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Inhibition of Simian varicella virus infection of monkeys by 1-(2-deoxy-2-fluoro-1-β-D-arabinofuranosyl)-5-ethyluracil (FEAU) and synergistic effects of combination with human recombinant interferon-β

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Summary

1-(2-Deoxy-2-fluoro-1-β-D-arabinofuranosyl)-5-ethyluracil (FEAU) has been shown to be a highly effective inhibitor of Simian varicella virus infection in African green monkeys. Administration of FEAU by either intravenous injection or gavage at doses as low as 1 mg/kg/day prevented the development of rash and reduced viremia. The effective dose could be further reduced to 0.2 mg/kg/day when administered in combination with a sub-effective dose of human recombinant interferon-β. No evidence of toxicity was seen in monkeys treated for 10 days with FEAU doses of 10 mg/kg/day when they were monitored by hematology and clinical chemistry tests and by clinical observations.

FEAU; Hu rIFNβ; Simian varicella virus; Monkey

Introduction

Prior studies with the fluorinated pyrimidines 1-(2-deoxy-2-fluoro-1-β-D-arabinofuranosyl)-5-iodouracil (FIAU) and 1-(2-deoxy-2-fluoro-1-β-D-arabinofurano-

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syl)-5-methyluracil (FMAU) showed them to be highly effective inhibitors of Simian varicella virus in vitro and in vivo (Soike et al., 1986). FIAU at oral doses of 3 mg/kg/day prevented development of Simian varicella rash and appreciably reduced viremia while FMAU was highly effective in preventing rash and viremia at doses as low as 0.2 mg/kg/day. Unfortunately, central nervous system toxicity observed in patients with advanced cancer treated with FMAU restricted its further consideration for antiviral applications (Fanucchi et al., 1985). Severe encephalopathy occurred in patients treated with 64 mg/m^2/day for 5 days.

Synthesis of analogs of FMAU resulted in the selection of the ethyl analog 1-(2-deoxy-2-fluoro-1-β-D-arabinofuranosyl)-5-ethyluracil (FEAU) as a potential antiviral with a more favorable therapeutic index (Chou et al., 1987). While FEAU was slightly less active than FMAU in inhibiting herpes simplex virus infection in VERO cells and in mice, it was also much less toxic (Fox et al., 1985).

We are reporting the results of studies evaluating FEAU in Simian varicella virus infected African green monkeys in an attempt to compare its efficacy with FIAU and FMAU. We have previously reported combination of an effective antiviral [9-(1,3-dihydroxy-2-propoxymethyl)guanine – DHPG] and human recombinant interferon-β (Hu rIFNβ) which permitted the use of lower doses of both compounds (Soike, et al., 1987). In an attempt to demonstrate enhanced efficacy of FEAU we have evaluated reduced doses of FEAU in combination with varying doses of Hu rIFNβ for antiviral activity against Simian varicella virus infection.

Materials and Methods

Monkeys

African green monkeys (*Cercopithecus aethiops*) were purchased as feral animals and stabilized during a 90-day quarantine period before use. Each of the monkeys was determined to be free of antibody to Simian varicella virus by a serum neutralization assay performed prior to virus inoculation.

Simian varicella virus

The stock virus was a pool of VERO cells infected by co-cultivation with lymphocytes from a monkey infected with the Delta herpesvirus (Allen et al., 1974). The cultures were subsequently expanded by 5 passages as 1 to 3 dilutions of infected cells to fresh VERO cell cultures. The stock was prepared as a large pool of infected VERO cells sedimented by centrifugation and suspended in a solution of 0.2 M sucrose, 0.01 M NaH_2PO_4 and 1% bovine serum albumin and stored frozen at −70°C in 1 ml aliquots.

Antiviral drugs

FEAU was prepared at concentrations for appropriate doses by dissolution in phosphate buffered saline (PBS) and sterilized by filtration through a 0.22 μm filter for intravenous administration. FEAU for oral administration was not filtered. Solutions of FEAU were prepared daily for use.

Human recombinant interferon-β (Hu rIFNβ) was provided by Triton Biosciences, Alameda, CA, in vials containing 54×10^6 IU per vial, and dissolved in PBS. Dilutions to desired concentrations were prepared in PBS, pH 7.2, containing 0.5% human serum albumin. Hu rIFNβ was administered by bolus infusion into the saphenous vein.

