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# Microbiological Screening of three Different Contraceptive Schedules Commonly Employed for Birth Control in Egypt for Potential Pathogenicity on the Microflora of the Lower Female Genital Tract

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**Abstract**

Comparative screening of three different schedules of birth control for adverse effect favouring microbial infection of the lower female genital tract was conducted in equl-sized (N = 30) and age matching groups of married multipara adopting oral intake of contraceptive pills (Gr.II), long-acting injectable MPA (Gr. III) and I.U.D wearers (Gr.IV). The microbiological results in each of these three instances were expressed in terms of the percentage incidence frequencies of positive (infected) cervical, endocervical and vaginal smears assessed with reference to parallel well fitting (Gr. I) normal control multipalra of the non-contractive users category. The outcome of this investigation revealed the occurrence of the most serious deleterious effect among (Gr. IV) I.U.D wearers reflected by 10% frequency rate for positive actinomycetes infection among cervical-endocervical smears compared to notable absence of such infection in (Gr. I) normal controls. In the meantime, the frequency rates of positive vaginal smears were markedly enhanced by variable magnitudes and probability levels of statistical significance for almost all varieties of microflora. On the other extreme, long acting injectable MPA contraceptive in (Gr. IV) multipara proved superior on account of its freedom from predisposition to enhanced microbial infection with virtually all types of microorganisms with the notable exception of bacteroids (20%). The drawbacks ascribed to oral intake of contraceptive pills involved markedly increased susceptibility to vaginal infection by aerobic micrococci (80%), anaerobic diphtheroids (30%) and candida albicans (23.3%).

## Introduction

THE search for a virtually safe, reliably effective easily practicable and ethically acceptable reversible means of birth control and family planning has been one of the major pre-occupations of specialized governmental institutes and challenging socioeconomic and health problem draining the financial resources of the country ever since the introduction into clinical use of the first combined oestrogen-progestogen hormonal oral conraceptive pills more than 30 years ago [1].

The rationale of birth control medication by the combined oral contraceptive "COC" pills entails daily regular intake of a single pill each evening over 20 days period beginning on the 5 the cycle day allowing 7 days rest for substitution by pharmacologically innert dummy tablets before resumption of the subsequent course of contraceptive schedule. Indeed, experience with long term use of the combined oral contraceptive "COC" pills yielded extremely low failure rate of the oder of 0.1 per 100 women years [1]. However, the frequent occurrence of untoward side-effects and complications attending prolonged intake of combined oral contraceptive "COC" pills encompassing gross impairment of hepatic function, metabolic aberrations involving carbohydrates, lipids and proteins disordered blood coagulation with consequent thrombo-embolic manifestations, hypertensive cardiovascular disease with myocrdial infarction and fatal cerebral accidents accounting for high mortality rate of the order of 20 per 100.000 users per year [2] has been ascribed to the high contents of both oestrogen and progestogen components in the early formulation of "COC" pills [3]. Moreover, a number of published reports called attention to the possible link between the fairly high oestrogen component in the early formulation of "COC" pills and subsequent development of breast cancer among young women users [4,5] and also as a predisposing cause of cervical neoplasia. The multiplicity of these hazardous complications and to obviate the potential risk of malignancy prompted the device of new formulation of the COC pills entailing reduction in the oestrogen component to 30 μg ethinyl oestradiol E.O coupled with 75 μg of the novel progestogen "Gestodene" currently available under the proprietary name, femodene [3].

Of the two long acting injectable progestogens that have been deviced for birth control schedules namely (L) medroxy progesterone acetate MPA (Depoprovera) Given by i.m. injection of 150 μg dose every 12 weeks and (2) orethisterone acetate (Noristerot) administered i.m. every 8 weeks, only the former agent has been licenced and approved for clinical use as long acting injectable contraceptive since the early 1960's. Experience with the use of this preparation over the last 30 years proved its extreme efficacy and safety with respect to its freedom from untoward side-effects [9]. The only drawback being the

liability to irregular uterine bleeding [7,8,9]. By virtue of the absence of oestrogen component in MPA, which is likely to supress milk secretions, this long acting injectable progestogen is recommended for use by lactating women.

The Cu-T(R) 380 IUD which has been approved by FDA in 1973 gained widespread applicability and favourable preference.

