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ISSN : 2584-010X (Online)

SAMHITA

Multi-Disciplinary Research Journal

Vol 1 Issue 2 (July - Dec 2023)

An official publication of Bhagwan Mahavir University, Surat

"SAMHITA" *Multi- Disciplinary Research Journal*

An official publication of Bhagwan Mahavir University, Surat

"In the vast landscape of academic publishing, 'SAMHITA', a Multi-Disciplinary Research Journal, holds a unique and esteemed position. Published under the aegis of Bhagwan Mahavir University, Surat, this journal is not merely a platform for scholarly discourse but also a reflection of the university's commitment to fostering research and intellectual growth. As an official publication of Bhagwan Mahavir University, Surat, 'SAMHITA' encapsulates the institution's vision and dedication to academic excellence. Every article, research paper, and editorial published within its pages is a testament to the rigorous standards and values upheld by the university. The ownership and rights to 'SAMHITA' are securely vested with Bhagwan Mahavir University, ensuring that its legacy of quality and integrity remains uncompromised."

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Vol 1 Issue 2 2023 (July - December 2023)

Editorial board

Message from Chief Patron

Shri. Anil Jain
Founder Member & Managing Trustee, Bhagwan Mahavir Education Foundation, Surat, Gujarat, India

I am honored to present the inaugural issue of the "SAMHITA" Multi-Disciplinary Research Journal, an official publication of Bhagwan Mahavir University, Surat. This journal embodies our commitment to academic excellence, fostering communication between scholars, and promoting innovative ideas in the sciences.

I'd like to commend the dedication of Bhagwan Mahavir University and the Editorial team in bringing this initiative to fruition. I anticipate fruitful collaborations in upholding the high standards of this journal. Our vision with SAMHITA is not just to present research, but to stimulate discussions, provoke thought, and inspire future investigations. We believe that in today's interconnected world, multi-disciplinary research is the keystone to innovation and progress.

Wishing everyone a valuable learning experience from this first issue!

A Message from Chief Patron

It is my pleasure and great privilege to present to you the introductory issue of "SAMHITA" Multi-Disciplinary Research Journal the official publication of Bhagwan Mahavir University, Surat. University Journals aims to provide high quality, reviewed, open access infrastructure for scholarly articles and other products of research.

"SAMHITA" Multi-Disciplinary Research Journal is a small step towards achieving our quality standard initiatives for academic excellence. Academic journals enable communication between scholars, form the basis for the development of further ideas, and track emerging ideas in the field of sciences. The journal provides an apt platform for reporting significant findings of research for both college teachers and students. The papers submitted to the journal undergo a rigorous peer review process.

Prof. Sanjay Jain
President, Bhagwan Mahavir University, Surat, Gujarat, India

I am delighted to congratulate Bhagwan Mahavir University and the Editorial team, for their commitment and drive in launching the journal. I also look forward to our teamwork in creating guideline to maintain the good standards of journal.

I wish you all have a good learning experience from this first issue of journal!!

A Message from Patron

Dr. Nirmal Sharma
Provost, Bhagwan Mahavir University, Surat, Gujarat, India

In this era of industrial development and the economic growth scenario, research has become a backbone of the progress. Industry and academia needs to go hand in hand for the entire process of the development. Our economy is required to be routed from developing country to the developed country.

Research publications by the scholars from the Universities can and will bring a desired change in industrial and economic growth. I am pleased to extend my most sincere congratulations to Bhagwan Mahavir University and the Editorial team, whose dedication, commitment, and scholarly excellence is reflected by publishing first issue of "SAMHITA" Multi-Disciplinary Research Journal under the umbrella of Bhagwan Mahavir University. "SAMHITA" Multi-Disciplinary Research Journal showcases the creative and multidisciplinary publication of Master's and Doctoral students in subject areas that include Engineering, Pharmacy, Science, Management, Commerce, Computer Application, Health Science, Education & Humanity. The journal culminates and disseminates the excellent research and scholarly contributions of faculties, research scholars & students.

I want to extend my most sincere congratulations to editorial team of Bhagwan Mahavir University and to faculties research scholars who, submitted their review article research work, reviewed the manuscripts, and managed the publication of this journal.

It is a privilege and a pleasure to promote and share this first issue of the first volume of the Journal.

Message from Editor-in-Chief

"**Learning gives creativity, creativity leads to thinking, thinking provides knowledge and knowledge makes you great.**" - Dr APJ Abdul Kalam.
We are very glad to present the first volume of multi-disciplinary research journal of Bhagwan Mahavir University with title quot ;SAMHITA" Multi-Disciplinary Research Journal.

Dr. Zarna Dedania
Editor-in-Chief, "SAMHITA" Multi-Disciplinary Research Journal

This volume contains a wide range of research papers covering different spectrums of Engineering, Pharmacy, Science, Management, Commerce, Commuter Application, Health Science, Education and Humanity. Authors from different areas like technical, personnel, academicians and Multi-Disciplinary Researchers contributed peer quality research papers to this Journal. This journal will be very helpful to develop a new breed of Entrepreneurs and Research Scholars.

We would like to place in record the patronage and support provided by Board of Management and our beloved, BMU Provost Dr. Nirmal Sharma, Research Dean, Dr. Vineet Jain and other authorities of BMU for their encouragement in publishing this pharmaceutical research journal. The journals aim to publish a broad ranging open access journal, eminent editorials from thought out the nation, rapid publication High visibility, Expert peer-reviewed research that will serve to create innovative information.

We invite the researchers to share knowledge and research activities in the form of review and research article for the publications at "SAMHITA" Multi-Disciplinary Research Journal and hence contribute to the field of innovative research that will serve to create a holistic understanding of the human dimension in these society.

Thankful to Dr. Pooja Desai amp ; Mr. Naishadh Solanki, Associate Editor of Journal for successfully bringing this issue at right time. At the end, we hope that this issue of "SAMHITA" Multi- Disciplinary Research Journal would fortify the bond between industry, researcher amp ; academia fostering evolution of the multi-disciplinary spectrums to nation.

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About Journal

"SAMHITA" Multi-Disciplinary Research Journal (SAMHITA) is a peer reviewed, open access, official journal of Bhagwan Mahavir University, Surat in India. Journal serves local as well as a global platform for Engineering, Pharmacy, Science, Management, Commerce, Computer Applications, Health Science, Education, Architecture & Humanity research which can generate and strengthen the scientific evidences. The journal's full text is available online at Website http ://journals.bmusurat.ac.in. The journal allows free access (Open Access) to its contents and permits authors to self-archive the final accepted version of the articles.

Journal Particulars

Title	**SAMHITA Multi-Disciplinary Research Journal**
Frequency	Biannually
ISSN	2584-010X (Online)
Publisher	Bhagwan Mahavir University
Chief Editor	Prof. (Dr.) Zarna Dedania
Copyright	SAMHITA Multi-Disciplinary Research Journal
Starting Year	2023
Subject	Multidisciplinary
Language	English
Format	Online
Email ID	editor.samhita@bmusurat.ac.in
Mobile No	9427239022
Website	`http://journals.bmusurat.ac.in`
Address	Bhagwan Mahavir University, BMU Campus, VIP Road, Vesu, Surat – 395007, Gujarat, India

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About Bhagwan Mahavir University

Bhagwan Mahavir University is committed to inclusion and innovation in education through philanthropy and pioneering initiatives. Bhagwan Mahavir University provides world-class education and empowering opportunities for all sections of society. As the world of business and job opportunities are changing rapidly, we are evolving to make our students not just job-ready but also life-ready, to help them see learning as a continuous process and become future-ready professionals.

Aim & Scope

"SAMHITA" Multi-Disciplinary Research Journal (SAMHITA) is issued under the patronages of Bhagwan Mahavir University, Surat in India. SAMHITA is a national journal which published six monthly in English. Journal publishes papers, review articles, and short communications dealing with all aspects of the Engineering, Pharmacy, Science, Management, Commerce, Commuter Application, Health Science, Education and Humanity subjects that are of interest to all professionals with strong emphasis on originality and scientific quality.

Editorial policy

"SAMHITA" Multi-Disciplinary Research Journal (SAMHITA) is committed to the timely publication of peer-reviewed articles in journals. SAMHITA requires all authors to comply fully with current ethical standards for publication in their disciplines. Manuscripts submitted to the journal must represent reports of original research, and the original data must be available for review by the editor if necessary. All authors of a manuscript must have agreed to its submission and are responsible for its content, including appropriate citations and acknowledgments, and must also have agreed that the corresponding author has the authority to act on their behalf in all matters pertaining to publication of the manuscript. By submission of a manuscript to the journal, the authors guarantee that they have the authority to publish the work and that the manuscript, or one with substantially the same content, was not published previously, and is not

being considered for publication elsewhere. When submitting papers for publication, it is expected that the authors will provide written assurance and describe the novelty of their work or in the approach taken in their research in a covering letter.

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Review policy

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Ethics

When reporting studies on human, indicate whether the procedures followed were in accordance with the ethical standards of the responsible (institutional or regional) and

When reporting experiments on animals, indicate whether the institution's or a national research national law on the care and use of laboratory animals was followed. Animal be as humane as possible and the details of anaesthetics and analgesics used should be clearly stated. The ethical standards of experiments must guidelines provided by the CPCSEA (animal) and ICMR (human). The journal will not consider any paper which is ethically unacceptable. A statement permission and ethical practices must be included in all research articles under the 'Materials and Methods' section.

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Vol 1 Issue 2 2023 (Jul - Dec 2023)

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A Review on Novel Approaches to the Treatment of Asthma

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Abstract: *Asthma is among the most prevalent disorders of the respiratory system, affecting more than 300 million people worldwide. It is a heterogeneous and intricate inflammatory disease of the airways with complicated underlying events yet to be unleashed. Despite of advances in the introduction of newer therapies, the symptoms and exacerbations caused by asthma continue to prevail. Hence, there is an emerging need for new drug delivery mechanisms to achieve therapy unrestricted to symptomatic relief as well as to overcome the hindrances faced by conventional drug delivery systems. The present review highlights a brief overview of the disease and the rising superiority of novel drug delivery systems over conventional systems in its treatment and prevention.*

***Key Words*:** Asthma, Novel drug delivery, metered dose inhaler, Dry powder inhaler, liposome, Nanoparticle, Solid lipid nanoparticle, Dendrimer

Received Date: 19/05/2023 Revised Date: 10/07/2023 Acceptance Date: 25/07/2023

1. INTRODUCTION

In his book entitled "On asthma and its treatment" published in 1860, Henry Hyde Salter defined asthma as peculiarly characterized paroxysmal dyspnoea that is typically periodic and separated by intervals of normal breathing [1]. Currently, asthma could be understood as a chronic inflammatory disease of the airways characterized by episodes of coughing, wheezing, difficulty in breathing, and chest tightness [2]. The impact of asthma on patients, families, and society is disproportionately high in low- and middle-income nations where access to competent treatment is limited. Despite a global decrease in asthma mortality in adults and children over the last 25 years, owing partly to greater usage of inhaled corticosteroids, there is still a significant global difference in years of life lost due to asthma [3]. Despite major advances in decoding the underlying pathogenesis, the exact series of events remain unclear hence, making it incurable. Two major reasons can be attributed to this, one, complexity in molecular mechanisms involved, and two, heterogenicity due to severity, differences in gene expression, genetic polymorphism, and variegated response to treatment [4]. Therefore, asthma refers to a general term that comprises a range of clinical presentations [5].

The most popular diagnostic treatments are mostly therapeutic agents such as antihistamines, corticosteroids, antileukotrienes, decongestants, mast cell stabilizers in the form of tablets, and sprays via oropharyngeal, intranasal, intratracheal routes, etc. However, these conventional dosage forms have some drawbacks mentioned as follows:

- To maintain the Drug Plasma Concentration, multiple doses are necessary.
- Drugs with a short half-life reduce patient compliance so repeated dosage is required.
- In the case of conventional dosage forms, a phenomenon known as the "typical peak-valley effect" or "see-saw effect" is observed due to variation in drug concentration in plasma, which hinders maintaining the ideal concentration of the medication.
- This modification may also result in over- or under-medication, both of which can have harmful adverse effects [6].

This led to the development of novel drug delivery systems, which offers advantages over traditional dosage forms, to overcome all of the above-mentioned restrictions.

2. ETIOLOGY

2.1. Genetics

It has been devised that genetics contribute to development of asthma in the range of 35%-95% [7]. The diversity in asthma phenotypes exists as a result of polygenic inheritance or varied combination of genes [8]. Multiple studies show that children with asthmatic parents are at increased risk of developing asthma [7]. For instance, the first Genome wide association studies (GWAS), published by Moffatt and colleagues in 2007 outlined that altered chromosome 17q12-21 is associated with childhood asthma [9]. Farzan et al. too showed that polymorphism in 17q21 locus enhanced the risk of

exacerbations in children despite treatment with inhaled corticosteroids [10]. Association studies also revealed that more than 100 genes were associated with allergy and asthma in 11 different populations [11]. ADAM33, an asthma gene, first recognized in Caucasian families from the United States and United Kingdom, was associated with bronchial hyperresponsiveness [12].

Table – 1: Major gene alterations identified in various asthma studies [7]

Possible functional groups	Genes
Atopy	HLAG, FCR1A, CD23, OPN3/CHML, CYF1P2, IL4, IL4RA, IL12, IL13, GATA3, STAT5, STAT6, TBX21, PHF11, IRAKM
Barrier function	FLG, SPINK5, CTNNA3, C11orf30, COL29A1, PNEDRIN, IL13
Eosinophils	MYB, WDR36, ILR1RL1, IL33
Epithelium	IRAKIM, TLR2, TLR4, CD14, GSTP1, GSTMI1,3,5, GSTT
Protein folding in endoplasmic reticulum	ORMDL3, GSDMB, ZPBPW, IKZFE
Tissue response	ADAM33, UPAR, NPSR1, IRAKM, IL13, COL29A1, TNC

2.2. Air pollution

Numerous epidemiological studies implicate air pollution as a major cause and aggravation of asthma [13]. Urbanization is, indeed, one underlying factor in the rising levels of air pollution leading to global burden of asthma and such respiratory ailments. Studies reveal those mothers exposed to air pollution had a detrimental effect in pulmonary development in utero [14]. Gaseous pollutants such as carbon monoxide, carbon dioxide, nitrogen dioxide, ozone, sulfur dioxide, heavy metals like lead, chromium, volatile organic compounds (VOC) and polycyclic aromatic hydrocarbons (PAH) are some of the significant ones [15]. NO2 in concentrations of more than 0.2ppb infiltrates deeper into the lungs and induce coughing, wheezing, bronchospasm, dyspnoea and even pulmonary edema [15]. SO2 is released from combustion of sulfur-containing coal and oil into the atmosphere [15]. Upon inhalation, it enters into the lungs, gets converted into bisulfite which interacts with sensory receptors to produce bronchoconstriction [15]. Among air pollutants, particulate matter (PM) has the greatest impact on human health [15]. PM can be defined as a complex blend of soot, smoke, dirt, dust and liquid droplets [15]. They act by activating oxidative stresses and initiate allergic inflammatory response [15]. Low-dose $PM_{2.5}$ has been shown to escalate Th-17 mediated cytokines, chemokines as well as neutrophil infiltration [16]. PM exposure may also result in apoptosis and autophagy in lung epithelial cells in asthma [15]. Diesel exhaust particle (DEP), is thought to enhance IL-33 signals in airway epithelium and aggravates severe airway hyperresponsiveness [10]. Indoor air contaminants, such as endocrine-disrupting chemicals (EDC) tend to increase parasympathetic activity that might enhance the risk of asthma [10].

2.3. Infections

One of the most common causes of Respiratory syncytial virus (RSV) infection in early life increases the risk of asthma [10]. Viruses such as rhinovirus, parainfluenza virus, coronavirus, influenza virus are associated with increased risk to development of asthma. Studies reveal that young children with severe symptoms during bronchiolitis are at increased risk of asthma [7]. Respiratory viruses significantly infect epithelial cells of the lungs [2]. In response to this, the healthy epithelial cells produce antiviral factors which aid in viral clearance [2]. However, in chronic inflammation of the airways of asthmatic patients, the antiviral response of epithelial cells become altered, leading to prolonged inflammation [2]. Such respiratory viruses could cause serious uncontrolled asthma exacerbations requiring urgent medical help and/or hospital admission [1]. It has been demonstrated using epithelial cells cultured in vivo that rhinoviruses render these cells resistant to apoptosis, thereby expanding the viral growth followed by their shedding leading to cytotoxic death of cells [1]. Single-cell RNA sequencing (RNA-seq) analysis on virus induced asthmatic patients revealed a gene core associated with IL-33 and epithelial cell repair [2].

2.4. Exposure to Allergens

More than 80% children with asthma are sensitized on exposure to environmental allergens, indoor allergen being most pivotal [17]. Allergens promote susceptibility to viral-induced wheezing [17]. The most common allergen in allergenic asthma is group-1 grass pollen [10]. Artemisia pollen allergy is a prominent cause of asthma in Northern China [10]. *Aspergillus fumigatus*, another potential allergen, is associated with reduced BAL macrophages, increased BAL levels of IL-4, IL-6, IL-10, IL-13 and TNF-α, and increased plasma levels of IL-4, IL-6, IL-10, IL-13, IL-17 and TNF-α [10]. However, no relationship between the presence of *A.fumigatus* and severity or control of the disease was established [10]. Cockroach allergens can activate the TLR2-trans-IL-6 signaling pathway in mice, which attracts neutrophils into the airways by increasing IL-17A production in CD-T cells, thereby contributing to neutrophilic asthma [16].

Examples of triggering agents in asthma:

- Allergic triggers- Pet dander, Dust mites, Pollens, Molds
- Physical triggers- Allergic rhinitis, Exercise

- Environmental triggers- Air pollutants, Tobacco smoke, Humidity, Cold air. [18]

Table - 2: List of aggravating factors in development of asthma [8]

Allergens	Airborne pollens House dust mites Animal dander Cockroaches Fungal spores
Drugs	Aspirin NSAIDs Non-selective β blocker
Emotions	Anxiety Stress Laughter
Environment	Cold air Fog Ozone Sulphur dioxide Nitrogen dioxide Tobacco smoke Wood smoke
Exercise	Particularly in cold, dry climate
Occupational stimuli	Bakers- Flour dust Farmers- Hay mould Printers- Arabic gum Chemical workers- azo dye, anthraquinone, ethylenediamine, toluene diisocyanates, polyvinyl chloride Plastics, rubber, wood workers- Formaldehyde, western cedar, dimethylethanolamine, anhydrides
Preservatives	Sulphites Benzalkonium chloride

2.5. Smoking

Noneosinophilic inflammation, with or without neutrophilic inflammation is commonly found in asthma patients associated with smoking [19]. Asthmatics who smoke tend to have uncontrolled symptoms, high risk of exacerbations, and increased emergency room visits [14]. Smoking during pregnancy predisposes to development of early onset asthma in the offspring [14]. It has been put forward that smoking during pregnancy affected lung functions more drastically than neonatal exposure [20]. In addition, exposure to tobacco smoke aggravates symptoms in a dose-independent manner [14]. It has been put forward that one puff of cigarette smoking contains 1016 oxygen radicals which activate inflammatory cells as well as growth factors and matrix metalloproteinases [20]. Smoking accelerates reduction in lung volume often linked with ageing [20]. In adults FEV1 diminishes after the age of 30 by roughly 30ml/year, whereas in smokers it is on average 40-45 ml/year [20].

2.6. Obesity

Obesity is considered to play a vital role in worsening asthma. A study on the same by Michael et al. showed that all patients who were on oral steroids were obese with mean BMI higher than those not taking oral steroids. One possible explanation could be increase in weight as a side effect of steroid therapy. Moreover, moderate to severe asthma tend to be prevalent in children who were obese [21]. Severity of the disease markedly affected the disease control and reduced response to inhaled corticosteroids. [22]. Another study by Schacter et al. Found that symptoms of dyspnea and wheezing in obese patients were often diagnosed to be suffering from asthma [21]. A recent study concluded that weight loss helped in improving asthma control in both children and adults, whereas improved lung function in adults. Further research, however, needs to be done on establishing a clear mechanism correlating obesity with asthma.

2.7. Age

Asthma is responsible for a significant rise in mortality of geriatric patients [23]. Asthma in elderly patients can be classified into two groups based on age of diagnosis of the disease [23]. "Late onset" asthma is diagnosed for the first time after the age of 65, and "early onset" persists into the older age [23]. In addition, late onset asthma patients tend to be less atopic with lower levels of serum IgE as well as reduced sputum and serum eosinophils [23]. Neetu et al. Demonstrated that rate of hospitalization of older patients were more than twice than that of young adults [24]. In their study, they also found that elderly patients suffered worse-short and long term control of asthma than young adult population [24].

2.8. Gender

In a study by Teague et al., it was found that children with asthma, irrespective of severity, were primarily male with normal body mass and lung function. However, the level of blood eosinophilia and allergen sensitization were relatively high in these patients. This was contrary to what was observed in adults in the same study, wherein regardless of severity, most asthmatics were females being more obese and airflow limitations [25].

3. TYPES OF ASTHMA

Classically, depending on the type of immune cells involved, asthma is divided into: (a) Type 2 or allergic asthma and (b) non -type 2 or non-allergic asthma [26]. Type 2 asthma is associated with T helper 2 (Th2) cell responses [2]. These cells, upon recognition of an allergen, produce type 2 cytokines IL-4, IL-5, IL-9, IL-13 [2]. This results in the accumulation of eosinophils, increased mucus production, whereas, allergen specific B-cells synthesize immunoglobulin E (IgE) [2]. This type of asthma generally begins in early life wherein exposure to allergens such as

pollen, dust, animal dander causes sensitization, but it can also be induced later in life, as seen in occupational asthma [2]. Non-type 2 asthma, on the other hand, does not involve Th2 cells or eosinophilic inflammation. It is often associated with obesity, ageing and smoking, thereby making it a late-onset disorder [2]. However, categorizing asthma in this manner is broad [2].

In response, experts established another way of classifying asthma that could aid in a better understanding of the disease. Woodruff et al. And Wenzel et al. Classified asthma into two endotypes (Th2 high and Th2 low) based on clinical features and biomarkers [16]. Endotypes are determined by underlying pathophysiological mechanisms that may result in direct variances in responsiveness to common therapy like inhaled corticosteroids or unique biologicals. Based on this, asthma can be divided into:

1. type 2-high or ultra-high (eosinophilic)
2. type 2-low (non-eosinophilic or neutrophilic) [2]

Hence, classification based on phenotypes and endotypes allows for the development of individualized and targeted medicines to address currently unmet requirements in the management and control of asthma, particularly severe asthma [27].

3.1. Type 2-high asthma

Type 2 immunity and inflammation are thoroughly researched and understood. It is distinguished by the production of type 2 cytokines such as IL-4, IL-5, and IL-13, as well as high levels of IgE antibodies. Type 2 cytokines are the primary regulators of type 2 inflammation [27]. More severe form is represented as ultra-type2-high asthma [2]. It is caused by allergen-induced IgE synthesis and Th2-associated cytokine production. IgE facilitates mast cell and basophil degranulation after allergen recognition via FcεRI cross-linking, and FcεRI activation results in the generation and release of inflammatory mediators. Eosinophil production, transport, and activation, goblet cell differentiation and mucus hypersecretion, airway hyperresponsiveness, and airway remodeling are all mediated by IL-4, IL-5, and IL-13 [16]. In response to TSLP, IL-25, and IL-33 signaling, ILC2s are activated and release type 2 cytokines (IL-4, IL-5, IL-9, IL-

13) and prostaglandins, boosting Th2-driven allergic reactions in the airways [26]. When exposed to allergens, epithelial cell-derived IL-25, IL-33, and thymic stromal lymphopoietin (TSLP) increase ILC2 amplification and release high amounts of type 2 cytokines, boosting airway

inflammation. Although corticosteroids may treat most Th2 asthma, in some cases of late-onset eosinophilic asthma, persistent eosinophilia is noted despite high doses of glucocorticoids, and these patients often present with severe asthma [16].

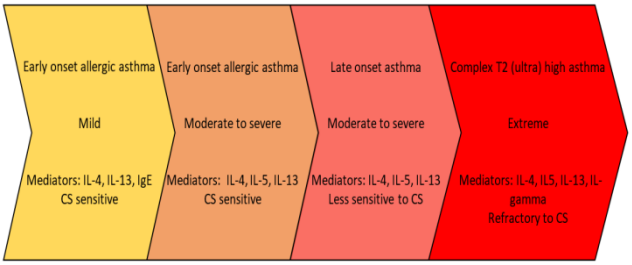

Fig -1: Progression of Type 2- high asthma [2]

3.2. Type 2-low asthma

The underlying pathophysiological mechanisms of type 2 low asthma are substantially less understood than those of type 2 high asthma [27]. The type 2-low endotype is more complex, with no biomarkers discovered thus far. As a result, type 2-low asthma encompasses all asthmatic individuals who do not have type 2-high inflammation [2]. One feature of such patients is a high amount of neutrophils (rather than eosinophils), which is generally associated with a high level of TH1 and TH17 cells. Resistance to steroid treatments is seen in neutrophilic asthma caused by TH17 inflammatory mediators, which appears to be a typical hallmark of type 2-low asthma [27].

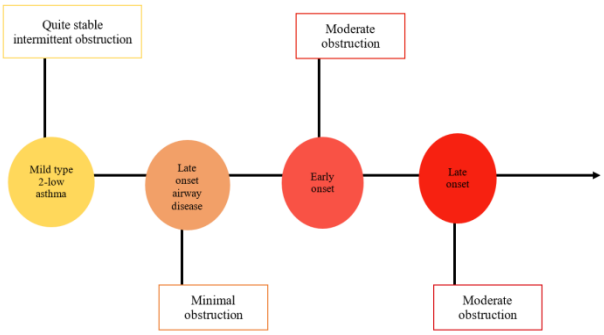

Fig -2: Progression of Type 2- low asthma [2]

3.3. Epidemic Thunderstorm asthma (ETSA)

Acute asthma exacerbations developed post-exposure to thunderstorms are referred to as Epidermic Thunderstorm Asthma (ETSA). On 21 November 2016, the most devastating epidermic thunderstorm asthma catastrophe in history occurred in Melbourne [28]. Thousands were hospitalized for severe respiratory ailments [29]. During the 12-hour storm, hospital admissions for asthma surged by over 1000%, a record breaking figure for which the health care system was unprepared [29]. Approximately 87% of the admitted patients suffered from allergic rhinitis, 28% were diagnosed with asthma, and 10 deaths have been reported [28]. Thunderstorms have the ability to bring airborne-allergens to the ground level, among which pollen grains, particularly grass pollen, are the most prominent [29]. Fungal spores may also be present in greater quantities in the atmosphere [29]. On exposure to moisture, as in rainfall, the pollen grains swell up and rupture due to osmotic shock, thereby releasing numerous fine allergenic

starch granules into the air [29]. These granules are fine enough to penetrate deep into the airways and trigger allergic responses, leading to asthma attacks in sensitized patients as well as in people without a history of asthma [29,8]. Unusual whether environmental factors brought on by climate change contribute to the global emergence of ETSA [29].

3.4. Aspirin-exacerbated respiratory disease (AERD)

Aspirin-exacerbated respiratory disease (AERD), a subcluster of Th2-high asthma, is induced by aspirin or any NSAID that inhibits the COX-1 enzyme [5,30]. This is characterized by a triad of asthma, eosinophilic rhinosinusitis, and nasal polyposis [30]. Patients with AERD are believed to have lower baseline lung function than those with aspirin-tolerant asthma, as indicated by the presence of airway remodeling [30]. When such patients ingest a COX-1 inhibitor, within 60-180 minutes an acute reaction develops involving symptoms comprising of coughing, nasal congestion, sneezing, rhinorrhea, wheezing, and fall in lung functions [30]. However, these are considered to be hypersensitivity reactions rather than allergic reactions [30]. Blocking of COX pathway redirects the arachidonate substrates to the 5-lipoxygenase pathway which produces cysteinyl leukotrienes (Cys-LTs) [31]. Mast cells, eosinophils and platelet adherent leukocytes have 5-lipoxygenase and leukotriene C4 synthase enzymes which function in the synthesis of cysLTs [31]. These cells and consequently, the cysLTs are elevated in AERD and are responsible for edema, bronchconstriction, smooth muscle hypertrophy, fibrous collagen depositions and mucus secretion into the airways [31,30]. Apart from this, patients with AERD have elevated levels of PGD2 (produced by mast cells), another inflammatory mediator responsible for extra-respiratory symptoms, particularly rashes and gastrointestinal distress [30]. In addition, eosinophils, basophils and platelets may be involved [30]. However, further research is required to understand a precise mechanism underlying the disease pathogenesis. Although rare in children, aspirin-exacerbated respiratory illness is more common in persons with severe asthma (15% vs 7% in the general population) and is typically accompanied by rhinitis and nasal polyposis [3].

4. MORPHOLOGIC FEATURES

In asthma, histopathological abnormalities in the bronchial and bronchiolar walls include the mucosa (the epithelium and lamina propria), submucosa (which includes airway smooth muscle (ASM) and mucus-secreting glands), and adventitia (the contact between the airway and the surrounding lung parenchyma). Airway epithelium seems weak in endobronchial biopsies, as evidenced by partially or totally denuded regions. Asthmatics have more epithelial cells in their bronchoalveolar lavage, indicating the possibility of epithelial desquamation in the airway lumen. This breakdown of the mechanical and biochemical dynamic barrier can result in submucosal cellular activation, which is referred to as an aberrant epithelial mesenchymal unit [32].

5. SYMPTOMS

Wheezing, shortness of breath, chest tightness, and cough are some of the symptoms of asthma. The most distinguishing elements of asthma are related to the pattern of symptoms, such as symptom kind, timing, triggers, and response to therapy. However, Exercise-induced bronchoconstriction may be the only symptom of asthma, especially when combined with high-intensity aerobic exercise, cold dry air, or chlorinated swimming pools. As a result, meticulous history taking is required to determine the likelihood that respiratory symptoms are caused by asthma rather than another diagnosis or condition [3].

6. PATHOPHYSIOLOY

As previously stated, asthma is the result of a complex interaction between genetics, environmental factors, as well as innate and adaptive immunity.

A collection of individuals with comparable clinically discernible traits are referred to as phenotypes because they lack a clear etiologic link to a particular pathophysiologic mechanism. Contrarily, an endotype describes a subgroup that exhibits the same pathophysiologic events that result in the onset, progression, and manifestation of a disease [33].

The following events play a significant role in the pathogenesis of asthma:

6.1. Inflammation

Airway inflammation is a multicellular process that involves CD4+ T lymphocytes, eosinophils, neutrophils and mast cells [34]. Although inflammation occurs largely in the airways, it may also involve terminal bronchioles and sometimes alveoli in severe cases [35,34]. The initiation of an inflammatory response in asthma begins when an allergen paves its way into the airways. Dendritic cells, which are antigen presenting cells present in the airway epithelium, assimilate these allergen with the aid of cathepsin S [35,34]. These peptides are then presented by HLA molecules (MHC class II). The dendritic cells then migrate to lymph node so as to initiate production of allergen-specific T-cells [35]. Hence, dendritic cells play a significant role in allergen-induced airway inflammation. There are roughly 500 dendritic cells per mm^2 within the epithelium [32].

6.2. T lymphocytes

By releasing particular cytokine patterns that promote eosinophil recruitment and survival as well as mast cell maintenance in the airways, T lymphocytes play a critical role in directing the inflammatory response in asthma [35]. Two subsets of effector CD4+ Th cells with distinct activities and cytokine secretion patterns were discovered and designated as type 1-Th (Th1) and type 2-Th (Th2) respectively [36]. Circulating memory Th2 cells localized in parenchyma function in eosinophil and T cell recruitment to the lung [2]. On the contrary, CD4+ resident memory T

cells (Trm) are seen in the lungs of patients with type 2-high asthma [2]. These cells release more Th2 cytokines than circulating Th2 cells and respond quickly on re-exposure to allergen albeit the mechanism is not clearly understood [2]. Trm cells induce mucus production, eosinophil activation, and bronchial hyperresponsiveness [2].

6.3. Mast cells

Mast cells are degranulated leukocytes which contain proinflammatory mediators such as histamine, proteases, lipid bronchoconstrictors such as cysteinyl- leukotrienes (LTD4, LTE4) and prostaglandins (PGD2) [37]. Although they are usually located in lamina propria in normal human airways, in asthma they are found in the airway epithelium, mucus glands and smooth muscles [32]. They are activated by allergens through IgE dependent mechanism [35]. Increased serum level of IL-33 is thought to be linked to activation of mast cells [10]. Mast cell counts are elevated in eosinophilic and Th2-high asthma, but normal in the submucosa of individuals with severe noneosinophilic asthma and the epithelium of nonsmokers with Th2-low asthma [19].

6.4. Neutrophils

The accumulation of neutrophils into the airways can cause significant tissue damage during inflammation [38]. This is because it is an inherent source of mediators such as prostaglandins, thromboxanes, LTB4 and PAF which aggravates inflammatory response [38]. Neutrophils may play a role in pathophysiology of asthma by releasing these mediators along with reactive oxygen species, enzymes namely elastase, cathepsin G, myeloperoxidases and non-enzymatic defensins [32]. Neutrophils are associated with non-allergic and steroid resistant asthma [5].

In severe neutrophilic asthma, antigen-presenting cells (APCs) encounter allergens and transport them to draining lymph nodes, where they activate naive Th0 cells. Th0 cells differentiate into Th17 cells and produce cytokines in the presence of particular cytokines (e.g. IL-17 and TNF-a). These cytokines then act on epithelial cells, promoting the release of IL-8, CXCL1, and granulocyte colony stimulating factor (G-CSF), all of which enhance neutrophil activation and recruitment. IL-8 is secreted by recruited neutrophils, exacerbating neutrophilic asthma. IL-8 is secreted by recruited neutrophils, exacerbating neutrophilic asthma. In the presence of specific stimuli, activated mucosa-associated invariant T (MAIT) and CD T cells create IL-17-secreting isoforms, which increase cytokine secretion (e.g., IFN-c, IL-17, and TNF-a), promote neutrophil recruitment, and aggravate asthma symptoms. The inflammasome (Nod-like receptor protein 3 [NLRP3]) self-oligomerizes to recruit apoptosis-associated speck-like proteins such as CARD (ASC) and pro-caspase-1. Activation caspase-1 causes pro-IL-1b and pro-IL-18 cleavage and activation, which results in a significant amount of IL-1b and IL-18 production and promotes the development of neutrophilic asthma. Certain miRNAs are secreted more often in neutrophilic asthma, and miRNAs enhance IL-6 and IL-8 production by airway epithelial cells. Asthmatics have higher exosome release, and exosomes enhance inflammatory cell activation and infiltration. They also contribute to airway remodeling [16].

6.5. Cytokines

Cytokines are a family of small glycosylated proteins that function in cell signaling, cell growth, differentiation, proliferation, chemotaxis, immunomodulation, immunoglobulin isotype switching and apoptosis [32]. Cytokines can exert their actions on adjacent cells, cells at distant sites, and cells which themselves produce them [32]. Since the functions of cytokines tend to imbricate, their individual roles in pathogenesis of asthma is arduous to characterize [32]. The cytokines which perpetuates chronic inflammatory response to asthma are those derived from T -lymphocytes [12]:

- IL-3 is crucial for the survival of mast cells in tissues and induces eosinophilia in vivo [35, 39].
- IL-4 is responsible for stimulating B lymphocytes to produce IgE, Th2 cell differentiation, enhances mucin expression on goblet cells, and expression of VCAM-1 on endothelial cells [35,39]
- IL-5 enhances eosinophil maturation and prevents apoptosis of inflammatory cells [12]
- IL-9 plays critical role in airway remodeling and eosinophilopoiesis, may mediate pathological functions along with IL-4 [12].

Apart from these, macrophages and epithelial cells too release cytokines like IL-1β, IL-6, TNF-α, and GM-CSF, which may amplify the inflammatory response [35]. Another cytokine worth mentioning is Thymic stromal lymphopoietin (TSLP), produced by airway epithelial cells and mast cells of asthmatic patients [12]. It is thought to play a significant role in stimulating dendritic cells to release CCL17 which attracts Th2 cells via CCR4 [12]. Recent studies on IL-25, IL-33 and TSLP reveal their role in initial priming of Th2 responses culminating in asthma [40]. It has been devised that IL-17 is associated with the development and severity of airway neutrophilia [41]. IL-6 is a pleiotropic cytokine produced by cells in response to inflammatory stimuli [4]. It is one such cytokine associated with neutrophilic airway inflammation [4]. It acts as an indicator of metabolic dysfunction along with asthma severity thereby making it a potential biomarker in obese asthmatic patients [4].

6.6. Eosinophils

Infiltration of eosinophils plays a prominant role in differentiating asthma from other non-infectious inflammatory disease of the airways [12]. In addition, asthma itself can be categorized into eosinophilic and noneosinophilic asthma based on presence or absence of airway eosinophils [41]. Hence, it could be inferred that eosinophils are not essential in clinical manifestations of asthma [41]. Mediators such as IL-5, granulocyte-macrophage colony stimulating factor (GM-CSF), eotaxin and CCL5 aid in the recruitment of eosinophils and lymphocytes into the airway wall of asthma patients. Their

recruitment initially involves adhesion of eosinophils to vascular endothelial cells by expression of specific glycoprotein molecules on the surface of eosinophils (integrins) and their expression of molecules such as ICAM-1 in the airway circulation [12]. They then migrate towards submucosa and subsequent activation take place [12]. The activated eosinophils release mediators near airway nerves, which changes the tone of parasympathetic and sensory nerves, thereby promoting BHR [2]. Apart from this, eosinophilic-associated fibrogenic factors such as TNF-β leads to airway remodeling characterized by smooth muscle thickening, goblet cell metaplasia and deposition of extracellular matrix protein [2]. In addition, persistent airway inflammation associated with eosinophils damages lung structural cells induced by the release of cytotoxic granule proteins (MBP, EPO, EDN) [2]. IL-5 plays a crucial role in maturation and recruitment of eosinophils into the airways [34]. Accordingly, anti-IL-5 therapy has demonstrated clinical efficacy in hypereosinophilic syndromes [41]. Recent studies on the clinical efficacy of mepolizumab, an anti-IL-5 monoclonal antibody was shown to reduce both blood and sputum eosinophil levels that reduced asthma exacerbations and facilitated reduction in oral corticosteroid therapy [41]. Eosinophils are linked to the development of AHR through the release of basic proteins and oxygen-derived free radicals [12]. Eosinophils from asthmatic patients show exaggerated responses to PAF and phorbol esters on comparison with eosinophils from atopic nonasthmatic individuals [12].

