CYTOKINES IN PERIODONTAL HEALTH AND DISEASES

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INTRODUCTION

- Inflammation is mediated by a variety of soluble factors, including a group of secreted polypeptides known as cytokines.
- Cytokines are small soluble proteins produced by a cell that alter the behavior or properties of another cell locally or systemically.
- Cytokine molecule group are interleukins, interferons, growth factors, cytotoxic factors, activating or inhibitory factors, colony-stimulating factors, and intercrines.
- Cytokines play an important role in numerous biological activities including proliferation, development, differentiation, homeostasis, regeneration, repair, and inflammation.
- A dynamic and ever-shifting balance exists between proinflammatory cytokines and antiinflammatory components of the human immune system.

DEFINITION

- ***Steve offenbacher 1996*** defined cytokines as mediator molecules, which direct and regulate inflammation and wound healing. The term cytokine meaning the cell protein is reserved for molecules, which transmit information or signals from one cell to another. It is part of a fundamental cell-to-cell communication network.

- Cytokines previously called as ***Lymphokines*** (cytokines produced by lymphocytes) or ***Monokines*** (cytokines produced by Monocytes). However it is now known that there is considerable overlap, e.g. TNF-, IL-6 are made by both cell types and so this nomenclature is not in wide use..

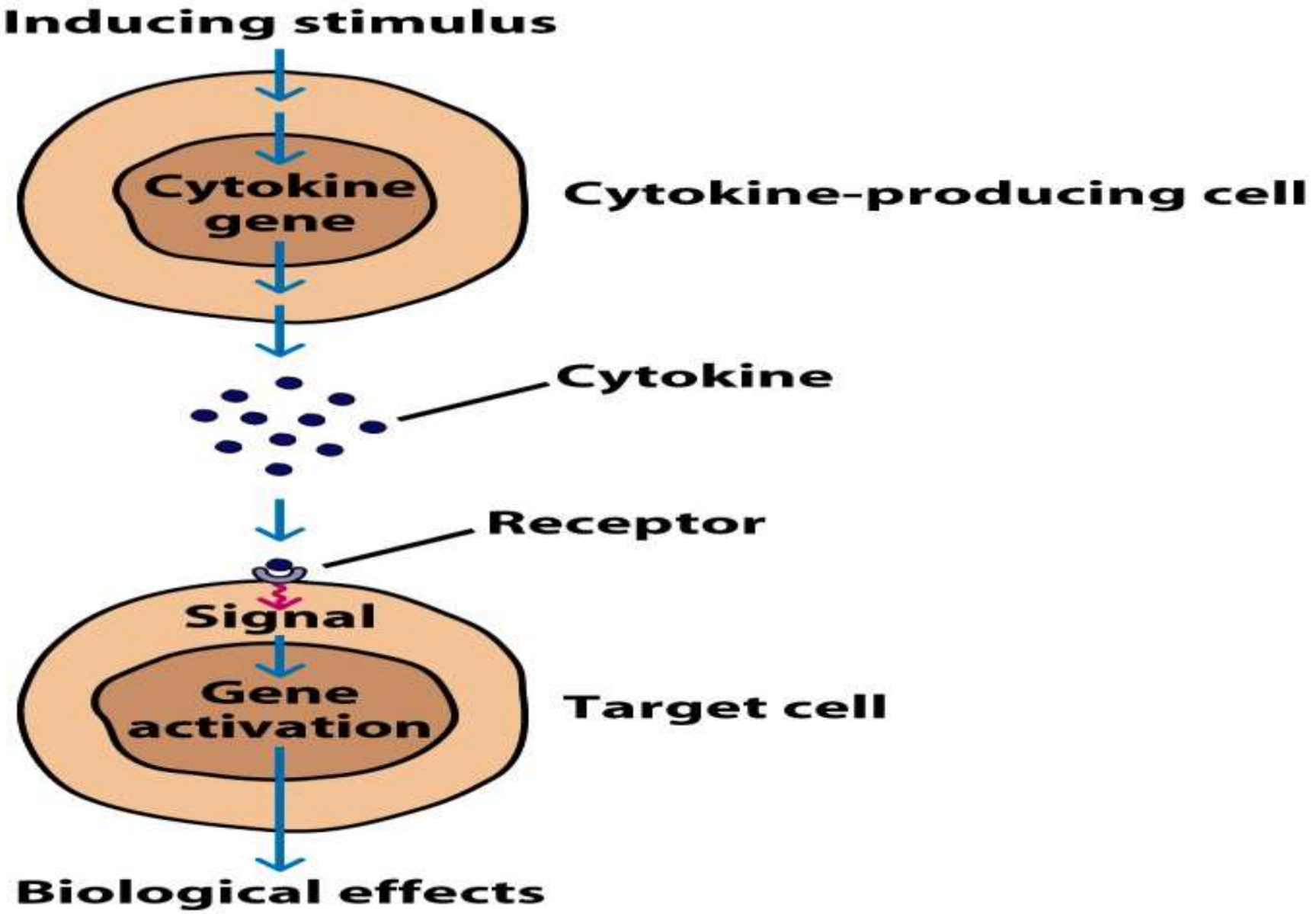


Figure 12-1a
Kuby IMMUNOLOGY, Sixth Edition

Antigens

↓

Induce stimulus of cytokine producing cells

↓

Releases cytokine

↓

Cytokine binds the receptors present on the target cell

↓

Gene activation occurs in the target cell

↓

which releases secondary mediators (i.e., MMPs and PGE_2)

↓

These mediators are responsible for loss of connective tissue and bone resorption

INTERLEUKINS - Molecular messengers acting between leukocytes.

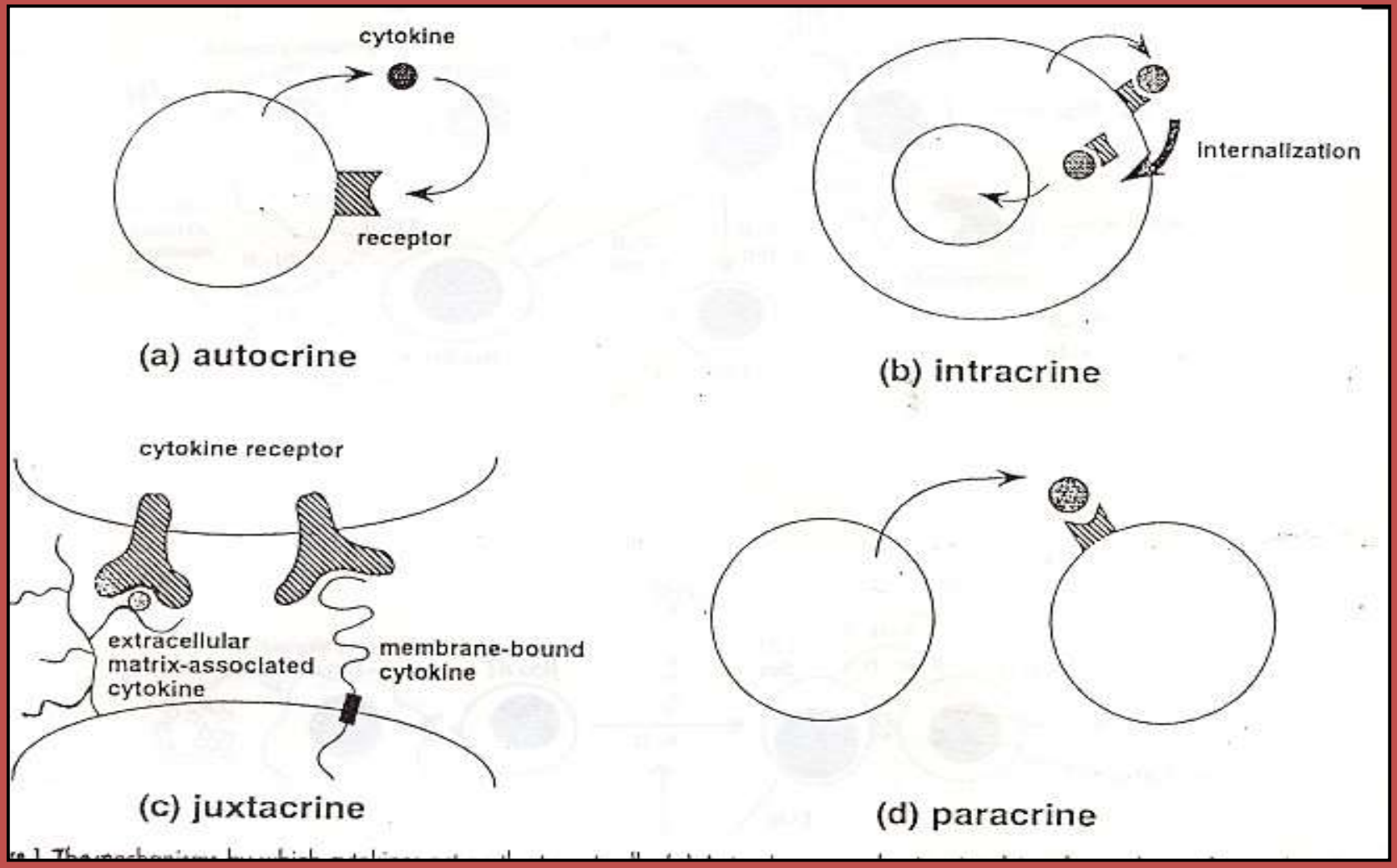
INTERFERONS - Molecules with non-specific antiviral effects.

COLONY STIMULATING FACTOR - Direct the division and differentiation of bone marrow cells and precursor of blood leukocytes.

CHEMOKINES - Direct the movement of leukocytes around the body, from blood stream into appropriate location within tissues.

GROWTH FACTORS - Naturally occuring proteins that function in the body to promote the proliferation , directed migration and metabolic activity of cells.

- cytokines act on the target cells are classified into four types: autocrine, intracrine, juxtacrine, and paracrine (Idelgaufts, 1995)

