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Regioselective Spirostan E-Ring Opening for the Synthesis of Dihydropyran Steroidal Frameworks

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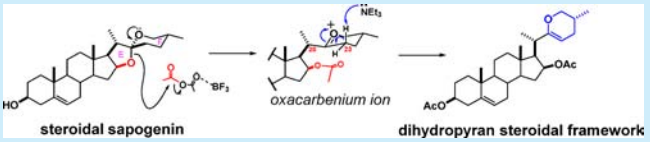
Supporting Information

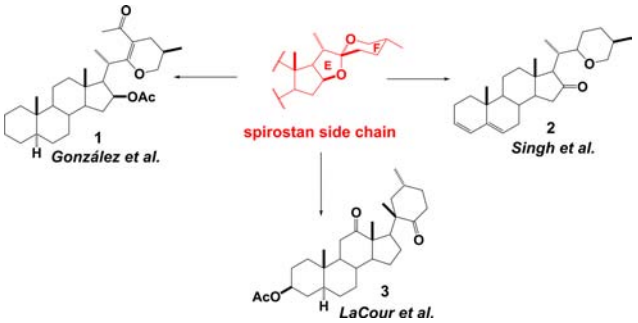
ABSTRACT: The regioselective opening of the ring E in spirostan sapogenins provides new dihydropyran derivatives. This novel side chain is obtained after a Lewis acid mediated acetolysis followed by an alkaline workup. The reaction mechanism is analyzed via density functional theory computations, and both experimental and computational data support the formation of an oxacarbenium intermediate. The behavior of the title skeletons under acidic conditions is also investigated.



Spirostan sapogenins constitute an important group of naturally occurring steroids commonly isolated from plants in their glycoside forms.[1] Several sapogenin-containing plants have been widely employed in folk medicine because of their extended properties, and consequently, great interest has arisen in their pharmacological and biological features as well as in the isolation of their active components and subsequent structural transformations.[2] Before the 1940s, several authors studied the structure of the spirostan side chain[3−5] until Marker finally proposed the spiroketal side chain with a tetrahydrofuran structure for the ring E and a tetrahydropyran structure for the ring F.[6] When Marker accomplished the synthesis of progesterone from sarsasapogenin in high yields, spirostans displayed high importance as starting materials for the synthesis of diverse drugs.[7,8] The key intermediate in the so-called "Marker's degradation" is the pseudosarsasapogenin. In such process, the spirostan ring F is selectively cleaved in moderate yield (around 70%) using strong reaction conditions. Many reports on spirostan side chain transformation have been published since the emblematic Marker's degradation where he showed its usefulness in steroid synthesis.[9−12]

There is a lack of general methods for the regioselective opening of the ring E. Before 1970, the attention was only focused on the selective opening of the ring F (or both E and F). In 1976, González et al. described the first opening of the ring E through the synthesis of a 22,26-epoxycholestane derivative (**1**), starting from the unsubstituted steroidal sapogenin (25*R*)-5α-spirostan using Ac_2O in the presence of BF_3 (Scheme 1).[13]

Scheme 1. Steroidal Frameworks Derived from the Opening of Ring E



In 1962, Zderic et al.,[14] working on tigogenin and using similar conditions to González et al., obtained a furostan compound (both compounds **1** and the furostan bear an α,β-unsaturated ketone on the side chain). Zderic et al. stated the possible formation of the side chain of **1**, but indicated that such transformation *would involve the unlikely possibility of ring E*

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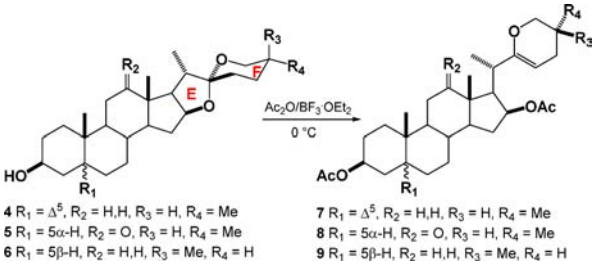
having undergone fission in preference to ring F. Notice that they catalogued the selective opening of ring E as *unlikely*. Later on, Singh et al. reported the synthesis of structure **2** from diosgenin,[15] employing several acidic media achieving the E-ring fission in 85% yield but with the elimination of the hydroxyl group at C-3. In 1999, LaCour et al. described several transformations of hecogenin including the 20,26-cyclocholestane structure **3**, which was obtained in 87% using triphenylphosphine, iodine, and a non-nucleophilic base.[16] Under similar conditions, it is also possible to obtain halogenated pseudosapogenins.

