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Facile synthesis of novel D-ring modified steroidal dienamides via rearrangement of 2*H*-pyrans

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ABSTRACT

A simple and practical method for synthesis of the D-ring modified steroidal dienamides (**4a–k**) from the steroidal α,α-dicyanoalkene **3** and aldehydes via vinylogous aldol reaction was first reported. By using NaOAc as a base, the desired products were obtained in moderate to good yields in ethanol under mild conditions. All the synthesized steroidal dienamides are new and are currently being evaluated for their biological activities.

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1. Introduction

Steroids are a class of important multi-cyclic compounds which exhibit diverse biological activities. Except for the naturally occurring substances, most of steroidal pharmaceuticals are semi-synthetic compounds [1–3]. Several steroidal derivatives have been investigated as new curative agents for cancers and other diseases. It is proved that a number of biologically important properties of modified steroids are dependent upon structural features of the steroid ring system and side chain [4–6]. Chemical modification of the steroid ring system and side chain provides a way to alter the functional groups, and numerous structure–activity relationships have been established by such synthetic alterations [7]. The development of new compounds to improve the selectivity and to minimize side effects of steroidal drugs has been a challenge for a long time and has drawn wide attention of medicinal chemists.

On the other hand, dienamides have always been recognized as key reactive intermediates due to their great diversities, potential synthetic values and commonly existence in nature [8–10]. Therefore, dienamides could be used as electron-rich or electron-deficient dienes in Diels–Alder reactions effectively [11], which have already been applied to asymmetric cycloaddition reactions regioselectively [12]. Dienamides are also key constituents in a number of biologically active natural products and pharmaceutically relevant units. Examples of these include Apicularen A [13,14], Salicylihalamide A [15], Crocacin C [16], and Zampanolide [17] (Fig. 1), of which Zampanolide represents potent cytotoxicity (IC_{50} = 1–5 ng/mL) against P388, A549, HT29 and MEL28 cell lines.

In view of the therapeutic importance of dienamides and in continuation of our previous work in developing new biologically active modified steroids [18–27], we are interested in the design, synthesis and biological evaluation of novel steroidal dienamides. Recently, Zhao [28] and Perumal [29] reported a novel one-pot approach to a variety dienamides from α,α-dicyanoalkenes and aldehydes via vinylogous aldol reaction by the electrocyclic ring opening of the initially formed pyran derivatives under mild basic catalysis, respectively. However, to the best of our knowledge, the synthesis of D-ring modified steroidal dienamides has not been reported. So herein we report the synthesis of a novel class of steroidal dienamides from steroidal α,α-dicyanoalkene and aldehydes via vinylogous aldol reaction, and then followed by rearrangement of the 2*H*-pyran intermediate.

2. Experimental

2.1. General remarks

All reagents and solvents used were of analytical grade purchased from commercial sources. Thin-layer chromatography (TLC) was carried out on glass plates coated with silica gel (Qingdao Haiyang Chemical Co., G60F-254) and visualized by UV light (254 nm). The products were purified by column chromatography

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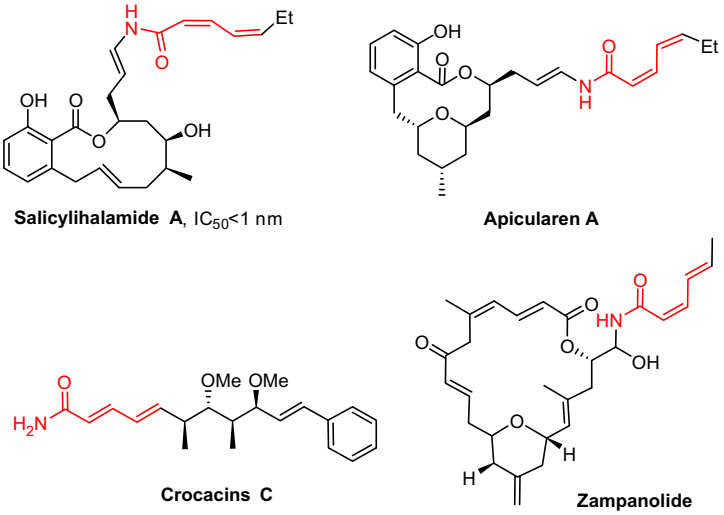


