

## Original Article

# Interleukin-10 inhibits the production of inflammatory cytokines by antigen-stimulated mononuclear cells from asthmatic patients

Toshiya Takahashi,<sup>1</sup> Ken-ichi Maeda,<sup>1</sup> Yoichi Nakamura,<sup>1</sup> Yoshio Okano,<sup>2</sup> Ning Ge<sup>1</sup> and Saburo Sone<sup>1</sup>

<sup>1</sup>Third Department of Internal Medicine, Tokushima University School of Medicine, Tokushima and

<sup>2</sup>Department of Internal Medicine, Toneyama National Hospital, Osaka, Japan

### ABSTRACT

Bronchial asthma, characterized by chronic airway inflammation, involves many inflammatory cytokines. Interleukin (IL)-10 is a potent inhibitor of cytokine synthesis. Thus, the effects of IL-10 were examined on the production of granulocyte–macrophage colony stimulating factor (GM-CSF), IL-5, IL-1 $\beta$ , IL-2 and interferon (IFN)- $\gamma$  by antigen (*Dermatophagoides farinae*, Df)-stimulated mononuclear cells obtained from asthmatic patients who were sensitized with the antigen and from healthy subjects *in vitro*. Production of IL-5 and IL-2 was enhanced by Df antigen in the asthmatic subjects, but not in the healthy controls. In contrast, levels of GM-CSF, IFN- $\gamma$  and IL-1 $\beta$  production were enhanced by the antigen in both groups. Exogenous IL-10 (10 ng/mL) inhibited the production of GM-CSF, IFN- $\gamma$  and IL-1 $\beta$  induced by Df antigen in both groups and also inhibited the production of IL-5 and IL-2 induced by the antigen in the asthmatic subjects. The inhibition of GM-CSF production by IL-10 was stronger than that by IL-4. These results indicated that the responsiveness to the inhibitory effect of IL-10 on the production of inflammatory cytokines is not abrogated in asthmatic patients and that IL-10 may be useful in the treatment of bronchial asthma.

**Key words:** bronchial asthma, granulocyte–macrophage colony stimulating factor, interferon- $\gamma$ , interleukin-1 $\beta$ , interleukin-2, interleukin-5, interleukin-10.

### INTRODUCTION

Bronchial asthma is characterized by chronic airway inflammation, a process involving many inflammatory cell types, such as mast cells, eosinophils, neutrophils, lymphocytes and macrophages.<sup>1–3</sup> These cells modulate inflammation by producing cytokines at the sites of airway inflammation.<sup>4–8</sup> Investigation of bronchoalveolar lavage fluid in asthmatic patients has revealed that levels of granulocyte–macrophage colony stimulating factor (GM-CSF), interleukin (IL)-5, IL-2 and IL-1 $\beta$  are higher in the patients compared with non-asthmatic controls.<sup>9–11</sup> Therefore, regulation of the production of inflammatory cytokines may be therapeutically beneficial in bronchial asthma.

Interleukin-10 is a cytokine initially described as an inhibitor of cytokine synthesis. This molecule is produced by T helper 2 (Th2) cells, B cells, macrophages, keratinocytes, mast cells and some tumor cells.<sup>12–14</sup> Interleukin-10 exhibits anti-inflammatory properties both *in vivo* and *in vitro* by directing Th populations toward a Th2-like cytokine production profile.<sup>15–17</sup> Furthermore, IL-10 inhibits the production of Th1 cytokines, such as IFN- $\gamma$  and IL-2.<sup>18–21</sup> In monocyte/macrophage lineage cells, IL-10 reduces the release of various inflammatory cytokines, such as IL-1, IL-6, IL-8, GM-CSF and tumor necrosis factor (TNF)- $\alpha$ .<sup>22–25</sup> Thus, IL-10 may be a potent regulator of allergic inflammation in the pathogenesis of bronchial asthma. However, there have been no previous reports of the effects of IL-10 on antigen-induced

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Correspondence: Saburo Sone MD, Third Department of Internal Medicine, Tokushima University School of Medicine, Kuramoto-cho 3, Tokushima 770-8503, Japan.

Email: ssone@clin.med.tokushima-u.ac.jp

Received 11 March 1999. Accepted for publication 27 September 1999.

production of inflammatory cytokines in asthmatic patients *in vitro*.

