

Biological Response Modifiers

T.E.G.K.Murthy², K. Sri Janaki^{1*}, S.Nagarjuna¹, P.Sangeetha¹, S.Sindhura¹

¹Department of Pharmacology, Sri Padmavathi School of Pharmacy, Tiruchanoor, Tirupathi, Andhra Pradesh, India.

²Department of Pharmaceutics, Bapatla College of Pharmacy, Bapatla, Andhra Pradesh, India.

*Corres. Author: ksrijanaki@gmail.com
Phone No: 9052368870

Abstract: Immune responses to infectious agents involve a complex set of interactions between cells and the factors they produce that culminate with disease resolution or death. Therefore, the manipulation of the immune system may have a great impact on the preservation and restoration of animal health. Biological response modifiers are agents that modify the host's response to pathogens with resultant beneficial prophylactic or therapeutic effects. The use of biological response modifiers or biologicals has rapidly expanded since the introduction of the first diagnostic antibodies; they are now widely employed in oncology, autoimmune disorders, inflammatory diseases and transplantation medicine. Biological response modifiers can also act passively by enhancing the immunologic response to tumor cells or actively by altering the differentiation/growth of tumor cells. Their widespread use has resulted in an increase in adverse drug reactions. Adverse effects result from both direct pharmacological actions and immunological actions, as well as through induction of a specific immune response. This article reviewed the role of interferons, interleukins, monoclonal antibodies, erythropoietin, tumor necrosis factor and colony stimulating factors as biological response modifiers. This review also sheds light on the various angiogenesis inhibitors, humanized antibodies, the increasing use of erythropoietin in anemia and the upcoming potential colony stimulating factors used to treat the infections in cancer patients.

Keywords:- Biological response modifiers, Peritubular cells, Cytokines, Glycoproteins, Neutropenia, Macrophages, White blood cells.

INTRODUCTION

Biological response modifiers are intrinsic products to the body that alter immune defenses to enhance, direct or restore the body's ability to fight disease. The body naturally produces small amounts of these substances¹. Biological response modifiers used in biological therapy include interferons, interleukins, monoclonal antibodies, erythropoietin, tumor necrosis factor and several types of colony-stimulating factors (Granulocyte Macrophage Colony Stimulating Factor, Granulocyte Colony Stimulating Factor).

INTERFERONS

Interferons are a group of proteins called cytokines produced by white blood cells, fibroblasts, or T-cells as part of an immune response to a viral infection or other immune trigger. The name of the proteins comes

from their ability to interfere with the production of new virus particles².

TYPES OF INTERFERON

TYPE I interferon

Interferon-alpha (leukocyte interferon) is produced by virus-infected leukocytes^{3,4}. Interferon-beta (fibroblast interferon) is produced by virus-infected fibroblasts, or virus-infected epithelial cells^{3,4}. Interferon-alpha (a family of about 20 related proteins) and interferon-beta are particularly potent as antiviral agents. They are not expressed in normal cells, but viral infection of a cell causes interferons to be made and released from the cell (that cell will often eventually die as a result of the infection). The interferon binds to target cells and induces an antiviral state. Both DNA and RNA viruses induce interferon but RNA viruses tend to induce higher levels. Double-stranded RNA produced during viral infection may be an important inducing agent.

Other stimuli will also cause these interferons to be made: e.g. exogenous double-stranded RNA, lipopolysaccharide, other components of certain bacteria.

TYPE II interferon

Interferon-gamma (immune interferon) is produced by certain activated T-cells^{3,4}. Interferon-gamma is made in response to antigen (including viral antigens) or mitogen stimulation of lymphocytes.

USES

Interferon alfa-2a (Roferon-A, Table 1.a) is FDA-approved to treat hairy cell leukemia, AIDS related Kaposi's sarcoma, and chronic myelogenous leukemia⁵. Interferon alfa-2b is approved for the treatment of hairy cell leukemia, malignant melanoma, condylomata acuminata, AIDS-related Kaposi's sarcoma, chronic hepatitis C, and chronic hepatitis B. Ribavirin combined with interferon alfa-2b, pegylated interferon alfa-2b, or pegylated interferon alpha-2a, has been approved for the treatment of chronic hepatitis C. Interferon beta-1b (Betaseron, Table 1.g) and interferon beta-1a (Avonex, Table 1.f) are approved for the treatment of multiple sclerosis⁶. Interferon β is also used in the treatment of rheumatic diseases. Interferon alfa-n3 (Alferon-N, Table 1.c) is approved for the treatment of genital and perianal warts caused by human papillomavirus (HPV). Interferon gamma-1B (Actimmune) is approved for the treatment of chronic granulomatous disease and severe malignant osteopetrosis.

