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Synthesis of Pteridine Derivatives as Anti-TB Agents

Sweta Desai[1*], Zarna Dedania[2], and Naishadh Solanki[3]

[1*]Research Scholar, Department of Pharmaceutical Sciences, Bhagwan Mahavir Centre for Advance research, Bhagwan Mahavir University, Sr. No. 149, VIP Road, Bharthana Road, Vesu, Surat, Gujarat 395007.

[2]Professor and HOD, Assistant Professor, Department of Quality Assurance, Bhagwan Mahavir Centre for Advance research, Bhagwan Mahavir University, Sr. No. 149, VIP Road, Bharthana Road, Vesu, Surat, Gujarat 395007.

[3]Assistant Professor, Department of Chemistry, Bhagwan Mahavir Centre for Advance research, Bhagwan Mahavir University, Sr. No. 149, VIP Road, Bharthana Road, Vesu, Surat, Gujarat 395007.

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***Corresponding Author Email:** 13swets@gmail.com

Abstract

Tuberculosis (TB) is a major global health issue with a high mortality rate, particularly due to the emergence of drug-resistant strains of Mycobacterium tuberculosis. Hence, the development of novel anti-TB agents is of great significance. In this regard, pteridine derivatives have emerged as a promising scaffold for the development of novel drugs. This study focuses on the synthesis of N-phenyl-6,7-dip-tolylpteridin-4-amine derivatives, and their evaluation for anti-TB activity. The three-step synthesis process involved the preparation of 6,7-dip-tolylpteridin-4-ol from 4,5-diamino-6-hydroxypyrimidine and 4,4-dimethylbenzil, followed by its reaction with phosphoryl chloride and dimethylformamide to yield 4-chloro-6,7-dip-tolylpteridine, and finally, the reaction of the latter with aniline in dimethylformamide to obtain N-phenyl-6,7-dip-tolylpteridin-4-amine derivatives. The synthesized compounds were evaluated for their anti-TB activity against standard drugs streptomycin, ciprofloxacin, and pyrazinamide. The results showed that all the compounds demonstrated significant anti-TB activity, with some displaying activity comparable to the reference drugs. Specifically, compounds E6, E8, E13, and E17 showed activity like that of streptomycin and ciprofloxacin, while compounds E3, E25, E9, E14, E19, and E4 demonstrated activity similar to pyrazinamide. In conclusion, the study highlights the potential of pteridine derivatives as promising anti-TB agents. The synthesized compounds exhibited significant anti-TB activity and could serve as an alternative to current standard therapies.

Keywords

Pteridine, Anti-Tb, Mycobacterium tuberculosis.

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INTRODUCTION

Tuberculosis, also known as TB, is one of the first diseases that have been documented to afflict people. It is also one of the leading causes of death on a global scale. The bacteria that cause TB are part of the Mycobacterium tuberculosis complex (1). Tuberculosis (TB) is a bacterial infection that most commonly affects the lungs but can also spread to other regions of the body, including the brain, spine, and kidneys. Mycobacterium tuberculosis, the bacterium responsible for TB, is transmitted from person to person through the air via coughing,

speaking, and sneezing. Symptoms of tuberculosis include weariness, weight loss, night sweats, and coughing (2). However, some TB sufferers may show no signs at all. Chest X-rays, sputum testing, and skin testing are just a few of the diagnostic options available for tuberculosis. Antibiotics are effective against tuberculosis, but therapy typically takes many medications over the course of several months to completely eradicate the bacterium. Drug-resistant TB strains are far more difficult to cure, and they might emerge if a patient does not finish the full course of treatment (3). Screening at-risk populations, detecting and treating those with active TB, and treating those with latent TB infection with preventive medication are all important steps in limiting the disease's spread. In addition, the Bacilli Calmette-Guerin (BCG) vaccination is used to protect children from contracting TB meningitis and other serious forms of the disease in some countries. According to the World Health Organization (WHO), tuberculosis is the second leading cause of death among humans after HIV/AIDS (4). This is even though tuberculosis continues to be a major threat to the human population. There is a disproportionately high incidence of tuberculosis in populations with lower socioeconomic status as well as in more marginalized parts of the community. One of the national goals outlined in India's National strategy plan (2017–2025) is the complete eradication of tuberculosis by the year 2025 (5).

Current treatment of TB

The most common way to treat tuberculosis (TB) is with a combination of antibiotics taken over a few months. For TB that can be treated with drugs, the standard treatment plan includes four drugs: isoniazid, rifampin, ethambutol, and pyrazinamide (6). The length of treatment can change based on how bad the disease is, but it usually lasts between six and nine months. If the disease is very bad or if the patient has a weak immune system, the treatment may take longer. Also, drug-resistant TB needs a different treatment plan and may need to be treated with several antibiotics for at least two years. Drug-resistant tuberculosis (TB) is caused by bacteria that are resistant to one or more of the drugs commonly used to treat TB (7). Drug-resistant tuberculosis is frequently more difficult to treat and necessitates a longer course of treatment than drug-susceptible tuberculosis. Drug-resistant tuberculosis is treated with a combination of several antibiotics over an extended period, typically 18 to 24 months or longer. The type and extent of drug resistance, as well as other factors such as the patient's overall health and the presence of other medical conditions, influence the drugs used and the duration of treatment (8). A combination of drugs such as bed aquiline, linezolid, and delamanid, which are newer drugs that have been shown to be effective against drug-resistant TB, is the standard treatment for drug-resistant TB (9). Surgery may be required in some cases to remove infected lung tissue. Treatment for drug-resistant tuberculosis can be difficult and requires close supervision by a healthcare provider. Even if patients begin to feel better, it is critical that they take their medications exactly as prescribed and complete the entire course of treatment. This helps to prevent the development of additional drug resistance and ensures the best possible outcome.

Recent development of drugs to treatment TB

Although progress has been gradual and problems persist, there have been some recent advancements in the discovery of new medications to treat tuberculosis. Bed aquiline, which was authorized by the US Food and Drug Administration (FDA) in 2012 for the treatment of drug-resistant TB, is one potential new medication (10). Bedaquiline functions by preventing the ATP synthase enzyme, which is necessary for the TB bacterium's energy metabolism. Bedaquiline has a lower rate of treatment failure than conventional treatment regimens, and clinical trials have demonstrated that it is successful in treating drug-resistant TB. Delamanid is a different brand-new medication that the FDA licensed in 2014 for the treatment of multidrug-resistant TB. Delamanid functions by preventing the production of mycolic acid, a crucial element of the TB cell wall. Delamanid is successful in treating multidrug-resistant TB, according to clinical trials, while its long-term safety and efficacy are still being examined (11). Pretomanid, which the FDA approved in 2019 for use in a new three-drug regimen for the treatment of severely drug-resistant TB, and clofazimine, which is being studied for use in combination therapy for TB, are other medications being developed for the treatment of TB (12). The complexity of the TB bacterium, the requirement for new therapeutic targets, the high cost and the protracted timetable of drug development are only a few of the important obstacles that must still be overcome in order to create new treatments for TB. Progress in this area has also been hampered by a lack of funding for TB research and development, notably from the commercial sector.

Need to discover other molecules to treat TB

There are various reasons why we need to create new TB drugs:

Drug resistance:

TB is known for developing drug resistance, and current treatments are losing effectiveness against the organism. The creation of novel compounds is critical for combating drug-resistant strains of tuberculosis and providing alternatives for individuals who do not react to present treatments (13).

Treatment time should be reduced:

The present TB treatment regimen lasts six to nine months, which can be taxing on patients and healthcare systems. The creation of novel compounds may result in shorter treatment durations, improving patient adherence and reducing the spread of tuberculosis.

Fewer adverse effects: Existing TB medicines, such as liver damage, can have considerable side effects, limiting their use. New compounds with fewer side effects might improve patient outcomes and quality of life.

New targets:

The discovery of new compounds enables the investigation of novel targets and mechanisms of action against the tuberculosis bacterium. This could result in the development of more effective medications with improved efficacy and safety profiles.

worldwide impact:

Tuberculosis (TB) is a serious worldwide health issue, particularly in low- and middle-income nations (14). The creation of novel compounds may assist to lower the burden of tuberculosis by making more effective and accessible medicines available to individuals in need.

Overall, developing novel compounds for the treatment of tuberculosis (TB) is critical for tackling the challenges posed by drug resistance, improving treatment results, and lowering the global burden of this lethal infectious disease.

