Comparison of Endotoxin-Induced Uveitis Model and Experimental Autoimmune Uveitis Model in Lewis Rats for Drug Screening

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ABSTRACT

Purpose: The study is to compare Endotoxin-Induced Uveitis (EUU) Model and Experimental Autoimmune Uveitis (EAU) model in Lewis Rats in order to benefit how to choose appropriate models for anti-inflammatory drug screening.

Methods: The EUU model was induced by endotoxin named lipopolysaccharides (LPS) through footpad injection on Day 9 in 49 Lewis rats included five groups: negative control of saline (Saline), positive control of demeclocycline + low dose lipopolysaccharides (Dex + LPS); positive control of demeclocycline + high dose lipopolysaccharides (Dex + H LPS); low dose lipopolysaccharides (LPS); high dose lipopolysaccharides (HLPS) and high dose lipopolysaccharides (HLP). The EAU model induced by a subcutaneous injection of peptide R16 of bovine Interphotoreceptor Retinoid Binding Protein (IRBP)-Microbacterium tuberculosis H37Ra on Day 1 to 30 Lewis rats included three groups: negative control of saline (Saline); positive control of demeclocycline + RBP (Dex + RBP) and RBP. Animals were sacrificed on Day 2 or 3 (EUU), and on Day 26 or 28 (EAU) for histopathological or cytokine analysis. During studies, animals were observed for clinical ophthalmic daily, ophthalmic examination and body weight daily (EUU) or weekly (EAU).

Results: In EUU, ophthalmic examination showed that both aqueous flare and vitreous body flare was in Dex + LPS, Dex + H LPS, HLPS and HLPS groups scored significantly higher than the Saline group in a time-dependent manner (p < 0.05). Both aqueous flare and vitreous body flare scores in LLPS and HLPS groups were significantly more severe than scores in Dex + LPS, Dex + HLPS, HLPS groups (p < 0.05). The results demonstrated that LPS induced inflammation in eyes which presented a peak exposure at 48 hours following administration, and Dex, inflicted to a certain extent inflammation. Pathological reports showed that the inflammatory cells in eyes increased in Dex + LPS, Dex + HLPS, LLPS and HLPS groups were significantly increased when compared to the Saline group in a time-dependent manner (p < 0.05). Cytokine analysis results showed that concentration levels of IL-1α, IL-6, MCP-1 and TNF-α in retina increased in various extents in Dex + LPS, Dex + HLPS, LLPS and HLPS groups. In EAU, ophthalmic examination showed signs of inflammation on Day 15, and the peak of inflammation was at Day 21 in RBP group. The clinical grading of EAU in RBP group was significantly different when compared to that in the Saline or Dex + RBP groups (p < 0.05). Pathological reports showed that mainly inflammatory and degenerative changes were detected in the retina of RBP group. Cytokine analysis reports that concentration levels of IL-17, TNF-α, MCP-1 and IL-6 were not significantly different between groups (p > 0.05).

Conclusion: The EUU and EAU models were successfully induced and the peak of inflammation was at 48 hours (EUU, Day 21) and 4 weeks (EAU, Day 28) following administrations. Ophthalmic examination effectively inhabited the inflammation in both models. Both of EUU and EAU models may provide a stable, effective and reliable method for anti-inflammatory drug screening.

METHODS & MATERIALS


EAU: 30 female Lewis rats, 250 – 350 grams. Groups: (1) Saline: balanced salt solution, (2) Dex + RBP: demeclocycline + RBP, (3) RBP: Administration: Day 1, injection of 25 µg/kg of peptide R16 of bovine Interphotoreceptor Retinoid Binding Protein, rats sacrificed on Day 1177 – 1191, sequence AD522669:EUVGPFOVFLKN or saline (BSS) subcutaneously at the base of the tail. Treatment: Day 1 – 10, 15 µg/kg of peptide R16 (Dox: 0.3%) topical ophthalmic, and 0.1% demeclocycline ophthalmic suspension.


RESULTS

1. Ophthalmic Examination in the EUU Model

2. Histopathological Images of Retina in the EAU Model

3. Comparison of EUU and EAU Models

4. Cytokines in Retina-

5. Pathological Grading in the EAU Model (Day 21)

6. Histopathological Images of Iris-Ciliary Body in the EUU Model

7. Comparison of EUU and EAU Models

8. Cytokines in Retina-

9. Histopathological Grading in the EAU Model (Day 21)

10. Histopathological Images of Iris-Ciliary Body in the EUU Model

11. Comparison of EUU and EAU Models

12. Cytokines in Retina-

13. Pathological Grading in the EAU Model (Day 21)

14. Histopathological Images of Iris-Ciliary Body in the EUU Model

15. Comparison of EUU and EAU Models

16. Cytokines in Retina-

17. Histopathological Grading in the EAU Model (Day 21)

18. Histopathological Images of Iris-Ciliary Body in the EUU Model

19. Comparison of EUU and EAU Models

20. Cytokines in Retina-

21. Pathological Grading in the EAU Model (Day 21)

22. Histopathological Images of Iris-Ciliary Body in the EUU Model

23. Comparison of EUU and EAU Models

24. Cytokines in Retina-

25. Histopathological Grading in the EAU Model (Day 21)

26. Histopathological Images of Iris-Ciliary Body in the EUU Model

27. Comparison of EUU and EAU Models

28. Cytokines in Retina-

29. Pathological Grading in the EAU Model (Day 21)

30. Histopathological Images of Iris-Ciliary Body in the EUU Model

31. Comparison of EUU and EAU Models

32. Cytokines in Retina-

33. Histopathological Grading in the EAU Model (Day 21)

34. Histopathological Images of Iris-Ciliary Body in the EUU Model

SUMMARY

1. The EUU and EAU models were successfully induced and the peak of inflammation was at 48 hours (EUU) or Day 21 (EAU) following administrations.

2. Demeclocycline significantly reduced the inflammation in both models.

3. Both of EUU and EAU models may provide a stable, effective and reliable method for anti-inflammatory drug screening.