Design, Synthesis and screening of some novel pyrimidines as Antihistaminic (H_1 antagonist).

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Abstract:

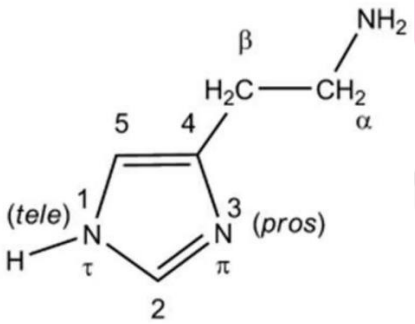
Novel pyrimidines were designed to meet the triangular pharmacophore requirement of histamine H1 receptor. Substituted Pyrimidine ring were prepared via two component synthesis method by the condensation of S, N Acetal with Symmetrical S-methyldiarylisothiourea. The structures of the synthesized compounds were confirmed using spectroscopic techniques. These compounds were also screened for anti-histaminic activity. The recorded pA_2 showed a significant anti-histaminic activity when compared to the reference anti-histaminic drug cetirizine. Compounds were also evaluated for their sedative potential as well as their anticholinergic activities as these two are known to be the common adverse effects of histamine H1-receptor antagonists.

Keywords: Pyrimidine, H_1 receptor, Antiallergic, Anticholinergic, Sedative**,** Pharmacophore

I. Introduction:

Heterocyclic compounds are those which have a cyclic structure with two, or more, different kinds of atom in the ring. [1] Heterocycles form by far the largest of the classical divisions of organic chemistry and are of immense importance biologically, industrially, and indeed to the functioning of any developed human society.[2] Heterocyclic compounds have been frequently found as a key structural motif in pharmaceuticals and agrochemicals. Heterocyclic structures having the ability to synthesize various compounds based on core structure and screen against a variety of biological activities which provide several active compound leads. Therefore, more combinations of heterocyclic structures can be designed, resulting in new structures with expected biological properties. [3].

In the family of heterocyclic compounds nitrogen containing heterocyclic are an important class of compounds in the medicinal chemistry and also contributed to the society from biological and industrial point which helps to understand life processes. [4].
Pyrimidine is a diazine i.e., six-membered heterocyclic with two nitrogen atoms in the ring. Pyrimidine is planer Six-member heterocyclic ring two N present in 1st and 3rd positions – *m* configuration. [5].
Substituted Pyrimidine constitute an important class of natural and non-natural products, any of which exhibit useful biological activities and clinical applications.[6]
Pyrimidine skeleton present in many natural products like DNA, RNA, vitamin B1 (thiamine) etc. Literature survey shows wide range of biological and pharmacological activities exhibited by pyrimidine derivatives like Antimicrobial Activity [7], Antifungal activity [8], antileishmanial [9], anti-inflammatory [10], analgesic [11], antipyretic [12], antiviral [13], antidiabetic [14], antioxidant [15], antihistaminic [16], anticancer activities [17] and many other.
Histamine generally referred to as an Autacoid, chemically histamine is [(1 H-imidazole-4-ethylamine.



(1) Histamine

Histamine is found in the most tissues, but is in highest concentration in skin, lungs and the gastrointestinal tract where it is stored in mast cells. In blood, it is found in basophils, in CNS, it appears as a neurotransmitter.[18]
Histamine upon release, its effects are principally local ones, as it functions as an autocoid. Histamine is one of the many mediators involved in allergic inflammatory responses, and it has an important role in the regulation of the secretion of gastric acid. These observations have led to development of many important drugs that antagonize its effects and are useful in treatment of allergic inflammatory disorders (H_1 antihistamines) and in the treatment of gastric hyper secretory disorders (H_2 antihistamines). [19].
In 1937, the first histamine receptor antagonist, Piperoxan, was discovered by Bovet [20]. Later Staub synthesized modified benzodioxan derivatives. Both of them won Nobel Prize for physiology and medicine in 1957 [21].