SIDE EFFECTS

The most frequent adverse effects are flu-like symptoms: increased body temperature, feeling ill, fatigue, headache, muscle pain, convulsion, dizziness, hair thinning, and depression. Erythema, pain and hardness on the spot of injection are also frequently observed. IFN therapy causes immunosuppression, in particular through neutropenia and can result in some infections manifesting in unusual ways⁷.

INTERLEUKINS

Interleukins are cell signalling molecules, and a part of the cytokine super family of signalling molecules. The interleukins were first described as signals for communication between (inter- between) white blood cells (leuk- from leukocytes). The term interleukin derives from (*inter-*) "as a means of communication", and (*-leukin*) "deriving from the fact that many of these proteins are produced by leukocytes and act on leukocytes". There are several types of interleukins which activate helper T cells (Table 2) which act on T cells to promote their division and activation of other cells of the immune system such as NK and B cells.

USES:

An IL-1 blocker, anakinra (Kineret), has been approved for treatment of rheumatoid arthritis. Interleukin-2 has been approved by the FDA for the treatment of some types of cancer, but has not yet been approved for the treatment of HIV disease. A recombinant form of IL-2 for clinical use is manufactured by Chiron Corporation with the brand name Proleukin. It has been approved by the Food and Drug Administration (FDA) for the treatment of cancers (malignant melanoma, renal cell cancer), and is in clinical trials for the treatment of chronic viral infections, and as a booster (adjuvant) for vaccines. IL-3 stimulates the differentiation of multipotent hematopoietic stem cells into myeloid progenitor cells (as opposed to lymphoid progenitor cells where differentiation is stimulated by IL-7) as well as stimulates proliferation of all cells in the myeloid lineage (erythrocytes, megakaryocytes, granulocytes, monocytes, and dendritic cells). It is secreted by activated T cells to support growth and differentiation of T cells from the bone marrow in an immune response. Interleukin-5 (IL-5) is a key cytokine for the production, differentiation, and activation of eosinophils.

TABLE-1 INTERFERONS AND THEIR TRADE NAMES

INTERFERONS	TRADE NAMES
a) Interferon alpha 2a	Roferon A
b) Interferon alpha 2b	Intron A/Reliferon
c) Interferon alfa-n3	(Alferon-N)
d) Human leukocyte Interferon-alpha (HuIFN-alpha-Le)	Multiferon
e) Interferon beta 1a, liquid form	Rebif
f) Interferon beta 1a, lyophilized	Avonex
g) Interferon beta 1b	Betaseron / Betaferon
h) PEGylated interferon alpha 2a	Pegasys
i) PEGylated interferon alpha 2b	PegIntron

TABLE-2 INTERLEUKINS AND THEIR EFFECTS

MAJOR SOURCE	ORIGIN	MAJOR EFFECT
Interleukin-1	Macrophages	Stimulation of T cells and antigen-presenting cells. B-cell growth and antibody production. Promotes hematopoiesis (blood cell formation) ⁸ .
Interleukin-2	Activated T cells	Stimulates growth and differentiation of T cell response. Can be used in immunotherapy to treat cancer or suppressed for transplant patients.
Interleukin-3	hematopoietic stem cells	Differentiation and proliferation of myeloid progenitor cells ⁸ . e.g. erythrocytes, granulocytes
Interleukin-4	B-cell and T-cell proliferation	Humoral and adaptive immunity ⁹ .
Interleukin-5	T cells and mast cells	Eosinophil growth ¹⁰ .
Interleukin -6	Activated T cells	Synergistic effects with Interleukin-1 or Tumor necrosis factor ¹¹ .
Interleukin-7	thymus and bone marrow stromal cells	Development of T cell and B cell precursors, homeostasis, increased proinflammatory cytokines ⁸ .
Interleukin-8	Macrophages	Chemo attracts neutrophils ⁸ .
Interleukin-9	Activated T cells	Promotes growth of T cells and mast cells.
Interleukin-10	Activated T cells, B cells and monocytes	Inhibits inflammatory and immune responses ¹² .
Interleukin-11	Stromal cells	Acute phase protein production, osteoclast formation ⁸ .
Interleukin-12	Macrophages, B cells	Promotes T _H 1 cells while suppressing T _H 2 functions ⁸ .
Interleukin-13	T _H 2 cells	Similar to Interleukin-4 effects ¹³ .
Interleukin-14	T cells and certain malignant B cells	controls the growth and proliferation of B cells, inhibits Ig secretion
Interleukin-15	mononuclear phagocytes (and some other cells), especially macrophages following infection by virus(es)	Induces production of Natural killer cells
Interleukin-16	CD8 T cells	Chemo attracts CD4 T cells ⁸ .
Interleukin-17	Activated memory T cells	Promotes T cell proliferation.
Interleukin-18	Macrophages	Induces Interferon production ⁸ .

