Frontiers in Drug Design and Discovery

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Frontiers in Drug Design & Discovery

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PREFACE

In view of emerging epidemics and infectious diseases as well as the increasing prevalence of metabolic and neurological disorders, it is important to identify new drug targets in order to discover and develop safer and more effective drugs. The 11th volume of Frontiers in Drug Design and Discovery is a collection of five scholarly written reviews, each focused on a specific disease, covering various aspects of development of therapeutics.

Vasconcellos *et al* have contributed a comprehensive review on the relationship of heme-oxygenase (HO) with autophagy. They have proposed to use this nexus as a drug development strategy against chronic inflammatory and stress related disorders. The chapter by Mufarrege *et al* discusses the challenges related to the high yield production of biopharmaceuticals in animal cells and related issues, including immunogenicity. Artemisinin has attracted scientific interest as a potent antimalarial in the past few decades. Chaudhary and Tiwari have contributed a comprehensive review on the synthesis of artemisinin peroxide analogues and their antimalarial activities as compared to the parent natural product and other commonly used antimalarial drugs.

In the next chapter, Boskabady *et al* discuss the effect and mechanisms of action of Allium cepa (onion) extracts, and the well-known phenolic and sulphur phytoconstituents in chronic inflammatory respiratory disorders, both for treatment and prevention. Sepsis remains a major cause of morbidity and mortality. Immunotherapy has therefore been explored extensively as a treatment option for sepsis. Soleimanpour and Dolati have contributed a comprehensive review on this emerging new mode of treatment of sepsis.

We would like to thank all the authors for their excellent contributions. We greatly appreciate the efforts of Miss. Mariam Mehdi (Assistant Manager Publications) and Mr. Mahmood Alam (Director Publications) for the efficient production of this volume. We are confident that this volume will be widely appreciated by the scientific community.

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CHAPTER 1

Heme-oxygenase and Autophagy connected as a Cytoprotective Mechanism: Potential Therapeutic Target

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Abstract: Heme-oxygenase (HO) is an enzyme that catalyzes the main step of heme degradation generating anti-inflammatory end products with protective roles in physiologic and pathological situations. The relevance of HO in inflammatory conditions is well reported through pharmacological and/or genetic modulation, pointing out its importance in several models of stress such as infection, inflammation, and oxidative disturbance. Under the referred situations, another well-known protective process triggered is autophagy, in which defective cytosolic components and organelles are eliminated *via* lysosomes. Besides its role on organelles and macromolecules recycling, autophagy also contributes to cellular homeostasis by generating the functional blocks required for anabolic reactions. Recently, different studies have demonstrated a link between HO activity and autophagy activation. In this chapter, we would like to draw the reader's attention to the interconnection between HO and autophagy regarding stress response mechanisms, highlighting its importance in homeostasis maintenance that might be useful in the therapy of inflammatory diseases in the future.

Keywords: Autophagy, Bilirubin and biliverdin, Carbon monoxide, Curcumin, Cytoprotection, Heme, Heme-oxygenase, HO-therapy, Inflammation, Iron, Macroautophagy, Rapamycin, Reactive oxygen species, Resveratrol.

HEME-OXYGENASE

Heme-oxygenases (HOs) are conserved enzymes found in almost all kingdoms [1]. They catalyze the oxidative cleavage of intracellular heme, allowing the tetra-

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pyrrole backbone for disposal or metabolic usage [2]. HO uses reduced ferredoxin or cytochrome p450 reductase as an electron source to cleave heme and generate biliverdin (which can be further reduced to bilirubin by biliverdin reductase), carbon monoxide (CO), and ferrous iron (Fe^{+2}) [3] (Fig. **1**). In humans, there are two isoforms characterized: HO-1 and HO-2 [4]. HO-1 is expressed in practically all tissues and cells submitted to a plethora of insults or different sorts of stress [5]. It suffers transcriptional level regulation, and although HO-1 can be phosphorylated by the Serine/Threonine kinase Akt/PKB, this post-translational modification does not seem to affect HO-1 activity [6]. HO-2 is expressed constitutively in different organs, including liver, kidney, gut, and vasculature, with the highest expression on the brain, central nervous system, and testes [7]. In contrast to HO-1, HO-2 suffers post-transcriptionally regulation through phosphorylation mediated by casein kinase 2 [8]. Unlike what is seen for HO-1, the broadly expressed HO-2 activity does not seem to be induced by external factors. In addition, HO-2 heme-affinity and its reaction speed are three times lower and one-tenth of the HO-1, respectively [9]. Studies suggest that HO-2 can be essential for neurotransmission and vascular homeostasis because its by-product CO acts as a neurotransmitter and promote vasodilatation through direct signalling in endothelial cells [5].

