Antiviral Research, 16 (1991) 17–28

ADONIS 016635429100237N

AVR 00496

Evaluation of infrequent dosing regimens with (*S*)-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]-cytosine (*S*-HPMPC) on simian varicella infection in monkeys

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(Received 8 August 1990; accepted 6 December 1990)

Summary

(*S*)-1-[3-Hydroxy-2-(phosphonylmethoxy)propyl]cytosine (*S*-HPMPC) was able to prevent simian varicella infection in African green monkeys inoculated intratracheally with virus. A dose of 50 mg/*S*-HPMPC/kg administered intravenously was shown to prevent the development of rash, reduce viremia and protect the monkeys from death. The 50 mg/kg dose was effective when treatments initiated on day 2 post-infection (p.i.) was given as ten daily doses of 5 mg/kg, as 10 mg/kg administered on five days on an alternate-day schedule, as two 25 mg/kg doses given on day 2 and on day 7 p.i., or as a single injection of 50 mg/kg on day 2. The single 50 mg/kg dose was also effective when treatment was delayed until four days p.i., but was ineffective when treatment was delayed until six days p.i. The 50 mg/kg dose was not effective when given orally by gavage. No evidence of toxicity was noted in daily clinical examinations, or in frequent hematology and clinical chemistry tests performed during the clinical evaluation of the infection.

Simian varicella virus; HPMPC; Monkey

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Introduction

The phosphonate nucleotide compounds first reported by Holý and Rosenberg (1987a,b) are of interest due to their uniqueness of structure and their broad range of antiviral activity. The prototype compound (*S*)-9-[3-hydroxy-2-(phosphonylmethoxy)propyl]adenine [(*S*)-HPMPA] was shown, in in vitro assays, to inhibit a variety of DNA viruses as well as some RNA viruses (De Clercq et al., 1986, 1987). Activity was impressive against the herpesviruses and, in particular, against cytomegalovirus, Epstein-Barr virus, and varicella-zoster virus (Lin et al., 1987, Baba et al., 1987). (*S*)-HPMPA was found to be an effective antiviral in several in vivo animal model systems employing herpes simplex type 1 (HSV-1) infection (De Clercq et al., 1987). Rabbits inoculated intraocularly with a thymidine-kinase-negative (TK^-) strain of HSV-1 promptly resolved their keratitis after topical HPMPA treatment was initiated. Mice infected by intraperitoneal injection of HSV-1 showed a reduced mortality following oral administration of HPMPA when compared with placebo-treated controls.

Another compound in this series (*S*)-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]cytosine [(*S*)-HPMPC], an analogue of HPMPA in which a cytosine replaces the adenine base, was also shown to be a highly effective antiviral, both in vitro and in vivo (De Clercq et al., 1987; Snoeck et al., 1988; Bronson et al., 1989a). Pharmacological data with HPMPC in MRC-5 cells showed the phosphorylated forms of HPMPC to have a long half-life (Bronson et al., 1989b). These data suggested that dosing with HPMPC might be done less frequently than with other rapidly metabolized antivirals and still induce a significant antiviral effect. In vivo studies were performed with mice infected with HSV type 2 and treated with HPMPC at the same total dose; however, treatment was administered as daily doses for five days, doses once on days 1, 3, and 5, doses at 1 and 5 days or as a single dose on day 1. All treatment regimens were efficacious, with the single dose of 50 mg/kg given on day 1 being the most effective.

This report details studies with HPMPC in the treatment of simian varicella virus infection in African green monkeys in which intermittent treatments were evaluated for ability to prevent the development of clinical disease.

