# Abstract

Liver toxicity remains a critical barrier in drug development and clinical therapeutics, with mitochondrial dysfunction (MD) recognized as a central mechanism driving drug-induced liver injury (DILI). Mitochondrial liabilities contribute substantially to compound attrition, post-marketing drug withdrawals, and regulatory restrictions. Chloramphenicol, despite its broad-spectrum antimicrobial efficacy, has limited clinical use owing to its mitochondrial toxicity and associated hepatotoxic effects. In light of these challenges, this research investigates whether strategic combination therapy specifically with potent antioxidants can mitigate such adverse effects, potentially rescuing otherwise valuable drugs from late-stage failure or regulatory rejection. The study evaluates the hepatoprotective potential of two well-characterized antioxidants, Astaxanthin and Quercetin, against chloramphenicol-induced mitochondrial toxicity using integrated *in-vitro* and *in-vivo* models to elucidate mechanistic pathways and therapeutic efficacy.

In the *in-vitro* component, HepG2 liver cells were cultured under galactose-adapted conditions to simulate enhanced mitochondrial reliance. Cells were exposed to chloramphenicol with or without co-treatment of Astaxanthin or Quercetin. Assays for ATP production, reactive oxygen species (ROS), and expression of key mitochondrial genes (SOD2, NRF1, SURF1, TFAM, and UCP2) were performed. Results demonstrated significant ROS attenuation and mitochondrial gene expression recovery with antioxidant treatment, indicating mitigation of chloramphenicol-induced toxicity.

In the *in-vivo* arm, male Wistar rats were administered chloramphenicol intraperitoneally, followed by oral antioxidant therapy. Biochemical markers including glutathione (GSH) and nitric oxide (NO) were quantified to assess oxidative stress. Both antioxidants significantly restored GSH levels and reduced NO, with Quercetin showing slightly superior efficacy.

This integrated study demonstrates that both Astaxanthin and Quercetin confer mitochondrial protection through modulation of oxidative stress and gene expression, suggesting their therapeutic potential as adjuncts in antibiotic-induced hepatotoxicity. Future investigations should focus on mechanistic insights, dose optimization, and clinical translation.