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PROLONGED THERAPY FOR PARACOCCIDIOIDOMYCOSIS: APPROACHES, COMPLICATIONS, AND RISKS[6]

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Paracoccidioidomycosis has a long incubation period and is hard to cure. This article discusses various types of available drug therapy and indicates general guidelines for treating the disease.

The successful management and treatment of the paracoccidioidomycosis patient depends, more than anything else, on the physician's knowledge of and personal experience with the disease.

The experience acquired to date in the endemic areas of this mycosis tends to support the theory that *Paracoccidioides brasiliensis* exists freely in nature, probably in soil, and that its portal of entry is the respiratory tract, with infection taking place in the lungs. As with coccidioidomycosis and other systemic fungus diseases with similar patterns, the primoinfection is usually asymptomatic and can only be diagnosed by skin testing.

For background on the development of the foregoing postulates, as well as on the classification of the clinical forms of the disease, we refer to our 1947 and 1964 publications on these subjects. In the first of these (*43*), we called attention to the long period of incubation observed on occasion with this mycosis—as much as 40 years or more—according to information provided in the patients' anamneses. Time-spans of this length between an asymptomatic period of residence in an endemic area, when the primoinfection was very probably acquired, and the appearance of the first clinical symptoms in a nonendemic area, have been recorded more than once. Further
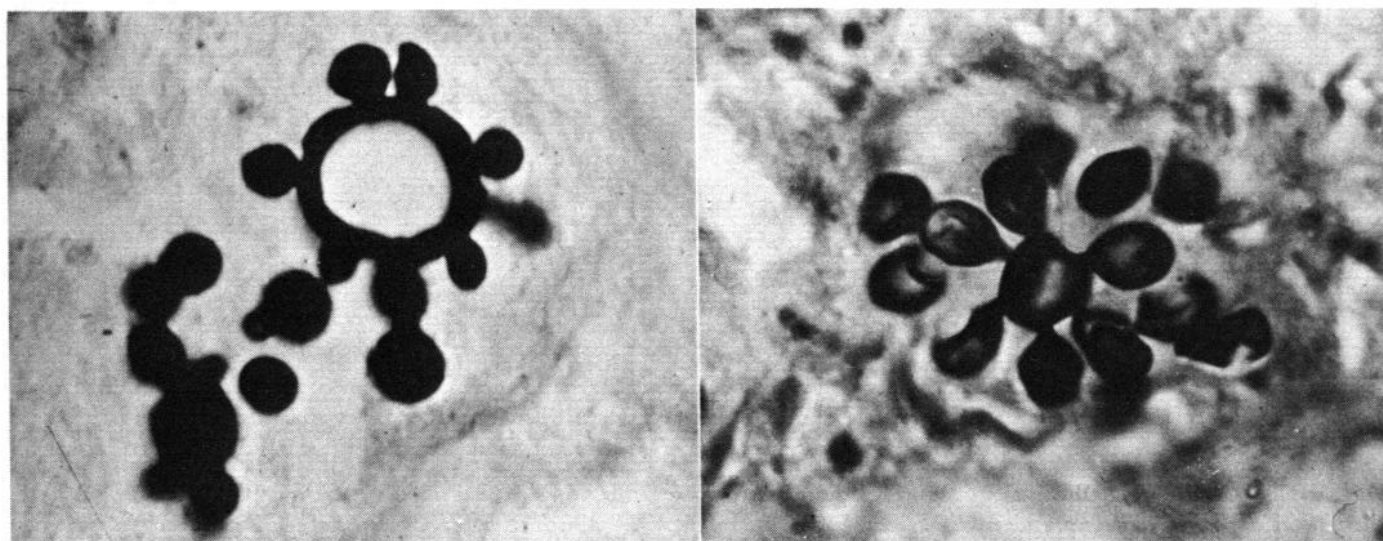
Two examples of the multiple-budding form of *Paracoccidioides brasiliensis*.

evidence of a subclinical infection was advanced by Pinto Lima in 1952, and corroborated by Machado Filho and Miranda in 1960, with the finding of *P. brasiliensis* in autopsied and biopsied lung material from patients clinically and radiologically free of lesions.

