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Synthesis and biological evaluation of new tetrahydro-β-carbolines as inhibitors of the mitotic kinesin Eg5

Nils Sunder-Plassmann,[a] Vasiliki Sarli,[a] Michael Gartner,[a] Mathias Utz,[b] Jeanette Seiler,[b] Stefan Huemmer,[c] Thomas U. Mayer,[c] Thomas Surrey[b] and Athanassios Giannis[a,*]

[a]*University of Leipzig, Institute for Organic Chemistry, Johannisallee 29, 04103 Leipzig, Germany*
[b]*European Molecular Biology Laboratory, Cell Biology and Biophysics Programme, Meyerhofstrasse 1, 69117 Heidelberg, Germany*
[c]*Max Planck Institute for Biochemistry, Chemical Genetics, Independent Research Group, Am Klopferspitz 18, 82152 Martinsried/Munich, Germany*

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Abstract—The mitotic kinesin Eg5 (or KSP) is a crucial player in the development and function of the mitotic spindle. Inhibition of this protein leads to cell cycle arrest and apoptosis without interfering with other microtubule-dependent processes. Therefore, it is a potential target in cancer therapy. Here, we report the synthesis and biological evaluation of a small library of molecules based on the structure of the known Eg5 inhibitor HR22C16. One of these derivatives (compound *trans*-**24**) proved to be a potent and specific Eg5 inhibitor.

1. Introduction

In the industrialized countries, cancer is one of the leading causes of death, claiming more than 2.5 million victims every year. Among the various successful approaches to treat cancer are drugs that perturb mitosis like vinca alkaloids, taxol, and epothilone. One of their major drawbacks is the fact that they all target the same protein, tubulin, the subunit of microtubules that are essential for the formation of the mitotic spindle.[1,2] As microtubules are also involved in many other cellular processes such as maintenance of organelles, cell shape, cell motility, synaptic vesicles, and intracellular transport phenomena,[3–5] interference with their formation or depolymerization often leads to dose-limiting toxic side effects like neurotoxicity, hair loss, and body weight loss.

The discovery of a new class of proteins, the mitotic kinesins, allows a novel approach to cancer treatment.[6] These proteins are exclusively involved in the formation and function of the mitotic spindle and some of them are only expressed in proliferating cells.[7,8] Their inhibition leads to cell cycle arrest and ultimately to apoptosis without interfering with other microtubule-dependent processes.[6]

The mitotic kinesin Eg5 (or KSP), a member of the kinesin-5 family, plays an important role in the early stages of mitosis. It mediates centrosome separation and formation of the bipolar mitotic spindle.[9] It cross-links and slides microtubules in relation to one another using energy from ATP-hydrolysis.[10] Inhibition of Eg5 leads to cell cycle arrest during mitosis and cells with a monopolar spindle, so called monoasters.[11] In 1999, Monastrol, the first small-molecule inhibitor of Eg5, was discovered in a phenotype assay.[12] Since then, more KSP-inhibitors have been found.[13–15,22]

Subsequently, after screening a compound library of 16,000 small molecules, the carboline derivative HR22C16 was also identified as an Eg5 inhibitor (Scheme 1).[16] Recently, it was proven that HR22C16 induces mitotic arrest and cell death in taxol-resistant cancer cells.[17] This shows that Eg5 inhibitors can be potentially used to overcome taxol-resistance in cancer treatment. However, a systematic investigation of the importance of the stereochemistry and the substituents as indicated in Scheme 2 had not been performed.

Here we describe the synthesis of a library of 60 small molecules based on the HR22C16-structure, in order to address the above-mentioned problem.

