Basic and preclinical experiments of IL 8 and MCAF

1992 Fiscal Year Final Research Report Summary

Basic and preclinical experiments of IL 8 and MCAF

Research Project

Project/Area Number

03044066

Research Category

Grant-in-Aid for international Scientific Research

Allocation Type

Single-year Grants

Section

Joint Research

Research Institution

Cancer Research Institute, Kanazawa University

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Keywords

Leukocyte Chemotactic factors / cytokine / interleukin 8 / MCAF / structural analysis / receptors / gene expression / pathophysialogical roles

Research Abstract

BASIC AND PRECLINICAL EXPERIMENTS OF IL 8 AND MCAF
Novel chemotactic cytokines, IL 8 and MCAF were identified, purified, and molecularly cloned by us at National Cancer Institute, USA in 1987-1989, belong a family of emerging chemotactic cytokine family, CHEMOKINE. In this international collabolative research, the following projects were performed.
1)Structural analysis of IL 8/MCAF and determining their active site(s)-----Human IL 8 was expressed in E coli and purified to homogeneity in a large scale. The structure of IL 8 was analyzed by both NMR and X-ray crystallography. The proposed model shows that IL 8 exists as dimer through hydrogen bonding and two symmetry-related anti-parallel a-helices, 24A long and separated by 14A, lie on top of a six-stranded anti-parallel b-sheet platform. The active site of IL 8 was presumed to be the region surrounding His at 33 as well as N-terminal region based on the mutation studies and structural analyses.
2)Structure of the receptors for IL 8 and M CAF-----The MW of the receptors for IL 8 on human nutrophils was estimated to be 60,000 with Kd of lnM. The MW of the receptors for MCAF on human monocytes was estimated to be 40,000 with Kd of 25nM. Although the cDNA for IL 8 was cloned by other groups, we have cloned several novel cDNA which belong to the family of chemotactic cytokine receptors.
3)Molecular analysis of the regulation of the production of IL 8-----After cDNA cloning of human IL 8, we revealed the induction of the production of IL 8 by various types of cells after stimulating with IL 1, TNF, endotoxin, exotoxin, viral proteins, heavy metals, and superoxide generating substances. We also showed the suppression of the production of IL 8 by glucocorticoids, vitamin D3, lipooxygenase inhibitors, and FK506. We have determined the enhancer of the human IL 8 gene to be consisted of AP-1+NFKB. IL 8 production suppressive agents seem to inhibit the activation of nuclear factors which bind to these regions.
4)Establishment of pathophysiological roles of IL 8/MCAF and examining possible clinical application of IL 8/MCAF-----We have revealed the production of IL 8/MCAF in various human inflammatory diseases including ulcerative colitis, psoriasis, glomerulonephritis, urinary tract infection, and arthritis. Essential involvement of IL 8 in recruiting neutrophils in acute inflammation models in rabbits, such as dermatitis, arthritis, and lung reperfusion was established using monoclonal neutralizing antibody against rabbit IL 8. IL 8/MCAF cDNA transfection into tumor cells showed significant anti-tumor effect of these cytokines in mice. MCAF is further shown to prime murine macrophages to be tumor cytocidal and also have anti-infectious activity in mice. Possible hematopoietic activity of these cytokines was not proved either in vitro or in vivo.▲ Less

Research Products (15 results)

All Other

All Publications (15 results)

[Publications] Mahe,Y.ef al.: "Hepatitis B virus X protein transactivates human interleukin 8 gene" J.Biol.Chem.266. 13759-13763 (1991)

[Publications] Smyth,M.J.,ef al.: "Interleukin 8 gene expression and production in human peripheral blood lymphocyte subsets." J.Immunol.146. 3815-3823 (1991)

[Publications] Ko,Y.,ef al.: "A sensitive enzyme-linked immunosorbent assay for-human interleukin 8." J.Immunol.Methods. 149. 227-235 (1992)

[Publications] YASUMOTO,K.,ef al.: "Tumor necrosis factor alpha and interferon gamma synergistically induce interleukin 8 production in a human gastric cancer cell line" J.Biol.Chem.267. 22506-22511 (1992)

[Publications] Thompson,H.L.,MATSUSHIMA,K.: "Human polymorphonuclear leukocytes stimulated by tumor necrosis factor-alpha show increased adherence to extracellular matrix proteins" Clim.Exp.Immunol.90. 280-285 (1992)

[Publications] Ko,Y.,ef al.: "Elevated IL-8 level in the urine of patients with urinary tract infections." Infect.Immun.(1993)

[Publications] Oppenheim,J.J., Zachariae,C.O.C., Mukaida,N., Matsushima,K.: "Properties of the novel proinflammatory supergene"intercrine"cytokine family." Annu.Rev.Immunol., 31 (1991)

[Publications] MATSUSHIMA,K., BALDWIN,E.T., MUJAIDA,N.: "Chemical Immunol.In INTERLEUKINS:MOLECULAR BIOLOGY AND IMMUNOLOGY Kishimoto,T.Ed.Novel leukocyte recruitment and activating cytokines." Karger Publ.,Basel,SW, 29 (1992)

[Publications] Mahe, Y.etal.: "Hepatitis B virus X protein transactivates human interleukin 8 gene" J.Biol. Chem.266. 13759-13763 (1991)

[Publications] Smyth, M.J., etal.: "Interleukin 8 gene expression and production in human peripheral blood lymphocyte subset" J.Immunol.146. 3815-3823 (1991)

[Publications] Ko, Y., etal.: "A sensitive enzyme-linked immunosorbent assay for human interleukin 8" J.Immunol. Methods. 149. 227-235 (1992)

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[Publications] Ko, Y., etal.: "Elevated IL 8 level in the urine of patients with urinary tract infections" Infect. Immun.

[Publications] Oppenheim, J.J. etal.: Annu.Rev.Immunol.Properties of the novel proinflammatory supergene"intercrine"cytokine family, 617-648 (1991)

[Publications] Matsushima, K., etal.: Kishimoto, T.Edt. "Novel leukocyte recruitment and activating cytokines". Chemical Immunology, In INTERLEUKINS : MOLECULAR BIOLOGY AND IMMUNOLOGY, 236-265 (1992)