Int. J. Pharm. Investigation, 10.5530/ijpi.20270097.
https://www.jpionline.org

Review Article

Drug Nanocrystals: From Formulation Design to Clinical Translation

Kamalesh Dilip Mali*, Sachin Yadav, Arya Nilesh Dubey, Gaurav Bharat Deore,

Department of Pharmaceutics, R. C. Patel Institute of Pharmacy, Shirpur, Dhule, Maharashtra, INDIA.

ABSTRACT

Poor aqueous solubility remains a critical challenge in pharmaceutical development, leading to limited drug absorption, high variability in systemic exposure, and reduced therapeutic efficiency. Conventional approaches such as salt formation, amorphous systems, and lipid-based formulations often face limitations related to stability, scalability, and regulatory complexity. Drug nanocrystals have emerged as a clinically established strategy to address these challenges by enhancing dissolution and bioavailability without altering the intrinsic pharmacological properties of the drug. This review provides a structured and critical overview of drug nanocrystal technology, encompassing fabrication methods, stabilization strategies, surface engineering, biological fate, and route-specific applications. Emphasis is placed on distinguishing mechanistic understanding from translational outcomes, supported by representative studies and marketed products. The review further discusses regulatory considerations, safety aspects, and factors influencing clinical translation. By integrating formulation design with *in vivo* behavior and regulatory perspectives, this work highlights the role of nanocrystals as a scalable and effective platform for poorly soluble drugs. Current challenges, including stability, targeting efficiency, and manufacturing reproducibility, are critically addressed, and future directions are outlined to support continued advancement and broader clinical adoption.

Keywords: Bioavailability, Biological Fate, Drug nanocrystals, Nanocrystal Preparation, Solubility.

Correspondence:
Prof. Kamalesh Dilip Mali
Department of Pharmaceutics, R. C. Patel Institute of Pharmacy, Shirpur, Dhule-425405, Maharashtra, INDIA.
Email: kamaleshmalipharma@gmail.com

Received: 12-04-2026;
Revised: 06-06-2026;
Accepted: 23-08-2026.

INTRODUCTION

Poor aqueous solubility remains one of the most significant challenges in contemporary pharmaceutical development and is a major contributor to formulation failure and inconsistent therapeutic outcomes. It is estimated that nearly 70-90% of Newly Discovered Chemical Entities (NCEs) fall under Biopharmaceutics Classification System (BCS) class II or IV, where drug absorption is primarily limited by dissolution rather than permeability (Rathod *et al*., 2025; Reza Pahlevi *et al*., 2023). As a result, many promising drug candidates exhibit low and variable oral bioavailability, ultimately affecting clinical efficacy and increasing the risk of attrition during development (Deshmukh *et al*., 2017).

To address solubility-limited absorption, a variety of formulation strategies have been investigated, including salt formation, pH modification, co-solvent systems, cyclodextrin complexation, amorphous solid dispersions, and lipid-based delivery systems. While these approaches have proven effective in specific circumstances, their broader application is often restricted by intrinsic limitations. For example, salt formation and prodrug strategies are highly dependent on molecular structure and may introduce chemical instability or regulatory complexity (Noreen *et al*., 2016; Varma *et al*., 2026). Amorphous solid dispersions, while capable of improving dissolution, are thermodynamically unstable and prone to recrystallization during storage or after administration, which can compromise long-term performance. Similarly, lipid-based and polymeric nanocarrier systems often require high excipient concentrations, leading to reduced drug loading, increased formulation complexity, and variability *in vivo* performance. These challenges highlight the need for formulation approaches that can improve bioavailability while maintaining stability, scalability, and regulatory feasibility (B and Ghosh, 2025).

In this context, drug nanocrystals have emerged as a potential and clinically confirmed approach for increasing the bioavailability of poorly soluble drugs while preserving their chemical structure. Nanocrystals are submicron-sized crystalline particles composed predominantly of the drug, typically stabilized by a small amount of surfactant or polymer to prevent aggregation (Eckert *et al*., 2020; Gigliobianco *et al*., 2018). This carrier-free nature distinguishes nanocrystals from conventional nanosystems and enables high drug loading, reduced excipient burden, and simplified formulation. Importantly, several nanocrystal-based

OPEN ACCESS

Manuscript TECHNOMEDIA

DOI: 10.5530/ijpi.20270097

Copyright Information :
Copyright Author (s) 2027 Distributed under Creative Commons CC-BY 4.0

Publishing Partner : Manuscript Technomedia. [www.mstechnomedia.com]

products have been successfully translated into clinical use, showing improved pharmacokinetic profiles and enhanced therapeutic consistency across different routes of administration (Zhou *et al.*, 2016).

The improved performance of nanocrystals (Figure 1) is mostly due to particle size reduction to the nanometer range, which alters drug behavior using well-established physicochemical principles. Nanosizing increases surface area and apparent saturation solubility, thereby accelerating dissolution and generating higher concentration gradients at absorption sites (Pinarbasli and Doganay, 2021). In addition, nanocrystals exhibit improved adhesion to biological membranes and prolonged residence time at absorption interfaces, which can further enhance drug uptake. Unlike amorphous systems, nanocrystals retain their crystalline structure, offering improved physical stability and lowering the possibility of performance loss due to recrystallization (Sun *et al.*, 2018) .

From a translational perspective, nanocrystals represent a practical balance between conventional formulations and complex nanocarrier systems. They can be produced using scalable and well-established techniques such as wet media milling and high-pressure homogenization, which are compatible with existing pharmaceutical manufacturing infrastructure (Srivalli and Mishra, 2015). Furthermore, regulatory evaluation of nanocrystal-based products is generally based on established quality attributes, including particle size distribution, crystallinity, and dissolution performance, facilitating their clinical translation (Dhaval *et al.*, 2020).

Despite these advantages, the successful development of nanocrystal formulations requires a comprehensive understanding of multiple interrelated factors, including fabrication technologies, stabilizer selection, surface characteristics, biological interactions, and safety considerations. These aspects collectively determine the *in vivo* fate and therapeutic performance of nanocrystal systems (Dhibar *et al.*, 2023).

In this context, the present review provides a systematic and critical overview of drug nanocrystals as a strategy for enhancing bioavailability. Emphasis is placed on fabrication technologies, mechanistic basis of dissolution and absorption enhancement, biological fate, regulatory considerations, and clinical translation. By integrating fundamental principles with practical and translational insights, this review provides a comprehensive framework for the rational design and development of nanocrystal-based drug delivery systems.

Search Strategy

A systematic search was conducted across primary electronic databases, including PubMed, Scopus, and Web of Science, to identify relevant literature published up to 2025. Search terms included "drug nanocrystals," "nanosuspensions," "bioavailability enhancement," and "clinical translation." Articles were selected based on their relevance to fabrication technologies, stability mechanisms, and route-specific applications. Preference was given to peer-reviewed research papers and recent systematic reviews that provided quantitative outcomes and translational insights.

Limitations of Conventional Bioavailability Enhancement Techniques

In pharmaceutical development, increasing the bioavailability of weakly water-soluble drugs has long been a problem, especially for substances with dissolution-limited absorption. Several formulation approaches have been studied, including molecular modification, solid-state manipulation, and colloidal delivery systems. Despite the fact that these methods have proven effective in certain situations, their wider applicability is still limited by intrinsic physicochemical and practical constraints (Verma, 2022).

Molecular-level methods such as salt creation, prodrug design, and cyclodextrin complexation try to improve apparent solubility by altering drug-solvent interactions (Wankhade and Dudhe, 2024). However, their efficacy is dependent on drug chemistry. For example, salt formation is restricted to ionizable drugs, but co-solvent systems may precipitate upon dilution, resulting in variable *in vivo* efficacy and potential toxicity. Prodrug methods, while effective, frequently increase regulatory complication (Gamsiz *et al.*, 2010; Jornada *et al.*, 2015).

Solid-state approaches, such as amorphous solid dispersions and cocrystals, improve solubility by enhancing the drug's energy state. Despite their advantages, these complexes are thermodynamically unstable and susceptible to recrystallization, which could risk long-term stability and bioavailability. This requires tight control over formulation and storage conditions, which limits their industrial robustness (Bookwala and Wildfong, 2023).

Colloidal and lipid-based systems, such as emulsions and polymeric nanoparticles, improve solubilization through carrier-mediated delivery. However, these systems typically require high excipient concentrations, resulting in low drug loading and increased formulation complexity. Additionally, variability in *in vivo* results and challenges related to scale-up and stability further restrict their widespread application (Losada-Barreiro *et al.*, 2024).

Overall, the limitations of conventional approaches highlight (Table 1) the need for alternative strategies that offer improved stability, scalability, and drug loading. Drug nanocrystals solve many of these issues by enhancing dissolution and absorption through particle size reduction without altering the drug's chemical structure.

Fundamentals of Drug Nanocrystals and Mechanistic Basis of Bioavailability Enhancement

Poor aqueous solubility remains a critical limitation in drug development, particularly for Biopharmaceutics Classification System (BCS) class II and IV compounds, where dissolution governs absorption. Drug nanocrystals address this limitation by reducing particle size to the nanometer scale while preserving the crystalline structure of the active pharmaceutical ingredient. Unlike carrier-based nanosystems, nanocrystals consist predominantly of the drug itself, enabling high drug loading with minimal excipient requirement (Deljavan Ghodrati and Turk, 2024).

Nanocrystals are generally defined as particles in the size range of 50-500 nm. Reduction to this scale alters drug behavior through both thermodynamic and kinetic mechanisms. According to the Ostwald-Freundlich equation, decreasing particle size increases surface tension and surface free energy, resulting in improving apparent saturation solubility, especially below ~200 nm. In parallel, the Noyes-Whitney equation explains the increase in dissolution velocity due to more surface area and reduced diffusion layer thickness. Together, these effects provide high concentration gradients at the absorption site, facilitating drug diffusion across biological membranes (Sivanathan *et al.*, 2024).

Beyond dissolution, nanosizing influences cellular uptake and biodistribution. Wang *et al.* (2023) demonstrated a clear size-dependent uptake, where ~70 nm nanocrystals showed significantly higher internalization compared to ~200 nm particles, with rapid cellular entry observed within minutes in both *in vitro* and zebrafish models (Wang *et al.*, 2023). Similarly, Hollis *et al.* reported that paclitaxel nanocrystals (~200 nm) accumulated in tumor tissue within 1-6 hr, although substantial distribution in liver and kidneys highlighted limitations of passive targeting mechanisms such as the EPR effect (Hollis *et al.*, 2013).

Formulation-based studies further support the performance advantages of nanocrystals. Mauludin *et al.* showed improved dissolution of Rutin nanocrystal tablets compared to conventional formulations (Mauludin *et al.*, 2008). Dolenc *et al.* reported enhanced dissolution of celecoxib nanocrystals stabilized with surfactants (Dolenc *et al.*, 2009), while Kocbek *et al.* reported a marked increase in drug release (>65% within 10 min) compared to micronized drug (<15%), confirming the impact of nanosizing on dissolution kinetics (Kocbek *et al.*, 2006). In addition, long *et al.* developed functionalized cellulose nanocrystals for doxorubicin delivery, achieving elevated cellular uptake and improved anticancer activity (Long *et al.*, 2021).

Collectively, these findings indicate that nanocrystals improve drug performance through integrated mechanisms involving enhanced solubility, faster dissolution, and improved cellular uptake. Representative quantitative outcomes are summarized in Table 2.

Fabrication Technologies for Drug Nanocrystals

Nanocrystal fabrication involves a variety of approaches to modifying particle size, morphology, and surface properties, all of which have a direct impact on drug solubility, dissolution rate, and bioavailability. These approaches have changed dramatically in response to advances in pharmaceutical engineering and nanotechnology, allowing for the creation of robust and scalable drug delivery systems. Nanocrystal fabrication approaches are broadly classified into top-down, bottom-up, and hybrid techniques, each offering distinct advantages and limitations depending on drug characteristics and process requirements (Bhaskar *et al.*, 2025).

Top-down approaches include mechanical size reduction of bulk drug particles, whereas bottom-up techniques rely on controlled nucleation and crystal growth from supersaturated solutions. More recently, emerging and hybrid technologies have been developed to combine the benefits of both approaches while resolving their individual limitations (Chen *et al.*, 2025). A comparative overview of major nanocrystal preparation methods is summarized in Table 3 and Figure 2.