Pterdine –scaffold motif

The pteridine scaffold motif is a heterocyclic compound that is widely distributed in nature and has been found to play important roles in various biological processes. Pteridines are characterized by a fused pyrimidine and pyrazine ring system, with a third nitrogen atom at position 7. In drug discovery, the pteridine scaffold motif has been utilized as a starting point for the development of a variety of pharmacologically active compounds, including antifolates, antitumor agents, antiviral agents, and antiparasitic agents (15). The antifolate class of drugs, in particular, is well-known for its use in the treatment of cancer and infectious diseases, such as malaria and bacterial infections (16). The pteridine scaffold has several advantages in drug discovery, including its structural diversity and ability to interact with multiple biological targets. Additionally, the pteridine scaffold has a relatively low toxicity and is metabolically stable, making it an attractive starting point for drug development (17). Overall, the pteridine scaffold motif is a versatile and valuable tool in drug discovery, and its use in the development of new drugs for the treatment of various diseases, including tuberculosis, is an active area of research. One of the most well-known therapeutic uses of the pteridine motif is in the development of antifolate drugs. These drugs are analogs of folic acid, a vitamin that is essential for DNA synthesis and cell division. Antifolate drugs work by inhibiting the enzyme dihydrofolate reductase (DHFR), which is involved in the synthesis of tetrahydrofolate, a form of folic acid that is needed for DNA synthesis. By inhibiting DHFR, antifolate drugs block the production of DNA, leading to the death of rapidly dividing cells, such as cancer cells or infectious agents like bacteria and parasites (18). Another therapeutic use of the pteridine motif is in the treatment of phenylketonuria (PKU), a metabolic disorder that results from a deficiency of the enzyme phenylalanine hydroxylase (PAH) (19). PAH is involved in the metabolism of the amino acid phenylalanine, which can accumulate in the blood and lead to severe neurological damage if left untreated. BH4 (tetrahydrobiopterin), a pteridine derivative, is used in the treatment of PKU to provide a cofactor that helps to activate PAH and improve the metabolism of phenylalanine. Pteridine derivatives have also been explored for their potential as anticancer agents, due to their ability to inhibit the activity of the enzyme thymidylate synthase, which is involved in DNA synthesis (20). In addition, some pteridine derivatives have been shown to have anti-inflammatory and antioxidant properties, which may be useful in the treatment of various chronic diseases (20). Overall, the pteridine motif is a versatile and valuable tool in drug development, and its therapeutic use is an active area of research.

MATERIAL AND METHODS

Chemicals were bought from companies including Spectrochem, Sigma-Aldrich, Loba Chem, Central Drug House (CDH) Ltd, and Merck India. The majority of the chemicals and solvents utilized were of LR grade. By employing precoated TLC plates, solvent systems of Benzene: Acetone (9:1), (8:2), T: E: F (5:4:1), and Chloroform: Methanol (9:1), thin layer chromatography was used to verify the compounds' purity. Under an ultraviolet lamp, the specks were seen. The uncorrected melting points were found in one-end open capillary tubes on a liquid paraffin bath. On a Perkin Elmer IR 4000-400 (v_{max} in cm^{-1})

Spectrophotometer in KBr Pellets and a Bruker Model Avance II 400 (400 MHz, 1H NMR) instrument, respectively, the compounds' IR and 1H nuclear magnetic resonance (1H NMR) spectra were captured. Tetramethylsilane (TMS) is used as an internal reference, and chemical changes are reported in parts per million (ppm).

Reaction Scheme

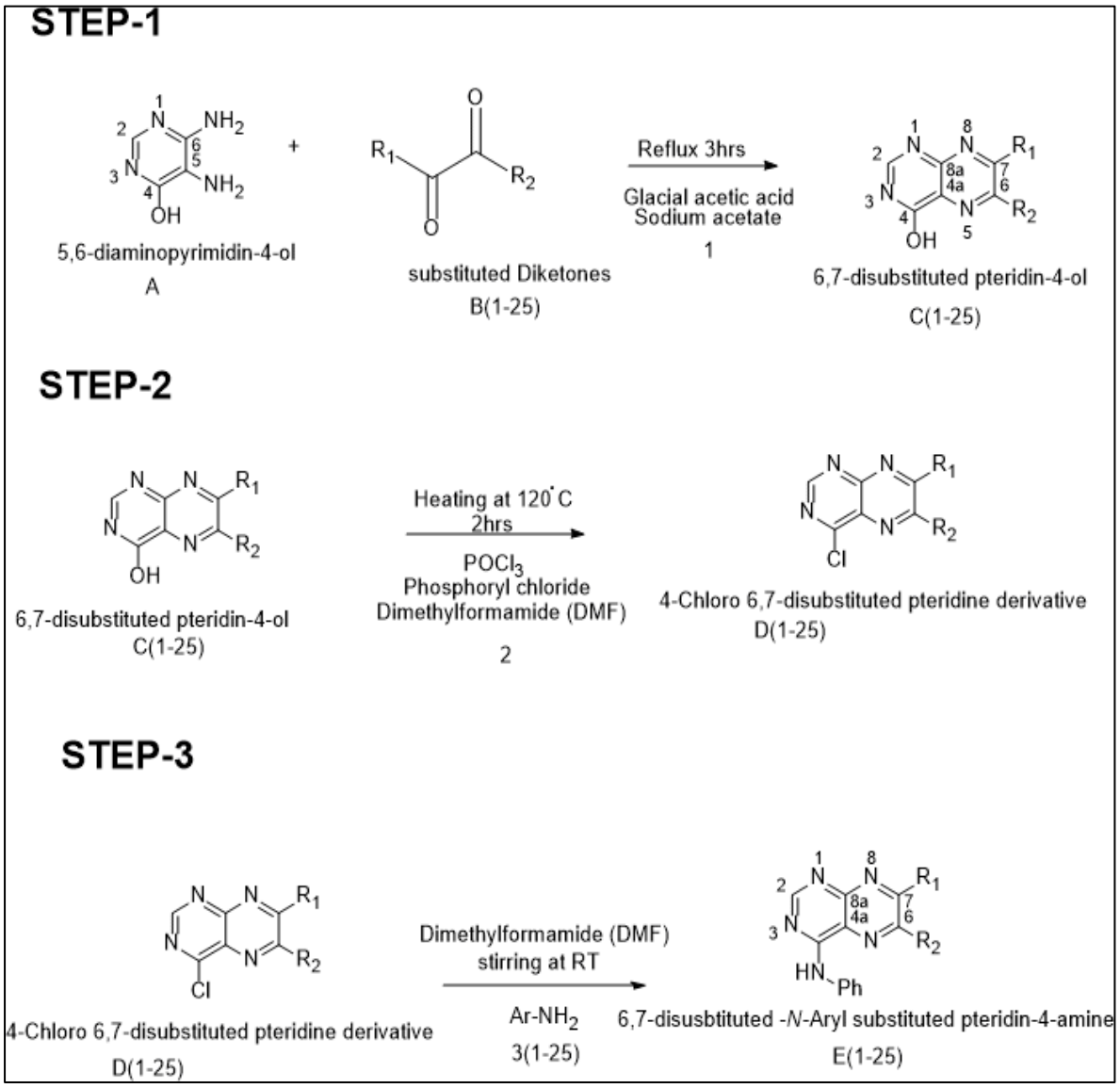

Figure -1: Reaction scheme for synthesis of substituted pteridine amine derivative E1-E25

