RESEARCH ARTICLE

Facile Synthesis of 5,6,7,8-tetrahydropyrimido[4,5-*d*] pyrimidine-2,4 (1*H*,3*H*) -dione Analogs via One Pot Multicomponent Reaction

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Abstract: Indeed, MCR technology is widely acknowledged now for its impact on drug discovery projects and is strongly supported by industry as well as academia. Uracil is an important one of the five nucleobases and significantly important because of their biological properties; of which 6-amino uracil is most important and can act as nucleophile and electrophile. 6-Aminouracils being rich are used as starting materials for the synthesis of heterocyclic compounds of biological significance such as pyrido-, pyrazolo, pyrimido and pyrimidines derivatives. 5,6,7,8-tetrahydropyrimido[4,5-d]pyrimidine-2,4(1H,3H)-dione have been synthesized by various conventional methods. However, these methods have drawbacks such as unsatisfactory yields and tedious work-up etc. In the present work, we would like to report a new route for the synthesis of 5,6,7,8-tetrahydropyrimido[4,5-d]pyrimidine-2,4 (1H,3H)-dione in MCRs.

Keywords: Multi Component Reaction (MCR), Green Synthesis, 6-Amino Uracil

Introduction

To construct various heterocyclic scaffolds, a number of advancements has introduced in implementation and design of multi component reactions (MCRs), as interest in preparation of diversity-oriented compounds has increased which cause an increase in consideration of this paradigm to synthesize a library of compounds [1].

MCRs that are easy to handle, can also be used to assemble the complex molecules in inert atmosphere without using dry conditions. In MCRs non-linear approach and convergent way is used and in one step all the moieties that determine the properties are introduced except sequentially. Thus, MCRs can be used to quickly generate the SARs (structure-activity relationships [2-8]. Any form of compounds can be synthesize with their high bond-forming efficacy and atom economy, unusual waste formation and the purification and separation of products are economical while using MCRs [9]. MCRs are the masterworks of reaction designs and synthetic efficiency [10, 11].

Many interesting pharmacological activities are exhibited by a variety of derivatives of pyrimidine [12-19]. A number of biological significant compounds were used to regulate the growth of plants in their protection area [20]. These studies revealed the introduction of expedient methods to synthesize pyrimido[4,5-*d*]pyrimidine-2,4-dione [21]. Now a days, pyrimido-pyrimidines (annulated uracil) is a very interesting class of compounds as they display a large

number of pharmacological activities with their effective inhibitory characteristics about tyrosine kinase area for the receptors of epidermal growth [22], dihydrofolate-reductase [23] and 5-phosphoribosyl-1-pyrophosphate synthetase [24]. A number of reports has given about their antiviral [25], antioxidant [26], antitumor [27], hepatoprotective and antifungal activities [28].

Uracil is an important constituent of nucleic acid and five nucleobases are a significant class of compound [29] due to their biological characteristics, from this class 6-amono uracil is an important compound that can act as an electrophile as well as nucleophile [30]. This compound being rich is used to synthesize biologically significant heterocyclic compounds as starting reagent such as pyrazolo, pyrimidine, pyrimido [31] and pyrido- derivatives [32]. 5,6,7,8-tetrahydropyrimido[4,5-*d*]pyrimidine-2,4(1*H*,3*H*)-dione have been synthesized by traditional methods. Thus there is a strong need to develop some better techniques to synthesize 5,6,7,8-tetrahydro pyrimido [4,5-*d*]pyrimidine-2,4(1*H*,3*H*)-dione that must be simple, economical, can give high yield in a short time. This method provides a new way to synthesize 5,6,7,8-tetrahydropyrimido[4,5-*d*]pyrimidine-2,4(1*H*,3*H*)-dione in MCRs.

