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Activity of Clindamycin with Primaquine against *Pneumocystis carinii* In Vitro and In Vivo

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The combination of primaquine with clindamycin is effective in both in vitro and in vivo models of *Pneumocystis* infection. Primaquine alone at concentrations from 10 to 300 μg/ml reduced the numbers of organisms in cultures to less than 7% of control. Significant inhibition was observed down to 0.1 μg/ml. Clindamycin at 5 μg/ml was ineffective alone. Combinations of clindamycin and primaquine in culture at various concentrations were effective, but there was no evidence of true synergy. In rats with established *Pneumocystis* pneumonia, clindamycin alone at 5 or 225 mg/kg was ineffective. Primaquine alone at 0.5 or 2 mg/kg did not significantly affect the numbers of organisms remaining. The combination of 0.5 mg of primaquine per kg and 225 mg of clindamycin per kg was effective for therapy, lowering the numbers of organisms in the lungs by about 90%. The combination of 2 mg of primaquine per kg and 225 mg of clindamycin per kg was more effective, lowering the numbers of organisms by almost 98%. In the in vivo prophylaxis model, primaquine at 0.1 or 0.2 mg/kg did not prevent the development of *Pneumocystis* pneumonia in immune-suppressed rats. Clindamycin at 50 mg/kg had a modest effect alone, but at 5 mg/kg all animals became heavily infected. At 0.5 mg/kg, primaquine alone reduced the severity of infection, but seven of eight rats were still infected. In contrast, the combination of 5 mg of clindamycin per kg and 0.5 mg of primaquine per kg prevented infection in 8 of 10 rats; 2 rats had minimal infection. These studies suggest that the combination of clindamycin and primaquine should be tested in therapy or prophylaxis of *Pneumocystis* infections in humans.

Treatment or prophylaxis of pneumonia caused by *Pneumocystis carinii* in patients with acquired immune deficiency syndrome (AIDS) has been hampered by the lack of effective, nontoxic agents. Pentamidine causes a variety of side effects which may limit therapy, and relapses are frequent (29, 34). Trimethoprim with sulfamethoxazole causes a surprisingly high incidence of side effects in AIDS patients, including severe hypersensitivity reactions that limit use for treatment (29, 33, 34). Other experimental agents have been tested with variable results. Difluoromethylornithine is effective in some patients, but thrombocytopenia may be a limiting toxicity (9, 34). Pyrimethamine with sulfadoxine (Fansidar) or dapsone with trimethoprim may be effective in individual patients, but significant adverse reactions still occur with both combinations (10, 21, 34). Trimetrexate, a lipid-soluble analog of methotrexate, is being investigated (1).

In the course of screening antimicrobial agents for potential effectiveness against *P. carinii*, we have evaluated combinations as well as individual agents. One of the combinations we chose to test was clindamycin with primaquine. This selection was based on the observation by Schmidt (25) that the curative action of primaquine in malaria was enhanced by mirincamycin. Mirincamycin, like clindamycin, is a lincosamide but has not been extensively tested in humans. Mirincamycin has no obvious advantages over clindamycin, which has been used successfully to treat a variety of bacterial infections (14). Moreover, clindamycin has been used successfully in AIDS patients for treatment of toxoplasmosis (20, 28). We therefore substituted clindamycin in our studies.

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We report here that the combination of clindamycin and primaquine is effective against *P. carinii* in culture and in both a treatment and a prophylaxis model of *Pneumocystis* pneumonia in rats. These models have predicted the clinical utility of trimethoprim-sulfamethoxazole and pentamidine (17, 18). For these reasons and because both clindamycin and primaquine are well-tested in humans, we suggest that the combination should be considered for trial in preventing and treating *P. carinii* pneumonia in AIDS patients.

MATERIALS AND METHODS

Cultures of *P. carinii* were prepared and evaluated as previously described (3, 23). Briefly, human embryonic lung fibroblastic cells (WI-38) were cultured in 12- or 24-well tissue cultures plates with minimum essential medium containing 10% fetal calf serum. Confluent monolayers were inoculated with homogenates of rat lungs infected with *P. carinii*. The infected rat lungs were obtained from rats immunosuppressed with cortisone acetate by the procedure of Frenkel et al. (8). Lungs from these animals were ground in minimum essential medium, and the inoculum was adjusted to give a final concentration in culture of 3×10^5 to 7×10^5 trophozoites per ml. Drugs were diluted in culture medium and added to the cultures at this time. The drug solution added was 10 μl or less per ml of culture medium; the inoculum was 0.1 ml/ml of culture medium. Plates were incubated at 35°C in 5% oxygen and 5 to 10% carbon dioxide, with the balance nitrogen. Separate plates were harvested for analysis at 1, 3, 5, 7, and 10 days after inoculation. At these times, 10 μl of culture supernatant was removed from each well, air dried onto 1 cm^2 of a slide, fixed with 100% methanol, and stained with Giemsa stain. These slides were