Simian varicella virus infection

Monkeys were infected by inoculation of 1.5 ml of a dilution of stock virus by intratracheal catheter and by injection of 1.5 ml of this virus dilution subcutaneously. The intended dose was approximately 1×10^5 PFU of virus and the inoculum was titrated at the time of each experiment. Treatment with FEAU and/or Hu rIFNβ was begun 48 h after virus inoculation and was administered in divided doses twice daily at 8 a.m. and 2 pm. for 10 days.

Virus infection was monitored by observation of rash and by quantitation of viremia as previously described (Soike et al., 1986, 1987). Rash development was scored daily on a scale of 1+ to 4+ in relation to severity. Viremia was determined 3, 5, 7, 9 and 11 days after virus inoculation by separating lymphocytes from a 3 ml heparinized blood specimen on a Lymphoprep® gradient (Organon-Teknika, Durham, NC). The separated lymphocytes were washed twice in RPMI-1640 medium containing 15% fetal bovine serum, 100 units penicillin, 100 μg streptomycin and 50 μg fungizone/ml. Following the second washing the cells were suspended in 10 ml of this medium. The 10 ml lymphocyte suspension was divided between two 25 cm^2 culture flasks containing VERO cells. Following 5 to 7 days incubation at 37°C in a CO_2 incubator, the cell monolayers were washed with PBS, fixed in methanol and stained with methylene blue-basic fuchsin. Developing plaques in each culture flask were counted and the mean number of plaques in the two flasks inoculated with each blood specimen was determined. Hematology and clinical chemistry tests were performed at 0, 3, 5, 7, 9 and 11 days after virus inoculation on all monkeys.

Blood was drawn for determination of antibody titers to Simian varicella virus at 14 and 21 days after virus inoculation. Monkeys dying during the course of the infection were given a complete necropsy and death as a consequence of Simian varicella virus infection was confirmed by gross and histologic examination.

Combination experiments with FEAU and Hu rIFNβ

FEAU and Hu rIFNβ were tested alone and in combination at three dose levels for each drug. FEAU was administered at 1.0, 0.2, or 0.05 mg/kg/day by gavage

and Hu rIFNβ was administered at 5.0×10^4, 1.0×10^4 or 0.25×10^3 IU/kg/day by intravenous bolus. All treatments as single or combination doses were begun 48 h after virus inoculation and given as divided doses at 8 a.m. and 2 p.m. daily for 10 days. The combination treatments maintained a constant ratio between FEAU and Hu rIFNβ with 1.0 mg/kg/day of FEAU given along with 5.0×10^4 IU/kg/day of Hu rIFNβ. Accordingly the mid-dose of FEAU was administered with the mid-dose of Hu rIFNβ while the low dose of FEAU was given with the low dose of Hu rIFNβ. Each monkey receiving combination treatment received both drugs within a minute of each other with the oral dose of FEAU followed immediately by the intravenous injection of Hu rIFNβ.

Analysis of synergy

A computer program was used for analysis of the combined effects of the drugs using the median effect equation and determination of the combination index (CI) (Chou and Talalay, 1984; Chou and Chou, 1986). The analysis employed constant ratios of FEAU to Hu rIFNβ and used data from the viremia values for days 5, 7 and 9. The analysis is based on the median effect equation $F_a/F_u = (D/D_m)^m$ where F_a and F_u are the fractions of the system which are affected and unaffected, D is the dose, D_m is the dose which produces the median effect and m is the slope of the dose-effect plots. The combination indices (CI) are calculated based on the classical isobologram equation (compounds being mutually exclusive or similar modes of action) as well as the conservative isobologram equation (mutually non-exclusive or different modes of action). The CI values determined from the median–effect plots of <1, $=1$, or >1 indicate synergism, additivism, or antagonism, respectively.

Results

An initial experiment to determine possible efficacy of FEAU comprised two groups of two monkeys each of which received either 10 or 3 mg/kg/day by intravenous injection. A control group of three monkeys received intravenous injections of PBS. Treatment was administered in divided doses at 8 a.m. and 2 p.m. beginning 48 h after virus inoculation and continuing for 10 days. Each of the three control monkeys developed severe infection with rash and high titered viremia resulting in death from systemic infection (Table 1). FEAU at 10 or 3 mg/kg/day prevented the development of rash and effectively reduced viremia. Antibody to Simian varicella virus was detected at moderate titers in the surviving monkeys. Administration of FEAU at doses of 10 mg/kg/day by either intravenous or oral routes of administration for 10 consecutive days resulted in no obvious toxicity evidenced by clinical examination or by hematology and clinical chemistry tests performed during the period of treatment. Treatment of two monkeys with intravenous injections of FEAU at 30 mg/kg/day as divided doses was also not toxic although highly effective in preventing clinical Simian varicella infection (data not reported).

