The Role of Cytokines in Inflammatory Bowel Disease

Maulana Suryamin, A. Aziz Rani***

ABSTRACT

The term "Inflammatory Bowel Disease" (IBD) is frequently used to denote two diseases, ulcerative colitis (UC) and Crohn's disease (CD). This condition is frequently recorded in the West, and along with development of diagnostic facilities, is beginning to be more commonly found in Indonesia.

The etiology of this disease is still unclear, but it is suspected that environmental, geographic, and genetic factors are involved. Cytokines play a great role in the pathogenesis of IBD, where in IBD there is an unbalance of pro-inflammatory cytokines and inhibitor cytokines. In IBD, there is an increase in pro-inflammatory cytokines, such as IL-1, IL-2, IL-6, IL-8, and alpha TNF in the intestinal mucosa. Such increase significantly correlates with the activity of ulcerative colitis through endoscopic examination.

At this moment, forms of therapy for IBD associated with cytokines are being developed, such as ways to inhibit cytokine synthesis, cytokine release, cytokine activity and the cytokine signaling pathway in the target cell.

INTRODUCTION

Inflammatory Bowel Disease (IBD) has been known for a long time in the West, especially in Europe and the United States. There is a high prevalence of IBD in several countries. It seems as though geographic, environmental, ethnic, and socio-cultural factors play an important role in the development of this disease.[1,2]

In Indonesia, IBD is still uncommon. Up to date there is still no available data on the epidemiology or prevalence of this disease, even though cases are being found more often, especially at the Division of Gastroenterology of the Department of Internal Medicine of Cipto Mangunkusumo Hospital, Jakarta. Even though cases are still rare, it is predicted that this disease will be more common, along with advances in diagnostic capabilities in the field of gastroenterology (such as colonoscopy) and the increase in the number of trained personnel.[2]

This literature review proposes to explain one of the many aspects of IBD, among which is the role of cytokines in the pathogenesis of IBD. With more detailed discussion, it is hoped that there is an increase in knowledge and awareness of IBD as a possible gastrointestinal diagnosis. Furthermore, we shall discuss the treatment options for IBD associated with the role of cytokines in the inflammation process that occurs in IBD.

DEFINITION OF INFLAMMATORY BOWEL DISEASE (IBD)

The term Inflammatory Bowel Disease (IBD) is commonly used for two separate disease entities, ulcerative colitis (UC) and Crohn's disease (CD). Up to date, the etiology of this disease is still unclear.[1,2,3] IBD is practically used for an intestinal inflammatory disease other than those with a clear etiology, such as infective colitis, ischemic colitis, and colitis due to radiation.[2]

Ulcerative colitis and Crohn's Disease are chronic inflammatory diseases of the gastrointestinal tract, identified/diagnosed from clinical symptoms, as well as from endoscopic/colonoscopic and histological findings.[1]

The inflammatory response of ulcerative colitis is limited to the mucosa and submucosa, and is usually only located in the colon. On the contrary, the inflammatory response that occurs in Crohn's Disease is more extensive, from the mucosa to the serosa, and could even affect the entire gastrointestinal tract.[1]

**Department of Internal Medicine, Faculty of Medicine of the University of Indonesia/Cipto Mangunkusumo Hospital, Jakarta, Indonesia*

***Division of Gastroenterology, Department of Internal Medicine, Faculty of Medicine of the University of Indonesia/Cipto Mangunkusumo Hospital, Jakarta, Indonesia*

EPIDEMIOLOGY

Up to now there is still no data on the epidemiology of IBD in Indonesia. Data from the Division of Gastroenterology of the Department of Internal Medicine of Cipto Mangunkusumo Hospital, Jakarta in the year 1995, recorded 11 cases of ulcerative colitis and 9 cases of Crohn's Disease out of 70 cases of colitis or out of 211 colonoscopy procedures.[2]