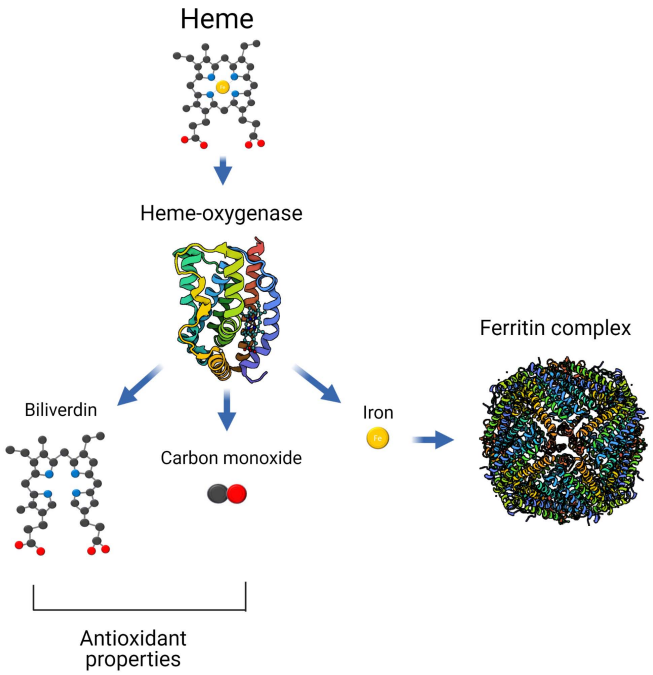

Fig. (1). Heme-oxygenase activity and by-products. Created with BioRender.com.

In mammals, the most studied isoform is HO-1, first described in 1968 and is also known as heat shock protein 32 [2]. HO-1 expression is affected by different transcription factors. These transcription factors bind to regulatory sequences in the HO-1 5'-promoter region [10 - 13], promoting HO-1 expression. The major transcription factors known to induce HO-1 are; specificity protein 1 (Sp1) regulated by phosphatidylinositol 3-kinase (PI3K); Hypoxia-inducible factor-1 (HIF-1); JunB and JunD; Nuclear Factor Kappa B (NF-κB); peroxisome proliferator-activated receptors (PPARγ) regulated by mitogen-activated protein kinase (MAPK); and nuclear factor erythroid 2-related factor2 (Nrf2). Nrf2 binds to enhancers termed antioxidant response elements (ARE) on the promoter region of HO-1 [14]. In the steady-state, Nrf2 is sequestered in the cytoplasm and targeted for proteasomal degradation *via* Kelch-like ECH-associated protein-1 (Keap-1) [15]. Furthermore, ARE is repressed by basic leucine zipper heterodimers, formed by either BTB domain and CNC homolog (Bach)-1 or Bach2 and musculoaponeurotic fibrosarcoma family (Maf) members [16]. Such complex regulation suggests the importance of HO-1 in different stress response mechanisms. In addition to its enzymatic activity, identifying a nuclear-truncated HO-1 (without the C-terminal domain), which seems to be enzymatically inactive, suggests a role of HO-1 in gene regulation [17]. It is believed that nuclear HO-1 upregulates genes associated with cytoprotection against oxidative stress and can enhance DNA repair [18].

It is important to consider that HO-1 mediates only part of the effects that confers cytoprotection. Many other crucial proteins and physiological mechanisms must be present, so the conjunction of factors could generate the proper response associated with adaptation and survival. In general, HO-1 is associated with the antioxidant response mechanism that counteracts the role of reactive oxygen species (ROS) on cell death and inflammation. Although many studies described several of these protective effects mediated by HO-1, the scavenging of free iron (generated through HO activity) has been described as crucial for the referred beneficial effects [19, 20]. Free iron binds to iron regulatory protein (IRP) and interrupts IRP association with transcripts involved in iron metabolism, allowing their translation [21]. In addition, HO-1 is associated with iron efflux and the exportation of iron-binding protein, reducing this critical source of intracellular reactive oxygen species (ROS) [22]. However, exacerbated expression of HO-1 is associated with cytotoxic effects mediated by uncontrolled iron levels [23].