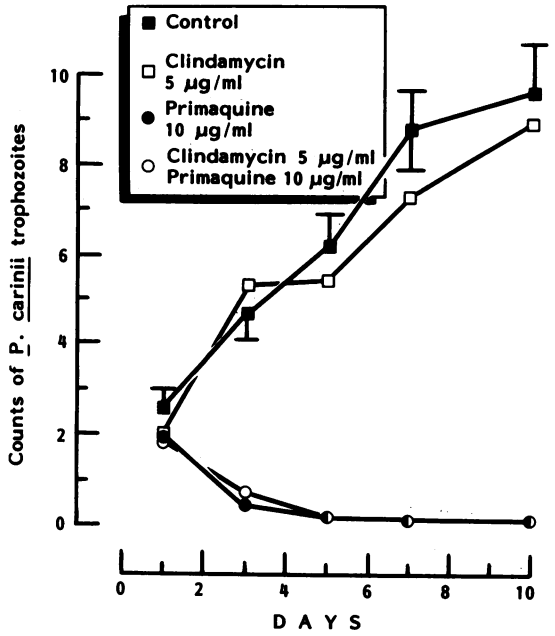

FIG. 1. Effect of primaquine and clindamycin on growth of cultured *P. carinii*. Primary cultures were grown in standard medium alone (control) or supplemented with clindamycin (5 μg/ml), primaquine (10 μg/ml), or both. Growth was assessed by counting the numbers of organisms in the culture supernatant at the times shown.

examined at ×1,000 magnification for quantitation of numbers of organisms (23). The data are reported as counts of trophozoites per field (see Fig. 1); multiplying these counts by a factor of 4×10^5 yields the number of organisms per milliliter of culture supernatant.

Female Sprague-Dawley rats (Harlan Laboratories, Indianapolis, Ind.) used in these studies weighed 120 to 150 g at the start of immune suppression. All rats received 25 mg of cortisone acetate subcutaneously twice weekly for 8 weeks. In one study (see Table 3), the animals were immune suppressed with dexamethasone (13) in the drinking water (1 mg/liter for 4 weeks, then 0.5 mg/liter for 4 weeks). Tetracycline was added to the drinking water of all animals to achieve a dose of 15 mg per rat per day. Rats were weighed weekly during the immunosuppression period. This model is similar to that originally described by Frenkel et al. (8) and used by Hughes et al. (17, 18) and by us (23, 27).

In the prophylaxis protocol, groups of 10 rats randomly assigned were started on drugs with the beginning of immunosuppression. These groups were maintained throughout the study. Rats for the treatment protocol were all immunosuppressed for 8 weeks. Any rats that gained weight during immune suppression were discarded (2). The remaining rats were randomly assigned to groups of 10 and then treated for 15 days with the test drugs as immune suppression continued. Doses and routes of administration for each study are indicated in the tables.

To evaluate infection at the end of the study, we anesthetized rats with ketamine hydrochloride and sacrificed them by exsanguination. Blood was collected with and without anticoagulant and used for hematology and cortisol assays. The lungs were removed aseptically. Portions of lung were fixed in Formalin for histology, and impression smears were made from other portions. Most of the lung tissue was frozen for subsequent culture (3).

Impression smears and sections were stained with a rapid methenamine-silver stain by the procedure of Brinn (4), modified as described earlier (3). Impression smears were also stained with Giemsa stain; sections were also stained with hematoxylin and eosin.

Slides were reviewed as unknowns by three examiners. Each evaluator rated the severity of infection using a scale of 0 to 5, with 5 representing >100 organisms per ×1,000 field, 4 representing 10 to 99 organisms per field, 3 representing 1 to 10 organisms per field, 2 representing 2 to 9 organisms in 10 fields, 1 representing 1 organism in 10 or more fields, and 0 representing no organisms seen in a search of more than 50 fields. As described, the evaluation scale is roughly logarithmic. The readings of the three evaluators were averaged to give a score for each slide. Means of these scores were separately determined for the Giemsa-stained impression smears, silver-stained impression smears, and silver-stained sections. Giemsa stains revealed living trophozoites and cysts. Silver stains revealed both living and dead cysts. For these reasons, we relied most heavily on Giemsa stains but confirmed our conclusions with silver stains.