6.7. Macrophage

Monocytes and macrophages are prevalent in airway mucosa in chronic asthma [34]. Macrophages can be activated by allergen via low-affinity IgE receptors particularly FcεRII [35]. Macrophages, along with epithelial cells produce mediators such as endothelin-1, profibrotic cytokines like TNF-β and PDGF [35]. Alveolar macrophages (AM) play a defensive role in maintaining pulmonary tissue homeostasis [5]. They also phagocytose apoptotic bodies and thus diminish inflammatory response in the airways [5.]. In asthma, however, these mechanisms are altered due to which inflammation is prolonged in the airways [5]. It is evident that the IL-10 secretion and gene transcription become defective in macrophages and monocytes, which may indicate severity of asthma by intensified inflammation [35].

6.8. Airway mucus

In asthma, the airway lumen is impaired and finally gets obstructed by localized mucus aggregation under conditions that cause mucous hypersecretion, including increased mucin production and secretion, growth of gel layer, and ineffective mucous clearance [41]. Impaired mucosal clearance is a potent contributor of fatal asthma [37]. In addition, infections with bacteria can worsen mucociliary clearance, increase mucus production and lead to persistent lower airway inflammation [17]. The most prevalent mucin released by goblet cells is Muc5AC which is also responsible for generating gels in airway secretions [42].

6.9. Airway remodeling

It has been observed the occurrence of structural alterations in asthmatic patient's airways. Collagen (type III and IV) and fibronectin deposition, increased thickness of the subepithelial basement membrane, goblet cell hyperplasia, increased ASM mass and size, angiogenesis, and fibrosis are all examples of these alterations contributing to a condition known as airway remodeling [32]. The thickness of subepithelial lamina reticularis increases with increasing severity of the disease, unrelated with duration [1]. In asthma, goblet cells take the place of ciliated cells, which results in airway remodeling [37].

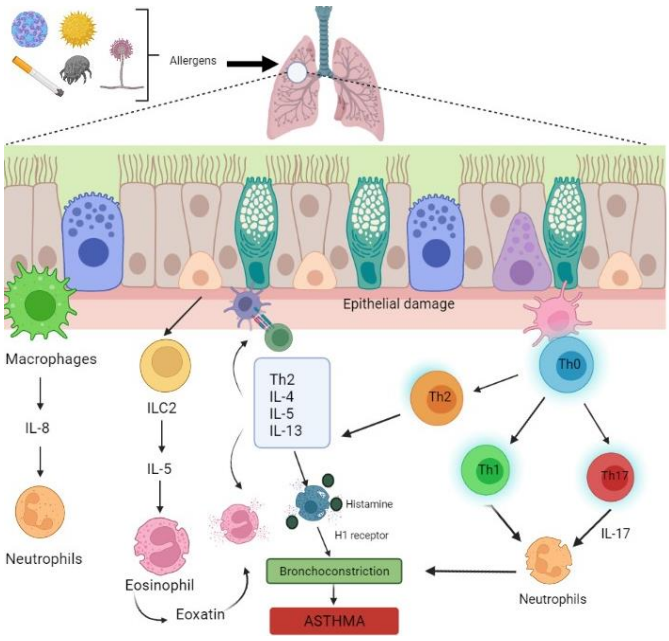

Fig - 3: Generalized pathogenesis of asthma [57]

7. TREATMENTS

As previously stated, asthma is a disease that affects airway epithelium cells, immunological cells, and various structural cell types. Details of the relationships between these cell types, as well as the interactions between currently available medicines and the host response, are being researched. Observation on the biologic reaction and the efficacy of various medicines in specific groups may one day lead to tailored therapeutics [43]. Treatment of comorbidities and modifiable risk factors, modification of lifestyle towards a healthier one by avoiding exposure to tobacco, promoting weight loss in obese patients, sublingual immunotherapy for some patients, removal from occupational exposures as well as ceasing the use of NSAIDs and inhaler training are all to be included in the individualized approach to treating asthma [3]. Medicines continue to be the mainstay of treatment, notwithstanding the importance of allergy avoidance and the control of comorbidities such as smoking and obesity. Despite modern therapy and management options, a considerable majority of asthma patients still have poorly managed illness and are vulnerable to acute exacerbations, which are mainly triggered by a respiratory viral infection. As a result,

innovative medicines to improve control and prevent asthma exacerbations remains needed. Inhaled corticosteroids β2 adrenergic receptor agonists, injectable immunoglobulin E antibodies, and quick relief medicines are used in combination treatment of asthma [43].

Table – 3: Overview of drugs used in asthma [27]

Drug	Category	Target	Mechanism of Action	Medical use	Examples
ICS (Inhaled Corticosteroids)	Long-term control medications	Glucocorticoid receptors	Suppression of airway inflammation	Mild/Moderate persistent asthma	Budesonide, beclomethasone, fluticasone
LABA (Long Acting β_2 agonists)	Long-term control medications	β_2AR	Relaxation of bronchial smooth muscle	Moderate to severe asthma	Salmeterol, formoterol, olodaterol
LAMA (Long Acting Muscarinic Antagonists)	Long-term control medications	MRs (functional selectivity for M3R)	Relaxation of bronchial smooth muscle	Uncontrolled asthma	Tiotropium, umeclidinium, glycopyrrolate
LTRA (Leukotriene Receptor Antagonists)	Long-term control medications	CysLT receptor 1	Antagonize actions of leukotrienes in the airway	Secondary option for mild/moderate persistent asthma	Montelukast, pranlukast, zafirlukast
Other leukotriene modifier	Long-term control medications	5-LOX	Inhibit the formation of leukotrienes	Secondary option for mild/moderate persistent asthma	Zileuton
Methylxanthines	Long-term control medications	Phosphodiesterase 3, adenosine receptors	Relaxation of bronchial smooth muscle	Secondary option for mild/moderate persistent asthma	Theophylline
Mast cell stabilizer	Long-term control medications	Calcium-activated potassium channels	Inhibit the release of inflammatory mediators	Mild persistent asthma	Cromolyn
Oral and intravenous corticosteroids	Quick-relief (rescue) medications	Glucocorticoid receptors	Suppression of airway inflammation	Severe asthma	Prednisolone, prednisone, methylprednisolone
SABA (Short Acting β_2 Agonists)	Quick-relief (rescue) medications	β_2AR	Relaxation of bronchial smooth muscle	Severe asthma	Albuterol, levalbuterol

SAMA (Short Acting Muscarinic antagonists)	Quick-relief (rescue) medications	MRs (no selectivity for M3R)	Relaxation of bronchial smooth muscle	Acute asthma exacerbation	Ipratropium, oxitropium
Antibodies	Quick-relief (rescue) medications	IgE and ILs	Reducing inflammation by blocking IgE and ILs	Allergic and severe asthma	Omalizumab, mepolizumab, dupilumab

8. NOVEL DRUG DELIVERY FOR ASTHMA

The term "novel drug delivery system" (NDDS) refers to the development of new pharmaceutical forms with modified properties such as lower particle size, greater permeability parameters, and selective site targeting [44]. When compared to their impact in traditional dosage forms, NDDSs can be employed to improve the performance of biotherapeutic drugs [44]. Some of the approaches that favors enhanced efficiency of asthma therapy with the aid of NDDS are described as follows:

8.1. Pulmonary drug delivery

Inhalation is a preferable route of drug administration in the treatment of asthma as well as COPD. The use of inhaled epinephrine for asthma treatment was first documented in England in 1929 [39]. Inhalational devices are classified into 4 categories: metered dose inhalers, soft mist inhalers (SMIs), dry powder inhalers (DPIs), and nebulizers. [45]. In truth, no inhalation device meets all of the requirements for delivering medications with greater patient compliance [46]. Dry powder inhalers (62.8%-88.5% of patients) and pressurized metered dose inhalers (18.9- 35.3% of patients) are most widely recommended inhalers for maintenance management of asthma and COPD [46].

Advantages of Pulmonary drug delivery:

- Enzymatic activity is relatively modest.
- Large surface area for absorption (100 m^2).
- A large vasculature.
- Short air-blood exchange channel due to weak alveolar epithelium [47].

8.1.1. Metered dose inhalers

The pressurized metered dose inhaler, often known as metered dose inhalers, is the most extensively used delivery device for medicine aerosol delivery. Metered dosage inhalers have several advantages, including mobility, the absence of an external power source, and the delivery of a set dose. pMDIs are favoured devices for administering drugs such as bronchodilators, steroids, anti-inflammatory compounds, and anticholinergics [46]. Hence, MDIs remains the most commonly used devices [45]. The pMDIs allow effective aerosolized administration of drugs. A pMDI is a pressurized system that contains roughly 1% of the total contents of propellants, flavouring agents, surfactants, preservatives, and active medicament [46]. Breath-actuated pMDIs, such as the Easibreathe® device detects the patient's breathing rate and automatically adjusts the trigger sensitivity for device activation. These are breath-controlled devices that synchronize the pace of inhalation with the release of the dose from the inhaler [46]. The issue of poor synchronization between the patient's breath and inhaler actuation can be avoided in this manner [46]. Azmacort®, which includes the inhalational corticosteroid triamcinolone acetonide, is an example of metered dose inhaler [46]. It requires a spacer device such as Aerochamber for proper administration [46]. It comes in the shape of a canister and should be thoroughly shaken before use [46]. Flovent HFA is another example of a pMDI; it has a metering valve linked to an aluminium canister and contains fluticasone, an inhaled corticosteroid suited for delivery [46]. The disadvantage of taking Flovent HFA is that it is ineffective in treating acute bronchospasm [46].

Advantages of pMDI:

- It is portable and simple to carry around [48].
- The patient's effort to inhale does not affect the amount of aerosol that is released [48].
- The inhaler canister's contents are shielded from contamination or oxidation [48].
- Suitable across age groups, with the aid of spacers or face masks [48].
- Can be used for both acute and chronic clinical conditions [48].

Disadvantages of pMDI:

- pMDIs necessitate perfect coordination between actuation and inhalation, which can be difficult, particularly in youngsters and the elderly [48].
- High oropharyngeal deposition [48].
- When delivered under pressure, the drug is at a low temperature and might induce pharyngeal discomfort [48].

8.1.2. Dry powder inhalers

Dry powder inhalers are (DPIs) yet another type of pulmonary delivery device that requires less synchronization between the actuation and breathing process to deliver powdered medications to the respiratory tract [49]. DPIs offer superior chemical stability since they

are made up of dried drugs as opposed to suspensions or solutions [49]. Dry powder is often composed of micronized medication particles in conjugation with large-sized excipients such as lactose, sucrose and glucose [49]. DPIs come in a variety of forms such as single-unit dose, multi-unit dose, and multiple reservoirs [49]. However, the formulation and production of dry powder particles with adequate properties for aerosolization and pulmonary administration is complicated [49]. Another challenge in the design of dry powder inhalers is the balance between flow rate and inhaler resistance in the device [46]. A quicker airflow is required in DPIs to promote particle deagglomeration, and greater impacts can yield a larger fine particle percentage [46]. Nevertheless, fast airflow increases the likelihood of deposition in the oropharynx and limits medication powder distribution to the lungs [46]. Depending on how reliant they are on the patient's inspiratory flow, DPIs can be either active or passive [50]. DPI formulations have been produced using a variety of standard procedures [51]. These approaches, however, have several drawbacks, including particle size, size dispersion, structure and insufficient control over powder crystallinity [51]. These issues can be resolved using specialized milling processes [51].

Table – 4: List of some common inhaler devices used in Canada [50]

Trade Name	Drug(s)	Device Type	Doses per unit	Drug Class
Onbrez®	Indacaterol	Breezhaler®	30	LABA
Breo®	Fluticasone furoate + vilanterol	Ellipta®	30	LABA+ICS
Flovent®	Fluticasone propionate	MDI	120	ICS
Ventolin®	Salbutamol	Dsikus®	60	SABA
Pulmicort®	Budesonide	Turbuhaler®	200	ICS
Spiriva®	Tiotropium	Handihaler®	30	LAMA

Advantages of DPI:

- DPI is easier to use and has benefits of enhanced medication stability and being free of propellant [52].
- DPI is suitable for drug delivery of hydrophobic formulations [52].
- Patient coordination is not required [48].

Disadvantages of DPI:

- The biggest problem with passive DPIs is the variation in inspiratory forces across patients of various ages and health conditions, which affects dose uniformity. One approach to overcome this is the use of active DPIs which incorporate internal energy sources so that dose uniformity is not reliant on the patient's inspiratory flow rate [49].
- Oropharyngeal deposition resulting in adverse effects such as oral candidiasis [48].
- It is vulnerable to both ambient humidity and exhaled air which might reduce DPI's efficiency [48].

8.1.3. Soft mist inhalers

These are the devices that create a soft mist of aerosol for the patient to inhale without using propellants [48]. They have the qualities of both pMDIs and nebulizers, with the liquid drug in is administered at a slower pace than a pMDI [48]. Soft-mist inhalers have a great rate of deposition (approximately 35-50%) than pMDIs, according to studies [48]. Respimat® 1 Soft MistTM inhaler (SMI) is a new, propellant-free inhaler designed to enhance inhaled drug over pMDIs or DPIs [53]. The major aims of designing the Respimat® SMI were to prevent the usage of propellants, lower the amount of inspiratory effort required by the patient, increase drug administration, and improve patient compliance [54]. These are currently unavailable in India [48].

8.1.4. Nebulizers

Nebulizers are devices that produce aerosol particles ranging in size from 1-5µm for inhalation based drug delivery [49]. There are two types of nebulizers that are often used based on the type of force to create aerosols from liquid suspension/solution: jet nebulizers and ultrasonic nebulizers [49]. Jet nebulizer operates by harnessing the gas flow from a compressor [46]. The formulation is atomized through a tiny opening in the nebulizer through which the gas travels [46]. Because of the leakage, there may be considerable loss of aerosol particle during exhale [46]. Ultrasonic nebulizers are often favored for aerosol treatment because they offer a higher production capability than jet nebulizers [46]. Aerosolized particles are generated using high frequency ultrasonic waves, with the needed vibration falling within the range of (1.2-2.4 MHz) of a piezo-electric crystal [46]. Mesh nebulizer is a novel nebulizer having a mesh of numerous small holes through which the medicine is fed to produce the desired aerosol [48]. These are further classified as static mesh nebulizers or vibrating mesh nebulizers [48]. The liquid drug is pushed through the mesh holes to form the aerosol in static mesh or passive mesh nebulizers [48]. On the other hand, in vibrating mesh or active nebulizers, a piezoelectric device causes vibrations, which force the drug through the mesh [48]. An example of vibrating mesh nebulizer is Aeroneb®Go [46]. Mesh nebulizers may deliver

both liquid medication formulation and suspensions; however, the performance of suspensions appears to diminished in terms of mass of inhaled aerosol and the output rate [46]. Akhuemokhan et al. developed a microemulsion (ME) of fluticasone propionate (FP) for pulmonary administration using mesh and jet nebulizers and compared it to the drug's commercial nebulizer suspension (Flixotide® nebules) [55]. The drug was administered well by the jet nebulizer, however the microemulsion was not delivered by mesh nebulizer [55]. These promising findings were demonstrated by the aerodynamic particle size distribution created by microemulsion using jet nebulizer [55]. The formulation and the device are both critical for the effective use of the nebulization system for pulmonary targeting [46].

Advantages of nebulizers:

- Nebulizers can deliver a rather high dose to the lungs which can be useful for respiratory disorders requiring high doses [52].

Disadvantages of nebulizers:

- High oral deposition [48].
- Nebulizers typically require 15-20 minutes for drug delivery [48].

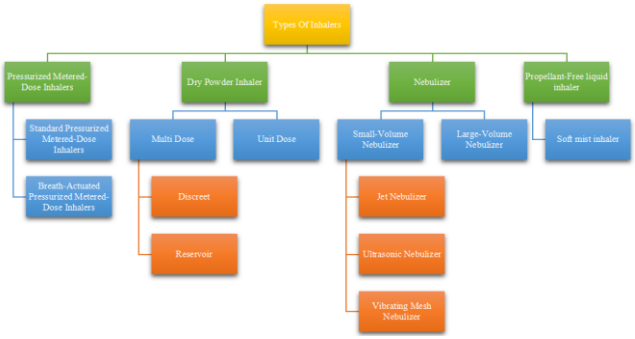

Fig -4: Types of inhalers

8.2. Particle-based drug delivery

Conventional drug therapy is typically hampered by low drug penetration and treatment response. To address this, nanoscale carriers have been employed as an advanced method to enhance pharmacokinetics of medicinal drugs. Nanoparticles, with particle size ranging from 1-100nm, are a novel technique of medication delivery [55]. These are generally intended to enhance encapsulated medication biodistribution by delivering them more effectively and selectively to the pathogenic site, also known as site-specific drug delivery [56]. The goal of this method is to improve therapeutic effectiveness while decreasing chemical toxicity [56]. Nanoparticles (NP) are classified into two types depending on their chemical composition: organic NPs and inorganic NPs [57]. Liposomes, polymeric NPs, dendrimers, and micelles are examples of organic nanoparticles, whereas inorganic nanoparticles include iron oxide NPs, gold NPs, silica NPs, quantum dots, graphene oxide, and carbon nanotubes [57].

Advantages of nanoparticles:

- The potential of nanoparticle systems to deliver targeted drugs is its primary benefit as a carrier vehicle [52].
- Nanoparticles have been employed in gene therapy to inhibit Th2 transcription factors, cytokines, and to increase the production of Th2 antagonists [58].
- Helps to enhance efficacy of existing therapies with minimal side effects [58].

Disadvantages of nanoparticles:

- Alveolar macrophages are monocyte-derived phagocytic cells that are prevalent in the lungs. Because of their small size, NPs can penetrate deep into the lungs and settle in the alveoli, where they can be consumed by macrophages. As a result, the half-life of drug-loaded NPs within the alveoli can only be a few hours, resulting in limited therapeutic efficacy and increasing dose frequency. [52].
- Distribution of NP into the lungs is difficult because their size does not allow for deep lung deposition and are hence primarily expelled through the respiratory system [52].
- Validating NP performance in vivo remains difficult. This is most likely owing to a lack of understanding of how NPs interact with complex physiological components such as pulmonary surfactants, phospholipids, and proteins in the lungs. For example, opsonin proteins found in blood serum bond to the surface of NPs, which makes them "visible" to immune cells [52].
- High production cost [49].

Some of the NPs significant in the treatment of asthma are described as follows:

8.2.1. Liposomes

Liposomes have sparked interest as a viable alternative to oral delivery for treating respiratory illnesses such as asthma since they are noninvasive and provide long-lasting therapy [57]. These are colloidal drug delivery methods made up of a lipid layer encircling an aqueous core [6]. Liposome size range from 50-100nm, depending on the cholesterol and phospholipid content [58]. The drug is distributed according to its solubility in the hydrophilic core or lipid layer [6]. Tahara et al. Developed egg phosphatidylcholine/cholesterol liposomes loaded with short acting pulmonary β2 agonist, which displayed a long term bronchoprotective effect (over 120 minutes) in a histamine induced guinea pig model as compared to the drug alone [52].

Budesonide encapsulated in stealth liposomes effectively reduced inflammation in experimental asthma, according to a study by Konduri et al. The animals accepted the treatment well and had no negative side effects. Stealth liposome encapsulation is therefore a safe and effective

vehicle for delivering inhaled steroids to the asthmatic lung [59].

An anti-asthmatic study of liposomal R-TBH conducted in guinea pigs showed that R-Terbutaline Hydrochloride (R-TBH) liposomal aerosol exerted anti-asthmatic effect 4 hours after administration, whereas no anti-asthmatic effect was observed with R-TBH. Encapsulation of R-TBH in liposomes likely protected the drug from rapid metabolism, and effective amounts of R-TBH released from liposomes were available in the lung. It was shown that sustained release of R-TBH from liposomes was achieved for at least 4 hours in vivo [60]. In a guinea pig asthma model, researchers used liposomes to treat the disease in vivo. Procaterol hydrochloride (PRO), a pulmonary β2-agonist with a short half-life, was encapsulated in liposomes and administered to the lungs. Liposomes improved drug retention in the lungs [47].

Advantages of liposomes:

- Non-toxic.
- Non-immunogenic.
- Biodegradable [6].
- Ability to incorporate wide range of active drugs.
- Ability to enhance bioavailability.
- Prevent rapid plasma clearance [61].

Disadvantages of liposomes:

- Tendency to form agglomerates [61].
- Susceptible to light, temperature, and metal ions [61].
- Difficult to sterilize [61].
- Limited potential to retain drugs for prolonged time. It can be resolved with hybrid LIP formulations [47].

Liposomes are unable to survive the shear force generated by the nebulizer; hence, physical stress is imparted to the liposomal bilayer when the aqueous dispersion is transformed into the respirable aerosol formulation. This results in the loss of the entrapped drug; however, SLN-based formulations can circumvent this [55].

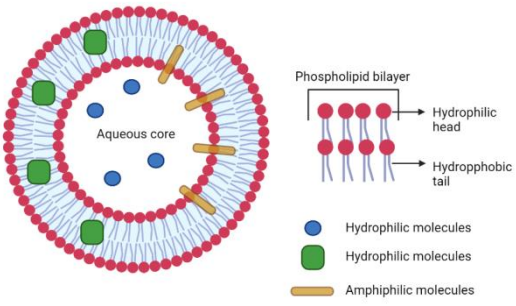

Fig -5: Generalized structure of liposomes [61]

8.2.2. Dendrimers

Dendrimers are branched macromolecules with well-defined structures [62]. They have three unique structural components: a core, interior layers made up of repeating units, and functional terminal groups [62]. Dendrimer-conjugated methyl prednisolone has been shown to improve methyl prednisolone's ability to attenuate allergen-induced inflammation [57]. In a study by Nasr et al., the solubility of poorly soluble drug beclomethasone dipropionate (BDP) was enhanced by complexing with PAMAM dendrimers [63]. The prolonged release profile, as well as the high aerosol production and Fine Particle Fraction(FPF) of BDP, imply that dendrimers are a potential nanocarrier system for pulmonary administration of BDP employing air-jet or vibrating-mesh nebulizers [63].

Advantages of Dendrimer:

- Improved cell membrane permeability [58].
- Improved bioavailability [58].

8.2.3. Solid lipid nanoparticles

Colloidal drug carriers with diameters ranging from 50nm to 1µm are known as solid lipid nanoparticles (SLN). They are composed of solid lipids or combination of liquid and solid lipids stabilized with the help of an emulsifier. The lipids often employed in the manufacture of SLN include biocompatible lipids such as waxes, fatty acids, triglycerides, and steroids that are well tolerated by the body [49].

It has following advantages:

- Ability to deliver both hydrophilic and lipophilic drugs.
- Enhanced drug stability.
- Relatively resistant to enzymatic degradation.
- Better drug delivery to the target site.
- Improved pharmacokinetic action [6].

Wang et al. used the solvent-injection approach to create curcumin-loaded SLN, which was then examined in ovalbumin (OVA)-induced asthma mice. The size of the optimized nanoparticles-based formulation was 190 nm, with a zeta potential of -20.7mV, and curcumin entrapment was determined to be 75%. Pharmacokinetic experiments were carried out in an allergic asthma model caused by ovalbumin. In the instance of SLN formulations tested in rats, a significant quantity of curcumin was identified in the lungs and liver. Curcumin-loaded SLNs inhibited T-helper-2 type cytokines, interleukin-4 and interleukin-13 significantly in vivo. In compared to curcumin alone, SLN-based formulations demonstrated active reduction of hyperresponsiveness and inflammatory cell infiltration. The study demonstrated that SLN-based formulations were superior delivery system techniques for the treatment of asthma [55].

6.8.3 Mucoadhesive drug delivery

Mucoadhesive drug delivery is mediated by the interaction of the mucus layer over the mucosal epithelium, mucin moieties, and polymer/co-polymer with prolonged dose residence time to the site of absorption [55]. The mucosa has a high blood supply and allows for quicker medication absorption than the oral route. Since it offers the chance to prevent either drug degradation by gastrointestinal contents or hepatic first-pass inactivation, pharmaceutical features of mucoadhesion have attracted a lot of attention in recent years [64].

Zhang et al. created mucoadhesive DDS-based budesonide in chitosan microparticle and assessed the characteristics of drug release in a mice model induced with allergic asthma by measuring the levels of IL-4 and IL-5 in bronchoalveolar lavage as well as mRNA [55]. The results of this study showed that, depending on the molecular weight of chitosan, drug release persisted for 12-18 hours [55]. Additionally, eosinophil number and IL-4 IL-5 mRNA levels were shown to decrease following a course of therapy lasting seven days in a row [55]. Another study reported that a mucoadhesive buccal patch containing 5 mg of Montelukast Sodium prepared by using Eudragit RL 100, and HEC, and Na CMC (F3 and F6 formulations) were best formulations [64]. these formulations of mucoadhesive buccal patches showed moderate swelling, convenient resident time, greater therapeutic efficacy, improved bioavailability and hence, turned out to be a promising one as a controlled drug delivery [64].

In a study by Lee et al., when theophylline was delivered intranasally as a complex with thiolated chitosan nanoparticles (TCN), the number of eosinophils in BAL fluid was greatly reduced primarily by inducing apoptosis or by preventing their migration. These outcomes unequivocally proved that chitosan nanoparticles delivered theophylline more effectively than unaltered chitosan or theophylline alone. One probable explanation could be the higher mucoadhesiveness of TCN is likely what caused theophylline to have stronger anti-inflammatory effects [42].

Drug delivery over mucosal surfaces have following benefits:

- Avoidance of first pass metabolism.
- Quicker absorption than oral administration.
- Drug localized at target site.
- Less expensive compared to injections.
- High patient compliance [65].

9. CONCLUSIONS

Asthma is one of the chronic respiratory diseases with a rising global burden. This demonstrates the ongoing need for new therapies. This also necessitates a thorough knowledge of the exact events taking place in the progression of the disease. While a recent breakthrough in nanotechnology has made treatment of a variety of potentially fatal lower respiratory tract infections possible, there is a need of addressing their limitations in clinical practice. This review aims to emphasize the superiority of novel drug delivery therapeutics over conventional ones, which may prove to be a more significant and prominent approach in the near future.

REFERENCES

1] Holgate ST, A Brief History of Asthma and Its Mechanisms to Modern Concepts of Disease Pathogenesis, Allergy, Asthma & Immunology Research, 2010; 2(3): 165-71.
2] Hammad H, Lambrecht BN, The Basic Immunology of Asthma, Cell, 2021; 184(6): 1469-85.
3] Papi A, Brightling C, Pedersen SE, Reddel HK, Asthma, Lancet, 2018; 391(10122): 783-800.
4] Kuruvilla ME, Lee F, Lee GB, Understanding Asthma Phenotypes, Endotypes, and Mechanisms of Disease, Clinical Reviews in Allergy & Immunology, 2019; 56(2): 219-33.
5] Hough KP, Curtiss ML, Blain TJ, Liu RM, Trevor J, Deshane JS, Thannickal VJ, Airway Remodeling in Asthma, Frontiers in Medicine, 2020; 7: 191.
6] Mehta M, Sharma N, Vyas M, Khurana N, Maurya PK, Singh H, de Jesus TP, Dureja H, Chellappan DK, Gupta G, Wadhwa R, Interactions with the Macrophages: An Emerging Targeted Approach Using Novel Drug Delivery Systems in Respiratory Diseases, Chemico-Biological Interactions, 2019; 304: 10-9.
7] Mims JW, Asthma: Definitions and Pathophysiology, In International Forum of Allergy & Rhinology, 2015; 5(S1): S2-S6.
8] Dipiro JT, Talbert RL, Yee GC, Matzke GR, Wells BG, Posey LM. Pharmacotherapy A Pathophysiologic Approach. 7th Edn. McGraw-Hill Medical New York; 2008, pp 463-493.
9] El-Husseini ZW, Gosens R, Dekker F, Koppelman GH, The Genetics of Asthma and the Promise of Genomics-Guided Drug Target Discovery, The Lancet Respiratory Medicine, 2020; 8(10): 1045-56.
10] Cevhertas L, Ogulur I, Maurer DJ, Burla D, Ding M, Jansen K, Koch J, Liu C, Ma S, Mitamura Y, Peng Y, Advances and Recent Developments in Asthma in 2020, Allergy, 2020; 75(12): 3124-46.
11] Subbarao P, Mandhane PJ, Sears MR, Asthma: Epidemiology, Etiology and Risk Factors, Cmaj, 2009; 181(9): E181-90.
12] Barnes PJ, Drazen JM, Pathophysiology of Asthma, Asthma and COPD, 2002: 343-59.
13] Vardoulakis S, Osborne N, Air Pollution and Asthma, Archives of Disease in Childhood, 2018; 103(9): 813-14.
14] Stern J, Pier J, Litonjua AA, Asthma Epidemiology and Risk Factors, In Seminars in Immunopathology 2020; 42(1): 5-15.
15] Tiotiu AI, Novakova P, Nedeva D, Chong-Neto HJ, Novakova S, Steiropoulos P, Kowal K, Impact of Air Pollution on Asthma Outcomes. International Journal of Environmental Research and Public Health, 2020; 17(17): 6212.
16] Zhang X, Xu Z, Wen X, Huang G, Nian S, Li L, Guo X, Ye Y, Yuan Q, The Onset, Development and Pathogenesis of Severe Neutrophilic Asthma, Immunology and Cell Biology, 2022; 100(3): 144-59.
17] Castillo JR, Peters SP, Busse WW, Asthma Exacerbations: Pathogenesis, Prevention, and

Treatment, The Journal of Allergy and Clinical Immunology: In Practice. 2017; 5(4): 918-27.
18] Vernon MK, Wiklund I, Bell JA, Dale P, Chapman KR What Do We Know about Asthma Triggers? A Review of the Literature, Journal of Asthma. 2012; 49(10): 991-98.
19] Thomson NC, Novel Approaches to the Management of Noneosinophilic Asthma, Therapeutic Advances in Respiratory Disease, 2016; 10(3): 211–34
20] Pietinalho A, Pelkonen A, Rytilä P, Linkage Between Smoking and Asthma, Allergy, 2009; 64(12): 1722-27.
21] Akerman MJ, Calacanis CM, Madsen MK, Relationship Between Asthma Severity and Obesity, Journal of Asthma, 2004; 41(5): 521-26
22] Peters U, Dixon AE, Forno E, Obesity and Asthma, Journal of Allergy and Clinical Immunology, 2018; 141(4): 1169-79.
23] Gillman A, Douglass JA, Asthma in the Elderly, Asia Pacific Allergy. 2012; 2(2): 101-08.
24] Talreja N, Baptist AP, Effect of Age on Asthma Control: Results from the National Asthma Survey, Annals of Allergy, Asthma & Immunology. 2011; 106(1): 24-29.
25] Teague WG, Phillips BR, Fahy JV, Wenzel SE, Fitzpatrick AM, Moore WC, Hastie AT, Bleecker ER, Meyers DA, Peters SP, Castro M, Coverstone AM, Bacharier LB, Ly NP, Peters MC, Denlinger LC, Ramratnam S, Sorkness RL, Gaston BM, Erzurum SC, Comhair SAA, Myers RE, Zein J, DeBoer MD, Irani AM, Israel E, Levy B, Cardet JC, Phipatanakul W, Gaffin JM, Holguin F, Fajt ML, Aujla SJ, Mauger DT, Jarjour NN, Baseline Features of the Severe Asthma Research Program (SARP III) Cohort: Differences with Age, The Journal of Allergy and Clinical Immunology: In Practice, 2018; 6(2): 545-54.
26] Morianos I, Semitekolou M, Dendritic Cells: Critical Regulators of Allergic Asthma, International Journal of Molecular Sciences, 2020; 21(21): 7930.
27] Wendell SG, Fan H, Zhang C, G protein–coupled receptors in asthma therapy: pharmacology and drug action, Pharmacological Reviews, 2020; 72(1): 1-49.
28] Hughes KM, Price D, Suphioglu C, Importance of Allergen–Environment Interactions in Epidemic Thunderstorm Asthma, Therapeutic Advances in Respiratory Disease, 2022; 16:17534666221099733.
29] Price D, Hughes KM, Thien F, Suphioglu C, Epidemic Thunderstorm Asthma: Lessons Learned from the Storm Down-Under, The Journal of Allergy and Clinical Immunology: In Practice. 2021; 9(4): 1510-15.
30] Laidlaw TM, Pathogenesis of NSAID-Induced Reactions in Aspirin-Exacerbated Respiratory Disease, World Journal of Otorhinolaryngology-Head and Neck Surgery, 2018; 4(3): 162-68.
31] Kim SH, Park HS, Pathogenesis of Nonsteroidal Antiinflammatory Drug-induced Asthma, Current Opinion in Allergy and Clinical Immunology, 2006; 6(1): 17-22.
32] Duangkham S, Welch N, Vega M, Shahbandar S, Tarbox J, Approaches to Studying the Pathogenesis of Asthma, The Southwest Respiratory and Critical Care Chronicles, 2022; 10(42): 7-11.
33] Holgate ST, Pathogenesis of Asthma. Clinical and Experimental Allergy, 2008; 38(6): 872-97.
34] Barnes PJ, Drazen JM, Pathophysiology of Asthma, Asthma and COPD, 2002: 343-59.
35] Hamid Q, Tulic M, Immunobiology of Asthma, Annual Review of Physiology, 2009; 71: 489-07.
36] Cosmi L, Liotta F, Maggi E, Romagnani S, Annunziato F, Th17 Cells: New Players in Asthma Pathogenesis. Allergy, 2011; 66(8): 989-98.
37] Castro M, Kraft M, Clinical Asthma; 1st Edn; Elsevier Mosby, Philadelphia, 2008, pp 23-34.
38] Kay AB, Asthma and Inflammation, Journal of Allergy and Clinical Immunology, 1991; 87(5): 893-10.
39] Rau JL, The Inhalation of Drugs: Advantages and Problems, Respiratory care, 2005; 50(3): 367-82.
40] Hansbro PM, Kaiko GE, Foster PS, Cytokine/Anti-cytokine Therapy – Novel Treatments for Asthma?, British Journal of Pharmacology, 2011; 163(1): 81-95.
41] Murphy DM, O'Byrne PM, Recent Advances in the Pathophysiology of Asthma, Chest, 2010; 137(6): 1417-26.
42] Lee DW, Shirley SA, Lockey RF, Mohapatra SS, Thiolated Chitosan Nanoparticles Enhance Anti-inflammatory Effects of Intranasally Delivered Theophylline, Respiratory Research, 2006; 7(1): 1-10.
43] Dua K, Wadhwa R, Singhvi G, Rapalli V, Shukla SD, Shastri MD, Gupta G, Satija S, Mehta M, Khurana N, Awasthi R, Maurya PK, Thangavelu L, Rajeshkumar S, Tambuwala MM, Collet T, Hansbro PM, Chellappan DK, The Potential of siRNA Based Drug Delivery in Respiratory Disorders: Recent Advances and Progress, Drug Development Research, 2019; 80(6): 714-30.
44] Seyfoddin A, Dezfooli SM, Greene CA, Engineering Drug Delivery Systems;Elsevier Science United Kingdom, 2020, pp1-16.
45] Melani AS, Inhaler Technique in Asthma and COPD: Challenges and Unmet Knowledge that can Contribute to Suboptimal Use in Real Life, Expert Review of Clinical Pharmacology, 2021; 14(8): 991-1003.
46] Chandel A, Goyal AK, Ghosh G, Rath G, Recent Advances in Aerosolised Drug Delivery, Biomedicine & Pharmacotherapy, 2019; 112:108601
47] Antimisiaris SG, Marazioti A, Kannavou M, Natsaridis E, Gkartziou F, Kogkos G, S Mourtas S, Overcoming Barriers by Local Drug Delivery with Liposomes, Advanced Drug Delivery Reviews, 2021; 174: 53-86.
48] Salvi S, Shevade M, Aggarwal A, Apte K, Barne M, Mohan MB, Ghoshal AG, Hadda V, Jaggi V, Jindal SK, Khosla I, Limaye S, Paliwal R, Rajan S, Roy S, Singh BP, Singh V, Thangakunam B, Thomas PK, Waghray P, Guleria R, A Practical Guide on the Use of Inhaler Devices for Asthma and COPD, J Assoc Physicians India, 2021; 69: 8-26.
49] Mishra B, Singh J, Novel Drug Delivery Systems and Significance in Respiratory Diseases, Academic Press, 2020: 57-95.
50] Shakshuki A, Agu RU, Improving the Efficiency of Respiratory Drug Delivery: A Review of Current Treatment Trends and Future Strategies for Asthma and Chronic Obstructive Pulmonary Disease, Pulmonary Therapy, 2017; 3(2): 267-81.
51] Patil JS, Sarasija S, Pulmonary drug delivery strategies: A concise, systematic review, Lung India: Official Organ of Indian Chest Society, 2012; 29(1): 44.
52] Kan S, Hariyadi DM, Grainge C, Knight DA, Bartlett NW, Liang M, Airway Epithelial-Targeted Nanoparticles for Asthma Therapy, American Journal of Physiology-Lung Cellular and Molecular Physiology, 2020; 318(3): L500-L509.
53] Kässner F, Hodder R, Bateman ED, A Review of Ipratropium Bromide/ Fenoterol Hydrobromide

(Berodual®) Delivered via Respimat® Soft Mist™ Inhaler in Patients with Asthma and Chronic Obstructive Pulmonary Disease, Drugs, 2004; 64(15): 1671-82.
54] Iwanaga T, Tohda Y, Nakamura S, Suga Y, The Respimat® Soft Mist Inhaler: Implications of Drug Delivery Characteristics for Patients, Clinical Drug Investigation, 2019; 39(11): 1021-30.
55] Dua K, Malyla V, Singhvi G, Wadhwa R, Krishna RV, Shukla SD, Shastri MD, Chellappan DK, Maurya PK, Satija S, Mehta M, Gulati M, Hansbro N, Collet T, Awasthi R, Gupta G, Hsu A, Hansbro PM, Increasing Complexity and Interactions of Oxidative Stress in Chronic Respiratory Diseases: An Emerging Need for Novel Drug Delivery Systems, Chemico-biological Interactions. 2019; 168-78.
56] Kang SH, Shin YS, Lee DH, Park IS, Kim SK, Ryu D, Park Y, Byun SH, Choi JH, Hong SJ, Interactions of Nanoparticles with Macrophages and Feasibility of Drug Delivery for Asthma, International Journal of Molecular Sciences, 2022; 23(3): 1622.
57] Ahmad A, Pharmacological Strategies and Recent Advancement in Nano-Drug Delivery for Targeting Asthma, Life, 2022; 12(4): 596.
58] Wadhwa R, Aggarwal T, Thapliyal N, Chellappan DK, Gupta G, Gulati M, Collet T, Oliver B, Williams K, Hansbro PM, Dua K, Maurya PK, Nanoparticle-Based Drug Delivery for Chronic Obstructive Pulmonary Disorder and Asthma: Progress and Challenges, Nanotechnology in Modern Animal Biotechnology, 2019; 59-73
59] Konduri KS, Nandedkar S, Düzgünes N, Suzara V, Artwohl J, Bunte R, Gangadharam PR, Efficacy of Liposomal Budesonide in Experimental Asthma, Journal of Allergy and Clinical Immunology, 2003; 111(2): 321-27.
60] Li Q, Zhan S, Liu Q, Su H, Dai X, Wang H, Beng H, Tan W, Preparation of a Sustained-Release Nebulized Aerosol of R-terbutaline Hydrochloride Liposome and Evaluation of Its Anti-Asthmatic Effects via Pulmonary Delivery in Guinea Pigs, AAPS PharmSciTech, 2018; 19(1): 232-41.
61] Guimarães D, Cavaco-Paulo A, Nogueira E, Design of Liposomes as Drug Delivery System for Therapeutic Applications, International Journal of Pharmaceutics, 2021; 601: 120571.
62] Mehta P, Kadam S, Pawar A, Bothiraja C, Dendrimers for Pulmonary Delivery: Current Perspectives and Future Challenges, New Journal of Chemistry, 2019; 43(22): 8396-409.
63] Nasr M, Najlah M, D'Emanuele A, Elhissi A, PAMAM Dendrimers as Aerosol Drug Nanocarriers for Pulmonary Delivery via Nebulization, International Journal of Pharmaceutics, 2014; 461(1-2): 242-50
64] Raghavendra R, Suryakar VB, Formulation and Evaluation of Montelukast Sodium Mucoadhesive Buccal Patches for Chronic Asthma Attacks, International Journal of Pharma and Bio sciences, 2010; 1: 2.
65] Jain KK, An overview of drug delivery systems, Drug delivery systems, 2020: 1-54.