Materials and Methods

In vitro antiviral assays

(*S*)-HPMPC (provided by Bristol Myers-Squibb) was dissolved at 1000 μg/ml in tissue-culture medium (minimum essential medium with 10% fetal bovine serum, 100 units/penicillin/ml, 100 μg/streptomycin/ml and 50 μg/fungizone/ml and sterilized by filtration through a 0.2-μ membrane filter. Vero cells grown in 24-well tissue culture plates were inoculated with a dilution of simian varicella

virus containing approximately 100 PFU of virus. After 3 h incubation at 37°C in a CO_2 incubator, the virus inoculum was removed and each well washed with 1 ml of tissue-culture medium. Dilutions of HPMPC were made from the stock solution to provide two-fold dilutions from 100 μg/ml to 1.5 μg/ml. Each of the HPMPC concentrations was inoculated into triplicate wells along with three virus control wells receiving culture medium without HPMPC. The plates were reincubated for five days, at which time the culture fluids were discarded and the monolayers fixed with methanol, stained with methylene-blue/basic-fuchsin stain. The numbers of plaques were counted, and the mean number of plaques was determined for the three wells at each drug concentration.

Simian varicella infection of monkeys

African green monkeys (*Cercopithecus aethiops*) were determined to be free of antibody to simian varicella virus by testing a 1:10 dilution of serum against 100 PFU of virus in a neutralization assay (Soike et al., 1987). Infection was accomplished by intratracheal inoculation of each monkey with 1.5 ml of a dilution of stock virus containing between 10^4 and 10^5 PFU of simian varicella virus. The course of simian varicella infection was monitored as described previously (Soike et al., 1986, 1989). In brief, rash was evaluated by daily examination and scoring the severity of rash on a scale of ± to 4+.

Viremia was quantified by collection of lymphocytes from a 2-ml heparinized blood specimen taken at days 3, 5, 7, 9 and 11 after virus inoculation. The lymphocytes were separated on a Ficoll-hypaque gradient, washed twice in tissue-culture medium (RPMI-1640, plus 15% fetal bovine serum, 100 units penicillin and 100 μg streptomycin/ml). The collected lymphocytes were washed twice in medium and suspended in 10 ml of the medium. This suspension was equally divided between two 25-cm^2 tissue culture flasks containing Vero cells. Following incubation at 37°C for five to seven days, the cell monolayers were fixed and stained as described above, and the numbers of plaques counted in each flask. The mean number of plaques present in the two flasks inoculated with each blood specimen allows quantification of the viremia. At 14 and 21 days post virus inoculation, blood was drawn from each monkey and the sera tested for antibody in a plaque reduction assay. Dilutions of each serum were mixed with a dilution of virus to provide 100–200 viral plaques in control cultures without serum. The virus serum mixtures were incubated for 1 h at room temperature, after which, each virus serum mixture was equally divided between two flasks of Vero cells and the cultures incubated. After five to seven days incubation the monolayers were stained, and the number of plaques counted. The antibody titer was expressed as the serum dilution resulting in the reduction in number of plaques by 80% or more, from the number present in the control cultures without added serum.

In addition, hematology and clinical chemistry tests were done on blood taken at days 0, 3, 7, 9 and 11 after virus inoculation for determination of possible toxicity of drug treatment.

In vivo antiviral activity

In an initial experiment, HPMPC was dissolved in phosphate-buffered saline (PBS) at concentrations of 5 and 1 mg/ml. Groups of four monkeys were treated daily by intravenous (i.v.) administration of 1 ml of drug solution per kg, providing treatment groups receiving 5 or 1 mg/kg/day. Treatment began 48 h after virus inoculation and was given for ten days. A group of four untreated control monkeys received similar daily i.v. treatments with PBS. The monkeys were evaluated for clinical expression of simian varicella infection, as described above.

In a second experiment, groups of three infected monkeys received HPMPC i.v. as a total dose of 50 mg/kg, with the dose given as ten daily treatments of 5 mg/kg, as five treatments of 10 mg/kg given on days 2, 4, 6, 8 and 10 post infection, or as two 25 mg/kg doses on days 2 and 7 post infection. Another group of three infected monkeys which received PBS by daily i.v. injection served as controls. All treatments began 48 h after virus inoculation.

