CHAPTER-1
INTRODUCTION

1.1 Malaria: The Prevalence

In 2020, an estimated 241 million malaria cases were reported in 85 malaria-endemic countries, with the majority of the rise coming from the WHO African Region. There were 224 million malaria cases estimated at the baseline of the Global Technological Strategy (GTS) for malaria 2016–2030 in 2015. Twenty-nine countries accounted for 96% of worldwide malaria cases, with Nigeria (27%), the Democratic Republic of the Congo (12%), Uganda (5%), Mozambique (4%), Angola (3.4%), and Burkina Faso (3.4%) accounting for nearly 55% of all cases. The WHO South-East Asia Region was responsible for around 2% of all malaria cases worldwide. Malaria cases have decreased by 78%, from 23 million in 2000 to around 5 million in 2020. Malaria cases have reduced by 83% in this region, from approximately 18 per 1000 people at risk in 2000 to roughly three cases in 2020. In the area, India accounted for 83% of the cases (Usui *et al.* 2019). Malaria mortality increased by 12% in 2020 compared to 2019, reaching an estimated 627000; of the additional 69000 deaths, an estimated 47000 (68%) were attributable to service delays during the COVID-19 epidemic. The malaria endemic countries are represented in Fig. 1.1. Despite COVID-19's issues, we've observed some good tendencies this year. Two nations – China and El Salvador – were declared malaria-free by the WHO in 2021, and another 25 are on pace to eradicate the disease by 2025. Sri Lanka was certified malaria-free in 2016 and remains malaria-free (https://www.who.int/ publications/i/item/9789240040496).

Between 2000-2020, an estimated 1.7 billion malaria cases and 10.6 million malaria deaths were avoided globally. The WHO African Region has seen the most cases (82%) and fatalities (95%) avoided, followed by the WHO South-East Asia Region (cases 10% and deaths 2%). There were an expected 33.8 million pregnancies in 33 moderate and high transmission countries in the WHO African Region in 2020, with 11.6 million (34%) of those exposed to malaria during pregnancy (https://www.who.int/ publications/i/item/9789240040496). Total malaria infections decreased by 84% in the 21 nations participating in the "Eliminating Countries for 2020" (E-2020) effort from 2010 to 2020.

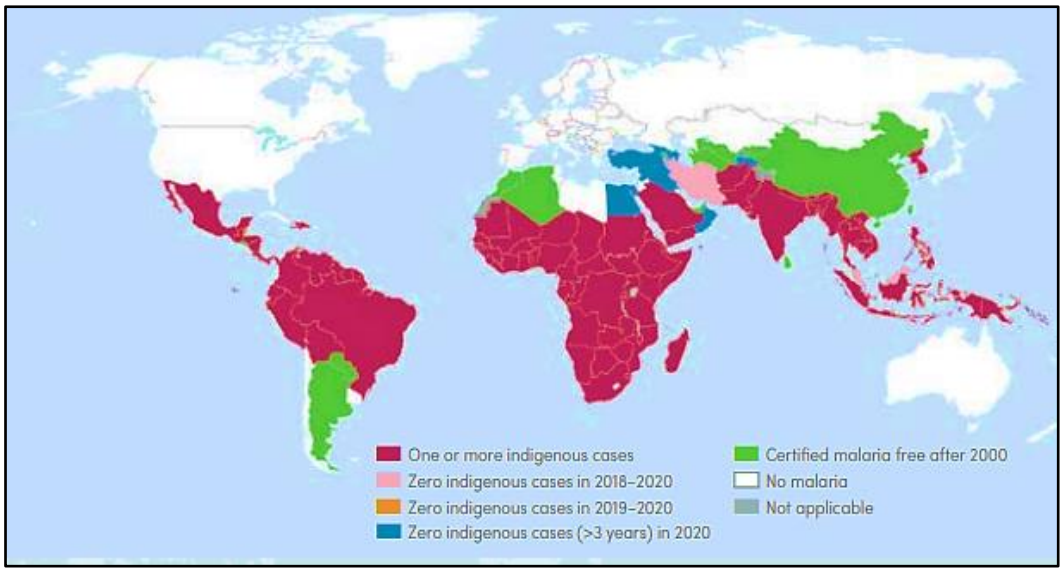

Figure 1.1: Map of malaria endemic countries (Source: WHO Malaria Report, 2021)

1.2 Malaria: The Disease

Malaria is a protozoan ailment of red blood cells induced by four Plasmodium species (*Plasmodium falciparum, Plasmodium vivax, Plasmodium malariae,* and *Plasmodium ovale*. Most malaria-related fatalities are caused by *P. falciparum* (Rougeron *et al.,* 2022). Malaria is transmitted most commonly by the bite of an infected female anopheles, although it can also be contracted through blood transfusions or infected needles, as well as congenital infection. Only 40 of the 430 identified species in this genus are important malaria vectors.

The malaria vector is the female Anopheles mosquito, which injects roughly 15–20 sporozoites into the bloodstream. When these sporozoites reach the liver, they increase and develop into hepatic schizonts, eventually exploding. Hundreds of merozoites enter the circulation and infect red blood cells quickly. In 1 to 2 weeks, the hepatic schizont ruptures. Young trophozoites consume hemoglobin from red blood cells and mature into schizonts, which rupture and release additional merozoites. For *falciparum, vivax, and ovale malaria,* the asexual life cycle takes 48 h, but *P. malariae* takes 72 h (Mancio-Silva *et al.,* 2022). Certain parasites evolved into sexual forms after a lot of asexual life cycles. In *falciparum malaria*, gametocyte formation takes around ten days, but just four days in *vivax malaria*. The life cycle of the female anopheles mosquito is completed when male and female gametocytes are consumed in the midgut, resulting in the formation of the zygote and the subsequent production

of new sporozoites (Fig. 1.2). The hypnozoite stage is absent in *P. malariae*, although it can survive in the blood for years if not treated adequately (Rougeron *et al.,* 2022).

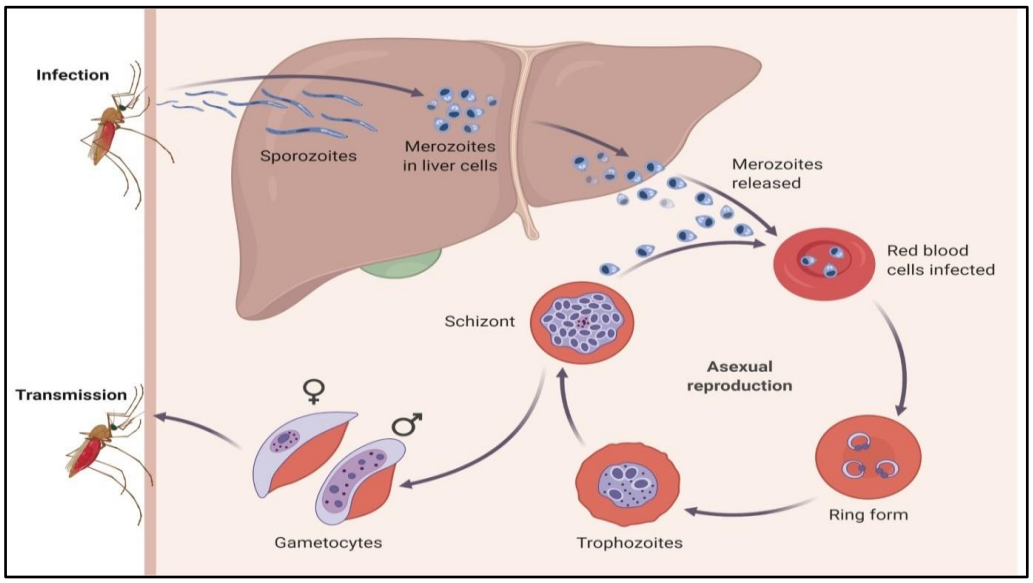

Figure 1.2: The life cycle of the malaria parasite.

