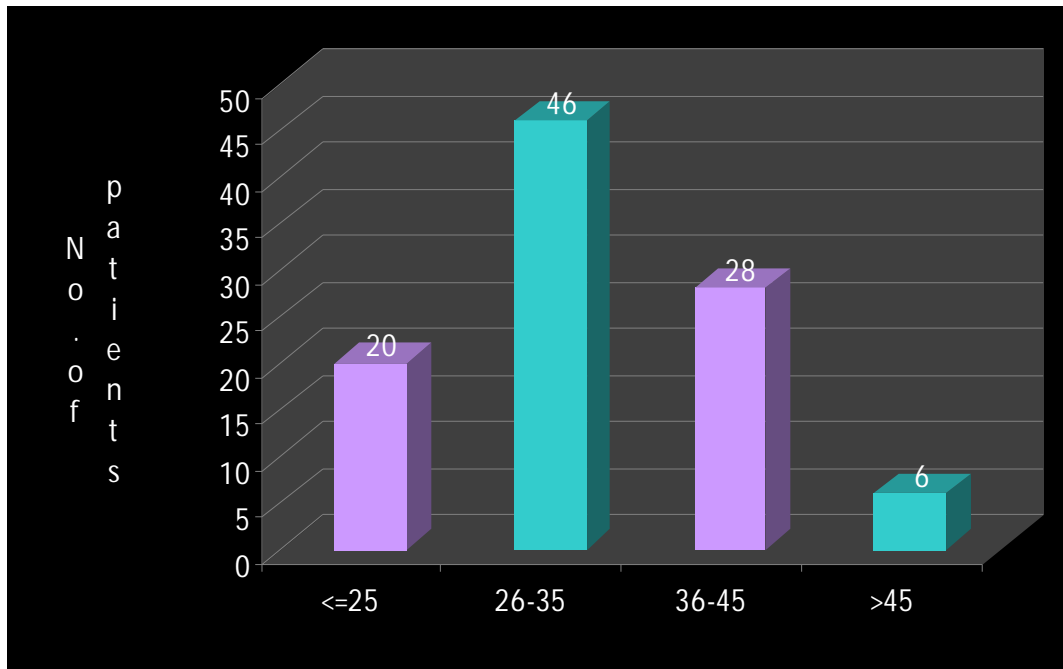
Out of 100 patients 92 were married, the remaining 8 of them were unmarried. P value was not significant in between the two trial groups. (P=1.00).

Among the married individuals 57 got married before the age of 20 yrs, 26 in between 21- 25 years of age and 9 got married above the age of 25 years.

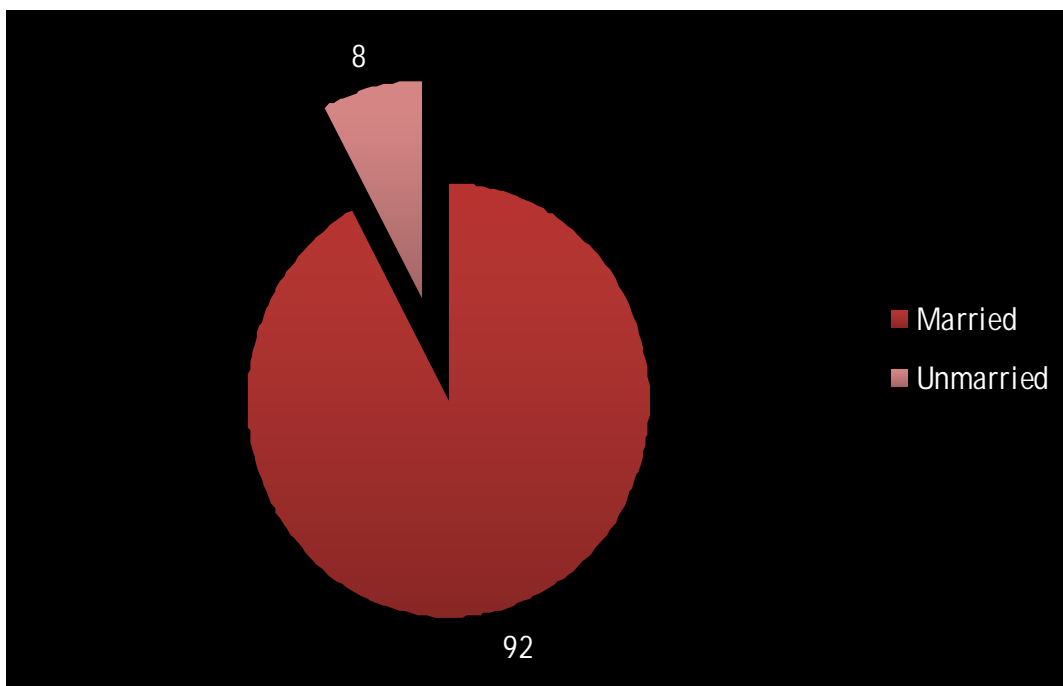
INFERENCE

- There was no significant difference in the distribution of marital status in between group 1 and group 2 (P=1.00).

AGE ASSOCIATION



MARITAL STATUS & ASSOCIATION



OBSTETRIC HISTORY

Out of the 100 patients 8 patients were unmarried, of this 6 individuals denied history of having sex. The rest 2 patients had no issues.

Among the remaining 92 married patients H/ O normal delivery was noted among 58 patients, LSCS among 12 patients, abortion in 3 patients, normal delivery + LSCS in 1 patient, normal delivery + abortion in 11 patients while nullipara were about 7 in number.

In the study population 15 had no children (including unmarried), 21 had only one pregnancy , 34 had 2 pregnancies , 22 had 3 pregnancies and 8 had > 3 Pregnancies.

There was no significant difference in the distribution of obstetric history between Group 1 and Group 2 (P=0.25).

FAMILY PLANNING

On analysing the family planning methods permanent sterilization was done in 61 patients , condoms were used by 2 patient's partners, IUCD was used in 18 individuals while no method was adopted by 19 individuals

- There was a significant difference in the distribution of FP between group 1 and group 2 (P=0.01).

NUMBER OF SEXUAL PARTNERS

No. of partners	Group 1	Group 2	P-value*
	N0 (%)	N0 (%)	
Mono	36 (72.0)	41 (82.0)	0.47 (N.S.)
Multiple	10 (20.0)	7 (14.0)	
Nil	4 (8.0)	2 (4.0)	

*Chi-square test was used to calculate the P-Value

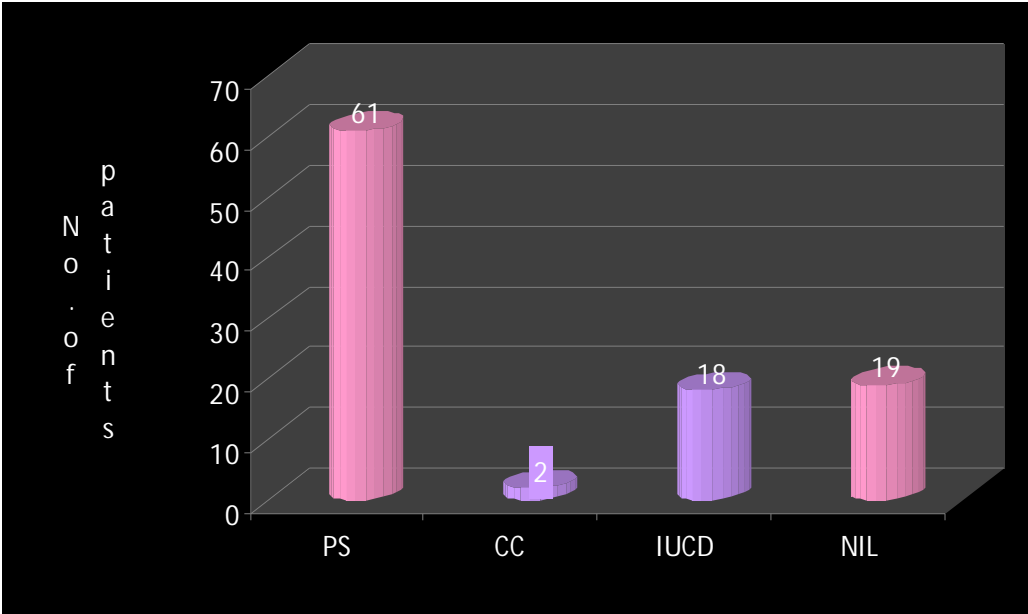
The number of sexual partners were calculated and analyzed. Those who had mono partner were 77, multiple sex partners observed in 17 patients, while 6 patients denied having sexual contact.