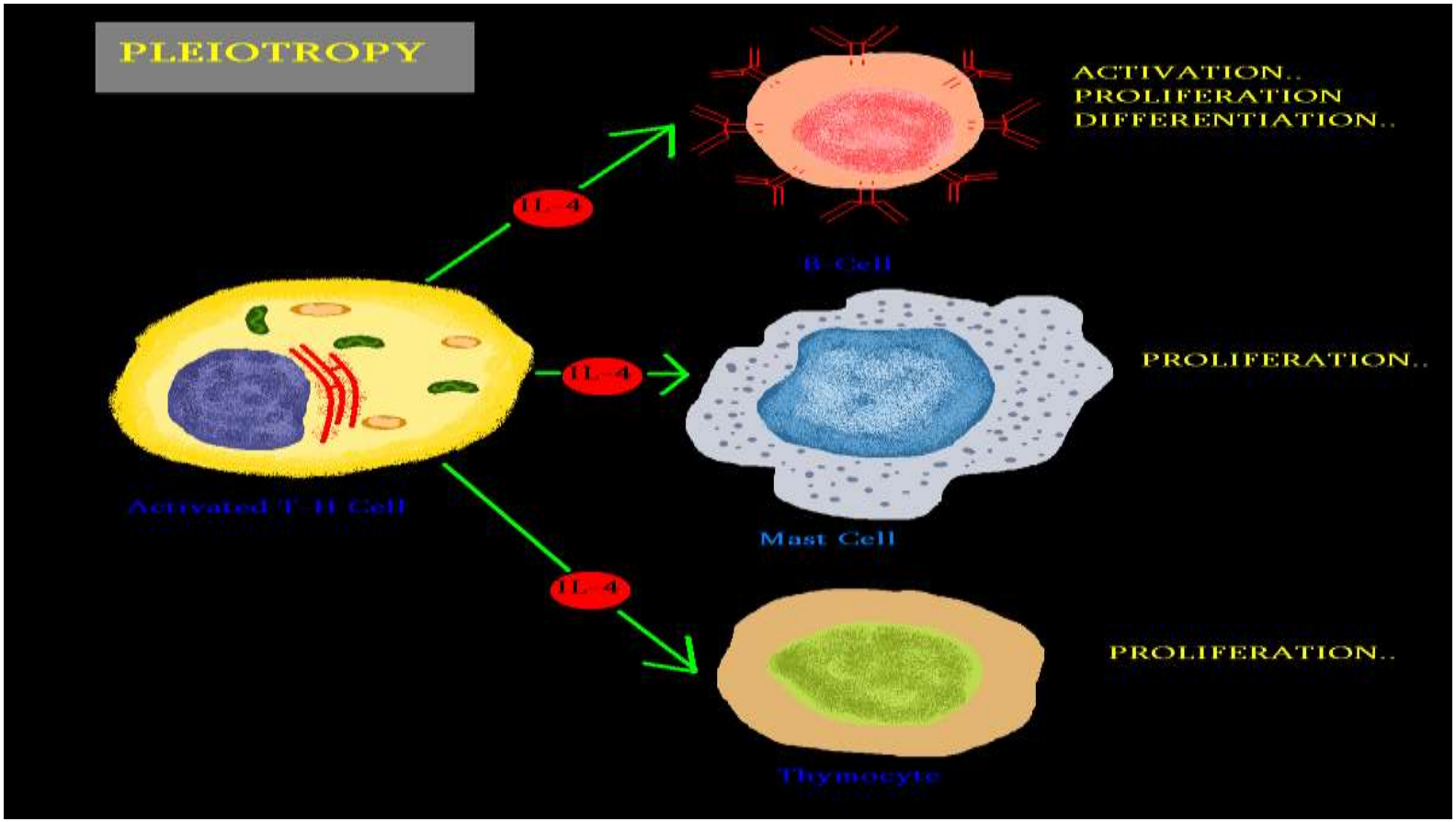


FUNCTIONS

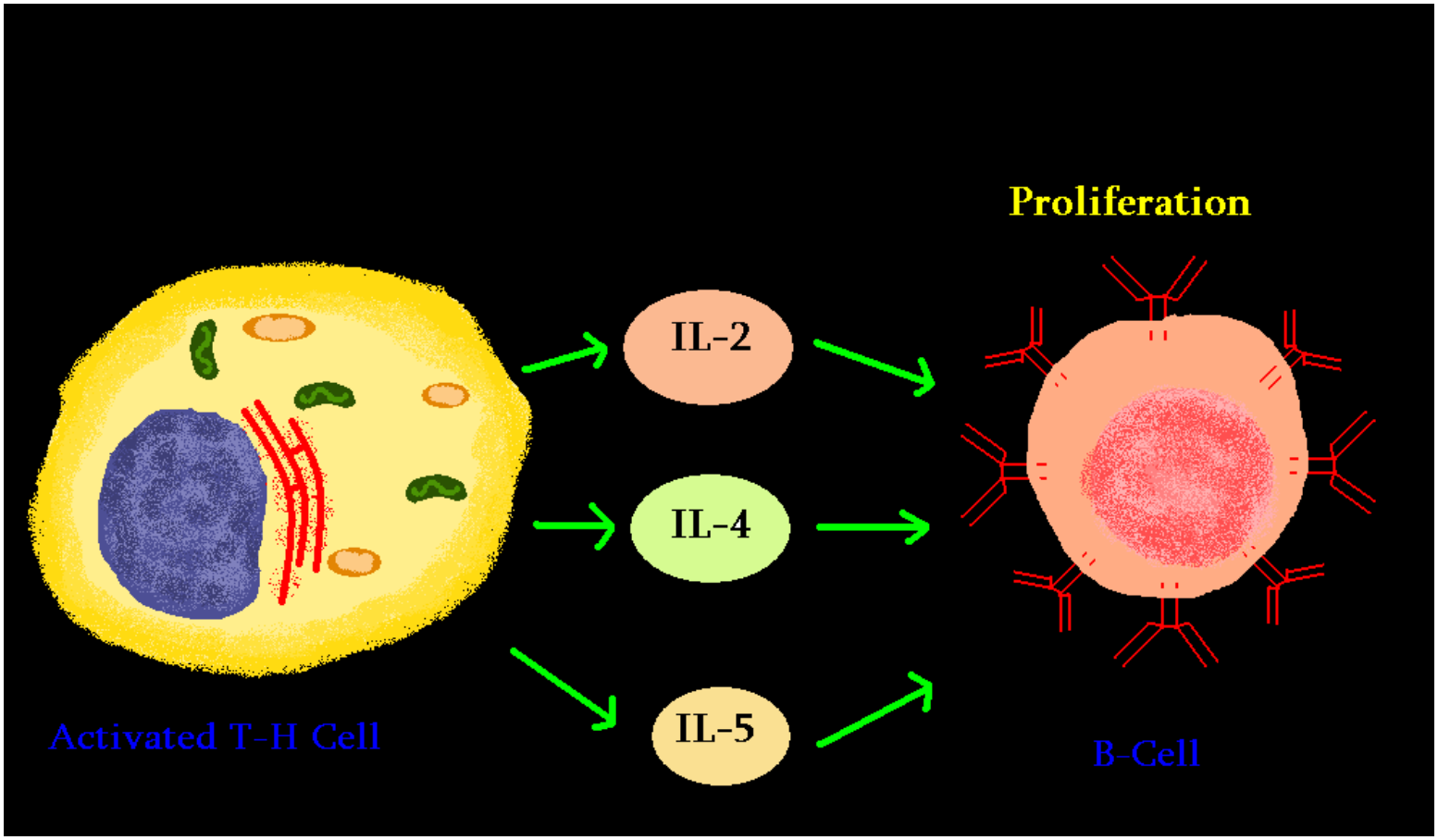
- Cytokines mediate natural immunity
- Cytokines regulate lymphocyte activation growth, and differentiation
- Cytokines mediate immune mediated inflammation
- Cytokines stimulate hematopoiesis

PROPERTIES

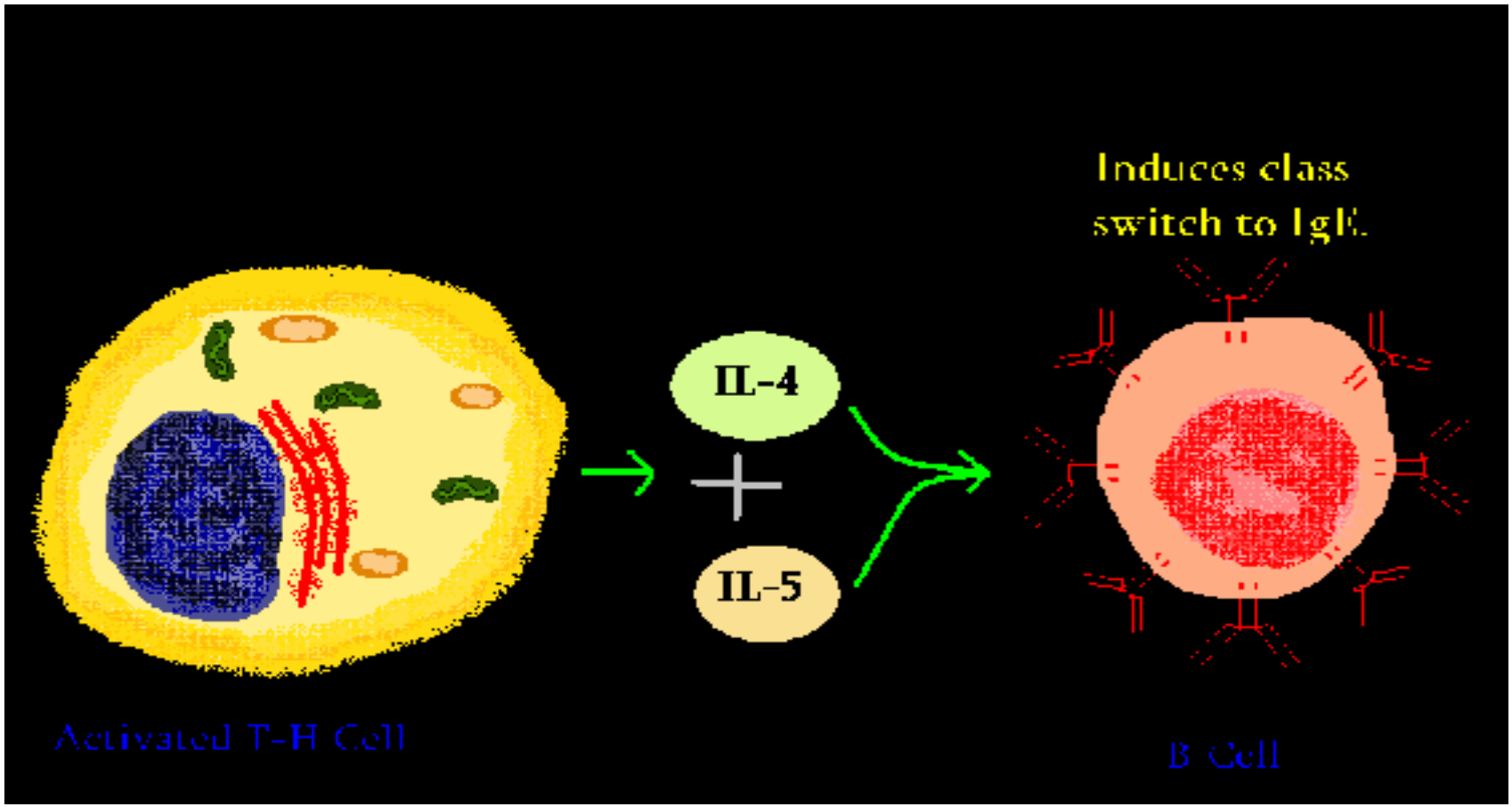
- Pleiotrophy -Different biological effects on different target cells