These observations already tell us that paracoccidioidomycosis is not comparable to coccidioidomycosis in other aspects of its pathogenesis. It cannot be said, unfortunately, that the asymptomatic primoinfection is benign and heals spontaneously, leaving sure protection against future disease. Quite the contrary: the parasite continues to be potentially active and threatening for many years. Moreover, even when the symptomatic forms are treated successfully with up-to-date drugs and cured clinically, radiologically, and serologically, the fungus can remain alive in some focus not accessible to exploratory biopsy and become active again when the biological equilibrium— including especially the immunoallergic state— of the human host is disturbed.

Other factors that complicate the management of paracoccidioidomycosis are the general circumstances of the patient himself. He comes to treatment with several strikes already against him. He is likely to be a male agricultural worker in his fourth to sixth decade, bowed down by hard work over the years, insufficient rest, undernutrition, alcoholism, illiteracy, lack of perspectives, and socioeconomic and hy-

gienic disadvantages. Worst of all, as far as hope for a successful cure is concerned, he appears at the clinic months or years after the disease has begun its course, either because he was negligent and did not seek consultation earlier or because no one had succeeded in identifying the etiologic agent. Thus, the host and his circumstances (surrounding climate, nutrition, hygiene, and socioeconomic situation) are of outstanding importance among the factors that enter into achieving the necessary favorable biological balance within the host to halt dissemination of the parasite.

In the present discussion of therapeutic norms, we will use the classification of clinical forms of paracoccidioidomycosis first published by us in 1965 (*38*), as follows: (1) asymptomatic or subclinical infection; (2) acute primary pulmonary infection leading subsequently to acute or chronic disseminated disease; (3) the chronic pulmonary form; (4) the acute disseminated form, corresponding to the septicopyemic form described by Pereyra and Vianna in 1918; (5) the chronic disseminated form most frequently observed with secondary lesions of the skin, mucous and semimucous membranes, epididymis, ocular globes, central nervous system, or internal organs; (6) forms occurring in association with pulmonary tuberculosis (12 per cent of all cases), leishmaniasis, other mycoses, etc.

The toxic signs are marked in the acute

disseminated disease and can include prostration, anorexia, anemia, jaundice, asthenia, weight loss, and sporadic fever. In the subacute or chronic forms the signs are reminiscent of those seen in tuberculosis: progressive weight loss, asthenia, anorexia, and sometimes fever and nocturnal sweating. Malnutrition occasioned by oropharyngeal lesions is often largely responsible for the anemia and loss of weight. Fatigue may result from pain and salivation produced by the mucocutaneous lesions or from a secondary bacterial infection, while difficulties in breathing can be caused by the presence of mediastinal adenopathies or of lesions in the bronchial tree.

P. brasiliensis lives and reproduces itself in the intercellular spaces, being observed within giant cells only in the granulomatous processes. Unless there are osseous lesions, it is not found in bone marrow. The exudative and proliferative phenomena coexist, the latter having a somewhat tuberculoid structure and a giant cell reaction similar to that which occurs with a foreign body. When the exudative reaction is predominant, abscesses are formed. Histopathology may reveal the presence of *P. brasiliensis* in its rapidly multiple-budding form, along with numerous small forms sometimes resembling bacilli. Areas of necrosis or necrobiosis are also seen. Fibrosis, either more or less intense, usually indicates that the disease is in its final stage, with residual calcification occurring only exceptionally.

Management and Treatment

Any patient found to have paracoccidioidomycosis should be hospitalized under the care of a physician fully familiar with the disease. The first step should be to obtain from the patient a precise and complete anamnesis, including a description of his living conditions, the climate(s) in which he has lived, his occupation, history of alcoholic consumption, habits of hygiene, and socioeconomic background, as well as a detailed record of the clinical evolution of the disease. It is most important that the physician be able to fully

evaluate the complex relationship between the individual patient and his environment.

In the hospital, all the clinical, radiological, mycological, histopathological, and serological examinations that may provide any information on the extent of the disease and on the host's immunoallergic state and overall defensive equilibrium should be performed. If there are any active superficial lesions, it is especially important that they be biopsied.

