# Two Spices, One Carrier: Curcumin–Piperine Co-Loaded Nanostructured Lipid Carriers

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***Abstract*—Curcumin has one of the widest preclinical activity profiles of any natural product and one of the worst pharmacokinetic records. In healthy volunteers given a single 10 or 12 g oral dose, only one of twelve subjects had detectable free curcumin in serum at any of fourteen sampling points; glucuronide and sulfate conjugates were detected in all. The barrier is not one problem but three: aqueous solubility of the order of nanograms per millilitre, extensive intestinal and hepatic glucuronidation and sulfation, and intestinal efflux, each capable of limiting exposure independently. A formulation that addresses only the first will plateau. Piperine, the pungent alkaloid of black pepper, addresses the second and third: it inhibits UDP-glucose dehydrogenase and glucuronidation in liver and intestine, and it inhibits human P-glycoprotein and CYP3A4, with reported $IC_{50}$ values of 15.5 and 74.1 µM for P-glycoprotein-mediated digoxin and cyclosporine transport in Caco-2 monolayers. Co-administered with 2 g of curcumin in humans, 20 mg of piperine raised bioavailability by a reported 2000%. Nanostructured lipid carriers address the first and contribute to the third: a binary solid–liquid lipid matrix whose crystal imperfections accommodate a high payload and resist expulsion on storage, presenting curcumin pre-solubilised, protecting it from the alkaline degradation that destroys it in intestinal fluid, and offering access to the intestinal lymphatic route that bypasses the liver entirely. Co-loading the two in one carrier is therefore not a convenience but a mechanistic complementarity: the carrier and the second active attack different barriers, and co-encapsulation ensures they arrive at the enterocyte together. This review assembles the pharmacokinetic evidence, examines each mechanism, surveys reported curcumin and curcumin–piperine lipid nanocarriers, sets out the formulation variables and their optimisation, and argues that intestinal permeability rather than particle size is the response that should drive the design.**

***Index Terms*—Curcumin; piperine; nanostructured lipid carriers; oral bioavailability; intestinal permeability; P-glycoprotein; glucuronidation; Box–Behnken design**

## I. INTRODUCTION

Curcumin is the most studied and least absorbed of the major dietary polyphenols. Isolated from the rhizome of Curcuma longa, it has documented anti-inflammatory, antioxidant, antineoplastic, antidiabetic and neuroprotective activity across a very large preclinical literature, and it has reached clinical trial in indications from colorectal adenoma to pancreatic cancer [1,2,3]. Phase 1 work established that it is remarkably well tolerated: doses up to 12 g daily have been given without dose-limiting toxicity [3,7].

That tolerability is itself diagnostic. A compound that can be given at twelve grams without adverse effect is a compound that is not reaching the systemic circulation, and the pharmacokinetic data confirm it. The review by Anand and colleagues framed the problem as poor absorption, rapid metabolism and rapid systemic elimination acting together [4], and two decades of formulation work have followed from that diagnosis.

Most of that work has targeted solubility. Nanoemulsions, solid dispersions, phospholipid complexes, cyclodextrin inclusion complexes, polymeric nanoparticles, liposomes and lipid nanoparticles have all been applied, and most improve exposure. But solubility is only the first of three barriers, and a formulation that dissolves curcumin more efficiently simply presents more substrate to the conjugating enzymes of the enterocyte. The ceiling moves; it does not disappear.

Piperine attacks the other two barriers. The pungent alkaloid of Piper nigrum is a classical bioenhancer, and its mechanisms are well characterised: inhibition of glucuronidation in liver and intestine, and inhibition of P-glycoprotein and CYP3A4 [15,16,17]. Co-administered with curcumin in healthy volunteers it produced one of the largest bioavailability enhancements reported for any oral combination [14].

The proposition examined here is that these two interventions are complementary rather than alternative, and that co-encapsulating both actives in a single nanostructured lipid carrier does something that administering them separately does not: it guarantees that the metabolic inhibitor is present at the enterocyte at the same time and place as the substrate it is protecting. This review assembles the evidence for each component, examines what nanostructured lipid carriers contribute that simpler lipid systems do not, and sets out how such a system should be designed and — more importantly — what should be measured.

## II. CURCUMIN: THREE BARRIERS, NOT ONE

### 2.1 What the human data show

Table 1 collects the human pharmacokinetic studies, and they tell a consistent story.