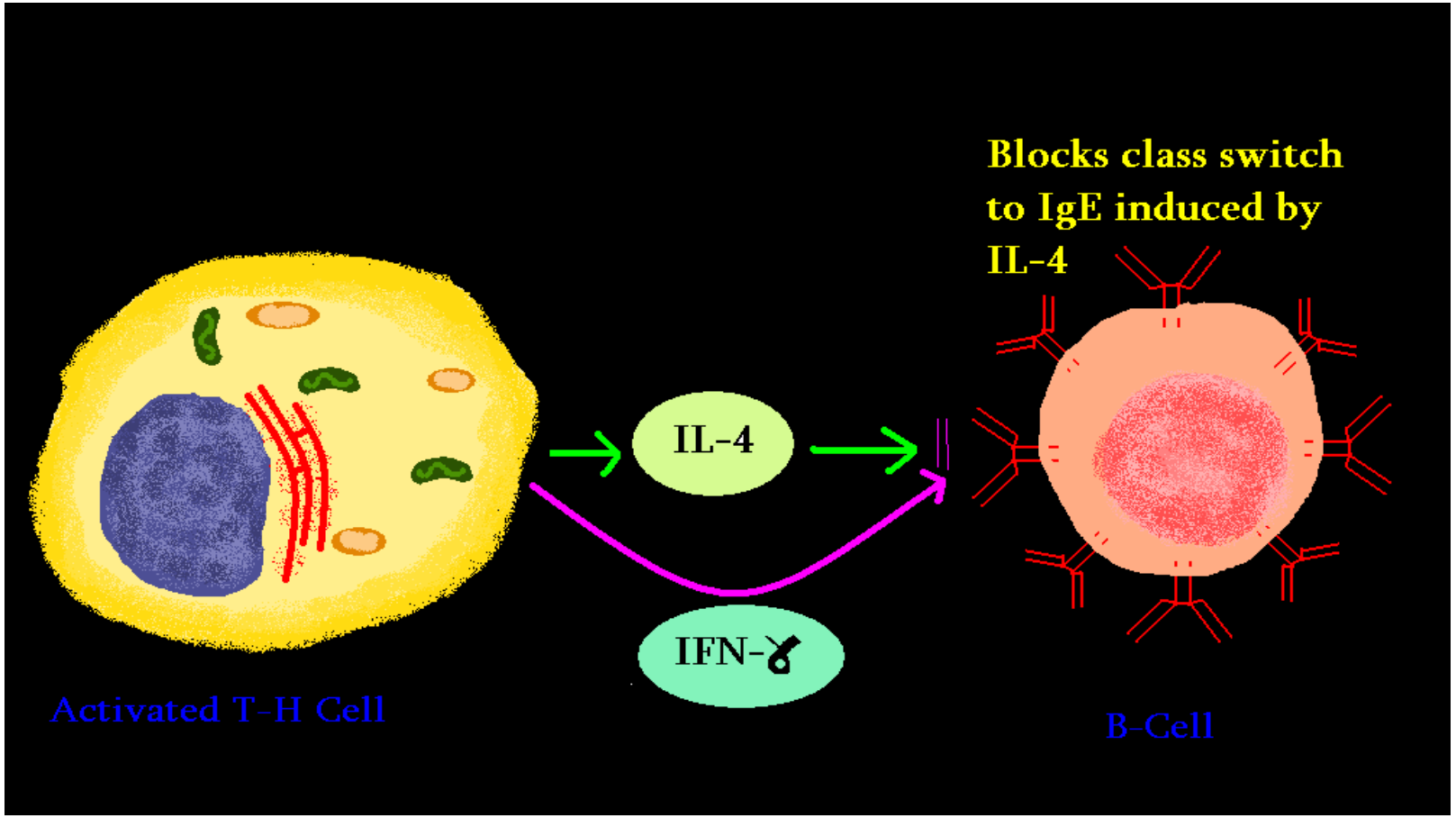
- Redundancy-Different cytokines often evoke similar biologic responses



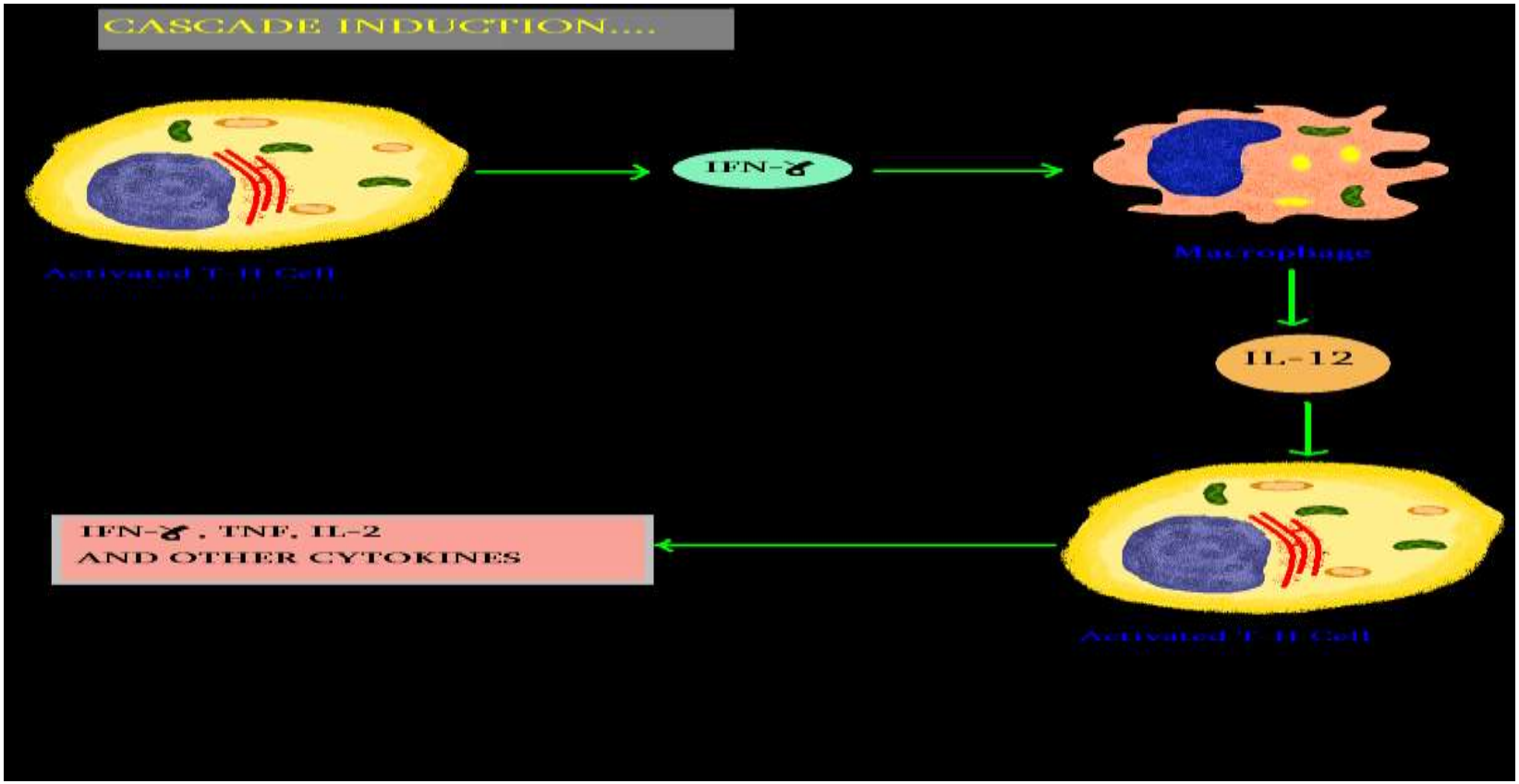
Synergy-Combination of two cytokines on cellular activity is greater than the additive effects of individual cytokines



- Antagonism- Effect of one cytokine inhibit the effects of another cytokine



Cascade Induction-when action of one cytokine on a target cell induces that cell to produce one or more other cytokines ,which in turn may induce other target cells



STRUCTURE

- Cytokines exhibit wide variety of primary structures, sizes, post transitional modification and conformation.
- They are glycoproteins of molecular weight in the range of 5000-10,000 D (15-25KDA).
- Some cytokines are monomeric proteins, whereas others are covalent / non-covalent dimers, trimers.
- X-ray crystallographic techniques revealed that there are relatively few 3 dimensional (tertiary) structures into which the polypeptide chains of different cytokines are folded.

CYTOKINE RECEPTORS AND RELATED MOLECULES

- Cytokines require specific cell surface receptors through which to mediate their range of actions on different cells.

- Frequently the action of a cytokines on a cell wall, include the up regulation of surface expression of its receptor as well as release of the molecule itself.

- Receptors vary in their form, some being single and other multiple chain complexes.

- **Cytokine Receptor Super Family** - Receptors for cytokines such as IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-9, IL-12, granulocyte stimulating factor and granulocyte-macrophage colony stimulating factor belong to this family.

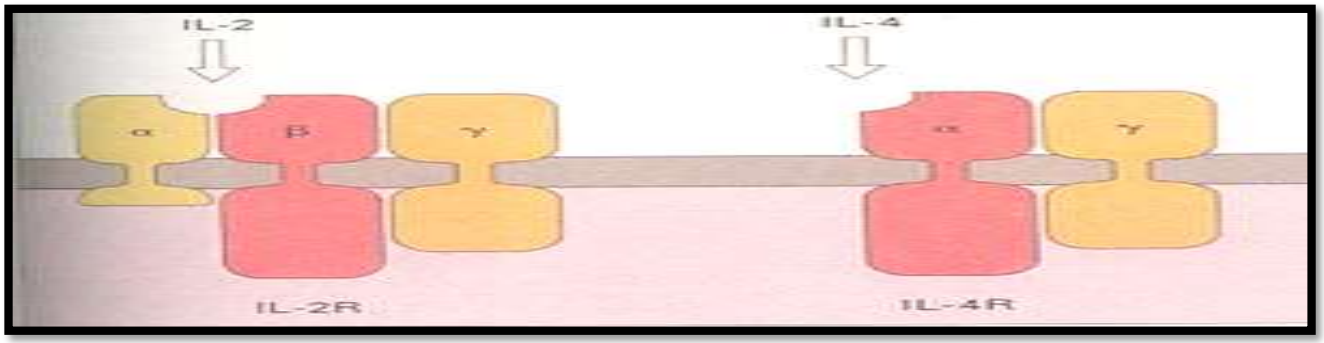
- **Immunoglobulin Super Family** - Receptors for all IFN types in addition to the two IL-1 receptors, and M-CSF.

- **Haemopoietic Growth Factor Family** - IL-2 receptor belongs to this chain.

- **Tumor Necrosis Factors (TNF) Family** - TNF-α, TNF- β and a number of related cytokines bind to this family of receptors.

- **Seven Transmembrane Glycoproteins Family** – Chemokines

- **Cytokine Receptors** *:* Two polypeptide chains
  Alpha chain for cytokine binding and signal transduction
  Beta chain for signaling but with only a minor role in binding**.**



cytokine binds to its receptor on the cells

↓

induces aggregation of receptor components

↓

cytoplasmic regions of subunits interact to initiate signaling cascade

↓

tyrosine phosphorylation and activation of janus kinase (jaks)or ras /map kinase pathway

↓

tyrosine phoshorylation of signal transducers and activators of transcription

↓

stat dimmers migrate to nucleus and bind to the enhancer region of gene

CLASSIFICATION

JAN LINDHE

Proinflammatory cytokines

E.g: IL-1, IL-6 and TNF

Chemotactic cytokines

E.g: IL-8

Lymphocytes signaling cytokines

E.g: Cytokines released by Th1, IL-2, IFN.

Cytokines released by Th2, IL-4,IL-5,IL-10 and IL-13.

