Résumé

ON THE FOUR CASES OF PRIMARY BRONCHO-PULMONARY CANDIDOSIS

HITOSHI YAMATO, TAKASHI NAKANE, HISASHI HITOMI, TADASHI TERAO & HIROSHI SATO

Internal Clinic of the Saiseikai Hospital of Okayama

Four cases of primary broncho-pulmonary candidosis were observed in Saiseikai Hospital of Okayama. One case was generalised candidosis, and resulted in fatal state. Another three cases were broncho-pulmonary forms. Two cases of them were restored to good health, one got better. All were treated with enteric-coating trichomycin per os.

Many cases of severe form have been reported from various parts of the world, but intermediate and mild forms are comparatively rare. It is commonly believed that the administration of antibiotics is the principal cause of candidosis since the discovery of penicillin.

Some author in this country does not even believe the existence of mild and intermediate form of the primary candidosis, but these forms are not so rare as they thought. In general, the patients of mild or intermediate form may be diagnosed and treated as acute pneumonia, chronic bronchitis, bronchial asthma, chronic tuberculosis, pulmonary emphysem etc.

Even when the candidosis of severe form were discovered after the misuses of antibiotics, the real agent may be the candida group from the beginning. This point is discussed in detail. It is very difficult to recover from severe form, but it is not hopeless to cure intermediate or mild form by suitable treatment. In first place, the rapid and right diagnosis is necessary to make the exact treatment and not to misuse antibiotics, treating the patients complaining cough.

The roentogenograms of intermediate or mild bronchopulmonary candidosis show many fine mottled shadow (or snow-fall), especially in the lower parts of the lung with relatively clear apex. This X-ray figure is thought as the most important characteristic for diagnosing broncho-pulmonary candidosis. Two typical X-ray figures were obtained.

STUDIES ON THE GROWTH MECHANISM OF CANDIDA ALBICANS

HIDEO OIKI

Osaka Municipal Medical College, Momoyama Municipal Hospital, Pediatrics Clinic

(Director : Dr. TETSUJI KINOSHITA, M. D.)

1) The cells of *Candida albicans* when grown on glucose containing medium become extremely large compared to the control cells and stain deep brown when treated with iodine in potassium iodide solution while the control cells turn but slightly yellowish.

2) One milligram of *Candida albicans* consume 19.5 mg, 38.5 mg and 44.5 mg of glucose per cc in 3, 5 and 7 days of culture.

3) *Candida albicans* can utilize not only glucose but fructose, galactose, maltose and saccharose during growth. Lactose is also used to some extent.

From these results together with the results of my previous experiments, the explosive proliferation of *Candida albicans* within the intestinal canal during the oral administration of antibiotics may be ascribed to the diminution of the intestinal flora susceptible to the antibiotics, in consequence of which the growth inhibitive metabolic products of the intestinal flora decrease and the nitrogenic and energy sources, especially glucose which otherwise would be consumed by the flora accumulate within the intestinal canal, thereby bringing about a favorable condition for its growth.

ON THE ANTITUBERCULOUS ACTION OF SULPHATHIAZOLE (1)

MICHIO TSUKAMURA, TETSUZO KIMINO, MASASHI HASHIMOTO & RYOZABURO SUZUKI

The Obuso National Sanatorium, Obu, Chita, Aichi

(A) The *in vitro* antituberculous action of sulfathiazole against *Mycobacterium tuberculosis* var. *avium* (the Jucho strain).

A 5-day culture of the organism was tested against sulfathiazole by the dilution method using the SAUTON medium, and it was observed it was necessary for the organism to multiply a number of generations before the drug could inhibit growth. Its inhibition appeared, therefore, to be a so-colled partial inhibition, as the growth of the organism was never completely but only incompletely inhibited. By the inhibition the organism showed an appearance of a flocculous growth. The flocculous submerged growth was brought about as follows : The organism multiplyed a number of generations in early stage of the growth (for 48 to 72 hours) even in highest concentrations of the drug as well as in the medium containing no drug and thereafter its growth was inhibited. However, if the size of in-

oculum were very small, a complete inhibition could be also observed. Under conditions used by us the growth of the organism was inhibited in the concentrations of 1.6 to 6.3 mcg per ml of the drug after 10 day incubation. The minimal concentration for inhibition was influenced by the size of inoculum. The larger the size of inoculum, the larger the amount of the minimal concentration for inhibition.