1.3 MALARIA: The therapeutic armamentarium

Conventional malaria chemotherapy has proved beneficial, but not significantly because it is linked to recurrent failures (Gujjari *et al.,* 2022). As it becomes more challenging to regulate the disease's spread throughout tropical areas, the disease's distinctive character in connection to its transmission circumstances becomes crucial (Espíndola *et al.*, 2022).

The parasite's complicated life cycle and the development of medication resistance are thought to be the primary factors in novel drug research in this disease segment. Another important aspect is the alarmingly high number of immune-compromised individuals who have co-infections with malaria.

Each year, these illnesses claim the lives of millions of people. Furthermore, the complex prescribed regimen, based on combination therapy, raises treatment costs and reduces patient compliance owing to the related adverse effects (Zhou *et.*, 2021). Effective disease prevention by eliminating mosquitoes with insecticidal spray and giving treatment based on artemisinin combination therapy are among the tools and approaches used to halt the spread of malaria. In cases of pregnancy, intermittent antimalarial medication has been utilized as a last resort to lessen and reduce the negative consequences of malaria infection on the foetus (Arya *et al.*, 2021).

Treatment protocols and regimens for malaria are closely related to the parasite's medication resistance and the government's effort to prevent and control fatality rates (Tindana *et al.,* 2021). Given the low number of new antimalarial medications licensed since 1990, the hunt for more effective and less toxic antimalarial continues, and the development of nano-based drug delivery formulations is likely to be a crucial tactic in the fight against malaria.

Antimalarial drugs can be classified based on antimalarial activity and structure as mentioned in Table 1.1 (Jones *et al.,* 2020).

Table 1.1: Classification of antimalarial drugs.

Based on antimalarial activity	According to the structure
Tissue schizonticides for causative prophylaxis: These medications attack the plasmodia's initial tissue forms, which commence the erythrocytic stage after growing within the liver. Further infection development can be avoided by inhibiting this step. Pyrimethamine and primaquine, for example. *Tissue schizonticides for relapse prevention:* These medications target the hypnozoites of *Plasmodium vivax* and *Plasmodium ovale* in the liver, which causes symptoms to resurface after reactivation. The prototypical drug is primaquine. However, pyrimethamine also has this effect. *Blood schizonticides*: These medications target the parasite's blood forms, thereby ending clinical malaria attacks. The most effective medicines in antimalarial chemotherapy are these. Examples include chloroquine, mefloquine, halofantrine, pyrimethamine, sulfadoxine, sulfones, tetracyclines, and other antimalarials.	*Aryl amino alcohols*: mefloquine, QN, halofantrine, quinidine (Cinchona alkaloids). *Folate synthesis inhibitors*: Type 1 – competitive inhibitors of dihydropteroate synthase – sulphones, sulphonamides; Type 2 – inhibit dihydrofolate reductase – biguanides like proguanil and chloroproguanil; diaminopyrimidine like pyrimethamine 8-aminoquinolines: WR238, 605, primaquine. Naphthoquinones: atovaquone Antimicrobials: azithromycin, tetracycline, doxycycline, clindamycin, fluoroquinolones 4-aminoquinolines: amodiaquine, chloroquine. Peroxides: artemisinin (qinghaosu) derivatives like artemether, artesunate, arteether (ART), artelinic acid iron chelating

Gametocytocides: These medications kill the parasite's sexual forms in the bloodstream, preventing infection transfer to the mosquito. Chloroquine and QN are gametocytocidal against *P. vivax* and *P. malaria*, but not against *P. falciparum.* All plasmodia, including *P. falciparum*, are gametocytocidal to primaquine. *Sporontocides:* These medications stop mosquitos from developing oocysts and thereby stop transmission. This is how primaquine and chloroguanide work.	agents: desferrioxamine

Quinine (QN), a cinchona alkaloid has been the most popular medication to treat malaria for over 300 years. QN was then substituted with chloroquine, although it is prohibited due to widespread resistance. Antimalarial medication therapy has changed dramatically since Chinese scientists discovered artemisinin derivatives. These medications are parasiticidal, killing immature circulating parasites and preventing them from maturing further (Adams *et al.,* 2022).

1.4 The artemisnins

The most significant artemisinin derivatives are artesunate, ART, and dihydroartemisinin. Semisynthetic and synthetic versions are now being produced as well. Artemisinin derivatives are fast-acting and have a short-beginning effect (Agrawali *et al.,* 2022). The artemisinin derived from the plant qinghaosu is represented in Fig. 1.3. Artemisinin and its derivatives are a relatively safe and well-tolerated series of drugs, with the majority of reports of side effects in clinical studies being anecdotal. However, reports of toxicity in laboratory animals and cell lines have raised questions about its safety. However, this might be due to extended exposure to greater doses of artemisinin's (Alam *et al.,* 2018).

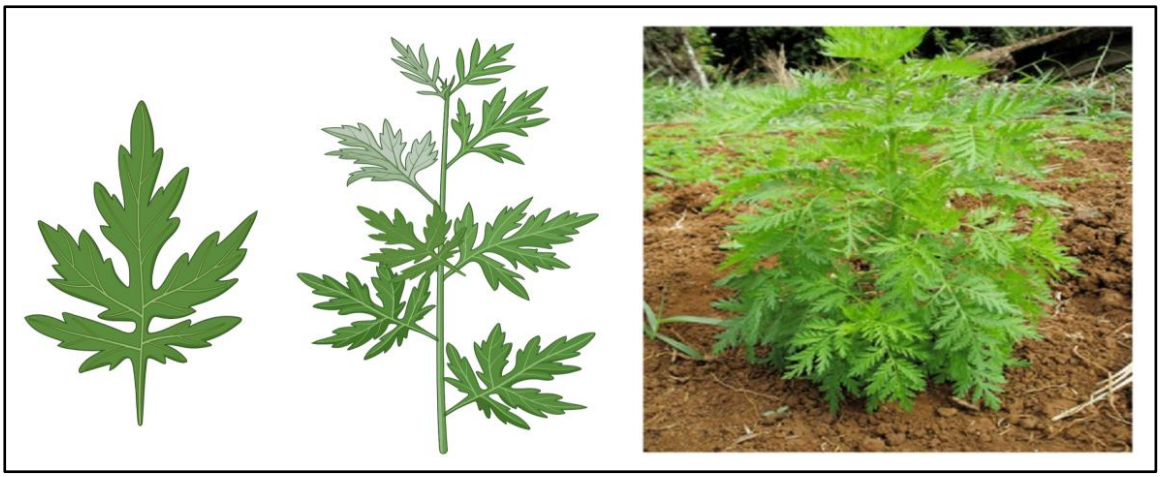

Figure 1.3: Artemisinin is derived from the plant qinghaosu.