In the present study, we examined whether IL-10 could suppress the production of inflammatory cytokines, such as GM-CSF, IL-5, IL-2, IL-1 $\beta$  and IFN- $\gamma$ , by specific antigen (house dust mite) stimulation in mononuclear cells (MNC) of asthmatic patients.

## METHODS

### Subjects

Studies were performed on cells from 30 patients with bronchial asthma (as defined by the criteria of the American Thoracic Society) with a mean age of  $42.4 \pm 6.3$  years and seven healthy subjects with a mean age of  $40.0 \pm 7.3$  years. The scores of asthmatic patients in a radioallergosorbent test (RAST) against *Dermatophagoides farinae* (Df) were  $> 2$  ( $> 2$  is positive on a 0–4 score) and those of healthy subjects against Df were all negative, as measured with a Phadebas RAST kit (Pharmacia Diagnostics, Uppsala, Sweden). None of the patients had received steroid therapy. Peripheral blood was collected during periods free from asthmatic attacks. All patients gave their informed consent for participation in the study.

### Reagents

The following reagents were used: Df antigen (Torii Pharmaceutical Co., Tokyo, Japan), recombinant human IL-10 (ED50 with TF-1 cells, 1 ng/mL; PeproTech Inc., Rocky Hill, NJ, USA), monoclonal antihuman IL-10 antibody (R & D Systems Inc., Minneapolis, MN, USA). Recombinant human IL-4 (specific activity  $1.0 \times 10^6$  U/mg protein) was kindly provided by Ono Pharmaceutical Co. (Osaka, Japan). *Dermatophagoides farinae* antigen, recombinant human IL-10 and recombinant human IL-4 used in the present study did not contain levels of endotoxin detectable by the Limulus test (sensitivity limit 0.3 ng/mL; Seikagaku Kogyo, Tokyo, Japan).

### Isolation of peripheral blood mononuclear cells

Heparinized peripheral blood samples were obtained from patients, who had given their informed consent to participation in the study, and were diluted with an equal volume of phosphate-buffered saline (PBS). The cells were centrifuged at  $400 \times g$  for 30 min in lymphocyte separation medium (density at 20°C 1.077–1.080 g/mL;

Organon Teknika Corp., Durham, NC, USA). The cells at the gradient interface were collected and washed twice in PBS with centrifugation at  $150 \times g$  for 10 min. These cells were mainly mononuclear cells (96%) and their viability was  $> 98\%$ , as determined by the trypan blue dye exclusion test.

### Preparation of cell-conditioned media

Mononuclear cells were washed twice with RPMI-1640 (Nissui Pharmaceutical Co., Tokyo, Japan) and resuspended in RPMI-1640 supplemented with 10% fetal calf serum (FCS). They were plated in triplicate at a final concentration of  $2 \times 10^6$  cells/mL, with or without Df antigen (10 mg/mL, unless otherwise stated), IL-10 (10 ng/mL) and/or IL-4 (10 ng/mL); 10 mg/mL of Df antigen was chosen as the optimal concentration based on a previous report.<sup>26</sup> The MNC were then incubated for 3 days at 37°C in an atmosphere of 5% CO<sub>2</sub> and 95% humidity in air. Then, cell-free supernatants were obtained by centrifugation at  $150 \times g$  for 10 min and stored at  $-70^\circ\text{C}$  until use.

### Measurement of cytokines

Interleukin-5 and GM-CSF were measured with commercial enzyme-linked immunosorbent assay (ELISA) kits (Quantikine Human Immunoassay; R & D Systems Inc.), which specifically detected human IL-5 and GM-CSF, respectively. Interferon- $\gamma$ , IL-1 $\beta$  and IL-2 were measured by ELISA (Otsuka Pharmaceutical Co., Tokushima, Japan), which specifically detected human IFN- $\gamma$ , IL-1 $\beta$  and IL-2, respectively. In these assays, the minimum detectable concentrations of human IL-5, GM-CSF, IFN- $\gamma$ , IL-1 $\beta$  and IL-2 were 1.0, 1.5, 20, 8 and 25 pg/mL, respectively.