The inhibition was completely abolished by the presence of only a very little amount of *p*-aminobenzoic acid.

A curve of emergence of resistance to the drug was followed by the GRAESSLE's method. The emergence of resistance was not significant. After 24 transfers every 10 days we could not obtain the resistance of populations over 200 mcg per ml.

(B) The *in vitro* antituberculous action of sulfathiazole against *Mycobacterium tuberculosis* var. *hominis* (the Frankfurt strain and the Aoyama-B strain).

The tuberculostatic action of the drug on the OKA-KATAKURA's medium (an egg-yolk medium) was tested in the range of 0 to 100 mcg per ml. 100 mcg of the 8-week culture of the organism in wet weigh was inoculated and it was observed for 12 weeks. A delay of growth was observed in concentrations between 1.57 and 50 mcg per ml of the drug. The delay of growth was especially significant in the range between 12.5 to 50 mcg per ml and the appearance of colonies was found 4 weeks later. This delay of growth seemed to result from a wide fluctuation of sensitivity of the organism to the drug. The growth of the organism was completely inhibited by the presence of 100 mcg per ml of the drug.

The inhibitory action of the drug was also abolished by the presence of 1 mcg per ml of *p*-aminobenzoic acid.

SOME OBSERVATIONS OF THE IN VITRO TUBERCULOSTATIC ACTION OF SULFONAMIDES

MICHIO TSUKAMURA

The Obuso National Sanatorium, Obu, Aichi

The *in vitro* tuberculostatic action of sulfonamides on *Mycobacterium tuberculosis* var. *hominis* (2 strains) and *Mycobacterium avium* (3 strains) was observed. Sulfanilamide (SA), 6-sulfanilamide-2, 4-dimethylpyrimidine (SD), sulfaguanidine (SG), sulfameradine (SR), sulfathiazole (ST), and 3, 4-dimethyl-5-sulfanilamide-isoxazole (SZ) were tested in the present report. Among these sulfonamides ST and SZ were highly active, SR and SD considerably active, and SG and SA only slightly active. ST=SZ>SR≧SD>SG≧SA. The succession of intesity of the tuberculostatic action was constant and independent of types and strains of the organisms tested and kinds of the media tested.

The growth inhibitory action of ST was considerably antagonized by addition of *p*-aminobezoic acid, *p*-aminobenzoylglutamic acid, and folic acid, and slightly by methionine, formate, purines. The mechanism of action of sulfonamides on mycobacteria was also, therefore, suggested as being similar to that of other sulfonamides-sensitive bacteria. The antagonistic ratio of ST/PABA was very large (1,000). The *in vitro* inactivity of ST seems to be interpreted by this finding.

p-Nitrobenzoic acid, *p*-nitrophenol and nitrobenzene were also considerably antagonistic to ST.

STUDIES ON THE COMBINED ACTION OF THE ANTIMICROBIAL SUBSTANCES

On the Phenomenon of the Repression of the Development of the Resistance "in Appearance"

YUTAKA KOBAYASHI

Department of Pediatrics, Medical Faculty, Kyoto University
(Director Prof. H. NAGAI)

Diviision of Childhood Tuberculosis, Tuberculosis Research Institute, Kyoto University
(Chief Prof. I. SAGAWA)

In the previous reports, when the author studied on the repression of the development of the resistance to streptomycin (SM) by guanofuracin (GF), the phenomenon, named the repression of the development of the resistance "in appearance", was observed remarkably. As this phenomenon was very interesting, the author studied on the mechanism of this phenomenon, and the results were summarized as follows.

The used strain was *Sh. flexneri* var. X, and this wild population did not include the resistant mutant more than 1.0 mcg/cc.