SIDE EFFECTS:

IL-2 can cause mood changes including irritability, insomnia, confusion, or depression. Recombinant human IL-3 had stimulatory hematopoietic effects, flu-like symptoms and fever. IL- 4 causes diarrhea, gastric ulceration, headache with nasal congestion, fluid retention, and arthralgia, in addition to constitutional symptoms such as fatigue, anorexia, nausea, and vomiting.

MONOCLONAL ANTIBODIES

Monoclonal antibodies (mAb or moAb) or monospecific antibodies that are the same because they are made by one type of immune cell which are all clones of a unique parent cell.

The first monoclonal antibody to receive FDA approval was rituximab. Monoclonal antibodies approved by the FDA include: ¹⁴

- Bevacizumab

- Cetuximab
- Panitumumab

USES

Monoclonal antibodies are widely used as diagnostic and research reagents. Their introduction into human therapy has been much slower. In some in vivo applications, the antibody itself is sufficient. Once bound to its target, it triggers the normal effector mechanisms of the body. In other cases, the monoclonal antibody is coupled to another molecule, for example:

- A fluorescent molecule to aid in imaging the target.
- A strongly-radioactive atom, such as Iodine-131 to aid in killing the target.

Some monoclonal antibodies that have been introduced into human medicine to suppress the immune system

- Muromonab-CD3 (OKT3)¹⁵ and two humanized anti-CD3 monoclonals: Bind to the CD3 molecule on the surface of T cells. Used to prevent acute rejection of organ, e.g., kidney, transplants. The humanized versions show promise in inhibiting the autoimmune destruction of beta cells in Type 1 diabetes mellitus.
- Infliximab (Remicade)¹⁵: Binds to tumor necrosis factor-alpha (TNF- α). Shows promise against some inflammatory diseases such as rheumatoid arthritis (by blunting the activity of Th1 cells). Side-effects: can convert a latent case of tuberculosis into active disease; can induce the formation of autoantibodies (by promoting the development of Th2 cells).
- Omalizumab (Xolair)¹⁵: Binds to IgE thus preventing IgE from binding to mast cells. Shows promise against allergic asthma.
- Daclizumab (Zenapax)¹⁵: Binds to part of the IL-2 receptor produced at the surface of activated T cells. Used to prevent acute rejection of transplanted kidney. Has also showed promise against T-cell lymphoma.
- A monoclonal antibody that binds CD33, a cell-surface molecule expressed by the cancerous cells in acute myelogenous leukemia (AML) but not found on the normal stem cells needed to repopulate the bone marrow.
- Calicheamicin, a complex oligosaccharide that makes double-stranded breaks in DNA. Mylotarg is the first immunotoxin that shows promise in the fight against cancer.
- **Alemtuzumab (MabCampath)¹⁵**: Binds to CD52, a molecule found on white blood cells. Has produced complete remission of chronic lymphocytic leukemia (for 18 months and counting).

Angiogenesis Inhibitors

- Vitaxin: Binds to a vascular integrin (alpha-v/beta-3) found on the blood vessels of tumors but not on the blood vessels supplying normal tissues. In Phase II clinical trials, Vitaxin has shown some promise in shrinking solid tumors without harmful side effects.
- Bevacizumab (Avastin)¹⁵: Binds to vascular endothelial growth factor (VEGF) preventing it from binding to its receptor. Approved by the US FDA in February 2004 for the treatment of colorectal cancers.

Others:

- Abciximab (ReoPro)¹⁵: Inhibits the clumping of platelets by binding the receptors on their surface that normally are linked by fibrinogen. Helpful in preventing reclogging of the coronary arteries in patients who have undergone angioplasty.

SIDE EFFECTS:

The main difficulty is that mouse antibodies are "seen" by the human immune system as foreign, and the human patient mounts an immune response against them, producing HAMA ("human anti-mouse antibodies"). These not only cause the therapeutic antibodies to be quickly eliminated from the host, but also form immune complexes that cause damage to the kidneys.

Monoclonal antibodies raised in humans would lessen the problem, but few people would want to be immunized in an attempt to make them and most of the attempts that have been made have been unsuccessful. Using genetic engineering it is possible to make mouse-human hybrid antibodies in an attempt to reduce the problem of HAMA.