Keywords: Carbolines; Kinesins; KSP; Pictet–Spengler reaction.
* Corresponding author. E-mail: giannis@chemie.uni-leipzig.de

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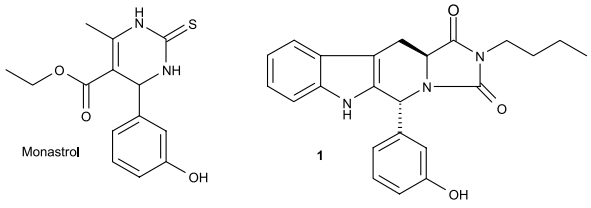
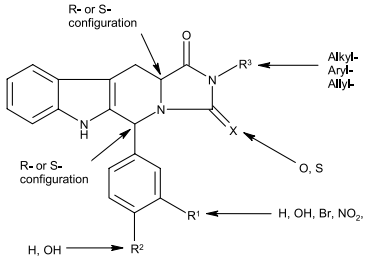


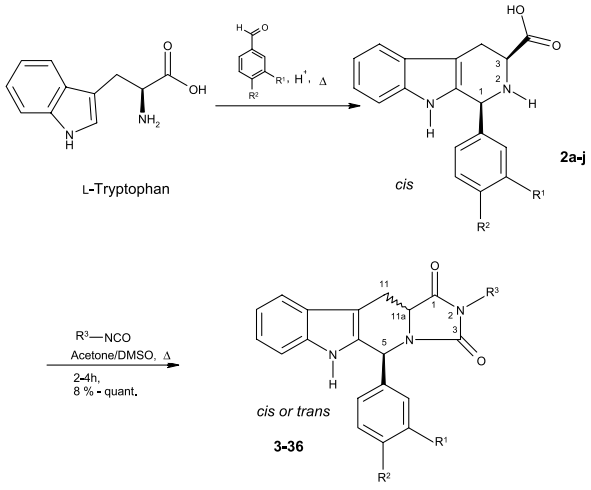
Scheme 1. The Eg5 inhibitors Monastrol and HR22C16 **1**.



Scheme 2. Basic scaffold of the tetrahydro-β-carboline library.

2. Results and discussion

Various ways to synthesize chiral tetrahydro-β-carbolines have been published in recent years. Interesting examples are the asymmetric Pictet–Spengler reaction[18] or the usage of a chiral ruthenium complex.[19] To obtain chiral 3-substituted tetrahydro-β-carbolines we decided to exploit the chiral pool, and used the cheap and commercially available amino acid tryptophan as a starting material.[20] Depending on the desired stereochemistry of the final product, we either used the D- or the L-form. For the initial step, we added two equivalents of an aldehyde to a suspension of tryptophan in diluted sulfuric acid (0.1 N). After refluxing for 4–5 h, the product **2** was filtered off. It was then transformed into the tetrahydro-β-carbolines **3**–**36** by the reaction with an isocyanate or an isothiocyanate (Scheme 3). In most cases, especially when R^1 = OH, R^2 = H and reaction times were short (<4 h), the resulting product was *cis*-configured. In some cases, however, a mixture of *cis*- and *trans*-product was formed which could easily be separated by column chromatography. In cases where there was no or little *trans*-product, the *cis*-form could be obtained by simply refluxing in acetonitrile with a small excess of potassium carbonate. The reactions with isothiocyanates exclusively yielded the *trans*-adduct.



Scheme 3. Synthesis of the tetrahydro-β-carbolines.

The yields of the first two steps varied from 22 to 84% and 8 to 100%, respectively, depending on the substituents R^1, R^2, and R^3. To synthesize compounds **31** and **32**, we first transformed L-leucine methylester into the corresponding isocyanate **37** (Scheme 4).[21] Schemes 5a and b show the complete library.