The defining dataset is that of Vareed and colleagues, who gave healthy volunteers a single oral dose of 10 g (n = 6) or 12 g (n = 6) and sampled serum from 0.25 to 72 h. Using an HPLC assay with a detection limit of 50 ng $mL^{-1}$, only one subject had detectable free curcumin at any of the fourteen time points assayed, while curcumin glucuronides and sulfates were detected in all subjects. Time to peak was 3.29 ± 0.43 h and the half-life 6.77 ± 0.83 h, with a glucuronide-to-sulfate ratio of 1.92:1, and the conjugates were present as either glucuronide or sulfate rather than as mixed conjugates [6].

Earlier studies are consistent. A Phase 1 trial in Taiwan reported serum concentrations peaking at 0.41 to 1.75 μM one hour after oral doses of 4 to 8 g [7]. In a UK trial, plasma concentrations of curcumin and its sulfate and glucuronide were of the order of 0.01 μM one hour after 3.6 g, and nothing was detectable below that dose [8]. In advanced pancreatic cancer patients given 8 g daily, circulating curcumin was detectable only as glucuronide and sulfate conjugates at low steady-state levels, peaking at 22 to 41 ng $mL^{-1}$ [9].

Animal data locate the loss. Early work showed that most of an oral dose of curcumin in rats is recovered in the faeces with only traces in urine [12,13], and metabolism studies established reduction to tetrahydro-, hexahydro- and octahydrocurcumin alongside glucuronidation and sulfation as the principal biotransformation routes [10,11].

Three distinct obstacles emerge, and distinguishing them is the whole basis of a rational formulation strategy.

The first is dissolution and stability. Curcumin is practically insoluble in water at acidic and neutral pH, and — less often emphasised — it degrades rapidly at the mildly alkaline pH of the small intestine. A delivery system that keeps curcumin out of contact with intestinal fluid is therefore protecting as well as solubilising it.

The second is pre-systemic conjugation. Curcumin is a substrate for UDP-glucuronosyltransferases and sulfotransferases in both the enterocyte and the hepatocyte, and the human data show that essentially everything that is absorbed appears in plasma already conjugated [6,10,11]. Whether the conjugates retain activity is contested; what is not contested is that free curcumin scarcely circulates.

The third is efflux. Curcumin interacts with intestinal efflux transporters, so a fraction of what enters the enterocyte is returned to the lumen before it can cross the basolateral membrane.

A nanocarrier addresses the first. It does not, by itself, address the second or third — and that is precisely the gap a co-loaded bioenhancer fills.

Table 1. Human pharmacokinetic studies of oral curcumin. Across dose levels from 3.6 to 12 g, free curcumin is at or below the limit of detection while conjugates are consistently measurable.

| Study population and dose | Analytical finding | Parameters reported | Ref. |
|---|---|---|---|
| Healthy volunteers, single oral 10 g (n = 6) or 12 g (n = 6); serum 0.25–72 h | HPLC limit of detection 50 ng $mL^{-1}$. Only one subject had detectable free curcumin at any of 14 time points; glucuronides | $T_{max}$ 3.29 ± 0.43 h; $t_{½}$ 6.77 ± 0.83 h; glucuronide : sulfate 1.92 : 1; conjugates present singly, not mixed | [6] |

| Study population and dose | Analytical finding | Parameters reported | Ref. |
|---|---|---|---|
| | and sulfates detected in all subjects | | |
| Patients with high-risk or pre-malignant lesions, 4–8 g orally | Serum curcumin measurable but low | Peak serum 0.41–1.75 μM at 1 h | [7] |
| Phase 1 oral curcumin, 3.6 g | Curcumin and its sulfate and glucuronide detectable only at the highest dose | Plasma concentrations of the order of 0.01 μM at 1 h; nothing detectable below 3.6 g per day | [8] |
| Advanced pancreatic cancer, 8 g daily | Circulating curcumin detectable as glucuronide and sulfate conjugates only | Low steady-state levels peaking at 22–41 ng $mL^{-1}$, relatively constant over 4 weeks; biological activity observed in some patients | [9] |
| Healthy volunteers, 2 g curcumin ± 20 mg piperine (n = 10, crossover) | Curcumin alone undetectable or very low; much higher concentrations with piperine from 0.25 to 1 h | Bioavailability increased by 2000% in humans and by 154% in rats (2 g $kg^{-1}$ curcumin with 20 mg $kg^{-1}$ piperine); no adverse effects | [14] |

## III. PIPERINE: A SECOND ACTIVE RATHER THAN AN EXCIPIENT

### 3.1 Three mechanisms

Piperine's status as a bioavailability enhancer rests on mechanisms that have been characterised individually, and it is worth separating them because they act at different points.

