Review Article

Heparin: A Conventional Drug with a New Prototype

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ABSTRACT

Heparin, a long-established anticoagulant, has evolved into a versatile therapeutic agent with diverse applications beyond thrombosis management. This review explores the expanding repertoire of heparin's pharmacological activities, delving into its interactions with various proteins. As a highly sulfated glycosaminoglycan, heparin demonstrates efficacy in antineoplastic activities, offering new avenues for cancer treatment. Additionally, recent research highlights its potential in addressing COVID-19, inflammation, infertility, malaria, Alzheimer's disease, and infectious diseases. The article provides insights into the discovery, structure, and bioengineered preparation methods of heparin derivatives, showcasing both established and emerging applications. Challenges in current clinical treatments, including dosage optimization and adverse effects, are discussed. The dynamic landscape of heparin-based therapies emphasizes the need for continued exploration, underscoring exciting opportunities for innovative therapeutic strategies. In conclusion, this concise review illuminates the expanding horizons of heparin's therapeutic potential, emphasizing the ongoing quest for refined applications. As research progresses, heparin and its derivatives stand poised to revolutionize treatment paradigms across a spectrum of medical conditions, promising improved patient outcomes and paving the way for novel therapeutic interventions.

Keywords: Heparin, anticoagulant, glycosaminoglycan, pharmacological activities, antineoplastic, COVID-19, inflammation, infertility, malaria, Alzheimer's disease, infectious diseases, bioengineered preparation methods, clinical treatments, dosage optimization, adverse effects, therapeutic potential, innovative strategies, patient outcomes, medical research.

INTRODUCTION

For over a decade, heparin has been used as an anticoagulant. Most mammalian mast cells contain heparin, a highly sulfated glycosaminoglycan (GAG). Glycosaminoglycan is extraordinarily acidic and harmful in nature. Its acidic nature and presence of sulfate tend to bind with various proteins like coagulating and fibrinolytic proteins, which are related to growth and proteins like cytokines and chemokines[1]. Heparin has a distinctive Penta saccharide structure which causes anticoagulation by binding with antithrombin and inhibits factor Xa and IIa activation in the coagulation cascade[3]. Recently, there was an eruption of covid-19 caused by severe acute respiratory syndrome SARS-CoV-2, which become a remarkably public health concern since the influenza pandemic happened in 19 18. The notable clinical characteristic of the Covid-19 pandemic is respiratory collapse followed by infection with SARS-CoV-2.

Microcirculation dysfunction and thrombotic microangiopathy frequently accompany severe instances of COVID-19 with increased mortality risk, particularly in elderly, diabetic, hypertensive, and other cardiovascular disease patients[6]. Recently developed Heparin and others have significant pharmacological effects on antiangiogenic, anti-inflammatory, antitumor, etc treatment. Being a natural water-soluble polysaccharide, heparin is an excellent biocompatible substance that opens up a more comprehensive range of clinical value through the invention of nanomedicines[3].

1. Discovery of Heparin:

Heparin is employed widely in recent clinical treatment, which reduces blood clotting. Heparin was discovered accidentally in 1915 by Jay McLean under the guidance of Prof. William Henry Howell, a prominent physiologist. Howell devised the hypothesis of blood coagulation, which describes the binding of the phosphatide thromboplastin or cephalin to antithrombin. Also, prothrombin converts into thrombin which causes clotting. Although the main component of Howell's theory is Cephalin, he allocates McLean to study its purity and show it was phosphatide itself, was responsible for clotting, not causing contamination. By 1916, McLean had investigated not just cephalin but also the characteristics of similar phospholipid extracts. Precise further research leads to the discovery of heparin. Howell soon discovered its significance and researched it further in Baltimore after McLean departed for a fellowship at the University of Pennsylvania. Between 1918 and 1924, Howell replaced the organic solvent-based separation process with more effective water extraction. From the Greek word "hepan" means liver Howell named it Heparin and examined its chemical constituents. Howell said that his extract was not a phosphatide but a glucuronic acid, a carbohydrate. He knew this material wasn't like McLean's since its chemical composition was different.

Howell acknowledged McLean's groundbreaking research on anticoagulant phosphatides but did not credit his pupil with discovering heparin. Apart from sporadic attempts throughout his years as a medical student, McLean did not

research heparin until 1919, following his graduation. In the early 1940s, the results of his failed preclinical investigations and two human clinical trials were published in scholarly publications. These allegations were part of a campaign launched by McLean to alter the common idea that Howell discovered heparin. McLean was confident that his phosphatide preparations in 1916 already included Howell's carbohydrate, so he asserted primacy. His argument, supported by letters to researchers, lectures, and even an autobiography, initially encountered opposition from the scientific community but ultimately prevailed. As Marcum noted in his paper on discovering heparin, scientific discoveries are the product of a dynamic interaction between many things. In this regard, the works of McLean and Howell were complimentary. McLean's decision to extract cephalin from various tissues made it possible to see the anticoagulant effect of phosphatides. Nonetheless, Howell's idea provided McLean with a scientific framework for appreciating the significance of his discovery, and its unexpected nature prompted Howell to enhance the extraction technique[2].