Artemisinins produce neurotoxicity in the brainstem areas of animals at larger dosages (Alemad *et al.,* 2020). However, this has yet to be shown in humans. Another potential risk is the use of artemisinin to treat malaria during pregnancy. Parenteral injection of artemisinins caused embryo loss in rats, rabbits, and monkeys, perhaps due to erythropoiesis suppression (Alhasani *et al.,* 2019). Artemisinins are not advisable for antimalarial therapy during the first trimester of pregnancy.

Artemisinin clears the blood of parasites quickly. However, it is ineffective against parasite liver stages. The medication cannot be used as a preventative measure. Artemisinin and its derivatives have been discovered to destroy *P. falciparum's* asexual blood and early stages of gametocytes (Amasya *et al.,* 2021). Although clinical resistance to artemisinin and its derivatives in malaria parasites collected from patients has not yet been demonstrated, there have been isolated cases of artemisinin therapeutic failures (Archontaki *et al.,* 2002).

Plasmodium strains have a unique capacity to dodge and overcome the unfavorable circumstances imposed on medication therapy and develop sophisticated mechanisms for drug resistance due to their genetic diversity. As a result, many commonly used antimalarial medicines have recently lost their effectiveness. CQ and MDR remain the most severe hurdles in malaria therapy that have recently appeared. Mutations might cause resistance in the *P. falciparum* MDR gene (pfmdr1) or the *P. falciparum* CQ transporter gene (pfcrt). The CQ resistance transporter (CRT) and the P-glycoprotein homologue 1(Pgh1), two potential protein transporters found in *P. falciparum*, are the

result of gene expression and may be involved in the CQ-resistance (CQR) phenomena (Ayoub *et al.,* 2016).

Another essential consideration in malaria treatment is the mode of delivery. The oral route is better for clinically uncomplicated malaria. Parenteral therapy is recommended in severe instances of complicated malaria, and *i.v.* The administration is required in the treatment of cerebral malaria. The parenteral administration of QN or artemisinin, both efficient antimalarials, is still the backbone of cerebral malaria treatment. QN has higher toxicity, and artemisinin derivatives are lipophilic, which means they have short plasma half-lives (Baka *et al.,* 2008).

Artesunate, artemether, ART, and dihydroartemisinin are the most significant artemisinin derivatives. ART, an ethyl ether derivative of dihydroartemisinin, is one of the most widely used therapeutics worldwide because its lipophilicity is higher than that of other artemisinin derivatives when compared to artemether, allowing it to accumulate in patient brain tissues and effectively control cerebral malaria (Banik *et al.,* 2022). It is effective against *P. falciparum* strains because it exhibits erythrocytic schizonticidal action.

ART is effective against chloroquine-sensitive and chloroquine-resistant Plasmodium strains in various trials (Gurumukhi *et al.,* 2022). ART kills malarial parasites by reducing the peroxide moiety with hemin, which produces additional harmful chemicals such as free radicals and reactive aldehydes. The parasites' hemin-rich internal milieu is thought to cause artemisinin's toxicity to the species. *In vitro*, without cells, it has been demonstrated that trioxane cleavage by ferrous ions follows a distinct molecular path and produces different compounds than nonferrous reducing agents. Erythrocyte deformability was reduced when ART and heam were combined. Only when ART was incubated in the presence of iron did electron paramagnetic resonance reveal a spin-trapped free radical signal (Beg *et al.,* 2021). As a result, iron is essential in both the mechanism of action and the toxicity of ART. The mechanism of action of ART is represented in Fig. 1.4.

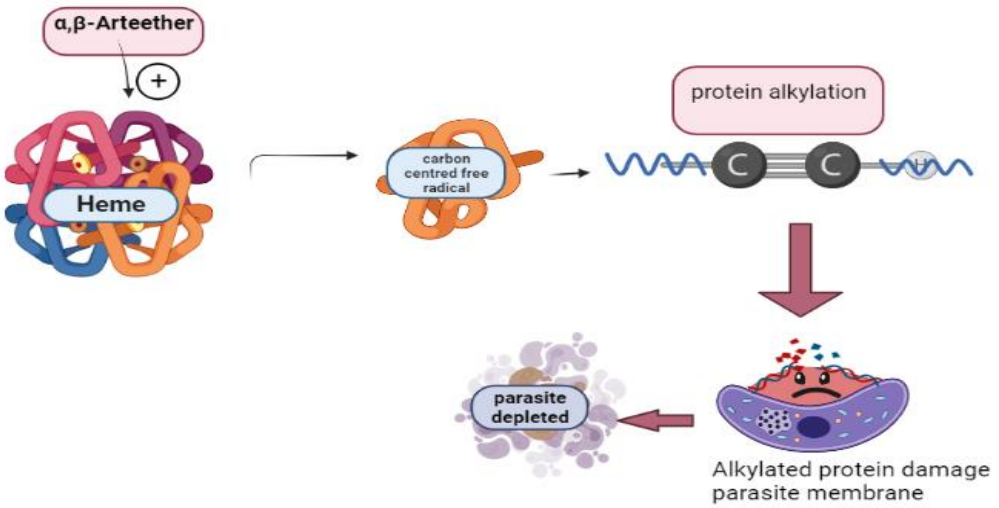

Figure 1.4: The mechanism of action of ART.

1.5. Solubility Enhancement: The integrated approaches for formulation development.

The main problem with ART is its low water solubility (17 μg/mL) and limited stability in the stomach media, where it degrades at pH 1.2 leading to poor bioavailability (Beloqui *et al.,* 2017). After oral delivery, the bulk of the α, β -ART (40%) is degraded in the stomach (Kumar *et al.,* 2021). The medicine belongs to the biopharmaceutical categorization system (BCS) Class II, which indicates poor solubility and high permeability. Due to these constraints, only intramuscular injection of/ ART is now available on the market, which has drawbacks such as patient noncompliance, injection site discomfort. ART is a chemical classed as BCS class II. As a result of the drug's solubility constraint, it's essential to raise the drug's solubility, which increases its bioavailability (Bhise *et al.,* 2017)

In quantitative terms, the proportion of a solute in a saturated solution at a given temperature is known as solubility. In qualitative terms, solubility is the spontaneous interaction of two or more substances to form a homogenous molecular dispersion. A saturated solution is one in which both the solute and the solvent have reached equilibrium. Parts, percentages, molarity, molality, volume fractions, and mole fractions can quantify drug solubility (Bonthagarala *et al.,* 2013). Solubility is the result of simultaneous as well as opposing processes of solutes and thereby combining, known as dynamic equilibrium (e.g., precipitation of solids). When the

two functions run at the same speed, solubility equilibrium occurs. Under some conditions, equilibrium solubility can be surpassed, resulting in a metastable supersaturated solution (Jain *et al.,* 2007). Solubility should not be confused with the capacity to dissolve or liquefy a material since this can happen as a result of a chemical reaction as well as dissolution. Putting one or more amphiphilic components into a solvent to generate a thermodynamically stable solution of a chemical that is ordinarily insoluble or only marginally soluble is known as solubilization.