Cortisol levels in serum were determined by using the GammaCoat radioimmunoassay (Travenol-Genentech Diagnostics, Cambridge, Mass.). Leukocyte counts and differentials were determined on a Coulter Counter (model Plus IV; Coulter Electronics, Inc., Hialeah, Fla.).

Comparisons between means were by an unpaired *t* test.

For electron microscopy, pellets of organisms centrifuged from culture supernatants were fixed in 3% glutaraldehyde in 0.1 M cacodylate (pH 7.2) buffer with 5% sucrose for 2 to 3 h and then postfixed in 1% osmium tetroxide in the same buffer without 5% sucrose for 1 h. Samples were then

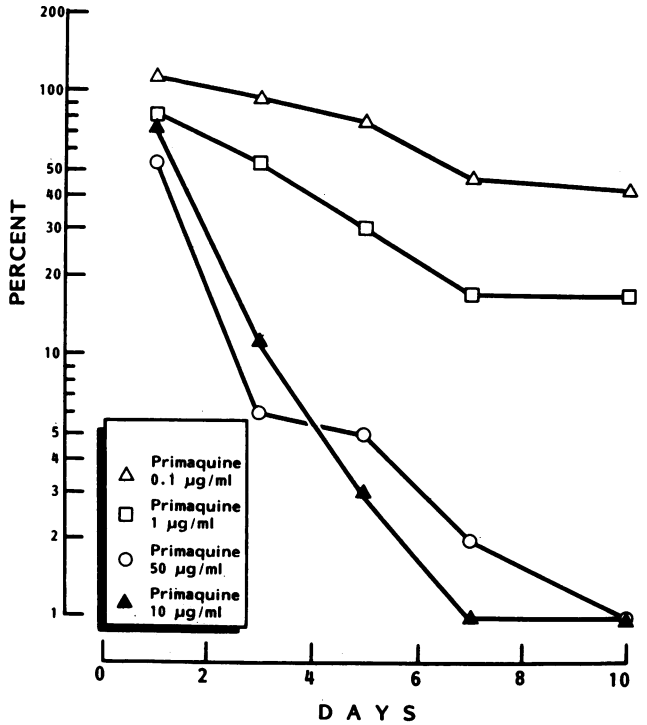

FIG. 2. Dose response of primaquine inhibition of growth of cultured *P. carinii*. Data are expressed as percentage of control growth in each individual experiment; the curves represent averages from four experiments. Controls increased an average of 3.5-fold in these experiments.

TABLE 1. Infectivity scores of rats treated for *P. carinii* pneumonia: study 1[a]

Treatment (mg/kg)[b]	*n*	Immune suppression[c]		Wt loss during treatment (g)[d]	Infectivity scores		
		Initial wt (g)	Wt loss (g)		Impression smears: Giemsa	Impression smears: Silver	Sections (silver)
Control	10	152 ± 1	28 ± 2	7.3 ± 1.3	3.4 ± 0.2	3.6 ± 0.1	3.2 ± 0.3
Primaquine (0.5)	9	152 ± 1	34 ± 3	0.6 ± 1.9	2.7 ± 0.4	2.7 ± 0.2[e]	3.0 ± 0.3
Clindamycin (5)	10	152 ± 2	31 ± 2	7.6 ± 3.0	2.9 ± 0.3	3.2 ± 0.3	3.0 ± 0.3
Primaquine (0.5) + clindamycin (5)	10	152 ± 1	31 ± 2	5.9 ± 2.8	2.6 ± 0.3	2.8 ± 0.2[e]	2.5 ± 0.4

[a] Results are expressed as mean ± standard error in this and all subsequent tables. Infectivity scores are described in Materials and Methods.
[b] Both drugs were administered in the drinking water to achieve the daily doses shown in the table.
[c] Initial weights are the body weights at the start of the 8-week period of immune suppression with cortisone acetate; weight loss is also reported for that period.
[d] Weight loss during therapy is reported for the 15-day period when the previously immune-suppressed rats received clindamycin or primaquine or both. Immune suppression continued throughout the treatment period.
[e] Statistically different from control, $P < 0.025$.

dehydrated in graded ethanol solutions and embedded in Spurr resin overnight. Thin sections were mounted on 200-mesh uncoated copper grids and stained with uranyl acetate and lead citrate. Grids were examined in a Zeiss EM109 transmission electron microscope.