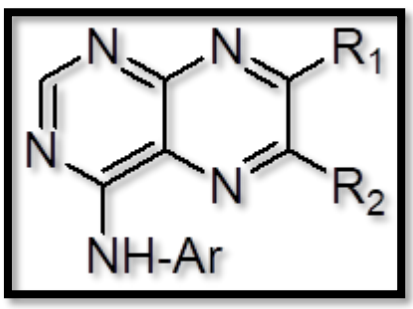

Figure-2: Final Structure of substituted pterdine molecules

Table 1- substitution of derivatives 1-25

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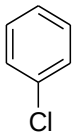
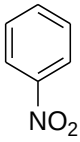
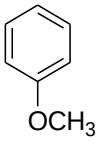
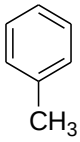
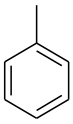
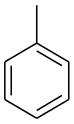
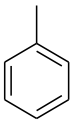
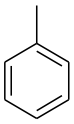
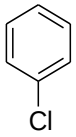
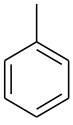
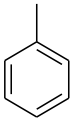
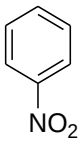
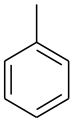
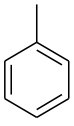
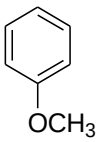
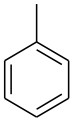
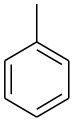
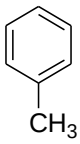
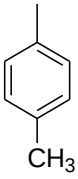
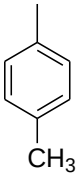
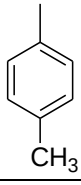
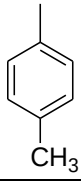
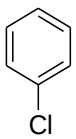
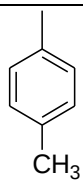
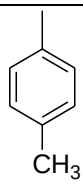
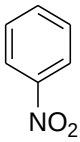
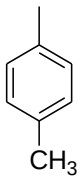
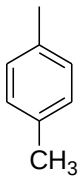
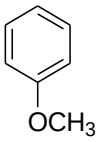
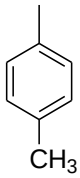
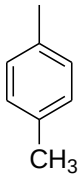
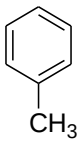
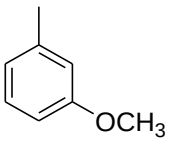
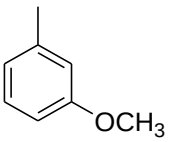
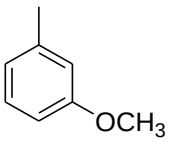
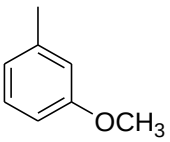
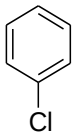
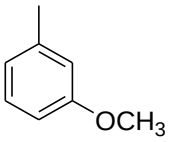
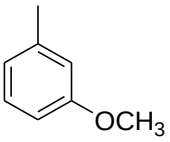
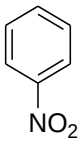
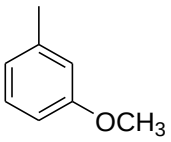
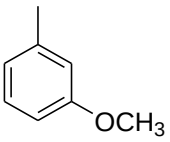
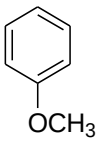
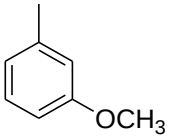
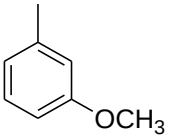
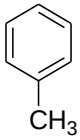
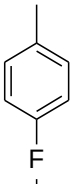
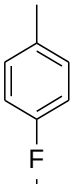
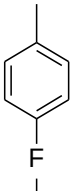
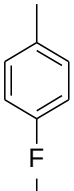
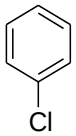
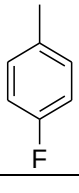
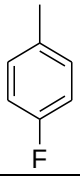
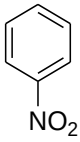
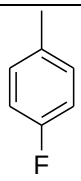
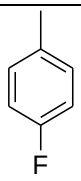
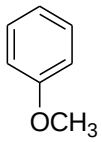
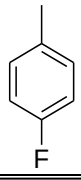
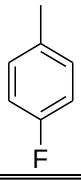
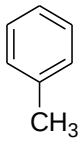
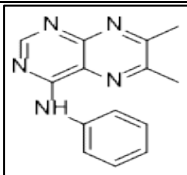
2	$-CH_3$	$-CH_3$	
3	$-CH_3$	$-CH_3$	
4	$-CH_3$	$-CH_3$	
5	$-CH_3$	$-CH_3$	
6			
7			
8			
9			
10			
11			
12			

13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			

24			
25			



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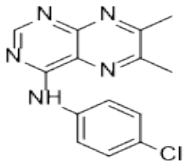

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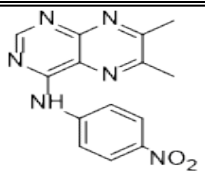

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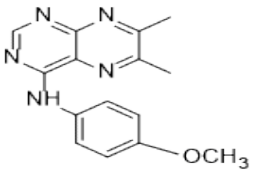

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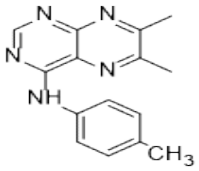

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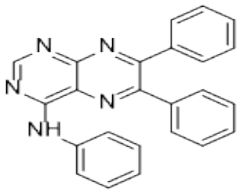

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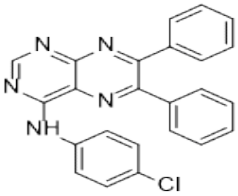

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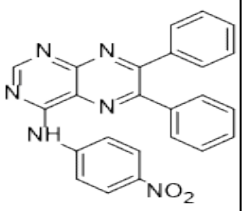

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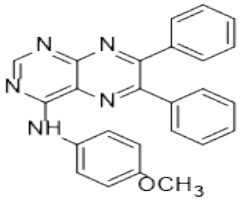

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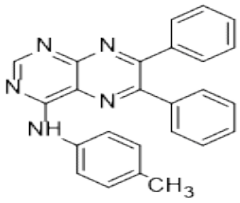

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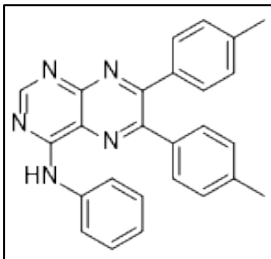

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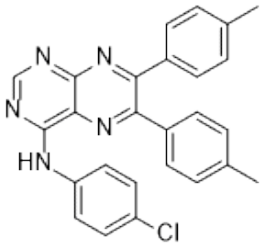

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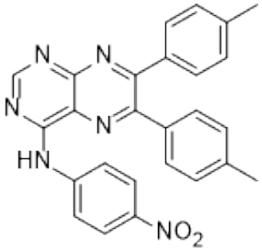

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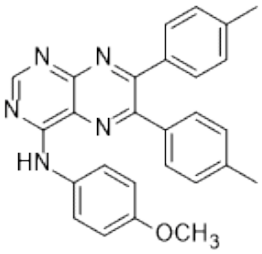

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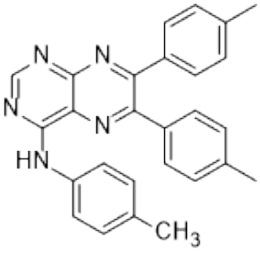

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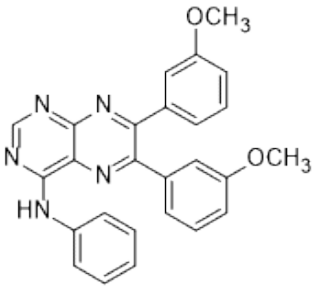

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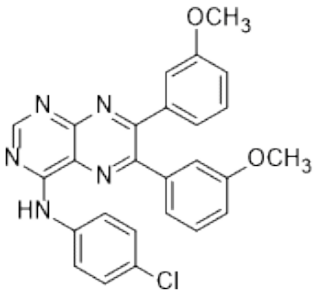

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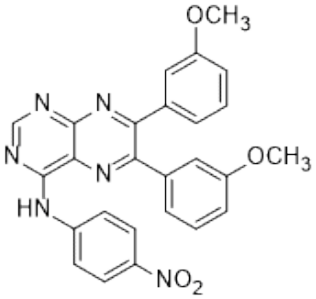

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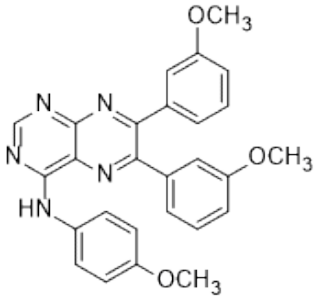

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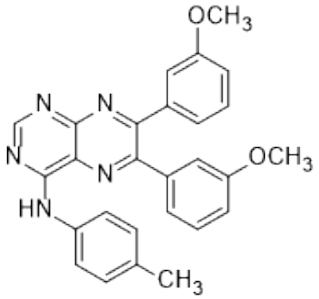

E20

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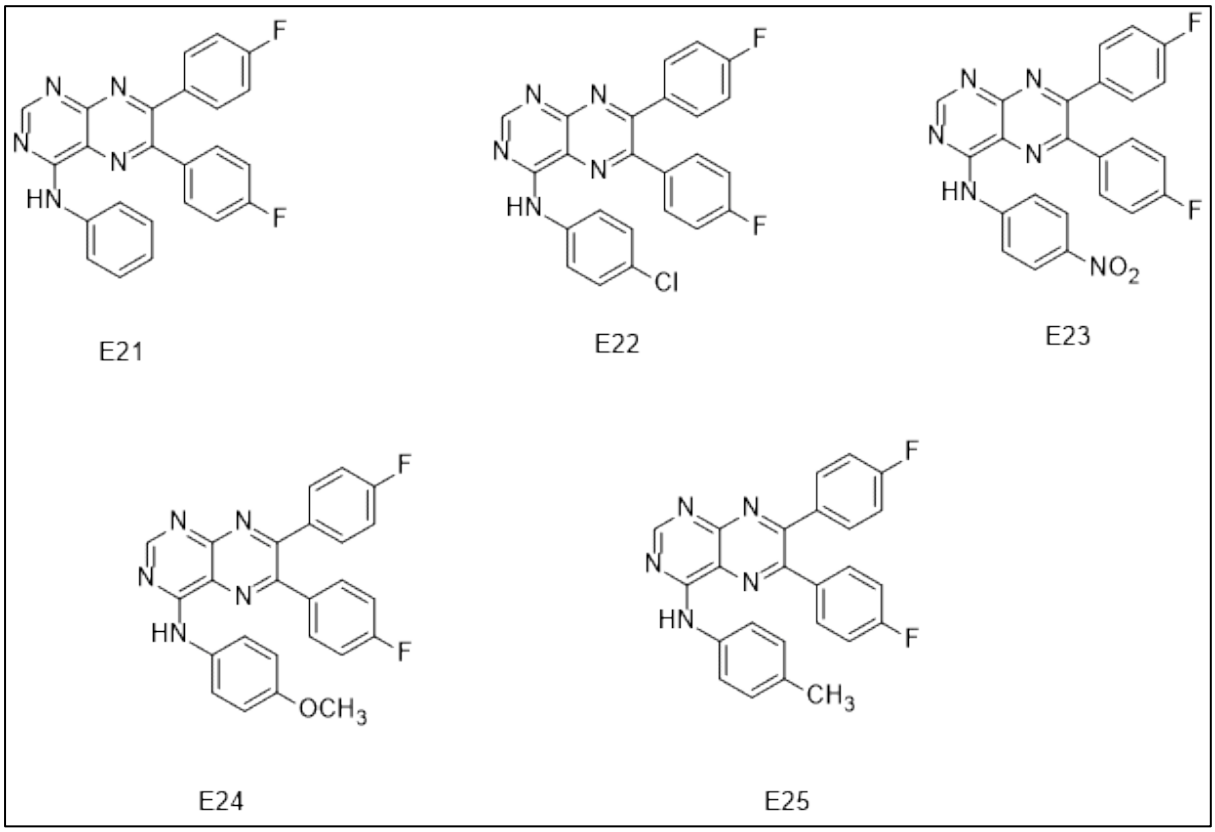