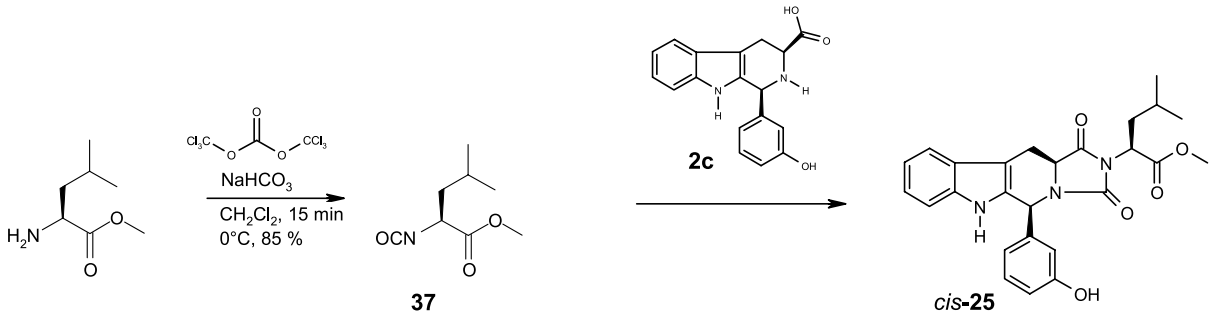
All synthesized tetrahydro-β-carbolines were tested for their activity against Eg5. To identify potential hits, we first screened the library with a pyruvate kinase/lactate dehydrogenase assay.[22] A list of the compounds with a relative inhibition of at least 50% at a concentration of 46 μM is shown in Figure 1. Monastrol, the first known Eg5 inhibitor, is shown for comparison.

Furthermore, we tested all compounds with the malachite green assay[23] for inhibition of nine different kinesins. Apart from Eg5, these were MKLP1, MKLP2, MPP1, KIF4, KIF5A, KIF5B, CenpE, and MCAK. All these kinesins were not or only weakly inhibited at the above-mentioned concentrations. Therefore, the synthesized tetrahydro-β-carbolines show specificity against Eg5.

We found that the potent inhibitors possess certain structural features. In most cases, the *trans*-isomers were more active than the *cis*-isomers, with the absolute stereochemistry at carbon C11a being *S*-configured. The activity was also very much dependent on the substituents R^1 and R^2. Nearly all substitution patterns other than R^1 = OH and R^2 = H lead to inactive molecules. Thus, the *meta*-hydroxy function seems to be important. However, a variety of substituents are possible for R^3. Linear alkyl chains with two to five carbon atoms (and R^1 = OH, R^2 = H) all lead to comparably active inhibitors. Benzyl-, phenyl- and *o*-trifluoromethoxyphenyl substituents also gave strong inhibitors when C11a was *S*-configured. The substitution of the oxygen at C3 with a sulfur atom did not result in an increase in activity.

We then chose nine inhibitors to determine their IC_{50} values. To obtain quantitative results concerning the effect of the stereochemical configuration we included one complete set of four diastereomers of HR22C16 (compounds **9** and **10**). Seven compounds were examined with the malachite green assay,[23] one with the EnzCheck® (Molecular Probes) Phosphate Assay and one with both tests.

The determined IC_{50} values are shown in Table 1. The most potent diastereomers of HR22C16 is compound *trans*-**10**. However *trans*-**9** and *cis*-**10** display significant inhibitory activity against Eg5 whereas derivative *cis*-**9** is almost inactive. Furthermore, derivative *trans*-**24** with



Scheme 4. Synthesis of the leucine derivatives with *cis*-**25** as example.

a *N*-benzyl side chain is the most active inhibitor with an IC_{50} value of 0.65 ± 0.30 μM. The other three stereoisomers of *trans*-**24** were inactive (IC_{50} > 45 μM). Finally, we examined the effect of *trans*-**24** on HeLa cells. We examined the morphology of the cells treated with *trans*-**24** by immunofluorescence with an anti-tubulin antibody (tubulin, green fluorescence) and Hoechst 33258 (DNA, blue fluorescence).[22] Figure 2 shows the normal spindle of an untreated HeLa cell and a cell treated with *trans*-**24**. As expected a monoastral phenotype is induced.