A structural observation of so-called first-generation histamine receptor antagonists or classical reveals that the two aromatic rings linked by a short chain of atoms to a tertiary amino group. This structural similarity gave a better approach in development of a large number of highly potent H1 receptor antagonists.[22]

Physicochemical properties that correlate most commonly with high potency as histamine H_1 receptor antagonists are the basicity of the side chain, the lipophilicity of the aryl rings and their spatial arrangement. [23]

Bioactive conformations of several (semi-)rigid classical histamine H_1-receptor antagonists have been investigated (cyproheptadine, phenindamine, triprolidine, epinastine, mequitazine, IBF28145, and mianserine). In general, these antihistamines contain two aromatic rings and a basic nitrogen atom. A previously derived pharmacophoric model with the nitrogen position fixed relative to the two aromatic rings is now found not to be suitable for describing the H_1-antagonist binding site. [25,26]

The classical pharmacophoric model of H_1 receptor antagonists suggest that the presence of a ring heteroatom adjacent to the aminoalkyl side-chain, a nitrogen with at least one proton and the site for heteroatom capable of hydrogen bonding are the basic structural requirements for H_1 antagonists. Although the classical structures explain the pharmacophoric requirements, it has been observed that even structures falling outside the classical general formula can exhibit potent activity (e.g. temelastine, levocarbastine, epinastine etc.). Borea et al. further refined this model to a triangular pharmacophoric model [27].

The Putative Pharmacophore of Histamine H_1 receptor antagonist suggests that out of two aromatic ring one preferred as trans ring, an unsubstituted phenyl (or -pyridyl) and the other the cis ring a p-substituted phenyl. Several structural modifications on trans ring are tolerated, whereas modification of cis ring is quite limited.[28]

II. Aim of Present work:

From X-ray crystallography data of 14 Histamine H_1 receptor antagonists it was discovered that there was a significant consistency among the crystal structure on the distance between the protonated nitrogen (N^+) and the centroid of the one of the aromatic rings (d_1 = 6.20 ± 0.15 A°). The distance between this aromatic group is also consistent (d_2 = 4.90 ± 0.24 A°), while the distance (d_3) between the second aromatic ring and the protonated nitrogen varies from 5.3 to 6.8 A°.

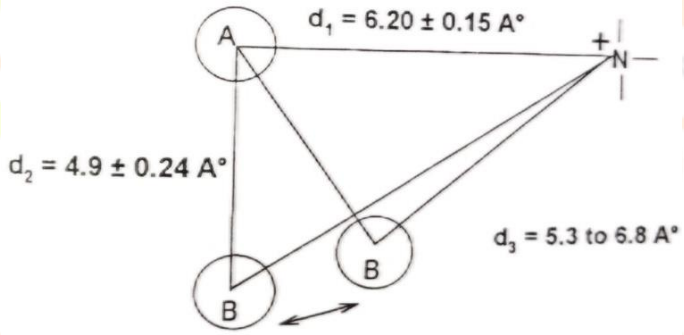
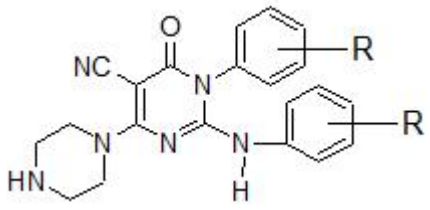


Fig 1: Geometry of H_1-receptor antagonists based on crystal srtucture.

Several substituted pyrimidines, have been designed, synthesised and reported to have potent antihistaminic activity. [29,30. Substituted pyrimidinones were designed in which distance measured were d_1= 6.56 A°. d_2 = 4.09 A° and d_3 = 7.4 A° which nearly satisfies the requirement of triangular pharmacophore for histaminic H1 receptor antagonist. The distances were measured using a PC based software, CS Chem-office and Chem. D Pro 3.0 (Cambridge Soft-USA).



Designed series

Table 1: Comparison of distances of designed compound with triangular Pharmacophore

Distance	d_1 (A°)	d_2 (A°)	d_3(A°)
Required	6.20 ± 0.15	4.90 ± 0.24	5.3 to 6.8
Found	6.565	4.09	7.4

III. Materials and methods

III A. Chemistry:

Melting points of all the compounds were determined in open capillary and are uncorrected. Infrared spectra were recorded in potassium bromide disc on Perkin-Elmer Model-841 spectrophotometer. spectra were recorded on Shimadzu 640-A UV-Visible spectrophotometer. Nuclear magnetic spectra were taken on Varian A-60 Spectrophotometer at 60 MHz and the chemical shifts are given in parts per million (6), down field from Tetramethyl silane (TMS) as internal standard. Splitting patterns are designated as follow. s = singlet, d = doublet, t = triplet, q — quartet and m = muptiplet. Mass spectra were obtained on Perkin Elmer LC-MS PE SCIEX API 165 spectrophotometer. The thin layer chromatography was performed on microscopic slides (2 x 7.5 cms.) coated with silica gel G and spots were visualized by UV radiation and exposure to iodine.

Synthetic grade chemicals were used.