The proportion of number of partners were comparable in between the two groups.(P = 0.47)

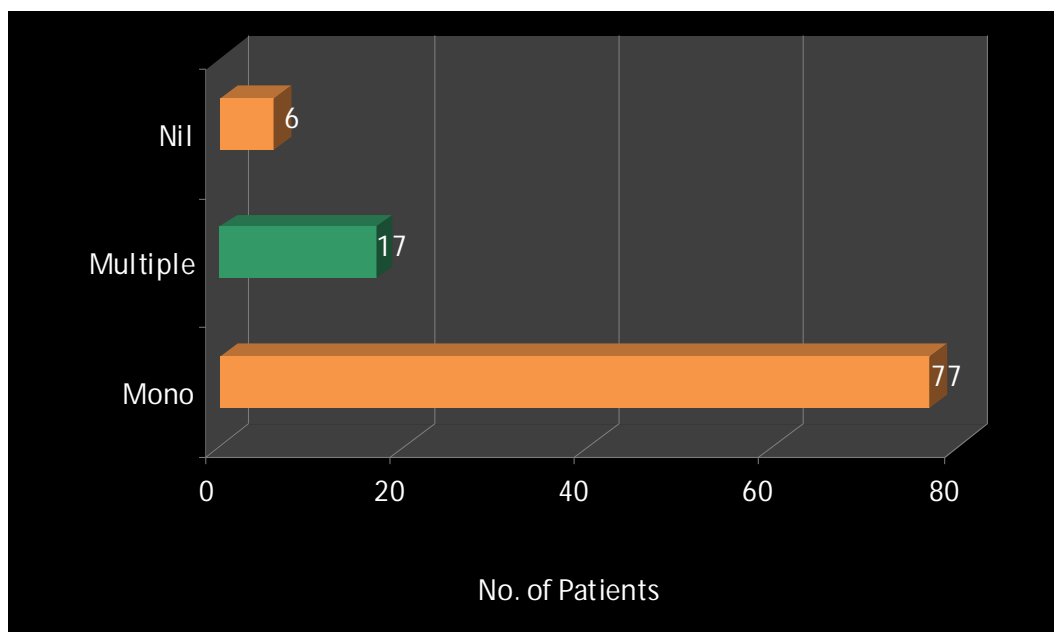
PARTNER ASSESSMENT

On analyzing the sex partners of the affected individuals, non specific urethritis was noted in 10 patient's partners, balanitis among 6

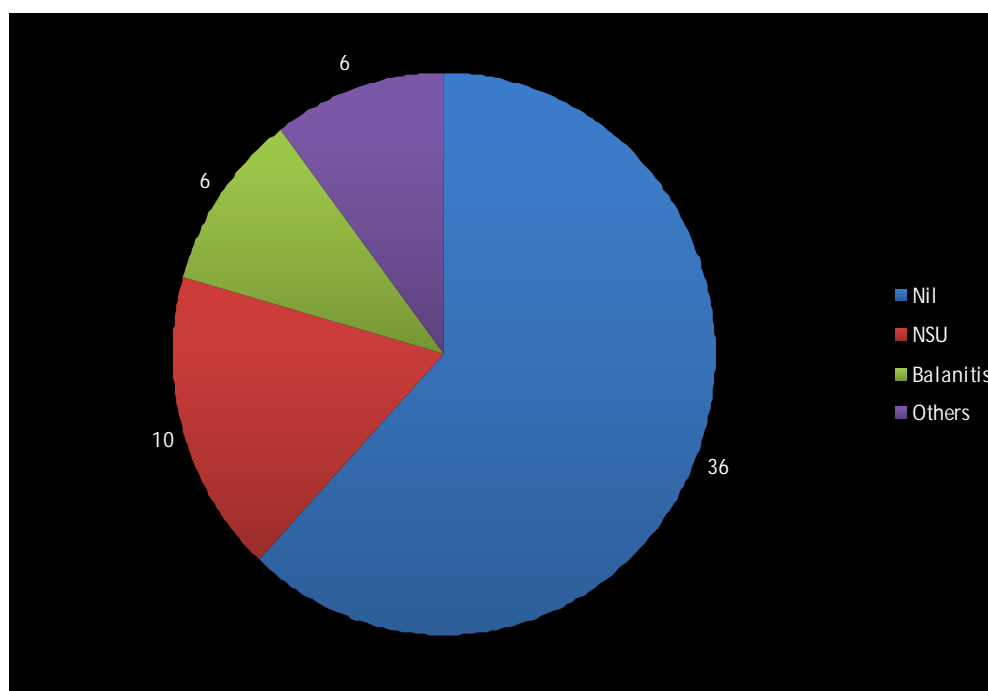
CONTRACEPTIVES & ASSOCIATION OF BV



NUMBER OF PARTNERS & BV ASSOCIATION



PARTNER TRACING



partners while no complaints among partners. 2 patient's partner had genital wart, 3 of the partners were PLHA, 1 positive for TPHA? 6 patients had no sexual partners. 30 partners did not come for evaluation.

- There was no significant difference in the distribution of partner assessment in between group 1 and group 2 (P=0.78).

CLINICAL FEATURES

	Group 1	Group 2	P value
Vaginal discharge	28 + 20	25 + 17	0.23
Saline mount	48	44	0.13
KOH mount	42	38	0.45
Vaginal pH	50	55	0.28
Cervix- Normal	36	35	0.36

VAGINAL DISCHARGE:

Vaginal discharge was profuse among 53 patients, moderate in 37 patients. P= 0.23 in between the two groups (in significant). 90 % of the study population showed vaginal discharge, while 84 % came with the complaint of vaginal discharge. 16 % were unaware or asymptomatic about the vaginal discharge.

There was no significant difference in the distribution of vaginal discharge in between group 1 and group 2 (p=0.23).

SALINE MOUNT FOR CLUE CELLS

Saline mount showed clue cells among 90 patients, clue cells and pus cells seen in 2 patients, clue cells absent in 8 patients. (p = 0.13 in significant) Clue cells were positive in 92 % of the study population

There was no significant difference in the distribution of saline mount between group 1 and group 2 (P=0.13).

KOH EXAMINATION

80 Patients showed positive whiff test and 20 patients were negative for whiff test. Both the group were comparable based on amine test.

VAGINAL pH

Vaginal pH was < 5 in 11 patients, 5- 5.5 in 72 patients and > 5.5 in 17 patients. (P = 0.08).

There was no significant difference in the distribution of vaginal pH between group 1 and group 2 (P=0.08).

CERVIX

There was no significant difference in the distribution of cervical status between group 1 and group 2 (P=0.36).

ENDOCERVICAL SMEAR

All the study patients were negative for Gonococci in Gram staining and culture.

URINE EXAMINATION

Urine samples were examined for sugar, albumin, pus cells and cast. 1 patient in metronidazole group & 5 patients in the lactobacillus.

Group showed positivity for urine sugar. Of them 4 patients were known diabetic and 2 were newly diagnosed. They were sent to the diabetology op.