3. Conclusion

We synthesized a small library of 60 compounds based on the known Eg5 inhibitor HR22C16. We identified certain structural features that characterize the most potent inhibitors of this class of molecules. All compounds were screened for inhibition of Eg5 and eight other kinesins. Twenty two showed more than 50% inhibition of Eg5 at 46 μM, 15 were more active than the first Eg5 inhibitor Monastrol. The other kinesins were not significantly inhibited. For nine of the Eg5 inhibitors, the IC_{50} values were determined. In most cases, the *trans*-isomers were more active than the *cis*-isomers, with the absolute stereochemistry at carbon C11a being *S*-configured. The activity was also very much dependent on the substituents R^1 and R^2. The *meta*-hydroxyphenyl group seems to be important for activity.

Compound *trans*-**24** proved to be the most active inhibitor of Eg5-ATPase activity with an IC_{50} of 0.65 μM. This novel inhibitor has the potential for being a potent anti-cancer drug candidate.

4. Experimental

4.1. Chemistry

All reactions were performed with commercially available reagents and they were used without further purification. Solvents were dried by standard methods and stored over molecular sieves. All reactions were monitored by thin-layer chromatography (TLC) carried out on Merck silica gel 60 F_{254} aluminum sheets and viewed with UV light. Flash-chromatography was performed on Merck silica gel 60. Melting points were determined in open capillaries using a Büchi Melting Point B-540 apparatus and are uncorrected. ^1H and ^{13}C NMR spectra were recorded on Varian VXR-200, Varian VXR-300, and Bruker DRX-600 NMR spectrometers at room temperature. The solvents stated were used as internal standards. High-resolution (HR) mass spectra were obtained with a 7 T APEX II mass spectrometer. Yields are not optimized.

4.2. General procedure for the Pictet–Spengler reaction

To a suspension of L- or D-tryptophan (3 g, 14.3 mmol) in sulfuric acid (0.1 N, 44 ml), the desired aldehyde (2 equiv, 28.6 mmol) was added. After heating to reflux for 4–5 h, the solid phase was filtered off, thoroughly rinsed with diethylether, and dried in vacuo.

4.2.1. (1*S*,3*S*)-1-Phenyl-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2a). Yield: 80%, brown solid.

^1H NMR (300 MHz, DMSO-d_6): δ = 2.95 (m, 1H, CH_aH$_b$), 3.18 (m, 1H, CH$_a$$H_b$), 3.86 (m, 1H, C*H*COOH), 5.46 (s, 1H, C*H*Ph), 6.99–7.51 (m, 9H, ar), 10.50 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.20 (s), 57.21 (t), 57.70 (t), 108.04 (q), 111.30 (t), 117.94 (t), 118.73 (t), 121.22 (t), 126.29 (q), 128.43 (2× t), 129.13 (t), 129.60 (2× t), 132.55 (q), 136.69 (q), 138.61 (q), 172.03 (q).

