Introduction

Interferon (IFN) was discovered in the 1950’s during research on viral interference, and its antitumor and other effects were reported from the 1960’s. During the 1970’s, attention was paid to IFN as an anti-cancer drug because its anti-tumor effect was reported in clinical studies. Now, it is clinically used to treat Type C hepatitis, multiple sclerosis, and various tumors including renal tumor, malignant melanoma, and brain tumor.

IFN is classified by its properties into 3 types: IFN-α, IFN-β, and IFN-γ. IFN-α and IFN-β code common gene loci and have common cellular surface receptors, while IFN-γ has different dynamics. Therefore, the former and latter are called Types I and II IFN, respectively. In the clinical application of IFN for brain tumor, IFN-α is mainly used in Western countries, while IFN-β was approved by the Ministry of Health, Labor, and Welfare and has been clinically used in Japan.

This paper describes the history of the clinical application of different types of IFN for brain tumor, current issues, and prospects for...
new therapeutic techniques.

Interferon Single Therapy

In studies of the antitumor effect of IFN-\(\alpha\) and IFN-\(\beta\) using brain tumor cells, Lundblad\(^5\) and Wakabayashi\(^7\) reported a direct inhibitory effect on brain tumor, and Otsuka et al.\(^9\) reported an indirect inhibitory effect through immunocompetent cells. Nakagawa and Ueda reported the clinical effect of IFN-\(\alpha\) on malignant brain tumor: they achieved partial response in patients with glioblastoma and medulloblastoma by the systemic and local administration, respectively, of IFN-\(\alpha\). Subsequent phase II studies on the use of recombinant IFN-\(\alpha\) for malignant glioma showed a response rate of 10.3 to 20%.

Nagai et al., who performed the systemic and local administration of IFN-\(\beta\), reported an overall response rate of 22.2% in 54 evaluable patients, and the response rates of 16.7% and 42.9% in patients with glioblastoma and medulloblastoma, respectively.\(^9\) In 10 patients with malignant glioma, Yoshida et al. systemically administered 3 \(\times 10^5\) to 3 \(\times 10^6\) units of IFN-\(\beta\) for 16 to 50 days continuously via an intravenous route or locally administered 5 \(\times 10^4\) to 3 \(\times 10^6\) units into the tumors for 7 to 73 days continuously via an Ommaya reservoir implanted when a tumor was removed. The result showed the size of the tumor was reduced by 50% or higher in 2 of 7 systemically treated patients and 1 of 3 locally treated patients.

However, it was reported that, in any case, the antitumor effect after the administration period lasted for only a short period, and that the administration of IFN alone would not eventually prolong survival, although it might provide remission during the administration period. Therefore, investigators started to attempt various regimens with IFN.

Interferon Combination Therapy

To improve the therapeutic results of the interferon single therapies for brain tumor, combinations with other therapies or drugs were attempted. So far, the following combinations have been examined.

1. Combination with radiotherapy

For the combination with radiotherapy, which has been the most effective adjuvant therapy for malignant brain tumor, Miyoshi et al.\(^5\) and Korosue et al.\(^6\) performed basic research with IFN-\(\alpha\)H9251 and IFN-\(\beta\)H9252, respectively. The following hypotheses were obtained: partially synchronized radiotherapy with IFN in relation to the DNA synthesis inhibiting effect of IFN might be effective; IFN might play a role by sensitizing patients to radiation; and there might be an interaction between sublethal damage by radiation and the direct antitumor effect of IFN.