Quest for Identity in That Long Silence by Shashi Deshpande

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Abstract: *Feminism plays pivotal role to lift up the life of Indian women in man dominated society. The true spirit of feminism is to look at man and woman as equal human being. The present research focuses on dilemma of woman in man dominated world, the plight of women and problems faced by women in India, especially their position in family and society. The present paper attempts to study Shashi Deshpande's women characters, her portrayal of women needs to be studied from a feminist angle. As an author of the '70s and 80s', she mirrors a realistic picture of the contemporary middle-class, educated, urban Indian woman through the character of Jaya. Through her novel That Long Silence, she portrays the miserable plight of the contemporary middleclass, urban Indian woman and also analyze how their life has not changed much even in the twentieth century. This paper discusses the dilemma of a married woman living in anguish and helplessness. In her loneliness, Jaya goes through a self- evaluation of her life and identity. This paper tries to illuminate the impact of silence in Indian women's life and Jaya's overcoming of it. The study tries to understand what makes a woman to stay silent and what eventually forces her to speak up.*

***Key Words*:** That Long Silence, Indian tradition, Male dominated society, Marriage, identity, silence.

Received Date: 18/06/2023 Revised Date: 04/08/023 Acceptance Date:17/08/2023

Feminism began to have equal space for man and woman and to look at them as equal human being. There are many feminist movements focused on equal rights and equal opportunities for women. According to Indian tradition, wives have to step in the shoes of their husband in all situations and everything after marriage. They are always subservient to their husbands and family. Even women's life is completely controlled either by her family or her husband. Women sacrifice their dreams and desires for the sake of their families and centre rounds the necessities of husband and children. They are not allowed to share their thoughts or opinions on anything. All they can do is to accept the decision taken by a man with great silence, which reflects through the female protagonist- Jaya. The writer wants such women who suffer to break their silence in the wake of feminist movement.

The novel illustrates the image of women in the middle-class family and the way she is sandwiched between the tradition and modernity. The sacrifice made by women counterpart is hardly noticed by the male dominated society.

Shashi Deshpande, a creative post-colonial writer, has wonderfully presented the problems, issues, and challenges of a middle-class married woman of modern India. Jaya the protagonist of That Long Silence represents the 90's women in our society. The title of the novel itself refers to the conditions of women in a patriarchal setup. Deshpande's novel directly deals with the dilemmas of housewives. The novel itself can be said to be an autobiographical novel of Deshpande. Deshpande has mainly focused on the life of middle-class educated women in the late 90s Indian society.

In her novel, That Long Silence Deshpande has portrayed educated middle-class Indian women who get entangled in marriage and traditions. Jaya is the protagonist of the novel. Jaya gets married to Mohan who is an educated man working as an engineer in a company. Jaya and Mohan are married for seventeen years and have two children Rahul and Rati. Jaya is not happy in her married life due to the dominance of Mohan in each phase of life.. Even in these seventeen years of married life, they do not grow close to one another to understand each-other. However the long silence grew between them which affect Jaya the most. Jaya is in despair in a male-dominated family. Mohan is a man who plays a dominant and leading role in the family. Jaya is dependent on him as a married lady, wife, and mother. Mohan controls her personal and professional life decisions. Like typical Indian mentality Jaya has to follow everything without sharing her thoughts. At one point in the novel she expressed her thoughts by saying,

> "I remember now that he had assumed I would accompany him,had taken for granted my acquiescence in his plans. So had I. Sita following her husband into exile, Savitri dogging Death to reclaim her husband, Draupadi stocically sharing her husband's travails…" (TLS 11)

Jaya in That Long Silence is educated girl with a sense of being unique and extraordinary. In spite of, education she stuck in the house and feels suffocated and trapped in the traditional Sita-role defined in patriarchal society. After marriage Mohan changed her name from Jaya to Suhasini, which affects her a lot. But she silently accepts it and becomes a so called good Indian wife.

> "Suhasini who was distinct from Jaya, a soft, smiling, placid, motherly woman. A woman who lovingly nurtured her family. A woman who coped. "(TLS 15-16)

However being an educated girl, Jaya follows her husband's command silently without any kind of revolt. Jaya accept this without protest because quite at an early age she was taught that her husband is a tree of protection, a security. Jaya, therefore, shuts her doors of dreams and accept life as it comes to her. She is a good writer. But her freedom of thoughts through her imagination also destroyed by Mohan. She, like a puppet performs a perfect role of Indian wife to meet the every desire of her man. Even in their sexual life Jaya has to accept everything what Mohan wants. Throughout the novel, she constantly compromises her dreams which made her a typical Indian wife. She remembers the story of the crow and sparrow very well and thought that no girl can forget the end of the story. It ironically reflects the cage life of Indian woman.

> "they will become that damnably, insufferably priggish sparrow looking after their homes, their babies…and to hell with the rest of the world. Stay at home, look after your babies, keep out the rest of the world, and you are safe."(TLS 17)

The poor Suhasini believes in this story, but Jaya would not think to be like it. Deshpande tries to focus not only on the patriarchal set up which is responsible for silencing the women but also the responsibility of women lies within the victim to refuse, to raise a voice and to break that silence. The novel traces the dilemma and the quest for identity of Jaya from the feelings of existence, freedom, resilience and adjustments. In the quest for identity, Jaya is trapped in the dilemma, firstly trying to be a suitable wife for Mohan and secondly, struggling to express the emotions of women's experience in the male-dominated society. She thought that woman's life becomes a waiting game to express her desires and willingness. Since she got married she had done nothing for herself except to wait to and fulfill needs and wishes of family. She expresses her inner thought that this waiting game begins at the very point when a girl child born.

> "Wait until you got married. Wait until your husband comes. Wait until you go to your in-law's home. Wait until you have kids."(TLS 30)

She is a silent sufferer in finding out herself very different with noble vision as a writer. She represents the middle class educated woman in India during 1990s who tries to find her identity throughout the novel. She searches her identity as an individual and where her emotions are getting subdued. It suggests that the endemic imbalance in a marriage causes the frustrations, disappointments, failures rather than the endurance and solace.

The novel is also set against the theme of identity crisis that Jaya is searching her own identity and is set ideally against the Indian backdrop. The novel raises eternal question whether a woman lives for her husband or children or for someone else. The protagonist raises her voice against the straight-jacketed role models of daughter, sister, wife and mother, and refuses to be the objects of cultural or social oppression of the age old patriarchal society.

To conclude, Shashi Deshpande, an eminent novelist, has emerged as a writer possessing deep insight into the female psyche. Focusing on the marital relation, she seeks to expose the tradition by which a woman is trained to play her subservient role in the family. Her novels reveal the man-made patriarchal traditions and uneasiness of the modern Indian woman in being a part of them. Shashi Deshpande uses this point of view of the present social reality as is experienced by women. To present the world of mothers, daughters and wives is also to present indirectly the fathers, sons and husbands, the relation between men and woman, and between women themselves. Her young heroines rebel against the traditional way of life and patriarchal values. The words which we always associate with what we consider to be the concept of an ideal woman are self-denial, sacrifice, patience, devotion and silent suffering. Shashi Deshpande's fiction is an example of the ways in which a girl child's particular.

This paper discusses the dilemmas of a married woman living in anguish and hopelessness. In her loneliness, Jaya goes through a self-evaluation of her life. It's in this fragmented state of the trauma she realizes that her silence cannot solve her marriage. So she decides to break her silence and speak with her husband Mohan with the hope to find a solution and restore their marriage.

Towards the end of the present novel she consciously acknowledges her writing as a kind of fiction and quotes Defoe's description of fiction as a king of lying which may make 'a great hope in the heart'. So she decides to plug that hole 'as said earlier by speaking and listening and erasing the silence between her and Mohan, her erasing of the silence stands for her assertion of her feminine voice, a voice with hope and promise, a voice that articulates her thoughts. The novel does not depict Jaya's life as a totally dismal and hopeless struggle. It suggests "hope" and "change" for the better. She pines for better life where there is no shackle to tie the legs forwarding towards career and success.

> "We don't change overnight. It's possible that we may not change even over long periods of time. But we can always hope without that, life would be impossible."(TLS 193)

REFERENCES

1] Mr.L.Aruldoss, Siles Appollo ,Dr.R.Vijayalakshmi, The Quest For Female identity with reference Shashi Deshpande's 'That Long Silence", International Journal of Pure and Applied Mathematics, 2018, 119 (18),2899-2908

2] Deshpande Shashi. That Long Silence. Penguin Books India,1989.

3] Sharma Siddharth. Shashi Deshpande's Novel:A Feminist Study. Atlantic Publishers and Distributers; New Delhi; 2005.

4] Aashima. A feminist revision of Shashi Deshpande_s_ that long silence and the dark holds no terror. Department of English- Maharaja Ganga Singh University,2009, http://hdl.handle.net/10603/288163

5] Krishna Priya J., The Quest for Identity in Shashi Deshpande's "That Long Silence" (1988), Munich, GRIN Verlag, 2020, https://www.grin.com/document/942013

Nanosuspension: Novel Approach for Drug Delivery

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Abstract: *Nanosuspensions have emerged as a encouraging strategy for the efficient delivery of hydrophobic drugs because of their multipurpose features and unique advantages. Techniques such as media milling and high pressure homogenization have been used commercially for producing nanosuspensions. Recently, the engineering of nanosuspensions employing emulsions and microemulsions as templates has been addressed in the literature. The unique features of nanosuspensions have enabled their use in various dosage forms, including specialized delivery systems such as mucoadhesive hydrogels. Rapid strides have been made in the delivery of nanosuspensions by parenteral, peroral, ocular and pulmonary routes. Currently, efforts are being directed to extending their applications in site-specific drug delivery.*

***Key Words*:** Nanosuspension, Nanosuspension, drug moieties, bioavailability, solubility, dosing frequency, nanosuspension technology.

Received Date: 27/07/2023 Revised Date: 12/09/2023 Acceptance Date: 26/09/2023

1. INTRODUCTION

Modern drug discovery is very successful at choosing promising molecules and rejecting non-promising ones in a very short time,1 but the same discovered molecules may not satisfy both therapeutic and formulation requirements.2 Up to 40% of new chemical entities (NCEs)3 discovered in the pharmaceutical industry today are compounds with low solubility. Conventional formulations of poorly water-soluble compounds are often associated with low and variable bioavailability. The formulation approaches available for drugs with low solubility include aqueous mixtures with an organic solvent4, 5 (e.g., water-ethanol), solubilization,6formation of complexes (e.g. using β-cyclodextrins),7-9 solid dispersions,10-12 and pH control or salt form.13 These formulation approaches have limitations, and their limited success is clearly demonstrated by the relatively low number of products in the market that are based on such technologies. The growing percentage of NCEs displaying solubility issues demands the development of new technologies for enhancing drug dissolution. Some approaches involve either chemical or mechanical modification of the environment surrounding the drug molecule (in a solution) or physically altering the characteristics of coarse drug particles.

The saturation solubility and dissolution rate of a drug substance can be mainly altered on two levels, through material engineering of a drug substance or through formulation approaches.14 Nanotechnology has been applied to develop drug delivery systems like microemulsions, solid lipid nanoparticles, liposomes, and polymeric nanoparticles. But the cost involved in their preparation and their large-scale production feasibility is a primary concern. Thus, it would be much smarter to have a simple, universal formulation approach for such molecules. Micronization has been a universal approach widely used to improve bioavailability of drugs with poor solubility. The significant increase in surface area obtained by particle size reduction greatly improves the dissolution properties of a drug, thereby allowing a wider range of formulation approaches and delivery technologies. The recent advances in particle-size engineering methods have widened the formulation opportunities for relatively water-insoluble drugs. Several reports have shown significant improvement in saturation solubility and dissolution rate when the drug was reduced to nanometer size.15-18 The nanosuspension of drugs has rapidly evolved into a mature drug-delivery strategy, and research interest is increasing in this area. This upward growth can be seen from the number of research articles and patents filed in the recent past. Consequently, drug nanosuspensions have arisen as a nanotechnology-based formulation approach, and they have been given increased attention due to their pharmaceutical advantage and pharmacoeconomic value, i.e., better performance in terms of cost and effect as compared to other approaches. Figure 1 shows the decision tree[19] for various formulation approaches for a drug, highlighting the importance of nanosuspensions, which comprise the only options for drugs with high melting points, high log P, and high dose requirements. However, considering the fact that more than 40–60 % of NCEs are low water solubility, have high log P, and have high melting points, nanosuspension is now preferred as a simple and universal formulation approach for a large number of drugs.16,20 Nanosuspensions involve colloidal dispersion of nanosized drug particles that are produced using an appropriate method and are stabilized by

a suitable stabilizer. 21,22 Nanosuspension technology offers major advantages of general applicability to most drugs and simplicity of method; it could become a universal formulation approach to processing drugs with low solubility. An important advantage of drug nanosuspensions is that they can be applied to various administration routes: oral,23 parenteral,24,25 ocular,26,27 pulmonary,28 dermal. In addition, they have shown great superiority over their traditional formulation counterparts.

Various articles have explained the production and applications of nanosuspensions,29-31 and some have focused on their stability.32, 33 But the complete picture of evolution of this system remains unclear. The aim of this review is to provide an overview of various approaches for preparation, characterization, applications, and advances in the field of nanosuspension technology, with special emphasis on the latest research developments and patents on nanosuspensions. document is template. We ask that authors follow some simple guidelines. In essence, we ask you to make your paper look exactly like this document. The easiest way to do this is simply to download the template and replace(copy-paste) the content with your own material.

Number the reference items consecutively in square brackets (e.g. [1]). However the authors name can be used along with the reference number in the running text. The order of reference in the running text should match with the list of references at the end of the paper.

2. PREPARATION OF NANOSUSPENSIONS

Several techniques are used to produce nanosuspensions. The existing technologies can be divided into "bottom-up" and "top-down" technologies, or a combination of both. A simple schematic representation of the two technologies is given in Fig. 1. The topdown technologies are disintegration methods that basically rely on mechanical attrition to render large crystalline particles into nanoparticles. The bottom-up approaches rely on controlled precipitation and/or crystallization. These processes involve dissolving the drug in a solvent and precipitating it in a controlled manner to nanoparticles through the addition of an antisolvent. Top-down technologies are more widely used than bottom-up technologies because top-down technologies yield better control over the processing parameters. This conclusion has been drawn based on a survey of marketed products based on nanosuspension technology.

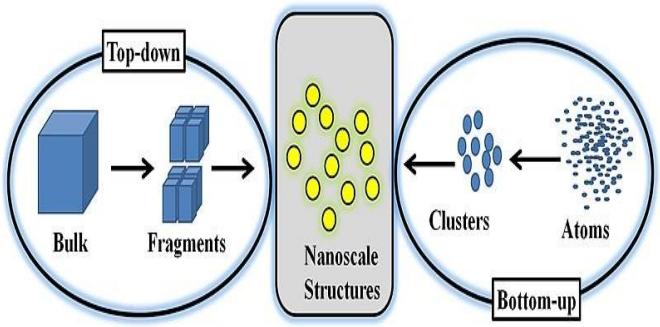
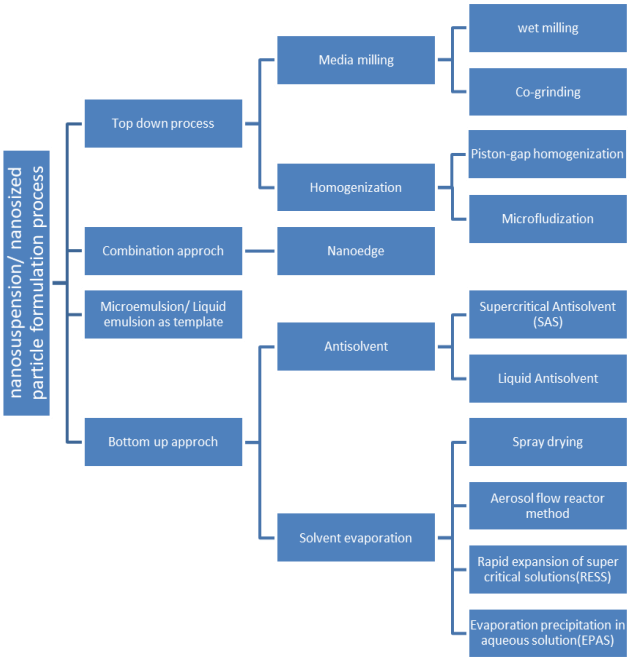


Fig -1: Schematic representation of top-down and bottom-up approaches.

The performance of nanosuspensions mainly depends on the particle size and the polydispersity index (PI) of the system, which is greatly affected by the method of preparation. The size and polydispersity obtained using various methods are not predictable, and obtained particle size and PI depends upon drug and process parameters. The PI is an important parameter governing the physical stability of nanosuspensions; it should be as low as possible for the long-term stability of nanosuspensions. A PI value of 0.1–0.25 indicates a fairly narrow size distribution, whereas a PI value greater than 0.5 indicates a very broad distribution. The acceptable PI values may differ with method of administration. For intravenous administration, a small mean particle size and a narrow particle-size distribution are necessary. Specifically, the number of particles greater than 5 μm should be sufficiently low after production and should remain within a limit during storage to avoid capillary blockade after intravenous injection. Various methods reported for preparation of nanosuspensions are depicted in Fig. 2. Of these, a few methods have already demonstrated a scaling-up possibility a prerequisite for introduction of a product to the market. The recovery and yield of these methods at a small scale has been established by many researchers. Niwa et al. developed simple and easy method of preparing oral nanosuspension of pharmaceutical candidates with low water solubility to support the drug discovery and preclinical studies using animals (for 50 mg to 30 g of drug). The

nanoparticles were successfully recovered with high yield (>95%) using this method.34In this section, various existing nanosuspension methods are briefly described. Technical notes have been provided regarding recent research work and patents employing these techniques.



PRECIPITATION

Bottom-up technology starts at the molecular level and proceeds (via molecular association) to the formation of a solid nanosized particle. In bottom-up techniques, the drug is dissolved in an organic solvent, and this solution is mixed with a miscible antisolvent to initiate fast precipitation of a finely dispersed product. Drug solubility plays an important role in the precipitation technique. Nucleation and growth kinetics dictate the final particle size and size distribution and are controlled via supersaturation, which can be achieved by controlling process parameters and modifying API solubility.35 Reproducibility and control over particle size in precipitation techniques are the major factors considered. The key factors to controlling the size and stability of drug nanoparticles are the choices of solvents and stabilizers and the mixing process. The mixing step is crucial to producing a rapid and uniform supersaturated solution that facilitates the formation of uniform drug nanoparticles. Other crucial factors include drug concentration, volume ratio of antisolvent to solvent, temperature, and viscosity. The precipitation method, a bottom-up method, has been employed successfully for preparing nanosuspensions. 36, 37 Examples of the precipitation technique include hydrosols, Nanomorph, supercritical fluid (SCF) technology, and other precipitation approaches. The precipitation method has the advantages of technical simplicity, simple equipment requirements, and easy scalability. SCF methods require special equipment, but the particle size control is better with these methods. The disadvantages of the precipitation method include the requirement that a drug be soluble in at least one solvent, the miscibility of that solvent with nonsolvent, solvent residues, and the preservation of the nanoparticle structure.

HYDROSOLS

The hydrosols were developed by Sucker et al. of the Sandoz Company (now Novartis, Inc.).[38] In this approach, the drug is dissolved in a solvent, and this solvent is added to a nonsolvent to initiate rapid precipitation of a finely dispersed product. To achieve this, it is necessary to pass the so-called Ostwald–Mier area very quickly, which means reducing the solvent quantity very quickly.39 This is achieved by adding the solvent to a nonsolvent (i.e., doing it the other way around would lead to the formation of larger crystals). The formed nanocrystals need to be stabilized by surfactants or polymers to avoid growth of the nanocrystals to microcrystals. In general, it is recommended that the product be lyophilized to preserve the nanosize of the particles. This technology is, to some extent, complex (e.g., the preservation of particle size), and it excludes all molecules that are poorly soluble in aqueous and organic media. But unfortunately, no products based on this technology have been marketed. An additional problem is solvent residue; its removal makes the process more costly.

NANOMORPH

The precipitation method developed by Sucker generally yields crystalline particles, whereas another precipitation technique developed by Knoll (now owned by Abbott, Inc.) reportedly creates amorphous particles.40 The product is called Nanomorph. The special feature of Nanomorph is an increase in the dissolution velocity due to the amorphous character of the product. The precipitation in the amorphous form is achieved by an aqueous polymer solution. However, for commercialization of this technique, the major challenge is to preserve the amorphous character during the shelf life, as any polymorphic change will result in changes in bioavailability. 3. Supercritical Fluid Technology Nanosized drug particles can be produced with supercritical fluids using various methods, such as the rapid expansion of supercritical solution process (RESS), the gas antisolvent (GAS) process, and the supercritical antisolvent process (SAS). The RESS process involves expanding the solution of the drug in a supercritical fluid through a nozzle. Upon expansion, supercritical fluid loses its solvent power, leading to precipitation of the dissolved drug as fine particles. Cyclosporine nanoparticles in the size range of 400 to 700 nm have been produced using this technique.41 The GAS process involves pressurizing a solution of drug in a common solvent with CO2. As the solvent is removed and the solution gets supersaturated, the drug precipitates and forms fine crystals. The SAS process uses a supercritical fluid (in which the drug is poorly soluble) and a solvent for the drug (which is also miscible with the supercritical fluid). The method involves injecting a solution of the drug into the supercritical fluid. As the drug solution becomes supersaturated, the drug precipitates as fine crystals. Chattopadhyay et al. have applied this method for preparing nanosuspension of griseofulvin, an antifungal agent with poor aqueous solubility. The particle size and morphology of the nanoparticles were further controlled by subjecting the drug solution to an ultrasound field generated by a vibrating surface inside the supercritical media. The frequency of the vibration was varied to obtain

particles with different sizes and morphologies.42 The SAS method can lead to particle nucleation overgrowth due to transient high supersaturation. With these methods, high supersaturating may also result in the development of an amorphous form or other undesired polymorphs. This is particularly true in the case of organic molecular crystals, in which the forces holding the molecules together in the lattice are relatively weak.

Other Precipitation Approaches

Recently, some new precipitation methods have been reported for preparing nanosized particles. These comprise.

- High gravity controlled precipitation (HGCP),
- Sonoprecipitation,
- The aerosol flow reactor method,
- Evaporative precipitation in aqueous solutions (EPAS), and
- Spray drying.

One of the most promising nanoprecipitation techniques available at the commercial production scale is HGCP. This technique comprises a rotating packed bed to strengthen mass and heat transfer by several orders in a multi phased system. The technique has subsequently been applied to the production of nanoparticulate drugs like cefuroxime axetil, azithromycin, danazol, cephadrine, and salbutamol sulphate.[43] Recently, sonoprecipitation (or sonocrystallization), a crystallization process which is mainly assisted by ultrasound, has been developed. The principle involves development of bubbles (cavitation) followed by collapse, which releases shock waves, thereby stimulating nucleation powered by changes in temperature and pressure. This technique has been successfully used for preparing nanosized particles of cefuroxime axetil.44 The aerosol flow reactor method is a single-step continuous process like spray drying that involves atomizing the drug solution into a carrier gas for drying. The method has the potential to control the particle morphology and polymorphic form because this method provides more precise control over the temperature history and residence time of droplets. This method has been used to produce spherical nanoparticles of beclomethasone dipropionate.45 Another precipitation method reported for nanoparticle preparation is EPAS. In this method, an organic solution of drug is preheated through a coil and injected under the surface of a heated aqueous solution with surfactant(s) added to stabilize the particles. Intensive atomization occurs below the liquid surface, which produces a large interface between the organic and aqueous solutions, causing rapid evaporation of the organic solvent and precipitation of particles. Cyclosporine A nanoparticles have been produced using this technique.46 In spray drying, a drug solution (aqueous or organic) is atomized to fine droplets, which are evaporated in a warm air current to form dry particles. This method is not suitable for production of nanoparticles because spray drying has low cyclone collection efficiency for nanoparticles. With an electrostatic collector, it is now possible to collect spray-dried nanoparticles. The applicability of this approach has been demonstrated using bovine serum albumin solution.47

HIGH-PRESSURE HOMOGENIZATION

High-pressure homogenization (HPH) is a disintegration method based on the top-down approach. Two homogenization principles are applied: piston gap fluidization and microfluidization. In the first method, the suspension of a drug and surfactant is forced under pressure through a nanosized aperture valve of a high-pressure homogenizer, and particle-size reduction is based on the cavitation principle. The particle cavitation forces are sufficiently high to convert the drug microparticles into nanoparticles. Particles are also reduced due to high shear forces and the collision of the particles against each other. The major concerns associated with this method are the need for drug particles to be in micronized state before loading and the number of homogenization cycles required.48 The size of the drug nanocrystals that can be achieved depends mainly on the power density of the homogenizer, the number of homogenization cycles, and temperature. Drug factors also have an important effect on the HPH process. An important determining factor for the final size of the drug nanocrystals is the hardness of the drugs. For soft drugs such as paclitaxel, it is possible to obtain a particle size as low as 250 nm; but it is very difficult to achieve a particle size of 250 nm for harder drugs, such as azodicarbonamide, irrespective of homogenization cycles and pressure. During the size reduction process, the particles and/or crystals break preferentially at weak points (i.e., imperfections). The number of imperfections decreases with decreasing particle size; thus the remaining crystals become more and more perfect. Therefore, the force required to break the crystals increases with decreasing particle size. The particles will not further diminute, even when additional homogenization cycles are applied, if the force (i.e., power density) in the homogenizer is equal to the interaction forces in the crystal. The second method is microfluidization, which is based on a jet stream principle. Here, the suspension is accelerated and passes with a high velocity through a specially designed homogenization chamber (either Z shaped or Y shaped), leading to a reduction in particle size due to the collision of particles and the shear forces generated. Techniques based on the HPH processes include hydrosol, Nanomorph, nanocrystals, dissocubes, Nanopure and Nanoedge. The HPH method has been used successfully by various researchers to achieve nanosuspensions.49-51

THE LIPID EMULSION/ MICROEMULSION TEMPLATE

Lipid emulsion and/or microemulsions are also used as templates for the preparation of nanosuspensions. These systems are applicable for drugs that are soluble in either volatile organic solvents or that are partially water-miscible solvents. In this technique, an organic solvent or mixture of solvents loaded with the drug is added slowly to an aqueous

medium with stirring at a high speed that leads to formation of small droplets (containing drug dissolved in organic solvent) emulsified in the aqueous vehicle. As the stirring progresses at high speed, the droplet size is further reduced. The process is also accompanied by slow evaporation of the organic solvent from the droplets. Once the organic solvent is evaporated completely, pure drug particles stabilized by surfactant are left behind, suspended in the aqueous vehicle. The advantages of lipid emulsions/microemulsions as templates for nanosuspension formation are that they are easy to produce by controlling the emulsion droplet and that this procedure is easy to scale up. However, the use of organic solvents affects the environment, and large amounts of surfactant or stabilizer are required. This approach was investigated for improving dissolution properties of drugs with low water solubility.52

MILLING TECHNIQUES

Milling techniques are now being widely used for preparation of nanosuspensions. Nanosuspensions obtained using milling techniques are prepared either by wet milling or dry co-grinding.

WET MILLING

Nanocrystal is a patent-protected technology developed by Liversidge et al.[53] This technique involves mechanical attrition of suspended drug particles using suitable milling media like glass, (yttrium-stabilized) zirconium oxide, or highly crosslinked polystyrene resins. The milling chamber is charged with drug, surfactant and milling media, and a milling medium (usually water), and the contents are subjected to a very high shear rate. The high energy and shear forces generated as a result of impaction of the milling media with the drug provide the necessary energy input to disintegrate the microparticulate drug into nanosized particles. A majority of nanosized products in the market are based on this technique. The advantages of this method include easy scale up, the possibility of handling a large quantity of drugs, and little batch-to-batch variation. The limitations include lengthy process, possible contamination with milling media, and chances of formation of unstable drug particles due to prolonged milling. This method has been used for formulating nanosuspensions of glyburide, itraconazole, fluticasone, etc.54-56

DRY CO-GRINDING

Preparation of nanosuspensions by dry-milling techniques has been reported.[57] It involves dry grinding of a drug with additives (i.e., co-grinding) and is a simple, organic, solventfree preparation process. Formation of drug nanoparticles occurs when the ground mixtures of drug with polymer and/or surfactant are dispersed in water. Water-soluble polymers and surfactants have been used as additives for effective size reduction of drug particles as well as to inhibit particle agglomeration and to improve their dissolution. Improvement in physicochemical properties and dissolution of poorly water-soluble drugs due to cogrinding can be attributed to an improvement in the surface polarity and transformation from a crystalline to an amorphous form.3 Dry co-grinding can be performed easily and economically without organic solvents and can reduce particles to the submicron level, often yielding a stable amorphous solid. This method is promising for preparation of nanosuspensions of dihydroartemisinin, albendazole, danazol, and felodipine.58, 59

COMBINATION TECHNOLOGIES

The previous nanosuspension methods are combined in some cases to gain better size reduction and improved stability of the system. The combination of these methods has resulted in improved advantages associated with nanosuspension technology. Nanoedge is a patented technology based on the combination of precipitation and homogenization.

NANOEDGE

The basic principles of Nanoedge are the same as those of precipitation and homogenization nanoprocesses. A combination of these techniques results in smaller particle size and better stability in a shorter time. The major drawbacks of the precipitation technique, crystal growth and long-term stability, can be resolved using Nanoedge technology. In this technique, the precipitated suspension is further homogenized, leading to a reduction in particle size and avoiding crystal growth. Precipitation is performed in water using water-miscible solvents such as methanol, ethanol, and isopropanol. It is desirable to remove those solvents completely, although they can be tolerated to a certain extent in formulation. For effective production of nanosuspensions using Nanoedge technology, an evaporation step can be included to provide a solvent-free, modified starting material followed by HPH.60

3. EVALUATION OF NANOSUSPENSIONS

The unique qualities and performance of nanoparticulate systems as devices for drug delivery arises directly from their physicochemical properties. Hence, determining such characteristics is essential for achieving the mechanistic understanding of their behavior; this understanding allows prediction of in vivo performance as well as allowing particle designing, formulation development, and process troubleshooting to be conducted in a rational fashion. Nanosuspensions are generally characterized according to the following parameters: particle size, surface charge (i.e., zeta potential), crystalline state, saturation solubility, and stability.

PARTICLE SIZE

The most basic and important property of any nanoparticulate system is particle size. The saturation

solubility, dissolution velocity, physical stability, and even biological performance of these systems depend on particle size. Saturation solubility and dissolution velocity vary considerably with change in particle size of the drug.61 The most frequently used techniques for size measurement of nanosized particles are dynamic light-scattering techniques, static light-scattering techniques, and microscopy. Each method has advantages as well as disadvantages. The mean size and width of distribution (polydispersity index, PI) is typically determined by photon correlation spectroscopy (PCS). This technique can be used for rapid and accurate determination of the mean particle diameter in nanosuspensions.62 PCS records the variation in the intensity of scattered light on the microsecond time scale.63, 64 The measuring range of PCS is limited to approximately 3 nm to 3 mm. Therefore, laser diffractometry (LD) is also used to detect any particles in the micrometer range or aggregates of drug nanoparticles. For nanosuspension intended for intravenous use, particle-size determination by Coulter Counter is also essential, as even a few particles with particle size greater than 5 μm may cause blockage of blood vessels. Depending on the type of equipment employed, the measured size range of particles is approximately 0.01–80 μm. The instrument used and the material to be analyzed are important parameters that affect accurate particle size measurement. The stability of the sample during analysis is the most important requisite for correct and reproducible results.65 Thus, all of these factors must be considered when selecting the appropriate technique for particle-size determination for a particular sample. A few examples that illustrate the importance of particle size on in vivo performance follow. Nowacek et al. demonstrated that physical characteristics such as particle size, surfactant coating, surface charge, and most importantly, shape are predictors of cell uptake and antiretroviral efficacy.66 In vivo studies conducted by Ghosh et al. showed superior systemic exposure of drug in case of nanosuspension compared to non-micronized coarse suspension in dogs.67 Detroja et al. showed enhanced antihypertensive activity of candesartan cilexetil compared to the plain drug in rats.68

SURFACE CHARGE (ZETA POTENTIAL)

Particle charge is a stability-determining parameter in aqueous nanosuspensions. It is measured by electrophoresis and is typically expressed as phoretic mobility [(mm/S)/(V/cm)] or zeta potential (mV). Zeta potential is used as a surrogate for surface charge and is often measured by observing the oscillations in signal that result from light scattered by particles located in an electric field.69,70 A number of instrumental configurations with different approaches have been implemented in different equipment; the most commonly used is the Doppler shift. The zeta potential of a nanosuspension is governed by both the surfactant and the drug itself. For a physically stable nanosuspension solely stabilized by electrostatic repulsion, a minimum zeta potential of ±30 mV is required. In combined electrostatic and steric stabilization, ±20 mV is sufficient as a rough guideline.71 Cerdeira et al. stabilized a miconazole nanosuspension by combining electrostatic and steric stabilization (-19±1 mV) for 6 months.72 Zhang et al. showed that all-trans retinoic acid (ATRA) nanosuspensions with zeta potential of -37.9 ± 2.0 mV had sufficient electrostatic stabilization for 6-month stability. El-Shabouri studied the effect of surface charge on bioavailability of cyclosporine-A nanoparticles. The relative bioavailability of positively charged nanoparticles increased, while it decreased for negatively charged nanoparticles.73 Crystalline State Drug particles of amorphous form are likely to be generated when nanosuspensions are prepared. Hence, it is essential to investigate the extent of amorphous drug particles generated during nanosuspension production. The crystalline status of the nanosuspension can be assessed using differential scanning calorimetry (DSC).74 This is particularly important when a drug exhibits polymorphic forms, some of which may be toxic. The changes in the physical state of the drug particles as well as the extent of amorphous fraction can be determined by X-ray diffraction analysis 75, 76 and can be supplemented by DSC studies.77 The assessment of the crystalline state and particle morphology together furthers understanding of the polymorphic and morphological changes that a drug undergoes when subjected to nanosizing. Yang et al. studied the effect of supersaturation on bioavailability of inhaled, nebulized aerosols for amorphous versus crystalline nanoparticulate dispersions. Pulmonary delivery of the nanoparticulate amorphous ITZ composition resulted in significantly higher systemic bioavailability than for the nanocrystalline ITZ composition, as a result of the higher supersaturation that increased the permeation.78 Lai showed that the drug dissolution rate in nanosuspensions is strongly affected by drug solubility and depends on the crystal form.79 The results of a study by Sigfridsson et al. showed that AZ68 was absorbed at a lower rate for crystalline nanosuspensions compared to amorphous nanosuspensions.[23]

SATURATION SOLUBILITY

The increase in saturation solubility and consequently an increase in dissolution rate of the compound decides its applications. Although saturation solubility is defined as a Compound-specific, temperature-dependent constant, it also depends on particle size. The increase in saturation solubility can be explained by the Kelvin–Gibbs equation (Eq. 1) and the Ostwald–Freundlich equation (Eq. 2). In the Kelvin–Gibbs equation, the vapor pressure increases with increasing curvature of the droplet of a liquid in gas. If this is extended to a solid, it implies that the dissolution pressure increases with decrease in particle size. According to the Ostwald-Freundlich equation, the increased saturation solubility is due to creation of high-energy surfaces when the more or less ideal drug microcrystals are disrupted to nanoparticle.[61]

$$In\frac{Pr}{P\infty} = \left(\frac{2yM}{rRT\rho}\right) \quad (1)$$

$$S = S\infty \left(\frac{2yM}{rRT\rho}\right) \quad (2)$$

In these equations, Pr is the dissolution pressure of a particle with radius r; P∞ is the

dissolution pressure of infinitely large particle; S is saturation solubility of the nanosized drug; S∞ is saturation solubility of an infinitely large drug crystal; γ is the crystal medium interfacial tension; M is the compound molecular weight; r is the particle radius; ρ is the density; R is a gas constant; and T is the temperature. Particle-size reduction of drug particles leads to an increase in surface area, resulting in an increased dissolution rate, according to the Noyes–Whitney equation:

$$\frac{dX}{dT} = \frac{DS}{h}\left(Cs - \frac{Xd}{v}\right) \quad (3)$$

where dX/dt is dissolution rate, Xd is amount dissolved, D is diffusion coefficient, S is particle surface area, v is volume of fluid available for The equation shows that the dissolution rate of a drug is proportional to the surface area available for dissolution. This principle has been extensively used in the micronization of drug for improving oral bioavailability. Obviously, decrease in particle size to nanometer range will further increase the dissolution rate due to a significant increase in effective particle surface area. According to the Prandtl equation (Eq. 4), Nanosizingresults in the decrease of the diffusion-layer thickness surrounding the particles and an increased concentration gradient between the surface of the particle and bulk solution, which facilitates particle dissolution by increasing dissolution velocity.

$$hH = k.\left(\frac{L\frac{1}{2}}{V\frac{1}{2}}\right) \quad (4)$$

In this equation, hH is the hydrodynamic boundary layer thickness; k is a constant; V is the relative velocity of the flowing liquid against a flat surface; and L is the length of the surface in the direction of flow. It is clear from Eqs. (3) and (4) that nanosizing is a suitable approach for increasing bioavailability of poorly soluble drugs where dissolution is the rate limiting step in systemic absorption.80 The theoretical backgrounds of the Kelvin–Gibbs, Ostwald–Freundlich, and Prandtl equations support the fact that below a size of approximately 1–2 μm, the saturation solubility is a function of the particle size. In view of this information, determination of saturation solubility remains an important investigation parameter for nanosuspension because it determines performance. The saturation solubility of the drug in different physiological buffers as well as at different temperatures should be assessed using different methods described in literature. For example, saturation solubility can be determined at different temperatures by shaking experiments until equilibrium has been reached. The improvement in dissolution rate of nanosuspension compared to conventional formulations reflects the advantages achieved by nanosizing. Apart from adhesiveness, increased dissolution velocity and increased saturation solubility are the special benefits of nanosuspensions. These two parameters mainly determine the in vivo fate of nanosuspensions. Showing a saturation solubility that was five times greater, improved bioavailability of oral nanocrystals of anthelmintic drug albendazole was observed compared to the raw material.[81] After nanosizing, increase in saturation solubility of nifedipine was observed, resulting in improved dissolution characteristics that resulted in improved bioavailability.[82]

SURFACE MORPHOLOGY

Nanoparticles can be directly observed by scanning electron microscopy (SEM) and transmission electron microscopy (TEM), with the former method being better for morphological examinations.83, 84 TEM has a smaller size limit of detection and provides structural information via electron diffraction, but staining is usually required. Researchers must be cognizant of the statistically small sample size and the effect of applied vacuum on the particles during analysis. Very detailed images can be obtained using the freeze–fracture approach in which a cast is made of the original sample.85 Sample corruption resulting from the extensive sample preparation is always a possibility, though lower vacuum instrumentation reduces this manipulation, albeit at the loss of some resolution.[86] Atomic force microscopy (AFM) can also be used to confirm the size and shape of nanosized particles. AFM is capable of scanning the surfaces in controlled environmental conditions and is complementary to SEM imaging.