2. Structure and biosynthesis of Heparin:

Heparin is an acidic carbohydrate in the glycosaminoglycan (GAGs) family[2]. Heparin can form salts with metals due to its positive optical rotation. Sulfate-rich components such as chondroitin sulphate, hyaluronic acid, and keratan sulphate are interspersed with glucosamine, hexuronic acid, and hexosamine disaccharide[2,5]. Heparin has unusual properties for a polysaccharide. To begin with, it has a polydispersed molecular structure, unlike simple low molecular weight medications like aspirin. On the other hand, Heparin is a polysaccharide that may comprise anywhere from 10 to over 1000 monosaccharide units and has a molecular weight of anywhere from 14 to 18 kDa. Heparin is synthesised as a serglycin glycoprotein in the endoplasmic reticulum and Golgi apparatus of connective tissue-type mast cells[9]. Heparin biosynthesis[2,7] is a multi-enzyme, multi-step process that begins with heparosan synthesis. After a tetrasaccharide linker is attached to a serine residue on a core protein, serglycin, the D-glucuronic acid[14], N-acetyl-D-glucosamine disaccharide units are added. In addition to being altered by N-deaccetylation/N-sulfation of the glucosamine units, C-5 epimerisation of the glucuronic, and O-sulfation at various points throughout the chain, this polymer is also subject to several other modifications[2,7]. Most of these changes are imperfect, leading to notable structural variation. Unknown cellular mechanisms regulate enzyme expression and activity at each stage. Heparin chains are deposited in the cytoplasmic secretory granules after being randomly broken by endo—D-glucuronidase[2].

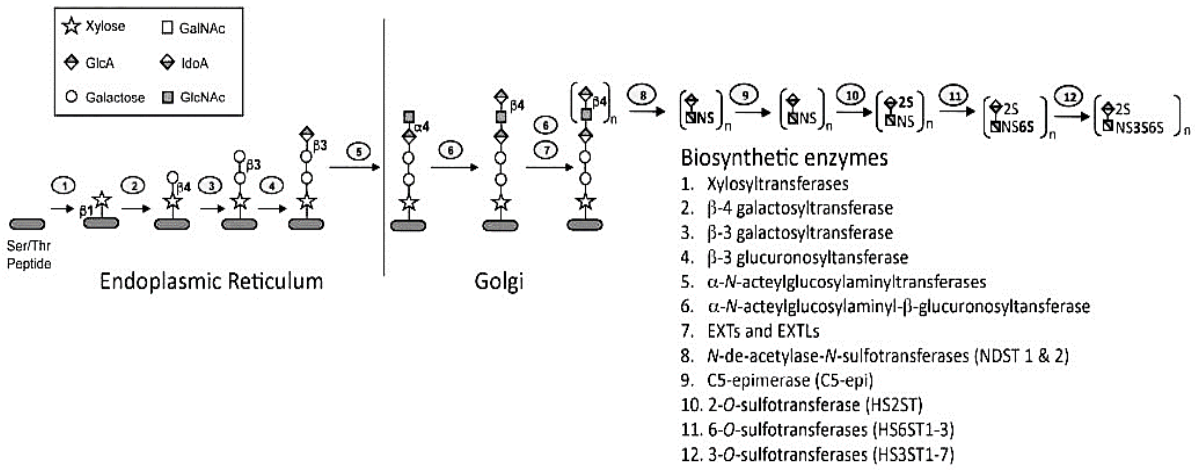

Figure 1: Biosynthesis of heparin

Heparin, a common anticoagulant, comes in three main varieties: 1. Unfractionated heparin (UFH), 2. Low molecular weight heparin (LMWH), and 3. Ultra-low molecular weight heparin (ULMWH). Separated from animal tissues, UFH has a molecular weight of 3,000 to 30,000 Da and is natural heparin. Heparin is principally made up of the disaccharides D-glucosamine-N,6-disulfate and L-iduronic acid-2-sulfate, both Tri sulfated. Heparin is a polymer, and around 70% comprises repeating Tri sulfated disaccharides that form highly sulfated and regular domains. Glucuronic acid is used instead of iduronic acid, and N-acetylglucosamine is used instead of N-sulfated glucosamine to create low-sulfated heparin. Heparin differs depending on the source of its extraction; for example, heparin obtained from bovine lungs has a lower N-acetyl glucosamine concentration but a greater sulfation level[10]. However, there are conserved sequence forms across the board in heparins. The core glycogen in one-third of UFH contains 3-O-sulfated glucosamine, indicating the presence of a unique five-sugar sequence. By binding to antithrombin (AT), this sequence has anticoagulant effects[17,18]. LMWHs are made by partial depolymerization of UFH by chemical or enzymatic processes[6]. LMWH has an average chain length of around 4500 Da (ranging from 2000 Da to 8000 Da)[11]. In addition, the unique depolymerization technique gives LMWHs their own pharmacokinetics and anticoagulant properties. This property makes LMWHs the preferred medicine for several indications[12-14], including treating venous thromboembolism and pulmonary embolism through individualized dosing. In addition to their