TABLE 1

Effect of intravenous administration of FEAU on the development of rash and viremia in African green monkeys infected with Simian varicella virus

FEAU[1] (mg/kg/day)	Monkey No.	Rash[2] – (Severity on days p.i.)								Viremia[3] – (Mean PFU on days p.i.)					Antibody titer[4]	
		6	7	8	9	10	11	12	14	3	5	7	9	11	14 days	21 days
Control	G029	1+	1+	2+	Dead					1	140	>400	Dead		Dead	Dead
	G030	–	1+	3+	3+	Dead				3	163	>400	>400	Dead	Dead	Dead
	G031	–	–	Dead						1	99	>400	Dead		Dead	Dead
10	G025	–	–	–	–	–	–	–	–	1	14	5	0	0	1:80	1:160
	G026	–	–	–	–	–	–	–	–	0	1	1	0	0	1:10	1:160
3	G027	–	–	–	–	–	–	–	–	1	8	0	0	0	1:40	1:160
	G025	–	–	–	–	–	–	–	–	0	1	1	0	0	1:20	1:160
Control	G250	–	1+	2+	3+	3+	2+	1+	±	0	34	68	29	0	1:160	1:160
	G252	–	–	–	±	2+	2+	Dead		2	25	>400	>400	>400	Dead	Dead
	G253	–	–	1+	2+	3+	2+	1+	–	1	11	107	36	1	1:320	1:640
1.0	G254	–	–	–	–	–	–	–	–	1	2	134	20	1	1:320	1:320
	G255	–	–	–	–	–	–	–	–	3	8	109	14	2	1:10	1:40
	G256	–	–	–	–	–	–	–	–	1	3	7	6	6	1:40	1:80
0.2	G258	–	1+	2+	2+	3+	1+	1+	±	2	8	16	36	1	1:160	1:320
	G259	–	±	±	±	2+	2+	1+	–	1	6	365	68	0	≥1:320	1:640
	G260	1+	2+	3+	3+	4+	4+	4+	2+	2	2	>400	159	2	1:160	1:640
0.04	G261	2+	4+	4+	4+	4+	Dead			3	3	>400	>400	Dead	Dead	Dead
	G262	–	1+	1+	2+	2+	±	–	–	4	53	92	46	2	1:320	≥1:640
	G263	–	±	1+	2+	2+	3+	3+	3+	12	44	>400	>400	2	1:160	≥1:640

[1]Administered as a bolus intravenous injection as divided doses twice daily beginning 48 h after virus inoculation and continuing for 10 days.
[2]Rash scored on a scale of 1+ to 4+ in relation to severity (note text).
[3]Viremia expressed as mean number of plaque forming units (PFU) developing in pairs of flasks of VERO cells inoculated with lymphocytes collected from 3 ml of heparinized blood (see text).
[4]The titer of antibody was the dilution of serum neutralizing 80% or more of plaques present in control cultures in a plaque reduction assay.

In a second experiment, groups of three monkeys received FEAU at 1.0, 0.2 or 0.04 mg/kg/day also given by intravenous injection as divided doses (Table 1). The three control monkeys developed moderate to moderately severe rash and moderate or severe viremia. One of the three control monkeys died with disseminated varicella. The two surviving monkeys showed good antibody titers. FEAU at 1.0 mg/kg/day prevented the development of rash but was only minimally effective in inhibiting viremia with two of the three monkeys shown to have moderate titers of virus in the blood. Antibody titers to Simian varicella virus were depressed in two of the monkeys. The lower doses of FEAU, 0.2 or 0.04 mg/kg/day, were ineffective and did not reduce either rash or viremia. One monkey at the lowest dose died of Simian varicella infection. Appreciable antibody titers were observed in each of the surviving monkeys.