STABILITY

Physical stability is crucial in the formulation of drug nanosuspension. Because nanosuspensions have a mean particle diameter in nanometer range, they are prone to aggregation. The aggregation may be due to Ostwald ripening, which occurs due to different saturation solubilities in the vicinity of very small and larger particles. Nanosizing results in the creation of additional surface area and/or interfaces that lead to a change in free energy and become thermodynamically unstable while tending to minimize the free energy.[3] Hence, stabilizers like surfactants or polymeric macromolecules are required to stabilize the nanoparticles against interparticulate forces and to prevent them from aggregating. Surfactants are used to minimize the free energy and stabilize the system. The stabilization provided by surfactants is by steric or electrostatic processes or a combination of them. Steric stabilization is achieved by adsorbing surfactants/polymers onto the particle surface, while electrostatic stabilization is obtained by adsorbing charged molecules, which can be ionic surfactants or charged polymers, onto the particle surface. Generally, steric stabilization alone is sufficient to provide stability to the

nanosized particles, but it is often combined with electrostatic stabilization as an additional measure. Formulation of nanosuspension requires careful selection of stabilizers. The ability of a surfactant to associate with a solid surface is dependent on several factors, including pH, ionic strength, temperature, structure, charge density, and other properties of both the surface and the surfactant.[87, 88] Therefore, selection of appropriate stabilizer for nanosuspension is a challenging task. Also the FDA approval status (GRAS) of the stabilizers must be considered for selection. The most popular surfactants used for stabilization purposes are Polysorbate 80, lecithins, cholic acid derivatives, and sodium lauryl sulfate. The surfactant stabilizers can be non-ionic (polysorbates) or anionic (sodium lauryl sulfate (SLS) and docusate sodium (DOSS). Due to their antiseptic properties, cationic surfactants are typically not used as stabilizers for oral formulation. The polymeric stabilizers most frequently applied are povidones (PVP K 30), poloxamers (F68 and F127), and cellulosics (HPC and HPMC). The molecular weights of these polymers are usually between 50 kDa and 100 kDa, and their chains should be long enough to provide a steric layer but not too large to slow down dissolution. The most popular non-ionic surfactants applied are the poloxamers and Tween 80; while sodium lauryl sulfate is the typical ionic surfactant used for this purpose. To achieve the most stable nanosuspension formulation, the stabilizers can be used alone or in combination. The drug:stabilizer ratios (w/w) in the formulations vary widely, ranging from 1:3 to 50:1. Other stabilizers that were studied for stabilization of drug nanoparticles include cyclodextrins89 and amphiphilic amino acid copolymers.88

Although the number of articles describing nanosuspension formulations is extensive; attempts to evaluate and compare the potential of different stabilizers are limited. A few studies that evaluated the potential of different stabilizers for providing stability to nanosuspensions are reported here. Verma et al. investigated the role of stabilizers (small molecules vs. polymeric) on Ostwald ripening for evaluating physical stability of indomethacin nanosuspension. They observed a lower rate of particle-size increase in small-molecule surfactant at higher concentrations compared to polymeric surfactant.90 In another study, the ability of povidone (PVP) and hydroxypropyl cellulose (HPC) to obtain nanosuspensions for seven model compounds by wet comminution was evaluated. Results showed that better nanosuspensions were produced when surface energy values of drug and stabilizer were comparable.91 Lee et al. reported the screening of 5 polymers (i.e., HPC, PVP, Poloxamer 407, polyethylene glycol (PEG) and Poloxamer 188) and 11 model drugs for nanosuspension stabilization. They found that poloxamer 188 was able to stabilize most of the model compounds. They also reported that drugs with lower aqueous solubility, higher molecular weight, and higher melting points were better candidates for nanosuspension production (i.e., easier to stabilize).92 Van Eerdenbrugh et al. evaluated nanosuspension production with 13 stabilizers of different classes; each was used in three concentrations for nine structurally different drug compounds. The performance of the surfactant was ranked in the following order: semi-synthetic polymers < linear synthetic < synthetic copolymer. Results showed that the hydrophobicity of the surfaces was decisive for the agglomeration tendency of the particles and hence the ease of nanosuspension stabilization.[93] In another study, the practically water insoluble drug miconazole was nanoground, and the stabilizing effects of a variety of surface active and polymeric excipients were tested. Hydroxypropylcellulose (HPC-LF) in combination with sodium dodecyl sulfate (SDS) were found to be the best stabilizers for the miconazole nanosuspensions. The study showed that excellent wetting of drug particles as well as their electrostatic and steric stabilization by excipients was necessary to produce stable nanosuspensions by nanogrinding.94 From all of these studies, it can concluded that selection of surfactant is a challenging task important to providing stability to nanosuspensions.

In addition to physical stability (Ostwald ripening), chemical stability of the active content in nanosuspension is affected in some cases by hydrolysis of the compound. The active content of the nanosuspension formulation must be studied, as some drugs have low stability in aqueous media. However, some examples have shown that formulation of nanosuspension prevents hydrolysis of a particular drug compared to solution.[95] Thus, drug content of the formulation must be studied immediately after preparation to verify the chemical stability of the drug. Formation of impurities due to process and formulation parameters must be studied. The impurities can be identified using various techniques such as infrared spectroscopy (IR), high performance liquid chromatography (HPLC), and mass spectroscopy (MS). In addition, impurities related to the process must also be tested, e.g., for the possibility of zirconium content in the formulation if the media-milling method using zirconium oxide beads was used for preparation. The techniques used for characterization of nanoparticulate systems are summarized in Table 1.

In addition to characterization of the properties mentioned, additional characterization of the nanosuspension is required if surface modification of particles is performed. The parameters for which surface-modified nanosuspensions are evaluated include adhesion properties, surface hydrophilicity/hydrophobicity, and interaction with body proteins. The adhesiveness of the drug nanoparticles is considered to be a major factor that contributes to increased bioavailability and reduced variability of absorption. Surface hydrophobicity determines the interaction with the cells prior to phagocytosis and is a relevant parameter for adsorption of plasma proteins. This is considered to be an important parameter affecting in vivo organ distribution after intravenous injection. Separation by hydrophobic interaction chromatography (HIC) depends on the reversible adsorption of biomolecules according to their hydrophobicity, and HIC is widely used for the separation and purification of proteins in their native states.96 The HIC technique is used for determining surface hydrophilicity/hydrophobicity;

hydrophilic particles pass through the column faster, and elution of hydrophobic particles is retarded.97

4. CONVERSION OF NANOSUSPENSION TO SOLID STATE

Nano sizing is now a well-established technique for enhancing the dissolution rate of drugs with low water solubility by increasing the surface area of particles. Despite their advantages, the stability of nanosuspension is a critical aspect that defines the safety and efficacy of the drug product. Stability issues related to nanosuspension include sedimentation or creaming, agglomeration, and crystal growth. The transformation of a nanosuspension into a solid product is often required for physical stability and/or patient convenience.[30] Particle growth may occur in a nanosuspension during manufacturing, storage, or shipping. To maintain particle size, surfactants are often used but they might not be able to maintain the particle size for the required period. Therefore, conversion of a nanosuspension to a solid form becomes essential if stable nanosuspension is unattainable. Solid dosage forms are convenient with regard to marketing. Thus, there is a current need to develop an efficient technique for converting nanosuspensions to solid forms while maintaining their performance characteristics. Various methods are available for this purpose. The acceptability of the method generally depends on its effectiveness in preserving particle size after processing. Methods that are commonly used for conversion of nanosuspension to dry state include spray drying, freeze drying (lyophilization), pelletization, and granulation.[95] The most common processes are freeze-drying and spray-drying.[22] Spray drying is widely used due to its simplicity and cost-effectiveness; it is generally preferred over lyophilization by the pharmaceutical industry to transform liquid nanosuspensions to dry products because it is faster and consumes less energy. In spray drying, nanoparticulate dispersion is atomized to fine droplets that evaporate in a warm air current to form dry particles. The driving force for drying is controlled by the liquid content and the difference between the inlet and outlet temperatures of the drying air. Chemical degradation of the drug due to heating is the foremost concern in spray drying. Nanosuspension of drugs like celecoxib and itraconazole successfully convert into nanocrystals by spray drying.98, 99 Freeze drying is another convenient method of converting nanosuspension to a solid powder.100 Freeze drying involves the nucleation and propagation of ice crystals (freezing) and a following sublimation process. When nanoparticulate dispersion is freeze dried, a temperature gradient inevitably develops, and nucleation begins in the area at the lowest temperature. After nucleation, the temperature gradient leads more liquid water molecules to rearrange themselves into the open structure of a solid lattice, resulting in the propagation of the freezing interface. While fast freezing results in polycrystalline structures that sometimes have defects, slow freezing allows water molecules to exclude foreign particles and eventually causes them to aggregate.101 The freezing step is more important than the subsequent sublimation step, as the steric stabilization of polymers becomes inactive when nanocrystal dispersions are freeze dried. In the freeze-drying process, a cryoprotectant can be used to prevent aggregation. Lee and Cheng found that freezing rate is a critical factor in freeze drying and that it depends on the API concentration of the dispersion.102 For a conversion of nanosuspension of drugs like 2-methoxyestradiol, naproxen, and loviride in its dry form Freeze drying is successfully used.103-105

Table - 1: Methods for Assessing Properties of Nanoparticulate System

Property Relevant Analytical Technique	
Property	**Relevant Analytical Technique**
Particle size Dark field optical microscopy, dynamic light scattering, static light	Dark field optical microscopy, dynamic light scattering, static light, ultrasonic spectroscopy, turbidimetry, NMR, single particle optical sensing
Morphology	TEM, SEM, Atomic force microscopy
Surface charge	Electrophoretic light scattering, U-tube electrophoresis
Surface hydrophobicity	Hydrophobic interaction chromatography
Surface adsorbates	Electrophoresis
Density	Isopycnic centrifugation, sedimentation-FFF
Interior structure	Freeze fracture SEM, DSC, X-ray diffraction, NMR

NMR, nuclear magnetic resonance spectroscopy; TEM, transmission electron microscopy; SEM, scanning electron microscopy; FFF, fast-freezing fixation; DSC, differential scanning calorimetry.

To improve the patient's convenience, the powder can be filled in capsules or converted to tablets. A few reports are available on the conversion of nanosuspension into tablets.[97, 98, 106, 107] These studies have shown that the tablet formulations containing nanosized drug particles dissolve faster than the unmilled drugs. This suggests that the conversion of a nanosuspension does not affect the properties of the drug. Although conversion to a dry state may provide the advantages of stability and/or patient convenience, it should not adversely affect the dissolution properties of the nanosuspension. Generally, various matrix forms are used in the process of converting a nanosuspension to a solid state. The selection of these matrix formers determines the utility

of this approach. Thus, due consideration must be given to the excipients and processes which are used to enhance the stability of the nanosuspension formulation.

5. APPLICATIONS OF NANOSUSPENSIONS

Nanosuspensions are used to advantage in diverse dosages forms. Their small size and increased surface area leads to increased dissolution rates and increased bioavailability. In contrast, the particulate nature of nanosuspension can result in targeting of monocyte phagocytic systems (MPSs), with unusual pharmacokinetic significances. Nanosuspensions can play a critical role as an enabling technology for molecules of low water solubility or permeability having significant activity as observed in in vitro studies. These molecules may pose problems at one or both of the following stages during new drug development processes:

Formulation of an intravenously injectable product for preclinical in vivo evaluation to measure its toxicity and other pharmacokinetic characteristics.

Improving absorption of the drug candidate from the gastrointestinal tract (GIT) which showed poor bioavailability during preclinical as well as clinical development studies.

As the particle size of nanosuspension is the range of 1–1000 nm, these formulations are suitable for application through various routes of administration like parenteral, oral, topical, pulmonary, and other targeted drug-delivery systems.

ORAL

A route most which is most widely favored for drug administration is Oral drug delivery. But some drugs show very limited bioavailability due to their poor solubility and absorption, and which eventually lowers their efficacy. For such cases, nanosuspension is the most suitable option as it helps to improve the dissolution rate and absorption due to enhanced surface area and improved adhesiveness. Nanosuspension can make possible increased mucoadhesion, which may improve gastrointestinal transit time and cause increased bioavailability. When administration with the conversion of atovaquone micronized drug in atovaquone as a nanosuspension resulted in a 2.5-fold increase in oral bioavailability. The improvement in oral bioavailability can be accredited to the adhesiveness of the drug nanosuspension, increased surface area and saturation solubility.[108] Taste masking of a particulate system in oral delivery is also easy. Administration of the nanosuspension has been reported to enhance oral bioavailability of BMS-488043 in dogs compared to the conventional formulation containing the micronized crystalline drug substance.[109] Nanosuspension formulation also helps to avoid the effect of food on absorption, as observed for Emend (aprepitant) formulation in another study.[110]

PARENTERAL

A nanosuspension is an approach by which one can convert poorly soluble non-injectable drugs into a formulation suitable for IV administration. Although the production of nanosuspension for parenteral use is very precarious, recent developments in nanosuspension technology have proven its usefulness for injectable formulations. Nowadays techniques for preparation of nanosuspensions are now accurately controlled and have a capacity to develop uniform particles with greater control over maximum particle size. Injectable formulation of nimodipine nanosuspension proved better than commercial product (ethanol based) in terms of local irritation and phlebitis risks.111 Lou et al. demonstrated that when formulated as a nanosuspension, oridonin showed stronger antitumor activity in mice compared to the free oridonin solution.112 Zakir et al. demonstrated that the pharmacokinetic profiles of Amp B, when given in the nanosuspension formulation, were different compared to the corresponding raw drug.[113]

OCULAR DELIVERY

Nanosuspension approach is a beneficial for drugs which shows very low solubility in lachrymal fluids. Nanosuspension is an ideal approach for ocular delivery of hydrophobic drugs due to their characteristic ability to improve saturation solubility of drugs. Kassem et al. developed a nanosuspension delivery system for certain glucocorticoid drugs.28 Gupta et al. designed a study to improve the bioavailability of forskolin via the influence of precorneal residence time and dissolution characteristics. They proved that the pH and thermoreversible polymeric in situ gel-forming nanosuspension with controlled drug release exhibited a greater potential for glaucoma therapy than the original formulation of the drug.[114] Ali et al. assessed ocular bioavailability of hydrocortisone nanosuspensions in albino rabbits using hydrocortisone solution as a control. Significantly higher bioavailability of hydrocortisone nanosuspensions was observed compared to the hydrocortisone solution. A sustained drug action was maintained for up to 9 h with the nanosuspensions compared to 5 h with the drug solution.115

PULMONARY

Nanosuspensions have confirmed beneficial for delivering drugs that exhibit low solubility in pulmonary secretion. Nowadays , available tactics for pulmonary delivery such as aerosols or dry powder inhalers hold some drawbacks, such as incomplete diffusion at required site, less residence time, etc., That all problems can be overcome by nanosuspensions. Success formulation which overcome aforementioned problem of pulmonary delivery is Fluticasone and budesonide.116 Po-Chang Chiang et al. evaluated aerosol delivery of fluticasone nanosuspension for pre-clinical pulmonary delivery. Results showed that the aerosol delivery of fluticasone with nanosuspension was as effective as intranasal (IN) dosing and was able to attain dose-dependent lung deposition. Using lipopolysaccharide model, Po-Chang

Chiang et al. demonstrated pulmonary-targeted preclinical efficacy and differentiated the side effects after intratracheal administration of nanosuspensions of inhaled corticosteroids. However, it was only suitable at sub maximum efficacy levels.117

DERMAL

The nanocrystalline form shows enhanced saturation solubility, resulting in improved diffusion of the drug into the skin. With the properties, such as increased penetration into a membrane, enhanced permeation, and bio adhesiveness, this might be very suitable for dermal application. Pio et al. showed increased permeability of Diclofenec sodium nanosuspension across the skin compared to the control for transdermal delivery.118 Kobierski et al. developed nanosuspensions of the anti-oxidant resveratrol for dermal application.119 Pardeike and Muller studied dermal application of PX-18 and PX-13 nanosuspension for psoriasis treatment. The results of the EPISKIN test indicated that the nanosuspension was not irritating to the skin.120

TARGETING

Particle size is the main factor on which uptake of drug nanoparticles depends. By changing the surface properties of nanoparticles, there in vivo behavior can be altered; thus, they may use in targeted delivery systems. With the preparing stealth of nanocrystals or by preparing smartcrystals (i.e., drug particles less than 100 nm) We can solve a problem of phagocytotic uptake of nanocrystals. Due to the simplicity of these methods, nanosuspension is a commercially viable option for targeted drug delivery. Mucoadhesive nanosuspension has been reported for targeting Cryptosporidium parvum.[121] Surface properties of particle e.g., surface hydrophobicity, surface charge, presence, and concentration of certain functional groups are responsible for their structure distribution. Thus, nanocrystals coated with Tween 80 can be used for brain targeting. Atovaquone nanocrystals coated with Tween 80 were used to treat toxoplasmosis, and the parasites were efficiently eradicated in brain.[122] Shegaonkar and Singh reported using nevirapine nanosuspension for HIV reservoir targeting. Macrophage uptake studies have confirmed enhanced cellular uptake for nanosized nevirapine with no added cytotoxicity, while gamma scintigraphy studies have shown that nanosuspensions can be used to target the spleen, thymus and lungs, which represent anatomical viral reservoirs.123 In addition to these examples, clinical applications of nanocrystal technology have been summarized in an excellent review by Junghanns and Muller.136 Applications of nanosuspensions have been reported for many routes of administration, and a few examples are summarized in **Table 2.**

Table - 2: Reported Examples of Various Routes for Nanosuspension Technology.

Active	Route	Method Used	Used References
Amphotericin	Oral	High-pressure homogenization	[124]
Fenofibrate	Oral	High-pressure homogenization	[125]
1,3-dicyclohexylurea	Oral	Wet milling	[126]
AC88 and BA99	Oral	Wet milling	[124]
Nitrendipine	Oral	Precipitation-Ultrasonication	[127]
Hydroxycamptothecin I	Intravenous	Precipitation combined high pressure homogenization	[128]
Curcumin	Intravenous	High-pressure homogenization	[129]
Asulacrine (ASL)	Intravenous	High-pressure homogenization	[130]
Ornidone	Injectable	High-pressure homogenization	[131]
Fluticasone	Pulmonary	Wet milling	[116]
Budesonide	Pulmonary	High-pressure homogenization	[132]
Forskolin	Ocular	Wet crushing of crystals using a highperformance disperser	[114]
Amphotericin A	Ocular	Solvent displacement process	[133]
Hydrocotison	Ocular	Microfluidic nanoprecipitation and Wet milling	[115]
Diclofenec sodium	Transdermal	Homogenization followed by freeze-drying and ultrasonication	[134]
Hesperetin	Dermal	High-pressure homogenization	[135]

6. CONCLUSIONS

Nanosuspensions looking to be a exclusive and yet commercially viable approach to opposing problems such as poor bioavailability which are mainly connected with the delivery of hydrophobic drugs, including those which are poorly soluble in aqueous as well as organic media. Developing techniques such as media milling and high-pressure homogenization have been successfully employed for large-scale production of nanosuspensions. The advances in developing techniques using emulsions or microemulsions as templates have provided still simpler approaches for production but with limitations. Additional investigation in this regard is still required. Some attractive features have widened the applications of nanosuspensions for various routes, such as increased dissolution velocity, increased saturation solubility, improved bioadhesivity, versatility in surface modification and ease of post-production processing. The applications of nanosuspensions in parenteral and oral routes have been very well investigated and applications in pulmonary and ocular delivery have been become conscious. However, their applications in

buccal, nasal and topical delivery are still awaiting exploration. The development of stealth nanosuspensions fastened with functionalized surface coatings capable of eliciting passive or active targeting as per the requirement can be regarded as the future step in nanosuspension research.

REFERENCES

1. Aronov AM. Predictive in silico modeling for hERG channel blockers. Drug Discov Today. 2005;10(2):149-155.
2. Hoever M, Zbinden P. The evolution of microarrayed compound screening. Drug Discov Today. 2004;9(8):358-365.
3. Chavhan SS, Petkar KC, Sawant K. Nanosuspensions in drug delivery: recent advances, patent scenarios, and commercialization aspects. CRIT REV THER DRUG. 2011;28(5).
4. Nachname V. In silico prediction of drug solubility in water-dioxane mixtures using the Jouyban-Acree model. Pharmazie. 2007;62(1):46-50.
5. Stovall DM, Givens C, Keown S, Hoover KR, Barnes R, Harris C, et al. Solubility of crystalline nonelectrolyte solutes in organic solvents: mathematical correlation of 4-chloro-3-nitrobenzoic acid and 2-chloro-5-nitrobenzoic acid solubilities with the Abraham solvation parameter model. Physics Chemistry of Liquids. 2005;43(4):351-360.
6. Dulfer WJ, Bakker MW, Govers HA. Micellar solubility and micelle/water partitioning of polychlorinated biphenyls in solutions of sodium dodecyl sulfate. TECHNOL CANCER RES T. 1995;29(4):985-992.
7. Tokumura T, Muraoka A, Machida Y. Improvement of oral bioavailability of flurbiprofen from flurbiprofen/β-cyclodextrin inclusion complex by action of cinnarizine. Eur J Pharm Biopharm. 2009;73(1):202-204.
8. Reddy MN, Rehana T, Ramakrishna S, Chowdary K, Diwan PV. β-Cyclodextrin complexes of celecoxib: molecular-modeling, characterization, and dissolution studies. AAPS PharmSciTech. 2004;6(1):68-76.
9. Yu CD, Sweetana SA, Chu NI, Lee GJ, Massey I. Enhancement of solubility, dissolution rate, and oral bioavailability of RS-82856 by complex formation with cyclodextrins. Drug Development Industrial Pharmacy. 1989;15(4):609-620.
10. Park Y-J, Kwon R, Quan QZ, Oh DH, Kim JO, Hwang MR, et al. Development of novel ibuprofen-loaded solid dispersion with improved bioavailability using aqueous solution. Archives of Pharmacal Research. 2009;32(5):767-772.
11. Nepal PR, Han H-K, Choi H-K. Enhancement of solubility and dissolution of Coenzyme Q10 using solid dispersion formulation. Int J Pharm. 2010;383(1-2):147-153.
12. Liu C, Wu J, Shi B, Zhang Y, Gao T, Pei Y. Enhancing the bioavailability of cyclosporine a using solid dispersion containing polyoxyethylene (40) stearate. Drug Dev Ind Pharm. 2006;32(1):115-123.
13. Gwak H-S, Choi J-S, Choi H-K. Enhanced bioavailability of piroxicam via salt formation with ethanolamines. Int J Pharm. 2005;297(1-2):156-161.
14. Stegemann S, Leveiller F, Franchi D, De Jong H, Lindén H. When poor solubility becomes an issue: from early stage to proof of concept. Eur J Pharm Biopharm. 2007;31(5):249-261.
15. Muller R, Bohm B, Grau J. Nanosuspensions: a formulation approach for poorly soluble and poorly bioavailable drugs. J Drug Deliv Sci Technol. 2000:345-357.
16. Müller RH, Benita S, Bohm B. Emulsions and nanosuspensions for the formulation of poorly soluble drugs: CRC Press; **1998**.
17. Liversidge GG, Cundy KC. Particle size reduction for improvement of oral bioavailability of hydrophobic drugs: I. Absolute oral bioavailability of nanocrystalline danazol in beagle dogs. Int J Pharm. 1995;125(1):91-97.
18. Mosharraf M, Nyström C. The effect of particle size and shape on the surface specific dissolution rate of

microsized practically insoluble drugs. International journal of pharmaceutics. 1995;122(1-2):35-47.
19. Rabinow BE. Nanosuspensions in drug delivery. Nat Rev Drug Discov. 2004;3(9):785-796.
20. Merisko-Liversidge E. Nanocrystals: Resolving pharmaceutical formulation issues associated with poorly water-soluble compounds. Nuclear Particle Sci. 2002.
21. Böhm BH, Müller RH. Lab-scale production unit design for nanosuspensions of sparingly soluble cytotoxic drugs. PDA J Pharm Sci Technol. 1999;2(8):336-339.
22. Patravale V, Date AA, Kulkarni R. Nanosuspensions: a promising drug delivery strategy. J Pharm Pharmacol. 2004;56(7):827-840.
23. Sigfridsson K, Forssén S, Holländer P, Skantze U, de Verdier J. A formulation comparison, using a solution and different nanosuspensions of a poorly soluble compound. Eur J Pharm Biopharm. 2007;67(2):540-547.
24. Peters K, Leitzke S, Diederichs J, Borner K, Hahn H, Müller R, et al. Preparation of a clofazimine nanosuspension for intravenous use and evaluation of its therapeutic efficacy in murine Mycobacterium avium infection. J Antimicrob Chemoth. 2000;45(1):77-83.
25. Gao L, Zhang D, Chen M, Duan C, Dai W, Jia L, et al. Studies on pharmacokinetics and tissue distribution of oridonin nanosuspensions. Int J Pharm. 2008;355(1-2):321-327.
26. Pignatello R, Bucolo C, Ferrara P, Maltese A, Puleo A, Puglisi G. Eudragit RS100® nanosuspensions for the ophthalmic controlled delivery of ibuprofen. Eur J Pharm Biopharm. 2002;16(1-2):53-61.
27. Kassem M, Rahman AA, Ghorab M, Ahmed M, Khalil R. Nanosuspension as an ophthalmic delivery system for certain glucocorticoid drugs. Int J Pharm. 2007;340(1-2):126-133.
28. Jacobs C, Müller RH. Production and characterization of a budesonide nanosuspension for pulmonary administration. Trop J Pharm Res. 2002;19(2):189-194.
29. Chingunpituk J. Nanosuspension technology for drug delivery. Walailak J Sci & Tech. 2007;4(2):139-153.
30. Keck CM, Müller RH. Drug nanocrystals of poorly soluble drugs produced by high pressure homogenisation. Eur J Pharm Biopharm. 2006;62(1):3-16.
31. Müller RH, Jacobs C, Kayser O. Nanosuspensions as particulate drug formulations in therapy: rationale for development and what we can expect for the future. Adv Drug Deliv Rev. 2001;47(1):3-19.
32. Wu L, Zhang J, Watanabe W. Physical and chemical stability of drug nanoparticles. Adv Drug Deliv Rev. 2011;63(6):456-469.
33. Van Eerdenbrugh B, Van den Mooter G, Augustijns P. Top-down production of drug nanocrystals: nanosuspension stabilization, miniaturization and transformation into solid products. Int J Pharm. 2008;364(1):64-75.
34. Niwa T, Miura S, Danjo K. Universal wet-milling technique to prepare oral nanosuspension focused on discovery and preclinical animal studies–development of particle design method. Int J Pharm. 2011;405(1-2):218-227.
35. D'Addio SM, Prud'homme RK. Controlling drug nanoparticle formation by rapid precipitation. Adv Drug Deliv Rev. 2011;63(6):417-426.
36. Kakran M, Sahoo N, Li L, Judeh Z, Wang Y, Chong K, et al. Fabrication of drug nanoparticles by evaporative precipitation of nanosuspension. International Journal of Pharmaceutics. 2010;383(1-2):285-292.
37. Zhang X, Xia Q, Gu N. Preparation of all-trans retinoic acid nanosuspensions using a modified precipitation method. Drug Dev Ind Pharm. 2006;32(7):857-863.
38. Gassmann P, List M, Schweitzer A, Sucker H. Hydrosols: alternatives for the parenteral application of poorly water soluble drugs. Eur J Pharm Biopharm. 1994;40(2):64-72.
39. Muller R. Drug nanocrystals of poorly soluble drugs. J Nanosci Nanotechnol. 2004:627-638.
40. Violante MR, Fischer HW. Method for making uniformly-sized particles from insoluble compounds. Google Patents; **1991**.
41. Young TJ, Mawson S, Johnston KP, Henriksen IB, Pace GW, Mishra AK. Rapid expansion from supercritical to aqueous solution to produce submicron suspensions of water-insoluble drugs. Biotechnol. 2000;16(3):402-407.
42. Chattopadhyay P, Gupta RB. Production of griseofulvin nanoparticles using supercritical CO2 antisolvent with enhanced mass transfer. Int J Pharm. 2001;228(1-2):19-31.
43. Chan H-K, Kwok PCL. Production methods for nanodrug particles using the bottom-up approach. Advanced drug delivery reviews. 2011;63(6):406-416.
44. Dhumal RS, Biradar SV, Yamamura S, Paradkar AR, York P. Preparation of amorphous cefuroxime axetil nanoparticles by sonoprecipitation for enhancement

of bioavailability. Eur J Pharm Biopharm. 2008;70(1):109-115.
45. Eerikäinen H, Watanabe W, Kauppinen EI, Ahonen PP. Aerosol flow reactor method for synthesis of drug nanoparticles. Eur J Pharm Biopharm. 2003;55(3):357-360.
46. Chen X, Young TJ, Sarkari M, Williams III RO, Johnston KP. Preparation of cyclosporine A nanoparticles by evaporative precipitation into aqueous solution. Int J Pharm. 2002;242(1-2):3-14.
47. Lee SH, Heng D, Ng WK, Chan H-K, Tan RB. Nano spray drying: a novel method for preparing protein nanoparticles for protein therapy. International journal of pharmaceutics. 2011;403(1-2):192-200.
48. Dubey R. Impact of nanosuspension technology on drug discovery and development. Drug Deliv Technol. 2006;6(6).
49. Li W, Yang Y, Tian Y, Xu X, Chen Y, Mu L, et al. Preparation and in vitro/in vivo evaluation of revaprazan hydrochloride nanosuspension. International journal of pharmaceutics. 2011;408(1-2):157-162.
50. Wang Y, Liu Z, Zhang D, Gao X, Zhang X, Duan C, et al. Development and in vitro evaluation of deacety mycoepoxydiene nanosuspension. Colloids Surf B. 2011;83(2):189-197.
51. Lemke A, Kiderlen AF, Petri B, Kayser O. Delivery of amphotericin B nanosuspensions to the brain and determination of activity against Balamuthia mandrillaris amebas. Nanomedicine. 2010;6(4):597-603.
52. Trotta M, Gallarate M, Carlotti ME, Morel S. Preparation of griseofulvin nanoparticles from water-dilutable microemulsions. International journal of pharmaceutics. 2003;254(2):235-242.
53. Merisko-Liversidge E, Liversidge GG, Cooper ER. Nanosizing: a formulation approach for poorly-water-soluble compounds. Eur J Pharm Biopharm. 2003;18(2):113-120.
54. Singh SK, Srinivasan K, Gowthamarajan K, Singare DS, Prakash D, Gaikwad NB. Investigation of preparation parameters of nanosuspension by top-down media milling to improve the dissolution of poorly water-soluble glyburide. Eur J Pharm Biopharm. 2011;78(3):441-446.
55. Yang W, Johnston KP, Williams III ROJEjop, biopharmaceutics. Comparison of bioavailability of amorphous versus crystalline itraconazole nanoparticles via pulmonary administration in rats. 2010;75(1):33-41.
56. Chiang P-C, Alsup JW, Lai Y, Hu Y, Heyde BR, Tung D. Evaluation of aerosol delivery of nanosuspension for pre-clinical pulmonary drug delivery. Nanoscale Res Lett. 2009;4(3):254.
57. Itoh K, Pongpeerapat A, Tozuka Y, Oguchi T, Yamamoto K. Nanoparticle formation of poorly water-soluble drugs from ternary ground mixtures with PVP and SDS. Chem Pharm Bull. 2003;51(2):171-174.
58. Chingunpitak J, Puttipipatkhachorn S, Chavalitshewinkoon-Petmitr P, Tozuka Y, Moribe K, Yamamoto K. Formation, physical stability and in vitro antimalarial activity of dihydroartemisinin nanosuspensions obtained by co-grinding method. Drug Dev Ind Pharm. 2008;34(3):314-322.
59. Vogt M, Kunath K, Dressman JB. Dissolution improvement of four poorly water soluble drugs by cogrinding with commonly used excipients. Eur J Pharm Biopharm. 2008;68(2):330-337.
60. Arunkumar N, Deecaraman M, Rani C. Nanosuspension technology and its applications in drug delivery. Asian J Pharm. 2014;3(3).
61. Müller RH, Peters K. Nanosuspensions for the formulation of poorly soluble drugs: I. Preparation by a size-reduction technique. Int J Pharm. 1998;160(2):229-237.
62. Müller B, Müller R. Particle size analysis of latex suspensions and microemulsions by photon correlation spectroscopy. Int. J Pharm Sci Rev Res. 1984;73(7):915-918.
63. Boluk Y, Danumah C. Analysis of cellulose nanocrystal rod lengths by dynamic light scattering and electron microscopy. Journal of nanoparticle research. 2014;16(1):2174.
64. Chu B, Liu T. Characterization of nanoparticles by scattering techniques. J Nanoparticle Res. 2000;2(1):29-41.
65. Keck CM. Particle size analysis of nanocrystals: improved analysis method. Int J Pharm. 2010;390(1):3-12.
66. Nowacek AS, Balkundi S, McMillan J, Roy U, Martinez-Skinner A, Mosley RL, et al. Analyses of nanoformulated antiretroviral drug charge, size, shape and content for uptake, drug release and antiviral activities in human monocyte-derived macrophages. Journal of controlled release. 2011;150(2):204-211.
67. Ghosh I, Bose S, Vippagunta R, Harmon F. Nanosuspension for improving the bioavailability of a poorly soluble drug and screening of stabilizing agents to inhibit crystal growth. Int J Pharm. 2011;409(1-2):260-268.

68. Detroja C, Chavhan S, Sawant K. Enhanced antihypertensive activity of candesartan cilexetil nanosuspension: formulation, characterization and pharmacodynamic study. Sci Pharm. 2011;79(3):635-652.
69. Hunter RJ. Zeta potential in colloid science: principles and applications: Academic press; **2013**.
70. Yang SC, Zhu JB. Preparation and characterization of camptothecin solid lipid nanoparticles. Drug development industrial pharmacy. 2002;28(3):265-274.
71. Müller R, Jacobs C. Buparvaquone mucoadhesive nanosuspension: preparation, optimisation and long-term stability. Int J Pharm. 2002;237(1-2):151-161.
72. Cerdeira AM, Mazzotti M, Gander B. Miconazole nanosuspensions: influence of formulation variables on particle size reduction and physical stability. International journal of pharmaceutics. 2010;396(1-2):210-218.
73. El-Shabouri M. Positively charged nanoparticles for improving the oral bioavailability of cyclosporin-A. Int J Pharm. 2002;249(1-2):101-108.
74. Muller RH, Jacobs C, Kayser O. Nanosuspensions as particulate drug formulations in therapy: rationale for development and what we can expect for the future. Eur J Pharm Biopharm. 2001;47(1):3-19.
75. Chavhan SS, Petkar KC, Sawant K. Nanosuspensions in drug delivery: recent advances, patent scenarios, and commercialization aspects. Critical Reviews™ in Therapeutic Drug Carrier Systems. 2011;28(5).
76. Muller R, Grau M, editors. Increase of dissolution rate and solubility of poorly water soluble drugs as nanosuspension. Proceedings. World Meeting APGI/APV, Paris; 1998.
77. Shanthakumar T, Prakash S, Basavraj R, Ramesh M, Kant R, Venkatesh P, et al., editors. Comparative pharmacokinetic data of DRF-4367 using nanosuspension and HP--CD formulation. Proceedings of the International Symposium on Advances in Technology and Business Potential of New Drug Delivery Systems, Mumbai; 2004.
78. Yang W, Johnston KP, Williams III RO. Comparison of bioavailability of amorphous versus crystalline itraconazole nanoparticles via pulmonary administration in rats. Eur J Pharm Biopharm. 2010;75(1):33-41.
79. Lai F, Sinico C, Ennas G, Marongiu F, Marongiu G, Fadda AM. Diclofenac nanosuspensions: influence of preparation procedure and crystal form on drug dissolution behaviour. Int J Pharm. 2009;373(1-2):124-132.
80. Sharma P, Garg S. Pure drug and polymer based nanotechnologies for the improved solubility, stability, bioavailability and targeting of anti-HIV drugs. Adv Drug Deliv Rev. 2010;62(4-5):491-502.
81. Ravichandran R. In vivo pharmacokinetic studies of albendazole nanoparticulate oral formulations for improved bioavailability. Int J Med Nano Res. 2010;2(1):B46-B53.
82. Hecq J, Deleers M, Fanara D, Vranckx H, Amighi K. Preparation and characterization of nanocrystals for solubility and dissolution rate enhancement of nifedipine. Int J Pharm. 2005;299(1-2):167-177.
83. Jores K, Mehnert W, Drechsler M, Bunjes H, Johann C, Mäder K. Investigations on the structure of solid lipid nanoparticles (SLN) and oil-loaded solid lipid nanoparticles by photon correlation spectroscopy, field-flow fractionation and transmission electron microscopy. J Control Release. 2004;95(2):217-227.
84. Molpeceres J, Aberturas M, Guzman M. Biodegradable nanoparticles as a delivery system for cyclosporine: preparation and characterization. Journal of microencapsulation. 2000;17(5):599-614.
85. Mosqueira VCF, Legrand P, Gulik A, Bourdon O, Gref R, Labarre D, et al. Relationship between complement activation, cellular uptake and surface physicochemical aspects of novel PEG-modified nanocapsules. Biomaterials. 2001;22(22):2967-2979.
86. Nizri G, Magdassi S, Schmidt J, Cohen Y, Talmon Y. Microstructural characterization of micro-and nanoparticles formed by polymer– surfactant interactions. Langmuir. 2004;20(11):4380-4385.
87. Zatz J, Kushla G, Lieberman H, Lachman L, Schwatz J. Pharmaceutical Dosage Form: Dysperse System. Vol. 2. Marcell Dekker Inc. New York; **1996**.
88. Lee J, Lee S-J, Choi J-Y, Yoo JY, Ahn C-H. Amphiphilic amino acid copolymers as stabilizers for the preparation of nanocrystal dispersion. Eur J Pharm Biopharm. 2005;24(5):441-449.
89. Makhlof A, Miyazaki Y, Tozuka Y, Takeuchi H. Cyclodextrins as stabilizers for the preparation of drug nanocrystals by the emulsion solvent diffusion method. International journal of pharmaceutics. 2008;357(1-2):280-285.
90. Verma S, Kumar S, Gokhale R, Burgess DJ. Physical stability of nanosuspensions: investigation of the role of stabilizers on Ostwald ripening. Int J Pharm. 2011;406(1-2):145-152.
91. Choi J-Y, Yoo JY, Kwak H-S, Nam BU, Lee J. Role of polymeric stabilizers for drug nanocrystal dispersions. Curr Appl Phys. 2005;5(5):472-474.

