Development and validation of a novel capillary electrophoresis method for simultaneous analysis of ribociclib, letrozole and ibuprofen

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INTRODUCTION

Breast cancer is the most common cancer worldwide while the HR+/HER2- subtype is the most prevalent.[1] Locally advanced or metastatic HR+/HER2- breast cancers are treated with the combination of cyclin-dependent kinase (CDK) 4 and 6 inhibitors and drugs with anti-estrogen effects. One of the common combinations used in clinical practice is ribociclib with letrozole (Figure 1).[2] Many breast cancer patients suffer from various comorbidities and side effects. Thus, different drug classes are regularly used in clinical practice, painkiller ibuprofen being one of the most common. Because of that, we developed a capillary electrophoresis (CE) method for the simultaneous determination of ribociclib, letrozole, and ibuprofen.

Capillary electrophoresis is a cost-efficient technique often considered an alternative to liquid chromatography as it offers a greener, cheaper, and simpler analyses since it requires significantly smaller sample volumes and reagent consumption and produces less waste.[3]

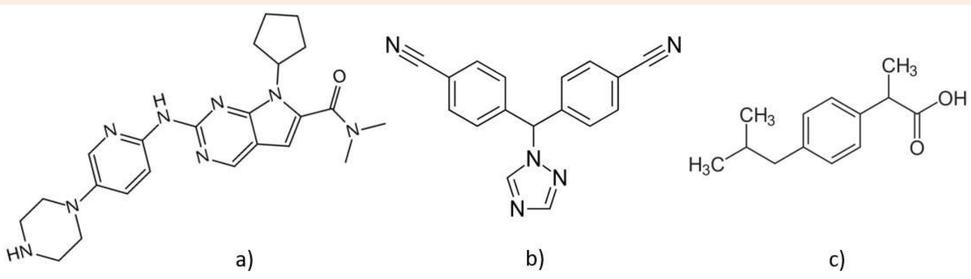
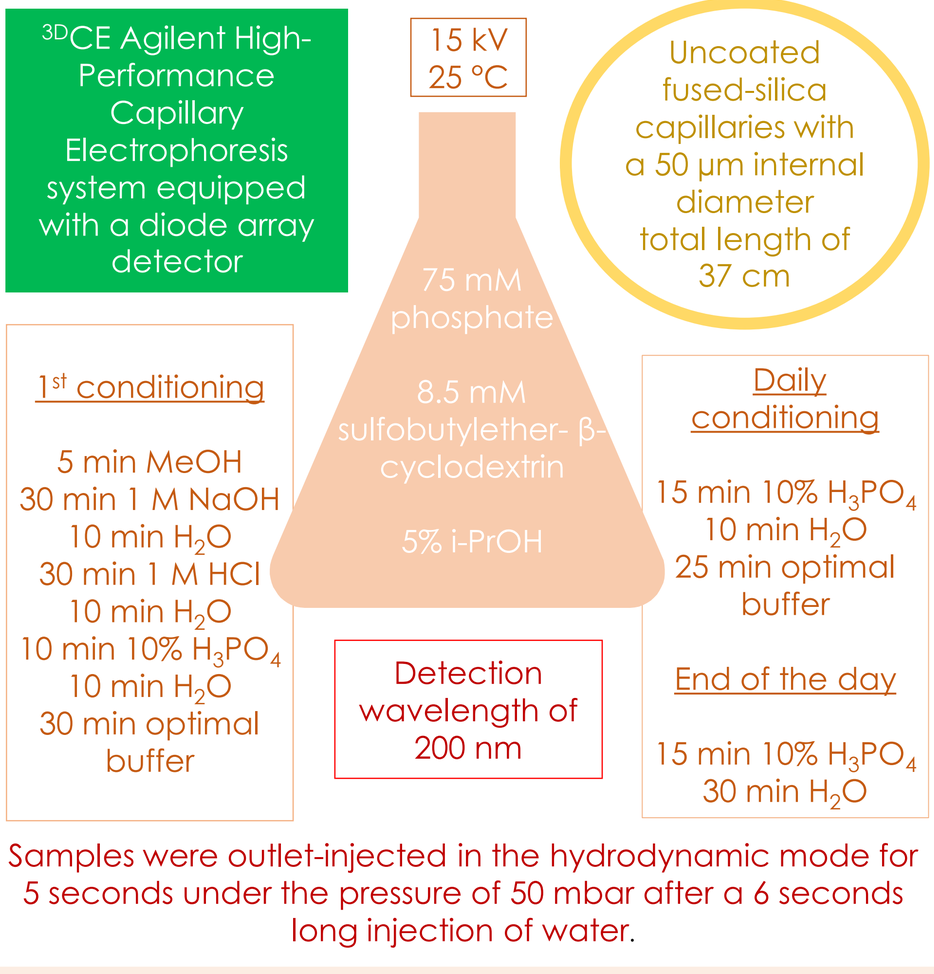


Figure 1. Chemical structures of **a)** ribociclib, **b)** letrozole and **c)** ibuprofen

EXPERIMENTAL



Samples were outlet-injected in the hydrodynamic mode for 5 seconds under the pressure of 50 mbar after a 6 seconds long injection of water.

