

patients with Chronic Active Hepatitis B caused by the hepatitis B virus (HBV), patients treated for a year with THN α_1 (5 patients) or with TF-5 (2 patients) showed a marked decrease in serum ALT; 6 of the 7 patients also showed reduced levels of serum HBV DNA, and 5 of 6 patients initially positive for se-rum hepatitis B surface antigen (HBsAg) subsequently cleared this antigen. Mutchnick, M. C., et al., *Hepatology*, 10:Abs. 575 (1989). No suggestion was made in these abstracts that the thymosins would be effective against any other hepatitis viruses.

[0017] There remains, therefore, an important need in the art for a new modality for the treatment of HCV infections in mammals, this modality is disclosed below.

SUMMARY OF THE INVENTION

[0018] A treatment modality for HCV infections has been devised comprising the administration to mammals of immune system-potentiating doses of one or more thymosins in combination with interferon therapy.

[0019] It is thus an object of this specification to disclose compositions and methods for the treatment of acute or chronic HCV infections in mammals comprising combination therapy with one or more thymosins and one or more interferons.

[0020] This and other objects will become apparent by reference to the specification and to the appended claims.

DESCRIPTION OF THE INVENTION

[0021] A novel modality for treating HCV infection in mammals has been devised, comprising the administration to such mammals of one or more thymosins at doses which potentiate immune responses, in combination with anti-viral doses of one or more interferons.

[0022] By the term "thymosins" is meant any or all of the immune system potentiating polypeptides naturally occurring in the thymus gland or produced by chemical or recombinant means, or fragments derived from any of these polypeptides. By the term "mammals" is meant any mammalian subject, including human and animal patients, requiring treatment for hepatitis C infection. "Mammal" and "subject" are used interchangeably.

[0023] Thymosin preparations suitable for treating HCV infections include TF-5, THN α_1 , and fragments thereof, e.g., C-terminal 4-28 and 15-28, and N-terminal 1-8, 1-14 and 1-20 fragments. These may be obtained from Alpha-1 Biomedicals Inc., Foster City, Calif.

[0024] Subjects, e.g., human patients, may receive the thymosin by subcutaneous injection or infusion, at appropriate intervals for an appropriate period of time. The thymosin is administered to mammals infected with hepatitis C virus in amounts which facilitate or promote in vivo inactivation of hepatitis C virus. A pharmaceutical dosage unit of an immune system-potentiating amount of a thymosin, such as TF-5, can be from about 900 to about 1200 mg/m² body surface area in a pharmaceutically acceptable carrier. A pharmaceutical dosage unit of an immune system-potentiating amount of a thymosin, such as THN α_1 or immune system-potentiating fragments thereof, can be from about 900 to about 1200 μ g/m² body surface area in a pharmaceutically-acceptable carrier. Lyophilized prepara-

tions of thymosins or fragments which contain mannitol and phosphate buffer are dissolved in diluent period to dispensing. Thymosins in diluent should remain stable for at least six months when stored in a refrigerator. It is convenient to dispense thymosin solutions in one ml dose vials per month.

[0025] For a typical human patient, an administration regimen of twice weekly (e.g., Monday and Thursday) subcutaneous injection of about 1500 to about 1700 μ g of THN α_1 or fragments therefrom is convenient. Dosages and length of treatment can be flexible, and can be determined by the subject's clinical response to the thymosins.

[0026] The course of the disease and its response to drug treatments may be followed by clinical examination and laboratory findings. As elevated serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are known to occur in uncontrolled hepatitis c, and as a complete response to treatment is generally defined as the normalization of these serum enzymes, particularly ALT (Davis, G. L., et al., *New England Journal of Medicine*, 321:1501-6 (1989.)), progress of treatment with thymosins is conveniently followed by this art-recognized test performed, e.g., on a sequential multiple analyzer.

[0027] Another means of evaluating subjects having antibodies to HCV (not all subjects with hepatitis C have detectable antibody to HCV—Weiner, A. J., et al., *Lancet*, 335:1-3 (1990)) is to periodically test subjects' sera for the titer of these antibodies. Anti-HCV antibodies may be tested by the currently available C 100-3 test (Kuo, G., et al., *Science*, 244:362-4 (1989)), by an Elisa test (Ortho Diagnostic Systems, Raritan, N.J.) or by a recombinant assay (RIBA-1 and RIBA-2, Chiron Corporation, Emeryville, Calif.). Any suitable test may be used.

[0028] In order to follow the course of HCV replication in subjects in response to drug treatment, HCV RNA may be measured in serum samples by, for example, a nested polymerase chain reaction assay that uses two sets of primers derived from the NS3 and NS4 non-structural gene regions of the HCV genome. Farci, P., et al., *New England Journal of Medicine*, 325:98-104 (1991); Ulrich, P. P., et al., *J. Clin. Invest.*, 86:1609-14 (1990).

[0029] Other appropriate laboratory tests to follow the course of treatment are listed in Example 1 below.

[0030] thymosin therapy is preferably used in combination with interferon therapy, thereby combining the immune system potentiating effect of thymosins with the anti-viral effects of the interferons. An improved response rate at the currently used interferon doses would be beneficial particularly in the light of dose-limiting side effects at higher doses of these proteins. An offshoot of this concept is the ability to achieve comparable efficacy with interferon plus thymosin at lower doses than would be required with interferon alone.

[0031] In this combination therapy regimen, one or more interferons (for example, recombinant interferon α -2b, intron-A, Schering-Plough, Kenilworth, N.J.) is (are) administered subcutaneously to subjects, e.g., human patients, at doses ranging between about 1 MU and 3 MU along with or sequentially with one or more thymosins, preferably including THN α_1 , at a dose of about 900 to about 1200 μ g/m² body surface area.

[0032] Although the example above speaks in terms of recombinant interferon α -2b, other anti-HCV-effective inter-