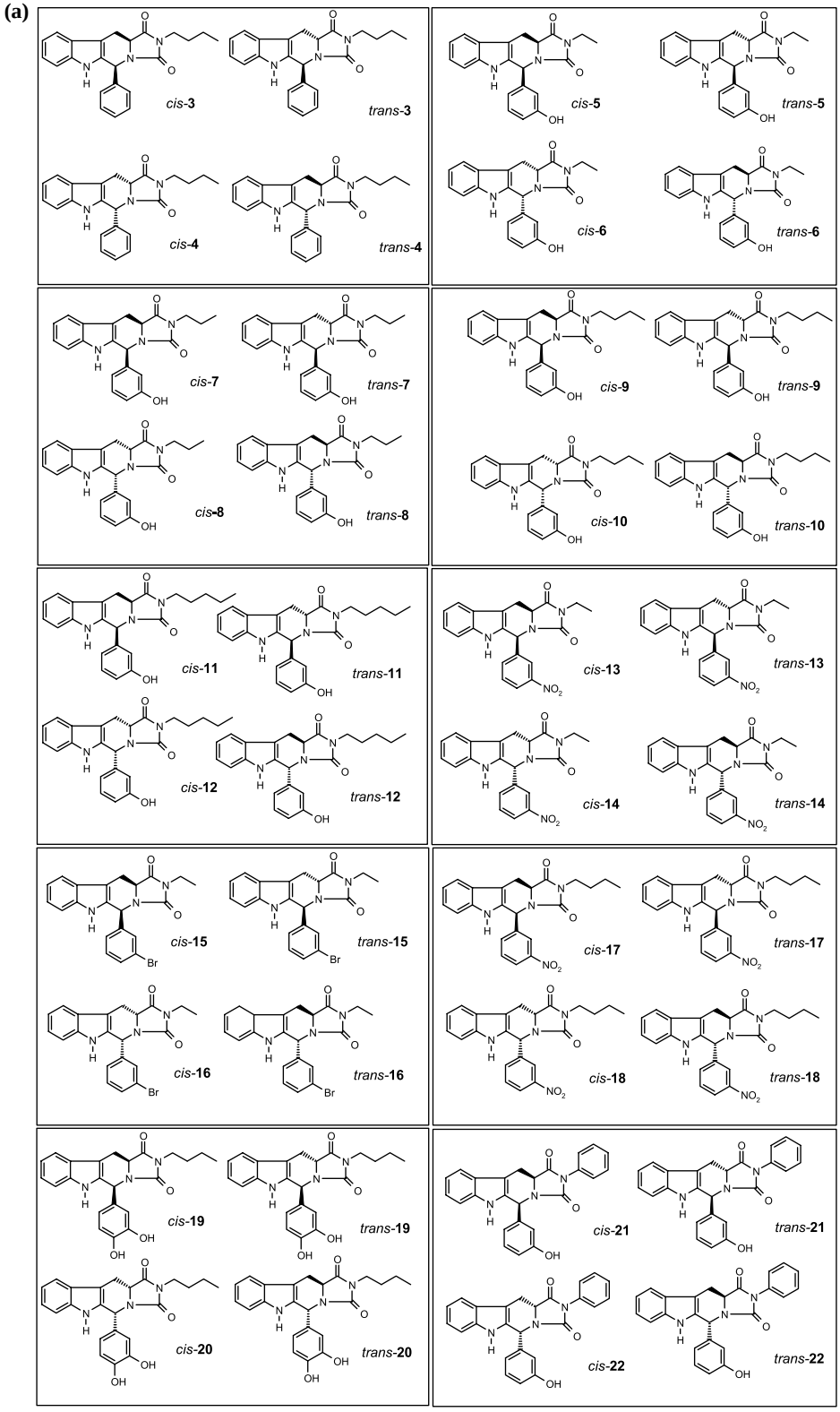
HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_2$: calcd: 293.1285; found: 293.1283.

4.2.2. (1*R*,3*R*)-1-Phenyl-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2b). Yield: 66%, brown solid.

^1H NMR (300 MHz, DMSO-d_6): δ = 2.95 (m, 1H, CH_aH$_b$), 3.18 (m, 1H, CH$_a$$H_b$), 3.86 (m, 1H, C*H*COOH), 5.46 (s, 1H, C*H*Ph), 6.99–7.51 (m, 9H, ar), 10.50 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.29 (s), 57.11 (t), 57.65 (t), 108.13 (q), 111.28 (t), 118.01 (t), 118.68 (t), 121.18 (t), 126.31 (q), 128.44 (2× t), 129.20 (t), 129.54 (2× t), 132.48 (q), 136.67 (q), 138.56 (q), 171.99 (q).

HRMS (ESI, pos.): $C_{18}H_{16}N_2O_2$: calcd: 293.1285; found: 293.1283.