Inhibition of glucuronidation came first. Atal and colleagues established the biochemical basis of piperine's bioenhancement, showing it to be a potent inhibitor of drug metabolism [17], and Reen and colleagues subsequently demonstrated non-competitive impairment of UDP-glucose dehydrogenase and of glucuronidation activity in liver and small intestine [16]. Since glucuronidation is the dominant route of curcumin elimination, this is the mechanistically closest match.

Inhibition of P-glycoprotein and CYP3A4 came later and is the best quantified. Bhardwaj and colleagues showed that piperine inhibits P-glycoprotein-mediated transport of digoxin and cyclosporine A across Caco-2 monolayers with $IC_{50}$ values of 15.5 and 74.1 μM respectively, and inhibits CYP3A4-catalysed verapamil metabolism in human liver microsomes in a mixed fashion with $K_i$ values of 36 to 77 μM. Because both proteins are expressed in enterocytes and hepatocytes and contribute substantially to first-pass elimination, the authors concluded that dietary piperine could affect plasma concentrations of their substrates [15]. Later work found that oral piperine altered intestinal and hepatic P-glycoprotein expression differentially in rats [20].

Effects on the absorptive surface itself are the third and least quantified. Piperine has been reported to modify intestinal permeability characteristics [18], and increased splanchnic blood flow and slowed gastrointestinal transit have both been proposed as contributors. These are plausible and probably real, but they are supported by less direct evidence than the enzyme and transporter effects and should be presented as such.

Clinically, piperine's bioenhancement is not confined to curcumin: it increases the bioavailability of propranolol and theophylline in healthy volunteers [19], and of phenytoin and rifampicin — the observation that prompted the Caco-2 work in the first place [15].

### 3.2 How to read the 2000% figure

The landmark study is unambiguous in what it reported and is frequently over-read. Shoba and colleagues gave rats 2 g $kg^{-1}$ curcumin alone, achieving moderate serum concentrations over 4 h; adding 20 mg $kg^{-1}$ piperine increased bioavailability by 154%, with significantly increased time to maximum and significantly decreased elimination half-life and clearance. In ten healthy male volunteers

given 2 g of curcumin, serum levels were undetectable or very low; concomitant administration of 20 mg piperine produced much higher concentrations from 0.25 to 1 h, and the increase in bioavailability was 2000%, with no adverse effects [14].

Three qualifications belong alongside that figure in any serious treatment of the subject.

The baseline was near zero. A 2000% increase means a twenty-one-fold rise from a starting point that was at or below the limit of quantitation, so the ratio is large partly because the denominator is very small. A twenty-one-fold rise on an undetectable baseline may still leave concentrations far below those active in vitro.

The study was small and has not been reproduced at that magnitude. It involved ten volunteers in a single crossover, and subsequent assessments have generally reported considerably more modest enhancements under routine conditions. Figure 1A plots the reported values alongside other strategies with that caveat attached.

The mechanism is a drug-interaction mechanism. Inhibiting CYP3A4 and P-glycoprotein is exactly how clinically significant interactions occur, and a formulation whose performance depends on that inhibition carries the same liability. Narrow-therapeutic-index co-medication is a genuine caution for any curcumin–piperine product, and it deserves to be stated rather than omitted.

None of this undermines the strategy. It does mean that a co-loaded formulation should be justified by the mechanistic complementarity set out in Figure 2, and evaluated against a physical mixture of the same two actives at the same doses — which is the comparison that isolates what co-encapsulation contributes.