The other HO-1 by-products, CO and biliverdin (bilirubin), exert antioxidant effects through different mechanisms. HO-1 activity is the source of 80-85% of the body's bilirubin and almost all endogenous CO generated [24]. Bilirubin showed superior antioxidant activity compared to biliverdin in a cell-free system [25]. Jansen and collaborators also demonstrated that biliverdin reductase is

CHAPTER 2

Development of Recombinant Therapeutic Proteins in Animal Cells: Challenges and Solutions

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Abstract: Recombinant therapeutic proteins, also referred to as biotherapeutics or biologics, are widely used for the treatment of numerous diseases such as cancer, diabetes and other non-communicable diseases. In addition, they also provide a solution to several chronic and emerging viral diseases.

Most biologics are produced in animal host cells as they provide relevant protein features, such as folding and post-translational modifications, similar to those produced by humans.

However, these products usually face great challenges mainly related to high manufacturing costs that may limit their use. To circumvent these issues, numerous strategies have been proposed. While some approaches focus on improving protein yield by introducing regulatory elements into the gene sequence, others specifically aim to optimize cell protein expression performance through cell engineering. Here, we will review the state of the art in the field of protein expression, focusing on the most promising strategies to improve protein yield in animal cells.

Product safety constitutes another concern that may restrict the use of biotherapeutics. Indeed, the development of unwanted immune responses against these drugs has been extensively reported. These immunogenicity events are triggered as a consequence of a breakdown in immune tolerance to the product. When a given biologic is administered to a patient, it encounters a variety of innate and adaptive immune cells that ultimately may orchestrate pro-inflammatory responses. Consequently, different scenarios may take place ranging from the development of anti-drug neutralizing antibodies to tissue damage. Hence, product immunogenicity is the main concern and should be closely monitored during product manufacturing and also in the clinic. In this respect, numerous *in vitro* and *in vivo* strategies are currently available, and more are still in development. These experimental platforms include the use of specific human immune cells, transgenic animals and even lymphatic micro-organoids. In this chapter, we will comprehensively describe these experimental strategies and their use in pre-clinical studies and clinical trials.

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Keywords: Animal Cells, Cell Engineering, Clinical Trials, Experimental Platforms, Immunogenicity, Lymphatic Micro-Organoids, Neutralizing Antibodies, Protein Yield, Regulatory Elements, Therapeutic Proteins, Transgenic Mice.

INTRODUCTION

For decades, therapeutic proteins have been proposed for the treatment of numerous chronic and emerging diseases. These products include antibodies, hormones, enzymes, growth factors, clotting factors and cytokines [1]. In most cases, these molecules are identical to the natural protein, and thus exert the same or similar biological function to that of their natural counterparts.

Numerous cell platforms are currently available for therapeutic protein production and include bacteria, yeast, insect cells, plant cells and animal cells.

Bacterial gene expression systems allow achieving high protein yields in short production periods. However, the use of these cells has been limited to the production of nonglycosylated proteins. Also, proteins produced from bacteria usually form aggregates. As a result, they must be isolated from inclusion bodies and then refolded to recover their natural structure and biological function.

Some biologics can also be produced with a high yield in yeast. Nevertheless, there are reports showing that these cells may produce glycan structures that can be immunogenic in humans [2, 3]. A similar scenario was observed for plant and insect cells. Plant cells produce immunogenic glycan structures such as core α1,3-fructose and β1,2-xylose [4, 5], and insect cells decorate their proteins with high mannose or paucimannose structures [6].

For all the above, a significant proportion of therapeutic proteins are currently produced in animal cells [1]. However, there are still some relevant limitations when producing therapeutic proteins in animal cells. The greatest challenges associated with the use of animal cells for biologic production are reaching high protein yield and reduce product immunogenicity.

Protein expression in animal cells is a complex process that involves multiple concatenated stages at a specific time and cellular location. In addition, most of these proteins require post-translational modifications such as glycosylation, phosphorylation, acetylation, hydroxylation, among others, required for protein maturation. Consequently, protein production in animal cells usually allows reaching low yields when compared with other cell platforms. For this reason, in the last years, numerous efforts have been made to enhance *in vitro* protein productivity. These approaches have focused on both optimizing RNA

transcription and translation through the use of specific regulatory elements and cell engineering to improve specific cellular pathways.

Another important aspect to be considered during product quality control and safety is protein immunogenicity. Like almost all proteins in the human body, therapeutic proteins are subject to immune screening when administered to patients. Product immunogenicity is related to several factors. Among them, the presence of T cell epitopes has shown to play an important role in breaking down immune tolerance [7 - 9]. In addition, some impurities or contaminants derived from different sources (host cell or manufacturing process) that may escape the purification strategy are found in the final product, increasing immunogenicity risk [10, 11]. As a result, these responses may lead to the development of anti-drug antibodies (ADA) and an exacerbated production of pro-inflammatory cytokines and chemokines.