Figure-3: structures of final derivatives E1-E25

Synthesis of compound-E1

Step-1

Synthesis of 6,7-dimethylpteridin-4-ol

A mixture of 4, 5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with butane2,3-dione (0.76 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 3 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 54.32%; mp: 196-198 ˚C; $R_{f:}$ 0.41 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_8H_8N_4O$.

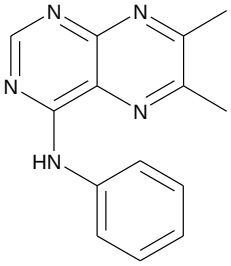
Step-2

Synthesis of 4-chloro-6,7-dimethylpteridine

A compound of 6,7-dimethylpteridin-4-ol (0.44 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1.5 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 58.23.12%; mp: 212-214 ˚C; $R_{f:}$ 0.37 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_8H_7ClN_4$.

Step-3

Synthesis of compound-1: 6,7-dimethyl-N-phenylpteridin-4-amine



4-chloro-6,7-dimethyl pteridine (0.0025mol, 0.485 g) was taken. Dimethylformamide (DMF) and aniline (0.0025mol, 0.232 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{14}H_{13}N_5$, White Solid, Yield: 61.58%; mp: 212-214 ˚C; $R_{f:}$ 0.34 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1516 (C=N) 1585 (C=C), 3038 (C–H aromatic), 3382 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.19 (s, 6H, -CH3), 4.0 (s, 1H, D2O exchangeable -NH,), 6.62-7.15 (m, 5H, Ar-CH), 8.1 (1H,s, Ar-CH) MS (EI) m/z: 252.15 (M + 1).

Synthesis of compound-E2

Step-1

Synthesis of 6,7-dimethylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with butane2,3-dione (0.76 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 3 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 54.32%; mp: 196-198 ˚C; $R_{f:}$ 0.41 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_8H_8N_4O$.

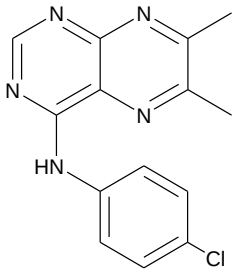
Step-2

Synthesis of 4-chloro-6,7-dimethylpteridine

A compound of 6,7-dimethylpteridin-4-ol (0.44 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1.5 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 58.23.12%; mp: 212-214 ˚C; $R_{f:}$ 0.37 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_8H_7ClN_4$.

Step-3

Synthesis of N-(4-chlorophenyl)-6,7-dimethylpteridin-4-amine



4-chloro-6,7-dimethyl pteridine (0.0025mol, 0.485 g) was taken. Dimethylformamide (DMF) and 4-chloro aniline (0.0025mol, 0.317 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{14}H_{12}ClN_5$, White Solid, Yield: 58.23%; mp: 234-236 ˚C; $R_{f:}$ 0.42 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1534 (C=N) 1596 (C=C), 3067 (C–H aromatic), 3346 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.14 (s, 6H, -CH3), 4.0 (s, 1H, D2O exchangeable -NH,), 6.58-7.84 (m, 4H, Ar-CH), 8.2 (1H,s, Ar-CH) MS (EI) m/z: 286.12 (M + 1).

Synthesis of compound-E3

Step-1

Synthesis of 6,7-dimethylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with butane2,3-dione (0.76 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 3 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 54.32%; mp: 196-198 ˚C; $R_{f:}$ 0.41 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_8H_8N_4O$.

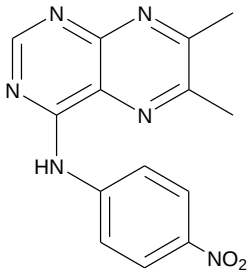
Step-2

Synthesis of 4-chloro-6,7-dimethylpteridine

A compound of 6,7-dimethylpteridin-4-ol (0.44 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1.5 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 58.23.12%; mp: 212-214 ˚C; $R_{f:}$ 0.37 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_8H_7ClN_4$.

Step-3

Synthesis of 6,7-dimethyl-N-(4-nitrophenyl) pteridin-4-amine



4-chloro-6,7-dimethyl pteridine (0.0025mol, 0.485 g) was taken. Dimethylformamide (DMF) and 4-nitro aniline (0.0025mol, 0.345 g) was added and reaction mixture was stirred at room temperature for 1.5 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{14}H_{12}N_6O_2$, White Solid, Yield: 67.41%; mp: 267-269 ˚C; $R_{f:}$ 0.61 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1534 (C=N), 1550 (N-O), 1596 (C=C), 3067 (C–H aromatic), 3346 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.11 (s, 6H, -CH3), 4.11 (s, 1H, D2O exchangeable -NH,), 6.58-8.27 (m, 4H, Ar-CH), 8.43 (1H,s, Ar-CH) MS (EI) m/z: 297.16(M + 1).

Synthesis of compound-E4

Step-1

Synthesis of 6,7-dimethylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with butane2,3-dione (0.76 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 3 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 54.32%; mp: 196-198 ˚C; R_f: 0.41 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_8H_8N_4O$.

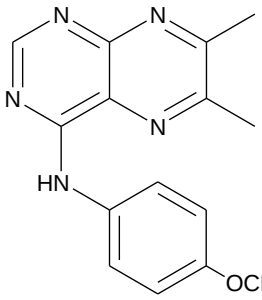
Step-2

Synthesis of 4-chloro-6,7-dimethylpteridine

A compound of 6,7-dimethylpteridin-4-ol (0.44 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1.5 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 58.23.12%; mp: 212-214 ˚C; R_f: 0.37 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_8H_7ClN_4$.

Step-3

Synthesis of N-(4-methoxyphenyl)-6,7-dimethylpteridin-4-amine



4-chloro-6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.485 g) was taken. Dimethylformamide (DMF) and 4-methoxy aniline (0.0025mol, 0.307 g) was added and reaction mixture was stirred at room temperature for 1.5 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{15}H_{15}N_5O$, White Solid, Yield: 60.36%; mp: 243-245 ˚C; R_f: 0.54 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1526 (C=N), 1587 (C=C), 3072 (C–H aromatic), 3334 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.14 (s, 6H, -CH3), δ 3.20 (s, 3H, -OCH3) 4.15 (s, 1H, D2O exchangeable -NH,), 6.58-7.96 (m, 4H, Ar-CH), 8.10 (1H,s, Ar-CH) MS (EI) m/z: 282.23(M + 1).

Synthesis of compound-E5

Step-1

Synthesis of 6,7-dimethylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with butane2,3-dione (0.76 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 3 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 54.32%; mp: 196-198 ˚C; R_f: 0.41 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_8H_8N_4O$.

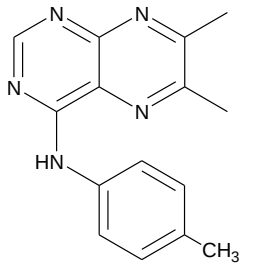
Step-2

Synthesis of 4-chloro-6,7-dimethylpteridine

A compound of 6,7-dimethylpteridin-4-ol (0.44 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1.5 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 58.23.12%; mp: 212-214 ˚C; R_f: 0.37 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_8H_7ClN_4$.

Step-3

Synthesis of N-(4-methoxyphenyl)-6,7-dimethylpteridin-4-amine



4-chloro-6, 7-bis (3-methoxyphenyl) pteridine (0.0025mol, 0.485 g) was taken. Dimethylformamide (DMF) and 4-methyl aniline (0.0025mol, 0.267 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{15}H_{15}N_5$, White Solid, Yield: 52.45%; mp: 220-222 ˚C; R_f: 0.62 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1531 (C=N), 1575 (C=C), 3065 (C–H aromatic), 3350 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.10 (s, 6H, -CH3), δ 2.20 (s, 3H, -OCH3) 4.13 (s, 1H, D2O exchangeable -NH,), 6.58-7.73 (m, 4H, Ar-CH), 7.90 (1H,s, Ar-CH) MS (EI) m/z: 266.15(M + 1).