### Statistical analysis

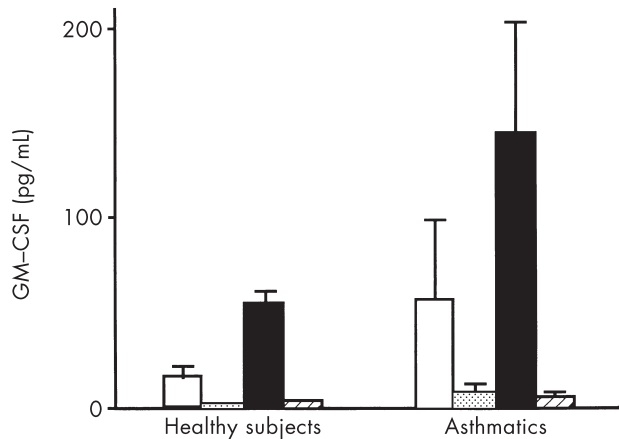
Data are presented as the mean  $\pm$  SEM. Individual experimental values were compared by paired two-tailed Student *t*-tests. Concentration dependency was examined by one-way analysis of variance of repeated measurements. A level of  $P < 0.05$  was considered significant.

## RESULTS

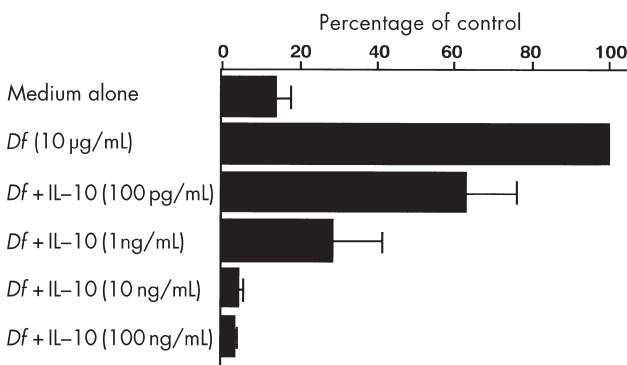
### Inhibitory effect of IL-10 on GM-CSF production by blood MNC from asthmatic subjects

The effects of IL-10 on GM-CSF production were examined in blood MNC of asthmatic patients and healthy

subjects (Fig. 1). The basal level of GM-CSF production without Df stimulation was higher in asthmatic patients than in healthy subjects ( $56.9 \pm 40.7$  vs  $15.6 \pm 6.1$  pg/mL,  $P < 0.05$ ). Although the addition of Df

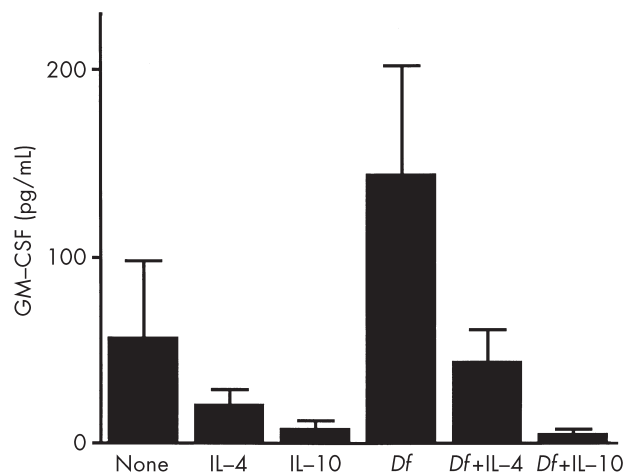


**Fig. 1** Down-regulatory effect of interleukin (IL)-10 on granulocyte-macrophage colony stimulating factor (GM-CSF) production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 30$ ) and healthy subjects ( $n = 7$ ). The MNC were incubated with *Dermatophagoides farinae* (Df) antigen ( $10 \mu\text{g/mL}$ ; ■), IL-10 ( $10 \text{ ng/mL}$ ; ▩), both Df and IL-10 (▨) or medium alone (□) for three days and the concentrations of GM-CSF in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section.