In this chapter, we will thoroughly review the main strategies that might be considered when producing biotherapeutics in animal cells in order to enhance protein yield. Also, we will analyze the state of the art about the most relevant strategies aimed to predict and mitigate therapeutic protein immunogenicity.

CHALLENGES IN THE PRODUCTION OF THERAPEUTIC PROTEINS IN ANIMAL CELLS

In the last decade, the number of recombinant proteins approved for therapeutic treatment in humans has grown exponentially. Considering the top ten best-selling pharmaceutical products in the world, eight of them are therapeutic proteins. It is then not surprising that this global market, which accounted for over $140 billion in 2016, is expected to steadily grow by 6% annually to reach over $200 billion by 2023 [12].

The success of therapeutic proteins in the pharmaceutical field is due to strong investments in research and development, led by innovative manufacturing systems that use animal cells for recombinant protein production. The protein yield reached through these systems has increased over the decades by the combined improvement of both cell-related and process-related features. Among process-related features, it is worth noting the development of new bioreactors, customization of cell culture conditions for each particular cell line, the massification of single-use media bags, improvements in harvesting methods and adoption of innovative purification techniques. All these factors were essential for significantly incrementing the mass of protein products obtained by cell unit and per dollar spent.

CHAPTER 3

Artemisinin Analogues as a Novel Class of Antimalarial Agents: Recent Developments, Current Scenario and Future Perspectives

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Abstract: The comprehensive chronological advancement and statistical account of the recent developments and current scenario on antimalarial chemotherapy, in particularly, based on various types of artemisinin analogues and their potential as antimalarial agents have been schematically presented in this book chapter. These studies had compared efficacies of various semi-synthetic artemisinin analogues with that of standard reference drug artemisinin, *β*-Arteether and various other anti-malarial drugs. Thus, this book chapter highlights the recent advancements and historical developments observed in the medicinal chemistry of artemisinin since its discovery during the past 49 years (**1971-2020**). Hence, this book chapter will serve as first-hand information on artemisinin-based antimalarial peroxides to organic and medicinal chemists, as well as biologist/pharmacologists working on anti-malarial drug discovery and development.

Keywords: Antimalarial peroxides, Arteether, Artemisinin, Artesunate, Malaria, Multi-drug Resistant.

INTRODUCTION

The present-day population of over ninety countries from WHO Africa, Asia and

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South America regions are at high risk of getting being affected by a communicable protozoan disease "*Malaria*" [1]. WHO has categorized malaria infection into two categories; severe and uncomplicated malaria. The severe malaria infection is the most fatal form of the disease, generally signified as vital organ failure, seizures, coma and prompts the demise of the patient within 48 h of its appearance if untreated [2]. Both the causative agent (*Plasmodium*) and transmission vector (female *Anopheles* mosquito) of the disease were discovered in the late nineteenth century [3]. At present, six *Plasmodium* species: *P. falciparum*, *P. vivax*, *P. malariae*, *P. ovale curtisi*, *P. ovale wallikeri* and *P. knowlesi* are mainly transmitted through six primary (*A. baimaii*, *A. culicifacies*, *A. stephensi*, *A. minimus*, *A. sundaicus*, *A. fluviatilis*) and four local (*A. annularis*, *A. nivipes*, *A. philippinensis*, *A. varuna)* species of female *Anopheles* mosquito (primary host) in the Indian subcontinent and produce symptoms of pernicious malaria disease in the secondary human host [4, 5]. Out of all parasite species, *P. falciparum* (96%) and *P. vivax* (3.4%) share the largest proportion of malaria infections (99% of overall malaria clinical cases) across the globe [6].

Quinine (classical *N*-containing) and artemisinin (endoperoxides) are two important class of antimalarials of modern-day malaria chemotherapy, which delivered various promising antimalarial drugs for both curative and prophylaxis responses. These medications have played a vital role in saving millions of lives from past to present and changes the scenario of the current antimalarial drug discovery program (Fig. **1**) [7]. The era of classical *N*-containing drugs began in the early nineteenth century with the isolation of quinine from '*Chinchona*' tree bark, which was traditionally being used by the Peruvian Indians for relieving the symptoms of fever and severe chills [8]. Later, Hans Andersag discovered an effective, economically cheap, synthetic substitute of quinine *i.e.*, chloroquine (**2**, Resochin®) in **1934,** which had been also categorized under the same 4-amino quinoline class of antimalarials. Chloroquine (**2**) was found susceptible to both uncomplicated and severe forms of malaria [9 - 11]. Along with chloroquine (**2**), quinoline-based antimalarials became the frontline medications for the treatment of malaria and were responsible for the worldwide annihilation of the disease until the emergence of the first chloroquine-resistant *P. falciparum* strain in **1957** from the Thai-Cambodia border. The parasite initiated an up-gradation in developing resistant and cross-resistant strains against various first-line quinoline derived drugs in different parts of the world, including tropical and subtropical regions and made a strong reappearance on the world map in the early nineties [12]. The lack of administrative guidance in various poor countries and the non-alignment of pharmaceutical industries in the research and development of the novel antimalarials directly diminished the advancement in the antimalarial drug development programme during the period **1975-1999**. As a result, exclusively four medications were developed as antimalarials out of 1,400 new drugs [13]. On