Synthesis of compound-E6:

Step-1

Synthesis of 6,7-diphenylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with benzil (2.10 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 68.41%; mp: 202-204 ˚C; R_f: 0.54 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{12}N_4O$.

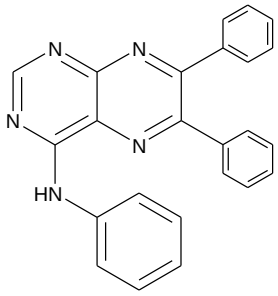
Step-2

Synthesis of 4-chloro-6,7-diphenylpteridine

A compound of 6,7-diphenylpteridin-4-ol (0.75 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 51.84.12%; mp: 240-242 ˚C; R_f: 0.45 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_{11}ClN_4$.

Step-3

Synthesis of N,6,7-triphenylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.795 g) was taken. Dimethylformamide (DMF) and aniline (0.0025mol, 0.232 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{17}N_5$, White Solid, Yield: 70.14%; mp: 208-210 ˚C; R_f: 0.56 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1515 (C=N), 1590(C=C), 2857 (C–H aliphatic), 3134 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.1 (s, 1H, D2O exchangeable -NH,), 6.62-7.79 (m, 15H, Ar-CH), 8.35 (1H,s, Ar-CH) MS (EI) m/z: 376.34 (M + 1)

Synthesis of compound-E7:

Step-1

Synthesis of 6,7-diphenylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with benzil (2.10 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 68.41%; mp: 202-204 ˚C; R_f: 0.54 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{12}N_4O$.

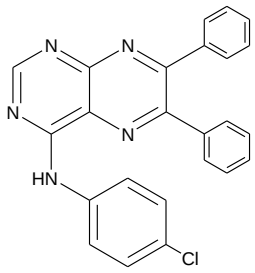
Step-2

Synthesis of 4-chloro-6,7-diphenylpteridine

A compound of 6,7-diphenylpteridin-4-ol (0.75 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 51.84.12%; mp: 240-242 ˚C; R_f: 0.45 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_{11}ClN_4$.

Step-3

Synthesis of N-(4-chlorophenyl)-6,7-diphenylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.795 g) was taken. Dimethylformamide (DMF) and 4-chloro aniline (0.0025mol, 0.317 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{16}ClN_5$, White Solid, Yield: 65.21%; mp: 213-215 ˚C; R_f: 0.52 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1509 (C=N), 1578 (C=C), 2876 (C–H aliphatic), 3113 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.1 (s, 1H, D2O exchangeable -NH,), 6.62-8.02 (m, 14H, Ar-CH), 8.13 (1H,s, Ar-CH) MS (EI) m/z: 410.27 (M + 1)

Synthesis of compound-E8:

Step-1

Synthesis of 6,7-diphenylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with benzil (2.10 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature.

Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 68.41%; mp: 202-204 ˚C; R_f: 0.54 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{12}N_4O$.

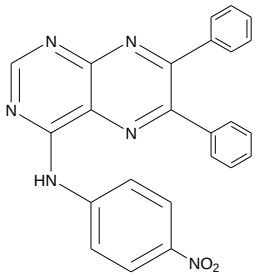
Step-2

Synthesis of 4-chloro-6,7-diphenylpteridine

A compound of 6,7-diphenylpteridin-4-ol (0.75 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 51.84.12%; mp: 240-242 ˚C; R_f: 0.45 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_{11}ClN_4$.

Step-3

Synthesis of N-(4-nitrophenyl)-6,7-diphenylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.795 g) was taken. Dimethylformamide (DMF) and 4-nitro aniline (0.0025mol, 0.345 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{16}N_6O_2$, White Solid, Yield: 62.12%; mp: 273-275 ˚C; R_f: 0.50 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1339 (N-O), 1510 (C=N), 1596 (C=C), 2872 (C–H aliphatic), 3101 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.0 (s, 1H, D2O exchangeable -NH,), 6.62-7.89 (m, 14H, Ar-CH), 8.41 (1H,s, Ar-CH) MS (EI) m/z: 421.22 (M + 1)

Synthesis of compound-E9:

Step-1

Synthesis of 6,7-diphenylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with benzil (2.10 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 68.41%; mp: 202-204 ˚C; R_f: 0.54 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{12}N_4O$.

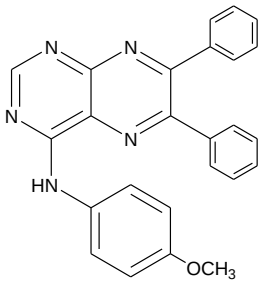
Step-2

Synthesis of 4-chloro-6,7-diphenylpteridine

A compound of 6,7-diphenylpteridin-4-ol (0.75 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 51.84.12%; mp: 240-242 ˚C; R_f: 0.45 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_{11}ClN_4$.

Step-3

Synthesis of N-(4-methoxyphenyl)-6,7-diphenylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.795 g) was taken. Dimethylformamide (DMF) and 4-methoxy aniline (0.0025mol, 0.307 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{25}H_{19}N_5O$, White Solid, Yield: 57.13%; mp: 255-257 ˚C; R_f: 0.59 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1145 (C-O), 1514 (C=N), 1589 (C=C), 2894 (C–H aliphatic), 3143 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.8 (s, 3H, -OCH_3,), δ 4.2 (s, 1H, D2O exchangeable -NH,), 6.71-8.12 (m, 14H, Ar-CH), 8.17 (1H,s, Ar-CH) MS (EI) m/z: 406.11 (M + 1)

Synthesis of compound-E10:

Step-1

Synthesis of 6,7-diphenylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with benzil (2.10 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 68.41%; mp: 202-204 ˚C; R_f: 0.54 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{12}N_4O$.

Step-2

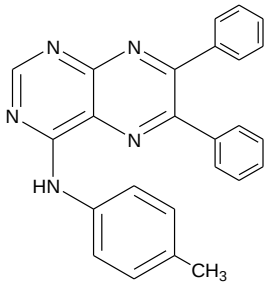
Synthesis of 4-chloro-6,7-diphenylpteridine

A compound of 6,7-diphenylpteridin-4-ol (0.75 gm, 0.0025mol) was mixed with Phosphoryl chloride

($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 51.84.12%; mp: 240-242 °C; R_f: 0.45 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_{11}ClN_4$.

Step-3

Synthesis of 6,7-diphenyl-N-p-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.795 g) was taken. Dimethylformamide (DMF) and 4-methyl aniline (0.0025mol, 0.267 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{25}H_{19}N_5$, White Solid, Yield: 48.27%; mp: 226-228 °C; R_f: 0.46 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1534 (C=N), 1605 (C=C), 2903 (C–H aliphatic), 3154 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.1(s, 3H, -CH_3,), δ 4.2 (s, 1H, D2O exchangeable -NH,), 6.56-7.87 (m, 14H, Ar-CH), 8.03(1H,s, Ar-CH) MS (EI) m/z: 390.23 (M + 1)

Synthesis of compound-E11:

Step-1

Synthesis of 6,7-dip-tolylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4-dimethyl benzil (2.38 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 72.34%; mp: 218-220 °C; R_f: 0.58 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O$.

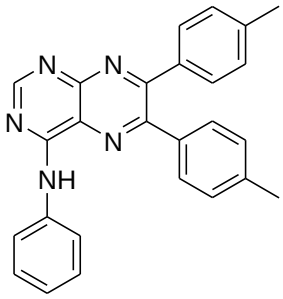
Step-2

Synthesis of 4-chloro-6,7-dip-tolylpteridine

A compound of 6,7-dip-tolylpteridin-4-ol (0.82 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 55.63.12%; mp: 253-255 °C; R_f: 0.49 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4$.

Step-3

Synthesis of N-phenyl-6,7-dip-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.865 g) was taken. Dimethylformamide (DMF) and aniline (0.0025mol, 0.232 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{21}N_5$, White Solid, Yield: 70.14%; mp: 208-210 °C; R_f: 0.56 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1523 (C=N), 1596(C=C), 2864 (C–H aliphatic), 3142 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 1.81 (s, 6H, -CH_3), 4.1 (s, 1H, D2O exchangeable -NH,), 6.59-7.68 (m, 13H, Ar-CH), 8.29 (1H,s, Ar-CH) MS (EI) m/z: 404.13 (M + 1)

Synthesis of compound-E12:

Step-1

Synthesis of 6,7-dip-tolylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4-dimethyl benzil (2.38 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 72.34%; mp: 218-220 °C; R_f: 0.58 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O$.