Fig. 1. Some natural occurring dienamides.

over silica gel (Qingdao Haiyang Chemical Co., 200–300 mesh). Melting points were determined on a Beijing Keyi XT4A apparatus and are uncorrected. All NMR spectra were recorded with a Bruker DPX 400 MHz spectrometer with TMS as internal standard in $CDCl_3$. Chemical shifts are given as δ ppm values relative to TMS (Most of the peaks due to the steroidal skeleton are merged and could not be differentiated. Thus δ values of only those peaks that distinguish the product and could easily be differentiated are reported). Mass spectra (MS) were recorded on Esquire3000 mass spectrometer by electrospray ionization (ESI).

2.2. Synthesis of 3-acetyl dehydroepiandrosterone **2**

A mixture of dehydroepiandrosterone (4.0 mmol), acetic anhydride (4.4 mmol), 4-dimethyl aminopyridine (DMAP, 0.02 mmol) and Et_3N (8.0 mmol) in dichloromethane (50 mL) was stirred for about 5 h at room temperature. After completion of the reaction as evident from TLC, the organic phase was washed with water and brine, dried over Na_2SO_4. Removal of solvent afforded compound **2** quantitatively without further purification. White solid, mp 169.4–170.6 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.43 (*d*, *J* = 5.1 Hz, 1H, H-6), 4.63 (*m*, 1H, 3α-H), 2.06 (*s*, 3H, –$OCOCH_3$), 1.07 (*s*, 3H, H-18), 0.91 (*s*, 3H, H-19). HRMS (ESI): *m/z* calcd for $C_{21}H_{30}NaO_3$ $(M + Na)^+$, 353.2093; found, 353.2094.

2.3. Synthesis of the D-ring modified steroidal dicyanoalkene **3**

To a solution of compound **2** (3.0 mmol) in ethanol (10 ml) containing ammonium acetate (0.5 g), malononitrile (4.5 mmol) was added. The reaction mixture was heated under reflux for about 3 h until compound **2** disappeared as indicated by TLC. The solid product formed upon cooling at room temperature was washed with ethanol, collected by filtration to yield gray solid of compound **3**, yield 95%, mp 187.9–188.9 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.38 (*d*, *J* = 5.1 Hz, 1H, 6-H), 4.74–4.45 (*m*, 1H, 3α-H), 3.04–2.86 (*m*, 1H, 16α-H), 2.74 (*m*, 1H, 16β-H), 2.04 (*s*, 3H, –$OCOCH_3$), 1.05 (*s*, 6H, H-18 and H-19). ^{13}C NMR (100 MHz, $CDCl_3$): δ 196.08, 170.50, 139.93, 121.58, 112.26, 111.16, 79.73, 73.58, 55.22, 49.34, 48.91, 37.99, 36.83, 36.56, 34.73, 33.61, 31.39, 31.37, 27.63, 23.56, 21.40, 20.78, 19.27, 16.24. HRMS (ESI): *m/z* calcd for $C_{24}H_{30}N_2NaO_2$ $(M + Na)^+$, 401.2205; found, 401.2229.

2.4. General procedure for the synthesis of the steroidal dienamides **4a–k**

To a solution of compound **3** (1.0 mmol) in ethanol, aldehydes (1.0 mmol) and sodium acetate (2.0 mmol) were added. The reaction mixture was heated under reflux for about 3–7 h. The solvent was removed and CH_2Cl_2 was added, the organic phase was washed with water and brine, dried over Na_2SO_4. After removal of the solvent, the residue was purified by silica gel chromatography with ethyl acetate/petroleum (1/2) to give the corresponding steroidal dienamides (Table 1).