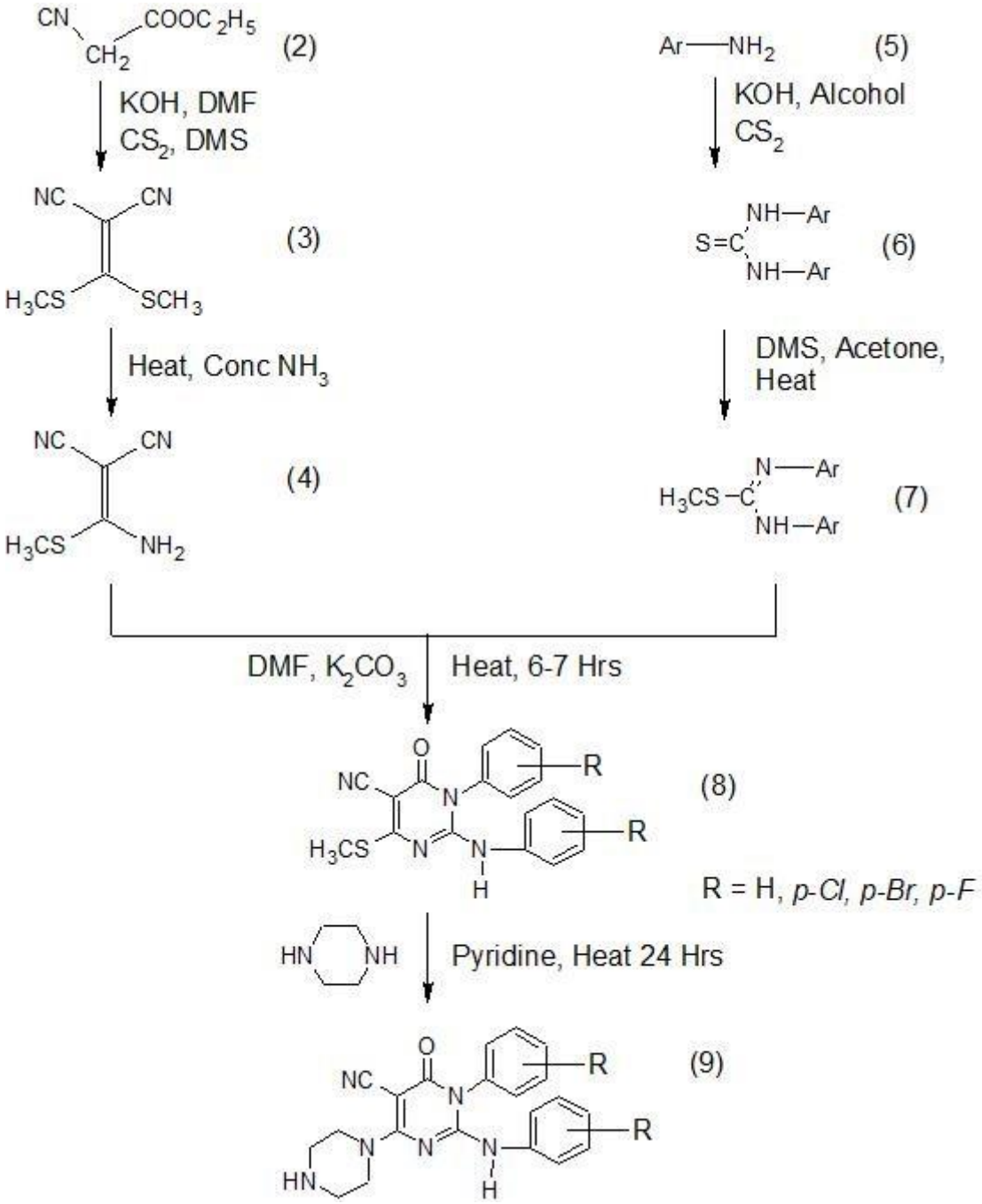


Figure 2: Scheme for synthesis of designed series

Sarting materials (3), (4), (6), (7) and (8) were prepared as reported method.

Synthesis of ethyl 2,2-di(methylmercapto)methylenecyanoacetate (3)

To an ice-cold solution of 13.2 gm (0.2 mole) of potassium hydroxide (85%) in 10 ml of water and 30 ml of dimethylformamide was added with cooling and stirring, 11.3 ml (0.1 mole) of ethylcyanoacetate followed by 7.6 gm (0.1 mole)

of carbon disulfide. The mixture was stirred for one hour at room temp., cooled and treated dropwise with 25.2 ml (0.2 mole) of dimethylsulphate, maintaining the temperature at 20 °C. The reaction mixture was allowed to stand at room temp. for 12 hours and poured in to ice-water. The solid obtained was filtered, washed with water and dried and recrystallised from methanol to get yellow crystalline compound.