Microfluidization

Microfluidization is a top-down method that uses high-pressure fluid dynamics to reduce particle size via shear, cavitation, and collision in microchannels. This approach produces nanocrystals with a narrow size distribution and good repeatability; however, its performance is highly dependent on process parameters like as pressure and cycle count. Liu *et al.* (2021) reported the production of siRNA-loaded lipid nanocarriers with an average size of ~164.2 nm and PDI of 0.23, demonstrating uniformity and stability for up to one month (Liu *et al.*, 2021). Similarly, Chen *et al.* achieved curcumin nanoliposomes of 68.1 nm with a PDI of 0.246, exhibiting enhanced stability under acidic conditions (Chen *et al.*, 2015; Khadke *et al.*, 2020). Further showed that increasing pressure from 15,000 to 20,000 psi and multiple processing cycles significantly reduced particle size and PDI, highlighting the sensitivity of this method to operational conditions. While microfluidization is scalable and suitable for lipid-based systems, its dependence on precise process optimization remains a limitation (Khadke *et al.*, 2020).

Milling Methods (Wet Media Milling)

Wet Media Milling (WBM), widely known as NanoCrystal technology, is one of the most established top-down techniques for nanocrystal production. It relies on mechanical attrition in the presence of stabilizers and is applicable to a broad range of poorly soluble drugs.

Mura *et al.* demonstrated that wet milling produced glyburide nanocrystals (~180 nm) with superior dissolution compared to dry milling (~120 nm), achieving 28% drug release within 10 min versus 14% for dry milling (Mura *et al.*, 2023). This indicates

that wettability and stabilizer interaction can outweigh particle size alone in determining performance. Similarly, Toziopoulou *et al.* (2017) reported that stabilizer such as HPMC and HPC significantly improved size reduction compared to PVP K30 (Toziopoulou *et al.*, 2017), underscoring the importance of formulation components. Despite its industrial stability and scalability, milling is energy-intensive process that can introduce contamination from milling media.

Hot-Melt Extrusion (HME)

Hot-Melt Extrusion (HME) is a hybrid technique that combines thermal and mechanical processing to create nanocrystal-based solid dispersions. Its solvent-free and continuous nature makes it attractive for industrial applications.

Baumgartner *et al.* reported that phenytoin nanocrystals remained uniformly dispersed within polymer matrices without loss of crystallinity, leading to improved dissolution. Patil *et al.* further reported 92% drug release within 5 hr for fenofibrate-loaded systems, compared to 62% from marketed formulations and 41% from pure drug (Baumgartner *et al.*, 2014), showing the efficiency of extrusion-based approaches. However (Ye *et al.*, 2015), showed that process parameters such as feed rate significantly influence energy input and product quality (Ye *et al.*, 2015). However, these advantages, thermal degradation and polymorphic transitions remain major limitations, particularly for heat-sensitive drugs.

Nanoprecipitation

Nanoprecipitation is a bottom-up technique based on rapid supersaturation and nucleation following solvent-antisolvent mixing. It offers precise control over particle size but requires strict control of process conditions.

Kumar *et al.* (2022) developed niclosamide nanocrystals using nanoprecipitation, stabilized with hyaluronic acid and PVP/PVA. The optimized formulation (173.3±10.2 nm; PDI 0.221±0.09) was incorporated into Carbopol 940 hydrogels for topical delivery. Using a quality-by-design approach, three formulations were optimized, exhibiting shear-thinning behavior and Higuchi-model drug release (≈71-86% over 12 hr). Nanocrystal conversion significantly improved efficacy, reducing the IC_{50} from 1.25 µM to ~0.017-0.023 µM in HaCaT cells, indicating improved bioavailability and anticancer potential (Kumar *et al.*, 2022). Similarly, Panwar *et al.* (2023) developed Hyaluronic Acid (HA)-modified Venetoclax Nanocrystals (HVNCs) using the nanoprecipitation method, enabling CD44-targeted delivery in breast cancer. HA coating increased particle size (~132 nm to ~173 nm) and improved targeting. The formulation showed Higuchi-type drug release and good hemocompatibility. HVNCs significantly enhanced anticancer activity, reducing IC_{50} ~9.7-fold compared to free drug and promoting higher apoptosis and cell cycle arrest (Panwar *et al.*, 2023). While nanoprecipitation is versatile and efficient, issues such as aggregation, solvent removal, and scale-up remain major challenges.

Spray Drying

Spray drying is a scalable bottom-up technique that converts liquid dispersions into dry powders via rapid solvent evaporation. It is particularly suitable for heat-sensitive compounds due to short exposure times. Shete *et al.* (2015) developed Hesperetin nanocrystalline solid dispersions using a bottom-up spray drying method with mannitol. The formulation significantly increased dissolution and oral bioavailability (≈1.8-fold C_{max}, 2.25-fold AUC). Improved intestinal uptake and cellular permeability were observed, along with elevated antioxidant and anti-inflammatory effects, highlighting its therapeutic potential (Shete *et al.*, 2015).

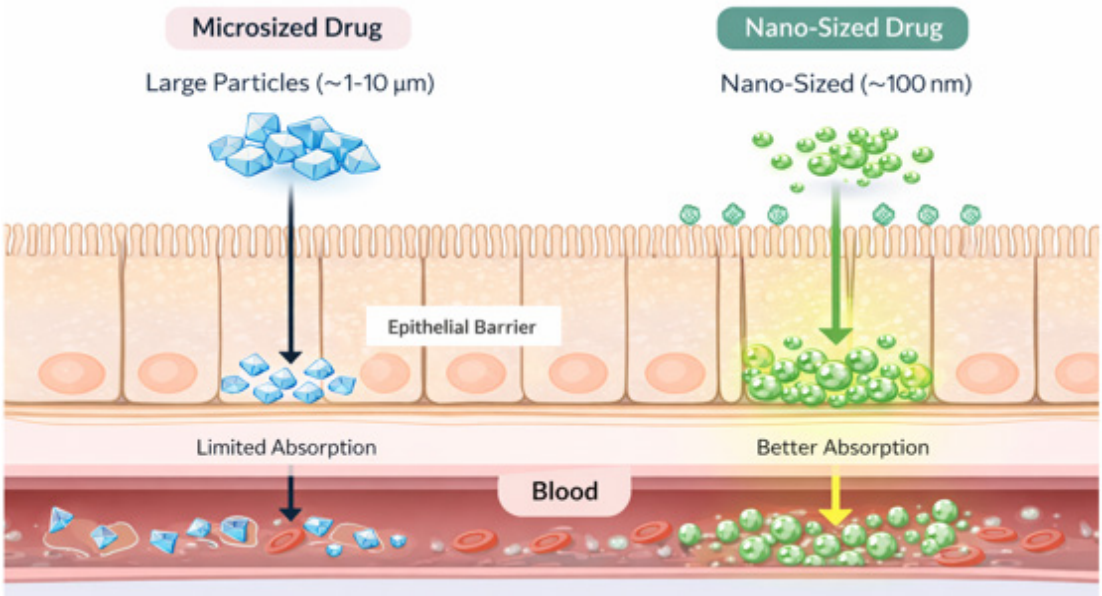


Figure 1: Absorption of nano sized and microsized drug.



Figure 2: Fabrication technologies for drug nanocrystals.

This method also decreases the need for stabilizers, which improves formulation simplicity. However, particle aggregation and process control remain critical concerns.

Supercritical Fluid Technology (SCF)

Supercritical Fluid (SCF) technology is a sophisticated and ecologically friendly technique to nanocrystal production. Supercritical CO_2 is commonly employed for its non-toxic properties and moderate critical conditions. Lu *et al.* created paclitaxel-loaded chitosan and HTCC nanoparticles using SCF-assisted processing and an O/W/O double emulsion with temperature-induced solidification. HTCC-NPs surpassed other nanoparticles (~130 nm, PDI<0.1) in terms of permeability, cell uptake, and tumor accumulation. Bioavailability increased by up to ~9.6%, with reduced hypersensitivity and higher cytotoxicity, suggesting promise for targeted cancer delivery (Lv *et al.*, 2011). Despite advantages such as solvent-free processing and high purity, limitations related to high equipment cost and limited drug solubility restrict widespread industrial application.

Drug Nanocrystal Stability

Drug Nanocrystals (NCs), due to their nanoscale size and increased surface area, have rapid dissolving rates and higher bioavailability than bulk drug forms. However, these benefits are coupled by thermodynamic instability, as increased surface free energy favors aggregation and crystal development. Ostwald ripening is an important instability mechanism in which smaller particles aggregated under supersaturated conditions, resulting in particle growth and loss of nanoscale advantages. This phenomenon may also raise safety concerns, particularly for intravenous formulations, where aggregation could potentially result in vascular complications (Shahiwala, 2025; Yang *et al.*, 2018).

To overcome these challenges, the incorporation of stabilizers is essential. Stabilizers reduce interfacial free energy and prevent aggregation through electrostatic or steric mechanisms. Ionic surfactants such as Sodium Dodecyl Sulfate (SDS) (Kojima *et al.*, 2018) provide electrostatic repulsion, whereas non-ionic surfactants (e.g., Tween, TPGS) (Rachmawati *et al.*, 2013; S. Sharma *et al.*, 2015) and polymeric stabilizers (e.g., Polyvinyl Alcohol (PVA), Polyvinylpyrrolidone (PVP) (Kumar and Siril, 2017), Hydroxypropyl Methylcellulose (HPMC) (Ueda *et al.*, 2019), Polyethylene Glycol (PEG)) (Wang *et al.*, 2020; Z. Zhang *et al.*, 2020) offer steric stabilization by forming protective layers around nanocrystals. Apart from physical stabilization, several excipients offer additional functional benefits. For example, TPGS has been shown to hinder P-glycoprotein (P-gp) efflux, increasing intracellular drug accumulation and overcoming multidrug resistance. Similarly, PEGylation lowers absorption by the Reticuloendothelial System (RES), extending systemic circulation; nevertheless, repeated exposure may result in Accelerated Blood Clearance (ABC), limiting long-term therapeutic efficacy. The Molecular Weight (MW) of polymeric stabilizers is particularly important, as lower MW polymers promote mucosal diffusion and oral absorption while higher MW polymers give stronger steric stabilization at lower doses. Additionally, stabilizers that improve both solubility and dissolution are wanted for maximum bioavailability. The growing interest in natural stabilizers like saponins indicates a trend toward safer and more biocompatible options. Table 4 summarizes the classification, processes, and functional functions of commonly employed stabilizers in nanocrystal formulations, emphasizing

Table 1: Limitations of Conventional Bioavailability Enhancement Techniques.

Approach	Key Limitation	Impact on Drug Delivery
Salt formation	Applicable only to ionizable drugs	Limited universality
Co-solvent systems	Risk of precipitation and toxicity	Unpredictable absorption
Amorphous dispersions	Thermodynamic instability	Recrystallization risk
Co-crystals	Stability and regulatory concerns	Variable performance
Lipid-based systems	High excipient requirement	Low drug loading

Table 2: Representative Studies on Bioavailability Enhancement by Nanocrystals.

Drug	Key Quantitative Outcome	References
Coumarin	Smaller particles (~70 nm) showed higher cellular uptake compared to larger (~200 nm), confirming size-dependent uptake	Wang *et al.*, 2023
Paclitaxel	Rapid tumor accumulation within 1 to 6 hr with evidence of EPR effect and off-target distribution	Hollis *et al.*, 2013
Rutin	Faster dissolution compared to conventional formulations, leading to improved oral performance	Mauludin *et al.*, 2008
Celecoxib	Higher dissolution compared to micronized drug, indicating enhanced solubility	Dolenc *et al.*, 2009
Ibuprofen	More than 65% drug release within 10 min compared to less than 15% for coarse drug, showing rapid dissolution	Kocbek *et al.*, 2006
Doxorubicin	Increased cellular uptake and targeting efficiency, demonstrating improved anticancer potential	Long *et al.*, 2021

their importance in preserving stability as well as impacting drug delivery performance.

Surface Engineering for Functional and Targeted Nanocrystals

Surface engineering of drug nanocrystals has emerged as an important technique for overcoming conventional systems' disadvantages, such as fast dissolution, non-specific distribution, and clearance by the mononuclear phagocyte system (MPS). While conventional stabilizers (surfactants and polymers) primarily maintain colloidal stability, they lack biological functionality. Thus, the incorporation of targeted ligands onto nanocrystal surfaces has become vital for improving pharmacokinetics and therapeutic specificity.

Because of their high surface energy, nanocrystals are intrinsically unstable and must be stabilized using steric or electrostatic methods. These interactions, guided by DLVO theory, keep the dispersion stable by balancing attractive and repulsive forces. However, effective surface engineering goes beyond stabilization to include rational ligand design, such as strong surface stabilization, resistance to protein adsorption, and controlled release of drug for targeting.

Folic acid (FA) is a frequently targeted ligand due to the activation of folate receptors in cancer cells. FA-functionalized nanocrystals have increased tumor accumulation and receptor-mediated uptake, as seen in sorafenib and paclitaxel systems, allowing for better intracellular drug release and therapeutic efficacy (Liang *et al.*, 2021; Setua *et al.*, 2009; Zhao *et al.*, 2021). Similarly, Hyaluronic Acid (HA), targeting CD44 receptors, has found to improve cellular internalization and tumor inhibition, particularly in lapatinib-based formulations (Agrawal *et al.*, 2017; Seo *et al.*, 2020).