Experimental

First of all, 0.39 mmol of 6-amino-uracil was taken, then dissolved it in H_2O and THF (1:1) proceeded this reaction on ultrasonic bath. Then in a separate beaker took formaldehyde (29 μl) and aniline in 2:1 ratio. Added this prepared mixture into the vial drop by drop and sonicated it on ultrasonic bath for 3 hours by monitoring it with TLC. Then the reaction mixture was cooled at room temperature to obtain the final product in vial. To get the pure crystals of the product, washed it by using 50% ethanol.

4a) 6-phenyl-5,6,7,8-tetrahydropyrimido[4,5-d] pyrimidine-2,4(1H,3H)-dione

IR: 3348(N-H), 1713(C=O), 1628(C=O), 1593(C=C), 1396 (C-H), 1230(C-N), 753 (benzene).1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.13 (s, 1H, NH), 9.97 (s, 1H, NH), 6.93-7.03 (m, 5H, ArH), 6.26 (s, 1H, NH), 5.13 (s, 2H, CH2), 4.06 (s, 2H, CH2). 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 149.6, 129.6, 121.9, 114.3, 83.3, 71.6, 52.1 Elemental Analysis: $C_{12}H_{12}N_4O_2$ C, 59.01; H, 4.95; N, 22.94. Found: C, 59.31; H, 4.99; N, 22.99.

4b) 6-(4-chlorophenyl)-5,6,7,8-tetrahydropyrimido [4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3320(N-H), 1710(C=O), 1628(C=O), 1591(C=C), 1388(C-H), 1020-1250 (C-N), 755 (benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.27 (s, 1H, NH), 10.05 (s, 1H, NH), 7.23 (d, 2H, ArH, J = 6.5 Hz), 7.11 (d, 2H, ArH, J = 6.5 Hz), 6.65 (s, 1H, NH), 5.17 (s, 2H, CH2), 4.12 (s, 2H, CH2). 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 147.7, 129.7, 127.2, 115.7, 83.3, 71.6, 52.1. Elemental Analysis: $C_{12}H_{11}ClN_4O_2$ C, 51.72; H, 3.98; Cl, 12.72; N, 20.10. Found: C, 51.91; H, 4.25; Cl, 12.86; N, 20.25

4c) 6-(3-chlorophenyl)-5,6,7,8-tetrahydropyrimido [4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3328(N-H), 1720(C=O), 1637(C=O), 1591(C=C), 1389(C-H), 1020(C-O), 1090, 1160(C-N), 755(benzene).1H-NMR (δ in ppm): (300 MHz, DMSO-d6) δ: 10.19 (s, 1H, NH), 10.11 (s, 1H, NH), 7.37 (s, 1H, ArH), 7.09 (m, 3H, ArH), 6.78 (s, 1H, NH), 5.23 (s, 2H, CH2), 4.35 (s, 2H, CH2). 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 151.0, 135.2, 131.0, 121.8, 114.7, 112.4, 83.3, 71.6, 52.1 Elemental Analysis: $C_{12}H_{11}ClN_4O_2$ C, 51.72; H, 3.98; Cl, 12.72; N, 20.10. Found: C, 51.78; H, 3.99; Cl, 12.75; N, 20.18.

4d) 6-(2-chlorophenyl)-5,6,7,8-tetrahydropyrimido [4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3350(N-H), 1712(C=O), 1630(C=O), 1590(C=C), 1388, 1450(N-O), 1299(C-H), 1018, 1112(C-N), 757(benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d6) δ: 10.34 (s, 1H, NH), 10.09 (s, 1H, NH), 6.97-7.37 (m, 4H, ArH), 6.29 (s, 1H, NH), 5.33 (s, 2H, CH2), 4.46 (s, 2H, CH2) 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 150.8, 130.8, 129.0, 127.7, 124.9, 123.7, 83.3, 71.1, 51.6 Elemental Analysis: $C_{12}H_{11}ClN_4O_2$ C, 51.72; H, 3.98; Cl, 12.72; N, 20.10; Found: C, 51.85; H, 3.99; Cl, 12.84; N, 20.24.