crucial function in thrombus therapy, LMWHs have several potential applications, such as preventing acute bronchial asthma spasms and preserving vascular strength during hemodialysis and arterial bypass grafting[15]. The oligosaccharides of ULMWH, the newest heparin, include anything between 5-10 saccharide units[16]. Fondaparinux, a Penta saccharide, is the sole ULMWH with FDA approval as of 2003. It specifically binds to FXa's AT-binding domain[19], making it a potent enzyme inhibitor. ULMWH has less deleterious effects on bleeding, more structural consistency, and better-quality control than UFH and LWMH. Due to its high price, ULMWH continues to account for just a negligible share of clinical HP products. Many challenges must be dealt with serially to make synthetic ULMWHs, such as the lengthy phases involved in synthesizing oligosaccharides, the intricacy of purification, and low synthetic yields. Many groups of scientists have worked tirelessly over the past few decades to find an effective way to synthesize fondaparinux, and they have succeeded in doing so by developing fondaparinux synthesis strategies that feature high stereoselectivity, straightforward purification, and high synthetic efficiency based on a one-pot glycosylation method; these strategies reduce the number of reaction stages required and increase the yield of glycan products. The ULMWH yields can be better by skipping the internal phases using chemoenzymatic synthesis[6].

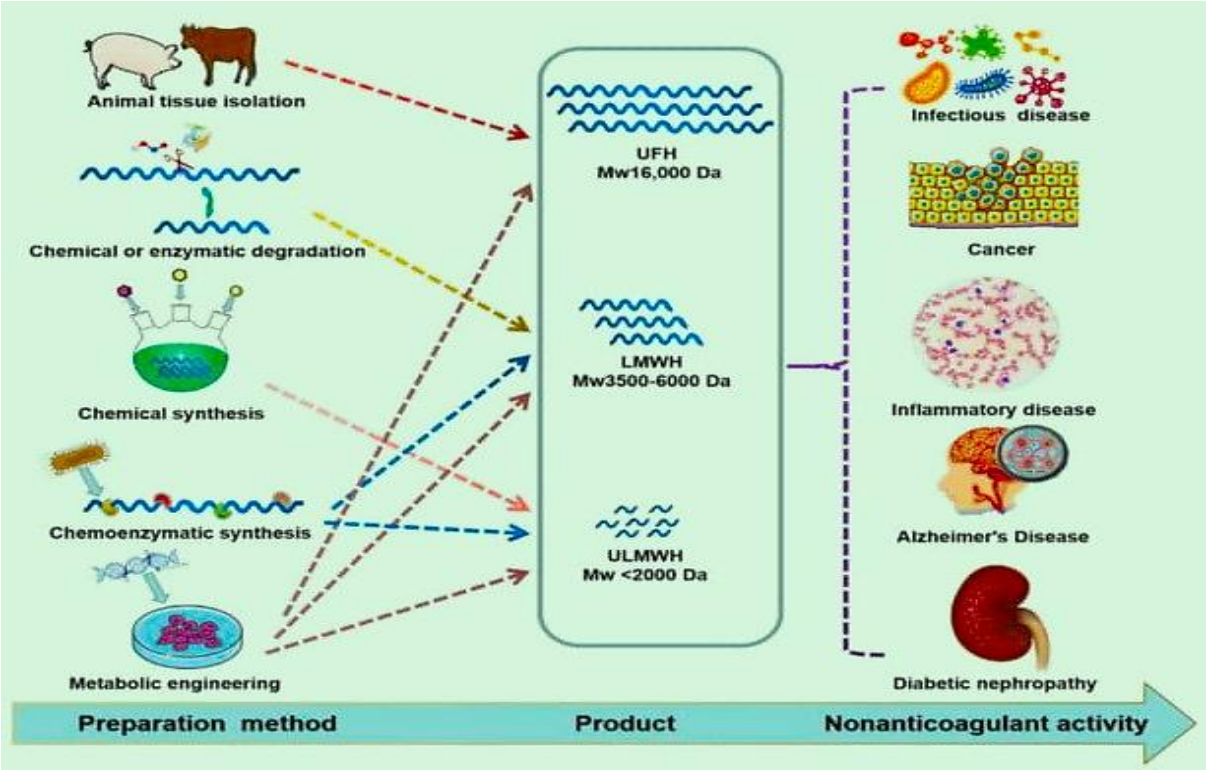

Figure 2: Procedures for preparing Heparin and its derivatives.

3. Applications of Heparin:

I. Non-anticoagulation activities of HP in clinical application:

The biological mechanism of coagulation is extraordinarily complex. By activating a sequence of coagulation elements in a particular order, insoluble polymers are generated during the coagulation process. Antithrombin III (AT-III) is the primary mediator of heparin's not clotting action, making it a key drug for treating thromboembolic illnesses. Heparin's ability to bind to the lysine residue of AT-III and form a reversible complex gives it its anticoagulant properties. This changes the configuration of AT-III, revealing its active arginine site and allowing it to combine quickly with the vibrant serine centres of factor IIIa (thrombin) and factors IXa, Xa, Xia, and XIIa to speed up their inactivation. To inactivate IXa/IIa, heparin forms a ternary complex with AT-III and coagulation factor while inactivating Xa requires a complex with AT-III. Once coupled with the coagulation factor AT-III, reclaimed heparin can be reused. Figure 3 depicts this procedure. The heparin chain needs at least 18 monosaccharide units to produce the heparin-AT-III thrombin ternary complex[3].

