ANALYTICAL DESIGN SPACE STRATEGY FOR THE DEVELOPMENT OF LC METHODS TO COMBAT POTENTIALLY COUNTERFEIT ANTIBIOTIC DRUGS

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The poor quality of medicines sold worldwide is a crucial problem of public health. The amount of counterfeit drugs is estimated at 10 % by the Food and Drug Administration. However, astonishing values (80%) were reported in some African countries[1]. One cause of the large diffusion of pharmaceuticals counterfeiting in developing countries is the lack of quality control at different levels (from formulation to delivery); making quite difficult the national regulatory bodies mission of ensuring the monitoring of medicines. Antibiotics are drugs commonly used in Africa and generic analytical methods are needed for the quality control of these medicines.

Following the context of pharmaceuticals guidelines ICH Q8 R2 [2], an innovative methodology based on design space (*DS*) was used to develop LC methods to trace, screen and determine antibiotic drugs. Briefly, *DS* could be defined as a subspace of the experimental domain providing chromatographic conditions that will ensure the quality of the separation [3-4-5]. For that purpose, retention times of each peak (at the beginning, the apex and the end) were recorded to model the chromatographic behaviour. The separation between the two peaks of the most critical pair was used as the quality criterion.

Firstly, a three factor D-optimal design was constructed in order to develop a reverse phase LC method to separate 19 antibiotics. Because of the close chemical structures of the molecules of interest and thus their similar chromatographic behaviour, the probability of separation was estimated at 10 %. A second D-optimal design with redefined experimental domain for each factor was set, allowing reaching an estimated probability to separate all the compounds close to 100%. The predicted optimal condition was successfully tested. This adjustable experimental design enabled to develop a robust method to screen a large panel of antibiotics.

Secondly, a geometric transfer of the optimized method to an ultra high performance liquid chromatography (UHPLC) was investigated in order to demonstrate the method robustness and to achieve high throughput separation. The quality of separation was not affected despite a slight loss of efficiency, thanks to the identified high guarantees given by the design space strategy. Moreover, a high speed and low cost (solvents consumption drastically decreased) method is a huge advantage for quality control laboratories.

Finally, the UHPLC method was validated in compliance with ICH Q2 R1 guideline using the accuracy profile approach.Then, several medicines marketed in Democratic Republic of Congo were analysed in order to test their conformity with the specifications. Two thirds of the samples tested were not conform because the drug content was higher than the limit; drug overdosing is harmful for the public health and can lead to serious damage of vital organs.

Thus, the main objective of this work was successfully reached. Indeed, a generic method able to trace, screen and quantify various antibiotic drugs in order to help detecting the potential poor quality medicines was proposed. Moreover, the interest of design space strategy was successfully demonstrated. This one should be used to develop and demonstrate robust method especially for any complex samples or generic method.

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