Step-2

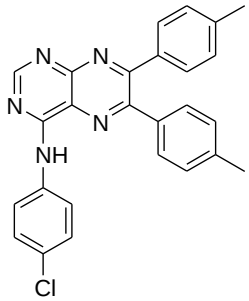
Synthesis of 4-chloro-6,7-dip-tolylpteridine

A compound of 6,7-dip-tolylpteridin-4-ol (0.82 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 55.63.12%; mp: 253-255 °C; R_f: 0.49 (*n*-hexane:

ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4$.

Step-3

Synthesis of N-(4-chlorophenyl)-6,7-dip-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.865 g) was taken. Dimethylformamide (DMF) and 4-chloro aniline (0.0025mol, 0.317 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{20}ClN_5$, White Solid, Yield: 64.67%; mp: 224-226 ˚C; $R_{f:}$ 0.61 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1534 (C=N), 1608(C=C), 2832 (C–H aliphatic), 3124 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 1.81 (s, 6H, -CH_3), 4.1 (s, 1H, D2O exchangeable -NH,), 6.67-7.92 (m, 12H, Ar-CH), 8.29 (1H,s, Ar-CH) MS (EI) m/z: 438.32 (M + 1)

Synthesis of compound-E13:

Step-1

Synthesis of 6,7-dip-tolylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4-dimethyl benzil (2.38 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 72.34%; mp: 218-220 ˚C; $R_{f:}$ 0.58 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O$.

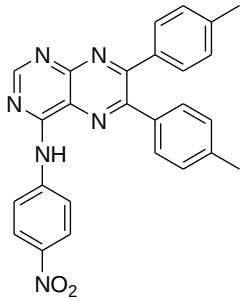
Step-2

Synthesis of 4-chloro-6,7-dip-tolylpteridine

A compound of 6,7-dip-tolylpteridin-4-ol (0.82 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 55.63.12%; mp: 253-255 ˚C; $R_{f:}$ 0.49 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4$.

Step-3

Synthesis of N-(4-nitrophenyl)-6,7-dip-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.865 g) was taken. Dimethylformamide (DMF) and 4-nitro aniline (0.0025mol, 0.345 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{20}N_6O_2$, White Solid, Yield: 54.31%; mp: 234-236 ˚C; $R_{f:}$ 0.54 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1343 (N-O), 1534 (C=N), 1594 (C=C), 2887 (C–H aliphatic), 3123 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 1.83 (s, 6H, -CH_3), 4.1 (s, 1H, D2O exchangeable -NH,), 6.65-8.10 (m, 12H, Ar-CH), 8.32 (1H,s, Ar-CH) MS (EI) m/z: 449.16 (M + 1)

Synthesis of compound-E14:

Step-1

Synthesis of 6,7-dip-tolylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4-dimethyl benzil (2.38 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 72.34%; mp: 218-220 ˚C; $R_{f:}$ 0.58 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O$.

Step-2

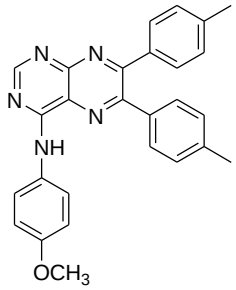
Synthesis of 4-chloro-6,7-dip-tolylpteridine

A compound of 6,7-dip-tolylpteridin-4-ol (0.82 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 55.63.12%; mp: 253-255 ˚C; $R_{f:}$ 0.49 (*n*-hexane:

ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4$.

Step-3

Synthesis of N-(4-methoxyphenyl)-6,7-dip-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.865 g) was taken. Dimethylformamide (DMF) and 4-methoxy aniline (0.0025mol, 0.307 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{27}H_{23}N_5O$, White Solid, Yield: 61.76%; mp: 254-256 ˚C; R_f: 0.61 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1136 (C-O), 1529 (C=N), 1598 (C=C), 2910 (C–H aliphatic), 3154 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 1.81 (s, 6H, -CH_3), δ 2.9 (s, 3H, -OCH_3,), 4.1 (s, 1H, D2O exchangeable -NH,), 6.56-7.72 (m, 12H, Ar-CH), 8.31 (1H,s, Ar-CH) MS (EI) m/z: 434.23 (M + 1)

Synthesis of compound-E15:

Step-1

Synthesis of 6,7-dip-tolylpteridin-4-ol

A mixture of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4-dimethyl benzil (2.38 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 1.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 72.34%; mp: 218-220 ˚C; R_f: 0.58 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O$.

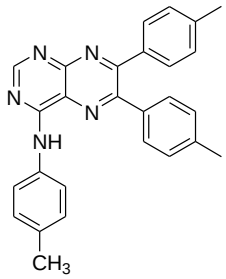
Step-2

Synthesis of 4-chloro-6,7-dip-tolylpteridine

A compound of 6,7-dip-tolylpteridin-4-ol (0.82 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 2 hours. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 55.63.12%; mp: 253-255 ˚C; R_f: 0.49 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4$.

Step-3

Synthesis of N,6,7-trip-tolylpteridin-4-amine



4-chloro-6,7-diphenylpteridine (0.0025mol, 0.865 g) was taken. Dimethylformamide (DMF) and 4-methyl aniline (0.0025mol, 0.267 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{27}H_{23}N_5$, White Solid, Yield: 52.43%; mp: 234-236 ˚C; R_f: 0.46 (DCM: MeOH (7:3)).IR (KBr, γmax cm− 1): 1523 (C=N), 1614 (C=C), 2914 (C–H aliphatic), 3162 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 1.86 (s, 9H, -CH_3), 4.2 (s, 1H, D2O exchangeable -NH,), 6.45-7.65 (m, 12H, Ar-CH), 8.12 (1H,s, Ar-CH) MS (EI) m/z: 418.34 (M + 1)

Synthesis of compound-E16

Step-1

Synthesis of 6,7-bis(3-methoxyphenyl)pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 3,3'- dimethoxy benzil. (1.70 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 59.67%; mp: 204-206 ˚C; R_f: 0.44 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O_3$.

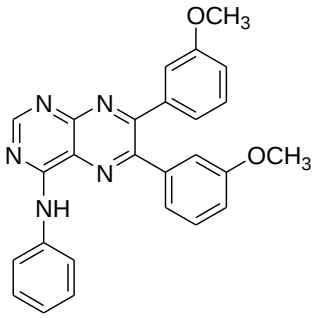
Step-2

Synthesis of 4-chloro-6,7-bis(3-methoxyphenyl) pteridine

A compound of 6,7-bis(3-methoxyphenyl) pteridin-4-ol (0.9 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4O_2$.

Step-3

6,7-bis(3-methoxyphenyl)-N-phenylpteridin-4-amine



4-chloro-6,7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.945 g) was taken. Dimethylformamide (DMF) and aniline (0.0025mol, 0.232 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{21}N_5O_2$, White Solid, Yield: 73.16%; mp: 238-240 ˚C; R_f: 0.37 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1138 (C-O), 1523 (C=N), 1578(C=C), 2865 (C–H aliphatic), 3156 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 3.76 (s, 6H, -CH3), 4.1 (s, 1H, D2O exchangeable -NH,), 6.89-7.92 (m, 13H, Ar-CH), 8.35 (1H,s, Ar-CH) MS (EI) m/z: 436.23 (M + 1)

Synthesis of compound-E17

Step-1

Synthesis of 6,7-bis(3-methoxyphenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 3,3'- dimethoxy benzil. (1.70 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 59.67%; mp: 204-206 ˚C; R_f: 0.44 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O_3$.

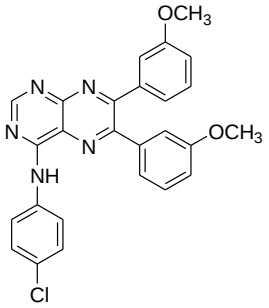
Step-2

Synthesis of 4-chloro-6,7-bis(3-methoxyphenyl) pteridine

A compound of 6,7-bis(3-methoxyphenyl) pteridin-4-ol (0.9 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4O_2$.

Step-3

N-(4-chlorophenyl)-6,7-bis(3-methoxyphenyl) pteridin-4-amine



4-chloro-6,7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.945 g) was taken. Dimethylformamide (DMF) and 4-chloro aniline (0.0025mol, 0.317 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{20}ClN_5O_2$, White Solid, Yield: 60.12%; mp: 238-240 ˚C; R_f: 0.43 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1123 (C-O), 1513 (C=N), 1584 (C=C), 2889 (C–H aliphatic), 3132 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 3.82 (s, 6H, -CH3), 4.2 (s, 1H, D2O exchangeable -NH,), 6.96-8.13 (m, 12H, Ar-CH), 8.46 (1H,s, Ar-CH), MS (EI) m/z: 470.23 (M + 1)

Synthesis of compound-E18

Step-1

Synthesis of 6,7-bis(3-methoxyphenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 3,3'- dimethoxy benzil. (1.70 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 59.67%; mp: 204-206 ˚C; R_f: 0.44 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O_3$.

Step-2

Synthesis of 4-chloro-6,7-bis(3-methoxyphenyl) pteridine

A compound of 6,7-bis(3-methoxyphenyl) pteridin-4-ol (0.9 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4O_2$.