- Chimeric antibodies:
The antibody combines the antigen-binding parts (variable regions) of the mouse antibody with the effector parts (constant regions) of a

To kill or inhibit malignant cells

- **Rituximab (Rituxan)**: Binds to the CD20 molecule found on most B-cells and is used to treat B-cell lymphomas¹⁶.
- **Zevalin¹⁵**: This is a monoclonal antibody against the CD20 molecule on B cells (and lymphomas) conjugated to either
 - The radioactive isotope indium-111 (¹¹¹In) or
 - The radioactive isotope yttrium-90 (⁹⁰Y). Both are given to the lymphoma patient, the ¹¹¹In version first followed by the ⁹⁰Y version (in each cases supplemented with Rituxan).
- **Bexxar (tositumomab)¹⁵**: This is a conjugate of a monoclonal antibody against CD20 and the radioactive isotope iodine-131 (¹³¹I). It, too, is designed as a treatment for lymphoma. Although both Bexxar and Zevalin kill normal B cells, they don't harm the B-cell precursors because these do not express CD20. So, in time, the precursors can repopulate the body with healthy B cells.
- **Herceptin (trastuzumab)¹⁵**: Herceptin binds to HER2, a receptor for epidermal growth factor (EGF) that is found on some tumor cells (some breast cancers, lymphomas). It is used in anti-cancer therapy for a specific kind of breast cancer.
- **Mylotarg¹⁵**: A conjugate of

human antibody. Infliximab, rituximab and abciximab are examples.

- Humanized antibodies:

The antibody combines only the amino acids responsible for making the antigen binding site (the hypervariable regions) of a mouse (or rat) antibody with the rest of a human antibody molecule thus replacing its own hypervariable regions. Daclizumab, Vitaxin, Mylotarg, Herceptin, and Xolair are examples.

In both cases, the new gene is expressed in mammalian cells grown in tissue culture (E. coli cannot add the sugars that are a necessary part of these glycoproteins).

ERYTHROPOIETIN

DEFINITION

Erythropoietin is a substance produced by the peritubular capillary endothelial cells of kidney that leads to the formation of red blood cells in the bone marrow. It is also called as hematopoietin or hemopoietin.

Erythropoietin is produced mainly by peritubular fibroblasts of the renal cortex. It is synthesized by renal peritubular cells in adults, with a small amount being produced in the liver^{17,18}. Regulation is believed to rely on a feed-back mechanism measuring blood oxygenation. Constitutively synthesized transcription factors for erythropoietin, known as hypoxia-inducible factors (HIFs), are hydroxylated and proteosomally digested in the presence of oxygen¹⁹. It binds to the erythropoietin receptor (EpoR) on the red cell surface and activates a JAK2 cascade. This receptor is also found in a large number of tissues such as bone marrow cells and peripheral/central nerve cells, many of which activate intracellular biological pathways upon binding with erythropoietin. The erythropoietin gene has been found on human chromosome 7 (in band 7q21). Erythropoietin (EPO) is a heavily glycosylated protein with a molecular weight of about 30,000 - 34,000 Daltons. Human erythropoietin is a polypeptide consisting of 165 amino acids, containing one O-linked and three N-linked carbohydrate chains. Binding of erythropoietin with sugars (called "glycosylation") slows the clearance of erythropoietin from the body thus allowing the hormone to last longer. Glycosylated erythropoietin comes in 3 forms: "alpha" (the most commonly used type in veterinary medicine), "beta" (of similar clinical efficacy to alpha), and Darbepoetin (which is particularly heavily glycosylated and lasts the longest). Normal levels of EPO are 0 to 19 mU/ml (milliunits per milliliter). The measurement of erythropoietin in the blood can indicate bone marrow disorders or kidney disease. Elevated levels can be seen in polycythemia rubra vera, a disorder characterized by an excess of red

blood cells. Lower than normal values of EPO are seen in chronic renal failure.

USES OF ERYTHROPOIETIN

1. Erythropoietin is used in patients who have anaemia resulting from chronic kidney disease and myelodysplasia.
2. In patients who require dialysis (have stage 5 chronic kidney disease (CKD)), iron should be given with erythropoietin²⁰. Dialysis patients in the US are most often given Epogen; outside of the US other brands of epoetin may be used.
3. Erythropoietin is also used for the treatment of cancer (chemotherapy and radiation).
4. Erythropoietin has been shown to be beneficial in certain neurological diseases like schizophrenia²¹.
5. More recently, a novel erythropoiesis-stimulating protein (NESP) has been produced²². This glycoprotein demonstrates anti-anemic capabilities and has a longer terminal half-life than erythropoietin. NESP offers chronic renal failure patients a lower dose of hormones to maintain normal hemoglobin levels.