Herein, we report the regioselective spirostan E-ring opening of steroidal sapogenins through a Lewis acid mediated acetolysis followed by a basic workup toward the synthesis of novel dihydropyran (DHP) steroidal frameworks. The reaction mechanism is analyzed via density functional theory (DFT) computations. Both experimental and computational data support the formation of an oxacarbenium intermediate. Their synthetic usefulness under various acidic media and the scope behind the mechanism are also discussed.

We have been interested in the selective opening of ring E in steroidal sapogenins because most reactions are directed toward the opening of the ring F or both rings E and F. The most important observation is that, under similar acidic conditions, the spiroketal behaves differently. Distinct products such as furostenes,[12] 22-oxocholestanes,[17−20] and epoxycholestanes[21] can be obtained with slight variations in temperature, reaction time, and/or reactants concentration. In those cases, the opening of ring F, ring E, or both are involved, and the formation of byproducts cannot be controlled. Special care must be taken during the performance of such reactions. That is why we have continued to try to discover the specific conditions for the selective transformation of the spirostan side chain and succeeded in obtaining the selective opening of the E-ring producing in excellent yields a dihydropyran in the side chain.

When the spirostan sapogenins diosgenin (**4**), hecogenin (**5**), and sarsasapogenin (**6**) were treated under Lewis acid mediated acetolysis using boron trifluoride etherate at 0 °C, followed by a basic workup with triethylamine, the dihydropyran skeletons **7**−**9** were obtained in 85−90% yield (see Scheme 2).

Scheme 2. Dihydropyran Derivatives (7−9) Obtained in One Step from Steroidal Sapogenins



4 R_1 = Δ⁵, R_2 = H,H, R_3 = H, R_4 = Me
5 R_1 = 5α-H, R_2 = O, R_3 = H, R_4 = Me
6 R_1 = 5β-H, R_2 = H,H, R_3 = Me, R_4 = H

7 R_1 = Δ⁵, R_2 = H,H, R_3 = H, R_4 = Me
8 R_1 = 5α-H, R_2 = O, R_3 = H, R_4 = Me
9 R_1 = 5β-H, R_2 = H,H, R_3 = Me, R_4 = H

In 1991, Szendi and Sweet[22] reported the synthesis of steroidal derivatives containing a DHP ring at the side chain but, via the condensation of a 3,4-(2*H*)-dihydropyran moiety with pregnenolone, so such methodology is not comparable to ours. The idea to trap the DHP derivative came from the observation of the mechanism proposed by some of us in 2010 for the synthesis of 26-hydroxy-22-oxocholestanes.[17] In this mechanism, the first step involves the opening of the ring E and the acetylation of the hydroxyl group at C-16. The regioselective *β*-elimination of a proton of C-23 in the tetrahydropyrilium intermediate, an oxacarbenium ion (***i***, Scheme 3), could be achieved by using the bulky non-

Scheme 3. Proposed Mechanism for the Synthesis of DHP Analogues through an Oxacarbenium Intermediate

4 R_1 = H,H, R_2 = H, R_3 = Me
5 R_1 = O, R_2 = H, R_3 = Me
6 R_1 = H,H, R_2 = Me, R_3 = H

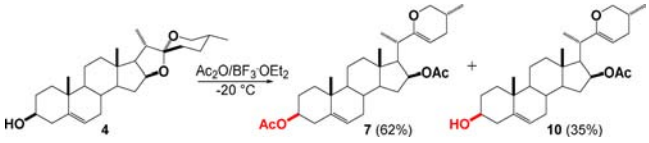
4*i* R_1 = H,H, R_2 = H, R_3 = Me
5*i* R_1 = O, R_2 = H, R_3 = Me
6*i* R_1 = H,H, R_2 = Me, R_3 = H

7 R_1 = H,H, R_2 = H, R_3 = Me
8 R_1 = O, R_2 = H, R_3 = Me
9 R_1 = H,H, R_2 = Me, R_3 = H

nucleophilic base triethylamine, leading to the formation of the desired DHP. The yield after purification was 85−90%. The formation of the exocyclic double bond via *β*-elimination of C-20 proton was not detected in any case likely due to the steric hindrance of both C-20 proton and triethylamine. The formation of the DHP side chain using the procedure described above confirms the existence of the proposed oxacarbenium ion.

These DHP derivatives are stable at pH ≥ 7 and extremely reactive under acidic media. Another interesting result came up from the performance of the reaction at low temperature. During the investigation of the optimal conditions for the selective synthesis of DHP analogues, we discovered that the same reaction provided a mixture of compounds **7** and **10** if the reaction is performed at −20 °C (see Scheme 4). Our results

Scheme 4. Synthesis of the Dihydropyran Analogue 10 Deprotected at C-3



suggest that the reactivity of the three oxygen atoms of diosgenin (and related sapogenins) is as follows: the tetrahydropyranyl oxygen is the most reactive, followed by the oxygen from the hydroxyl group at C-3 position, and finally, the tetrahydrofuranyl oxygen is the least reactive. When the reaction was carried out below −20 °C, the rate decreased considerably and the methodology lost practical applicability.