In a third experiment, each of the HPMPC treated monkeys received 50 mg/kg given as a single i.v. treatment. One group received the dose at 48 h post infection (day 2), a second group received this same dose at 96 h after virus inoculation (day 4), and a third group received HPMPC on day 6 after virus inoculation. The control group received PBS given on day 2 post injection. All groups contained three monkeys.

In another experiment, HPMPC as the single 50 mg/kg dose was evaluated as in the previous experiment, with the drug given orally by gavage rather than by intravenous infusion. The single oral treatment was given at either 2, 4, or 6 days after virus inoculation.

Results

S-HPMPC was shown to completely inhibit the replication of simian varicella virus in vitro at a concentration of 6.26 μg/ml with an ED_{50} of 2.3 μg/ml. This inhibitory dose compares favorably with other antivirals tested in vitro against simian varicella virus. An ED_{75} of 6.2 μg/ml was reported for acyclovir and 4.7 μg/ml for adenine arabinoside-5′-monophosphate (Soike et al., 1984). E-5-(2′-bromovinyl)-2′-deoxy-uridine (BVDU) and 1-β-D-arabinofuranosyl-E-5-(2-bromovinyl) uracil (BVaraU) were considerably more effective, with ED_{75} values of 0.0073 μg/ml and 0.0014 μg/ml, respectively.

In an initial experiment, an effective daily dose of HPMPC was determined which prevented the development of symptoms of simian varicella infection in inoculated monkeys. The virus inoculum contained 1.3×10^4 PFU per monkey. HPMPC was administered as i.v. doses of 5 mg/kg/day or 1 mg/kg/day. Each of four placebo-treated control monkeys became infected, with development of rash and viremia and death of three of the four monkeys with severe simian varicella (Table 1). However, rash was completely prevented in

TABLE 1

Evaluation of daily intravenous dosing of *S*-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]cytosine (HPMPC) for antiviral activity against simian varicella virus infection in African green monkeys

Treatment	Monkey	Appearance of Rash[c] – Days post-infection (p.i.)								Viremia[d] – Mean PFU on days post-infection					Antibody titer[e]	
group[a]	number[b]	7	8	9	10	11	12	13	14	3	5	7	9	11	14 days	21 days
Control	J252	1+	Died							5	225	>1000	Died		Dead	Dead
PBS. i.v.	J258	Died								2	510	Died			Dead	Dead
	J259	–	–	1+	2+	3+	Died			4	70	>1000	>1000	>1000	Dead	Dead
	J260	±	1+	2+	2+	3+	2+	2+	2+	1	15	657	149	19	1:160	1:640
HPMPC	J257	–	–	–	–	–	–	–	–	3	4	36	1	0	1:40	1:160
5 mg/kg/	J253	–	–	–	–	–	–	–	–	2	14	24	0	0	1:20	1:80
day, i.v.	J255	–	–	–	–	–	–	–	–	2	15	30	7	1	1:10	1:10
	J256	–	–	–	–	–	–	–	–	2	18	31	1	2	1:80	1:80
HPMPC	J247	–	–	±	2+	3+	4+	4+	4+	0	7	34	20	156	1:10	1:640
1 mg/kg/	J248	–	–	±	2+	Died				2	16	47	55	Died		
day, i.v.	J249	1+	1+	Died						0	6	70	Died			
	J250	–	–	±	±	1+	1+	1+	1+	0	3	49	14	7	1:80	1:320

[a] Treatment was begun 48 h after virus inoculation and continued once daily for ten days.
[b] Monkeys were infected by intratracheal inoculation of 1.3×10^4 PFU of simian varicella virus per monkey.
[c] Rash was scored daily on a score of ± (minimal) to 4+ (severe).
[d] Viremia was expressed as the mean number of plaques developing in Vero Cells co-cultured with lymphocytes separated from 1 ml of heparinized blood.
[e] Antibody titer was the serum dilution which neutralized 80% or more of the plaques present in control cultures without added monkey serum.