The oral route is the most desirable and preferred mode of administration for systemic effects; however, low drug solubility is a significant challenge for researchers (Homayun *et al.,* 2019). Water insolubility causes formulation problems in around 40% of orally delivered medicines. The rate of a drug's dissolving, absorption, distribution, and excretion is determined by its solubility properties (Vinarov *et al.,* 2021). Solubility is one of the key limiting requirements for orally given medicines achieving their concentration in systemic circulation for pharmacological response. As a result, increasing the solubility of a mixture enhances its bioavailability. There are numerous ways to improve a drug's solubility and bioavailability if it is water-insoluble. The techniques are chosen depending on various considerations, including the drug's properties, the substance's nature, and the intended dosage form (McGuckin *et al.,* 2022). Solubility enhancement strategies include physical adjustments, chemical modifications of the medicinal component, and other methods (Rinaki *et al.,* 2003).

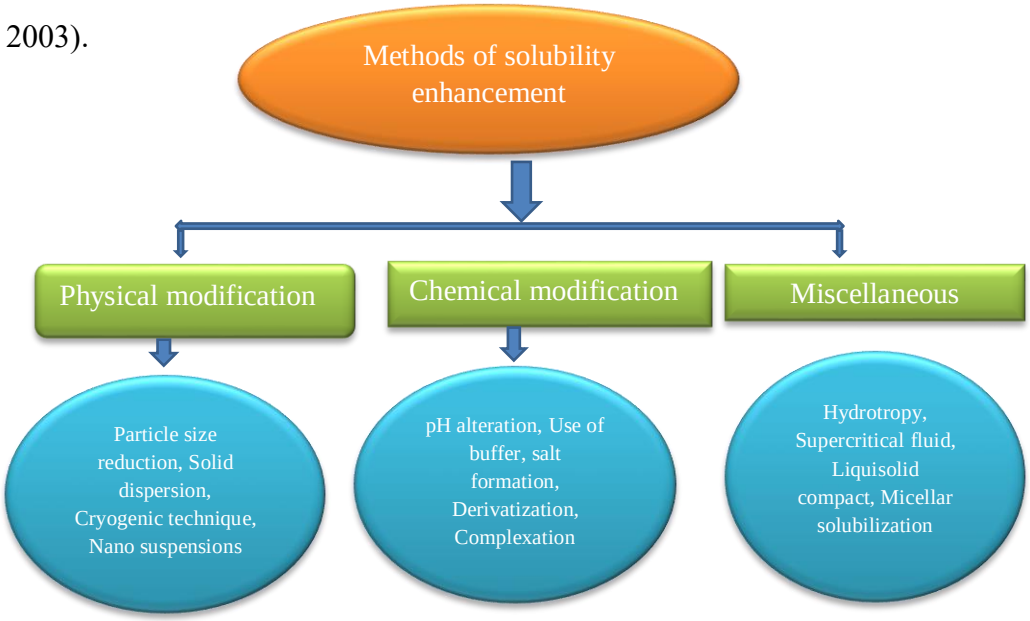

Figure 1.5: Various solubility enhancement techniques.

Various approaches for increasing solubility are employed are discussed in Fig 1.5. Physical alterations are the most widely used approach. These techniques include particle size reduction techniques like nanosuspensions, micronization, crystal lattice modification techniques like crystallization, amorphous form, and polymorphs, drug dispersion in carriers like solid dispersions, solid solutions, eutectic mixtures, and cryogenic techniques (Brough *et al,.* 2013). Chemical changes are the second most popular method for increasing solubility. It consists of pH modification that may cause a weakly water-soluble medicine to dissolve in water.

Inclusion complexes also improve the solubility of medications that are difficult to dissolve. When a nonpolar molecule or a nonpolar component of a molecule (known as a guest) is introduced into the cavity of another molecule or group of molecules, inclusion complexes (known as the host) are produced, which improve solubility. To boost the solubility of poorly soluble drugs, various techniques such as Hydrotropy (adding a significant amount of a second solute increases the first solute's water solubility) and cosolvency (weakly soluble drugs must be combined with a water-miscible solvent in which the drug is readily soluble to boost its solubility in water) are used.

Carl A. Neuberg, a physicist, was the first to coin the term "hydrotropy" in 1916. Hydrotropes having an amphiphilic chemical structure can make sparingly soluble organic compounds more soluble in water (Hodgdon *et al.,* 2007). Including a second solute (hydrotrope) helps improve the aqueous solubility of poorly soluble solutes. Hydrotropes are ionic organic salts that help to enhance or reduce the solubility of a given solute in a given solvent through salt in' or salt out' actions. A weak Van der Waals contact, such as attractive dipole-dipole interaction, occurs when a hydrotropic molecule interacts with a less aqueous-soluble molecule (Dhapte *et al.,* 2015). Hydrotropes have both hydrophobic and hydrophilic fractions. However, the hydrophobic portion is relatively minor in comparison to surfactant. The balance between the hydrophobic and hydrophilic sections of the hydrotrope determines the efficacy of hydrotropic solubilization. The higher the hydrophobic component of addiction, the better the hydrotropic, and the existence of a charge on the hydrophilic component are less relevant (Liu *et al.,* 2022). Table 1.2 illustrates various hydrotropic agents used for enhancing the solubility of drugs.

Table 1.2: Classification of hydrotropic agents.

S. No.	Hydrotropic agents	Examples
1.	Urea and its derivatives	Urea, N, N-dimethyl urea
2.	Aromatic alcohols	Resorcinol, pyrogallol, catechol, α,β-naphthols
3.	Derivatives of resorcinol, pyrogallol, catechol, α,β-naphthols	Sodium salicylate, sodium benzoate, sodium citrate, sodium acetate, sodium ascorbate, potassium citrate, citric acid, and benzoic acid.
4.	Aromatic hydrotropes	Caffeine, nicotinamide, N, N-diethyl nicotinamide, N, N-dimethyl benzamide
5.	Surfactants	Sodium dodecyl sulfate, dodecylated oxidibenzene
6.	Soluble drugs	Ibuprofen sodium, metformin hydrochloride

The mixed hydrotropic solubilization technique enhances the solubility of poorly water-soluble drugs in the blends of hydrotropic agents, which may give a synergistic enhancement effect on the solubility of poorly water-soluble drugs (Patil *et al.,* 2021). The concentrated solutions made by taking several hydrotropes (sodium benzoate, sodium ascorbate, sodium citrate, niacinamide, urea) in small concentrations show additive or synergistic enhancement in solubility (Namdev *et al.,* 2022).