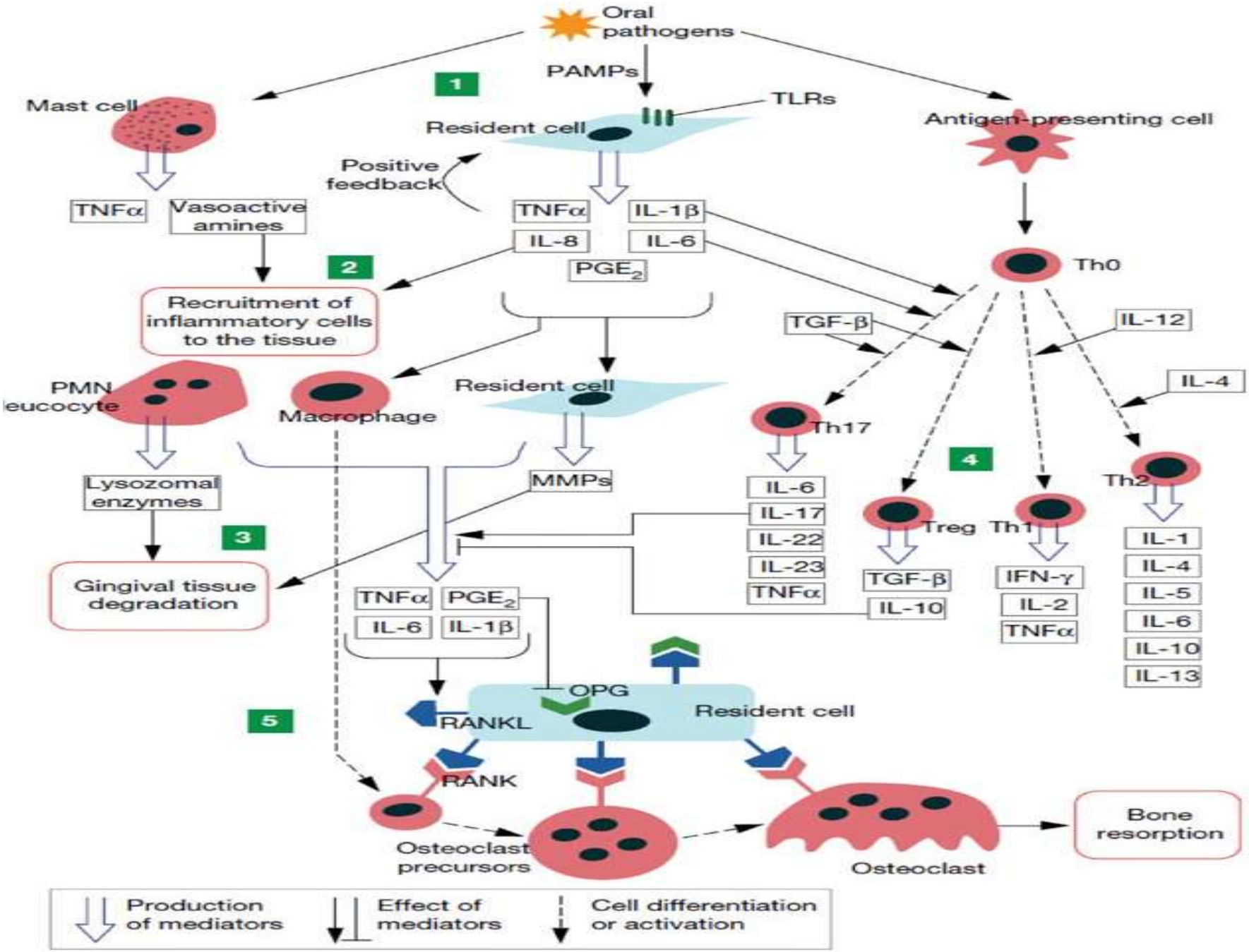
NIESENGARD AND NEWMAN

First group: Cytokines, which serve as mediators of innate immunity.
E.g. Interferon alpha and beta, TNF and IL-6.

Second group: Cytokines that regulate the growth and differentiation of lymphocytes.
E.g. IL-2, IL-4 and TGF-beta

Third group: cytokines regulate the hematopoietic activity in the bone marrow and are collectively referred to as Colony Stimulating Factors or CSF's produced by stromal cells or antigen stimulated T- lymphocytes.
E.g. GM-CSF, G-CSF, M-CSF, MULTI- CSF, IL-3, IL-7.

Fourth group: Cytokines which share the common property of being activators of inflammatory cell function.
E.g. Interferon α


Oral pathogens
PAMPs
1
Mast cell
TLRs
Resident cell
Antigen-presenting cell
Positive feedback
TNFα
Vasoactive amines
TNFα
IL-1β
IL-8
IL-6
PGE2
Th0
2
Recruitment of inflammatory cells to the tissue
TGF-β
IL-12
IL-4
PMN eucocyte
Macrophage
Resident cell
Th17
4
Th2
Lysozomal enzymes
MMPs
IL-6
IL-17
IL-22
IL-23
TNFα
Treg
Th1
IL-1
IL-4
IL-5
IL-6
IL-10
IL-13
3
Gingival tissue degradation
TNFα
PGE2
IL-6
IL-1β
TGF-β
IL-10
IFN-γ
IL-2
TNFα
OPG
RANKL
Resident cell
5
RANK
Osteoclast precursors
Osteoclast
Bone resorption
Production of mediators
Effect of mediators
Cell differentiation or activation

PRODUCTION

- Most of them are T cell products, produced by activation by presentation of antigen to T cell

- Small amounts but bind to receptors with high affinity

- The cell types involved in production of cytokines in periodontal destruction are

- MACROPHAGES
- T & B LYMPHOCYTES
- GINGIVAL FIBROBLASTS
- GINGIVAL EPITHELIAL CELLS
- ENDOTHELIAL CELLS

MACROPHAGES

- 30% of infiltrating cells in periodontal lesions
- Oppenhiem 1981 – to produce TNF α , INF α IL 1,6 ,10,12& 13
- Production of IL1 at inflammation site
- Matsuki – IL 1 α & IL1β mRNA expression cells in inflamed gingiva are produced by macrophages

T & B Lymphocytes

- Helper T cells secrete cytokines
- In PDL lesions these cytokines not only involve in protection but rather cause onset and progression of disease
- T cells – IL 1 ; TNF
- In established lesions the majority of infiltrating cells are B cells
- B cell population → production of IL1 → tissue destruction

GINGIVAL FIBROBLASTS

- They are the most abundant cells in the periodontal ligament
- When activated by stimuli they secrete -IL 1 α , IL 1 β , IL 6 , TNF α

GINGIVAL EPITHELIAL CELLS

- They are the first barriers for the invading foreign substance
- Isolated cells have expressed - IL 1 β , IL 6 , IL 8 , TNF α
- They secrete monocyte chemoattrant protein 1 (MCP 1)

ENDOTHELIAL CELLS

- In vitro studies showed that endothelial cells can secrete cytokines (including various chemokines such as IL-8), IL-1, and IL-6.

- when the cells were stimulated with LPS, TNF-alpha , IL- 1, IL-4, IL- 13, or the IL-6-soluble IL-6 receptor complex(Mantovani et al., 1997).

CYTOKINE EXPRESSION IN PERIODONTAL HEALTH

- Tissue homeostasis represents a delicate balance between anabolic and catabolic activities.

- The regulations of migration, proliferation, and differentiation of resident cells and of the production of tissue matrix in a healthy state are major aspects of periodontal tissue homeostasis.

- cytokines, which are secreted by fibroblasts (Moscatelli et al., 1986), endothelial cells, and epithelial cells, play a crucial role in tissue homeostasis.

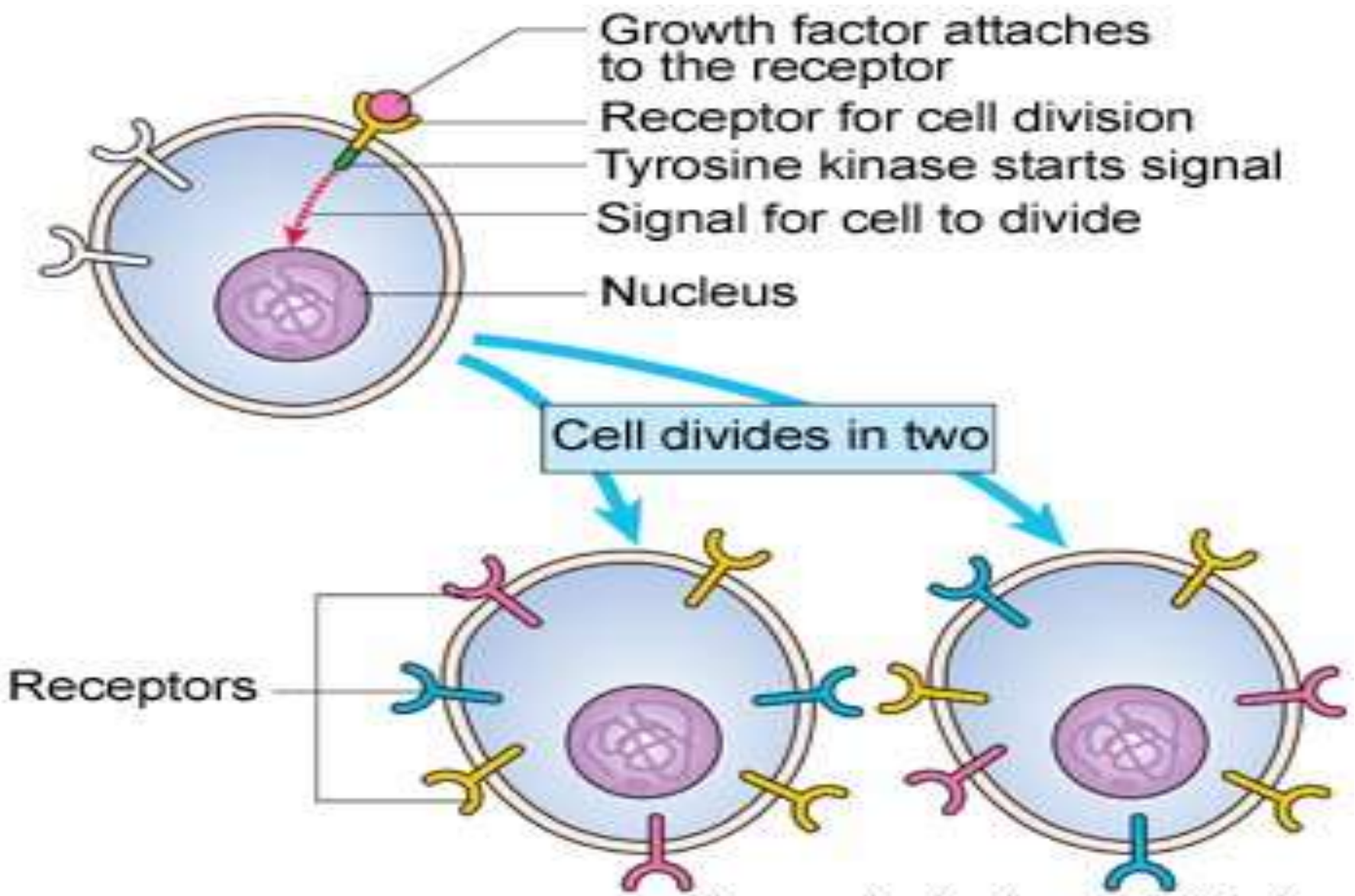


Diagram showing how growth factors signal to the cell to grow and divide

FIBROBLAST GROWTH FACTOR (FGF)

- Fibroblast growth factors (FGF) is one of the well-characterized cytokine families that can be found in many tissues.

- Two of the nine isoforms of FGF have been characterized in some detail: one is acidic FGF (aFGF; FGF-1) and the other is basic FGF (bFGF; FGF-2). Both FGFs bind to heparan sulfate, heparin, and fibronectin in the extracellular matrix.

- Acidic fibroblast growth factor (aFGF) is primarily known for its effect on endothelial cell replication and neovascularization.
- In bone tissue cultures, aFGF stimulates DNA synthesis and cell replication, which results in increased protein synthesis (Idelgaufts, 1995), especially of collagen type 1.
- Like aFGF, bFGF has angiogenic properties and is highly chemotactic and mitogenic for a variety of cell types.

