NOVEL GRADIENT ELUTION (U)HPLC METHOD DEVELOPMENT THAT ENABLES SUCCESSFUL METHOD TRANSFER

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Nowadays, one of the ultimate requirements in drug analysis is rapid method development, prompt chromatographic run time and long lasting method's lifecycle. Special attention should be paid to development of gradient elution (U)HPLC methods that are commonly related to troubleshooting during the inter-laboratory method transfer. The dwell volume difference between (U)HPLC systems is the main reason for this issue (1). The aim of this study was to propose new gradient method development methodology with integrated dwell volumes values in the optimization process. This is especially useful approach for industry where is frequently known which (U)HPLC instruments will be applied for drug analysis. Proposed methodology was tested on the model mixture which encompassed dabigatran etexilate mesylate and its nine impurities. Experimental design methodology and three different (U)HPLC instruments with significant dwell volume differences were utilized. Statistically significant variables were selected with Plackett-Burman design, and along with dwell volume values were included in D-optimal design. The separation criteria s between adjacent peaks was selected as output for method optimization. Indirect modelling together with Monte Carlo simulations enabled selection of optimal and robust chromatographic conditions joint for all instruments. The optimal conditions included 24% (v/v) of initial amount of acetonitrile, 54% (v/v) of the final amount of acetonitrile, 15 min of gradient elution run time and pH value equal to 4.9. The proposed method utility was proved since it was successfully validated and met all validation criteria (2).

References

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NOVI PRISTUP U RAZVOJU (U)HPLC GRADIJENTNIH METODA KOJI BI OMOGUĆIO USPEŠAN TRANSFER

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U današnje vreme, jedan od važnih zahteva prilikom razvoja hromatografskih metoda jeste da se razvoj uradi brzo, da vreme trajanja hromatografske analize bude kratko i da se obezbedi dug životni ciklus metode. Posebnu pažnju treba obratiti na (U)HPLC metode sa gradijentnim eluiranjem kod kojih se često javljaju problemi prilikom međulaboratorijskog transfera. Glavni razlog za zahtevan transfer gradijentnih (U)HPLC metoda je razlika u vrednostima *dwell volume* između (U)HPLC uređaja (1). Cilj ovog rada je bio da predloži novu metodologiju razvoja gradijentnih (U)HPLC metoda, pri čemu će vrednosti *dwell volumes* biti integrisane sa drugim ulaznim promenljivama u fazi optimizacije. Ovo bi bio posebno koristan pristup u industriji gde se obično zna na kojim će sve (U)HPLC instrumentima metoda biti korišćena. Metodologija je testirana na smeši dabigatran eteksilat mezilata i njegovih devet nečistoća. Za razvoj metode korišćen je eksperimentalni dizajn i tri različita (U)HPLC instrumenta sa značajnim razlikama u *dwell volume* vrednostima. Statistički značajne ulazne promenljive su dobijene primenom *Plackett-Burman* dizajna, i zajedno sa vrednostima dwell volume su bile uključene u *D-opimal* dizajn. Kriterijum razdvajanja *s* između susednih parova pikova odabran je kao odgovor u odnosu na koji je metoda optimizovana. Indirektno modelovanje zajedno sa Monte Karlo simulacijom omogućilo je izbor optimalnih i robusnih hromatografskih uslova zajedničkih za sva tri instrumenta. Oni su uključivali 24% (v/v) početnog udela acetonitrila, 54% (v/v) finalnog udela acetonitrila, vreme trajanja gradijenta od 15 minuta i pH vrednost 4,9. Primenljivost predložene metodologije je dokazana, jer je dobijena metoda uspešno validirana na sva tri (U)HPLC instrumenta (2).

Literatura

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