Tau is Required for Glucocorticoid-Induced Chondrocyte Apoptosis and Osteoarthritis Progression

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Disclosure: None

Background Glucocorticoids (GCs) are widely prescribed for autoimmune diseases; however, prolonged exposure can exacerbate osteoarthritis (OA) by inducing chondrocyte apoptosis and cartilage degradation. Our laboratory has long investigated the adverse effects of dexamethasone (Dex) and recently identified Tau as a low-affinity glucocorticoid receptor and a novel therapeutic target in GC-induced osteoporosis (Wenyu Fu et al., *Cell Research*, 2025). Although GCs are recognized as key contributors to OA progression, the precise mechanisms remain poorly understood. Therefore, the objectives of this study were: (1) to determine whether Dex directly accelerates OA development, if so, whether this is mediated through the low-affinity glucocorticoid receptor Tau, and (2) to evaluate whether inhibition of Tau phosphorylation exerts therapeutic effects in GC-enhanced OA.

Methods In vitro chondrocyte cultures and in vivo mouse models (GR ko and Tau ko mice) were employed to investigate the role of GR and Tau in Dex-induced effects (male, n=6). TRx0237, a Tau aggregation inhibitor, was tested for its protective role. Molecular changes (n=3) were assessed by Western blot, RT-qPCR, immunofluorescence, and TUNEL assays, while cartilage degeneration was evaluated by Safranin O staining and OARSI scoring.

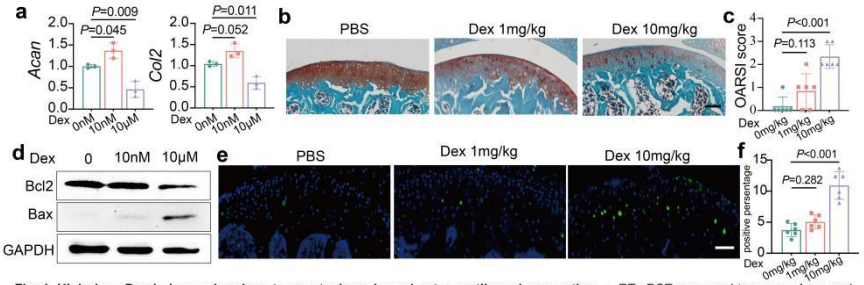

Fig. 1. High-dose Dex induces chondrocyte apoptosis and accelerates cartilage degeneration. a. RT-qPCR was used to assess Acan and Col2 expression under different dosage of Dex treatment (n=3); **b, c.** Safranin O staining and OARSI scoring were performed to evaluate cartilage structure and degeneration (male, n=6), scale bar= 100μm; **d.** Western blotting was conducted to detect Bax and Bcl2 expression (n=3); **e,f.** TUNEL staining **(e)** with quantification **(f)** was used to assess chondrocyte apoptosis **under** different dosage of Dex treatment (n=6), scale bar= 100μm.

Results **Dex treatment suppresses chondrocyte anabolism gene expression, and promotes chondrocyte apoptosis.** High dosage of Dex suppressed the transcription of ECM-related genes *ACAN* and *COL2* **(Fig. 1a)**. In vivo, high dosage of Dex injections resulted in proteoglycan loss and higher OARSI scores **(Fig. 1b,c).** Mechanistically, higher dosage of Dex treatment altered apoptotic signaling by reducing Bcl2 and elevating Bax **(Fig. 1d)**. In vivo, Dex injections increased TUNEL-positive chondrocytes **(Fig. 1e,f)**, confirming that high dosage of Dex treatment promotes cartilage degradation and chondrocyte apoptosis.

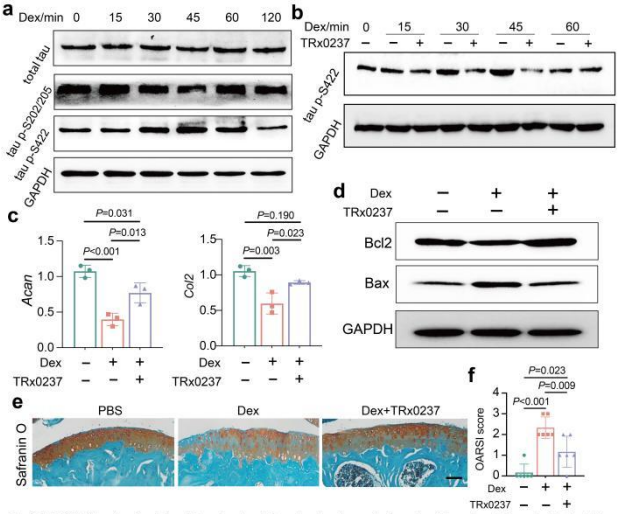

Fig. 2. TRx0237 attenuates Dex-induced tau phosphorylation, chondrocyte apoptosis, and cartilage degradation. a,b. Western blot was performed to examine total tau and site-specific phosphorylation in chondrocytes treated with Dex for the indicated times with or without TRx0237. **c.** RT-qPCR was used to assess the expression of Acan and Col2 in chondrocytes exposed to Dex and/or TRx0237 (n=3). **d.** Western blot was used to detect Bcl2 and Bax protein levels in chondrocytes under the indicated treatments (n=3). **e.** Mice were Intraperitoneal injected with PBS, Dex, or Dex+TRx0237, and articular cartilage sections were subjected to Safranin O staining (male, n=6), scale bar= 100μm. **f.** Cartilage damage was evaluated using OARSI scoring of stained sections (n=6).