Epidemiological data from Western countries, especially of caucasians in Northern Europe and North America demonstrate an incidence rate of 2-10 cases per population of 100,000 for ulcerative colitis and 1-6 cases per population of 100,000 for Crohn's Disease. The prevalence of Ulcerative Colitis is 35-100 cases per population of 100,000, while the prevalence of Crohn's Disease is 10-100 cases per population of 100,000. The male to female ratio is not very significant, even though there is a tendency for a higher frequency among females. The most frequent age range is from 15 to 25 years. The rate is a little less for blacks than for whites. Jews born and raised out of Israel have a higher rate of IBD, demonstrating a mixed etiological influence of environmental and genetic factors. The incidence and prevalence rate of IBD in Mid and Southern Europe is relatively lower, while the prevalence rate in South America, Asia, and Africa is even less.[1,3]

ETIOLOGY AND PATHOGENESIS

Up to date, the etiology of IBD is still unclear, and there is yet a satisfactory explanation for the population phenomena or geographical distribution of this disease. In general, many authors or researches classify the etiology of IBD into 2 (two) main categories, the first being environmental factors, such as smoking habits; oral contraception; bacterial, viral, and other infections, etc. The second category consists of genetic factors.[1,3]

Environmental Factors

Smoking

Smoking protects oneself against ulcerative colitis. Data shows that the incidence rate for ulcerative colitis is less in smokers, while the incidence rate in ex-smokers is moderate, while non-smokers have a high incidence rate for ulcerative colitis. The opposite is true for Crohn's Disease, where smokers have a much higher incidence rate of Crohn's Disease than non-smokers.[3,4]

Oral Contraception

There is a mild association between the use of oral contraception and the incidence of Crohn's Disease, but there is still inadequate data to prohibit its use of such.[1,3]

Infection

Several studies demonstrate data that support the fact that infection is an etiology towards ulcerative colitis and Crohn's disease. An infection that has a great influence on the incidence of IBD is the measles virus infection. The incidence rate for Crohn's disease is reported to have increased after measles immunization programs in those areas.[1,3]

In addition to that, an infection that has been reported to be associated with an increase in Crohn's disease is infection by *Mycobacterium paratuberculosis*. This microorganism is frequently found in milk and is found in the pathologic examination of patients with Crohn's disease.

Genetic Factors

Several theories explain a strong influence of genetic factors in the pathogenesis of IBD. Most authors admit a strong association of familial ties in the incidence of IBD. A significant risk for IBD is found in a family where there is a positive case of IBD in one of its members. Approximately 15% of IBD patients have other family members with IBD. The incidence rate of IBD for a

Table 1. Colonoscopy Data from the Division of Gastroenterology of the Department of Internal Medicine of Cipto Mangunkusumo Hospital, 1995.

1995	Number of cases	% of all cases of colitis	% of colonoscopy
Ulcerative colitis	11	15.7%	5.2%
Crohn's Disease	9	12.8%	4.3%
Total cases of colitis	70	100%	-
Total colonoscopy	211	-	100%

person whose family member has IBD is 30 to 100 times higher than the normal population.[1,2,3,5]

Up to now, the specific gene carrying the characteristic that makes one more susceptible to IBD is still undefined. However, it is known that the HLA gene class II is associated with Crohn's disease and ulcerative colitis. The DR1/DQw5 haplotype is associated with the incidence of Crohn's disease and HLA-DR2 is associated with the incidence of ulcerative colitis.

PATHOGENESIS[1,3]

The body's immune response plays an important role in the pathogenesis of IBD. The first immune response is when the antigen enters the physical/mechanical system of the intestines (mucose membrane, cilia, etc.). The antigen would be captured and processed (phagocytosis) by macrophages to be destroyed. Even though a great proportion of the antigen is destroyed, a portion remains undestroyed. This remaining antigen would then be presented on the surface of the macrophage and activate the T cell directly or with the help of Interleukin-1 (IL-1). The T-cell assists the macrophage in destroying the antigen by binding the antigen and activating T cytotoxic cells and the T helper cells with the help of Interleukin-2 (IL-2).