Step-3

Synthesis of (6, 7-bis (4-methoxy phenyl)-N-(4-nitrophenyl) pteridin-4-amine)

4-chloro-6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.945 g) was taken. Dimethylformamide (DMF) and 4-nitro aniline (0.0025mol, 0.345 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{26}H_{20}N_6O_4$, White Solid, Yield: 64.32%; mp: 253-255 ˚C; R_f: 0.31 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1130 (C-O), 1273(C–N), 1331 (N-O) 2965 (C–H aliphatic), 3008 (C–H aromatic), 3346 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 3.73 (s, 6H, -CH3), 4.0 (s, 1H, D2O exchangeable -NH,), 6.72-8.39 (m, 13H, Ar-CH), MS (EI) m/z: 481.15 (M + 1)

Synthesis of compound-E19

Step-1

Synthesis of 6,7-bis(3-methoxyphenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 3,3'- dimethoxy benzil. (1.70 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 59.67%; mp: 204-206 ˚C; R_f: 0.44 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O_3$.

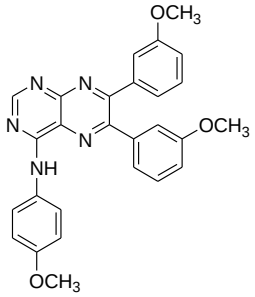
Step-2

Synthesis of 4-chloro-6,7-bis (3-methoxy phenyl) pteridine

A compound of 6,7-bis(3-methoxyphenyl) pteridin-4-ol (0.9 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4O_2$.

Step-3

Synthesis of 6, 7-bis (3-methoxy phenyl)-N-(4-methoxyphenyl) pteridin-4-amine



4-chloro- 6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.945 g) was taken. Dimethylformamide (DMF) and 4-methoxy aniline (0.0025mol, 0.307 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{27}H_{23}N_5O_3$, White Solid, Yield: 69.13%; mp: 249-251 ˚C; R_f: 0.52 (DCM: MeOH (8:2)).IR (KBr, γ_{max} cm− 1): 1141 (C-O), 1533 (C=N), 1596 (C=C), 2914 (C–H aliphatic), 3147 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 3.1 (s, 9H, -OCH_3,), δ 4.2 (s, 1H, D2O exchangeable -NH,), 6.67-8.05 (m, 12H, Ar-CH), 8.20 (1H,s, Ar-CH) , MS (EI) m/z: 466.21 (M + 1)

Synthesis of compound-E20

Step-1

Synthesis of 6,7-bis(3-methoxyphenyl) pteridin-4-ol

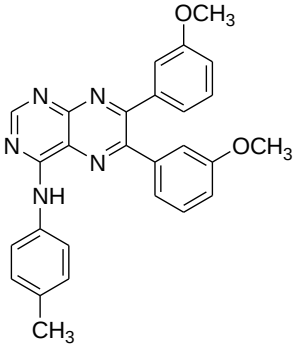
A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 3,3'- dimethoxy benzil. (1.70 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 59.67%; mp: 204-206 ˚C; R_f: 0.44 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{20}H_{16}N_4O_3$.

Step-2

Synthesis of 4-chloro-6,7-bis (3-methoxy phenyl) pteridine

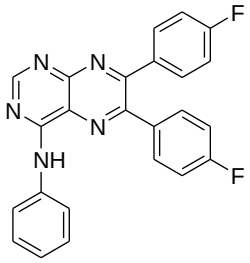
A compound of 6,7-bis(3-methoxyphenyl) pteridin-4-ol (0.9 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 59.36%; mp: 240-242 ˚C; R_f: 0.59 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{20}H_{15}ClN_4O_2$.

Step-3
Synthesis of 6,7-bis(3-methoxyphenyl)-N-p-tolylpteridin-4-amine



4-chloro-6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.945 g) was taken. Dimethylformamide (DMF) and 4-methyl aniline (0.0025mol, 0.267 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{27}H_{23}N_5O_2$, White Solid, Yield: 56.78%; mp: 250-252 ˚C; R_f: 0.52 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1130 (C-O), 1546 (C=N), 1622 (C=C), 2918 (C–H aliphatic), 3136 (–NH);1H NMR (400 MHz, DMSO d6) δ PPM: δ 2.1(s, 3H, -CH_3,), δ 3.73 (s, 6H, -CH3), 4.0 (s, 1H, D2O exchangeable -NH,), 6.61-7.83 (m, 12H, Ar-CH), 8.12 (1H,s, Ar-CH) MS (EI) m/z: 450.17 (M + 1)

Synthesis of compound-E21
Step-1
Synthesis of 6,7-bis(4-fluorophenyl) pteridin-4-ol
A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4'- diflouro benzil. (2.46 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 64.78%; mp: 211-213 ˚C; R_f: 0.51 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{10}F_2N_4O$.
Step-2
Synthesis of 4-chloro-6,7-bis(4-fluorophenyl) pteridin
A compound of 6,7-bis(4-fluorophenyl) pteridin-4-ol (0.84 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_9ClF_2N_4$.
Step-3
Synthesis of 6,7-bis(4-fluorophenyl)-N-phenylpteridin-4-amine



4-chloro-6,7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.885 g) was taken. Dimethylformamide (DMF) and aniline (0.0025mol, 0.232 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{15}F_2N_5$, White Solid, Yield: 61.39%; mp: 265-267 ˚C; R_f: 0.61 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1124 (C-F), 1532 (C=N), 1607 (C=C), 2934 (C–H aliphatic), 3152 (–NH);1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.1 (s, 1H, D2O exchangeable -NH,), 6.62-8.32 (m, 13H, Ar-CH), 8.24 (1H,s, Ar-CH) MS (EI) m/z: 412.23 (M + 1)

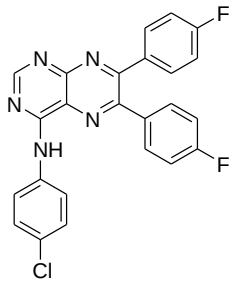
Synthesis of compound-E22
Step-1
Synthesis of 6,7-bis(4-fluorophenyl) pteridin-4-ol
A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4'- diflouro benzil. (2.46 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 64.78%; mp: 211-213 ˚C; R_f: 0.51 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{10}F_2N_4O$.

Step-2
Synthesis of 4-chloro-6,7-bis(4-fluorophenyl) pteridin
A compound of 6,7-bis(4-fluorophenyl) pteridin-4-ol (0.84 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane:

ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_9ClF_2N_4$.

Step-3

Synthesis of N-(4-chlorophenyl)-6,7-bis(4-fluorophenyl) pteridin-4-amine



4-chloro-6,7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.885 g) was taken. Dimethylformamide (DMF) and 4-chloro aniline (0.0025mol, 0.317 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{14}ClF_2N_5$, White Solid, Yield: 65.22%; mp: 269-271 ˚C; R_f: 0.63 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1124 (C-F), 1514 (C=N), 1588 (C=C), 2881 (C–H aliphatic), 3119 (–NH);1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.2 (s, 1H, D2O exchangeable -NH,), 6.62-8.37 (m, 12H, Ar-CH), 8.20 (1H,s, Ar-CH) MS (EI) m/z: 446.18 (M + 1)

Synthesis of compound-E23

Step-1

Synthesis of 6,7-bis(4-fluorophenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4'- diflouro benzil. (2.46 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 64.78%; mp: 211-213 ˚C; R_f: 0.51 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{10}F_2N_4O$.

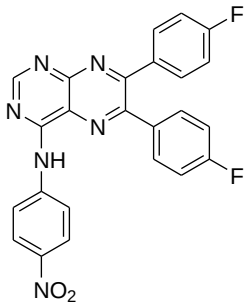
Step-2

Synthesis of 4-chloro-6, 7-bis (4-fluoro phenyl) pteridin

A compound of 6,7-bis(4-fluorophenyl) pteridin-4-ol (0.84 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_9ClF_2N_4$.

Step-3

Synthesis of 6,7-bis(4-fluorophenyl)-N-(4-nitrophenyl) pteridin-4-amine



4-chloro-6,7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.885 g) was taken. Dimethylformamide (DMF) and 4-nitro aniline (0.0025mol, 0.345 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{24}H_{14}F_2N_6O_2$, White Solid, Yield: 58.32%; mp: 251-253 ˚C; R_f: 0.56 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1124 (C-F), 1327 (N-O), 1502 (C=N), 1588 (C=C), 2858 (C–H aliphatic), 3121 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ, 4.2 (s, 1H, D2O exchangeable -NH,), 6.56-8.12 (m, 12H, Ar-CH), 8.21 (1H,s, Ar-CH) MS (EI) m/z: 457.09 (M + 1)

Synthesis of compound-E24

Step-1

Synthesis of 6,7-bis(4-fluorophenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4'- diflouro benzil. (2.46 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 64.78%; mp: 211-213 ˚C; R_f: 0.51 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{10}F_2N_4O$.