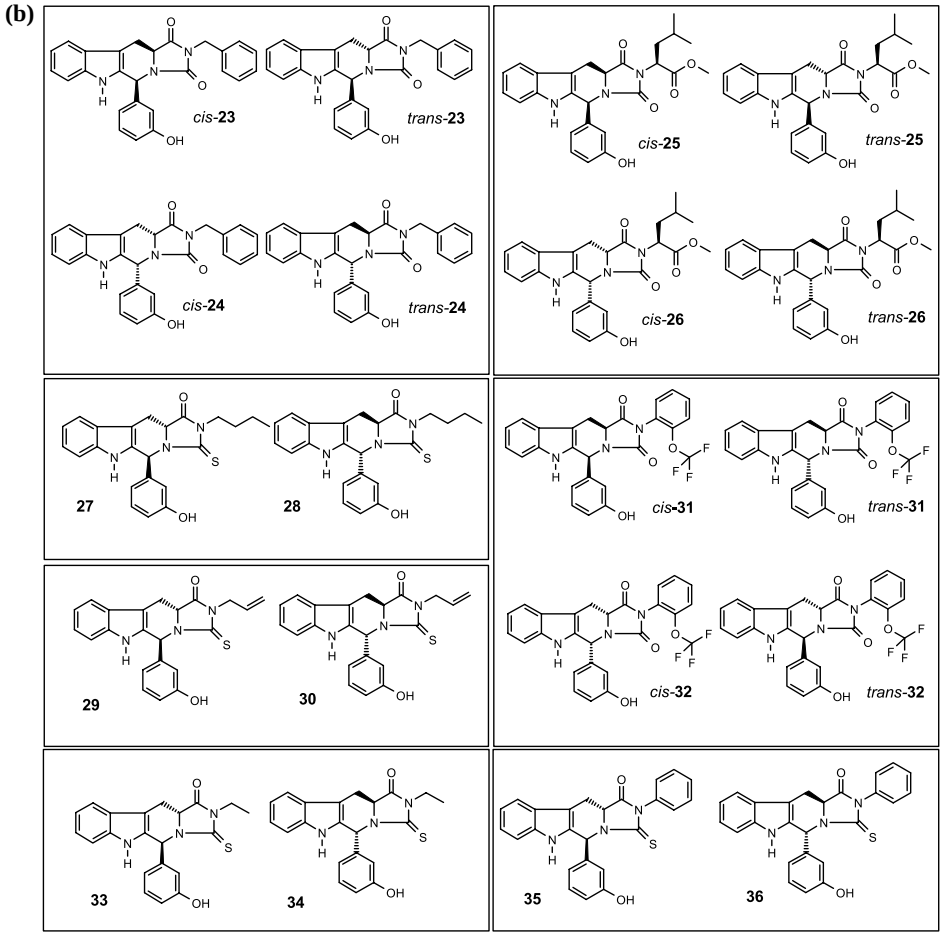


Scheme 5. (a): Tetrahydro-β-carboline library (1) (b): Tetrahydro-β-carboline library (2).

4.2.3. (1*S*,3*S*)-1-(3-Hydroxyphenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2c). Yield: 84%, brown solid.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.92 (m, 1H, CH_aH$_b$), 3.21 (m, 1H, CH$_a$$H_b$), 3.82 (m, 1H, C*H*COOH), 5.39 (s, 1H, C*H*Ph), 6.81–7.50 (m, 8H, ar), 10.51 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.03 (s), 57.37 (t), 57.66 (t), 108.02 (q), 111.51 (t), 115.90 (t), 116.51 (t), 117.98 (t), 118.77 (t), 120.16 (t), 121.28 (t), 126.26 (t),