As a result of the activation of the T cytotoxic cell and the T helper cell, beside the antigen being destroyed, local tissue is also destroyed. In addition, T helper cells activate the humoral immune system, which is the B cell, to secrete immunoglobulin (especially IgG). Together with macrophage and neutrophil activated by monokine, immunoglobulin G is able to increase prostaglandin and leukotrine production, and activate the complement system, which all together ignite an inflammatory reaction in the form of hyperemia, edema, and even tissue destruction (in the intestinal surface). If the inflammation process continues and becomes chronic, IBD ensues, with its various clinical symptoms.

WHAT ARE CYTOKINES?

Cytokines are glycosylated proteins secreted by various cells, including cells of the immune system (such as

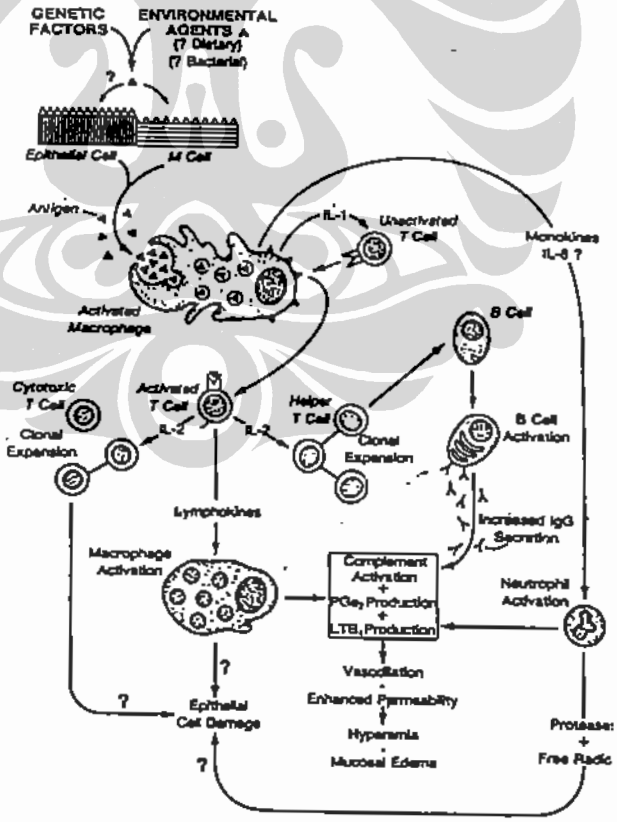

Figure 1. Pathogenesis of IBD.[1]

lymphocytes, macrophages, etc.) and cells that are not from the immune system (such as endothelial cells, epithelial cells, and smooth muscle cells).[1,3,6,7] Cytokines have autocrine, paracrine, and endocrine functions. The effect of each cytokine depends on the expression of the cytokine receptor on the target cell, as well as the differentiation of the exposed cell towards previous cytokines.[1,7]

Several authors classify cytokine function by the cell that produce them. The T helper cell consists of the T helper cell 1 (hT-1), which is more dominant in secreting IL-2, and gamma interferon (gamma IFN), which greatly assist the initial development of the T cell, B cell, and other immune mediator cells. The T helper cell-2 (hT-2) is more dominant in secreting IL-2, IL-4, IL-5, IL-6 and IL-10, which are greatly associated with the humoral immune system.[1]

Another classification categorizes cytokines by their

Table 2. Cytokine Classification.[8]