92. Lee J, Choi J-Y, Park C. Characteristics of polymers enabling nano-comminution of water-insoluble drugs. International journal of pharmaceutics. 2008;355(1-2):328-336.
93. Van Eerdenbrugh B, Vermant J, Martens JA, Froyen L, Van Humbeeck J, Augustijns P, et al. A screening study of surface stabilization during the production of drug nanocrystals. Journal of pharmaceutical sciences. 2009;98(6):2091-2103.
94. Cerdeira AM, Mazzotti M, Gander B. Miconazole nanosuspensions: influence of formulation variables on particle size reduction and physical stability. Int J Pharm. 2010;396(1-2):210-218.
95. Mu RH. Manufacturing of nanoparticles by milling and homogenization techniques. Nanoparticle technology for drug delivery: CRC Press; 2006. p. 45-76.
96. Catsimpoolas N. Methods of protein separation: Springer Science & Business Media; **2013**.
97. Stolnik S, Davies MC, Illum L, Davis SS, Boustta M, Vert M. The preparation of sub-200 nm biodegradable colloidal particles from poly (β-malic acid-co-benzyl malate) copolymers and their surface modification with Poloxamer and Poloxamine surfactants. J Control Release. 1994;30(1):57-67.
98. Dolenc A, Kristl J, Baumgartner S, Planinšek O. Advantages of celecoxib nanosuspension formulation and transformation into tablets. International journal of pharmaceutics. 2009;376(1-2):204-212.
99. Van Eerdenbrugh B, Froyen L, Van Humbeeck J, Martens JA, Augustijns P, Van Den Mooter G. Alternative matrix formers for nanosuspension solidification: dissolution performance and X-ray microanalysis as an evaluation tool for powder dispersion. Eur J Pharm Biopharm. 2008;35(4):344-353.
100. Konan YN, Gurny R, Allémann E. Preparation and characterization of sterile and freeze-dried sub-200 nm nanoparticles. Int J Pharm. 2002;233(1-2):239-252.
101. Goldblith SA, Rey LR, Rothmayr W. Freeze drying and advanced food technology: Academic Press; 1975.
102. Lee J, Cheng Y. Critical freezing rate in freeze drying nanocrystal dispersions. J Control Release. 2006;111(1-2):185-192.
103. Du B, Li X-T, Zhao Y, Zhang Z-Z. Preparation and characterization of freeze-dried 2-methoxyestradiol nanoparticle powders. Pharmazie. 2010;65(7):471-476.
104. Van Eerdenbrugh B, Froyen L, Martens J, Blaton N, Augustijns P, Brewster M, et al. Characterization of physico-chemical properties and pharmaceutical performance of sucrose co-freeze–dried solid nanoparticulate powders of the anti-HIV agent loviride prepared by media milling. International journal of pharmaceutics. 2007;338(1-2):198-206.
105. Kim S, Lee J. Effective polymeric dispersants for vacuum, convection and freeze drying of drug nanosuspensions. Int J Pharm. 2010;397(1-2):218-224.
106. Dolenc A, Govedarica B, Kocbek P, Srcic S, Kristl J. Nanosized particles of orlistat with enhanced in vitro dissolution rate and lipase inhibition. Int J Pharm. 2010;396(1-2):149-155.
107. Basa S, Muniyappan T, Karatgi P, Prabhu R, Pillai R. Production and in vitro characterization of solid dosage form incorporating drug nanoparticles. Drug Dev Ind Pharm. 2008;34(11):1209-1218.
108. Schöler N, Krause K, Kayser O, Müller RH, Borner K, Hahn H, et al. Atovaquone nanosuspensions show excellent therapeutic effect in a new murine model of reactivated toxoplasmosis. Antimicrob Agents Chemother. 2001;45(6):1771-1779.
109. Fakes MG, Vakkalagadda BJ, Qian F, Desikan S, Gandhi RB, Lai C, et al. Enhancement of oral bioavailability of an HIV-attachment inhibitor by nanosizing and amorphous formulation approaches. Int J Pharm. 2009;370(1-2):167-174.
110. Majumdar AK, Howard L, Goldberg MR, Hickey L, Constanzer M, Rothenberg PL, et al. Pharmacokinetics of aprepitant after single and multiple oral doses in healthy volunteers. Br J Clin Pharmacol. 2006;46(3):291-300.
111. Xiong R, Lu W, Li J, Wang P, Xu R, Chen T. Preparation and characterization of intravenously injectable nimodipine nanosuspension. Int J Pharm. 2008;350(1-2):338-343.
112. Lou H, Gao L, Wei X, Zhang Z, Zheng D, Zhang D, et al. Oridonin nanosuspension enhances anti-tumor efficacy in SMMC-7721 cells and H22 tumor bearing mice. Colloids Surf B. 2011;87(2):319-325.
113. Zakir F, Sharma H, Kaur K, Malik B, Vaidya B, Goyal AK, et al. Nanocrystallization of poorly water soluble drugs for parenteral administration. Journal of biomedical nanotechnology. 2011;7(1):127-129.
114. Gupta S, Samanta MK, Raichur AM. Dual-drug delivery system based on in situ gel-forming nanosuspension of forskolin to enhance antiglaucoma efficacy. AAPS PharmSciTech. 2010;11(1):322-335.
115. Ali HS, York P, Ali AM, Blagden N. Hydrocortisone nanosuspensions for ophthalmic delivery: a comparative study between microfluidic

nanoprecipitation and wet milling. J Control Release. 2011;149(2):175-181.
116. Yang JZ, Young AL, Chiang P-C, Thurston A, Pretzer DKJJops. Fluticasone and budesonide nanosuspensions for pulmonary delivery: preparation, characterization, and pharmacokinetic studies. 2008;97(11):4869-4878.
117. Chiang P-C, Hu Y, Blom JD, Thompson DC. Evaluating the suitability of using rat models for preclinical efficacy and side effects with inhaled corticosteroids nanosuspension formulations. Nanoscale Res Lett. 2010;5(6):1010-1019.
118. Piao H, Kamiya N, Hirata A, Fujii T, Goto M. A novel solid-in-oil nanosuspension for transdermal delivery of diclofenac sodium. Trop J Pharm Res. 2008;25(4):896-901.
119. Kobierski S, Ofori-Kwakye K, Müller R, Keck C. Resveratrol nanosuspensions for dermal application–production, characterization, and physical stability. Pharmazie. 2009;64(11):741-747.
120. Pardeike J, Müller RHs. Dermal and ocular safety of the new phospholipase A2 inhibitors PX-18 and PX-13 formulated as drug nanosuspension. Journal of Biomedical Nanotechnology. 2009;5(4):437-444.
121. Kayser O. A new approach for targeting to Cryptosporidium parvum using mucoadhesive nanosuspensions: research and applications. Int J Pharm. 2001;214(1-2):83-85.
122. Shubar HM, Dunay IR, Lachenmaier S, Dathe M, Bushrab FN, Mauludin R, et al. The role of apolipoprotein E in uptake of atovaquone into the brain in murine acute and reactivated toxoplasmosis. J Drug Target. 2009;17(4):257-267.
123. Shegokar R, Singh K. Nevirapine nanosuspensions for HIV reservoir targeting. Pharmazie. 2011;66(6):408-415.
124. Kayser O, Olbrich C, Yardley V, Kiderlen A, Croft S. Formulation of amphotericin B as nanosuspension for oral administration. Int J Pharm. 2003;254(1):73-75.
125. Hanafy A, Spahn-Langguth H, Vergnault G, Grenier P, Grozdanis MT, Lenhardt T, et al. Pharmacokinetic evaluation of oral fenofibrate nanosuspensions and SLN in comparison to conventional suspensions of micronized drug. Adv Drug Deliv Rev. 2007;59(6):419-426.
126. Wahlstrom JL, Chiang P-C, Ghosh S, Warren CJ, Wene SP, Albin LA, et al. Pharmacokinetic evaluation of a 1, 3-dicyclohexylurea nanosuspension formulation to support early efficacy assessment. Nanoscale Res Lett. 2007;2(6):291-296.
127. Xia D, Quan P, Piao H, Piao H, Sun S, Yin Y, et al. Preparation of stable nitrendipine nanosuspensions using the precipitation–ultrasonication method for enhancement of dissolution and oral bioavailability. Eur J Pharm Biopharm. 2010;40(4):325-334.
128. Zhao Y-X, Hua H-Y, Chang M, Liu W-J, Zhao Y, Liu H-M. Preparation and cytotoxic activity of hydroxycamptothecin nanosuspensions. Int J Pharm. 2010;392(1-2):64-71.
129. Gao Y, Li Z, Sun M, Li H, Guo C, Cui J, et al. Preparation, characterization, pharmacokinetics, and tissue distribution of curcumin nanosuspension with TPGS as stabilizer. Drug Dev Ind Pharm. 2010;36(10):1225-1234.
130. Ganta S, Paxton JW, Baguley BC, Garg S. Formulation and pharmacokinetic evaluation of an asulacrine nanocrystalline suspension for intravenous delivery. Int J Pharm. 2009;367(1-2):179-186.
131. Lou H, Zhang X, Gao L, Feng F, Wang J, Wei X, et al. In vitro and in vivo antitumor activity of oridonin nanosuspension. Int J Pharm. 2009;379(1):181-186.
132. Yang JZ, Young AL, Chiang P-C, Thurston A, Pretzer DK. Fluticasone and budesonide nanosuspensions for pulmonary delivery: preparation, characterization, and pharmacokinetic studies. Int. J Pharm Sci Rev Res. 2008;97(11):4869-4878.
133. Das S, Suresh PK. Nanosuspension: a new vehicle for the improvement of the delivery of drugs to the ocular surface. Application to amphotericin B. Nanomedicine. 2011;7(2):242-247.
134. Piao H, Kamiya N, Hirata A, Fujii T, Goto M. A novel solid-in-oil nanosuspension for transdermal delivery of diclofenac sodium. Pharmaceutical research. 2008;25(4):896-901.
135. Mishra PR, Al Shaal L, Müller RH, Keck CM. Production and characterization of Hesperetin nanosuspensions for dermal delivery. International journal of pharmaceutics. 2009;371(1-2):182-189.
136. Junghanns J-UA, Müller RH. Nanocrystal technology, drug delivery and clinical applications. Int J Nanomedicine. 2008;3(3):295.

Leveraging Machine Learning to Strengthen Cybersecurity: Addressing Challenges and Embracing Opportunities

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Abstract: *As the internet evolves, cyber threats are constantly changing, posing challenges to the cybersecurity landscape. This paper focuses on the significance of data in machine learning and deep learning within the context of cybersecurity. It explores common network datasets used in machine learning and deep learning and discusses the difficulties associated with applying these techniques in cybersecurity. The evolution of malware, particularly the rise of bot malware and the formation of botnets, underscores the importance of analyzing network traffic to identify compromised machines. The paper aims to provide security professionals with insights into the application of machine learning techniques for detecting intrusion, malware, and spam. The goal is to improve detection and response capabilities in the dynamic field of cybersecurity. While machine learning shows promise, its effective use in cybersecurity requires ongoing exploration and refinement. By staying informed about emerging threats and continuously improving machine learning algorithms, we can strengthen our defenses against cyber-attacks and protect critical systems and data.*

Key Words: Machine learning, Deep learning, Cyber security, Adversarial learning, Statistics KDD.

Received Date: 30/10/2023 Revised Date: 28/11/2023 Acceptance Date: 15/12/2023

1. INTRODUCTION

The rapid evolution of the Internet has brought about a dynamic shift in the landscape of cyber-attacks, painting a less than optimistic picture for cybersecurity. This survey report delves into the critical realm of machine learning (ML) and deep learning (DL) methods as applied to network analysis for intrusion detection. A comprehensive exploration of existing literature surveys is presented, accompanied by concise tutorial descriptions for each ML/DL method.

In the contemporary digital era, computer systems and web services are undergoing a trend towards increased centralization. This centralization is accentuated by the fact that many applications have evolved to cater to vast user bases, reaching into the millions or even billions. While this concentration of information makes entities susceptible to exploitation, it also positions them strategically to leverage their data and user base for enhanced security measures. This phenomenon is further amplified by the convergence of powerful data processing hardware and the continuous development of sophisticated data analysis and machine learning algorithms. Indeed, the present juncture stands as an opportune moment to harness the potential of machine learning for fortifying cybersecurity defences.

Machine learning, as a facet of artificial intelligence, encompasses algorithms and processes designed to "learn" by generalizing from past data and experiences, facilitating the prediction of future outcomes. At its core, supervised machine learning methods embrace a Bayesian approach to knowledge discovery. These methods leverage the probabilities of previously observed events to infer the probabilities associated with new events. Conversely, unsupervised methods draw abstractions from unlabelled datasets, applying these abstractions to new data without prior categorization. Both these methodological families find application in solving problems of classification, where observations are assigned to predefined categories, or regression, where numerical properties of an observation are predicted.

To illustrate the concept of supervised machine learning, consider a scenario involving a set of animals with explicitly defined categories. For instance, we might know definitively that a dog and an elephant fall into the category of mammals, while an alligator and an iguana are categorized as reptiles. In a supervised setting, the task is to extract features from each labelled data point, identifying similarities in their properties to distinguish between animals of different classes.

The underlying mathematics and statistics, coupled with the algorithms that unearth patterns and correlations, form the backbone of machine-learning methodologies. The complexity of these algorithms, as well as their capacity to detect anomalies within data, varies widely. Anomalies within datasets are critical indicators in the cybersecurity domain, signalling potential threats or breaches. Thus, the continuous refinement and exploration of machine-learning algorithms become imperative in adapting to the ever-evolving cybersecurity landscape.

In conclusion, this introduction sets the stage for a comprehensive exploration of machine learning and deep learning methods in the context of cybersecurity. As we

delve into the subsequent sections, a detailed analysis of intrusion detection, network analysis, and the application of various ML/DL techniques will unfold, shedding light on the challenges and opportunities within this critical domain. [1]

Table - 2: Myths and Reality of Machine Learning

Myth	Reality
Machine learning in cybersecurity can fully replace human experts.	While powerful, machine learning cannot replace skilled cybersecurity professionals who offer contextual knowledge, creativity, critical thinking, intuition, and a nuanced understanding of complex attack vectors and cybercriminals' thinking.
Machine learning can address all threats and vulnerabilities.	Certain types of attacks, such as zero-day exploits or highly targeted and sophisticated attacks, can be missed by machine learning models that lack training in that area.
Machine learning models in cybersecurity do not make mistakes.	Machine learning models are only as good as the datasets they are fed. The results will be subpar or incorrect if the data is incomplete or inaccurate.
Machine learning renders attacks ineffective.	While machine learning models can adjust defenses to counter cyberattack vectors, criminals continuously adjust their approaches with a high degree of efficacy.
Machine learning in cybersecurity is impervious to adversarial attacks.	Unfortunately, machine learning is susceptible to adversarial attacks. If an attacker can inject misleading or incorrect data into a training dataset, the machine learning model will generate inaccurate results or make erroneous predictions.
Machine learning is only available to large organizations.	Machine learning is available and in wide use. Any organization can use and benefit from machine learning at some level by leveraging user-friendly security tools, cloud-based security services, and pre-built models.
Machine learning in cybersecurity requires large datasets to provide value.	The efficacy of machine learning improves with the volume of data provided, but models can be used and trained with smaller quantities of quality data.

1.1. Additional machine learning cybersecurity use cases.

Below is a list of common examples (not exhaustive) of ways machine learning can be used in the cybersecurity space [2].

Table - 2: Use cases with Description.

Use Case	Description
Vulnerability Management	Provides recommended vulnerability prioritization based on criticality for IT and security teams
Static File Analysis	Enables threat prevention by predicting file maliciousness based on a file's features
Behavioral Analysis	Analyzes adversary behavior at runtime to model and predict attack patterns across the cyber kill chain
Static & Behavioral Hybrid Analysis	Composes static file analysis and behavioral analysis to provide advanced threat detection
Anomaly Detection	Identifies anomalies in data to inform risk scoring and to direct threat investigations
Forensic Analysis	Runs counterintelligence to analyze attack progression and identify system vulnerabilities
Sandbox Malware Analysis	Analyzes code samples in isolated, safe environments to identify and classify malicious behavior, as well as map them to known adversaries

2. PROBLEM DEFINITION

In this segment, we highlight various factors that should be taken into account before opting for the implementation of machine learning (ML) algorithms in Network Operations Centers (NOC) and Security Operations Centers (SOC). It is important to note that, as of the current state-of-the-art, no algorithm can be regarded as entirely autonomous without some level of human supervision. We support each consideration with findings from either existing literature or original experiments conducted within large enterprises.

We initiate by outlining the testing environments used in our experiments and detailing the metrics utilized for evaluation. Our experiments primarily concentrate on Network Intrusion Detection, specifically employing the K-Means Network Intrusion Detection algorithm. We utilize

three labeled real training datasets, comprising benign and malicious network flows. These datasets are gathered from a sizable organization with nearly 10,000 hosts. The labels assigned to the data are established by identifying flows that triggered alerts from the enterprise network Intrusion Detection System (IDS) and were subsequently reviewed by a domain expert, who flagged them as malicious.

The dataset in question was employed for The Third International Knowledge Discovery and Data Mining Tools Competition, which coincided with KDD-99, the Fifth International Conference on Knowledge Discovery and Data Mining. The competition's objective was to develop a network intrusion detector—an anticipatory model capable of discerning between undesirable connections, referred to as intrusions or attacks, and favorable, normal connections. This database comprises a predefined collection of data intended for auditing purposes, encompassing a diverse range of simulated intrusions within a military network environment.

KDD, which stands for Knowledge Discovery in Databases, encompasses the comprehensive procedure of discovering knowledge within data, emphasizing the advanced application of specific data mining techniques. This field is pertinent to researchers engaged in machine learning, pattern recognition, databases, statistics, artificial intelligence, knowledge acquisition for expert systems, and data visualization. The overarching objective of the KDD process is to derive meaningful knowledge from data within the framework of extensive databases.[3]

Steps Involved in a Typical KDD Process [4]

1. Goal setting and Application Understanding
This is the first step in the process and requires prior understanding and knowledge of the field to be applied in. This is where we decide how the transformed data and the patterns arrived at by data mining will be used to extract knowledge. This premise is extremely important which, if set wrong, can lead to false interpretations and negative impacts on the end-user.

2. Data Selection and Integration
After setting the goals and objectives, the data collected needs to be selected and segregated into meaningful sets based on availability, accessibility importance and quality. These parameters are critical for data mining because they make the base for it and will affect what kinds of data models are formed.

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3. Data Cleaning and Preprocessing
This step involves searching for missing data and removing noisy, redundant, and low-quality data from the data set in order to improve the reliability of the data and its effectiveness. Certain algorithms are used for searching and eliminating unwanted data based on attributes specific to the application.

4. Data Transformation
This step prepares the data to be fed to the data mining algorithms. Hence, the data needs to be in consolidated and aggregate forms. The data is consolidated on the basis of functions, attributes, features etc.

5,6,7. Data Mining

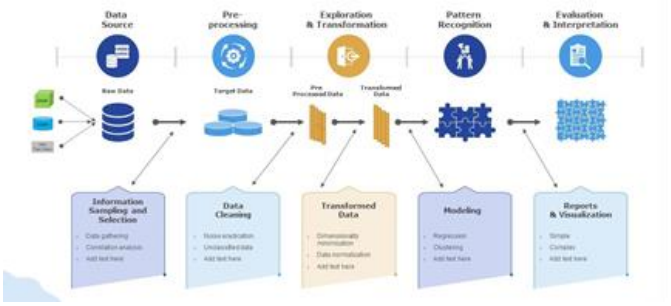

Fig -1: Data Mining Process Phases

3. THE KDD PROCESS

The knowledge discovery process (illustrates in the given **Figure - 2**) is iterative and interactive, comprises of nine steps. The process is iterative at each stage, implying that moving back to the previous actions might be required. The process has many imaginative aspects in the sense that one can't present one formula or make a complete scientific categorization for the correct decisions for each step and application type. Thus, it is needed to understand the process and the different requirements and possibilities in each stage [5,6,7]. The process begins with determining the KDD objectives and ends with the implementation of the discovered knowledge. At that point, the loop is closed, and the Active Data Mining starts. Subsequently, changes would need to be made in the application domain. For example, offering various features to cell phone users in order to reduce churn. This closes the loop, and the impacts are then measured on the new data repositories, and the KDD process again. Following is a concise description of the nine-step KDD process, Beginning with a managerial step [4]

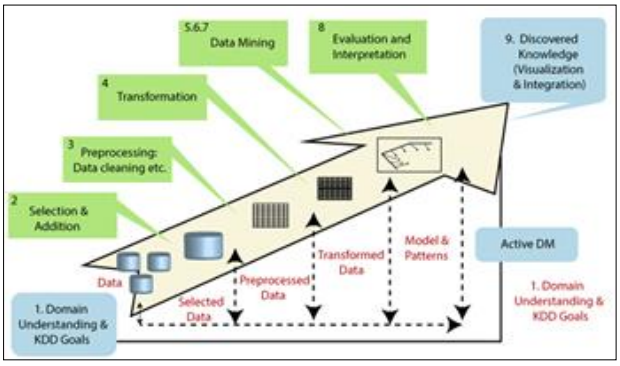

Fig -2: Steps in KDD Process

1 - Domain understanding and KDD Goals

This marks the initial preparatory phase, setting the stage for comprehending the necessary actions, such as transformations, algorithm selection, representation, and more. Those overseeing a Knowledge Discovery in Databases (KDD) project must grasp and define the objectives of the end-user, considering the environment in which the knowledge discovery process is set to unfold, which includes incorporating pertinent prior knowledge [9,10].

2 - Choosing and creating a data set

Once the objectives have been clearly defined, the next crucial step in the knowledge discovery process involves identifying the data that will be employed. This encompasses the exploration of available data, the acquisition of relevant information, and the subsequent integration of all gathered data into a unified set that embodies the characteristics intended for consideration in the knowledge discovery process [10,11].

This data determination process holds paramount significance due to the fact that Data Mining, a pivotal component of knowledge discovery, learns and derives insights from the available data. The data serves as the foundation for constructing models, and any omission of significant attributes can jeopardize the success of the entire study. The inclusion of a comprehensive set of attributes is vital for providing a rich evidence base that facilitates effective model development. However, [12,13] the inclusion of attributes needs to be balanced with considerations of cost and resource implications. Organizing, collecting, and managing extensive data repositories can be a resource-intensive undertaking. Therefore, there exists a delicate balance between the desire to incorporate a multitude of attributes for a nuanced understanding of phenomena and the practical constraints associated with resource utilization. This balance is a critical aspect where the interactive and iterative nature of the Knowledge Discovery in Databases (KDD) process comes into play.

The process commences with the identification of the best available datasets, acknowledging that these datasets may not encompass all conceivable attributes. Subsequently, the iterative aspect unfolds, characterized by an ongoing effort to expand the dataset by incorporating additional attributes. This expansion is guided by a continuous assessment of the impact on knowledge discovery and modeling. The iterative nature of this process allows for a dynamic and responsive approach, ensuring that the evolving needs and insights are accommodated.

In essence, the decision on which data to include is a strategic one that involves a careful consideration of the trade-offs between comprehensiveness and cost-effectiveness. It requires a thoughtful approach to strike a balance between the desire for a comprehensive dataset and the pragmatic constraints of resources. Moreover,[11] the selection of data is not solely about quantity but also about relevance. The data must align with the defined objectives of the knowledge discovery process and be reflective of the environment in which the insights will be applied. Understanding the context and purpose of the data is essential for ensuring that the selected dataset is meaningful and conducive to the achievement of the desired goals.

In conclusion, the determination of data for the knowledge discovery process is a pivotal stage that sets the foundation for subsequent analyses and model development. It involves a careful consideration of available data, the acquisition of pertinent information, and the strategic integration of attributes. The dynamic and iterative nature of this process allows for adaptability in response to evolving insights, striking a balance between data comprehensiveness and resource efficiency. Ultimately, the success of the knowledge discovery endeavor hinges on the thoughtful and purposeful selection of data that aligns with the defined objectives and contextual requirements.

3 - Preprocessing and cleansing

During this crucial phase, the focus is on enhancing the reliability of the data, encompassing processes such as data cleaning, handling missing values, and addressing noise or outliers. These tasks may involve the application of intricate statistical techniques or the utilization of Data Mining algorithms to ensure the data's quality and integrity. An exemplary approach involves treating specific attributes suspected of lacking reliability or containing substantial missing data as targets for Data Mining supervised algorithms.

The first facet of this enhancement process involves data cleaning, a meticulous procedure aimed at rectifying imperfections within the dataset. One of the prevalent challenges is handling missing quantities, where certain data points are absent or incomplete. To overcome this hurdle, sophisticated statistical techniques may be employed, or alternatively, Data Mining algorithms can be leveraged to impute missing values. For instance, if a particular attribute is deemed unreliable due to significant missing data, it becomes a prime candidate for the application of a Data Mining supervised algorithm [13,14].

This entails the creation of a prediction model for the unreliable attribute, enabling the estimation and filling in of missing data. Furthermore, the refinement process extends to addressing noise and outliers within the dataset. Noise, characterized by random and irrelevant variations in the data, can distort the accuracy of models and analyses. Outliers, on the other hand, are data points that deviate significantly from the overall pattern. Both phenomena can adversely impact the reliability of insights derived from the data. To mitigate these issues, advanced statistical methods or specialized Data Mining algorithms can be employed to identify and filter out noise and outliers, ensuring a more robust dataset. The decision on the extent to which attention is directed toward these data refinement efforts hinges on several factors. The nature and importance of the

data, as well as the specific objectives of the knowledge discovery process, play pivotal roles in guiding the level of scrutiny applied during this phase. While some datasets may require meticulous cleaning and noise reduction due to their critical role in decision-making processes, others may permit a more lenient approach. Regardless of the level of attention dedicated to data refinement, delving into these aspects is indispensable. The insights gained from studying and addressing issues such as missing data, noise, and outliers can be illuminating in and of themselves. This process not only contributes to the reliability of enterprise data systems but also fosters a deeper understanding of the intricacies within the dataset, thereby enhancing the overall effectiveness of subsequent analyses.

In essence, the step of enhancing data reliability is a multifaceted undertaking that involves meticulous cleaning, handling missing values, and mitigating noise or outliers. The application of advanced statistical techniques and Data Mining algorithms is instrumental in fortifying the dataset's integrity. The targeted prediction models for attributes with missing data exemplify a proactive approach to addressing data deficiencies [15,16,17]. The decision on the intensity of data refinement efforts is contingent upon various factors, and the insights gained from this process contribute not only to data reliability but also to a more profound comprehension of the dataset's intricacies. Ultimately, this phase plays a pivotal role in ensuring the robustness and accuracy of the data utilized in the knowledge discovery process.

4 - Data Transformation

In this phase, the groundwork and refinement of data tailored for Data Mining take center stage. Various techniques are employed, including dimension reduction through methods such as feature selection and extraction, as well as record sampling. Additionally, attribute transformation techniques, such as discretization of numerical attributes and functional transformation, are integrated into the process. The success of the entire Knowledge Discovery in Databases (KDD) project is contingent upon this step, and its specifics often vary based on the unique requirements of the project.

Dimension reduction techniques play a pivotal role in streamlining the dataset for more effective analysis. This involves selecting pertinent features and extracting relevant information, which is particularly critical in scenarios where certain attributes may carry more significance when considered in conjunction with others. For instance, in medical assessments, the interplay of attributes may hold greater importance than individual attributes alone. In the business domain, considerations may extend beyond individual factors to include external influences, efforts, and transient issues. An illustrative example is evaluating the impact of cumulative advertising efforts [8,18,19]. Attribute transformation, another key aspect, involves modifying the nature of attributes to facilitate a more meaningful analysis. This may encompass discretizing numerical attributes or applying functional transformations. The choice of transformation techniques is often project-specific and depends on the nature of the data and the objectives of the knowledge discovery process.

The importance of selecting the right transformation strategies at the outset cannot be overstated. A well-chosen transformation can yield significant insights, whereas an inappropriate one may lead to misleading results. Iterative refinement becomes crucial, as the KDD process builds upon itself. The insights gained from one iteration inform the need for subsequent transformations, creating a continuous feedback loop that refines the understanding of the data and the transformations required for optimal analysis.

In summary, this stage involves crafting and refining data specifically tailored for Data Mining. Techniques such as dimension reduction and attribute transformation are employed to streamline the dataset for effective analysis. The success of the overall KDD project hinges on the careful selection of transformation strategies, which are often project-specific. The iterative nature of the process underscores the importance of continuously refining the transformation approaches based on insights gained in each iteration, ensuring an evolving and nuanced understanding of the data.

5 - Prediction and description

At this juncture, the focus shifts to determining the type of Data Mining methodology to employ, considering options such as classification, regression, clustering, and more. This decision is intricately tied to the objectives set forth in the Knowledge Discovery in Databases (KDD) process and is heavily influenced by the insights gleaned from earlier stages. The primary objectives in Data Mining typically revolve around prediction and description, each aligning with specific methodologies.

Prediction, often synonymous with supervised Data Mining, entails the development of models capable of forecasting outcomes based on existing data patterns. This approach relies on the premise that the model, once trained on an ample dataset, can effectively generalize to predict future cases. [20,21,22] The supervised nature of this methodology involves the use of labeled training data, where the algorithm learns from explicit examples to make predictions or classifications. The objective is to create a predictive model that can be applied to new, unseen data to anticipate outcomes or trends. On the other hand, descriptive Data Mining encompasses unsupervised and visualization aspects, emphasizing the exploration and understanding of data patterns without explicit guidance from labeled examples. Unlike prediction, which is forward-looking, descriptive Data Mining is more retrospective in nature, seeking to unveil inherent structures and relationships within the data. Unsupervised techniques, such as clustering, aim to group similar data points together based on intrinsic patterns, facilitating the identification of natural clusters or categories.

Inductive learning serves as a foundational principle for many Data Mining techniques. In this approach, a model is constructed, either explicitly or implicitly, by generalizing from a substantial number of training examples. The underlying assumption is that the patterns identified in the training data are applicable to future cases, allowing for predictions or insights beyond the scope of the training set. The choice of Data Mining methodology is not solely dependent on the overarching objectives but is also influenced by the specific characteristics of the data and the insights gained in preceding steps. For instance, the nature of the available data, the level of granularity required for analysis, and the desired depth of understanding all play pivotal roles in shaping the selection of the appropriate technique. Meta-learning, an additional consideration, involves understanding the inherent characteristics of the available data. This meta-level understanding guides the selection and application of Data Mining techniques, ensuring a more nuanced and tailored approach. The effectiveness of the chosen methodology is contingent upon the alignment between the selected technique and the unique attributes of the dataset at hand.

In summary, the decision on which Data Mining approach to employ is a pivotal step in the KDD process. The distinction between prediction and description, guided by supervised and unsupervised methodologies, respectively, sets the stage for subsequent analyses. The inductive learning principle forms the backbone of many techniques, emphasizing the generalization from training examples to future cases. Moreover, the consideration of meta-learning factors in, ensuring that the chosen methodology aligns with the specific characteristics of the dataset, adds an additional layer of sophistication to the decision-making process. Ultimately, this step serves as a crucial bridge between the goals of the KDD process and the practical application of Data Mining techniques.

6 - Selecting the Data Mining algorithm

Now equipped with a chosen Data Mining technique, the next critical step involves determining the strategies for deploying that technique effectively. This stage entails selecting a specific approach for uncovering patterns, often involving multiple inducers. [23,24] The decision-making process is influenced by factors such as precision versus understandability, with considerations like neural networks being preferred for precision, while decision trees are favored for their interpretability. Precision, as a metric, emphasizes the accuracy and reliability of predictions, making it a critical factor in scenarios where the utmost accuracy is paramount. Neural networks, with their intricate architecture and capacity for capturing complex relationships, are often chosen when precision is the primary concern. However, this precision comes at the cost of interpretability, as neural networks can be perceived as "black box" models, making it challenging to comprehend the underlying decision-making processes.

On the other hand, understandability becomes a crucial criterion when the interpretability of the model is of greater significance. Decision trees, for example, are known for their transparent and easy-to-follow structure, making them an excellent choice in situations where comprehensibility is a priority. The trade-off between precision and understandability necessitates a thoughtful consideration of the specific goals and requirements of the Knowledge Discovery in Databases (KDD) project.

The incorporation of meta-learning strategies adds an additional layer of sophistication to this decision-making process. Meta-learning aims to elucidate the factors contributing to the success or failure of a Data Mining algorithm in a particular context. This approach strives to uncover the conditions under which a specific Data Mining algorithm is most effective. Each algorithm within the chosen technique comes with parameters and learning strategies, such as ten-fold cross-validation or alternative methods for dividing the dataset into training and testing subsets. Understanding the nuances of these parameters and strategies is crucial for fine-tuning the application of the chosen Data Mining technique. For instance, cross-validation methods, including ten-fold cross-validation, play a pivotal role in assessing the generalization performance of a model by partitioning the data into subsets for training and testing. The selection of an appropriate cross-validation strategy depends on factors such as dataset size, characteristics, and computational resources [25,26]. The iterative nature of meta-learning allows for an adaptive and informed approach to refining the parameters and strategies employed. By gaining insights into the specific conditions that influence the success of a Data Mining algorithm, practitioners can make informed adjustments to enhance performance in a given context.

In conclusion, the decision on strategies for deploying a chosen Data Mining technique is a multifaceted process that involves balancing precision and understandability. The selection of specific algorithms and their associated parameters is guided by the goals of the KDD project and the inherent characteristics of the data. Meta-learning adds a valuable dimension by seeking to understand the contextual factors influencing algorithmic success, enabling a more informed and adaptive approach. Ultimately, the effectiveness of the chosen strategies is pivotal in realizing the full potential of the Data Mining technique in uncovering meaningful patterns and insights within the dataset.

7 - Utilizing the Data Mining algorithm

Finally, the process culminates in the implementation of the chosen Data Mining algorithm. At this stage, it may be necessary to iterate through the utilization of the algorithm multiple times until a satisfactory outcome is achieved. This iterative approach involves adjusting algorithmic control parameters, such as the minimum number of instances in a

single leaf of a decision tree, to fine-tune the model and optimize its performance [27,28].

8 - Evaluation

In this phase, we evaluate and interpret the patterns and rules extracted by the Data Mining process in alignment with the objectives defined in the initial step. We carefully examine the preprocessing steps, acknowledging their influence on the results obtained from the Data Mining algorithm. For instance, we assess the impact of including a specific feature in step 4 and iterate from that point if necessary. The emphasis during this stage is on the clarity and usefulness of the generated model [29,30].

Comprehensibility and utility are key considerations, ensuring that the induced model is not only understandable but also valuable in addressing the goals set at the outset. Additionally, this step involves documenting the identified knowledge for future reference and utilization. Finally, the last phase involves the practical application of the insights gained through Data Mining, providing an overarching feedback loop that refines and enhances the overall discovery results.

9 - Using the discovered knowledge

Now, equipped with the extracted knowledge, we are poised to integrate it into another system for further action. The effectiveness of this integration lies in the ability to implement changes to the system and measure their impacts. The success of this step is pivotal in determining the overall effectiveness of the entire Knowledge Discovery in Databases (KDD) process. However, this phase poses several challenges, notably the transition from the controlled "laboratory conditions" under which the knowledge was discovered. Initially, the knowledge is derived from a specific static representation, often in the form of a dataset. However, as we move to implementation, the nature of the data becomes dynamic. This shift introduces complexities, as the conditions under which the knowledge was acquired may not fully align with the real-time, evolving nature of the system. [31,32,33] The transition from static depiction to a dynamic operational environment requires careful consideration and adaptation. One challenge encountered in this step involves the dynamic nature of data. Data structures may undergo changes, leading to the unavailability of certain quantities that were present during the knowledge discovery phase.