the other hand, the evolution of resistant Anopheles vector against pyrethroids was also terrifying the situation concurrently [14]. With a 30% overall mortality in the **1990s**, the picture of " malaria-specific mortality " in the case of children between the ages of 1-5 years from Africa had represented a massive ascent in " malaria-associated death rate " as compared to the 12% mortality observed during the period **1960-1990**. Similarly, the "world malaria death toll" frequently increased every year and touched its peak (half a million to one million annually) in the next two decades (**1990-2004**) [15, 16]. Besides, the macroeconomic level of poorer endemic countries from tropical and subtropical regions of the world was also enormously affected by malaria and in comparison, to the rest of the world, their one-half per-capita GDP (gross domestic product) loosed between the four decades (**1950-1990**) [17].

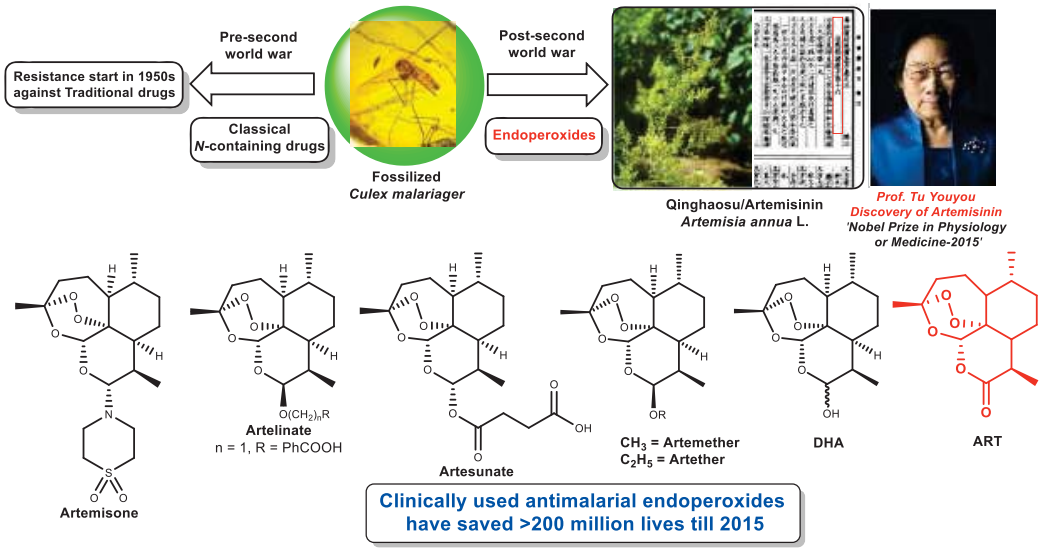

Fig. (1). From past to present, the evolution of antimalarial drugs.

During the '*Second Indochina War*'; an initial request of the Vietnamese president to the Chinese counterpart for help to fight against malaria formed the basis for the launch of the systematic screening programme (project-**523**) on traditional Chinese herbs and rediscovery of '*Qing hao*' as a potent antiplasmodial agent. The active ingredient artemisinin (**11**) ("*Qinghaosu*") was isolated from the plant *Artemisia annua* (Sweet wormwood) (Chemical Abstracts, Registry No. 63968-64-9) by the Chinese researchers in **1971** [18]. Since its isolation, the artemisinin class of endoperoxides have served as frontline medications for the treatment of drug-resistant malaria and saved more than 200 million lives till **2015** [19]. In early **2001**, the parasite started developing resistance against artemisinin in SE

CHAPTER 4

The Effects of *Allium Cepa* and their Derivatives on Respiratory Diseases and the Possible Mechanisms of these Effects

