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Efficient Multicomponent Reaction Synthesis of the Schistosomiasis Drug Praziquantel

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Schistosomiasis, also known as snail fever, is a very large but nevertheless neglected tropical disease (NTD) affecting more than 200 million people worldwide, with more than 90% occurring in sub-Saharan Africa.[1] More than 700 million people live in areas where schistosomiasis is endemic. The causing disease agents are the small worms *S. haematobium* and other species transmitted by skin contact in standing freshwater. Although schistosomiasis has a relative low mortality rate (≈300000 per year) it is highly debilitating, leading to damage of the internal organs, cancer, and impaired growth and cognitive development in children. Importantly, there is evidence for a strong correlation between schistosomiasis and HIV infection in Africa.[2] Thus, the urinary form of schistosomiasis, which affects up to 50% of women in parts of Africa, damages the lining of the vagina, the first defensive barrier against HIV. Thus, an affordable $0.32 (US) solution per treatment for preventing HIV/AIDS has recently been proposed based on the highly effective and low-cost anti-schistosomal drug praziquantel (PZQ, **1**).[3] The discovery of PZQ represents the most important progress in the treatment of schistosomiasis since the discovery and reporting of the disease etiology by Theodor Maximilian Bilharz in the 19th century in the seminal papers "Ein Beitrag zur Helminthographia humana, aus brieflichen Mittheilungen des Dr. Bilharz in Kairo, nebst Bemerkungen von C. Th. v. Siebold" and "Über die Eingeweidewürmer Ägyptens".[4,5] PZQ was discovered in a collaborative effort between the pharmaceutical companies Merck and Bayer and has been developed for human use together with the World Health Organization (WHO).[6] It is now the only effective drug to treat schistosomiasis and is one of the few drugs on the WHO's list of essential medicines.[7] PZQ is administered as a racemate and it is not only very effective, it is also very safe. A single treatment with one tablet is typically enough to eradicate the parasite. It is regularly administered to small children and pregnant women. In the privately financed schistosomiasis control initiative (SCI), millions of sub-Saharan Africans at high risk of serious disease are currently treated with PZQ.[8] Cleary, cost-of-goods (COG) is key in reaching as many schistosomiasis suffering people as possible in the poorest countries of the world.

We present here a three-step synthesis of PZQ, which to the best of our knowledge comprise the shortest access to this important drug: three steps from bulk starting materials by using multicomponent reaction (MCR) technology. Currently, the generic form of PZQ is produced by many companies according to a few major processes. The original Merck process uses bulk and cheap isoquinoline as starting material to yield PZQ in a sequential five-step synthesis.[4] A key step in this synthesis, a Reissert reaction, uses a several-fold excess of KCN and therefore, large volumes of aqueous cyanide waste is produced.[9] This comprises a significant environmental threat due to special waste-processing needs, since PZQ is produced on a multi-ton scale per year. Other small- and large-scale processes are sequential five-step and longer syntheses and are described in the Supporting Information.

PZQ is a tetrahydroisoquinoline derivative containing an α-aminoacylamide moiety. This fragment can be advantageously assembled by the classical Ugi multi-component reaction.[10] Therefore, our synthesis strategy uses the Ugi reaction as a key transformation (Scheme 1). Thus, (2-isocyanoethyl)benzene (**2**) reacts with paraformaldehyde (**3**), cyclohexylcarboxylic acid (**4**), and 2,2-dimethoxy ethylamine (**5**) to yield the advanced precursor **6** quantitatively. A Pictet–Spengler reaction under acidic conditions should yield PZQ. However, heating a solution of **6** in dichloromethane to reflux with different Lewis and Brønsted acids, yielded only the Δ^5-2-oxopiperazines. Therefore, we investigated the higher boiling 1,2-dichloroethane as solvent and treated the Ugi intermediate **6** with methanesulfonic acid under reflux temperature overnight as has been reported

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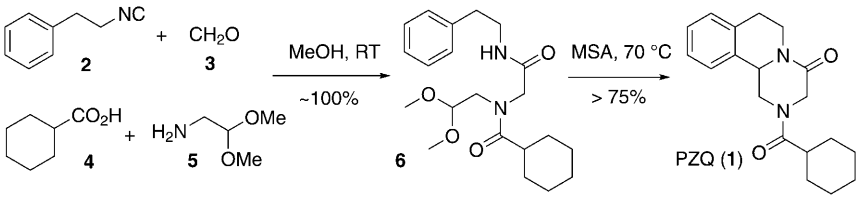

Scheme 1. Stepwise Ugi four-component reaction and Pictet–Spengler reaction to yield the schistosomiasis drug praziquantel (MCA = methanesulfonic acid).