**Fig. 2** Dose-dependent effects of interleukin (IL)-10 on granulocyte-macrophage colony stimulating factor (GM-CSF) production by peripheral mononuclear cells (MNC) from asthmatic patients. The MNC were incubated in the presence of *Dermatophagoides farinae* (Df) antigen ( $10 \mu\text{g/mL}$ ) with various concentrations of IL-10 ( $0.1$ – $100 \text{ ng/mL}$ ) for 3 days and the concentrations of GM-CSF in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section. Values are the mean + SEM from triplicate cultures.

antigen to the cultures enhanced the production of GM-CSF in both groups, the level of the induction was much higher in asthmatic patients ( $143.9 \pm 59.4$  vs  $56.9 \pm 40.7$  pg/mL,  $P < 0.05$ ) than in healthy subjects ( $54.6 \pm 6.5$  vs  $15.6 \pm 6.1$  pg/mL,  $P < 0.05$ ). Interleukin-10 ( $10 \text{ ng/mL}$ ) markedly inhibited the elevated production of GM-CSF by Df stimulation to a level less than that under unstimulated control conditions in both asthmatics ( $5.1 \pm 2.4$  vs  $143.9 \pm 59.4$  pg/mL,  $P < 0.05$ ) and healthy subjects ( $3.4 \pm 1.0$  vs  $54.6 \pm 6.5$  pg/mL,  $P < 0.05$ ). The inhibition by IL-10 was dose-dependent and reached the lowest level at  $10 \text{ ng/mL}$  IL-10 in three experiments with different asthmatic patients (Fig. 2 shows representative data from one asthmatic patient). Because IL-4 is also known as a cytokine synthesis inhibitory factor, the inhibitory effect of IL-10 on GM-CSF production was compared with that of IL-4 (Fig. 3). Although IL-4 ( $10 \text{ ng/mL}$ ) also inhibited GM-CSF production by MNC from asthmatic patients in both the presence and absence of Df antigen, the inhibitory effect of IL-10 was much stronger ( $6.3 \pm 2.6$  vs  $134.3 \pm 79.5$  pg/mL,  $P < 0.05$ ) than that of IL-4 ( $40.2 \pm 17.7$  vs  $134.3 \pm 79.5$  pg/mL,  $P < 0.05$ ).



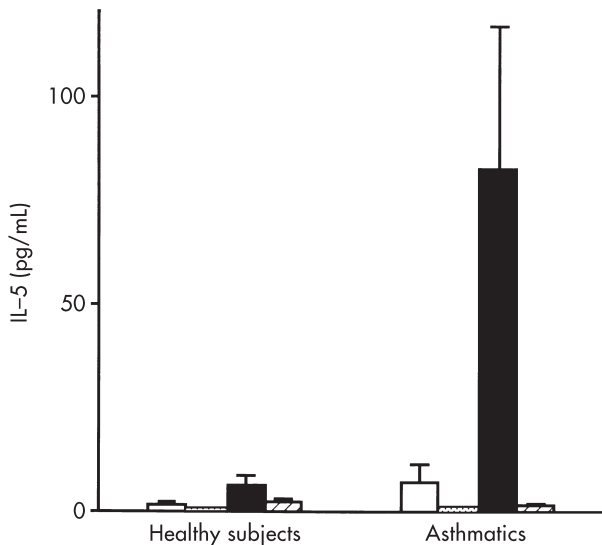
**Fig. 3** Comparison of the effects of interleukin (IL)-4 and IL-10 on granulocyte-macrophage colony stimulating factor (GM-CSF) production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 7$ ). The MNC were incubated with or without *Dermatophagoides farinae* (Df) antigen ( $10 \mu\text{g/mL}$ ), IL-4 ( $10 \text{ ng/mL}$ ) and IL-10 ( $10 \text{ ng/mL}$ ) for 3 days and the concentrations of GM-CSF in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section. Values are the mean + SEM from seven patients.

### Inhibitory effect of IL-10 on IL-5 production by blood MNC from asthmatic subjects

Levels of IL-5 production by MNC without Df stimulation were low and were not different between asthmatic patients and healthy subjects ( $7.1 \pm 4.0$  vs  $1.6 \pm 4.0$  pg/mL, not significant (NS)). The Df antigen stimulated the production of IL-5 by MNC from asthmatic patients at significantly higher levels ( $82.5 \pm 32.8$  vs  $7.1 \pm 4.0$  pg/mL,  $P < 0.05$ ) compared with those from healthy subjects ( $6.0 \pm 2.5$  vs  $1.6 \pm 0.3$  pg/mL, NS). Under the same experimental conditions, addition of IL-10 (10 ng/mL) resulted in significant inhibition of IL-5 production by Df-stimulated MNC in asthmatic patients ( $1.4 \pm 0.3$  vs  $82.5 \pm 32.8$  pg/mL,  $P < 0.05$ , Fig. 4). The inhibitory effect of IL-10 was dose-dependent and reached the lowest level at 10 ng/mL of IL-10 in three experiments in different asthmatic patients (Fig. 5 shows representative data from one asthmatic patient).