Cytokine	Source	Target	Effect
Natural Immunity			
IFN α / β	Macrophage (α)	All cells	increased Class I MHC
	Fibroblast (β)		increased NK activity
			reduced viral replication
IL-1	Macrophage	T cell and B cell	Co-stimulator
		Endothelial cell	active phase response
		Hepatocyte	
		Hypothalamus	Fever
IL-6	Macrophage	T cell and B cell	T cell & B growth
	Endothelial cell	Hepatocyte	acute phase response
	T cell		
IL-8	Macrophage	Neutrophil	Activation and chemotaxis
(Chemokine)	Endothelial cell	T cell	
	T cell		
TNF	Macrophage	Endotel	acute phase response
	T cell and NK	Neutrophil,	Fever
		hepatocyte	
	mast cell	hypothalamus	Catabolism
Regulation Lymphocyte			
IL-2	CD4, CD8	T cell	Growth
	T cell	B cell	antibody synthesis
		NK cell	Activation
IL-4	CD4, T cell	T cell	Growth
		B cell	Activation and growth
IL-10	CD4, T cell	CD4, T cell	Inhibition of cytokine production
IL-12	Macrophage	T cell	Growth
	B cell	NK cell	Activation
TGF β	T cell	T cell	Regulation growth
	Macrophage	Macrophage	General inhibition
	Other cells	Other cells	
Activation of Inflammation Cell s			
IFN γ	T cell	Macrophage	Activation
	NK cell	Endothelial cells	Activation
		NK cell	Activation
		All cells	Increased expression of Class I/II MHC
IL-5	T cell	Eosinophil	Activation
		B cell	Growth and activation
Leukocyte Growth			
IL-3	T cell	myeloid progenitor	Differentiation
		B cell	Growth and Differentiation
IL-7	Fibroblast	T cell	Growth
		B cell	Growth and Differentiation
		Macrophage	Activation

association with natural immunity, lymphocyte regulatory function, activation of inflammatory cells, and regulation of leukocyte growth.[8]

THE ROLE OF CYTOKINES IN INFLAMMATORY BOWEL DISEASE

We have discussed earlier that the etiology of ulcerative colitis and Crohn's disease (Inflammatory Bowel Disease = IBD) is still unclear. However, experts believe that the pathogenesis of IBD occurs through a series of immune processes that involve cytokines as mediators of inflammation.

In the human intestine, we encounter what is known as Gut-Associated Lymphoid Tissue (GALT), which protects the bowel from all forms of stimulation from various antigens in the intestinal lumen. If the immune hemostasis of GALT is disturbed, such as deregulation due to unknown foreign substances, the intestines become inflamed. In general, such inflammation occurs for a good cause, since the inflammation cells attempt to assist the elimination of such foreign substances. However, continuous inflammation would cause destruction of surrounding cells.

We know that during such inflammation, there is an increase in pro-inflammatory cytokine cells. **Ishiguro** et al mentioned a significant increase in the production of Interleukin-1 (IL-1), IL-2, IL-6, IL-8, and alpha TNF in the mucosa of patients with ulcerative colitis and Crohn's disease.[9,10] The increase in these cytokines also has a significant correlation with ulcerative colitis activity through endoscopic examination.[11]

The following cytokine act like hormones through receptors on the surface of the following target cells[1,7]:
Directly:

1. More than one effect on various cells (pleotrophy)
2. Autoregulation (autocrine function)
3. On distant cells (paracrine function)

Indirectly:

1. Induction of receptor expression on other cytokines or together with other cytokines in stimulating cells (synergism)
2. Preventing receptor expression or cytokine production (antagonism)

Aside from the production of pro-inflammatory cytokines, cytokine that suppress the inflammation process are also produced, as to establish a balance. Aside from suppressing excessive inflammation, cytokines also fix tissue destruction due to IL-1, IL-2, IL-6, IL-8 and alpha TNF. Such cytokines are the IL-1 receptor antagonist (IL-1ra), soluble alpha TNF receptor (s alpha TNF r), IL-10, and beta TGF.[9,10]

PRO-INFLAMMATORY CYTOKINES INHIBITORS

Interleukin-1 (IL-1)

IL-1 is the first cytokine to be secreted, and initiates the whole cytokine-meidated inflammatory process. IL-1 is produced by monocytes/macrophages, epithelial cells and fibroblasts to activate T cells, to assist macrophages in eliminating foreign matter (antigen). Aside from activating T cells, IL-1 also stimulates chemotaxis from granulocytes and could initiate fever.[9,12,13]