Oral administration of FEAU was highly effective in protecting the monkeys from Simian varicella infection. Groups of three monkeys were treated with 0 (control), 10, 3, or 1 mg/kg/day with treatment by gavage at 8 a.m. and 2 p.m. begun 48 h after virus inoculation (Table 2). Two of the control monkeys had severe rash and the third had a moderate rash. Viremia was severe in one control monkey that died as a result of infection. The two remaining monkeys had a mild viremia. FEAU at each of the three doses completely prevented the appearance of rash. Viremia was also inhibited by FEAU at each of the three dose levels. Only one monkey at the lowest dose (1 mg/kg/day) showed a mild viremia.

Experiments to determine the effects of combination of FEAU with Hu rIFNβ were performed in a series of six experiments and the data pooled for analysis. Each treatment group receiving FEAU or Hu rIFNβ alone or the various combinations of FEAU and Hu rIFNβ contained four or five monkeys. The mean viremia values for each of these groups are presented in Table 3. High titers of virus appeared in the blood of the control monkeys on days 5, 7 and 9 after virus inoculations. FEAU administered alone at each of the three doses resulted in appreciable viremia on post-infection days 7 and 9. Similarly Hu rIFNβ given alone permitted substantial viremia on days 7 and 9. The combination of FEAU and Hu rIFNβ employing the lowest doses resulted in high titered viremia on days 5 and 7. The combinations at the higher doses were highly effective in reducing viremia on each of the days of assay.

Results of analysis of these data for synergy with determination of the combination index are reported in Table 4. The low doses of FEAU and Hu rIFNβ administered in combination did not result in synergy. Data obtained from analysis of the low dose monkeys on day 5 or day 9 essentially showed a moderately antagonistic effect with a CI of 1.53 and 1.34, while data from day 7 showed strong antagonism with a CI of 4.72. Analysis of viremia data from the mid- or high-dose combinations showed strong synergism (CI less than 1) especially on days 7 and 9. Results on day 5 indicated synergism which was less pronounced.

No adverse effects of the combination treatment were evident in daily clinical observations of the monkeys or in hematology and clinical chemistry tests performed during the entire course of the study. Similarly, neither FEAU or Hu rIFNβ had any adverse effects when administered alone.

TABLE 2

Effect of oral administration of FEAU on the development of rash and viremia in African green monkeys infected with Simian varicella virus

FEAU[1] (mg/kg/day)	Monkey No.	Rash[2] – (Severity on days p.i.)								Viremia[3] – (Mean PFU on days p.i.)				Antibody titer[4]	
		6	7	8	9	10	11	12	14	3	7	9	11	14 days	21 days
Control	F644	±	3+	4+	4+	Dead				38	>400	>400	Dead	Dead	Dead
	G668	–	–	±	2+	1+	±	±	–	8	32	29	25	1:320	1:640
	G604	–	±	3+	4+	4+	2+	1+	1+	24	16	39	34	≥1:320	≥1:640
10	G249	–	–	–	–	–	–	–	–	2	0	0	0	<1:10	1:20
	G267	–	–	–	–	–	–	–	–	5	1	1	0	1:80	1:160
	G264	–	–	–	–	–	–	–	–	15	1	3	5	1:40	1:80
3	G265	–	–	–	–	–	–	–	–	7	13	9	10	1:160	1:320
	G257	–	–	–	–	–	–	–	–	3	5	1	4	1:80	1:160
	G268	–	–	–	–	–	–	–	–	5	1	0	1	1:80	1:160
1	G269	–	–	–	–	–	–	–	–	0	0	2	0	1:80	1:80
	G270	–	–	–	–	–	–	–	–	0	2	2	3	1:160	1:320
	G274	–	–	–	–	–	–	–	–	7	47	7	7	1:160	1:320

[1]Administered orally as divided doses twice daily beginning 48 h after virus inoculation and continuing for 10 days.

[2]Rash scored on a scale of 1+ to 4+ in relation to severity (note text).

[3]Viremia expressed as mean number of plaque forming units (PFU) developing in pairs of flasks of VERO cells inoculated with lymphocytes collected from 3 ml of heparinized blood (see text).

[4]The titer of antibody was the dilution of serum neutralizing 80% or more of plaques present in control cultures in a plaque reduction assay.

