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Short Communication

Drug testing for activity against varicella-zoster virus in hairless guinea pigs

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Summary

Inoculation of congenitally hairless guinea pigs with varicella-zoster virus (VZV) (Oka strain) results in a self-limited exanthematous infection analogous to varicella in children. Administration of acyclovir or 6-methoxypurine arabinoside modified the course of infection. This model should facilitate pre-clinical testing of putative anti-VZV agents

Varicella, Antiviral testing; Animal model; Guinea pig; Acyclovir; 6-Methoxypurine arabinoside

Because varicella-zoster virus (VZV) has an extremely limited host range, much of what we understand of the pathophysiology of infection has been deduced from epidemiologic studies and by analogy to herpes simplex virus (HSV) (Myers et al., 1987) In the absence of a small animal model, development and preclinical testing of candidate antiviral drugs have been limited to in vitro testing and studies in primates employing a simian varicella virus (Soike et al , 1981) Studies with non-human primates are expensive and, because they do not utilize the human virus, may not accurately predict the efficacy of a drug in the treatment of VZV infection

To facilitate testing of antiviral agents, we have developed a model of varicella utilizing congenitally hairless Hartley guinea pigs and VZV adapted to guinea pigs

Note Animals used in this study were maintained in accordance with the guidelines of the Institutional Animal Care and Use Committee of the Children's Hospital Research Foundation

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TABLE 1

Effects of antivirals on expression of VZV exanthem

Drug[a]	Number[b] of animals	Number with exanthem	Onset of exanthem	Duration of exanthem[c]
Placebo	20	17	4 6 ± 0 3	2 6 ± 0 3
Acyclovir	19	9[d]	4 0 ± 0 4	2 8 ± 0 5
Ara-M	20	10[d]	3 5 ± 0 3	3 4 ± 0 7

[a]Administered sc three times/day for 10 days, beginning 4 hours after inoculation
[b]H_o/H_A guinea pigs inoculated im with $7\ 5 \times 10^4$ PFU VZV, Oka strain
[c]Mean days ± SEM
[d]Different from placebo treated animals $P < 0\ 02$ by chi square

cells in vitro (Myers et al , 1991) Guinea pigs inoculated with VZV undergo a self-limited viremic infection, developing both humoral and cell-mediated immune responses (Jenski and Myers, 1987, Matsunaga et al , 1982, Myers et al , 1980, 1985) Congenitally hairless animals develop an erythematous papular exanthem, the frequency of which can be reduced with passively administered antibody given prophylactically (Myers et al , 1991). In the studies reported here we sought to utilize exanthem reduction as a measure of antiviral effectiveness in vivo

VZV, Oka strain (ATCC No 795), was further passaged in human fibroblasts four times IAF/H_A-H_o Hartley guinea pigs weighing approximately 425 g (Charles River Breeding Laboratories, Wilmington, MA) were inoculated by the intramuscular route with $7\ 5 \times 10^4$ PFU and examined daily for exanthem Animals were bled from the heart for serum before infection and on day 28 VZV antibody was measured by ELISA as previously described (Myers et al , 1991). Sixty hairless guinea pigs were randomized into three treatment groups to receive drug three times a day for ten days Examination daily was performed by an observer blinded as to treatment The three treatment groups consisted of placebo (5 ml/kg/dose), acyclovir (20 mg/kg/dose as 2 ml/kg/dose) and 6-methoxypurine arabinoside (ara-M) (50 mg/kg/dose as 5 ml/kg/dose) administered subcutaneously beginning 4 h after infection Ara-M is an experimental anti-VZV compound developed by the Burroughs Wellcome/Company (Averett et al , 1991) Drugs were provided encoded by the Wellcome Research Laboratories (Research Triangle Park, NC) In vitro evaluation revealed an ID_{50} against VZV, Oka strain, in human foreskin fibroblasts of 5.4 μM acyclovir and 0 22 μM ara-M (Stanberry and Myers, 1988)

Both acyclovir ($P = 0\ 013$) and ara-M ($P = 0\ 017$) reduced the frequency of exanthem compared to placebo treated animals (Table 1). Animals receiving acyclovir developed inflammation at the site of drug administration. The onset and duration of exanthem in animals that developed exanthem was similar for all study groups All animals seroconverted to VZV There were no differences in the mean ± SD ELISA titers for placebo and acyclovir treated groups (0 417 ± 0 163 and 0 373 ± 0 115, respectively) However, the ELISA titer for animals treated with ara-M (0 306 ± 0 108) was less then for the placebo ($P = 0\ 03$ by Student's t-test) The reduced VZV antibody response seen in ara-M treated animals is similar to the reduction of HSV antibody response seen in humans and guinea pigs with genital

HSV infection treated with acyclovir (Bernstein et al., 1984, 1986) The reduced VZV antibody response by this nucleoside analog could be a consequence of a direct immunosuppressive effect of the drug or, more likely, an effect on the magnitude of viral replication There was no difference in ELISA titers between animals that did or did not develop exanthem

Although a self-limited childhood illness, varicella may be a devastating infection in the adult or immunocompromised host (Myers, 1977). In the otherwise healthy child, varicella may also cause significant morbidity and, uncommonly, mortality (Preblud, 1986). Herpes zoster, a consequence of reactivation of latent VZV, occurs commonly among the immunocompromised and the elderly with the potential for dissemination and post-herpetic neuralgia (Myers, 1977). Thus, the need for treatment of some patients with VZV infections has promoted evaluation of compounds with potential anti-VZV activity. Prophylactic treatment of hairless guinea pigs with 28-day VZV-convalescent guinea pig serum has previously been shown to reduce the frequency of exanthem in animals (Myers et al., 1991) as does zoster immune plasma when administered to children (Geiser et al., 1975). In these studies we have demonstrated that acyclovir, which has previously been shown to ameliorate varicella in children (Balfour et al , 1990) also can modify exanthem in euthymic, hairless guinea pigs. Further, an additional candidate antiviral drug, ara-M, was shown to be as effective as acyclovir in reduction of exanthem in this model system This caviid model of varicella should facilitate preclinical studies of candidate anti-VZV agents

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