Inasmuch as the patient will probably have lived in a rural area for the greater part of his life, a systematic search should be made for concurrent infection with *Trypanosoma cruzi* or associated parasitic diseases such as schistosomiasis, uncinariasis, ascariasis, or other intestinal helminthiasis. Tuberculosis should be considered in patients with pulmonary lesions, since the two infections occur simultaneously in approximately 12 per cent of all cases. Naturally, the presence of such chronic conditions as diabetes, hypertension, tumors, etc., will make it necessary to prepare a special program.

Any plan of patient management should provide for bed rest, proper diet with emphasis on protein intake, supplemental vitamins, and whole blood transfusions (250 to 300 ml) two or three times a week, in order to help create favorable conditions for immunological resistance.

The chemotherapeutic drugs and antibiotics currently in use are fungistatic *in vivo*. Ultimate control of the infection, however, depends on the defensive capacity of the host, particularly with regard to formation of a productive granuloma which cuts off the parasite's ability to reproduce, curtails its dissemination, and tends to destroy it.

Signs of a severe case are: a generally poor state of health, symptoms of the acute disseminated form, meningeal lesions, multiple nodular or exudative lesions in the lungs, lymphatic involvement, a negative paracoccidioidin skin-test reaction accompanied by increasingly high serum titers, and histopathologic features showing predominance of the exudative reaction with presence of *P. bra-*

siliensis in its multiple-budding form together with numerous small forms—indicating its unhindered proliferation and readiness to disseminate.

Chemotherapy with Sulfonamides

In 1940 Ribeiro *(46)* demonstrated the effectiveness of sulfadiazine in the treatment of paracoccidioidomycosis. Later, Padilha-Gonçalves *(40, 41)* used sulfamethoxypyridine and Nohemi *et al. (39)* used sulfisoxazole.

During the period 1964 to 1967, Ferreira Lopes *et al. (19)*, Hutzler *et al. (26)*, Barbosa and Daher *(6)*, and Rubinstein and R. Negroni *(48)*, among others, reported on their results with a new sulfonamide derived from sulfamethoxypyridazine,4-sulfanilamido-5,6-dimethoxypyridine, synthesized by Roche Laboratories (Ro 4-4393, "Fanasil"). The advantage of this new product is that it can be administered in a single dose, either orally, intramuscularly, or intravenously, just once a week. For an adult weighing 60 kg or more the dosage is 1.5 to 2 g. In the mouse the median lethal dose (LD_{50}) is 5 g/kg, while there is a complete absence of toxicity at 0.25 g/kg. The drug has an average life in human plasma of from 100 to 200 hours, and it is excreted from the system in acetylated form (60 per cent), as intact sulfamide (30 per cent), and in glucoronic form (10 per cent).

Padilha-Gonçalves *(41)* has observed that sulfanilamide and the sulfones are not active in the treatment of paracoccidioidomycosis.

The initial optimism over sulfonamide therapy began to decline over the years as relapses and resistance to treatment were observed. Ferreira Lopes and Armond *(17)* recorded resistance in 9.8 per cent of the cases in a series of 122 patients; Machado Filho and Miranda *(32)* had a rate of 4.3 per cent in 394 cases; and Restrepo *et al. (44)* observed six cases among 16 patients treated with sulfonamides alone, plus four in a group of 22 who received amphotericin B and sulfonamides in combination.

Sampaio *(49)*, reporting the results of sulfonamide therapy in 338 cases studied over the period 1948 to 1958 at the Hospital das Clínicas in São Paulo, Brazil, saw that 69.5 per cent of the patients improved or were actually clinically cured, 8.9 per cent got worse, and 21.6 per cent died. Of 109 cases on which it was possible to gather data over a 10-year span, 46 patients continued satisfactorily, 19 got worse, and 44 died. Twenty of the deaths took place during the first year after discharge, nine between one and three years, five between three and five years, seven between five and seven years, and three between eight and nine years.