previously.[11] However, only traces of PZQ were obtained in our hands, whereas most of the starting material was decomposed. We then carefully monitored the reaction course and reduced the reaction time to 6 h, which resulted in a poor yield of PZQ of 37%. Next, we found that the reaction could be carried out at lower temperature (70°C) with $MgSO_4$ as an additive to give PZQ in 75% yield in a very clean reaction. In an attempt to render the synthesis more environmentally friendly, we finally eliminated the usage of the chlorinated solvent 1,2-dichloroethane and could isolate PZQ in 76% yield (65% after recrystallization). The required isocyanide **2** is commercially available or can be synthesized from inexpensive and bulk-available phenylethylamine by using the classical Hoffmann's (one step, >60%) or Ugi's formamide method (over two steps, 77%).[12] The herein obtained PZQ is identical with commercial samples in all physical measures and shows the same LD_{50} for schistosome killing in vivo (see the Supporting Information).

Overall, our finding comprises a short and convergent synthesis yielding PZQ (**1**) in an overall yield of ≈45% from readily available materials. The reactions were carried out under very mild conditions and the sequence is atom economic, yielding only water and two equivalents of methanol as side products.[13] Our synthetic pathway for PZQ, with only three steps and inexpensive, bulk-available starting materials could potentially represent an improvement of current processes and has the potential to lower the cost-of-goods of the production of this essential drug.[14,15]

Experimental Section

All reactions were performed under air atmosphere. Analytical thin-layer chromatography (TLC) was performed on SiO_2 plates on alumina available from Whatman. Visualization was accomplished by UV irradiation at 254 nm, or by staining with any one of the following reagents: iodine, ninhydrin (0.3% w/v in glacial acetic acid/*n*-butyl alcohol 3:97), Vaughn's reagent ($(NH_4)_6Mo_7O_{24}\cdot 4H_2O$ (4.8 g) and $Ce(SO_4)_2\cdot 4H_2O$ (0.2 g) in conc. H_2SO_4 (10 mL) and H_2O (90 mL)). Flash column chromatography was performed by using SiO_2 60 (particle size 0.040–0.055 mm, 230–400 mesh, EMD science distributed by Bioman), Preparative TLC was conducted using preparative silica gel TLC plates (1000 μm, 20 cm × 20 cm).

Proton and carbon NMR spectra were obtained on a Bruker Avance 600 MHz NMR spectrometer. Chemical shifts are reported as δ values in parts per million (ppm) as referenced to residual solvent. ^{1}H NMR spectra are tabulated as follows: chemical shift, multiplicity (s = singlet, bs = broad singlet, d = doublet, t = triplet, q = quartet, m = multiplet), coupling constant(s), and number of protons. High-resolution mass spectra were obtained at the University of Pittsburgh Mass Spectrometry facility. LC-MS analysis was performed on an SHIMADZU instrument, using an analytical C18 column (Dionex Acclaim 120 Å, 2.1 × 50 mm, 3.0 μm, 0.2 mL min^{-1}).

Synthesis of (2-isocyanoethyl)benzene (2) according to a modified Hoffmann procedure: A 500 mL round-bottomed flask equipped with a mechanic stirring bar, a reflux condenser, and a pressure-equalizing dropping funnel was charged with 60 mL of water. Sodium hydroxide (60.0 g, 1.5 mol) was added portionwise. A mixture of 2-phenylethylamine (24.3 mL, 0.19 mol), chloroform (16.0 mL, 0.19 mol), and benzyltriethylammonium chloride (400 mg, 1.8 mmol) in dichloromethane (60 mL) was added dropwise over 10 min. The reaction mixture was refluxed for 4 h. After the reaction mixture was diluted with ice and water (50 mL), the organic layer was separated and retained, the aqueous layer was extracted with dichloromethane (2 × 50 mL). The combined organic layers were washed with cooled aqueous solution of hydrochloride (1 N, 3 × 100 mL) and brine (100 mL), and dried over anhydrous magnesium sulfate. The drying agent was removed by filtration. The filtrate was passed through a short column (Ø = 6 cm, *l* = 5 cm). After concentration, (2-isocyanoethyl)benzene (**2**) was afforded as yellowish oil (15.5 g, 61%). ^{1}H NMR (600 MHz, $CDCl_3$): δ = 7.38 (t, *J* = 7.5 Hz, 2H), 7.32 (t, *J* = 7.5 Hz, 1H), 7.27 (d, *J* = 7.8 Hz, 2H), 3.62 (tt, *J* = 1.8, 7.2 Hz, 2H), 3.00 ppm (tt, *J* = 1.8, 7.2 Hz, 2H); ^{13}C NMR (150 MHz, $CDCl_3$): δ = 156.57, 136.73, 128.83, 128.75, 127.29, 43.03, 35.66 ppm.

