

LipoEdge™

LIPOSOMAL TECHNOLOGY

US-FDA REGD. · FSSAI · WHO-GMP · cGMP · ISO · HACCP

Inside This Brochure

	How We prove your Liposome is safe in Living System
	3 Things We Confirm About Every Liposomal Formulation
	From Sample to Validated Dossier — 4 Steps
	Common Problems & Our Solution
	Practical support

How We Prove Your Liposome Is Safe in Living Systems

A complete guide to the in vitro biological testing that proves your liposomal formulation is biocompatible, non-toxic, and genuinely effective at the cellular level.

Safety Confirmed

Cytotoxicity, hemocompatibility and apoptosis profiling confirm your formulation does not harm living cells.

Biocompatibility Verified

ISO 10993-5 compliant cell viability studies across multiple cell lines verify true biocompatibility.

Regulatory-Ready Data

Full technical dossier with assay results, compliance certificates and pathway mapping delivered.

Contact: info@thelipoedge.com · www.thelipoedge.com · +91 33 4025 1539

3 Things We Confirm About Every Liposomal Formulation

And what each result means for your product's safety, efficacy, and regulatory compliance.

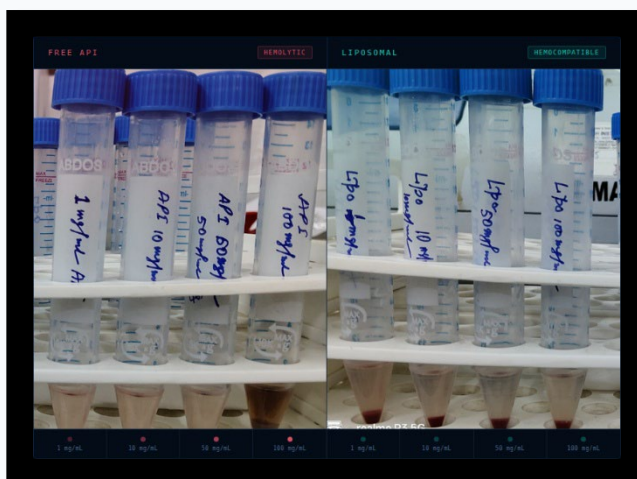
01 · Is your liposome safe in blood? — HEMOCOMPATIBILITY

Hemolysis Rate	≤5%	What this means for your product: ≤5% hemolysis confirms your formulation will not rupture red blood cells intravenously — a mandatory criterion for any injectable liposomal product.
Cell Viability (MTT)	≥80%	What this means for your product: ≥80% cell viability supports good cellular compatibility under the tested conditions
IC ₅₀ (Cytotoxicity)	High value preferred	What this means for your product: A high IC ₅₀ means a much larger dose is needed to kill 50% of cells — confirming a wide safety window between effective and toxic concentrations.
Osmotic Fragility	Within physiological range	What this means for your product: Confirms red blood cell membranes are not destabilised by the formulation — essential proof that the liposomal carrier itself is biocompatible.

Hemocompatibility Assay

Visual Comparison: Free API (Hemolytic) vs. Liposomal API (Hemocompatible)

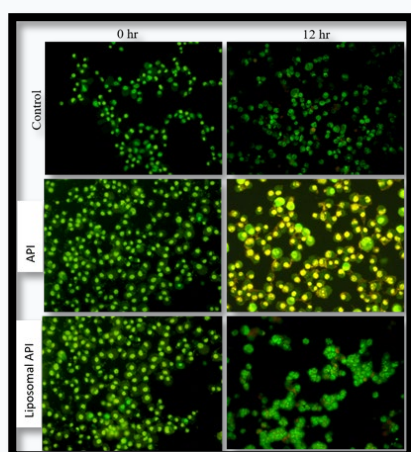
From Image : LEFT: Free API — red pellet at each dose confirms RBC rupture. RIGHT: Liposomal API — minimal pellet across all concentrations (1–100 mg/mL), confirming encapsulation eliminates hemolytic activity.



02 · Does it harm cells? — CYTOTOXICITY & CELL VIABILITY

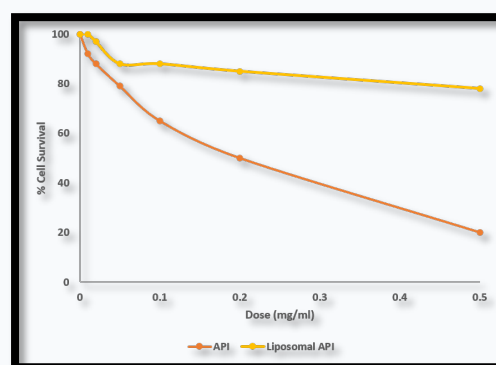
MTT Assay (L929, CHO, HepG2)	ISO 10993-5	What this means for your product: Three internationally validated cell lines screen for cytotoxicity. Multiple lines eliminate false negatives, giving robust, regulator-accepted safety data.
Colony Formation Assay	Proliferation rate	What this means for your product: Tests whether sub-lethal doses inhibit cell replication — identifies toxicity missed by short-term viability assays.
JC-1 Mitochondrial Function	$\Delta\Psi_m$ ratio	What this means for your product: A drop in mitochondrial membrane potential is the earliest marker of cell death — catches toxicity before conventional assays detect it.