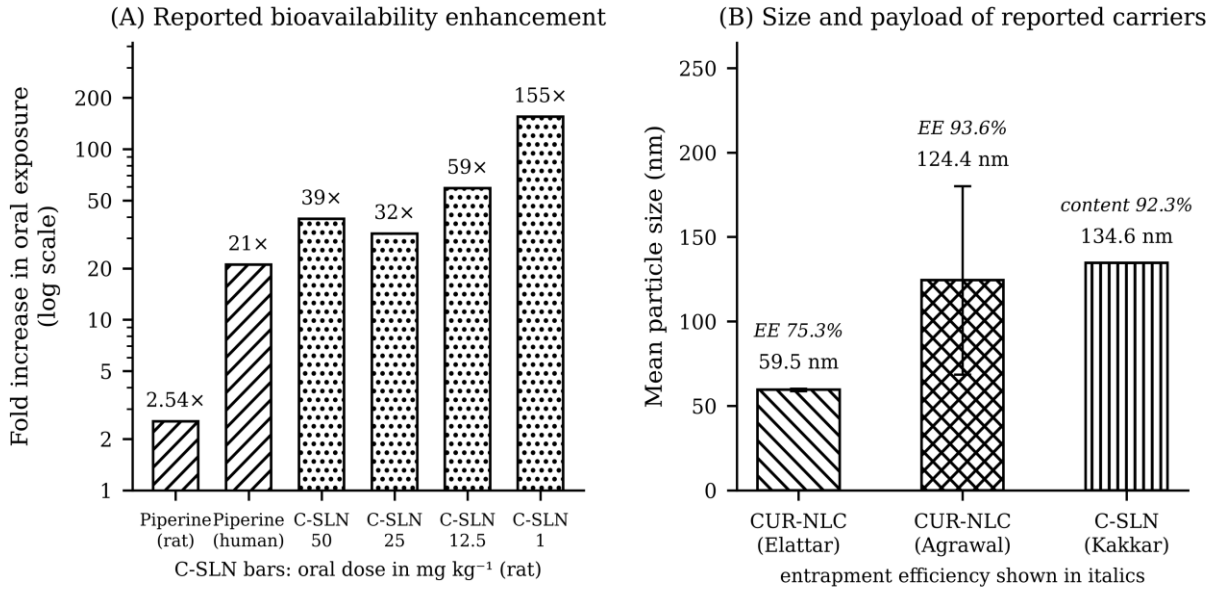



Fig. 1. (A) Reported fold increases in oral curcumin exposure. Piperine co-administration gave a 2.5-fold increase in rats and a 21-fold increase (reported as 2000%) in ten healthy volunteers [14]; curcumin-loaded solid lipid nanoparticles gave 39-fold at 50 mg kg$^{-1}$ and 155-fold at 1 mg kg$^{-1}$ in rats, the enhancement rising as the dose falls [28]. Values come from different species, doses and assays and are indicative rather than strictly comparable; the human piperine figure rests on a baseline at or below the limit of quantitation. (B) Particle size of reported curcumin lipid nanocarriers, with entrapment efficiency or total drug content annotated above each bar [26,28,30].

## IV. WHY A NANOSTRUCTURED LIPID CARRIER

Lipid nanoparticles arrived in two generations, and the distinction matters for a high-dose, poorly soluble payload like curcumin. Table 2 compares the options.

Solid lipid nanoparticles, developed in the early 1990s, consist of a single solid lipid stabilised by surfactant and offer biocompatible excipients, organic-solvent-free manufacture and scalability [21]. Their weakness is crystallinity: a single lipid crystallises into a highly ordered modification with little room between the

chains, which limits drug loading, and continued polymorphic transition during storage expels drug that was loaded successfully at manufacture [21,27].

Nanostructured lipid carriers were the response. By blending a solid lipid with a liquid lipid — an oil — the matrix is deliberately disordered: spatially dissimilar molecules cannot pack into a perfect lattice, and the resulting imperfections accommodate more drug in amorphous clusters and resist expulsion on storage, while the matrix remains solid at room and body temperature [22,23]. Three structural types are described: the imperfect crystal type, the amorphous type, and the multiple oil-in-fat-in-water type in which oil nanocompartments are enclosed within the solid matrix [24,25,26].

For curcumin–piperine co-loading this matters in a specific way. Curcumin is a crystalline solid with modest solubility even in lipids, while piperine is markedly more soluble in liquid lipids than in solid ones. A binary matrix with a liquid lipid fraction therefore offers a compartment in which both can be accommodated, which a single solid lipid does not — and the oil fraction becomes a formulation variable with a direct bearing on how much of each active can be loaded.