Synthesis of *N*-(2,2-dimethoxyethyl)-*N*-(2-oxo-2-(2-phenethylamino)-ethyl)cyclohexanecarboxamide (6) (Ugi reaction): To a mixture of paraformaldehyde (**3**) (3.33 g, 0.11 mol), 2,2-dimethoxyethylamine (**5**) (11.67 g, 0.11 mol), and cyclohexyl carboxylic acid (**4**) (14.22 g, 0.11 mol) in methanol (110 mL) (2-isocyanoethyl)benzene (**2**) (15.0 g, 0.11 mol) was added dropwise at 0°C. After stirred at room temperature for 48 h, the mixture was concentrated. The residue was dissolved in diethyl ether (150 mL) and washed with water (100 mL) and brine (100 mL), and dried over anhydrous magnesium sulfate. The drying agent was removed by filtration. After concentration, pale yellowish oil was obtained, which upon standing crystallizes to yield **6** (40.9 g, 98%). R_f = 0.33 (hexane/ethyl acetate, 1:2, v/v); ^{1}H NMR (600 MHz, $CDCl_3$): δ = 7.17–7.31 (m, 5H), 7.01 (br s, 0.5H), 6.50 (br s, 0.5H), 4.58 (dd, *J* = 4.8, 4.8 Hz, 0.5H), 4.39 (dd, *J* = 4.8, 4.8 Hz, 0.5H), 3.99 (s, 1H), 3.98 (s, 1H), 3.55 (dd, *J* = 6.6, 13.2 Hz, 1H), 3.48 (dd, *J* = 6.6, 13.2 Hz, 1H), 3.42 (t, *J* = 7.2 Hz, 2H), 3.37 (s, 3H), 3.33 (s, 3H), 2.82 (t, *J* = 7.2 Hz, 1H), 2.78 (t, *J* = 7.2 Hz, 1H), 2.60 (tt, *J* = 3.0, 11.4 Hz, 0.5H), 2.25 (tt, *J* = 3.0, 11.4 Hz, 0.5H), 1.59–1.78 (m, 5H), 1.44 (m, 2H), 1.23 ppm (m, 3H); ^{13}C NMR (150 MHz, $CDCl_3$): δ = 178.0, 177.8, 169.5, 169.2, 138.7, 138.5, 128.72, 128.6, 128.5, 126.6, 126.4, 103.4, 102.6, 55.4, 55.0, 54.04, 52.1, 51.4, 50.3, 41.0, 40.6, 40.4, 40.2, 35.6, 35.5, 29.3, 29.2, 25.7, 25.6, 25.5 ppm.

Synthesis of 2-(cyclohexanecarbonyl)-2,3,6,7,11b-hexahydro-pyrazino[2,1-a]isoquinolin-4-one (praziquantel, 1) (Pictet–Spengler reaction): *N*-(2,2-Dimethoxyethyl)-*N*-(2-oxo-2-(2-phenethylamino)ethyl)cyclohexanecarboxamide (**6**) (30.0 g, 79.8 mmol) was added portionwise to methanesulfonic acid (104.0 mL, 1.6 mol) at 0°C. After heating to 70°C for 6 h, the reaction mixture was poured into an ice–water mixture and adjusted to pH 8 with an aqueous solution of NaOH (20%). The solution was extracted with diethyl ether (4 × 100 mL). The combined organic layers were washed with brine (100 mL), dried and concentrated to afford **1** (19.0 g, 76%) as yellowish solid. The residue was recrystallized from ethyl acetate/hexane (1:1) to afford **1** (16.2 g, 65%) as a white solid. M.p. 132–134°C (lit.[16] 132–135°C, lit.[17] 132–133°C); ^{1}H NMR (600 MHz, $CDCl_3$): δ = 7.18–7.29 (m, 4H), 5.17 (dd, *J* = 3.0, 13.8 Hz, 1H), 4.81 (m, 2H), 4.47 (d, *J* = 17.4 Hz, 1H), 4.08 (d, *J* = 17.4 Hz, 1H), 2.78–2.99 (m, 4H), 2.47 (tt, *J* = 3.0, 12.0 Hz, 1H), 1.72–1.83 (m, 5H), 1.53–1.57 (m, 2H), 1.27–1.28 ppm (m, 3H); ^{13}C NMR (150 MHz, $CDCl_3$): δ = 174.7, 164.3, 134.7, 132.7, 129.2, 127.4, 126.9, 125.4, 54.9, 48.9, 45.1, 40.7, 39.0, 29.2, 28.9, 28.6, 25.7 ppm.

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Keywords: drug discovery · isocyanides · multicomponent reactions · Pictet–Spengler reaction · Ugi reaction

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