The next approach is to enhance solubility in solid dispersions. Solid dispersion is one of the most promising and cost-effective methods for increasing the solubility of BCS class II and IV compounds. Solid dispersions are reliable goods with at least two components: a hydrophobic medicament and a hydrophilic matrix. Amorphous or crystalline matrixes are possible. Crystalline or amorphous clusters may be used to spread the drugs molecularly throughout the matrix (Frizon *et al.,* 2013). The drug is released as extremely minute colloidal particles when exposed to aqueous fluids, and the carrier dissolves. The most widely used strategy for enhancing the dissolving behavior of poorly soluble medications is to include them in polymer matrices and produce so-called amorphous solid dispersions. Three primary techniques were established for the creation of amorphous solid dispersions formulations: solvent evaporation (spray drying) and (ii) thermal evaporation (hot melt extrusion). The dissolution rate and bioavailability of poorly water-soluble medications are expected

to be high due to the increased surface area acquired in this manner (Zhao *et al.,* 2013). Surface-active and self-emulsifying carriers can aid in forming solid dispersions, although their commercial usage has been limited.

The most extensively used approach for increasing solubility is solid dispersion complexation. Complexation is known as the association of two or more molecules to produce a nonbonded entity with well-defined stoichiometry. London forces, hydrogen bonding, and hydrophobic interactions are all used in the complexation process (Li *et al.,* 2022). Physical blending, kneading, co-precipitation, solution/solvent evaporation, neutralization precipitation, milling/co-grinding, spray drying, lyophilization, and supercritical antisolvent technique are some of the technologies used to prepare inclusion complexes of poorly water-soluble drugs with cyclodextrin (CD). The inclusion complex in CD cavity is represented in Fig. 1.6.

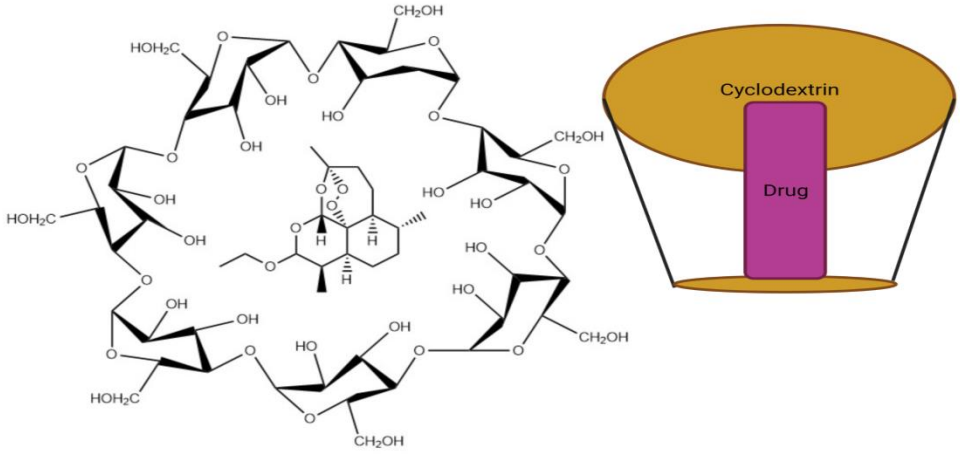

Figure 1.6: Inclusion complex in CD cavity.

The inclusion complex creation approach has been used more accurately than any other solubility enhancement technique to increase the aqueous solubility, dissolving rate, and bioavailability of weakly water-soluble medicines (Jansook and Loftsson 2022). Inclusion complexes are created when a nonpolar molecule or a nonpolar section of a molecule (known as the guest) is inserted into the cavity of another molecule or a group of molecules (known as the host). CDs are the most prevalent host molecules. CDs are cyclic oligomers produced by the enzymatic breakdown of starch by CD glycosyltransferase (CGT). CDs are cyclic oligosaccharides that are non-reducing, crystalline, and water-soluble. CDs are made up of glucose monomers

organized in a ring. α-CD, β-CD, and γ- CD are three naturally occurring CDs (Schwarz *et al.,* 2022).

CDs are cyclic oligosaccharides having a lipophilic inner chamber and a hydrophilic outer surface. They have a lipophilic core chamber and are made up of (α-1,4-) connected α-D-glucopyranose units. CD molecules are structured like cones, with secondary hydroxyl groups extending from the broader edge and primary groups extending from the narrow edge due to the chair creation of the glucopyranose units. This results in a hydrophilic outer surface for CD molecules, whereas the lipophilicity of their core cavity is equivalent to that of an aqueous ethanolic solution (Mu *et al.,* 2022).

The random replacement turns crystalline CDs into amorphous mixes of isomeric derivatives, which increases their solubility. HP-β-CD and β-CD, RAMEB (randomly methylated β-CD), sulfobutylether β-CD, and branched CD such as glucosyl-β-CD are all CD derivatives of medicinal relevance (Sun *et al.,* 2022).

Unlike CD, natural α-CD and β-CD cannot be digested by human salivary and pancreatic amylases, but they are all fermented by intestinal bacteria. At low to moderate oral doses, hydrophilic CDs are non-toxic. In topical and oral formulations, native CDs and their derivatives are employed. Still, only α-CD and the hydrophilic products of β-CD and α-CD may be used in parenteral formulations (Jansook *et al.,* 2021). CD creates visible aggregates in aqueous solutions, making it unsuitable for parenteral formulations. β-CD cannot be utilized in parenteral formulations due to its nephrotoxicity. Lipophilic CD derivatives, such as methylated CDs, are absorbed into the systemic circulation to some extent and have been proven hazardous following parenteral administration. M-β-CD oral dosing is currently limited due to its possible toxicity (Loftsson *et al.,* 2005).

By directing drugs to specific sites of action, nanotechnology-based systems give a better solution and allow for a better therapeutic outcome. When the parasitic count is large, low medication concentrations are frequently used, resulting in the development of drug resistance in malaria parasites (Ochola *et al.,* 2006).

Furthermore, nanotechnology has the potential to revive the usage of old, dangerous medications by altering and modifying their bio-distribution and thereby reducing their toxicity.

This is notably useful in malaria treatment, as new dosage forms for delivering medications to specific sites, such as parasite-infected cells, are urgently needed, particularly for antimalarials in clinical usage. Nanocarriers not only allow the use of potentially harmful but effective medications (Entezar-Almahdi *et al.,* 2021) but also improve and augment the efficacy of vaccination formulations (Carcaboso *et al.,* 2003).

