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Development and Validation of a RP-HPLC Method for Determination of Chondroitin Sulphate and Curcumin in Topical Formulation

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***SUMMARY*. A very simple, reproducible, accurate and precise method was developed for simultaneous determination of chondroitin sulphate (CS) and curcumin (Cur) using reverse phase High performance liquid chromatography (RP-HPLC) at 195nm in drug solutions and cream formulation. The chromatographic conditions for developed method were optimized by changing mobile phase compositions over different reverse phase columns. The developed method was validated by various parameters like specificity, linearity, limit of detection (LOD), limit of quantification (LOQ), range, precision, accuracy and system suitability were performed and calculated. Stability studies under stressed conditions were carried out to evaluate the effects of acid, alkali, oxidation, heat and degradation by UV light. The validated method was found linear over the concentration range 192.05 to 1500μg/mL for CS and 2.9 to 1500 μg/mL for Cur, respectively, with a correlation coefficient ($r^2 > 0.999$) for both analytes. The inter-day and intra-day precision were found to be 1.80 and 0.72 for CS and Cur respectively. The accuracy calculated as recovery was between 95-105% for both CS and Cur. The stability studies showed that CS was highly sensitive to acid, base and oxidative degradation and to lesser extent to heat and UV, whereas no considerable effects of acid, heat and oxidation were observed in case of Cur, while curcumin shows a significant degradation under alkali and UV light exposure. In conclusion, the developed and validated method was found to be simple, precise, accurate and can be used for routine analysis of CS and Cur in drug solution and cream formulation.**

RESUMEN. Se desarrolló un método muy simple, reproducible, exacto y preciso para la determinación simultánea de condroitín sulfato (CS) y curcumina (Cur) mediante cromatografía líquida de alta resolución (RP-HPLC) de fase inversa a 195 nm en soluciones de fármacos y formulación en crema. Las condiciones cromatográficas para el método desarrollado se optimizaron cambiando las composiciones de la fase móvil en diferentes columnas de fase inversa. El método desarrollado fue validado por varios parámetros como especificidad, linealidad, límite de detección (LOD), límite de cuantificación (LOQ), rango, precisión, exactitud e idoneidad del sistema que se realizaron y calcularon. Se realizaron estudios de estabilidad en condiciones de estrés para evaluar los efectos del ácido, álcali, oxidación, calor y degradación por luz ultravioleta. El método validado se encontró lineal en el rango de concentración de 192,05 a 1500 μg / ml para CS y de 2,9 a 1500 μg / ml para Cur, respectivamente, con un coeficiente de correlación ($r^2 > 0,999$) para ambos analitos. Se encontró que la precisión entre días e intradiarios era de 1,80 y 0,72 para CS y Cur respectivamente. La precisión calculada como recuperación fue entre 95-105% tanto para CS como para Cur. Los estudios de estabilidad mostraron que el CS era altamente sensible a la degradación ácida, básica y oxidativa y, en menor medida, al calor y a los rayos UV, mientras que no se observaron efectos considerables de ácido, calor y oxidación en el caso del Cur, mientras que la curcumina muestra una degradación significativa bajo álcali. y exposición a la luz ultravioleta. En conclusión, se descubrió que el método desarrollado y validado es simple, preciso, exacto y se puede utilizar para el análisis de rutina de CS y Cur en la formulación de la solución de fármaco y la crema.

KEY WORDS: curcumin, chondroitin sulphate, stability, system suitability

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