Other polysaccharides such as chondroitin sulfate also use CD44-mediated uptake to achieve selective tumor targeting (Liang *et al.*, 2020; *et al.*, 2022). Natural polymers like alginate and chitosan provide additional benefits by allowing pH-responsive behavior and protecting nanocrystals in adverse physiological environments, hence increase oral bioavailability (Lin *et al.*, 2011; Ryu *et al.*, 2020).

Protein-based ligands, such as Transferrin (Tf), promote receptor-mediated uptake in proliferating cells while also delaying systemic circulation (Han *et al.*, 2020; Yong *et al.*, 2019). Triphenylphosphonium (TPP) allows for mitochondrial targeting, which is very useful in decreasing multidrug resistance by interrupting cellular energy pathways. Emerging techniques, such as entropic ligands, broaden functionality by modifying thermodynamic features to increase nanocrystal solubility (Yang *et al.*, 2016; Yang *et al.*, 2016).

Despite these improvements, obstacles remain, such as ligand detachment, protein corona formation, and heterogeneity in receptor expression, all of which can limit targeting efficacy *in vivo* conditions. Furthermore, repeatability and large-scale

production of surface-functionalized nanocrystals remain major obstacles. Addressing these obstacles will necessitate the creation of multifunctional, stimuli-responsive systems that combine stability, targeting, and controlled drug release. As shown in Table 5, diverse ligand systems enable the functionalization of nanocrystals for targeted and responsive drug delivery.

Biological Fate and *in vivo* Performance of Drug Nanocrystals

The *in vivo* behavior of drug nanocrystals has traditionally been interpreted based on rapid dissolution at the site of administration, leading to enhanced concentration gradients and improved drug absorption. Early pharmacokinetic studies supported this assumption, reporting comparable profiles between nanocrystal formulations and drug solutions, suggesting near-complete dissolution in biological environments (Liu *et al*., 2012). However, recent advances in imaging and tracking techniques have significantly revised this understanding.

Following oral administration, nanosizing has been found to significantly increase bioavailability. For example, danazol nanocrystals (~169 nm) showed an approximately 16-fold increase in oral bioavailability, due to improved dissolution (Liversidge & Cundy, 1995). Despite this traditional rationale, more recent research suggests that nanocrystals can also be absorbed in their intact form. Imaging data has identified nanocrystals in intestinal villi and mesenteric lymph, implying transcellular transport via endocytosis processes such as caveolae-mediated uptake (Fu *et al*., 2013; G. Zhang *et al*., 2021). These findings showing that oral absorption is governed by a dual mechanism involving both dissolution and particle-mediated transport.

In the case of intravenous administration, early interpretations similarly emphasized rapid dissolution. However, subsequent studies have demonstrated prolonged persistence of intact nanocrystals *in vivo*. For instance, itraconazole nanocrystals were detected in splenic macrophages up to 7 days post-administration, highlighting the dominant role of the Mononuclear Phagocyte System (MPS) in determining biodistribution (Rabinow *et al*., 2007). Particle size further influences pharmacokinetics, where larger nanocrystals exhibit increased Area Under the Curve (AUC), Mean Residence Time (MRT), and half-life due to slower dissolution and enhanced phagocytic uptake (Liu *et al*., 2012). Moreover, it has been estimated that nanocrystals require 5-10 min for complete dissolution, whereas systemic circulation occurs within seconds, allowing repeated circulation before dissolution. Consequently, up to ~95% of the administered dose can accumulate in liver and spleen within minutes, indicating rapid clearance by the MPS (Manjunath and Venkateswarlu, 2006).

For dermal delivery, nanocrystals predominantly accumulate within hair follicles rather than diffusing directly through the skin. Studies have demonstrated follicular penetration depths of approximately 196 μm, where nanocrystals act as drug reservoirs and release the drug gradually (Pelikh and Keck, 2020). Similarly, in ocular delivery, nanocrystals exhibit enhanced adhesion and prolonged retention, resulting in approximately two-fold higher bioavailability and extended therapeutic duration (up to 9 hr compared to 5 hr) (Donia *et al*., 2020). At the cellular level, nanocrystals undergo endocytosis-mediated uptake and intracellular trafficking through organelles such as the endoplasmic reticulum and Golgi apparatus, enabling controlled intracellular drug release (Deng *et al*., 2017).

Table 3: Overview of Nanocrystal Preparation Techniques.

Method (Approach)	Principle	Key Advantage	Limitation	References
Microfluidization (Top-down)	High-pressure shear, cavitation, and collision in microchannels	Narrow size distribution; high reproducibility; scalable	Sensitive to pressure and cycle parameters	Liu *et al*., 2021; Chen *et al*., 2015; Khadke *et al*., 2020
Wet Media Milling (Top-down)	Mechanical attrition using milling media with stabilizers	Industrially established; applicable to poorly soluble drugs	Energy-intensive; contamination risk	Mura *et al*., 2023; Toziopoulou *et al*., 2017
Hot-Melt Extrusion (Hybrid)	Thermal + mechanical processing to form solid dispersions	Solvent-free; continuous and scalable process	Thermal degradation; polymorphic changes	Baumgartner *et al*., 2014; Ye *et al*., 2015
Nanoprecipitation (Bottom-up)	Supersaturation via solvent-antisolvent mixing	Precise particle size control; versatile	Aggregation; solvent removal; scale-up issues	Kumar *et al*., 2022; Panwar *et al*., 2023
Spray Drying (Bottom-up)	Rapid solvent evaporation to form dry nanocrystals	Scalable; suitable for heat-sensitive drugs	Particle aggregation; process control challenges	Shete *et al*., 2015
Supercritical Fluid (SCF) (Advanced)	Rapid precipitation using supercritical CO_2	Solvent-free; high purity; controlled particle size	High equipment cost; limited drug solubility	Lv *et al*., 2011

Table 4: Stabilizers and their Role in Enhancing Stability of Drug Nanocrystals.

Stabilizer Type	Examples	Key Role / Benefit	References
Ionic surfactants	SDS	Electrostatic stabilization: improves dispersion but may cause irritation	Kojima *et al.*, 2018
Non-ionic surfactants	Tween, TPGS	Steric stabilization; improves solubility; TPGS helps overcome drug resistance (P-gp inhibition)	Rachmawati *et al.*, 2013; Sharma *et al.*, 2015
Polymeric stabilizers	HPMC, PVP, PVA	Steric hindrance; enhances stability, bioavailability, and controlled release	Kumar and Siril, 2017; Ueda *et al.*, 2019
PEGylated systems	PEG conjugates	Stealth effect: prolongs circulation and reduces RES uptake	Wang *et al.*, 2020; Zhang *et al.*, 2020
Natural stabilizers	Saponins	Biocompatible stabilization with low toxicity	Chen *et al.*, 2017; Jin *et al.*, 2018

Overall, the biological fate of nanocrystals is determined by a complicated interaction of dissolution kinetics and particle-mediated transport mechanisms. This dual-purpose sets nanocrystals apart from conventional formulations and points out their potential for controlled and targeted drug delivery. The important mechanistic discoveries from various administration methods are given in Table 6 and Figure 3.

Drug Nanocrystals: Translational Performance and Clinical Utility

Drug nanocrystals have been effectively converted into clinically useful dosage forms via a variety of modes of administration, including oral and parenteral delivery. Their primary advantage is improved bioavailability, reduced pharmacokinetic variability, and constant drug administration without affecting intrinsic pharmacological efficacy. Nanocrystals in oral delivery reduce food effects and improve dissolution-limited absorption, whilst in parenteral systems they eliminate the requirement for hazardous solubilizing excipients and allow for controlled drug release. Collectively, these characteristics help to increase therapeutic performance and patient compliance (Chary *et al.*, 2024). Several marketed products illustrate flexibility and translated success of nanocrystal technology across various administration routes, as shown in Table 7.

Applications of Drug Nanocrystals

Drug nanocrystals have emerged as a clinically important strategy (Figure 4) for increasing the therapeutic effectiveness of poorly water-soluble drugs in a variety of delivery routes. Unlike mechanistic explanations that focus on *in vivo* fate, their applications are generally defined by increases in pharmacokinetic uniformity, therapeutic efficacy, dose reduction, and patient compliance (Dhibar *et al.*, 2023). Table 8 summarizes the route-specific advantages of nanocrystal-based systems, allowing them to be translated into realistic drug delivery solutions.

Nanocrystals in oral delivery of drugs have significantly boosted the efficiency of poorly soluble drugs by increasing dissolution-limited absorption and decreasing gastrointestinal variability. For example, baicalein nano co-crystals showed a 6.09-fold increase in bioavailability compared to pure drug and 3.32-fold compared to conventional nanocrystals, highlighting the advantage of combining crystal engineering with nanosizing (Pi *et al.*, 2018). Similarly, Silybin nanocrystals (HM40) produced by wet milling showed improved solubility, prolong stability over 24 months, and significantly improved oral bioavailability (2.61-fold in rats; 1.51-fold in humans), demonstrating effective clinical translation potential (Seo *et al.*, 2024). Lutein nanocrystals (~110.7 nm) produced by antisolvent precipitation significantly enhanced solubility (7.3 to 215.7 µg/mL) and dissolution, resulting in improved oral bioavailability in rat models, with ~3.24-fold increase in C_{max} and ~2.28-fold increase in AUC. These findings highlight nanosizing as an effective strategy for overcoming solubility-limited absorption (Chang *et al.*, 2017). Lutein nanocrystals (~230 nm) significantly improved dissolution and oral bioavailability (~15.2-fold) in Sprague-Dawley rats, with enhanced ocular accumulation (~3.4-fold), this suggesting nanosizing as an effective strategy for improving systemic and ocular delivery of poorly soluble nutraceuticals (Seto *et al.*, 2019). Collectively, these findings show that oral nanocrystals provide constant drug release and reduce interpatient variability while maintaining pharmacological activity.

Parenteral nanocrystals represent an important milestone in systemic drug administration, particularly in oncology and long-acting drugs that require fast exposure and controlled pharmacokinetics. These systems, which typically vary in size from 100 to 300 nm, have prolonged circulation due to delayed dissolution, acting as particle reservoirs which allow controlled drug release. Surface functionalization with Polyethylene Glycol (PEG) and targeted ligands promotes tumor accumulation through Enhanced Permeability and Retention (EPR), while lowering premature clearance (Lu *et al.*, 2016).

Nanocrystals show significant improvements in therapeutic efficiency and dose modification. Anlotinib nanocrystals (~200 nm) showed strong anticancer activity in a nude mouse hepatocellular carcinoma model, achieving tumor suppression at a significantly lower dose (50 µg/kg) compared to oral solution (1

mg/kg), resulting in a ~20-fold dose reduction (Luo *et al.*, 2021). Similarly, paclitaxel nanocrystals stabilized with TPGS showed sustained release and enhanced cytotoxicity in resistant H460 cells, as well, improved tumor suppression in xenograft models (Liu *et al.*, 2010), while camptothecin nanocrystals showed superior efficacy in MCF-7 tumor-bearing mice (Zhang *et al.*, 2011).

The clinical potential of nanocrystals is further revealed by prolonged-acting uses. In mice and rats, cabotegravir nanocrystal prodrug (NM2CAB) maintained plasma levels over 90% inhibitory concentration for up to a year after a single dosage, with sustained retention in muscle and lymphoid tissues and no obvious toxicity (Gautam *et al.*, 2021). Surface-engineered systems such as nevirapine nanocrystals also showed improved organ accumulation and reduced RES clearance, improving systemic retention (Shegokar and Singh, 2011). These findings highlight parenteral nanocrystals' ability to provide sustain release, reduce doses, and improve therapeutic effects.

Ocular drug delivery remains quite difficult due to physiological obstacles such as tear turnover, blinking, and nasolacrimal drainage, which result in rapid drug clearance and limited bioavailability. Conventional formulations sometimes need frequent dosing, which leads to reduced patient compliance and dose-related adverse effects. In this context, nanocrystals present an attractive approach for improving ocular retention and therapeutic efficacy by improving adhesion, controlling release, and increasing local drug concentration (Geng *et al.*, 2024).

Recent research has shown the translational potential of nanocrystal-based ocular systems. In albino rabbits, hydrocortisone nanosuspensions (~300 nm) produced using nanoprecipitation and wet milling showed significantly improved bioavailability. The drug effect lasted up to 9 hr, compared to 5 hr for conventional solutions, and the AUC values were higher (~28-31 vs 15.86) (Ali *et al.*, 2010). Similarly, ion-paired moxifloxacin nanocrystals allowed sustained intraocular drug release following a single subconjunctival injection, compared

Table 5: Surface Engineering Strategies for Functional Nanocrystals.