4e) 6-(4-nitrophenyl)-5,6,7,8-tetrahydropyrimido[4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3339(N-H), 1709(C=O), 1630(C=O), 1595(C=C), 1389, 1440(N-O), 1299(C-N), 1018-1115(C-N), 755and 885(benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.39 (s, 1H, NH), 10.18 (s, 1H, NH), 8.33 (d, 2H, ArH, J = 6.2 Hz), 8.14 (d, 2H, ArH, J = 6.2 Hz), 6.69 (s, 1H, NH), 5.21 (s, 2H, CH2), 4.19 (s, 2H, CH2). 13C-NMR (δ in ppm): 163.7, 162.4, 155.7, 154.8, 137.4, 124.8, 112.3, 83.3, 71.6, 52.1 Elemental Analysis: $C_{12}H_{11}N_5O_4$ C, 49.83; H, 3.83; N, 24.21; Found: C, 49.87; H, 3.91; N, 24.43.

4f) 6-(3-nitrophenyl)-5,6,7,8-tetrahydropyrimido[4,5-d] pyrimidine-2,4(1H,3H)-dione

IR: 3335(N-H), 1715(C=O), 1621(C=O), 1594(C=C), 1389(N-O), 1019 and 1093(C-N), 755 (benzene).1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.25 (s, 1H, NH), 10.16 (s, 1H, NH), 8.46 (s, 1H, ArH), 8.17-8.03 (m, 3H, ArH), 6.83 (s, 1H, NH), 5.31 (s, 2H, CH2), 4.37 (s, 2H, CH2). 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 150.5, 148.8, 130.5, 120.4, 113.4, 107.2, 83.8, 71.6, 52.1 Elemental Analysis: $C_{12}H_{11}N_5O_4$ C, 49.83; H, 3.83; N, 24.21; Found: C, 49.85; H, 3.85; N, 24.31.

4g) 6-(2-nitrophenyl)-5,6,7,8-tetrahydropyrimido[4,5-d] pyrimidine-2,4(1H,3H)-dione

IR: 3335(N-H), 1715(C=O), 1621(C=O), 1594(C=C), 1389(N-O), 1019 and 1093(C-N), 755(benzene).1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.47 (s, 1H, NH), 10.13 (s, 1H, NH), 8.05-7.37 (m, 4H, ArH), 6.32 (s, 1H, NH), 5.41 (s, 2H, CH2), 5.16 (s, 2H, CH2) 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 143.3, 140.0, 135.1, 126.0, 119.0, 110.7, 83.3, 70.6, 51.1 Elemental Analysis: $C_{12}H_{11}N_5O_4$ C, 49.83; H, 3.83; N, 24.21; Found: C, 49.91; H, 3.92; N, 24.35;

4h) 6-(2-methyl-5-nitrophenyl)-5,6,7,8-tetrahydro pyrimido[4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3350(N-H), 1712(C=O), 1630(C=O), 1590(C=C), 1388, 1450(N-O), 1299(C-H), 1018, 1112(C-N), 757(benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d6) δ: 10.29 (s, 1H, NH), 10.17 (s, 1H, NH), 8.42 (s, 1H, ArH), 8.25 (d, 1H, ArH, , J = 6.3 Hz), 7.55 (d, 1H, ArH, , J = 6.3 Hz), 6.68 (s, 1H, NH), 5.29 (s, 2H, CH2), 4.43 (s, 2H, CH2), 2.21 (s, 3H, CH3). 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 148.2, 145.8, 138.9, 132.6, 124.7, 107.1, 83.3, 71.9, 52.4, 17.9 Elemental Analysis: $C_{13}H_{13}N_5O_4$ C, 51.48; H, 4.32; N, 23.09; C, 51.52; H, 4.42; N, 23.15.

