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Effects of polysaccharides (FI_0-b) from mycelium of *Ganoderma tsugae* on proinflammatory cytokine production by THP-1 cells and human PBMC (I)[1]

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KEY WORDS *Ganoderma tsugae*; polysaccharides; interleukin-1; interleukin-6; interleukin-8; tumor necrosis factor

ABSTRACT

AIM: To compare the effects of water-soluble polysaccharides, FI_0-b, and its formic acid-modified derivative, FI_0-b-H, on production of human proinflammatory cytokines. **METHODS**: The polysaccharides were modified by formic acid. Cytokine production was quantitated by radioimmunoassay. mRNA for cytokines was measured by semi-quantitative RT-PCR. **RESULTS**: FI_0-b and FI_0-b-H 4, 40, and 400 mg/L significantly downregulated interleukin-1α (IL-1α) production by THP-1 cells induced by lypopolysaccharide (LPS) 1 or 10 mg/L and phorbol myristate acetate (PMA) 200 nmol/L. At lower stimulation with LPS 10 mg/L and PMA 200 nmol/L, both polysaccharides significantly upregulated tumor necrosis factor α (TNFα) production by THP-1 cells. However, at higher stimulation with LPS 100 mg/L and PMA 200 nmol/L, they downregulated TNFα production. FI_0-b-H downregulated interleukin-8 (IL-8) production by THP-1 cells at a lower-dose of LPS 1 mg/L and PMA 200 nmol/L, but upregulated IL-8 production at a higher-dose of LPS 10 mg/L and PMA 200 nmol/L. Production of cytokines (IL-1α and TNFα) was transcriptionally or post-transcriptionally regulated by FI_0-b and FI_0-b-H. **CONCLUSION**: The water-soluble polysaccharides of *Ganoderma tsugae* mycelium have bidirectional immunomodulatory effects on cytokine production in different stimulatory conditions in a dose-dependent manner. Compared with FI_0-b, FI_0-b-H has more marked effects on human proinflammatory cytokine production.

INTRODUCTION

The fruit body and cultured mycelium of *Ganoderma tsugae*, SongShan LingZhi, a widely used Chinese traditional herb, were demonstrated to inhibit the growth of murine sarcoma cells, S-180[1-3]. Some investigators have suggested that the extract of *G tsugae* mycelium increased both the splenic NK cell activity and serum interferon (IFN) titers in mice[4,5]. However, the effects of the polysaccharide from *G tsugae* mycelium on production of human proinflammatory cytokines remains unclear.

We have previously established a systematic procedure for the extraction, fractionation and the purification of polysaccharides from *G tsugae*. Based on these results, FI_0-b[1] (tumor inhibition ratio 46.8 %), a β-*D*-(1,3) linked glucan (M_r 10 000), was selected.

Some reports showed that the formic acid degradation products, chemically modified from the active polysaccharide, had a tumoricidal activity[6]. In this study, FI_0-b was also chemically modified by formic acid to increase hydrophilicity. We attempted to compare the effects of FI_0-b with its formic acid-modified derivative, FI_0-b-H, on the production of proinflammatory cytokines

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and their mRNA expression by human myelomonocytic leukemic cells, THP-1, and human peripheral blood mononuclear cells (PBMC).

MATERIALS AND METHODS

Chemical modification of polysaccharides from cultured mycelium of *G tsugae* Polysaccharide fraction of FI_0-b (100 mg) was dissolved in 60 mL of 88 % HCOOH at 80 ℃ for 40 min, precipitated with 5 v of 99 % EtOH (over night) and centrifuged at 400 × *g* for 30 min. The precipitate was again dissolved in 30 mL of H_2O at 100 ℃ for 2 h, precipitated with 5 v of 99 % EtoH and centrifuged at 400 × *g* for 30 min. Then, the precipitate was dialyzed, and lyophylized and 36 mg of precipitate was obtained[6]. In this study, contaminated endotoxin amounts were determined by *Limulus* amebocyte lysate test (Seikagarukogyo, Tokyo, Japan). The endotoxin levels in the two samples were below 2.5 μg/L.