TABLE 2

Comparison of daily intermittent intravenous dosing of *S*-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]cytosine (HPMPC) for antiviral activity against simian varicella virus infection in African green monkeys

Treatment group[a]	Monkey number[b]	Appearance of Rash[c] – Days post-infection (p.i.)								Viremia[d] – Mean PFU on days post-infection					Antibody titer[e]	
		7	8	9	10	11	12	13	14	3	5	7	9	11	14 days	21 days
Control	J911	1+	2+	3+	4+	4+	4+	Died		79	178	>1000	>1000	>1000	Dead	Dead
PBS. i.v.,	J912	1+	3+	4+	4+	Died				23	110	>1000	>1000	Died	Dead	Dead
q.d.	J913	±	±	Died						15	32	>1000	Died		Dead	Dead
HPMPC	J573	–	–	±	±	–	–	–	–	1	0	3	0	0	<1:10	1:20
5 mg/kg	J261	–	1+	±	±	±	–	–	–	0	1	8	2	0	1:80	1:80
i.v., q.d.	J904	–	±	±	1+	±	–	–	–	2	1	5	3	0	1:20	1:20
HPMPC	J571	–	–	±	±	–	–	–	–	3	3	5	0	0	<1:10	1:20
10 mg/kg	J907	–	–	±	–	–	–	–	–	0	2	0	0	0	<1:10	1:10
i.v., q.o.d.	J903	–	–	–	–	–	–	–	–	6	0	0	1	0	1:20	1:160
HPMPC	J908	–	–	–	–	–	–	–	–	2	3	2	0	0	1:40	1:80
25 mg/kg	J574	–	–	–	–	–	–	–	–	2	2	4	0	0	<1:10	<1:10
i.v., days 2 & 7	J909	–	–	–	–	–	–	–	–	2	3	0	1	0	<1:10	<1:10

[a] Treatment was begun 48 h after virus inoculation with one group receiving daily dosing of 5 mg/kg, one group receiving 10 mg/kg every other day, and a third group receiving 25 mg/kg initially and again five days later (i.e. the total dose was 50 mg/kg for each treatment group).
[b] Monkeys were infected by intratracheal inoculation of 7.7×10^4 PFU of simian varicella virus per monkey.
[c] Rash was scored daily on a score of ± (minimal) to 4+ (severe).
[d] Viremia was expressed as the mean number of plaques developing in Vero Cells co-cultured with lymphocytes separated from 1 ml of heparinized blood.
[e] Antibody titer was the serum dilution which neutralized 80% or more of the plaques present in control cultures without added monkey serum.

the monkeys receiving 5 mg HPMPC/kg, and viremia was appreciably reduced from that seen in the untreated controls. Infection occurred in each of the four monkeys treated with the 1 mg/kg dose. Viremia, while present in each of these monkeys, was appreciably less than the viremia which was present in the control monkeys. Two of the four monkeys died from systemic simian varicella infection. Antibody titers to simian varicella virus were slightly depressed in the monkeys receiving the 5 mg/kg dose when compared with the titer in the single surviving control monkey or with monkeys treated with 1 mg/kg/day.

In the initial experiment, a daily dose of 5 mg/kg for ten days, with a resulting total dose of 50 mg/kg, was effective in preventing the development of severe simian varicella. In a second experiment, this same dose of 50 mg/kg was administered i.v. using different treatment regimens. Monkeys were inoculated with 7.7×10^4 PFU of virus. Severe disease was evident in the three control monkeys, with development of marked rash, severe viremia and death of each of the monkeys (Table 2). HPMPC as a daily 5 mg/kg dose resulted in a greatly reduced rash appearing in each of the three monkeys and viremia was essentially prevented. The 10 mg/kg HPMPC given on an alternate-day schedule resulted in a minimal rash of one or two days duration in two of the three monkeys, with no rash in the third monkey. Viremia was reduced. HPMPC at 25 mg/kg given on post-infection days 2 and 7 prevented rash completely. Viremia was detected in low titer in repeated samplings during the early post-infection period. Antibody titers to simian varicella virus were generally low or less than 1:10 in many of the monkeys treated with HPMPC.