Step-2

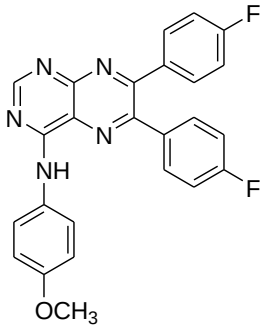
Synthesis of 4-chloro-6, 7-bis (4-fluorophenyl) pteridin

A compound of 6,7-bis(4-fluorophenyl) pteridin-4-ol (0.84 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 ˚C; R_f: 0.52 (*n*-hexane:

ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_9ClF_2N_4$.

Step-3

Synthesis of 6,7-bis(4-fluorophenyl)-N-(4-methoxyphenyl) pteridin-4-amine



4-chloro-6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.885 g) was taken. Dimethylformamide (DMF) and 4-methoxy aniline (0.0025mol, 0.307 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{25}H_{17}F_2N_5O$, White Solid, Yield: 55.18%; mp: 248-250 °C; R_f: 0.56 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1110 (C-F), 1139 (C-O), 1519 (C=N), 1593 (C=C), 2895 (C–H aliphatic), 3147 (–NH);1H NMR (400 MHz, DMSO d6) δ 2.9 (s, 3H, -OCH_3,), δ PPM: δ, 4.3 (s, 1H, D2O exchangeable -NH,), 6.55-8.12 (m, 12H, Ar-CH), 8.26 (1H,s, Ar-CH) MS (EI) m/z: 442.11 (M + 1)

Synthesis of compound-E25

Step-1

Synthesis of 6,7-bis(4-fluorophenyl) pteridin-4-ol

A mixture of of 4,5-Diamino-6-hydroxypyrimidine (1.26gm, 0.01mol) was mixed with 4,4'- diflouro benzil. (2.46 gm, 0.01mol). 1 gm sodium acetate and 50 ml of glacial acetic acid were added to above mixture. It was refluxed for 2.5 hours. After reflux, the resulting reflux mixture was cooled at room temperature. Precipitates was coming out at room temperature, and it was collected by vacuum filtration. White Solid Yield: 64.78%; mp: 211-213 °C; R_f: 0.51 (*n*-hexane: ethyl acetate-3:2). Molecular formula: $C_{18}H_{10}F_2N_4O$.

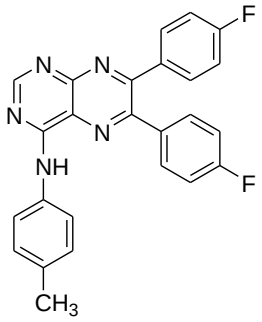
Step-2

Synthesis of 4-chloro-6, 7-bis (4-fluorophenyl) pteridin

A compound of 6,7-bis(4-fluorophenyl) pteridin-4-ol (0.84 gm, 0.0025mol) was mixed with Phosphoryl chloride ($POCl_3$) (0.38 gm, 0.0025mol) and 0.5 ml dimethylformamide (DMF) was added. The reaction mixture was heated at 120 °C for 1 hour. After heating, the mixture was cooled at room temperature and reaction mixture was poured in cool water to get precipitate of product. White Solid Yield: 54.12%; mp: 225-228 °C; R_f: 0.52 (*n*-hexane: ethyl acetate: methanol -2:2:1). Molecular formula: $C_{18}H_9ClF_2N_4$.

Step-3

Synthesis of 6,7-bis(4-fluorophenyl)-N-p-tolylpteridin-4-amine



4-chloro-6, 7-bis (3-methoxy phenyl) pteridine (0.0025mol, 0.885 g) was taken. Dimethylformamide (DMF) and 4-methyl aniline (0.0025mol, 0.267 g) was added and reaction mixture was stirred at room temperature for 2 hours. A reaction mixture was poured in ice cold water to obtain product. The product was filtered collected and dried. Molecular formula: $C_{25}H_{17}F_2N_5$, White Solid, Yield: 52.18%; mp: 244-246 °C; R_f: 0.48 (DCM: MeOH (8:2)).IR (KBr, γmax cm− 1): 1110 (C-F), 1516 (C=N), 1622 (C=C), 2895 (C–H aliphatic), 3148 (–NH); 1H NMR (400 MHz, DMSO d6) δ PPM: δ 1.9 (s, 3H, -CH_3,), δ 4.0 (s, 1H, D2O exchangeable -NH,), 6.45-7.95 (m, 12H, Ar-CH), 8.06 (1H,s, Ar-CH) MS (EI) m/z: 426.17(M + 1)

Biological Activity

Using the M. tuberculosis H37 Rv strain, the anti-tubercular potency of pteridine derivatives was examined using the Microplate Alamar Blue Assay (MABA) method (21). Using ciprofloxacin, pyrazinamide, and streptomycin as reference medicines, the activity was assessed. Potent compounds were examined using the Lowenstein-Jensen medium. 4 ml of sterilized malachite green solution was added to the mineral salt solution, which contained potassium phosphate (4.0 g), magnesium sulphate (0.4 g), magnesium citrate (1.6 g), asparagine (6.0 g), and 20 ml of glycerol, and the volume was then increased to 1000 ml with distilled water. Preliminary screening identified powerful compounds that were dissolved in DMSO and transferred 0.8 ml of each concentration using various McCartney bottles. Bottles were incubated at 70-75 C for the following 4 days with the addition of 7.5 ml of Lowenstein-Jansen medium. Using the Lowenstein Jensen proportion approach and the genotype MTBDR DNA strip assay, it is possible to identify multidrug-resistant tuberculosis (21).

Table 2- Biological activities of derivatives E1-E25 against standard Drugs

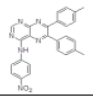
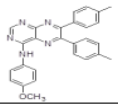
Derivatives E1-E25	MIC (µM)
Streptomycin standard	12.47 ± 3.49
Ciprofloxacin standard	10.26 ± 3.37
Pyrizinamide standard	32.59 ± 2.44
Compound E1	16.46 ± 1.35
Compound E2	46.53 ± 2.56
Compound E3	25.83 ± 2.64
Compound E4	32.47 ± 3.64
Compound E5	59.05 ± 2.48
Compound E6	10.26 ± 1.53
Compound E7	64.56 ± 4.74
Compound E8	14.58 ± 4.62
Compound E9	28.94 ± 3.59
Compound E10	41.58 ± 3.62
Compound E11	78.34 ± 2.64
Compound E12	33.13 ± 2.63
Compound E13	11.48 ± 4.63
Compound E14	29.05 ± 3.94
Compound E15	42.58 ± 2.11
Compound E16	55.39 ± 3.04
Compound E17	14.58 ± 0.49
Compound E18	82.56 ± 2.35
Compound E19	31.79 ± 3.69
Compound E20	49.05 ± 2.62
Compound E21	49.03 ± 3.74
Compound E22	22.87 ± 2. 05
Compound E23	31.74 ± 3.48
Compound E24	20.94 ± 2.75
Compound E25	26.73 ± 2. 49

Table 3- Synthesized compounds E1-E25 showing MIC in range with the standard drugs where Y= in range with standard drug and - = not in range with standard drug

STANDARD DRUGS COMPOUNDS	STREPTOMYCIN	CIPROFLOXACIN	PYRIZINAMIDE
E1	Y	Y	-
E2	-	-	-
E3	-	-	Y
E4	-	-	Y
E5	-	-	-
E6	Y	Y	-
E7	-	-	-
E8	Y	Y	-
E9	-	-	Y
E10	-	-	-
E11	-	-	-
E12	-	-	Y

E13	Y	Y	-
E14	-	-	Y
E15	-	-	-
E16	-	-	-
E17	Y	Y	-
E18	-	-	-
E19	-	-	Y
E20	-	-	-
E21	-	-	-
E22	-	-	-
E23	-	-	-
E24	-	-	-
E25	-	-	Y

Table 4: There are three set of compounds, one having activity like that of standard drug, streptomycin and ciprofloxacin other class pyrazinamide and remaining not falling in the range for Anti-TB activity.

Strept+Cip	Pyrizinamide	Not in the range
E6 	E3 	E2 
E13 	E25 	E5 
E8 	E9 	E7 
E17 	E14 	E10 
E1 	E19 	E11 
-	E4 	E15 
-	E12 	E16

CONCLUSION

In conclusion, the pteridine derivatives E1-E25 have shown promising results in their anti-TB activity, as evaluated against standard drugs streptomycin, ciprofloxacin, and pyrazinamide. All the compounds demonstrated anti-TB activity, with some showing activity close to the reference drugs. Specifically, compounds E6, E8, E13, and E17 showed activity close to streptomycin and ciprofloxacin, while compounds E3, E25, E9, E14, E19, and E4 showed activity close to pyrazinamide. These findings suggest that pteridine derivatives could be further explored as potential anti-TB agents, with the potential to offer an alternative to current standard therapies. Further studies could focus on optimizing these compounds to improve their potency and selectivity, as well as investigating their mechanism of action against TB.

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