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Short communication

The role of cocrystallization screening for the assessment of structure-activity relationship in drug development

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Introduction

The selection of the crystalline phases in a form of molecular co-crystals has become scientific challenge at the early stage of drug development of pharmaceutical formulations and in the late stage of synthesis and isolation of active pharmaceutical ingredients (APIs) in desirable defined crystalline forms. Optimal crystal form of API interactively interrelates and impacts its aqueous solubility and dissolution rate that are benchmark for drug delivery and absorption determining the extent of its bioavailability and pharmacokinetics profile. Determining the crystal structure and revealing the crystal packing forces and geometry of the API has impact its physicochemical properties. This approach is the criteria for assessment of the performance of the API. The range of crystal forms in which molecular co-crystals of APIs may exist is advantageous comparing to their polymorphs, salts, solvates and hydrates due to the vast number of potential co-formers which extend the limited counterions for salt formation implying the existence of more complex intermolecular interactions based on different H-bonding patterns with API that lead to conformational changes and flexibility for crystal packing in process of co-crystallization.

Co-crystallization became well known bottom-up approach starting from intermolecular interactions among either selected neutral, ionic or zwitterionic molecules to design and control the properties of the multicomponent crystals (Braga, 2004). In the scope of interest for drug design and formulation, Good et al. (2009) and Cheney et al. (2011) emphasized that the main advantage for designing pharmaceutical co-crystals (PCCs) is, through their modulating properties, to improve the performance of the native APIs such are: biopharmaceutical profile (solubility and dissolution rate), thermodynamical stability (phase transition of polymorphs, solvate/ hydrate formation, decomposition) or bulk powder processability (flowability, compressibility, particle size and shape control). Childs et al. (2007) has pointed out the necessity co-crystals (CCs) semantically to be classify based on accomplishments in research of supramolecular chemistry. This approach enlightens the complex reality of multi-component systems and the wide scope associated between salts and co-crystals, and their differences based on the location of the transferred proton within the salt - co-crystal continuum.

Biguanide drugs are well known and wide used oral antidiabetic drugs for oral therapy of diabetes type-2 that directly improve insulin action. Recent studies in the research work carried out by Vujic et al. (2015) has pointed out that biguanides in combination with targeted inhibitors in order to obtain synergy in reduction cell viability, inhibited tumor growth in the mutated neuroblastoma rat sarcoma oncogene (NRAS) protein from melanoma cells. Hence, it is expected that combination of biguanides which affect activation of the AMP-activated protein kinases (AMPK) and the regulation of energy metabolism with outcome to cell's energy sparing, in combination with other anti-cancer drug-models would influence direct blocking cell's signaling and hinder the resistance.

Materials and methods

Co-crystallization screening reveals protocol was undertaken in order to grown single crystalline phases of PCCs composed of drug model metformin (MET), selected from biguanides class of drugs and coformers that belong to different pharmacotherapy and functional group classes, respectively.

Co-crystallization screening was carried out on applying slow-rate solvent evaporation method for growing sin-

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gle crystalline phases at room temperature with quality absolute structure to be determined. An equimolar quantity of MET and co-crystal partner was dissolved in the minimum quantity of ethanol and left for slow evaporation at room temperature. Colorless crystals were observed after a few days.

For four MET PCC models, the methods of preparation reproducible batches were optimized. The quality of batches was controlled by Powder x-ray diffractometer comparing the obtained experimental diffractograms with the same one that was theoretically generated from the single crystal for each of four PCC models.

Single-crystal diffraction data were collected on a Nonius Kappa diffractometer equipped with a CCD detector with graphite-monochromatized MoKα radiation (λ = 0.71069 Å). Intensities were corrected for Lorentz and polarization effects. The structures were solved by direct methods with the SIR97 suite of programs and refinement were performed on F2 by full-matrix least-squares methods with all non-hydrogen atoms anisotropic.

Flow-cytometry was applied for measuring viability of the two PCC models.

Results and discussion

Vujic et al. (2015) has carried out research for both pro-cancer and anti-cancer effects of biguanides on cancer cells, indicating existence of association of the antidiabetic therapy and reduced risk of cancer in diabetic patients. Because biguanide represents the π-conjugated system, MET can exist in three resonance-stabilized forms, i.e. as neutral molecule (MET), monoprotonated (MET+) or diprotonated (MET2+) cation, with dissociation constants in water in range from pKa1 ≈ 12.00 to pKa2 ≈ 2.00.

A search of the biguanide fragments in the structural literature, both in the CCDC (Cambridge Crystallography Database Center) database and in patents, shows that in crystals MET exists as monoprotonated (MET+) or deprotonated (MET2+) but never in its neutral form MET.

We have undertaken a systematic study of the crystal chemistry of MET with the aim of understanding its properties in the solid state and finding relationships with its biopharmaceutical profile. We have determined the structures of the 29 MET PCCs. Four of this MET PCC models are "drug-drug" type of co-crystals. The ligand used for co-crystallyzation was from the following classes: inorganic acids (nitric, phosphoric and carbonic acid); organic NH-type acids (saccharine and acesulfame); organic OH-type acids (squaric and picric acid); monocarboxylic acids (fumaric, acetic, trifluoroacetic, trichloroacetic, dichloroacetic, monochloroacetic, glycolic, salicylic, diclofenac) and dicarboxylic acids (oxalic, malonic, maleic, fumaric, succinic, adipic acid).

Conclusion

In the paper are presented structure analyses for "drug-drug" type of PCCs where both API and CF exhibit pharmacological effect. This approach of designing "drug-drug" type of PCC aligned to the strategy for drug repositioning, the idea for use of a drug for treating diseases other than the drug-specified. This concept was prompted in 2012 through the Discovering New Uses for Existing Molecules program, initiated by US's National Institute of Health (NIH).

The case study underlines the crystal growth and the method of preparation for "drug-drug" type of PCCs wherein two different APIs cocrystallized in single crystal cell, and that represent new paradigm for approaching in development of "fixed-doses" or "combo" pharmaceutical formulations. Preliminary results of the Structure-Activity Relationship study on the co-crystals composed of MET with dichloroacetic acid indicate dual and complementary anti-cancer activities of the two selected drug models for co-crystallization.

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