VDRL & TPHA

2 patients in the lactobacillus group were reactive to VDRL in 1: 1 & 1: 2 dilutions respectively. But the TPHA reading was negative

VCTC

In both groups 2 patients were found newly positive for HIV and were sent for CD₄ count were > 500 cells / ml and they were under regular observation in ART centre after the study period. 2 patients were already positive for HIV and were on ART before the study period itself.

ULTRA SONO GRAM

Ultra sonogram was done at the end of 2 months. 41 patients in the lactobacillus group and 44 patients in the metronidazole group showed no abnormality in USG. While 8 patients in the lactobacillus group and 1 patient in metronidazole group showed fatty changes in the liver. As we did not take initial USG we were not able to correlate this finding with treatment outcome. 4 patients showed other abnormality like fibroid uterus, endometrial hyperplasia.

There was a significant difference in the distribution of USG between group 1 and group 2 (P=0.01).

AMSELS CRITERIA BETWEEN TWO STUDY GROUPS AT DIFFERENT TIME POINTS

Patients were assessed based on the following response grading in their follow up visits.

Amsels criteria

1 & 2 - Good response

3- 5 - No response

AMSEL'S CRITERIA

1 Week

In metronidazole group good response was noted in 46 patients (92%). In lactobacillus group 45 patients (90 %) showed good response.

1 month

48 patients in metronidazole group (100 %) and 50 in lactobacilli (100%) group showed good response. 2 patients in metronidazole group were lost from follow up.

2 month

43 patients (97.7%) showed good response in metronidazole group, while all the 50 patients (100%) in the lactobacilli group showed good response. Recurrence seen in 1 patient in

metronidazole group and 6 patients lost from follow up in metronidazole group.

6 months

32 patients (76.2%) in metronidazole group, 47 patients (94%) in lactobacillus group showed good response. Recurrence seen in both groups, 10 (23.8%) in metronidazole group and 3 (6%) in lactobacillus group. 8 patients lost from follow up in metronidazole group.

Time point	Amsels criteria	Group 1	Group 2	P-value
		No (%)	No (%)	
Day – 0	Good response	0 (0.0)	0 (0.0)	-
	No response	50 (100.0)	50 (100.0)	
Week – 1	Good response	46 (92.0)	45 (90.0)	1.00 (N.S.) [#]
	No response	4 (8.0)	5 (10.0)	
Month – 1	Good response	48 (100.0)	50 (100.0)	-
	No response	0 (0.0)	0 (0.0)	
Month – 2	Good response	43 (97.7)	50 (100.0)	0.47 (N.S.) [#]
	No response	1 (2.3)	0 (0.0)	
Month – 6	Good response	32 (76.2)	47 (94.0)	0.01 (Sig.) [@]
	No response	10 (23.8)	3 (6.0)	

[#]Fishers Exact Test (2-tailed) was used to calculate the P-Value

[@] Chi-square test with Yates' correction was used to calculate the P-Value

COMPARISON OF NUGENT'S SCORING BETWEEN TWO STUDIES GROUPS AT DIFFERENT POINTS OF TIME:

Patients were assessed based on the Nugent's scoring system during their follow up visits by the following response grading

Grade 1 - 0-3 score as Good response

Grade 2 - 4-6 score as moderate response

Grade 3 – 7-10 score as no response to treatment.

Time point	Nugent's scoring	Group 1	Group 2	P-value*
		No (%)	No (%)	
Day – 0	Good response	0 (0.0)	0 (0.0)	-
	Moderate response	0 (0.0)	0 (0.0)	
	No response	50 (100.0)	50 (100.0)	
Week – 1	Good response	20 (40.0)	28 (56.0)	0.16 (N.S.)
	Moderate response	24 (48.0)	20 (40.0)	
	No response	6 (12.0)	2 (4.0)	
Month – 1	Good response	39 (81.3)	42 (84.0)	0.56 (N.S.)
	Moderate response	8 (16.6)	8 (16.0)	
	No response	1 (2.3)	0 (0.0)	
Month – 2	Good response	27 (61.4)	49 (98.0)	<0.0001 (Sig.) [@]
	Moderate response	17 (38.6)	1 (2.0)	
	No response	0 (0.0)	0 (0.0)	
Month – 6	Good response	13 (30.9)	39 (78.0)	<0.0001 (Sig.)
	Moderate response	25 (59.5)	11 (22.0)	
	No response	4 (9.5)	0 (0.0)	

*Chi-square test was used to calculate the P-Value

[@] Chi-square test with Yates' correction was used to calculate the P-Value

NUGENT'S SCORING SYSTEM

1 Week:

In metronidazole group, good response seen in 20 patients (40%), moderate response in 24 patients (48%) and no response to treatment in 6 patients (12%).

Under lactobacilli group 28 patients (56%) showed good response, 20 patients (40%) showed moderate response and no response to treatment seen in 2 patients (4%).

Two groups were comparable ($P = 0.16$)

1 month:

In metronidazole group 39 patients (81.3%) showed good, 8 patients (18.4%) showed moderate and 1 patient (2%) did not respond to treatment. 2 patients were lost from follow up.

In the lactobacilli group the response was good in 42 patients (84%) and moderate response in 8 patients (16%).

Two groups were comparable ($P = 0.56$)

2 months:

27 patients (61.4%) showed good and 17 patients (38.6%) moderate response to treatment. 6 patients were lost from follow up in metronidazole group.

In lactobacilli group 49(98%) showed good and 1 patient (2%) showed moderate response to treatment.

Lactobacillus group was better than metronidazole group ($P = < 0.0001$)

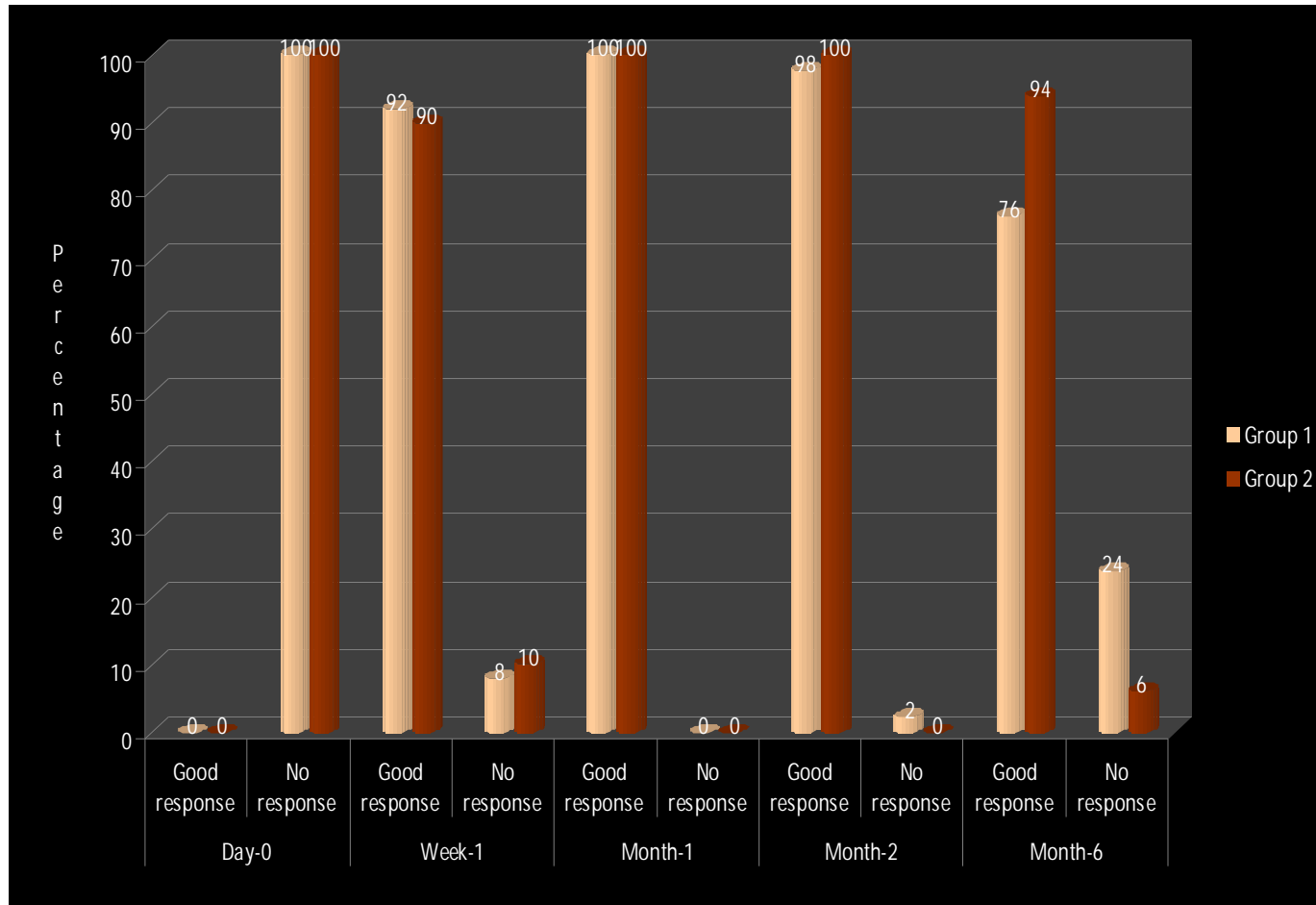
6 months:

Under metronidazole group 13 patients (30.9%) showed good, 25 patients (59.5%) elicited moderate and 4 patients (9.5%) showed no response to treatment. 8 patients were lost from follow up.

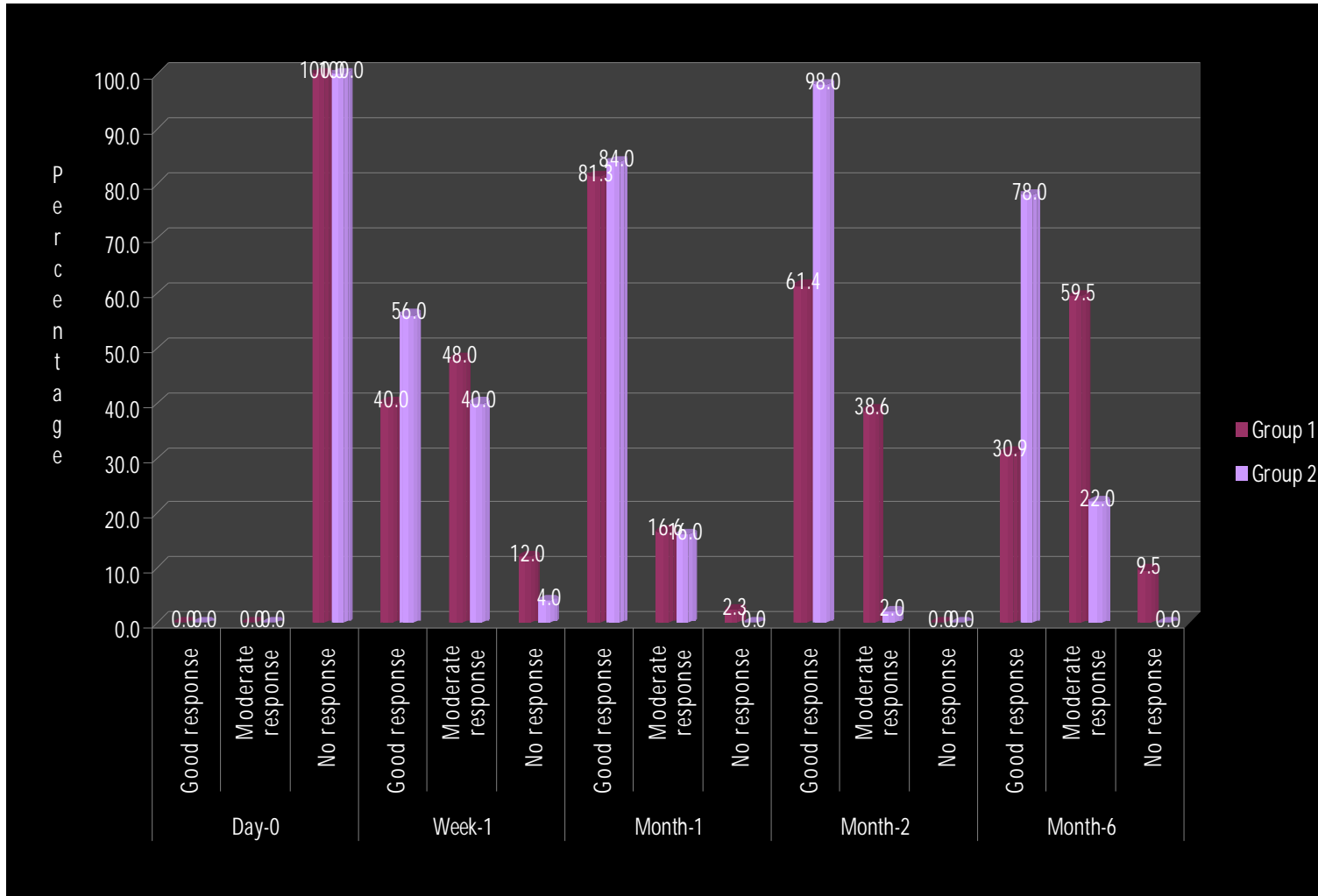
In lactobacillus group 39 (78%) showed good and 11(22%) showed moderate response to treatment respectively.

Lactobacillus group showed better therapeutic response in this period. ($P < 0.0001$)

AMSEL'S CRITERIA



NUGENT'S SCORING SYSTEM



Discussion

DISCUSSION

DEMOGRAPHIC CHARACTERS

AGE

The mean age of patients in the study group was 32.4± 8.4. This correlated with the earlier reported finding that BV was more common in the sexually active, reproductive age group.⁵⁹

DURATION

The mean duration of symptoms in the study was 2 months. The longest period observed was 10 years and the shortest period, 1 week. This depends upon the individual's sensory adaptation with the existing disease.

COMPLAINTS

Genital discharge was given as a chief complaint in 67 patients, genital itching was noticed in 10 individuals and genital odour in 7 patients.

16 patients were asymptomatic.

MENSTRUAL HISTORY

Most of them (86 %) had regular menstrual cycles. There were no menstrual disorders in the patients.

MARITAL STATUS

In our study 92% were married. In the previous Patiala study done in 80 randomly selected patients, 37.5% were married. Our study confirms the alteration of vaginal ecoflora after commencing sexual activity.⁶⁰

This study coincides with the previous South Indian study which stated the high incidence of Bacterial vaginosis in married, sexually active individuals.⁶¹

57 / 100 got married before the age of 20 years. This study confirms that younger age at marriage has association with

Bacterial vaginosis which was already quoted by Australian study and the study by Larrson et al.⁶¹

PARTNER TRACING

58 patient's sexual partners were examined and investigated. 36 individuals showed no abnormality. Non specific arthritis was seen among 10 patients, Balanitis noted in 6 partners, 2 with genital wart, 3 PLHA and 1 showed TPHA positivity.

We were not able to trace all the partners of the patients included in the study group.

PREVIOUS VENEREAL DISEASE

35 patients had come with recurrent episodes of Bacterial vaginosis, 2 patients in the Metronidazole group (genital ulcer -1, Vaginal candidiasis -1) and one patient in the Lactobacillus group presented with past history of cervicitis. One patient in the metronidazole group had a past history of genital herpes and bacterial vaginosis.

Our study consisted of (15 + 20) 35 recurrent cases and 65 newly detected Bacterial vaginosis patients.