Solid Lipid Nanoparticles (SLNs) has been introduced as an alternative drug carrier system suitable to enhance the solubility of water-insoluble drugs. SLNs have attracted significant concerns from scientists for their outstanding advantages over other colloidal carriers in terms of solubilized features, high stability, low bio-toxicity, and large-scale production. Therefore, applications of SLNs for oral, skin, ocular, pulmonary, and parenteral administrations have been widely developed, some of which present attractive results. Disadvantages that remain for SLNs include entrapment efficiency and encapsulation of hydrophilic materials. Lately, the novel idea of combining methods of SLNs and other polymeric systems has been initiated (Dikmen *et al.,* 2011).

Overall, SLNs have been investigated for various pharmaceutical applications, including improving the bioavailability of poorly water-soluble drugs. The SLNs systems have several advantages, including the fact that they are not readily taken up by cells in the Reticulo Endothelial System (RES) and thus bypass liver and spleen filtering, the fact that the lipid matrix is made from physiological lipid, which reduces the risk of acute and chronic toxicity, and the fact that they are easier to manufacture than bi-polymeric nanoparticles (Dikmen *et al.,* 2022). The SLNs drug delivery system enables the controlled and targeted release of encapsulated drugs. By coating or attaching ligands to SLNs, a wider range of drugs can be targeted. Moreover, the stability of the drug has been enhanced, resulting in the development of SLNs with a three-year shelf life, which is a notable improvement compared to other colloidal carrier systems (Vyas *et al.,* 2022). Chemical release kinetics are better regulated with encapsulated chemicals. SLNs may enhance the bioavailability of encapsulated

bioactive. There is excellent reproducibility when numerous procedures are used during the preparation phase. Combining hydrophilic and hydrophobic medications is a possibility. The carrier lipids are deemed safe because they are biodegradable. Organic solvents should be avoided wherever possible. Manufacturing and sterilizing on a large scale are possible. Chemical protection is offered for the labile material. SLNs, on the other hand, have several disadvantages, including Limited capacity to load drugs. Drug ejection happens during storage as a result of a polymeric transition. The water content of the dispersions is rather considerable (70- 99.9%). The capacity to load water-soluble pharmaceuticals is limited due to partitioning effects during production (Hangargekar *et al.,* 2021).

Solid lipids, emulsifiers, and water/solvent make up SLNs. Triglycerides (tri-stearin), partial glycerides (Imwitor), fatty acids (stearic acid, palmitic acid), steroids (cholesterol), and waxes are some of the lipids employed (cetyl palmitate). Several emulsifiers and their combinations (Pluronic F 68, F 127) were utilized to stabilize the lipid dispersion. The use of several emulsifiers may help to avoid particle aggregation. Hot homogenization, cold homogenization, microemulsion, solvent emulsification-diffusion, precipitation method, W/O/W double emulsion method, and spray drying method are the most common SLNs manufacturing techniques (Hosseini *et al.,* 2022).

Because of factors such as poor drug solubility and absorption, rapid metabolism, high drug plasma level fluctuation, and food effects, oral drug delivery is always looking for new ways to improve. All of these factors contribute to disappointing in vivo results and the failure of the traditional delivery system. Oral drug delivery has taken on a new dimension with the rising usage of SLNs as a carrier for administering poorly water-soluble, lipophilic medications over the last decade. Nanolipid carriers outperform all other solubility improvement approaches regarding medication solubility, dissolution, and bioavailability (Müller *et al.,* 2000). Nanostructured lipid carriers (NLCs), unlike SLNs, is constructed of a solid lipid matrix that encases a liquid lipid (oil) in their nano shell which will protect the drug (Pardeike *et al.,* 2009). The backbone of such nanocarrier systems comprises a unique architectural composition of solid and liquid lipids as well as phospholipids, which provides the ideal environment for the drug to improve therapeutic effect and also by reducing the systemic toxicity. Actretin (Agarwal *et al.,* 2010), indomethacin (Ricci *et al.,* 2005), triamcinolone acetonide (J. Araújo *et al.,* 2011) methotrexate (Garg *et al.,* 2017), and

other medications have recently been reported to be delivered using NLCs. NLCs are made up of a mixture of solid and liquid lipids, with the liquid lipid improving the solubility of poorly water-soluble medications. NLCs are commonly utilized for lipophilic medicines due to their lipophilic properties. Homogenization and melt emulsification are the most common methods for making NLCs (Garg *et al.,* 2022).

Because of their excellent drug-loading capacity and long-term stability, NLCs are preferred. Because the medicine can be placed between lipid layers or fatty acid chains, this is the case. A randomly ordered lipid matrix also aids high drug loading. Lipid nanoparticles are well-tolerated lipids such as fatty acids, triglycerides, waxes, etc. Compared to pure lipids, adding monoglycerides or triglycerides improves medication solubility (Hansapaiboon *et al.,* 2022). Nanostructured lipid-based systems provide several benefits, including drug targeting, customized drug release, a simple manufacturing technique, and flexibility for large-scale manufacture. Because of several issues with SLNs, such as poor drug loading capacity, drug ejection during storage, and high-water content.

After being synthesized through heat homogenization, SLNs undergo a process of crystallization and transformation towards a more organized structure during storage (Attama *et al.,* 2011). This transformation into a more organized form lowers crystal structural defects, which leads to drug ejection. As a result, limiting or eliminating the formation of crystals can help to avoid drug ejection. NLCs of the second generation solve this problem by having a high drug-loading capacity and long-term stability. Furthermore, lipid nanoparticles with highly concentrated dispersions demonstrated improved physical strength than those with low concentrated dispersions because highly concentrated dispersions maintain their size during storage. Again, because particles in low-concentrated dispersions are free to migrate, the risk of collisions rises, resulting in aggregation (Rodrigues *et al.,* 2021).

SMEDDS (Self Micro Emulsified Drug Delivery Systems) are formulations based on lipids. SMEDDS is an isotropic combination of medicine, oil/lipid, surfactant, and co-surfactant that, when diluted with physiological fluid, produces fine emulsion/lipid droplets ranging from roughly 100 nm to 50 nm. As a result, the medication remains in solution in the gut, bypassing the dissolving phase that slows the absorption of hydrophobic medicines in their crystalline condition (Tran *et al.,* 2021).

The nature of the oil/surfactant combination, oil/surfactant ratio, surfactant concentration, and self-emulsification temperature are all elements that influence self-emulsification. SMEDDS have been developed using a few highly particular excipient combinations. Drug integration efficiency in a SMEDDS is mainly determined by the drug/physicochemical system's compatibility. Pre-formulation solubility and phase diagram investigations are thus required to get an appropriate design (Jain *et al.,* 2022).

The primary element of SMEDDS is self-emulsification, which is mainly examined visually. Determining the emulsification rate and size distribution can be used to estimate self-emulsification efficiency (of the droplets). In a SMEDDS, the charge of the oil droplets must also be evaluated. Techniques such as XRD and differential scanning calorimetry (DSC) can assess the polymorphism and melting properties of lipids/drug in these systems (Mishra *et al.,* 2022).