Cell culture Human myelomonocytic leukemic cells, THP-1, were used in this study. The cells were cultured in RPMI-1640 (Gibco NY, USA) medium supplemented with 10 % heat-inactivated newborn bovine serum (Da Lian Biological Regent Factory, Dalian, China), benzylpenicillin 1×10^5 IU/L, streptomycin 100 mg/L, HEPES 10 mmol/L, and L-glutamine 5 mmol/L. THP-1 cells 3.2×10^8/L were added to the 96 well microtiter plates. The cells were stimulated by lypopolysaccharides (LPS) (*E coli* serotype 0111: B4, Sigma Chemical Co, MO, USA) 1 or 10 mg/L and phorbol myristate acetate (PMA) (Sigma) 200 nmol/L for induction of IL-1α, IL-6, IL-8, or tumor necrosis factor α (TNFα). FI_0-b or FI_0-b-H was administered 30 min before treatment of the cells with LPS and PMA. As a control experiment, culture medium with the same volume of the polysaccharide solution or LPS-PMA solution was added. The final volume was 200 nL/well. After incubation at 37 ℃ with 5 % CO_2 for 20 h, the cells were lysed by three times freeze and thaw cycles.

Separation of human PBMC Blood was obtained from three healthy adult volunteers. PBMC were separated by density centrifugation at 400 × *g* for 10 min in a Ficoll-Hypaque solution (Histopaque*, 1.077 kg/L), washed three times with phosphate buffer salined (PBS), then suspended in RPMI-1640 medium containing 2 % human AB serum. PBMC 5×10^9 cells/L were added to the 24 well culture plates, then stimulated by LPS 1 mg/L. FI_0-b or FI_0-b-H was added 30 min before LPS treatment. The final volume was 1 mL/well. After incubation at 37 ℃ with 4 % CO_2 for 20 h, the cell suspension was lysed by three times freeze and thaw cycles.

Cytokine quantitation After cell lysis and centrifugation at 12 000 × *g* in 1.5 mL Eppendorf tube, IL-1α, IL-6, IL-8, and TNFα in the supernatants were quantitated by human cytokine specific raidoimmunoassay (RIA). Methods for RIA for human cytokines were followed as previously reported[7,8].

Analysis of mRNA production for IL-1α and TNFα by THP-1 cells using RT-PCR Method THP-1 cells (3.2×10^8 cells/L) were added to the 24-well culture plates. The cells were cultured as previously described and were stimulated with LPS 10 mg/L or 100 mg/L and PMA 200 nmol/L. The final volume was 1 mL/well. We obtained maximal production of TNFα mRNA 3 h after incubation and IL-1α mRNA after 5 h. The cells were collected by centrifugation at 450 × *g* for 10 min. Total RNA was extracted by using ISOGEN® (Nippon Gene Co, Toyama, Japan) cell lysis buffer. The production of IL-1α and TNFα mRNA was detected by RT-PCR method[9].

1 Primers for TNFα and β-actin Primers for TNFα and β-actin were synthesized as below. TNFα Primer (sense): 5′-ATGAGCACTGAAAGCATGATCT-NFα Primer (antisense): 3′-TCACAGGGCAATGATCC-CAAAGTAGACCTGCCCβ-actin Primer (sence): 5′-TCCTGTGGCATCCACGAAACTβ-actin Primer (antisence): 3′-GAAGCATTTGCGGTGGACGAT.

2 Reaction conditions 24 cycles: 94 ℃ 45 s, 60 ℃ 45 s, 72 ℃ 90 s; 30 cycles: 94 ℃ 45 s, 60 ℃ 45 s, 72 ℃ 90 s.

Statistics Data were expressed as $\bar{x} \pm s$ and assessed by unpaired Student's *t*-test.

RESULTS

Effect of FI_0-b and FI_0-b-H on proinflammatory cytokine production by THP-1 cells Both FI_0-b and FI_0-b-H had no significant effect on IL-1α production induced by LPS 1 mg/L and PMA 200 nmol/L, however, FI_0-b 4 mg/L upregulated IL-1α production in non or low stimulatory conditions (LPS 1 mg/L and PMA 200 nmol/L). On the other hand, FI_0-b and FI_0-b-H obviously downregulated IL-1α production induced by LPS 10 mg/L and PMA 200 nmol/L. Higher doses