2.4.1. 3β-Acetoxyl-5-en-16-benzylidene –androstano-17-(2-amino-1-cyano-2-oxoethyli-dene) (**4a**)

Yellow solid, yield 76%, mp 134.1–135.8 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.55–7.29 (*m*, 5H, ArH), 7.25 (*s*, 1H, Ar–CH=), 6.02 (*s*, 1H, –NH_2), 5.77 (*s*, 1H, –NH_2), 5.40 (*d*, *J* = 4.7 Hz, 1H, 6-H), 4.61–4.50 (m, 1H, 3α-H), 2.04 (s, 3H, –$OCOCH_3$), 1.06 (*s*, 3H, 18-H), 1.05 (*s*, 3H, 19-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 175.44, 170.54, 164.92, 140.09, 137.42, 136.19, 135.78, 129.90, 128.90, 128.67, 121.61, 116.36, 98.51, 73.70, 52.54, 49.51, 47.13, 38.05, 36.79, 36.61, 34.73, 31.48, 31.42, 31.29, 29.70, 27.66, 21.42, 21.10, 19.31, 16.51. HRMS (ESI): *m/z* calcd. for $C_{31}H_{37}N_2O_3$ $(M + H)^+$, 485.2804; found, 485.2802.

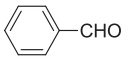
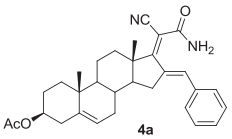
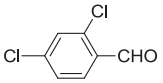
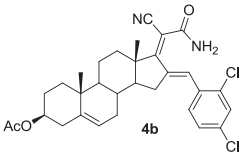
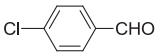
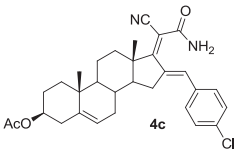
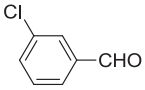
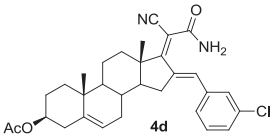
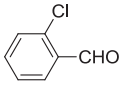
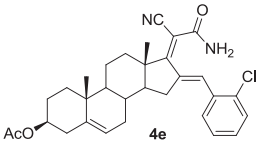
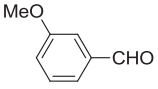
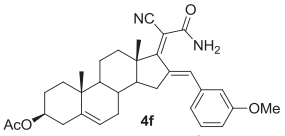
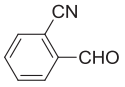
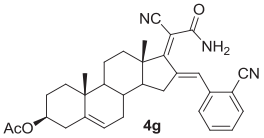
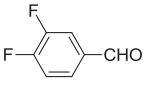
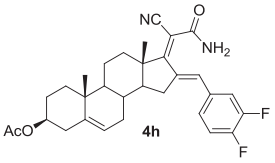
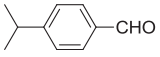
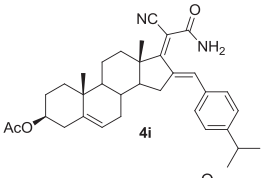
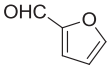
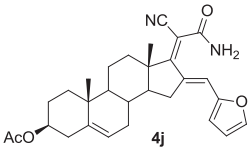
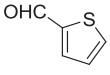
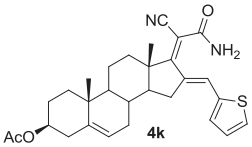
2.4.2. 3β-acetoxyl-5-en-16-(2, 4-dichlorobenzylidene)-androstano-17-(2-amino-1-cyano-2-oxo-ethylidene) (**4b**)

Yellow solid, yield 87%, mp 146.5–147.5 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.47 (*s*, 1H, Ar–CH=), 7.38 (*dd*, 2H, ArH), 7.25 (*dd*, 1H, ArH), 6.55 (*s*, 1H, -NH_2), 6.38 (*s*, 1H, –NH_2), 5.35 (*d*, *J* = 4.7 Hz, 1H, 6-H), 4.68–4.52 (*m*, 1H, 3α-H), 2.03 (*s*, 3H, –$OCOCH_3$), 1.08 (*s*, 3H, 18-H), 1.04 (*s*, 3H, 19-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 172.51, 170.60, 165.08, 140.04, 135.38, 134.70, 133.19, 130.36, 130.16, 129.64, 127.01, 121.55, 116.08, 100.11, 73.70, 52.33, 49.41, 47.31, 38.00, 36.75, 36.57, 34.67, 31.34, 31.24, 31.03, 27.63, 21.41, 21.22, 19.27, 16.59. HRMS (ESI): *m/z* calcd. for $C_{31}H_{35}Cl_2N_2O_3$ $(M + H)^+$, 553.2025; found, 553.2007.