Ligand	Target Mechanism	Key Functional Outcome	References
Folic Acid (FA)	Folate receptor targeting	Enhanced tumor uptake and receptor-mediated internalization	Liang *et al.*, 2021; Setua *et al.*, 2009; Zhao *et al.*, 2021
Hyaluronic Acid (HA)	CD44 receptor targeting	Increased cellular uptake and tumor inhibition	Agrawal *et al.*, 2017; Seo *et al.*, 2020
Chondroitin sulfate	CD44-mediated targeting	Selective tumor accumulation	Liang *et al.*, 2020; *et al.*, 2022
Alginate / Chitosan	pH-responsive protection	Improved oral bioavailability and controlled release	Lin *et al.*, 2011; Ryu *et al.*, 2020
Transferrin (Tf)	Transferrin receptor targeting	Enhanced uptake and prolonged circulation	Han *et al.*, 2020; Yong *et al.*, 2019
Triphenylphosphonium	Mitochondrial targeting	Increased cytotoxicity and reversal of drug resistance	Han *et al.*, 2018; Zhang *et al.*, 2017
Entropic ligands	Thermodynamic modulation	Improved solubility and system stability	Yang *et al.*, 2016; Yang *et al.*, 2016

Table 6: Biological Fate of Drug Nanocrystals Across Administration Routes.

Route	*In vivo* behavior	References
Oral	Dual behavior involving dissolution and intact particle uptake via endocytosis, contributing to enhanced absorption (~16-fold increase in bioavailability).	Fu *et al.*, 2013
Intravenous	Rapid uptake by mononuclear phagocyte system with organ accumulation (~95% in liver/spleen) and prolonged retention (up to 7 days) due to phagocytosis and size-dependent dissolution.	Liu *et al.*, 2012
Dermal	Accumulation within hair follicles (~196 μm depth) forming localized drug reservoirs enabling sustained release.	Pelikh and Keck, 2020
Ocular	Enhanced mucosal adhesion leading to prolonged retention (~9 hr) and localized depot formation with improved bioavailability (~2-fold).	Donia *et al.*, 2020
Cellular	Endocytosis-mediated uptake followed by intracellular trafficking through ER-Golgi pathways enabling controlled drug release.	Deng *et al.*, 2017

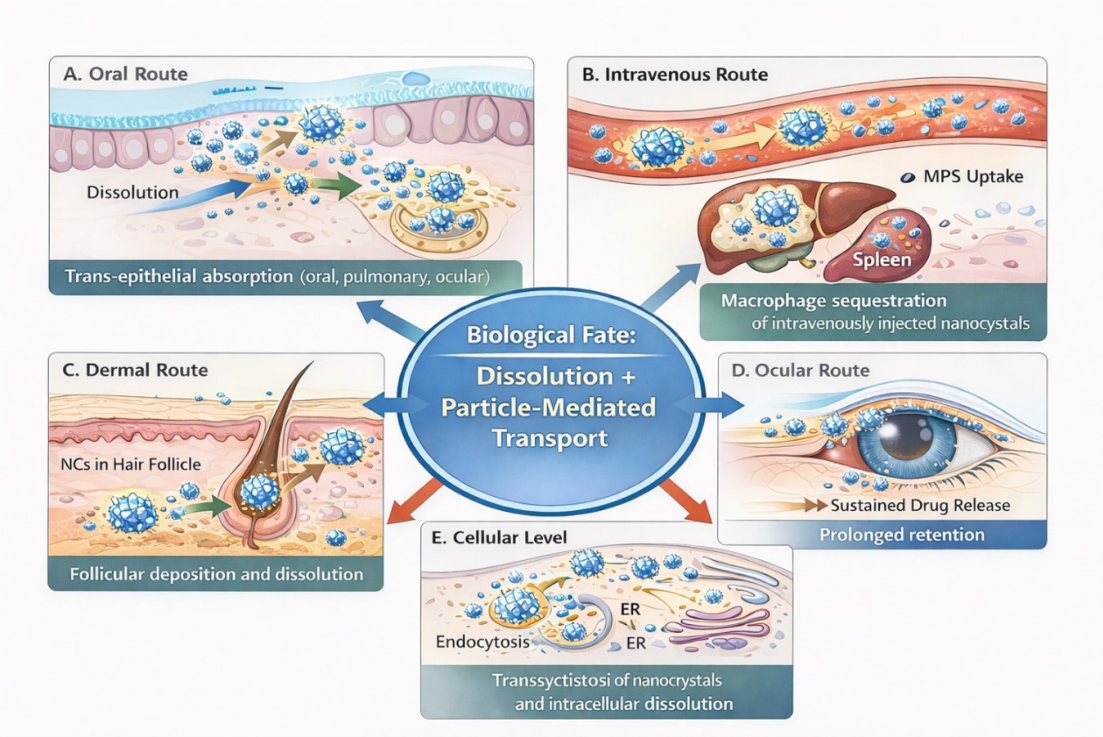


Figure 3: Biological Fate of Drug Nanocrystals Across Different Delivery Routes.

Table 7: Marketed Nanocrystal Products and Production Technologies.

Drug / Product	Route	Technology	Key Benefit	References
Aprepitant (Emend)	Oral	NanoCrystal (WBM)	Improved absorption window	Litou *et al.*, 2019
Sirolimus (Rapamune)	Oral	NanoCrystal (WBM)	Increased bioavailability (~21%)	Jeon *et al.*, 2018
Megestrol acetate (Megace ES)	Oral	NanoCrystal (WBM)	Higher bioavailability at lower dose	Guk *et al.*, 2016
Fenofibrate (Tricor)	Oral	NanoCrystal (WBM)	Reduced food effect	Ling, 2013
Fenofibrate (Lipanthyl)	Oral	NanoCrystal (WBM)	Improved exposure consistency	Vogt *et al.*, 2007
Fenofibrate (Triglide)	Oral	IDD-P (HPH)	Enhanced dissolution	Ling, 2013
Paliperidone palmitate (Invega)	Parenteral	NanoCrystal (WBM)	Sustained release profile	Chue and Chue, 2012
Paliperidone palmitate (Xeplion)	Parenteral	NanoCrystal (WBM)	Improved patient adherence	Lamb and Keating, 2016

with conventional eye drops in both rat and rabbit infection models (Appell *et al.*, 2025).

Furthermore, forskolin nanocrystal-based *in situ* gel systems showed prolonged intraocular pressure reduction (31% for 12 hr) compared to conventional formulations (18%, 4-6 hr), highlighting improved therapeutic outcomes (Gupta *et al.*, 2010).

Collectively, these findings indicate that nanocrystals improved ocular drug delivery by prolonging residence time, improving bioavailability, and reducing dosing frequency, thereby offering a clinically alternative to conventional therapies.

Pulmonary drug delivery is a very efficient route for both local and systemic therapy because of its huge surface area and lack of first-pass metabolism, allowing for rapid absorption that may match intravenous pharmacokinetics. However, traditional inhalation methods are frequently hampered by low solubility, heterogeneous deposition, and fast mucociliary clearance (Kee *et al.*, 2026; Peng *et al.*, 2024). Nanocrystals solve these challenges by improving dissolution kinetics and allowing uniform distribution within the deep lung region, particularly when formulated as aerosolizable systems (Yue *et al.*, 2022).

Recent studies showed that formulation engineering key role in pulmonary performance. Zheng *et al.* reported polysorbate-stabilized nintedanib nanocrystals with optimized nebulization behavior, effectively overcoming solubility and oral

bioavailability limitations in pulmonary fibrosis therapy (Zheng *et al.*, 2024). Similarly, Kumar *et al.* developed lipid-coated paclitaxel nanocrystals that surpassed uncoated systems in terms of aerosolization efficiency, sustained release, and lung retention, indicating the importance of surface modification in controlling release and clearance (Kumar *et al.*, 2024).

Critically, whereas nanocrystals increase local drug concentration and lower systemic toxicity, their efficacy is still dependent on aerodynamic features and mucus interactions. As a result, rational design that incorporates carrier systems and particle engineering is crucial. Overall, pulmonary nanocrystals are a potential approach to targeted lung delivery, notably in cancer and fibrotic disorders.

Topical drug delivery provides localized therapeutic effect while avoiding first-pass metabolism, making it ideal for dermatological diseases. However, medication penetration and efficacy may be limited by the epidermal barrier and enzymatic activity (e.g., P450s, N-acetyltransferases) (Xue *et al.*, 2024). Nanocrystals solve these issues by increasing drug solubility, preserving high concentration gradients, and improving penetration into deeper epidermal layers with smaller particles achieve superior permeation (Singh *et al.*, 2022).

Recent studies showed the translational potential of nanocrystal-based topical systems. Kumar *et al.* (2022) reported niclosamide nanocrystals (~173 nm) incorporated into a hyaluronic acid-modified hydrogel, achieving sustained release (~12 h) and significantly improved cytotoxic efficacy in HaCaT cells (IC_{50} reduced from 1.25 μM to 0.017 μM), showing improved cellular uptake and targeting (Kumar *et al.*, 2022). Curcumin nanocrystals incorporated into clay-based hydrogels enhanced skin penetration and antioxidant activity, demonstrating improved topical bioavailability through prolonged contact and optimized rheological properties (Ruggeri *et al.*, 2022). Similarly, Vasanth (2024) found that ketoconazole nanocrystals (48-83 nm) in a gel system with high entrapment efficiency (~85-93%) and prolonged drug release (~12 hr), resulting in improved local bioavailability and reduced dosing frequency (Nayak *et al.*, 2020).

Collectively, nanocrystal-based topical systems enable improved penetration, sustained release, and improved therapeutic outcomes in skin disorders.

Overall, drug nanocrystals are used to increase drug performance while retaining the drugs pharmacological characteristics. Their adaptability across various modes of administration showing their importance as a formulation-driven approach for overcoming solubility and delivery issues. As shown in Table 8, the integration of nanocrystal technology with sophisticated formulation methodologies is expanding its therapeutic value. Future development is intended to focus on optimizing formulation design, increasing targeting efficiency, and improving scalability to support wider industrial and clinical application.



Figure 4: Nanocrystal across different administration routes.

Table 8: Applications of Drug Nanocrystals Across Different Delivery Routes.

Route	Drug	Outcomes	References
Oral	Baicalein	~6.09-fold ↑ bioavailability vs coarse drug; 3.32-fold vs NCs; improved dissolution and absorption	Pi *et al.*, 2018
Oral	Silybin	2.61-fold (rats) and 1.51-fold (humans) ↑ bioavailability; stable for 24 months	Seo *et al.*, 2024
Oral	Lutein	Solubility ↑ (7.3 → 215.7 μg/mL); C_{max} ↑ 3.24-fold; AUC ↑ 2.28-fold (rats)	Chang *et al.*, 2017
Oral	Lutein	~15.2-fold ↑ bioavailability; ~3.4-fold ↑ ocular accumulation (rats)	Seto *et al.*, 2019
Parenteral	Anlotinib	~20-fold dose reduction (50 μg/kg vs 1 mg/kg); strong tumor inhibition (mice)	Luo *et al.*, 2021
Parenteral	Paclitaxel	Sustained release; enhanced cytotoxicity in MDR cancer cells; improved tumor inhibition	Liu *et al.*, 2010
Parenteral	Camptothecin	Superior tumor suppression vs solution (MCF-7 xenograft)	Zhang *et al.*, 2011
Parenteral	Cabotegravir	Plasma levels maintained ≥1 year; long-acting PK; no toxicity (mice, rats)	Gautam *et al.*, 2021
Parenteral	Nevirapine	Enhanced organ accumulation; prolonged systemic retention	Shegokar and Singh, 2011
Ocular	Hydrocortisone	AUC ↑ (~28-31 vs 15.86); prolonged action (9 hr vs 5 hr) in rabbits	Ali *et al.*, 2010
Ocular	Moxifloxacin	Single-dose sustained intraocular delivery vs multiple eye drops (rats, rabbits)	Appel *et al.*, 2025
Ocular	Forskolin NC	IOP reduction 31% (12 hr) vs 18% (4-6 hr); improved glaucoma therapy	Gupta *et al.*, 2010
Pulmonary	Nintedanib	Improved nebulization and lung delivery; overcomes oral limitations	Zheng *et al.*, 2024
Pulmonary	Paclitaxel	Sustained release; improved aerosolization and lung retention	Kumar *et al.*, 2024
Topical	Niclosamide	IC_{50} reduced (1.25 μM → 0.017 μM); sustained release ~12 hr	Kumar *et al.*, 2022
Topical	Curcumin	Enhanced skin penetration and antioxidant activity	Ruggeri *et al.*, 2022
Topical	Ketoconazole	Sustained release (~12 hr); improved bioavailability; reduced dose	Vasanth, 2024

Regulatory Considerations in the Development of Drug Nanocrystals

The expanding adoption of nanocrystal technology into pharmaceutical development is shown in the number of regulatory applications and approvals over the last several decades. Retrospective evaluations of regulatory submissions show that nanocrystal-based formulations account for a sizable number of nanotechnology-enabled drug items examined by the US Food and Drug Administration. From 1973 to 2015, a total of 82 nanocrystal-related submissions were reported, comprising 55% Investigational New Drug (IND), 32% Abbreviated New Drug Applications (ANDAs), and 13% New Drug Applications (NDAs), highlighting both innovation potential and reformulation opportunities (Chen *et al.*, 2017; Kapoor *et al.*, 2017).