RESULTS

The effects of clindamycin and primaquine against *P. carinii* were initially tested in culture. In four studies, 5 μg of clindamycin per ml showed little effect on control growth (Fig. 1). In contrast, primaquine reduced the numbers of *Pneumocystis* organisms in culture in a strong, dose-dependent manner (Fig. 2). Addition of clindamycin did not further reduce the numbers of organisms; one of four similar experiments is shown in Fig. 1.

Therapy of *P. carinii* pneumonia in rats with clindamycin, primaquine, or a combination of the two was evaluated in three independent studies. In the first study, animals received 0.5 mg of primaquine per kg, 5 mg of clindamycin per kg, or both drugs for 15 days. The combination of primaquine and clindamycin was possibly effective (Table 1). Primaquine alone at 0.5 mg/kg may have caused small reductions in infectivity scores in these moderately infected animals, but the changes were too small to be considered significant. Clindamycin alone also had little effect. No hematological toxicity was noted in any group; leukocyte counts, erythrocyte counts, platelet counts, and hemoglobin levels remained in the range of controls.

In the second therapy study, the dose of clindamycin was increased to 225 mg/kg, but the primaquine dose remained at 0.5 mg/kg. Doses were administered for 18 days, primaquine orally as in the previous study and clindamycin by subcutaneous injection three times daily. In these heavily infected animals, neither clindamycin nor primaquine alone had significant activity, but the combination reduced infectivity scores an average of one unit, representing about a 90% drop in the number of organisms detected in the lungs (Table 2).

In the third study, the clindamycin dose remained at 225 mg/kg but the primaquine dose was increased to 2 mg/kg. Doses were administered for 18 days, primaquine orally and clindamycin by subcutaneous injection three times daily, just as in the previous study. Neither clindamycin nor primaquine alone had significant activity (Table 3). The combination was effective, reducing infectivity scores almost two units in the Giemsa-stained impression smears, which corresponds to decreases approaching 98% in the numbers of trophozoites remaining in the lungs.

Prophylaxis of *P. carinii* pneumonia was evaluated in two independent studies. In the first, primaquine alone at 0.5 mg/kg reduced the severity of infection in all eight surviving animals in the group (Table 4). Clindamycin alone had no effect in the seven animals surviving the study. In sharp contrast, all 10 of the rats receiving the combination survived the study and 8 of the 10 showed no sign of infection with *P. carinii*. Gross examination of the living animals also revealed the differences between groups. Only the animals receiving the combination of drugs retained the glossy fur of a normal rat and a nearly normal appearance; in contrast, control animals or animals receiving only one of the drugs had ruffled fur, hunched backs, and a general appearance of disease. Cortisol concentrations were measured at sacrifice in all the animals. The average concentrations were not statistically significantly different among the four groups, and all groups were in the range expected for rats, based on previous experience with this model (13 to 21 μg/ml). Hema-

TABLE 2. Treatment of *P. carinii* pneumonia in rats: study 2[a]

Treatment (mg/kg)	*n*	Initial wt (g)	Wt loss (g)	Infectivity scores		
				Impression smears: Giemsa	Impression smears: Silver	Section (silver)
Control	10	141 ± 1	52 ± 4	4.4 ± 0.4	3.6 ± 0.2	3.8 ± 0.3
Clindamycin (225)	9	141 ± 2	45 ± 4	3.9 ± 0.3	3.5 ± 0.2	3.6 ± 0.2
Primaquine (0.5)	10	143 ± 1	52 ± 3	4.1 ± 0.3	3.2 ± 0.3	3.7 ± 0.2
Clindamycin (225) + primaquine (0.5)	9	141 ± 1	40 ± 4	3.2 ± 0.4[b]	2.8 ± 0.3[b]	2.8 ± 0.3[b]

[a] Primaquine was given in drinking water to achieve the desired daily dosage. Clindamycin hydrochloride was administered three times daily by subcutaneous injection (75 mg/kg per dose). Immune suppression was with dexamethasone (30 μg daily for 4 weeks, then 15 μg daily for the rest of the study).
[b] Statistically significantly different from control, $P < 0.05$.