A fourth advantage is specific to the oral route and is frequently underexploited. Lipid nanoparticles of appropriate composition can be taken up into the intestinal lymphatics rather than the portal circulation, and lymphatic transport bypasses the liver and therefore hepatic first-pass metabolism entirely [34,35]. Lymphatic uptake is favoured by long-chain rather than medium-chain lipids, since long-chain fatty acids are re-esterified to triglycerides and packaged into chylomicrons [36]. For a molecule whose principal loss is conjugative first-pass metabolism, this is arguably the most interesting property of the carrier, and lipid selection is therefore not merely a question of drug solubility.

Table 2. Lipid and particulate carriers for oral curcumin, with the property that distinguishes each.

| Carrier | Matrix | Strengths | Limitations for curcumin–piperine co-loading |
|---|---|---|---|
| Nanoemulsion | Liquid oil droplets stabilised by surfactant | Simple, high solubilisation capacity, large interfacial area | No solid matrix, so little control of release; thermodynamically unstable; usually needs high surfactant |
| Solid lipid nanoparticle (SLN) | Single solid lipid, solid at body temperature | Biocompatible excipients, organic-solvent-free manufacture, scalable, controlled release | Ordered crystal lattice limits payload; polymorphic transition on storage expels loaded drug |
| Nanostructured lipid carrier (NLC) | Binary blend of solid and liquid lipid; disordered but solid matrix | Crystal imperfections raise payload and resist expulsion; liquid fraction suits a second lipophilic active; lymphatic uptake accessible with long-chain lipids | Requires solid–liquid lipid miscibility screening; oil fraction must be bounded or the matrix softens toward a nanoemulsion |
| Polymeric nanoparticle | Biodegradable polymer, e.g. PLGA | Robust, tunable release; reported ≥ 9-fold higher curcumin bioavailability than curcumin with piperine | Organic solvent in manufacture; polymer degradation products; no intrinsic lymphatic advantage |
| Free drug plus bioenhancer | No carrier; co-administered curcumin and piperine | Simple; the basis of the largest reported human enhancement | No protection from intestinal degradation; no guarantee that both actives reach the same enterocyte together |

4.1 What reported curcumin lipid nanocarriers achieve
The literature is substantial and consistent. Figure 1B plots the reported particle sizes and entrapment efficiencies.

Kakkar and colleagues loaded curcumin into solid lipid nanoparticles of 134.6 nm mean size with a total drug content of 92.33 ± 1.63% by a microemulsification technique, and reported improvements in oral bioavailability in rats of 39-, 32-, 59- and 155-fold at doses of 50, 25, 12.5 and 1 mg $kg^{-1}$ relative to solubilised curcumin [28]. The inverse relationship between dose and fold-enhancement is itself informative: the carrier helps most where absorption is most solubility-limited.

Shaikh and colleagues prepared curcumin-encapsulated polymeric nanoparticles of 264 nm with a polydispersity index of 0.31 and 76.9% entrapment at 15% loading, which withstood ICH accelerated stability conditions for three months, and reported at least a nine-fold increase in oral bioavailability compared with curcumin administered with piperine as absorption enhancer [29]. That comparator choice makes the study unusually relevant here: it indicates that a particulate carrier can outperform piperine co-administration alone.

Among nanostructured lipid carriers specifically, Agrawal and colleagues optimised a curcumin-loaded system by a three-factor three-level Box–Behnken design using solid-to-liquid lipid ratio, surfactant concentration and ultrasonication time as independent variables, preparing the particles by modified melt emulsification followed by ultrasonication. The optimised carrier had a size of 124.37 ± 55.81 nm, polydispersity index 0.201, zeta potential −17.2 ± 2.35 mV, entrapment efficiency 93.62 ± 0.68% and drug release 92.73 ± 0.06% [30]. Other work has reported curcumin NLC pharmacokinetics and anticancer activity [31] and curcumin NLCs of 59.49 ± 0.71 nm with 75.33 ± 5.06% entrapment and prolonged release over 72 h [26].

Work on the specific combination exists and is recent. Bhairy and colleagues developed and validated a bivariate ultraviolet spectroscopic method for simultaneous estimation of curcumin and piperine in a combined nanoparticulate system, reporting a formulation containing glyceryl distearate as solid lipid and medium-chain triglycerides as liquid lipid [33], and subsequently assessed ex vivo intestinal permeability and lymphatic uptake of curcumin and piperine-loaded nanostructured lipid carriers, finding higher permeation from the co-loaded carriers than from a suspension of the two actives [32].