Some of the erythropoietin available forms as biomedicine are:

- Epogen which is made by Amgen
- Epotin which is made by Gulf Pharmaceutical Ind. (JULPHAR)
- Betapoietin which is made by CinnaGen and Zahravi
- ReliPoietin which is made by Reliance Life Sciences Pvt. Ltd
- Epokine which is made by Intas Biopharmaceutica Pvt. Ltd
- Shanpoietin which is made by Shantha Biotechnics Ltd

SIDE EFFECTS OF ERYTHROPOIETIN:

Erythropoietin is generally well tolerated. Side effects can include chest pain, swelling due to retention of fluid, fast heart beat, headache, high blood pressure, increase in number and concentration of circulating red blood cells, seizures, shortness of breath, skin rash, pain in joints, diarrhea, nausea, fatigue, or flu-like syndrome after each dose.

TUMOR NECROSIS FACTOR:

TNF is produced mainly by macrophages, but they are produced also by a broad variety of other cell types including lymphoid cells, mast cells, endothelial cells, cardiac myocytes, adipose tissue, fibroblasts, and neuronal tissue. Large amounts of TNF are released in response to lipopolysaccharide, other bacterial products, and Interleukin-1 (IL-1). It has a number of actions on various organ systems,

generally together with IL-1 and Interleukin-6 (IL-6):

- On the hypothalamus:
 - Stimulating of the hypothalamic-pituitary-adrenal axis by stimulating the release of corticotropin releasing hormone (CRH)
 - Suppressing appetite
 - Fever
- On the liver: stimulating the acute phase response, leading to an increase in C-reactive protein and a number of other mediators. It also induces insulin resistance by promoting serine-phosphorylation of insulin receptor substrate-1 (IRS-1), which impairs insulin signaling.
- It is a potent chemoattractant for neutrophils, and helps them to stick to the endothelial cells for migration.
- On macrophages: stimulates phagocytosis, and production of IL-1 oxidants and the inflammatory lipid prostaglandin E₂ PGE₂.
- On other tissues: increasing insulin resistance.

A local increase in concentration of TNF will cause the cardinal signs of Inflammation to occur: heat, swelling, redness, and pain. Whereas high concentrations of TNF induce shock-like symptoms, the prolonged exposure to low concentrations of TNF can result in cachexia, a wasting syndrome. This can be found, for example, in cancer patients.

TYPES OF TUMOR NECROSIS FACTOR

- Tumor necrosis factor alpha (TNF- α) is the most well-known member of this class, and sometimes referred to when the term "tumor necrosis factor" is used.
- Tumor necrosis factor-beta (TNF- β), also known as lymphotoxin is a cytokine that is inhibited by interleukin 10.

USES

Tumor necrosis factor promotes the inflammatory response, which, in turn, causes many of the clinical problems associated with autoimmune disorders such as rheumatoid arthritis, ankylosing spondylitis, Crohn's disease, psoriasis and refractory asthma. These disorders are sometimes treated by using a TNF inhibitor. This inhibition can be achieved with a monoclonal antibody such as infliximab (Remicade) or adalimumab (Humira), or with a circulating receptor fusion protein such as etanercept (Enbrel).

SIDE EFFECTS OF TUMOR NECROSIS FACTOR

Tumor necrosis alpha may produce side effects such as malignancy, injection site reactions, infusion reactions, demyelinating disease, heart failure, induction of autoimmunity, infections.

COLONY STIMULATING FACTOR

Colony-stimulating factors (CSFs) are secreted glycoproteins which bind to receptor proteins on the surfaces of hemopoietic stem cells and thereby activate intracellular signaling pathways which can cause the cells to proliferate and differentiate into a specific kind of blood cell (usually white blood cells, for red blood cell formation see erythropoietin)²³.

Colony-stimulating factors include:

- *CSF1* - Macrophage colony-stimulating factor
- *CSF2* - Granulocyte macrophage colony-stimulating factors (also called GM-CSF and sargramostim)
- *CSF3* - Granulocyte colony-stimulating factors (also called G-CSF and filgrastim)
- Synthetic - Promegapoeitin