This phenomenon depended of the fact that highly SM-resistant population developed, even when the wild population was subcultured in the media containing the low concentration of SM(0.4~1.6 mcg/cc) alone. This subculture was carried out in that manner which the order of SM-concentration and that increase was as completely same as that of SM in the co-existed series. That is, the next explanation was possible : owing to the co-existence of GF, the highly developed SM-resistance was not manifested in the co-existed media at the same rate of both antimicrobials, and therefore, the population, from the co-existed media, manifested the high resistance, in the media containing SM alone. Thus, it was considered that this phenomenon was entirely differed from the synerigsm.

But the development of SM-resistance, subcultured in the coexisted media, was slightly delayed, compared with the subcultures in the media containing

the low concentration of SM alone. On the other hand, when SM combined with chloramphenicol (CM), this phenomenon was not observed.

So, the next was concluded, that the co-existence of GF interfered a little with the development of SM-resistance of this kind, and the co-existence of CM repressed such development of the resistance almost completely.

From the difference of the results between the combination of SM with GF and that of SM with CM, it might be suggested that the combination did not only diminish the mutation rate which occurred the dual resistant mutant, but also acted upon the process of the mutation itself. In other words, the co-existed GF or CM was not the more mechanical selector regarding the repression of the development of SM-resistance.

On basis of the above mentioned results, the mechanism of the development of the resistance of the bacteria to antimicrobials was discussed.

THE ANTAGONISTIC EFFECT OF PARA-AMINOSALICYLIC ACID ON THE TUBERCULOSTATIC ACTION OF SULFATHIAZOLE AGAINST MYCOBACTERIUM AVIUM

MICHIO TSUKAMURA

The Obuso National Sanatorium, Obu, Chita, Aichi

The tubrculostatic action of sulfathiazole against *Mycobacterium avium*, strain Jucho, was tested by the serial dilution method, in which the medium used was the SAUTON medium (2 ml in each tube) and 5.5×10^5 viable cells were inoculated to each tube, and the effect of addition of para-aminosalicylic acid (PAS) was observed. Under these conditions the maximal concentration of sulfathiazole in which the organism grew as well as the contrast was 1.25 mcg per ml in the absence of PAS after 8 days incubation at 37°C, whereas this was considerably increased by addition of PAS (2.5, 5.0 and 40.0 mcg per ml in the presence of PAS in concentrations of 1, 10 and 100 mcg per ml, respectively).

It appeared that the antagonistic effect of PAS on the tuberculostatic action of sulfathiazole was much significant in concentrations of 100 mcg per ml of the drug, comparing with that in concentrations of the drug smaller than 10 mcg per ml and the antagonistic effect of PAS was 0.01 times smaller than that of PABA. The contamination of PABA in the PAS-preparation used was paper-chromatographically excluded. PABA could not been found out by paper chromatography,* although PAS was added to the culture of the organism. The minimal inhibitory concentration of PAS against the test strain was 0.5 per cent.

Considering the fact that PABA was substituted by PAS in a PABA-requiring mutant of *E. coli* (TOBIE, W. C. & JONES M. J. J. Bact., 57 : 573, 1949), it seems to be very probable that the antagonistic effect of PAS on the tuberculostatic action of sulfathiazole on *Mycobaterium avium* is due to the possibility that PAS can be utilized in a manner similar to that in which PABA is utilized.

In summary, PAS has an antagonictic effect on the tuberculostatic action of sulfathiazole against *Mycobacterium avium*.

* (N-Butanol saturated with M/15 KH_2PO_4, ascending method, and Toyo filter paper No. 50 were used. Rf of PAS 0,087-0.097. Rf of PABA 0.39-0.41.)

STUDIES ON THE APPEARANCE OF DRUG RESISTANT STRAINS OF TUBERCLE BACILLI IN EXPERIMENTAL PULMONARY TUBERCULOSIS OF RABBITS

OSAMU SUZUKI

The Third Clinic of Internal Medicine, Osaka University School of Medicine

(Director : Prof. IMASATO DONOMAE)

Rabbits weighing about 2.5 kg, previously sensitized with 5 mg of the virulent human strain of tubercle bacilli (strain TADA), were inoculated with 25 mg of the same strain suspended in 10~15 % gelatin solution, which was injected into the right lung directly through the 4th intercostal space of the side chest. Thus experimental tuberculous were produced in their lungs. These rabbits were divided into 3 groups, and 50 mg of streptomycin (SM) per day, 10 mg of isonicotinic acid hydrazide (INAH) per day, and 500 mg of para-aminosalicylic acid (PAS) per day were consecutively given intramuscularly to each of them respectively. Each group had the untreated control of the same number.