PLATELET-DERIVED GROWTH FACTOR

- PDGF, which was originally detected in the α-granules of platelets, is a potent growth factor for various connective tissue cells.
- Other cell types also synthesize PDGF including macrophages, endothelial cells, fibroblasts, astrocytes, myoblasts, and smooth-muscle cells.
- PDGF functions as a local autocrine and paracrine growth factor.
- PDGF is a powerful promoter of cell migration and proliferation.

INSULIN-LIKE GROWTH FACTORS (I AND II)

- Two different IGFs (IGF-I and IGF-II). Both were isolated initially as serum factors with insulin-like activities that could not be inhibited by anti-insulin antibodies.

- The structure of both IGFs is homologous to human pro-insulin.

- In periodontal research, it was shown that IGF-I is chemotactic and mitogenic for PDL cells.

- IGF-I only slightly induces periodontal tissue regeneration, several lines of evidence suggest that IGF-I combined with other growth factors such as bFGF, PDGF, and TGF-3 may augment the osseous wound-healing process.

TRANSFORMING GROWTH FACTOR-B

- TGF-β appears to be synthesized by all normal cells .
- The different isoforms of TGF-β (TGF-β1, TGF-β2, and TGF-β3) are encoded by different genes.
- TGF-β is the most potent known growth inhibitor for epithelial cells, endothelial cells, fibroblasts, neuronal cells, lymphocytes, and hepatocytes.
- It stimulates the synthesis of connective tissue matrix components, such as collagen, fibronectin, proteoglycan, glycosaminoglycan, osteonectin, and osteopontin in many cell types, including PDL cells.
- It also inhibits the degradation of matrix proteins by inhibiting the synthesis of metalloproteinases such as collagenase and by increasing the synthesis of proteinase inhibitors

CEMENTUM-DERIVED GROWTH FACTOR

Cementum-derived growth factor (CGF) was detected exclusively in cementum and was shown to be the major cementum mitogen for PDL cells and gingival fibroblasts.

CGF may promote the migration and growth of progenitor cells present in structures adjacent to the dentin matrix and participate in their differentiation into cementoblasts.

- mRNA expression of cytokines in clinically healthy gingival tissues by reverse-transcription/ polymerase chain-reaction (RT-PCR), mRNA expression of a variety of growth factors-such as epidermal growth factor (EGF), platelet-derived growth factor(PDGF), and transforming growth factor-r (TGF-1)-was observed because these cytokines have been relatively well-characterized through the research of periodontal regeneration. (T. Nozaki et al)

- IL-1,IL-6, and TNF was also detected in the clinically healthy gingival tissues, although their density was relatively low compared with that in the inflamed sites.

CYTOKINES AND PERIODONTAL DISEASE

INTERLEUKINS

- The term interleukin signifies substances produced by leukocytes (Leukin) that regulate other (inter) cells.

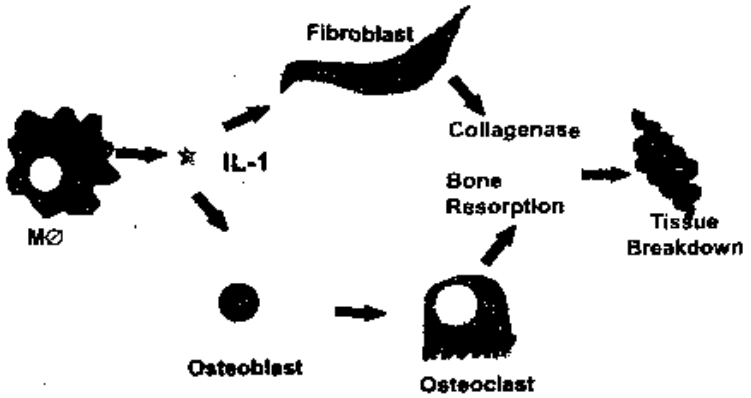
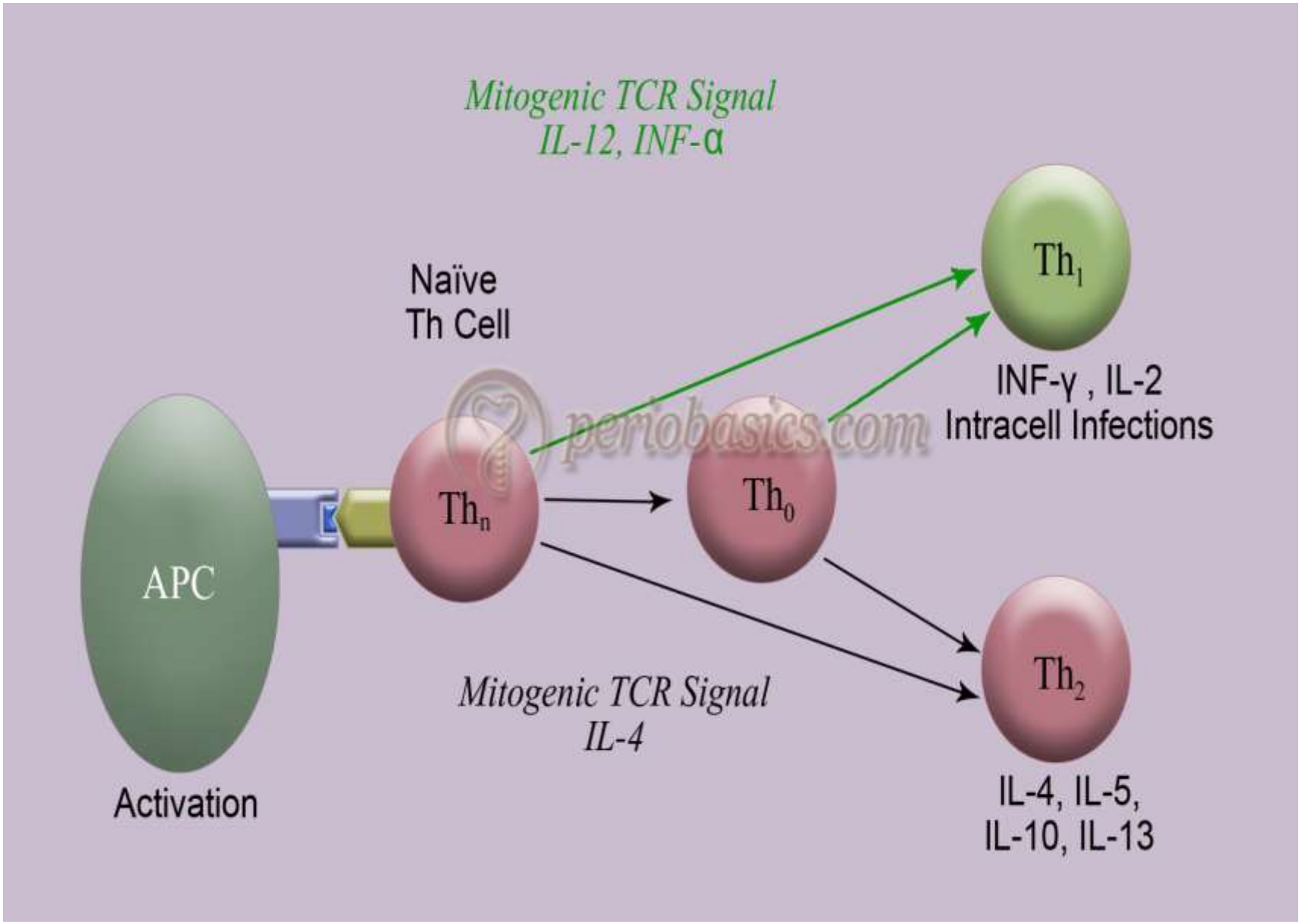
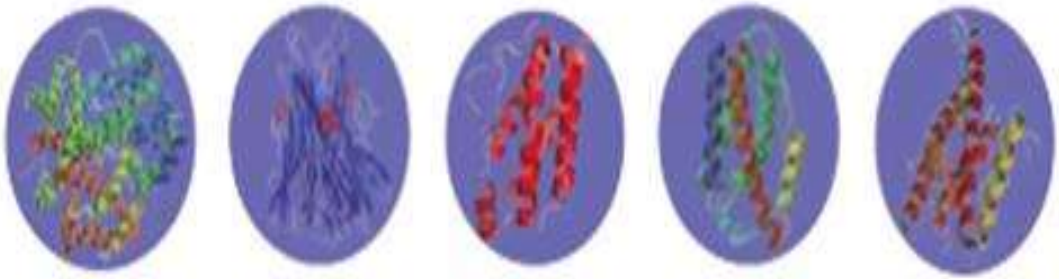


Fig. 7. IL-1 produced by macrophages (MØ) as the major mediator of tissue breakdown in periodontal disease


Mitogenic TCR Signal
IL-12, INF-α
Naïve
Th Cell
Th1
INF-γ , IL-2
Intracell Infections
periobasics.com
Thn
Th0
APC
Mitogenic TCR Signal
IL-4
Th2
Activation
IL-4, IL-5,
IL-10, IL-13

Pro-inflamatory Cytokines : stimulate the immune system

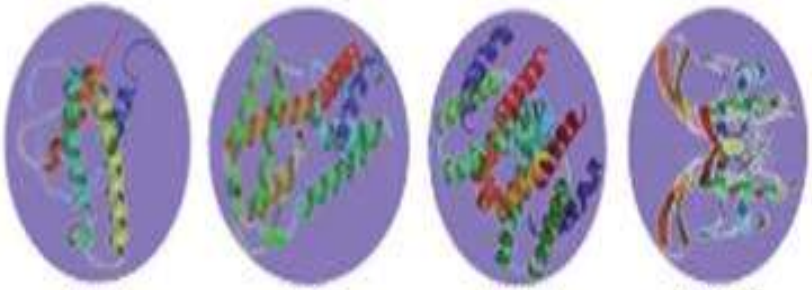
Th1



INF-γ TNF-α IL-2 IL-6 IL-12

Anti-inflamatory Cytokines : supress the immune system

Th2



IL-4 IL-5 IL-10 TGF-β

Crystallography derived from RCSB Protein Data Bank.[1]

INTERLEUKIN-1

- IL-1 was discovered by Gery *et al*. in 1972

- synthesized by - macrophages, monocytes, lymphocytes, vascular cells, brain cells, skin cells, and fibroblasts.

- IL-1 is known to stimulate the proliferation of keratinocytes, fibroblasts, and endothelial cells and to enhance fibroblast synthesis of type I procollagen, collagenase, hyaluronate, fibronectin, and prostaglandin E2 (PGE2).

- IL-1 has been suggested to play a key role in promoting alveolar bone destruction in periodontal disease.

- There are three IL-1 ligands, IL-1β, IL-α, and IL-1 receptor antagonist (IL-1ra). IL-1α and IL-1β have similar biological activity, while IL-1ra binds to IL-1 receptors, but does not have agonist activity and acts as a competitive inhibitor.

- The pro-inflammatory effects of IL-1 can be inhibited by IL-1 receptor antagonist (IL-1Ra), originally referred to as IL-1 inhibitor.

- IL-1ra is produced by monocytes and macrophages.

- The anti-inflammatory cytokines IL-4, IL-5, IL-10, and IL-13 inhibit the synthesis of IL-1b, yet they stimulate the synthesis of IL-1ra.

- Kornman KS et al to periodontal disease is influenced by genetic polymorphism of IL-1 gene. association between IL-1 and severity of periodontal disease genotype.