Nanocrystal formulations are especially important for Biopharmaceutics Classification System (BCS) class II drugs, as poor water solubility limits absorption despite acceptable permeability. According to regulatory data, more than 60% of nanocrystal-based products are intended for oral administration, demonstrating their efficiency in enhancing dissolving and lowering food-dependent variability (Chen *et al.*, 2017). Comparative studies show that nanocrystal formulations have better pharmacokinetic uniformity and less affinity to physiological variables than conventional micronized systems.

Nanocrystal drugs are regulated using recognized Chemistry, Manufacturing, and Controls (CMC) frameworks. Critical quality attributes include Particle Size Distribution (PSD), polymorphic form and surface features, which all have significant impact on dissolving behavior, bioavailability, and stability (Kapoor *et al.*, 2017). According to regulatory guidelines, in order to ensure reliable efficacy in therapy, nanocrystal size should typically stay below 1000 nm with controlled distribution (Chen *et al.*, 2017). In order to prevent Ostwald ripening, which causes instability and unpredictability in drug performance as smaller particles dissolve and accumulate onto greater sized particles, narrow PSD is very important.

One of most important regulatory issues is still stability. Nanocrystals are prone to aggregation and recrystallization because of their high surface energy. Formulation creation requires careful consideration of both chemical stability (degradation of labile drugs like omeprazole and paclitaxel) and physical stability (aggregation, crystal formation) (Müller *et al.*, 2011). Stabilizers play vital role by providing steric and electrostatic repulsion, thereby maintaining dispersion stability and preventing particle growth (Goodman *et al.*, 2021). In addition, the presence of amorphous fractions frequently produced during processing might improve dissolution but requires strict control to ensure long-term stability (Gao *et al.*, 2008).

Regulatory approval is also influenced by manufacturing factors. While top-down techniques like milling and High-Pressure Homogenization (HPH) must deal with contamination and process reproducibility, bottom-up approaches like precipitation techniques require precise control over residual solvents and nucleation conditions (Hu *et al.*, 2016; Malamatari *et al.*, 2018). Among these, HPH is widely favoured due to its solvent-free and scalability, making it suitable for industrial production. The comparative regulatory and manufacturing considerations across different fabrication methods are summarized in Table 9.

Compatibility with the 505(b)(2) regulatory process, which permits reformulation of current drugs using already documented safety and efficacy data, is another benefit of nanocrystal technology. Compared to more complex nanocarrier-based systems, this approach facilitated quicker clinical translation (Chandra and C, 2021). Overall, as Table 9 summarizes, effective development necessitates strict control of particle size, stability, and manufacturing methods to assure consistent product quality and therapeutic performance, even if nanocrystals offer a relatively simplified path to regulatory approval.

Toxicological and Safety Considerations

The toxicological profile of drug nanocrystals is mostly affected by particle size, biodegradability, and formulation composition. Particle size is a key factor in determining cellular interaction and potential toxicity. Nanocrystals smaller than 100 nm have a higher cellular internalization rate, which may increase intracellular exposure and associated toxicity risks, whereas particles between 100 and 1000 nm are more likely to undergo controlled clearance via the mononuclear phagocyte system, resulting in a lower overall risk.

Biodegradability is a significant advantage of drug nanocrystals. Unlike non-degradable nanomaterials, drug nanocrystals are mostly made up of the active pharmaceutical ingredient and dissolve *in vivo*, reducing long-term accumulation and associated toxicity risks (Müller *et al.*, 2011). However, temporary interactions with immune cells and excipient-related effects must be carefully assessed because stabilizers and surfactants may affect biocompatibility.

Experimental and translational examinations support nanocrystals' excellent safety profile. For example, PX-13 and PX-18 nanocrystals (130-280 nm) had equivalent or lower cytotoxicity than traditional formulations, with no

Table 9: Regulatory and Manufacturing Considerations in Nanocrystal Development.

Aspect	Regulatory Requirement / Challenge	Regulatory Significance	References
Submission trends	IND, ANDA, NDA pathways with increasing submissions	Indicates strong translational and generic potential	Chen *et al.*, 2017; Kapoor *et al.*, 2017
Particle size control	Defined size (<1000 nm) with narrow distribution	Critical for batch consistency, dissolution, and bioavailability	Chen *et al.*, 2017
Physical stability	Prevention of aggregation and Ostwald ripening	Ensures product quality and shelf-life	Gao *et al.*, 2008
Chemical stability	Protection of labile drugs	Maintains drug integrity and safety	Müller *et al.*, 2011
Manufacturing (CMC)	Control of contamination and residual solvents	Essential for regulatory approval and quality compliance	Hu *et al.*, 2016; Malamatari *et al.*, 2018
Scalability and reproducibility	Validated large-scale processes (e.g., HPH)	Ensures consistent industrial production	Hu *et al.*, 2016
Regulatory pathway	Use of 505(b)(2) or generic pathways	Enables faster approval and lifecycle management	Chandra and C, 2021

Table 10: Determinants of Toxicological Behavior of Drug Nanocrystals.

Factor	Safety Consideration	Toxicological Outcome / Risk	References
Particle size (<100 nm)	Higher cellular internalization	Increased intracellular exposure may elevate toxicity risk	Yu *et al.*, 2023
Particle size (100-1000 nm)	Limited penetration; MPS clearance	Controlled distribution with relatively low toxicity	Srinivas *et al.*, 2021
Biodegradability	API-based nanocrystals dissolve *in vivo*	Minimal long-term accumulation; generally safe	Müller *et al.*, 2011
Physical stability	Aggregation / Ostwald ripening	May alter dose uniformity and safety profile	Gao *et al.*, 2008
Immune interaction	Recognition by macrophages	Transient immune activation possible	Wen *et al.*, 2025
Formulation excipients	Surfactants and stabilizers	Potential irritation or toxicity depending on type and concentration	Kojima *et al.*, 2018
Cytotoxicity studies	PX-13, PX-18 nanocrystals (130-280 nm)	Comparable or lower toxicity than conventional formulations	Pardeike, 2009
Dermal safety	Nanosuspension formulations (5%)	No irritation in EPISKIN and HET-CAM models	Chinthaginjala *et al.*, 2023; Llorente *et al.*, 2023
Translational safety	Cosmetic nanocrystals (rutin, hesperidin)	Safe in human patch and ocular studies	Hassan and Soliman, 2022; Stahr *et al.*, 2021

irritation seen in EPISKIN and HET-CAM models (Pardeike, 2009). Similarly, hesperetin nanocrystals (~100 nm) reduced inflammatory markers (TNF-α, IL-6) and oxidative stress while promoting cellular compatibility (Stahr *et al.*, 2021). *In vivo*, rutin nanocrystals (270-500 nm) demonstrated improved therapeutic effectiveness without evidence of nanocrystal-induced toxicity (Hassan and Soliman, 2022). Comparable findings in cosmetic nanocrystal systems support their safety in human-relevant environments (Hassan and Soliman, 2022). Comparable findings in cosmetic nanocrystal systems further confirm their safety in human-relevant conditions (Sharma and Medhi, 2025).

Overall, these findings show that pharmacological nanocrystals have a predictable and tolerable safety profile when important issues like size, stability, and excipient composition are well regulated. Table 10 summarizes the important toxicological factors.

CHALLENGES AND FUTURE OUTLOOK

Despite significant advancements, various formulation and translational hurdles continue to restrict the clinical use of therapeutic nanocrystals. Physical instability, particularly aggregation and Ostwald ripening caused by high surface energy, is still a major hurdle for stability and efficiency. The variability in particle size distribution delays drug dissolution and the reproducibility of *in vivo* consequences. In surface-engineered systems, additional restrictions such as ligand detachment, protein corona formation, and heterogeneity receptor expression may impair targeting efficacy under physiological environments. Manufacturing and regulatory problems, including as scalability, process reproducibility, and polymorphic transition control, persist to be major obstacles to industrial translation. Furthermore, consistent critical quality features and strong *in vitro-in vivo* correlations are required for regulatory approval. Although nanocrystals are typically regarded as safe, long-term toxicity, excipient-related effects, and immunological interactions must be further investigated.

Future research should focus on rational design strategies that combine stability, targeting, and controlled drug release on a single platform. Green manufacturing, continuous processing, and AI-assisted formulation optimization are examples of emerging technologies that show promise for enhancing scalability and repeatability. Furthermore, the development of predictive models that link physicochemical characteristics to *in vivo* performance will be essential to accelerating clinical translation.

CONCLUSION

Drug nanocrystals have emerged as one of the most effective formulation-based techniques for increasing the effectiveness of drugs that are poorly water soluble. Their carrier-free nature, high drug loading, and potential to improve dissolution without affecting pharmacological efficacy set them apart from typical nanocarriers. Importantly, clinically authorized products exhibit translational feasibility via oral and parenteral routes.

Advances in fabrication methods, surface engineering, and understanding of *in vivo* behavior have increased their therapeutic utility. However, obstacles like as stability, production control, and regulatory standards remain important to broad implementation.

Overall, drug nanocrystals are a clinically established and scalable technology with significant potential for future drug development, especially when combined with increased process control and mechanistic understanding.

ACKNOWLEDGEMENT

The authors are thankful to the Principal of R.C. Patel Institute of Pharmaceutical Education and Research and R.C. Patel Institute of Pharmacy, Shirpur, Dist.: Dhule (MS) India- 425 405, for providing necessary library facilities.

ABBREVIATIONS

ABC: Accelerated Blood Clearance; **API:** Active Pharmaceutical Ingredient; **AUC:** Area Under the Curve; **BCS:** Biopharmaceutics Classification System; **CMC:** Chemistry, Manufacturing, and Controls; **CNTs:** Carbon Nanotubes; **DLVO:** Derjaguin-Landau-Verwey-Overbeek Theory; **EPR:** Enhanced Permeability and Retention; **ER:** Endoplasmic Reticulum; **FA:** Folic Acid; **HA:** Hyaluronic Acid; **HET-CAM:** Hen's Egg Test-Chorioallantoic Membrane; **HME:** Hot-Melt Extrusion; **HPC:** Hydroxypropyl Cellulose; **HPMC:** Hydroxypropyl Methylcellulose; **HPH:** High-Pressure Homogenization; **HVNCs:** Hyaluronic Acid-Modified Venetoclax Nanocrystals; **IC_{50}:** Half Maximal Inhibitory Concentration; **IND:** Investigational New Drug; **IOP:** Intraocular Pressure; MDR**:** Multidrug Resistance; **MPS:** Mononuclear Phagocyte System; **MRT:** Mean Residence Time; **MW:** Molecular Weight; **NCs:** Nanocrystals; NCEs**:** New Chemical Entities; **NDAs:** New Drug Applications; ANDAs**:** Abbreviated New Drug Applications; **PEG:** Polyethylene Glycol; **PDI:** Polydispersity Index; **P-gp:** P-glycoprotein; **PSD:** Particle Size Distribution; **PVA:** Polyvinyl Alcohol; **PVP:** Polyvinylpyrrolidone; **RES:** Reticuloendothelial System; SCF**:** Supercritical Fluid; **SDS:** Sodium Dodecyl Sulfate; **Tf:** Transferrin; **TPGS:** D-α-Tocopheryl Polyethylene Glycol Succinate; **TPP:** Triphenylphosphonium; WBM**:** Wet Bead Milling.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

Agrawal, S., Dwivedi, M., Ahmad, H., Chadchan, S. B., Arya, A., *et al.* (2017). CD44 targeting hyaluronic acid coated lapatinib nanocrystals foster the efficacy against triple-negative breast cancer. Nanomedicine: Nanotechnology, Biology and Medicine, 14(2), 327-337. https://doi.org/10.1016/j.nano.2017.10.010

Ali, H. S. M., York, P., Ali, A. M. A., & Blagden, N. (2010). Hydrocortisone nanosuspensions for ophthalmic delivery: A comparative study between microfluidic nanoprecipitation and wet milling. Journal of Controlled Release, 149(2), 175-181. https://doi.org/10.1016/j.jconrel.2010.10.007

Appell, M. B., Malmberg, K., Pasupathy, A., Josyula, A., Ortiz, J., *et al.* (2025). Ion-paired moxifloxacin nanocrystal formulation improves treatment and prevention of ocular infection. Journal of Controlled Release: Official Journal of the Controlled Release Society, 381, 113634. https://doi.org/10.1016/j.jconrel.2025.113634

