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Prevention from naturally acquired viral respiratory infection by interferon nasal spray

Hitoshi Saito, Hiroshi Takenaka, Sachio Yoshida, Toshihito Tsubokawa, Akira Ogata, Fukui and Jiro Imanishi, Kyoto, Japan

SUMMARY

Human leukocyte interferon (IFN-α), 50,000 IU per day, was sprayed into the nasal cavity of 73 volunteers twice a day from January 9 till March 4, 1984. The rise in complement fixation antibody titers against influenza A virus was not significantly different between the interferon group and the placebo group. However, the number of subjects without elevated antibody titers and without symptoms in the interferon group was significantly higher than that in the placebo group ($p < 0.05$). Prophylactic nasal spray of IFN-α seems to protect against upper respiratory viral infection.

INTRODUCTION

There have been only a few double-blind studies concerning the preventive effects of interferon (IFN) on upper respiratory viral infection (Solov'ev, 1968; Panusarn et al., 1974; Imanishi et al., 1980). Furthermore, those findings failed to conclusively demonstrate a prophylactic effect of IFN against naturally acquired infection of influenza A virus, especially inhibition of antibody titers. Therefore, a new double-blind study was carried out.

SUBJECTS

Seventy-three healthy students from a nursing school volunteered to participate in this study. They consisted of 69 females and 4 males and ranged in age from 18 to 32 years (mean age 21 years). The IFN was given to 37 subjects and the placebo to 36. The subjects were divided on the basis of key codes randomized by Prof. Ogata, Department of Environmental Health. They were required to record on a form the administration, body temperature and subjective symptoms of common cold such as nasal, pharyngeal and bronchial symptoms.

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Administration of IFN and placebo

Purified human interferon-alpha produced by Japan Red Cross was administered as a spray. The dose was adjusted to 1,000,000 IU/9.5 ml (80 sprays on an average) with saline. One spray contained approximately 12,500 IU of IFN in 0.12 ml. One spray was administered into each nasal cavity in the morning and evening, for a total daily dose of 50,000 IU. As a placebo, human degammaglobulined plasma was diluted to the same protein concentration as the IFN preparation. As it was found that IFN activity did not decrease after refrigeration for two weeks, the spray container was filled with new IFN every two weeks. The study was conducted for eight weeks from January 9 till March 4, 1984. The total maximum amount of IFN administered was 2,800,000 IU.

Antibody titration

Blood was drawn before, during and after the experiment. Sera were stored at $-80\,^{\circ}C$ until assay. Serum antibody titers were measured by single radial complement fixation (CF) method. CF titers were calculated by comparing the ring formed with a standard curve. Microbes examined in this study were influenza virus A, B, respiratory synsytial virus (RS) and mycoplasma pneumoniae.

Exclusion from analysis

The following subjects were excluded from analysis:

1. those suffering from a common cold within three days after entering the study;
2. those having an abnormal rise in antibody titers before this study;
3. those whose rates of the nasal instillation were under 50%.

RESULTS

Rate of nasal instillation

On the basis of the exclusion criteria, eight subjects from each group were excluded. In addition, some of the remaining 57 subjects failed to comply with the dosing protocol. The compliance rate in the IFN group ($n = 29$) was $75.2 \pm 15.1\%$ (mean $\pm$ SD), and in the placebo group ($n = 28$) $67.8 \pm 14.0\%$. However, these differences were not statistically significant.

Table 1. Rise in complement fixation (CF) antibody titers.

viruses	CF≧8	CF≧16	paired	significant rise (%)
influenza A	43 cases	14	16	30 (41%)
influenza B	4	5	0	5 (7%)
RS	20	3	0	3 (4%)
mycoplasma	0	0	0	0 (0%)

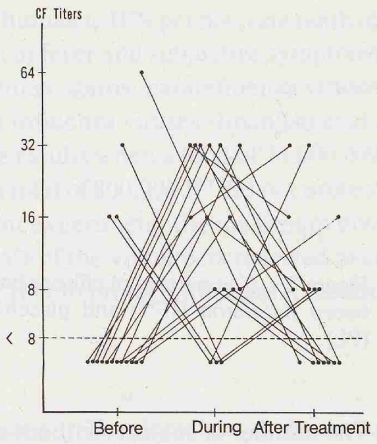

Figure 1. Changes of influenza A antibodies in the paired sera.

Complement fixation antibody titers

The rise in CF antibody titers during the double-blind test is summarized in Table 1. Significant rises of more than 16 were found in 41% of those with influenza A, 7% with influenza B and 4% with RS. Changes of influenza A antibodies in the paired sera are shown in Figure 1. These changes indicate that an influenza A virus epidemic occurred during this study.

Fever and subjective symptoms

Pertinent data were obtained from only 25 of the 73 volunteers after application of our criteria. Twelve of them were given IFN and 13 the placebo. Fever over 37 °C was observed in 33% (4 out of 12) in the IFN group,and 62% (8 out of 13) in the placebo group. However, this difference was not statistically significant because of the small number of the cases.

