# The Isolation and Synthesis of Neodolastane Diterpenoids

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The Isolation and Synthesis of Neodolastane Diterpenoids†

Dean Marković,a,b,* Maria Kolympadi,b Brigitte Deguin,a François-Hugues Porée,a Māris Turksc

The neodolastane diterpenoids comprise a group of 44 compounds including guanacastepenes, heptemerones, plicatilisins, radianspenes, 2,15-epoxy-5,13-dihydroxyneodolast-3-en-14-one and sphaerostanol. These fungal and marine natural products are characterized by tricyclic neodolastane skeleton which consists of fused five-, seven- and six-membered rings. The reported antibiotic activity against antibiotic-resistant bacteria together with strong antifungal and anticancer activities and their novel structures render these compounds as interesting synthetic targets. The aim of this account is to summarise the progress in the isolation, characterisation and synthesis of these diterpenoids as well as to review their biogenetic origin and diverse biological activities since their discovery in 2000.

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1 Introduction

Natural products have served as a rich source of biologically active substances and as a precious wealth in drug discovery processes.1–4 Approximately 50% of the small pharmaceutically important molecules during the period from 2000 to 2010 have been connected to the field of natural products.5 Since their early history and Ruzička’s discovery of “Biogenetic Isoprene Rule” in 1953,6 terpenoids7 have emerged as the largest family among natural molecules, with about 360 diverse and complex skeletal types and over 55,000 members.8 In the past, most of these compounds have been isolated from plants, whereas in recent years, novel skeletons have emerged mostly from fungal9 and marine resources.10–13

Due to their unique structures and interesting biological activities, tricyclic C7-C7-C6 diterpenoids14 represent excellent targets for total synthesis.15 In particular, neodolastanes, a structurally diverse family of 44 diterpenoids which includes guanacastepenes, heptemerones, plicatilisins, radianspenes, 2,15-epoxy-5,13-dihydroxyneodolast-3-en-14-one and sphaerostanol,16, 17 inspired numerous synthetic research groups. The studies towards their syntheses were the subject of Mischne’s (2005),18 Hierseman’s (2005 & 2006),15, 19 and Lee’s (2006),20 reviews. The excellent review by Baran (2007) on modern synthesis of biologically active terpenoids also shortly address the synthesis of guanacastepenes.21

Neodolastane skeleton 1 was firstly proposed by Vidari as an intermediate in the biosynthesis of trichoaurentianolide A in 1995.22, 23 Due to the geographical origin of the first characterised member of the family, guanacastepene A (2) found in Guanacaste region of Costa Rica, Clardy and co-workers rename the skeleton to guanacastane skeleton 1 (2000).16 Similarly to 2, guanacastepenes B-O (3-16) were
isolated a year later form endophytic fungus CR115 growing on the branches of Daphnopsis americana (Thymelaeaceae) tree.\textsuperscript{17} Isoskeletal heptemerones A-G (17-23) were extracted from a fungus, Coprinus heptemerus (Psathyrellaceae) (2005),\textsuperscript{24} and 2,15-epoxy-5,13-dihydroxyneodolast-3-en-14-one (24) from basidiomycetes of Trametes corrugata (Polyporaceae) (2009).\textsuperscript{25} Sphaerostanol (45) was obtained from the red algae (Sphaerococcaceae) (2010),\textsuperscript{26} radianspenes A-M (25-36) from fungal strains Coprinus radians M65 (Coprinaceae) (2012),\textsuperscript{27} and plicatilisins A-H (25-36) from basidiomycete of macrofungi, Coprinus plicatilis 82 (Coprinaceae) (2014).\textsuperscript{28, 29} In the context of this review, we summarise the advances in the isolation and characterisation of diterpenoids with neodolastane skeleton. The diversity of structural motifs in connection to the detailed conformational analysis of the guanacastepene A are disclosed as well as biogenetic pathway and the biological activities. Lastly, synthetic studies and achievements in the total synthesis of guanacastepenes and heptemerones published after 2006 are discussed. The work reported earlier was summarized in previous reviews\textsuperscript{15, 18-21} and is briefly reported here (vide supra).

2 Structural diversity of neodolastanes
The structures of guanacastepenes A–O (2-16) were determined by X-ray crystallography.\textsuperscript{16, 17} The family is characterized by the typical tricyclic C5-C7-C6 neodolastane skeleton 1. Many members have supplementary heterocyclic and carbocyclic rings. Fused furan or pyrrole systems whose heteroatom is positioned at C2, are present in guanacastepenes D–O, (5-16). 1,4-Oxazepine motif exists in guanacastepenes D (5) and H (9). Guanacastepenes L (13) and M (14) possess an additional fused tetrahydrofuran arriving from cyclisation at C13 while guanacastepene K (12) contains the more complex norbornane ring system.

In 2005, Sterner and co-workers isolated and characterised seven new neodolastanes, named heptemerones A-G, (17-23).\textsuperscript{24} The heptemerone family was presented with the same C8- and C11-configurations as reported for the guanacastepenes although only the relative configurations were determined (the same numbering as for neodolastane skeleton (1)). The final
proof of the absolute configuration was obtained by the first total synthesis of (-)-heptemerone B (18) conducted by Trauner’s group,30 presuming that other members of the family have the same absolute configuration. Indeed, configuration and substitution patterns of heptemerones are similar to guanacastepenes. For example, the structure of heptemerone G (23) resembles the structure of guanacastepene A (2), but is missing an acetoxy group at C13. Other members of this family possess an additional furane ring connecting C2-C3-C4 and C15 atoms similarly to guanacastepenes E-G, (6-8). A supplementary hydroxyl group at C1 can be found for heptemerones C-F, (19-22) and interestingly, the A ring of heptemerone A (17) is oxidatively cleaved. In 2009, Liu and co-workers reported the isolation of a new diterpenoid named 2,15-epoxy-5,13-dihydroxyneodolast-3-en-14-one (24) together with guanacastepene E (6).25 The similar spectroscopic data of 24 to those of 6 and 18 indicated that compound 24 is their 5-O-and 13-O-deacetylated version. Shen et al. (2012) isolated thirteen diterpenoids named radianspenes A-M, (25-36).27 Their structures were compared to the guanacastepenes. Radianspene A (25), B (26) and C (27) resemble guanacastepene A (2) structure, whereas radianspenes E-I, (29-32) are related to guanacastepene E (6), with O-atoms at the same positions, but at different oxidation stages. Interestingly, the additional oxidation at C-19/C-20 in the terminal position of the isopropyl group is present in the radianspene D (28). Thus, this compound can be considered as the “missing link” in the biogenesis of guanacastepenes K-M, (12–14) (vide infra).

More recently, the same group isolated eight new neodolastanes named plicatilisins A-H (37-44) from fungi Coprinus plicatilis.28, 29 The relative configuration of plicatilisin A (37) was suggested from NOESY experiments and compared to published data for guanacastepene N (15). The lactone ring of plicatilisin B (38) is opened and reduced to diol, whereas C14 and C13, which bear hydroxyl moieties in plicatilisin A (37), are replaced by methylene and ketone groups, respectively. Interestingly, plicatilisin C (39) is the only member of guanacastepenes-like diterpenoids with an anti-relation between the two hydroxyl groups at C13- and C14-atoms. Compared to other members of the family, plicatilisin D (40), G
(43) and H (44) have a supplementary furan-derived ring, similar to guanacastepene E (6), F(7), J (11) or N (15). Plicatilisin E (41) is structurally related to radianspene C (27), with an additional double bond between C6-C7. Plicatilisin F (42) undergoes the keto-enol tautomerisation and can be correlated to guanacastepene A (2).