Regarding clinical applications, Mahaley et al. reported that the combination of radiotherapy and IFN significantly prolonged the median survival time in patients with malignant glioma, and that the combination provided better results than the combination of radiotherapy and BCNU (carmustine), which was previously the standard therapy for brain tumor patients in the institution.\(^7\)

2. Combination with chemotherapy

Various combinations of IFN and anticancer agents have been examined. The Mayo Clinic reported that the combination of BCNU and IFN-\(\alpha\) caused a significantly higher synergistic effect than that with other drugs in 35 patients with recurrent glioma: the combination achieved an efficacy rate of 29% and a period of 10.1 months, and blocked the progression of the disease for 6 months or longer in 37% of the patients. Nitrosourea anticancer drugs, such as ACNU (nimustine hydrochloride) and MCNU (ranimustine), are available in Japan, but single therapy with any of the drugs has been effective for only 30 to 50% of patients with brain tumor.

Examination of the combination of IFN-\(\beta\) and ACNU with 13 human glioma cell lines showed the combination 5 mg of ACNU and
$1 \times 10^7$ IU of IFN-$\beta$ provided a tumor proliferation inhibiting effect of at least 2 log cell kill, and that the effect was obtained in 9 cell lines, as compared with 2 and 1 cell line by the single therapy with ACNU and IFN, respectively. Further, the effect was higher than that of at least 2 log cell kill observed in 7 cell lines treated with 10 mg of ACNU alone.\(^5\) When ACNU is clinically applied at a usual dose of 2 to 3 mg/kg body weight, the concentration obtained in brain tumors is approximately 1 to 5 mg. It is practically impossible to increase the dose because of possible adverse effects, such as bone marrow suppression. Therefore, the results indicating the potentiation of the antitumor effect more than the addition of the effect of each anticancer drug and IFN at a usual dose suggest the effectiveness of the combination therapy.

### 3. Combination with radio-chemotherapy

Since the combination of IFN-$\beta$ and ACNU showed high antitumor activity in a basic experimental study with a human glioma cell line, a clinical study was started in Japan by combining the IFN-$\beta$ and ACNU combination therapy with radiotherapy (IFN-$\beta$-ACNU-Radiation [IAR] therapy) as an adjuvant therapy for malignant glioma. Yoshida et al. reported that the prognosis as determined by the mean survival period was significantly improved with IAR therapy (25.3 months) as compared with radiation alone (15.2 months) and radiation + ACNU (19.7 months), and that the initial response rate by IAR therapy was higher than that by radiation + ACNU (60.5% vs. 35.7%). Further, Yoshida et al. also confirmed the efficacy of IAR therapy in 175 malignant glioma patients followed for a long time.\(^9\) Hatano et al. reported that increasing the administration frequency of IFN-$\beta$ to twice daily increased its antitumor effect.\(^10\) A U.S. study on the combination of IFN-\(\alpha\), BCNU, and radiation for malignant brain tumor reported a median survival time of 12.7 months and a mean survival time of 16.1 months for Grade IV astro-

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**Fig. 1** 54-year-old male patient: An enhanced lesion on CT scan indicating a tumor was observed in the right frontal lobe (a). It was diagnosed as glioblastoma. Since the postoperative image showed a residual tumor (b), IMR therapy was performed. A marked reduction of the tumor was observed at the end of the initial induction therapy (c). This patient received 3-month maintenance therapy, with no tumor recurrence for the subsequent 2 years.
Interferon Maintenance Therapy

Although up to 60% of patients with malignant brain tumor could achieve remission by initial induction therapy, most of them experienced recurrence. For example, the remission and mean survival periods of patients with glioblastoma were reported to be as short as 11.2 and 13.9 months, respectively. Therefore, various maintenance therapies following initial induction therapy are being examined.