Synthesis of ethyl (1-methylmercapto-l-amino)methylenecyanoacetate (4)To a paste of ethyl 2,2-di(methylmercapto) methylenecyanoacetate (10 gm., 0.04 mole) (3) in rectified spirit was added 50 ml (1 mole) of conc. ammonia solution. The mixture was then refluxed for 6 hours on water bath cooled, and poured in ice-water. The solid separated was filtered, washed with water and dried. The crude product was recrystallised from ethanol-chloroform mixture.

GENERAL PROCEDURE for Synthesis of N,N-diarylisothioureas (6)

To a solution of potassium hydroxide (5.6 gm, 0.1 mole) in ethanol (30 ml, 100%), was added the appropriate monoarylamine (0.2 mole) and carbondisulphide (20 ml). The mixture was refluxed on a steam bath until the crystals of N,N-diarylisothiourea starts separating out. Excess of organic solvent was removed by distillation under reduced pressure. The residue was washed with HC1 (10% v/v) and water, dried and recrystallised from ethanol.

GENERAL PROCEDURE for Synthesis of S-methyldiarylisothioureas (7)

To the ice-cold suspension of the appropriate diarylisothiourea (6, 0.1 mole) in minimum quantity of acetone (30 ml), dimethylsulphate (0.1mole) was added dropwise with continuous stirring over a period of half an hour. The mixture was there after stirred at room temp. for an additional hour, and then after refluxed for three hours. The acetone was removed by distillation. The heavy oil residue was poured in ice cold water and filtered. The filtrate was basified using Na2CO3 (10% aq. Na_2CO_3). The solid separated was filtered, washed with cold water, dried and recrystallised

GENERAL PROCEDURE for Synthesis of 5-Cyano-6-methylmercapto-3-(substituted)-phenyl-2-[(substituted aryl)amino]-pyrimidin-4(3H)-ones (8)

A solution of of ethyl (1-methylmercapto-l-amino)methylenecyanoacetate (4, 0.1 mole) and S-methyldiarylisothioureas (7, 0.1 mole) in DMF was refluxed for 6-7 hrs with intermittent addition of pinch of K_2CO_3. The mixture was allowed to cool and poured in 4000 ml ice-cooled water. The solid separated was filtered washed with cold water, dried and recrystallised with DMF-Methanol.

The designed series was synthesized by nucleophilic attack of the piperazine on C-6 of the pyrimidine with concomitant loss of methylmercaptan to give targeted compounds

Synthesis of 5-Cyano-6-pierazino-3-(substituted)-phenyl-2-[(substituted aryl)amino]-pyrimidin-4(3H)-ones (9) :

A solution of Synthesis of 5-Cyano-6-methylmercapto-3-(substituted)-phenyl-2-[(substituted aryl)amino]-pyrimidin-4(3H)-ones (8, 0.1 mole) an piperazine (0.3 moles) in pyridine was refluxed for 24 hours. The reaction mixture was allowed to cool and poured in ice-cooled water. The solid separated was filtered washed with cold water, dried and recrystallised with chloroform-n hexane.

The structure of all the synthesized compounds was characterized by IR, 1HNMR, Mass spectra and elemental analysis.

III B. Pharmacological screening [32-35]

III B. 1) In vitro H_1-receptor antagonistic activity:

The *in vitro* antihistaminic activity was determined by using the inhibition of the isotonic contraction induced by histamine on isolated guinea pig ileum. Test compounds were dissolved in propelyne glycol with a little warming and further dilutions were made using distilled water to get 10^{-4} and 10^{-5} molar solution. Standard histamine solution was prepared in physiological salt solution. Cetirizine was used as a standard drug. Overnight fasted guinea pig were scarified by cutting neck blood vessel, prior to sacrifice they were stunned. Responses were taken on a 2 cm long piece of ileum stimulated by physiological salt solution at 37°C. The method involves the blocking of the histamine induced contraction by the antagonists at different logarithmically increasing dose levels. Each response was repeated 2–3 times. Graphs of log dose of the antagonist *vs* percent inhibition were plotted and EC_{50} values were calculated and pA_2 values were determined.