B, S., & Ghosh, A. (2025). Mechanistic Insights into Amorphous Solid Dispersions: Bridging Theory and Practice in Drug Delivery. Pharmaceutical Research, 42(1), 1-23. https://doi.org/10.1007/s11095-024-03808-w

Baumgartner, R., Eitzlmayr, A., Matsko, N., Tetyczka, C., Khinast, J., & Roblegg, E. (2014). Nano-extrusion: A promising tool for continuous manufacturing of solid nano-formulations. International Journal of Pharmaceutics, 477(1-2), 1-11. https://doi.org/10.1016/j.ijpharm.2014.10.008

Bhaskar, R., Ola, M., Pingle, V., Bari, V., & Patil, R. (2025). Nanocrystal System: A Comprehensive Review of Method of Preparation and their Characterization, Patents and Marketed Products. Journal of Drug Delivery and Therapeutics, 15(2), 171-185. https://doi.org/10.22270/jddt.v15i2.6982

Bohlouli, S., Jafarmadar Gharehbagh, F., Dalir Abdolahinia, E., Kouhsoltani, M., Ebrahimi, G., *et al.* (2021). Preparation, Characterization, and Evaluation of Rutin Nanocrystals as an Anticancer Agent against Head and Neck Squamous Cell Carcinoma Cell Line. Journal of Nanomaterials, 2021(1), 1-8. https://doi.org/10.1155/2021/9980451

Bookwala, M., & Wildfong, P. L. D. (2023). The Implications of Drug-Polymer Interactions on the Physical Stability of Amorphous Solid Dispersions. Pharmaceutical Research, 40(12), 2963-2981. https://doi.org/10.1007/s11095-023-03547-4

Chandra, A., & C, N. (2021). Review on nanoparticles technology and applications based on drug delivery. IP International Journal of Comprehensive and Advanced Pharmacology, 6(3), 117-120. https://doi.org/10.18231/j.ijcaap.2021.021

Chang, D., Ma, Y., Cao, G., Wang, J., Zhang, X., *et al.* (2017). Improved oral bioavailability for lutein by nanocrystal technology: formulation development, *in vitro* and *in vivo* evaluation. Artificial Cells, Nanomedicine, and Biotechnology, 46(5), 1018-1024. https://doi.org/10.1080/21691401.2017.1358732

Chary, P. S., Shaikh, S., Bhavana, V., Rajana, N., Vasave, R., & Mehra, N. K. (2024). Emerging role of nanocrystals in pharmaceutical applications: A review of regulatory aspects and drug development process. Applied Materials Today, 40, 102334. https://doi.org/10.1016/j.apmt.2024.102334

Chen, J., Zhao, Z., Yathavan, B., Mirajkar, A., Guo, D., & Huang, J. (2025). Recent Advancement of Nanocrystal Dosage Forms. In Pharmaceutical science. Intechopen. https://doi.org/10.5772/intechopen.1007776

Chen, M.-L., John, M., Lee, S. L., & Tyner, K. M. (2017). Development Considerations for Nanocrystal Drug Products. The AAPS Journal, 19(3), 642-651. https://doi.org/10.1208/s12248-017-0064-x

Chen, X., Zou, L.-Q., Niu, J., Liu, W., Peng, S.-F., & Liu, C.-M. (2015). The Stability, Sustained Release and Cellular Antioxidant Activity of Curcumin Nanoliposomes. Molecules, 20(8), 14293-14311. https://doi.org/10.3390/molecules200814293

Chen, Y., Liu, Y., Xu, J., Xie, Y., Zheng, Q., Yue, P., & Yang, M. (2017). A Natural Triterpenoid Saponin as Multifunctional Stabilizer for Drug Nanosuspension Powder. AAPS PharmSciTech, 18(7), 2744-2753. https://doi.org/10.1208/s12249-017-0756-7

Chinthaginjala, H., Bogavalli, V., Hindustan, A. A., Pathakamuri, J., Pullaganti, S. S., Gowni, A., & Baktha, B. (2023). Nanostructured Lipid Carriers: A Potential Era of Drug Delivery Systems. Indian Journal of Pharmaceutical Education and Research, 58(1), 21-33. https://doi.org/10.5530/ijper.58.1.3

Chue, P., & Chue, J. (2012). A review of paliperidone palmitate. Expert Review of Neurotherapeutics, 12(12), 1383-1397. https://doi.org/10.1586/ern.12.137

Deljavan Ghodrati, A., & Turk, C. T. S. (2024). Nanocrystals and their applications in pharmaceutical technology: An up-to-date overview. Journal of Research in Pharmacy, 28(6), 1921-1935. https://doi.org/10.29228/jrp.865

Deng, F., Zhang, H., Wang, X., Zhang, Y., *et al.* (2017). Transmembrane Pathways and Mechanisms of Rod-like Paclitaxel Nanocrystals through MDCK Polarized Monolayer. ACS Applied Materials & Interfaces, 9(7), 5803-5816. https://doi.org/10.1021/acsami.6b15151

Deshmukh, A. S., Tiwari, K. J., & Mahajan, V. R. (2017). Solubility Enhancement Techniques for Poorly Water-Soluble Drugs. International Journal of Pharmaceutical Sciences and Nanotechnology, 10(3), 3701-3708. https://doi.org/10.37285/ijpsn.2017.10.3.1

Dhaval, M., Makwana, J., Sakariya, E., & Dudhat, K. (2020). Drug Nanocrystals: A Comprehensive Review with Current Regulatory Guidelines. Current Drug Delivery, 17(6), 470-482. https://doi.org/10.2174/1567201817666200512104833

Dhibar, M., Chakraborty, S., Kundu, A., & Laha, P. (2023). Chemistry Characterization and Application of Nanocrystals-based Drug Delivery System: Present to Future Perspective. Pharmaceutical Nanotechnology, 11(3), 265-275. https://doi.org/10.2174/2211738511666230109151410

Dolenc, A., Kristl, J., Baumgartner, S., & Planinšek, O. (2009). Advantages of celecoxib nanosuspension formulation and transformation into tablets. International Journal of Pharmaceutics, 376(1-2), 204-212. https://doi.org/10.1016/j.ijpharm.2009.04.038

Donia, M., Osman, R., Awad, G. A. S., & Mortada, N. (2020). Polypeptide and glycosaminoglycan polysaccharide as stabilizing polymers in nanocrystals for a safe ocular hypotensive effect. International Journal of Biological Macromolecules, 162, 1699-1710. https://doi.org/10.1016/j.ijbiomac.2020.07.306

Eckert, R. W., Hartmann, S. F., Knoth, D., Pelikh, O., Müller, R. H., & Keck, C. M. (2020). Nanocrystals: The Universal Formulation Principle to Improve the Bioactivity of Poorly Soluble Actives (pp. 453-487). Jenny Stanford. https://doi.org/10.1201/9781003045151-10

Fu, Q., Sun, J., Zhang, D., Li, M., Wang, Y., *et al.* (2013). Nimodipine nanocrystals for oral bioavailability improvement: Preparation, characterization and pharmacokinetic studies. Colloids and Surfaces B: Biointerfaces, 109, 161-166. https://doi.org/10.1016/j.colsurfb.2013.01.066

Gamsiz, E. D., Thombre, A. G., Ahmed, I., & Carrier, R. L. (2010). Drug Salts and Solubilization: Modeling the Influence of Cyclodextrins on Oral Absorption. Annals of Biomedical Engineering, 39(1), 455-468. https://doi.org/10.1007/s10439-010-0169-1

Gao, L., Zhang, D., & Chen, M. (2008). Drug nanocrystals for the formulation of poorly soluble drugs and its application as a potential drug delivery system. Journal of Nanoparticle Research, 10(5), 845-862. https://doi.org/10.1007/s11051-008-9357-4

Gautam, N., Mcmillan, J. M., Kumar, D., Bade, A. N., Pan, Q., *et al.* (2021). Lipophilic Nanocrystal Prodrug-Release Defines the Extended Pharmacokinetic Profiles of a Year-Long Cabotegravir. Nature Communications, 12(1), 3453. https://doi.org/10.1101/2021.01.18.427147

Geng, F., Fan, X., Liu, Y., Lu, W., & Wei, G. (2024). Recent advances in nanocrystal-based technologies applied for ocular drug delivery. Expert Opinion on Drug Delivery, ahead-of-print(ahead-of-print), 21(2), 211-227. https://doi.org/10.1080/17425247.2024.2311119

Gigliobianco, M. R., Casadidio, C., Censi, R., & Di Martino, P. (2018). Nanocrystals of Poorly Soluble Drugs: Drug Bioavailability and Physicochemical Stability. Pharmaceutics, 10(3), 134. https://doi.org/10.3390/pharmaceutics10030134

Goodman, E. D., Carlson, E. Z., Dietze, E. M., Tahsini, N., *et al.* (2021). Size-controlled nanocrystals reveal spatial dependence and severity of nanoparticle coalescence and Ostwald ripening in sintering phenomena. Nanoscale, 13(2), 930-938. https://doi.org/10.1039/d0nr07960j

Guk, J., Son, H., Chae, D. W., & Park, K. (2016). Quantitative Assessment of Food Effect on the Pharmacokinetics of Nano-Crystallized Megestrol Acetate. Basic & Clinical Pharmacology & Toxicology, 120(3), 270-277. https://doi.org/10.1111/bcpt.12677

Gupta, S., Samanta, M. K., & Raichur, A. M. (2010). Dual-Drug Delivery System Based on *In Situ* Gel-Forming Nanosuspension of Forskolin to Enhance Antiglaucoma Efficacy. AAPS PharmSciTech, 11(1), 322-335. https://doi.org/10.1208/s12249-010-9388-x

Han, S., Li, X., Zhou, C., Hu, X., Zhou, Y., *et al.* (2020). Further Enhancement in Intestinal Absorption of Paclitaxel by Using Transferrin-Modified Paclitaxel Nanocrystals. ACS Applied Bio Materials, 3(7), 4684-4695. https://doi.org/10.1021/acsabm.0c00599

Han, X., Su, R., Huang, X., Wang, Y., Kuang, X., Zhou, S., & Liu, H. (2018). Triphenylphosphonium-modified mitochondria-targeted paclitaxel nanocrystals for overcoming multidrug resistance. Asian Journal of Pharmaceutical Sciences, 14(5), 569-580. https://doi.org/10.1016/j.ajps.2018.06.006

Hassan, A. S., & Soliman, G. M. (2022). Rutin Nanocrystals with Enhanced Anti-Inflammatory Activity: Preparation and *ex vivo*/*In vivo* Evaluation in an Inflammatory Rat Model. Pharmaceutics, 14(12), 2727. https://doi.org/10.3390/pharmaceutics14122727

Hollis, C. P., Weiss, H. L., Leggas, M., Evers, B. M., Gemeinhart, R. A., & Li, T. (2013). Biodistribution and bioimaging studies of hybrid paclitaxel nanocrystals: Lessons learned of the EPR effect and image-guided drug delivery. Journal of Controlled Release, 172(1), 12-21. https://doi.org/10.1016/j.jconrel.2013.06.039

Hu, C., Qian, A., Wang, Q., Xu, F., He, Y., *et al.* (2016). Industrialization of lipid nanoparticles: From laboratory-scale to large-scale production line. European Journal of Pharmaceutics and Biopharmaceutics, 109, 206-213. https://doi.org/10.1016/j.ejpb.2016.10.018

Huang, D., Gui, J., Chen, X., Yu, R., Gong, T., *et al.* (2022). Chondroitin Sulfate-Derived Paclitaxel Nanocrystal via π-π Stacking with Enhanced Stability and Tumor Targetability. ACS applied materials & interfaces, 14(46), 51776-51789. https://doi.org/10.1021/acsami.2c15881

Jeon, H. J., Lee, H.-E., & Yang, J. (2018). Safety and efficacy of Rapamune\xae (Sirolimus) in kidney transplant recipients: results of a prospective post-marketing surveillance study in Korea. BMC Nephrology, 19(1), 201. https://doi.org/10.1186/s12882-018-1002-6

Jin, X., Luo, Y., Chen, Y., Ma, Y., Yue, P., & Yang, M. (2018). Novel breviscapine nanocrystals modified by panax notoginseng saponins for enhancing bioavailability and synergistic anti-platelet aggregation effect. Colloids and Surfaces B: Biointerfaces, 175, 333-342. https://doi.org/10.1016/j.colsurfb.2018.11.067

Jornada, D. H., Dos Santos Fernandes, G. F., Chiba, D. E., De Melo, T. R. F., Dos Santos, J. L., & Chung, M. C. (2015). The Prodrug Approach: A Successful Tool for Improving Drug Solubility. Molecules, 21(1), 42. https://doi.org/10.3390/molecules21010042

Kapoor, M., Lee, S. L., & Tyner, K. M. (2017). Liposomal Drug Product Development and Quality: Current US Experience and Perspective. The AAPS Journal, 19(3), 632-641. https://doi.org/10.1208/s12248-017-0049-9