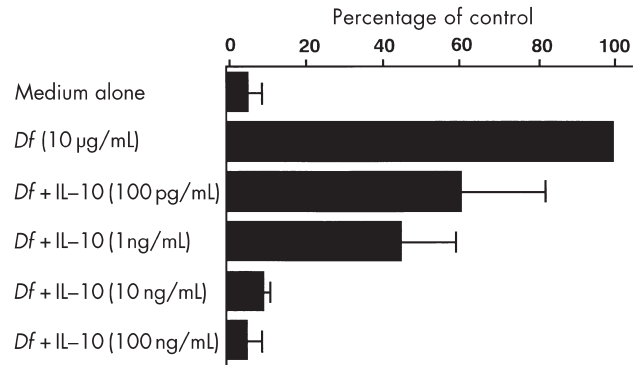
### Inhibition of IFN- $\gamma$ , IL-1 $\beta$ and IL-2 production by IL-10 in asthmatic subjects and healthy volunteers

We also examined the inhibitory effects of IL-10 on the production of IFN- $\gamma$  (Fig. 6), IL-1 $\beta$  (Fig. 7) and IL-2 (Fig. 8).

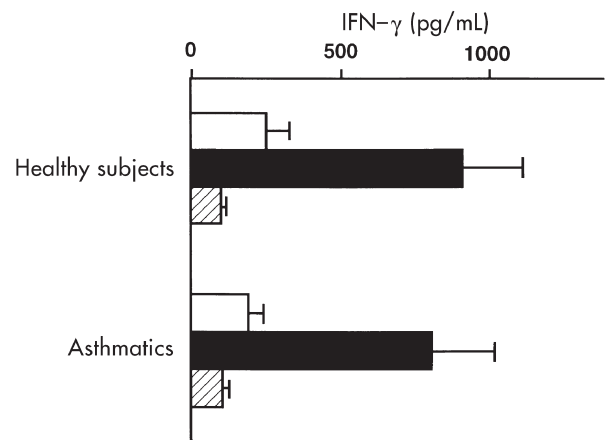


**Fig. 4** Down-regulatory effect of interleukin (IL)-10 on IL-5 production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 30$ ) and healthy subjects ( $n = 7$ ). The MNC were incubated with *Dermatophagoides farinae* (Df) antigen (10  $\mu$ g/mL; ■), IL-10 (10 ng/mL; ▨), both Df and IL-10 (▩) or medium alone (□) for 3 days and the concentrations of IL-5 in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section.

Interferon- $\gamma$  production was detected in the supernatants of MNC from both asthmatic patients ( $192.3 \pm 47.3$  pg/mL) and from healthy subjects ( $256.7 \pm 46.7$  pg/mL) without Df stimulation. Induction of IFN- $\gamma$  production by Df stimulation was not significantly different between the two groups ( $810.9 \pm 276.4$  vs  $192.3 \pm 47.3$  pg/mL,



**Fig. 5** Dose-dependent effects of interleukin (IL)-10 on IL-5 production by peripheral mononuclear cells (MNC) from an asthmatic patient. The MNC were incubated in the presence of *Dermatophagoides farinae* (Df) antigen (10  $\mu$ g/mL) with various concentrations of IL-10 (0.1–100 ng/mL) for 3 days. The concentrations of IL-5 in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section. Values are the mean + SEM from triplicate cultures.



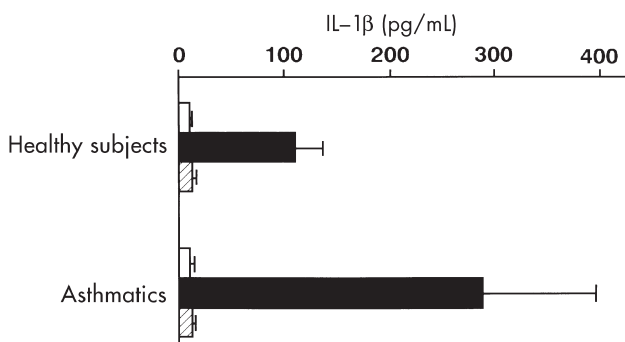
**Fig. 6** Down-regulatory effects of interleukin (IL)-10 on interferon (IFN)- $\gamma$  production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 30$ ) and healthy subjects ( $n = 7$ ). The MNC were incubated with *Dermatophagoides farinae* (Df) antigen (10  $\mu$ g/mL; ■), Df and IL-10 (10 ng/mL; ▨) or medium alone (□) for 3 days and the concentrations of IFN- $\gamma$  in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section.

