and mass spectra identical with those of an authentic sample. The above findings are compatible only with the presence of a free phenolic hydroxyl and the attachment of the succinoyl moiety via the alcoholic hydroxyl group at C-6.

The above conclusion was confirmed by the preparation of the methyl ester methyl ether of **1** from codeine as follows. Succinoylation of codeine with succinic anhydride in refluxing pyridine for 3 hours furnished codeine 6-hemisuccinate, which after purification by precipitation of its methanol solution with excess ether melted at 165° to 170°C. On methylation with diazomethane this latter acid was converted to a methyl ester, which proved identical with the methyl ester methyl ether of **1** by comparison of their infrared and nuclear magnetic resonance spectra, and their mobilities on thin-layer chromatography.

Simon, Dole, and Hiller (*2*) have described a succinoyl derivative of morphine by a procedure different from ours, which they likewise showed to be morphine 6-hemisuccinate.

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Gene Therapy for Human Genetic Disease?

In contrast to Friedmann and Roblin, who state, "Storage diseases associated with lysosomal enzyme deficiencies do not appear to respond to enzyme therapy" (*1*), we believe that there is considerable hope for treatment of some lysosomal enzyme deficiencies by this means. This is so because proteins can be transported into cells through the process of pinocytosis, followed by a merger of the pinocytic vesicles with lysosomes and a mixing of their contents. Therapeutic replacement of deficient enzymes with plasma, leukocytes, or through transplanted kidneys (rather than with purified proteins which are not presently available) has already shown promise in those systemic storage disorders in which the affected cells have marked pinocytic activity (*2*).

Let us examine the two cases responsible for the pessimistic outlook of Friedmann and Roblin. In one instance (*3*), the disease selected for treatment, metachromatic leukodystrophy, may indeed be among those not amenable to enzyme replacement. In this as in other neurological disorders, the blood-brain barrier may prevent the circulating enzyme from reaching the cells of the brain.

In the second instance (*4*), plasma was infused into a patient with Fabry's disease. The purpose of the test was to monitor the relevant enzyme, ceramide trihexosidase, after it entered the patient's circulation. The quantity of enzyme was far too small to expect a notable therapeutic benefit, but could not be increased because of the deleterious effect of administering large amounts of plasma to Fabry patients. Clearly, the experiment gives no information on the therapeutic potential of purified enzyme supplied in appropriate dosage.

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Friedmann and Roblin (*1*) have been critical of the efforts of therapy for two argininemics. We are in agreement with Friedmann and Roblin concerning what must be done to satisfy any prospective techniques for gene therapy, and indeed have more than met the set criteria, as they apply to the Shope virus.

As to the criterion of prior experience with the disease, the patients were studied jointly and intensively by three groups of investigators, both from the point of view of genetics and biochemistry (*2*). The three children affected by this disease are from the same family and are of widely differing ages. The progress of the deterioration of the disease has been identical in the two older children; the third is a baby and has undergone little change as yet.

The virus used was purified by successive sedimentation in the preparative centrifuge, and subsequent separation in cesium chloride gradients and rate density gradients. It was homogeneous, as judged by electron microscopy, and produced no harmful effects on repeated blind passages in tissue culture. The virus was tested immunologically for possible contamination by other viruses, and none was found. Ultimately it was filtered through Millipore filters.

The virus has been studied for 40 years without having produced any known harmful effects on any investigator, including one who inoculated himself in 1933. Massive doses have been given to many kinds of animals without discernible effect except for, as pointed out, the decrease in blood arginine concentration. Inoculated into the skin of wild or domestic rabbits, it produces warts. The wild-type virus is not propagable in the domestic rabbit and produces no change in any tissue other than squamous epithelium. If massive doses are given the rabbit intravenously, no warts appear; only a low blood arginine develops.

The most direct evidence that the arginase induced is virus information, since the mutation of the virus is associated with a change in the structure of the enzyme, was published in 1971 (*3*). Although this evidence had been available in abstract for several years (*4*), the complete data were not available to the authors in time for inclusion in their article.

The use of the virus was said to have been perhaps premature; nevertheless, when one has a patient with a progressively deteriorating disease that is known not to respond to dietary or

other known measures, and one has a possible means of stopping the progression of the disease with an agent that has been extensively investigated for 40 years, there appears to be little alternative other than to try it.