Interleukin-2 (IL-2)

IL-2 is secreted by IL-1 activated T cells. IL-2 activates and stimulates the proliferation of T cells and B cells that would activate the cellular and humoral immune system. In addition, IL-2 also helps in stimulating macrophage activity at the site of inflammation.[13,14]

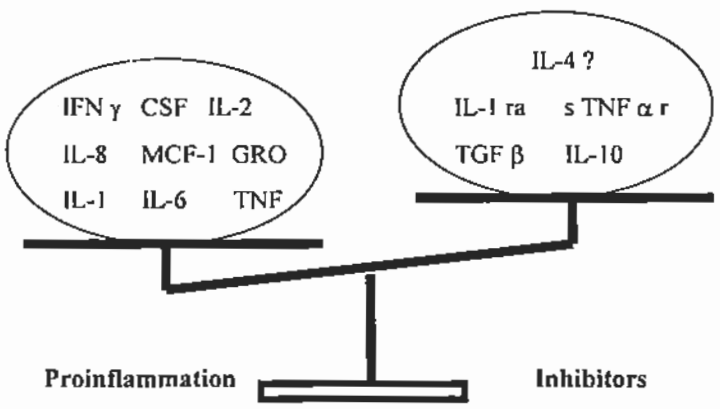

Figure 2. The Balancing Between Proinflammation of Cytokines and its Inhibitors.[8]

Interleukin-6 (IL-6)

Both IL-6 and IL-1 are produced/secreted by monocytes/macrophages and epithelial cells. However, IL-6 differs in function, to stimulate proliferation and B cell differentiation to furthermore activate the humoral immune system in working together in synergism with IL-2 to produce immunoglobulins that bind foreign matter (antigen) to be destroyed.

Interleukin-8 (IL-8)

In the body, IL-8 is secreted by monocytes/macrophages, epithelial cells, granulocytes, and activated endothelial cells. During the inflammation process, IL-8 acts to stimulate chemotaxis from granulocytes and express/facilitate molecular adhesion.[13,16]

Alpha TNF

Alpha TNF is produced by monocytes/macrophages, T cells, and Natural Killer cells (NK cells). Alpha TNF plays an autocrine function by stimulating the macrophage to play a more active role in the inflammation process. Furthermore, alpha TNF also enhances cell metabolism.[13,17]

Interleukin-1 Receptor Antagonist (IL-1 ra)

IL-1 ra could also be called as IL-1 inhibitor, which is a protein size 23-25 kDa. IL-1 ra inhibits IL-1 by blocking receptors on IL-1 target cells. Several studies demonstrate that administration of IL-1 significantly reduces the inflammation response.[9,12,13]

Interleukin-10 (IL-10)

IL-10 has been known to be the cytokine synthesis inhibitory factor (CSIF), in other words acts as an "anti-cytokine". IL-10 produces murine T helper-2 and inhibits the synthesis of cytokines (especially gamma IFN) that is secreted by T helper-1 cells.[6,9,18] **Autschbach** et al reported that IL-10 (production) deficiency is a general cause in the development of IBD.

Soluble Alpha TNF Receptor (s alpha TNF r)

Soluble alpha TNF receptor could also be called alpha TNF inhibitor. Just as IL-1 ra is an IL-1 inhibitor, s alpha TNF acts by blocking receptors that are targeted by alpha TNF. A balanced ratio (near 1) between alpha TNF secretion and r alpha TNF secretion could significantly reduce the inflammation response.[9,13,17,20]

Based on several studies, imbalance between proinflammatory cytokines and inhibitory cytokines play an important role in establishing the degree of inflammation, such as the balance between IL-1 and IL-1 receptor antagonist and alpha TNF and alpha TNF receptor. In other words, the ratio of proinflammatory cytokine secretion and its inhibitor should be approximately one (balanced). But in actuality, in ulcerative colitis and Crohn's disease (Inflammatory Bowel Disease) the secretion of proinflammatory cytokines is always more than inhibitory cytokines. Based on this, scientists reason on the possibility of the role of genetic factors in the development of IBD. In Japan, it has been discovered that IBD is closely associated with the HLA DR2 antigen.