Kee, P. E., Tan, C. S., Liew, K. B., Lee, S.-K., *et al.* (2026). Unlocking the Potential of Nanotechnology in Advancing Pulmonary Drug Delivery Systems. Current Pharmaceutical Design, 32. https://doi.org/10.2174/0113816128421567251110095308

Khadke, S., Roces, C. B., Donaghey, R., Giacobbo, V., Su, Y., & Perrie, Y. (2020). Scalable solvent-free production of liposomes. Journal of Pharmacy and Pharmacology, 72(10), 1328-1340. https://doi.org/10.1111/jphp.13329

Kocbek, P., Baumgartner, S., & Kristl, J. (2006). Preparation and evaluation of nanosuspensions for enhancing the dissolution of poorly soluble drugs. International Journal of Pharmaceutics, 312(1-2), 179-186. https://doi.org/10.1016/j.ijpharm.2006.01.008

Kojima, T., Karashima, M., Yamamoto, K., & Ikeda, Y. (2018). Combination of NMR Methods To Reveal the Interfacial Structure of a Pharmaceutical Nanocrystal and Nanococrystal in the Suspended State. Molecular Pharmaceutics, 15(9), 3901-3908. https://doi.org/10.1021/acs.molpharmaceut.8b00360

Kumar, A., Valamla, B., Thakor, P., Chary, P. S., Rajana, N., & Mehra, N. K. (2022). Development and evaluation of nanocrystals loaded hydrogel for topical application. Journal of Drug Delivery Science and Technology, 74, 103503. https://doi.org/10.1016/j.jddst.2022.103503

Kumar, M., Jha, A., Bharti, K., Manjit, M., Kumbhar, P., *et al.* (2024). Lipid-coated nanocrystals of paclitaxel as dry powder for inhalation: Characterization, *in vitro* performance, and pharmacokinetic assessment. Colloids and Surfaces B: Biointerfaces, 237, 113865. https://doi.org/10.1016/j.colsurfb.2024.113865

Kumar, R., & Siril, P. F. (2017). Enhancing the Solubility of Fenofibrate by Nanocrystal Formation and Encapsulation. AAPS PharmSciTech, 19(1), 284-292. https://doi.org/10.1208/s12249-017-0840-z

Lamb, Y. N., & Keating, G. M. (2016). Paliperidone Palmitate Intramuscular 3-Monthly Formulation: A Review in Schizophrenia. Drugs, 76(16), 1559-1566. https://doi.org/10.1007/s40265-016-0645-5

Liang, H., Zou, F., Liu, Q., Wang, B., Fu, L., *et al.* (2021). Nanocrystal-loaded liposome for targeted delivery of poorly water-soluble antitumor drugs with high drug loading and stability towards efficient cancer therapy. International Journal of Pharmaceutics, 599, 120418. https://doi.org/10.1016/j.ijpharm.2021.120418

Liang, Y., Fu, X., Du, C., Xia, H., Lai, Y., & Sun, Y. (2020). Enzyme/pH-triggered anticancer drug delivery of chondroitin sulfate modified doxorubicin nanocrystal. Artificial Cells, Nanomedicine, and Biotechnology, 48(1), 1114-1124. https://doi.org/10.1080/21691401.2020.1813741

Lin, N., Huang, J., Chang, P. R., Feng, L., & Yu, J. (2011). Effect of polysaccharide nanocrystals on structure, properties, and drug release kinetics of alginate-based microspheres. Colloids and Surfaces B: Biointerfaces, 85(2), 270-279. https://doi.org/10.1016/j.colsurfb.2011.02.039

Ling, H. (2013). A Review of Currently Available Fenofibrate and Fenofibric Acid Formulations. Cardiology Research, 4(2), 47-55. https://doi.org/10.4021/cr270w

Litou, C., Patel, N., Turner, D. B., Kostewicz, E., Kuentz, M., *et al.* (2019). Combining biorelevant *in vitro* and *in silico* tools to simulate and better understand the *in vivo* performance of a nano-sized formulation of aprepitant in the fasted and fed states. European Journal of Pharmaceutical Sciences, 138, 105031. https://doi.org/10.1016/j.ejps.2019.105031

Liu, G., Zhang, D., Jiao, Y., Guo, H., Zheng, D., *et al.* (2012). *In vitro* and *in vivo* evaluation of riccardin D nanosuspensions with different particle size. Colloids and Surfaces B: Biointerfaces, 102, 620-626. https://doi.org/10.1016/j.colsurfb.2012.09.006

Liu, X., Bahloul, B., Lai Kuen, R., Andrieux, K., Roques, C., & Scherman, D. (2021). Cationic lipid nanoparticle production by microfluidization for siRNA delivery. International Journal of Pharmaceutics, 605, 120772. https://doi.org/10.1016/j.ijpharm.2021.120772

Liu, Y., Huang, L., & Liu, F. (2010). Paclitaxel Nanocrystals for Overcoming Multidrug Resistance in Cancer. Molecular Pharmaceutics, 7(3), 863-869. https://doi.org/10.1021/mp100012s

Liversidge, G. G., & Cundy, K. C. (1995). Particle size reduction for improvement of oral bioavailability of hydrophobic drugs: I. Absolute oral bioavailability of nanocrystalline danazol in beagle dogs. International Journal of Pharmaceutics, 125(1), 91-97. https://doi.org/10.1016/0378-5173(95)00122-y

Llorente, X., Esteruelas, G., Bonilla, L., Agudelo, M. G., Filgaira, I., *et al.* (2023). Riluzole-Loaded Nanostructured Lipid Carriers for Hyperproliferative Skin Diseases. International Journal of Molecular Sciences, 24(9), 8053. https://doi.org/10.3390/ijms24098053

Long, W., Ouyang, H., Zhou, C., Wan, W., Yu, S., *et al.* (2021). Simultaneous surface functionalization and drug loading: A novel method for fabrication of cellulose nanocrystals-based pH responsive drug delivery system. International Journal of Biological Macromolecules, 182, 2066-2075. https://doi.org/10.1016/j.ijbiomac.2021.05.193

Losada-Barreiro, S., Celik, S., Sezgin-Bayindir, Z., Bravo-Fernández, S., & Bravo-Díaz, C. (2024). Carrier Systems for Advanced Drug Delivery: Improving Drug Solubility/Bioavailability and Administration Routes. Pharmaceutics, 16(7), 852. https://doi.org/10.3390/pharmaceutics16070852

Lu, Y., Li, Y., & Wu, W. (2016). Injected nanocrystals for targeted drug delivery. Acta Pharmaceutica Sinica. B, 6(2), 106-113. https://doi.org/10.1016/j.apsb.2015.11.005

Luo, M., Sun, H., Jiang, Q., Chai, Y., Li, C., Yang, B., & Hong, Z. (2021). Novel Nanocrystal Injection of Insoluble Drug Anlotinib and Its Antitumor Effects on Hepatocellular Carcinoma. Frontiers in Oncology, 11(6), 777356. https://doi.org/10.3389/fonc.2021.777356

Lv, P.-P., Wei, W., Yue, H., Yang, T.-Y., Wang, L.-Y., & Ma, G.-H. (2011). Porous Quaternized Chitosan Nanoparticles Containing Paclitaxel Nanocrystals Improved Therapeutic Efficacy in Non-Small-Cell Lung Cancer after Oral Administration. Biomacromolecules, 12(12), 4230-4239. https://doi.org/10.1021/bm2010774

Malamatari, M., Taylor, K. M. G., Malamataris, S., Douroumis, D., & Kachrimanis, K. (2018). Pharmaceutical nanocrystals: production by wet milling and applications. Drug Discovery Today, 23(3), 534-547. https://doi.org/10.1016/j.drudis.2018.01.016

Manjunath, K., & Venkateswarlu, V. (2006). Pharmacokinetics, tissue distribution and bioavailability of nitrendipine solid lipid nanoparticles after intravenous and intraduodenal administration. Journal of Drug Targeting, 14(9), 632-645. https://doi.org/10.1080/10611860600888850

Mauludin, R., Müller, R. H., & Keck, C. M. (2008). Development of an oral rutin nanocrystal formulation. International Journal of Pharmaceutics, 370(1-2), 202-209. https://doi.org/10.1016/j.ijpharm.2008.11.029

Müller, R. H., Gohla, S., & Keck, C. M. (2011). State of the art of nanocrystals - Special features, production, nanotoxicology aspects and intracellular delivery. European Journal of Pharmaceutics and Biopharmaceutics, 78(1), 1-9. https://doi.org/10.1016/j.ejpb.2011.01.007

Mura, P. A., Cirri, M., Rossetti, A., Allemandi, D. A., Paredes, A. J., & Palma, S. D. (2023). Preparation of glyburide nanocrystals with improved dissolution properties by dry-ball- and wet-bead- milling: Systematic comparison by experimental design of the performance of the two methods. Journal of Drug Delivery Science and Technology, 91, 105222. https://doi.org/10.1016/j.jddst.2023.105222

Nayak, D., Tawale, R. M., Aranjani, J. M., & Tippavajhala, V. K. (2020). Formulation, Optimization and Evaluation of Novel Ultra-deformable Vesicular Drug Delivery System for an Anti-fungal Drug. AAPS PharmSciTech, 21(5), 140. https://doi.org/10.1208/s12249-020-01681-5

Noreen, S., Maqbool, I., Ijaz, M., & Tanveer, S. (2016). Cyclodextrin Inclusion Complexes: Novel Techniques to Improve Solubility of Poorly Soluble Drugs: A Review. Global Pharmaceutical Sciences Review, 1(1), 29-34. https://doi.org/10.31703/gpsr.2016(i-i).04

Panwar, D., Thakor, P., Sharma, M., Bakshi, A. K., Bhavana, V., *et al.* (2023). Hyaluronic acid-engineered Bcl-2 inhibitor nanocrystals for site-specific delivery to breast tumor cells. Nanomedicine, 18(15), 1005-1023. https://doi.org/10.2217/nnm-2023-0132

Pardeike, J. (2009). Nanosuspensions and nanostructured lipid carriers for dermal application [Die Deutsches Institut Fur Entwicklungspolitik]. In Refubium (Universitätsbibliothek der Freien Universität Berlin). https://doi.org/10.17169/refubium-4394

Pelikh, O., & Keck, C. M. (2020). Hair Follicle Targeting and Dermal Drug Delivery with Curcumin Drug Nanocrystals-Essential Influence of Excipients. Nanomaterials, 10(11), 2323. https://doi.org/10.3390/nano10112323

Peng, S., Wang, W., Zhang, R., Wu, C., Pan, X., & Huang, Z. (2024). Nano-Formulations for Pulmonary Delivery: Past, Present, and Future Perspectives. Pharmaceutics, 16(2), 161. https://doi.org/10.3390/pharmaceutics16020161

Pi, J., Wang, S., Li, W., Kebebe, D., Zhang, Y., *et al.* (2018). A nano-cocrystal strategy to improve the dissolution rate and oral bioavailability of baicalein. Asian Journal of Pharmaceutical Sciences, 14(2), 154-164. https://doi.org/10.1016/j.ajps.2018.04.009

Pinarbasli, O., & Doganay, A. (2021). Prospects of Nanomedicine with Nanocrystal Technology (pp. 681-703). Jenny Stanford. https://doi.org/10.1201/9781003164739-25

Rabinow, B., Kipp, J., Papadopoulos, P., Wong, J., Glosson, J., *et al.* (2007). Itraconazole IV nanosuspension enhances efficacy through altered pharmacokinetics in the rat. International Journal of Pharmaceutics, 339(1-2), 251-260. https://doi.org/10.1016/j.ijpharm.2007.02.030

Rachmawati, H., Shaal, L. A., Müller, R. H., & Keck, C. M. (2013). Development of Curcumin Nanocrystal: Physical Aspects. Journal of Pharmaceutical Sciences, 102(1), 204-214. https://doi.org/10.1002/jps.23335

Rathod, V. G., Deshmukh, N. B., & Deshmukh, S. P. (2025). Advanced Formulation Strategies for Enhancing the Bioavailability of Poorly Soluble Drugs: A Comprehensive Review. Asian Journal of Research in Pharmaceutical Sciences, 15(4), 429-432. https://doi.org/10.52711/2231-5659.2025.00064

Reza Pahlevi, M., Sopyan, I., & Gozali, D. (2023). Technique Development in Improving the Solubility of Poorly Water-Soluble Drugs (BCS II and IV): a Review Study. Jurnal Farmasi Galenika (Galenika Journal of Pharmacy) (e-Journal), 9(2), 147-164. https://doi.org/10.22487/j24428744.2023.v9.i2.15969

Ruggeri, M., Sánchez-Espejo, R., Casula, L., Barbosa, R. D. M., Sandri, G., *et al.* (2022). Clay-Based Hydrogels as Drug Delivery Vehicles of Curcumin Nanocrystals for Topical Application. Pharmaceutics, 14(12), 2836. https://doi.org/10.3390/pharmaceutics14122836