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Abstract: *Allium cepa* (*A. cepa*) or onion is an important condiment plant consumed worldwide. The plant and its constituents, such as quercetin (Qt), thiosulphinates, and phenolic acids, show various pharmacological and therapeutic effects, including the effects on respiratory disorders. In this chapter, various pharmacological and therapeutic effects of *A. cepa* and its constituents on respiratory disorders are reviewed. Using keywords such as onion, *A. cepa*, *A. cepa* constituents, therapeutic effects, pharmacological effects, and respiratory disorders in different online databases were searched until the end of July 2020. *A. cepa* showed a significant relaxant effect in tracheal smooth muscle (TSM) comparable to the effect of theophylline, indicating the bronchodilatory or reliving effect on obstructive pulmonary diseases. The possible mechanisms of this effect were reported as β2-adrenergic stimulatory and/or calcium channel blockade. The plant and its constituents also demonstrated a preventive effect on inflammatory respiratory diseases by improving lung pathological changes and

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tracheal responsiveness in various lung diseases. The possible effect of preventive effects of onion and its constituents on lung disorders is their inhibitory effects on inflammatory cells, inflammatory mediators, cytokines, cyclooxygenase and lipoxygenase activity, and oxidative stress markers. Therefore, *A. cepa*, and its constituents could be of therapeutic value in treating respiratory disorders both through reliving and prevention of lung diseases.

Keywords: *Allium cepa*, Onion, Pharmacological properties, Phenolic acids, Quercetin, Respiratory disorders, Therapeutic effects, Thiosulphinates.

INTRODUCTION

Onion or *Allium cepa* L., (*A. cepa*) is a herb of the Liliaceae family [1] over 250 genera and 3700 species which is one of the oldest cultivated plants [2] originated from central Asia [3, 4] and grows all over the world mainly in the moderate climates zones [5, 6]. Onion is characterized as sweet and non-sweet based on its taste and as yellow, red, or white based on its color [3, 4] and is used as a spice due to its odor and taste [7]. Onion is consumed as a food additive as powder, essential oil, or fresh [8, 9].

There is a long time history of using *A. cepa* in traditional medicine [10] in various parts of the world, including Egypt, where it has been used for various healing properties [11]. In China, it is used for treatment of fever, headache, cholera, dysentery, common cold, and arthritis [12] and in other parts of the world to treat stomach diseases, throat infection and hepatitis [1].

Various pharmacological effects were reported for *A. cepa* including anti-inflammatory [13], antioxidant, anti-diabetic [5, 14, 15], anti-fungal [2], antimicrobial, anti-mutagenic [16], anti-cancer, antiplatelet [17], anti-hyperglycemic, antispasmodic [3, 18], and anti-asthmatic [9] properties. The preventive effects of long-term consumption of *A. cepa* on vascular and heart diseases, neurodegenerative disorders and cataract formation [3], improvement of kidney function [6], antihelmintic, aphrodisiac, carminative, emmenagogue, expectorant activities, as well as beneficial effects on dysmenorrhea, vertigo, fainting, migraine, wounds, scars, keloids, pain, and swelling after bee, treatment of bruises, earache, jaundice and pimples [16] as well as anti-carcinogenic properties [19] were also reported.

The therapeutic effects of *A. cepa* and its constituents on respiratory and allergic diseases, such as inhibitory effects on bronchoconstriction [20], were also shown. The relaxant effect of flavonoids from *A. cepa* on TSM, indicated their possible bronchodilator effect [21 - 23]. Onion showed immunomodulatory [24] effects on the tracheal responsiveness and lung inflammation [25] in sensitized rats. In addition, the anti-asthmatic effects of *A. cepa* extract (AcE) and its constituent,

quercetin (Qt), were demonstrated in a murine model of asthma [26]. World Health Organization (WHO) recommended using AcE to treat several diseases such as common colds, coughs, asthma, bronchitis, and allergic diseases [10]. The anti-allergic properties of the extracts of *A. cepa* [27] and its flavonoid Qt [28, 29] were showed in a mouse model asthma.

Due to the adverse effects of currently used drugs for treating respiratory disorders and the absence of their full therapeutic efficacy, new drugs should be developed to treat inflammatory obstructive respiratory diseases [30]. Two types of drugs are used to treat inflammatory and obstructive respiratory diseases, including relieving drugs to reduce airway constriction and preventive drugs to reduce lung inflammation [31]. Several adverse side effects were reported for typically used drugs to treat asthma and allergic rhinitis, such as antihistamines, decongestants, anticholinergic, and corticosteroid drug including sedation, impaired learning and memory as well as cardiac arrhythmias [30]. Therefore, therapeutic strategies should seek to decrease these side effects of the present prescribed drugs. Several safe, natural therapies such as urtica dioica, bromelain, Qt, N-acetylcysteine, and vitamin C are already introduced to treat the above disorders [15]. The anti-allergic effect of polyphenols found in foods and plants in different disease models and human clinical trials was shown. These agents showed anti-inflammatory, anti-oxidant, and immunomodulatory effects. They can modulate the process of allergic sensitization by the interaction of polyphenols with proteins and inhibit mediator release [16]. Several studies also showed the preventive effect of derivatives from *A. cepa* such as Qt on respiratory disorders [26 - 29].