Because of its anticoagulant characteristics, HP is most widely used for preventing DVT and PE[6]. Maternal mortality rates rise when hypercoagulation manifests as venous thrombosis during pregnancy. In addition, HP's hypolipidemic effect has been documented for some time now[3]. The risk of developing atherosclerosis and severe pancreatitis dramatically increases in those with hyperlipidemia, a common metabolic condition. HP and insulin have been shown to activate lipoprotein lipase to reduce plasma triglyceride levels. Treatment for acute pancreatitis caused by hypertriglyceridemia appears well-tolerated, helpful, and reasonably priced. This section focuses on HP and its derivatives' potential therapeutic use outside of anticoagulation for treating infectious diseases, cancer, dementia, and kidney disease caused by diabetes.

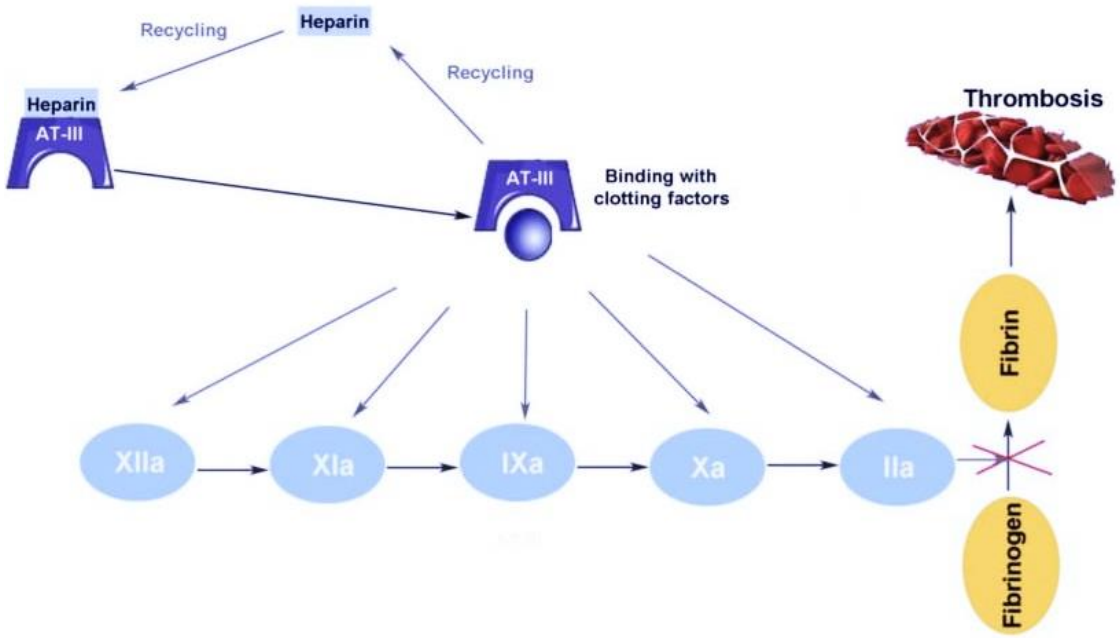

Figure 3: Role of Heparin in Anticoagulation.

I.I Heparin in infectious disease:

Novel infections and viral types constantly evolve due to environmental and human activity changes, making infectious illnesses a considerable danger to health worldwide[20]. A pathogen's first order of business is to undermine the body's immune system. Heparan sulphate (HS), which is similar in structure to heparan phosphate (HP) but has a comparatively low amount of sulfation and IdoA level, is a critical connection between pathogens and hosts. While mast cells are the only source of HP, HS may be isolated from virtually any animal cell and has a significant role in various physiological processes, such as cell identification, adhesion, signal transduction, and host-pathogen interactions[21-29]. HP, an HS mimic, acts as a competitive inhibitor by preventing the pathogen protein from attaching to HS on the host cell's surface. This finding suggests that HP may have clinical utility for treating or preventing infectious disorders. The prospect of using HP to treat COVID-19 and other viruses, germs, and parasites is particularly intriguing is the prospect of using HP to treat COVID-19 and other viruses, germs, and parasites[6]

I.I.I Heparin and Covid-19

Covid-19 patients with primary infections, thrombotic events, or organ damage were some of the essential candidates for therapy with heparin[30]. As a result, exhaustive studies on Heparin's antiviral activities are currently underway. The clinical and experimental data gathered over the past few years point to Heparin being beneficial for Covid-19 patients. Morbidity from Covid-19 infection is elevated because of coagulopathy[31]. Countrywide retrospective research in the United States of 4297 Covid-19 patients has shown that treating HP as a preventive anticoagulant reduces risk from the lack of anticoagulant[32].