The dynamic nature of data introduces a level of uncertainty, and adjustments must be made to ensure that the knowledge remains applicable and relevant in the evolving data landscape. Moreover, the transformation from a controlled environment to the operational system introduces the potential for unexpected modifications in the data domain. Attributes that were previously stable may now exhibit values that were not anticipated during the knowledge discovery process. This unpredictability underscores the need for a robust and adaptable implementation strategy to accommodate these changes seamlessly. Implementing knowledge into another system involves more than just technical considerations [34,35]. It requires a comprehensive understanding of the broader context, encompassing organizational dynamics, user needs, and the intended impact on business processes. The success of the implementation is not solely measured by technical efficiency but also by its alignment with organizational goals and the ability to enhance decision-making and operations. The challenges in this phase emphasize the importance of continuous monitoring and adaptation. As the system undergoes changes and the data environment evolves, there is a need for ongoing evaluation to ensure that the implemented knowledge remains effective. This iterative process involves feedback loops, allowing for adjustments and refinements based on real-world outcomes.

In conclusion, the transition from knowledge discovery to knowledge implementation is a critical phase in the KDD process. The challenges in this step, such as the shift from laboratory conditions to dynamic operational settings, underscore the need for adaptability and continuous monitoring. The success of this phase is contingent on the seamless integration of knowledge into the system, considering the evolving nature of data, potential structural changes, and unanticipated modifications in the data domain. Ultimately, the effectiveness of the entire KDD process is realized when the extracted knowledge contributes to tangible improvements in the targeted system and decision-making processes.

4. THE KDD CUP 99 DATA SET

Network security is becoming more and more crucial due to the massive rise in the number of applications operating on computer networks and the enormous growth in their usage. It is evident from [36,37,38] that every computer system has security flaws that are costly for manufacturers to fix due to their technical difficulty.

As a result, intrusion detection systems' (IDSs') function as specialized tools for identifying irregularities and network attacks is growing in significance.

5. K-MEANS CLUSTERING ALGORITHM

The mean algorithm, which divides n observations into k clusters, is the most basic unsupervised learning algorithm for solving the clustering problem. Each observation is assigned to a cluster, with the cluster prototype being the closest mean [39,40, 41].

Algorithm Steps:

1 - Indicate the number of clusters (K).
2 - Choose K data points at random for the centroid without replacing them after first rearranging the dataset.
3 - Continue iterating until the centroid remains unchanged. i.e., the distribution of data points among clusters remains constant.

4 - Sum the squared distances between each centroid and each data point.
5 - Assign each data point to the centroid, or nearest cluster.
6 - By averaging all of the data points that are a part of each cluster, find the centroid for each cluster.

6. CONCLUSIONS

The independence of machine learning algorithms shouldn't be overstated, as the lack of human oversight can make it easier for a determined adversary to compromise an organization, steal information, and even carry out acts of sabotage. Because of the internet, machine learning is a very broad area of computer science in modern technology. In this world of evolution, communication, data passing, and security are the main problems. As machine and deep learning techniques are being used more and more for a variety of purposes, including cyber security, it is critical to determine which category of algorithms is capable of producing satisfactory results at what time. We examine these methods for three pertinent cyber security issues: spam, malware, and intrusion detection.

REFERENCES

1] https://www.sailpoint.com/identity-library/how-ai-and-machine-learning-are-improving-cybersecurity
2] https://www.crowdstrike.com/cybersecurity-101/machine-learning-cybersecurity/
3] https://www.upgrad.com/blog/kdd-process-data-mining/
4] https://www.javatpoint.com/kdd-process-in-data-mining
5] https://www.geeksforgeeks.org/kdd-process-in-data-mining/https://www.scaler.com/topics/data-mining-tutorial/kdd-in-data-mining/
6] https://www.datascience-pm.com/kdd-and-data-mining/
7] https://www.sciencedirect.com/topics/computer-science/knowledge-discovery-in-database
8] Anita S. Kini, A. Nanda Gopal Reddy, Manjit Kaur, S. Satheesh, Thomas Martinetz, Hammam Alshazly, "Ensemble Deep Learning and Internet of Things-Based AutomatedCOVID-19 Diagnosis Framework", Contrast Media & Molecular Imaging, vol. 2022, Article ID 7377502, 10 pages, 2022. https://doi.org/10.1155/2022/7377502
9] Aditi Sharan, "Term Co-occurrence and Context Window based Combined Approach for Query Expansion with the Semantic Notion of Terms", International Journal of Web Science(IJWS), Inderscience, Vol. 3, No. 1, 2017.
10] Saurabh Kumar, S.K. Pathak, "A Comprehensive Study of XSS Attack and the Digital Forensic Models to Gather the Evidence". ECS Transactions, Volume 107, Number 1, 2022.
11] Yadav, C.S.; Pradhan, M.K.; Gangadharan, S.M.P.; Chaudhary, J.K.; Khan, A.A.; Haq, M.A.; Alhussen, A.; Wechtaisong, C.; Imran, H.; Alzamil, Z.S.; Pattanayak, H.S. "Multi-Class Pixel Certainty Active Learning Model for Classification of Land Cover Classes Using Hyperspectral Imagery". Electronics 2022, 11, 2799. https://doi.org/10.3390/electronics11172799.
12] Yadav, C.S.; Yadav, A.; Pattanayak, H.S.; Kumar, R.; Khan, A.A.; Haq, M.A.; Alhussen, A.; Alharby, S. "Malware Analysis in IoT & Android Systems with Defensive Mechanism". Electronics 2022, 11, 2354. https://doi.org/10.3390/electronics11152354.
13] A Goswami, D Sharma, H Mathuku, SMP Gangadharan, CS Yadav, "Change Detection in Remote Sensing Image Data Comparing Algebraic and Machine Learning Methods", Electronics,
14] Singh, J. "An Efficient Deep Neural Network Model for Music Classification", Int. J. Web Science, Vol. 3, No. 3, 2022.
15] Vijay Kumar Bohat, "Neural Network Model for Recommending Music Based on Music Genres", In 10th IEEE International Conference on Computer Communication and Informatics (ICCCI - 2021), Jan. 27-29, 2021, Coimbatore, INDIA.
16] Singh, J., "Learning based Driver Drowsiness Detection Model", In 3rd IEEE International Conference on Intelligent Sustainable Systems (ICISS 2020), , pp. 1163-1166, Palladam, India, Dec. 2020.
17] A. Sharan, "Rank fusion and semantic genetic notion based automatic query expansion model",Swarm and Evolutionary Computation, Vol-38, Elsevier, 2018.
18] R. Singh, "Ranks Aggregation and Semantic Genetic Approach based Hybrid Model for Query Expansion ", International Journal of Computational Intelligence Systems, Vol. 10 (2017) 34–55.
19] A. Sharan, "A new fuzzy logic based query expansion model for efficient information retrieval using relevance feedback approach", Neural Computing And Applications, Vol 28, Springer, 2017.
20] Chin-Teng Lin, Mukesh Prasad, Chia-Hsin Chung, Deepak Puthal, Hesham El-Sayed, Sharmi Sankar, Yu-Kai Wang, Jagendra Singh, Arun Kumar Sangaiah, "IoT-based Wireless Polysomnography Intelligent System for Sleep Monitoring", IEEE Access, Vol 6, Oct 2017
21] Mukesh Prasad, Yousef Daraghmi, Prayag Tiwari, Pranay Yadav, Neha Bharill, "Fuzzy Logic Hybrid Model with Semantic Filtering Approach for Pseudo Relevance Feedback- based Query Expansion", 2017 IEEE Symposium Series on Computational Intelligence (SSCI), 2017.
22] Rakesh Kumar, "Lexical Co-Occurrence and Contextual Window-Based Approach with Semantic Similarity for Query Expansion", International Journal of Intelligent Information Technologies (IJIIT), IGI, Vol. 13, No. 3, pp. 57-78, 2017.
23] Aditi Sharan, "Term Co-occurrence and Context Window based Combined Approach for Query Expansion with the Semantic Notion of Terms", International Journal of Web Science(IJWS), Inderscience, Vol. 3, No. 1, 2017.
24] Mukesh Prasad, Om Kumar Prasad, Er Meng Joo, Amit Kumar Saxena and Chin- Teng Lin, "A Novel Fuzzy Logic Model for Pseudo-Relevance Feedback-Based Query Expansion", International Journal of Fuzzy Systems, Springer, Vol 18, 2016.
25] A. Sharan, "Ranks aggregation and semantic genetic approach based hybrid model for query expansion", International Journal of Computational Intelligence Systems, Taylor & Francis, Vol. 10, Issue 1, 2017, Pages 34 - 55
26] A. Sharan, "Relevance Feedback based Query Expansion Model using Ranks Combining and Word2vec Approach", IETE- Journal of Research, Taylor & Francis, Vol 62, 2016.
27] K. Singh and Aditi Sharan, "Relevance feedback based query expansion model using Borda count and semantic similarity

approach", Computational Intelligence and Neuroscience, Article ID: 568197, pp. 1-14, 2015.

28] A. Sharan, "Context Window based Co-occurrence Approach for ImprovingFeedback based Query Expansion in Information Retrieval", International Journal of Information Retrieval Research, IGI, Vol. 5, No. 4, pp. 32-46, 2015.

29] Yadav, A., Kumar, S., Singh, J. (2022). A Review of Physical Unclonable Functions (PUFs) and Its Applications in IoT Environment. In: Hu, YC., Tiwari, S., Trivedi, M.C., Mishra, K.K. (eds) Ambient Communications and Computer Systems. Lecture Notes in Networks and Systems, vol 356. Springer, Singapore. https://doi.org/10.1007/978-981-16-7952-0_1

30] Saurabh Kumar, Suryakant Pathak (2022) An enhanced digital forensic investigation framework for XSS attack, Journal of Discrete Mathematical Sciences and Cryptography, 25:4, 1009-1018, DOI: 10.1080/09720529.2022.2072424

31] Sharan, A., "A novel model of selecting high quality pseudo-relevance feedback documents using classification approach for query expansion," 2015 IEEE Workshop on Computational Intelligence: Theories, Applications and Future Directions (WCI), 2015, pp. 1-6, doi:10.1109/WCI.2015.7495539.

32] A. Sharan, "Lexical Ontology based Computational Model to Find SemanticSimilarity", Intelligent Computing Networking and Informatics, Advances in Intelligent Systems and Computing, AISC Series, Springer, Vol. 243, pp. 119-128, 2014.

33] A. Sharan, "A new fuzzy logic based query expansion model for efficient information retrieval using relevance feedback approach", Neural Computing And Applications, Vol 28, Springer, 2017.

34] Aditi Sharan, "Term Co-occurrence and Context Window based Combined Approach for Query Expansion with the Semantic Notion of Terms", International Journal of Web Science(IJWS), Inderscience, Vol. 3, No. 1, 2017.

35] R. Aggarwal, S. Tiwari and V. Joshi, "Exam Proctoring Classification Using Eye Gaze Detection," 2022 3rd International Conference on Smart Electronics and Communication (ICOSEC), 2022, pp. 371-376, doi: 10.1109/ICOSEC54921.2022.9951987.

36] Himansu Sekhar Pattanayak, " Multi-class Pixel Certainty Active Learning Model for Remote Sensing Applications", Electronics, Article id - 1791849, 2022.

37] Kamal Upreti, Aditya Kr. Gupta, Nandan Dave, Arihant Surana and Durgesh Mishra, "Deep Learning Approach for Hand Drawn Emoji Identification", In 2022 IEEE International Conference on Current Development in Engineering and Technology (CCET), 23-24 Dec, SAGE University, Bhopal, India, 2022.

38] Kamal Upreti, Snigdha Shrivastava, Aabhas Garg, Anupam Kumar Sharma, "Prediction & Detection of Cardiovascular Diseases using Machine Learning Approaches", In 2022 IEEE International Conference on Communication, Security and Artificial Intelligence (ICCSAI-2022), 24-25 Dec, Galgotia University, Greater Noida, India, 2022.

39] Aruna Yadav, A., Kumar, S., Singh, J. (2022). A Review of Physical Unclonable Functions (PUFs) and Its Applications in IoT Environment. In: Hu, YC., Tiwari, S., Trivedi, M.C., Mishra, K.K. (eds) Ambient Communications and Computer Systems. Lecture Notes in Networks and Systems, vol 356. Springer, Singapore. https://doi.org/10.1007/978-981-16-7952-0_1

40] Pramod G. Musrif, J Singh, Amol More, Ashish Shankar, Ramkrishna (2023), "Design of Green IoT for Sustainable Smart Cities and Ecofriendly Environment", European Chemical Bulletin Journal, Volume 12, issue 6, 2023.

HPLC Method Development and Validation of Artesunate and Amodiaquine in Tablet Dosage Form

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Abstract: *An accurate and precise HPLC method was developed and validated for simultaneous estimation of Amodiaquine and Artesunate in tablet dosage form. The column was used was C18 column (150 x 4.6mm, 5µm) column and a mobile phase composed of Potassium Dihydrogen o-Phosphate Buffer (pH-5): Acetonitrile (50:50 v/v). The flow rate was kept 1 ml/min and the detection wavelength was 280nm. The retention time for Amodiaquine and Artesunate was found to be 3.1 min and 5.3 min respectively. The linearity range was found to be 12.5-37.5 µg/ml for Artesunate with corelation coefficient 0.999 and 32.5-97.5 µg/ml for Amodiaquine corelation coefficient 0.9999 for Artesunate and Amodiaquine respectively. The LOD and LOQ for Artesunate 0.32 and 0.97 µg/ml was found to be and for Amodiaquine was found to be 0.50 and 0.82 respectively. The repeatability, intraday and interday precision was found to be less than 2%. The proposed methods for estimation of Amodiaquine and Artesunate were found to be selective, precise and accurate. The developed validated method is applicable for the simultaneous determination of Amodiaquine and Artesunate in tablet dosage form.*

***Key Words*:** HPLC, Amodiaquine, Artesunate

Received Date: 18/07/2023 Revised Date: 22/09/2023 Acceptance Date: 27/09/2023

1. INTRODUCTION

Malaria is a life-threatening disease caused by parasites of the Plasmodium genus. Malaria is caused by 4 species of the protozoa parasite. Plasmodium is endemic. In the most parts of India four species of plasmodium cause human malaria: Plasmodium falciparum, Plasmodium Vivax, Plasmodium Malariae and Plasmodium Ovale. Malaria parasites are transmitted from one person to another by the female anopheles' mosquitoes. Artemisinin based combination therapy is based on the use of two drugs with different modes of action. Artemisinin-derivative that causes rapid and effective reduction of parasite biomass and gametocyte carriage and second drug that has a longer duration of action.

1.1. Artesunate

Artesunate is an antimalarial agent of chemical class a hemisuccinate derivative of dihydroartemisinin and is chemically known as 4-oxo-4-{[(1R,4(3R,5aS,6R,8aS,9R,10S,12R,12aR)-Decahydro-3,6,9-trimethyl-3,12-epoxy-12H-pyrano[4,3-j]-1,2-benzodioxepin-10-ol, hydrogen succinate, which is itself obtained by the reduction of artemisinin, a sesquiterpene lactone endoperoxide. Amodiaquine is class of synthetic amino 4-quinoline and chemically known as chloroquinolin-4-yl)amino]-2-[(diethyl amino)methyl]phenol. Its activity is characterized by a schizonticidal action on all Plasmodium species. So it is used to treat acute illnesses by destroying intra erythrocytic forms.2

The mechanism of action of Artesunate has been widely studied and appears well established. The Artesunate endoperoxyde's bridge is split by heme within the infected erythrocyte, generating singlet oxygen. Parasite proteins, particularly in membranous structures, are thus alkylated, leading to parasite death.3,4 The amodiaquine, penetrate the infected red blood cells in a specific way and prevent the parasite from polymerizing heme into an insoluble product called hemozoin, leading to parasite death.

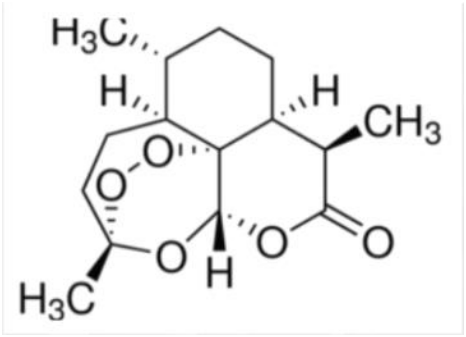

Fig - 1: Chemical structure of Artesunate

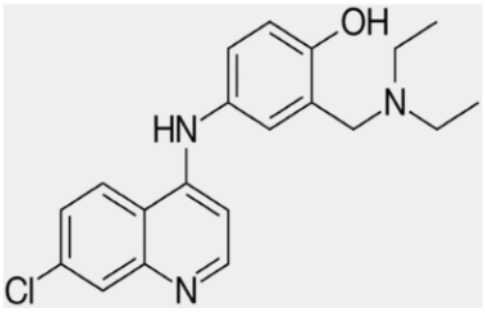

Fig - 2: Chemical structure of Amodiaquine

For Artesunate is official HPLC methods are reported in IP 5 and USP 6. Literature survey reveals that, Spectrophotometric method for Artesunate with curcumin in liposomal formulation 7, HPLC method for the simultaneous determination of Artesunate and mefloquine hydrochloride in fixed dose combination Tablets 8 and simultaneous estimation of Artesunate and Mefloquine hydrochloride in bulk and marketed formulation by UV spectroscopic 9, Stability Indicating Method Development and Validation for Simultaneous Estimation of Mefloquine and Artesunate in Tablet Dosage Form 10 were reported.

Compendial methods for estimation amodiaquine are not available. The literature survey reveals that Stability Indicating HPLC Method for the Determination of Amodiaquine Hydrochloride 11 and UV Spectrophotometric and HPLC Methods for Quantitative Determination of Chloroquine and Amodiaquine in Pharmaceutical Formulations 12 were reported in research articles.

Very few methods reported for the determination of Artesunate and Amodiaquine in combined dosage form by different instrumental techniques are Simultaneous Determination of Artesunate and Amodiaquine in Human Plasma Using LC-MS/MS 13, RP-HPLC Method for Simultaneous Estimation of Artesunate and Amodiaquine in Combined Tablet Dosage Form 14, RP-HPLC Method for Simultaneous Estimation of Artesunate and Amodiaquine HCL in their Combined Pharmaceutical Dosage Form 15 Spectrophotometric method determination of Artesunate and Amodiaquine in combined dosage form16 were reported.

The aim of present work was to develop and validate an accurate, cost effective and precise HPLC method.

2. MATERIAL AND METHODS

Instruments and Apparatus

The instruments were used; HPLC System- Liquid Chromatograph: LC-2010 CHT (Shimadzu), Detector-UV VIS Detector (UV-2487) – Dual Absorbance Detector, YMC C-18 column (150 x 4.6mm, 5µm), UV-Visible Spectrophotometer: Shimadzu UV double beam spectrophotometer 1800, Electronic analytical balance (ME204) and Digital melting Point Apparatus. The calibrated volumetric apparatus were used for preparation and dilutions.

Reagents and Chemicals

API was provided as a gift sample from IPCA Laboratory ltd, Athal, Silvassa. All Chemicals and reagents used were of Analytical Grade and HPLC Grade. Marketed Formulation used for assay was by IPCA LABORATORIES LIMITED having Label Claim 25mg of Artesunate and 67.5mg Amodiaquine.

Standard stock solution of Artesunate and Amodiaquine

Weigh an accurately about 25 mg Artesunate in 100 ml volumetric flask and dissolved with 100 ml of methanol. (250 µg/ml). Weigh an accurately about 65 mg Artesunate in 100 ml volumetric flask and dissolved with 100ml of methanol. (650 µg/ml).

Working standard preparation (Combine standard preparation)

Pipette out 1 ml from Artesunate standard stock solution and 1ml from Amodiaquine standard stock solution in 10 ml with Mobile phase (Potassium Dihydrogen o-Phosphate Buffer (pH-5): Acetonitrile (50:50 v/v)) (ART-25 µg/ml), (AMO-65 µg/ml).

System Suitability Studies

System suitability testing is an integral part of many analytical procedures. The tests are based on the concept that the equipment, electronics, analytical operations and samples to be analyzed constitute an integral system that can be evaluated as such. The system suitability study was evaluated from the standard chromatogram by three replicate injections of Artesunate and Amodiaquine. The %RSD, theoretical plate was calculated for standard solution (ART-25 µg/ml -25µg/ml, AMO -65 µg/ml).

Method validation

The analytical method validation was performed as per ICH guidelines ICH Q2 (R1): Validation of Analytical Procedures: Text and Methodology (2005)

3. LINEARITY AND RANGE

Preparation of calibration curve for Artesunate

The standard stock solution of 250 µg/ml was prepared by dissolving 25 mg of Artesunate in 100 ml methanol. The standard sub-stock solution of concentrations 12.5, 18.75, 25, 31.25 and 37.5 µg/ml were prepared from above standard solution with methanol.

Preparation of calibration curve for Amodiaquine

The standard stock solution of 650 µg/ml was prepared by dissolving 65 mg of Amodiaquine in 100 ml methanol. The standard sub-stock solution of concentrations 32.5, 48.75, 65, 81.25 and 97.5 µg/ml were prepared from above standard solution with methanol.

Each sample injected in triplicate for each concentration level and calibration curve was constructed by plotting the peak area versus the drug concentration.

The HPLC chromatogram and area were shown in Figure 3 and Table 1 respectively.

Table -1: Calibration Data of Artesunate and Amodiaquine

Sr no.	Conc. (µg/ml)		Peak area ± SD (n=3)		%RSD	
	ART	AMO	ART	AMO	ART	AMO
1	12.5	32.5	174.759 ± 1.39	1808.692 ± 14.32	0.79	0.79
2	18.75	48.75	266.284 ± 5.23	2752.823 ± 32.56	1.96	1.18
3	25	65	357.079 ± 6.69	3698.559 ± 25.88	1.87	0.69
4	31.25	81.25	444.168 ± 3.25	4600.067 ± 45.56	0.73	0.99
5	37.5	97.5	534.26 ± 5.20	5533.136 ± 56.17	0.97	1.01

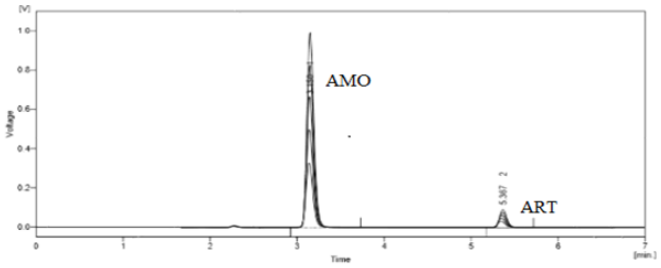

Fig - 3: Chromatogram of Linearity of Artesunate (12.5-37.5 µg/ml) and Amodiaquine (32.5-97.5µg/ml)

Precision

The injection system precision was determined by performing 6 replicate injection for Repeatability and 3 replicate injections for Intra-day and Inter-day Precision. The precision expressed as standard deviation or relative standard deviation.

Repeatability.

The data for repeatability of peak area measurement for Artesunate and Amodiaquine based on six times measurement of same concentration (100µg/ml). The % RSD was found to be 0.19 and 0.12 for Artesunate and Amodiaquine. The HPLC chromatogram was shown in Figure 4.

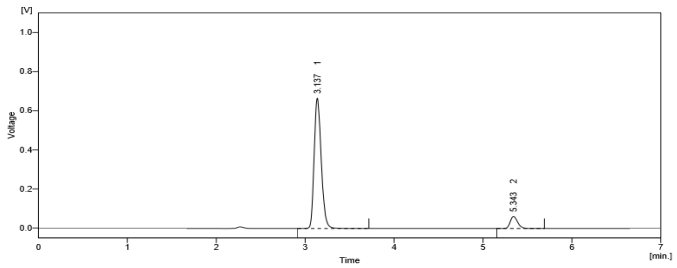

Fig – 4: Chromatogram for Repeatability 1 (100 µg/ml ART and AMO)

Intraday Precision

Artesunate (12.5, 25 and 37.5 µg/ml) and Amodiaquine (32.5, 65, 97.5 µg/ml) were taken in a ratio was analyzed at three levels of mention concentration for three times in a day.

Interday precision

The Artesunate (12.5, 25 and 37.5 µg/ml) and Amodiaquine (32.5, 65, 97.5 µg/ml) were taken in a ratio was analyzed at three levels of mention concentration on three different consecutive days.

The Results of Intraday Precision and Interday Precision for Artesunate and Amodiaquine were shown in Table 2.

Table - 2: Results of Intraday Precision and Interday Precision

		Intraday Precision			Interday Precision		
Name of drug	Conc. (µg/ml)	Area (n=3)	SD (n=3)	%RSD	Area (n=3)	SD (n=3)	%RSD
ART	12.5	179.641	1.93	0.10	173.86	0.35	0.20
	25	354.96	0.69	0.19	355.31	0.71	0.25
	37.5	531.09	1.05	0.20	531.61	1.07	0.20
AMO	32.5	1796.41	1.94	0.11	1797.56	1.86	0.10
	65	3673.61	4.078	0.11	3677.63	4.32	0.11

	97.5	5496.23	6.45	0.12	5501.79	6.42	0.12

Accuracy

Accuracy was done by % Recovery Study. The assay sample solutions were prepared by spiking the API at 3 levels i.e. 80%, 100% and 120%. The Percent Recovery data obtained by the proposed RP-HPLC method. The results were shown in Table 3.

Table -3: Result of Accuracy (%Recovery)

Sr no.	Tablet content Taken eq. to (mg)		Standard Added (mg)		Total Drug Recovered mg		%Recovery of Standard Added	
	ART	**AMO**	**ART**	**AMO**	**ART**	**AMO**	**ART**	**AMO**
Blank	25	67.5	-	-	24.96	67.35	-	-
	25	67.5	-	-	25.00	67.50	-	-
	25	67.5	-	-	24.60	66.90	-	-
	Mean ± SD				24.85 ± 0.22	67.25 ± 0.31		
80%	25	67.5	20	54	44.94	121.50	99.84	99.78
	25	67.5	20	54	49.90	119.56	100	100
	25	67.5	20	54	45.00	120.56	94.4	99.11
	Mean ± SD				35.61 ± 0.21	88.45 ± 0.31	98.93 ± 0.59	98.28 ± 0.34
100%	25	67.5	25	67.5	50.10	131.23	100.86	99.38
	25	67.5	25	67.5	49.96	134.86	99.92	98.92
	25	67.5	25	67.5	51.23	132.66	101.25	102.46
	Mean ± SD				39.67 ± 0.44	101.52 ± 1.21	99.17 ± 1.10	101.52 ±1.21
120%	25	67.5	30	81	54.96	147.44	99.92	99.28
	25	67.5	30	81	55.00	148.97	100.00	100.31
	25	67.5	30	81	55.06	149.02	100.01	100.35
	Mean ± SD				44.81 ± 0.21	108.35 ± 0.70	101.83 ± 0.48	98.50 ± 0.63

Table - 4: Results of Robustness

Sr. No	Parameter	Mean area (n=3)		SD (n=3)		%RSD	
	Potassium dihydrogen o-phosphate buffer: Acetonitrile	**ART**	**AMO**	**ART**	**AMO**	**ART**	**AMO**
1	48:52v/v	349.22	3614.34	2.13	18.32	0.61	0.51
2	52:48 v/v	370.69	3836.70	2.17	18.26	0.58	0.48
	Flow rate						
1	1.2 ml/min	349.22	3614.34	2.13	18.32	0.61	0.51
2	0.8 ml/min	370.69	3836.70	2.17	18.26	0.58	0.48
	pH						
1	5.2	341.40	3533.37	2.12	18.67	0.62	0.53

2	4.8	366.61	3797.60	2.34	17.83	0.64	0.47

Robustness

The Robustness of the method was evaluated by the change in following parameters such as by changing the flow rate: ± 0.2 ml/min, by changing the Mobile Phase: ± 2.0% solvent in Mobile Phase, by changing the pH: ± 0.2. The results were shown in Table 4.

4. LOD AND LOQ

LOD:

Analyte must reliably differentiate from background noise. It is the lowest concentration of analyte in a sample that can be detected but not necessarily quantified.

LOQ:

The Limit of Quantitation is the minimum injected amount that gives precise measurements in chromatography typically requiring peak height 10 times higher than the baseline noise. The results of LOD and LOQ for Artesunate and Amodiaquine was shown in Table 5.

Table - 5: Results of LOD and LOQ

Parameter	Artesunate	Amodiaquine
LOD (µg/ml)	0.32	0.50
LOQ (µg/ml)	0.97	1.51

System Suitability Studies

A standard solution for Artesunate and Amodiaquine was prepared as per the test method and was injected six times into HPLC System.

The system suitability parameters were evaluated from standard chromatogram by calculating %RSD from six replicate injections for Artesunate and Amodiaquine.

Table - 6: System suitability test parameters

Parameter	Artesunate	Amodiaquine
Number of Theoretical Plates (N)	7443	18339
Retention Time	5.3	3.1
Asymmetry	1.318	1.333
Resolution	13.933	0.00

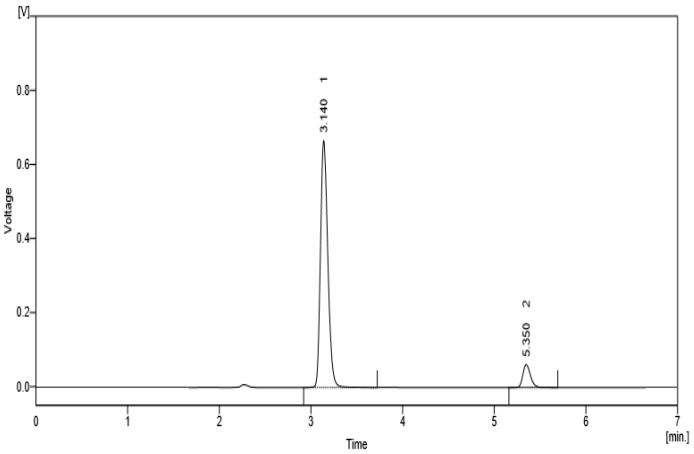

Fig - 5: Chromatogram of Artesunate (25 µg/ml) and Amodiaquine (65 µg/ml) Standard Solution

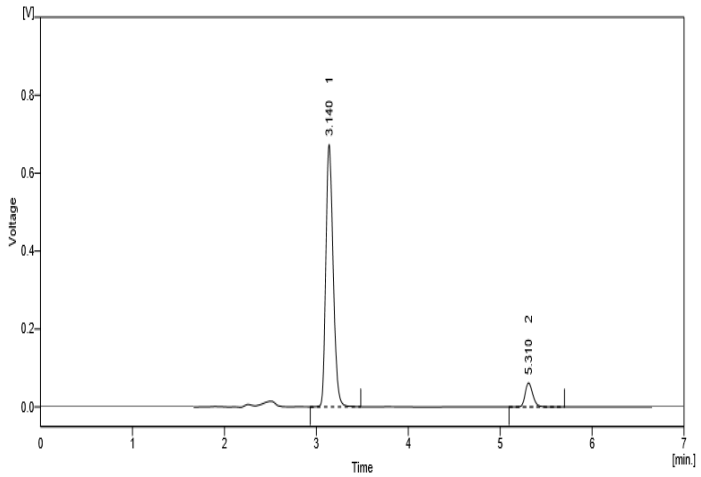

Fig -6: Chromatogram of Artesunate (25 µg/ml) and Amodiaquine (65 µg/ml) Tablet solution

Table - 7: Assay of Artesunate and Amodiaquine

Sr. no	Drug	Label Claim (mg)	Amount found (mg)	Area of samples	%Assay	%ASSAY ± SD	%RSD of assay
1	ART	25	24.46	350.38	98.22	98.42 ± 0.19	0.20
2		25	24.60	351.07	98.42		

3		25	24.65	351.76	98.61		
		Average			98.42		
1	AMO	67.5	67.87	3715.18	100.55	100.70 ± 0.13	0.13
2		67.5	68.01	3722.67	100.75		
3		67.5	68.03	3723.87	100.79		
		Average			100.70		

5. RESULTS AND DISCUSSION

The RP-HPLC method was validated in terms of linearity and range, precision, robustness, LOD, LOQ, assay and accuracy. The develop RP- HPLC method was found to be linear for the range from 12.5-37.5 µg/ml for Artesunate and 32.5-97.5 µg/ml for Amodiaquine. The precision of the method was studied as an intra-day, inter-day variations and repeatability. The % RSD value was found to be less than 2 indicates that the method was precise. The limits of detection were found to be 0.32 and 0.82 µg/ml for Artesunate and Amodiaquine respectively. Limit of Quantitation were found to be 0.50 and 1.51 µg/ml for Artesunate and Amodiaquine respectively. The % accuracy was found to be 99.41± 0.88 % - 100.86±1.39 % and 99.62±0.46 % - 100.86±.1.39% for Artesunate and Amodiaquine respectively. The developed method was found to be selective and specific as the assay results indicate there is no interference of excipients.

6. CONCLUSIONS

The proposed HPLC method was developed and validated according to ICH Guidelines and was found to be precise, accurate and reproducible for the determination of Artesunate and Amodiaquine. The recovery studies revealed excellent accuracy and high precision than previous reported method. The developed selective and validated method can be applied for quality control and routine analysis.

REFERENCES

1] Rang HP., Dale MM., Ritter JN. and Moore PK. In Pharmacology; 5th Edn Churchill Livingstone, New York, 2003, pp 702-709.
2] https://pubchem.ncbi.nlm.nih.gov/compound/amodiaquine
3] www.chemicalland21.com/Lifescience/phar/ARTESUNATE.htm.
4] www.drugbank.ca/drugs/DB00613www.chemicalland21.com/Lifescience/phar/ARTESUNATE.htm.
5] Indian Pharmacopeia; sixth Edn, The Indian Pharmacopoeia commission, Ghaziabad, pp 838-839.
6] The Official Compendia of Standard USP 24NF 19; Asian Edn, Rockville, 2000, pp 126-127.
7] Sharma S, Saraog GK, Kumar V, Development of Spectrophotometric Methods for Simultaneous Determination of Artesunate and Curcumin in Liposomal Formulation, Int. J. App. Pharm., 2015;7:18-21.
8] Fernando H, Andrade N, Fernandes N, Araujo R, Development and Validation of an HPLC method for the simultaneous determination of Artesunate and mefloquine hydrochloride in fixed dose combination Tablets, Braz. J. Pharm. Sci. 2013; 49: 837-843.
9] Akshay D, Sapakal I, Wadkar KA, Mohite SK, Magdum CS, Development and validation of UV Method for simultaneous estimation of Artesunate and Mefloquine hydrochloride in bulk and marketed formulation, Sch. Acad. J. Pharm, 2013; 2(4): 293-297.
10] Rao UM, Ramarao N, Stability Indicating Method Development and Validation for Simultaneous Estimation of Mefloquine and Artesunate in Tablet Dosage Form, Sch. Acad. J. Pharm, 2014; 3: 411-417.
11] Lakhani SN, Desolate UA, Natalee RB, A Stability Indicating HPLC Method for the Determination of Amodiaquine Hydrochloride, Int. J. Pharm. Sci. Rev. Res, 2014; 28: 196-199.
12] Lawal A, Umar RA, Development and Validation of UV Spectrophotometric and HPLC Methods for Quantitative Determination of Chloroquine and Amodiaquine in Pharmaceutical Formulations, Int. J. Chem. Res, 2012; 4: 669-676.
13] Maddela R, Pilli NR, Simultaneous Determination of Artesunate and Amodiaquine in Human Plasma Using LC-MS/MS and Its Application to a Pharmacokinetic Study, Int. J. Ph. Pharm. Sci, 2015; 7: 105- 112.
14] Gandhi S, Deshpande P, Jagdale P, A Simple and Sensitive RP-HPLC Method for Simultaneous Estimation of Artesunate and Amodiaquine in Combined Tablet Dosage Form, J. Chem. Pharm. Res, 2010; 2: 429-434.
15] Odedara MH, Faldu SD, Dadhania KP, RP-HPLC Method for Simultaneous Estimation of Artesunate and Amodiaquine HCL in their Combined Pharmaceutical Dosage Form. J. Pharm. Sci. Biosci. Res. 2012; 2: 114-117.
16] Kushwaha R, Dedania ZD, Dedania RD, Padsala J, Mauraya J, Spectrophotometric method determination of Artesunate and Amodiaquine in combined dosage form, International Journal of Research and Analytical Reviews, 2018; 4(5): 781-786
17] ICH Harmonized Tripartite Guideline. Validation of Analytical Procedures: Text and Methodology Q2 (R1), International Conference on Harmonization, Geneva: Switzerland; 2005. 1-13.

A Study on Regulatory Requirement for Development of Cardiac Pacemaker In USA

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Abstract: *Cardiac pacing is an emerging lifesaving procedure that is being widely used in the recent times. Therefore, it is considered vital for the healthcare professionals to be aware of patients' knowledge and experience after the cardiac device implantation and also the impact these implanted devices have on their day-to-day life. This study was conducted with an aim to assess the knowledge and attitude of patients regarding permanent pacemakers (PMs) and their quality of life (QOL) after the permanent PM implantation. A descriptive cross-sectional study design was used in this study. A total of seventy patients were chosen by total enumerative sampling technique among those patients attending the cardiology outpatient department, PM clinic and selected cardiology wards of a tertiary care centre in South India. A significant association between attitude and age was found. Conscious effort must be taken to help patients cope better and experience good QOL through systematic teaching after the PM implantation. This will help patients to function maximally and live life to their best capacities in the family and society.*

Key Words: Attitude, knowledge, permanent pacemaker, quality

Received Date: 02/08/2023 Revised Date: 26/10/2023 Acceptance Date: 20/11/2023

1. INTRODUCTION OF CARDIAC PACEMAKER[1-4]

The true beginning of the concept of a pacemaker began over 200 years ago. In the late 1700s, Luigi Galvani discovered that he could cause contraction of a frog heart simply by passing an electrical current through the heart. This concept was further realized nearly 100 years later with the first successful resuscitation of a child by Gilliam de Boulogne utilizing electricity. He was able to accomplish this by introducing an electrical current to the patient's chest with a return electrode on the leg after a drowning. After this feat, many successful resuscitations were reported, leading to the term "artificial cardiac pacemaker" by Dr. Hyman in 1932.

Pacemakers are adjustable artificial electrical pulse generators, frequently emitting a pulse with a duration between 0.5 and 25 milliseconds with an output of 0.1 to 15 volts, at a frequency up to 300 times per minute. The cardiologist or pacemaker technologist will be able to interrogate and control the pacing rate, the pulse width, and the voltage, whether the device is temporary or permanent. Pacemakers are typically categorized as external or internal. The external variety is almost always placed for temporary stabilization of the patient or to facilitate some type of surgical procedure and the implantable type is usually permanent and often, significantly more complex than the temporary, external variety.