Scheme 5. *(continued)*

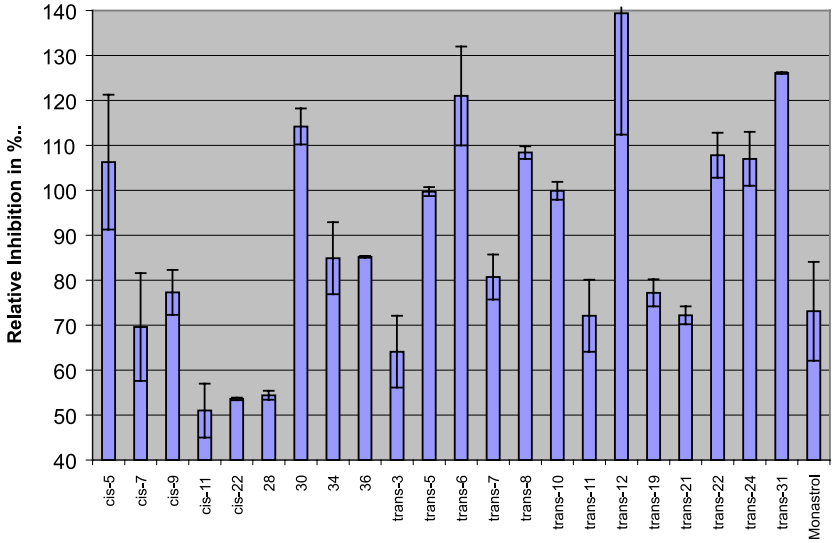


Figure 1. Eg5 inhibitors and their relative inhibition of Eg5-ATPase activity.

129.44 (q), 132.25 (q), 136.72 (q), 139.37 (q), 157.65 (q), 171.81 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_3$: calcd: 309.1234; found: 309.1232.

4.2.4. (1*R*,3*R*)-1-(3-Hydroxyphenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2d). Yield: 71%, brown solid.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 2.86 (m, 1H, CH_aH_b), 3.16 (m, 1H, CH_aH_b), 3.76 (m, 1H,

Table 1. IC_{50} values of selected Eg5 inhibitors

Compound	IC_{50} value (μM)
cis-**9**[a]	36 ± 9.9
trans-**9**[a]	14 ± 5.8
cis-**10**[a]	21 ± 16.4
trans-**10**[a] (HR22C16)	4.3 ± 1.18
trans-**24**[b]	0.65 ± 0.30
29[a]	18 ± 2.5
30	13 ± 0.6[a] 15 ± 1.1[b]
33[a]	11 ± 5.9
34[a]	17 ± 2.8

[a] EnzChek-Assay.
[b] Malachite green assay.

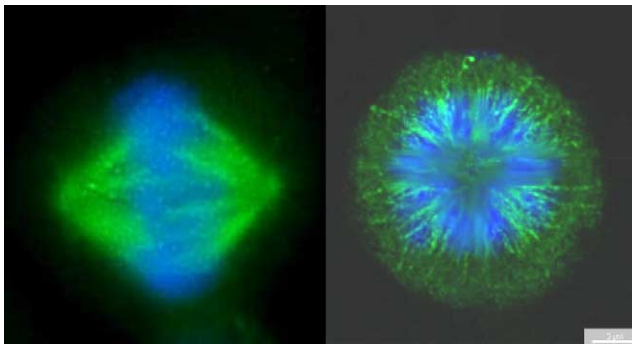


Figure 2. HeLa cells with normal spindle (left) and with monoasters after treatment with *trans*-**24** (20 μM) (right).

C*H*COOH), 5.33 (s, 1H, C*H*Ph), 6.76–7.46 (m, 8H, ar), 10.45 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.25 (s), 57.19 (t), 57.65 (t), 107.86 (q), 111.40 (t), 115.65 (t), 116.24 (t), 117.82 (t), 118.62 (t), 119.95 (t), 121.09 (t), 126.26 (q), 129.33 (t), 132.70 (q), 136.61 (q), 139.99 (q), 157.53 (q), 172.02 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_3$: calcd: 309.1234; found: 309.1230.

