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# Histologic comparison of the fallopian pathologic effects of erythropoietin and U-74389G

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## Abstract

**Background:** This study compared the effects of erythropoietin (Epo) and the antioxidant lazaroid U-74389G (L) on fallopian pathology in a rat model of fallopian ischemia-reperfusion (FIR). The comparison was based on two preliminary studies evaluating the effects of each drug in an FIR animal model.

**Methods:** Fallopian pathology was assessed at two endpoints: 60 minutes (groups A, C, E) and 120 minutes (groups B, D, F) of reperfusion. Groups A and B received no drugs, groups C and D received Epo, and groups E and F received U-74389G.

**Results:** The Epo study showed a non-significant reduction in total fallopian pathology within the "lesion-free" grade by -0.01 (95% CI: -0.09 to 0.06). The U-74389G study showed a non-significant increase in pathology by 0.01 (95% CI: -0.05 to 0.06). Co-evaluation indicated that Epo slightly ameliorated fallopian pathology, while U-74389G slightly worsened it, though the difference was non-significant.

**Conclusion:** Epo demonstrates a slight, non-significant superiority over U-74389G in restoring fallopian pathology, potentially applicable in clinical settings.

**Keywords:** Erythropoietin, Fallopian tubes, Ischemia, Reperfusion, U-74389G

## Introduction

U-74389G, a lazaroid-class antioxidant, is not widely recognized for its effects on fallopian pathology (FP) (1). Of 265 published studies on U-74389G, approximately 19.6% focus on ischemia-reperfusion (IR) models, but its tissue-specific effects remain unclear. Chemically, U-74389G (21-[4-(2,6-di-1-pyrrolidinyl-4-pyrimidinyl)-1-piperazinyl]-pregna-1,4,9(11)-triene-3,20-dione maleate salt) inhibits lipid peroxidation, both iron-dependent and arachidonic acid-induced. It has shown protective effects in kidney, liver, brain microvascular endothelial cells, and heart IR models, reducing leukocyte activity, downregulating proinflammatory genes, counteracting endotoxin shock, modulating cytokine production, enhancing mononuclear cell immunity, protecting endothelium, and exhibiting antishock properties (2).

Studies report 4.9% to 62.4% of pregnant women use multiple medications, with 4.9% to 33.7% doing so in the first trimester, reflecting diverse medication use patterns (5).

Erythropoietin (Epo), a cytokine, is less studied in FP but serves as a reference drug for comparison. Of over 34,000 studies on Epo, only 3.9% address IR models, warranting further investigation of its FP effects (3, 4).

This study compares the histopathological effects of U-74389G and Epo on fallopian pathology in a rat FIR model.

## Materials & Methods

Animal Preparation:

This study was conducted under veterinary licenses (3693/12-11-2010, 14/10-1-2012), with details on licensing, experimental location, and ethical approvals provided in preliminary studies (1, 5). Female Albino Wistar rats (16–18 weeks) were housed under standard conditions with ad libitum food and water for 7 days pre-experimentation. Anesthetic

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protocols, acid-base monitoring, electrocardiography, oxygen supplementation, and euthanasia procedures were previously described.

Rats were randomly assigned to six groups (n=10/group), all subjected to 45 minutes of ischemia via laparotomy and infrarenal aortic clamping. Groups were:

• Group A: 60 minutes reperfusion, no drug.
• Group B: 120 minutes reperfusion, no drug.
• Group C: Epo (10 mg/kg IV) at reperfusion onset, 60 minutes reperfusion.
• Group D: Epo (10 mg/kg IV) at reperfusion onset, 120 minutes reperfusion.
• Group E: U-74389G (10 mg/kg IV) at reperfusion onset, 60 minutes reperfusion.
• Group F: U-74389G (10 mg/kg IV) at reperfusion onset, 120 minutes reperfusion.

Ischemia was induced by clamping the infrarenal aorta for 45 minutes, followed by clamp removal to initiate reperfusion. Drugs were administered via inferior vena cava catheterization at reperfusion onset. Fallopian tubes were harvested at 60 minutes (groups A, C, E) or 120 minutes (groups B, D, F) for histopathological evaluation.

Fallopian pathology was assessed using four variables:

• Endosalpingeal edema (EE)
• Fallopian congestion (FC)
• Endosalpingeal karyorrhexis (EK)
• Salpingitis (S)

Each variable was scored by lesion severity:

• 0.0–0.499: No lesions
• 0.5–1.499: Mild lesions
• 1.5–2.499: Moderate lesions
• 2.5–3.0: Severe lesions

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

The Wilcoxon signed-rank test assessed histologic score differences between groups based on reperfusion time and drug administration. Paired t-tests analyzed difference values (DG) for each histologic endpoint. Statistical analyses used STATA 6.0.

## Results

Paired t-test analysis showed Epo was not significantly superior to U-74389G in promoting FP restoration within the “lesion-free” grade (mean difference: 0.03, 95% CI: -0.09 to 0.15).

Epo Treatment:

The comparison of histologic scores between groups following erythropoietin treatment revealed no statistically significant differences. Pairwise analyses using the Wilcoxon Signed-Rank Test showed only minimal, non-significant changes in scores (Table 1).

U-74389G Treatment:

**Table 1** Differences in histologic scores after erythropoietin treatment (Wilcoxon Signed-Rank Test)

| DG Comparison | Difference | p-value |
|---|---|---|
| A vs. B | +0.03 | 0.872 |
| A vs. C | +0.08 | 0.360 |
| A vs. D | +0.05 | 0.317 |
| B vs. C | +0.05 | 0.563 |
| B vs. D | +0.03 | 0.655 |

Analysis of post-treatment histologic variable mean scores using the Wilcoxon Signed-Rank Test revealed no significant differences among most group comparisons. However, a statistically significant difference was observed between groups E and F (+0.12, p=0.047). All other pairwise comparisons, including A-B, A-E, A-F, B-E, and B-F, did not reach statistical significance (Table 2).

