Soy-Based Nanoemulsion of Griseofulvin: Formulation, Antifungal Efficacy and Toxicity Studies in Rats

U. E. Osonwa¹, E. C. Okpana², C. D. Nwakile², K. I. Chukwu³, C. O. Esimone²
¹University of Houston, ²Nnamdi Azikiwe University, ³University of Saskatchewan

Purpose
Griseofulvin, a fungistatic agent with poor aqueous solubility and oral bioavailability, which is less than 25% and this limits its efficacy. The purpose of this work was to formulate griseofulvin into nanosized formulation to overcome the poor solubility and improve the efficacy.

Methods
Griseofulvin powder was formulated as a nanoemulsion using a simple homogenization technique, with soya bean oil, soya lecithin and distilled water as components. The nanoemulsion was characterized by particles size and size distribution, viscosity, and electrical conductivity after formulation and after storage for 90 days at shelf conditions. The antifungal activity against typed cultures of Candida albicans, Aspergillus niger, and Trichophyton rubrum was established in vitro. In vivo antifungal activity was determined with A. niger by establishing systemic mycosis on 20 eight-month old male white albino rats of the Wister strain weighing 200-250 g. The rats were divided into 4 groups (n=5). The nanoemulsion formulation and a commercial brand (a suspension) of the drug were administered orally at 0.77 mcg/g and 10 mcg/g, respectively. The percentage parasitemia was determined by microbial count. The effect of the nanoemulsion on the hematopoietic system was determined by administering the two products at the same dose of 10 mcg/g ip daily for 90 days to six weeks old albino mice of both sexes(n=5). Blood was sampled once a day for the first 7 days and then weekly for a total of 90 days and analyzed for hematological parameters. The results were compared to values gotten with the commercial suspension at the same dose. The effect of the nanoemulsion and commercial suspension on the liver was evaluated by determining liver enzymes levels and histopathological changes using standard procedures.

Results
The optimized formulation had a soy oil –lecithin ratio of 7:3 with a drug concentration of the nanoemulsion was 0.48 μg/ml. The nanoemulsion 24 h after formulation had the following parameters- average drop size of 150.30 ± 2.23 nm, pH of 6.30 ± 0.00, conductivity of 84.00 ± 0.58 ms/cm and viscosity of 0.89 ± 0.05. The values after 90 days of shelf storage at room temperature were 162.20±3.14 nm (pdi 0.203), pH of 6.29±0.01, conductivity of 89.00±0.00 (ms/cm) and viscosity of 0.89±0.01 cps. The in vitro antifungal activity showed that the nanoemulsion was more active against all three organisms at a much lower dose (0.48 μg/ml) than the commercial preparation (5 mg/ml). In vivo antifungal activity showed that the nanoemulsion produced a significantly reduced blood bacterial counts compared to the commercial suspension. The dose of the nanoemulsion used was less than 1/10 th that of the commercial preparation, parasite clearance was still higher. The nanoemulsion showed higher hematopoietic parameters of the mice compared to the commercial suspension. The liver histological results showed less prominent lesions with the nanoemulsion than with the commercial preparation.

Conclusion
Griseofulvin was successfully formulated as nanoemulsion for oral delivery. The efficacy was markedly improved while systemic toxicity was reduced.