of FI_0-b-H (40 or 400 mg/L) slightly enhanced IL-1α production wiuhout stimulants, however, decrease the IL-1α production by LPS 10 mg/L and PMA 200 nmol/L (Tab 1). Both FI_0-b and FI_0-b-H upregulated IL-8 production without LPS or PMA stimulation (medium), however, downregulated IL-8 production at a low dose of stimulants (LPS 1 mg/L and PMA 200 nmol/L). At a higher dose of stiumlants (LPS 100 mg/L and PMA 200 nmol/L), FI_0-b 4 mg/L downregulated IL-8 production from (24 ± 5) μg/L to (6.4 ± 0.7) μg/L, on the other hand, FI_0-b-H 4 mg/L upregulated IL-8 production [(46 ± 8) μg/L]. FI_0-b-H, especially at 400 mg/L, significantly increased IL-8 production [(160 ± 40) μg/L] at a high dose of LPS and PMA, whereas FI_0-b 400 mg/L had no obvious effect on IL-8 production at the same stimulatory condition (Tab 1).

Tab 1. Effect of FI_0-b and FI_0-b-H on IL-1 α and IL-8 production by THP-1 cells (μg/L). $n = 3$ samples. $\bar{x} \pm s$. $^bP < 0.05$, $^cP < 0.01$ *vs* control.

Polysaccharides mg/L	Medium	LPS 1 mg/L PMA 200 nmol/L	LPS 10 mg/L PMA 200 nmol/L
		IL-1 α	
Medium	0.13 ± 0.04	0.5 ± 0.3	4.8 ± 2.5
FI_0-b 4	4.20 ± 0.16^c	3.6 ± 0.6^c	0.38 ± 0.09^b
FI_0-b 40	0.30 ± 0.10	1.0 ± 0.4	0.08 ± 0.03^b
FI_0-b 400	0.20 ± 0.05	0.081 ± 0.017	0.20 ± 0.03^b
FI_0-b-H 4	0.10 ± 0.0	0.124 ± 0.014	0.28 ± 0.08^b
FI_0-b-H 40	0.74 ± 0.14^c	0.52 ± 0.14	1.2 ± 0.4
FI_0-b-H 400	0.54 ± 0.10^c	0.38 ± 0.26	0.08 ± 0.03^b
		IL-8	
Medium	1.00 ± 0.10	15.0 ± 2.2	24 ± 5
FI_0-b 4	15 ± 4^c	3.6 ± 1.0^c	6.4 ± 0.7^c
FI_0-b 40	2.0 ± 0.4	8.8 ± 1.0^b	9.8 ± 1.2^c
FI_0-b 400	12 ± 4^c	4.0 ± 0.6^c	15 ± 4
FI_0-b-H 4	24 ± 4^c	4.6 ± 0.6^c	46 ± 8^c
FI_0-b-H 40	21 ± 7^c	5.2 ± 1.2^c	30 ± 5
FI_0-b-H 400	3.8 ± 0.9^c	1.003 ± 0.006^c	160 ± 40^c

FI_0-b and FI_0-b-H themselves had no obvious suppressive effects on IL-6 production by THP-1 cells in the non-stimulatory condition. At a low dose of LPS 10 mg/L and PMA, FI_0-b-H 4 or 40 mg/L significantly upregulated IL-6 production [(48.2 ± 1.0), (19 ± 6) μg/L, respectively] compared to medium control [(6.2 ± 2.5) μg/L]. However, these doses of FI_0-b-H slightly reduce the IL-6 production by higher dose of LPS 100 mg/L and PMA, therefore, the formic acid degradation derivative, FI_0-b-H, demonstrated significant bidirectional regulation of the IL-6 production (Tab 2). FI_0-b-H slightly upregulated TNFα production without stimulus and significantly upregulated TNFα production at a lower level of stimulation by LPS 10 mg/L and PMA 200 nmol/L from (2.16 ± 0.26) in medium control to (1280 ± 190) μg/L (LPS 100 mg/L or PMA 200 nmol/L). but with the highest stimulation by LPS 100 mg/L and PMA, FI_0-b-H downregulated TNFα production. FI_0-b possessed similar but weaker effects on TNFα production.

Tab 2. Effect of FI_0-b and FI_0-b-H on IL-6 production by THP-1 cells (μg/L). $n = 3$ samples. $\bar{x} \pm s$. $^bP < 0.05$, $^cP < 0.01$ *vs* control.

