M1430-03-20

Enhancing Oral Bioavailability in Preclinical Dog PK Studies Using Ball Milling Technology

Yifan Yang, Jianping Sun, Chao Zhang, Yu Chen, Jacob Zhi Chen, Shoutao Liu, Yi Tao, Liang Shen

DMPK Department, WuXi AppTec, 31 Yiwei Rd., Waigaoqiao Free Trade Zone, Shanghai, China

CONTACT INFORMATION: liu_shoutao@wuxiapptec.com

PURPOSE

Improving oral bioavailability of poorly water-soluble drugs remains a major challenge in preclinical PK studies because limited dissolution often restricts systemic exposure. Reducing particle size increases surface area and dissolution rate, which frequently translates into higher absorption and improved PK profiles[1]. High-energy wet ball milling is a robust lab technique to generate micro- and nanosized drug particles; by applying mechanical shear and impact, it produces fine suspensions or powders with faster dissolution and demonstrated bioavailability gains in animal studies[2]. Milling outcomes are controlled by variables such as solvent viscosity, bead size and material, milling speed and energy input, duration, and temperature management[3,4]. These factors jointly influence the final particle size distribution, physical stability, and potential solid-state changes, all of which determine in vivo performance. In this work, we applied wet ball milling to prepare an aprepitant nanosuspension and systematically optimized key process parameters to develop a reproducible formulation aimed at enhancing aprepitant oral bioavailability in dog PK studies.

METHOD(S)

1.Materials

- Compound: Aprepitant (Sigma-Aldrich, BCS Class II, solubility <5 μg/mL).
- Vehicle: 0.5% HPC (w/v) + 0.2% SLS (w/v) in purified water.
- Equipment: Ball milling (Retsch PM100)

  Mastersizer 3000, PLM, HPLC-UV.

Formulation

- Nanoparticle suspension
- Stock suspension: Aprepitant 50 mg/mL .
- Milling: Beads: 1.0 mm zirconia; speed 350 rpm; cycle 10 min on/5 min off for a total of 18 h; maintain <40°C—ball-mill the concentrate until the target particle size is achieved.
- Dilution: dilute milled concentrate to 0.4 mg/mL with vehicle.

2. Dog PK study

- Animals: Male Beagle dogs (N=10 total; n=5/group)

  Group 1: Raw drug suspension

  Group 2: Nanoparticle suspension
- Dosing: oral, single dose, 2 mg/kg
- Sampling times: predose, 0.25, 0.5, 1, 2, 4, 6, 8, 12, 24, 48h and 72h
- Bioanalysis: LC-MS/MS, PK parameters (Cmax, Tmax, AUC)

RESULT(S)

1. Particle size results

- Raw drug powder: particle size 10-100 μm (Figure 1)
- Raw drug suspension: particle size < 5 μm (Figure 2)
- Nanoparticle suspension: Dv90 = 0.258 μm (258 nm) (Figure 3)

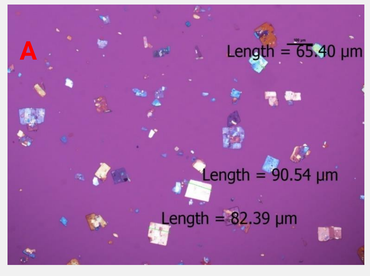
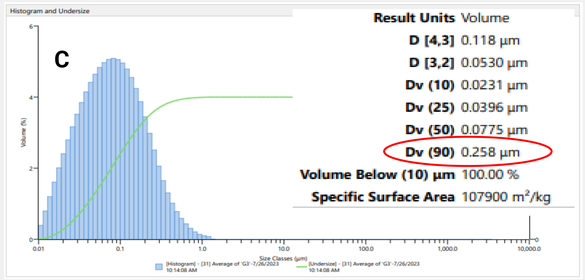


Figure 1. Particle morphology of aprepitant. (A) Raw drug powder (10× magnification, PLM)

Figure 2. Particle morphology of aprepitant. (B) Raw drug suspension (20× magnification, PLM).

Figure 3. (C) Particle size distribution of drug nanoparticle suspension analyzed by Mastersizer 3000

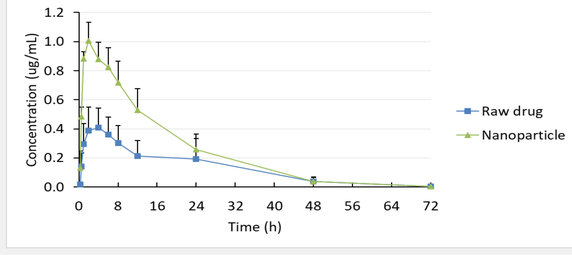


2. PK results of the nanoparticle suspension group in Beagle dogs showed a 2.31-fold increase in Cmax and a 1.85-fold increase in AUC compared to the raw drug suspension group.

Table 1.Comparison of PK parameters of Aprepitant following oral administration of nanoparticle versus raw drug in beagle dogs

Suspensions	Dose (mg/kg)	AUC 0-72 (ug/ml*h)	C_{max} (ug/ml)	T_{max} (h)
Raw drug	2	8.76±4.59	0.441±0.150	2.80±1.10
Nanoparticle	2	16.6±4.60	1.02±0.109	1.75±0.500

Figure 4. Mean (± SD) plasma concentration–time profiles of Aprepitant following oral dosing of raw drug and nanoparticle in beagle dogs.



CONCLUSION(S)

- Ball milling effectively reduces poorly soluble drugs to the nanoparticle range, improving dissolution rate and systemic exposure; In the case of heat-sensitive compounds use intermittent milling and controlled speed to avoid thermal degradation.
- Preventing post-milling aggregation requires the selection of an appropriate stabilizer (SLS) in combination with a dispersant (HPC).
- We screened multiple stabilizers and dispersants and optimized key milling parameters (bead size, bead: liquid ratio, speed, time, temperature). The combined “equipment + stabilizer + dispersant” optimization is critical to produce high-performance nanosuspensions and improve oral bioavailability.

ABBREVIATION

PK: Pharmacokinetic; HPC: Hydroxypropyl Cellulose;
SLS: Sodium Lauryl Sulfate; PLM: Polarized Light Microscopy; HPLC: High Performance Liquid Chromatography; AUC: Area Under the Curve;

REFERENCES

[1] Bhalani DV, Nutan B, Kumar A. Bioavailability Enhancement Techniques for Poorly Aqueous Soluble Drugs and Therapeutics. Biomedicines. 2022 Aug 23;10(9)
[2] Alshora D, Ibrahim M, Elzayat E, Almeanazel OT, Alanazi F. Defining the process parameters affecting the fabrication of rosuvastatin calcium nanoparticles by planetary ball mill. Int J Nanomedicine. 2019;14:4625-4636.
[3] Sumei Zeng, Wenjian Cai, Shaoqun Zeng , et al. Research progress on the preparation of nanosuspensions for injection by wet media grinding technology and its quality evaluation[J].China Journal of Pharmaceutical Industry,2022,53(09):1251-1261.
[4] Burmeister CF, Kwade A. Process engineering with planetary ball mills. Chem Soc Rev. 2013 Sep 21;42(18):7660-7.