

배양 비점막 상피세포에서 IFN- γ 와 IL-4에 의한 ICAM-1의 발현양상

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Expression of ICAM-1 by IFN- γ and IL-4 in Cultured Normal Human Nasal Epithelial Cells

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— ABSTRACT —

Background and Objectives : Intercellular adhesion molecule-1 (ICAM-1) is an important molecule in immune and inflammatory responses. It is regulated by IL-1, IL-4, TNF- α , and IFN- γ on many cell types. The aim of this study was to investigate the effects of IFN- γ and IL-4 on expression of ICAM-1 in cultured normal human nasal epithelial (NHNE) cells. **Materials and Methods** : NHNE cells were prepared by primary culture method of monolayer culture of dissociated cells from inferior turbinate mucosa. Primary cultured cells were characterized as an epithelial cell type by immunofluorescence assay and transmission electron microscopy. We analyzed the quantitative expression of ICAM-1 on cultured NHNE cells by treated with IFN- γ and IL-4 using fluorescence activated cell sorter (FACS). **Results** : Treatment of cultured NHNE cells with IFN- γ (1ng/ml) for 24 hours induced 4-fold increase of the surface ICAM-1 compared with constitutive expression by mean intensity fluorescence (MIF). However, IL-4 showed no effect even though the concentration of it was increased gradually. IFN- γ induced ICAM-1 expression according to the time course was significantly highest level at 24 hours. There were no significant changes in expression of ICAM-1 by IL-4 according to the time course. **Conclusion** : These findings suggest that IFN- γ is a potent ICAM-1 inducer, but IL-4 have no effects on expression of ICAM-1 on cultured NHNE cells. (*J Clinical Otolaryngol* 2000;11:261-266)

KEY WORDS : Human nasal epithelial cells · ICAM-1 · IFN- γ · IL-4.

서 론

cyto-
kine), (growth factor)

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ICAM - 1
 IL - 1 , IFN - , TNF - IL - 4³⁾⁴⁾
 IFN - ICAM - 1 가
 IL - 4 vascular cell adhe -
 sion molecule - 1 (VCAM - 1)
 ICAM - 1
 가⁵⁻⁷⁾
 IL - 4 ICAM - 1 가
 IFN - IL - 4가 ICAM - 1
 IFN - IL - 4가
 ICAM - 1

재료 및 방법

비점막 상피세포의 분리 및 배양

3
⁸⁾⁹⁾
 0.01M
 100 U/ml
 penicillin, 100 µg/ml streptomycin sulfate 0.25
 µg/ml Fungizone 가 1
 4 30
 5 × 5 mm
 dispase(Boehringer Mannheim, Ma -
 nnheim, Germany) 37 90
 200 mesh
 1500 rpm 10
 12 well pl -
 ate (36 , 5% CO₂)
 Medium 154(Cascade Biologics,

Portland, OR) 100U/ml penicillin, 100 µg/ml st -
 reptomycin sulfate, 0.25 µg/ml Fungizone 가
 18
 4
 상피세포 동정
 가
⁸⁾ (Ni -
 kon, Tokyo, Japan)
 (Hitachi, Naka, Japan)
 (desmosome)
¹³⁾
 (tonofilament)
 int -
 ermediate filament anti - cytokeratin
 No. 8(Boehringer Mannheim, Mannheim, Germany),
 anti - cytokeratin pan(Boehringer Mannheim, Man -
 nheim, Germany), anti - vimentin(Sigma, St. Louis,
 MO) von Willebrand factor
 antivon Willebrand factor(Boehringer Mannheim, Mannheim, Germany)
⁷⁾¹⁴⁾
 ICAM-1의 발현 유도 및 정량
 ICAM - 1 IFN -
 IL - 4 4
 0.01, 0.1 1 ng/ml recombinant human IFN -
 (Endogen, Woburn, MA) 0.01, 0.1, 1, 10 100
 ng/ml recombinant human IL - 4(Endogen, Wob -
 urn, MA) 24 1 ng/ml IFN -
 100 ng/ml IL - 4 24, 48, 72 96
 ICAM - 1
 anti - ICAM - 1 PE(Becton Dickinson, Fr -

anklin Lakes, NJ) 4 40

(fluorescence activated cell sorter, FACS) ELI - TE(Coulter, Harbor Boulevard, PO) 488 nm (mean intensity fluorescence, MIF)

SPSS SAS software ANOVA Repeated Measures ANOVA

결 과

비점막 상피세포의 형태 관찰

가

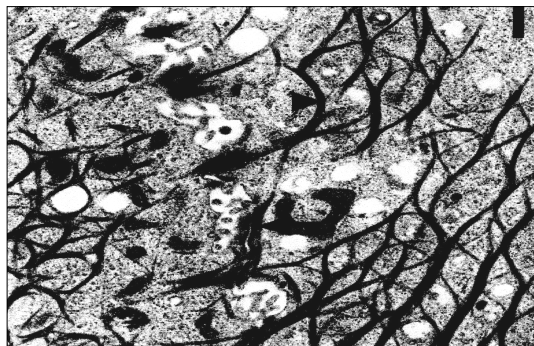


Fig. 1. Photograph of horizontal section of cultured normal human nasal epithelial cells by transmission electron microscopy. Tonofilaments (arrowhead) and desmosomes (arrow) were observed. Unit of bar scale : 10 μ m.