In addition to its anticoagulant effects, HP has a direct antiviral impact by working on SARs-CoV-2 proteins, inhibiting viral adherence and replication. SARS-CoV-2's surface-anchored trimeric spike (Sp) glycoprotein enters the host cell with the help of the Angiotensin-converting enzyme 2 (ACE2) receptor on the respiratory epithelial cell. Notably, HS interacts with Sp-ACE2's Receptor-binding domain (RBD) to serve as a co-receptor and strengthen the connection between the two molecules. Using HP or analogues as bait receptors can reduce viral attachment by binding this Sp protein and preventing it from connecting with the virus to HS[6]. To investigate the structural space of this interaction, we have prepared a library of HP analogues and size-defined pieces. According to the findings, 2S and 6S groups are far more critical to RBD binding than NS groups. Secondary structural changes in the RBD can be induced using a molecule as small as a hexasaccharide[33]. Further, the viral precursor protein is cleaved by the SARS-CoV-2 virus' major protease (M-pro or 3CLpro) during replication in host cells. The M-pro protein is another prime focus for research into antiviral therapies. Recent studies have demonstrated a robust attachment between M-pro and HP, with a dissociation constant KD value of 17 and 32 M at 25 and 350C, respectively. The M-pro's proteolytic activity is also inhibited by HP, with an IC50 value of 83 nM and an inhibition constant of 7 nM. Heparin suppresses the SARS-CoV-2 virus infection, as suggested by these investigations. The rapid advancement of Heparin and its derivatives has led many to believe it should be the primary antiviral medication for treating SARS-CoV-2 and other coronaviruses. According to research, patients with Covid-19 who experience "cytokine storm syndrome" are at risk for fatal systemic inflammation disorder and lung tissue destruction from IL-6 activation, which leads to disseminated intravascular coagulation (DIC). Heparin binds to inflammatory cytokines, lowering their concentration, and stimulating the release of nitric oxide, increasing its

synthesis. This is achieved by decreasing the ability of inflammatory cells to stick to and invade target tissues. HP inhibits the signalling pathway of the proinflammatory transcription factor nuclear factor B (NF- B), demonstrating a direct anti-inflammatory impact on human endothelial cells and human monocytes exposed to lipopolysaccharide (LPS). Researchers observed that LMWH dramatically decreased the plasma level of IL-6 in COVID-19 patients, a significant trigger in the "cytokine storm" associated with this disease's severe symptoms. Recent research has argued that HP can effectively be an anti-inflammatory medicine for Covid-19 patients with myocarditis. According to a study by Sun et al., the level of heparin-binding protein (HBP), also known as Catatonic microbial protein of molecular weight 37 kDa, was significantly higher in severely ill Covid-19 patients. By establishing HBP as a disease marker and prospective therapeutic target in COVID-19 sickness, the high correlation between HBP and deteriorating Covid-19 reveals the critical function of HBP in inflammation-related responses in persons with severe Covid-19.

In conclusion, Hp has several beneficial impacts on patients with the Covid-19 phenotype. However, additional research on the optimal dosage, timing, and duration of administration is required.