Polysaccharides mg/L	Medium	LPS 10 mg/L PMA 200 μmol/L	LPS 100 mg/L PMA 200 μmol/L
		IL-6	
Medium	1.62 ± 0.17	6.2 ± 2.2	6.8 ± 2.1
FI_0-b 4	1.5 ± 0.3	4.3 ± 1.5	1.46 ± 0.20^b
FI_0-b 40	4.20 ± 0.16^c	3.6 ± 0.6^c	0.38 ± 0.09^b
FI_0-b 400	1.4 ± 0.4	1.76 ± 0.04^b	2.54 ± 0.06
FI_0-b-H 4	0.55 ± 0.19^b	48.2 ± 1.0^c	3.4 ± 0.3
FI_0-b-H 40	1.75 ± 1.11	19 ± 6^b	0.186 ± 0.004^c
FI_0-b-H 400	1.7 ± 0.5	2.70 ± 0.28^b	2.7 ± 1.0^b
		TNFα	
Medium	0.22 ± 0.04	2.16 ± 0.26	640 ± 25
FI_0-b 4	0.80 ± 0.28^b	2.64 ± 0.15	2.6 ± 0.3^c
FI_0-b 40	0.30 ± 0.13	3.2 ± 0.5	5.20 ± 0.23^c
FI_0-b 400	0.56 ± 0.19^b	112.2 ± 2.4^c	280 ± 30^c
FI_0-b-H 4	3.4 ± 0.9^c	1280 ± 190^c	6.00 ± 0.5^c
FI_0-b-H 40	1.9 ± 0.9^b	400 ± 117^c	160 ± 15^c
FI_0-b-H 400	0.88 ± 0.28^b	160 ± 25^c	2.8 ± 0.3^c

Effects of FI_0-b and FI_0-b-H on proinflammatory cytokine production by PBMC Non-stimulated PBMC produced low amounts of cytokines, however, some cytokine production was augmented after administration of FI_0-b and FI_0-b-H without the stimulant LPS (Tab 3). When PBMC were added with LPS at 1 mg/L, higher levels of IL-1α production was detected [(92 ± 18) μg/L, Volunteer # 1]. Addition of FI_0-b or FI_0-b-H significantly inhibited IL-1α production [(1.50 ± 0.21), (2.00 ± 0.06) μg/L respectively, Volunteer # 1] to the lowest levels. However, when LPS induced PBMC to produce low amounts of IL-1α [(0.524 ± 0.011) μg/L, Volunteer # 2; (2.4 ± 1.4) μg/L, Volunteer # 3], FI_0-b and FI_0-b-H significantly upregulated IL-1α production. Both FI_0-b and FI_0-b-H augmented

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IL-6 production in three volunteers' PBMC without LPS, on the other hand, FI_0-b and FI_0-b-H had no significant effect on LPS-induced IL-6 production in human PBMC.

Both FI_0-b and FI_0-b-H stimulated IL-8 production by PBMC without LPS (Volunteer # 2 or Volunteer # 3). LPS induced remarkably higher amount of IL-8 production by PBMC [(13 000 ± 500) μg/L, Volunteer # 1; 18 000 ± 6000 μg/L, Volunteer # 3], and addition of FI_0-b significantly reduced IL-8 production [(1000 ± 300) μg/L, Volunteer # 1; (1500 ± 700) μg/L, Volunteer # 3] in the same stimulatory condition. Both FI_0-b and FI_0-b-H upregulated production of TNFα by PBMC induced by LPS at 1 mg/L (Volunteer # 1, Volunteer # 3), whereas FI_0-b itself induced TNFα production [Volunteer # 1: (9.6 ± 1.9) μg/L; Volunteer # 2: (14.1 ± 1.0) μg/L] compared with medium control [(0.23 ± 0.05), (0.48 ± 0.03) μg/L respectively].