The initial results with Fanasil were likewise not very impressive. Ferreira Lopes and co-workers *(19)* treated 14 patients with this drug and observed good results in nine, some improvement in four, and failure in one. In a series of eight patients, Hutzler *et al., (26)* had the following results: very good in one, good in one, mediocre in four, and poor in two. Barbosa and Daher *(6)* reported the following results in nine patients: good in six, mediocre in two, and poor in one. In most instances the clinical observations were supported by radiological and serological findings, and sometimes even by results from biopsies of cicatrized lesions or lymph nodes.

In studies of resistance to sulfonamides, Ferreira Lopes and Armond *(17)* observed a beneficial effect when 2,4-diamino-5-(3,4,5-trimethoxybenzyl)-pyrimidine (Ro-6846, "Trimethoprim") was used in combination with Fanasil, which seems to potentiate the sulfa drug. Trimethoprim alone has no therapeutic effect in the treatment of paracoccidioidomycosis. With the two drugs in combination, however, the authors were able to successfully treat three patients whose infections were resistant to sulfonamides. Equally favorable was R. Negroni's experience in two cases treated with Bactrim. This latter drug, synthesized by Roche Laboratories, comes in scored tablets for adults, each tablet containing 80 mg of Trimethoprim and 400 mg of sulfamethoxazole; it is administered in an average dose of two tablets twice a day, followed by a maintenance

dose of one twice a day. There are also pediatric tablets, which contain 20 mg of Trimethoprim and 100 mg of sulfamethoxazole. A 5-ml teaspoonful (tsp) of the pediatric syrup contains 40 mg and 200 mg, respectively. The recommended dosage for children six weeks to five months old is a half tsp of the syrup twice a day; for those six months to five years, 1 tsp twice a day; and for those between six and twelve years, 2 tsp a day. If the tablets are used, two should be given twice a day to children between two and five years of age, and four twice a day to those between six and twelve.

The sulfonamides have low toxicity and are excellently tolerated. In the case of 2-hydroxystilbamidine, Lacaz *et al.* (*30*) have reported a minimum inhibitory concentration (MIC) *in vitro* for *P. brasiliensis* of from 2.5 to 10 mg per 100 ml of culture medium. This medication appears to be less toxic than amphotericin B. It is administered in daily doses of 225 mg in a solution of 5 per cent glucose in water, using a graduated dropper. A total of 8 g is sufficient to effect a cure. Most of the drug is deposited in the liver and kidneys, only a small quantity being excreted. During the treatment, both the drug solution and the patient himself must be kept out of the sunlight. Despite the immediate and effective results obtained in a case treated by Miranda and co-workers in 1956, the drug has not been extensively used with paracoccidioidomycosis, and Padilha-Gonçalves (*40, 41*) is of the opinion that it does not offer any special advantages.

The Bayer Company has synthesized a product, biphenyl-(2-chlorophenyl)-1-iminazolylmethane (Bay b 5097), with an MIC *in vitro* for pathogenic fungi, including *P. brasiliensis,* of from 4 to 8 μg/ml. A dosage of 100 mg/kg of body weight gives a concentration in serum of 10 to 20 μg/ml. The average recommended dosage for an adult is 60 to 100 mg a day, given in two or three doses and taken orally in capsule form. R. Negroni used this drug in two cases of paracoccidioidomycosis but had to abandon it because the patients were clearly getting worse.

Treatment with Antifungal Antibiotics

Amphotericin B

This is the drug most widely used in the treatment of paracoccidioidomycosis and any of the other systemic fungus diseases. The MIC *in vitro* observed by Lacaz *et al.* (*31*) for *P. brasiliensis* is from 0.06 to 0.243 μg/ml, depending on the strain. It is essentially fungistatic, but in doses four to ten times greater than the MIC it is also fungicidal. Drouhet (*15*) and other researchers have observed that it stimulates endogenous respiration of the fungi, which leads to rapid consumption of hydrocarbon reserves; it also acts on the patient's metabolism, blocks the synthesis of nucleic acid and proteins, and alters cellular permeability when it settles in the sterols, resulting in the flight of the K-ion and other essential metabolites. In the mouse, this last-mentioned property is considered responsible for lytic action on the red corpuscles.