The present research publication is primarily concerned with comparative screening of three different contraceptive schedules currently adopted for birth control practice for their adverse effects on the cytological integrity and the microflora harbouring the lower female genital tract. The three selected contraceptive schedules comprised (1) low oestrogen combined hormonal progestogen pills, (2) long acting injectable MPA and (3) Cu-T 380A coated IUD's.

Reliable assessment and precise evaluation of the effects of the three selected contraceptive schedules on microbial infections of the lower female genital tract obviously entails comprehensive knowledge of the nature of microflora harbouring the vagina of adult normal married control multipara not resorting to any form of birth control device. Detailed and classified information concerning this subject along with sources of references and documentation in the literature have been compiled in tables $(1)_A$ and $(1)_B$.

## Material and Methods

*Test Subjects :*

The presently reported clinical studies conducted on a total of 120 young adult fertile female multipara aged 25 < 35 years aimed at comparative assessment of the likelihood of three different contraceptive schedules conventionally adopted for birth control and family planing to evoke local noxious tissue damage and untoward adverse reactions in the lower female genital tract reflected by the incidence frequencies of histopathological lesions and coexisting alterations in the prevalence and/or pathogenicity of he microflora. The total number of female subjects was classified into four equal sized groups each comprising 30 multipara encompassing the following categories:

1. Gr I- Normal controls not resorting to any form of contraceptive regimens.

2. Gr II- multipara placed on specified courses of oral intake of combined low-oestrogen dose. Progestogen hormonal contraceptive formulation issued in the form of pills under the proprietary name femodene which has been introduced in clinical use as an oral contraceptive since 1987 [3]. This pill incorporates 30 µg eshinyl oestradiol "E.D." along with 7.5 µg gestodene as the active progestogen. Clinical experience with the use of this combined hormonal conraceptive formulation in U.K. revealed satisfactory efficacy in the supression of ovulation and inhibition of menstrual cycle periodicity coupled

with low incidence of untoward side effects [10].

3. Gr III multipara subjected to long acting injectable progestational hormonal contraceptive viz medroxy progesterone acetate "MPA" issued in the market under the proprietary name depoprovera (UPJOHN) dispensed in 150 μg dose contained in 1 ml vial for I.M. injection every 12 weeks [3]. Medroxy progesterone acetate "MPA" proved to be extremely effective for long term contraceptive use displaying failure rate of less than 1% [8].

At the same time "MPA" was shown to be safe with respect to the incidence of untoward side-effects, the only significant drawback being the liability to irregular bleeding episodes [9].

4. Gr IV- multipara wearing copper-containing CuT-380 intrauterine contraceptive device "IUA". Intrauterine contraceptive devices act via a multiplicity of mechanisms.

The female multipara complying with the previously specified criteria were chosen from those attending the birth control clinic, Cairo University Hospital seeking serial check-up of the adopted contraceptive schedules. Thorough medical examination was carried out on each of the participants in this study to ensure freedom from any systemic disease notably diabetes mellitus, arterial hypertension, hepatorenal dysfunction, and gross endocrinological disorders likely to invalidate the reliability of the results of explorative study.

*Procedure of procuring cytomicrobial smears from the lower female genital tract:*

The procedural steps and the equipment used for this purpose conformed to the descriptive account given by Pharr in 1963. The subjects were instructed not to perform cleansing vaginal douches and to avoid the use of intravaginal medication for 24 hours period before specimen collection which was carried out in the premenestrual phase of the cycle. The speculum introduced into the vagina for visualization of the cervix and allowing its necessibility to smear collection and swabbing may be mositened with few drops of sterile isotonic saline solution without resorting to the use of any lubricant.

Three types of smear specimens listed hereafter were conventionally obtained from each female subjects:

1. The entire portiovaginalis of the cervix was swabbed with a cottonwool tipped applicator stick introduced into the external os. The specimen thus obtained was smeared rapidly by rolling motion on the glass slide.

2. Endocervical smear was collected by aspiration or the cervical mucus plug and endocervical secretion by means of a pipette moistened with saline and subsequently thinly spread on glass slide and

immediately immersed in 95% Ethyl Alcohol for fixation [12].

