RESULTS

The resutls obtained lead to the following considerations:

1. All the immunoglobulin fractions increase during pregnancy, in primary foeto-placental insufficiency; the most significant increase was that of the IgG, which was much above the values found in normal pregnancies (and these values in their turn are greater than in non-pregnant subjects) (Fig. 1);
2. Increases were recorded even in IgM and IgA levels, though to a much lesser extent;
3. The increases seemed to be proportional to the nature of the foeto-placental dysfunction (the greater the degree of insufficiency, the higher the level of immunoglobulin);
4. The mean values reached by the IgG immunoglobulin were around 3000 mg%, by the IgA, 400 mg% and by the IgM, 300 mg%;
5. Drug therapy (with vasodilators, and with a view to stimulating tissue oxygenation: xanthenol nicotinate and taurine) brought about a diminution in values of the immunoglobulins, which tended to return to normal after 30 days of treatment (Fig. 2);
6. The immunoglobulin pattern remained unchanged even if irreversible placental lesions occurred.

SUMMARY

After drawing a distinction between primary and secondary foeto-placental insufficiency, and between their acute and chronic forms, the authors take into consideration the behaviour of the separate immunoglobulin pattern in the different forms of primary chronic insufficiency. Finally they report on variations after adequate therapy.

Attachment of M. hominis to human spermatozoa

by

F. BUSOLO*, G. BERTOLONI*, L. CONVENTI*, G. A. MELONI*

In 1973, the observation of T-mycoplasma (*Ureaplasma urealyticum*) cells adhering to spermatozoa of men with reproductive failure emphasized the importance of the relationship between these organisms and infertility ([1]). Epidemiological studies on this subject, carried out before and after these observation ([2,3,4,5,6,7,8]), have not yielded univocal results. While working on the presence of mycoplasmas in the ejaculates and cervical secretions of infertile couples, we obtained some data which seem to suggest for *M. hominis* the same kinds of features previously described for T-mycoplasmas.

MATERIALS AND METHODS

Ejaculates from 15 men, belonging to infertile couples were examined. The media used for culturing mycoplasma were the same as described by McCormack et al. ([9])

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Aliquots of 0,1 ml of sperm samples were inoculated into urea broth, arginine broth and on corresponding solid media. The positive cultures in liquid media (in which the pH had risen 0,5 units or more) were subcultured in solid media and studied for identification. The remainder of each sperm sample was washed three times in phosphate buffered saline. From the obtained pellets slides were prepared and: a) directly examined by interference phase-contrast microscopy, b) observed after staining with Giemsa solution, c) fixed with cold acetone for immunofluorescence. Antisera to *M. hominis* used for this method were prepared in white rabbits according to the immunization method described by Razin ([10]) and were heated at 56° for 30 min before use. Fluorescein cojugated anti rabbit IgG produced in goats (Hyland) was used for indirect immunofluorescence. One drop of anti-*M hominis* antiserum was placed on each smear, allowed in a moist chamber at 37° for 30 minutes, rinsed with PBS for 15 minutes and covered with fluorescent antibodies for 30 minutes at 37°. The smears were again rinsed both with PBS (for 15 minutes) and with distilled water. The air dried slides mounted with cover slip with PBS-glycerol (1 : 1) were finally examined with a Leitz Orthoplan fluorescence microscope. Smears only treated with fluorescein conjugated antisera were used as negative controls.

RESULTS AND DISCUSSION

The results of isolation of *M. hominis* and *U. urealyticum* from ejaculates are summarized in the following table.

Table. *Isolation of M. hominis and U. urealyticum from men belonging to infertile couples.*

U. urealyticum	M. hominis	Both	positive n	positive %N	negative	Total (N)
4	1	2	7	47	8	15

U. urealyticum was recovered from 4 specimens and *M. hominis* from 1 specimen, whereas association of these two species occurred in two ejaculates. Interference microscopy showed that in a specimen (the same from which successively pure cultures of *M. hominis* are obtained) about 50% of the spermatozoa had one or more swelling, which were constant in size and located on the neck region (Figs. 1 and 2).

The examination of Giemsa-stained slides corresponding to this case presented the same features and also showed the basophilic of these elements adhering to the sperm cell heads (Figs. 3 and 4).