USES

1. It is used for bone marrow stimulation.
2. It is also used in stem cell mobilization.

GRANULOCYTE MACROPHAGE COLONY STIMULATING FACTOR

Colony-stimulating factors are a family of glycoproteins that modulate the haematopoietic process and control survival, proliferation, differentiation, and functional ability of mature haematopoietic progenitors, with frequent overlapping activities²⁴. The human granulocyte-macrophage colony-stimulating factor (hGM-CSF) belongs to this family. It is produced by a variety of cells including lymphocytes, endothelial cells, fibroblasts, monocytes, as well as some malignant cells²⁵. Human GM-CSF cDNA codes for a protein with 144 amino acids, with a leading peptide of 17 amino acids, two N-linked glycosylation sites, four potential O-linked glycosylation sites, and two disulfide bridges²⁶. Due to the glycosylation, the apparent molecular mass of the protein varies from 18 to 32 kDa. It is classified as a multilineage colony-stimulating factor because it stimulates the proliferation and differentiation of haematopoietic progenitor cells of neutrophil, eosinophil, and monocyte origin. Recombinant human granulocytemacrophage colony-stimulating factor (rhGM-CSF) is approved by the US Food and Drug Administration (FDA) for the acceleration of myeloid recovery following autologous bone marrow transplantation and in patients who fail to engraft after bone marrow transplantation. Evidence also supports the use of GM-CSF in many other clinical settings in which patients are neutropenic^{27, 28}.

Three synthetic human GM-CSFs have been produced by using recombinant DNA technology, employing different expression systems: bacteria, yeast, and mammalian cells²⁹. Sargramostim belongs to a class of drugs called colony-stimulating factors because of

their ability to stimulate cells in the bone marrow to multiply and form colonies. Sargramostim is man-made. It is a product of the genetic engineering of genes from fungi and is produced by recombinant DNA technology in bacteria. Other colony stimulating factors are epoetin alfa (Epoen, Procrit) and filgrastim (Neupogen).

USES

Sargramostim is used to increase the number of white blood cells when the white blood cell count is low, a condition called neutropenia. A reduced level of white blood cells causes an increased susceptibility to infections. Sargramostim is used in cancer patients with non-Hodgkins lymphoma, acute lymphoblastic leukemia and Hodgkins lymphoma who have received a bone marrow transplant to rapidly increase the white blood cell count. It also is used to manage patients with acute myelogenous leukemia over the age of 55 years who develop neutropenia because of chemotherapy. Sargramostim also may be used in cancer patients or healthy patients who have low white blood cell counts to boost the counts and thereby improve the bone marrow they will be donating for use in bone marrow transplantation.

SIDE EFFECTS

The most common side effects while taking sargramostim are mild to moderate fever, weakness, chills, headache, nausea, diarrhea, muscle and bone pain. Side effects less often seen are shortness of breath, leg and arm swelling or a mild rash at the site of injection.

GRANULOCYTE COLONY STIMULATING FACTOR

Granulocyte colony stimulating factor is produced in the body by the immune system and stimulates the formation of one type of white blood cell, the neutrophils. Neutrophils take part in the inflammatory reaction. They are responsible for detecting and destroying harmful bacteria and some fungi. G-CSF is also known as colony-stimulating factor 3 (CSF 3). Granulocyte colony stimulating factor is produced by endothelium, macrophages, and a number of other immune cells. The natural human glycoprotein exists in two forms, a 174- and 180-amino-acid-long protein of molecular weight 19,600 grams per mole. The more-abundant and more-active 174-amino acid form has been used in the development of pharmaceutical products by recombinant DNA (rDNA) technology. The Granulocyte colony stimulating factor receptor is present on precursor cells in the bone marrow, and, in response to stimulation by Granulocyte colony stimulating factor, initiates proliferation and differentiation into

mature granulocytes. Granulocyte colony stimulating factor is also a potent inducer of HSCs mobilization from the bone marrow into the bloodstream, although it has been shown that it does not directly affect the hematopoietic progenitors that are mobilized³⁰.

Beside the effect on the hematopoietic system, Granulocyte colony stimulating factor can also act on neuronal cells as a neurotrophic factor. Indeed, its receptor is expressed by neurons in the brain and spinal cord. The action of Granulocyte colony stimulating factor in the central nervous system is to induce neurogenesis, to increase the neuroplasticity and to counteract apoptosis^{31,32}. These properties are currently under investigations for the development of treatments of neurological diseases such as cerebral ischemia. The gene for Granulocyte colony stimulating factor is located on chromosome 17, locus q11.2-q12. It is also found that the Granulocyte colony stimulating factor gene has 4 introns, and that 2 different polypeptides are synthesized from the same gene by differential splicing of mRNA³³.

USES

1. Granulocyte colony stimulating factor stimulates the production of white blood cells (WBC). In oncology and hematology, a recombinant form of Granulocyte colony stimulating factor is used with certain cancer patients to accelerate recovery from neutropenia after chemotherapy, allowing higher-intensity treatment regimens.
2. Chemotherapy can cause myelosuppression and unacceptably low levels of white blood cells, making patients prone to infections and sepsis. Granulocyte colony stimulating factor is also used to increase the number of hematopoietic stem cells in the blood of the donor before collection by leukapheresis for use in hematopoietic stem cell transplantation. It may also be given to the receiver, to compensate for conditioning regimens.
3. G-CSF is currently under investigation for cerebral ischemia in a clinical phase IIb and several clinical pilot studies are published for other neurological disease such as amyotrophic lateral sclerosis³⁴.