3. Smear from pooled vaginal secretion in the posterior fornix was obtained by means of an applicator stick tipped with non-absorbent cottonwool which was then rapidly and evenly spread into a film on the glass slide. These three types of specimens viz. vaginal, cervical and endocervical were most conveniently smeared on separate partitions of a single specially designed slide designated "VCE" slide.

- Fixation and staining of gynaecological cyto-microbial smears.

- The cytomicrobial smears mounted on glass slides were fixed while still moist by immersion in 95% ehtyl alcohol for 20 minutes. Dryness of the smears before fixation should be avoided since it produces distortion of the structural organization and nuclear morphology of the cellular components besides evoking loss of the specificty of their staining reactions. Optimal dehydration of the smears also ensures their firm adhesion to the glass slide.

The fixed gynaecological smears were eventually subjected to a variety of staining solutions designed for differential diagnosis of the nature of microbacterial flora in accordance with the type and intensity of their straining reactions depending on the biological properties of the causative microorganisms.

- Gram stain - conferring purple colour on gram positive microorganisms in contrast to red colour on gram negative bacteria [13].

Kopeloff-Bermann modification of Gram stain excellent for staining vaginal smears.

Ziehle-Neelsen carbolfunchisin stain conferring red colour on this fast bacteria which is slow to develop.

Papanicolaou Stain; This is the most convenient stain adopted for the confirmatory detection of anaerobic filamentous actinomyces infection in cervical and endocervical smears.

*Method of statistical analysis of microbial screening results :*

The results of microbial screening of each of the three types of gynaecological smears viz. cervical, endocervical and pooled vaginal were most appropriately evaluated by transforming the absolute numbers of negative (sterile) and positive (infected) smears in (Gr. I) normal control multipara not using any contraceptive and in each of the three other test groups resoting to different birth control schedules. The percentage incidence frequencies of negative versus total positive smears in each of the four test groups were arranged in 2 x 2 (fourfolds) table and subjected to binomial chi-square test [14] to determine the probability levels of statistical significance.

## Results

Records of the microbial screening of cervical - endocervical and of vagina

smears from the individual subjects included in each of the four groups of multipara have been expressed as percentage incidence frequencies of positive (infected) smears for each type of micro-organisms among the 30 specimens procured from each of the four test groups. These frequency values have been collectively displayed in table (2) and (3) for cervical endocervical and vaginal smears respectively. (Gr. I) normal control multipara and (Gr III) married female subjects adopting long acting injectable MPA contraceptive schedule exhibited total freedom from anaerobic filamentous actinomycetes infection verified by 100% incidence frequency of negative (sterile) specimens. The low incidence of 3.3% for positive smears of mild score $(+)_1$, actinomycetes infection among (Gr. II) multipara oral contraceptive pills, proved by Chi-square test to be of minor and insignificant probability level $p > 0.05$ for the difference from the zero percentage. In contrast, (Gr. IV) married multipara displayed a conspicuously raised incidence frequency of the order of 10% for positive moderate score (+)2 infection by actinomycetes which proved by $x^2$-test to exceed the Zero% level in (Gr. I) normal controls and (Gr. III) under long acting injectable MPA by highly significant probability grade ($p < 0.005$) and to surpass the low incidence frequency of 3.3% in (Gr. II) married multipara adopting oral contraceptive pills by a merely significant probability grade ($p < 0.05$).

The compiled data in table (3) display the incidence frequency values of positive (infected) vaginal smears among the four groups of married multipara demonstrable for the major varieties of microflora and arranged from above downwards in decreasing order of frequency values. The micrococci attained peak incidence frequency of 60% for positive vaginal smears in (Gr. I) normal control control multipara, followed in oröder of decreasing frequencies B. Proteus (26.7%) Doderlein's lalctobacilli (20%), diphtheroids (16.7%), bacteroids (10%), candida albicans (10%) and reaching down a minimal nadir of 3.3% for trichomonas vaginalis infection.

Comparative appraisal of the percentage frequency values of positive vaginal smelars in Grl normal control multipara and each of the three other groups adopting different schedules of birth control revealed as shown in table (3) highly significant increase in the rate of infection by gram +ve aerobic cocci in GrII multipara under oral contraceptive pills (80%) rising to a peak increase by very highly significant magnitude attaining 86.7% in GrIV I.U.D. wearers. The percentage frequency rate of vaginal infection by Doderlein's lactobacilli was significantly enhanced up to 33.3% in GrIV female subjects. Compared to 20% in Grl normal control noncontraceptive.