Pacemakers are one type of cardiac implantable electronic devices (known as CIED). The first implantable ICD was developed in 1980, and since that time, it has become more difficult to differentiate between pacemakers and ICDs. This is because every ICD currently implanted has an anti-bradycardia pacing function. It is critical for the patient and any health care provider to understand which device has been implanted to prevent unnecessary ICD therapy. This is most likely to occur with any electromagnetic interference (EMI) and could lead to activation of the device. Most types of CIED use several insulated lead wires with non-insulated tips that are implanted in the heart, either by percutaneous vein insertion or directly by a cardiac surgeon. Cardiac pacemakers are made up of two parts: the pulse generator and the leads or electrodes. The North American Society of Pacing and Electrophysiology (NASPE) and the British Pacing and Electrophysiology Group (BPEG) jointly developed a generic pacemaker code, utilized worldwide, that would allow provider and manufacturers to describe the characteristics of the device, and this was last updated in 2002 and is shown below in the Pacemaker Table. The first letter in the code indicated which chamber is paced; the second letter indicates which chamber is being sensed by the device; the third letter indicates if there is a response to sensing; the fourth position indicates whether the device will modulate or change the programmed rate independent of the patient's cardiac activity, the fifth and last letter of the

code indicates additional multisite pacing. The last two letters of the code (in the fourth and fifth position) are rarely used in typical nomenclature. The simplest settings are AAI and VVI. The AAI mode paces and senses in the atrium and each sensed event triggers the generator to fire

within the P wave and the VVI mode paces and senses the ventricle and is suppressed by a sensed ventricular event.

2. REGULATORY FRAMEWORK IN USA

2.1. Applicable Regulation and Guideline[5]

There are two important aspects of long-term care of patients with implantable pacemakers. Clinical follow-up involves complete evaluation of the patient as an individual and is generally the responsibility of the referring physician. Technical follow-up, concerned with pacemaker function and detection of impending pacemaker failure, should be the responsibility of a physician with extensive pacemaker experience. Follow-up surveillance should include:

- A program for pacemaker replacement and a system for its implementation.
- telephone number of the responsible physician. Complete instructions for every patient, preferably written. They should include information about suitable physical and social activities, the frequency and nature of postoperative visits, and a procedure for obtaining medical care in emergency situations. The patient should be given an identification card to carry with him at all times; it should show, as a minimum, the date of implantation and the type of pulse generator and electrode used, and the name, address and
- Regularly scheduled follow-up appointments. The frequency with which a patient receives follow-up examinations will depend upon the type of surveillance system utilized, the complication rate and anticipated longevity of the specific pacemaker used, the age and medical condition of the patient and other variable social and geographic factors. Optimally patients should be seen within one month after discharge from the hospital to detect early problems, at least three times during the first year and approximately every two months thereafter until one approach the anticipated end-of-life of the pacemaker. With standard output units, this will be 24 months and with low output pacemakers, 36 months. Approximately six months before anticipated end of-life, the patient should begin to receive closer surveillance either with monthly visits, weekly trans telephone monitoring, or a suitable combination of both.
- Record of pacemaker function. The important parameters that have been selected for measurement of pacemaker function should be recorded in an easily retrievable form so that all individuals responsible for follow-up have ready access to the information.

2.2. Regulation that Strikes and right Balance5

A variety of reform options are available to policy makers to strengthen and streamline FDA's approval process and improve oversight and safety of implantable devices. As previously discussed, without endorsing or ranking them, these options include the following:

- Strengthen the premarket approval process for the riskiest implantable devices.
- Strengthen the market clearance process for devices of moderate risk through increased use of the de novo approach.
- Eliminate "grandfathered" market clearance for implantable devices and require testing of devices that were in use prior to 1976.
- Prohibit recalled devices from serving as predicate devices—that is, older devices that have been recalled should not serve as the basis for clearance of newer implantable devices.
- Impose limits on the time that a device can serve as a predicate device.
- Strengthen post market oversight and reporting for implantable devices through the use of more post market surveillance studies, innovative monitoring techniques, and additional funding for these activities.
- Make better use of implantable device patient registries.
- Expand use of unique device identifiers.
- Improve communication with stakeholders.
- Strengthen quality controls by giving FDA authority to conduct premarket inspection of all facilities that make implantable devices.
- Strengthen FDA enforcement activities through improved targeting of recalls and other actions.

Implantable devices can and do save lives. They improve the quality of life for millions of Americans. Sometimes, they fail. When this happens, people can sustain serious injury or death. Careful regulation and oversight are essential to ensure the safety and effectiveness of these devices both before and after they reach the market. Regulatory oversight needs to safeguard patients while still encouraging innovation that makes implants safer, more effective, and more affordable.

3. STATUS OF CARDIAC PACEMAKER

3.1. Status of Nomenclature for Implantable Cardiac Pacemakers6

When there was only one type of pacemaker, no matter what it was called, everyone knew it was a device that discharged at a fixed rate. To obviate any such confusion in this review, we have designed the following nomenclature code to identify the mode of operation of the pulse generator.

Table 1: Three letter Identification Code

1st Letter	2nd letter	3rd letter
Chamber Paced	Chamber Sensed	Mode of Response

V – VENTRICLE	A - ATRIUM
I– INHIBITED	T - TRIGGERED
D - DOUBLE CHAMBER	O - NOT APPLICABLE

First letter: The paced chamber is identified by V for ventricle, A for atrium or D for double both atrium and ventricle.

Second letter: The sensed chamber, if either, is again V for ventricle, A for atrium.

Third letter: The mode of response, if any, is either:

I for inhibited, a pacemaker whose output is blocked by a sensed signal, or

T for triggered a unit whose output is discharged by a sensed signal.

The letter "O" indicates that a specific comment is not applicable.

3.2. Electrodes, Leads and Connectors[6]

By definition, the electrode is the uninsulated portion of a lead, anode or cathode, in direct electrical contact with the body. In unipolar electrode systems only one electrode, the cathode, is in the heart. The other, an "indifferent" electrode remote from the heart, is a large metal plate often the external capsule of the pulse generator. In a bipolar electrode system both electrodes, cathode and anode, lie against or near responsive myocardial tissue. The electrode may take the form of a hemisphere, cylindrical ring, coil, or a screw-in wire.

The most popular electrode materials are platinum iridium and a cobalt-nickel alloy, chosen because of their resistance to corrosion, satisfactory electrical conductivity, compatibility with body tissues, and limited polarization effects. New electrode materials should be compared to these as standards.

The electrode is in continuity with the metallic lead wire from the pulse generator. Sometimes it is merely the bare end of the wire; in most cases, however, it is a separate metallic element joined to the lead wire. This junction must be strong, permanent, and of a similar metal so that corrosion will not occur at the connection. The most common lead wire configuration is the helical coil in which no one point will be subjected to excessive flexion and thus to ultimate fracture. Wires are typically composed of steel, cobalt-nickel alloys or platinum-iridium, with the last somewhat less resistant to flexion fatigue. Despite the sophisticated design, wire fractures continue to occur at a rate of 1% to 2% a year. With the development of long-term power sources efforts will have to be made to develop still better leads. Manufacturers should provide information regarding the electrical resistance of the lead system, its dimensions, materials, electrode surface area, and anticipated threshold of stimulation.

Unipolar electrodes are smaller in diameter than bipolar, have superior R wave sensing, are easier to repair, and may necessitate less surgery during pulse generator replacement. Bipolar electrodes, on the other hand, are less sensitive to interfering signals and in some cases if one lead fractures, may be converted to a unipolar system. Both types are used widely and it is impossible at this time to recommend one over the other.

When pacemakers were first developed, one of the principal considerations was that electrodes should be large enough to prevent corrosion by high current density. It soon became evident that there was unequally strong argument for the use of small electrodes. The threshold of stimulation of the heart is directly related to the effective electrode size; the smaller the electrode surface area the lower the stimulation threshold. Lower stimulation thresholds require less pulse generator output and less current drain from the battery, so battery life can be prolonged by smaller electrodes. Earlier fears that the small electrode system would corrode have not been realized because of the use of reduced current output pulse generators, and because appropriate materials have been found with low corrosion effects. Most manufacturers now provide pacemakers with small area electrodes, and these are to be recommended. Although the electrodes could be made smaller still, it remains to be demonstrated that further gains can be made in increasing the life of conventional batteries. The fear that small electrodes would be associated with a high incidence of perforation has not been borne out by practical experience.

Table 2: Suggested Nomenclature code for Implantable Cardiac Pacemakers

Chamber paced	Chamber sensed	Mode of response	Generic description	Previously used designation
V	O	O	Ventricular pacing; no sensing function	Asynchronous; fixed rate; set rate
A	O	O	Atrial pacing; no sensing function	Atrial fixed rate; atrial asynchronous
D	O	O	Atrioventricular pacing; no sensing function	AV sequential fixed rate
V	V	I	Ventricular pacing and sensing, inhibited mode	Ventricular inhibited; R inhibited ; R blocking ; R suppressed; non-competitive inhibited
V	V	T	Ventricular pacing and sensing; triggered mode	Ventricular triggered; R triggered; R wave stimulated; non-competitive triggered

A	A	I	Atrial pacing and sensing; inhibited mode	Atrial inhibited; P inhibited; P blocking
A	A	T	Atrial pacing and sensing; triggered mode	Atrial triggered; P triggered; P stimulated; P synchronous
V	A	T	Ventricular pacing: atrial sensing, triggered mode	Atrial Synchronous, atrial synchronized, AV synchronous
D	V	I	Atrioventricular pacing, ventricular sensing, inhibited mode	Bifocal sequential demand, AV sequential

The lead system is attached to the pulse generator by a connector or "plug-in." Connection and disconnection are made by physical compression or by various types of screw arrangements and all connectors currently available are physically and electrically secure when properly used. Because corrosion at the connector can occur if dissimilar metals come in contact with each other, materials of the connecting elements should be identical. Manufacturers have not agreed upon a universal connector system but when one pacemaker model is exchanged for another, they will provide adapter kits to accommodate the pulse generator of one model to the lead wire of another; there should also be a description of how to convert a bipolar to a unipolar system.

Problems with both connection and disconnection have occurred. In connecting the pulse generator and the lead wire, lack of adequate proof of a secure contact has led to faulty connection and later disruption of the elements. Freezing of the connection by metallic corrosion, mechanical jamming, and silicone cement at times have made disconnection difficult or impossible. Further work in design of connectors is necessary. The connector should provide positive indication of proper contact, the metallic elements should be protected from tissue fluids, and disconnection should be accomplished by a simple manoeuvre that will not require destruction of any of the components. Although a universal connector for all pacemakers would be a highly desirable feature, this cannot be recommended strongly at this time because it would be restrictive in the development of new designs and devices.

3.3. Pulse Generator[6, 7]

Pulse generators are 100 to 200 gm. discoid or rectangular packages with rounded edges. Weight and size are largely dependent on the power source, which usually consists of a battery of four or five individual mercury zinc cells. The battery and the electronic components are usually potted in epoxy resin which provides protection against component movement and, to some extent, against body fluids. The outer surface of the epoxy pot may be the surface of the pulse generator. Alternately the entire unit may be covered with silicone rubber or by a titanium or stainless steel case. The metallic coverings are designed to provide protection against electromagnetic interference (EMI) and, if hermetically sealed, additional protection against the body environment. Although it has always seemed desirable to encase the pulse generator or at least its circuitry in a hermetically sealed case, this has only recently become practical. All currently available pulse generators are well accepted by the body, and component materials, including the anode of the pacemaker system, are no allergenic, non-toxic, and non-carcinogenic. The ideal implant would be a flat package, with rounded smooth edges, as small as possible and with a specific gravity the same as body tissues.

3.4. Pacemaker Output[6]

At first the electrical output of the pacemaker was established empirically. The upper limit was determined in part by pacemaker size which, in turn, was related to the number of cells in the batteries. The lower limit of output was not known. The moder pulse generator typically contains four or five mercury zinc cells in series or series-parallel configuration. In an attempt to increase longevity and reduce size, the standard pulse generator output has been reduced to a level somewhat closer to the excitation threshold of the heart, leaving acceptable margins for threshold rise after the first few weeks of implantation and for the diminishing voltage toward the end of battery life. The stimulating rate also is voltage dependent. Thus, as the battery wears out and voltage drops, the output of the pacemaker is still sufficient to stimulate the heart, but its rate gradually decreases. A sudden rate change of several beats per minute may be a signal for replacement.

The output pulse varies in configuration and duration from model to model, but in general it is a rectangular "monophasic" waveform 0.5 to 1.7 ms. in duration.

The rate of implantable pacemakers is set by the manufacturer at approximately 70 beats per minute. Some pacemakers have rate adjustment controls. One such control is activated by turning a potentiometer in the pacemaker with a special percutaneous needle. Another delivers bursts of magnetic impulses over the pacemaker which activate an internal decoder that adjusts the rate, or the output, or both, to one of a number of predetermined settings.

Most pulse generators consist of a stimulating circuit and a sensing circuit, both of which draw current from the battery. In the presence of complete heart block, an asynchronous pulse generator (VOO) with only a stimulating circuit may be used. As circuit efficiency of the non-competitive triggered or inhibitory (VVT or VVI) pulse generators increases, the need for an asynchronous (VOO) unit decreases.

With available equipment pulse generator life can be prolonged by the use of:

- Small surface area electrodes
- Variable output (voltage, current and pulse duration) pulse generators, adjustable to the level required to pace the heart.
- Asynchronous (VOO) pulse generators where suitable
- Reduced output pulse generators for replacement of old units

- Replacement of pulse generators just before exhaustion rather than at an elective predetermined interval.

4. NONCLINICAL TESTING[8]

The following series is intended to identify issues that need to be addressed to qualify a "new" pacemaker lead and to identify some of the non-clinical tests which may be used to support a pacemaker lead submission. Sponsors should examine this listing to determine testing appropriate for their device. Since new lead designs may experience failure modes not previously seen, this guidance document may not reflect the complete battery of non-clinical testing necessary to qualify all pacing leads/designs. It's the responsibility of the lead manufacturer to define a comprehensive testing methodology for a particular lead design.

4.1. Biocompatibility

Biocompatibility evaluation depends, in part, on the full characterization of all sterilized device materials in contact with tissue and/or body fluids. In order to accurately identify these materials, the material specifications from the manufacturer, qualitative and quantitative information concerning all constituent materials used in the manufacturing of the lead should be provided. All protocols, test results and identification of control materials should be provided in order that an independent evaluation of the study conclusions can be made. Protocols do not need to be submitted if standard methods are utilized (e.g., USP methods) and complete references for the methods are provided.

Biocompatibility testing may not be necessary if a material has a long history of use in currently marketed pacemaker leads. If there is sufficient knowledge about the biocompatibility/toxicity of every constituent of the lead, then it need not be subjected to further biocompatibility tests. It is incumbent upon the device submitter to provide sufficient evidence to establish that further biocompatibility testing is not necessary. A sponsor may submit information and data available in publications or from other legitimate sources which show that the material is non-toxic in tests identical to or equivalent to the biological tests listed below. Any changes in formulation, manufacturing or processing between the tested and submitted products which might affect biocompatibility should be identified.

The effects of sterilization on device materials and potential leachable, as well as toxic by-products resulting from sterilization should be considered when conducting biocompatibility tests. Therefore, testing should be conducted on the sterilized final product and any leachable material from the sterilized final product. The exact chemical analysis of device extracts may be omitted if the extracts are subject to toxicity testing. But, as stated above, the qualitative and quantitative description of all constituent materials in the device before extraction should be provided and the material specifications for the device should be comprehensive.

4.2. Animal Studies

The purpose of animal studies is to assess the structural integrity, biostability, electrical performance, biocompatibility, handling characteristics and/or mechanical performance of the fully assembled lead. Animal studies should be designed to closely approximate the intended use of the device in humans. Electrical data should consist of measurement of the following parameters:

- R and P wave amplitudes at implant and at appropriate intervals following implant
- Voltage stimulation threshold at a 0.5 ms pulse width at implant and at appropriate intervals following implant.
- Strength duration (pulse width versus stimulation threshold)
- Pacing impedance at implant and at appropriate intervals following implant.

4.3. Bench Testing

Electrical and mechanical tests should be conducted on components, subassemblies and/or finished leads, as appropriate. All tests should be performed on leads fabricated by representative manufacturing processes and subjected to the final validated sterilization procedures intended for the device. If test samples are subjected to either no sterilization or other sterilization procedures, the rationale for the procedure used should be supplied.

An adequate number of samples should be tested. If sample devices of different lead models are tested, it should be clearly indicated which models were used for each test.

Testing of leads or subassemblies should be performed after sterilization. Testing should include, but not necessarily be limited to the following, as appropriate:

1. Verify the electrical continuity of each conduction path by measuring the DC resistance.
2. Measure leakage current during voltage application (before drying, after soaking).
3. Determine the strength of each bond, joint, etc, in the lead (lower 95 percent confidence bound) as well as the composite lead strength. Leads should be subjected to a tensile test which simulates the stress it may experience during the implant procedure as well as after implant. Before pull testing, the lead should be soaked in saline for 10 days to simulate any effects of body fluids on the lead body.
4. For leads that are hermetically sealed at the distal end, verify that the lead is leak-proof when immersed in isotonic saline at 37°C under physiological pressure for a minimum period of ten days.
5. Document the corrosion resistance of all conductors and electrode materials in the condition of the finished lead. Address current pulsing when appropriate.
6. Evaluate the performance of the sty let intended to be used during lead placement. Measure the sty let insertion and removal forces.
7. Fatigue resistance of the conductor(s) should be verified. Intact leads should be used for this testing. Loading conditions that are utilized should be able to be extrapolated to worst-case physiological conditions, for example, stresses and ranges of motions etc. Different areas of the lead are subjected to different stresses; this

factor should be taken into consideration in the design of an appropriate test protocol. Test methods designed to accelerate fatigue of conductors should be shown to be able to produce characteristic fracture morphologies that may have been documented previously in vivo.

8. Connectors intended to be used for joining pulse generators and leads should withstand the mechanical forces that might occur after implantation. Generally, most lead connectors are designed to comply with ISO 5841-3 (IS-1). This standard outlines the appropriate testing for lead connectors. If the connector is labelled as "IS-1" compatible, it should meet all ISO 5841-3 testing and dimensional requirements.

9. Evaluate the performance of the anchoring sleeve packaged with the lead. Testing should assure that the lead will be held securely in place and not damage the lead body when the anchoring sleeve is sutured according to the Instructions for Use.

10. Measure the pressure exerted by lead tip and express in units of pressure.

5. CLINICAL TESTING8

If the design of the lead is novel enough or new indications/claims are being sought for the lead, a clinical trial may be needed. Examples where clinical data may be appropriate include:

- Incorporation of an electrode that has not been approved for use on another lead body.
- Changes to a marketed lead which might alter the handling characteristics.
- Change in indication from atrial to ventricular pacing.

The success of a clinical trial is based on the overall coordination of three steps: the design of the study; the conduct of the study; and the analysis of the results. Each step of the initial overall study plan should be executed and carefully considered by the sponsor. The clinical study should be ultimately capable of demonstrating the effectiveness and safety of the device in terms of:

- Prescribed, recommended, suggested and other conditions of use in the labelling or advertising.
- Probable benefit to health weighed against any probable injury or illness.
- Reliability of the device (see 21 CFR 860.7(b))
- Intended patient population.

5.1. Clinical Study Design

A detailed protocol for a clinical trial should include:

1. A well-defined, clear question (hypothesis) or set of questions that are to be answered about the lead by the clinical study.

2. A statement of the study type, i.e., randomized, case control, concurrent control etc. In all cases, the data intended to be used as a control should be identified and comparability discussed with respect to critical study variables including indications, outcome variables, definitions, baseline characteristics, inclusion/exclusion criteria.

3. A sample size of all study groups calculated to demonstrate that a sufficient number of patients will be enrolled to adequately address the study hypotheses. Sample size is primarily a function of the pre-determined level of significance and the power of the study to detect a treatment effect of a predetermined magnitude. As a general rule should not be greater than 0.05 and b should not be greater than 0.20. Any deviation from this range of values should be clearly justified. The greater the difference to be detected between treatment and control groups in the study, the lower the number of subjects needed, provided the a and b remain unchanged. It is imperative that the sponsor seek the assistance of a statistician familiar with clinical trial methodology in order to develop the protocol and determine the appropriate number of subjects to be enrolled in the study.

4. A specification of the outcome variables or clinically relevant endpoints that will be measured to support the study hypotheses. The measure of each primary endpoint should be objective and concisely defined.

5. A description of the means to eliminate selection bias should be included in the protocol. Sequential screening of all potential subjects for the study, with a record of the patients not enrolled and the reason for non-enrollment is one way of avoiding selection bias.

6. A specification of all baseline and follow-up assessments consistent with the study objectives. Follow-up assessments should include the allowable time window.

5.2. Study endpoints

Endpoints commonly used for the evaluation of permanent pacing leads include the following:

1. Effectiveness

- Battery longevity
- Pacing impedances
- Voltage stimulation threshold
- Sensing characteristics

2. Safety

Lead related adverse events (observations and complications). The following should be addressed regarding complications and observations:

- Observations are lead-related adverse events which are corrected by non-invasive measures, e.g., reprogramming.
- Complications are lead-related adverse events that are corrected using invasive measures to correct or which result in the loss of a significant device function, e.g., lead dislodgment; and
- deaths, all deaths and lead-related deaths

5.3. Criteria for Lead – Related Complication and Failures

WHEN: The following condition occurs:

- Insulation Breach
- Dislodgement
- Pacing impedance less than 200 ohms (describe how impedance was measured)
- Loss of capture
- Perforation
- Conductor failure
- Extra cardiac Stimulation
- Pacing impedance greater than 3000 ohms or beyond the measuring capabilities of the device (describe how impedance was measured)
- Over sensing
- Loss of sensing/Under sensing AND: The condition was not:
- Corrected by reprogramming of the pulse generator (except for reprogramming of mode or polarity)
- Caused by a pulse generator malfunction.

THEN: The occurrence should be reported along with the following interventions/interactions in which the lead was:

- Abandoned Surgically
- Abandoned Electrically
- Modified Surgically
- Modified Electrically
- Tolerated (based on medical judgment)
- Removed/Explanted (full of partial)

6. THE EVOLUTION OF PACEMAKERS[9-20]

6.1. Excitation and Conduction System[9-13]

The heart is composed of atrial and ventricle muscles that make up the myocardium and specialized fibres that can be subdivided into excitation and conduction fibres. Once electrical activation is initiated contraction of the muscle follows. An orderly sequence of activation of the cardiac muscle in a regularly timed manner is critical for the optimal functioning of the heart. The excitation and conduction system responsible for the control of the regular pumping of the heart is presented in Figure 1. It consists of the intermodal tracks, the bundle if HIS, Sino atrial Node (SA), Bachmann's bundle, bundle branches, the Atria Ventricular (AV) node, Purkinje Fibres. Cardiac cells are able to depolarize at a rate specific for the cell type. The intrinsic rate of AV-nodal cells is about 50 beats per minute (bpm), whereas Purkinje fibres depolarize at a rate of no more than 40 bpm. During normal sinus rhythm, the heart is controlled by the SA node having the highest intrinsic rate of 60–100 bpm depending on the hemodynamic demand. The right atrial intermodal tracks and Bachmann's bundle conduct the SA-nodal activation throughout the atria, initiating a coordinated contraction of the atrial walls. The impulse reaches the AV node, which is the only electrical connection between atria and ventricles. The AV node introduces an effective delay, allowing the contraction of the atria to complete before ventricular contraction is initiated. Due to this delay, an optimal ventricular filling is achieved. Subsequently, the electrical impulse is conducted at a high velocity by the His- Purkinje system comprising the bundle of His, Purkinje fibres and bundle branches. Once the bundle of His is activated, the impulse splits into the right bundle branch, which leads to the right ventricle and the left bundle branch serving the left ventricle. Both bundle branches terminate in Purkinje fibres so The Purkinje fibres are responsible for spreading the excitation throughout the two ventricles, enabling a coordinated and massive contraction.

6.2. Cardiac Signals[9, 16]

Surface Electrocardiogram

The electrocardiogram (ECG) is a recording from the body surface of the electrical activity generated by the heart. In 1899, the ECG was originally observed by Waller. In 1903, Einthoven introduced electrophysiological concepts still in use today, including the labelling of the waves characterizing the ECG. He assigned the letters P through U to the waves avoiding conflicts with other physiologic waves studied at that time Figure 2 depicts a typical ECG signal.

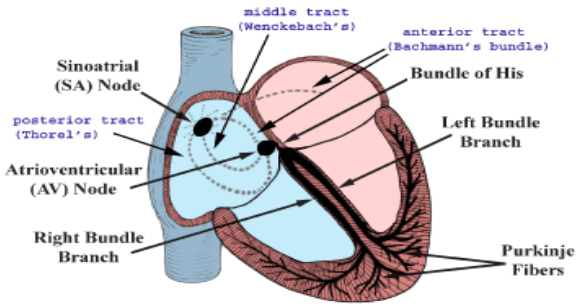

Fig - 1: The cardiac conduction system

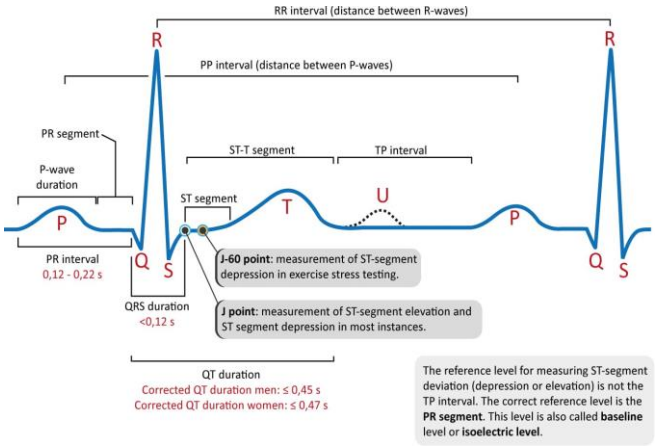

Fig - 2: Typical electrocardiogram

6.3. The History and Development of Cardiac Pacing

Artificial Pacemakers[9, 18]

An artificial pacemaker is a device that delivers a controlled, rhythmic electric stimulus to the heart muscle in order to maintain an effective cardiac rhythm for long periods of time.

ensuring effective hemodynamic performance. The indication for implanting a permanent pacemaker and selection of the appropriate mode of operation are mainly based on the type of cardiac disease involved such as failure of impulse formation (sick-sinus syndrome) and/or impulse conduction (AV-block).

Functionally, a pacemaker comprises at least three parts: an electrical pulse generator, a power source (battery), and an electrode (lead) system.

Different types of output pulses (e.g., monophasic, and biphasic) can be used to stimulate the heart. The output stimulus provided by the pulse generator is the amount of electrical charge transferred during the stimulus. For effective pacing, the output pulse should have an appropriate width and sufficient energy to depolarize the myocardial cells close to the electrode. Generally, a pacemaker can provide a stimulus in both chambers of the heart. During AV block, ventricular pacing is required because the seat of disease is in the AV node or His-Purkinje system. However, in case of a sick sinus syndrome, the choice of pacemaker will be one that will stimulate the right atrium.

A pacemaker utilizes the energy stored in batteries to stimulate the heart. Pacing is the most significant drain on the pulse generator power source. The battery capacity is commonly measured in units of charge (ampere hours). Many factors will affect the longevity of the battery, including primary device settings like pulse amplitude and duration and pacing rate. An ideal pulse generator battery should have a high energy density, low self-discharge rate, and sufficient energy reserve between early signs of depletion and full depletion to allow for safe replacement of the device.

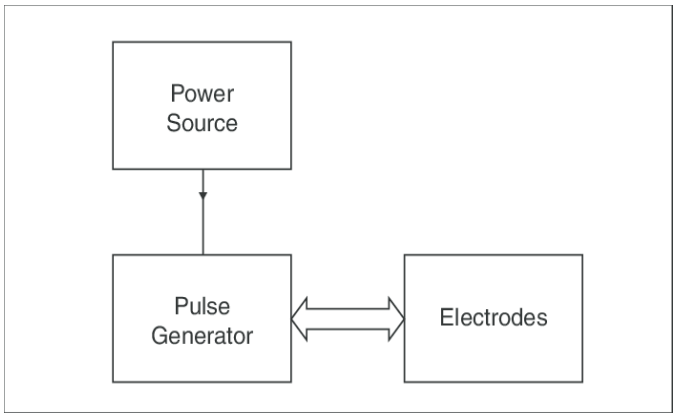

Fig - 3: Basic pacemaker functional block diagram

6.4. Hyman's Pacemaker[9]

In the early 20th century, many experiments such as drug therapy and electrical cardiac pacing had been conducted for recovery from cardiac arrest. Initial methods employed in electrically stimulating the heart were performed by applying a current that would cause contraction of the muscle tissue of the heart. Albert S. Hyman stated that the introduced electric impulse serves no other purpose than to provide a controllable irritable point from which a wave of excitation may arise normally and sweep over the heart along its accustomed pathways.

6.5. Demand Pacemaker[9, 19]

These pacemakers, called asynchronous or fixed-rate pacemakers compete with the natural heart activity and can sometimes even induce arrhythmias or Ventricular Fibrillation (VF). By adding a sensing amplifier to the asynchronous pacemaker in order to detect intrinsic heart activity and thus avoid this competition, one obtains a demand pacemaker, and it provides electrical heart-stimulating impulses only in the absence of natural heartbeats. The other advantage of the demand pacemaker compared to the fixed rate system is that now the battery life of the system is prolonged because it is only activated when pacing stimuli are needed.

6.6. Dual-Chamber Pacemaker[9, 22]

A dual-chamber pacemaker typically requires two pacing leads: one placed in the right atrium and the other placed in the right ventricle. A dual-chamber pacemaker monitors (senses) electrical activity in the atrium and/or the ventricle to see if pacing is needed. When pacing is needed, the pacing pulses of the atrium and/or ventricle are timed so that they mimic the heart's natural way of pumping. Dual-chamber pacemakers were introduced in the 1970s. One of the first descriptions of a dual-chamber pacemaker was given by Berkovits in 1971 and Berkovits announced a bifocal (AV sequential) pacer that sensed only in the ventricle but paced both chambers. In the presence of atrial standstill or sinus-node syndrome plus AV block, the bifocal pacemaker could deliver a stimulus to the atrium and then, after an appropriate interval, to the ventricle. In accordance with the principles of the demand pacemaker design, a sense amplifier is provided to detect intrinsic ventricular activity. The timing control circuits determine both atrial and ventricular time-out stimulating period. However, the atrial-stimulating impulse is generated first and after a predetermined time interval (200 Ms), the ventricular-stimulating impulse is generated. Three electrodes are provided: a neutral electrode, an electrode for atrial stimulation and an electrode for ventricular pacing and sensing. The Field-Effect Transistor (FET) Switch (S FET) is inserted in the feedback path of the ventricular electrode in order to avoid erroneous detection because of the atrial contraction. The S FET is normally conducting. The negative pulse generated at the atrial electrode is transmitted through the diode Da, charging the capacitor Ca, and turning off the switch. When the atrial stimulating terminates, Ca discharges through resistor Ra and turns on the switch again. In this manner, the sense amplifier is disabled during each atrial stimulation and for a short interval thereafter.

7. IMPLANTABLE PACEMAKERS TESTING GUIDELINES[21]

This guideline describes a general framework for design verification testing of a safe and effective implantable cardiac pulse generator. The tests are designed to

reasonably assure safe and effective functioning of the pacemaker in the patient, according to written specifications of performance, and its survival under expected environmental conditions in the body and during storage, shipping, and handling.

This guideline is intended to apply to bradycardia pacemakers which are to be commercially marketed and are manufactured using standard production techniques and methods. It may not apply to devices which are used in limited research applications.

The testing is that referred to in the premarket approval regulation 21 CFR Part 814 and must be reported as described in 21 CFR Part 814 articles 814.20(b) (3) (v) and 814.2020(b) (6). The testing requirements include any and a1 l additional requirements imposed or referred to in the "Good Manufacturing Practice for Medical Devices: General" Regulation (21 CFR Part 820). This guideline represents practices which have been developed over several years and are generally understood by the pacemaker industry.

The tests are grouped into (A) in vitro component tests,(B) in vitro device tests(C)animal tests, (D)biocompatibility tests,(E)clinical investigation and (F) Manufacturing. An appendix provides a sample protocol for preclinical pacemaker testing.

7.1. In vitro component tests:

Component parts shall be tested for design verification by the pulse generator manufacturer of its supplier according to written specifications of performance and testing (such as Military Standard=. or their equivalent).

The component parts referred to shall include, but not be limited to:

Hybrid Integrated Circuit and/or Chip Carrier
Battery
Connector
Other components where necessary to assure reliable operation.

7.2. In vitro finished Device Testing:

The following testing shall be performed:

Electrical Characterization

This testing shall be designed to verify the proper functioning of the pulse generator within specified tolerances in the human body during the device's expected operational life. All parameters such as rate, pulse width, sensitivity, and timing cycles and periods; and all features such as intra-cardiac, electro-grams, remote measurements, hysteresis, rate fall-back, and elective replacement indicators, must be characterized for functioning under expected temperatures (30C^{o} to 40C^{o}), loads (300 ohms to 2000 ohms), and battery voltage's Beginning of. Life (BOL) to End of Service (EOS). The device shall be programmed to each (node and feature and to the lowest, nominal, and highest values of programmed parameters. Analysis of the effect of worst-case combinations of load, temperature and battery voltage must be made.

Environmental

The pulse generator shall be subjected to a sequence of mechanical and environmental tests to assure that the device will meet its labelled specification after being subjected to condition that exceed those normally have seen in handling, shipping, storage or clinical use. Test shall include:

Temperature storage or cycling
Mechanical Vibration
Mechanical Shock

Interference

The pacemaker shall be evaluated for effects on its functioning and/or programming by external sources of interference. Sources of interference can be from the general environment, in this clinical setting, occupational environment or from the human anatomy. The following sources of interference shall be evaluated for all devices as appropriate to the specific device design:

Conducted and Radiated Electromagnetic interference.
Electrosurgical Units
Defibrillators

Reliability

The device must be tested and analysed from a reliability standpoint. Testing of the device or, where appropriate, its component, must include accelerated life testing which will demonstrate the excepted real time longevity performance and failure rate of the device.

Programmer

Programmers built for verification testing shall be representative of marketable products and subjected to functional, environmental, interference, software, and reliability testing. This testing must be designed to assure its operation according to written specification in conjunction with any and all of its intended pulse conditions; under specified, expected environmental condition; and its survival in use as well as in storage, shipping and handling.

Animal Testing

Animal Testing should be performed where appropriate to verify functions, features, or other characteristics of the device.

Biocompatibility Testing

For a material which has been tested and used previously in direct blood contacting devices, a sponsor may submit information available in publications or other legitimate sources which show that the material is nontoxic in tests identical or equivalent to those listed below. All new materials in the non-hermetic portion of the pulse generator must pass the tests below to insure safety for use in permanent implant.

The required toxicity tests for implantable device are listed as follows:

1. United States Pharmacopeia
2. (U.S.P.) XXI (Class V) Biological Tests for plastics and U.S.P. XXI Intramuscular implant tests and U.S.P. XXI pyrogen test.
3. Sensitization Assay:

4. Estimate the potential for sensitization of a material by using a test such as the guinea pig maximization test.
5. Cytotoxicity Test:
6. Determine the lysis of cells, the inhibition of growth, and other toxic effects on cells caused by material and extracts from the materials using cell culture techniques.
7. Haemolysis:
8. Determine the degree of red cell lysis and the separation of haemoglobin caused by materials in-vitro. Describe the test methodology.
9. Clinical Investigation
10. Objectives
11. The objective of the study must be defined such that the study will constitute a demonstration of reasonable assurance of the safety and efficacy for the device. The study must establish a list of indications and contraindications and, if any, warnings, and precautions for the use of the device. Generally, pacemakers must show pacing and sensing capabilities with modes, parameters, features, and logical combinations of these shown to be safe and effective within the meaning of the Federal. Food, Drug and Cosmetic Act, as amended, 1980.

Patient Selection

Patients should be selected. For the clinical studies who can be expected to benefit from the device's capabilities and whose conditions can demonstrate its effectiveness. The patient should be psychologically stable, cooperative and available for follow-up and have a reasonable life expectancy so that a proper clinical evaluation of the device might be conducted.

Investigators

Investigators for pacemaker studies shold be selected who are qualified by training or experience in cardiovascular disease at a minimum. In the case of devices with special features, trained physicians with special skills should be specified in a representative sample of the investigators.

Manufacturing

A descriptian of the testing during the manufacturing process must be included in the PMh Application in the section required by 21 CFR 814.20(b)4(v). This testing should complement the design verification testing to ensure that each manufactured unit will operate within the specifications of the design with respect to tolerances, environmental considerations, and interfaces. Summary descriptions of the following tests should be included:

Tests

Component

Screen

Burn-in

Assembly

Final Product

Special QC

8. POST-MARKET SURVEILLANCE8

One of the provisions of the Safe Medical Devices Act of 1990 (SMDA) provided for Discretionary Post-Market Surveillance (DPS) studies. The FDA has decided to use this provision to require the submission of additional data about the safety and effectiveness of permanent implanted cardiac pacemaker electrodes. FDA has determined that the legal entity who has received clearance to market through-submission of the premarket notification (510(k)) or premarket approval (PMA) application for a particular lead will have primary responsibility for conducting post-market surveillance of that lead. All others who are involved in the distribution of these devices will be responsible for ensuring that any data or information in their possession is made available to the sponsor of a DPS protocol.

9. CONCLUSIONS

Permanent pacemaker implantation is a very vital part of Cardiology and Cardiac surgery. It is a safe procedure with low complication rates. There is a gradual increase in the number of cases performed annually in Accra, with a slight male preponderance. Most patients are elderly, with complete heart block as the commonest indication. It is lifesaving, improves the quality of life and enhances survival.

REFERENCES

1] Putte JA, Malek R and Ellison MB, "Pacemaker", September-2020, https://www.ncbi.nlm.nih.gov/books/NBK526001/
2] Crofoot M, Sarwar M and Weir AJ, "External Pacemaker", September-2020, https://pubmed.ncbi.nlm.nih.gov/30137851/
3] Ivanchina AE, Kopylov FY and Volkova AL, Clinical value of algorithms of minimization of Right Ventricular pacing in Patients with Sick Sinus syndrome and history of Atrial Fibrillation, 2018; (8): 58-63.
4] Bernstrin AD, Daubert J and Flether RD, The revised NASPE/BPEG generic code for antibradycardia, adaptive-rate and multisite pacing, 2002; 25(2): 260-264.
5] Lind KD, Implantable Devices: Regulatory framework and Reform options, 2017; 1-14.
6] Parsonnet V, Furman S and Nicholas PD, Implantable Cardiac Pacemakers Status report and Resource Guideline, 1974; (50): 21-35.
7] Mulpuru SK and Madhavan M, Cardiac Pacemaker function, Troubleshooting and Management, 2017; 69(2): 189-210.
8] CDRH, Guidance for the Submission of Research and Marketing Applications for Permanent Pacemaker Leads and for Pacemaker Lead Adaptor 510(k) Submission, 2000: 1-17.
9] Haddad SA, Houben RP and Serdijn WA, The Evolution of Pacemakers, 2006: 38-48.
10] Woollons DJ, To beat or no to beat: The history and development of Heart Pacemakers, Engineering Science and Education, 1995, 4(6): 259-268.
11] Geddes LA, Historical highlights in cardiac pacing, IEEE Enginnering in Medicine and Biology, 1990, 2(2): 12-18.

