環形 RNA 在蛋白尿引起之發炎反應的功能性探討

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環形 RNA (CircRNA) 是利用反向剪接所形成的長鏈非編碼 RNA，可與 miRNA 反應，抑制 miRNA 功能、或與蛋白質結合調控轉錄或轉譯等，近期的研究之中發現了環形 RNA 的異常調控與發炎反應和多種疾病相關。

末期腎臟病 (End stage renal disease; ERSD)，為不可逆的腎臟受損，而台灣是全球 ERSD 發病率和患病率最高的國家。導致 ERSD 的原因有糖尿病、高血壓和慢性腎炎 (chronic kidney disease; CKD) 等，CKD 中蛋白尿 (albuminuria) 的持續存在會導致腎小管間質發炎和纖維化，讓 CKD 惡化成 ESRD。

先前實驗室的研究當中發現長非編碼 RNA (long non-coding RNA; lncRNA) 在 CKD 中扮演了重要的角色。但 CircRNA 在 CKD 中是否擁有類似的功能則尚不清楚。

因此，我們利用了 HK-2 腎小管上皮細胞作為平台。藉由給予細胞過量的白蛋白，我們可以模擬蛋白尿誘發的發炎反應。我們將有發炎反應的 HK-2 細胞做全轉錄體定序 (Whole transcriptome sequencing)，發現了共有 6291 個 circRNA 的表現量被過量的白蛋白影響。

我們選了其中表現量差異最大的 29 個 circRNA，利用 RNase R 與 RT-PCR 實驗，還有 circular junction 定序，確定了這些 circRNA 表現。此外我們藉由 qRT-PCR 定量，確立了這些 circRNA 的變化。為了探討 circRNA 是否有功能，我們選擇了曾經被報導過具有功能的 circRNA，針對了這些 circRNA 設計了 overexpression (OE) 與 Knockdown (KD) 的實驗，藉由觀察 OE 或 KD circRNA 之後的發炎反應，我們可以明確知道有哪些 circRNA 可調控蛋白尿所引發的發炎反應。

Functional Investigation of Circular RNA in Albuminuria-Induced Inflammatory Response

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Circular RNAs (circRNAs) is a long non-coding RNA generated through back-splicing, which can interact with miRNA, inhibiting miRNA function, or bind to proteins to regulate transcription or translation. Recent studies showed that the abnormal regulation of circRNAs are associated with inflammatory responses and various diseases.

End-stage renal disease (ESRD) represents an irreversible kidney damage, with Taiwan having the highest incidence and prevalence rates. ESRD can be caused by risk factors such as diabetes, hypertension, and chronic kidney disease (CKD). In CKD, the persistent albuminuria leads to tubulointerstitial inflammation and fibrosis, exacerbating CKD into ESRD.

Previous studies in our laboratory have highlighted the significant role of long non-coding RNA (lncRNA) in CKD. However, it remains unclear whether circRNAs exhibit similar functions in CKD.

Therefore, we utilized HK-2 renal tubule epithelial cells as a platform. By treating cells with excessive albumin, we could mimic the inflammatory response induced by albuminuria. We subjected the albumin-treated HK-2 cells into next generation sequencing for whole transcriptome analysis, showing that the expression levels of 6291 circRNAs were affected by excessive albumin.

We then selected the top 29 circRNAs with the most significant expression differences for validation using RNase R treatment, RT-PCR assay, and circular junction sequencing. Further, we quantified the expression changes of these circRNAs through qRT-PCR in albumin-treated HK-2 cells. To investigate the functional roles of circRNAs, we chose 9 circRNAs with the most substantial upregulation and downregulation and constructed overexpression (OE) and knockdown (KD) experiments. By observing the inflammatory response following OE or KD of these circRNAs, we can conclusively identify which circRNAs can regulate the inflammation response induced by albuminuria.