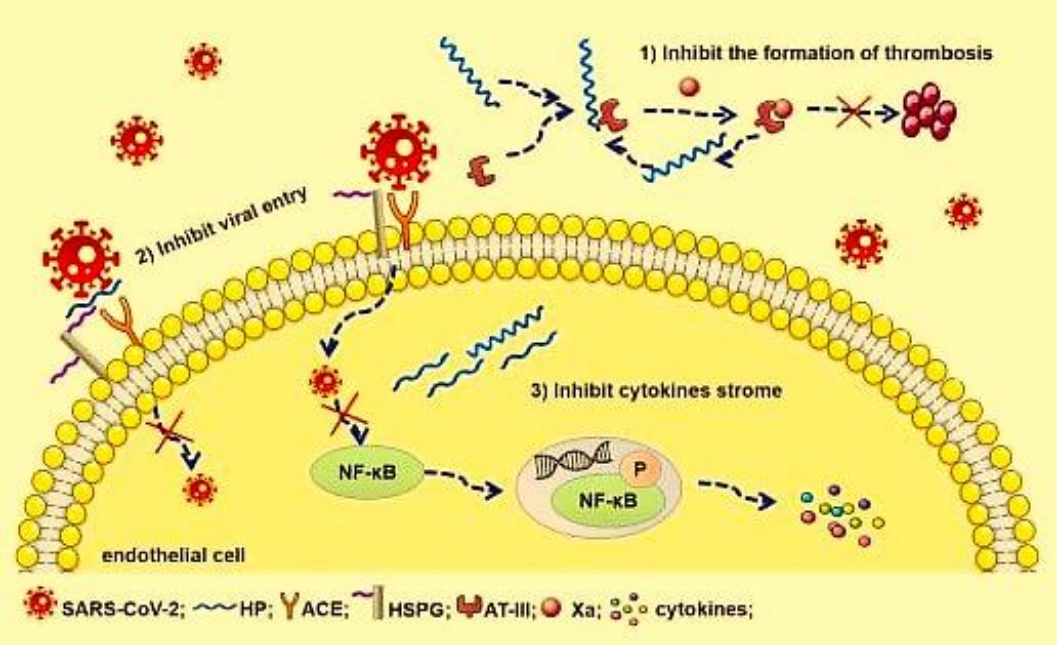

Figure 4: Action of Heparin and its derivatives on the SARS-CoV-19 virus

I.I.II Heparin in Malaria

Malaria is still a widespread infectious pandemic throughout the world's tropics and subtropics. Plasmodium spp., a genus of protists, is responsible for malaria. Malaria is spread when a female Anopheles mosquito infected with the Plasmodium falciparum parasite feeds on a human and injects the parasite's sporozoites into her host's blood. After transforming into merozoites in the liver, schistosomes enter the circulation to begin attacking RBC. It maintains its blood-stage cycle by asexually replicating through trophozoite and schizont phases to generate invading daughter cells for fresh RBCs. Mosquitoes feed on sexually mature parasites, known as gametocytes, that circulate in the peripheral blood. Mosquitoes feed on the peripheral blood of hosts infected with sexually mature parasites, either female or male gametocytes. After fertilisation in the insect midgut, the zygote develops into an ookinete and travels through the midgut epithelium to produce an oocyst, from which sporozoites are eventually released. Malaria re-entered the human body after the following mosquito bite after sporozoites moved to the salivary glands from the bloodstream. To a large extent, GAGs are responsible for binding Plasmodium-infected RBCs (pRBCs) to other molecules. Sequestration of pRBCs' poor clinical outcome in pregnancy-associated malaria has been related to malaria infection in the microvasculature and the placenta[34,35], which is thought to be caused by binding to the GAG chondroitin 4-sulfate (CSA). Plasmodium falciparum is prevented from sticking to RBCs and generating rosettes by negatively charged polysaccharides such as heparin, chondroitin and dextran sulphates, fucoidan, and the non-sulfated GAG hyaluronan[36,37]. When red blood cells (RBCs) are exposed to heparan sulphate (HS) or a chemical comparable to HS, resetting occurs. Heparin's significant anticoagulation and bleeding qualities and the potential hazards of infection as certain heparins are sourced from animal sources have limited its possible utility in treating malaria. However, it has been found that in vitro, in animal models, and in vivo, in fresh parasite isolates, depolymerised heparin without anticoagulant action inhibits rosette growth and pRBC cytoadherence. In contrast to heparin's IC50 of around 4 g/ml, shorter heparin fragments consisting of hexa- and octal saccharides with minor anticoagulant action[38] exhibited much lower antimalarial activity in vitro. Nonmammalian sea creatures provide an alternate supply of various sulphated

polysaccharides, some of which are structurally similar to pRBC-binding GAGs[41-43].

These marine sulfated glycans have great potential as a replacement for future antimalarial therapy since they inhibit P. falciparum cytoadhering and in vitro growth as efficiently as heparin at doses where their anticoagulant action is exceedingly low. While natural immunity to malaria is predominantly targeted at extracellular merozoites[39], no medications currently target Plasmodium invasion of erythrocytes[40], despite specific candidates being proposed.

I.II Heparin in cancer

In the 20th century, researchers began investigating the utilisation of heparin and its derivatives in cancer treatment after discovering that heparin might slow the expansion of tumour tissue that had been transplanted. Malignancy and thrombosis have been linked since Armand Trousseau, in 1985, classified superficial thrombophlebitis as a prelude to a strange visceral malignancy. First, heparin must be given as an anticoagulant during cancer treatment since the patients' full-grown cancer blood is often hypercoagulable. Thirdly, cancer-associated thrombosis (CAT) often results in venous thromboembolism (VTE). For almost a decade, LMWH has been an effective therapy for CAT[6]. Angiogenesis and cell adhesion are also linked to cancer spread. Some studies have shown that recombinant tissue factor pathway inhibitors (TFPIs) and LMWH can block the effects of angiogenesis-inducing substances such as vascular endothelial growth factor (VEGF). Essential for controlling blood vessel growth. In addition to CAT linked to cancer, other studies and meta-analyses have been found. Mousa and Norrby et al[33,34] showed the potential of LMWH for the angiogenesis of newly formed tumours using a chorioallantois membrane assay (CAM) angiogenesis model. Heparin also affects cancer metastasis by altering how chemokines interact with their receptors. Breast cancer metastasis relies on the chemokine CXCL12 and its receptor CXCR4. To lessen the metastasis of breast cancer cells in mice, LMWH blocked the connection between CXCL12 and CXCR4[35]. Although LMWH did not improve overall survival in people with solid tumours[37], it did decrease tumour cell adhesion and metastatic load in animal cancer models. The beta-galactoside-binding protein galectin three is upregulated in many different kinds of cancer. Heparin has been shown in recent studies to block this enzyme, halting the spread of cancer cells. Although the mechanism by which heparin's linking inhibits galectin-3-mediated cancer cell metastasis is not fully understood, it is clear that this effect is significant[3].

More than fifty ongoing clinical trials specifically target using HP and HP-like compounds in cancer. Unfortunately, these therapeutic studies had less than ideal-results. For example, in two randomised phase III lung cancer studies (RASTEN), the LMWH-adherent subgroup did not show significantly improved survival. These findings ran counter to a meta-analysis of lung cancer published in 2016 that found that patients with lung cancer who have no other reason to take anticoagulants fare significantly better when given HP or LMWH as primary thromboprophylaxis, especially those with limited-stage small-cell lung cancer (SCLC). Furthermore, when tested for hepatocellular carcinoma (HCC) in Phase III studies, the HP mimic PI-88 did not improve survival over standard treatment.

PI-88 induced everyday bleeding-related side events, including thrombocytopenia and thrombosis[37]. The HP PG545 is a second-generation PI-88 counterpart, providing another point of view. PG545 has immunomodulatory and antiangiogenic effects because, like other HP mimetics, it suppresses heparinase production and interferes with various angiogenic factors (including VEGF and FGF). Evidence of immune cell stimulation and cancer progression control in advanced solid tumours[38] was found in Phase I research. In addition, the drug showed tolerable safety data and adequate pharmacokinetic (PK) features. It is now being tested alongside nivolumab in Phase II clinical studies (NCT05061017).