ASSOCIATED OTHER DISORDERS

7 patients in each group had associated systemic disorders. In the Metronidazole group 1 patient had diabetes mellitus, 2 had systemic hypertension, 1 had hypo thyroidism, 1 had rheumatic fever, 1 had associated skin disorder, 1 patient was a PLHA on ART -1.

In the Lactobacillus group 5 patients had Diabetes mellitus , 1 had Oral candidiasis, 1 was a PLHA on ART.

In each group 2 patients found newly positive for HIV and were referred to ART centre . 2 patients in the Lactobacillus group were reactive to VDRL in 1:1 and 1:2 dilution but none of them showed TPHA positivity .

OBSTETRIC HISTORY

This study outlines the direct correlation of the number of pregnancies and the incidence of Bacterial vaginosis. Australian study also correlated the above point.⁶³

The two trial groups had comparable values (P=0.25)

FAMILY PLANNING

Majority had undergone permanent sterilization process 61/100. Intra uterine devices were used in 18 patients. Only 2 patients used barrier methods.

As barrier methods are cited to have some protective effects against acquisition of Bacterial vaginosis , this study correlated the preventive effect of condoms as barrier users had less prevalence compared to other modes of contraception.

CLINICAL DETERMINANTS

SYMPTOMS

In the study done by Mathew R et al in pregnant individuals, those who were in the 2nd trimester out of 200, 50 were asymptomatic.⁶⁴ Though it included only pregnant patients, the study showed that 25% were asymptomatic. In our study 16% were asymptomatic.

VAGINAL DISCHARGE

Vaginal discharge was profuse in 53 patients and moderate among 47 patients. 90% showed abnormal vaginal discharge.

CERVICAL STATUS

Cervix was normal in 71 patients, eroded cervix noted among 21 patients and neobothian follicle in 2 patients. (P = 0.36)

LABORATORY INVESTIGATION

Saline mount:

93 / 100 individual's vaginal discharge showed positivity for clue cells on saline mount.

Whiff test

80 % showed positivity for Whiff test.

Vaginal pH

All of them had alkaline vaginal discharge. Vaginal pH was 4.5 – 5 in 11 , 5-5.5 in 72 patients and > 5.5 in 17 individuals.

ULTRA SONOGRAM

USG was normal in 85 patients , 9 patients showed fatty liver 1 showed L adnexal cyst , 2 had fibroid uterus , 1 patient had adeno cystosis and endometrial hyperplasia (P = 0.01 significant)

Out of 9 individuals with fatty liver, 8 were in the Lactobacillus group, 1 from the Metronidazole group. This association has to be looked for in further studies with lactobacillus.

THERAPEUTIC EFFICACY

Therapeutic efficacy was observed at the end of 1 week, 1 month , 2 months and 6 months of starting treatment.

Analysis of treatment response was based on Amsels criteria and Nugent's scoring system.

On comparing the two groups based on Amsel's criteria at 1 week, 1 month and 2 months period both the drugs groups had comparable therapeutic benefits. But after 6 months the P value was significant 0.01. Metronidazole group showed good response in 76.2% and lactobacillus group showed good response in 94%

On comparing the two groups based on Nugent's scoring system at 1 week and 1 month period both the groups were comparable. While at 2 months and 6 months period lactobacillus group was better than metronidazole group ($P = < 0.0001$ in both time period) At the end of study period in metronidazole group 30.9 % showed good therapeutic response and 59.5 % showed moderate response.

In lactobacilli group, 78% showed good and 22% showed moderate response. In the metronidazole group 8 patients were lost from follow up.

The follow up period was very long. The treatment course also differed. For metronidazole 7 days course while for lactobacilli

group the treatment period was 2 months. The cost was also high for lactobacilli.

After 2 months of therapy, USG showed fatty liver changes in 8 patients under lactobacilli group and 1 in metronidazole group. In literature few cases of liver abscess reported after lactobacillus ingestion. Whether our finding of fatty changes in liver was due to lactobacilli was questionable. It would have been a valid finding if USG was done at the initial visit and repeated at follow up visits. This is the constraint in our study. But this is to be carefully looked for in further studies.

In the study done by Reid G et al, oral lactobacillus for 60 days was compared with placebo. The study showed no adverse effects in lactobacillus users. Culture findings suggested a significant depletion in yeast and coliforms at day 28, 60 and 90 for lactobacillus treated group versus controls. Microscopy revealed restoration of normal lactobacillus colonized microflora in 37% women during lactobacilli treatment compared to 13% in the placebo group. (P=0.02)⁶⁵

The Nigerian study which compared the efficacy of oral lactobacillus for 7 days with 0.75 % metronidazole gel it showed 90 % efficacy in lactobacillus group and 55 % improvement in metronidazole group. This report was taken at the end of 1 month.⁶⁶

In another study by Kingsley Anukkam et al which included 2 study groups both groups initially had a course of Metronidazole for 1 week . After that group 1 patient were treated with Probiotics and Group 2 with a Placebo. 88% in the probiotic group and 40 % in the Placebo group were cured at the end.⁶⁷

These studies like our study showed the effectiveness of probiotics in the management of Bacterial vaginosis. In this era of emerging antibiotics resistance ,chronicity and the recurring nature of BV they are the definite doubtless alternative to antibiotics.

SIDE EFFECTS NOTED

In Metronidazole group 4 patients complained of nausea and 5 patients complained of metallic taste.

3 in the Lactobacillus group and 1 in the Metronidazole group were treated for Candidiasis in the follow up period.

On doing ultra sonogram 8 patients (16 %) in the Lactobacillus group and 1 (2 %) in the Metronidazole group 25 showed fatty change in the liver. This is to be associated in further studies.

RECURRENCE

Based on Amsel's criteria at the end of 1 month both the groups showed good response in all the patients. But recurrence noted in 1 and 10 patients at 2 months and 6 months period in metronidazole group and in 3 patients in the lactobacillus group at 6 months period.

Based on Nugent's scoring system ,in metronidazole group at 2 months of completing treatment good and moderate response to treatment was noted in 61.4% and 38.6% respectively. But at the end of 6 months period no response to treatment seen in 9.5% . While lactobacillus group showed nil recurrence at the end of 6 months.

Conclusion

CONCLUSION

1. Bacterial vaginosis was seen commonly in the age group of 24 – 40 years.
- Menstrual cycle irregularities had no association with prevalence. BV was seen in both married and unmarried as well as those who never had sexual intercourse. But the incidence in unmarried & those who never had sexual intercourse is minimal.
 - The earlier age at marriage was directly proportionate to incidence of BV.
 - 77 %, Majority had mono partners, 17 % had multiple sexual partners and 6% denied sexual exposure.
 - The mode of delivery had no significant association. Number of deliveries was directly proportionate to acquisition of BV.
 - Moderate to profuse genital discharge was the commonest observed finding.
 - In saline mount 90 % showed clue cells positivity. Whiff test was positive in 80% , Vaginal pH was > 5 in all the patients.

2. By Amsel's criteria, the two groups were comparable in the 1 week, 1 month, and 2 months period ($P = 1, 0.56, 0.47$) while at 6 months period lactobacillus group was better than metronidazole group

By Nugent's criteria, the two groups were comparable in the 1 week, 1 month period. But at 2 months and 6 months period lactobacillus group showed better therapeutic benefits.

3. Few tolerable side effects were noticed in the metronidazole group. Though both the groups completed their treatment schedule, 8 patients lost during follow up in the metronidazole group.

4. Recurrence was noted in both the study groups at the end of 6 months, based on Amsel's criteria. In metronidazole group 1 and 10 patients showed recurrence at 2 months and 6 months period. In lactobacillus group 3 patients showed recurrence at the end of 6 months.

Recurrence was noted in 4 patients (9.5%) in the metronidazole group at the end of 6 months, while no recurrence seen in lactobacillus users based on Nugent's scoring system.