**Table 2** The values difference for groups (DG) after Wilcoxon signed-rank test for all postL histologic variables mean scores. +0.004 + 0.080

| DG | Difference | p-value |
|---|---|---|
| A-B | –0.02 | 0.879 |
| A-E | –0.09 | 0.114 |
| A-F | +0.04 | 0.799 |
| B-E | –0.07 | 0.059 |
| B-F | +0.05 | 0.575 |
| E-F | +0.12 | 0.047* |

*Significant at p<0.05

Comparative Analysis:

The comparison of mean histologic scores across postL, postEpo, and postL–postEpo conditions revealed variable changes among the groups (Table 3). PostL differences ranged from –0.09 (A vs. E) to +0.12 (E vs. F), with a mean difference of +0.005. PostEpo differences showed a narrower range (–0.025 to +0.075), with a mean of +0.033. The postL–postEpo difference, reflecting the net change after erythropoietin treatment, varied from –0.16 (A vs. E)



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to +0.15 (E vs. F), with an overall mean of –0.029 ± 0.11. These findings suggest group-specific variations in histologic response following treatment.

Meta-analysis

The meta-analysis of U-74389G/erythropoietin efficacy across 34 hematologic variables showed variable treatment effects over time (Table 4). The mean efficacy ratio at 1 hour was 5.04 (p=0.060), suggesting a strong but not statistically significant effect. At 1.5 hours, the ratio was 1.86 and reached statistical significance (p<0.001). The 2-hour ratio was 2.02 with borderline significance (p=0.053). The overall reperfusion time ratio was 1.18 (p=0.001), indicating a consistent positive treatment effect. The interaction p-value was 2.20, suggesting variability in treatment response over time.

**Table 3:** Differences in mean histologic scores between groups for PostL, PostEpo, and PostL–PostEpo conditions (Mean ± SD: –0.03 ± 0.11)

| Group Comparison (DG) | PostL Difference | PostEpo Difference | PostL–PostEpo Difference |
|---|---|---|---|
| A vs. B | –0.02 | +0.025 | -0.04 |
| A vs. E | –0.09 | +0.075 | -0.16 |
| A vs. F | +0.03 | +0.05 | -0.02 |
| B vs. E | –0.07 | +0.05 | -0.12 |
| B vs. F | +0.05 | +0.025 | +0.02 |
| E vs.F | +0.12 | -0.025 | +0.15 |
| **Mean** | +0.005 | +0.0333 | -0.029 |

**Table 4:** Meta-Analysis of U-74389G/Erythropoietin Efficacy Ratios on 34 Hematologic Variables

| Endpoint Variable | 1h Ratio | p-value | 1.5h Ratio | p-value | 2h Ratio | p-value | Overall Reperfusion Time Ratio | p-value | Interaction p-value |
|---|---|---|---|---|---|---|---|---|---|
| Mean | 5.04 | 0.060 | 1.86 | <0.001 | 2.02 | 0.053 | 1.18 | 0.001 | 2.20 |

## Discussion

The preliminary study on U-74389G's effect on fallopian tubes showed a slight, non-significant worsening of pathology (5). U-74389G accumulates in cell membranes, protecting vascular endothelium from peroxidative damage but poorly penetrates the blood-brain barrier. It exhibits neuroprotection, membrane stabilization, and benefits in ototoxicity, Duchenne muscular dystrophy, and septic states. It increases γ-glutamyl transferase, superoxide dismutase, and glutathione levels, acts as an immunosuppressant in flap survival, prevents learning impairments, delays synaptic transmission decay during hypoxia, and shows antiproliferative properties in brain cancer cells, positioning it as a promising anti-inflammatory drug for IR injuries. Epo showed a slight, non-significant improvement in FP in non-iron-deficient rats (5).

Studies on transgenic chickens producing human Epo (hEPO) in egg whites via lentiviral vectors demonstrated oviduct-specific expression (6, 7). EphA2, a tyrosine kinase receptor, is implicated in fallopian tube epithelial cell adhesion, potentially affecting tubal pregnancy (8). EphB3 and EphA1 expression in fallopian tubes and ovarian tissues suggests roles in ovarian serous carcinoma grading (9, 10). Oviductal organoids under heat stress and mycotoxin exposure highlight redox and inflammatory pathways relevant to FP (11, 12). Glutathione's role in preimplantation embryos supports its potential in FP restoration (13). High-grade serous ovarian carcinoma (HGSOC) pathways, including those involving glutathione, further contextualize FP (14). Antimony exposure studies indicate oxidative stress impacts on oocyte quality and FP (15).

Table 3 confirms Epo's non-significant superiority over U-74389G in FP restoration. However, meta-analysis of 34 serologic variables suggests U-74389G's biochemical activity is approximately twice that of Epo ($p < 0.001$), though this attenuates short-term (16).

## Conclusion

Despite U-74389G's biochemical superiority (2.2-fold activity vs. Epo, $p < 0.001$), FP findings show Epo's slight, non-significant advantage in restoration. Further



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studies may align biochemical and histopathological outcomes, offering potential for clinical applications in genetic engineering, tubal implantation, fertilization, preimplantation, and HGSOC management.

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## Conflicts of Interest

No declared conflicts of interest.

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