In order to better understand the reaction mechanism of the title opening, a series of DFT computations were carried out using the PBE0 approach[23] in conjunction with a 6-311+G(d,p) basis set using **4** as the model (Figure 1). Three important factors should be included to properly describe the mechanism. The first is the solvent, the second one is the dispersion, and finally the thermal factors (see the computational details in the SI). Thus, the discussion is based on the Gibbs free energies computed at 273.15 K, using CH_2Cl_2 as solvent. The interaction of **4** with $Ac_2O{\cdot}BF_3$ provokes the opening of ring E and the acetylation of C-16 via the transition state **TS1**. In **TS1**, while a C−O single bond of the acetic anhydride is breaking (2.441 Å), a bond is forming between the carbon of carbonyl from the acetyl moiety and the oxygen atom in C-16 (2.043 Å). The computed barrier for this step is 19.3 kcal/mol.

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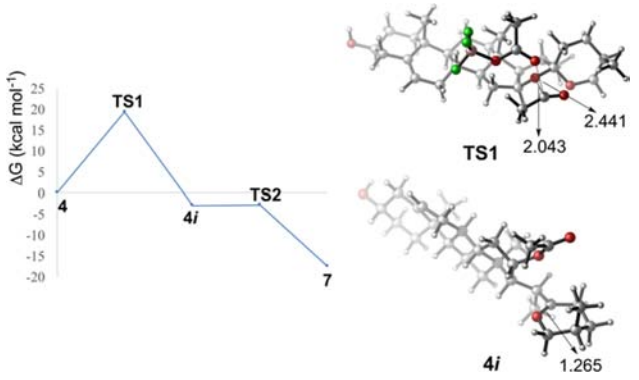


Figure 1. Full energy profile of the Lewis acid mediated acetolysis of steroidal sapogenins and structures of **TS1** and **4*i***.

The formed intermediate **4*i*** is an oxacarbenium ion with a short C−O bond in the F ring of 1.265 Å. Intermediate **4*i*** is lower in energy than the reactants by 2.9 kcal/mol. Finally, the transformation of **4*i*** to the final product **7** via **TS2** implicates the proton transfer from C-23 to an amine. This exergonic acid−base reaction (ΔG = −14.6 kcal/mol) has a negligible barrier of less than 1.0 kcal/mol at 273.15 K. Overall, our computational results are consistent with the experimental observations, supporting the formation of an oxacarbenium intermediate in the Lewis acid mediated acetolysis of steroidal sapogenins as previously proposed by several authors, including us.

All target compounds showed high stability in neutral or slightly basic media, though samples in $CDCl_3$ were stable enough to collect the NMR data. A complete assignment for the ^{1}H- and ^{13}C NMR signals was possible using a combination of COSY, HSQC, and HMBC experiments. Briefly, the ^{1}H NMR data of compound **7** shows the characteristic signal for the vinyl proton H-6 at 5.35 ppm. The vinylic proton H-23 is observed as a dd at 4.34 ppm, shifted highfield with regard to H-6. This effect is explained given that the electronic density of the π-system of the Δ^{22} becomes less important due to the inductive effect of the oxygen atom in the dihydropyran ring. The diastereotopic protons H-26eq and H-26ax are at 3.90 and 3.38 ppm, respectively, these data confirm the existence of the ring F. In ^{13}C NMR, C-22 is found at 157.8 ppm, shifted downfield in comparison with other vinyl carbons since it is directly bonded to the dihydropyranylic oxygen. C-23 is at 93.4 ppm. In the typical region for carbons bearing an oxygen atom, C-26 (71.7 ppm), C-3 (73.8 ppm), and C-16 (75.0 ppm) are detected. Similar shifts are found for compounds **8**−**10**. The full characterization of all products is in the SI.

Compound **10** (CCDC 1438444) crystallized in the chiral monoclinic space group *P*2(1), and the molecular structure shown in Figure 2 reflects the absolute configuration proposed by NMR, corroborating the correct assignment of all asymmetric carbons. The Flack and Parson's parameters of 0.177(35) and 0.078(58), respectively, are consistent with the absolute structure shown in Figure 2.[24]

The new DHP derivatives are envisaged as starting points for the synthesis of numerous compounds already reported in the literature. We used compound **7** as our model. Under acidic conditions (protic acid), **7** must regenerate the oxacarbenium ion **4*i*** (Scheme 5). Hence a question arises: which nucleophilic attack is preferred, the one at C-26 or the one at C-22? Employing a series of acids, we obtained the 26-substituted 22-oxocholestanes **11**−**13** (Scheme 5). All yields and conditions are described in Table 1. The structures of compounds **11** and **13** were confirmed by comparing their NMR with authentic samples.

