Drug–Drug Molecular Salt Hydrate of an Anticancer Drug Gefitinib and a Loop Diuretic Drug Furosemide: An Alternative for Multidrug Treatment

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ABSTRACT: A 1:1 monohydrate salt containing gefitinib, an orally administrated chemotherapy treatment for lung and breast cancers and furosemide, a loop diuretic drug, commonly used in the treatment of hypertension and edema, has been prepared. The molecular salt crystallized in triclinic *P*-1 space group. The C–O bond lengths (~1.26 Å) in the COOH group show that proton transfer has occurred from furosemide to morpholine moiety of the gefitinib suggesting cocrystal to be ionic. The morpholine moiety of the gefitinib showed significant conformational change because of its involvement in conformation dictating the strong N–H···O hydrogen bonding interaction. The strong hydrogen bonding interaction between gefitinib and furosemide places their benzene rings in stacking mode to facilitate the generation of π-stack dimers. The neighboring dimers are bridged to each other via water molecule through N–H···O, C–H···O, O–H···N, and O–H···O interactions. The remarkable stability of the salt hydrate could be attributed to the strong hydrogen bonding interactions in the crystal structure. Interestingly, release of water from the lattice at 140°C produced new anhydrous salt that has better solubility and dissolution rate than salt hydrate. The drug–drug molecular salt may have some bearing on the treatment of patient suffering from anticancer and hypertension. J Pharm Sci 104:4207–4216, 2015
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INTRODUCTION

Preparation of multicomponent crystals or cocrystals[1–6] and the study of their physicochemical properties have evolved into a contemporary area of research comprising pharmaceutical solids,[7–12] agrochemicals,[13] high energy materials,[14–17] and so on in the last two or three decades. Constant and consistent attempt to develop active pharmaceutical ingredient (API) cocrystals with suitable cocrystal excipient is gaining widespread research interest because of its exploitation in tuning the physicochemical properties of an API that could be economically beneficial and intellectually stimulating.[8,18–20] Thus, preparing pharmaceutical cocrystals is an innovative strategy to enhance the performance of the API without affecting their therapeutic efficiency. Furthermore, cocrystals are less prone to exhibit polymorphism,[21] hence reduces the possibility of polymorphic transition during storage that is the biggest problem in the pharmaceutical industry. The cocrystal former can be an API or any another substance that is generally recognized as safe (GRAS) compound prescribed by US Food and Drug Administration (FDA).[22] Multi-API cocrystals have potential relevance in the context of delivery of combination drug that can be experimented to prevail over the issues associated with traditional combination drugs. Furthermore, tablet of multi-API cocrystal boost clinical effectiveness and reduce production cost.

In recent times, the design and synthesis of cocrystals are gaining considerable attention first because of their involvement in tuning the physicochemical properties of a substance and second an academic interest in molecular recognition-driven self-assembly process.[11] Intermolecular interactions that are central to the molecular recognition process are responsible for the generation of families of molecular networks either with the same molecular components (single component crystals and their polymorphs) or with different molecular components (cocrystals) in the crystalline state and cocrystallization is a result of competing molecular associations between the two. The components in a cocrystal assemble via noncovalent interactions such as hydrogen bonds, halogen bonds, π···π, or van der Waals. Here, we report the molecular salt (cocrystal ion) formation of gefitinib (tyrosine kinase inhibitor), an orally administrated chemotherapy treatment for lung and breast cancers that inhibits tyrosine kinase,[23] an enzyme that transport phosphates from ATP to protein's tyrosine residue and furosemide,[24,25] a loop diuretic drug, commonly used in the treatment of hypertension and edema (Fig. 1a).[26,27] Gefitinib is currently marketed in over 64 countries for patients with advanced nonsmall cell lung cancer who have received at least one previous chemotherapy regime.[28] Earlier, we reported the novel metastable polymorph of gefitinib that is isostructural to its stable crystals form and heating the same at the transition temperature converts it to thermodynamically stable crystal form.[29] We started our research work with the aim to improve the solid properties of gefitinib and develop its

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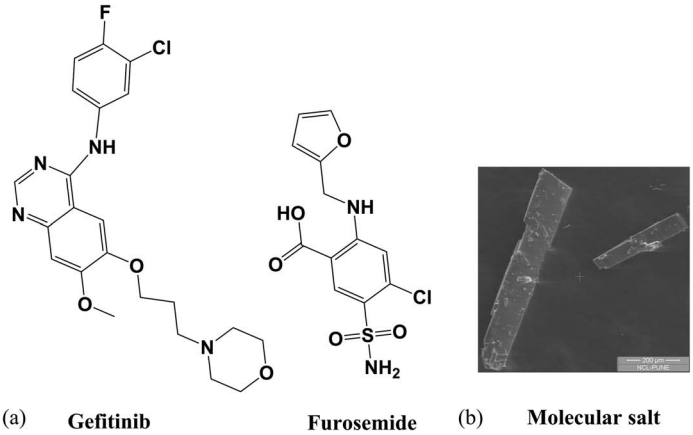

Figure 1. (a) Structural diagram of gefitinib and furosemide and (b) Scanning Electron Microscope (SEM) image of crystals of gefitinib–furosemide molecular salt hydrate.