The results thus obtained were as follows .

1) The organs of the rabbits which were given SM for 60 consecutive days and those of the rabbits which were given INAH for 55 consecutive days produced a great number of colonies on the OGAWA's medium containig 10 mcg of SM per cc and on the same medium containing 10 mcg of INAH per cc respectively, and made a significant contrast with the controls. This phenomenon was more remarkably noted concerning the pulmonary cavities compared with the other parts of the lung and other organs.

2) The above-mentioned fact was more evidently observed when the rabbits were treated with SM for 80 days or with INAH for 90 days. As for the former, some colonies were observed to grow

even on the medium containing 100 mcg of SM per cc.

3) As for PAS, any colony could not be cultivated from the organs of the rabbits treated with PAS for 60 and 90 days on the medium containing 1 mcg of PAS per cc.

ON THE TRICHOMYCIN TREATMENT OF RABBITS EXPERIMENTALLY INFECTED WITH NICHOLS STRAIN OF TREPONEMA PALLIDUM

SEIGO HOSOYA, MOMOE SOEDA, SUSUMU IMAMURA & KAZUO OKADA

The 1 st Department of Bacteriology of the Institute for Infectious Diseases, Tokyo University

By the subcutaneous injection of 1~10 mg of trichomycin daily to rabbits with experimental syphilis, treponema in the infected testicle disappeared completely, the swelling and induration of the lesion recovering to the normal state.

Serologic reactions of syphilis also proved to become negative.

ON THE TREATMENT OF EXPERIMENTAL SYPHILIS IN RABBITS BY ORAL ADMIMISTRATION OF TRICHOMYCIN

KAZUO OKADA

The 1 st Department of Bacteriology of the Institute for Infectious Diseases (Director Prof. SEIGO HOSOYA)

1) *In vitro* activity of trichomycin against *Treponema pallidum* (NICHOLS strain) proved to be far stronger than that of crystalline potassium salt of penicillin G.

2) Trichomycin inhibited the growth of obligately anaerobic REITER strain of *Treponema* in a concentration of 0.047 mcg/ml. Penicillin inhibits the growth of the same organism in a concentration of 0.19 mcg/ml. On the contrary, oxytetracycline, chlortetracylcine, chloramphenicol, erythromycin, leucomycin and streptomycin were ineffective even in a concentration of 50 mcg/ml.

3) When enteric coated oral tablets of trichomycin (10,000 *Candida* units 1 tablet) were orally administered once daily (the daily dosage : 5~10 tablets) to more than 10 rabbits with exprimental syphilis for 10~20 days, the swelling and induration of the testicle infected with *Treponema* disappeared completely and the organisms could could not be detected from the lesion. Serological tests containing WASSERMANN reaction proved to become negative before long.

EFFECT OF ANTITUBERCULOUS AGENTS ON THE VITAMIN C

Metabolism in Patients with Pulmonary Tuberculosis. Report III. Effect of Tibione

MASATO IKEUCHI & HIROYUKI MIZUNUMA

Dept. of Internal Medicine, Tokyo Jikeikai School of Medicine

Experiments were made on the metabolism of vitamin C (VC) in rabbits and in patients with pulmonary tuberculosis administered with tibione (TB 1). After oral administration of 100 mg of tibione daily to rabbits four weeks, the degree of saturation of total VC in urine was measured by adminstering intravenously 80 mg of l-ascorbic acid (l-AA) daily. The rabbits administered with TB 1 showed an extension of the saturation degree twice as the normal one.

A long-term administration of TB 1 to patients with pulmonary tuberculosis showed an extension of the saturation degree of VC two to three times as the normal ones.