## Discussion

Though the majority of published reports on the subject of this scientific docu-

Table (1): Microflora Harbouring the Lower Female Genital Tract and Incidence Frequency Rates of thier Isolation in Smears from Adult Normal Female Multipara. Microbcterial flora (Anaerobic and Aerobic and Bacilli)

| Scheme of classification of invading microorganisms and prominent morphological features | Percentage frequency rates of isolation of normal cervical, endocervical and vaginal smears. | Names of authors; sources of References and dates of literature Publication |
|---|---|---|
| 1) Aerobic gram positive micrococci (Diplococci, Staphylococci and Staphylococci Non Pathogenic | 90% (Micrococci)<br>35% (Streptoccoci) | Morris & Morris (1969); **[15]**<br>Barrlett et al (1978) **[16]**<br>Corbishley (1977) **[17]** |
| 2) Anaerobic Peptococci and peptostreptococci | 7.2% | Watt & Associates (1981) **[18]** |
| 3) Aerobic gram negative Bacilli Motile Bacillus Proteus Glucose but non lactose fermenter | 31% | Corbisley (1977) **[17]** |
| 4) Anaerobic Gram positive Bacilli viz Doderlein's bacilli (Lactobacilli) | 75% (In clumps of > 10 Microorganisms) | Taylor et al (1982); **[19]**<br>Croickshank Sharman (1984) |
| 5) Anaerobic Gram positive Rod Shaped Corynebacteria (Diphterosids) and Eubacteria | 15% - 30% (Diphteroids)<br>4% (Eubacteria) | Osborne et al (1979); **[20]**<br>Larsen Galask (1982); **[21]**<br>Taylor et al. (1982) **[19]** |

Table (2): Comparative Incidence Frequencies of Negative and Positive Cervical and Endocervical Smears among Age Matching groups of Married Multipara Adopting Three Different Contraceptive Schedules Assessed with Reference to Parallel Control Non Contraceptive Users.Equal Sized Group of Multipara, N = 30 each..

| Varities of test groups of mulitpara | Numbers of cervical and endocervical smears (expressed as Percentage incidence frequencies) conforming to specied categories hereblow. *Nature and grading scores of severity also indicated* Negative smears (sterile) | Positive smears (infected) | Chi-square test of statistical significance of differences between perentage frequency values arranged in 2x2 tabulation Calculated $x^2$; d.f. = 1 Probability level "Q" |
|---|---|---|---|
| Gr. I normal control (non contraceptive users) | 100% (30 30) | Zero % (- / 30) Nil $p > 0.05$ | I. control and Gr. II oral pills $x^2$ - 3.359 @, d.f. =1 $p > 0.5$, insignificant |
| Gr. II oral pills | 96.67% | 3.3% (1 / 30) *Actinomyectes* (+) $p < 0.005$** Mild score | Gr. III injectable MPA No difference Gr. IV local IUD $x^2$ = 1.5263**; d.f.= 1 $p < 0.005$; H. sign. |
| G. III injectable MPA (Long acting) | 100% (30 / 30) | Zero% (- / 30) Nil $p < 0.05$ | Gr III injectable MPA $x^2$ - 3.3559 @ d.f. =1 $p > 0.05$; insignificant Gr IV local IUD $x^2$ = 38630*, d.f.=1 $p < 0.05$; Sign. |
| Gr IV Cu Containing IUD | 90 % (29 / 30) | 10% (3 / 30) *Actinomyectes* (+) Mild score | *Bet Gr III injectable MPA and;* Gr IV local IUD $x^2$ = 10.5263**, d.f. = 1 $p < 0.005$; H. sign |

Table (3): Comparative Incidence Frequencies of Negative and Positive Cervical and Endocervical Smears among Age Matching groups of Married Multipara Adopting Three Different Contraceptive Schedules Assessed with Reference to Parallel Control Non Conntraceptive Users.Equal sized Group of Multipara, N = 30 each.