The structure of a new bromoditerpenoid alcohol, spicaherostanol (45), and its relative stereochemistry, was elucidated by Roussis and co-workers.

Oppositely from molecules of terrestrial origin, this marine natural product is fully saturated and possesses the angular methyl groups in a cis-relationship with the halogen atom located at C7.

3 Conformational analysis of guanacastepene A structure

Pseudo-twist-boat TB-2 and pseudo-chair C-2 conformations of guanacastepene A with their corresponding energies as computed by the MM2 force field calculations are shown in Scheme 1. In the pseudo-twist-boat conformation TB-2 the molecule is bent. This conformation was observed in the X-ray crystal structure and surprisingly, it is 0.14 kcal/mol less stable than the corresponding pseudo-chair conformation C-2 with an energy barrier of 15 kcal/mol at r.t., as computed by Clardy et al. Although there has been some disagreement in the literature, in the case of non-substituted cycloheptene, the calculations of several groups indicate the preference of the chair over the twist conformation by 0.57-1.10 kcal/mol. The observed conformational isomerism can explain the missing $^{13}$C-NMR signals as the isomers can slowly interconvert at r.t. and populate both conformational states, thus masking many of the NMR signals at the NMR time scale. The cycloheptene ring is fused to cyclohexene via sp$^2$ C3- and all carbon quaternary C8-atoms. The cyclohexene ring is in a half-chair conformation with the hydroxyl group in pseudo-axial position. Interestingly, alkene orbitals of α,β-unsaturated aldehyde and α,β-unsaturated ketone are almost perpendicular to each other, greatly diminishing electron exchange and, thus, these groups can be considered as two distinct entities in a pseudo-twist-boat conformation TB-2. However, the rotation around C5-C6 and C6-C7 bonds allows the inter- conversion from one to another half-chair that permits better overlap of orbitals in the 1,3-dienyl system.

As computed by Clardy and co-workers using MM2 force field calculations at r.t. and as confirmed by X-ray analysis.

4 Biogenesis of guanacastepenes

Neodolastane skeleton $^{12}$ is related to marine origin dolastane skeleton $^{32, 33}$ by the transannular cyclisation or to the neodolabellanes by a Wagner Meerwein migration of methyl group from the dolabellane skeleton 46 (Fig. 1). $^{34-37}$ Although no biosynthetic studies on this family of compounds were conducted, Clardy $^{16, 17}$ and Hiegermann$^{15}$ proposed biogenesis of neodolastane skeleton as a series of enzyme-catalysed ring closures and Wagner Meerwein migrations from geranylgeranyl pyrophosphate (47) (Schemes 2 and 3).

![Scheme 1](image-url)
Scheme 2. Proposed biogenesis of the neodolastane skeleton 1.

Two sequential cyclisations of 47 with the loss of the pyrophosphate group led to the formation of dolabellyl carbocation 48, which undergoes [1,2]-rearrangements of hydrides and affords the carbenium ion 50. Subsequent Wagner Meerwein migration of the methyl group gives the neodolabelly cation 51. The elimination of proton at C2 and reprotonation at C7 produces carbocation 52 that is a substrate for a transannular cyclisation to give neodolastyl carbocation 53. The latter eliminates the proton at C4 and forms an unsaturated version of neodolastane skeleton 1, neodolasta-12,14-diene, (54).

Scheme 3. Proposed biogenesis within the guanacastepene family. Scheme 3 shows possible further modification of the skeleton within the guanacastepene family. The proposed biogenesis includes transformations I-VI. The simplest members are guanacastepenes B (3) and C (4) and their oxidised counterpart.
guanacastepene A (2), (transformation I). The functionalisation and oxidation of these compounds or compounds similar to them leads to the formation of more complex skeletal systems of guanacastepenes D-O, (5-16). For instance, guanacastepenes E-G, (6-8) are the products of direct Michael addition of C15-OH to the α,β-unsaturated ketone. Guanacastepenes I (10), J (11), N (15) and O (16) or guanacastepenes H (9) and D (5) can be produced by the oxidation of the C15-angular group followed by intermolecular Michael addition of water (transformation III) or ethanamine (transformation IV), respectively. Oxidation of the isopropyl methyl group into an alcohol followed by intramolecular cyclisation provides skeleton typical for guanacastepenes L (13) and M (14) (transformation V) whereas guanacastepene K (12) can be formed by the aldol reaction of aldehyde at C19/C20 with enolate at C1 (transformation VI). Tautomeric and conformational equilibria may still lead to a greater structural diversity.

5 Biological activities

The action of neodolastanes on living organisms are diverse. These natural products have essentially been the object of preliminary biological studies while the comprehensive biological assays or studies concerning structure-activity relationships were not conducted. As depicted in Tables 1-3, these diterpenoids were tested as antibiotic, antifungal and anticancer agents. Importantly, guanacastepene A (2) showed antibiotic activity against antibiotic-resistant Gram-positive bacteria (Table 1a).16, 17, 38 This compound also exhibited moderate activity against a panel of Gram-positive and Gram-negative bacteria as well as Candida albicans. Although the antibacterial activities of heptemerones were weak (Table 1b) this family of compounds showed potent activity against the fungal germination (Table 2).39 Cytoxicities of these compounds were moderate (Table 3a) and apart from heptemeron D (20), other members of the family were not phytotoxic. In vitro IC₅₀ growth inhibitory values of radianspenes,27 plicatilisins,28, 29 and phaerostanol (45)26 against different tumour cell lines were also determined (Table 3b-d). Among them, the most important is the cytotoxicity activity of radianspene C (27) with an IC₅₀ value of 0.91 μM against breast adenocarcinoma.

In further text, the results of antibiotic, antifungal and cytotoxic tests concerning the mentioned diterpenoids are systematically summarised.

5.1 Antibiotic activities

The screening of biological activities of guanacastepene A (2) revealed antibacterial properties against methicillin-resistant Staphylococcus aureus (MRSA) and vancomycin-resistant Enterococcus faecalis (VREF) (Table 1a).16, 38 When agar plates streaked with MRSA were treated with 50 µg of guanacastepene A (2) or 30 µg of vancomycin, 7-10 mm and 17 mm zones of growth inhibition were observed, respectively.

The same treatment against VREF produced 8 mm antibacterial inhibition zones for guanacastepene A (2) while vancomycin was not active. Guanacastepene A also showed activity against Escherichia coli and Candida albicans. Nevertheless, further biological studies severely diminished the potential of guanacastepene A (2) to be employed as a potential antibiotic agent due to its moderate activity against bacteria and the haemolytic activity against human red blood cells, most likely by nonspecific membrane lysis.39 To the best of our knowledge, biological assays of other members of the guanacastepene family, B-O (3-16), have never been reported.
Magnaporthe grisea or E. coli by heptomerone G (23) and guanacastepene A (2), respectively.

5.2 Antifungal activities

Anke and co-workers also examined the antifungal activities of heptomerones against Bipolaris victoriae, Botrytis cinerea, Colletotrichum graminicola, Drechslera oryzae, Fusarium solani, Magnaporthe grisea, Ascochyta pisi, Cladosporium cladosporioides, Penicillium notatum and Septoria tritici (Table 2a).

Heptomerone G (23) was the most active compound with minimum inhibitory concentration (MIC) in the range of 1µg/ml. Interestingly, MIC is highly influenced by the composition of the assay medium. In water heptomerones were 5-10 times more active than in complex YMG-media (0.4% glucose and Czapek yeast medium). Heptomerone G (23) also inhibited the growth of M. grisea at 1 µg/ml in the leaf segment assay with Oryza sativa and Hordeum sativum (Table 2b).