12] Greatbatch W and Holmes CF, History and Implantable Devices, IEEE Enginnering in Medicine and Biology, 1991, 10(3): 38-49.
13] Sanders RS and Lee MT, Implantable Pacemakers, Proceedings of the IEEE, 1996, 84(3): 480-486.
14] Mond HG, Recent advances in pacemaker lead technology, Cardiac Electrophysiology, 1999, 3(1): 5-9.
15] Guyton AC and Hall JE. Textbook of Medical Physiology. 9th Edition. Saunders WB; Toronto; 1996.
16] Bronzino JD. The Biomedical Engineering Handbook. 2nd Edition. Boca Raton, FL; 2000,
17] Sutton R and Bourgeois I. The foundation of Cardiac Pacing; Part 1. Mount Kisco; NY; 1991.
18] Schaldach M and Furman S, Advances in Pacemaker Technology. Springer Verlag; Berlin; 1975.
19] Berkovits BV; American Optical Corp. Demand Pacer. United State Patent US3528428, 1970.
20] Berkovits BV; American Optical Corp. Atrial and ventricular demand pacer. United State Patent US 3595242A. 1971.
21] Dahmas DF, Implantable pacemaker testing guidance, 2007: 1-9.
22] Funke HD, Cardiac pacing with the universal DDD pulse generator: Technological and electrophysiological considerations. 1982: 191-223.

Unfolding: Mixed-Used Vertical Neighborhood in Mumbai

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Abstract: *Indian metropolitan cities are shifting towards taller buildings and compact living spaces due to rapid urbanization and influx of population. Previously, cities developed horizontally but are now developing vertically. This is due to the high rate of population migration to metro cities, leading to higher population density. According to a survey by World Bank, India is expected to experience a surge in urban population by 2050. In order to improve living conditions in densely populated areas and meet future needs, steps must be taken to bring together essential human necessities in a sustainable manner, including incorporating more green spaces and urban farming into development projects.*

The goal is to create a mixed-use development that meets the essential needs of people in the future. This development will include three types of spaces: a private space for personal needs, a space for economic engagement, and a more flexible space for building social bonds and community identity. The development will be organized vertically, with multiple layers that provide easy access to all necessary resources.

The concept of a vertical city involves creating a high-end housing infrastructure that also provides a complex network of functions typically found in a horizontal city. The goal is to balance comfort and elegance in the user's lifestyle. This concept is a prototype solution for future Indian cities and can be replicated to more efficiently face upcoming urban challenges.

Key Words: Vertical skyline, urbanization, vertical city, multi-dimensional, housing

Received Date: 10/10/2023 Revised Date: 05/11/2023 Acceptance Date: 08/12/2023

1. INTRODUCTION

As Mumbai according to this research study, this research encountered many problems like traffic congestions, emergency facilities, living habitation, higher footprint, daily requirements and many more. So, to solve these problems which can make every facility by their steps. By taking vertical city steps this problem can be solved and catered in a much better and progressive way in times of pandemic and crisis over that per city population that can be said as 2000 people with 100% of citizen facilities. So, this research paper conclusion came up with a solution of vertical city that can make small solutions for this problem in this rapid urbanization in Mumbai. The future of the mega-cities is a dense, compact, mixed-use and vertical urban growth. Flexibility, adaptability, technological advancements and implementation of social, economic & ecological aspects - are major challenges for getting ideal solution of vertical city. Technology and medical advances enable people to live longer, healthier lives. However, this brings up problems and concerns with overpopulation and overcrowding. As the population increases, so does the need for living space. Increasing the number of homes and neighbourhood often results in the destruction of forests and other habitats. This limits our natural resources, endangers wildlife, and threatens to disrupt our ecosystem. This problem cannot be overlooked.

Otherwise, the destruction of the planet and its inhabitants is inevitable. The human population continues to rise at an alarming rate. If action is not taken, a critical mass will eventually be reached. In other words, the environment will no longer be able to support us. You might be wondering how to live a sustainable life. People are starting to look to the vertical city concept as a solution to this growing and unavoidable problem.

In short, a vertical city is an entire human habitat contained in a massive skyscraper. Vertical cities hold the key to solving overpopulation and overcrowding. Rather than destroying forests and swamps to build houses, shopping centres, and factories, they can be placed in a vertical tower, serving to preserve the environment. Sky high construction enhances available living and working space, which reduces the impact of overpopulation. In a vertical city, people would live, work, and go to school.

Vertical towers also hold the key to preserving natural resources. As the population increases, it will become increasingly difficult for farmers to grow enough food to feed everyone. There simply won't be enough land to farm on. But vertical towers can be used for farming and agriculture. The possibilities offered by vertical cities are absolutely stunning and breathtaking. Some people believe vertical cities are impossible, but this is untrue. With the proper planning, these towers are can easily become a reality. The key to making this concept successful is

spreading awareness. As the dangers of overpopulation become apparent, so does the demand for a solution. Unless someone discovers a better answer, vertical cities currently hold the key to sustainability.

Fig - 1: City affected by calamities.

Fig - 2: Future for vertical city

Fig - 3: Future for vertical city

1.1. Concepts v/s Reality

If vertical cities are to become a reality, it's vital to separate concept from reality. In other words, the difference between what's possible and what isn't must be distinguished. While we have no actual cities to base facts on, there is growing demand for apartments contained within skyscrapers. The biggest concern at the moment seems to be funding. Creating and maintaining a vertical city will be a costly and time-consuming project. The issue of construction finance must be addressed. Those with the resources must be convinced this solution is viable before proceeding, which might prove difficult and challenging. But this is not an impossible task, a little planning and research is all that's needed. The next question is one of emotional well-being. A lack of sunlight can cause sadness and depression. Most people naturally enjoy the outdoors.

So, how will vertical cities affect the mood and emotional state of its inhabitants? There is not much doubt that remaining indoors can have an adverse impact on emotional well-being, but this can be solved. Open areas near the outer parameters can provide access to fresh air and sunlight. It would even be possible to install outdoor swimming pools and recreational areas. It's also important to remember living in a vertical tower does not mean you are confined to it. You will be free to come and go as you please.

Fig - 4: Pie chart showing rapid growth in population.

1.2. The Pros of Vertical Urbanization

Used effectively, building upwards allows you to urbanise using a smaller area of land. Urbanisation is often seen as damaging to the environment, destroying nature. While horizontal urbanisation naturally contributes to the disappearance of agricultural areas, vertical urbanisation limits the damage done by a huge margin. New York City is a prime example of a city that takes advantage of vertical urbanisation to limit the impact on the environment. Despite being one of the most popular cities in the world, Central Park can run for 2.5 miles through the centre of the city thanks to clever use of vertical urbanisation in the buildings surrounding it. The rise of global warming is one of the biggest environmental problems facing the planet at the moment, and continued horizontal urbanisation does little to solve the issue. Single-family homes are more difficult to insulate than multiple occupancy buildings, such as flats, and as such, energy loss is far higher. Vertical urbanisation allows for more efficient energy usage and conservation, at least when it comes to heating because the area is more localised. There are also economic and social benefits. It is far cheaper to build and develop on something you already have a foundation for, rather than to urbanise new areas completely.

1.3. The Cons of Vertical Urbanization

Unfortunately, vertical urbanisation does come with its share of problems too. When everything becomes closely packed together, the opportunities to maintain a healthy lifestyle become somewhat limited, when travelling becomes more vertical focused than horizontal. Walking down the road to your office may net you a few steps, but if you work on the top floor of the building, you are likely to opt to take the elevator rather than the stairs. Elevation also brings about problems with providing utilities to high-up areas. All floors of a building are going to require pipes for water usage and removal. The higher up you are, the more power is needed to pump water to the upper floors. Hygiene can also pose an issue, particularly with waterways, when hundreds of people are operating out of a single building. This puts pressure on architects and the water companies themselves to ensure the building can cope with the additional people. Plus, putting people close together may help promote social cohesion, but it reduces the amount of individual space available for people, which can impact one's quality of life.

1.4. Are vertical city feasible?

Right now, we have some very tall buildings, but nothing nearly as ambitious as a vertical city. Architects have come up with some theoretical designs, though. Italian firm Luca Curci Architects, for example, created an idea for a 180-floor vertical city that could support up to 25,000 people. The building would be zero-impact, include plenty of green spaces and let in natural light and air. It would be in the water and accessible via boat, helicopter, or a semi-submerged bridge.

For now, it is a concept—one that looks increasingly appealing as populations continue to grow and our planet's resources become more stressed. Building vertical cities would take extensive amounts of research and planning, but they could help solve some of humankind's biggest challenges, so the effort may be worthwhile. What would be the benefits of a vertical city? As populations expand, we need to find ways to house people without destroying what's left of our natural environment.

Building upward instead of outward enables us to host vast amounts of people in a small footprint, while conserving land and natural resources. We could then use the land for food production, recreation, or natural resources, or leave it as a natural area.

Vertical cities could also have other environmental benefits. Having numerous services and amenities in these mega towers would reduce the need for driving, reducing emissions associated with cars and saving residents money.

These buildings could also save energy and even generate their own electricity. These towers would have a multitude of surfaces where people could install solar panels, and their height and size make them ideal for wind turbines. The new Shanghai Tower, one of the world's tallest buildings, has already implemented this idea and has 270 wind turbines built into its facade. These buildings may also foster social connectedness by making it easier for individuals to socialize in common areas and visit friends. To do so, they would not even have to leave the building, although of course, they could if they wanted to.

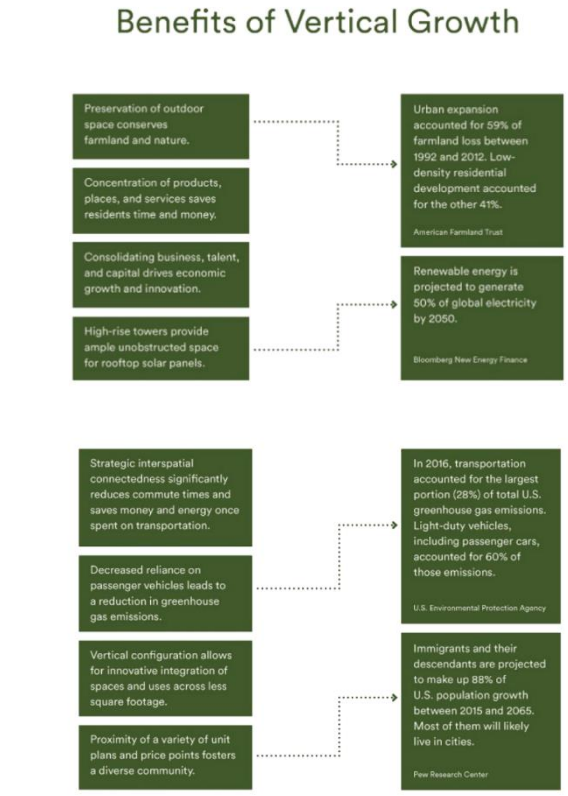

Fig - 5: Showing benefits of vertical city.

Utopian idea of 'Vertical City' is a future of modern mega-cities. By redevelopment of Mumbai as a vertical city, Mumbai's current urban problems could be resolved. The implementation of appropriate category of vertical city is important to get the expected result.

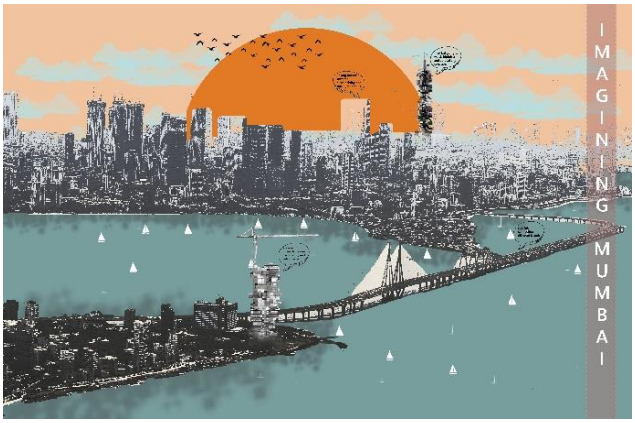

Fig - 6: Worli-sea link illustration.

2. OBJECTIVE AND METHODOLOGY

An approach to architectural innovation for vertical development in denser cities of India.

2.1. Objective

1. To fulfill all the requirements for citizens with their daily needs without moving out can give solution to many urban problems.
2. To study the diverse architectural style of the whole state and its representation.
3. To increase the economy by providing market spaces and eateries.

2.2. Methodology

Fig - 7: Working Methodology.

3. CITY NAMED MUMBAI

In this chapter of thrust study this research paper has tried to know city as Mumbai in a better vision and have closely noticed each problem faced by their citizens. Also have identified city's growth and evolution by different ruler and how architectural changes evolved during period of time.

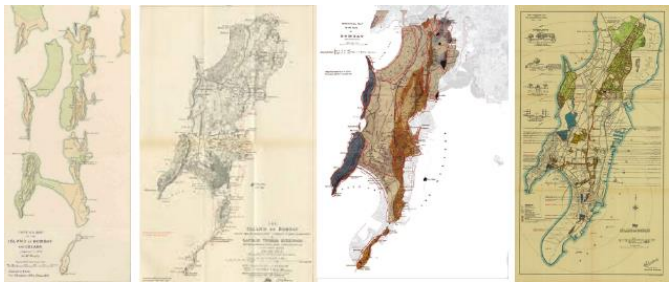

Fig - 8: *Image showing Evolution of Mumbai city according to timeline.*

3.1. Why Mumbai as urban fabric?

This offers many opportunities for discovery and exploration. There are virtually no vacant lots or surface parking. Also, as there are more intersections, traffic is slower and safer. Fine grained urban fabric is not imposed on a community like its coarse cousin.

Rather, it evolves over time in a piecemeal way, responding to what came before, and adapting to what came afterwards. This evolutionary process creates places that are not frozen in the era when they were built but are dynamic and reflective of a neighborhood's changing needs.

This creates an urban fabric that can seamlessly evolve over time from lightly developed residential areas to mixed-used retail to dense urban core if that's what the community desires. In this way, there are far more resilient than the mega projects mentioned above who, when they lose a single tenant, often fail.

3.2 Urban issue

Urban issues showing problems of Mumbai city with rapid growth of change.

Traffic congestion:

With trains arriving at the station every 3 minutes and transporting approximately 6 million people a day, the trains in Mumbai are heavily packed each day. Not only are the trains packed, but the roads are commonly seen excessively congested in peak hour traffic. Mumbai has invested in new technologies to prevent congestion. They are currently changing the lights where needed to make traffic flow smooth. Although this is preventing traffic congestion from being as bad as it has previously been, it isn't eradicating it either.

Pollution:

There are approximately 7,000 metric tons of rubbish being disposed of each day by Mumbaikars. With no mandatory recycling system in place by the government to date, Mumbai's waste is accumulating each day, and rubbish dumps are already filling up, polluting the area. On top of that, 700,000 cars are travelling on the roads of Mumbai each day. Creating air pollution with the rest of the manufacturing companies. Driverless cars have been considered to prevent air pollution, as private transport contributes to the air pollution the most. The driverless cars also prevent traffic congestion, but the cost to fill the whole of Mumbai with these cars is too large. A group called URBAIR, Urban Air Quality Management Initiative, has been created to increase the quality of air in Asia.

Poor sanitation:

It is said that most preventable diseases are spread through poor sanitation. This is due to there not being running water facilities in many houses or lavatories. As a result of

poor sanitation in slums, many children suffer from preventable diseases.

Due to diseases, mostly spread through poor toiletry sanitation, many foundations, including the Bill and Melinda Gates Foundation, have launched new campaigns to create awareness about the issue. Some campaigns have even started a competition to find a portable and affordable toilet like structure that can increase sanitation levels in India and other Asian countries.

Overpopulated slums:

Approximately 60% of Mumbai's population live in slums with no running water, electricity and gas. The shelters are made out whatever materials that can be sourced for a cheap price. The living standards in slums are very low and rubbish pollutes the area.so much that, large pipes are used as footpaths because the actual footpaths are too littered to walk on.

The Slum Rehabilitation Authority has been implementing multiple plans to rebuild slums. The current program being implemented today is rebuilding the slums into high rise buildings and offering 65% of the apartments to the previous slum residents and leasing the remaining 35% of apartments for commercial use. To go ahead with the plan, 70% of the previous slum residences have to okay the plan as they will have a housing issue in the building process.

Mumbai healthcare condition:

Doctors and healthcare workers who are responding to a global health crisis—trying to protect individuals, families, and communities in adverse situations with stretched resources, shortage of personal protective equipment (PPE) and other equipment's—have found themselves as unexpected targets in the fight against covid-19.6 There have been several reported incidences of such violence against them during this pandemic time in India. Although the exact numbers of such cases cannot be determined, there are a few glaring examples: on 8 April 2020, two trainee doctors in New Delhi were allegedly assaulted by a neighbor who accused them of spreading the disease. On19 April 2020, the burial of a neurosurgeon who had died after contracting covid-19 in Chennai was disrupted by a mob who attacked the undertakers. The citizens' opposition was due to a misconception that the contagion may spread in the neighborhood if the surgeon was buried there. A group of public health workers in Indore, a city in central India, who were trying to 'contact-trace' a person, were descended upon by a group of 100 people pelting stones and drove them away. Increasingly, reports pour in of doctors being spat on, hurled abuses at and driven away.

4. STUDY

Site analysis for area lower Parel with residence and commercial area. Detailed analysis is done for the study of urban fabric and its future growth need and usage.

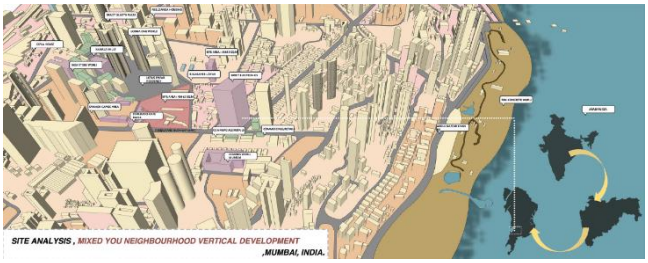

Fig - 9: Site analysis

Many mill plots have been used for Development and many structures have been developed so far and few of them are in abandoned condition which can be considered for redevelopment.

4.1. Site layout with immediate context

The site is adjoining to a busy street with a lot of commercial activities taking place. Heavy pedestrian movements are seen on the streets around the site.

4.2. Site description: why mill plot?

The site is flat land with wild vegetation and abandoned mill structures. The structures have been partly demolished by N.T.C. There are several high Rises in the lower Parel area close to site such as World one 117 story high, omkar 1973, Raheja Imperia, Lodha trump, and many more.

Connectivity from outside:

The site is very well connected by a good Road Network & it is very much reachable by both the locals & the tourists. The site is very well connected with City Level Transit Nodes, i.e., Airport, Railways & Bus Stations, from where the city welcomes its tourists. easy connectivity with surrounding and residential and commercial area with high profile offices makes site more prominent.

Fig - 10: Urban mapping for site fabric

Table - 1: Percentage for area distribution

	AREA (sq.m.)	PER. (%)
PUBLIC STREET/ROOD	TOTAL AREA	29.7%
WATER BODIES/RIVER	12,000	12%
BUILDING FOOTPRINT	50,000	50%
PUBLIC OPEN SPACES	3,300	3.3%
PRIVATE OPEN SPACE	5,000	05%
TOTAL AREA	1,00,000	100%

Strength
- Worli metro station is proposed 900-meter walk from the site.
- Considerably large site area for development.
- Easy approach to Bandra - Worli sea link.
- Aerial view of sea above 75 feet. (As per world one).
- Majorly surrounded by residential and commercial spaces.
- F.S.I availability.

Weakness
- Far from the lower Parel railway station (Central line).
-Limited approach to site from main road.

Opportunity
- Site located in the vicinity of tall structure like world one.
- Work space requirement.

Challenges
- Highly congested area.
- Lack of open green spaces.
- Strong wind force.
- Rainfall amount in Mumbai.
- High summer temperature.

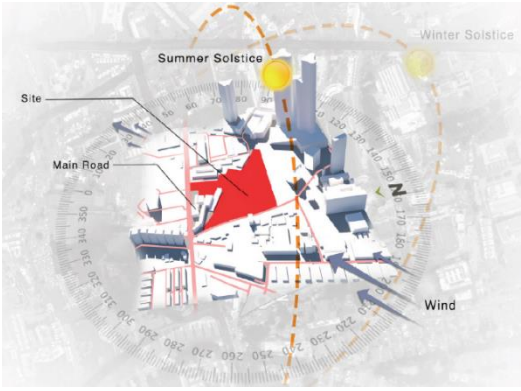

Fig - 11: Site analysis diagrams

5. DESIGN EXECUTION AND DESIGN SOLUTION.

The urban lung allows the building to breathe conceptually through a large full height atrium that is naturally ventilated by air inlets located at the sky garden levels. The gardens create natural cross and stack ventilation, along with purifying the air quality through the use of specific plant species.

Author wants to build a new environmentally friendly town, where environment is considered as an important part of everyday life. This research paper proposes Spiral Garden system: a public sustainable place like a green heart, easy to maintain and self-sufficient, coated by a joint population work that will cause a greater sociability among neighbours. Like a spiral, a light structure protected by a transparent and suggestive mesh, raises the city to create sustainable exchange places in different ways.

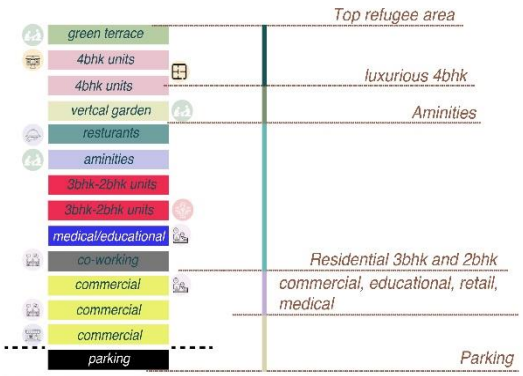

Fig - 12: Diagram for area distribution

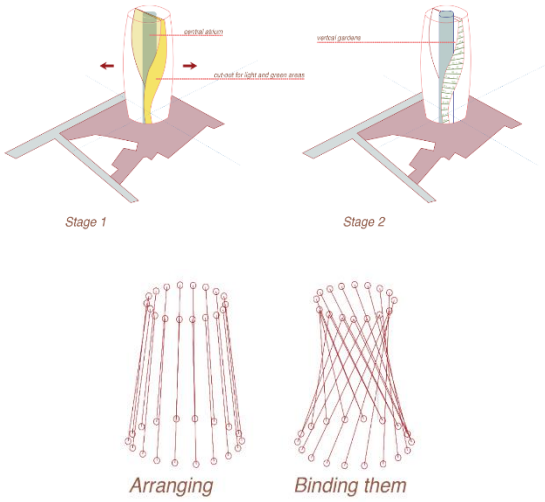

Fig - 12: Conceptual Diagram

The author wants to build a new environmentally friendly town, where the environment is considered an important part of everyday life. This research paper proposes **Spiral Garden system**: a public sustainable place like a green heart, easy to maintain and self-sufficient, coated by a joint population work that will cause a greater sociability among neighbors. Like a spiral, a light structure protected by a transparent and suggestive mesh, raises the city to create sustainable exchange places in different ways.

This spiral contains an ascendant garden where native vegetation areas coexist with urban orchards shared and planted for the neighbors in a way to establishing a green outdoor walk and making easy the involvement of neighborhood residents in their creation and maintenance, as well as increasing social relations between people exchanging the natural products gowned and causing an environmental exchange. To sum up, we propose an ecological project in a way to give sustainable change to dally city lies, where humans and nature can coexist.

Fig - 13: Site layout

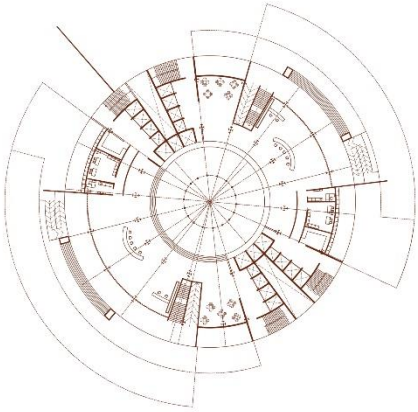

Fig - 14: Ground floor layout

The distribution of different typologies was done in a way that the lower portion was provided for offices as well as small apartments, the central portion was provided for recreational activities and the top floors were given for luxury apartments.

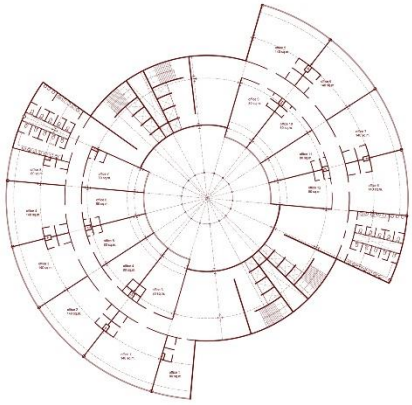

Fig - 15: Commercial floor layout

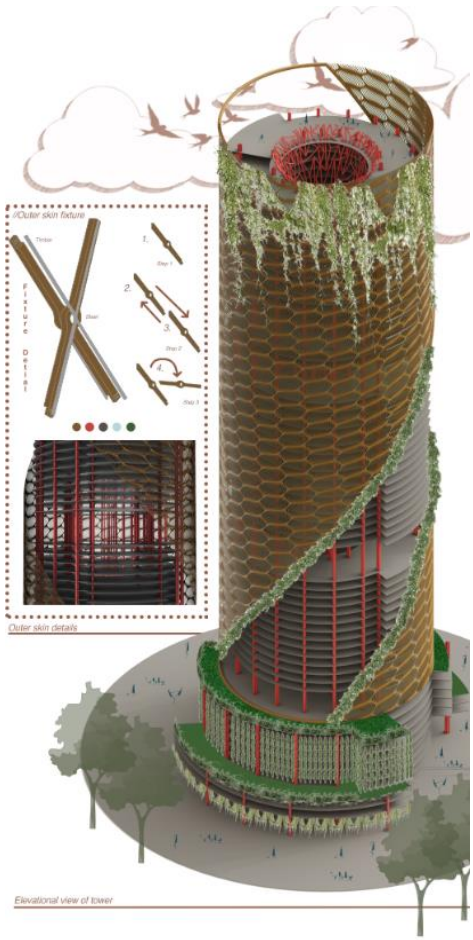

Fig - 16: Isometric view and details

An additional core was added to the building so that residential and office spaces can have different vertical movements respectively also skewing the building to get maximum view towards the sea and even creating an urban edge for the people nearby on the western side of the site.

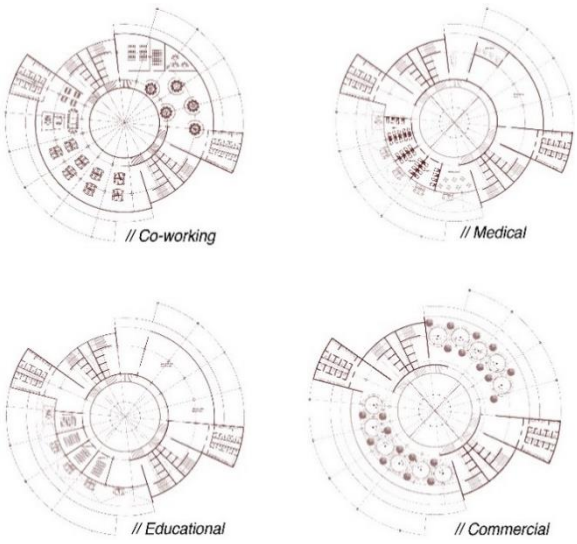

Fig - 17: Co-working, Medical, Educational and Commercial layout

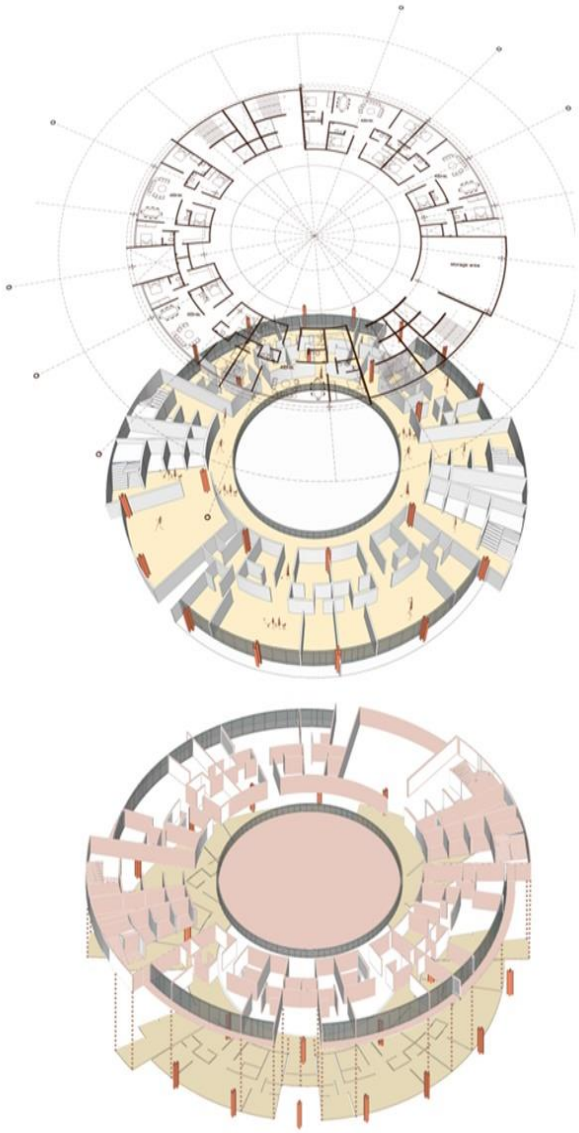

Fig - 18: Axonometric view for floor

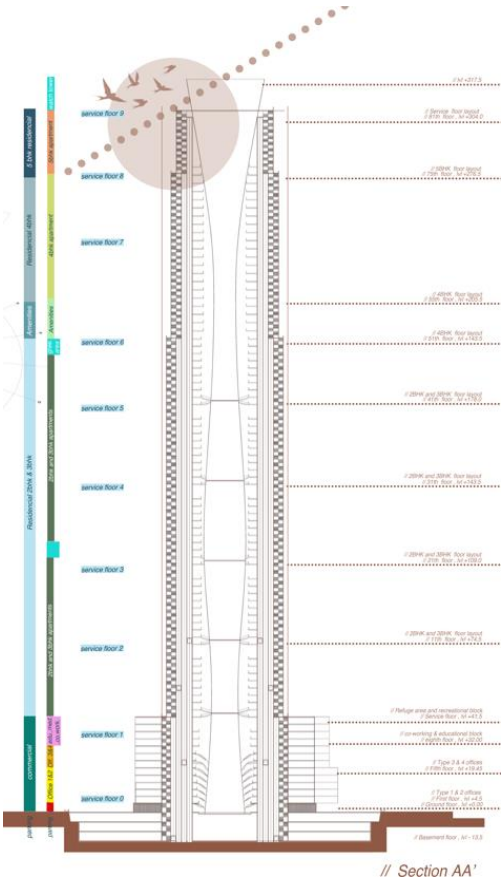

Fig - 19: Section of tower

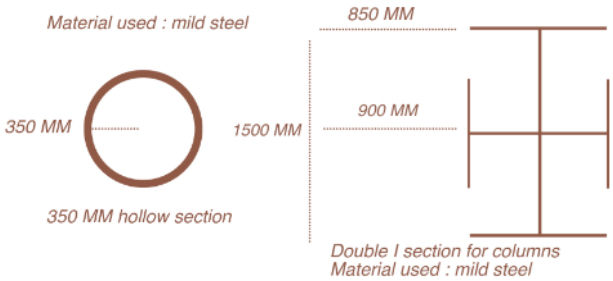

Fig - 20: Structural details

Suspended structure

- Central core with horizontal cantilevers at roof level, to which vertical hangers.
- Floor slabs are suspended from hangers.

Outrigger truss

The outrigger trusses or girders are connected directly to shear walls or braced frames at the core and to columns located outboard of the core.

Tubular framework

Creating outer skin as structural element with steel sections that can hold weight of skyscraper structure. Having core type structure surrounding the building.

Fig - 21: Building views.

Fig - 22: Building views.

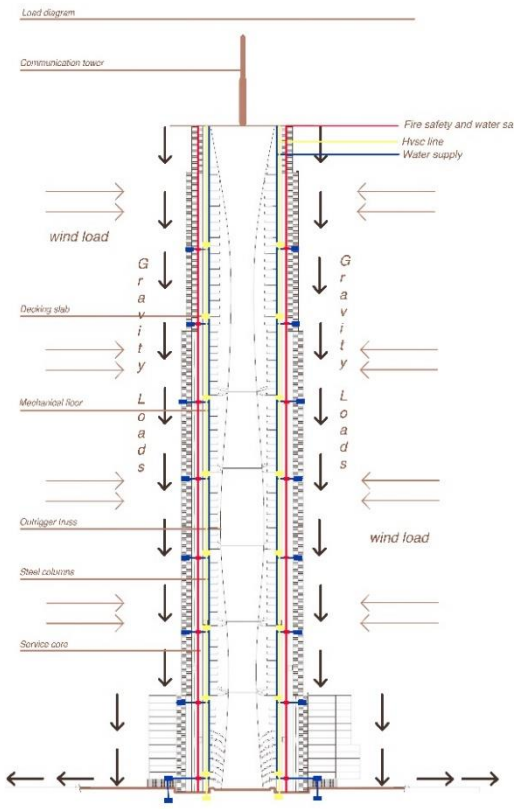

Fig - 23: Load calculation

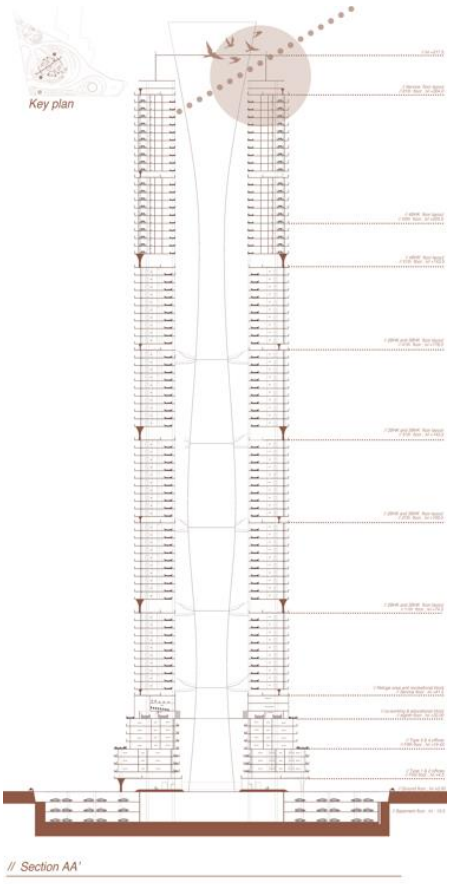

Fig - 24: Section for tower

6. CONCLUSIONS

It is now clear that current urban problems of Mumbai are not decreasing by the luxurious single-used vertical developments, new transportation systems (like monorails), proposals of new satellite towns like Navi Mumbai or by slum relocation plans. A new city-planning strategy of vertical city for the redevelopment of Mumbai is required to protect Mumbai from further declination and decaying by urban problems.

My own approach to sustainable city form for Mumbai is - reinterpretation and reinvention of the dense, compact, three-dimensionally connected & vertical mixed-use model of a city, which not only includes luxurious lifestyle & economical sustainability for rich & middle-income groups but also includes economic growth of the poor and even socio-cultural & environmental sustainability of the city.

It is clear from the intentions of the above diagram that Mumbai as vertical city is leading towards the third category of vertical city (Series of mixed-use towers). But this category of vertical city cannot apply directly in Mumbai, by providing a singular vertical city center with gradually decreasing height due to its linear geographical area. Mumbai's geographical constraints are leading towards another category of vertical city, where

decentralized vertical nodes/clusters are linearly or irregularly generated in the whole city.

It will be feasible to develop all important areas of Mumbai as vertical nodes of mixed-use & connected towers, surrounded by factories and urban farmlands.

REFERENCES

1] **Standard Journal Article**
Deep Gala Research paper on vertical neighborhood published at Issue on 2 March 2018.

2] **Standard Journal Article**
Razjan Hassan Research paper on co-working hub published at Issue Nov 7, 2018.

3] **Research thesis,**
Nirav Khandwala at Nottingham university on Feasibility of Vertical city in Mumbai Published on September 2018

4] Christopher Alexander research paper on a city is not tree published in magazine Design in 1966

5] Ype Cuperus an introduction to open building published in Delf University in 2000.

6] Vertical city: A solution for sustainable living by Kenneth King., Book.

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Shah DP, Jani GK, Modification and Characterization of Gellan Gum, Pharmaceutical Technology, 2009; 33(7): 48-58.

Books and other Monographs

Author(s) of Book (Surname initials). Title of Book. Edition. Publisher; Place of publication; Year of publication.

Personal author(s)

Eisen HN. Immunology: an introduction to molecular and cellular principles of the immune response. 5th Edition. New York: Harper and Row; 1974.

Chapter or Article in a book

Author(s) of Chapter (Surname initials). Title of Chapter. In: Editor(s) name, Editors. Title of Book. Place of publication: Publisher; Year of publication, Page numbers.

Kelly HW and Sorknes CA. Asthma, Dipiro JT, Talbert RL, Yee GC, Matzke TR, Wells BG, Posey LM, Pharmacotherapy- A Pathophysiological Aproch, Sixth Edition, The McGraw-Hill; 2005.504

Patent

Larsen CE, Trip R, Johnson CR, inventors; Novoste Corporation, assignee. Methods for procedures related to the electrophysiology of the heart. US patent 5529 067. 1995.

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SAMHITA

Multi-Disciplinary Research Journal

Vol 1 Issue 1 (Jan - June 2023)

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