$pA_2 = -\log[M] + \log [(A_2/ A_1) - 1]$

M = Molar Conc. Of drug used

A_1 = Conc. Of agonist at 50% height in absence of antagonist

A_2 = Conc. Of agonist at 50% height in presence of antagonist

III B. 2) Sedative potential:

As most of 1st generation antihistamine are associated as Sedation to be main side effect, so, the sedative potential of the compounds was evaluated. The drug may decrease spontaneous motor activity in animal, hence a photo actometer was used to find out sedative potential. Albini mice weighing 20–25 g were divided into 6 groups of 5 each. The first group was kept as a control group, a second group received vehicle, and the third, fourth and fifth and sixth group received the test compound (i.p.) at a dose of 1 mg/kg. Test compounds were suspended in 10 ml 10% w/v Sodium CMC and further diluted with 10% w/v Sodium CMC to get required conc. Of 100µg/ml .Each group of animals were separately placed in the photo actometer and the number of cut offs were recorded for 10 min, at an interval of 30 min, for hrs.

III B. 3) Anticholinergic activity:

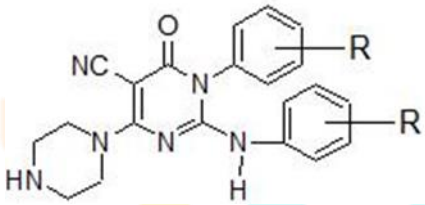
Antihistamines are reported to have an anticholinergic activity, so the anticholinergic activity of the compounds was tested *in vitro* by inhibition of the Acetylcholine induced contraction of an isolated rat ileum. Test compounds were dissolved in propelyne glycol with a little warming and further dilutions were made using distilled water to get 10^{-4} and 10^{-5} molar solution. Adult male rats weighing 200–250 g were sacrificed by cervical dislocation. Segments of ileum (2 cm in length), were obtained, flushed of their contents and trimmed of mesentery. Preparations were suspended from the transducer lever in the axis of the longitudinal muscle with fine thread, mounted in the 50 mL internal chamber of organ bath containing Tyrode's solution, maintained at 37°C and bubbled with 95% O2 and 5% CO_2. Then, it was allowed to stabilize for 60 minutes prior to drug addiction. The method involves the blocking of the Acetylcholine induced contraction by the antagonists at different logarithmically increasing dose levels.

IV. Results and discussion:

IV A, Chemistry:

Synthesis of designed derivatives was accomplished as per scheme in Figure 2.

IV A 1) Physical characteristic: All designed compounds were colourless, amorphic solid compound, melting point ranging from 210^0 – 254^0 C. The title compounds are freely soluble in dimethylformamide, dimethylsulfoxide, chloroform, acetone, methanol and sparingly soluble in benzene and insoluble in n — hexane, petroleum ether, dil. NaOH and dil. HC1 solution.



(9a-9d)

Table 2: Physical characteristic of target compounds

No	R	Mot. Formula	Mol. Wt.	Recryst. Solvent*	M. P. (°C)	Rf**	Yield (%)
9a	H	$C_{21}H_{20}N_6O$	372.4	C : H	215-217	0.25	87
9b	p-F	$C_{21}H_{18}F_2N_6O$	408.4	C : H	228-230	0.26	83
9c	p - C l	$C_{21}H_{18}Cl_2N_6O$	441.3	C : H	210-212	0.28	76
9d	p-Br	$C_{21}H_{18}Br_2N_6O$	530.2	C : H	251-253	0.32	79

C = Chloroform H = n-hexane ** TLC [Benzene : Methanol (4.5 : 0.5)]

IV A 2) Spectral characteristic:

The structures of the target compounds were confirmed by the spectral analysis.

INFRARED SPECTRA :

The I.R. spectrum of all the compounds shows the characteristic stretching peaks of the secondary amino, cyano and carbonyl functional group. The presence of broad/sharp peak at 3440-3260 cm^{-1} shows the presence of secondary amino function. The sharp peak at 2220-2200 cm^{-1} confirms the presence of -CN group. The sharp peak at 1690-1660 cm-1 typically indicates the

presence of carbonyl group. The spectrum shows the multiple peaks of -CH stretching for CH_2 of the piperazine between 3000-2800 cm-[1].

U.V. SPECTRA :

The U.V. spectra (in methanol) of all the compounds show two peaks. One in range of 235-244.6 nm and other in 290-292.8 nm. All the compounds have shown identical pattern of U.V. spectra, which confirms that all the compounds belong to same series.

