Effect of Clindamycin on Pneumonia from Reactivation of *Toxoplasma gondii* Infection in Mice

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Received 31 August 1990/Accepted 29 January 1991

Clindamycin was used to treat the reactivation of a chronic *Toxoplasma gondii* infection in mice. Clindamycin reduced mortality by 44% when used prophylactically (*P* < 0.001) but appeared to be less effective when used to treat clinically apparent reactivation. Further studies should be conducted to establish the efficacy of clindamycin for the treatment of toxoplasmosis in humans.

The reactivation of a *Toxoplasma gondii* infection is a potentially devastating complication of immunosuppression, especially in persons with AIDS. Pneumonia is a manifestation of toxoplasmosis in immunosuppressed persons, including those with transplanted organs, malignancies, and AIDS (4). In addition, some 3,000 cases of congenital toxoplasmosis occur each year in the United States alone, and pneumonia is a manifestation of the infection in congenitally infected infants (26). The combination of sulfadiazine and pyrimethamine, standard therapy for toxoplasmosis, may be toxic or only partially effective. Persons with AIDS are often unable to take sulfadiazine because of hypersensitivity. Effective alternative therapies for toxoplasmosis are needed.

Limited published observations suggest that clindamycin is effective in humans (9, 23). Clindamycin has usually been given in combination with other drugs, making evaluation of the contribution of clindamycin difficult. For example, a trial comparing the combination of clindamycin and pyrimethamine with the combination of sulfadiazine and pyrimethamine suggested that the efficacies of the two combinations were not significantly different (10). Clindamycin therapy has produced conflicting results in animal models of primary *T. gondii* infections (3, 12, 13, 15, 21, 25) and in cell cultures challenged with *T. gondii* tachyzoites (14, 20). Clindamycin has not been used to treat reactivated chronic infections in experimental animals.

We have described the clinical reactivation of a *T. gondii* infection in mice, the model used in these experiments (22). After injection with the C56 strain of *T. gondii*, BALB/c or DBA/2NCr mice suffer a primary systemic infection. With sulfadiazine treatment, the mice recover within 2 weeks. Pneumonia is not detectable histologically during the primary infection (22). Approximately 3 weeks later, the *T. gondii* infection is reactivated. The mice exhibit tachypnea, cyanosis, lethargy, ruffled fur, and weight loss. Pneumonia is the sole histological manifestation of reactivation. Other organs, including the heart and brain, have no histological evidence of a *T. gondii* infection during reactivation (22; unpublished observations). Most of the BALB/c mice recover from the *T. gondii* pneumonia without treatment, but most of the DBA/2NCr mice die. To further define the efficacy of clindamycin for the treatment of toxoplasmosis, we administered clindamycin and, for comparison, sulfadiazine and pyrimethamine, to DBA/2NCr mice to prevent or treat pneumonia caused by the reactivation of a chronic *T. gondii* infection.

Female DBA/2NCr mice were obtained from contract facilities of the National Cancer Institute (Bethesda, Md.) and maintained under specific-pathogen-free conditions. They were fed standard rodent chow and water ad libitum. Two hundred tachyzoites of the C56 strain of *T. gondii* were given intraperitoneally to 6- to 9-week-old mice. Sodium sulfadiazine was added to drinking water (400 mg/liter) from 3 to 21 days after inoculation to treat the initial systemic *T. gondii* infection (22).

Two approaches were used to determine the effect of clindamycin on the reactivation of the *T. gondii* infection. To treat established pneumonia, we began drug therapy after the mice developed the clinical signs mentioned above, usually 35 days after inoculation (range, 35 to 45 days). To prevent death from pneumonia, we began therapy 30 days after the injection of *T. gondii*, a time point determined in preliminary experiments to precede the clinical signs of pneumonia. The combination of sulfadiazine and pyrimethamine was given for comparison in all experiments. Therapy was continued for 14 days.

Powders of clindamycin, sulfadiazine, and pyrimethamine were mixed with ground rodent chow and offered to the mice ad libitum. Clindamycin (The Upjohn Co., Kalamazoo, Mich.) was offered in doses of 4 and 2 mg/g of food. Sodium sulfadiazine (City Chemical Corp., New York, N.Y.) and pyrimethamine (Burroughs Wellcome Co., Research Triangle Park, N.C.) were given in doses of 1.25 and 0.0112 mg/g of food, respectively (25). Since 20-g mice eat approximately 4 g of food per day (11; unpublished observations), we estimated that the mice ingested 800 or 400 mg of clindamycin per kg of body weight per day or 250 mg of sulfadiazine and 2.24 mg of pyrimethamine per kg of body weight per day. All experiments were performed at least twice, with similar results. Preliminary experiments to determine the effectiveness of parenteral therapy were discontinued because clindamycin proved irritating when injected intraperitoneally.

Survival was estimated with the product-limit method (17), and the significance of differences between treatments was estimated with the Tarone-Ware statistic (24). Calculations were performed with statistical software from BMDP Statistical Software, Inc., Los Angeles, Calif.

Clindamycin therapy clearly prevented mortality from *T.
Gondii pneumonia when begun 30 days after T. gondii inoculation, before pneumonia was clinically apparent (Fig. 1). Survival of groups that received clindamycin (2 or 4 mg/g of food) and the combination of pyrimethamine and sulfadiazine was significantly greater than survival of untreated mice (P < 0.001).

Clindamycin appeared to be less effective when begun 35 days after T. gondii inoculation, after signs of pneumonia were detectable (Fig. 2), and survival was not significantly different from that in untreated mice (P = 0.11 for 2 mg of clindamycin per g of food and P = 0.15 for 4 mg of clindamycin per g of food). In contrast, survival was significantly greater in mice given pyrimethamine and sulfadiazine than in untreated mice (P = 0.03).

In separate experiments, it was determined that drug therapy alone had no detectable effect on mice. Representative mice that died despite clindamycin therapy were autopsied and found to have T. gondii pneumonia similar to that which was previously described (22).

Clindamycin therapy has shown variable effectiveness in experimental animals with a primary T. gondii infection and treated with clindamycin alone (3, 12, 15, 21) or in combination with other drugs (13, 25). In contrast, clindamycin has not prevented the replication of T. gondii in tissue culture systems (14, 20).

In postnatally infected humans, severe toxoplasmosis usually results from the reactivation of an infection in the presence of immunosuppression rather than from a primary infection (19). In an effort to more closely model human infections, we used clindamycin to prevent or treat the reactivation of a T. gondii infection in mice. Further support for the relevance of this model is the observation that the primary manifestation of reactivation in mice in this study was pneumonia. Pneumonia is a manifestation of toxoplasmosis in both immunosuppressed (4) and congenitally infected (26) humans.

Toxoplasmosis has been a major problem in persons with AIDS. The combination of sulfadiazine and pyrimethamine is standard therapy, but many persons with AIDS are sensitive to sulfadiazine. Several newer agents are under development (1, 2, 5–8, 16, 18), but their efficacy in humans has not been documented. The evidence that clindamycin is effective against the reactivation of an infection in mice emphasizes the need for further evaluation of its effectiveness in humans.

This work was supported by grants from The Upjohn Co. and the Department of Veterans Affairs.

We are grateful for the excellent technical assistance of Kathleen Carroll.

REFERENCES


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