4i) 6-(2,4-dichlorophenyl)-5,6,7,8-tetrahydro pyrimido[4,5-d]pyrimidine-2,4(1H,3H)-dione

IR: 3340(N-H), 1713(C=O), 1620(C=O), 1592(C=C), 1388(C-H), 1018 and 1092(C-N), 755(benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 10.55 (s, 1H, NH), 10.29 (s, 1H, NH), 7.98 (s, 1H, ArH), 7.65 (d, 1H, ArH, , J = 6.5 Hz), 7.52 (d, 1H, ArH, , J = 6.5 Hz), 6.37 (s, 1H, NH), 5.34 (s, 2H, CH_2), 4.76 (s, 2H, CH2) 13C-NMR (δ in ppm): 163.7, 162.4, 154.8, 148.9, 131.3, 127.8, 125.2, 124.6, 117.1, 83.3, 71.1, 51.6 Elemental Analysis: $C_{12}H_{10}Cl_2N_4O_2$ C, 46.03; H, 3.22; Cl, 22.64; N, 17.89; Found: C, 46.14; H, 3.32; Cl, 22.71; N, 17.92.

4j) Ethyl 4-(5,7-dioxo-1,4,5,6,7,8-hexahydropyrimido [4,5-d]pyrimidin-3(2H)-yl)benzoate

IR: 3348 (N-H), 1713(C=O), 1613(C=O), 1594(C=C), 1389(C-H), 1018 and 1093(C-N), 755(benzene). 1H-NMR (δ in ppm): (300 MHz, DMSO-d_6) $_\delta$: 10.46 (s, 1H, NH), 10.19 (s, 1H, NH), 8.2 (d, 2H, Ar-H, J = 6.4 Hz), 7.47 (d, 2H, Ar-H, J = 6.4 Hz), 6.71 (s, 1H, NH), 5.53 (s, 2H, CH_2), 4.18 (s, 2H, CH_2), 4.07 (q, 2H, CH_2), 2.12 (t, 3H, CH_3) ^{13}C-NMR (δ in ppm): 165.9, 163.7, 162.4, 154.8, 153.9, 130.8, 119.6, 111.7, 83.3, 71.6, 60.9, 52.1, 14.1 Elemental Analysis $C_{15}H_{16}N_4O_4$
C, 56.96; H, 5.10; N, 17.71; Found: C, 56.99; H, 5.17; N, 17.74.

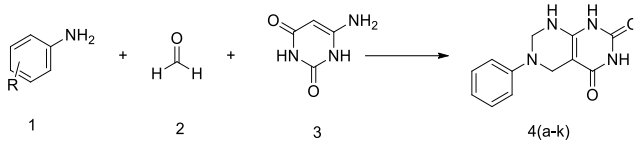
4k) 3-(5,7-dioxo-1,4,5,6,7,8-hexahydropyrimido[4,5-d] pyrimidin-3(2H)-yl)benzoic acid
IR: 3345(N-H), 1720(C=O), 1610(C=O), 1596(C=C), 1389(C-H), 1019 and 1100(C-N), 755(benzene). ^{1}H-NMR (δ in ppm): (300 MHz, DMSO-d_6) δ: 11.35 (s, 1H, OH), 10.27 (s, 1H, NH), 10.05 (s, 1H, NH), 7.40 (s, 1H, Ar-H), 7.17 (m, 3H, Ar-H), 6.73 (s, 1H, NH), 5.34 (s, 2H, CH2), 4.37 (s, 2H, CH2) ^{13}C-NMR (δ in ppm): 166.3, 163.7, 162.4, 154.8, 149.5, 129.0, 119.8, 119.5, 112.6, 83.3, 71.6, 52.1 Elemental Analysis: $C_{13}H_{12}N_4O_4$ C, 54.17; H, 4.20; N, 19.44; Found: C, 54.19; H, 4.28; N, 19.47.

Result and Discussion

By using this method a great variety of 6-amino-uracil derivatives were obtained via following reactions. This one pot, catalyst free reaction of formaldehyde with a variety of anilines yield 6-aminouracil derivatives containing different substituents with altered positions is generally given bellow. The product obtained then react with 6-aminouracil giving the derivatives of 6-aminouracil (4a-k).