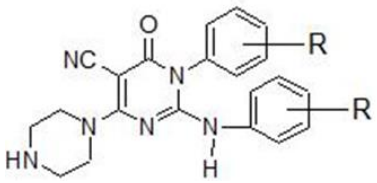
MASS SPECTRA :

All compounds were also characterized by mass spectral analysis. All the analyzed compounds gave M+ peak at the molecular weight. Compound containing halides (Cl and Br) on aryl ring shows M+2 peaks also.

NMR SPECTRA :

The 1H NMR Spectra of the compounds were studied in $CDCl_3$ The compound exhibited the following shifts.

The protons of piperazine show two multiplets at δ 2.96 and δ 3.91. The aryl protons resonate around δ 7.1-7.3 as complex multiplets corresponding to 8 protons. The secondary amino proton resonates at δ 6.07 as singlet corresponding to a single proton. The secondary proton of piperazine ring resonates at δ 7.6 as multiplet.



(9a-9d)

Table 3: Spectral characteristicanalysis of target compounds

No	**R**	**U.V. (nm) in methanol**	**Mass Peak (m/e)**	I.R. Spectra **(cm^{-1})**	^{1}H NMR shift **(δ ppm)**
9a	H	244.4, 291.0	372 (M^+)	3280 (NH), 2200 (CN), 1670 (C=O)	2.94 (m, 4H, -H_2C-N-CH_2), 3.86 (m, 4H, -H_2C-N-CH_2), 6.04 (s, 1H, -NH-C_6H_4), 7.0-7.4 (m, 10H, -C_6H_4-), 7.6 (m, 1H, -H_2C-NH-H_2C)
9b	p-F	234., 290.7	408 (M^+)	3280 (NH), 2200 (CN), 1670 (C=O)	2.94 (m, 4H, -H_2C-N-CH_2), 3.86 (m, 4H, -H_2C-N-CH_2), 6.04 (s, 1H, -NH-C_6H_4), 7.1-7.3 (m, 8H, -C_6H_4-), 7.6 (m, 1H, -H_2C-NH-H_2C)
9c	p-Cl	235.0, 291.2	441(M^+), 443 (M^{+2})	3280 (NH), 2200 (CN), 1670 (C=O)	2.96 (m, 4H, -H_2C-N-CH_2), 3.91 (m, 4H, -H_2C-N-CH_2), 6.07 (s, 1H, -NH-C_6H_4), 7.1-7.3 (m, 8H, -C_6H_4-), 7.6 (m, 1H, -H_2C-NH-H_2C)
9d	p-Br	236.6, 290.0	530 (M^+), 532 (M^{+2})	3240 (NH), 2200 (CN), 1670 (C=O)	2.92 (m, 4H, -H_2C-N-CH_2), 3.87 (m, 4H, -H_2C-N-CH_2), 6.07 (s, 1H, -NH-C_6H_4), 7.1-7.3 (m, 8H, -C_6H_4-), 7.6 (m, 1H, -H_2C-NH-H_2C)

Table 4: Elemental analysis of target compounds

No	**R**	% C		% H		% N	
		Calculated	Found	Calculated	Found	Calculated	Found
9a	H	67.72	67.85	5.41	5.48	22.57	22.66
9b	p-F	61.76	61.88	4.44	4.51	20.58	20.68
9c	p - C l	57.15	57.28	4.11	4.23	19.04	19.13
9d	p-Br	47.57	47.74	3.42	3.56	18.85	18.97

IV B. Pharmacological screening

IV B. 1) In vitro H_1-receptor antagonistic activity:

All the compounds were screened for In vitro H_1-receptor antagonistic activity by *in vitro* model on the guinea pig ileum using cetrizine as standard drug. All the compound have exhibited significant H_1-receptor antagonistic activity with PA_2 value ranging from 7.6 to 8.2, while cetirizine has shown PA_2 value 8.2

Table 5: In vitro H_1-receptor antagonistic activity (PA_2) value of target compounds

No	R	A_2	A_1	$(A_2/ A_1) - 1$	PA_2
9a	H	0.46	0.38	0.21	7.6
9b	p-F	0.63	0.35	0.8	8.2
9c	p-Cl	0.59	0.33	0.78	8.1
9d	p-Br	0.42	0.32	0.31	7.9
Cetrizine		0.67	0.40	0.67	8.4

IV B. 2) Sedative potential:

All the compounds were screened for sedative potential on mice using a photoactometer at a dose level of 1 mg/kg i.p. and recording 'cut off' for 10 min duration at the interval of 30 minutes. % Sedation was calculated for each group. All compounds exhibited comparable sedation to that of the standard drug cetirizine.

