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SYNTHESIS OF NOVEL STEROIDAL 1, 2-DISUBSTITUTED PYRIDAZINE-3, 6-DIONES: A NEW ENTRY TO NITROGEN HETEROCYCLES.

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ABSTRACT

Condensation products obtained from Cholesterol and stigmasterol with 3, 6-dichloropyridazine, underwent double Chapman rearrangement under conventional as well as microwave irradiation to afford corresponding N-substituted heterocyclic compounds.

KEYWORDS: Cholesterol, stigmasterol, steroidal, pyridazine, imidates, microwave irradiation, Chapman rearrangement.

INTRODUCTION

Number of steroidal heterocyclic compounds have been reported to posses various therapeutic activities.[1] Pyridazine ring represents molecular frameworks that serve as a platform for developing pharmaceutical agents for various applications.[2]

It is expected that the fusion of heterocycles with steroids led to a change in their physiological activities and the appearance of new interesting biological behavior. Thus, several steroidal heterocycles have been obtained exhibiting activity as potential inhibitors of cytochrome P450 enzyme aromatase.[3]

Steroidal heterocycles containing anellated rings in the steroidal moiety have prompted a great interest in the biological study of many heterocyclic compounds. New compounds were synthesized in which the androstane ring A was condensed with a great variety of heterocyclic rings.[4, 5, 6, 7] A steroid containing a ring-A fused pyridazone is also described.[8] The reaction of 4-amino-1, 2, 4-triazole with the 2-hydroxymethylene-3-oxo-Δ^4-steroid and three 16-hydroxymethylene-17-oxo-steroids afforded androst-4-eno[3, 2-*f*]-(*s*-triazolo [4, 3-

b] pyridazine), androstano[17, 16-*f*]-(*s*-triazolo [4, 3-*b*]pyridazines) and 3-methoxyestra-1, 3, 5-(10)-trieno[17, 16-*f*]-(*s*-triazolo[4, 3-*b*]pyradazine) respectively. Although conditions favourable for Dimroth Rearrangement were employed, no such transformations were encountered during the synthesis of the steroidal triazolopyridazines.[9]

Many steroidal 1, 4-diketone derivatives were synthesized by acid-catalyzed condensation of 2-acetylestradiol-17-beta-acetate with substituted phenylglyoxals. Conversion of the products into the corresponding pyridazine derivatives was achieved by reaction with hydrazine hydrate. The synthesized compounds were evaluated for their uterotrophic, antiuterotrophic, and antifertility activities.[10]

Condensation of 5-α-androstan-17-β-ol-3-one with glyoxylic acid in the presence of base yields the corresponding 2-carboxy-2-hydroxymethyl derivative, which readily lactolizes upon treatment with acid to give the ring A-fused lactone (III). Acetylation of this compound yields the corresponding diacetate which is converted by refuxing in hydrazine solution to the ring A-fused pyridazone having an acetate group at 17 position. Mild alkaline hydrolysis converts the acetate to the corresponding free hydroxyl derivative.[11]

Attempts at direct N-substitution of 3, 6-pyridazinedione derivatives are tedious and involve substrates that are not easily accessible.[12]

Although most of the methodologies have their own synthetic values, some limitations mainly due to the use of as solvent like dimethyl formamide with cumbersome workup of the reaction mixture, the long reaction times, harsh reaction conditions, tedious preparation procedures; which could represent significant drawbacks for preparative purposes. Thus there is scope for their synthesis by simple and eco-friendly method. The present invention describes the synthesis of 1, 2-disterylpyridazine-3, 6-dione derivatives in two steps through double *Chapman rearrangement* under microwave irradiation in absence of solvent in second step.

MATERIALS AND METHODS

The melting points were determined using capillary tube and are uncorrected. The FTIR spectra were recorded on Spectrum One Perkin Elmer (US). The 1H-NMR spectra were recorded on a Bruker AVANCE (300MHz) spectrometer (with TMS as internal references). 13C-NMR spectra were recorded on Bruker AVANCE (75 MHZ) spectrometer. Mass spectra

were recorded on API-3000MD-series (US). UV spectra were recorded on Shimadzu 2401 PC and Shimaduz 2450, Japan, Spectrophotometer. Elemental analyses were carried out in EA 3000, Euro Vector, Italy. The purity of the compounds was checked by TLC on pre-coated SiO_2 gel (200mesh). Modified LG microwave laboratory oven was used for microwave irradiation. The solvents were purified by distillation before use.

RESULTS AND DISCUSSION

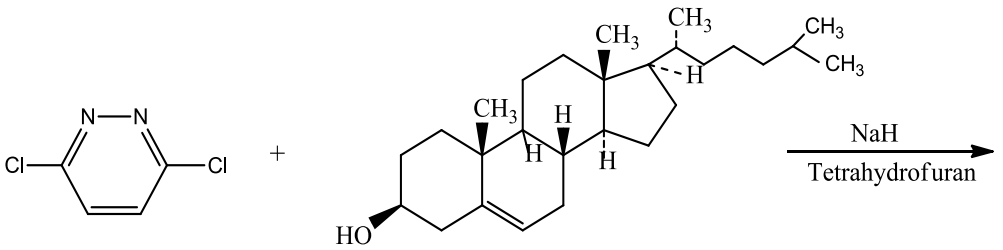
The present paper reports synthesis of 1, 2-dicholesterylpyridazine-3, 6-dione and 1, 2-distigmasterylpyridazine-3, 6-dione by subjecting 3, 6-dicholesteroxypyridazine and 3, 6-distigmasteroxypyridazine respectively to double *Chapman rearrangement*.

The thermal conversion of aryl N-arylbenzimidates to N-aroyldiphenylamines is known as the *Chapman rearrangement.*[13] Though imidates of many classes of compounds have been subjected to *Chapman rearrangement*, 3, 6-distigmasteroxypyridazine and 3, 6-dicholesteroxypyridazine have not been investigated.

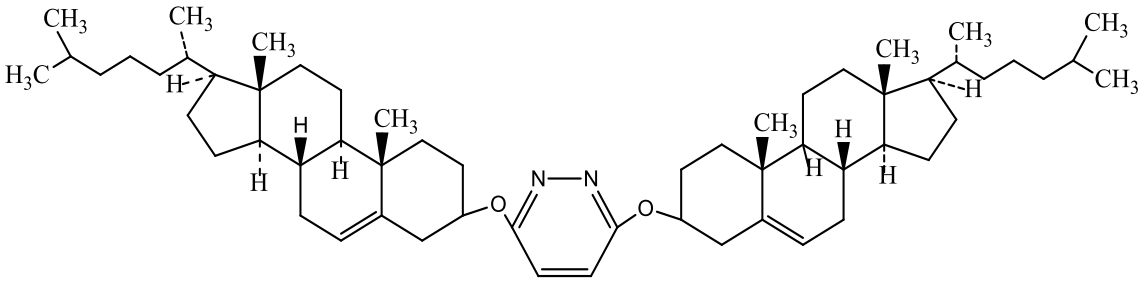
In light of the observations from literature survey as well as our interest in evolving new, simpler, ecofriendly, convenient methodologies in organic synthesis and absence of reports on the *Chapman rearrangement* of 3, 6-distigmasteroxypyridazine and 3, 6-dicholesteroxypyridazine led us to undertake the present work in continuation with earlier work.[14, 15, 16, 17, 18, 19,20, 21]

For this purpose, 1, 2-dihydropyridazine-3, 6-dione was visualized as the starting substrate. This on chlorination followed by condensation with cholesterol **(2a)** and stigmasterol **(2b)** yielded the respective imidates, **3a and 3b**. These were then subjected to double *Chapman rearrangement* to afford the corresponding 1, 2-di-(cholesteryl)-pyridazine-3, 6-dione **(4a)** and 1, 2-di-(stigmasteryl)-pyridazine-3, 6-dione **(4b)** respectively. **(Scheme 1 and 2)** 3, 6-dichloropyridazine **(1)** has been synthesized as per literature procedure from 1, 2-dihydroxypyridazine-3, 6-dione.[22]

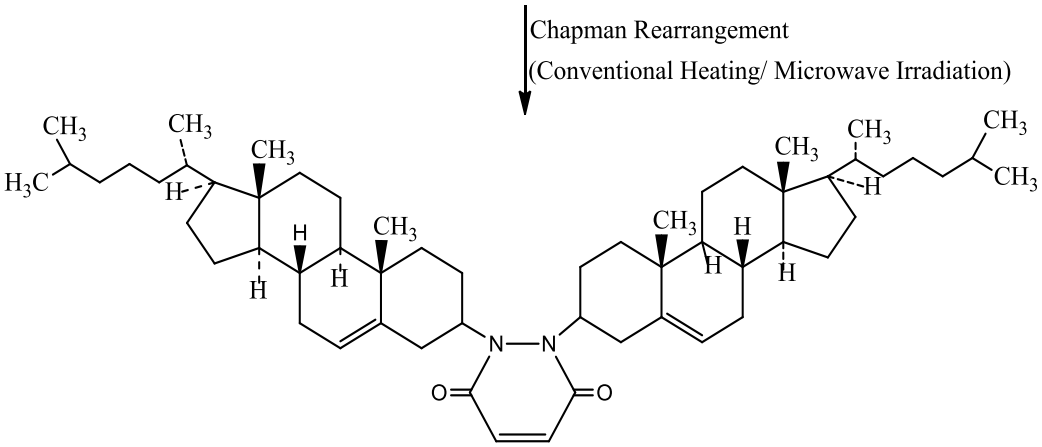
Scheme1: 1, 2-di-(cholesteryl)-pyridazine-3, 6-dione (4a)



(1) Cholesterol (2a)

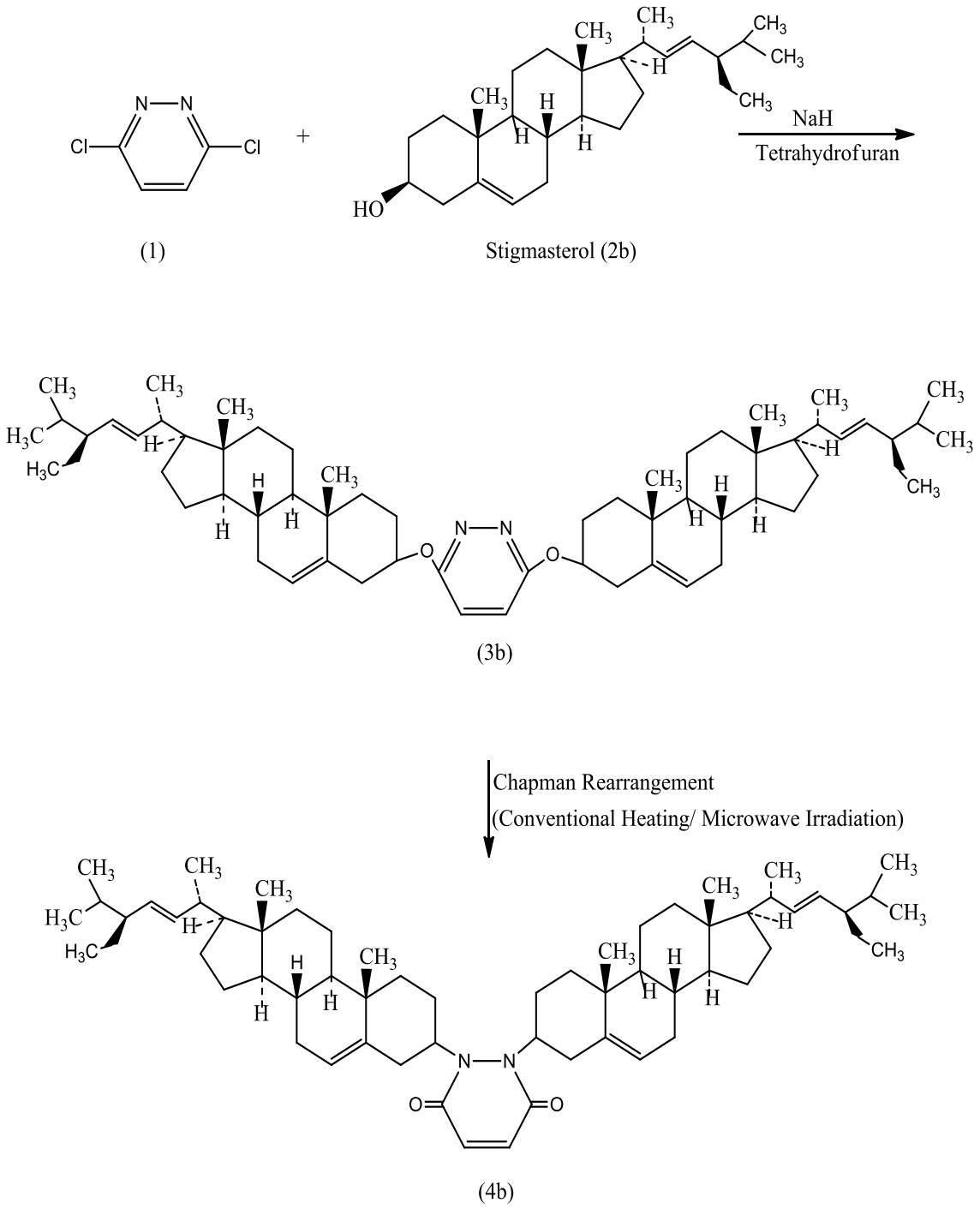


(3a)



(4a)

Scheme 2: 1, 2-di-(stigmasteryl)-pyridazine-3, 6-dione (4b)



General Procedure for preparation of 3a and 3b.

To a solution of cholesterol **(2a)** / stigmasterol **(2b)** (0.025M) in dry tetrahydrofuran (100 ml), Sodium hydride (0.03M, 60% dispersion in oil) is added in portions to the flask under nitrogen atmosphere. The mixture was gently refluxed for 30 minutes followed by cooling to room temperature. A solution of 3, 6-dichloropyridazine (0.025M) **(1)** in tetrahydrofuran is added dropwise under stirring in 15-20 minutes. Tetrabutylammonium iodide (0.0025M) was

added in one portion and the solution is stirred at 50°C-60°C for 8-10 hours. After completion of the reaction (TLC), the solvent was removed under reduced pressure and the residue was treated with of saturated NaCl solution (50 ml). The mixture was extracted with dichloromethane (4× 25 ml) and the combined extracts were dried over anhydrous sodium sulfate. The solvent was removed under reduced pressure the residue was flash chromatographed (chloroform:petroleum ether, 20:80) to afford 3, 6-distigmasteroxypyridazine **(3a) /** 3, 6-distigmasteroxypyridazine **(3b)** as viscous oil.

3, 6-dicholesteroxypyridazine (3a).

Yield: 58%. Oil, ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.69 (m, 6H), 0.87 (m, 6H), 0.88 (m, 6H), 0.93 (m, 6H), 0.98 (m, 6H), 1.02 (m, 6H), 1.13 (m, 6H), 1.32 (m, 10H), 1.51 (m, 8H), 1.84 (m, 12H), 1.99 (m, 8H), 2.26 (m, 4H), 3.54 (m, 2H), 5.37 (m, 2H), 6.43 (s, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 12.01, 19.02, 19.71, 22.02, 22.82, 22.87, 23.32, 24.51, 26.89, 28.32, 28.73, 30.87, 32. 42, 36.02, 36.01, 36.63, 37.59, 39.23, 39.87, 41.32, 50.27, 55.59, 57.03, 75.11, 103.42, 122.21, 131.77, 156.83. IR (KBr, cm^{-1}): 639, 786 (-CH bend. Ar), 839, 928 (=C-H bend. Alkene), 1032, 1061, 1105 (C-H cycloalkane), 1197 (C-O-C stretch.), 1337 (C-N stretch.), 1360, 1381, 1462 (C-H bend. Alkane), 1635 (C=C stretch. Ar), 1671 (C=C stretch. Alkene), 2871-2926 (-CH stretch. Aliphatic). UV spectrum:λ_{max} 221 Abs. 0.297. Optical activity: $[\alpha]^{20}_{D}$ = -21.62. HRMS: *m/z* cal. mass for $C_{58}H_{92}N_2O_2$ $[M+H]^+$ =849.3632, obs. mass $[M+H]^+$ = 849.3640. Molecular formula: $C_{58}H_{92}N_2O_2$. Elemental analysis: Calculated: C (82.08%), H (10.85%), N (3.30%). Found: C (82.15%), H (10.72%), N (3.18%).

3, 6-distigmasteroxypyridazine (3b).

Yield: 61%. Oil, ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.93(m, 6H), 1.04(m, 6H), 1.08(m, 6H), 1.22(m, 6H), 1.29(m, 6H), 1.37(m, 6H,), 1.62 (m, 16H), 1.91 (m, 16H), 1.97 (m, 14H), 2.40 (m, 6H), 4.17(m, 2H), 4.19(m, 2H), 5.20(m, 2H), 6.39 (s, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 12.09, 13.11, 17.06, 19.32, 22.12, 24.32, 27.02, 28.43, 31.02, 32.96, 37.25, 40.98, 42.32, 49.45, 52.40, 57.04, 58.39, 71.07, 102.61, 121.89, 127.57, 130.56, 140.78, 157.61. IR (KBr, cm^{-1}): 661, 769 (-CH bend. Ar), 904, 1038 (=C-H bend. Alkene), 1069, 1093 (C-H cycloalkane), 1188 (C-O-C stretch.), 1343 (C-N stretch.), 1465 (C-H bend. Alkane), 1607 (C=C stretch. Ar), 1652 (C=C stretch. Alkene), 2831-2942 (-CH stretch. Aliphatic). UV spectrum:λ_{max} 214 Abs. 0.284. Optical activity: $[\alpha]^{20}_{D}$ = -23.64. HRMS: *m/z* cal. mass for $C_{62}H_{96}N_2O_2$ $[M+H]^+$ = 901.4436, obs. mass $[M+H]^+$ = 901.4429. Molecular formula:

$C_{62}H_{96}N_2O_2$. Elemental analysis: Calculated: C (82.67%), H (10.67%), N (3.11%). Found: C (82.74%), H (10.74%), N (3.04%).

Procedure for preparation of 1, 2-dicholesterylpyridazine-3, 6-diones (4a) and 1, 2-distigmasterylpyridazine-3, 6-diones (4b) by double *Chapman rearrangement* of 3, 6-dicholesteroxypyridazine (3a) and 3, 6-distigmasteroxypyridazine (3b) under conventional heating.

In a flask, equipped with water condenser 3, 6-dicholesteroxypyridazine **(3a)** / 3, 6-distigmasteroxypyridazine **(3b)** (0.01M) was heated in nitrogen atmosphere at 180^{o}C-190^{0}C for 80-100 minutes. After completion, (TLC) the reaction mass was cooled to room temperature and petroleum ether (25 ml) was added. It was purified to afford viscous oil.

Thus, compounds **3a** and **3b** smoothly underwent double *Chapman rearrangement* but the reaction times were larger and percentage yields were moderate. It was therefore thought worthwhile to carryout the double *Chapman rearrangement* of these compounds under microwave irradiation.

Reduced reaction times, less effect on the environment and better reaction yields are some of the common advantages of using microwave irradiation for chemical reactions.[23]

Procedure for preparation of 1, 2-dicholesterylpyridazine-3, 6-diones (4a) and 1, 2-distigmasterylpyridazine-3, 6-diones (4b) by double *Chapman rearrangement* of 3, 6-dicholesteroxypyridazine (3a) and 3, 6-distigmasteroxypyridazine (3b) under microwave irradiation.

In a flask, equipped with water condenser 3, 6-dicholesteroxypyridazine **(3a)** / 3, 6-distigmasteroxypyridazine **(3b)** (0.01M) was irradiated (900 W) in a microwave oven for 17-20 minutes. After completion (TLC), the reaction mass was cooled to room temperature and petroleum ether (25 ml) was added under stirring. It was purified to afford viscous oil.

Percentage Yield and reaction time under conventional heating and microwave irradiation are presented in the **Table**.

1, 2-di-(cholesteryl)-pyridazine-3, 6-dione (4a).

Viscous oil, ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.59 (m, 6H), 0.80 (m, 6H), 0.87 (m, 6H), 0.92 (m, 6H), 0.97 (m, 6H), 1.03 (m, 6H), 1.13 (m, 6H), 1.30 (m, 10H), 1.54 (m, 8H), 1.83 (m, 12H), 1.94 (m, 8H), 2.34 (m, 4H), 3.60 (m, 2H), 5.37 (m, 2H), 6.41(s, 2H) ^{13}C NMR (75

MHz, $CDCl_3$): δ 11.53, 19.32, 20.03, 20.81, 22.52, 22.77, 23.53, 24.61, 27.06, 27.68, 28.82, 30.81, 32.14, 35.42, 36.21, 36.75, 37.64, 38.91, 39.92, 42.53, 51.05, 55.72, 57.51, 74.01, 101.53, 121.07, 142.17, 134.93, 153.04. IR (KBr, cm^{-1}): IR (KBr, cm^{-1}): 668, 781(-CH bend. Ar), 846, 931(=C-H bend. Alkene), 1029, 1053, 1112 (C-H cycloalkane), 1333 (C-N stretch.), 1371, 1379, 1471(C-H bend. Alkane), 1611 (C=C stretch. Ar), 1615 (C=C stretch. Alkene), 1692 (N-C=O stretch.), 2873-2930 (-CH stretch. Aliphatic). UV spectrum: λ_{max} 253 Abs. 0.341. Optical activity: $[\alpha]^{20}_D$ = -19.74. HRMS: *m/z* cal. mass for $C_{58}H_{92}N_2O_2$ $[M+H]^+$ =849.3632, obs. mass $[M+H]^+$ = 849.3641. Molecular formula: $C_{58}H_{92}N_2O_2$. Elemental analysis: Calculated: C (82.08%), H (10.85%), N (3.30%). Found: C (81.98%), H (10.91%), N (3.19%).

1, 2–di-(stigmasteryl)-pyridazine-3, 6-dione (4b).

Viscous oil, ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.98 (m, 6H), 1.02 (m, 6H), 1.05 (m, 6H), 1.19 (m, 6H), 1.31 (m, 6H), 1.37 (m, 6H,), 1.52 (m, 16H), 1.94 (m, 16H), 1.99 (m, 14H), 2.38 (m, 6H), 4.15 (m, 2H), 4.18 (m, 2H), 5.30 (m, 2H), 6.57 (s, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 12.16, 13.20, 16.88, 19.27, 22.04, 24.71, 27.12, 28.29, 30.81, 33.05, 37.72, 41.10, 42.63, 49.12, 52.23, 56.58, 58.71, 70.66, 102.10, 122.01, 127.73, 135.85, 140.96, 154.41. IR (KBr, cm^{-1}): 692, 754 (-CH bend. Ar), 1033, 1067 (=C-H bend. Alkene),1146, 1192 (C-H cycloalkane), 1341 (C-N stretch.),1380, 1458(C-H bend. Alkane), 1612 (C=C stretch. Alkene), 1687 (N-C=O stretch.), 2873-2988(-CH stretch. Aliphatic). UV spectrum:λ_{max} 224 Abs. 0.263. Optical activity: $[\alpha]^{20}_D$ = -20.52. HRMS: *m/z* cal. mass for $C_{62}H_{96}N_2O_2$ $[M+H]^+$ = 901.4436, obs. mass $[M+H]^+$ = 901.4445. Molecular formula: $C_{62}H_{96}N_2O_2$. Elemental analysis: Calculated: C (82.67%), H (10.67%), N (3.11%). Found: C (82.77%), H (10.74%), N (3.02%).

Table: Time and yield of the synthesized compounds 4a and 4b

	Conventional heating		Microwave irradiation	
	Time (minutes)	**% Yield**	**Time (minutes)**	**% Yield**
4a	80	41	17	52
4b	100	39	20	50

CONCLUSION

3, 6-dicholesteroxypyridazine **(3a)** and 3, 6-distigmasteroxypyridazine **(3b)** for the first time underwent facile double *Chapman rearrangement* to afford 1, 2-dicholesterylpyridazine-3, 6-diones **(4a)** and 1, 2-distigmasterylpyridazine-3, 6-dione **(4b)** respectively under conventional heating as well as microwave irradiation.

Microwave assisted method of synthesis provides a simpler and environmental-friendly alternative for the conventional procedures.

The synthesis of novel heterocycles reported in this paper has the potential of exhibiting pharmacological and agrochemical activities.

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