Table 2. a) Inhibition of germination of conidia by heptomerones C (19), D (20), F (22) and G (23). b) Antifungal activities of same compounds in the leaf segment assay with Oryza sativa and Hordeum sativum. M. grisea was used as plant-pathogenic fungus. Values given are MIC [µg/ml].

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<th>F (22)</th>
<th>G (23)</th>
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<td>&gt;100</td>
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<tr>
<td>Hordeum sativum</td>
<td>20</td>
<td>50</td>
<td>50</td>
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*YMG-medium consists of glucose 1%, malt extract 1% and yeast extract 0.4% in tap water at 22°C.

5.3 Cytotoxic activities

The cytotoxic activities of heptomerones were examined in vitro against four human cancer cell lines including T cell leukaemia (Jurkat), monocyctic leukaemia (Mono-Mac-6), cervical cancer (HeLa) and hepatocellular carcinoma (HepG2) (Table 3a). Heptomerone G (23) was again the most active against Mono-Mac-6 with an IC50 value as low as 2.8 µM. Although no activities of heptomerone A (17) in the other tests were observed, this compound possessed a cytotoxic activity in a range of 10.8-216.3 µM. Heptomerones A (17) and C (19) also displayed an interesting cytotoxic activities.

The in vitro cytotoxicity of radianspene A27 and plicatilisin A28, 29 were evaluated against human cancer cell lines using cisplatin or doxorubicin as positive controls (Table 3b and c). The tests were conducted using breast adenocarcinoma (MDA-MB), hepatocellular carcinoma (HepG2), cervical cancer (HeLa), stomach adenocarcinoma (BGC-823), colon cancer (HCT 116), and osteosarcoma (U2OS) cell lines. Acetate exchange for radianspene A (25), B (26), E (29), H (31) and I (32) was observed under testing conditions. Notably, the radianspene C (27) exhibited significant cytotoxic activity with IC50 of 0.91 µM against the human breast carcinoma MDA-MB-435 cell line (Table 3b).

The in vitro anti-tumour activity of sphaerostanol (45) was probed together with 15 other isolated secondary metabolites against four human apoptosis-resistant (U373, A549, SKMEL-28, OE21) and two human apoptosis-sensitive (PC-3, LoVo) cancer cell lines (Table 3d).

6 Synthetic approaches

A wide range of original synthetic approaches to guanacastepene and heptomerone tricyclic skeletons have been developed and can be broadly divided into three groups: a) synthesis of hydroazulene core, b) synthesis of tricyclic guanacastepene scaffolds and c) total synthesis of...
guanacastepenes and heptomerones. Thus, the results of the syntheses are accordingly summarised in further text.

### 6.1 Synthesis of hydroazulene core

Disconnection of C3-C4 and C7-C8 bonds has proven to be the most popular approach to the neodolastane skeleton 1 leading to hydroazulene core 55 (Fig. 2). The bicycle 55 was successfully synthesised by Magnus, Tius, Chiu, Srikrishna and Greaney, while the approach via this synthon was also used in the total synthesis of guanacastepenes A, C and heptemerone G by Danishefsky, Snider, Mehta and Wicha.

Table 4 and Scheme 4 summarise methodologies, key disconnections, number of steps and yields relevant to the construction of hydroazulene core 55. Magnus’ synthesis of synthon 55 relays on the C1-C11 and C9-C10 bond disconnections and was accomplished in 18 steps from commercially available isobutylmethyl ketone as shown in Table 4. The key step of the synthesis, a pyrylium-ylide [5+2] intramolecular cycloaddition, is highlighted in Scheme 4a. Elimination of acetic acid from the acetal provided zwitterionic pyrylium-ylide that smoothly underwent the stereoselective cycloaddition giving enone 58 in 80% yield.

![Fig. 2 The disconnection of guanacastane 1 to hydroazulene core 55.](image-url)
This compound was further transformed into fully functionalised enone 59. Tius\(^{42}\) and Srikrishna's groups\(^{44}\) employed the ring closing metathesis approach to close the seven-membered ring disconnecting C3-C8 or C8-C9 bonds, respectively (Entries 2 and 4; Table 4; Scheme 4b and d). Metathesis of 60 under high dilution with second generation Grubbs catalyst afforded 61 (82%), which was further converted into epoxy ketone 62 in 65% yield. Chiu and co-workers reported an elegant synthesis of hydroazulene core 66 using a diastereoselective Nazarov cyclisation (Scheme 4c).\(^{43}\) Ketoester 63 was first converted into dieneone 64 in 4 steps with 77% yield. Lewis acid mediated Nazarov cyclisation of 64 provided hydroazulene core 65 (98%) as a single diastereomer in 98% yield. Thus, hydroazulene core 66 was obtained readily.

**Table 4.** The disconnection of guanacastane 1 to hydroazulene core 55 and overview of the used methodologies.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Group</th>
<th>Year</th>
<th>Methodology</th>
<th>Key Disconnect</th>
<th>Number of steps</th>
<th>Total yield/%</th>
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<td>2001</td>
<td>pyrylium ylide [5+2] cycloaddition</td>
<td>C1-C11, C9-C10</td>
<td>18</td>
<td>1.9</td>
<td>34, 35</td>
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<tr>
<td>2</td>
<td>Tius</td>
<td>2002</td>
<td>RCM</td>
<td>C3-C8</td>
<td>12</td>
<td>7.1</td>
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<td>3</td>
<td>Chiu</td>
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<td>Nazarov cyclisation</td>
<td>C11-C12</td>
<td>6</td>
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<tr>
<td>4</td>
<td>Srikrishna</td>
<td>2004</td>
<td>RCM</td>
<td>C8-C9</td>
<td>9(^{a})</td>
<td>(^{b})</td>
<td>44</td>
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<td>5</td>
<td>Greaney</td>
<td>2007</td>
<td>photochemical rearrangement</td>
<td>C1-C11</td>
<td>13</td>
<td>1.7</td>
<td>45</td>
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</table>

\(^{a}\)asymmetric; \(^{b}\)yield not available.
Table 5 summarises the synthetic approaches to guanacastepene core. In 2002, Lee utilised reductive ring closure as the key step to form C2-C3 bond. Kwon (2003) employed Mukaiyama aldol reaction to construct C1-C2 bond and Brummond (2003) used Pauson-Khand reaction for the formation of C1-C11, C12-C13 and C13-C14 bonds. Finally, Sato (2010) used his rhodium-catalysed hydroacylation cycloisomerisation methodology. The synthesis were achieved in eleven to nineteen steps with 2 to 34% overall yield. One of the first asymmetric synthesis of guanacastepene tricycle was reported by Lee and co-workers (Scheme 5a). They synthesised intermediate 78 by copper-catalysed 1,4-addition of (1S)-citronellyl-derived Grignard reagent to (1S)-verbenone 76. Both reactants are readily available chiral pool materials. The formation of C2-C3 bond was performed by intramolecular Sn2 reductive coupling of the aldehyde function of 79 onto the α,β-unsaturated ketone that provided guanacastepene I precursor 80 in 90% yield. This methodology also allowed the correct installation of stereochemistry at C8-quaternary carbon because thermodynamically more favoured cyclisation leading to trans-relationship of the two quaternary methyl groups.