Effects of FI_0-b and FI_0-b-H on cytokine mRNA expression by THP-1 cells In order to investigate the mechanism of immunomodulatory effects of FI_0-b and FI_0-b-H, we studied their effects on TNFα and IL-1α mRNA expression induced by THP-1 cells. We selected the samples that could significantly increase or decrease cytokine production by THP-1 cells. Both 24 and 30 cycles of cDNA amplification were tried by PCR. At 30 cycles, each band on the agarose gel was more obvious than that at 24 cycles (Fig 1, 2). This result showed that at twenty-four cycles, cDNA amplification had not reached the plateau stage. Therefore, at twenty-four cycles we could compare the effect of FI_0-b and FI_0-b-H on the amount of cDNA which might reflect original amount of mRNA. At lower-dose of LPS and PMA, FI_0-b-H 4 mg/L induced significant upregulation in TNFα mRNA production and at the same time TNFα protein production by THP-1 cells was markedly high [(1280 ± 190) μg/L] (Tab 2 and Fig 1 Lane C). But at higher-doses of LPS and PMA, FI_0-b-H 4 mg/L had no obvious effect on TNFα mRNA amount (Lane D, E), whereas it reduced TNFα protein production in the same condition. FI_0-b 400 mg/L induced higher amount of IL-1α mRNA in LPS and PMA-stimulated THP-1 cells (Fig 2 Lane C), but significantly downregulated IL-1α protein production [(0.20 ± 0.03) μg/L] (Tab 1). Results from RT-PCR for IL-1α did not coincide with IL-1α protein amount measured by RIA.

万方数据

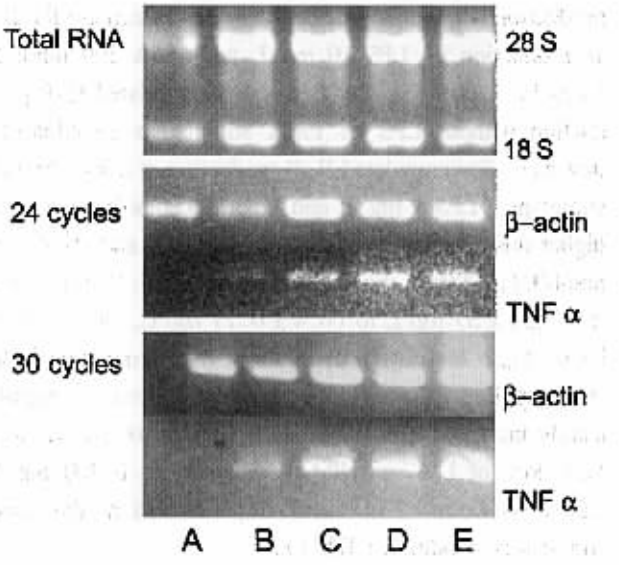

Fig 1. RT-PCR for TNF_α produced by THP-1 cells with LPS 10 mg/L and PMA 200 nmol/L or LPS 100 mg/L and PMA 200 nmol/L. Lane A: FI_0-b-H 4 mg/L; Lane B: LPS 10 mg/L + PMA; Lane C: LPS 10 mg/L + PMA + FI_0-b-H 4 mg/L; Lane D: LPS 100 mg/L + PMA; Lane E: LPS 100 mg/L + PMA + FI_0-b-H 4 mg/L.

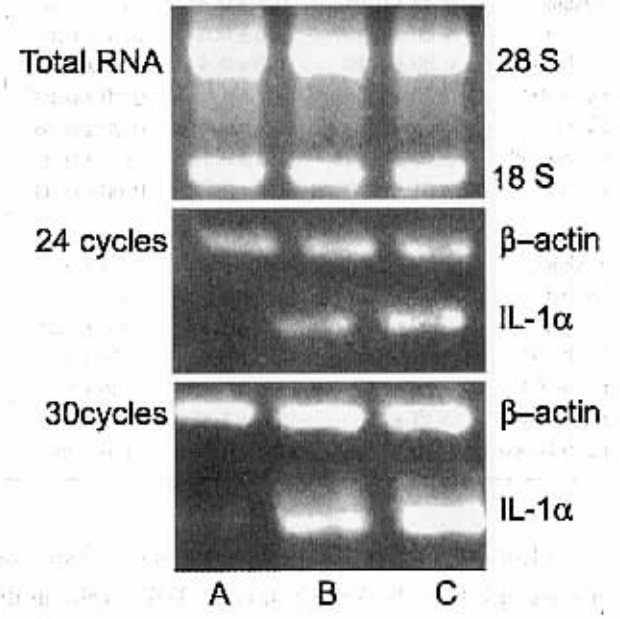

Fig 2. RT-PCR for $IL\text{-}1_\alpha$ produced by THP-1 cells with LPS 10 mg/L and PMA 200 nmol/L. Lane A: FI_0-b 400 mg/L; Lane B: LPS + PMA; Lane C: FI_0-b 400 mg/L + LPS + PMA.