TABLE 3. Treatment of *P. carinii* pneumonia in rats: study 3

Treatment (mg/kg)[a]	n	Body wt (g)[b]		Infectivity scores of impression smears	
		Initial	Loss	Giemsa	Silver
Control	8	128 ± 2	26 ± 4	4.7 ± 0.2	3.8 ± 0.1
Trimethoprim (50) + sulfamethoxazole (250)	9	126 ± 2	23 ± 2	1.9 ± 0.5[c] ($P < 0.001$)	2.0 ± 0.3[c] ($P < 0.001$)
Clindamycin (225)	10	124 ± 2	29 ± 3	4.4 ± 0.2	3.8 ± 0.1
Primaquine (2)	10	127 ± 2	33 ± 3	4.3 ± 0.3	3.4 ± 0.3
Clindamycin (225) + primaquine (2)	9	124 ± 2	25 ± 2	2.8 ± 0.4[c] ($P < 0.001$)	3.0 ± 0.3[c] ($P < 0.05$)

[a] Primaquine, trimethoprim, and sulfamethoxazole were given in drinking water to achieve the daily doses shown in the table. Clindamycin hydrochloride was administered three times daily by subcutaneous injection (75 mg/kg per dose). Immune suppression was achieved with cortisone acetate.
[b] Initial weights are the body weights at the start of the 8-week period of immune suppression with cortisone acetate; weight loss is also reported for that period.
[c] Statistically significantly different from control.

tology revealed no signs of significant toxicity with these agents used in the prophylaxis protocol.

The second prophylaxis study tested the effects of low doses of primaquine (0.1 or 0.2 mg/kg) with low or intermediate doses of clindamycin (5 or 50 mg/kg, respectively). Alone at these low doses, primaquine was without effect (Table 5). At 5 mg/kg, clindamycin alone was ineffective, but at 50 mg/kg, infectivity scores were slightly lower than control, suggesting a modest effect alone. The combinations of 0.1 or 0.2 mg of primaquine per kg with 50 mg of clindamycin per kg were no more effective than clindamycin alone in this study.

Electron microscopy was done to determine whether effective combinations of clindamycin and primaquine caused characteristic morphological changes in *P. carinii*. Organisms from animals that were not treated with drugs displayed typical morphology (12), with electron-dense cytoplasm, nuclear bodies, thin cell membranes, and tubular arrays (Fig. 3). Treatment with clindamycin and primaquine in combination altered the appearance of the trophozoites, which showed clearing of the cytoplasm and some surface alterations. Electron-dense particles were also noted in these organisms.

DISCUSSION

Based on our experiments, clindamycin with primaquine is a potentially useful combination for prophylaxis or therapy of *Pneumocystis* pneumonia. Primaquine alone at the doses tested had little or no activity in the therapy model but was active in the prophylaxis model and in culture. Clindamycin alone had no effect in culture or in therapy and only modest activity alone in one prophylaxis study. Previous studies by Hughes et al. (17) with clindamycin alone in a prophylaxis model of *P. carinii* pneumonia in rats suggested the drug at 400 mg/kg per day was ineffective. These results are largely confirmed in our studies.

TABLE 4. Prophylaxis against *P. carinii* pneumonia in rats

Treatment (mg/kg)[a]	n	Initial wt (g)[b]	Wt loss (g)[c]	Infectivity scores of impression smears	
				Giemsa	Silver
Control	10	144 ± 2	27 ± 3	3.2 ± 0.2	3.2 ± 0.2
Primaquine (0.5)	8	146 ± 1	28 ± 3	0.9 ± 0.2	1.4 ± 0.2
Clindamycin (5)	7	143 ± 5	25 ± 3	3.7 ± 0.1	3.7 ± 0.2
Clindamycin (5) + primaquine (0.5)	10	149 ± 2	24 ± 1	0.1 ± 0.1	0.2 ± 0.1

[a] Both drugs were administered in the drinking water to achieve the daily doses shown in the table.
[b] Initial weights are the body weights at the start of immune suppression with cortisone acetate and drug administration.
[c] Weight loss is over the 8-week period of immune suppression and drug administration.