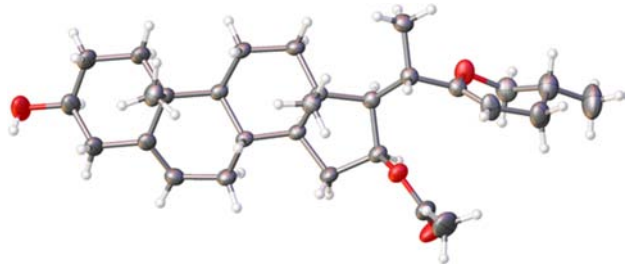
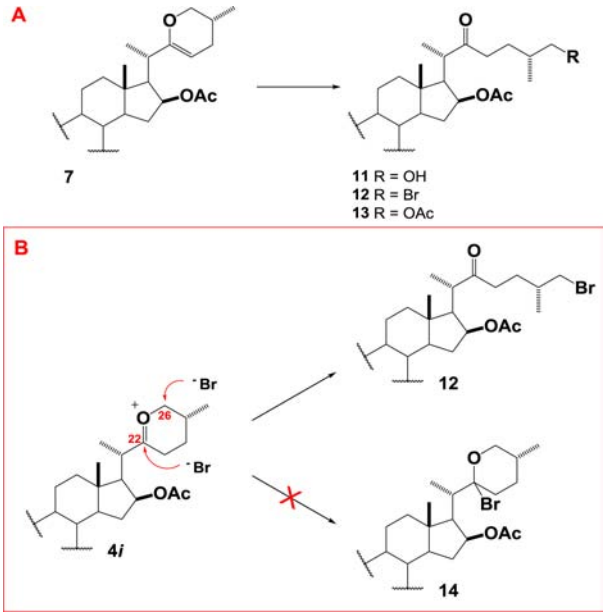


Figure 2. Molecular structure of **10** with thermal ellipsoids at the 30% probability level.

Scheme 5. (A) Transformation of the DHP Side Chain into 22-Oxocholestanes. (B) Reactivity of the DHP Ring under Protic Acid Media[a]



[a]A regioselective nucleophilic attack takes place at C-26.

Table 1. 22-Oxocholestane 26-Substituted Compounds Obtained from the Transformation of DHP Derivative 7 under Protic Acid Media at 25 °C

entry	acidic conditions	reaction time (h)	products yield (%)
1	HCl 5%	2	**11** (100)
2	HBr 5%	3	**11** (98)
3	HBr/AcOH 5%	4	**12** (30), **13** (30)
4	AcOH	2	**11** (98)

In entry 3, some starting material was recovered. The presence of compound **12** allowed us to solve the dilemma of the nucleophilic attack since the mechanism must go necessarily by the attack toward C-26 as illustrated in Scheme 5. Compound **14** was not obtained at all. Let us underline that in entries 1, 2, and 4 the 22-oxocholestane-26-hydroxy compound **11** is obtained in almost quantitative yields. Although this synthetic route requires two steps, it represents

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an excellent method for the synthesis of this precious side chain.

In summary, we report the regioselective opening of the ring E of spiroketal sapogenins toward dihydropyran skeletons. The target DHP side chain is obtained in excellent yields from the spiroketal moiety using a one-step convenient procedure. This procedure works well with diosgenin, hecogenin, and sarsasapogenin as starting materials and can be extended to other spirostans. We were able to crystallize the derivative (25*R*)-22,26-epoxy-3β-hydroxycholesta-5,22-diene-16β-yl acetate and to corroborate the proposed structure of dihydropyranic steroids. In addition, a series of DFT computations provided the theoretical evidence of key oxacarbenium ion intermediate that was formed in our reaction conditions. Furthermore, we have proven the synthetic usefulness of the DHP side chain under protic acid media and obtained the precious 22-oxocholestane side chain in excellent yields. The use of HBr/AcOH 5% allowed us to determine that the regioselective nucleophilic attacks takes place at C-26.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.orglett.6b00492.

X-ray crystallographic data of **10** (CIF)

Experimental section; complete characterization of compounds **7**−**10**; computational details; details of X-ray collection, solution, and refinement of **10**; NMR spectra of compounds **7**−**10**; Cartesian coordinates of all the stationary points involved in the mechanism of transformation of **4**−**7** (PDF)

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Notes

The authors declare no competing financial interest.

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