The development of HP mimetics with antitumor activity but decreased anticoagulant activity and optimisation of administration[40] are also required, despite many studies showings that Cancer patients may benefit from HP and its compounds. The anticancer effects of HP and its derivatives and combination therapies, including these and chemotherapy, need more study[6].

I.III Heparin in Anti-inflammatory Treatment

The local circulatory and immunological systems are often involved in the chain of defensive responses known as inflammation, which is triggered by harmful stimuli. HP has been demonstrated to offer anti-inflammatory benefits in addition to its anticoagulant effects. Multiple papers have detailed the protein interactions between HP and its mimetics, resulting in anti-inflammatory effects. These interactions include proteins from the selectin, chemokine, and complement systems[3,6]. Heparin protected mice against the shock and death produced by lipopolysaccharide (LPS) in a model study conducted by Filkin et al. in 1968. Antithrombin's strong affinity for heparin is due to the specific Penta saccharide sequence on its surface[42] is the only case of "specific binding" for which there is adequate proof. As a result, the complex and only partly known mechanism underlying heparin's anti-inflammatory activity remains a mystery. The HP's anti-inflammatory responses were proven methodically by Poterucha et al. HP may decrease inflammatory responses by inhibiting neutrophil production and activity, decreasing eosinophil migration, enhancing vascular permeability, and interacting with cytokines in the vascular endothelium[44,45]. Heparin suppresses new smooth muscle cell growth in the lining of blood arteries[46]. Patients with inflammatory bowel disease, bronchial asthma, and acute pancreatitis have benefited from HP and similar therapies. Recent research has shown that heparin and its analogues may effectively treat asthma, COPD, critical lung damage, and septic shock because of its anti-inflammatory characteristics[47]. Anticoagulant-free heparin is effective in treating sepsis.

When the immune system fails to combat an infection effectively, a condition known as sepsis may develop. One of the leading causes of death in people with clinically severe conditions, it has the "three high characteristics" of being very common, extremely deadly, and extremely expensive to treat. Histone has a regulatory role in sepsis. Heparin, a negatively charged, highly sulfated polysaccharide structure, may help treat sepsis because it suppresses inflammation[48]. Heparin prevents the positively charged histones from binding to the negatively charged platelets. Notably, heparin could have a role in controlling inflammation, repairing damaged endothelium cells, restoring vascular barrier function, and in endothelial and cross-endothelial effects of inflammatory cytokines[49]. It is generally known that the glycocalyx[50], a glycoprotein present on the surface of endothelial cells, relies heavily on syndecan-1. To repair the residual glycocalyx on the cell surface and stop its shedding, new research suggests that UFH, an HS analogue, may mobilise the glycocalyx core protein syndecan-1[51,52]. These aids in maintaining a healthy cellular membrane and blood vessel wall.

Since Hala et al. were curious about the anti-inflammatory effects of polyanion heparin and the potential emission of naproxen, they looked into creating and characterising a polyelectrolyte multilayer (PEM) coating. Polyanion-free (PEM) media include heparin and interleukin (IL)-[53], two substances that prevent cell adhesion.

II. Application of heparin as Nanomaterials

Nano formulated heparin or heparin derivatives have various benefits for treating several illnesses[54]. However, it wasn't until 2008 that heparin nanocomposites were developed. Hollow capsules were synthesized by the first study's researchers utilizing an iron heparin and multilayer film complex, dramatically reducing the time needed for anticoagulation. The tablets can be opened to treat iron deficiency, and the iron is administered intravenously[54]. Since then, scientists have been working to improve the medicinal applications of heparin nanomaterial composites. Heparin is a naturally occurring, biocompatible, and versatile water-soluble polysaccharide. Stability and water solubility increase and molecular directionality is enhanced when nanoparticles are included. According to research[55-57], heparin nanoparticles are an efficient cytokine delivery system due to their extended half-life and excellent capacity for loading growth factors. The potential of heparin nanoparticles in drug delivery is enormous Heparin-based nanoparticles have applications beyond preventing blood loss. Qi Tan et al.[58] demonstrated superior vascular endothelial growth factor (VEGF) localization and release in vitro and enhanced fibroblast infiltration, extracellular matrix formation, and accelerated angiogenesis in a mouse subcutaneous implantation paradigm. Yasutaka et al. engineered LMWH protamine nanoparticles (lmwh-h/P NPs) to deliver the heparin-binding growth factor. The fibroblast growth factor (FGF-2) half-life was dramatically prolonged after being conjugated to nanoparticles[59]. The PEG-LMWH-taurocholate conjugate (LHT7) developed by Jeong et al. included a PEG-LHT7/TRAIL/Protamine nano complex. Cancer cells are preferentially eliminated without affecting normal cells by tumor necrosis factor-related apoptosis-inducing ligand (TRAIL).it represents a promising treatment alternative. However, TRAIL's physicochemical instability and short half-life limit its therapeutic application. The drug's short half-life, anti-angiogenesis efficacy, and uniform capacity to cause apoptosis in cancer cells[60] were significantly enhanced after encapsulation in a heparin nanoparticle.

When used together, the therapeutic benefits of several heparin-based nanoparticles become clear. Since nanotechnology and heparin-based biomolecules are rising, future research should concentrate on heparin nano formulations[8].