Solid-SMEDDS (S-SMEDDS) refers to solid dosage forms with self-emulsification capabilities in the context of dosage forms. S-SMEDDS focuses on solidification techniques such as nanoparticle technology, adsorptions to reliable carriers, melt extrusion, and spray drying to convert liquid/semisolid simple Emulsion (SE) materials into powders/nanoparticles. Powders/nanoparticles, also known as SE nanoparticles/solid dispersions/dry emulsions, can be formed into different solid SE dosage forms or put into capsules (SE capsules) (Suram *et al.,* 2022). SE capsules, on the other hand, include capsules that are directly filled with liquid/semisolid SMEDDS without any solidifying excipients. S-SMEDDS is, in some ways, clever mixtures of SMEDDS and solid dosage forms because many of the qualities of S-SMEDDS (such as specificity, excipient selection, and characterization) are cumulative of both SMEDDS and solid dosage forms. In the case of SE pellet characterizations, self-emulsification is assessed, and attributes such as surface roughness, friability, and others are calculated. In the 1990s, S-SMEDDS such as SE solid dispersions, dry emulsions, and SE capsules were introduced. In contrast, other solid SE dosage forms such as SE microspheres/nanoparticles, SE pellets/tablets, and SE suppositories/implants were just recently introduced. Solidification procedures are utilized to convert liquid/ semisolid SMEDDS to solid SMEDDS (Yang *et al.,* 2022).

Wet mass extrusion is followed by spheronization to produce evenly sized spherical particles known as spheroids, pellets, beads, or matrix pellets, depending on the material and process. This strategy can get around bioavailability and site-specific pharmaceutical delivery issues. Extrusion spheronization is a standard method for making multi particulates for oral controlled medication delivery systems like pellets and beads (Krogars *et al.,* 2000). This technology has gained prominence in recent years due to its ease and processing speed. Long-acting, controlled-release delivery methods are frequently developed using extrusion spheronization. Extrusion spheronization produces pellets or beads that have the following benefits over traditional drug delivery systems (Gayke *et al.,* 2020).

The quality of pellets is highly influenced by the process parameters associated mainly with the extrusion stage. Parameters such as morphology, size distribution, porosity, sphericity, etc. affect the release profile and stability of pellets, while the formulation parameters such as presence and absence of soluble or insoluble fillers, surface-active agents, pH adjusters, drug load, the ratio of filler and drug influences release profile (Abbaspour *et al.,* 2014).

Enteric coating is a beneficial method for the oral administration of medications that rapidly break down in the stomach, such as insulin, since it prevents the drug from being released in the stomach's acidic environment before reaching the intestine (Rehman *et al.,* 2021).

Enteric coatings are primarily utilized to keep APIs stable when the gastrointestinal environment's acidic conditions are exposed. Erythromycin, pancreatin, and proton pump inhibitors like omeprazole are examples of APIs. They minimize the adverse effects of APIs like aspirin and some nonsteroidal anti-inflammatory drugs, such as nausea, stomach irritation, and bleeding (Khan *et al.,* 2022). Provides potential for "night-time dosing" techniques, in which the dosage form is ingested before bedtime, and adequate blood levels of the API are achieved immediately before awakening. Colonic medication delivery became easier. Enteric coatings' functioning is primarily mediated by a change in the pH of the environment in which the enteric-coated product is exposed. At low pH values, enteric polymers stay unionized (insoluble), but at pH values of 5.0–5.5, they begin to dissolve (Wang *et al.,* 2022).

Enteric film-coating polymers are poly acids that typically only dissolve in water with a pH of 5.0–6.0. These polymers are chosen for their ability to form robust coatings that adhere strongly to tablet surfaces and allow rapid drug release from dosage forms once they pass from the stomach into the small intestine (Wathoni *et al.,* 2020). The examples of aqueous enteric-coated systems is mentioned in Table 1.3.

Table 1.3: Examples of aqueous enteric-coated systems.

Product	Form	Polymer
Eudragit L30D[a]	Latex dispersion	Poly(methacrylic acid-co-ethyl acrylate) Poly (MA-EA)
Eudragit L100-55[a]	Spray-dried latex	Poly (MA-EA)
HP-F	Micronized dry powder	HPMCP
Sureteric	Dry powder system	PVAP
Acryl-Eze	Dry powder system	Poly (MA-EA)
Aquarius Control ENA	Dry powder system	Poly (MA-EA)
Aquateric	Spray-dried pseudo latex	CAP
Aquacoat ECD	Pseudo latex dispersion	CAP
Aquasolve	Micronized dry powder	HPMCAS
CAP	Dry powder	CAP

1.6 Quality by Design

Novel drug delivery systems (NDDS) is gaining popularity for treating many disorders. As a result, producing high-quality formulations by optimizing constituent compositions and manufacturing processes is a difficult challenge. The traditional method for achieving high-quality medication manufacture is altering one variable at a time (OVAT) while maintaining all other variables constant. This strategy may produce a high-quality product once or twice, but it cannot ensure that following a specific procedure will always result in high-quality output. The method and development may be suitable, but they cannot be suboptimal, and there is always the possibility of improving product quality (Gurumukhi *et al.,* 2022). Based on the Design of Experiment (DoE) concept, Quality by Design (QbD) is gaining favor in the industry for product development. For the manufacture of safe, cost-effective, and

acceptable medication production for customers, numerous industries use QbD. According to Q8 R2, QbD is "a systematic approach to development that begins with established objectives and stresses product and process understanding and control, based on strong science and quality risk management" (Gurumukhi *et al.,* 2022). It is a scientific, risk-free, proactive approach to pharmaceutical product development that begins with defining product quality and stresses understanding and regulating numerous process parameters to achieve a quality profile. QbD analyses the aspects that are crucial for product quality from the customer's perspective and convert them into qualities that the product should have, as well as how the different parameters may be adjusted in a limited way to obtain the intended product/drug target profile (Bastogne *et al.,* 2017). QbD found the links between process variables, formulation, and product characteristics and the elements that cause variability. This information may also create a more flexible, resilient, and dependable manufacturing process that consistently delivers high-quality products over time (Vogt *et al.,* 2011). Because QbD is a broad idea that extends outside the pharmaceutical business, a new vocabulary, "Formulation by Design," has been developed based on the QbD integrated principle and, more particularly, in the development of pharmaceutical dosage forms. Formulation by Design is a more precise approach to producing high-quality pharmaceuticals. The five phases are primarily responsible for achieving the objective of high-quality medication manufacture (Ainurofiq *et al.,* 2022).