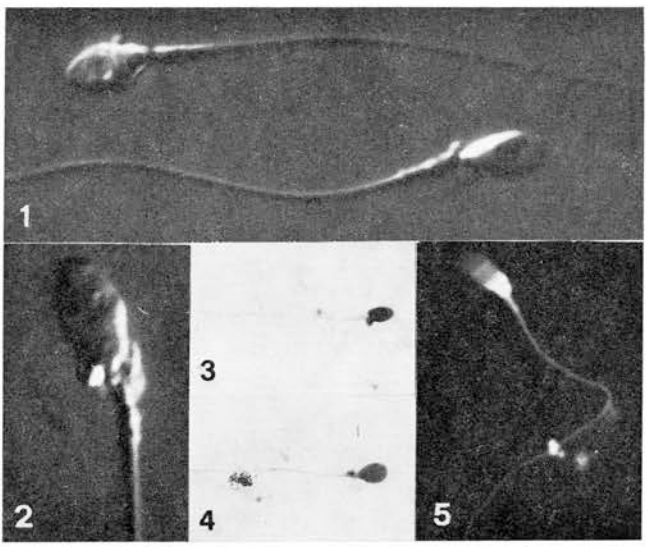
From the same ejaculate only *H. hominis* was isolated, as confirmed by growth -inhibition and metabolic inhibition tests.

Referring to the above mentioned data of Gnarpe and Friberg for T-strains, we thought that the formations observed under microscopy could be due to the presence of *M. hominis* cells.

This interpretation was confirmed by the positivity of indirect immunofluorescence tests carried out with anti-*M hominis* antiserum. In fact positively stained areas were seen in correspondence to the sperm cell necks and were pratically identical in size and localization to the formations previously observed by other methods (Fig. 5).

We feel that anomalous formations such as cytoplasmic residuals or other alterations of the spermatozoa neck can be excluded. This interpretation is basically supported by the positivity of immunofluorescence results.

Since epidemiological studies concerning the eventual relationship between *M. hominis* and infertility are lacking, perhaps our observations should be confined to the biological problem of adhesivity, peculiar to mycoplasma cells. These



FIGS. 1 (×2,000) and 2 (×2,500). Interference phase-contrast microscopy. - FIGS. 3 and 4 (×550). Giemsa-stained preparations. - FIG. 5 (×1,300): immunofluorescence.

properties of *Mollicutes* also express themselves both in morphogenesis and morphology of the colonies ([11,12]) and in attachment of mycoplasma cells to inert surfaces ([13,14]). It should be mentioned that erythrocytes ([17]), leukocytes ([15,16]), spermatozoa and other animal cells ([16,18,19,20,21,22]) adhere to mycoplasma colonies. Moreover, the fact that some mycoplasma agglutinate erythrocytes ([23]) and sperm cells ([20]) suggests the existence of specific mechanisms not sufficiently ascertained.

Nevertheless the adhesion of *M. pneumoniae*, *M. gallisepticum*, *M. pulmonis* and *M. agalactiae* to respiratory epithelial cells and to erythrocytes is considered to be an important event in the pathogenesis of diseases caused by these organisms ([23]).

SUMMARY

About 50% of the spermatozoa present in an ejaculate from a man belonging to an infertile couple (from which *M. hominis* was recovered) showed one or more small ovoidal formations adhering to the neck region.

These formations which were evident in interference microscopy, and after Giemsa staining, appeared positively stained in indirect immunofluorescence carried out with anti-*M. hominis* antiserum. These observations are discussed in comparison with the data obtained by other authors for *U. urealyticum*.

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Monotherapy with mepartricin versus combined amphotericin b plus tetracycline in mycotic and protozoal vaginitis

by

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INTRODUCTION

In the last years the etiologic pattern of infectious vaginitis has undergone deep transformations ([6]). In fact mycetes of the Candida genus (*C. albicans*) and protozoa such as *T. vaginalis*, are among the principal pathogenic agents encountered nowadays ([1]).

This evolution depends on several factors, previously unknown or scarcely represented, which favour infectious colonization of the vagina such as the use of hormonal contraceptives ([5,15]), of anti-inflammatory steroids ([14]) and broad spectrum chemobiotics ([4,12,13,14,16]).

Other well known factors are chronic endocrinopathies such as diabetes and also pregnancy ([7]). At vaginal level this problem is further complicated by some mechanisms of pathogenic interconversion recently identified. It appears that mycotic infection can coexist with protozoal infection or arise secondarily ([2]), especially if the latter was not adequately treated. In this regard we must point

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