- Laine ML et al In a meta-analysis, it is demonstrated that IL-1α and IL-1β genetic variation are significant contributors to chronic periodontitis

INTERLEUKIN-2

- IL-2 (α andβ) was originally called T-cell growth factor because of its effect on mitogen or antigen activating T-cells and is known to play a general role in immune responses.
- IL-2 also stimulates macrophage functional activity, modulates natural killer function and induces natural killer proliferation.
- It is secreted by Th cells and NK cells.
- It is increased in periodontal tissue in periodontitis.

INTERLEUKIN-3

- It is secreted by activated helper T cells, NK cells.
- Stimulates the formation of mast cells, neutrophils, macrophages, eosinophils, and megakaryocytes.
- Possesses neurotrophic activity, and may be associated with neurologic disorders.

INTERLEUKINS-4

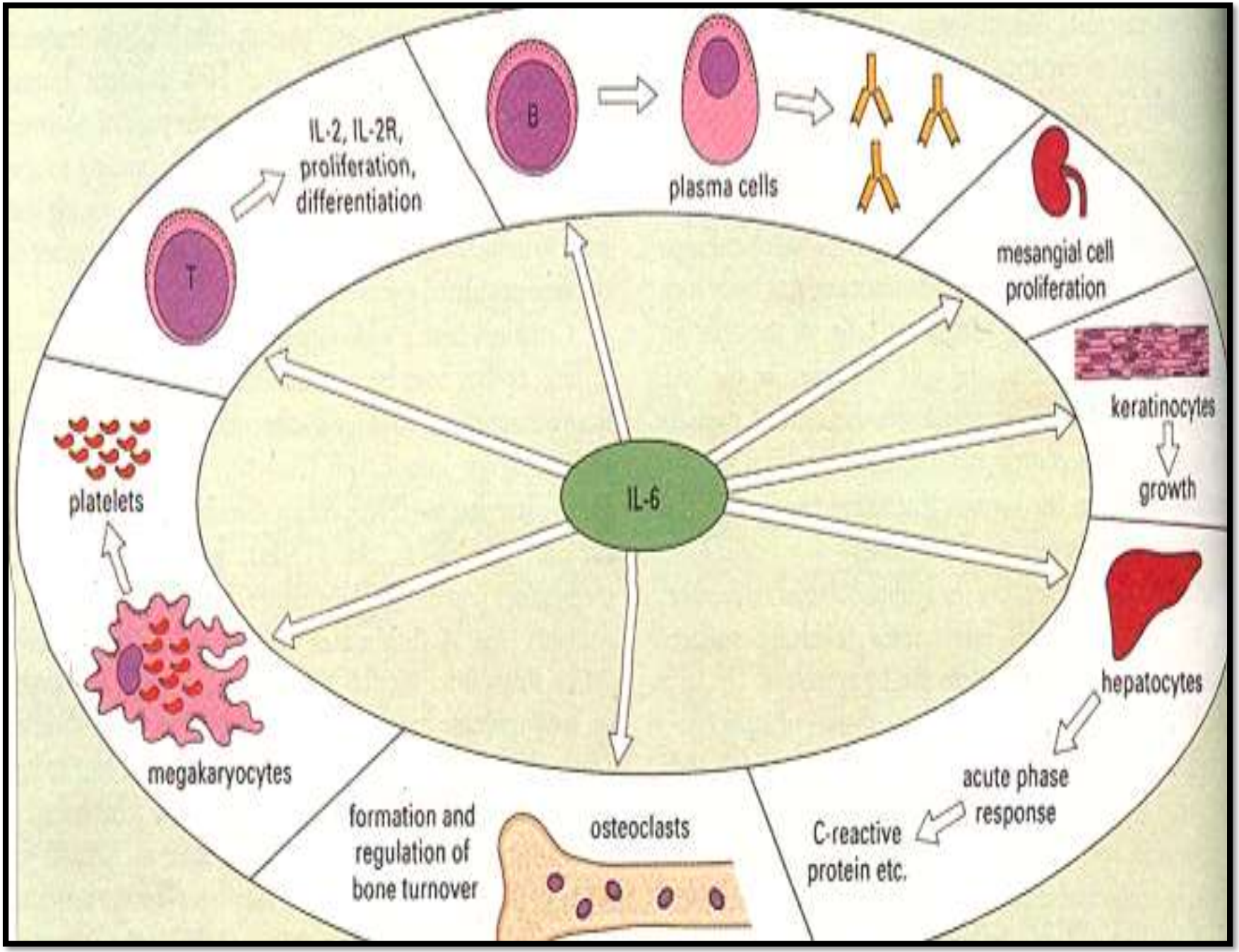
- originally called T-cell-derived B-cell growth factor (BCGF-1)
- Also called as migration inhibition factor, secreted by helper Tcells.
- Role in the activation, proliferation and differentiation of B cells, T-cell growth, macrophage function, and growth of mast cells.
- Induces IgE synthesis by B cells
- Receptors found on T-cells, B-cells, mast cells, myeloid cells, fibroblasts, stromal cells, endothelial cells and monocytes.
- Its receptor also binds to IL13, contributing to overlapping functions with IL13.
- It suppresses the synthesis of proinflammatory cytokines, such as IL-1, IL-6, IL-8 and TNF- α and activated monocytes.

INTERLEUKIN -5

- Produced by lymphocytes
- Coffman - IgA enhancing factor, eosinophil colony stimulating factor.
- The beta subunit of its receptor is shared with the receptors for IL3 and CSF.
- Stimulate the production and function of eosinophils. The elevated production is reported to be related to asthma.
- Enhances the activities of basophils by priming them to release mediators such as histamine in response to other signals.

Interleukins-6

- Formerly known as B- cells stimulatory factor II interferon B2 and plasmacytoma growth factor.
- produced by activated monocytes or macrophages, endothelial cells, activated T-cells, and fibroblasts.
- induction of the final maturation of B-cells into immunoglobulin-secreting plasma cells(serum IgG levels can rise 120-400-fold).
- Tamura, 1993 simultaneous treatment of mouse osteoblastic cells and bone marrow cells with IL-6 and soluble IL-6 receptor strikingly induced osteoclast formation


IL-2, IL-2R,
proliferation,
differentiation
T
B
plasma cells
mesangial cell
proliferation
keratinocytes
growth
IL-6
platelets
megakaryocytes
hepatocytes
acute phase
response
C-reactive
protein etc.
formation and
regulation of
bone turnover
osteoclasts

- **Reinhardt 1993** - detected elevated level in the GCF of patients with refractory periodontitis.

- **Gillespie,2007-** increases in sites of gingival inflammation and plays a role in bone resorption via induction of RANKL.

- **Saxilin, 2009** - serum IL-6 was associated with periodontal infection.

- **Nibali, 2008-** reported association between IL6 polymorphisms and (AgP).

- **Riccardo 2016-** Modest evidence of association has been found between interleukin-6 gene polymorphisms and periodontal disease.

- Meta-analysis showed no association between the IL-6 (–572) GG genotype and periodontitis

INTERLEUKIN -7

- Formerly known as lymphopoitin 1 based on its capacity to influence early lymphopoiesis.
- Secreted by thymus, spleen and bone marrow stromal cells that functions as a growth factor for T and B cells precursors.
- Enhances the function of mature activated lymphocytic cells, particularly those with cytotoxic activity.
- It induces T cell proliferation by expressing IL2.

INTERLEUKIN -8

- Is chemotactic for neutrophils and increases its adherence to endothelium
- They contribute to inflammatory response
- Produced by – T cells
  Endothelial cells fibroblasts
  Platelets

- **Kurdowska 2003-** Increased IL 8 detected in Chronic periodontitis anti IL8: IL8 complex present in 90%chronic and 50% refractory pts .

- **Kim 2009-** SNP (rs4073) in the IL8 gene is not associated with susceptibility to periodontitis in Brazilian individuals

INTERLEUKIN-9

- It is a T cell growth factor which acts in synergy with other cytokines
- Secreted by IL2 activated T cells and Hodgkin`s lymphoma cells

INTERLEUKIN-10

- Produced by TH2 cells , CD8 T cells ,activated B cells.
- Called cytokine synthesis inhibitory factor for its ability to inhibit production of cytokines by T cells and NK.
- Inhibits antigen presenting capability of monocytes.
- It synergizes with other to stimulate proliferation of B cells and mast cells.
- Along with TGF – IgA by B cell.
- Down regulates IL1
- **Anti-inflammatory effects:** inhibits the secretion of other proinflammatory cytokines, suppresses phagocytosis, oxidative burst, intracellular killing.

Gonzales 2002 - genetic polymorphisms at the interleukin-10 loci are not associated with aggressive and chronic periodontitis.

Sumer 2007 - IL-10 gene polymorphism at position -597 seems to be associated with severe generalized CP.

Claudino, 2008 - IL-10 promoter-592 polymorphism decrease the expression TIMP-3, and OPG expression in periodontitis.

INTERLEUKIN-11

- Secreted by bone marrow and stromal cells
- Down regulates IL1
- Stimulate the T-cell-dependent development of immunoglobulin-producing B cells.
- Stimulates and increases in numbers and size of megakaryocyte colonies in combination with IL3.
- May play an important role in in vitro platelet production.

INTERLEUKIN-12

- Called as cytotoxic lymphocyte maturation factor (CLMF).
- B cells and macrophages.
- IL12 + IL2 = INF by T cells and NK.
- Develops Th1.
- Suppress Th2 functions.
- induce production of – CSF , TNF , IL 2 and IL 16
- Orozco 2006 - Very little IL-12 was detected in the gcf at both gingivitis and periodontitis sites with levels decreasing with increasing inflammation.

INTERLEUKIN-13

- Structural homology between IL-4 and IL-13
- Expressed in activated Th2 cels
- Regulates B cell and monocytes activity
- Masada 1990 detected more frequently than IL-l in GCF from untreated patients with periodontitis.
- Wilton 1993 IL- 13 concentrations did not correlate with GCF volume or clinical measurements of plaque index, bleeding index.
- Gonzales 2007 the IL-13 polymorphisms not associated with AgP.

INTERLEUKIN -14

- known as B cell growth factor.
- It is produced by follicular dendritic cells and activated T cells.
- role in the development of B cell memory.
- It enhances the proliferation of activated B cells and inhibits the synthesis of immunoglobulin.
- Its receptors are found only in cells of the B cells lineage.

INTERLEUKIN-15

- Widely expressed in liver , kidney , heart , lung , bone marrow.
- Produced by monocytes and epithelial cells
- Role in T cell generated immune response

INTERLEUKIN-16

- Lymphocyte chemoattractant factor.
- Produced by lymphocyte.
- Inhibitor of hiv replication.
- Induces the migration of cd4+ t cells, eosinophils and monocytes.

INTERLEUKIN-17

- Key cytokine produced by newly identified subset of Th cells called Th17 lymphocytes.
- It is important in stimulating chemokines to recruit neutrophils.
- Receptor (IL-17R) composed of two subunits, IL-17RA and IL-17RC.
- Stimulate the expression of IL6 and cyclooxygenase-2 as well as enhance the production of nitric oxide (NO).
- It shares properties with IL1 and TNF and mediate bone destruction.
- Yu,2007 found that when IL17 signal was ablated in mice there was greater periodontal bone loss in response to P.g.

INTERLEUKIN -18

- Secreted by activated T cells and macrophages.
- Induces INF production.
- Increase cytotoxicity of NK .
- Noack , 2008 rejected the hypothesis that functionally relevant IL-18 have a major effect on aggressive periodontitis susceptibility.

INTERLEUKIN-19,20

IL-19

- Produced by monocytes
- Modulate Th1 activity

IL-20

- Produced by monocytes and keratinocytes
- Regulate inflammatory responses to skin.
- Induces keratinocyte differentiation and proliferation.