The duration of subjective symptoms associated with a common cold was also compared. Pertinent data were obtained from 24 volunteers. The symptoms continued for more than 4 days in 46% in the IFN group (6 out of 13), 64% in the placebo group (7 out of 11). This difference also was not significant but subjective symptoms appeared to be reduced in the IFN group.

Antibody titers

Significant rises in CF of more than 16 were predominately seen in those with influenza A. Therefore, the antibody titers of these subjects were compared between the IFN and placebo groups. In the IFN group, the antibody titers were elevated in 15 out of 26 subjects, and in the placebo group in 18 out of 26. No significant inhibition by IFN was observed (Figure 2).

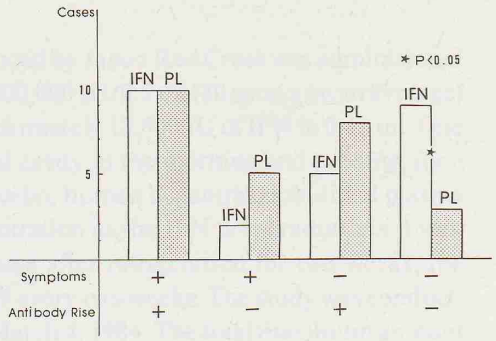

Figure 2. Comparison of efficacy between interferon (IFN) and placebo (PL).

The number of subjects without elevated antibody titers and symptoms, indicating absence of a cold, in the IFN group was significantly ($p < 0.05$) larger than that in the placebo group, as assessed by a Fisher's exact test (Figure 2, Table 2).

Table 2. Comparison of the number completely free from catching cold.

	interferon	placebo
no symptoms and no antibody rise	9*	3
others	17	23
total	26	26

* $p < 0.05$ by a Fisher's exact test.

DISCUSSION

Several double-blind tests have been conducted on the effect of IFN on respiratory infection, but some of them were experimental. Infection and clinical prophylactic effects of the agent were only partially demonstrated. Therefore, a new double-blind test was carried out. However, we observed a statistically significant preventive effect of IFN in only one item. The reason for this was the small number of subjects available for analyses.

Another important problem was dose compliance. In this study, 16 volunteers were excluded because of a low compliance rate of less than 50%. On the other hand, only three subjects received the target dose of 2,800,000 IU of IFN during the eight weeks. Administration between 100% and 90% in the eight weeks was noted in eight subjects. Therefore, volunteers administered over 90% were only 11 out of 73 (15%). Even among those whose compliance rate was more than 50%, timing of the nasal spray is important. It has been reported that the best protective action of IFN was found when the agent was given 24 h and 2 h before infection. Therefore, even those receiving IFN will be infected when the dose is not properly given 24 h before infection.

A previous double-blind test using 5,000 IU of human α-IFN per day, one tenth of the present dose, revealed a marked reduction in fever and subjective symptoms by the IFN, and that the rise in HI antibody titers against parainfluenza viruses were significantly suppressed, but not against influenza viruses (Imanishi et al., 1980). Merigan et al. (1973) obtained effective results when a total of 14,000,000 IU of human α-IFN was administered, while a total of 800,000 IU did not protect against influenza virus infection. In the present experiment, the maximum dosage of IFN was set at 2,800,000 IU but only 15% of the volunteers received over 2,520,000 IU. Therefore, the proper dosage of IFN to protect influenza infection is not clear.

RÉSUMÉ

Interféron leucocyte humain (IFN-α) a été testé sur 73 volontaires, du 9 janvier au 4 mars 1984; 50.000 U.I. par jour étant vaporisées deux fois par jour, dans la cavité nasale. En augmentation de titres d'anticorps de fixation complémentaire contre le virus A de la grippe, la différence entre le groupe interféron et le groupe placebo n'était pas significative. Cependant, le nombre de sujets sans anticorps augmentés et sans symptômes était significativement plus élevé dans le groupe interféron que dans le groupe placebo ($p < 0{,}05$). Une vaporisation prophylactique d'IFN-α semble protéger contre l'infection virale de la voie respiratoire supérieure.

REFERENCES

1. Imanishi J et al. The preventive effect of human interferon-alpha preparation on upper respiratory disease. J Interferon Res 1980; 1:169–78.
2. Merigan TC et al. Inhibition of respiratory virus infection by locally applied interferon. Lancet 1973; 17 March: 563–7.
3. Panusarn C et al. Prevention of illness from rhinovirus infection by a topical interferon inducer. N Engl J Med 1974; 291:57–61.
4. Solov'ev VD. Some results and prospects in the study of endogenous and exogenous interferon. In: The Interferon. New York: Academic Press, 1968; 233.

Hitoshi Saito, M.D.
Professor and Chairman
Department of Otolaryngology
Fukui Medical School
Matsuokacho, Yoshidagun,
Fukui 910-11, Japan