$P < 0.05$  and  $908.7 \pm 196.4$  vs  $256.7 \pm 46.7$  pg/mL,  $P < 0.05$ ). In contrast, the stimulatory effect of Df antigen on IL-1 $\beta$  production in asthmatic patients ( $289.0 \pm 105.0$  vs  $10.3 \pm 1.5$  pg/mL,  $P < 0.05$ ) was stronger than that in healthy subjects ( $110.0 \pm 25.8$  vs  $9.6 \pm 1.6$  pg/mL,  $P < 0.05$ ). Interleukin-2 was not detected in the supernatant of MNC from subjects without Df stimulation and the antigen stimulation of IL-2 production was observed only in asthmatic patients ( $61.5 \pm 13.7$  pg/mL) and not in healthy subjects. The levels of production of these three

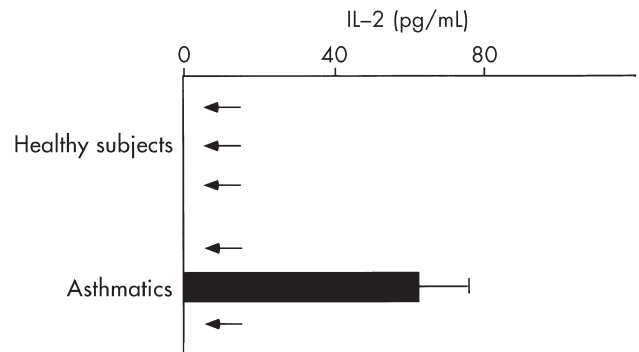
cytokines were completely reduced by the addition of IL-10 to the unstimulated control level.

**Blocking effect of anti-IL-10 antibody on IL-10 inhibition of GM-CSF production in an asthmatic subject and a healthy volunteer**

To assess whether IL-10 inhibition of cytokine production by Df-stimulated MNC is mediated by IL-10 itself, we examined the blocking effect of anti-IL-10 antibody

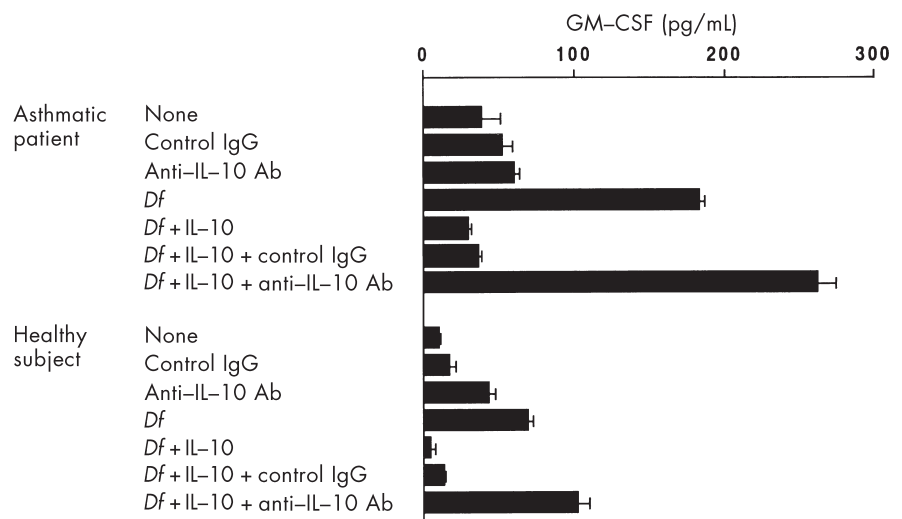


**Fig. 7** Down-regulatory effects of interleukin (IL)-10 on IL-1 $\beta$  production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 30$ ) and healthy subjects ( $n = 7$ ). The MNC were incubated with *Dermatophagoides farinae* (Df) antigen ( $10 \mu\text{g/mL}$ ; ■), Df and IL-10 ( $10 \text{ ng/mL}$ ; ▨) or medium alone (□) for 3 days and the concentrations of IL-1 $\beta$  in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section.