METHOD DEVELOPMENT AND VALIDATION

During method development, different buffer types (borate and phosphate with and without SDS) and additives and surfactants (SBE-β-CD and i-PrOH) were tested. Separation was achieved with 75 mM phosphate, 8.5 mM sulfobutylether-β-cyclodextrin, and 5% i-PrOH (Figure 2). The optimal conditions were 15 kV, 25 °C and 5 s hydrodynamic outlet injection under 50 mbar. Preconditioning consisted of 10 min rinse with 10% H3PO4 and 15 min with the optimal buffer, while postconditioning was done under -15 kV voltage for 8 min. The developed method was validated with ketoprofen as the internal standard according to the ICH guidelines and was shown to be linear in the range of 1 to 75 µg mL^{-1} (Table 1). Intra-day and inter-day repeatabilities were evaluated for method precision with acceptable RSD values. Corrected peak area (ratio of peak area of the analyte and of the internal standard) and of the corrected migration time (ratio of migration time of the analyte and of the internal standard) were determined. The robustness of the method was tested by changing applied voltage (± 0.5 kV), sulfobutylether-β-cyclodextrin concentration (± 0.5 mM), and temperature (± 2 °C) one variable at a time, and the changes in peak areas and migration times were shown to be in the range of 0.34 to 7.27 %.

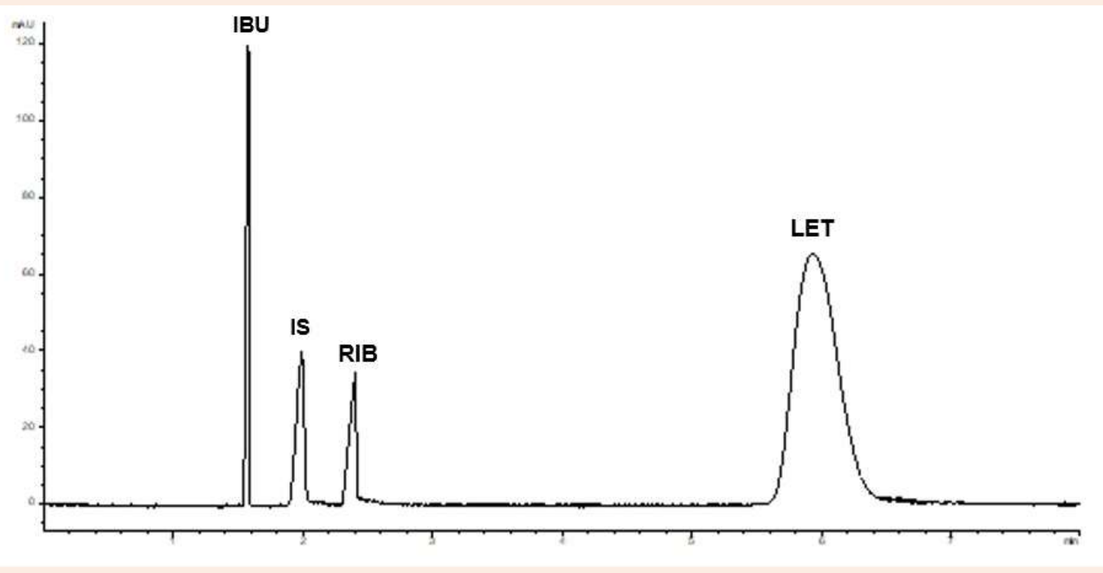


Figure 2. Electropherograms of IBU, RIB and LET (all 50 µg mL^{-1}) under optimal conditions (75 mM phosphate, 8.5 mM sulfobutylether-β-cyclodextrin, and 5% i-PrOH, V = 27.5 kV, λ = 200 nm, 25 °C, injection time 5 s after a 6 seconds long injection of water, pressure 50 mbar)(IBU = ibuprofen, RIB = ribociclib, LET = letrozole and IS = internal standard)

Table 1. Regression and correlation data for the calibration curves in concetration range 1 to 75 µg mL^{-1}

Analyte	Regression equation	R^2
Ribociclib	y = 0.0195x – 0.0112	0.9970
Letrozole	y = 0.1489x + 0.2755	0.9914
Ibuprofen	y = 0.0258x – 0.0202	0.9978

CONCLUSION

A novel, fast, cheap, and green CE method for simultaneous determination of ribociclib, letrozole and ibuprofen in less than 7 min is proposed. The validated method is the basis for the further development of an even more sensitive method capable of detecting low plasma concentrations of these drugs using innovative sample preparation techniques and coupling to the mass detector which could then be applied to standard clinical practice.

References

1. Shah et al., *The Oncologist,* 2020, 25, 900–908.
2. Im et al., *New England Journal of Medicine,* 2019, 381 (4), 307–316.
3. Deeb et al., *Electrophoresis,* 2014, 35 (1), 170–189.

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