Apoptosis Detection — AO/EtBr Staining on L929 Cells



□ Green: Healthy □ Yellow: Pre-apoptotic □ Orange: Apoptotic
 At 12 hr, free API (row 2) shows marked yellow/orange shift confirming apoptosis. Liposomal API (row 3) shows controlled delivery with far less apoptotic activity.

MTT Assay — % Cell Survival: API vs. Liposomal API



Liposomal API (yellow) maintains $\geq 78\%$ cell survival at 0.5 mg/ml. Free API (orange) drops to $\sim 20\%$ — confirming the carrier dramatically reduces cytotoxicity while preserving biological activity.

03 · Does it work inside cells? — CELLULAR UPTAKE & EFFICACY

Cellular Uptake — Caco-2	Fluorescence intensity	What this means for your product: Measures how efficiently your liposome enters intestinal cells. High uptake proves the formulation actively delivers the active inside — not just to the cell surface.
Endocytic Pathway ID	Inhibitor profiling	What this means for your product: Chemical inhibitors reveal exactly which pathway internalises your liposome (clathrin, caveolae, macropinocytosis). Required mechanistic data for regulatory dossiers.
Antioxidant Activity (DCFH-DA)	ROS reduction %	What this means for your product: Fluorometric ROS measurement confirms antioxidant efficacy is retained after encapsulation — critical for nutraceutical and neuroprotective formulations.

From Sample to Validated Dossier — 4 Steps

Every LipoEdge™ biological assessment goes through this validated pipeline before you receive your certificate.

01

Study Design & Cell Line Selection

- ✓ Customised protocol developed for your specific application
- ✓ Cell lines selected: L929, CHO, HepG2, Caco-2 (as appropriate)
- ✓ Control groups defined — empty liposome, free API, vehicle
- ✓ Concentrations and timepoints established

→ Your formulation is assessed with the right cells and controls before any test begins.

02

Cellular Safety Screening

- ✓ Hemolysis & hemocompatibility confirmed ($\leq 5\%$ threshold)
- ✓ MTT cytotoxicity run across all selected cell lines
- ✓ IC_{50} values calculated for API vs. liposomal API
- ✓ Osmotic fragility and RBC integrity measured

→ We confirm your formulation is safe before testing for efficacy.

03

Intracellular Delivery & Pathway Mapping

- ✓ Cellular uptake tracked by fluorescence microscopy
- ✓ Endocytic pathway identified using chemical inhibitors
- ✓ Apoptosis profiled by AO/EtBr staining (0 hr vs 12 hr)
- ✓ Mitochondrial function checked by JC-1 assay

→ We trace exactly how your liposome enters the cell and what it does inside.

04

Technical Dossier & Regulatory Package

- ✓ ISO 10993-5 compliance certificate issued
- ✓ Full assay data compiled with statistical analysis
- ✓ Mechanistic pathway summary included
- ✓ Regulatory-ready documentation package delivered

→ You receive a complete, regulator-ready document package.

Common Problems & Our Solution

Four biological failures we diagnose and fix.

PROBLEM: Free API is hemolytic — red blood cells are rupturing

OUR FIX: Hemocompatibility assay confirms dose-dependent RBC rupture with free API. Liposomal encapsulation eliminates direct membrane contact, reducing hemolysis to $\leq 5\%$.

PROBLEM: Your liposome isn't entering target cells efficiently

OUR FIX: Caco-2 uptake studies and endocytic pathway mapping identify the barrier. We adjust surface charge, PEGylation or particle size until intracellular delivery is confirmed.

PROBLEM: You cannot separate carrier toxicity from API toxicity

OUR FIX: Side-by-side MTT comparison of empty liposomes vs. API-loaded liposomes isolates each component's cytotoxic contribution. Carrier lipid composition is reformulated if needed.

PROBLEM: Biological results are inconsistent between batches

OUR FIX: We combine in vitro bio data with nanoparticle characterisation (size, PDI, EE%) to pinpoint the batch variable driving the biological inconsistency.

Practical Support

YOUR BIOLOGICAL QUESTION	OUR SUPPORT
Is your liposome safe for intravenous use?	Hemocompatibility assay · Osmotic fragility · Hemolysis rate ≤5%
Is your formulation toxic to cells?	MTT cytotoxicity (L929, CHO, HepG2) · IC ₅₀ profiling · ISO 10993-5
Does the liposome actually enter cells?	Caco-2 uptake study · Fluorescence microscopy · Endocytic pathway ID
Does your active still work after encapsulation?	Antioxidant activity (DCFH-DA) · MIC assay · ELISA mechanistic mapping
Does your formulation induce apoptosis in target cells?	AO/EtBr staining · JC-1 mitochondrial membrane potential
Need regulatory-ready biological data?	Full technical dossier · ISO certificate · Pathway summary report

Your Next Step

☐ Test an Existing Formulation Send us your sample. We run the full biological safety and efficacy suite and deliver your certificate.	 Develop a New Product We design the study protocol and validate your liposomal formulation end-to-end for biological safety.	 Talk to Our Team Discuss your biological testing requirements with our scientists and get a customised study plan.
--	---	---

Let's Prove Your Formulation Performs

Talk to our team about your liposomal product. We'll handle the science. You'll get the proof.

www.thelipoedge.com

info@thelipoedge.com · +91 33 4025 1539

Bangur Avenue, Kolkata 700089 · Salt Lake City, Kolkata 700091, India

© 2026 LipoEdge™ · West Bengal Chemical Industries Limited (WBCIL)