Tab 3. Effect of FI_0-b and FI_0-b-H on proinflammatory cytokine production by PBMC (μg/L). $n=3$ samples. $\bar{x} \pm s$. $^bP<0.05$, $^cP<0.01$ *vs* control. NA: non applicable.

Polysaccharides	Volunteer # 1	Volunteer # 2	Volunteer # 3
		IL-1 α	
Medium	2.2 ± 0.3	0.0083 ± 0.0006	0.31 ± 0.03
FI_0-b 400 mg/L	0.92 ± 0.04^c	2.2 ± 0.3^c	1.50 ± 0.09^c
FI_0-b-H 400 mg/L	1.2 ± 0.7	3.0 ± 1.4^b	1.05 ± 0.10
LPS 1 mg/L	92 ± 18	0.524 ± 0.011	2.4 ± 1.4
FI_0-b 400 mg/L + LPS	1.50 ± 0.21^c	8.4 ± 0.8^c	10.4 ± 1.9^c
FI_0-b-H 400 mg/L + LPS	2.00 ± 0.06^c	28.80 ± 0.23^c	40 ± 6^c
		IL-6	
Medium	0.02 ± 0.09	0.013 ± 0.007	0.002 ± 0.0003
FI_0-b 400 mg/L	15.6 ± 1.0^c	15.2 ± 1.4^c	2.2 ± 0.4 ^{c}
FI_0-b-H 400 mg/L	37 ± 5^c	0.0042 ± 0.0014^c	0.0121 ± 0.0017^c
LPS 1 mg/L	20 ± 3	16.8 ± 2.4	16 ± 5
FI_0-b 400 mg/L + LPS	17 ± 3	39 ± 26	25 ± 7
FI_0-b-H 400 mg/L + LPS	30 ± 6	78 ± 17^c	2.6 ± 2.3^b
		IL-8	
Medium	242 ± 22	140 ± 40	410 ± 12
FI_0-b 400 mg/L	190 ± 40	710 ± 140^c	1400 ± 500^b
FI_0-b-H 400 mg/L	156 ± 12	560 ± 140^b	2400 ± 1000^b
LPS 1 mg/L	13000 ± 500	150 ± 40	18000 ± 6000
FI_0-b 400 mg/L + LPS	1000 ± 300^c	1180 ± 40^c	1500 ± 700^c
FI_0-b-H 400 mg/L + LPS	8700 ± 1900	810 ± 100^c	NA
		TNF α	
Medium	0.23 ± 0.05	0.48 ± 0.03	0.23 ± 0.05
FI_0-b 400 mg/L	9.6 ± 1.9^c	14.1 ± 1.0^c	0.08 ± 0.12^c
FI_0-b-H 400 mg/L	0.24 ± 0.06	0.50 ± 0.09	1.32 ± 0.08^c
LPS 1 mg/L	5.1 ± 2.2	19.8 ± 1.8	0.57 ± 0.13
FI_0-b 400 mg/L + LPS	38 ± 13^b	22 ± 6	1.92 ± 0.15^c
FI_0-b-H 400 mg/L + LPS	23 ± 6^b	15 ± 5	1.68 ± 0.08^c

DISCUSSION

THP-1 cell line has human myelomonocytic leukemic cell origin and its reactions to a variety of stimulation have some similarity to those of human macrophages[10,11]. Administration of LPS and PMA induced IL-1α, IL-6, IL-8 and TNFα production by THP-1 cells, however, LPS and PMA regulated the cytokine gene expression by distinct signal pathways in these monocytic cells at both the transcriptional and post transcriptional levels.

Our previous study[1,2] demonstrated that the water-soluble extract of *G tsugae* mycelium (FI_0-b) had anti-proliferative effect on murine S-180 sarcoma cells *in vivo*, however, its effect was not obvious *in vitro*. This study showed that FI_0-b effect on cytokine production by THP-1 cells was not significant at lower or higher doses of LPS and PMA except for TNFα production. FI_0-b-H, with higher hydrophilicity compared with FI_0-b, showed stronger positive effects on these proinflammatory cytokine production by THP-1 except for IL-1α. At lower LPS stimulation, TNFα production was extremely low, but addition of FI_0-b-H tremendously augmented the cytokine production. At a low FI_0-b-H dose, high quantity of TNFα was induced, but at high FI_0-b-H doses, TNFα production was low. When higher doses of LPS and PMA induced elevated levels of TNFα production, FI_0-b-H significantly inhibited its production. Regarding IL-6 production, there existed similar bidirectional regulation of TNFα release production. On the other hand, regarding IL-8, addition of FI_0-b-H at higher LPS doses still increased the cytokine production. This might be attributed to the fact that the IL-8 production did not reach the maximal production even at higher LPS doses. Increase in IL-8 production at higher LPS doses also rules out the speculation that decrease in other cytokine' production at

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higher stimulation was due to cell toxicity.