| Classified varities of vaginal microflora | Numbers of positive (infected) pooled vaginal smears among test groups of mulitpara *expressed as percentage incidences for types of specified microflora.* | | | |
|---|---|---|---|---|
| | Gr. I Control Non contraceptive users | Gr. II Oral Pills | Gr. III Injectable MPA | Gr. IV Local IUD |
| 1) Micrococci Aerobic; Geram +ve | 60% (18 / 30) | 80% (24 / 30) | 70% (21 / 30) | 86.7% (26 / 30) |
| 2) Bacilli Aerobic gram -ve B. proteus | 26.7% (8 / 30) | 3.33% (10 / 30) | 30% (9 / 30) | 40% (12 / 30) |
| 3) Doderlein's lactobacilli Anaerobic gram +ve | 20% (6 / 30) | 23.3% (7 / 30) | 26.7% (8 / 30) | 33.3% (10 / 30) |
| 4) Coryne bacteria (Diphtheroids) Anaerobic, gram+ve | 16.7% (5 / 30) | 30% (9 / 30) | 23.3% (7 / 30) | 46.7% (14 / 30) |
| 5) Fusobacteria and bacteroids | 10% (3 / 30) | 16.7% (5 / 30) | 2.0% (6 / 30) | 36% (11 / 30) |
| 6) Yeasts and fungi Candida (monilia) albicans | 10% (3 / 30) | 23.3% (7 / 30) | 3.3% (1 / 30) | 30% (9 / 30) |
| 7) Protozoal oganisms Trichomonalis | 3.3% (1 / 30) | 10% (3 / 30) | 6.7% (2 / 30) | 20% (6 / 30) |
| Percentage frequency values of total positive (infected) smears in each group | 20.95% (44 / 210) | 30.95% (65 / 210) | 25.71% (54 / 210) | 41.91% (88 / 210) |
| Chi-square test of statistical significance of differences between percentage frequency values of total psoitive (infected) smears in various test groups | $p > 0.10$@ | $p > 0.40$@ | $p > 0.20$@ | $p > 0.40$@ |
| | $p > 0.40$@ | | | $p < 0.005$** |

** Highly significant

@ Insignificant

mentation provided very useful information, no the pathogenicity of different contraceptive schedules on the microflora harbouring the lower female genital tract, yet they could not be considered as satisfactorily re-liable basis for drawing precise conclusions concerning their relative pathogenic hazards as they were not conducted on properly designed and adequately controlled observations amenable to conventional tests of statistical analysis.

In contrast, the present microbiological screening procedure was carried out on three virtually large sized groups (N = 30) of married fertile female multipara using three different methods of contraception along with parallel well matching normal control non-contraceptive users of comparable age groups, homogeneity in socio-economic and hygeinic standards and equality in frequency of parity. In the mean time, standardized procedure and aseptic precautions were undertaken in procuring the endocervical and cervical as well as vaginal smears and in detailed procedure steps of preparation, fixation and staining of smears.

Reference may be made at first to the extent of conformity and consistency as opposite the discordance and divergence between the relative percentage proportions of positive swabs in the normal control group of multipara and those reported in previous publications on normal vaginal microflora. The endocervical and cervical smears from normal non-contraceptive users controls revealed total absence of positive infection with branched filamentous actinomycetes. This finding which reflected absolute resistance of intact columnar epithelial living and glandular mucus secretory components of the cervix to infection by an invasive filamentous growth of actinomycetss is strikingly different from low incidence of the order of 5% for positive actinomycetes infection reported [22, 23].

The frequency distribution pattern of positive vaginal smears for infection by the diverse varieties of normal vaginal microflora displayed highest peak prevalence of 60% for aerobic gram +ve micrococci followed in decreasing order of positive swabs by profound drop to 26.7% for aerobic gram -ve bacilli eg. B. Proteus, and successively reaching down to 20% for anaerobic gram +ve Doderlein's lactobacilli, 16.7% for anaerobic diphtheroids, 10% for Bacteroids, 10% for Candida (Monilia) Albicans and yeasts and ultimately declining to a nadir minimum of 3.3% for Trichomonas vaginalis.