A single dose of 50 mg HPMPC/kg administered by intravenous injection was evaluated, with the drug being given at different times after infection. Monkeys were infected by inoculation of 1.7×10^5 PFU of simian varicella virus. Two of the three placebo-treated control monkeys developed rash and viremia (Table 3). The third control monkey exhibited only a minimal rash and no viremia. One of the three control monkeys died of simian varicella infection. The 50 mg/kg dose of HPMPC given as a single injection on either day 2 or 4 post infection was equally effective, causing markedly reduced viremia and resulting in limited rash in one of the three monkeys in each treatment group. In monkeys in which the administration of HPMPC was delayed until days 6 after virus injection, treatment did not prevent the development of rash. Instead, two of the three monkeys developed moderate to moderately severe rash which persisted for seven or eight days of observation. The third monkey had a reduced rash, with several vesicles present on only one day. Viremia was present in two of the three monkeys in this treatment group. Viremia was seen on day 3, increased in number of plaques on day 5 and then appeared to resolve after treatment was given on day 6. This is in contrast to the two controls, as well as to controls in prior experiments where viremia increased between days 3 and 5 p.i. to maximum severity on day 7 p.i. Viremia typically declines on days 9 and 11 as antibody appears.

Again, antibody to simian varicella virus was of low titer in the monkeys receiving HPMPC on days 2 or 4 following infection. Moderate antibody titers

TABLE 3

Evaluation of delayed dosing of *S*-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]cytosine (HPMPC) administered by intravenous injection for antiviral activity against simian varicella virus infection in African green monkeys

Treatment	Monkey	Appearance of rash[c] – Days post-infection (p.i.)								Viremia[d] – Mean PFU on days post-infection					Antibody titer[e]	
group[a]	number[b]	7	8	9	10	11	12	13	14	3	5	7	9	11	14 days	21 days
Control	K147	–	±	2+	3+	2+	1+			2	117	538	0	0	1:20	1:640
PBS, i.v.	K145	–	–	–	1+	±	±	–	–	0	0	0	0	0	1:40	1:160
	K150	1+	2+	4+	Died					9	564	>1000	Died		Dead	Dead
HPMPC	K111	–	–	–	±	±	–	–	–	0	0	0	0	0	<1:10	1:20
50 mg/kg	K143	–	–	–	–	–	–	–	–	1	0	0	0	0	<1:10	<1:20
i.v., day 2	K142	–	–	–	–	–	–	–	–	0	0	0	0	0	<1:10	<1:20
HPMPC	K105	–	–	–	–	–	–	–	–	8	1	0	0	0	1:10	1:40
50 mg/kg	K097	–	±	1+	±	–	–	–	–	1	0	0	0	0	1:20	1:160
i.v., day 4	K144	–	–	–	–	–	–	–	–	0	0	0	0	0	<1:10	<1:20
HPMPC	K146	–	–	1+	–	–	–	–	–	4	22	0	0	0	1:20	1:160
50 mg/kg	K149	–	1+	1+	2+	2+	2+	2+	±	2	7	1	0	0	1:10	1:160
i.v., day 6	K148	±	2+	3+	3+	3+	3+	2+	1+	0	0	0	0	0	1:10	1:160

[a] Treatment at 50 mg/kg was given as a single intravenous injection with treatment administered on day 2, day 4 or day 6 after virus inoculation.
[b] Monkeys were infected by intratracheal inoculation of 1.7×10^5 PFU of simian varicella virus per monkey.
[c] Rash was scored daily on a score of ± (minimal) to 4+ (severe).
[d] Viremia was expressed as the mean number of plaques developing in Vero Cells co-cultured with lymphocytes separated from 1 ml of heparinized blood.
[e] Antibody titer was the serum dilution which neutralized 80% or more of the plaques present in control cultures without added monkey serum.