When administered intravenously, amphotericin B can be nephrotoxic, producing hyposthenuria, uremia, and reduction in the clearance of inulin, para-aminohippuric acid, and creatinine. According to Winn (*57*), the best guide as to its tolerance is the clearance rate per minute observed in the serum of a patient under treatment. Calcium deposits were seen in the renal tubules when the total dosages exceeded 7 g. Anemia (diminution of both hemoglobin and red blood corpuscles) and hypocalcemia were other toxic manifestations.

With intravenous administration, the drug appears to be taken up rapidly by certain tissues and then slowly released. It attaches itself to β-lipoprotein in the serum and tends to decrease its extravascular diffusion. Concentrations in the parotid saliva and the spinal fluid are very low, tending to be higher in the latter instance when proteins are elevated. Battock *et al.* (*7*), using the spectrophotometer to measure the growth curves, reported average concentrations of 1.21, 0.62, and 0.32 μg/ml at 1, 18, and 42 hours, respectively, after infusion of 1.2 mg/kg of amphotericin B, Squibb strain 1647.

Administered orally in experimental animals, the drug lacks toxicity, even in a total dosage as high as 8 g/kg, but the concentrations recorded in plasma are minimal, ranging from 0.03 to 0.08 μg/ml.

Workers in this field essentially follow two different methods for determining the dosage of amphotericin B to be used in the treatment of the systemic mycoses. The first is to work up to a daily dosage that produces a peak level in the serum corresponding to twice the MIC *in vitro* for the causal agent. Once this level has been reached and is tolerated, it is then maintained for 10 weeks. In the case of paracoccidioidomycosis, when the dosage gets as high as 1.2 mg/kg, it is considered advisable to fall back to one intravenous injection every two days. No more than 7 g is recommended for the total cure. Brummer *et al.* (*10*) have suggested starting with 1 mg and increasing the dosage daily by 5 to 10 mg in order to avoid any hypersensitive reaction. The other approach consists in working up to the maximum level the patient can tolerate and maintaining it with daily doses of the antibiotic. We prefer to use the first of these methods.

For *intrathecal* administration, an aqueous solution of 0.25 mg/ml is used, starting with a dosage of 0.1 ml of the solution diluted in 2 to 3 ml of spinal fluid. The injections are repeated two to three times a week, increasing the dosage progressively until the level of 0.5 mg—that is, 2 ml of the mother solution—is reached. The cisternal approach is tolerated best, even though it invariably gives the patient a headache.

With administration by *aerosol,* the drug is given in concentrations of 5 mg/ml every six hours.

When circumstances indicate the need to give the drug by *irrigation,* an aqueous solution of 1 mg/ml is administered once every four days, starting with 5 ml of solution and going as high as 25 ml. Sometimes 5 to 20 mg of heparin are added to prevent phlebitis, and methylmelubrine, antihistamines, antiemetics, and corticosteroids may also be included.

Frequently the immediate results achieved with amphotericin B are spectacular. However, although resistance has not been recorded, relapses do occur. In the series of 61 cases treated by Sampaio (*49*), 32 of which were resistant to sulfa drugs, good results were observed in all the cases except those with pulmonary and/or lymphatic involvement. In seven cases of the cutaneous/lymphatic form, observed for a period of 12 to 24 months, he recorded three clinical cures and four relapses. The total doses of amphotericin B ranged from 2.05 to 5.4 mg. Of four lymphatic cases, three failed to respond after receiving 3.3 mg of the drug; and 8 of 11 cases with cutaneous/lymphatic/pulmonary involvement relapsed.

The simultaneous administration of amphotericin B and sulfonamides does not appear to enhance the action of either. However, many authors recommend using the one followed by the other. Rubinstein and R. Negroni (*48*) treated 10 cases with amphotericin B and Fanasil with very good immediate results in nine cases and good results in one. Restrepo *et al.* (*44*) recorded only four relapses in 22 cases treated with both amphotericin B and sulfa drugs.