Wakabayashi et al. performed IFN-β-MCNU-Radiation (IMR) therapy as an initial induction therapy in patients who developed malignant glioma for the first time, and compared the remission period between those treated with a maintenance therapy consisting of $1 \times 10^6$ units of IFN-β every 2 weeks and $80 \text{mg/m}^2$ of MCNU every 6 weeks for at least 3 months after the end of the induction therapy and those not treated with it. The patients registered into the initial induction therapy were randomly divided into 2 groups with and without the maintenance therapy, and they were compared for time to tumor progression (TTP) and total survival period. The results showed a significantly prolonged survival period in the maintenance therapy group (Fig. 2). Particularly, the patients who achieved complete remission by the initial induction therapy appeared to achieve a significantly prolonged remission period by receiving the maintenance therapy. It was also suggested that a certain therapeutic effect could be expected from the maintenance therapy in patients who developed the disease for the first time at 47 years or younger, or who achieved a partial response or a better response with the initial induction therapy. These results suggest that both initial induction and maintenance therapies may be important for the treatment of malignant glioma.\textsuperscript{12)

New Developments in Interferon Therapy

1. Drug delivery system (DDS)-combined interferon (liposome)

Although IFN-β has been clinically applied for treating brain tumor, the clinical efficacy of IFN-β alone is less than expected when it was introduced in an uncombined form. It seems necessary to combine it with other therapies or drugs to fully realize its potential. In fact, IFN-β shows a marked antitumor effect at as low as 100 units \textit{in vitro}, while it has been reported to produce tumor reduction by 50% or higher only in 15% of clinical cases, even at 10 million units. This difference in the efficacy of IFN may be explained by the pharmacokinetics of IFN, its stability in blood or tissue, or the blood brain barrier.

In an effort to overcome the problem of the...
low in vivo effect of IFN, liposome has been examined as a drug delivery system (DDS). Epstein et al. examined the embedding of IFN in liposome and successfully changed the biological activities and pharmacokinetics of IFN. Kato et al. added sulfatide to liposome as a component to deliver IFN through the blood brain barrier into the cerebral parenchyma, and compared the stability, pharmacokinetics, intraorgan distribution, and antitumor effect between the embedded and free IFN. The result showed the blood titer of the free IFN became undetectable as early as 2 hours after intravenous administration, while the liposome-embedded IFN was detected at as high as 10^3 IU/ml or higher even 8 hours after administration. Further, an IFN titer of 100 IU/g tissue or higher was confirmed in the brain and subcutaneously implanted brain tumor tissue where no IFN was detected after the intravenous administration of the free IFN. It is expected that the clinical application of DDS will progress to increase the effectiveness of IFN for brain tumor.\(^\text{13}\)

2. Interferon gene therapy

Larsson et al. reported that endogenous IFN-\(\beta\) was produced from glioma cells using a super-induction technique. This glioma-derived endogenous IFN-\(\beta\) has an antitumor effect on human glioma cells. We have been developing IFN gene therapy in which human IFN-\(\beta\) genes are embedded in the liposome with an affinity for glioma cells to selectively introduce the liposome into glioma cells and locally generate a large amount of endogenous IFN-\(\beta\), thereby causing an antitumor effect on glioma. Since this technique ensures the secretion and maintenance of a much higher local concentration of IFN than administration from outside, the so-called paracrine effect can be expected. Further, the technique has been reported to cause phenomena that have not been observed with exogenous IFN, such as the induction of apoptosis of transgenic glioma cells. Finally, we expect an association with the immune system to indirectly enhance the antitumor effect.\(^\text{14}\)

A clinical study on the gene therapy for brain tumor (malignant glioma) using this positively-charged liposome embedded-human IFN-\(\beta\) gene (local injection of the IFN-\(\beta\) gene-embedding liposome into brain tumor) was started on April 3, 2000 at the Nagoya University Hospital. So far, 5 patients have been registered and examined for the safety and efficacy of the therapy. The results of the study will be reported soon.

Conclusions

This paper outlines the current clinical application of interferon to brain tumor. There remains much to be examined about the use of IFN, such as appropriate administration regimens and the importance of maintenance therapy. However, together with the new developments in IFN therapy including the use of DDS and gene therapy, it is expected that the therapeutic results of antitumor therapies with cytokines including IFN will be improved.

REFERENCES

6) Korosue, K., Tamaki, N. and Matsumoto, S.:


