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SYNTHESIS AND ANTI-INFLAMMATORY ACTIVITY OF SOME FUSED PYRIMIDINES

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لما وجد أن بعض مشتقات البيريميدوبيريميدين المستبدلة في حلقة البولي مثيلين بذرة كلور لها تأثير فعال كمضادات للإلتهاب، فقد تم تحضير بعض مشتقات البيريميدو (٦،١ :أ) أزيبين والبيريميدو (٤:٥،٤،٥′) بيريميدو (٦،١ :أ) ازيبين الجديدة التى تم استبدال ذرة الكلور بها بجزئي أكبر حجما مثل أمين ثنائي ألفاني حلقي (٣). وبتفاعل مركب (٣) مع الأيزوسينات المختلفة حصلنا على مركب (٤) الذى تم اختزاله إلى ٣ أمينو (٥) الذى أستخدم لتحضير مشتقات عديدة من ٣ ميثيل سلفانيل ميثيل (٨)، ميثيل ثيو حمض الخليك، ٣ ميثيل ثيو خلات (٩) و٣ ·اسيتيل سلفانيل ميثيل، ٣ · بينزويل سلفانيل ميثيل (١٠) عن طريق مشتق ٣ سلفانيل ميثيل (٧) الناتج من تفاعل ٣ كلورو ميثيل (٦) مع الثيويوريا. كما تم تحضير ٣ ·كلورو بيوتيل الثنائي الحلقات (١١) وتحويله إلى مركب ثلاثي الحلقات (١٢) ومنه تم الحصول على المركب الرباعى الحلقات (١٣). بالإضافة إلى تفاعل كلوريد الأوكساليل مع المركب (٥) ثم تكثيفه مع الكوحليات أو الأمينات ليعطى مركبات (١٤)، (١٥) التى تم حلقنتها إلى مركبات (١٦) ، (١٧) على التوالى. ولقد تم اختبار كفاءة ثماني عشر مركبا من بين المركبات الجديدة كمضادات للالتهاب وتبين من خلال تطبيق الطرق الإحصائية والتمثيل البياني على النتائج التى حصل عليها أن كل المركبات المختارة لها تأثير فعال كمضادات للالتهاب عند (ب <٠،٠٥)، وكذلك وجد لكل المركبات فعالية معتبرة عند (ب<٠،٠١) ماعدا (١٣ب)، (١٥د)، (١٦ب). ولقد وجدت المركبات (٤ج) ، (٨ج) ، (١٤د) لها فعالية أعلى من الدكلوفيناك صوديوم. ولقد أستخدمت النمذجة الجزيئية لإيجاد العلاقة بين فعالية كل مركب وتركيبه.

*Seeking new anti-inflammatory agents and based on molecular modeling studies, design and synthesis of pyrimidoazepine and pyrimidopyrimidoazepine derivatives substituted in the polymethylene ring with different alicyclic secondary amines was performed. Thus, reacting 2-dicyanomethylidenoperhydroazepine **1** with sulphuryl chloride furnished the 6-chloro derivative **2**.The o-iminonitriles **3** were obtained via the reaction of **2** with isopropyl or*

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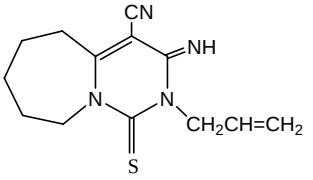
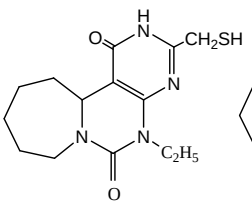
*phenyl isocyanate. Refluxing **3** with morpholine or piperidine afforded the 5-morpholino or piperidino 3-imino derivatives **4**. Reduction of the latter compounds furnished the corresponding 3-amino derivatives **5**. Preparation of the 3-chloromethyl pyrimidoazepine derivatives **6** was achieved via the reaction of **5** with chloroacetyl chloride. Reacting **6** with thiourea and further decomposition of the methyl isothiourea salts gave the 3-thiomethyl derivatives **7**. Preparation of thioethers **8** and **9** was done through the reaction of **7** with methyl iodide, chloroacetic acid or ethyl chloroacetate. Refluxing the 3-thiomethyl compounds **7** with acetyl or benzoyl chloride yielded the 3-thioester compounds **10**. The uncyclized (4-chlorobutanamide) derivatives **11** were obtained through the reaction of the enaminonitriles **5** with chlorobutyryl chloride. Refluxing compounds **11** with alcoholic hydrochloric acid solution yielded the 3-chloropropyl pyrimidopyrimidoazepine derivatives **12**.These tricycles when reacted with different primary or secondary amines yielded the tetracyclic ring system pyrrolopyrimidopyrimidoazepines **13**. Reacting **5** with oxalyl chloride gave the 3-chlorocarbonyl derivatives, which were reacted without separation with different alcohols and amines affording the corresponding 3-carbamoylformate **14** and 1-N-substituted -2-oxoacetamide **15**, respectively. Intramolecular cyclization of these latter compounds yielded their tricyclic counterparts **16** and **17**, respectively. Eighteen representative compounds were screened for their anti-inflammatory activity using diclofenac as reference drug. Also, molecular modelling was performed.*

INTRODUCTION

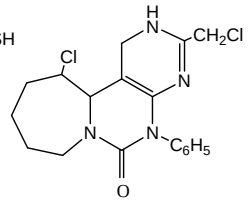
COX-inhibitors (antiinflammatory and antipyretic drugs) are widely used classes of therapeutic agents. Previous studies done by Ebeid et al on the nitrogen bridgehead condensed pyrimidine derivatives led to the discovery of new active anti-inflammatory compounds belonging to pyrimido[1,6-a]azepines **I**[1] and pyrimido[4',5':4,5]pyrimido[1,6-a]azepines **II** ring system.[1] Moreover, further studies on several pyrimido [4',5':4,5]pyrimido[1,6-a]azepine derivatives revealed that introduction of chlorine atom in the polymethylene ring at position 12 affording compound **III**[2] resulted in significant improvement in anti-inflammatory activity.



I



II



III

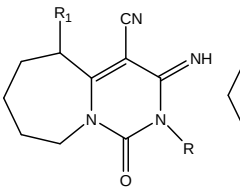
Based on these findings and for further exploration of the pyrimido-pyrimidoazepine ring system in the field of anti-inflammatory agents, molecular modeling calculations have been carried out on compounds **II** and **III** in order to correlate the spatial changes of the molecule with its activity.

It was observed that twisting increases between the polymethylene ring and the rest of the molecule by about 15^{o} upon insertion of the chlorine atom, leading to increased activity (Table I).

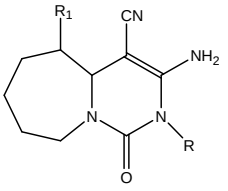
Moreover, a slight decrease in torsion angle H(Cl)(N)-12-12a-12b was perceived, with retention of the activity in the significant range, when a bulky moiety such as a alicyclic secondary amine was substituted for the chlorine atom.

It was of interest to extrapolate this concept to the bicyclic pyrimido[1,6-a]azepine derivative **I** substituted by an alicyclic secondary amine in position 5 affording compound **4c** where this modification was accompanied by a significant increase in twisting of torsion angle and activity (Table II).

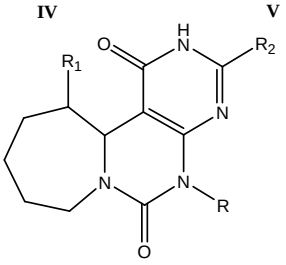
On the light of these findings and as a part of ongoing search for new potent anti-inflammatory agents, this work pertains to the design and synthesis of new series of pyrimido [1,6-a]azepine **IV** and **V,** and pyrimido[4',5':4,5]pyrimido[1,6-a]azepine derivatives **VI** substituted in the polymethylene ring with piperidine or morpholine moiety in order to investigate the influence of the conformational changes of the studied pharmacophores on the anti-inflammatory activity.



IV

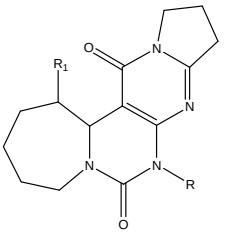


V



VI

In the same vein, the research aims to investigate the influence of increasing the bulkiness of the molecule on the twisting of torsion angles 12-12a-12b-(CO) and Cl(N)-12-12a-12b, as well as on the anti-inflammatory activity. Consequently, annulation of an additional ring on the tricyclic pyrimido[4',5':4,5] pyrimido[1,6-a]azepine ring system **VI** was done yielding compound **VII**.



VII

Table I: Torsion angles of atoms 5-4a-4-C(CN) and H(N)5-4a-4 and percentage activity.

Compound No.	Torsion angle of Atoms ° 5-4a-4-C(CN)	Torsion angle of atoms ° H(N)5-4a-4	Activity %
I	3.0353	46.2324 (-67.3853)	23
4c	-7.9882	-88.6645	88
5c	3.4387	-58.8528	70
14c	2.7157	-57.5781	84.2
14d	3.4157	-56.5781	81
14e	4.0708	-58.0348	60
15c	5.5007	-58.5448	64
15d	6.3661	-59.3181	55.7
15e	6.1151	-58.9613	46.4

Table II: Torsion angles of atoms 12-12a-12b-CO and Cl(N)-12-12a-12b and percentage activity.

Compound No.	Torsion angle ° 12-12a-12b-CO	Torsion angle ° Cl(N)-12-12a-12b	Activity %
II	72.3555	48.3943 (- 66.5016)	38
III	87.3819	-63.7448	76
6c	89.6907	-52.0906	60
7c	90.2238	-43.8014	57.1
8c	88.9228	-43.1943	81.4
10b	89.4528	-51.7408	74.2
10c	89.8137	-52.8788	71.4
12b	89.8111	54.6651	53.5
13b	91.4670	-49.9437	41.4
16b	89.2502	-54.4143	49.2
17a	84.1508	-52.1845	55.7

MATERIALS AND METHODS

Melting points (°, uncorrected) were recorded on an Electrothermal 1 A 9100 Digital Melting Point Apparatus. IR spectra as KBr disk were recorded on a Bruker Vector 22 Germany. ^{1}H-NMR spectra were recorded in DMSO-d_6 or $CDCl_3$ on Jeol FT 90Q, 300 MHz using TMS as internal standard. Mass spectra were performed on Fennigan MAT, SSQ 7000, Mass Spectrometer with Electron Impact at 70 eV. Elemental analyses were carried out at the Microanalytical Center, Faculty of Science, Cairo University. TLC was performed in chloroform using TLC aluminium sheet protected with silica gel 60 F254- layer thickness 0.2 mm.

Synthesis of compounds

2-Dicyanomethylidenoperhydroazepine **1**,[3] 3-chloro-2-dicyanomethylidenoperhydroazepine **2**[2] and 5-chloro-3-imino-2-isopropyl or phenyl-1,2,3,5,6,7,8,9-octahydro-1-oxopyrimido[1,6-a]azepine-4-carbonitrile **3**[2] were prepared according to the reported methods.

3-Imino-2-isopropyl- or phenyl-5-morpholino- or piperidino-1-oxo-1,2,3,5,6,7,8,9-octahydropyrimido-[1,6-a]azepine-4-carbonitrile (4a-c)

A mixture of **3a,b** (10 mmol) and morpholine or piperidine (15 mmol) in absolute ethanol (10 ml) was heated under reflux for 6 hours and evaporated under vacuum to dryness. The remaining residue was triturated with ether; the separated crystals were filtered and recrystallized from aqueous ethanol (Table III). **4a:** IR (KBr) 3225 (NH), 2217 (CN), 1595 (NCON). **4b**: IR (KBr) 3277, 2217, 1600. **4c**: IR (KBr) 3247, 2225, 1630. **4a:** ^{1}H-NMR ($CDCl_3$) δ= 1.31-1.39 (m, C-6(H_2), C-7(H_2), 4H); 1.85 (d, CH($\underline{CH_3}$)$_2$, 6H); 1.96-2.21 (m, C-8(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.55 (t, 2C-a(H_2), 4H); 3.53 (t, C-9(H_2), 2H); 4.21 (septet, $\underline{CH}(CH_3)_2$, 1H); 4.73 (t, C-5(H), 1H); 8.91 (s, NH, 1H). **4c**: ^{1}H-NMR ($CDCl_3$) δ= 1.18-1.35 (m, C-6(H_2), C-7(H_2), 4H); 1.96-2.21 (m, C-8(H_2), 2H); 2.49 (t, 2C-a(H_2), 4H); 3.13 (t, C-9(H_2), 2H); 3.71 (t, C-5(H), 2C-b(H_2), 5H); 7.17-7.41 (m, C_6H_5, 5H); 9.41 (s, NH, 1H). **4a**: MS: m/z (%)= M^+ 329 (0.08), 133 (100).

3-Amino-2-isopropyl- or phenyl-5-morpholino- or piperidino-1-oxo-1,2,4a,5,6,7,8,9-octahydropyrimido [1,6-a]azepine-4-carbonitrile (5a-c)

To a stirred mixture of **4a-c** (10 mmol) in absolute ethanol (20 ml), sodium borohydride (0.2 g, 5 mmol) was added in portions and stirring was continued for one hour. The reaction mixture was left to stand overnight at room temperature; the separated crystals were filtered, washed with water and recrystallized from aqueous ethanol (Table III) .**5a**: IR (KBr) 3280, 3166 (NH_2), 2217 (CN), 1620 (N-CO-N). **5b**: IR (KBr) 3210, 3123, 2220, 1645. **5c**: IR (KBr) 3277, 3260, 2215, 1600. **5a:** ^{1}H-NMR (DMSO-d_6): δ= 1.31-1.39 (m, C-6(H_2), C-7(H_2), 4H); 1.85 (d, CH($\underline{CH_3}$)$_2$, 6H); 1.96-2.21 (m, C-8(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.51(t, 2C-a(H_2), 4H); 3.75 (t, C-9(H_2), 2H); 4.35 (septet, $\underline{CH}(CH_3)_2$, 1H); 4.73 (d, C-4a(H), 1H); 5.13 (q, C-5(H), 1H); 8.53 (s, NH_2, 2H). **5c**: ^{1}H-NMR (DMSO-d_6): δ= 1.29-1.46 (m, C-6(H_2), C-7(H_2), 4H); 1.86-2.22 (m, C-8(H_2), 2H); 2.33 (t, 2C-a(H_2), 4H); 3.20 (t, C-9(H_2), 2H); 3.79 (t, 2C-b(H_2), 4H); 4.56 (q, C-5(H), 1H); 4.80 (d, C-4a(H),1H); 6.75-7.26 (m, C_6H_5, 5H); 9.51(s, NH_2, 2H). **5a**: MS: m/z (%)= M^+ 331 (18), 210 (100).

3-Chloromethyl-5-isopropyl- or phenyl-12-morpholino- or piperidino-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4',5':4,5]pyrimido [1,6-a]azepine-1,6-dione (6a-c)

A mixture of **5a-c** (10 mmol), chloroacetyl chloride (15 mmol) and triethylamine (2.5 ml) in dry benzene

Table III: Physical and analytical data of the prepared compounds **4a-c, 5a-c, 6a-c** and **7a-c**.

No	R	R^1	Yield % M.P°	Mol. formula (M.wt.)	Microanalytical data		
						Calcd.	Found
4a	$CH(CH_3)_2$		70 200-4	$C_{18}H_{27}N_5O$ 329.44	C H N	65.62 8.26 21.26	65.91 8.53 21.21
b	C_6H_5		65 170-3	$C_{21}H_{25}N_5O$ 363.46	C H N	69.40 6.93 19.27	69.25 6.34 19.56
c	C_6H_5		80 155-7	$C_{20}H_{23}N_5O_2$ 365.43	C H N	65.73 6.34 19.16	65.23 6.45 19.42
5a	$CH(CH_3)_2$		60 167-9	$C_{18}H_{29}N_5O$ 331.46	C H N	65.23 8.82 21.13	64.75 8.32 21.14
b	C_6H_5		70 180-3	$C_{21}H_{27}N_5O$ 365.47	C H N	69.01 7.45 19.16	68.71 6.95 19.66
c	C_6H_5		85 197-9	$C_{20}H_{25}N_5O_2$ 367.46	C H N	65.37 6.86 19.06	65.10 6.62 19.57
6a	$CH(CH_3)_2$		65 170-3	$C_{20}H_{30}ClN_5O_2$ 407.94	C H N	58.89 7.41 17.17	58.55 7.66 16.72
b	C_6H_5		70 200-4	$C_{23}H_{28}ClN_5O_2$ 441.96	C H N	62.51 6.39 15.85	62.72 5.95 15.43
c	C_6H_5		85 183-5	$C_{22}H_{26}Cl\,N_5O_3$ 443.93	C H N	59.52 5.90 15.78	59.41 5.83 16.15
7a	$CH(CH_3)_2$		60 200-3	$C_{20}H_{31}N_5O_2S$ 405.56	C H N	59.23 7.70 17.27	58.81 7.83 16.72
b	C_6H_5		75 187-9	$C_{23}H_{29}N_5O_2S$ 439.58	C H N	62.84 6.65 15.93	62.44 6.31 15.82
c	C_6H_5		80 195-7	$C_{22}H_{27}N_5O_3S$ 441.55	C H N	59.84 6.16 15.84	60.21 5.62 15.35

(10 ml) was refluxed at a temperature not exceeding 70° for two hours and evaporated to dryness, under vacuum. The residue was triturated with petroleum ether; the separated crystals were filtered and recrystallized from aqueous ethanol, (Table III). **6a**: IR (KBr) 3259, 1650 1595. **6b**: IR (KBr) 3320, 1645, 1599. **6c**: IR (KBr) 3326, 1648, 1596. **6a**: ^{1}H-NMR ($CDCl_3$) δ= 1.31-1.39 (m, C-10(H_2), C-11(H_2), 4H); 1.85 (d, CH(CH_3)$_2$, 6H); 1.96-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2),8H); 2.95 (t, 2C-a(H_2), 4H); 3.33 (septet, CH(CH_3)$_2$, 1H); 3.60 (s, CH_2Cl, 2H); 4.13 (t, C-8(H_2), 2H); 5.13 (d, C-12a(H), 1H); 5.36 (q, C-12(H), 1H); 8.53 (s, NH, 1H). **6b**: ^{1}H-NMR ($CDCl_3$) δ= 1.01-1.20 (m, C-10(H_2), C-11(H_2), 4H); 1.82-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.38 (t, 2C-a(H_2), 4H); 3.73 (t, C-8(H_2), 2H); 3.99 (s, CH_2Cl, 2H); 4.25 (q, C-12(H), 1H); 5.19 (d, C-12a(H), 1H); 7.16-7.66 (m, C_6H_5,5H); 8.54 (s, NH, 1H). **6a**: MS: m/z (%)= M^++2 409 (0.05); M^+ 407 (8.22), 133 (100).

5-Isopropyl- or phenyl-12-morpholino- or piperidino-3-sulfanylmethyl-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4`,5`:4,5]pyrimido[1,6-a]azepine-1,6-dione (7a-c)

A mixture of **6a-c** (10 mmol) and thiourea (0.76 g, 10 mmol) in ethanol (10 ml) was heated under reflux for about three hours then left to cool. The separated crystals were filtered, dissolved in ice cooled solution of 10٪ NaOH and filtered; the alkaline filtrate was acidified with dilute hydrochloric acid to pH 4. The formed precipitate was filtered, washed with water and crystallized from aqueous ethanol (Table III). **7a:** IR (KBr) 3212, 1665, 1612. **7b:** IR (KBr) 3310, 1653, 1615. **7c**: IR (KBr) 3364, 1665, 1630. **7a**: ^{1}H-NMR ($CDCl_3$) δ= 1.31-1.39 (m, C-10(H_2), C-11(H_2), 4H); 1.45 (d, CH(CH_3)$_2$, 6H); 1.59 (s, SH, 1H); 1.96-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2),8H); 2.53 (s, CH_2SH, 2H); 2.55 (t, 2C-a(H_2), 4H); 3.73 (t, C-8(H_2), 2H); 4.35 (septet, CH(CH_3)$_2$,1H); 4.73 (q, C-12(H),1H); 5.21(d, C-12a(H), 1H); 8.53 (s, NH, 1H). **7c**: ^{1}H-NMR ($CDCl_3$) δ= 1.09-1.19 (m, C-10(H_2), C-11(H_2), 4H); 1.69 (s, SH, 1H); 1.86-2.20 (m, C-9(H_2), 2H); 2.39 (s, CH_2SH, 2H); 2.85 (t, 2C-a(H_2), 4H); 3.44 (q, C-12(H), 1H); 3.85 (t, C-8(H_2), 2H); 4.15 (t, 2C-b(H_2), 4H); 5.18 (d, C-12a(H), 1H); 7.26-7.71 (m, C_6H_5, 5H); 8.74 (s, NH, 1H). **7a:** MS: m/z (%)= M^+ 405 (2.34), 160 (100).

3-Methylsulfanylmethyl-12-morpholino-5-phenyl-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4`,5`:4,5]pyrimido[1,6-a]azepine-1,6-dione (8a)

A mixture of **7c** (10 mmol), methyl iodide (1.4 g, 10 mmol) and anhydrous potassium carbonate (1.9 g, 20 mmol) in dry acetone (15 ml) was refluxed for about 6 hours and filtered while hot. The filtrate was distilled under vacuum; the residue was triturated with ethanol and the obtained crystals were filtered, dried and recrystallized from aqueous

ethanol (Table IV). IR (KBr) 3236, 1620, 1595. ^{1}H-NMR ($CDCl_3$) δ= 1.22-1.29 (m, C-10(H_2), C-11(H_2), 4H); 1.55-1.82 (m, C-9(H_2), 2H); 2.21 (s, SCH_3, 3H); 2.33 (q, C-12(H), 1H); 2.51 (s, CH_2S, 2H): 2.87 (t, 2C-a(H_2), 4H); 3.26 (t, C-8(H_2), 2H); 3.67 (t, 2C-b(H_2), 4H); 4.25 (d, C-12a (H), 1H); 7.12-7.63 (m, C_6H_5, 5H); 9.35 (s, NH, 1H).

5-Isopropyl- or phenyl-12-morpholino- or piperidino-1,6-dioxo-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4',5':4,5]pyrimido[1,6-a] azepine-3-methyl thioacetic acid (8b,c)

A mixture of **7a,c** (10 mmol), chloroacetic acid (0.94 ml, 10 mmol) and potassium hydroxide (0.72 g, 20 mmol), in absolute ethanol (20 ml) was refluxed for about 8 hours then filtered while hot. The filtrate was evaporated to dryness; the formed residue was dissolved in water then acidified with dilute hydrochloric acid. The separated crystals were filtered, washed with water and recrystallized from aqueous ethanol (Table IV). **8b:** IR (KBr) 3421, 3275, 1652, 1598. **8c:** IR (KBr) 3433, 3280, 1645, 1612. **8b**:^{1}H-NMR ($CDCl_3$): δ= 1.16-1.34 (m, C-10(H_2), C-11(H_2), 4H); 1.70 (d, CH($\underline{CH_3}$)$_2$, 6H); 1.96-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.73 (s, $\underline{CH_2}$S, 2H); 2.95 (t, 2C-a(H_2), 4H); 3.53 (septet, $\underline{CH}(CH_3)_2$, 1H); 4.35 (t, C-8(H_2), 2H); 4.56 (s, $\underline{CH_2}$COOH, 2H); 5.07 (d, C.12a(H), 1H); 5.53 (q, C-12(H), 1H); 6.91 (s, COOH, 1H); 8.49 (s, NH, 1H).

Ethyl 5-phenyl-12-piperidino-1,6-dioxo-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4',5':4,5]pyrimido [1,6-a]azepine-3-methyl thioacetate (9)

A mixture of compound **7b** (4 g, 10 mmol), ethyl chloroacetate (0.9 ml, 10 mmol) and anhydrous potassium carbonate (1.9 g, 20 mmol), in dry acetone (15 ml) was treated according to the method used for the preparation of compounds **8a** (Table IV). IR (KBr) 3273, 1712, 1650, 1610. ^{1}H-NMR ($CDCl_3$) δ= 1.21-1.31 (m, C-10(H_2), C-11(H_2), 4H); 1.73 (t, $COOCH_2\underline{CH_3}$, 3H); 1.96-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.55 (s, $\underline{CH_2}$S, 2H); 2.95 (t, 2C-a(H_2), 4H); 3.23 (s, $CH_2S\underline{CH_2}$, 2H); 3.61 (t, C8(H_2), 2H); 4.10 (q, $COO\underline{CH_2}CH_3$, C-12(H), 3H); 4.73 (d, C-12a(H), 1H); 6.93-7.46 (m, C_6H_5, 5H); 8.53 (s, NH, 1H). MS: m/z (%)= M^+ 525 (3.97), 97 (100).

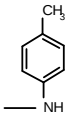
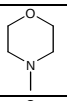
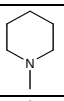
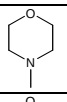
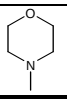
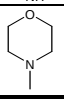
3-Acetyl or benzoyl sulfanylmethyl-12-morpholino- or piperidino-5-phenyl-1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4',5':4,5]pyrimido[1,6-a]azepine-1,6- dione (10a-c)

A mixture of **7b,c** (10 mmol), acetyl or benzoyl chloride (11 mmol) and triethylamine (0.5 ml) in dry benzene (10 ml) was refluxed for about 4 hours. The solvent was removed under vacuum and the obtained residue was triturated with ether; the separated crystals were filtered and recrystallized from aqueous ethanol (Table IV). **10a:** IR (KBr) 3235 (NH), 1705, 1610 (2CO). **10c**: IR (KBr) 3385 (NH), 1702 (CO),

Table IV: Physical and analytical data of the prepared compounds **8a-c**, **9**, **10a-c**, **14a-e** and **15a-e**.

No	R	R^1	R^2	Yield % M.P°	Mol. formula (M.wt.)	Microanalytical data		
							Cald.	Found
8a	C_6H_5		CH_3	65 165-7	$C_{23}H_{29}N_5O_3S$ 2 H_2O 491.61	C H N	56.19 6.77 14.25	55.85 6.52 14.36
b	$CH(CH_3)_2$		CH_2COOH	50 205-7	$C_{22}H_{33}N_5O_4S$ 463.60	C H N	57.00 7.17 15.11	57.31 7.30 15.6 4
c	C_6H_5		CH_2COOH	60 210-11	$C_{24}H_{29}N_5O_5S$ 499.58	C H N	57.70 5.85 14.02	57.31 5.38 14.53
9	C_6H_5		$CH_2COO\ C_2H_5$	70 175-8	$C_{27}H_{35}N_5O_4S$ 525.66	C H N	61.69 6.71 13.32	61.51 6.91 12.92
10a	C_6H_5		CH_3CO	60 184-8	$C_{25}H_{31}N_5O_3S$ 481.61	C H N	62.35 6.49 14.54	62.42 6.25 14.81
b	C_6H_5		CH_3CO	65 180-5	$C_{24}H_{29}N_5O_4S.$ ½H_2O 492.59	C H N	58.52 6.14 14.22	58.29 5.81 14.63
c	C_6H_5		C_6H_5CO	70 195-7	$C_{29}H_{31}N_5O_4S$ 545.66	C H N	63.83 5.73 12.83	63.44 5.28 12.53
14a	$CH(CH_3)_2$		CH_3	45 185-7	$C_{21}H_{31}N_5O_4$ 417.50	C H N	60.41 7.48 16.77	60.51 7.63 17.15
b	C_6H_5		C_2H_5	65 175-8	$C_{25}H_{31}N_5O_4$ 465.55	C H N	64.50 6.71 15.04	64.16 6.22 15.60
c	C_6H_5		CH_3	75 135-9	$C_{23}H_{27}N_5O_5$ 453.49	C H N	60.92 6.00 15.44	60.51 5.85 15.42
d	C_6H_5		C_2H_5	80 155-7	$C_{24}H_{29}N_5O_5$ 467.52	C H N	61.66 6.25 14.98	61.90 6.78 15.22
e	C_6H_5		$CH(CH_3)_2$	85 177-9	$C_{25}H_{31}N_5O_5.$ ½H_2O 490.55	C H N	61.21 6.58 14.28	61.52 6.31 13.94
15a	C_6H_5			62 203-5	$C_{28}H_{36}N_6O_3$ 504.63	C H N	66.64 7.19 16.65	66.22 7.50 16.35

Table IV: (Cont.)

No	R	R^1	R^2	Yield % M.P°	Mol. formula (M.wt.)	Microanalytical data		
							Cald.	Found
15b	C_6H_5			70 183-5	$C_{30}H_{34}N_6O_3$ 526.63	C H N	68.42 6.51 15.96	68.14 6.91 16.38
c	C_6H_5			75 190-3	$C_{27}H_{34}N_6O_4$ 506.60	C H N	64.01 6.76 16.59	64.30 6.25 16.37
d	C_6H_5			55 205-8	$C_{28}H_{30}N_6O_4$ 514.58	C H N	65.35 5.88 16.33	65.10 5.45 16.17
e	C_6H_5			65 190-3	$C_{26}H_{32}N_6O_5$ 508.58	C H N	61.40 6.34 16.52	61.83 5.94 16.30

1615 (N-CO-N). **10a**: ^{1}H-NMR ($CDCl_3$) δ= 1.19-1.36 (m, C-10(H_2), C-11(H_2), 4H); 1.96-2.21 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.33 (s, CO$\underline{CH_3}$, 3H); 2.56 (s, CH_2S, 2H); 2.85 (t, 2C-a(H_2), 4H); 3.55 (q, C-12(H), 1H); 4.03 (t, C-8(H_2), 2H); 4.73 (d, C-12a(H), 1H); 7.43-8.26 (m, C_6H_5, 5H); 8.53 (s, NH, 1H). **10c**: ^{1}H-NMR (DMSO-d_6) δ= 1.24-1.30 (m, C-10(H_2), C-11(H_2), 4H); 1.96-2.21 (m, C-9(H_2), 2H); 2.50 (t, 2C-a(H_2), 4H); 2.81 (s, $\underline{CH_2}$S, 2H); 3.24 (t, C-8(H_2), 2H); 3.73 (t, 2C-b(H_2), 4H); 4.38 (q, C-12(H),1H); 4.76 (d, C-12a(H), 1H); 7.13-7.66 (m, 2C_6H_5, 10H); 8.53 (s, NH,1H). **10a**: MS: m/z (%)= M^+ 481 (6.22); 159 (100).

4-Chloro-N-[4-cyano-5-morpholino or piperidino-2-phenyl-1,2,4a,5,6,7, 8,9-octahydro-1-oxopyrimido[1,6-a] azepin-3-yl] butanamide (11a,b)

A mixture of **5b,c** (10 mmol), chlorobutyryl chloride (2.1 ml, 15 mmol) and triethylamine (2.5 ml) in dry benzene (10 ml) was stirred for one hour. The reaction mixture was then refluxed for two hours then evaporated to dryness, under vacuum. The left residue was triturated with ether; the separated crystals were collected and recrystallized from aqueous ethanol (Table V). **11a**: IR (cm^{-1}) 3277 (NH), 2205 (CN), 1729, 1650 (2CO). **11b:** IR (cm^{-1}) 3317 (NH), 2215 (CN), 1725, 1633 (2CO). **11a**: ^{1}H-NMR (DMSO-d_6) δ= 1.15-1.36 (m, C-6(H_2), C-7(H_2), 4H); 1.75 (quintet, HNCO$CH_2\underline{CH_2}$-,2H); 1.96-2.26 (m, C-8(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.59 (t, HNCO$\underline{CH_2}$-,2H); 2.78 (t, 2C-a(H_2), 4H); 3.29 (t, C-9(H_2), 2H); 3.71(t, -$\underline{CH_2}$Cl, 2H); 4.34 (q, C-5(H), 1H); 4.73 (d, C-4a(H), 1H); 6.63-7.21(m, C_6H_5, 5H); 7.93 (s, NH,1H). **11a** MS: m/z(%)= M^+-1 469 (16.3); M^+ 470 (5.33), 133 (100).

Table V: Physical and analytical data of the prepared compounds **11a**,**b**, **12a**,**b** and **13a**,**b**.

No	R	Yield % M.P°	Mol. formula (M.wt.)	Microanalytical data		
					Calcd.	Found
11a		60 13-5	$C_{25}H_{32}ClN_5O_2$ 470.01	C H N	63.89 6.86 14.90	63.42 6.51 14.55
b		73 190-4	$C_{24}H_{30}ClN_5O_3$ 471.98	C H N	61.07 6.41 14.84	61.33 6.56 14.44
12a		65 125-8	$C_{25}H_{32}ClN_5O_2$ 470.01	C H N	63.89 6.86 14.90	63.72 6.33 14.61
b		70 150-3	$C_{24}H_{30}ClN_5O_3$ 471.98	C H N	61.07 6.41 14.84	61.35 6.96 14.53
13a		65 177-9	$C_{25}H_{31}N_5O_2$ 433.55	C H N	69.26 7.21 16.15	69.31 7.55 16.37
b		80 180-5	$C_{24}H_{29}N_5O_3$ 435.52	C H N	66.19 6.71 16.08	66.33 7.14 16.50

3-Chloropropyl-12-morpholino- or piperidino-5-phenyl-1,2,5,6,8,9,10, 11,12,12a-decahydropyrimido[4', 5':4,5]pyrimido[1,6-a]azepine-1,6-dione (12a,b)

Compounds **11a,b** were refluxed in alcoholic/HCl solution (15 ml) for three hours. The solvent was evaporated under vacuum; the left residue was triturated with ether and the separated crystals were collected and recrystallized from aqueous ethanol (Table V). **12a**,: IR (KBr) 3244 (NH), 1725, 1637 (2CO). **12b:** IR (KBr) 3205 (NH), 1710, 1650 (2CO). **12a:** ^{1}H-NMR (DMSO-d_6) δ= 1.28-1.31 (m, C-10(H_2), C-11(H_2), 4H); 1.42 (t, -$\underline{CH_2}CH_2CH_2Cl$, 2H); 1.55-1.66 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 1.82 (quintet,-$CH_2\underline{CH_2}CH_2Cl$, 2H); 2.83 (t, 2C-a($H_2$), 4H); 2.98 (q, C-12(H), 1H); 3.24 (t, C-8(H_2), 2H); 3.62 (t, $CH_2CH_2\underline{CH_2}Cl$, 2H); 4.41 (d, C-12a(H), 1H); 7.25-7.71 (m, C_6H_5, 5H); 9.50 (s, NH, 1H). **12b:** ^{1}H-NMR (DMSO-d_6) δ= 1.15-1.36 (m, C-10(H_2), C-11(H_2), 4H); 1.41 (t, -$\underline{CH_2}CH_2CH_2Cl$, 2H); 1.66 (quintet,-$CH_2\underline{CH_2}CH_2Cl$, 2H); 1.96-2.26 (m, C-9(H_2), 2H); 2.73 (t, 2C-a(H_2), 4H); 3.29 (t, 2C-b(H_2), 4H); 3.55 (t,-$CH_2CH_2\underline{CH_2}Cl$, 2H); 3.70 (q, C-12(H), 1H); 4.35 (t, C-8(H_2), 2H); 4.82 (brs, C-12a(H),1H); 6.63-7.21 (m, C_6H_5, 5H); 7.93 (s, NH, 1H).

1-Morpholino- or piperidino-8-phenyl-1,2,3,4,5,7,8,10,11,12,14, 14b-dodecahydropyrrolo[1'',2'':1',2'] pyrimido[4',5':4,5]pyrimido[1,6-a]azepine-7,14-dione

or

12-Morpholino- or piperidino-5-phenyl-1,2,3,5,6,8,9,10,11,12,12a, 13-dodecahydro-pyrrolo[2'',1'':2',3'] pyrimido[4',5':4,5]pyrimido[1,6-a] azepine-6,13-dione, (13a,b)

A mixture of **12a,b** (10 mmol) and the appropriate amine (11 mmol) in dry benzene (15 ml) was refluxed for three hours. The reaction mixture was evaporated under vacuum and the obtained residue was triturated with ether. The separated crystals were collected and recrystallized from aqueous ethanol (Table V). **13a**: IR (KBr) 1625, 1590 (2CO). **13b**: IR (KBr) 1650, 1610 (2CO). **13a**: ^{1}H-NMR (DMSO-d_6) δ= 1.15-1.36 (m, C-10(H_2), C-11(H_2), 4H); 1.56 (t, C-1(H_2), 2H); 1.75 (quintet, C-2(H_2), 2H); 1.96-2.26 (m, C-9(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.4 (t, 2C-a(H_2), 4H); 2.89 (t, C-3(H_2), 2H), 3.73(t, C-8(H_2), 2H); 4.35 (q, C-12(H), 1H); 4.73 (d, C-12a(H), 1H); 6.63-7.21 (m, C_6H_5, 5H). **13a:** MS m/z (%): M^+ 433 (14.03), 210 (100).**13b:** MS m/z (%)= M^+ -1 434.1 (0.02), 126.9 (100).

Alkyl 4-cyano-2-isopropyl- or phenyl-5-morpholino or piperidino-1-oxo-1,2,4a,5,6,7,8,9-octahydro-pyrimido[1,6-a]azepin-3-ylcarbamoylformate (14a-e)

A mixture of **5a-c** (10 mmol) and oxalyl chloride (1.9 g, 15 mmol) in dry benzene (10 ml) was heated under reflux at a temperature 60-70° for about two hours. The solvent and excess oxalyl chloride were evaporated under reduced pressure and the residue was washed with three portions of dry benzene (5 ml) each. The appropriate alcohol (20 ml) was added and the reaction mixture was heated under reflux for about two hours. The solvent was removed by distillation under reduced pressure and the obtained residue was crystallized from aqueous ethanol (Table IV). **14a:** IR (KBr) 3221, 2211, 1738, 1652, 1598. **14b:** IR (KBr) 3277, 2205, 1729, 1603. **14c:** IR (KBr) 3217, 2215, 1735, 1613. **14d:** IR (KBr) 3310, 2216, 1710, 1630. **14e:** IR (KBr) 3250, 2218, 1734, 1620. **14a:** ^{1}H-NMR ($CDCl_3$) δ= 1.35-1.46 (m, C-6(H_2), C-7(H_2), 4H); 1.52(d, CH($\underline{CH_3}$)$_2$, 6H); 1.82-1.93 (m, C-8(H_2), 2C-b(H_2), C-c(H2), 8H); 2.53 (t, 2C-a(H_2), 4H); 3.24 (t, C-9(H_2), 2H); 3.45 (q, C-5(H), 1H); 3.67(s, OCH_3, 3H); 4.13(septet, $\underline{CH}(CH_3)_2$, 1H); 4.52 (d, C-4a (H), 1H); 8.72 (s, NH, 1H). **14c:**^{1}H-NMR (DMSO-d_6) δ= 1.09-1.23 (m, C-6(H_2), C-7(H_2), 4H); 1.63-1.81 (m, C-8(H_2), 2H); 2.45 (t, 2C-a(H_2), 4H); 2.88 (q, C-5(H), 1H); 3.52 (s, O$\underline{CH_3}$, 3H); 3.69 (t, 2C-b(H_2), 4H); 4.19 (t, C-9(H_2), 2H); 4.82 (brs, C-4a(H), 1H); 7.08-7.60 (m, C_6H_5, 5H); 9.38 (s, NH, 1H). **14a:** MS m/z (%): M^+ 417 (4.21), 114 (100).

1N-(4-Cyano-5-morpholino- or piperidino-2-phenyl-1-oxo-1,2,4a,5,6,7,8,9-octahydropyrimido[1,6-a] azepin-3-yl)-2-morpholino- or piperidino-2-oxoacetamide (15a-e)

A mixture of **5b,c** (10 mmol) and oxalyl chloride (1.9 g, 15 mmol) in dry benzene (5 ml) was refluxed at a

temperature 60-70° in a water bath for about two hours. The solvent and excess oxalyl chloride were evaporated under reduced pressure; the formed residue was dissolved in dry benzene (15 ml) and the appropriate amine (11 mmol) was added. The reaction mixture was refluxed for about three hours and evaporated to dryness; the residue was triturated with ethanol, filtered and recrystallized from aqueous ethanol (Table IV). **15a:** 3315, 2220, 1656, 1598. **15b:** IR (KBr) 3209, 2218, 1635, 1596. **15c:** IR (KBr) 3280, 2217, 1649,1599. **15d**: IR (KBr) 3285, 2221, 1655, 1610. **15e:** IR (KBr) 3279, 2215, 1655,1598. **15a:** ^{1}H-NMR ($CDCl_3$) δ= 1.21-1.39 (m, C-6(H_2), C-7(H_2), 4H); 1.96-2.21 (m, C-8(H_2), 2C-b(H_2), 2C-b'(H_2), C-c(H_2), C-c'(H_2),14H); 2.95 (t, 2C-a(H_2), 4H); 3.26 (t, 2C-a'(H_2), 4H); 3.73 (q, C-5(H), 1H); 4.35 (t, C-9(H_2), 2H); 4.73 (d, C-4a(H), 1H); 6.93-7.46 (m, C_6H_5, 5H); 8.54 (s, NH, 1H). **15b:** ^{1}H-NMR ($CDCl_3$) δ= 1.21-1.41 (m, C-6(H_2), C-7(H_2), 4H); 1.89 (s, C_6H_4-$\underline{CH_3}$, 3H); 1.96-2.21 (m, C-8(H_2), 2C-b(H_2), C-c(H_2), 8H); 2.95 (t, 2C-a(H_2), 4H); 3.71(t, C-9(H_2), 2H); 4.30 (q, C-5(H), 1H); 4.83 (brs, C-4a(H), 1H); 7.03-7.66 (m, C_6H_5, C_6H_4, 9H); 9.33 (s, NH,1H). **15e:** ^{1}H-NMR (DMSO-d_6): δ= 1.29-1.59 (m, C-6(H_2), C-7(H_2), 4H); 1.66-1.80 (m, C-8(H_2), 2H); 2.50 (t, 2C-a(H_2),4 H); 3.05 (t, 2C-a'(H_2), 4H); 3.45 (t, C-9(H_2), 2H); 3.73 (t, 2C-b(H_2), 2C-b'(H_2), 8H); 4.35 (q, C-5(H), 1H); 4.80 (d, C-4a(H), 1H); 7.23-7.96 (m, C_6H_5, 5H); 8.83 (s, NH, 1H). **15a:** MS m/z (%): M^+ 504 (2.11), 149 (100).

Alkyl 12-morpholino-5-phenyl-1,6-dioxo-1,2,5,6,8,9,10,11,12,12a-deca-hydropyrimido[4`,5`:4,5]pyrimido [1,6-a]azepine-3-carboxylate(16a,b)

Compounds **14c,d** (10 mmol) were refluxed with sodium ethoxide (sodium metal 0.2 g in ethanol 15 ml) for 10 hours then left to cool at room temperature.The separated crystals were filtered, washed with water and recrystallized from aqueous ethanol (Table VI). **16a:** IR (KBr) 3245, 1734, 1625. **16b:** IR (KBr**)** 3277, 1729, 1603. **16a:** ^{1}H-NMR ($CDCl_3$) δ= 1.12-1.32 (m, C-10(H_2), C-11(H_2), 4H); 1.53-1.86 (m, C-9(H_2), 2H); 2.34 (q, C-12(H), 1H); 2.87 (t, 2C-a(H_2), 4H); 3.16 (t, C-8(H_2), 2H); 3.67 (s, OCH_3, 3H); 4.07 (t, 2C-b(H_2), 4H); 4.43 (d, C-12a(H), 1H): 7.13-7.64 (m, C_6H_5, 5H); 9.22 (s, NH, 1H). **16b:** ^{1}H-NMR (DMSO-d_6) δ= 1.01-1.23 (m, C-10(H_2), C-11(H_2), 4H); 1.47 (t, $OCH_2\underline{CH_3}$, 3H); 1.83-2.01 (m, C-9(H_2), 2H); 2.47 (t, 2C-a(H_2), 4H); 3.13 (t, C-8(H_2), 2H); 3.53 (t, 2C-b(H_2), 4H); 4.31 (q, $O\underline{CH_2}CH_3$, C-12(H), 3H); 4.82 (d,C-12a(H), 1H); 7.16-7.60 (m, C_6H_5, 5H); 9.18 (s, NH, 1H). **16a:** MS m/z (%): M^+ 453 (5.21), 161 (100).

12-Morpholino- or piperidino-5-phenyl-1,6-dioxo-1,2,5,6,8,9,10,11, 12,12a.decahydro-pyrimido[4`,5`:4, 5]pyrimido[1,6-a]azepine-3-sub-stituted carboxamide (17a,b)

Compounds **15c,e** (10 mmol) were refluxed in alcoholic/HCl solution (15 ml) for about three hours then left to

Table VI: Physical and analytical data of the prepared compounds **16a,b** and **17a,b**.

No	R^1	R^2	Yield % M.P°	Mol. formula (M.wt.)	Microanalytical data		
						Calcd.	Found.
16a		CH_3	70 202-5	$C_{23}H_{27}N_5O_5$ 453.49	C H N	60.92 6.00 15.44	60.53 6.20 14.95
b		C_2H_5	65 195-7	$C_{24}H_{29}N_5O_5$ 467.52	C H N	61.66 6.25 14.98	61.44 6.13 15.27
17a			65 165-8	$C_{27}H_{34}N_6O_4$ 506.60	C H N	64.01 6.76 16.59	64.41 6.65 16.81
b			80 180-3	$C_{26}H_{32}N_6O_5$ 508.57	C H N	61.40 6.34 16.52	61.22 6.49 16.73

cool. The separated crystals were filtered and recrystallized from aqueous ethanol (Table VI). **17a:** IR (KBr) 3245 (NH), 1637, 1598 (2CO). **17b:** IR (KBr**)** 3233 (NH), 1621, 1596 (2CO). **17a:** ^{1}H-NMR (DMSO-d_6) δ= 1.01-1.23 (m, C-10(H_2), C-11(H_2), 4H); 1.63-1.81 (m, C-9(H_2), 2C-b'(H2), C-c(H2), 8H); 2.45 (t, 2C-a(H_2), 4H); 3.18 (t, 2C-a'(H_2), 4H); 3.72 (t, C-8(H_2), 2H); 4.20 (quintet, 2C-b(H_2), 4H); 4.76 (q, C-12(H), 1H); 5.27 (d, C-12a(H), 1H); 7.16-7.64 (m, C_6H_5, 5H); 9.25 (s, NH, 1H).

Anti-inflammatory screening

The anti-inflammatory activity of 18 selected newly synthesized compounds **4c**, **5c, 6c, 7c, 8c, 10b,c, 11b**, **12b, 13b, 14c,d,e, 15c,d,e, 16b, 17a** was evaluated using the method of "rat paw carrageenan oedema" as described by Winter *et al*.[4]

Procedure

Adult albino rats of both sexes weighing between 120-150 g were used. Rats were uniformly hydrated by giving 3 ml water/rat through gastric inculation to reduce variability to oedema response. Animals were divided into 20 groups each of six animals. The control group was given saline solution containing few drops of Tween 80. Diclofenac sodium (10 mg/Kg) and drugs under examination (10 mg/Kg) were suspended in distilled water by the aid of few drops of Tween 80 (to improve wettability of the particles) and were given intra-peritoneally one hour before induction of inflammation. Induction of inflammation was performed by s.c injection of 50 µl of 1% carargeenan-sodium gel into the subplantar region. The volume of the inflamed paw was measured just before and three hours after induction of inflammation (Table VII).

Table VII: Anti-inflammatory effects of the tested compounds using the rat paw carrageenan oedema technique.

Compound	Dose mg/kg	Decrease in oedema weight (g)						Mean oedema weight ±SE	% Inhibition
		1	2	3	4	5	6		
Control	0	0.8	0.9	0.5	0.3	0.9	0.9	0.7±0.23	
Diclofenac-Na	10	0.2	0.1	0.2	0.2	0.1	0.2	0.16±0.05	77.1
4c	10	0	0	0.1	0	0.2	0.2	0.08±0.09	88.5
5c	10	0.2	0	0.4	0.2	0.3	0.2	0.21±0.13	70.0
6c	10	0	0.2	0.5	0.4	0.4	0.2	0.28±0.18	60.0
7c	10	0.4	0.3	0.0	0.3	0.4	0.4	0.3±0.15	57.1
8c	10	0.0	0.3	0.1	0.1	0.2	0.1	0.13±0.10	81.4
10b	10	0.2	0.1	0.2	0.2	0.3	0.1	0.18±0.08	74.2
10c	10	0.2	0.2	0.0	0.3	0.4	0.1	0.2±0.13	71.4
11b	10	0.4	0.2	0.4	0.3	0.1	0.2	0.26±0.12	63.3
12b	10	0.4	0.3	0.3	0.2	0.4	0.4	0.33±0.08	53.5
13b	10	0.7	0.3	0.5	0.4	0.3	0.3	0.41±0.16	41.4
14c	10	0.1	0.1	0.1	0.1	0.2	0.1	0.11±0.04	84.2
14d	10	0.1	0	0.2	0.2	0.3	0	0.13±0.12	81.0
14e	10	0.5	0.3	0.1	0.4	0.1	0.3	0.28±0.14	60.0
15c	10	0.3	0.2	0.3	0.3	0.2	0.2	0.25±0.05	64.2
15d	10	0.8	0.3	0.4	0.1	0.3	0.0	0.31± 0.27	55.7
15e	10	0.6	0.4	0.2	0.5	0.2	0.4	0.38±0.16	46.4
16b	10	0.5	0.2	0.6	0.6	0.3	0.0	0.36±0.24	49.2
17a	10	0.4	0.2	0.4	0.3	0.1	0.2	0.26±0.12	55.7

The percentage inhibition of inflammation was calculated according to the following equation:
% Inhibition= $(1- w_t/w_c) \times 100$.

RESULTS AND DISCUSSION

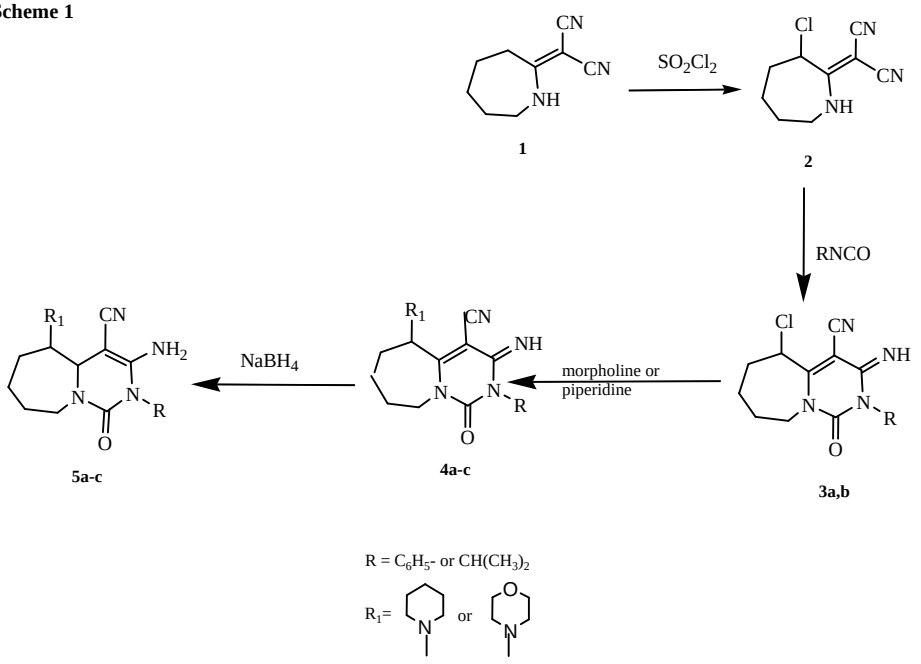
Synthesis

The starting compound 2-dicyanomethylidenoperhydroazepine **1**[3&5] was treated with sulphuryl chloride according to a previously reported method[2&6] yielding the 3-chloro-derivative (**2**).

In previous work, the preparation of the o-iminonitriles from the dicyanomethylidene derivative of azepine was described.[1,3,7&8] Thereby, in the present work refluxing compound **2** with isopropyl or phenyl isocyanate in methylene chloride using triethylamine as a catalyst provided compounds **3a,b**[2] (Scheme 1).

Good yields of the 3-imino-2-isopropyl or phenyl-5-morpholino or piperidino-1,2,3,5,6,7,8,9-octahydro-1-oxopyrimido[1,6-a]azepine-4-carbonitrile **4a-c** were obtained when the chloro derivatives **3a,b** were reacted with either morpholine or piperidine in absolute ethanol.

Scheme 1



The corresponding 3-amino analogues **5a-c** were obtained through 1,4-addition reaction[1,9&10] of one molecule of hydrogen, achieved by the action of sodium borohydride on the 3-imino derivatives **4a-c** in absolute ethanol (Scheme 1).

Preparation of 3-chloromethyl-5-isopropyl or phenyl-12-morpholino or piperidino 1,2,5,6,8,9,10,11,12,12a-decahydropyrimido[4',5',4,5]pyrimido[1,6-a]azepine-1,6-diones **6a-c** was achieved[1,11-14] by refluxing the enaminonitrile derivatives **5a-c** with chloroacetyl chloride, in dry benzene, using triethylamine as a base catalyst. The data drawn from IR spectra of **6** supported the proposed tricyclic structure due to the disappearance of the nitrile absorption band. The suggested mechanism was previously reported.[1&15]

Decomposition of the methyl isothiourea salts, obtained from the interaction of thiourea with the 3-chloromethyl derivatives **6a-c** yielded the corresponding 3-thiomethyl derivatives **7a-c**.

Preparation of the corresponding methyl thioether **8a** was done through the reaction of the thiol, compound **7c** with methyl iodide and anhydrous potassium carbonate, in dry acetone.

The methylthioacetic acid derivatives **8b,c** were prepared through reacting the mercapto derivatives **7a**,**c** with chloroacetic acid and potassium hydroxide, in absolute ethanol. Moreover, refluxing **7b** with ethylchloroacetate and anhydrous potassium carbonate in dry acetone afforded the methyl-thioacetate derivative (**9**).

A simple method was done to prepare the thioesters **10a-c** through refluxing the thiol, compounds **7b,c** with acetyl or benzoyl chloride, in dry benzene and using triethylamine as a catalyst (Scheme 2).

Applying our previous method of chloroacylation, the enaminonitriles **5b,c** were reacted with chlorobutyryl chloride in dry benzene, using triethylamine as a catalyst, affording unexpectedly the uncyclized 3-(4-chlorobutanamide) derivatives **11a,b**. Evidence that intramolecular cyclization didn't occur was drawn from the fact that IR scanning of the obtained products showed a strong and sharp nitrile stetching band at 2205 cm^{-1}.

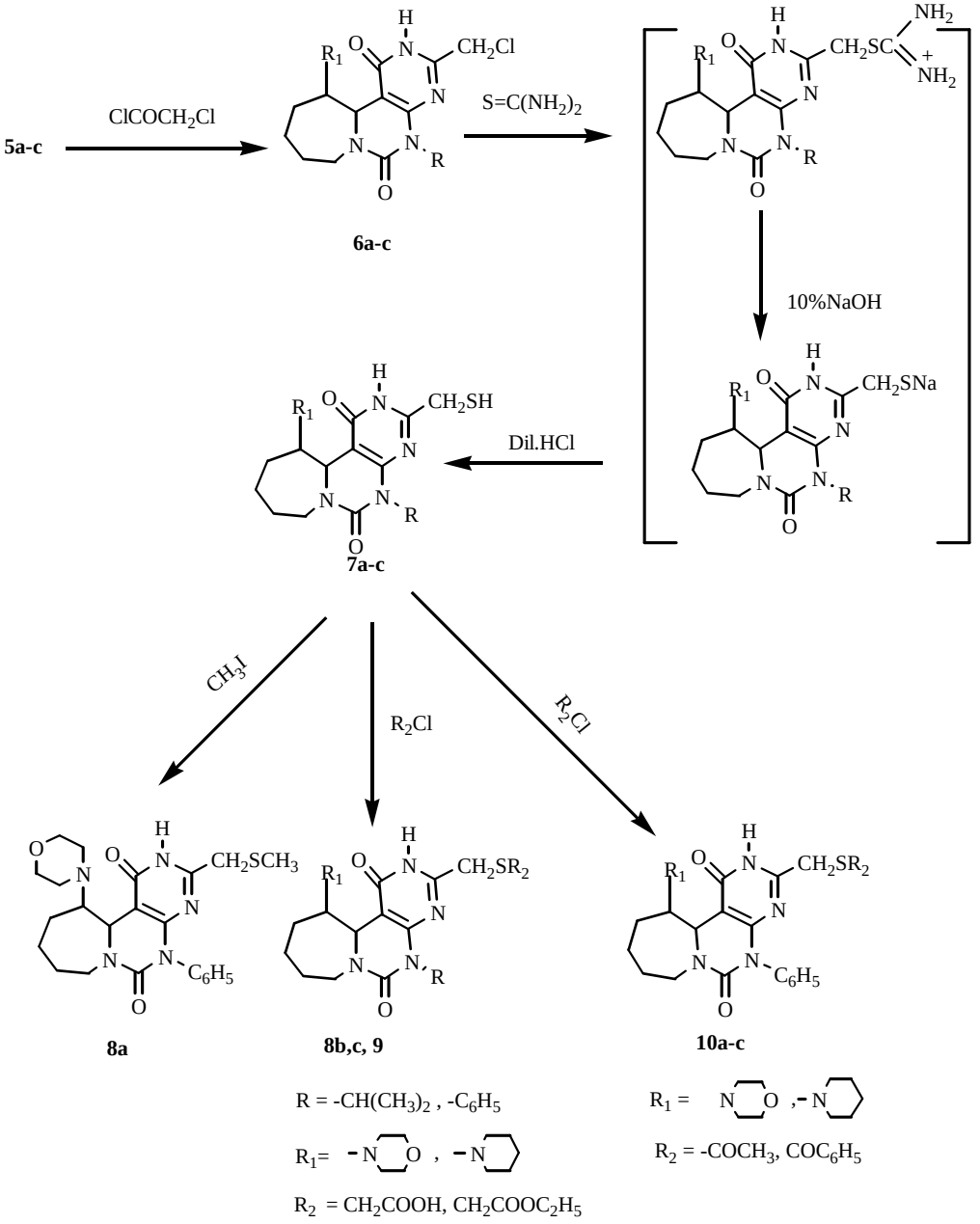
Thereby, in a successful trial to obtain the tricyclic derivatives, compounds **11a,b** were refluxed with ethanolic hydrochloric acid solution for three hours yielding 3-chloropropyl-12-morpholino or piperidino-5-phenyl-1,2,5,6,8,9,10,11, 12,12a-decahydropyrimido[4',5':4,5] pyrimido[1,6-a]azepine-1,6-diones, **12a,b**.The proposed structure of **12** was inferred from their IR spectra, which revealed the absence of the cyano group.

Furthermore, refluxing the tricyclic compounds **12a,b** with different alicyclic secondary amines as well as primary aromatic amines afforded[16] a single crystalline compound identified as the tetracyclic ring system pyrrolopyrimido-pyrimido-azepines **13a,b** as confirmed by physical properties, thin layer chromatography, elemental, as well as spectral analyses (Scheme 3). The lack of a stretching band around 3200 cm^{-1} gave us a support for the intramolecular cyclization onto the imidic nitrogen leading to formation of the tetracycles. Additionally, the ^{1}H-NMR spectrum of **13a** revealed a triplet at 2.89 ppm, quintet at 1.75 ppm and a triplet at 1.56 ppm related to the methylene protons of the pyrrole ring as compared to a triplet at 3.62 ppm, quintet at 1.82 ppm and a triplet at 1.42 ppm related to the corresponding methylene protons of the 3-chloropropyl moiety observed in the ^{1}H-NMR scan of **12a**. Moreover, mass spectrum of **13b** showed the molecular ion peak at 434.1 constituting the molecular weight of the compound -1.

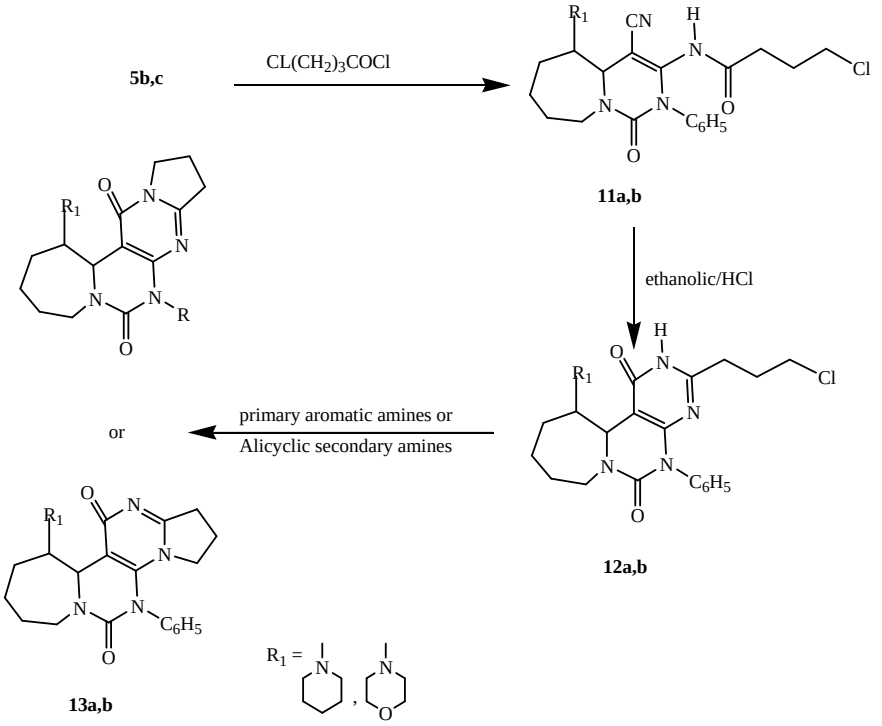
In the above reaction two possible structural isomers (linear and angular) could be obtained.

As previously discussed and similar to other acid chlorides, oxalyl chloride was reacted with the enaminonitriles **5a-c** in dry benzene aiming for an additional fused pyrimidine ring as previously reported.[17] However, the high instability and rapid hydrolysis of the formed chlorocarbonyl derivatives made their isolation difficult. Accordingly, the crude products, obtained from the reaction of **5a-c** with oxalyl chloride, were smoothly reacted with different alcohols yielding crystalline compounds identified, unexpectedly, as the uncyclized carbamoylformate derivatives **14a-e** as drawn from the IR spectra of these compounds which demonstrated a strong nitrile absorption band.

Scheme2


5a-c
ClCOCH2Cl
6a-c
S=C(NH2)2
10%NaOH
Dil.HCl
7a-c
CH3I
R2Cl
R2Cl
8a
8b,c, 9
10a-c
R = -CH(CH3)2 , -C6H5
R1= -N O , -N
R2 = CH2COOH, CH2COOC2H5
R1 = N O , -N
R2 = -COCH3, COC6H5

Scheme 3



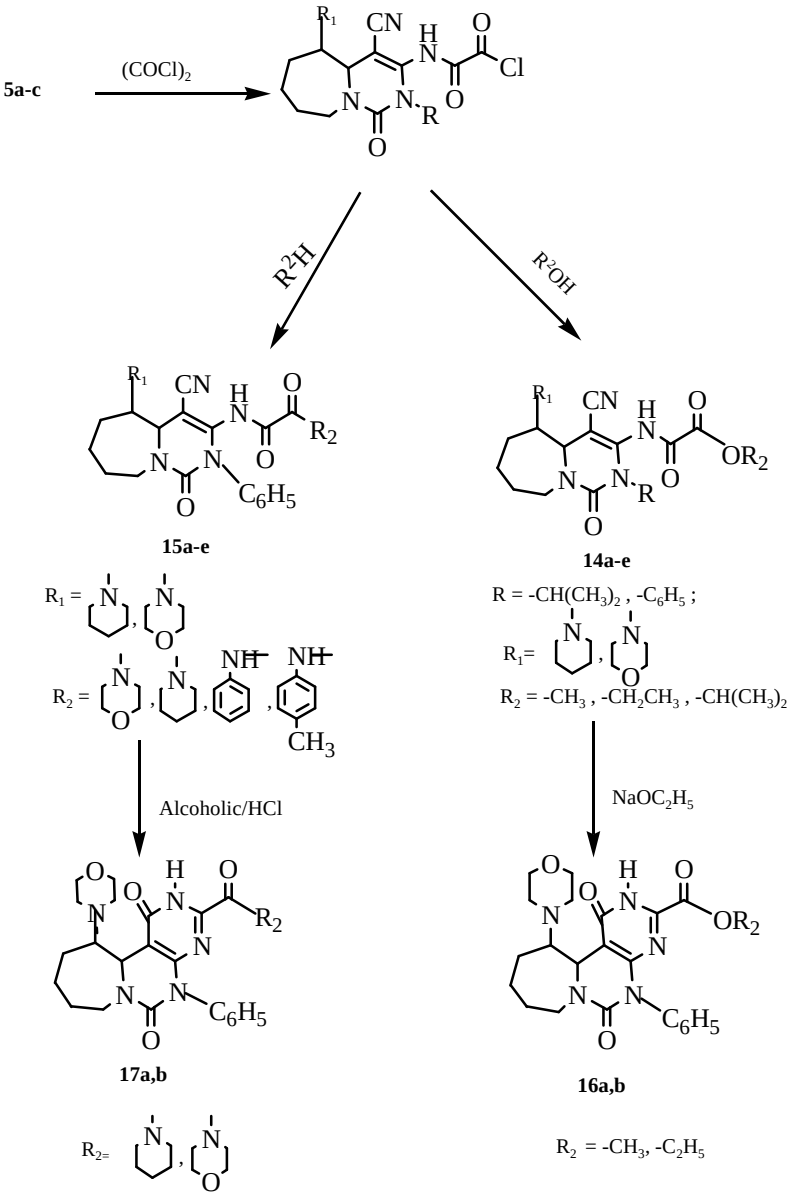
Furthermore, amide formation was carried out via the reaction of the acid chloride intermediates with excess of the appropriate amine using dry benzene, as a solvent, to produce the corresponding N-substituted-2-oxoacetamide derivatives **15a-e**. The IR spectra of these compounds revealed the presence of the characteristic nitrile stretching band.

The carbamoylformate derivatives **14c,d** were cyclized via refluxing with sodium ethoxide to produce the desired tricycles **16a,b**. Moreover, preparation of N-substituted carboxamides **17a,b** was achieved when the corresponding N-substituted-2-oxoacetamide derivatives **15c,d** were refluxed with alcoholic solution of hydrochloric acid (Scheme 4). IR spectra of compounds **16** and **17** were complying with their structures.

Pharmacological properties

Application of one way ANOVA statistical test on the above tabulated data as well as the graphical representation (Fig. 1) of these data indicate that all the 18 newly synthesized compounds have significant anti-inflammatory effect at $P<0.05$. On the other hand, significant activity at $P<0.01$ was assigned to all the tested compounds except **13b, 15e** and **16b**.

Scheme 4


5a-c
(COCl)2
R2H
R2OH
15a-e
14a-e
R1 = , ,
R2 = , , , ,
R = -CH(CH3)2 , -C6H5 ;
R1= , ,
R2 = -CH3 , -CH2CH3 , -CH(CH3)2
Alcoholic/HCl
NaOC2H5
17a,b
16a,b
R2= , ,
R2 = -CH3, -C2H5

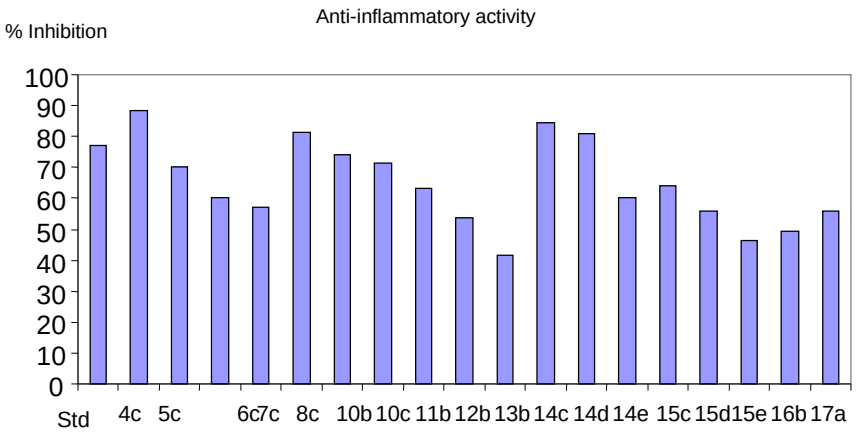


Fig. 1: Graphical representation of the data.

It is noteworthy that compounds **4c, 8c, 14c** and **14d** displayed potential anti-inflammatory effect as compared to the standard (diclofenac-sodium).

Molecular modeling and structure-activity relationship (SAR) of the newly prepared compounds

A comparative measurement of torsion angles of the well known active compounds **I, II** and **III** and that of the prepared compounds **4c, 5c, 6c, 7c, 8c, 10b,c, 12b, 13b, 14c,d,e 15c,d,e, 16b, 17a** revealed certain 3D structural similarities. A comparison of the electrostatic potentials of these compounds have been constructed and presented. For the sake of molecular modelling study HyperChem software[18] had been used. The structure models were generated, then fully minimized to obtain the optimum structures using RMS gradient of 0.1 and Fletcher-Reeves algorithm. The electrostatic potential maps were constructed first by calculating the charges by Modified Neglectof Differential Overlap (MNDO) method. The potential maps were calculated derived from a single point charge models at horizontal grid points= 100 and vertical grid points= 100 and contour level of 50 and an increment of 0.1. The obtained isopotential surfaces were moved in space together with the underlying molecules in order to obtain maximum overlap.

Torsion angles of atoms 5-4a-4-C(CN), H5-5-4a-4, 12-12a-12b-CO and H(Cl)(N)-12-12a-12b and the corresponding activity for the chosen compounds are presented in Tables I and II.

It has been observed that the tested compounds fall into two main categories. The first comprises the bicyclic ring system pyrimido [1,6-a] azepine derivatives while the tricyclic counterparts, the pyrimido[4',5':4,5] pyrimido[1,6-a]azepines, represent the second one. Within the first series, it was observed from the data obtained, that introduction of a morpholine moiety at position 5 of the bicyclic pyrimido[1,6-a]azepine ring system **I** resulting in formation of compound **4c** caused significant increase in twisting of torsion angles 5-4a-4-C(CN) and H5-5-4a-4 accompanied with improved activity.

On the other hand, reduction of the imino group at position 3 of compound **4c** resulting in the formation of the corresponding 3-amino derivative **5c** led to decreased activity to a large extent probably due to the decrease of twisting of torsion angles as indicated by molecular modeling study.

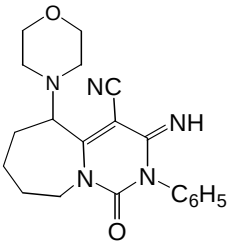
Different substitution on the amino derivative **5** was done in order to see the influence of these changes on the torsion angles and hence on the activity. When substitution of this amino group was effected with different ester or amide carbonyl group yielding compounds **14c,d,e** and compounds **15c,d**; no significant difference in the torsion angles was observed**.** Nevertheless, the difference in activity noted may be due to difference in the electronic factors.

In view of the results obtained for the second series, it was observed that torsion of angle 12-12a-12b(CO) increases upon insertion of chlorine atom at position 12 of the tricycle **II** leading to great improvement in activity of the product **III**. The tricycle **6c** bearing morpholine moiety at position 12 (instead of the chlorine atom) retained activity within significant range. Further substitution of the tricycle **6c** at position 3 with different reagents or subjecting it to cyclization was effected in order to see the relevance of these changes on the torsion angles and hence on activity. With substitution no significant difference in torsion angles was perceived accompanied with difference in activity. To our interest, compound **8c** bearing acetic acid moiety at position 3 showed improved activity as compared to the standard.

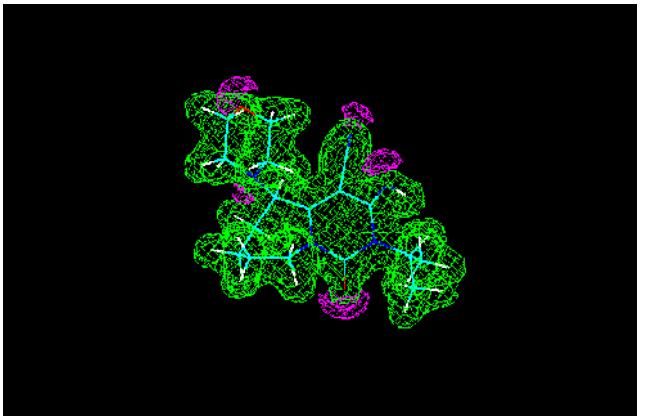
Additionally, cyclization of the 3-chloropropyl derivative **12b** resulting in the formulation of the tetracycle **13b** led to decreased activity; whilst no significant change in angle 12-12a-12b-CO was observed, a slight decrease in twisting of angle H(Cl)(N)-12-12a-12b was noted.

Finally, the bicyclic carbamoyl ester and amide derivatives **14c**, **15c** revealed higher twisting of torsion angle 5-4a-4-C(CN) and higher activity compared to their cyclized counterparts **16b, 17a** confirming our concept.

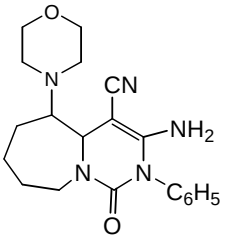
3D Electrostatic potential maps of some selected compounds



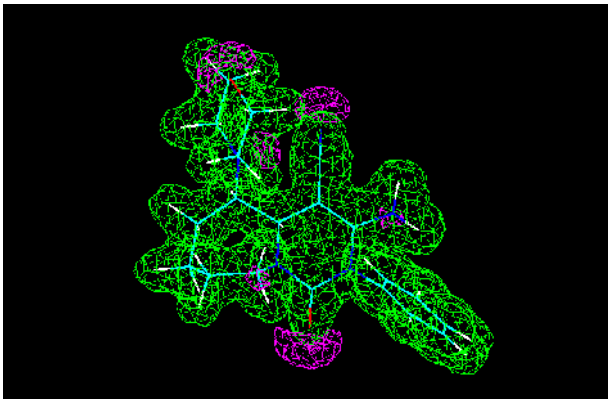
(4c)



3D Electrostatic potential map of compound (4c) derived from a single point charge model using MNDO charges – 5kcal/mol green, +5kcal/mol pink
Torsion angle C = - 7.98822°
Torsion angle D = - 88.6645°

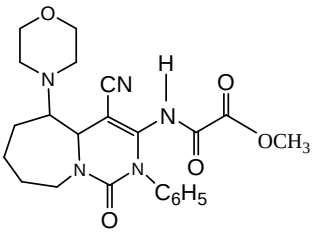


(5c)

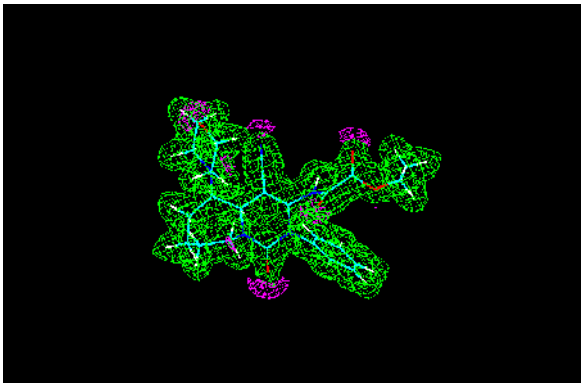


3D Electrostatic potential map of compound (**5c**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle C = 3.4387°
Torsion angle D = - 58.8528°

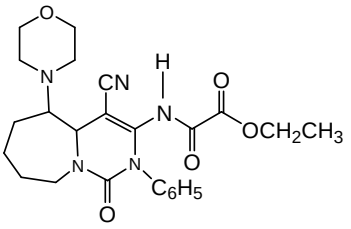
N.B.: For simplicity letter A will be used to express torsion angle at atom : 12-12a-12b-C(CO) , letter B to express the torsion angle at atoms H(Cl)(N)-12-12a-12b, letter C to express torsion angle at atoms 5-4a-4-C(CN) and letter D to express torsion angle at atoms H(N)-5-4a-4.



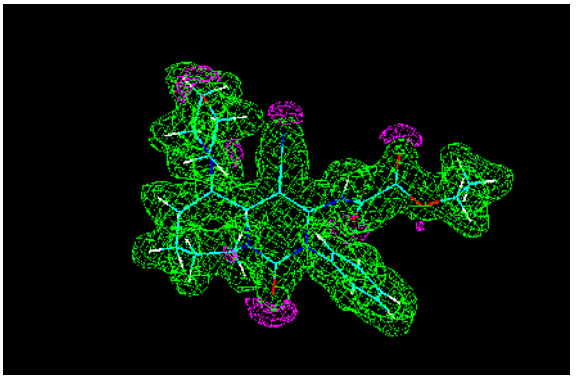
(14c)



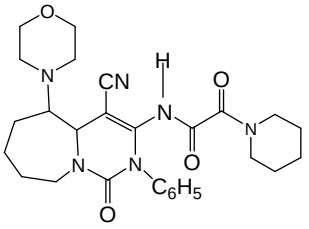
3D Electrostatic potential map of compound (**14c**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle C = 2.71576^{o}
Torsion angle D = - 57.5781^{o}



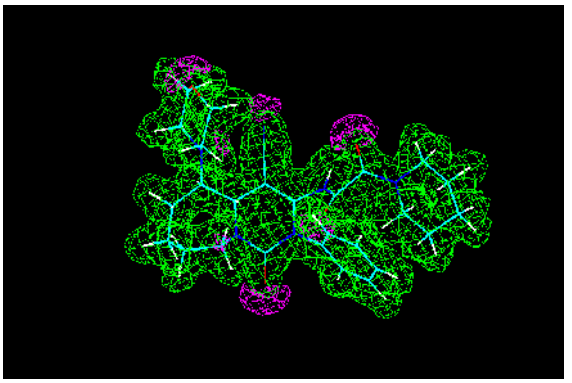
(14d)



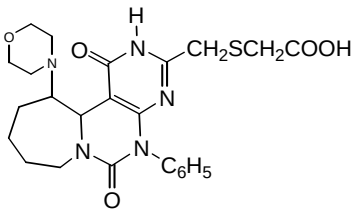
3D Electrostatic potential map of compound (**14d**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle C = 3.41576^{o}
Torsion D = - 56.5781^{o}



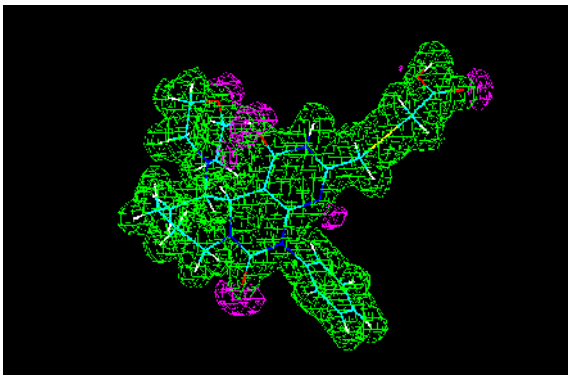
(15c)



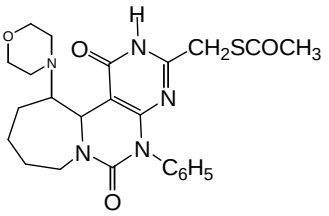
3D Electrostatic potential map of compound (**15c**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle C = 5.50074°
Torsion angle D = - 58.5448°



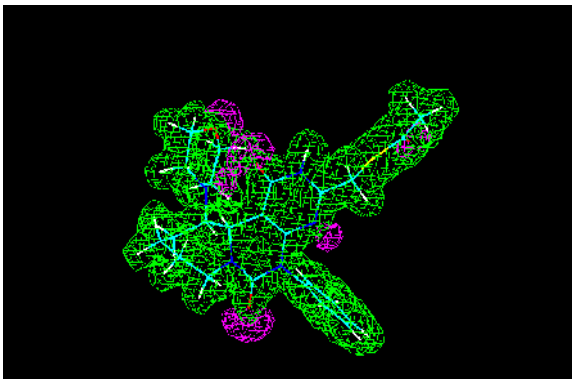
(8c)



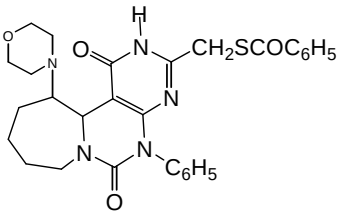
3D Electrostatic potential map of compound (**8c**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle A = 88.9228°
Torsion angle B = - 43.1943°



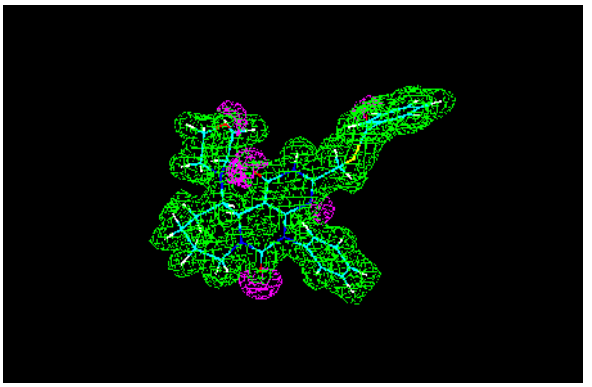
(10b)



3D Electrostatic potential map of compound (**10b**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle A = 89.4528°
Torsion angle B= - 51.7408



(10c)



3D Electrostatic potential map of compound (**10c**) derived from a single point charge model using MNDO charges – 5 kcal/mol green, +5 kcal/mol pink
Torsion angle A = 89.8137°
Torsion angle B = - 52.8788°

REFERENCES

1- M.Y. Ebeid, Z. I. Gad, N. M. El-Sayed, EL. S. Ahmed and B. El-Sayeh, Egypt. J. Pharm. Sci., 32, 441 (1991) (and references therein).

2- M.Y. Ebeid, H. H. Hassanein and N. Obidan, Bull. Fac. Pharm. Cairo, Univ., 33**,** special issue, 35 (1995).

3- M. Ebeid and I. Bitter, Egypt. J. Pharm. Sci., 19, 349 (1978).

4- C. A. Winter, E. A. Risley, G. W. Nuss, Proc. Soc. Exp. Biol. Med., 111, 544 (1962).

5- P. Schalck and J. Rieker, Angew. Makromol. Chem., 15, 203 (1971).

6- N. M. El-Sayed and M. M. Hussein, Egypt. J. Pharm. Sci., 44, 319 (2003)

7- N. M. El-Sayed and H. A. Abdel Latif, Bull. Fac. Pharm. Cairo, Univ., 31, 55 (1993).

8- N. M. El-Sayed, D. A. Abou El Ella, A. E. Farag, M. Y. Ebeid and N. A. Monieb, ibid., 38**,** 19 (2000).

9- A. I. Mayers and N. Nazarenko, J. Am. Chem. Soc., 94, 3243 (1972).

10- O. A. Reutov, "Fundamentals of Theoritical Organic Chemistry", 2nd, Scripta Technica Inc., Appleton century Crofts, New York, 1967, p. 216.

11- M. T. Bogert and W. F. Hand, J. Am. Chem. Soc., 24, 1031 (1902).

12- M. Y. Ebeid, N. M. El-Sayed and El-S. Ahmed, "Condensed Pyrimidines XII". Proceedings of XXI Conference of Pharmaceutical Sciences, 1990, pp. 105-112.

13- E. C. Taylor and A. McKillop, “The Chemistry of Cyclic Enaminonitriles and o-Aminonitriles”,7, Interscience Publishers, a Division of John Willey and Sons, New York, London, Sydney, Toronto, 1970, p. 227.

14- E. C. Taylor and Y. Shvo, J. Org. Chem., 33, 1719 (1968).

15- A. A. Harb, Egypt. J. Pharm. Sci., 33, 283 (1992).

16- N. M. El-Sayed, M. M. Hanna, N. M. Abdel Gawad, H. H. Hassanein and M. Y. Ebeid, Bull. Fac. Pharm., Cairo Univ., 31, 171 (1993).

17- M. Y. Ebeid, H. H. Hassanein and M. Riad, ibid., 31**,** 187 (1993).

18- HyperChem ™ version 6.03. Molecular modelling system (2000).