Filgrastim: Filgrastim is produced by bacteria through the use of genetic engineering and recombinant DNA technology. Filgrastim belongs to a class of drugs called colony-stimulating factors because of their ability to stimulate cells in the bone marrow to multiply and form colonies.

USES

Filgrastim is used to prevent infectious complications associated with a decrease in the number of neutrophils in the body (neutropenia). Neutropenia may develop in cancer patients

receiving chemotherapy or undergoing bone marrow transplantation. Neutropenia also may occur for unknown reasons in adults and infants. Filgrastim is used in healthy patients who will be donating bone marrow if their white blood cell counts are low.

SIDE EFFECTS

Filgrastim is well-tolerated. The most common side effect in cancer patients receiving chemotherapy and

patients with severe, chronic neutropenia due to cyclical chemotherapy, a birth defect or an unknown cause, is mild to moderate bone pain. Patients receiving filgrastim after bone marrow transplantation commonly experience nausea and vomiting. Patients preparing to donate bone marrow who receive filgrastim commonly experience bone and muscle pain.

REFERENCES

- Roth, J. A. 1988. Enhancement of nonspecific resistance to bacterial infection by biologic response modifiers, p. 329-342. In J. A. Roth (ed.), *Virulence mechanisms of bacterial pathogens*. American Society for Microbiology, Washington, D.C.
- Lotze, M. T., R. M. Dallal, J. M. Kirkwood, and J. C. Flickinger. "Cutaneous Melanoma." In *Principles and Practice of Oncology*, edited by V. T. DeVita, S. A. Rosenberg, and S. Hellmon. Philadelphia: Lippincott, 2001.
- Aguilar, R. F. "Interferons in Neurology." *Rev Invest Clin* 52, no. 6 (2000): 665 - 679.
- Polman, C. H., and B. M. J. Uitdehaag. "Drug Treatment of Multiple Sclerosis." *BMJ* 321 (2000): 490 - 494.
- Goldstein, D; Laszlo, J (Sep 1988). "The role of interferon in cancer therapy: a current perspective" (Free full text). *CA: a cancer journal for clinicians* 38 (5): 258 - 77.
- Aolicelli, D; Direnzo; Trojano (14 September 2009). "Review of interferon beta-1b in the treatment of early and relapsing multiple sclerosis". *Biologics: targets & therapy* 3: 369 - 76.
- Bhatti Z, Berenson CS (2007). "Adult systemic cat scratch disease associated with therapy for hepatitis C". *BMC Infect Dis* 7: 8. doi:10.1186/1471-2334-7-8. PMID 17319959.
- Lippincott's Illustrated Reviews: Immunology. Paperback: 384 pages. July 1, 2007, Page 68.
- Howard M, Paul WE (1982). "Interleukins for B lymphocytes". *Lymphokine Res.* 1 (1): 1-4.
- Dubucquoi S, Desreumaux P, Janin A, Klein O, Goldman M, Tavernier J et al. Interleukin 5 synthesis by eosinophils: association with granules and immunoglobulin-dependent secretion. *J Exp Med* 194; 179(2):703-708.
- Nishimoto N (May 2006). "Interleukin-6 in rheumatoid arthritis". *Curr Opin Rheumatol* 18 (3): 277-81.
- Pestka S, Krause CD, Sarkar D, Walter MR, Shi Y, Fisher PB (2004). "Interleukin-10 and related cytokines and receptors". *Annu. Rev. Immunol.* 22: 929-79
- Wynn TA. IL-13 effector functions. *Annu Rev Immunol.* 2003; 21:425-5
- Takimoto CH, Calvo E. "Principles of Oncologic Pharmacotherapy" in Pazdur R, Wagman LD, Camphausen KA, Hoskins WJ (Eds) *Cancer Management: A Multidisciplinary Approach*. 11 ed. 2008.
- Waldmann, Thomas A. (2003). "Immunotherapy: past, present and future". *Nature Medicine* 9: 269-277
- Riley JK, Sliwkowski MX. CD20: a gene in search of a function. *Semin Oncol.* 2000;27:17-24.
- Jacobson LO, Goldwasser E, Fried W, Plzak L (March 1957). "Role of the kidney in erythropoiesis". *Nature* 179 (4560): 633-4
- Fisher JW, Koury S, Ducey T, Mendel S (October 1996). "Erythropoietin production by interstitial cells of hypoxic monkey kidneys". *British journal of haematology* 95 (1): 27-32.
- Jelkmann W (March 2007). "Erythropoietin after a century of research: younger than ever". *Eur. J. Haematol.* 78 (3): 183-205.
- Macdougall IC, Tucker B, Thompson J, Tomson CR, Baker LR, Raine AE (1996). "A randomized controlled study of iron supplementation in patients treated with erythropoietin". *Kidney Int.* 50 (5): 1694-9.
- Ehrenreich H, Degner D, Meller J, et al. (January 2004). "Erythropoietin: a candidate compound for neuroprotection in schizophrenia" (PDF). *Molecular psychiatry* 9 (1): 42-54.
- Macdougall IC (July 2000). "Novel erythropoiesis stimulating protein". *Semin. Nephrol.* 20 (4): 375-81.
- Alberts, Bruce; et al. (2002). *Molecular Biology of the Cell* (4th ed.). New York, NY: Garland Science.
- ARMITAGE, James O. Emerging applications of recombinant human granulocyte-macrophage colony stimulating factor. *Blood*, December 1998, vol. 92, no. 12, p. 4491-4508.
- FLEETWOOD, Andrew J.; COOK, Andrew D. and HAMILTON, John A. Functions of granulocyte macrophage colony-stimulating factor. *Critical*

- Reviews in Immunology, 2005, vol. 25, no. 5, p. 405-428.
26. KAUSHANSKY, Kenneth; O'HARA, Patrick J.; HART, Charles E.; FORSTROM, John W. and HAGEN, Frederick S. Role of carbohydrate in the function of human granulocyte-macrophage colony-stimulating factor. *Biochemistry*, July 1987, vol. 26, no. 15, p. 4861-4867.
27. TRINDADE, Eunice; MATON, Pierre; REDING, Raymond; DE VILLE DE GOYET, Jean; OTTE, Jean Bernard; BUTS, Jean Paul and SOKAL, Etienne M. Use of granulocyte macrophage colony stimulating factor in children after orthotopic liver transplantation. *Journal of Hepatology*, June 1998, vol. 28, no. 6, p. 1054-1057.
28. BILGIN, Kemal; YARAMIS, Ahmet; HASPOLAT, Kenan; TAS, M. Ali; GUNBEY, Sacit and DERMAN, Orhan. A randomized trial of granulocyte-macrophage colonystimulating factor in neonates with sepsis and neutropenia. *Pediatrics*, January 2001, vol. 107, no. 1, p. 36-41.
29. HUSSEIN, Atif M.; ROSS, Maureen; VREDENBURGH, James; MEISENBERG, Barry; HARS, Vera; GILBERT, Colleen; PETROS, William P.; CONIGLIO, David; KURTZBERG, Joanne; RUBIN, Peter and PETERS, William P. Effects of granulocyte-macrophage colony stimulating factor produced in CHO cells (regramostim), *Escherichia coli* (molgramostim) and yeast (sargramostim) on priming peripheral blood progenitor cells for use with autologous bone marrow after high-dose chemotherapy. *European Journal of Haematology*, November 1995, vol.54, no. 5, p. 281-287.
30. Thomas J, Liu F, Link DC (May 2002). "Mechanisms of mobilization of hematopoietic progenitors with granulocyte colony-stimulating factor". *Curr. Opin. Hematol.* 9 (3): 183-9.
31. Schneider A, Krüger C, Steigleder T, Weber D, Pitzer C, Laage R, Aronowski J, Maurer MH, Gassler N, Mier W, Hasselblatt M, Kollmar R, Schwab S, Sommer C, Bach A, Kuhn HG, Schäbitz WR (August 2005). "The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis". *J. Clin. Invest.* 115 (8): 2083-98.
32. Pitzer C, Krüger C, Plaas C, Kirsch F, Dittgen T, Müller R, Laage R, Kastner S, Suess S, Spoelgen R, Henriques A, Ehrenreich H, Schäbitz WR, Bach A, Schneider A (December 2008). "Granulocyte-colony stimulating factor improves outcome in a mouse model of amyotrophic lateral sclerosis". *Brain* 131 (Pt 12): 3335-47.
33. Nagata S, Tsuchiya M, Asano S, Kaziro Y, Yamazaki T, Yamamoto O, Hirata Y, Kubota N, Oheda M, Nomura H (1986). "Molecular cloning and expression of cDNA for human granulocyte colony-stimulating factor". *Nature* 319 (6052): 415-8.
34. Zang Y et al. *Amyotroph Lateral Scler.* 2008 Dec 4:1-2 Preliminary investigation of effect of granulocyte colony stimulating factor on amyotrophic lateral sclerosis.