Ryu, J. H., Yoon, H. Y., Sun, I.-C., Kwon, I. C., & Kim, K. (2020). Tumor-Targeting Glycol Chitosan Nanoparticles for Cancer Heterogeneity. Advanced Materials (Deerfield Beach, Fla.), 32(51), 2002197. https://doi.org/10.1002/adma.202002197

Seo, J.-H., Lee, S. Y., Hwang, C., Yang, M., Lee, J., *et al.* (2020). Multi-layered cellulose nanocrystal system for CD44 receptor-positive tumor-targeted anticancer drug delivery. International Journal of Biological Macromolecules, 162, 798-809. https://doi.org/10.1016/j.ijbiomac.2020.06.193

Seo, S., Kim, G.-Y., Kim, M.-H., Lee, K. W., Kim, M.-J., *et al.* (2024). Nanocrystal Formulation to Enhance Oral Absorption of Silybin: Preparation, *In vitro* Evaluations, and Pharmacokinetic Evaluations in Rats and Healthy Human Subjects. Pharmaceutics, 16(8), 1033. https://doi.org/10.3390/pharmaceutics16081033

Seto, Y., Ueno, K., Suzuki, H., Sato, H., & Onoue, S. (2019). Development of novel lutein nanocrystal formulation with improved oral bioavailability and ocular distribution. Journal of Functional Foods, 61, 103499. https://doi.org/10.1016/j.jff.2019.103499

Setua, S., Menon, D., Asok, A., Nair, S., & Koyakutty, M. (2009). Folate receptor targeted, rare-earth oxide nanocrystals for bi-modal fluorescence and magnetic imaging of cancer cells. Biomaterials, 31(4), 714-729. https://doi.org/10.1016/j.biomaterials.2009.09.090

Shahiwala, A. (2025). Large-Scale Production of Nanocrystals for Pharmaceutical Use (pp. 293-311). Crc. https://doi.org/10.1201/9781003482826-9

Sharma, M., & Medhi, B. (2025). Advances in Nanoformulations for Cosmetic Applications. International Journal of Pharmaceutical Sciences and Nanotechnology (IJPSN), 18(3), 8035-8039. https://doi.org/10.37285/ijpsn.2025.18.3.1

Sharma, S., Verma, A., Teja, B. V., Shukla, P., & Mishra, P. R. (2015). Development of stabilized Paclitaxel nanocrystals: *In vitro* and *in vivo* efficacy studies. European Journal of Pharmaceutical Sciences, 69, 51-60. https://doi.org/10.1016/j.ejps.2014.11.012

Shegokar, R., & Singh, K. K. (2011). Surface modified nevirapine nanosuspensions for viral reservoir targeting: *In vitro* and *in vivo* evaluation. International Journal of Pharmaceutics, 421(2), 341-352. https://doi.org/10.1016/j.ijpharm.2011.09.041

Shete, G., Pawar, Y. B., Thanki, K., Jain, S., & Bansal, A. K. (2015). Oral bioavailability and pharmacodynamic activity of hesperetin nanocrystals generated using a novel bottom-up technology. Molecular Pharmaceutics, 12(4), 1158-1170. https://doi.org/10.1021/mp5008647

Singh, B., Upadhyay, P. K., & Kumar, M. (2022). Nanocrystal for Dermatological Application: A Comprehensive Review. Current Nanoscience, 18(1), 48-60. https://doi.org/10.2174/1573413717666210121143038

Sivanathan, G., Rajagopal, S., Mahadevaswamy, G., Angamuthu, G., & Dhandapani, N. V. (2024). Pharmaceutical Nanocrystals: An Extensive Overview. International Journal of Applied Pharmaceutics, 16(6), 1-9. https://doi.org/10.22159/ijap.2024v16i6.52257

Srinivas, M., Sharma, P., & Jhunjhunwala, S. (2021). Phagocytic Uptake of Polymeric Particles by Immune cells Under Flow Conditions. Cold Spring Harbor Laboratory, 18(2), 4501-4510. https://doi.org/10.1101/2021.04.01.438153

Srivalli, K., & Mishra, B. (2015). Drug nanocrystals: four basic prerequisites for formulation development and scale-up. Current Drug Targets, 16(2), 136-147. https://doi.org/10.2174/1389450115666141120114036

Stahr, P.-L., Grewal, R., Eckert, G. P., & Keck, C. M. (2021). Investigating hesperetin nanocrystals with tailor-made sizes for the prevention and treatment of Alzheimer\u2019s disease. Drug Delivery and Translational Research, 11(2), 659-674. https://doi.org/10.1007/s13346-020-00888-0

Sun, L., Hu, Y., & Zhang, L. (2018). Recent Trends in Nanocrystals for Pharmaceutical Applications. Current Pharmaceutical Design, 24(21), 2394-2402. https://doi.org/10.2174/1381612824666180524103341

Toziopoulou, F., Malamatari, M., Nikolakakis, I., & Kachrimanis, K. (2017). Production of aprepitant nanocrystals by wet media milling and subsequent solidification. International Journal of Pharmaceutics, 533(2), 324-334. https://doi.org/10.1016/j.ijpharm.2017.02.065

Ueda, K., Iwai, T., Sunazuka, Y., Chen, Z., Kato, N., Higashi, K., & Moribe, K. (2019). Effect of molecular weight of hypromellose on mucin diffusion and oral absorption behavior of fenofibrate nanocrystal. International Journal of Pharmaceutics, 564, 39-47. https://doi.org/10.1016/j.ijpharm.2019.04.033

Varma, T. S., Gavhane, Y. N., & Shamkuwar, P. B. (2026). Solubility Enhancement Strategies for Poorly Soluble Drugs: Recent Trends. Journal of Drug Delivery and Therapeutics, 16(3), 356-365. https://doi.org/10.22270/jddt.v16i3.7623

Verma, N. (2022). A Review Article On Solubility Enhancement Techniques. Zenodo (CERN European Organization for Nuclear Research). https://doi.org/10.5281/zenodo.7227422

Vogt, M., Kunath, K., & Dressman, J. B. (2007). Dissolution enhancement of fenofibrate by micronization, cogrinding and spray-drying: Comparison with commercial preparations. European Journal of Pharmaceutics and Biopharmaceutics, 68(2), 283-288. https://doi.org/10.1016/j.ejpb.2007.05.010

Wang, J., Lv, F.-M., Wang, D.-L., Du, J.-L., *et al.* (2020). Synergistic Antitumor Effects on Drug-Resistant Breast Cancer of Paclitaxel/Lapatinib Composite Nanocrystals. Molecules, 25(3), 604. https://doi.org/10.3390/molecules25030604

Wang, J., Xue, X., & Miao, X. (2023). Antioxidant Effects of Quercetin Nanocrystals in Nanosuspension against Hydrogen Peroxide-Induced Oxidative Stress in a Zebrafish Model. Pharmaceuticals, 16(9), 1209. https://doi.org/10.3390/ph16091209

Wankhade, V., & Dudhe, P. (2024). Basic Concepts and Application of Prodrug Design. International Journal of Advanced Research in Science, Communication and Technology, 57-65. https://doi.org/10.48175/ijarsct-22811

Wen, J., Lei, C., Hua, S., Cai, L., Dai, H., *et al.* (2025). Regulation of macrophage uptake through the bio-nano interaction using surface functionalized mesoporous silica nanoparticles with large radial pores. Journal of Materials Chemistry. B, 13(1), 137-150. https://doi.org/10.1039/d4tb01124d

Xue, L., Ding, J., Liu, Y., Ma, Y., Yang, C., *et al.* (2024). Strategies and methods of nanocrystal technology for targeting drug delivery. Journal of Nanoparticle Research, 26(6). https://doi.org/10.1007/s11051-024-06025-z

Yang, H., Kim, H., Jung, S., Seo, H., Nida, S. K., Yoo, S.-Y., & Lee, J. (2018). Pharmaceutical Strategies for Stabilizing Drug Nanocrystals. Current Pharmaceutical Design, 24(21), 2362-2374. https://doi.org/10.2174/1381612824666180515125247

Yang, Y., Qin, H., & Peng, X. (2016). Intramolecular Entropy and Size-Dependent Solution Properties of Nanocrystal-Ligands Complexes. Nano Letters, 16(4), 2127-2132. https://doi.org/10.1021/acs.nanolett.6b00737

Yang, Y., Qin, H., Jiang, M., Lin, L., Fu, T., Dai, X., *et al.* (2016). Entropic Ligands for Nanocrystals: From Unexpected Solution Properties to Outstanding Processability. Nano Letters, 16(4), 2133-2138. https://doi.org/10.1021/acs.nanolett.6b00730

Ye, X., Patil, H., Feng, X., Tiwari, R. V., Lu, J., *et al.* (2015). Conjugation of Hot-Melt Extrusion with High-Pressure Homogenization: a Novel Method of Continuously Preparing Nanocrystal Solid Dispersions. AAPS PharmSciTech, 17(1), 78-88. https://doi.org/10.1208/s12249-015-0389-7

Yong, J. M., Mantaj, J., Cheng, Y., & Vllasaliu, D. (2019). Delivery of Nanoparticles across the Intestinal Epithelium via the Transferrin Transport Pathway. Pharmaceutics, 11(7), 298. https://doi.org/10.3390/pharmaceutics11070298

Yu, B.-X., Zhang, Y.-H., Li, C.-Y., Xian, J.-Y., *et al.* (2023). Cell Protection and Crystal Endocytosis Inhibition by Sulfated *Laminaria* Polysaccharides Against Nano-COM-Induced

Oxidative Damage in Renal Epithelial Cells. ACS Omega, 8(8), 7816-7828. https://doi.org/10.1021/acsomega.2c07584

Yue, P., Zhou, W., Huang, G., Lei, F., Chen, Y., *et al.* (2022). Nanocrystals based pulmonary inhalation delivery system: advance and challenge. Drug Delivery, 29(1), 637-651. https://doi.org/10.1080/10717544.2022.2039809

Zhang, G., Wang, Y., Zhang, Z., He, Z., Liu, Y., & Fu, Q. (2021). FRET imaging revealed that nanocrystals enhanced drug oral absorption by dissolution rather than endocytosis: A case study of coumarin 6. Journal of Controlled Release, 332, 225-232. https://doi.org/10.1016/j.jconrel.2021.02.025

Zhang, H., Hollis, C. P., Zhang, Q., & Li, T. (2011). Preparation and antitumor study of camptothecin nanocrystals. International Journal of Pharmaceutics, 415(1-2), 293-300. https://doi.org/10.1016/j.ijpharm.2011.05.075

Zhang, W., Taheri-Ledari, R., Ganjali, F., Mirmohammadi, S. S., *et al.* (2023). Effects of morphology and size of nanoscale drug carriers on cellular uptake and internalization process: a review. RSC Advances, 13(1), 80-114. https://doi.org/10.1039/d2ra06888e

Zhang, Y., Zhang, C., Chen, J., Liu, L., Hu, M., Li, J., & Bi, H. (2017). Trackable Mitochondria-Targeting Nanomicellar Loaded with Doxorubicin for Overcoming Drug Resistance. ACS Applied Materials & Interfaces, 9(30), 25152-25163. https://doi.org/10.1021/acsami.7b07219

Zhang, Z., Chu, Y., Li, C., Tang, W., Qian, J., *et al.* (2020). Anti-PEG scFv corona ameliorates accelerated blood clearance phenomenon of PEGylated nanomedicines. Journal of Controlled Release, 330, 493-501. https://doi.org/10.1016/j.jconrel.2020.12.047

Zhao, J., Du, J., Wang, J., An, N., Zhou, K., Hu, X., Dong, Z., & Liu, Y. (2021). Folic Acid and Poly(ethylene glycol) Decorated Paclitaxel Nanocrystals Exhibit Enhanced Stability and Breast Cancer-Targeting Capability. ACS Applied Materials & Interfaces, 13(12), 14577-14586. https://doi.org/10.1021/acsami.1c00184

Zheng, H., Li, J., & Leung, S. S. Y. (2024). Inhalable polysorbates stabilized nintedanib nanocrystals to facilitate pulmonary nebulization and alveolar macrophage evasion. Biomaterials Advances, 166, 214084. https://doi.org/10.1016/j.bioadv.2024.214084

Zhou, Y., Du, J., Wang, L., & Wang, Y. (2016). State of the art of nanocrystals technology for delivery of poorly soluble drugs. Journal of Nanoparticle Research, 18(9). https://doi.org/10.1007/s11051-016-3575-y

Cite this article: Mali KD, Yadav S, Dubey AN, Deore GB. Drug Nanocrystals: From Formulation Design to Clinical Translation. Int. J. Pharm. Investigation. 10.5530/ijpi.20270097.