Table 6: Sedative potential of target compounds:

No	**R**	**Time in minutes** % Sedation			
		30	**60**	**90**	**120**
9a	H	26.35	46.37	54.58	52.66
9b	p-F	38.56	52.34	57.33	53.68
9c	p - C l	27.11	43.88	53.93	51.02
9d	p-Br	24.63	42.97	51.36	53.27
Cetrizine		11.62	23.24	31.49	34.25
Vehicle		2.87	3.87	2.94	3.54

IV B. 3) Anticholinergic activity: when tested for their Anticholinergic activity in vitro by their ability to inhibit acetylcholine-induced contraction on isolated rat ileum, all were found to be devoid of any anticholinergic activity.

References:

1. Alan R. Katritzky, Charles W. Rees, Comprehensive Heterocyclic Chemistry, Volume 1, Elsevier Publication, 1984, Pages 1-6
2. Alan R. Katritzky, Introductin: Heterocycles, Chem. Rev. 2004, 104, 5, 2125–2126.
3. Vijayendar Venepally, Ram Chandra Reddy Jala, An insight into the biological activities of heterocyclic–fatty acid hybrid molecules, European Journal of Medicinal Chemistry Volume 141, 1 December 2016, Pages 113-137.
4. García-Valverde M, Torroba T. Special issue: sulfur-nitrogen heterocycles. *Molecules*. 2005;10(2):318–320.
5. Joule, John A.; Mills, Keith, Heterocyclic Chemistry (5th ed.). 2010, Oxford: Wiley publication.
6. Elderield RC. Heterocyclic Compounds. Vol. 6. New York, NY, USA: John Wiley & Sons; 1957.
7. Russo, F., Romeo, G., Caruso, A., Cutuli, V., Amore, D., and Santagati, N.A. (1999). A Review on Pharmacological Aspects of Pyrimidine Derivatives, Eur. J. Med. Chem., 29, 569
8. Agarwal N, Raghuwanshi SK, Upadhyay DN, Shukla PK, Ram VJ. Suitably functionalised pyrimidines as potential antimycotic agents. Bioorganic and Medicinal Chemistry Letters. 2000;10(8):703–70.
9. Ram VJ, Haque N, Guru PY. Chemotherapeutic agents XXV: synthesis and leishmanicidal activity of carbazolylpyrimidines. European Journal of Medicinal Chemistry. 1992;27(8):851–85.
10. Amir M, Javed SA, Kumar H. Pyrimidine as anti-inlammatory agent: a review. Indian Journal of Pharmaceutical Sciences. 2007;68:p. 337.
11. Sondhi SM, Jain S, Dwivedi AD, Shukla R, Raghubir R. Synthesis of condensed pyrimidines and their evaluation for anti-inlammatory and analgesic activities. Indian Journal of Chemistry B. 2008;47(1):136–143.
12. Smith PAS, Kan RO. Cyclization of isothiocyanates as a route to phthalic and homophthalic acid derivatives. Journal of Organic Chemistry. 1964;29(8):2261–2265.
13. Balzarini J, McGuigan C. Bicyclic pyrimidine nucleoside analogues (BCNAs) as highly selective and potent inhibitors of varicella-zoster virus replication. Journal of Antimicrobial Chemotherapy. 2002;50(1):5–9.
14. Lee HW, Bok YK, Joong BA, et al. Molecular design, synthesis, and hypoglycemic and hypolipidemic activities of novel pyrimidine derivatives having thiazolidinedione, European Journal of Medicinal Chemistry. 2005;40(9):862–874.