Annexure

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Proforma

PROFORMA

COMPARATIVE STUDY IN THE MANAGEMENT OF BACTERIAL VAGINOSIS LACTOBACILLUS Vs METRONIDAZOLE

NAME AGE OCCUPATION OP/ IP NO

ADDRESS

COMPLAINTS:

Abnormal genital discharge	Burning micturition
Genital ulcer	Menstrual irregularities
Genital swelling	Inguinal swelling
Genital itching	Others

HISTORY OF PRESENTING ILLNESS

1. Genital discharge

Duration	Smell : Foul smelling / not so
Itching: Present / Absent	Associated with menstruation /
	Sexual intercourse / Nil

2. H/O Recent intake of antibiotics for long duration (More than 1 month)

3. Others if any specify

TREATMENT HISTORY:

Underwent any treatment / Nil

L/E:

Inguinal nodes

just palpable / Enlarged Discrete / Matted Tender / Nontender
Signs of inflammation Present / Absent

S/ E: 1. External urethral orifice Congested / Normal

2. Vagina Mass / Ulcer / Growth / scar / Vesicles
Genital ulcer Single / multiple
Painful/ Painless

3. Vaginal discharge Scanty / moderate/ profuse
Foul smelling/ Not so
Mucoid / Mucopurulent/ frothy /
thick curdy white/ Homogenous uniformly
adherent

4. Cervix Healthy / Eroded / IuCD / discharge/ Polyp/
Nebothian follicle / Growth

EXAMINATION OF ANAL ORIFICE

ORAL ORIFICE

SKIN / MUCOSA

BONES / JOINTS

INVESTIGATION

1. Urine Albumin Hb
 Sugar

2. VDRL TPHA

3. VCTC

4. Vaginal discharge

Vaginal pH	Sniff / Whiff	Saline mount	10 % KOH

5. Smears Taken

Vaginal				Cervical	Urethral
Epithelial	Clue cell	Pus cell	Lactobacillus		

AMSELS CRITERIA

Vaginal discharge

2. Vaginal pH

3. Sniff / Whiff

4. Clue cells

5. Lactobacillus

NUGENTS SCORING

(Gardnerella, Mobiluncus, Prevotella / oil field)

1	2	3	4
< 1	1- 5	6- 30	> 30

(Lactobacilli / oil field)

> 30	6- 30	1-5	<1
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BV = Score 7-10 : Nil BV = Score 0-

TREATMENT MODALITIES TRIED

1. Tab. Metronidazole 400 mg b.d x 7 days

2. Cap. Lactobacillus 1 bd x 1 month, followed by 1 o d x Next 1 month

FOLLOW UP

1 week 1 month 2 months 6 months

1. Vaginal discharge

2. Odor

3. Vaginal pH

4. Amine test (Sniff / Whiff)

5. Saline mount

6. Gram stain

சுய ஒப்புதல் படிவம்

ஆய்வு செய்யப்படும் தலைப்பு

பெண் பிறப்புறுப்பில் வெள்ளைப் பருதல் - மெட்ரோனிடிசால்,
லாக்டோ பாசிலஸ் ஒரு ஒப்பீடு

ஆராய்ச்சி நிலையம் : பால்வினை நோய் துறை,
அரசு பொது மருத்தவமனை,
சென்னை-600 003.

பங்கு பெறுபவரின் பெயர் :
பங்கு பெறுபவரின் எண் :
பங்கு பெறுபவர் இதனை (✓) குறிக்கவும்

மேலே குறிப்பிட்டுள்ள மருத்துவ ஆய்வின் விவரங்கள் எனக்கு விளக்கப்பட்டது. என்னுடைய சந்தேகங்களை கேட்கவும், அதற்கான தகுந்த விளக்கங்களை பெறவும் வாய்ப்பளிக்கப்பட்டது.

நான் இவ்வாய்வில் தன்னிச்சையாகதான் பங்கேற்கிறேன். எந்த காரணத்தினாலோ எந்த கட்டத்திலும் எந்த சட்ட சிக்கலுக்கும் உட்படாமல் நான் இவ்வாய்வில் இருந்து விலகி கொள்ளலாம் என்றும் அறிந்து கொண்டேன்.

இந்த ஆய்வு சம்மந்தமாகவோ, இதை சார்ந்த மேலும் ஆய்வு மேற்கொள்ளும் போதும் இந்த ஆய்வில் பங்குபெறும் மருத்துவர் என்னுடைய மருத்துவ அறிக்கைகளை பார்ப்பதற்கு என் அனுமதி தேவையில்லை என அறிந்து கொள்கிறேன். நான் ஆய்வில் இருந்து விலகிக் கொண்டாலும் இது பொருந்தும் என அறிகிறேன்.

இந்த ஆய்வின் மூலம் கிடைக்கும் தகவல்களையும், பரிசோதனை முடிவுகளையும் மற்றும் சிகிச்சை தொடர்பான தகவல்களையும் மருத்துவர் மேற்கொள்ளும் ஆய்வில் பயன்படுத்திக் கொள்ளவும் அதை பிரசுரிக்கவும் என் முழு மனதுடன் சம்மதிக்கிறேன்.

இந்த ஆய்வில் பங்கு கொள்ள ஒப்புக்கொள்கிறேன். எனக்கு கொடுக்கப்பட்ட அறிவுரைகளின்படி நடந்து கொள்வதுடன் இந்த ஆய்வை மேற்கொள்ளும் மருத்துவ அணிக்கு உண்மையுடன் இருப்பேன் என்றும் உறுதியளிக்கிறேன். என் உடல் நலம் பாதிக்கப்பட்டாலோ அல்லது எதிர்பாராத வழக்கத்திற்கு மாறான நோய்க்குறி தென்பட்டாலோ உடனே அதை மருத்துவ அணியிடம் தெரிவிப்பேன் என உறுதி அளிக்கிறேன்.

பங்கேற்பவரின் கையொப்பம் இடம் தேதி

கட்டைவிரல் ரேகை

பங்கேற்பவரின் பெயர் மற்றும் விலாசம்

ஆய்வாளரின் கையொப்பம் இடம் தேதி

ஆய்வாளரின் பெயர்

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INSTITUTIONAL ETHICAL COMMITTEE
GOVERNMENT GENERAL HOSPITAL & MADRAS MEDICAL COLLEGE,
CHENNAI-600 003.

Telephone: 044-2530 5000

Fax : 044 - 25305115

K.Dis.No.006859/P & D3/Ethics/Dean/GGH/09

Dated:23-03-2009

Title of the work

" comparative study of management of Bacterial vaginosis with Lactobacillus vs Metronidazole "

Principal Investigator

Dr. Narmadha . S

Department

M.D.D.V.L., post graduate student
Institute of venerology, MMC, Ch-3.

The request for an approval from the Institutional Ethical Committee (IEC) was considered on the IEC meeting which is held on 31st March at 2 P.M in Government General Hospital, Deans, Chamber, Chennai-3.

The members of the Committee, the Secretary and the Chairman are pleased to approve the proposed work mentioned above, submitted by the principal investigator .

The principal investigator and their term are directed to adhere the guidelines given below.

1. You should get detailed informed consent from the patients/participants and maintain confidentiality.
2. You should carry out the work without detrimental to regular activities as well as without extra expenditure to the Institution or Government.
3. You should inform the IEC in case of any change of study procedure, site and investigation or guide.
4. You should not deviate form the area of the work for which I applied for ethical clearance.
5. You should inform the IEC immediately, in case of any adverse events or serious adverse reactions.
6. You should abide to the rules and regulations of the institution(s)
7. You should complete the work within the specific period and if any extension of time is required, you should apply for permission again and do the work.
8. You should submit the summary of the work to the ethical committee on completion of the work.
9. You should not claim funds from the Institution while doing the work or on completion.
10. You should understand that the members of IEC have the right to monitor the work with prior intimation.