The results of LPS-induced cytokine production by PBMC were generally similar to those of lower-dose LPS and PMA-induced cytokine production by THP-1 cells. However, there were no unified responses to FI_0-b or FI_0-b-H, reflecting different individual genetic backgrounds.

FI_0-b and FI_0-b-H may increase antitumor function *in vivo* by stimulation of proinflammatory cytokines at relatively lower stimulatory conditions. IL-1, IL-6, IL-8, and TNF are multifunctional cytokines having widely overlapping functions involving immune response, inflammation, hematopoiesis, and tumoricidal activity[12,13]. In inflammation, IL-1 and TNF induce fever, shock, cytokine and prostaglandin syntheses, and bone and cartilage resorption, therefore they play a key role in the manifestation of diseases including cachexia, extensive burns and chronic infections. IL-6 stimulates IgG production by B-cells, IL-8 has chemotactic activity towards neutrophils, furthermore they also play a role in a variety of inflammatory diseaes. FI_0-b 400 mg/L increased IL-1α mRNA amount, on the other hand, it downregulated IL-1α protein production, indicating that FI_0-b 400 mg/L regulated the IL-1α expression on the post-transcriptional levels. FI_0-b-H 4 mg/L elevated TNFα mRNA amount induced by LPS and PMA at two different doses and also upregulated TNFα protein production by LPS 10 mg/L and PMA 200 nmol/L, indicating that TNFα protein production was reflected by its mRNA quantity. Cytokine gene expression might be regulated by these herbal polysaccharides at both transcriptional and post-transcriptional steps. FI_0-b-H is genarally more effective than FI_0-b and the chemical modification method can enhance this polysaccharides' immunomodulatory function. The water-soluble polysaccharides of *G tsugae* mycelium have significant bidirectional immunomodulatory effects on proinflammatory cytokine production in different stimulatory conditions.

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松杉灵芝菌丝体多糖(FI_0-b)对人组织瘤细胞和人外周血白细胞产生炎症性细胞因子的影响 (I)[1]

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关键词 松杉灵芝; 多糖类; 白介素-1; 白介素-6; 白介素-8; 肿瘤坏死因子

目的: 比较研究松杉灵芝水溶性多糖(FI_0-b)及其甲酸修饰产物(FI_0-b-H)对人炎症性细胞因子产生的影响. **方法**: 用放射免疫分析法(RIA)及逆转录聚合酶链反应(RT-PCR)分析法研究 FI_0-b 和 FI_0-b-H 对人组织瘤细胞(THP-1)和人外周血单核细胞(PBMC)分泌的各种与炎症有关的细胞因子(白介素-1 α, 6, 8, 肿瘤坏死因子 α)及对白介素-1 α, 肿瘤坏死因子 α mRNA 表达的影响. **结果**: FI_0-b 和 FI_0-b-H (浓度分别为 4, 40, 400 mg/L)显著抑制 LPS 10 mg/L 协同 PMA 200 nmol/L 诱导的 THP-1 细胞和 PBMC 细胞产生 IL-1 α 和 TNF α 的量. 当低浓度刺激剂(LPS 1 mg/L 协同 PMA 200 nmol/L)刺激 THP-1 细胞时, FI_0-b-H 明显提高白介素-8 的产生; 但当高浓度刺激剂(LPS 10 mg/L 协同 200 nmol/L PMA)刺激 THP-1 细胞时, FI_0-b-H 却明显抑制白介素-8 的产生. FI_0-b 和 FI_0-b-H 对 THP-1 细胞白介素-1α 和肿瘤坏死因子 α 的 mRNA 表达有显著作用. **结论**: 与 FI_0-b 相比, FI_0-b-H 在人炎症性细胞因子的产生方面表现出较强的活性. 松杉灵芝菌丝体水溶性多糖在不同的刺激条件下, 具有双向免疫调节作用, 并具有一定的剂量依赖关系.

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