TRx0237 alleviates Dex-induced Tau phosphorylation and chondrocyte apoptosis. High dose Dex preferentially phosphorylated Tau at residue Ser422 in osteoclastogenesis progression in GC-induced osteoporosis model. To examine the role of Tau phosphorylation in OA development, we repeated the experiments in chondrocyte and found that with high dosage Dex treatment, Tau at residue Ser422 were phosphorylated **(Fig. 2a).** TRx0237 treatment reduced Dex-induced Tau Ser422 phosphorylation **(Fig. 2b)**, partially restored *Acan* and *Col2* expression **(Fig. 2c)**, and rebalanced the Bcl2/Bax ratio (**Fig. 2d**). In vivo, TRx0237 preserved proteoglycan content, decreased OARSI scores **(Fig. 2e,f)**. These findings suggest that Tau inhibition mitigates Dex-driven apoptosis and cartilage degeneration.

GR and Tau knockout modulate Dex-induced chondrocyte apoptosis and cartilage degradation. GR KO and Tau KO mice were employed to examine Dex responsiveness. GR or Tau knockout Chondrocytes were generated using CRISPR–Cas9 technology **(Fig. 3a)**. Notably, Tau deficiency markedly attenuated the Dex-induced suppression of *ACAN* and *COL2* expression (**Fig. 3b**). In vivo, Tau KO mice displayed reduced cartilage degradation, lower OARSI scores **(Fig. 3c,d)**, and decreased chondrocyte apoptosis compared with wild-type (WT) controls following Dex injection **(Fig. 3e,f)**. These findings indicate that Tau acts as an important low affinity receptor, alongside high affinity receptor GR, and plays a major role in mediating Dex-induced chondrocyte apoptosis and OA progression.

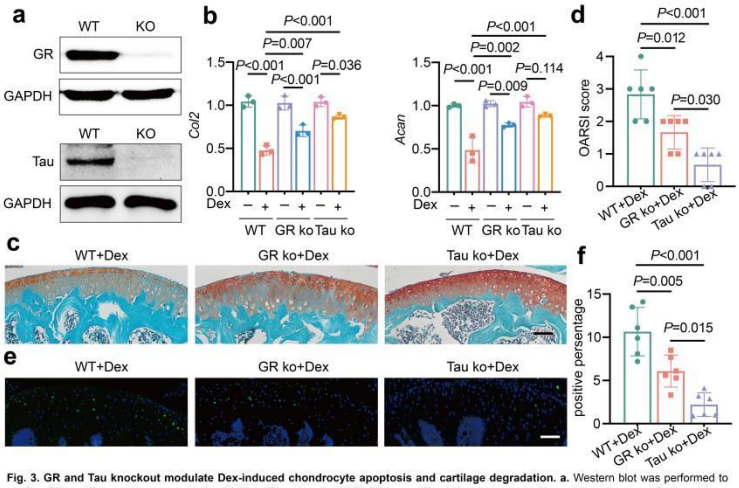

Fig. 3. GR and Tau knockout modulate Dex-induced chondrocyte apoptosis and cartilage degradation. a. Western blot was performed to confirm GR and Tau protein expression in c28/I2. **b.** Chondrocytes (WT, GR ko and Tau ko) were treated with or without Dex, and Col2 and Acan mRNA levels were measured by RT-qPCR. **c,d.** Articular cartilage from WT, GR ko, and Tau ko mice receiving Dex injections was processed for Safranin O staining (scale bar= 100μm), evaluated by OARSI scoring. **e,f.** Apoptosis analyzed by TUNEL staining (e) with quantification of positive signals (f), scale bar= 100μm.

Discussion This study reveals that Dex accelerates chondrocyte apoptosis and cartilage degradation through Tau besides GR, with pharmacological inhibition (TRx0237) of Tau effectively mitigates these deleterious effects, highlighting Tau as a promising therapeutic target to reduce glucocorticoid-associated cartilage damage in OA. Subsequent investigations will utilize the destabilization of the medial meniscus (DMM model to delineate the role of Tau in OA progression.

Conclusion The Tau plays a central role in driving chondrocyte apoptosis and osteoarthritis progression. Targeting Tau pharmacologically offers a potential strategy to protect cartilage from glucocorticoid-induced damage.