TABLE 3

Viremia in mean plaque forming units (PFU) from blood of African green monkeys infected with Simian varicella virus and treated with FEAU and Hu rIFN-β

Combined treatment		Viremia – Mean PFU on days p.i.			
FEAU[1]	Hu rIFN-β[2]	Day 3	Day 5	Day 7	Day 9
0	0	10.4	111.4	710.0	589.1
0.05	0	2.7	23.3	383	261
0.2	0	11.2	95.8	295	197
1.0	0	55.3	40.8	256	251
0	2.5×10^3	34.3	199	370	561
0	1.0×10^4	10.8	136	169	200
0	5.0×10^4	6.6	28	611	41
0.05	2.5×10^3	20.3	131	394	25.3
0.2	1.0×10^4	3.6	36.6	32.3	3.2
1.0	5.0×10^4	2.1	4.3	0.5	0.5

[1]FEAU was administered orally by stomach tube as divided doses at 8 a.m. and 2 p.m. beginning 48 h after virus inoculation and continuing as divided daily doses for 10 days. Values are doses in mg/kg/day.
[2]Hu rIFN-β was given by intravenous bolus injection as divided doses at 8 a.m. and 2 p.m. beginning 48 h after virus inoculation. Values are doses in IU/kg/day.

Discussion

FEAU is a highly effective inhibitor of Simian varicella virus infection in the African green monkey. Intravenous treatment of Simian varicella virus-infected

TABLE 4

Combination indices (CI)[1] for combination of FEAU and Hu rIFN-β doses in the treatment of Simian varicella virus infection of African green monkeys

Combined treatment		Viremia					
FEAU[2]	Hu rIFN-β[3]	Day 5		Day 7		Day 9	
		Fa	CI	Fa	CI	Fa	CI
0.05	2.5×10^3	<0.02	1.28 (1.53)	0.445	4.72 (4.72)	0.571	1.12 (1.34)
0.2	1.0×10^4	0.672	0.404 (0.445)	0.955	0.125 (0.125)	0.995	0.035 (0.035)
1.0	5.0×10^4	0.962	0.664 (0.745)	0.999	0.006 (0.006)	0.999	0.049 (0.049)

[1]CI values <1, = 1, >1 indicate synergism, additivism and antagonism, respectively. Values given are based on classical isobologram equation and values in parentheses are based on conservative isobologram equation. Fa is the fractional inhibition which is percent inhibition/100.
[2]FEAU administered orally in mg/kg/day.
[3]Hu rIFN-β administered by intravenous injection in IU/kg/day.

monkeys with 3 mg/kg/day beginning 48 h after virus inoculation prevented development of rash as well as inhibition of viremia. A dose of 1 mg/kg/day was partially effective by preventing rash while having only a minimal effect on appearance of viremia. Oral administration of FEAU at 10, 3, or 1 mg/kg/day could protect monkeys from development of rash and appreciably reduce viremia. Activity of FEAU against Simian virus was comparable to that reported for FIAU but was less effective than FMAU (Soike et al., 1986). FIAU at 1 mg/kg/day p.o. prevented rash but permitted a moderate viremia while FMAU at 0.2 mg/kg/day inhibited both rash and viremia.

Similar levels of activity have been reported for the fluorinated pyrimidines in the treatment of herpes simplex virus infection in other animal model systems. Trousdale et al. (1983) employed topical treatment of rabbit eyes infected with the McKrae strain of HSV-1 with solutions of 0.02% FMAU. Corneal infection was reduced by treatment begun 4 h after virus inoculation which was administered 5 times daily for 11 days. Mice infected by intracerebral inoculation of HSV-1 (KOS strain) were protected from death by FIAC at 15 mg/kg/day and by FMAU at 0.5 mg/kg/day (Schinazi et al., 1983). Considerably higher doses of FIAC and FMAU were required to affect HSV type 2 infection in guinea pigs (Mayo and Hsuing, 1984). FIAC or FMAU doses of 100 mg/kg/day given intraperitoneally reduced mean lesion score but had little effect on virus titer. Doses of 50 mg/kg/day had no effect on the number of animals with lesions and no effect on lesion scores. Evaluation of FEAU in these animal model systems has not been reported.

The fluorinated pyrimidines are highly effective antivirals and would be expected to have great potential for the treatment of human herpesvirus infections. While FEAU has been reported to be less toxic than FIAU and FMAU methods to lower the effective dose of FEAU would provide an even more favorable therapeutic index. It therefore was of interest to determine whether the effective antiviral dose of FEAU might be further reduced by combination of FEAU with another antiviral active against Simian varicella. Previous studies with the nucleoside, DHPG, showed that low doses of Hu rIFNβ lowered the effective dose of DHPG tenfold. In experiments with varying doses of FEAU administered together with sub-effective doses of Hu rIFNβ, it was possible to demonstrate synergism with FEAU at a dose equal to one fifth of the effective dose when used alone.