SECRETARY
IEC, GGH, CHENNAI

CHAIRMAN
IEC, GGH, CHENNAI

DEAN
GGH & MMC, CHENNAI

DM 30-03 (3)

Master Chart

GROUP - 1 METRONIDAZOLE

No.	AG	Com	Dur	PVD	O	Men	Mar	NP	Con	Obs	FP	VD	Cx	Sal	KOH	pH	A0D	A1w	A1m	A2m	A6m	N0d	N1w	N1m	N2m	N6m	USG
1	36	1	2	1	2	1	1	1	1	1	1	1	1	1	1	5	2	1	1	1	2	3	3	2	2	2	1
2	48	1	6	1	2	3	1	1	3	1	1	1	1	1	1	4.5	2	1	1	1	1	3	2	2	1	1	1
3	51	1	1	1	1	3	1	1		1	4	1	1	4	1	5	2	2	1	1	1	3	2	1	1	1	1
4	23	1	120	1	2	2	1	1		1	1	1	1	1	1	4.5	2	1	1	1	1	3	1	1	1	1	1
5	27	1	0.3	3	2	1	1	1		1	1	1	1	1	2	5	2	1	1			3	2	1			1
6	20		0.3	1	2	1	1	1	1	1	2	1	1	1	1	4.5	2	1	1	1	1	3	2	1	1	2	1
7	22	1	2	3	2	1	1	1		3	4	1	1	1	1	5	2	1	1	1	1	3	2	1	1	2	1
8	35	3	0.3	3	2	2	1	2	1	1	4	1	1	1	1	4.5	2	1	1	1	1	3	1	1	2	2	1
9	26		120	2	2	1	1	1	3	1	4	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	2
10	45		0.1	3	2	1	1	1	1	1	1	1	1	1	1	6	2	1	1	1	1	3	3	3	2	3	2
11	46	2	24	3	2	1	1	1		1	1	1	2	1	1	5.5	2	2	1	1	1	3	3	2	2	2	1
12	41	2	1	1	2	1	1	1		1	1	2	1	1	1	4.5	2	1	1	1	1	3	2	1	1	1	1
13	33	1	1	3	2	1	1	1	4	1	3	1	1	1	2	5.5	2	1	1	2	2	3	1	1	2	2	1
14	21	1	6	3	2	1	1	1	1	6	4	1	1	1	2	5.5	2	1	1	1	2	3	2	1	2	2	2
15	21	1	0.3	3	2	1	2	3		6		2		1	1	6	2	1	1	1	1	3	2	1	1	1	1
16	33	1	0.3	3	2	1	1	2	1	1	1	1	2	1	1	4.5	2	1	1	1	1	3	1	1	2	2	1
17	35		0.3	3	2	1	1	2	2	1	4	1	1	1	1	6	2	1	1	1	2	3	1	1	1	1	1
18	29	1	0.5	3	2	1	1	2	1	2	1	1	1	1	2	5	2	1				3	1	1			1
19	27	1	2	3	2	1	1	1	3	1	1	1	1	1	1	5	2	1	1	1	1	3	1	1	1	1	1
20	36	2	0.1	3	2	1	1	1	1	1	1	2	1	1	2	5	2	1	1	1	2	3	1	1	2	3	1
21	22	1	0.3	3	2	1	1	1		6	4	1	1	4	1	6	2	1	1	1	1	3	1	1	2	2	1
22	43	2	48	1	2	1	1	1	1	1	4	1	1	1	1	5	2	1	1	1	2	3	2	1	2	2	1
23	21	1	0.6	3	2	1	1	1	4	1	4	2	1	1	2	6	2	1	1	1	1	3	1	1	1	1	3
24	26	2	2	1	2	1	1	1	1	3	4	2	1	1	1	5.5	2	1	1	1	1	3	3	1	1	2	1
25	43	1	1	3	2	2	1	1		1	1	2	2	1	1	5	2	1	1	1	2	3	1	1	1	2	1
26	26	1	1	3	2	1	1	1	1	1	4	2	1	1	1	4.5	2	1	1	1	1	3	2	2	2	2	1
27	27	1	2	3	2	1	1	2		1	1	2	2	1	1	6	2	1	1	1	2	3	2	1	1	2	1
28	35	1	2	3	2	1	2	3		6		2		1	1	4	2	2	1	1	1	3	2	1	1	2	1
29	45		1	1	1	1	1	1	1	6	4	1	1	1	1	5	2	1	1			3	1	1			1
30	32	1	3	3	2	1	1	2		2	1	1	1	1	1	5.5	2	2	1	1	1	3	3	2	1	1	1
31	35	1	2	3	1	2	1	1	4	1	1	1	1	1	1	5	2	1	1	1	1	3	2	1	1	1	1
32	45	3	2	3	1	1	1	1		2	3	1	1	1	1	5	2	1	1	1	1	3	1	1	1	1	1
33	27	2	3	3	2	1	1	1	1	2	3	1	1	1	1	5	2	1	1	1	2	3	2	1	1	2	1
34	30		1	3	2	1	1	1	2	5	1	1	2	1	1	6	2	1				3	1				1
35	51		1	1	2	3	1	2	3	1	1	2	1	1	1	5	2	1	1	1	1	3	1	1	1	2	1
36	38	1	6	1	2	1	1	2	1	1	1	1	1	1	1	6	2	1	1	1	2	3	2	2	2	2	1
37	47	1	0.3	1	2	1	1	1		1	3	1	1	1	1	5	2	1	1	1	1	3	2	1	1	2	1
38	26	1	36	1	1	1	1	1		5	4	2	2	1	1	5	2	1	1	1	1	3	2	1	1	2	1
39	32		6	1	2	1	1	1	2	5	1	2	1	1	1	6	2	1	1	1	2	3	2	1	2	2	1
40	45	1	2	1	2	2	1	2	1	1	1	2	1	1	1	5	2	1	1	1	1	3	1	1	2	2	1
41	36	1	66	1.2	2	1	1	1		5	1	2	2	1	1	5	2	1	1	1	2	3	2	1	2	2	1
42	30		60	3	2	1	1	1		2	1	2	1	1	1	6	2	1	1			3	1	1			3
43	37	3	7	3	2	1	1	1		5	1	2	2	1	1	5	2	1	1	1	1	3	1	1	1	1	1
44	41	1	1	1	1	1	1	1	2	4	1	2	1	1	1	5.5	2	1	1	1	1	3	2	1	1	1	1
45	23	1	24	3	1	1	2	3				2		1	1	5	2	1	1	1	1	3	1	1	1	1	1
46	30	1	3	3	2	1	1	1	1	2	3	2	1	1	1	6.5	2	1	1	1	1	3	2	2	2	3	1
47	27	1	1	3	2	1	1	1		1	1	2	2	1	1	5.5	2	1	1	1	1	3	3	2	1	2	1
48	20	1	2	1	2	1	1	2		5	3	1	2	1	1	5.5	2	1	1			3	2	1			2
49	40	1	1	1	2	1	1	1	1	2	4	1	1	2	2	5.5	2	1	1	1	1	3	2	1	1	3	1
50	32	1	2	1	2	1	2	3				1		2	2	5	2	1	1	1	1	3	2	2	2	2	1