The highest incidence rate of 60% of positive infection of normal vaginal smears by aerobic gram +ve saprophytic non-pathogenic micrococci proved to be appreciably lower than the isolation rate of 90% among smears of normal vaginal microflora reported [15, 16]. Among the species of Aerobic gram -ve bacilli, infection by B. Proteus yielded 26.7% positive swabs

among normal controls, a percentage frequency rate which some authors [17] proved to be insignificantly lowered than 31% estimate given by the value value of 16.7% for positive frequency rate of vaginal infection by anaerobic diphtheroids in normal controls well coincided with the lower limit of positive swabs extending between 15%-30% in normal vaginal microflor repoted by others. [20, 21].

The fairly high estimate of 75% incidence frequency for positive infection by anaerobic gram +ve Doderlein's lactobacilli in normal vaginal microflora according to some publications [19, 24] proved to be 3.75 folds greater than the presently reported percentage rate of 20% for positive vaginal smears in normal controls. This marked discrepancy between the two sets of estimates might well be ascribed to racial differences between foreigners and Egyptians with consequent dissimilarity in prevailing neuroendocrine balance and in the defensive mechanism against bacterial infection mostly implicating immune system. Equality at a frequency rate of 10% for positive infection by anaerobic gram-ve bacteroid species of Gardnerella vaginalis strain was demonstrable for the present records of vaginal swabs from normal controls and published report by others [18,19]. The value of 10% incidence frequency of vaginal candidiasis among normal control multipara amounted to half the rate of 20.8% for positive infection by Candida albicans among normal vaginal microflora given [25]. The lowest incidence rate of 3.3% for positive infection by trichomonas vaginalis in swabs form normal controls showed perfect consistency and approximate equality to 3% incidence frequency for positive infection by this organism in normal vaginal microflora [26, 27].

The modulatory influence of the adopted contraceptive schedules on the prevalence pattern of positive vaginal smears from normal controls is most readily appreciated by critical survey and evaluation of the alterations evoked by each birth control regimen separately. The oral intake of combined low oestrogen progestogen hormonal contraceptive-pills induced de novo actinomycetes infection of the endocervix and cervix detectable as low frequency rate of 3.3% for positive swabs. However, this estimate yielded insignificant probability level at $p > 0.05$ from zero positive record denoting total absence of infection in cervical smears from normal controls. Moreover, oral contraceptive pills elicited marked enhancement of the incidence frequencies of positive vaginal smears for virtually all sorts of microflora reflected by variable magnitudes of conspicous increases in percentage frequencies of positive swabs attaining significant increment by almost two folds (30%) for infection by diphtheroids and rising by well over two folds to attain highly significant increase up to 23.3% for positive vaginal candidiasis infection. This estimate for positive vaginal

moniliasis among contraceptive pill users showed close proximity to corresponding incidence freuqency rates for vaginal infection by Candida albicans of the order of 20% - 25% [28, 29] rising up to 27% according to some data [30]. However, discordant estimates amounting to 13.5% for positive vaginal candidiasis in combined hormonal pills was given by Morris and Morris [15] dropping down further to 7% [31]. In addition, combined hormonal contraceptive pills evoked well pronounced increase in the incidence frequency of infected vaginal swabs by gram +ve aerobic micrococci attaining 80% there by exceeding the corresponding estimate of 60% in normal controls by highly significant magnitude. The potentiated enhancement of the frequency rates of positive infection by vaginal microflora seemed to be perfectly consistent and in good keeping with the well recognized ability of the oestrogen component of the combined hormonal pill to promote proliferative mitotic multiplication of the invading microorganisms. The finding of approximately three folds augmentation in the incidence frequency of positive infection by Trichomonas vaginalis to attain 10% in multipara under oral contraceptive pills could well be explained and perfectly fitted on the basis of revelation [31] demonstrating the possession by Trichomonas vaginalis of specific endogenous oestrogen receptors capable of selecltive uptake and binding of 17-*B* oestradiol component of the pill. The frequency rate of 10% for positive infection of vaginal smears by Trichomonas vaginalis under the modulatory influence of combined hormonal contraceptive pills displayed equality to 10% estimate for positive Trichomonas vaginalis infection given [32, 33, 34].

The use of long acting injectable was shown to afford complete protection against cervical actinomycetes infection and conspicuously reduced frequency rate of positive vaginal candidiasis from 10% in normal control to 3.3% presumably due to the chemical nature of injectable long-acting MPA being essentially a progesterone derivative and hence capable in conformity to statements by Catarell [35] evoking heavy glycogen deposition in the vaginal mucosa thus creating a protective barrier against invasive spread of fungal infection by Candida albicans.