In another study, Kwon and co-workers utilised a Diels–Alder reaction involving diene 81 which can be prepared from ethoxycetylene in two steps with 74% yield, and maleic acid under high pressure and temperature to obtain the precursor of highly functionalised intermediate 82 (32%), as shown in Scheme 5b. Copper-catalysed conjugate addition of organozinc reagent of 82 to α,β-unsaturated ketone 83 gave 84 in an 82% yield. Sequential conjugate addition of Me2CuLi to enone 84 and TiCl4-mediated intramolecular Mukaiyama type aldol reaction was followed by the elimination catalysed by p-toluenesulfonic acid (p-TsOH) and produced two tricyclic epimers with methyl groups in cis- and trans-relationships, cis-85 and trans-85 (cis-85/trans-85=1:1, 30% for 2 steps). The methyl magnesium bromide addition catalysed by copper bromide dimethyl sulfide complex followed by elimination with p-TsOH improved the ratio to 1:2:1. By this synthetic route, guanacastepene core 85 was obtained in 13 steps with 6% overall yield (Entry 2; Table 5).

Brummond’s group reported the construction of tricycle 89 by employing a rhodium-catalysed Pauson–Khand reaction (Scheme 5c). Smith’s enone 86 was transformed into dialkyne 87 in 14% over 8 steps. Mesylation of the pendant propargylic alcohol of 87 followed by the direct addition of (Me2PhSi)2CuCNLi2 afforded the desired allene 88 in 90% yield over two steps. Rhodium-catalysed Pauson-Khand reaction successively formed C1-C11, C12-C13 and C13-C14 bonds of the hydroazulene system of skeleton 89 in 65% of yield. Due to steric hindrance of the isopropyl group, all further attempts to install the quaternary C11-stereocentre by copper-catalysed conjugate addition were not successful and led to fulvene containing product resulting from the dehydration of the 1,2-adduct. The overall yield of this reaction sequence was 8.2% for eleven steps (Entry 3; Table 5).

In 2010, Sato and co-workers reported a rhodium(I)-catalysed cascade reaction consisting of the hydroacylation cycloisomerisation steps leading to the easy formation of skeleton 93. The synthesis of the substrate syn-91 was performed in a stereoselective manner from hemiacetal syn-90 (Scheme 6). When syn-91 was treated with a catalytic amount of [Rh(dppe)]ClO4 (dppe = 1,2-bis(diphenylphosphino)ethane), generated in situ, the desired C3–C7–C8 tricyclic compound syn-93 (54%) and bicyclic syn-94 (12%) were obtained via the intermediary of eight-membered rhodacycle 92. This procedure allowed the formation of three carbon-carbon bonds in one step and yielded syn-93 in 34% overall yield over eleven steps (Entry 4, Table 5). Similarly, the treatment of anti-91 produced the mixture of desired anti-93 in 9% yield with contaminant formation of bicyclic anti-94 (32%).
6.3 Total synthesis of guanacastepenes and heptomerones

Table 6 summarises the methodologies, key disconnections, number of steps and yields of the conducted total syntheses of guanacastepenes and heptomerones. En route to guanacastepenes A (2) and C(4), Danishefsky,54,61 Snider,62-64 Mehta73-76 and Wicha70-72 formed the C3-C4 bond of the six-membered ring by Knoevenagel or intramolecular condensations while Hanna65,66,84 used tandem ring closing metathesis reaction to build three new C-C bonds. Sorensen created the C10-C11 bond of central seven membered ring of guanacastepenes A (2) and E (6) by [2+2]-cycloaddition fragmentation methodology.57,85 Yang’s group used intramolecular Diels–Alder (IMDA) cycloaddition to construct the C3-C8 and C4-C5 bonds of the C8-epi-guanacastepene O, C8-epi-(16).77 Overman employed the rare 7-endo-Heck cyclisation as a key step in his synthesis of guanacastepene N (15)81 and Trauner developed electrochemical oxidation leading to formation of C1-C2 bond of heptomerone B (18) and guanacastepene E (6).30 Finally, Carreira utilised his original methodology consisting of cycloinsertion and fragmentation steps to construct guanacastepene N (15) and guanacastepene O (16).83 Statistically, 23 steps were needed to accomplish total synthesis of guanacastepenes or heptomerones with average yield of 2.7%.

The pioneering work in total synthesis of guanacastepene A (2) was performed by Danishefsky’s group.54,61 Many of the following synthetic studies developed reaction sequences to Danishefsky’s intermediates or adopted his chemistry to solve the problematic synthetic steps. The synthesis of (+)-guanacastepene A (2) is outlined in Scheme 7. Ketone 96 was obtained in two steps with 71% overall yield from commercially available 2-methyl-2-cyclopentenone (95).86 The intramolecular reductive cyclisation of vinyl iodide of 96 onto the pendant ketone was achieved via the vinyllithium intermediate generated in situ by halogen-metal exchange. The corresponding allylic alcohol underwent an oxidative rearrangement with PCC providing hydroazulene core 97 in 70% yield over 2 steps. From this intermediate, Danishefsky group developed different synthetic routes to correctly install the relative stereochemistry of C8 centre in anti-relationship respect to the angular methyl group at C11. Hydroazulene core 97 was finally transformed to β-ketoester 99 in 10 steps with 19% yield via intermediate 98. The eliminative epoxide opening and intramolecular Knoevenagel condensation...
furnished tricyclic intermediate 100 (74%) with completed neodolastane skeleton 1. Further functionalisations were directed towards correct installation of the oxygenation and unsaturation groups at the north edge of the skeleton of guanacastepene core to provide acetonide 101 (50%) over four steps. Rubottom oxidation of silylenol ether derived from 101 with dimethylsulfoxide followed by acetylation of the corresponding alcohol provided 102 with a correctly installed C13-acetoxy substituent. Acetonide deprotection followed by rapid oxidation of an unstable diol completed the synthesis of (±)-guanacastepene A (2). Snider and co-workers utilised an intramolecular Prins type reaction and an aldol condensation as the key step in their formal synthesis of guanacastepene A (2) (Table 3 and Scheme 8). The synthetic intermediate 104 was obtained in 60% yield over 4 steps. The EtAIICl-promoted cyclisation followed by sequential [1,2]-hydride and methyl shifts of 104 provided the cyclopentanone 106 in 69% yield as the only cyclic product with correct syn-stereochemistry of methyl and isopropyl groups. The latter in two steps was transformed into triene 107 (65%). The ring closing metathesis of 107 furnished a hydroazulene core 108 (88%) that was further elaborated to enone 109 (29%). Under basic conditions, the aldol condensation precursor 109 gave the desired tricyclic structure 110 that was transformed in 3 additional steps in Danishefsky’s acetonide 101 with 42% overall yield. Interesting formal total synthesis of guanacastepene A (2) based on Danishefsky’s acetonide 101 using a tandem ring-closing-metathesis (RCM) was reported by Hanna and co-workers. As shown in Scheme 9, trienylene 112 was obtained from cyclopentanone 111 (9%) in 9 steps. This relatively simple substrate 112 underwent tandem ene-yne-ene RCM and rapidly built up tricyclic skeleton 113 (82%). Further functionalisation provided Danishefsky’s acetonide 101 over seven steps. Afterwards Hanna and co-workers prepared a series of tricyclic guanacastepene like structures 115a-e (Table 7). Authors probed the influence of substituents R1 and R2 on ring closing metathesis using second generation Grubbs catalyst. Except for 114c that remained unchanged, all other dienynes were easily converted into the expected products in high yields (70-93%) regardless the R1 and R2 substitution.

Mehta’s total synthesis of guanacastepene C (4) is disclosed in Scheme 10. The authors used ring-closing-metathesis and Knoevenagel condensation as the key methodologies. Readily available endo-eneone 116 was employed as a masked dieneone to control the stereochemistry during the preparation of cyclopentanone 117 by exclusive reactivity on its exo face. Compound 116 was transformed into enone 117 in 6 steps with 27% overall yield. The ring closing metathesis of terminal alkenyl groups of 117 readily provided hydroazulene core 118 in 95% yield. β-Ketoester 119 was obtained in eighteen steps and underwent an intramolecular Knoevenagel condensation to provide the tricycle that was further transformed to guanacastepene C (4) in an eight step sequence.

In 2006, Sorensen’s group reported an asymmetric formal synthesis of guanacastepene A (2) and a first total synthesis of guanacastepene E (6) (Schemes 11 and 12). They employed (S)-(+)-carvone (67) as a chiral pool starting material to introduce C12-stereocentre of cyclopentanone portion 123 of the molecule as a single enantiomer in an 8 steps sequence with 11-14% yield. The α-acyloxy nitrile 121, obtained in 5 steps (41%) from 67, was deprotonated with lithium bis(trimethylsilyl)amide (LiHMDS) to afford the enol tautomer, dione 122 in 58% yield via the intermediacy of postulated epoxy alkoxide ion 121. The exchange of vinyl nonafluorobutanesulfonate (NF) from 122 to vinylstannane 123 by palladium-catalysed coupling with hexamethylditin was challenging due to the steric effect of isopropyl group and leads to the formation of 123 in 59% yield over 2 steps. The ring C fragment was prepared by the classical resolution method using S-(-)-O-acetyl mandelic acid (Scheme 11). Cyclohexenone 124 was transformed to Danishefsky diene that underwent Diels-Alder reaction with dimethyl...
acetylenedicarboxylate (DMAD) giving cyclic adduct 125 in 97% yield for 2 steps. This compound 125 was further transformed to racemic allylic alcohol 126 (87% yield over two steps), which was esterified with inexpensive O-acetyl (S)-(+) -mandelic acid and the diastereoisomers were easily separated by flash column chromatography. Further exchange of mandelic ester 127 to acetic acid ester 128 was performed in 2 steps (97%).

Table 6. Different approaches to the total syntheses of guanacastepenes and heptemerones.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Group</th>
<th>Year</th>
<th>Compound</th>
<th>Methodology</th>
<th>Key Disconn.</th>
<th>Number of steps</th>
<th>Total yield (%)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Danishefsky</td>
<td>2002</td>
<td>(+)-guanacastepene A</td>
<td>Knoevenagel condensation</td>
<td>C3-C4</td>
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<td>54, 61</td>
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<td>(+)-guanacastepene A</td>
<td>Intramolecular condensation</td>
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<td>19</td>
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<td>62</td>
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<td>3.</td>
<td>Hanna</td>
<td>2004</td>
<td>(+)-guanacastepene A</td>
<td>tandem RCM</td>
<td>C2-C3, C4-C5</td>
<td>17</td>
<td>.6</td>
<td>65</td>
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<tr>
<td>4.</td>
<td>Mehta</td>
<td>2005</td>
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<td>Knoevenagel condensation</td>
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<td>33</td>
<td>0.13</td>
<td>73, 74, 76</td>
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<td>Yang</td>
<td>2006</td>
<td>(+)-8-epi-guanacastepene O</td>
<td>IMDA</td>
<td>C3-C8, C4-C5</td>
<td>16</td>
<td>1.9</td>
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<td>Overman</td>
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<td>(+)-guanacastene N</td>
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<td>C2-C3</td>
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<td>Trauner</td>
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<td>(-)-heptemerone B &amp; (-)-guanacastepene E</td>
<td>electrochemical oxidation</td>
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<td>3.2</td>
<td>30</td>
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<td>Wicha</td>
<td>2010</td>
<td>(+)-guanacastepene G &amp; (+)-guanacastepene A</td>
<td>Knoevenagel condensation</td>
<td>C3-C4</td>
<td>38</td>
<td>4.0</td>
<td>71</td>
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<tr>
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<td>Carreira</td>
<td>2011</td>
<td>(+)-guanacastepene N</td>
<td>cycloinsertion</td>
<td>C2-C3, C8-C9</td>
<td>23</td>
<td>0.09</td>
<td>83</td>
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</table>

*longest sequence; ^formal total synthesis; ^asymmetric; ^yield not available

Scheme 7. First total synthesis of (+)-guanacastepene A (2) by Danishefsky and co-workers. PCC = pyridinium chlorochromate; DMDO = dimethyldioxirane.

Construction of the central seven-membered ring commenced by a Stille coupling under Corey conditions (CuCl additive) which unified allylic acetate 128 with stannane 123 and furnished cycloaddition substrate 129 in 91% yield (Scheme 12). Allylic mandelate 127 was also a good substrate for this reaction, slightly diminishing yield to 78%. The irradiation-mediated intramolecular [2+2]-cycloaddition between pendant olefin and the tetrasubstituted cyclopentene double bonds of 129 proved exclusively adduct 130 in 82% yield. Observed diastereofacial selectivity is enforced by the presence of the isopropyl group which largely dictates the configuration at the second all carbon quaternary centre providing the desired [2 + 2] adduct 130.
Scheme 8. Snider's formal synthesis of (±)-guanacastepene A (2). Grubbs I cat. = \( \text{Cl}_2(\text{PCy}_3)_2\text{Ru} = \text{CHPh} \).

Scheme 9. Hanna's formal synthesis of (±)-guanacastepene A (2). Grubbs II cat. = \( \text{Cl}_2(\text{PCy}_3)(\text{NHC})\text{Ru} = \text{CHPh} \).

Table 7. Tandem RCM of dienynes 114a-e. Grubbs II cat. = \( \text{Cl}_2(\text{PCy}_3)(\text{NHC})\text{Ru} = \text{CHPh} \).

A selective reductive fragmentation of ketone 130 was achieved with samarium diiodide and the resulting putative samarium enolate was trapped with phenylselenenyl bromide giving the corresponding organoselenide that was oxidised with meta-chloroperoxybenzoic acid (m-CPBA) and eliminated to generated the complete tricyclic [5-7-6] ring system of the guanacastepenes 131 in 43% overall yield. Enone 131 was transformed to advanced Danishefsky's intermediate 101 by exchange of the benzylidene acetal to the isopropylidene ketal of 101 completing the formal synthesis of (+)-guanacastepene A (2). From tricycle 131, Sorensen also performed the first total synthesis of (+)-guanacastepene E (6) in five steps with 31% yield.

Scheme 10. Mehta's total synthesis of (±)-guanacastepene C (4). Grubbs II cat. = \( \text{Cl}_2(\text{PCy}_3)(\text{NHC})\text{Ru} = \text{CHPh} \).
Sorensen's asymmetric synthesis of A and C ring fragments of guanacastepenes. DCC = N,N'-dicyclohexylcarbodiimide; DMAD = dimethyl acetylenedicarboxylate; DMAP = 4-dimethylaminopyridine; dppf = 1,1'-bis(diphenylphosphinoferrocene); LDA = lithium diisopropylamide; LiHMDS = lithium bis(trimethylsilyl)amide; Nf = nonafluorobutanesulfonate; NMP = N-methyl-2-pyrrolidone; PMP = para-methoxyphenol.

Yang and co-workers developed an intramolecular Diels-Alder (IMDA) strategy for the construction of the tricyclic core of guanacastepenes (Scheme 13). Although this approach is attractive in light of its concise and convergent nature leading to the fast construction of the guanacastepene skeleton, the IMDA reaction of E- or Z-precursors E-132 or Z-132 led to formation of thermodynamically controlled product 133 with the inverted stereochemistry of C8 all carbon quaternary centre during the model studies.

Scheme 11. Sorensen's asymmetric synthesis of A and C ring fragments of guanacastepenes.

Scheme 12. Sorensen's asymmetric formal synthesis of (+)-guanacastepene A (2) and total synthesis of (+)-guanacastepene E (6). DMSO = dimethyl sulfoxide; HMPA = hexamethylphosphoramide; m-CPBA = meta-chloroperoxybenzoic acid; PPTS = pyridinium para-toluenesulfonate.

Scheme 13. Yang and co-workers IMDA strategy for the construction of the tricyclic core of guanacastepenes.
As presented in Scheme 14, this approach was further applied for the total synthesis of C8-epi-guanacastepene O, C8-epi-(16). Furane 134 was transformed into cyclopentenone 135 corresponding to the ring A of the guanacastepene skeleton, in four steps with 53% yield. A conjugate addition of cyanocuprate 136 to 135 followed by the aldol reaction produced propargylic alcohol 137 as a mixture of diastereoisomers in 62% yield for 2 steps. Further chemoselective reduction/oxidation reactions using BH3 and (2,2,6,6-tetramethylpiperidin-1-yl)oxy(TEMPO)/bis(acetoxy)iodobenzene (BAIB) gave the substrate for the IMDA reaction, ketone 138 (54%). Similarly to model studies, refluxing of 138 afforded expected guanacastepene core with C8-epimeric stereocentre that was oxidized with Dess-Martin periodinane (DMP) to yield a dione 139 in 64% yield. The construction of additional lactone ring was achieved under the catalytic action of t-BuOK and further pyridinium dichromate (PDC) oxidation installed the required keto function at C5-position. Lactone 140 thus obtained in 43% yield for two steps. Further protecting group manipulation afforded compound 141 (68%, 3 steps) which was treated with diisobutylaluminium hydride (DIBAL-H) to region- and stereoselectively reduce C5-keto function furnishing C8-epi-guanacastepene O, C8-epi-16 in 58% yield.

Scheme 14. Yang’s total synthesis of (±)-C8-epi-guanacastepene O (16). BAIB = bis(acetoxy)iodobenzene; DIBAL-H = diisobutylaluminium hydride; DMP = Dess-Martin periodinane; LDA = lithium disopropylamide; PDC = pyridinium dichromate; TEMPO = (2,2,6,6-tetramethylpiperidin-1-yl)oxy.

In 2006, Overman’s group performed the first asymmetric total synthesis of (+)-guanacastepene N (15). This convergent synthesis used a rare 7-endo Heck cyclisation to close the central ring of the guanacastepene core as outlined in Schemes 15 and 18. The five-membered ring fragment 144 was obtained from dienone 142 using cheap (+)-menthol as a means to control the stereochemical outcome of the Stork-Danheiser reaction of 143 and to separate the formed diastereoisomers by HPLC. Optically active (R)-3-methylcyclohex-2-yl acetate (145) was prepared in 95% e.e. by lipase-catalysed kinetic resolution of racemic 3-methyl-2-cyclohexen-1-ol. The C8 quaternary centre of iodide 146 was formed by Ireland–Claisen rearrangement with erosion of enatiomeric excess (80% e.e.) and with 69% yield over 4 steps. Linkage of five- and six-membered cyclic building blocks was achieved by means of a diastereoselective conjugate addition of the organocuprate reagent deriving from 146 to the enone 144 affording C11 all carbon quaternary centre (Scheme 16). The formed enolate was trapped as the enoxysilane 147. Further functionalisation of the α-carbon was achieved by Eschenmoser methylation providing exocyclic enone 148 in 58% yield from 147. The key step of the synthesis is a Heck cyclisation. Thus, 148 was converted in nine steps in unsaturated triflate 149 with 39-44% yield. The palladium-catalysed intramolecular Heck cyclisation of dienyl triflate 149 as expected took place in

Scheme 15. Overman’s asymmetric preparation of five and six membered ring portions of (+)-guanacastepene N (15). p-TsOH = p-toluenesulfinic acid.
a 7-endo mode affording tricycle 150 in 75% yield. The installation of C13-acetoxy group was achieved by Rubottom oxidation firstly elaborated by Danishefsky (vide supra) and led to the fully functionalised A ring of tricycle 151. Interestingly, the deprotection of benzyl ester of 151 was challenging and required the utilisation of Pd(OAc)₃ in the presence of Et₃SiH to provide all four possible diastereomers of lactone 152 (77%). Allylic bromination of lactone 152 with N-bromosuccinimide (NBS) and benzoyl peroxide installed a bromine atom at a position C5 with a high β-stereoselectivity (α/β=1:20) and simultaneously re-established the central unsaturation in 49-64% yield. As the authors suggested, the observed selectivity is a result of the steric influence of α-oriented pseudoaixial-methyl substituent at C8. The reinstallation of unsaturation at the C1-position likely occurred by allylic bromination followed by elimination of HBr. The retentive replacement of the α-Br substituent with OH was realised by air oxidation of the allylic radical derived from 153 using (n-Bu)₃SnH followed by in situ reduction of the corresponding peroxide intermediate with Ph₃P. (+)-Guanacastepene N (15) and its C5-epimer C5-epi-15 were generated in 47% yield with α,β-diastereoselectivity of 10:1.

Scheme 16. Overman’s total synthesis of (+)-guanacastepene N (15). dba = dibenzylideneacetone; DMF = dimethylformamide; DMA = dimethylacetamide; DMP = Dess-Martin periodinane; dppb = 1,4-bis(diphenylphosphino)butane NBS = N-Bromosuccinimide.

Trauner’s group reported an asymmetric total syntheses of the unnatural enantiomers of (-)-heptemerone B (-)-(18) and (-)-guanacastepene E (-)-(6) (Schemes 18 and 19).³⁰, ⁷⁹, ⁸⁰ The key reaction of these concise and convergent syntheses was an uncommon electrochemical oxidation that closed the central seven-membered ring (Scheme 18). Cyclopentenone building block 157 was obtained from known chiral glyoxalate 154. A chiral auxiliary-mediated carbonyl-ene reaction yielded the desired anti-diastereoisomer 155 as the major product (anti/syn=10:1) that was further transformed to enone 156 in four additional steps with 60% overall yield. Cyclisation of 156 via RCM catalysed by Grubbs’ second generation catalyst afforded the enantiomerically pure enone 157 (86%).

The synthesis of the six-membered ring portion 161 started by the addition of mono-lithiated 3,4-diodofuran (158) to (E)-4-methylhex-4-enal (159) (62%). Oxidation of the obtained alcohol 160 using DMP followed by the enantioselective reduction with (+)-B-chlorodisopinocampheylborane regenerated optically active (R)-160 in 94% e.e. Palladium-catalysed intramolecular Heck reaction formed the first all carbon quaternary centre of the target molecule with a diastereomeric ratio of 5:1:1. The intermediate was further transformed in a four steps sequence to the primary iodide 161 finishing the synthesis of six-membered ring fragment of the molecule.

To link the A and C ring fragments of the molecule the authors combined the protocols of Lipshutz and Yamamoto for cuprate conjugate addition. The organocuprate arriving from 161 was added to the enone moiety of 157 in the presence of BF₃·Et₂O providing 162 in 54% yield and forming C11 all carbon quaternary stereocentre (Scheme 18). Intramolecular formation of C1-C2 bond was achieved by employing Moeller’s and Wright’s anodic oxidation of exoxyxilane generated from 162. This umpolung reaction coupled silyl enol ether with the furane moiety and closed the central seven-membered ring of the ketone 163 in 76% overall yield. Further elaboration of 163 afforded the unnatural enantiomer of (-)-heptemerone B (-)-(18) (35%) in seven steps and after methanolic hydrolysis of acetate, (-)-guanacastepene E (-)-(6) (28%).
Scheme 17. Trauner’s asymmetric synthesis of A and C ring fragments of (-)-heptemerone B (-)-(18) and (-)-guanacastepene E (-)-(6). DMP = Dess-Martin periodinane; [(+)-DIP-Cl] = (+)-B-chlorodiisopinocampheylborane; Grubbs II cat. = Cl₂(PCy₃)(NHC)Ru=CHPh; TBDPS = tert-butyldiphenylsilyl.

Scheme 18. Trauner’s total synthesis of (-)-heptemerone B (-)-(18) and (-)-guanacastepene E (-)-(6) using anionic oxidation as key step. KHMDS = potassium bis(trimethylsilyl)amide; TBSOTf = trimethylsilyl trifluoromethanesulfonate.

In 2010, Wicha and co-workers reported the first total synthesis of (±)-heptemerone G (30) and a formal total synthesis of (±)-guanacastepene A (2) based on Danishefsky’s late-stage acetonide 101.70, 71 Similarly to Tius42 and Srikrishna,44 authors used the RCM strategy for the synthesis of hydroazulene bicyclic core 165 (Scheme 19).72 Cyclopentanone 95 was employed as the starting material and transformed into diene 164 in 4 steps with 49% yield. Ring-closing-metathesis catalysed by Grubbs second generation catalyst formed bicycle 165 in excellent yield. Its further conversion to enone 166 was achieved in seventeen additional steps and 26% overall yield. Lithium enolate derived from 166 was successfully generated using LiHDMDS and hexamethylphosphoramide (HMPA). This procedure, firstly established by Danishefsky, allowed the stereoselective installation of the methyl group of 166. Eight steps homologation sequence of 167 led to β-ketoester 168 that underwent the intramolecular Knoevenagel condensation and afforded tricycle 169 (60% yield over nine steps). An additional three step synthetic sequence via diol 170 provided Danishefsky’s intermediate 101 and thus virtually completed the formal synthesis of (±)-guanacastepene A (2). Three steps transformation of diol 170 also allowed the first total synthesis of (±)-heptemerone G (30) in 55% overall yield. In 2013, Wicha and co-workers disclosed a synthesis of the optically active hydroazulene core 165 using 2-furylmethyl carbinol (171) as the starting material (Scheme 20).49 One-pot Piancatelli rearrangement–isomerisation sequence using 171 and MgSO₄ (10% aq.) at 160°C provided enone 172 in 56% yield. The enzymatic kinetic resolution of rac-172 with Novozym 435® and isopropenyl acetate afforded (S)-172 in 99% e.e. and 45% yield after column chromatography separation. Acetate (R)-173 obtained in 81% ee and 55% yield, was converted into optically active 172 by an adapted Tanis three step sequence based on a hydrolysis, Mitsunobu inversion and an enantiomeric enrichment enhancing the total yield of (S)-172 to 80%. Optically active intermediate of previous synthesis, ketone 165 was obtained in six steps with 39% yield.
Scheme 19. Wicha’s first total synthesis of (±)-heptemerone G (30) and formal total synthesis (±)-guanacastepene A (2). LHMDs = lithium bis(trimethylsilyl)amide; Grubbs II cat. = Cl\textsubscript{2}(PCy\textsubscript{3})(NHC)Ru=CHPh; HMPA = hexamethylphosphoramide; p-TsOH = para-toluensulfonic acid.

Scheme 20. Wicha’s preparation of optically active hydroazulene fragment 165. MTBE = methyl tert-butyl ether.

In 2011, Carreira and Gampe reported the total syntheses of (±)-guanacastepenes N (15) and O (16).

As shown in Scheme 21, the synthesis commenced with the preparation of fused cyclopentanone 175 by a sequence of seven steps that correctly installed C11 all carbon quaternary stereocentre. To obtain guanacastepene tricyclic scaffold, authors employed their original annulative ring expansion cascade consisting of a one-pot addition and ring opening reactions.

Thus, the enolate derived from 175 was reacted with cyclohexyne generated in situ from iodonium salt 176. The obtained cyclobutenol 177 (74%) underwent [Fe\textsubscript{2}(CO)\textsubscript{5}] initiated ring opening giving guanacastepene tricylic core 178 in 51% yield (for further discussion see Schemes 25 and 26). The DIBAL-H reduction followed by cyclopropanation using Shi’s modification of the Furukawa conditions and a re-oxidation back to the ketone stage, afforded 1:1 mixture of diastereomeric cyclopropanes 179 in 60% yield over three steps (Scheme 21). Cleavage of the cyclopropyl ring under Birch conditions installed C8 quaternary centre with correct stereochemistry and was followed by three step sequence to furnish enone 179 (75%) that was further transformed to lactone 181 in 35% yield. Treatment of 181 to TBSOTf/Et\textsubscript{3}N afforded the corresponding bis-TBS-enol ether. Interestingly, exposure of this compound to OsO\textsubscript{4}/NMO triggered the oxidative cascade reaction producing ketone 184 and α-hydroxyketone 183 in 69% overall yield. As proposed by Carreira, the postulated intermediate, osmate 182 first undergoes γ-elimination (H-C3) followed by oxidation to give 183 (29%) or HO-elimination to afford 184 (40%). Although not suggested, the formation of product 184 can also arrive from non-selective osmylation of C4-C15 olefine functionality of bis-silyl enol ether arriving from 181 followed by elimination of H-C3 proton. Similarly, the product 183 may be formed by the dihydroxylation of C13-C14 and C4-C15 olefinic bonds of the same intermediate and elimination of proton at C3. In contrast to β-selectivity of Rubottom oxidation, the installation of C13-hydroxyl group of 183 occurred with a high α-diastereofacial selectivity (α/β=4:1).

From hydroxyketone 183, the total synthesis of guanacastepene O (16) was terminated in three additional steps with 48% overall yield. On the other hand, ketone 184 was transformed to guanacastepene N (15) via Overman’s late intermediate 153.
The treatment of 184 with Mn(OAc)$_3$ installed the acetoxy group at β-side (β/α=4:1, 68%) and was followed by Wohl–Ziegler bromination that gave the desired β-bromide 153 in 58% yield (see also Scheme 16).

In addition to the synthesis of (±)-guanacastepenes N (15) and O (16), Carreira and Gampe synthesised precursors of (±)-guanacastepenes D (5) and H (9), as shown in Scheme 22. The intermediate of the previous synthesis, enone 180, was converted to five aminated diterpenoids 185-189. The reaction of 180 with Nagata’s reagent (Et$_2$AlCN) followed by treatment with HClO$_4$ transformed intermediate nitrile, to 2:1 mixture of C4-epimers of α- and β-185 in 16-25% yields over two steps. The diversification of the carbon scaffold 185 was achieved by the oxidation with high excess of NBS providing dienyl lactame 186 (40%) or with NCS (2.0 equiv.) giving allylic chloride 188 in 95% yield (d.r. α/β=95:5). Interestingly, the use of Me$_3$NPhBr$_3$ led to C13-bromination of lactames 185 and 186 and produced guanacastepene scaffolds 187 (d.r.=4:1, 90%) and 189 (d.r. α/β=1:1; 90%), respectively.

Scheme 21. Carreira’s total syntheses of (±)-guanacastepenes N (15) and O (16). NBS = N-bromosuccinimide; TBSOTf = trimethylsilyl trifluoromethanesulfonate.

