様　式　Ｃ－１９、Ｆ－１９－１、Ｚ－１９　（共通）

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研究課題名（和文）miRNA-9 delivery using lactosome, a newly-developed drug delivery system, in the treatment of rheumatoid arthritis

研究課題名（英文）miRNA-9 delivery using lactosome, a newly-developed drug delivery system, in the treatment of rheumatoid arthritis

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交付決定額（研究期間全体）：（直接経費）　　3,200,000円

研究成果の概要（和文）：ラクトソーム-ミR-9をコラーゲン誘発関節炎マウスモデルに静脈注射し、ラクトソーム-コントロールsiRNAを対照群として使用した。平均関節炎スコアは、対照群に比べてmiR-9群で比較的低かった。ラクトソーム-miR-9群の関節の組織学的評価では、炎症の減少が示された。STIAマウスを使用して関節炎を誘発し、異なる関節炎モデルにおけるラクトソーム-miR-9の有効性を確認するために実験を繰り返した。
ラクトソームを使用してRasGRP4を標的とする可能性を調査した。RasGRP4の欠損は、適応免疫応答を抑制することなく、K/BxN STIAおよびAIAの両方を軽減した。

研究成果の学術的意義や社会的意義
我々の研究成果の科学的および社会的意義は、関節炎治療における新たなアプローチを提供する点にあります。ラクトソームを用いたmiR-9の投与は、炎症を効果的に抑制し、既存の治療法に対する耐性を克服する可能性を示した。さらに、RasGRP4の標的化は、免疫応答を維持しつつ関節炎の症状を軽減する新たな治療戦略となり得る。これにより、関節炎患者の生活の質向上が期待され、社会全体の健康増進に寄与する。

研究成果の概要（英文）：Lactosome-miR-9 was injected intravenously into a collagen-induced arthritis mouse model, with lactosome-control siRNA serving as the control group. The mean arthritic score was relatively lower in the miR-9 group compared to the control group. Histological assessments of the joints in the lactosome-miR-9 group showed reduced inflammation. STIA mice was used to induce arthritis and repeated the experiments to confirm the efficacy of lactosome-miR-9 in a different arthritis model. We investigated the potential of targeting RasGRP4 using lactosome: RasGRP4 deficiency attenuated both K/BxN STIA and AIA without suppressing adaptive immune responses.

研究分野：膠原病リウマチ内科

キーワード：Rheumatoid Arthritis　Lactosome　Drug Delivery System　miRNA-9　RasGRP4

様　式　Ｃ－１９、Ｆ－１９－１、Ｚ－１９（共通）

１．研究開始当初の背景

Although multiple biological agents as well as JAK inhibitors have been used to treat rheumatoid arthritis (RA), only half of the patients achieve remission and the side effects such as opportunistic infection cannot be neglected. Rheumatoid arthritis synovial fibroblasts (RASFs) play a major role in promoting the pathogenesis of RA. Studies have been focused on targeting RASFs for decades, but the results are under satisfactory. microRNA-9 (miR-9) expression in serum has been reported low in RA patients and miR-9 suppresses inflammatory NF-kB pathway and bone destructive molecules RANKL in RASFs. The nanocarrier, poly(L-lactide)-co-polypeptide (lactosome) has been widely studied in cancer recently. The biodegradability and low immunogenicity profile make lactosome a great candidate in treating autoimmune diseases, including RA.

２．研究の目的

To target RASFs using lactosome conjugated miR-9 to treat active RA model.

３．研究の方法

- Collagen-Induced Arthritis (CIA) Mouse Model:
    - Animals: DBA/1 mice aged 8-10 weeks were used.
    - Induction of Arthritis: Arthritis was induced by intradermal injection of bovine type II collagen emulsified in complete Freund's adjuvant.
    - Treatment Groups: Mice were randomly divided into two groups:
        - Experimental Group: Received intravenous injections of lactosome-miR-9.
        - Control Group: Received intravenous injections of lactosome-control siRNA.
- Serum Transfer-Induced Arthritis (STIA) Mouse Model:
    - Animals: C57BL/6 mice aged 8-10 weeks were used.
    - Induction of Arthritis: Arthritis was induced by intraperitoneal injection of K/BxN serum.
    - Confirmation of Efficacy: The experiments were repeated using the STIA model to confirm the efficacy of lactosome-miR-9.

      Treatment Administration

- Injection: Lactosome-miR-9 and lactosome-control siRNA were administered intravenously.

Assessment of Arthritis

- Clinical Scoring:
  - The severity of arthritis was evaluated using an established clinical scoring system. Each limb was scored on a scale of 0 to 4, with a maximum score of 16 per animal.
  - Mean Arthritic Score: The mean arthritic score was calculated for both groups and compared.
- Histological Analysis:
  - At the end of the treatment period, mice were sacrificed, and their joints were harvested for histological examination.
  - Inflammation Assessment: Joint sections were stained with hematoxylin and eosin (H&E) and assessed for inflammation, cartilage destruction, and bone erosion.

Investigation of RasGRP4 Targeting

- RasGRP4 Knockout Mice:
  - Animals: RasGRP4-deficient mice were used to evaluate the role of RasGRP4 in arthritis.
  - Induction of Arthritis: Both K/BxN STIA and Antigen-Induced Arthritis (AIA) models were used.
  - Assessment: The severity of arthritis was assessed as described above. Additionally, the impact on adaptive immune responses was evaluated by measuring T-cell and B-cell activity.

4．研究成果

- Arthritic Score: The mean arthritic score was significantly lower in the lactosome-miR-9 group compared to the control group in the CIA model.
- Histological Findings: The lactosome-miR-9 group showed reduced inflammation, cartilage destruction, and bone erosion in histological assessments.
- Efficacy in Different Models: Repeated experiments in the STIA model to confirm the efficacy of lactosome-miR-9.
- RasGRP4 Targeting: RasGRP4 deficiency attenuated arthritis in both K/BxN STIA and AIA models without suppressing adaptive immune responses.

５．主な発表論文等

〔雑誌論文〕　計0件

〔学会発表〕　計1件（うち招待講演　0件／うち国際学会　0件）

１．発表者名 Rihan Da
２．発表標題 Essential roles of RasGRP4 in synovial resident cells in serum transfer - induced arthritis
３．学会等名 Japan College of Rheumatology 2024
４．発表年 2024年

〔図書〕　計0件

〔産業財産権〕

〔その他〕

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６．研究組織

	氏名 （ローマ字氏名） （研究者番号）	所属研究機関・部局・職 （機関番号）	備考
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７．科研費を使用して開催した国際研究集会

〔国際研究集会〕　計0件

８．本研究に関連して実施した国際共同研究の実施状況

共同研究相手国	相手方研究機関