multi-API cocrystal,[30–33] to treat and manage the patient suffering from cancer and other diseases. Patients undergoing targeted chemotherapy with drugs such as gefitinib could generally have kidney toxicity such as renal dysfunction and nephrotic syndrome as its side effects along with common side effects such as diarrhea, rash, acne, dry skin, nausea, vomiting, and interstitial lung disease.[34,35] However, reports on gefitinib causing hypertension are less.[36] To overcome the effect of kidney toxicity and hypertension caused by gefitinib, there is a need of combination drugs therapy. In this regards, we chose furosemide, a loop diuretic drug used for the treatment of hypertension and edema as cocrystal former. All the three polymorphs of the drug furosemide suffer from poor aqueous solubility (~6 μg/mL).[25] API furosemide was also chosen because of its acidic nature and gefitinib is known to form cocrystals with acidic compounds[37] through acid-amide hydrogen bonds. Another advantage of preparing molecular complex of furosemide with gefitinib is that it could improve the solubility and dissolution rate of furosemide compared with its monotherapy treatment.[24] In this contribution, we report the crystallization of gefitinib with furosemide, the characterization of new molecular salt and correlation of its crystal structure with the stable crystalline form of gefitinib and furosemide.

EXPERIMENTAL

Crystallization

Cocrystallization was carried out from equimolar amounts of commercial samples of gefitinib and furosemide by grinding (dry grinding as well solvent-assisted grinding) and slow evaporation from the solution under ambient conditions. The grinding experiment was carried out manually using mortar and pestle. The 1:1 stoichiometric molar ratio of gefitinib and furosemide was grinded for about 15 min using both dry (neat) grinding and liquid-assisted grinding (or kneading) methods. In liquid-assisted grinding method, either small (catalytic) amount of water or the ethanol–water mixture was added to the grinding mixture in two separate experiments. The grinded sample was characterized using powder X-ray diffraction (PXRD) to verify the formation of salt by comparing it with simulated powder pattern from single-crystal X-ray diffraction (XRD) of molecular salt.

The same grinded material was used for solution crystallization. The grinded sample was dissolved in the ethanol–water mixture (1:1, v/v) and heated up to 70°C–75°C to dissolve the sample for about 10–20 min. The hot solution was then filtered into the conical flask to remove the traces of undissolved compound or any foreign material, and the solution was evaporated at room temperature.

Single-Crystal X-Ray Crystallography

Single-crystal structure of gefitinib–furosemide molecular salt was determined by measuring X-ray intensity data on a Bruker SMART APEX II single-crystal X-ray CCD diffractometer having graphite-monochromatized (Mo-K_α = 0.71073 Å) radiation at 150(2) K. The X-ray generator was operated at 50 kV and 30 mA. A preliminary set of cell constants and an orientation matrix were calculated from total 36 frames. The optimized strategy used for data collection consisted different sets of ϕ and ω scans with 0.5° steps in ϕ/ω. Data were collected with a time frame of 10 s keeping the sample-to-detector distance fixed at 5.00 cm. The X-ray data acquisition was monitored by APEX2 program suit.[38] All the data were corrected for Lorentz-polarization and absorption effects using SAINT and SADABS programs integrated in APEX2 package.[38] The structure was solved by direct method and refined by full matrix least squares, based on F^2, using SHELX-97.[39] The H-atoms bound to the N-atoms (N–H and NH_2) and oxygen of the water molecule were located in difference Fourier and refined isotropically. Other hydrogen atoms were placed in idealized positions (C-H = 0.95 Å for SP^2 hybridized C-atoms including H atoms in phenyl, furan, and pyrimidine groups, C-H = 0.99 Å for the methylene H atoms, C-H = 0.98 Å for the methyl H-atoms) and constrained to ride on their parent atoms [Uiso(H) = 1.2 Ueq(C) or 1.5 Ueq(methyl C)]. Crystallographic data for gefitinib-furosemide molecular salt is summarized in Table 1. Molecular diagrams were generated using Mercury programs.[40] Geometrical calculations were performed using SHELXTL[39] and PLATON.[41]

Powder X-Ray Diffraction

The experimental PXRD patterns were recorded on Rigaku Micromax-007HF instrument (high-intensity microfocus rotating anode X-ray Generator) with R-axis detector IV++ at a continuous scanning rate of 2° 2θ/min using Cu Kα radiation (40 kV, 30 mA) with the intensity of the diffracted X-ray being collected at intervals of 0.1° 2θ. A nickel filter was used to remove Cu Kβ radiation.

Infrared Spectroscopy

The solid-state infrared spectrum of the molecular salt was acquired by using BRUKER ALPHA Fourier transform infrared spectrophotometer at room temperature in Nujol from 500 to 4000 cm^{-1} range.

Differential Scanning Calorimetry

The thermal behavior of novel crystalline salt of gefitinib–furosemide was investigated by measuring the enthalpy change on a TA Q-100 Differential Scanning Calorimeter instrument. Crystals obtained from crystallization were first air-dried before they were used for differential scanning calorimetry (DSC)