Critics of gene therapy seem to forget that, when live viruses are used to immunize against a wide variety of diseases, virus genes are being inserted into man. The use of the Shope virus is different only in that we are taking advantage of this possibility. Another advantage of the Shope preparations used is their purity, unlike most live virus preparations given for immunological purposes. Also, man is continually being exposed and infected by a host of pathogenic and nonpathogenic viruses. This raises the additional question as to what a massive dose of virus is, as the total amount of virus present in the body in an active disease such as measles must be truly large. The Shope virions are known not to be completed to any significant degree in any animal other than the wild cottontail rabbit in Kansas or in the same cottontail rabbit brought to the laboratory and fed food raised in Kansas. It appears, then, to be a rather ideal passenger virus to which desired genetic information may be added in the future for further efforts in gene therapy.

The only other point relating to our work was that tissue cultured cells from the patients should be used to test out the virus in vitro. That arginase was induced with the Shope virus in fibroblasts from an argininemic came as quite a pleasant surprise, as the virus was not known to go in culture. No detectable virions have been produced (*5*).

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Largely as a result of the pioneering work of Neufeld and her colleagues, there are now several reports that, in tissue culture cells derived from patients with lysosomal storage diseases, beneficial phenotypic effects can be brought about by means of enzymic factors elaborated by other cells (*1*) or supplied in culture medium in a partially purified form (*2*). This important phenomenon of course suggests that similar corrective effects may occur in patients with some lysosomal storage diseases (*3*). However, we feel that important problems exist in ensuring optimum uptake into cells, such as neurons, where the enzyme is critically needed and not only into macrophages and cells of the reticuloendothelial system; in overcoming the blood-brain barrier where target cells are in the central nervous system; and in eliminating the possibility of immunological reaction to the administered enzyme preparation. We agree that further trials with purified enzyme preparations will be required to evaluate further the potential for enzyme therapy in lysosomal or other disorders.

Rogers indicates the many precautions that were taken before injection of Shope papilloma virus into the hyperargininemic patients. In our article, we did not intend to imply that the virus was administered without precautions. However, we still find ourselves in disagreement with Rogers on several points:

1) We believe that a major premise underlying the scientific rationale for the use of Shope virus, that is, that there is a viral gene which codes for a virus-specific arginase, is still questionable. Rogers cites his new evidence in support of this premise but omits mention of work by Orth and his colleagues (*4*) which shows that the Shope papilloma arginase has kinetic, molecular, and antigenic properties identical to those of the rabbit liver enzyme.

2) Rogers' comparison of live attenuated virus immunization with virus-mediated gene therapy strikes us as an unfair one. Surely there is a difference between injection of a nononcogenic, nonintegrating virus with the intent to stimulate a patient's antibody production and injection of a virus which is oncogenic under some conditions, which may be able to integrate its viral DNA into the DNA of the patient's cells, with the *intent* to alter permanently the patient's genetic constitution, albeit, hopefully, in a beneficial way.

3) The initial injections of the virus in the children were made almost 2 years ago and, at least to our knowledge, there have been no reports published in medical or scientific journals describing criteria for virus therapy, preliminary studies in vitro on the patients' cells, and even biochemical and clinical effects of the therapy. We feel that this kind of information would be helpful in evaluating the use of this agent.

4) Finally, the use by Rogers of the word wart tends to obscure the fact that the Shope virus is indeed an oncogenic virus, and it should be remembered that variable proportions of virus-induced papillomas in both domestic rabbits and cottontail rabbits develop into invasive malignant skin cancers (*5*).

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Origin of the Martian Chaotic Terrains

From considerations of atmospheric escape processes, McElroy (*1*) has estimated the rate of degassing of water from the martian surface relative to that for the earth. If one accepts his data, certain restrictions can be imposed on the theory that the martian "chaotic terrains" (rough, uncratered terrains, apparently caused by vertical subsidence) were produced by withdrawal of permafrost.

McElroy calculated that the ratio of the amount of water that has escaped from Mars over its geologic history to the amount of carbon dioxide currently present in the martian atmosphere is approximately 45 (when the quantities of H_2O and CO_2 are expressed in mole-