ESTABLISHED STRAINS OF TRANSGENIC MICE EXPRESSING ANTISENSE RNA AGAINST MOUSE HEPATITIS VIRUS N PROTEIN mRNA

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Antisense RNA can bind in a highly specific manner to complementary sequences, blocking the ability of the bound RNA to be processed or translated. Therefore, this technology has the potential to define the function of a gene product by specifically inhibiting its expression and also could be used to terminate viral or cancer genes yielding the prospect of novel therapies. This study is presented as a strategy against viral infections diseases in livestock and laboratory animals.

Mouse hepatitis virus (MHV), a member of the coronavirus group is associated with acute and chronic diseases in the laboratory mouse. MHV infection occurs at high prevalence in commercial and research colonies of laboratory mice throughout the world. The genome of MHV is a single plus (+) strand RNA of approximately 30kb and complexes with nucleocapsid (N) protein that is encoded by mRNA7 of MHV to form the helical nucleocapsid.

In this study, the transgenic mice strains that express antisense RNA against MHV N proteins mRNA were established, since the N protein may play an important role in the regulation of viral replication and transcription. These transgenic mice are expected to be resistant to MHV infection. A transgene, 2AI, was constructed to express MHV mRNA in reverse orientation directed by the long terminal repeat (LTR) of Rous sarcoma virus (RSV). LTR sequences of RSV contain the strong enhancer and promoter sequence for the gene in eukaryote cells and are anticipated to express highly the directed gene in various tissues of transgenic mice.

2AI was introduced into mouse eggs by microinjection into a male pronucleus after fertilization. The integration of 2AI into the genomic DNA of transgenic mice was analyzed by PCR and Southern blot hybridization. Of five 2AI integrated founder mice, four stably transmitted 2AI to their offspring. Recently two independent strains of transgenic mice were established and antisense mRNA7 was detected in the brain, lung, heart, spleen, liver, kidney and muscle in adult animals of these strains by reverse transcriptase (RT)-PCR. The author is at present examining viral strains, routes of infection and ages of mice for experimental infection.