INTERLEUKIN-21,22

IL-21

- Produced by Th1 cells

Regulate

- Haematopoiesis
- NK cell differentiation
- B-cell activation
- T-cell co-stimulation

IL-22

- Source is T-cells
- Inhibits IL-4 production

INTERLEUKIN-23,24

IL-23

- Produced by dendritic cells
- Induces proliferation of INF-gamma by Th2 cells

Saraiva AM et al 2013 -Insufficient studies exist that correlate IL-23 polymorphisms with AgP. Existing studies show that there is no significant trend in association of IL-23 polymorphisms with AgP

IL-24

- Produced by monocytes,melanocytes,vascular smooth muscle,NK cells and B-cells
- Inhibits melanoma cell growth
- Induces IL-6 and TNF-alpha by monocytes

INTERLEUKIN-25

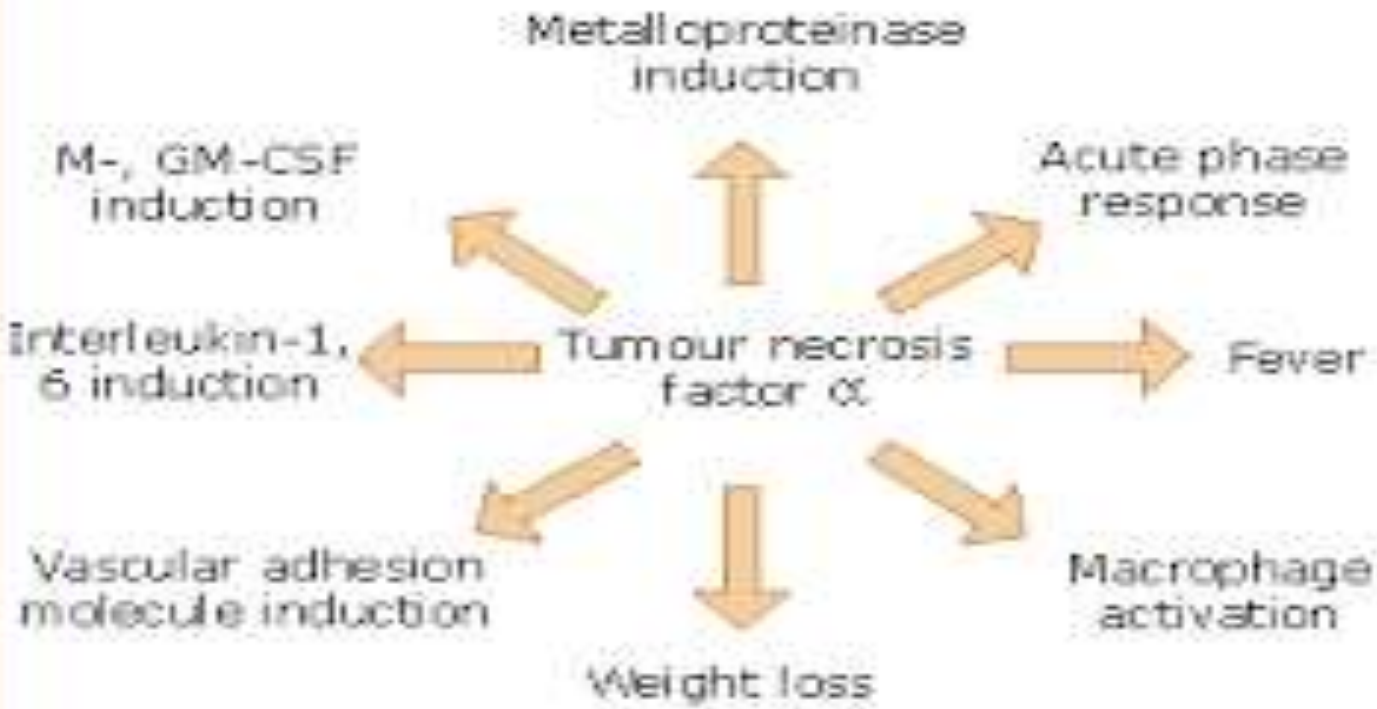
- Induces secretion of Th1 cytokine profile.
- Source is unknown

INTERLEUKIN-35

- IL-35 could have an important role in suppressing periodontal inflammation and maintaining periodontal health.

Fig. 1

Actions of tumour necrosis factor α



TUMOUR NECROSIS FACTOR

- T-cells play a crucial role in regulating a variety of immune responses by secreting various cytokine, formerly known as lymphokine

- There are two structurally and functionally similar forms of TNF α and β but they differ biochemically.

- TNF α is 17 KD is derived from stimulated macrophages and appears to have significant stimulatory activity on the cytoxic T lymphocytes (CTL) responsible for lysing tumor or virally infected cells.

- TNF-β is a 25 KD glycoprotein derived from activated T cells with a 28% homology to TNF-α

TUMOUR NECROSIS FACTOR- ALPHA

- Derived from stimulated macrophages
- Have significant stimulatory activity on the cytotoxic T cells responsible for lysing tumor or virally infected cells.
- Induces the adhesion molecules and chemokines on the endothelium
- Induces apoptosis in susceptible cells
- It acts by causing the activation of NFkB which is a master switch of immune system.
- TNFα aids leukocytes in their ability to adhere to endothelial cells and increases their phagocytosis and chemotaxis.

TUMOUR NECROSIS FACTOR- BETA

- Derived from activated T cells
- Occasionally known as lymphotoxin
- osteoclast activation and antiviral activity.

Functions (At low conc.)

Causes vascular endothelial cells to express adhesion molecules

Activates leukocytes and mononuclear phagocytes to produce cytokines

Interferon like protective effect against viruses

Functions (At high conc.)

An endogenous pyrogen

Acts on hepatocytes to increase synthesis of acute phase proteins

Activates coagulation system

Suppresses stem cell division

- **Galbraith 1998** - The periodontitis patients polymorphism of 308 of the TNFα gene, had higher TNF-α production by oral PMNs.

- **Meikle 1989** - demonstrated that gingival fibroblasts stimulated in vitro with TNF- α are able to degrade collagen.

- **Gillespie 2007-** It can induce osteoclastogenesis by both RANKL dependent and independent manner

INTERFERONS

- Interferons (IFNs)- are natural proteins produced by the cells of the immune system of most vertebrates in response to challenges by foreign agents such as viruses, bacteria, parasites and tumor cells.

- Interferons belong to the large class of glycoproteins known as cytokines.

- Interferons assist the immune response by inhibiting viral replication within other cells of the body.

- Interferons have been divided into two major classes according to the type of receptor through which they signal:

- Type I or viral interferon - further divided into alpha and beta subcategories.

- Type II or immune interferon - referred to as gamma interferon.

Lundqvist 1994 - reported IFN- Gamma - adult periodontitis

.

Type I interferons

IFN-α is also called leukocyte interferon

Sources: Fibroblasts, T-lymphocyte, macrophages

Functions:

- Inhibits viral replication
- Inhibits cell proliferation
- Increases lytic potentials of NK cells
- Modulates MHC expression

Type II interferon -IFN-gamma

Source: T-lymphocytes, NK cells (immune type)

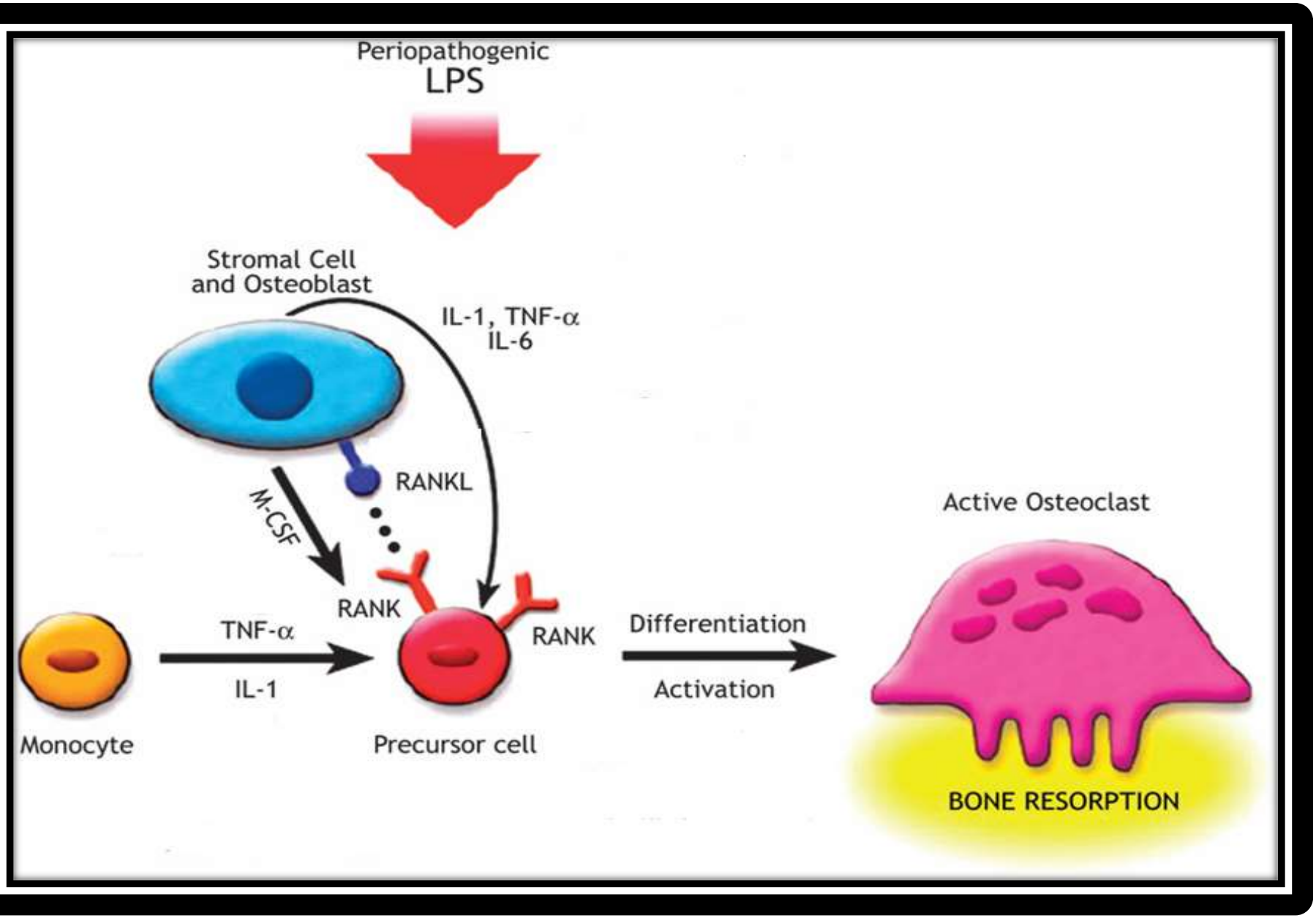
Functions:

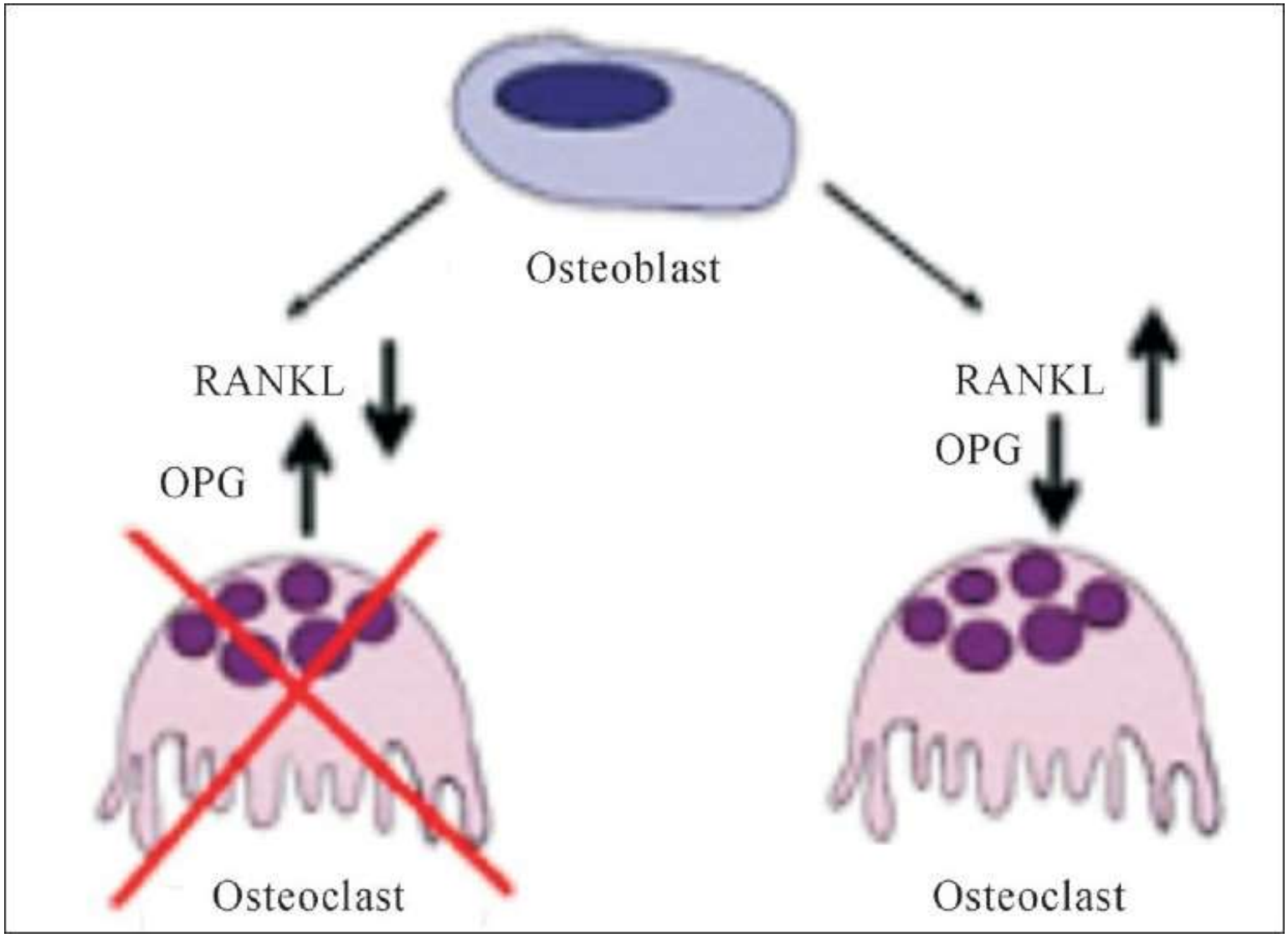
- Potent activator of mononuclear phagocytes
- Increases MHC expression
- Promote T & B lymphocyte differentiation
- Up regulates neutrophil respiratory burst
- Promotes T lymphocyte adhesion and extravasation

RANK, RANKL

RECEPTOR ACTIVATOR OF NUCLEAR FACTOR-KAPPA B LIGAND-RECEPTOR ACTIVATOR OF NUCLEAR FACTOR-KAPPA B-OSTEOPROTEGERIN AXIS

- RANK, RANKL, and OPG are cytokines that belong to TNF-α super family.
- RANK is a receptor found on the surface of osteoclast precursors.
- When RANK binds to its ligand RANKL, it stimulates the differentiation of these precursor cells into mature osteoclasts.
- OPG (osteoprotegerin)competes with rankl by binding to rank without stimulating any differentiation. it is the ratio of rankl and opg expressions that is important in inflammation-induced bone resorption, including periodontitis


Periopathogenic
LPS
Stromal Cell
and Osteoblast
IL-1, TNF-α
IL-6
RANKL
M-CSF
RANK
RANK
TNF-α
IL-1
Monocyte
Precursor cell
Differentiation
Activation
Active Osteoclast
BONE RESORPTION


Osteoblast
RANKL
OPG
RANKL
OPG
Osteoclast
Osteoclast

ROLE OF CYTOKINES IN BONE UNCOUPLING

- The inflammatory process that leads to osteoclastogenesis and bone resorption may also be responsible for the failure to form adequate amount of new bone, i.e., inflammation causes uncoupling of bone formation following bone resorption.

- Osteoblast survival is a key factor in bone formation.

- TNF-α stimulates the production of Dickkopf-1 (DKK-1), which suppresses bone formation by inhibiting the WNT (wingless WNT/beta catenin) pathway.

- DKK-1, a negative regulator of WNT pathway, is up-regulated by TNF stimulation through TNF-1 receptor and p38 mitogen-activating protein kinase signaling. The up-regulated DKK-1 not only promotes bone resorption but also blocks bone formation and repair in the diseased joint

CHEMOKINES IN PERIODONTAL DISEASE

- They generally have low molecular weights, ranging from 7-14 KDa, and stimulate recruitment of leukocytes.

- Chemokines are secondary proinflammatory mediators, i.e., they are typically induced by primary proinflammatory mediators such as Interleukin –1 or TNF.

- By recruitment of leukocytes, chemokine activity leads to activation of host defense mechanisms and stimulates the early events of wound healing.

- The chemokine family is divided into the following types

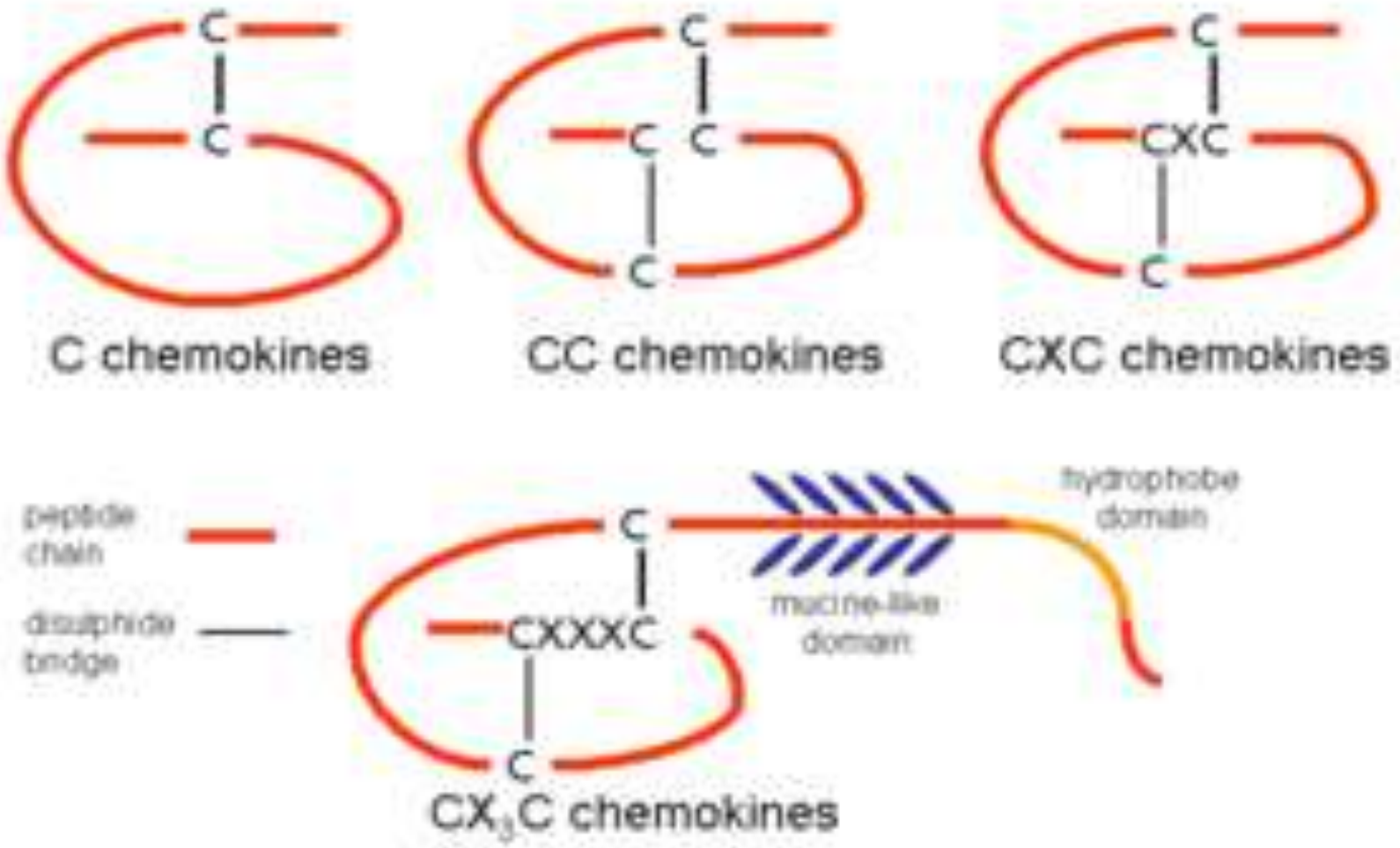
C Subfamily

CC Subfamily

CXC Subfamily

CX3C Subfamily

Structure of chemokine classes



- The two major chemokine subfamilies are based upon the portion of cystine residues i.e. CXC and CC.

- The CXC family members also known as the alpha Chemokines. They primarily stimulate neutrophils.

- C Chemokines are also known as the beta Chemokines. They stimulate Basophils, eosinophils, T-lymphocytes and NK cells.

- **Yu,2007-** examined that CXCR2 deficient mice infected with P.g showed increase in periodontal bone loss.

- **Silva,2007**- Some chemokines contribute to inflammation induced bone resorption as they can stimulate recruitment, differentiation or fusion of precursor cells to form osteoclasts or enhance osteoclast survival.

SPECIFIC CYTOKINES IN PERIODONTAL DISTRUCTION

- IL 1 α - Bone resorption and proliferation of B cells.
- IL 1 β - bone resorption.
- IL 2 - activation of macrophages and proliferation of T cells.
- IL 6 - T cell proliferation and B cell differentiation.
- IL 8 - T cell attraction and chemotatic migration.
- TNF -stimulate osteoclastic bone resorption and activate macrophages.
- TGF β – stimulates bone resorption.
- INF γ - activates T cells ; inhibit B cells and activates macrophages

ASSAYS FOR CYTOKINES

The assays to detect cytokines in laboratory include

- ELISA
- IRMA – Immuno radiometric assay
- Bioassay
- Radio Receptor Binding Assay
- **Instant ELISATM**

CYTOKINE THERAPY

- Anticytokine therapy for periodontal diseases especially targets proinflammatory cytokines, that is, TNF-α, IL-1, and IL-6, because these are essential for the initiation of the inflammatory immune reaction and are produced for prolonged periods in periodontitis.

- Neutralization of cytokines

- Blockage of cytokine receptors

- Activation of anti-inflammatory pathways, such as, immune-suppressive