For treatment studies, clindamycin was injected three times daily to elevate levels in serum transiently and mimic clinical use, but in our prophylaxis studies, clindamycin was given by continuous oral dosing, which would not be expected to produce large peaks in serum concentration. Not only was clindamycin given in these prophylaxis studies in such a way as not to produce peaks, but the dose was also quite low, based on comparisons for metabolic rates between humans and rats (5). In rats, clindamycin is rapidly cleared from serum with a half-life of about 30 min, but in humans the serum half-life is 2 to 3 h (5). Despite all these factors which brought levels in serum to below those expected to be effective, animals receiving clindamycin with primaquine had fewer organisms in their lungs than animals receiving primaquine alone. In the treatment protocols, the level of synergy seemed more dependent on the primaquine dose than on the clindamycin dose.

Two explanations for the effectiveness of clindamycin even at low doses in the combination regimens are possible. One explanation arises from the known ability of clindamycin to concentrate in alveolar macrophages: the concentration of drug at the site of infection may be much higher than concentrations in plasma (14). The second possible explanation has to do with the less well known actions of clindamycin.

Clindamycin has a broad range of effects beyond the ribosomal mechanism normally considered to be responsible of the antimicrobial activity of the compound and inhibits growth of a wide variety of microorganisms. The drug has been used against plasmodia and has also been used against *Toxoplasma* species (14, 15, 26, 28, 30). These activities show that the drug may affect protozoans as well as bacteria. The mechanism for the antiprotozoal effect of clindamycin is unknown, but may reside in the ability of the drug to penetrate these simple eucaryotic cells and affect mitochondrial activity (6). Clindamycin alters activity of polymorphonuclear leukocytes (7, 14, 26), changes release of toxins from staphylococci and other bacteria (14, 24), and alters binding of microbes to fibronectin (14, 19). Many of these actions require far lower concentrations of clindamycin than the MIC for gram-positive bacteria.

The only previous use of primaquine in the rat model for prophylaxis of *P. carinii* pneumonia was in fixed combination with chloroquine (18). The doses were 5.62 mg/kg daily

TABLE 5. Clindamycin-primaquine prophylaxis: study 2

Treatment (mg/kg)[a]	*n*	Initial wt (g)	Wt loss (g)	Infectivity scores		
				Impression smears		Sections (silver)
				Giemsa	Silver	
Control	9	126 ± 2	18 ± 3	2.6 ± 0.2	3.3 ± 0.1	2.1 ± 0.2
Clindamycin (5)	10	124 ± 3	12 ± 6	2.2 ± 0.2	2.6 ± 0.2[b] ($P < 0.025$)	2.3 ± 0.3
Clindamycin (50)	10	122 ± 2	12 ± 3	1.8 ± 0.3[b] ($P < 0.05$)	2.4 ± 0.2[b] ($P < 0.005$)	2.1 ± 0.2
Primaquine (0.1)	10	126 ± 2	21 ± 6	3.2 ± 0.2[c] ($P < 0.025$)	2.9 ± 0.2	2.9 ± 0.3[c] ($P < 0.05$)
Primaquine (0.2)	8	122 ± 2	19 ± 3	3.3 ± 0.3[c] ($P < 0.05$)	3.0 ± 0.2	3.1 ± 0.2[c] ($P < 0.005$)
Primaquine (0.1) + clindamycin (5)	10	121 ± 3	9 ± 3	2.2 ± 0.2	2.5 ± 0.3[b] ($P < 0.025$)	2.4 ± 0.3
Primaquine (0.2) + clindamycin (5)	10	127 ± 2	16 ± 2	2.5 ± 0.3	3.0 ± 0.3	2.1 ± 0.2
Primaquine (0.1) + clindamycin (50)	9	121 ± 4	12 ± 4	2.1 ± 0.3	2.4 ± 0.3[b] ($P < 0.005$)	1.8 ± 0.2
Primaquine (0.2) + clindamycin (50)	9	127 ± 4	10 ± 4	1.8 ± 0.1[b] ($P < 0.005$)	2.2 ± 0.2[b] ($P < 0.001$)	2.1 ± 0.1

[a] Primaquine and clindamycin were given in drinking water to achieve the daily doses shown in the table. Both drugs were administered throughout the 8 weeks of immune suppression with cortisone acetate.
[b] Statistically significantly lower than control.
[c] Statistically significantly higher than control.

for primaquine and 37.5 mg/kg daily for chloroquine. Used in this way, the drug was judged ineffective. In our tests, primaquine alone at 0.5 mg/kg daily was effective for prophylaxis, but the combination of primaquine with clindamycin was clearly superior. The combination is also clearly more effective for therapy.