TABLE 4

Evaluation of delayed dosing of *S*-1-[3-hydroxy-2-(phosphonylmethoxy)propyl]cytosine (HPMPC) administered by oral gavage

Treatment group[a]	Monkey number[b]	Appearance of rash[c] – Days post-infection (p.i.)								Viremia[d] – Mean PFU on days post-infection					Antibody titer[e]	
		7	8	9	10	11	12	13	14	3	5	7	9	11	14 days	21 days
Control	J925	±	2+	3+	2+	1+	–	–	–	6	50	151	9	0	1:20	1:40
PBS. p.o.,	J914	1+	3+	Died						19	547	>1000	Died		Died	Died
day 2 p.i.	J920	–	–	1+	Died					1	358	>1000	>1000	Died	Died	Died
	K114	+	1+	2+	2+	+	–	–	–	0	251	169	5	0	1:10	1:40
HPMPC	K094	–	–	±	1+	–	–	–	–	0	34	184	17	0	1:40	1:40
50 mg/kg	K092	–	1+	2+	2+	1+	–	–	–	7	70	75	13	0	1:20	1:80
p.o., day 2	J916	–	–	±	1+	Died				5	34	>800	>1000	Died	Died	Died
HPMPC	J924	1+	1+	2+	2+	1+	±	–	–	0	1	4	0	0	1:20	1:160
50 mg/kg	J915	1+	2+	2+	2+	1+	±	–	–	16	375	23	2	0	1:10	1:40
p.o., day 4	J919	+	2+	3+	3+	3+	3+	2+	2+	4	590	>800	>800	0	1:40	>1:640
HPMPC	J918	1+	3+	4+	4+	Died				8	305	>1000	>1000	Died	Died	Died
50 mg/kg	J917	1+	2+	3+	4+	3+	3+	2+	2+	2	100	665	23		1:320	1:640
p.o., day 6	J923	3+	Died							0	>1000	>1000	Died		Died	Died

[a] Treatment at 50 mg/kg was given as a single oral dose with treatment administered on day 2, day 4 or day 6 after virus inoculation.
[b] Monkeys were infected by intratracheal inoculation of 4.8×10^3 PFU of simian varicella virus per monkey.
[c] Rash was scored daily on a score of ± (minimal) to 4+ (severe).
[d] Viremia was expressed as the mean number of plaques developing in Vero Cells co-cultured with lymphocytes separated from 1 ml of heparinized blood.
[e] Antibody titer was the serum dilution which neutralized 80% or more of the plaques present in control cultures without added monkey serum.

were seen in the monkeys treated with HPMPC on day 6 post-infection, as well as in the two control monkeys.

To assess oral efficacy of HPMPC, a single dose of HPMPC was administered at 50 mg/kg by gavage on days 2, 4, or 6 post-infection. The virus inoculum contained 4.8×10^3 PFU of simian varicella virus. The data are presented in Table 4. The control group containing four monkeys developed infection with rash; two developed a moderate viremia and two developed a severe viremia. The two monkeys with severe viremia died from simian varicella pneumonia. In the group of monkeys treated with HPMPC given as a single 50 mg/kg oral dose on day 2 post-infection, the effect was equivocal, with rash possibly slightly less than that appearing in the control monkeys. Viremia was severe in one monkey and of moderate severity in the other two. The one monkey with severe viremia died as a consequence of infection. Oral treatment, when deferred until days 4 or 6 post-infection, had no effect on rash or viremia, and virus-related deaths occurred in two of the three monkeys treated at day 6 post-infection.