**Fig. 8** Down-regulatory effects of interleukin (IL)-10 on IL-2 production by peripheral mononuclear cells (MNC) from asthmatic patients ( $n = 30$ ) and healthy subjects ( $n = 7$ ). The MNC were incubated with *Dermatophagoides farinae* (Df) antigen ( $10 \mu\text{g/mL}$ ; ■), Df and IL-10 ( $10 \text{ ng/mL}$ ; ▨) or medium alone (□) for 3 days and the concentrations of IL-2 in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section. Arrows indicate negative results (i.e. concentrations below  $20 \text{ pg/mL}$ ).

**Fig. 9** Blocking effects of anti-interleukin (IL)-10 antibody (Ab) on the production of granulocyte-macrophage colony stimulating factor (GM-CSF) by peripheral mononuclear cells (MNC) from an asthmatic patient or a healthy subject. The IL-10 was incubated with anti-IL-10 antibody for 30 min and the mixture was added in the culture of *Dermatophagoides farinae* (Df)-stimulated ( $10 \mu\text{g/mL}$ ) MNC (final concentrations of IL-10 and anti-IL-10 antibody were  $10 \text{ ng/mL}$  and  $2 \mu\text{g/mL}$ , respectively). The concentrations of GM-CSF in the supernatants were determined by enzyme-linked immunosorbent assay as described in the Methods section. Values are the mean + SEM from triplicate cultures.



on GM-CSF production by Df-stimulated MNC in the presence of IL-10 (10 ng/mL). Interleukin-10 was incubated with anti-IL-10 antibody for 30 min and the mixture was added in the culture of MNC from an asthmatic patient or a healthy volunteer (final concentration of anti-IL-10 antibody in the culture was 2 mg/mL). Results showed that the antibody completely reversed the suppressive effect of IL-10 on GM-CSF production in MNC from both the asthmatic patient (Df,  $183 \pm 2.9$ ; Df + IL-10,  $26.8 \pm 0.7$ ; Df + IL-10 + anti-IL-10,  $262 \pm 12.1$  pg/mL;  $P < 0.05$ ) and the healthy subject (Df,  $70.1 \pm 2.8$ ; Df + IL-10,  $4.4 \pm 4.4$ ; Df + IL-10 + anti-IL-10,  $103 \pm 7.1$  pg/mL;  $P < 0.05$ ; Fig. 9). The antibody increased GM-CSF production in the absence of Df antigen and IL-10 in the culture of MNC from the healthy subject (none,  $10.4 \pm 0.5$ ; anti-IL-10,  $43.7 \pm 3.8$ ;  $P < 0.05$ ) but not the asthmatic patient (none,  $39.2 \pm 12.3$ ; anti-IL-10,  $60.9 \pm 3.0$ ; NS).

## DISCUSSION

The Df antigen enhanced the production of GM-CSF, IFN- $\gamma$  and IL-1 $\beta$  by MNC obtained not only from asthmatic patients, but also from healthy subjects. These observations indicated a possible division of stimulatory effects of Df antigen on cytokine production by MNC into three groups:

1. Levels of IL-5 and IL-2 production were up-regulated by Df antigen only in asthmatics who were clinically sensitized with the antigen, but not in healthy subjects, as reported by other investigators.<sup>27</sup>
2. Production of GM-CSF and IL-1 $\beta$  was up-regulated by the antigen in both groups, but this effect was stronger in asthmatic patients than in healthy subjects.
3. The stimulatory effect of Df antigen on the production of IFN- $\gamma$  was not different between healthy subjects and asthmatic patients.