Incorporating QbD into a formulation starts with Step I: determining the dosage form's goal. Several characteristics of Critical Quality Attribute (CQAs) also affect the aim or Quality Target Profile. The Ishikawa Fish Bone Diagram is used in Step II to prioritize the Critical Material Attributes (CMA) and Critical Process Parameters (CPP) that potentially have a direct impact on the response variable (CQA). It starts with determining the response variable that directly impacts product quality (e.g. homogenising time in formulation of SLNs etc.) using an experimental design (such as Plackett Burman Design, for example) to screen for relevant elements from a list of all conceivable parameters that might influence the target quality (Araújo *et al.,* 2021). This phase also contains a list of various Critical Process Parameters (CPP) required to meet the quality objectives. The range of variables is also defined using experimental design (higher and lower levels). Step III proposes an experimental design that optimizes the screening factor and level of factors (high, medium, or low)

for chosen answers that might impact the final product's quality. A well-designed matrix is created, which may aid in the development of the desired product (Prajapati *et al.,* 2021). The numerous formulations recommended by the experimental design are constructed, and the response variables chosen are assessed. Using the data acquired in Step III, an appropriate mathematical model is built in Step IV. The association between selected screening factors and response variables is discovered using response surface methodology. 2D Contour plots or 3D response surface plots graphically describe the design space. The design space is explored for optimal concentrations of the components for reaching the Quality Target Product Profile (QT PP) (Than *et al.,* 2021). The validation of the projected value of the planned experimental design is included in Step V of FbD. The optimal formulation is employed in industry for scaling-up processes and, eventually, large-scale production cycles.

The many exercises of the FbD are contained in an experimental design. The screening of critical parameters and response surface analysis of the improved formulations are part of the systematic optimization of nano-formulations. Fractional factorial design, complete fractional design, Taguchi design, and Box Behnken design are widely used for factor screening. The screening factors are typically optimized using central composite design (CCD), fractional factorial design (FFD), and box behnken design (BBD). The experiment's design is determined by the information available and the degree of control over making incorrect decisions (Type I and Type II mistakes for testing the hypothesis).

Screening aids in the identification of critical elements that assist in the achievement of the quality target product profile (QTPP). The next step is to determine the best value for the examined criteria. To accomplish the QTPP, optimization is required to obtain the optimal values of critical parameters (Dispas *et al.,* 2018). The flowchart how to select design is discussed in Fig.1.7. Factorial design, central composite design, box Behnken design, and mixture design are all options (simplex lattice, simplex centroid). A model defines the connections between the independent and response variables. Theoretical and empirical models are the most common forms of models. An empirical specifies the relationship between the variables and the response in the form of a polynomial of a specific order, commonly first, second, or third.

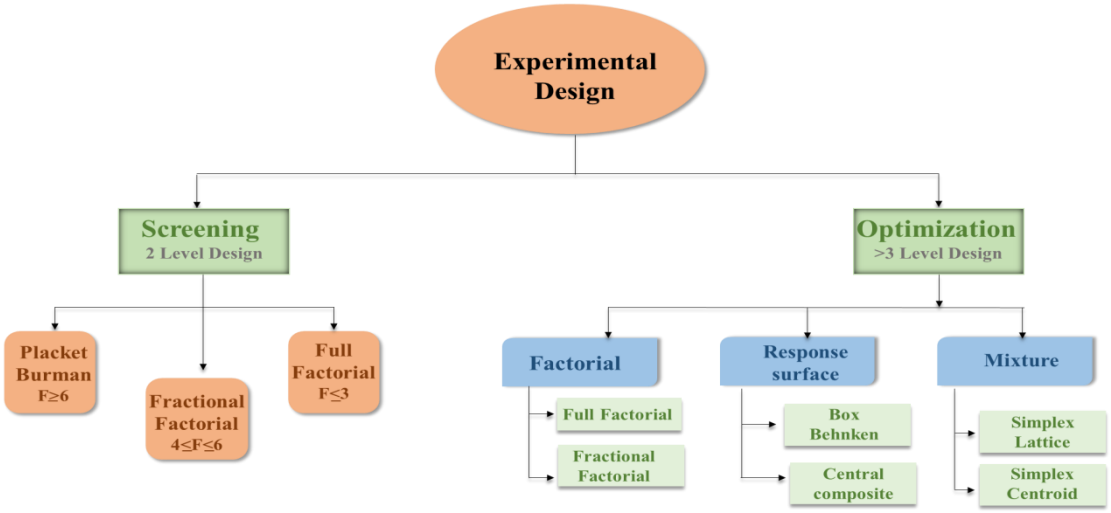

Figure 1.7: The flowchart to select experimental design.

Regression analysis is used to determine the coefficient of a quantitative component, but it is not utilized to determine the coefficient of a qualitative element. Multiple Linear Regression Analysis (MLRA) is commonly used to investigate a large number of components as well as their interactions. When a high number of variables are included in a multivariate investigation, the partial least square (PLS) approach is employed for regression analysis (Joshi *et al.,* 2021). The models are used to analyze the ANOVA, Student's t-test, and R^2 value. The graphs displayed are the main effects plots, half-normal plots, and response against time order plots. During FbD optimization, many graphical plots are commonly plotted as aids for assessing the factor's distribution. For the diagnosis of the FbD model, many graphical plots such as the Normal probability plot, Residuals vs. Predicted Plots, Residuals vs. Run, and Box-Cox Plots are shown (Namjoshi *et al.,* 2020). Graphical optimization aids in the selection of the most optimal concentration for the goal profile. To accomplish this, response variables are set to their desired limits, and the values of influencing factors are adjusted as needed.

2-D counter plots of several elements are placed on top of each other to determine the optimal design space. Overlay plots of mixed response contour plots are examples of this. The yellow hue is usually used to emphasize the permitted area (Joshi *et al.,* 2021).

Further, the response factor limitations are limited to a more acceptable range to determine the best combination that produces the needed quality target profile. Graphical optimization is promising in the case of single components, while

mathematical model optimization is more appropriate in the case of many variables. Techniques such as the desirability function and the objective function can be used. A desirability function ranges from zero outside the bounds to one at the objective. The numerical optimization locates a position where the desirability function is maximized (Dispas *et al.,* 2018).

Validation is the recorded act of establishing that a technique, process, or action will consistently provide the desired outcomes. It involves system and equipment qualification. Good manufacturing techniques and other regulatory requirements necessitate it. The goal of validation is to confirm that the product's user demands and planned applications can be met regularly (Aboushady *et al.,* 2020). The most compelling reason for validation is to ensure that all procedures and machinery in the pharmaceutical manufacturing process are employed to ensure the safety, integrity, quality, and strength of a dosage form to the greatest extent feasible (Kolekar *et al.,* 2021). If there is a substantial change to the premises, facilities, equipment, or process that may influence the quality of the product, either directly or indirectly, validation is critical. It is vital that the validation program is documented and the documentation is kept up to date. The method should be approved and released for use in ordinary production after thoroughly evaluating all validation documents, including data from equipment qualification and product/package performance testing to confirm compatibility with the process. It's critical to keep track of process details (such as time, speed, and equipment) and any changes that occur throughout ordinary production (Beg *et al.,* 2020).