CYTOKINE-ASSOCIATED THERAPY FOR INFLAMMATORY BOWEL DISEASE

Cytokine has been known to play a very important role in the pathogenesis of ulcerative colitis and Crohn's Disease (Inflammatory Bowel Disease). In IBD treatment/therapy, cytokines (particularly proinflammatory cytokines) play an important role in the patient's recovery.

Up to now, experts are still trying to find the best medications for IBD, including by controlling cytokine activity. There are 4 (four) major cytokine-related therapeutic approaches for IBD, namely: [21]

1. Inhibit cytokine synthesis
2. Inhibit cytokine release
3. Inhibit cytokine action
4. Inhibit cytokine intracellular signaling pathway

1. Inhibit Cytokine Synthesis

Up to now, there are many drugs known to inhibit cytokine release, both selectively or inhibiting all cytokines in general. These drugs inhibit genetic transcription or mRNA translation in cytokine-producing cells. In recent years, immunosuppressive agents such as cyclosporine and FK506 have been demonstrated to inhibit the synthesis of lymphokines such as IL-2, IL-3, IL-4, and gamma INF.[1,3,21]

2. Inhibit Cytokine Release

If cytokine transcription or translation inhibition were not possible, another effort would be to prevent formed cytokine from being released by cytokine-producing cells to target cells. Specific enzymes are needed for cytokines such as for pro-inflammatory cytokine IL-1 beta and alpha TNF to be released to target cells. In case of IL-1 beta, the enzyme converses the precursor 33 kDa IL-1 beta into active 17 kDa IL-1 beta that is released to the target cells. This enzyme is called the IL-1 beta Converting Enzyme (ICE). **Sterling Winthrop** and **Vertex** reported the development of a strong IL-1 beta inhibitor in vivo.[21,22]

3. Inhibit Cytokine Action

Cytokine activity can be inhibited by providing or activating its antagonistic cytokine, or with the use of soluble cytokine receptors. Several cytokines have been known to have antagonists that work by blocking target cell receptors. Among these is the IL-1 receptor antagonist (IL-1 ra) for IL-1.[9,21] While known soluble cytokine receptors include soluble alpha TNF receptor for alpha TNF and soluble IL-6 receptor for IL-6.[23] Administration of cytokine antagonist also has its risks. **Anis LA** et al reported that administration of recombinant cytokine therapy could produce a toxicity reaction towards various organs, especially in the skin. Such skin reaction could manifest locally at the site of injection in the form of pruritus, erythema, erythroderma, or even the formation of bullae.[24]

4. Inhibit Cytokine Intracellular Signaling Pathway

After a cytokine binds with a receptor at a target cell, a series of reaction occurs according to the signaling pathway that the cytokine expresses in the target cell. If the signaling pathway in the target cell were inhibited, the target cell is no longer active as expected by the cytokine. Inhibition of signaling pathways can be performed by immunosuppressive agents such as cyclosporine.[21]

CONCLUSION

1. Inflammatory Bowel Disease includes ulcerative colitis and Crohn's disease, which up to now still has no known etiology, but possibly associated with environmental and genetic factors.
2. Cytokines, especially pro-inflammatory cytokines such as IL-1, IL-2, IL-6, and alpha TNF, play an important role in the pathogenesis of IBD.
3. Imbalance between proinflammatory cytokines and their inhibitory cytokine in the intestinal mucosa is an important activity in the development of IBD.
4. Experts try to treat IBD by controlling the role of cytokine starting from inhibiting cytokine synthesis from cytokine-producing cells, up to inhibiting signaling pathways in cytokine target cells.