GROUP 2 LACTOBACILLUS

No.	AG	Com	Dur	PVD	O	Men	Mar	NP	Con	Obs	FP	VD	Cx	Sal	KOH	pH	A0D	A1w	A1m	A2m	A6m	N0d	N1w	N1m	N2m	N6m	USG
1	30	1	12	3	2	1	1	1	1	1	1	2	2	1	1	5.5	2	1	1	1	1	3	2	1	1	1	1
2	26		1	1	2	1	1	1	1	1	1	2	2	1	1	5	2	1	1	1	1	3	1	1	1	1	1
3	38	1	2	3	2	1	1	1	1	1	4	2	1	2	2	4.5	2	1	1	1	1	3	1	1	1	2	2
4	23	2	1	1	2	1	1	1	1	1	1	2	1	2	2	5	2	1	1	1	1	3	1	1	1	1	1
5	34	1	24	3	1	2	1	1	1	1	1	1	1	2	2	5	2	1	1	1	1	3	3	2	1	1	3
6	25	1	1	3	2	1	1	1	2	1	1	1	1	2	2	5.5	2	1	1	1	1	3	1	1	1	2	2
7	35		1	3	2	1	1	1	1	1		2	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	1
8	44	2	5	3	2	1	1	2		1	1	1	1	1	1	5	2	1	1	1	1	3	1	1	1	1	2
9	29	1	0.3	3	2	1	1	1		5	2	1	3	2	2	5	2	1	1	1	2	3	1	1	1	2	1
10	27	1	2	3	2	1	1	1	4	1	3	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	1
11	35	1	1	3	2	1	1	1	1	1	1	1	3	2	2	5.5	2	1	1	1	1	3	1	1	1	1	3
12	34	3	24	3	1	2	1	1	1	1	1	1	1	1	1	5	2	2	1	1	1	3	2	1	1	1	3
13	13	2	3	1	2	1	1	1	2	1	3	2	1	1	1	4	2	2	1	1	1	3	2	1	1	1	1
14	54	1	0.5	3	2	3	1	1	1	5		2	1	1	1	5	2	1	1	1	1	3	2	2	1	2	1
15	30	1	4	3	2	1	1	1	1			2	1	1	1	6	2	1	1	1	1	3	1	1	1	1	1
16	30	1	1	1	2	1	1	1	1	1	1	2	2	1	1	4.5	2	1	1	1	1	3	1	1	1	1	1
17	38	1	2	3	2	1	1	1	1	1	1	2	2	1	2	5	2	1	1	1	1	3	2	1	1	2	1
18	33	3	1	3	2	1	1	1	1	1	1	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	2	1
19	35		1	3	2	2	1	1	1	1		2	1	1	1	5	2	1	1	1	1	3	1	1	1	1	3
20	30	1	120	3	2	1	1	1	2	1	3	2	2	1	2	5	2	1	1	1	1	3	1	1	1	1	1
21	29	3	2	3	2	1	1	1	1	1	3	2	1	1	2	6	2	1	1	1	2	3	2	1	1	2	1
22	36	1	7	1	2	1	1	1	1	1	1	2	1	1	1	5.5	2	1	1	1	1	3	2	2	1	1	1
23	34	1	96	2	2	1	1	1	3	1	1	1	1	1	1	6	2	1	1	1	1	3	1	1	1	1	1
24	40	2	1	1	1	1	2	1	1			1	2	1	1	5	2	2	1	1	1	3	2	1	1	1	3
25	27	1	1	3	2	1	1	1	1	1	1	1	1	1	1	5.5	2	1	1	1	2	3	2	2	1	2	1
26	23	1	0.5	3	2	1	1	1	4	1	3	1	1	1	1	5	2	1	1	1	1	3	1	1	1	2	3
27	30		0.3	3	1	1	1	1	1	1	3	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	1
28	27	1	1	3	2	1	1	1	2	1	1	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	1
29	37	1	5	3	2	1	2	3				1		1	1	5	2	1	1	1	1	3	2	1	1	1	1
30	40	1	4	3	1	1	1	1	1	1		1	1	1	1	6	2	2	1	1	1	3	2	1	1	1	3
31	30	1	2	3	1	1	1	1	4	1	1	1	1	1	1	5	2	1	1	1	1	3	1	1	1	2	1
32	28	1	4	3	2	1	1	1	1	2	1	1	2	1	1	5.5	2	1	1	1	1	3	3	2	1	1	3
33	43	1	12	3	2	1	1	1	2	1	1	1	2	1	1	5	2	1	1	1	1	3	2	1	1	1	1
34	38	1	1.5	3	2	2	1	1	1	1	1	1	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	1
35	37	1	3	3	2	1	1	1	1	5		2	1	1	2	5.5	2	1	1	1	1	3	1	1	1	1	3
36	21	1	1.5	1	2	2	2	1	3			2	1	1	1	5	2	1	1	1	1	3	1	1	1	1	1
37	27	1	48	1	2	1	1	1	1	2	2	1	2	2	1	5	2	1	1	1	1	3	1	1	1	1	3
38	22	1	24	1	2	1	1	2	1	5	3	2	1	1	1	5.5	2	1	1	1	1	3	1	1	1	1	3
39	23	1	2	1	2	1	1	1		1	1	1	1	1	1	5	2	1	1	1	1	3	1	1	1	1	1
40	26	1	24	3	2	1	1	1	1	1		2	1	1	1	5	2	1	1	1	1	3	1	1	1	1	1
41	45	1	3	3	1	1	1	2		2	1	1	1	1	1	5	2	1	1	1	1	3	2	1	1	1	1
42	40		0.3	3	2	1	1	1	2	1	1	2	2	1	1	5	2	1	1	1	1	3	2	1	1	1	3
43	45	3	5	1	2	1	1	1	1	1		2	2	1	2	5	2	1	1	1	1	3	1	1	1	1	1
44	20	1	2	1	2	1	1	2		3		2	1	1	1	6	2	1	1	1	1	3	2	1	1	1	1
45	25	1	1	3	2	1	1	1	1	1	3	1	1	1	1	5.5	2	1	1	1	1	3	2	1	1	1	1
46	29		6	1	2	1	1	1	1	1	2	1	2	1	1	6	2	1	1	1	1	3	1	1	1	1	1
47	36	1	3	3	2	1	1	2		5	1	2	1	1	1	5.5	2	1	1	1	1	3	2	1	1	1	2
48	20	1	2	1	2	1	1	1		1	3	2	1	1	1	5	2	1	1	1	1	3	2	2	2	2	2
49	19		3	3	2	1	2	3				2		1	1	5.5	2	1	1	1	1	3	2	2	1	1	1
50	29	1	8	1	2	1	1	2	1	2	1	2	1	1	1	5.5	2	1	1	1	1	3	2	2	1	1	1

KEY TO MASTER CHART

- NO - Serial number
- Ag - Age in years
- Com - Complaint 1- Genital discharge ,2- Itching ,3- Malodor in genitals
- Dur - Duration in months
- PVD - Previous venereal disease 1- similar, 2- Other, 3- Nil
- O - Other illness 1- Present , 2- Absent
- Men - Menstrual history 1- Regular, 2- Irregular
- Mar - Marital history 1- Married , 2- Unmarried
- NP - Number of partner 1- Mono , 2- Multiple
- Con - Contact tracing 1- Nil , 2- Urethritis, 3 – Balanitis , 4- Others
- Obs - Obstetric history 1- normal delivery, 2- LSCS, 3- Abortion
- FP - Family planning 1- Sterilization, 2- barrier, 3- IUCD, 4- Nil
- VD - Venereal disease 1- Profuse, 2- Moderate
- Cx - Cervix 1- Normal, 2- Erosion, 3- Other
- Sal - Saline mount 1- Clue cells present, 2- Clue cells absent, 3- Other
- KOH - KOH mount 1- Odour present, 2- Odour absent
- Ph - Vaginal pH
- USG - Ultrasonogram 1- Nil abnormality detected, 2- Fatty change in liver ,
3- Other
- A - Amsel's criteria 1- <3 No BV, 2- 3 and 4 BV
- N - Nugent's scoring system 1- Score 0-3, 2- Score 4-6, 3- Score 7-10
0d- 0 Day 1w -1 week 1m – 1 month 2m- 2 months 6m- 6 months