Two observations follow. The platform works, repeatedly and across groups. And the analytical problem of a dual-loaded system — quantifying two actives simultaneously in the same matrix — has already been solved, which removes one practical obstacle to the work.

## V. FORMULATION VARIABLES AND THEIR OPTIMISATION

Table 3 sets out the variables and the quality attributes each governs.

Lipid selection is the first and most consequential decision, and it carries a trade-off that is often missed. The solid lipid determines matrix crystallinity and therefore loading and expulsion behaviour; the liquid lipid creates the imperfections that accommodate drug. But chain length also determines the absorption route: long-chain lipids favour chylomicron formation and lymphatic transport, while medium-chain lipids are absorbed predominantly into the portal blood [36]. A formulation optimised for maximum entrapment using medium-chain triglycerides may have optimised away the lymphatic bypass that gives the carrier its metabolic advantage. Whether lymphatic transport or payload matters more is an empirical question, and it should be asked explicitly rather than settled by default.

The solid-to-liquid lipid ratio is the central compositional variable. Too little oil and the matrix crystallises toward solid-lipid-nanoparticle behaviour with its loading limit and expulsion risk; too much and the particle softens toward a nanoemulsion, losing the solid matrix that controls release. Typical reported systems use oil fractions well below half the lipid phase.

Surfactant concentration controls interfacial tension and therefore attainable particle size and physical stability, against the usual constraint that surfactant load is a tolerability variable. Ultrasonication time or homogenisation energy controls droplet break-up with diminishing and eventually reversing returns, since prolonged sonication heats the dispersion and can promote coalescence as well as degrade a thermolabile polyphenol.

The variable unique to this system is the curcumin-to-piperine ratio. It has no counterpart in a single-drug formulation and no obvious default: piperine must be present in sufficient quantity to inhibit enterocyte glucuronidation and efflux, but it occupies matrix space that would otherwise carry curcumin, and it is pharmacologically active in its own right with the interaction liability noted above. Treating this ratio as a formal design factor rather than fixing it at a literature value is the single most useful methodological choice available in such a study.

The Quality by Design framework set out in ICH Q8(R2) provides the structure — a quality target product profile, critical quality attributes derived from it, and a design space within which performance is robust [42,47]. A three-factor three-level Box–Behnken design requires fifteen runs including three centre-point replicates and estimates a full quadratic surface without extreme corner combinations [40,41], and it has already been applied successfully to curcumin NLCs with exactly these factors [30].

The responses deserve more thought than they usually receive. Particle size, polydispersity index and entrapment efficiency are conventional, easily measured and weakly connected to performance. The response that matters for this formulation is apparent permeability, and including it as a modelled response rather than a final check on the optimised batch is what would convert a particle-size optimisation into a delivery optimisation.

Table 3. Formulation and process variables for a curcumin–piperine co-loaded NLC, and the quality attributes each governs.

| Variable | Effect | Attributes affected |
|---|---|---|
| Solid lipid identity | Determines matrix crystallinity and the degree of lattice order | Entrapment efficiency; drug expulsion on storage; release rate |
| Liquid lipid identity and chain length | Creates matrix imperfections; chain length decides portal versus lymphatic absorption | Payload; lymphatic uptake and therefore hepatic first-pass bypass |
| Solid : liquid lipid ratio | Too little oil gives SLN-like crystallinity and expulsion; too much softens the matrix toward a nanoemulsion | Entrapment; particle size; release control; physical stability |
| Curcumin : piperine ratio | Piperine must suffice to inhibit enterocyte glucuronidation and efflux, but occupies matrix space | Relative payloads; apparent permeability; interaction liability |
| Surfactant type and concentration | Lowers interfacial tension and controls attainable size and physical stability | Particle size; PDI; zeta potential; tolerability |
| Ultrasonication or homogenisation energy | Drives droplet break-up with diminishing and eventually reversing returns | Particle size and distribution; integrity of a thermolabile polyphenol |
| Process temperature and cooling rate | Governs the polymorphic form into which the matrix solidifies | Crystallinity; expulsion on storage; long-term entrapment |