REFERENCES

1. Stenson WF. Inflammatory bowel disease. In: Yamada T, editors. Textbook of gastroenterologi, 2nd ed. Philadelphia: JB Lippincott Company; 1995. p. 1748-806.
2. Djojoningrat D. Inflammatory bowel disease (Penyakit kolon inflamasi) problem diagnostik di Indonesia. Div Gastroenterologi Bag. IPD FKUI / RSCM. p. 1-8.
3. Kamm MA. Inflammatory bowel disease. 2nd ed. London: Martin Dunitz Ltd; 1999. p. 1-82.
4. Sher ME, Bank S, Greenberg R, Sardinha TC, Weissman S, et al. The influence of cigarete smoking on cytokine levels in patients with inflammatory bowel disease. Inflam Bowel Dis. May 1999. p. 73-8.
5. Lee JCW, Lennard-Jones JE. Inflammatory bowel disease in 67 families each with three or more affected first-degree relatives. Gastroenterol 1996; 111: 587-96.
6. Shaw AR. Molecular biology of cytokine: an introduction. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 19-45.
7. Baratawidjaja KG. Sitokin. In: Baratawidjaja KG. Imunologi dasar. 3rd ed. Jakarta: Balai Penerbit FKUI; 1996. p. 64-75.
8. Stenson WF, Blumberg RS. The imune system. In: Yamada T, editor. Textbook of gastroenterologi. 2nd ed. Philadelphia: JB Lippincott Company; 1995. p. 111-40.
9. Ishiguro Y. Proinflammatory cytokine in IBD. Gastroenterol 1999; 34: 149-51.
10. Rogler G, Andus T. Cytokines in inflammatory bowel disease. Word J. Surg 1998; 22: 382-9.
11. Ishiguro Y. Mucosal proinflammatory cytokine production correlates with endoscopic activity of ulcerative colitis. Gastroenterol 1999; 34: 66-74.
12. Dinarello CA. Interleukin-1. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 47-82.
13. Mac Dermot RP. Alteration in the mucosal imune system in ulcerative colitis and Crohn's disease. Med Clin of Nort Am 1994;78:1207-31.
14. Kuzial WB, Greene WC. Interleukin-2. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 83-102.
15. Hirano T. Interleukin-6. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 169-90.
16. Damme JV. Interleukin-8 and related molecules. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 201-14.
17. Monogue KR, Deventer SJH, Cerami A. Tumour necrosis factor alpha or cachectin. In: Thomson AW, editor. The cytokine handbook. London: Academic Press; 1991. p. 241-56.
18. Nikolaus S, Bauditz J, Gronchetti P, Witt C, Locks H, et al. Increased secretion of proinflammatory cytokines by circulating polymorphonuclear neutrophils and regulation by interleukin-10 during intestinal inflammation. Gut 1998; 42; 470-6.
19. Autschbach F, Braunstein J, Helmke B, Zuna I, Schurmann G, Neimir Z, et al. In situ expression of interleukin-10 in non inflamed human gut and in inflammatory bowel disease. Am J Pathol 1998;153; 121-30.
20. Noguchi M, Hiwatashi N, Liu Z, Toyota T. Secretion imbalance between tumour necrosis factor and its inhibitor in inflammatory bowel disease. Gut 1998;43;203-9.
21. Henderson B. Therapeutic modulation of cytokines. Annals of the Rheumatic Dis 1995;54;519-23.
22. Mc Alindon ME, Hawkey CJ, Mahida YR. Expression of interleukin-1b and interleukin-1b converting enzyme by intestinal macrophages in health and inflammatory bowel disease. Gut 1998;42;214-9.
23. Hosohawa T, Kusugami K, Ina K, Ando T, Shinoda M, et al. Interleukin-6 and soluble interleukin-6 reseptor in the colonic mucosa of inflammatory bowel disease. J of Gastroenterol hepatol 1999;987-96.
24. Asnis LA, Gaspari AA. Cutaneous reaction to recombinant cytokine therapy. J of the Am Acad of Dermatol 1995;33;393-409.