Table 1. Characteristics of cultured normal human nasal epithelial (NHNE) cells, mongolian gerbil fibroblast (MGF), and calf pulmonary arterial endothelial cell (CPAE) by immunofluorescence staining

Monoclonal antibody	Specificity	Results		
		NHNE	MGF	CPAE
Anti-cytokeratin pan	All epithelial cell	+	-	-
Anti-cytokeratin No. 8	Nonsquamous epithelial cell	+	-	-
Anti-vimentin	Cell of mesenchymal origin	-	+	-
Anti-von Willebrand factor	Endothelial cell	-	-	+

: IFN- IL-4 ICAM-1

(Fig. 1).

면역형광항체법에 의한 상피세포의 동정

cytokeratin No. 8 cytoke-
ratin pan

Mongolian gerbil fibroblasts
CPAE(calf pulmonary endothelial cell)
가

(Table 1).

사이토카인의 농도별 처리에 따른 ICAM-1의 발현

IFN- IC-
AM-1 3.94 \pm 1.42 IFN-
0.01, 0.1 1ng/ml 24
4.15 \pm 2.26, 7.16 \pm 2.65 15.
67 \pm 7.94 IFN- 가
(p=0.035), 1 ng/ml IFN-

Table 2. ICAM-1 expression on cultured normal human nasal epithelial cells after treatment of IFN- and IL-4 for 24 hours

Concentration (ng/ml)*	MIF (mean intensity fluorescence)	
	IFN-	IL-4
0.00 †	3.94 \pm 1.42	2.73 \pm 0.63
0.01	4.15 \pm 2.26	2.30 \pm 0.42
0.10	7.16 \pm 2.65	2.29 \pm 0.39
1.00	15.67 \pm 7.95	2.28 \pm 0.58
10.00	Not tested	2.75 \pm 0.78
100.00	Not tested	2.83 \pm 0.69

* : concentration of IFN- and IL-4

† : Constitutive expression of ICAM-1

Values are the mean \pm SD of five experiments in IFN- and three experiments in IL-4 treatment.

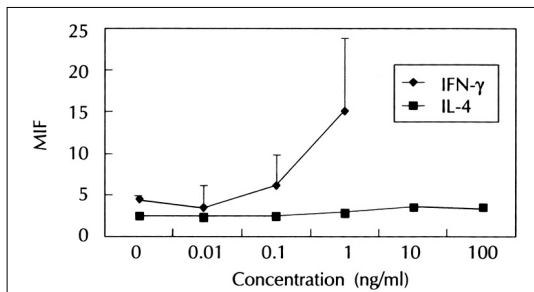


Fig. 2. Mean intensity fluorescence (MIF) of ICAM-1 on cultured normal human nasal epithelial cells according to the concentration of IFN- and IL-4. IFN- significantly increases ICAM-1 expression in a dose dependent pattern ($p < 0.05$), but IL-4 shows no effect.

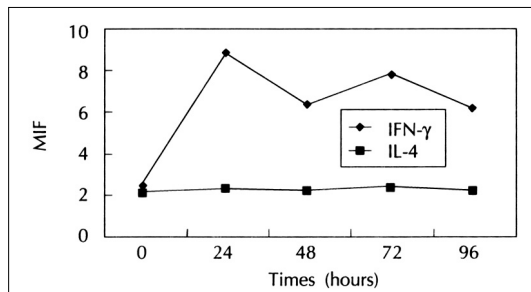


Fig. 3. ICAM-1 expression on cultured normal human nasal epithelial cells according to the time course after the treatment of 1 ng/ml of IFN- and 100 ng/ml of IL-4. Expression of ICAM-1 by IFN- is a highest level at 24 hours, but there is no changes in expression of ICAM-1 by IL-4. MIF : mean intensity fluorescence.

Table 3. ICAM-1 expression on cultured normal human nasal epithelial cells according to the time course after treatment of IFN- and IL-4

Time (hours)	MIF (mean intensity fluorescence)	
	IFN-	IL-4
0*	2.65 ± 0.11	2.60 ± 0.44
24	8.77 ± 2.73	2.68 ± 0.56
48	6.53 ± 2.38	2.44 ± 0.57
72	7.60 ± 1.58	2.65 ± 0.69
96	4.49 ± 1.21	2.61 ± 0.82

* : Constitutive expression of ICAM-1
 Values are the mean ± SD of three experiments, respectively

가 4 가
 (Table 2 and Fig. 2). IL-4 0.01, 0.1, 1, 10
 100 ng/ml 24
 가 , IL-4 1
 가 (Ta-
 ble 2 and Fig. 2).

사이토카인의 시간별 처리에 따른 ICAM-1의 발현
 1 ng/ml IFN- 100ng/ml IL-4
 24, 48, 72 96
 ICAM-1
 . IFN-
 24 96
 (Ta-
 ble 3 and Fig. 3).

. IL - 4

가 .

고 찰

ICAM - 1
 9- 12)
 IFN - IL - 4
 , ICAM -
 15) 16)
 ICAM - 1
 15- 17)
 가 , IL - 4
 ICAM - 1
 . IL - 4가 , ,
 ICAM - 1 가 ,
 6)
 (transcription)
 ICAM - 1 가 7)
 ICAM - 1 가 18)
 ICAM - 1 가 , IL - 4 ICAM - 1

: IFN- IL-4 ICAM-1

가
 ,
 IL-4가 ICAM-1
 Kim⁹⁾
 ICAM-1 가 , IL-1
 1
 IL-4
 4 ICAM-1
 ICAM-1
 가 IL-4
 ICAM-1
 4가 ICAM-1
 ICAM-1
 , 가
 (transepithelial mi-
 gration)
 결론
 IFN- IL-4
 ICAM-1 IFN-
 ICAM-1 가 , 24
 , IL-4 ICAM-1
 IFN- 가 ICAM-1
 중심 단어 : ICAM-1 · IFN- · IL-4

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