Conversely the growth of most bacterial strains of vaginal microflora was favourably influenced by injectable MPA contraceptive as verified by conspicuous increases in the frequency rates of positive vaginal smears which were as a rule insignificant with the sole exception of Gardnerella vaginalis in which case the incidence of positive smears displayed two folds significant augmentation to attain 20% estimate.

The use of IUD's emerged as the most hazardous of the three adopted contraceptive schedules with respect to its demosntrable ability of promoting considerably

enhanced grades of infection of the lower female genital tract along with concomitant accentuation of the pathogenicity of most strains of vaginal microflora. Corroborative evidence for this statement may be found in the de novo induction of a fairly high incidence rate of the order of 10% for positive actinomycetes infection of the endocervix involving extensive invasion of the mucosal lining with branched filamentous mycelial colonziation with consequently high risk of spread to the uterine endometrium thereby predisposing to the development of pelvic inflammatory disease as one of the seriously grave complications. This statement showed perfect consistency and good keeping with the findings and views of a number of researchers in this field in so far that they reported fairly high percentage frequency rates for filamentous actinomycetes infection and colonziation within the endocervix and recommended the use of immunofluorescent technqiue for improved positive detection among smears from IUD's wearers **[36, 37, 38, 22]**.

The noxious damage evoked by the presence of IUD's on the integrity and protective efficiency of the startified squamous epithelial lining of the vagina among IUD's wearers presumably mediated via its action as an impacted foreign body in the uterine cavity leading to altered nature of the vaginal discharge was evidenced in the present study by well pronounced vriable magnitudes of percentable increases in frequency rates of positive infection by practically all species of vaginal microflora attaining highest estimates amounting in descending order to 86.7% for aerobic gram +ve micrococci, 46.7% for anaeroic gram +ve diphtheroids, 40% for aerobic gram - ve bacilli vis B. proteus, 36.7% for fusobacteroids, 33.3% for anaerobic gram +ve Doderlein's lactobacilli, 30.3% for vaginal candidiasis and 20% for Trichomonas vaginalis. However, incidence frequencies of positive vaginal candidiasis among IUD's wearers reported by Pareuijck et al. **[39]** 20% and by Fahmy and coauthors 16% **[40]** proved to be appreciably lower than the estimae of 30% recorded in the present study. The incidence frequency of vaginal infection by diphtheroids was significantly increased by almost two folds reaching up to 30% in GrII oral pills and rising up further by nearly three folds to a peak of 46.7% in GrIV adopting I. U. D. Bacteroid infection was encountered in 20% of vaginal smears in (Gr. III) receiving injectable MPA and displayed a conspicously higher peak frequency of 36.7% for positive smears in (Gr. IV) I..U.D wearers. Appreciably higher frequency values for positive candida infection were demonstrable in vaginal smears from (Gr. II) multipara under oral pills (23.3%) and in (Gr. IV) I..U.D wearers (30%). I.U.D's in (Gr. IV) multipara favoured vaginal infection by trichomonas as verified by fairly raised frequency rate (20%) of positive smears. Despite the statistically significant increases in percentage frequencies of positive vagi-

nal smears for infections by certain varieties of microflora beyond the corresponding control values in (Gr. I) encountered in (Gr. II) under pills for infections by aerobic micrococci (80%) diphtheroids (30%) and Candida albicans (23.3%) and demonstrable solely for bacteroids (20%) in (Gr. III) receiving injectable MPA, yet the overall total sum of positive vaginal smears for infections by the seven specified varieties of microflora displayed in table (3) though apparently higher in both groups (30.95% in Gr. II and 25.71% in Gr. III) proved to be statistically insignificant from the corresponding control value in GrI (20.95%) verified by probability levels of $p > 0.10$ and $p > 0.40$ respectively it was only (GrI. V) I.U.D wearers who displayed peak rise in overall total frequency of positive smears for vaginal infection by all test varieties of microflora attaining two folds incremental increase up to 41.91% ($p > 0.005$) beyond the corresponding control value of 20.95% in GrI normal multipara.

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