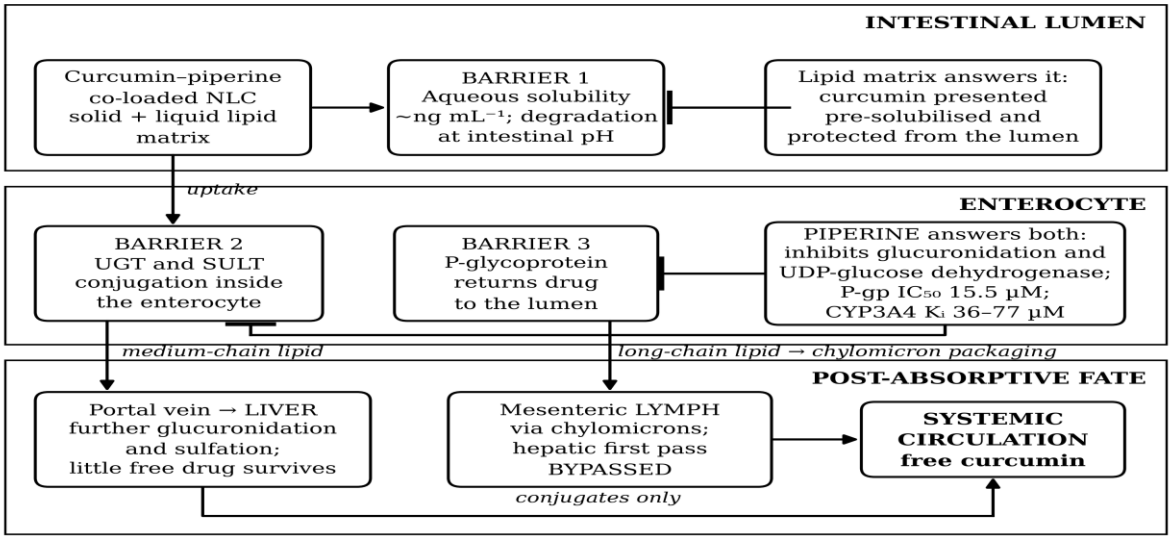


Fig. 2. The three barriers to oral curcumin absorption and the point at which each intervention acts. In the lumen, poor aqueous solubility and degradation at intestinal pH limit how much curcumin is available: the lipid matrix of the carrier addresses this by presenting curcumin pre-solubilised and protected. In the enterocyte, UDP-glucuronosyltransferases and sulfotransferases conjugate most of what enters while P-glycoprotein returns a further fraction to the lumen: piperine inhibits both. Absorbed drug then passes either to the portal vein and the liver, where conjugation continues, or — if the carrier is built from long-chain lipids and taken up into chylomicrons — into the mesenteric lymph, which drains to the systemic circulation and bypasses the liver entirely. Simplified; not all metabolic routes are shown.

## VI. MEASURING WHAT THE FORMULATION CLAIMS TO IMPROVE

The project is titled around intestinal permeability, and the methods for measuring it differ in what they can establish.

The Caco-2 monolayer is the standard in vitro model, with established protocols for determining apparent permeability and a long-validated correlation with human oral absorption [37,38]. For this system it has a specific advantage: because piperine's P-glycoprotein inhibition was itself characterised in Caco-2 cells [15], bidirectional transport experiments comparing apical-to-basolateral with basolateral-to-apical flux allow the efflux ratio to be calculated, and a fall in efflux ratio on co-loading piperine is direct evidence that the intended mechanism is operating. A curcumin-only NLC should be included alongside the co-loaded carrier for that comparison to mean anything.

The everted intestinal sac, described by Wilson and Wiseman seventy years ago and still serviceable, retains the full intestinal wall including the mucus layer and the enzymatic machinery of the enterocyte [39]. That last point is why it is particularly appropriate here: a model that lacks intestinal glucuronidation cannot report on a mechanism that depends on inhibiting it, and the fraction of curcumin appearing in the serosal fluid as free rather than conjugated drug is the observation that would most directly demonstrate what piperine is contributing.

Lymphatic uptake requires its own experiment — typically mesenteric lymph duct cannulation or a chylomicron-flow-blocking approach using cycloheximide — and is the measurement that would establish whether the carrier is in fact bypassing the liver [35,36]. It is rarely performed, which is why the claim that lipid nanoparticles exploit lymphatic transport is more often asserted than demonstrated. Recent work on curcumin–piperine NLCs has begun to address this directly [32].