In the present chapter, the effects of *A. cepa* and its constituents on respiratory disorders and the possible mechanisms of these effects will be reviewed.

METHODS

The keywords "*Allium cepa"*, "onion", "flavonoid ", "quercetin"," organosulfur" , "saponin","phenolic compounds","therapeutic effects", "pharmacological effects" and " respiratory disorders" were used to find appropriate articles in the present chapter. For this purpose, ISI Web of Knowledge, Medline/PubMed, Scopus, and Google Scholar were searched until the end of May 2020,

RESULTS

Onion Compounds and their Biologicals Effects

A. cepa contains vitamins and minerals, sulfur amino acids, and a variety of secondary metabolites such as flavonoids (particularly flavonols and

CHAPTER 5

Immunotherapy of Sepsis

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Abstract: Sepsis is a clinical condition that occurs due to dysregulated fundamental host reaction to an infection, resulting in organ dysfunction. Proinflammatory mediators are related to indications watched early in patients with sepsis. In spite of decades of investigation, the morbidity and mortality of sepsis stay exceptionally very high. Regardless of the recognition of numerous prospective targets in the complex immune response pathways, no specific immunotherapeutic medicines are at present accessible. Presently, immunosuppression in sepsis is a subject matter of strong research amongst various pre-clinical and clinical studies. In fact, an improved consideration of a patient's immune status on a constant basis will plausibly allow focused immunotherapy. This book chapter provides an overview of the epidemiology, etiology, pathogenesis of sepsis and gives an outline of the challenges in immunotherapy for patients suffering from sepsis.

Keywords: Immunotherapy, Inflammatory, Sepsis, Septic shock.

INTRODUCTION

Sepsis is a serious organ dysfunction caused by a dysregulated host response to associate infection and may be the main world health concern worldwide with an over $20 billion (5.2%) of total U.S.A. hospital expenses in 2011 [1]. The term "Sepsis" is originated from the Greek word "sepsin," which means to putrefy or "to make putrid." Sepsis is the primary reason behind death from infection among critically ill patients in intensive care unit (ICU), notably, if not recognized and treated promptly.It is calculable to have an effect on over thirty million individuals worldwide once a year, probably resulting in 6 million deaths [2].

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In 1992, a North American Consensus Committee formally outlined sepsis as infection with two or more systemic inflammatory response syndrome (SIRS) criteria (abnormalities in white blood cell count, temperature, heart rate, and respiratory rate), which might cause organ dysfunction, leading to a septic shock [3] (Fig. **1**).

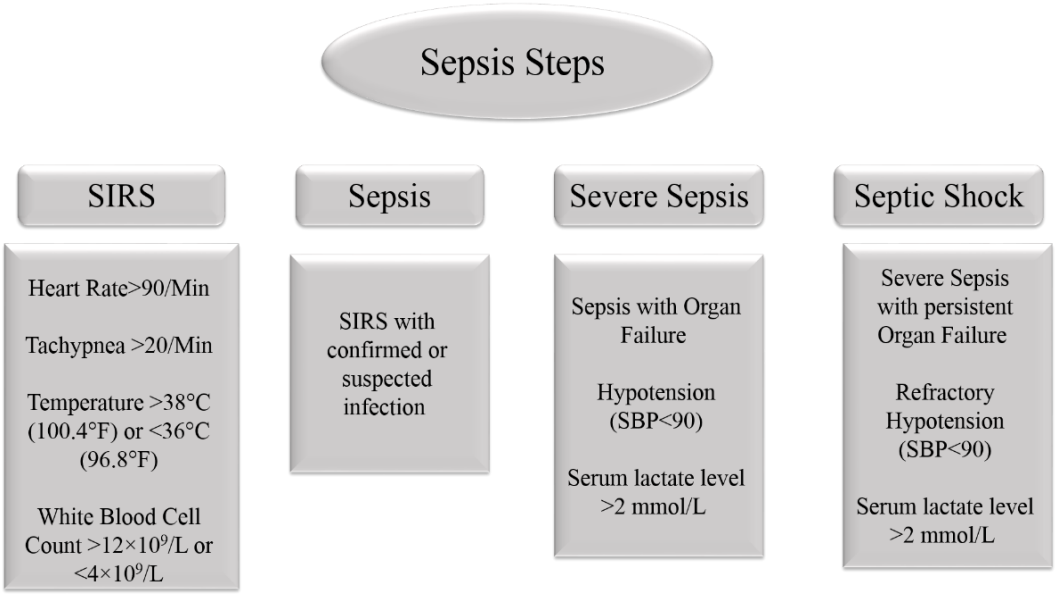

Fig. (1). Clinical definitions of sepsis. sirs; systemic inflammatory response syndrome.