Antibody titers to simian varicella virus in the two surviving control monkeys were unusually low at 21 days post-infection. Antibody titers in the HPMPC-treated monkeys showed variability.

In studies in monkeys infected with simian varicella virus, no evidence of toxicity was seen following treatment with 50 mg/kg as a single dose i.v. or oral dose, or when this same total dose was divided as ten daily 5 mg/kg doses. The monkeys appeared alert and normally responsive, with unaffected appetites. Toxicity was monitored by daily clinical examinations of the monkeys, and by hematology and clinical chemistry tests performed frequently through the period of clinical evaluation of the viral infection. Red and white blood-cell counts and hematocrits remained within normal limits. Clinical chemistry tests, which included alanine and aspartate amino-transferases, creatinine, blood urea nitrogen, albumin, globulin and total serum protein, were within normal ranges.

Conclusions

The phosphonyl methoxy alkyl derivatives of purine and pyrimidine bases are a new group of antiviral compounds that do not require phosphorylation by the virus thymidine kinase. As a consequence, they may exhibit antiviral activity against thymidine-kinase-negative viruses or viruses deficient in thymidine kinase. Synthesis of these compounds with the phosphorus atom linked to the carbon atom of the alkylside chain of the nucleoside protects the molecule from the host esterases, which would readily hydrolyse a phosphate group attached by an oxygen atom to the nucleoside. In addition, phosphorylation to the di- and tri-phosphates results in a compound which is slow to degrade, and therefore the active molecule persists and exerts a lengthy antiviral effect.

The activity of HPMPC has been demonstrated in an animal model system in which African green monkeys were infected with simian varicella virus. HPMPC, at a dose of 50 mg/kg, with treatment started 48 h after virus inoculation, effectively moderated the clinical course of simian varicella. HPMPC was highly effective when the dose was administered as ten daily doses of 5 mg/kg, as 10 mg/kg given on an alternate-day schedule, as 25 mg/kg administered on days 2 and 7 post-infection, or as a single 50 mg/kg dose given on day 2 post-infection. Even a single dose of 50 mg/kg injected i.v. on day 4 after virus inoculation appeared to be an efficacious treatment regimen.

It was significant that treatment with HPMPC as a single dose of 50 mg/kg could be deferred as late as four days after virus inoculation and still prevent the sequelae of infection. The data from monkeys treated as late as six days post-infection suggest that treatment at this time still may prevent progression of simian varicella infection and may hasten its resolution.

Oral administration of HPMPC was not effective in preventing simian varicella infection. This would suggest that HPMPC is poorly absorbed from the gastrointestinal tract, although oral HPMPA has been reported to protect mice from death following intraperitoneal inoculation of HSV-1 (DeClercq et al., 1987).

Antibody titers to simian varicella virus were noted to be appreciably lower in many of the monkeys treated with effective doses of HPMPC relative to titers in monkeys which served as untreated controls. This may reflect inhibition of virus replication by HPMPC, resulting in a reduced antigenic stimulus or possibly an effect on antibody-producing cells. Treatment with HPMPC at six days after virus inoculation, however, did not depress the antibody titers in the infected monkeys (Table 3). It therefore appears that this inhibition of antibody is a result of virus inhibition, and is not likely a direct effect on the antibody producing B-lymphocytes.

In conclusion, HPMPC at 50 mg/kg in this animal model is as effective given as a single dose as when divided into up to ten doses given over a ten-day period. This possibility for efficacious infrequent dosing would be important for the development of convenient schedules for a parenteral drug. The results obtained in a primate animal model would suggest that similar efficacy would be seen in humans

Acknowledgements

Appreciation is expressed to Mary Ann Bennett for typing of the manuscript. This study was supported by Contract NO1-AI-62521 with the Antiviral Testing Program of the National Institute of Allergy and Infectious Diseases.

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