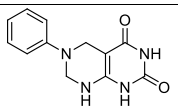
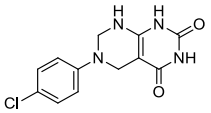
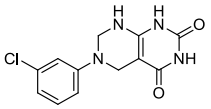
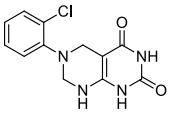
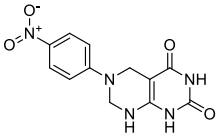
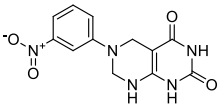
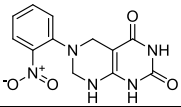
The compounds synthesized indicate N-H stretching of secondary amine in region 3350-3320cm^{-1} and two C=O stretching in region 1600-1750cm^{-1} as each compound contains two carbonyl functional groups.

Reaction:



R= H, 4-Cl, 3-Cl, 2-Cl, 4-NO_2, 3-NO_2, 2-NO_2, 2-CH_3, 5-NO_2, 2,4-di-Cl, 4- $COOCH_2CH_3$, 3-COOH
Scheme 1: synthesis of 6-aminouracil derivatives using MCRs

Table 1: Physical Parameters of Synthesized Compounds

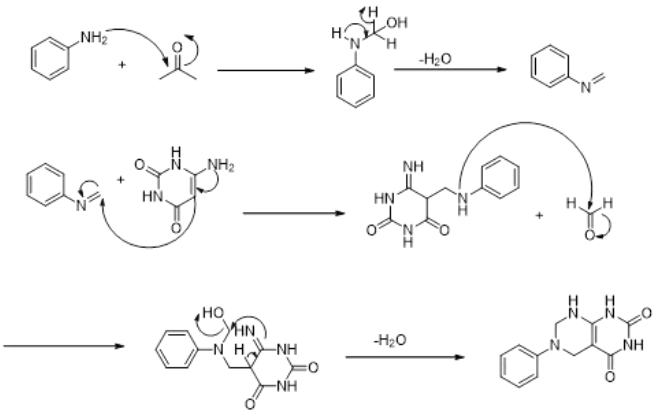
Serial No.	Compound Code	Structure of Compound	% age yield	Melting point (C^o)
1	4a		78%	240ºC
2	4b		75%	250ºC color change.
3	4c		74%	>265ºC
4	4d		79%	>255ºC slight change in color.
5	4e		74%	>270ºC
6	4f		75%	>265ºC
7	4g		73%	>250º C

These compounds also indicate the presence of aromatic ring stretching of C=C in region 1500-1650cm^{-1}, the stretching in the region of 1000-1250cm^{-1} is due to C-N in amine and the stretching in the region of 1300-1400cm^{-1} is due to C-H groups. The stretching in the region of 700-900cm^{-1} is due to aromatic ring

system. Few compounds that are synthesized showed strong bands in the region of 1330-1365cm^{-1} due to the presence of NO_2 group. According to proton NMR spectrum **(4a)** CH group showed one singlet peak at δ 4.06 ppm, NH groups showed three singlet peaks at δ 10.13, 9.97 and 6.26 respectively and a multiplet due to benzene ring. 10 separate resonances further confirmed this structure by ^{1}H-decoupled ^{13}C NMR spectrum **(4a)**.

Mechanism of reaction

Mechanism of this reaction is given bellow which explain every step of this reaction and how the product is generated by this reaction.



Scheme 1: Proposed mechanism for synthesis of amino-uracil derivatives

Conclusions

In this method we overcome the drawbacks of poor yields and reaction time. This method provides a new way to synthesize 5,6,7,8-tetrahydropyrimido[4,5-d] pyrimidine-2,4(1H,3H)-dione in MCRs which is simple, economical and give high yield in a short time. In future the synthesized compounds will be used in pharmacological and pharmaceutical applications.

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