FEAU is a highly effective antiviral compound against Simian varicella virus infection in African green monkeys. The active dose of FEAU is comparable to the dose of FIAU found to be effective in this same animal model infection (Soike et al., 1986). No toxicity was seen in monkeys treated with intravenous injections of FEAU at doses as high as 10 and 30 mg/kg/day. Monkeys treated with these doses appeared clinically healthy, ate well, and showed no abnormalities in hematology and clinical chemistry tests performed during their period of treatment. With low toxicity of FEAU also reported by Chou et al. (1987) in both in vitro and in vivo studies these results would suggest that FEAU has potential for treatment of human herpesvirus infections.

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References

Allen, W.P., Felsenfeld, A.D., Wolf, R.H. and Semtana, H.F. (1974) Recent studies in the isolation and characterization of Delta herpesvirus. Lab. Anim. Sci. 24, 222–228.

Chou, T.-C. and Talalay, P. (1984) Quantitative analysis of dose-effect relationships: the combined effects of multiple drugs or enzyme inhibitors. Adv. Enzyme Regul. 22, 27–55.

Chou, J. and Chou, T.-C. (1986) Dose-effect analysis with microcomputers: quantitation of ED50, LD50, synergism, antagonism, low-dose risk, receptor ligand binding and enzyme kinetics. IBM-PC Series. Elsevier-Biosoft, Elsevier Science Publishers, Cambridge, U.K.

Chou, T.-C., Kong, X.-B., Fanucchi, M.P., Cheng, Y.-C., Takahashi, K., Watanabe, K.A. and Fox, J.J. (1987) Synthesis and biological effects of 2-fluoro-5-ethyl-1-β-D-arabinofuranosyluracil. Antimicrob. Agents Chemother. 31, 1355–1358.

Fanucchi, M.P., Leylund-Jones, B., Young, C.W., Burchenal, J.H., Watanabe, K.A. and Fox, J.J. (1985) Phase I trial of 1-(2′-deoxy-2′-fluoro-1-β-D-arabinofuranosyl)-5-methyl uracil (FMAU). Cancer Treat. Rep. 69, 55–59.

Fox, J.J., Watanabe, K.A., Schinazi, R.F. and Lopez, C. (1985) Antiviral activities of some newer 2′-fluoro-5-substituted arabinosyl pyrimidine nucleosides. In: Herpes Viruses and Virus Chemotherapy, pp. 53–56. Elsevier Science Publishing, Inc., New York.

Mayo, D.R. and Hsuing, G.D. (1984) Treatment of primary acute genital herpes in guinea pigs by intraperitoneal administration of fluoropyrimidines. Antimicrob. Agents Chemother. 26, 354–357.

Schinazi, R.F., Peters, J., Sokol, M.K. and Nahmias, A.J. (1983) Therapeutic activities of 1-(2-fluoro-2-deoxy-β-D-arabinofuranosyl)-5-Iodocytosine and thymine alone and in combination with acyclovir and vidarabine in mice infected intracerebrally with herpes simplex virus. Antimicrob. Agents Chemother. 24, 95–103.

Soike, K.F., Cantrell, C. and Gerone, P.J. (1986) Activity of 1-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-5-iodouracil against simian varicella virus infections in African green monkeys. Antimicrob. Agents Chemother. 29, 20–25.

Soike, K.F., Eppstein, D.A., Gloff, C.A., Cantrell, C., Chou, T.-C. and Gerone, P.J. (1987) Effect of 9-(1,3-dihydroxy-2-propoxymethyl) guanine and recombinant human β interferon alone and in combination on simian varicella virus infection in monkeys. J. Infect. Dis. 156, 607–614.

Trousdale, M.D., Nesburn, A.B., Su, T.-L., Lopez, C., Watanabe, K.A. and Fox, J.J. (1983) Activity of 1-(2′-fluoro-2′-deoxy-β-D-arabinofuranosyl) thymine against herpes simplex virus in cell cultures and rabbit eyes. Antimicrob. Agents Chemother. 23, 808–813.