The mechanism of action of primaquine and related 8-aminoquinolines is not completely clear. Many steps in nucleic acid synthesis may be affected (11). How lincosamides such as mirincamycin and clindamycin interact with primaquine is unknown. Mirincamycin lowered the dose of primaquine required to produce radical cure of *Plasmodium cynomolgi* infections in rhesus monkeys (25). Our studies on *P. carinii* suggest that clindamycin enhances the effectiveness of doses of primaquine that would be expected to be low in rats (16, 31). Whether this effect is caused by a specific interaction or simply relates to the tendency of both drugs to concentrate in tissues and cells in the lung remains unknown.

We were unable to detect primaquine or the metabolite carboxyprimaquine in serum from rats receiving primaquine continuously in drinking water at doses of 1.9 or 7.7 μmol/kg (0.5 or 2 mg/kg, respectively). We used a high-pressure liquid chromatography assay with a lower limit of sensitivity of ca. 0.1 μg/ml for primaquine and 0.5 μg/ml for carboxyprimaquine (22). Other workers have shown levels of primaquine in the blood to fall rapidly but levels in tissues to be higher and to fall more slowly. In one study, levels in serum were 3 nmol/g 3 h after an intraperitoneal dose of 82 μmol/kg, but levels in lung were about 200 μmol/g (1.3 μg/g

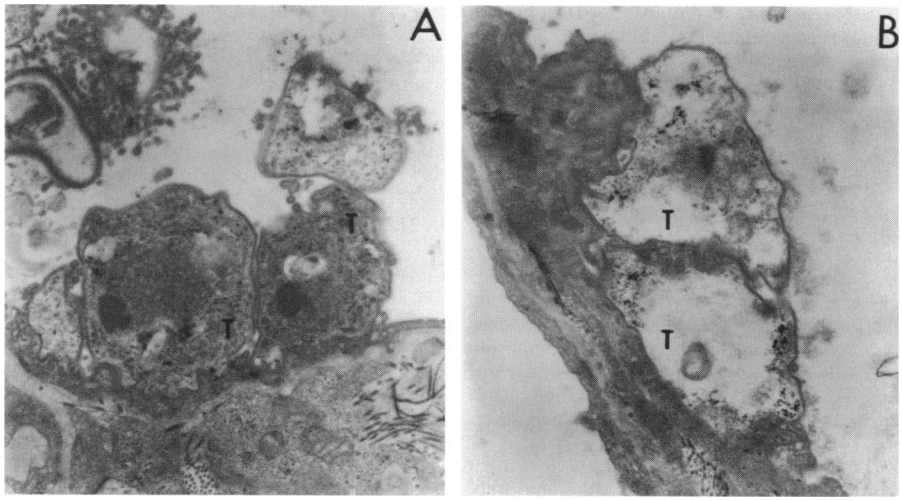

FIG. 3. Electron microscopy of *P. carinii*. Organisms in panels A and B are from control rats (A) or from rats treated with 225 mg of clindamycin per kg and 0.5 mg of primaquine per kg (B). T, Trophozoites attached to pneumocytes. Organisms from control rats show electron-dense cytoplasm, nuclear bodies, and thin plasma membranes, whereas those from treated animals have lost structural detail in the cytoplasm and show accumulation of electron-dense granular material.

of tissue) (16, 30). Of all tissues tested, lung tissue always contained the highest concentration of primaquine. Because the drug tends to concentrate in lung tissue, therapeutic efficacy may not be easily predicted by concentrations in serum.

Part of the appeal of the clindamycin-primaquine combination is that both drugs are well-known clinical entities. Primaquine has been widely used in treatment and prophylaxis of malaria (32). The drug is generally safe except for patients with glucose-6-phosphate dehydrogenase deficiency, who may suffer hemolytic anemia. Other hematological side effects are usually dose dependent. To our knowledge, primaquine has not been studied in AIDS patients. When used as an antibacterial agent, clindamycin is relatively nontoxic but can be associated with gastrointestinal symptoms requiring cessation of therapy. Clindamycin has been used to treat toxoplasmosis in mice (15) and in a very limited number of AIDS patients (20, 28). No evidence of unpredicted or unusual side effects was noted, but the drug has not been systematically evaluated.

Because clindamycin has been used safely in AIDS patients and because of the unusual efficacy of the combination of clindamycin with primaquine in our model system, we suggest cautious testing of clindamycin in appropriate combinations with primaquine in humans.

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