6.4 Computational Studies

Achievements in the synthesis of guanacastepenes have opened an avenue for computational research and demonstrated the necessity of their synergy. For instance, Danishefsky and co-workers employed Rubottom oxidation protocol to install C13 acetoxy function of guanacastepene A (vide supra, Scheme 7). Surprisingly, the epoxidation of silyl enol ether 190 with dimethyldioxirane (DMDO) occurred from more congested β-face providing 191 with the synperiplanar epoxy moiety to residual C12-isopropyl and C11-methyl functionalities. This intriguing stereoselectivity was a subject of computational studies performed by Houk, Danishefsky and co-workers.
The corresponding transition structures and energies leading to α- and β-selectivities were calculated employing density functional theory investigations as shown in Scheme 23. Because of the eclipsing interactions, the α-epoxidation transition structure 192α is less stable for 2.6 kcal/mol compared to staggered transition structure 192β for β-epoxidation as shown by Newman projections. This torsional steering is the most important factor and enhances the energetic difference of two transition states preferring β-epoxidation. The high asynchronicity of the formation of C14-O and C13-O bonds in the transition structures still intensifies the torsional interactions.

In another study, Houk, Wicha and co-workers examined the origin of the stereoselectivity of alkylations of lithium enolates of hydroazulenones 193 and 197 by computational methods. As shown in Scheme 24, methylation of the lithium enolates 193 and 197 proceeded with the opposite diastereofacial selectivities. While enolate 193 gives the β-methylated product 195 with unnatural syn-angular methyl groups in 80% d.e. and 72% yield, the reaction of enolate 197 which has an extra C1-C2 double bond, led to the exclusive formation of the α-methylated product 198 (98%). Previously, Danishefsky and Mehta reported the same preferential α-selectivity for intermediates closely related to hydroazulenone precursor of 197.

For both reactions, the steric effects rather than torsional factors were responsible for the formation of the observed products. The pseudo-chair conformation of enolate 193 was computed to be most stable. α-Attack on 193 leads to transition structure 194α that has significant steric repulsion between an H-atom of methyl bromide and two homoallylic Hs with the distances under the sum of their van der Waals radii (2.28Å and 2.31Å vs 2.37Å and 2.62Å Scheme 24). In contrast, only one steric interaction is present in the transition structure of the β-methylation 194β, with the distance 2.37Å. Thus, the transition state for β-methylation is 1.1 kcal/mol lower in energy than the α-methylated transition state, which also corresponds to the observed diastereoselectivity d.e. (195/196)≈80%. As predicted by calculations, the enolate 197 adopts the pseudo-twist-boat conformation with the C8-angular methyl on β-face of the molecule pointing towards the enolate moiety. Therefore, two major steric repulsions between the H-atom of methyl bromide and H-atoms of the substrate are present for β-alkylation with the distances 2.08 and 2.26Å leading to exclusive α-attack of the electrophile. The transition state of α-alkylation with methyl bromide on enolate 197 was calculated to favour α-attack for ∆∆G = 4.6 kcal/mol (>99% d.e.).

In 2010, Carreira and Gampe reported their seminal work on the insertion of cyclohexynes into cyclic ketones leading to the formation of medium-sized, fused rings. They further applied this new methodology in the synthesis of natural products (Scheme 25). Specifically, the tricyclic core of guanacastepene was synthesised using in situ generated enolate 200 and cyclohexyne 201. When cyclobutanol 177,
acquired as a single diastereomer, was treated with $t$-BuOK, a 2:1 mixture of enones $203$ and $204$ was obtained. The authors proposed that the potassium alkoxide of $177$ undergoes torquoselective electrocyclic ring opening via intermediate $202$ to afford

Scheme 24. Sterically controlled alkylations of lithium enolates of hydroazulenones $193$ and $197$, as computed by Houk, Wicha and co-workers, and their corresponding free energies at 298K. Using B3LYP/6-31G* method and solvent correction employing the CPCM polarizable conductor calculation model as implemented in Gaussian 03 at 298K. For $198\alpha$, $198\beta$, methanol was specified as the solvent and for $202\alpha$, $202\beta$, THF. Distances given in Å. HMPA = hexamethylphosphoramide.

Scheme 25. Carreira and Gambé's insertion of cyclohexynes into cyclic ketones in the synthesis of the tricyclic core of guanacastepenes.
non-conjugated enone 203. The trans-cis isomerisation of strained trans-cycloheptadiene 202 and protonation furnished 203 or tautomerisation to C1 gave rise to diene 204. To avoid the formation of a mixture of products, Carreira and Gampe subjected cyclobutanol 177 to iron-promoted electrocyclic ring opening that occurred more selectively via the intermediacy of Fe(CO)₅-cyclobutene complex 205, which afforded enone 203. Further treatment of 203 by DBU exclusively gave the desired enone 178. In 2012, the mechanism of this annulative cascade reaction was a subject of the computational studies performed by Houk and Sader. B3LYP and M06−2X calculations of potential energy surface revealed a stepwise [2+2] cycloaddition of cyclohexyne 201 to the enolate 200 via intermediacy of vinyl anion 206 (Scheme 26). The first step of the reaction, nucleophilic attack of the enolate 200 on cyclohexyne 201 occurs without an activation barrier and is highly exothermic with a ΔH of −36 to −48 kcal/mol, while the second, the ring closure to cyclobutene alkoxide, has an activation energy of just 2−5 kcal/mol with exothermicity of 6−7 kcal/mol as calculated for closely related model systems.

### Scheme 26
Mechanistic pathways of the insertion of cyclohexynes 201 into cyclic ketones 200 as calculated by Houk and Sader using B3LYP/6-31G(d) method. Values enclosed in parentheses are thermal energies, ΔH, shown in kcal/mol.

Theoretically, the ring opening of potassium cyclobutene alkoxide 207 to the guanacastepene core 211 may occur by torquoselective electrocyclic reaction in thermally allowed conrotatory or thermally forbidden disrotatory mode. The third distinct mechanistic pathway is based on the non-electrocyclic cleavage of C2−C9 bond. The conrotatory reaction of potassium alkoxide 207 gives a trans-cycloheptadiene intermediate 202 that is 9 kcal/mol less stable than starting potassium alkoxide 207 with an activation barrier of 19 kcal/mol (transition structure 208). Further trans-cis isomerisation of trans-198 to cis-211 cycloheptadiene occurs with an activation energy of less than 1 kcal/mol. On the other hand, non-pericyclic mechanistic pathway via transition structure 210 directly produces cis-211 cycloheptadiene that is 22 kcal/mol more stable than the starting potassium alkoxide 207. The activation barrier of non-pericyclic mechanistic pathway is only 11 kcal/mol, leading to the conclusion that a more stable disrotatory product can be formed directly through this pathway. A truly thermally forbidden disrotatory process has a planar transition structure that was shown to be 19 kcal/mol higher in energy than the allowed conrotatory transition structure, as calculated for the model system.

### 7 Conclusions
Traditionally, natural products or natural product-like compounds have been one of the major resources for the generation of new drugs and their current demand for pharmaceutical purposes remains high. Natural products have frequently played an important role in deciphering processes of biological interest. Their synthesis have often led to the development of new methodologies with broad synthetic applications.

As presented by this review, the interdisciplinary research related to guanacastepenes has been constantly increasing. New compounds bearing neodolastane skeleton were isolated and characterised from fungal and marine resources. Their therapeutic properties still have to be investigated in detail. In this light, guanacastepenes have been an interesting target for many synthetic groups and they have opened a door for the development of new and innovative methodologies for the construction of C₅-C₇-C₆ tricyclic skeletons. By undertaking a survey towards their synthesis, fundamental stereochemical and mechanistic questions were answered. To conclude, guanacastepenes are a recent example of how natural products stimulate progress in research in different fields and in general, the acquirement of valuable experimental and theoretical knowledge.

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### 9 Notes and References
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This article is dedicated to Prof. Reinhard Neier in occasion of his birthday.