In vivo, a four-arm oral pharmacokinetic study in rats is the minimum that would support the central claim: the co-loaded NLC, a curcumin-only NLC, a physical mixture of curcumin and piperine at the same doses, and a plain curcumin suspension. Without the third arm the study cannot distinguish the benefit of co-encapsulation from the benefit of co-administration, and that distinction is the entire rationale for the formulation. Plasma should be assayed for free curcumin and, separately, for total curcumin after enzymatic hydrolysis, since the ratio between them is the direct readout of whether glucuronidation has been inhibited.

Supporting work follows the standard pattern: release data fitted to zero-order, first-order, Higuchi and Korsmeyer–Peppas models [45,46]; analytical methods validated to ICH Q2(R1), with the bivariate approach already described for this drug pair [33,43]; and accelerated stability testing to ICH Q1A(R2) [44], monitoring particle size, zeta potential and — critically for a lipid matrix — drug expulsion, which is the failure mode nanostructured lipid carriers exist to prevent and the one that only appears on storage.

## VII. GAPS AND OUTLOOK

Several gaps are worth naming directly.

The comparator gap. Most co-delivery studies compare the co-loaded carrier with free drug or with a plain suspension. The arm that isolates the contribution of co-encapsulation is a physical mixture of the two actives at the same doses, and it is reported far less often than it should be.

The free-versus-conjugated gap. Plasma assays frequently report total curcumin after hydrolysis without reporting the free fraction separately. For a formulation whose stated mechanism is inhibition of glucuronidation, the free-to-total ratio is the primary

evidence, and reporting only the total obscures exactly the quantity of interest.

The lymphatic gap. Lymphatic transport is routinely invoked as a mechanism for lipid nanoparticles and rarely measured. Where it is measured, lipid chain length should be treated as a design variable rather than inherited from a previous formulation.

The ratio gap. The curcumin-to-piperine ratio is typically fixed by reference to the 2 g : 20 mg proportion of the 1998 human study, which was a co-administration study using free drug and has no necessary relevance to a co-encapsulated carrier delivering both actives to the same enterocyte at the same time. This ratio deserves to be optimised.

The safety gap. Piperine's bioenhancement is inhibition of CYP3A4 and P-glycoprotein, the same mechanism underlying clinically significant drug interactions [15]. A product designed around that inhibition should carry an explicit interaction assessment rather than treating piperine as an inert excipient.

The translational gap. No curcumin–piperine co-loaded lipid nanocarrier appears to have been evaluated clinically, despite both components having human pharmacokinetic data and despite curcumin's exceptional safety margin at high oral doses. Given that tolerability, a first-in-human pharmacokinetic comparison would be an unusually low-risk study.

## VIII. CONCLUSION

Curcumin's problem has three parts, and the common error is to treat it as one. Solubility and intestinal stability limit how much dissolves and survives; glucuronidation and sulfation in the enterocyte and liver remove almost all of what is absorbed, so that free curcumin was detectable in only one of twelve volunteers given 10 or 12 g; and efflux returns a further fraction to the lumen. A nanocarrier addresses the first and a bioenhancer addresses the second and third, which is why combining them is a mechanistic argument rather than an additive one.

Nanostructured lipid carriers are the right carrier for this particular pair. The binary solid–liquid matrix provides the crystal imperfections that accommodate a high payload and resist expulsion, the liquid lipid fraction offers a compartment suited to piperine's solubility behaviour, and — with appropriate lipid chain length — the carrier opens the intestinal lymphatic route that bypasses hepatic first-pass metabolism altogether. Published curcumin lipid nanocarriers have produced tens-of-fold improvements in oral exposure, and recent work on the co-loaded combination has shown higher ex vivo intestinal permeation than a suspension of the same two actives.

What would make such a study a contribution rather than another formulation report is the design of its comparisons. Treat the curcumin-to-piperine ratio as a design factor rather than inheriting it from a 1998 co-administration trial. Include apparent permeability as a modelled response rather than a final check. Use a model that retains intestinal metabolism, so that the mechanism being claimed can actually be observed. Report free and conjugated curcumin separately. And include a physical mixture arm, because without it the central claim — that co-encapsulation beats co-administration — remains untested. Each of these is straightforward; together they would answer the question the formulation is built around.

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