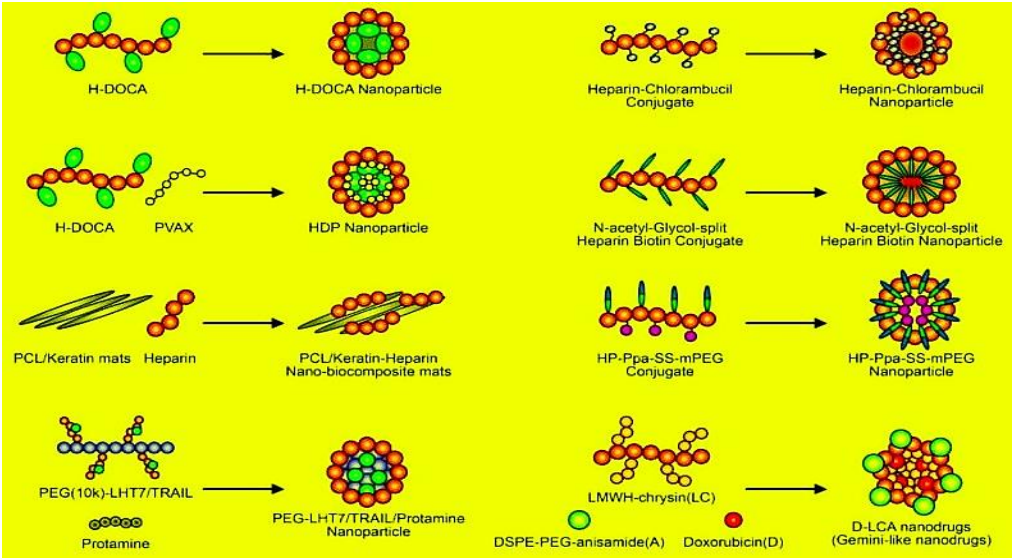

Figure 5: Nanoparticle structures derived from heparin derivatives. Different varieties of heparin conjugates are being studied as nano drug carriers or potential new drugs. The self-assembled heparin–drug conjugate that forms following nano formulation has demonstrated tremendous therapeutic potential for use in various treatments.

4. Challenges of Heparin Treatment

The effects of heparin are only sometimes advantageous. Heparin therapy has its share of side effects, some of which may be traced back to the drug's biological function. When used in therapeutic settings, heparin's short half-life means it must be administered often, which can be challenging for patients to remember. Slow-release formulations[61] are being developed to lengthen the half-life. Although heparin is the best anticoagulant currently in clinical usage, it does carry the potential for bleeding[62,63]. For older people and those with renal impairment, Injection site hematomas and cerebral hemorrhage are two examples of the milder symptoms associated with heparin-related bleeding. Some studies have found that when heparin is used for epidural anesthesia or spinal cord puncture, a hematoma can form at the puncture site, which is exceedingly risky since it can induce paralysis. Heparin's propensity to attach to positively charged molecules like platelet 4 (PF4) increases with chain length, leading to the formation of novel antigen complexes that can cause thrombocytopenia (HIT) by killing platelet and endothelial cell receptors. Heparin-induced thrombocytopenia (HIT) can potentially cause necrosis (dead skin) at the injection site. Relapse is common with HIT[64] because immunological memory is erased a few months after the onset of the disease.

Heparin-treated individuals have the same risk of HIT recurrence as those who were never given heparin. In addition, Heparin, according to some research, can also cause osteoporosis by binding to bone proteins, therefore inhibiting their production, increasing their absorption, and ultimately lowering bone mass[65]. This is another typical side effect of heparin and LMWH use for extended periods[66,67]. However, pregnant women, the elderly, and children should be warned that the impact is permanent and irreversible[68], despite some studies showing that short-term LWMH usage (3-6 months) does not impair bone mineral density.

Side effects include eosinophil increases, hyperkalemia, and others that are uncommon and often go away as heparin medication is stopped. In addition, patients with chronic renal failure, who have aberrant calcium and phosphorus levels in their bodies, are at high risk of developing calcium buildup at the injection site of heparin.

CONCLUSION AND FUTURE ASPECTS

Heparin has been the go-to anticoagulant for emergencies for well over a century. New anticoagulants are being developed, but HP is still valuable due to its many therapeutic applications. HP's high electronegativity and structural diversity are directly linked to its ability to bind to and operate as a significant regulator of many things. Its anti-viral and anti-inflammatory effects were crucial during the Corona Virus epidemic. Several animal and human investigations of HP without anticoagulants are currently being conducted. Before beginning such clinical studies, several potential constraints and difficulties should be considered. First, the development and use of HP as a non-anticoagulant medication are hampered by its significant anticoagulant action and adverse effects such as bleeding, alopecia, and increased liver enzymes. Second, more research is needed to understand better the HP mechanism's role in cancer, Alzheimer's disease, and other disorders. Third, further research is required to determine optimal dosing, optimal study duration, and the specific dose-response relationship to develop effective treatments with HP and its derivatives. Fourth, because of its chemical features and origins, HP and its products require extensive testing to ensure patient safety and determine optimal dosing, administration, and drug interactions. Fourth, Heparin is a cheap generic medicine. Therefore, there are few financial incentives to find new uses for it. Notably, new structural HP and HP derivatives are continually appearing due to the fast development of industry, biotechnology, and metabolic engineering. Eventually, the plans to make "designer" HP and similar variants with new medicinal potentials will be carried out.

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