2.4.3. 3β-Acetoxyl-5-en-16-(4-chlorobenzylidene) - androstano-17-(2-amino-1-cyano-2-oxoethy-lidene) (**4c**)

Yellow solid, yield 86%, mp 161.0–162.1 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.34 (*d*, 4H, ArH), 7.19 (*s*, 1H, Ar–CH=), 6.32–5.96 (*m*, 2H, –NH_2), 5.39 (*d*, *J* = 3.9 Hz, 1H, 6-H), 4.63–4.58 (*m*, 1H, 3α-H), 2.04 (*s*,

Table 1
Synthesis of steroidal dienamides (**4a–k**).

Entry	Aldehydes	Products[a]	Yields[b] (%)	Time (h)
1		 **4a**	76	5
2		 **4b**	87	3
3		 **4c**	86	3
4		 **4d**	84	3
5		 **4e**	81	3
6		 **4f**	90	3
7		 **4g**	55	7
8		 **4h**	85	3
9		 **4i**	70	4
10		 **4j**	86	3
11		 **4k**	84	3

[a] Reaction conditions: **3** (1.0 mmol), aldehydes (1.0 mmol), NaOAc (2.0 mmol), EtOH (10 mL), reflux, 3–7 h.
[b] Isolated yields.

3H, –$OCOCH_3$), 1.05 (*s*, 6H, 18-H and H-19). ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.96, 170.58, 164.98, 140.07, 138.03, 134.69, 134.21, 130.99, 128.89, 121.56, 116.31, 98.83, 73.69, 52.44, 49.46, 47.14, 38.02, 36.68, 34.70, 31.58–31.07, 27.64, 21.42, 21.07, 19.29, 16.51, 15.27. HRMS (ESI): *m/z* calcd. for $C_{31}H_{36}ClN_2O_3$ $(M+H)^+$, 519.2414; found, 519.2411.

*2.4.4. 3β-Acetoxyl-5-en-16-(3-chlorobenzylidene) – androstano-17-(2-amino-1-cyano-2-oxoethy-lidene) (**4d**)*

Yellow solid, yield 84%, mp 147.0–148.2 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 7.38 (*s*, 1H, ArH), 7.36–7.24 (*m*, 3H, ArH), 7.18 (*s*, 1H, Ar–CH=), 6.04 (*m*, 2H, –NH_2), 5.40 (*d*, *J* = 4.7 Hz, 1H, 6-H), 4.71–4.49 (*m*, 1H, 3α-H), 2.04 (*s*, 3H, –$OCOCH_3$), 1.05 (*s*, 6H, 18-H and H-19). ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.60, 170.51, 164.71, 140.02, 138.89, 137.91, 134.55, 133.83, 129.80, 129.43, 128.65, 127.81, 121.52, 116.16, 99.18, 73.64, 54.81, 52.39, 49.42, 47.13, 37.99, 36.65, 34.67, 31.51, 31.05, 29.66, 27.60, 21.37, 21.03, 19.25, 16.48. HRMS (ESI): *m/z* calcd. for $C_{31}H_{35}ClN_2NaO_3$ $(M+Na)^+$, 541.2234; found, 541.2230.

*2.4.5. 3β-Acetoxyl-5-en-16-(2-chlorobenzylidene) – androstano-17-(2-amino-1-cyano-2-oxoethy-lidene) (**4e**)*

Yellow solid, yield 81%, mp 227.4–228.0 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 7.55 (*s*, 1H, ArH), 7.45 (*d*, *J* = 7.2 Hz, 1H, ArH), 7.38 (*t*, *J* = 6.2 Hz, 1H, ArH), 7.33–7.17 (*m*, 2H, ArH and Ar–CH=, overlapped), 6.14 (*m*, 2H, –NH_2), 5.35 (*s*, 1H, 6-H), 4.70–4.50 (*m*, 1H, 3α-H), 2.03 (*s*, 3H, –$OCOCH_3$), 1.10 (*s*, 3H, 18-H), 1.05 (*s*, 3H, 19-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 173.27, 170.54, 164.76, 140.02, 139.50, 134.67, 131.61, 129.84, 129.70, 129.61, 126.60, 121.59, 116.17, 99.69, 73.69, 52.37, 49.44, 47.33, 38.02, 36.77, 36.59, 34.68, 31.37, 31.25, 30.96, 29.69, 27.64, 21.41, 21.06, 19.28, 16.60. HRMS (ESI): *m/z* calcd. for $C_{31}H_{35}ClN_2NaO_3$ $(M+Na)^+$, 541.2234; found, 541.2232.

*2.4.6. 3β-Acetoxyl-5-en-16-(3-methoxylbenzylidene)-androstano-17-(2-amino-1-cyano-2-oxo ethylidene) (**4f**)*

Yellow solid, yield 90%, mp 146.1–147.8 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 7.28 (*m*, 1H, ArH), 7.20 (*s*, 1H, Ar–CH=), 7.01 (*d*, *J* = 7.6 Hz, 1H, ArH), 6.93 (*s*, 1H, ArH), 6.86 (*d*, *J* = 8.2 Hz, 1H, ArH), 6.37 (*s*, 2H, –NH_2), 5.39 (*d*, *J* = 4.3 Hz, 1H, 6-H), 4.69–4.51 (*m*, 1H, 3α-H), 3.81 (*s*, 3H, –OCH_3), 2.03 (*s*, 3H, –$OCOCH_3$), 1.04 (*s*, 6H, 18-H and H-19). ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.67, 170.63, 165.60, 159.59, 140.00, 137.85, 137.55, 135.31, 129.60, 122.40, 121.66, 116.36, 115.29, 114.38, 98.80, 73.75, 55.32, 52.40, 49.44, 47.02, 38.02, 36.67, 34.69, 31.33, 29.68, 27.63, 21.41, 21.06, 19.28, 16.48. HRMS (ESI): *m/z* calcd. for $C_{32}H_{38}N_2NaO_4$ $(M+Na)^+$, 537.2729; found, 537.2740.

Table 2
Optimization of reaction conditions for formation of compound **4c**.[a]

Entry	Base	Solvent	Yield[b] (%)
1	Imidazole	EtOH	15
2	DMAP	EtOH	0
3	Basic Al_2O_3	EtOH	0
4	Et_3N	EtOH	20
5	NaOAc	EtOH	86
6	NaOAc	EtOH	65[c]
7	NaOAc	DCM	0
8	NaOAc	EtOAc	0
9	NaOAc	MeCN	Trace
10	NaOAc	Acetone	0
11	NaOAc	Toluene	0

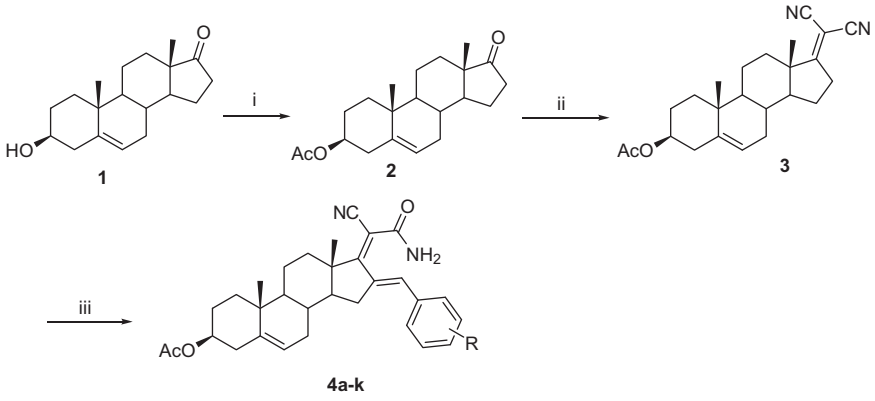
[a] Unless otherwise noted, the reaction was carried out with 3 (0.1 mmol) and 4-chlorobenzaldehyde (0.1 mmol) and Bases (0.2 mmol) under reflux in solvent (3 ml) for about 3 h.
[b] Isolated yields.
[c] Under the same conditions except that the amount of catalyst was increased to 0.6 mmol.

*2.4.7. 3β-Acetoxyl-5-en-16-(2- cyanobenzylidene) – androstano-17-(2-amino-1-cyano-2-oxoethy-lidene) (**4g**)*

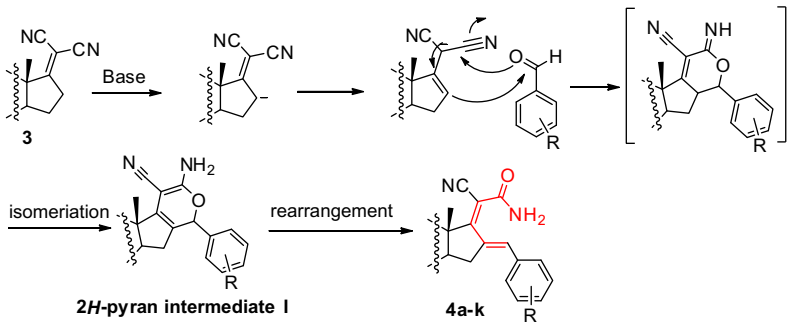
Brown solid, yield 55%, mp 211.7–212.5 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 7.77–7.52 (*m*, 4H, ArH), 7.47–7.36 (*m*, 1H, Ar–CH=), 6.27 (*s*, 1H, –NH_2), 6.06 (*s*, 1H, –NH_2), 5.38 (*d*, *J* = 5.1 Hz, 1H, 6-H), 4.73–4.50 (*m*, 1H, 3α-H), 2.05 (*s*, 3H, –$OCOCH_3$), 1.12 (*s*, 3H, 18-H), 1.07 (*s*, 3H, 18-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 172.42, 170.55, 164.21, 142.24, 140.12, 139.78, 133.21, 132.66, 129.67, 128.77, 128.49, 121.46, 117.68, 115.83, 112.96, 100.84, 73.67, 52.28, 49.42, 47.31, 38.03, 36.77, 36.60, 34.68, 31.41, 31.26, 29.70, 27.64, 21.41, 21.03, 19.30, 16.63. HRMS (ESI): *m/z* calcd. for $C_{32}H_{35}N_3NaO_3$ $(M+Na)^+$, 532.2576; found, 532.2578.

*2.4.8. 3β-Acetoxyl-5-en-16-(2, 4-difluorobenzylidene)-androstano-17-(2-amino-1-cyano-2-oxo-ethylidene) (**4h**)*

Yellow solid, yield 85%, mp 143.2–145.5 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 7.20 (m, 4H, ArH and Ar–CH=, overlapped), 6.38 (*s*, 2H, –NH_2), 5.40 (*d*, *J* = 4.6 Hz, 1H, 6-H), 4.60 (*m*, 1H, 3α-H), 2.04 (*s*, 3H, –$OCOCH_3$), 1.05 (*s*, 3H, 18-H), 1.04 (*s*, 3H, 19-H). ^{13}C NMR (10.0 MHz, $CDCl_3$): δ 174.28, 170.64, 165.23, 151.45, 148.96, 140.04, 138.41, 133.56, 132.96, 126.54, 121.55, 118.12, 117.94, 117.62, 117.45, 116.21, 99.23, 77.40, 77.08, 76.77, 52.36, 49.43, 47.10, 38.00, 36.76, 36.57, 34.69, 31.42, 31.24, 31.21, 27.62, 21.40, 21.05, 19.27, 16.49. HRMS (ESI): *m/z* calcd. for $C_{31}H_{35}F_2N_2O_3$ $(M+H)^+$, 521.2616; found, 521.2614.



Scheme 1. Synthesis of steroidal dienamides (**4a–k**). Reagents and conditions: (i) $(Ac)_2O$, DMAP, Et_3N, DCM, r.t; (ii) malononitrile, NH_4OAc, EtOH, reflux; (iii) Aldehydes, NaOAc, EtOH, reflux.



Scheme 2. Proposed mechanism for the formation of steroidal dienamides **4a–k.**

*2.4.9. 3β-Acetoxyl-5-en-16-(4-isopropylbenzylidene)-androstano-17-(2-amino-1-cyano-2-oxo ethyli-dene) (**4i**)*

Yellow solid, yield 70%, mp 142.2–143.6 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.38 (*d*, *J* = 8.2 Hz, 2H, ArH), 7.29–7.23 (*m*, 3H, ArH and Ar–CH=, overlapped), 6.37 (*m*, 2H, –NH_2), 5.41 (*d*, *J* = 4.4 Hz, 1H, 6-H), 4.60 (*m*, 1H, 3α-H), 2.93 (*m* 1H, –C<u>H</u>(CH_3)), 2.04 (*s*, 3H, –$OCOCH_3$), 1.26 (*s*, 3H, –CH(CH_3)), 1.25 (*s*, 3H, –CH(CH_3)), 1.06 (*s*, 3H, 18-H), 1.04 (*s*, 3H, 18-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 175.07, 170.63, 165.84, 150.03, 140.01, 136.52, 135.59, 133.88, 130.08, 126.80, 121.70, 116.51, 98.26, 73.76, 52.50, 49.49, 47.00, 38.03, 36.68, 34.70, 33.98, 31.36, 29.69, 27.64, 23.79, 21.42, 21.09, 19.29, 16.48. HRMS (ESI): *m/z* calcd. for $C_{34}H_{43}N_2O_3$ $(M+H)^+$, 527.3274; found, 527.3271.

*2.4.10. 3β-Acetoxyl-5-en-16-furylmethylene – androstano-17-(2-amino-1-cyano-2-oxoethylidene) (**4j**)*

Yellow solid, yield 86%, mp 156.8–157.5 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.51 (*s*, 1H, furyl-H), 7.11 (*s*, 1H, Ar–CH=), 6.50 (*m*, 4H, furyl-H and –NH_2, overlapped), 5.40 (*d*, *J* = 3.9 Hz, 1H, 6-H), 4.77–4.44 (*m*, 1H, 3α-H), 2.02 (*s*, 3H, –$OCOCH_3$), 1.04 (*s*, 3H, 18-H), 1.00 (*s*, 3H, 19-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.01, 170.64, 165.58, 152.49, 144.44, 139.95, 134.60, 122.23, 121.77, 116.61, 114.60, 112.49, 97.92, 77.47, 77.24, 76.99, 73.78, 52.00, 49.45, 47.80, 38.01, 36.76, 36.56, 34.73, 31.64, 31.41, 31.26, 27.63, 21.40, 21.11, 19.28, 16.40, 14.18. HRMS (ESI): *m/z* calcd. for $C_{29}H_{35}N_2O_4$ $(M+H)^+$, 475.2597; found, 475.2599.

*2.4.11. 3β-Acetoxyl-5-en-16-thienylmethylene – androstano-17-(2-amino-1-cyano-2-oxoethyli-dene) (**4k**)*

Yellow solid, yield 84%, mp 153.3–154.3 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.53 (*s*, 1H, Ar–CH=), 7.49 (*d*, *J* = 4.8 Hz, 1H, thienyl-H), 7.20 (*d*, *J* = 3.2 Hz, 1H, thienyl-H), 7.10 (*dd*, *J* = 4.8, 3.2 Hz, 1H, thienyl-H), 6.46 (*s*, 2H, –NH_2), 5.42 (*d*, *J* = 3.8 Hz, 1H, 6-H), 4.61 (*m* 1H, 3α-H), 2.04 (*s*, 3H, –$OCOCH_3$), 1.05 (*s*, 3H, 18-H), 1.02 (*s*, 3H, 19-H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.43, 170.65, 165.62, 140.50, 140.01, 134.64, 131.50, 129.58, 128.67, 127.79, 121.70, 116.61, 97.87, 77.48, 77.17, 76.85, 73.77, 52.13, 49.44, 47.98, 38.02, 36.76, 36.58, 34.76, 31.65, 31.44, 31.27, 27.64, 21.42, 21.10, 19.30, 16.43. HRMS (ESI): *m/z* calcd. for $C_{29}H_{35}N_2O_3S$ $(M+H)^+$, 491.2368; found, 491.2365.

3. Results and discussion

The protocol for the synthesis of steroidal dienamides (**4a–k**) is very simple and straightforward involving the vinylogous aldol reaction of steroidal α,α-dicyanoalkene **3** and aldehydes, and then followed by rearrangement of the 2*H*-pyran intermediate. The intermediate **3** was prepared in high yield via Aldol condensation of 3-acetyl dehydroepiandrosterone with malononitrile in ethanol catalyzed by ammonium acetate. The direct condensation reaction of dehydroepiandrosterone **1** with malononitrile was also performed in ethanol catalyzed by ammonium acetate, affording the corresponding product in low yield (about 50%) even with prolonged reaction time (>12 h) (Scheme 1).

Initially, the reaction of steroidal α,α-dicyanoalkene **3** and 4-chlorobenzaldehyde was selected as a model reaction for catalyst evaluation. First, with ethanol as solvent, several different bases as catalysts were evaluated under reflux (Table 2, entries 1–6). To our pleasure, when NaOAc was used, the desired product **4c** was obtained with 86% yield. While using imidazole or Et_3N as organic base, the yield decreased significantly (Table 2, entries 1 and 4). DMAP or Al_2O_3 as base was also screened, but no reaction was observed (Table 2, entries 2 and 3). When the amount of NaOAc was increased to 0.6 mmol, the yield decreased slightly due to the formation of the competitive dehydration product (Table 2, entry 6). Next, we examined the influence of different solvents on the isolated yield (Table 2, entries 7–11), unfortunately, no reaction was observed with the exception of EtOH, showing that alcoholic solvent could promote this kind of reaction. The present reaction was best performed with 200 mol% of NaOAc in EtOH under reflux for about 3–7 h.

Having established the optimal conditions, we then applied the protocol to the vinylogous aldol reaction of different aldehydes with **3**, affording the desired steroidal dienamides (**4a–k**) in moderate to good yields (Table 1). Besides, we also found that treatment **3** with aliphatic aldehydes or aromatic aldehydes with electron-withdrawing group did not afford the desired products, revealing that the electronic effect of the substituents on the aromatic ring had a remarkable effect on this reaction.

All the new compounds were characterized by ^{1}H, ^{13}C and mass spectra. Of which compound **4a** was fully characterized by 1D, 2D NMR and mass spectra. In the ^{1}H NMR spectra, the olefinic proton of compound **4a** appeared at δ = 7.25 ppm as a sharp singlet, where the amide NH_2 protons resonated at δ = 6.02 and 5.77 ppm as two broad singlets. The amide carbon in ^{13}C NMR signaled at δ = 164.92 ppm. In the HSQC spectra, the proton of the olefinic proton (δ = 7.25 ppm) had a direct correlation with the carbon (δ = 98.51 ppm), revealing that the olefinic carbon of **4a** resonated at 98.51 ppm. According to the previous reports of zhao and Perumal's group [28,29], the stereochemistry of **4a** should be the (2*Z*, 4*E*) configuration, as shown in Scheme 2.

A possible mechanism for the formation of compounds **4a–k** was proposed in Scheme 2. Under basic conditions, the steroidal α,α-dicyanoalkene **3** was first deprotonated to form a nucleophile that then attacked the aldehyde via vinylogous aldol reaction. Subsequent intramolecular nucleophilic addition and isomerization afforded the 2*H*-pyran intermediate I. ultimately, intermediate I furnished the electrocyclic ring-opened products (**4a–k**).

4. Conclusion

In summary, we have developed a simple method to synthesize a series of the steroidal dienamides from readily available starting materials such as the steroidal α,α-dicyanoalkene **3** and aldehydes via vinylogous aldol reaction followed by the rearrangement of 2*H*-pyran intermediate in one-pot procedure. By using NaOAc as catalyst, the desired products were obtained in moderate to good yields in ethanol under mild conditions. Efforts towards biological evaluation of these new compounds are in progress and will be reported elsewhere.

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