These results may be explained by differences in responsiveness of cytokine-producing cells to Df antigen. Cells producing GM-CSF, IFN- $\gamma$  and IL-1 $\beta$  may have been sensitized by Df antigen or may have responded strongly to the antigen in both atopic patients, such as asthma patients, and in healthy subjects and IL-5- and IL-2-producing cells may have been sensitized by the antigen or may have responded to the antigen selectively in atopic patients, but not in healthy subjects. This hypothesis is consistent with reports that IL-5 and IL-2 are essential for the mechanism of helper T lymphocyte-modulated allergic reactions.<sup>4-7</sup> Alternatively, Df antigen may have dual activities of antigen-specific and non-specific

stimulation, although the non-specific stimulatory effect of Df antigen was not due to contaminating endotoxin, because none was detected by Limulus test. Asthmatic-selective stimulation of the production of IL-5 and IL-2 may be induced by antigen-specific activity of Df antigen and the stimulation of IFN- $\gamma$  production in both asthmatic patients and healthy subjects may be induced by non-specific activity of the antigen. Moreover, the enhanced production of GM-CSF and IL-1 $\beta$  may be induced both by specific and non-specific activities of the antigen. Further investigations are needed to clarify the mechanism of these effects.

In the present study, we showed that IL-10 strongly inhibited the production of inflammatory cytokines, such as GM-CSF, IL-5, IL-1 $\beta$ , IL-2 and IFN- $\gamma$ , by Df antigen-stimulated blood MNC. Inhibitory effects of IL-10 on the production of GM-CSF, IFN- $\gamma$  and IL-1 $\beta$  were found in both asthmatic patients and in healthy subjects. Interleukin-10 also inhibited the production of IL-5 and IL-2 induced by Df antigen in asthmatic patients. This inhibition was confirmed to be due to IL-10 itself by neutralization of the inhibitory effect by anti-IL-10 antibody. These observations are consistent with reports showing that IL-10 inhibits the production of pro-inflammatory cytokines by monocytes/macrophages stimulated with non-specific factors and the production of IL-5, IL-2 and IFN- $\gamma$  by T cells.<sup>18-21,28</sup> In addition, the present neutralizing study showed that anti-IL-10 antibody increased GM-CSF production in the absence of IL-10 in culture of MNC from a healthy subject, but not from an asthmatic patient. This indicates that endogenous production of IL-10 in asthmatic patients does not reach a level capable of suppressing the overproduction of inflammatory cytokines and that the responsiveness to the inhibitory effect of IL-10 is not affected in asthmatic patients. There have been several previous reports regarding endogenous production of IL-10 in allergic diseases. Koning *et al.* have reported that IL-10 mRNA expression by purified T cells of children with allergic and non-allergic asthma and children with atopic dermatitis is strongly decreased compared with that of healthy controls.<sup>29</sup> Borish *et al.* have reported that the bronchial lavage fluid of asthmatic patients is characterized by diminished concentrations of IL-10 compared with that of normal and non-asthmatic subjects.<sup>30</sup> Moreover, the concentrations of GM-CSF, IL-5, IL-2 and IL-1 $\beta$ , but not of IFN- $\gamma$ , have been reported to be higher in asthmatic patients compared with non-asthmatic controls.<sup>9-11</sup> Interestingly, Grunig *et al.* have shown that eosinophilic airway inflammation, IL-5 levels in bronchoalveolar lavage fluid and

numbers of  $\alpha\beta$ T cells in the lung tissues are all heightened in IL-10 gene knockout mice compared with wild-type controls.<sup>31</sup> Thus, decreased IL-10 production may result in a lack of immunosuppression of allergic inflammation and exogenous IL-10 may have a biological function as an anti-allergic cytokine. Analysis of cytokine production by alveolar macrophages of asthmatic subjects has demonstrated that inhaled corticosteroid therapy results in the increased expression of IL-10 mRNA and protein by alveolar macrophages, while the levels of macrophage inflammatory protein-1 $\alpha$ , IFN- $\gamma$  and GM-CSF production are decreased after steroid treatment.<sup>32</sup> It has been reported previously that IL-10 administration inhibits inflammatory cell accumulation in the airways of ovalbumin-sensitized mice.<sup>33</sup>

In summary, the present findings indicate that the responsiveness to the inhibitory effect of IL-10 on the production of inflammatory cytokines is not abrogated in asthmatic patients. Therefore, regulation of production of inflammatory cytokines by IL-10 may be useful in the treatment of bronchial asthma and further investigation of the roles of IL-10 in airway of asthmatic patients is required.

## ACKNOWLEDGEMENTS

The authors thank Mr Yasukazu Ohmoto and Miss Kaori Murata (Cell Technology Institute, Otsuka Pharmaceutical Co., Tokushima, Japan) and Miss Naomi Morita (medical student, University of Tokushima School of Medicine) for technical assistance.

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