15. Abu-Hashem AA, El-Shehry MF, Badria FA. Design and synthesis of novel thiophenecarbohydrazide, thienopyrazole and thienopyrimidine derivatives as antioxidant and antitumor agents. Acta Pharmaceutica. 2010;60(3):311–323.
16. Rahaman SA, Pasad YR, Kumar P, Kumar B. Synthesis and anti-histaminic activity of some novel pyrimidines. Saudi Pharmaceutical Journal. 2009;17(3):255–258.
17. Kaldrikyan MA, Grigoryan LA, Geboyan VA, Arsenyan FG, Stepanyan GM, Garibdzhanyan BT. Synthesis and antitumor activity of some disubstituted 5-(3-methyl-4-alkoxybenzyl)pyrimidines. Pharmaceutical Chemistry Journal. 2000;34(10):521–524.
18. Bachert, C. Histamine - A major Role in Allergy? Clin. Exp. Allergy 1998, 28, 15-19.
19. Bella, M. De.; Braghiroli, D.; Witiak, D. T. Antiallergic and anti-ulcer drugs. In Principles of Medicinal Chemistry, Foye, W. O.; Lemke, T. L.; Williams, D. A., Eds.; Waverly, B. I. Pvt. Ltd., New Delhi, 1995; 416-443.
20. Fourneau, E.; Bovet, D. Recherches Sur laction sympathicolytique dun nouveau derive du dioxane. Arch. Int. Pharmacodyn. 1933, 46, 178.
21. Greaves, M. W. Antihistamines. *Dermatol. Clin.* 2001, 19, 53-62.
22. Laduron, P. M.; Jonsssem, P. P.; Gommeron, W.; Leysen, J. E. Studies on some histamine Hr receptor antagonist. Mol. Pharmacol. 1982, 21, 294.
23. Ter Lak , A. M.; Venhort, J.; Bast, G. M.; Kelder D.; Timmerman, H. The histamine H1-receptor antagonist binding site. A stereoselective pharmacophoric model based upon (semi)rigid Hi-antagonist and including a known interaction site on the receptor. J. Med. Chem. 1995, 38, 3351-3360.
24. A M ter Laak 1 , J Venhorst, G M Donné-Op den Kelder, H Timmerman; The histamine H1-receptor antagonist binding site. A stereoselective pharmacophoric model based upon (semi-)rigid H1-antagonists and including a known interaction site on the receptor. J Med Chem, 1995 ;38(17):3351-60.
25. Ter Laak, A. M.; Venhort, J.; Bast, G. M.; Kelder D.; Timmerman, H. The histamine Hi-receptor antagonist binding site. A stereoselective pharmacophoric model based upon (semi)rigid Hi-antagonist and including a known interaction site on the receptor. J. Med. Chem.1995, 38, 3351-3360.
26. Harms, A. F.; Hespe, W.; Rekker, R, F.; Timmerman, H.; Ariens, J. De. Drug Design, Ariens, E. J., Ed.; Academic Press: New York, 1975; 6 ,1-80.
27. Borea PA, Bertolasi V, Gill G. Crystallographic and conformational studies on histamine H1 receptor antagonists. Arzneim Forsch/ Drug Res 1986;36:895.
28. Zhang, M. Q.; Leurs, R.; Timmerman, H. Histamine Hi-receptor antagonists, in Burger's Medicinal Chemistry and Drug Discovery; 5th Ed., Interscince: New York, 1995; 5, 495-559.
29. Mahesh T Chhabria 1, Vimal T Patel, Kombu S Rajan, Pathik S Brahmkshatriya; Design, synthesis and histamine H1-receptor antagonistic activity of some novel 4-amino-2-(substituted)-5-(substituted) aryl-6-[(substituted aryl) amino] pyrimidines; Arzneimittelforschung, 2009;59(5):243-7.
30. Shishoo, Chamanlal & Shirsath, Vikas & Rathod, Ishwarsinh & Yande, Vikas. (2000). Design, synthesis and antihistaminic (H1) activity of some condensed 3- aminopyrimidin-4(3H)-ones. European journal of medicinal chemistry. 35. 351-8. 10.1016/S0223-5234(00)00128-8.
31. Saxena M, Agrawal SK, Patkaik AK, Saxena AK. Synthesis, biological evaluation and quantitative structure activity relationship analysis of [β-(aroylamino)ethyl]piperazines and piperidines and [2-[(arylamino)carbonyl]ethyl]piperazines and piperidines and pyrazinoisoquinolines. A new class of potent H1-antagonists. J Med Chem 1990; 33: 2970.
32. Shah GB, Parmar NS. Antiasthmatic property of polyherbal preparation E-721 B. Phytother Res 2003; 17: 1092–1097.
33. Goyal R, K., Patel N. M., Bhatt R.V., Mehta A. A.; "Practical in Pharmacology', B S Shah Prakashan; Ahmedabad, 1st edn., 81, 1994
34. Ghosh M. N.; "Fundamental of experimental Pharmacology', Scientific book agency; Calcutta, 2nd edn., 60, 1984
35. Seyed Hassan Hejazian-Y, Mohammad Hossein Dashti-R, Seyed Majid Mahdavi, Masood Anvar Qureshi,The Effect of Carum Copticum Extract on Acetylcholine Induced Contraction in Isolated Rat's Ileum, Journal of Acupuncture and Meridian Studies, Volume 2, Issue 1, 2009, Pages 75-78,