Systemic Inflammatory Response Syndrome (SIRS) was characterized by abnormalities in 2 or more among 4 clinical criteria of tachycardia (heart rate>90/min), tachypnea (respiratory frequency>20/min), temperature>38°C (100.4°F) or <36°C (96.8°F), and white cell count>12×10^9/L or <4×10^9/L [4].

Patients with septic shock can be clinically identified with a clinical construct of sepsis requiring vasopressor to maintain at a Mean Arterial Pressure (MAP) of 65 mmHg or greater and a serum lactate level >2 mmol/L (18mg/dL) against adequate volume resuscitation [5]. Sepsis-1 and Sepsis-2 descriptions define sepsis as an infectious source along with 2 or more SIRS criteria [6].

In February 2016, updated definitions for sepsis and septic shock, termed 'Sepsis-3,' were available by a Task Force organized by the European Society of Intensive Care Medicine and the Society of Critical Care Medicine [7]. As stated for Sepsis-3, sepsis is a severe, fatal, organic dysfunction triggered by dysregulated host response to infection [8]. In Sepsis-3, lactate levels are integrated as a proxy for cellular metabolic abnormality along with acute mortality [9]. In addition, the

task force counseled the employment of a simplified Sequential (sepsis-related) Organ Failure Assessment – known as quick SOFA (qSOFA) – as an ancillary tool to promptly recognize adult patients who are at higher risk of death or stay in the ICU for 3 or more days if they are infected [10]. The qSOFA score is positive if the patient has at least 2 of the clinical criteria given in Table **1** [11].

Table 1. Quick sequential organ assessment score (qSOFA).

qSOFA	
Respiratory Rate ≥ 22 breaths/min Systolic Blood Pressure ≤100 mm Hg Acute mental status change (Glasgow Coma Scale < 15)	0=Mortality < 1% 1= Mortality = 2-3% ≥2=Mortality ≥ 10%

Sepsis diagnosis depends on nonspecific physiological criteria and culture-based pathogen detection [12]. For early detection of sepsis, different markers are used, such as cytokine/chemokine biomarkers, acute-phase protein biomarkers, coagulation biomarkers, polymorphonuclear CD64 index, *etc* [13]. In terms of immunological alterations, a role has been proven for immunosuppression in the pathogenesis of sepsis [14]. Immunotherapeutic approaches targeted at motivating the immune system hold a significant potential for inverse sepsis-induced immunosuppression and increase in patient outcomes [15]. The immunotherapy of sepsis is divided into intravenous immunoglobulin (IVIG), corticosteroids, and inhibitory treatments [16]. The following sections provide an overview of epidemiology, etiology, pathogenesis, neonatal sepsis, and treatment of sepsis with an emphasis on immunotherapy approaches.

EPIDEMIOLOGY OF SEPSIS

Sepsis and epidemic diseases, has had a considerable effect on the history of humanity [17]. Sepsis is today the most important cause of morbidity and mortality worldwide. With treatment costs of about $50,000 per patient and 650,000 to 750,000 annual cases, severe sepsis and septic shock rank amongst the highest costly ailments in adults worldwide [18]. Sepsis has emotional impact on 1.7 million adults in the United States every year and is possibly involved in more than 250,000 deaths [19]. Numerous studies suggest that sepsis contributes to 30% to 50% of hospitalizations that end in death, with the majority of patients being admitted to hospital with sepsis rather than getting sepsis at hospital [20]. In 2017, an estimated 48.9 million incident cases of sepsis were documented worldwide and 11.0 million sepsis-related deaths were recorded, comprising 19.7% of all global deaths [21]. The most recent worldwide approximations of sepsis incidence and mortality were made on the basis of information on patients admitted to hospital in 7 High-Income Countries (HIC), indicating 19.4 million

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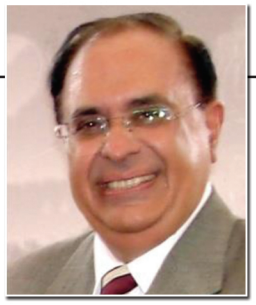

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