Saramycetin

This agent (X-5079 C or Ro-27758) is fungistatic *in vitro* for *Blastomyces dermatitidis* in a concentration of 1 mg/ml. Albornoz *et al.* (*2*) used it with paracoccidioidomycosis in a daily dosage of 4 mg/kg, administered subcutaneously in four sessions of six hours each over a period of six weeks. Ferreira Lopes and Furtado (*18*) treated seven patients with total doses of from 8.4 to 21.6 mg and found that the drug was perfectly well tolerated, with no local pain, and that it gave good results in four cases and reasonable improvement in three.

Pymaricin

Like other related polyenes, this antibiotic acts on the endogenous respiration of the fungi. The MIC for *P. brasiliensis* is 20 μg/ml. Furtado (*24*) tried it out in the treatment of two cases

of paracoccidioidomycosis but had to discontinue it because of its toxic side-effects.

It may be given in spray form two to three times daily in a dosage of 0.1 ml in a 2.5 per cent aqueous solution containing 1 ml of Tyloxapal (Alesaire).

Hamycin

This drug, produced in India, is a heptane similar to amphotericin B. It is given orally. Studies in the future will tell more about its benefits.

Conclusions

Experience acquired to date in the treatment of paracoccidioidomycosis has led us to the following general conclusions:

- We do not believe that complete sterilization of *P. brasiliensis* is achieved, at least not as an immediate result of the therapy currently in use.
- The chemotherapeutic agents available at present are fungistatic *in vivo;* however, control of the mycotic infection depends largely on the capacity of the host's defensive system to combat the parasite.
- The natural defenses of the host, together with the acquired specific resistance to paracoccidioidomycosis, are the elements that the physician must cultivate and take the greatest possible advantage of. He can do this by assuring that the patient gets adequate rest in bed and proper food; by carefully watching out for toxic effects from the medication; and by building up the patient's strength, if necessary, with whole blood transfusions.
- At the present time sulfonamides are the therapy of choice for paracoccidioidomycosis, except in the cases indicated in the next point below. Nevertheless, it may be useful to intercalate one or two series of intravenous injections of amphotericin B, given on alternate days, not to exceed a total dose of 3 g.
- Treatment with amphotericin B should be reserved for cases in which the prognosis is grave or where there is resistance to sulfonamides. We are in favor of using moderate dosages (given intravenously in drops on alternate days) when a level of 1 to 1.2 mg/kg is reached. We also prefer to supplement it with sulfonamide therapy.
- We consider that clinical, serological, and radiological cure is a sufficient criterion for discharging a patient, although this picture does not assure that he has been cured mycologically. He should remain under observation for the rest of his life and periodically undergo treatment with sulfonamides during the first few years after his discharge.
- We believe that specific vaccination therapy is useful as a supplement to chemotherapy and the use of antibiotics.
- Naturally, when the disease occurs in association with diabetes, tuberculosis, etc., these other conditions must be appropriately treated. The simultaneous use of antifungal medication and tuberculostatics is beneficial, but isoniazid alone aggravates paracoccidioidomycosis. Potassium iodide is contraindicated in the treatment of this mycosis.
- In cases of pulmonary localization, it is important to have the assistance of a specialist in chest diseases in order to know the extent of the damage and to outline the most suitable program for recuperation and rehabilitation.
- Sometimes surgery is indicated—as, for example, when there are abscesses or in the event of certain abdominal or pulmonary lesions, including fibrous sequelae.
- Finally, and most important of all, early diagnosis of this mycosis provides a great advantage for successful treatment, making it possible to halt dissemination in vital organs such as the lungs and adrenals, as well as in the central nervous system, and to minimize or prevent altogether the dreaded fibrous sequelae.

SUMMARY

Successful diagnosis and treatment of paracoccidioidomycosis hinges on resolution of several problems. Primoinfection generally produces no recognizable symptoms and can be diagnosed only by skin testing; the incubation period may span 40 years or more; and there is presently no way of assuring a complete cure.

However, a number of drugs have been shown effective against the disease. The author recommends that treatment with sulfonamides

(perhaps with one or two series of intravenous amphotericin B injections) be considered the treatment of choice. However, treatment with amphotericin B should be reserved for cases where there is a grave prognosis or resistance to sulfonamides. The article also indicates general guidelines for treatment, and mentions three other drugs (saramycetin, pymaricin, and hamycin) that offer some possibilities for the future.

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