4.2.5. (1*S*,3*S*)-1-(3-Nitrophenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2e). Yield: 71%, auburn solid.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 2.95–3.91 (m, 2H, CH_aH_b), 4.13 (m, 1H, C*H*COOH), 5.55 (s, 1H, C*H*Ph), 7.00–8.34 (m, 8H, ar), 10.43 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 22.39 (s), 57.02 (t), 59.03 (t), 107.13 (q), 111.40 (t), 117.94 (t), 118.63 (t), 118.95 (t), 121.25 (t), 122.08 (t), 123.08 (t), 124.05 (t), 125.93 (q), 134.16 (q), 136.30 (q), 136.95 (q), 147.75 (q), 171.67 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_3$: calcd: 338.1135; found: 338.1140.

4.2.6. (1*R*,3*R*)-1-(3-Nitrophenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2f). Yield: 50%, auburn solid.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 2.92–3.89 (m, 2H, CH_aH_b), 4.07 (m, 1H, C*H*COOH), 5.50 (s, 1H, C*H*Ph), 7.00–8.20 (m, 8H, ar), 10.43 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.34 (s), 57.05 (t), 57.62 (t), 107.78 (q), 111.36 (t), 115.52 (t), 116.08 (t), 117.77 (t), 118.59 (t), 119.93 (t), 121.05 (t), 126.27 (q), 129.34 (t), 132.98 (q), 136.57 (q), 140.34 (q), 157.45 (q), 172.05 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_3$: calcd: 338.1135; found: 338.1142.

4.2.7. (1*S*,3*S*)-1-(3-Bromophenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2g). Yield: 12%, auburn solid.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.92–4.32 (m, 3H, CH_aH_b, C*H*COOH), 5.39 (s, 1H, C*H*Ph), 6.91–7.95 (m, 8H, ar), 10.41 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.34 (s), 59.94 (t), 60.62 (t), 105.68 (q), 112.13 (t), 112.63 (t), 118.74 (t), 119.52 (t), 119.84 (t), 120.03 (t), 122.08 (t), 127.52 (q), 129.42 (t), 132.79 (q), 136.11 (q), 138.17 (q), 146.85 (q), 192.80 (q).

HRMS (ESI, 70 eV): $C_{18}H_{15}BrN_2O_2$: calcd: 369.0244; found: 369.0232.

4.2.8. (1*R*,3*R*)-1-(3-Bromophenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2h). Yield: 22%, auburn solid.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.92–4.32 (m, 3H, CH_aH_b, C*H*COOH), 5.39 (s, 1H, C*H*Ph), 6.91–7.95 (m, 8H, ar), 10.41 (s, 1H, CN*H*C).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 24.26 (s), 60.45 (t), 61.32 (t), 105.97 (q), 111.23 (t), 112.63 (t), 118.11 (t), 118.33 (t), 118.44 (t), 119.89 (t), 122.37 (t), 127.73 (q), 129.22 (t), 132.83 (q), 136.33 (q), 138.12 (q), 146.95 (q), 191.91 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_3$: 369.0244; found: 369.0237.

4.2.9. (1*S*,3*S*)-1-(3,4-Dihydroxyphenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2i). Yield: 46%, brown solid.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 3.02 (m, 1H, CH_aH_b), 3.32 (m, 1H, CH_aH_b), 3.89 (m, 1H, C*H*COOH), 5.53 (s, 1H, C*H*Ph), 6.62–7.53 (m, 7H, ar), 10.53 (s, 1H, CN*H*C).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 23.94 (s), 55.00 (t), 58.08 (t), 107.82 (q), 112.21 (t), 116.34 (t), 117.90 (t), 118.11 (t), 118.81 (t), 119.51 (t), 121.70 (t), 126.73 (q), 131.88 (q), 137.24 (q), 137.42 (q), 145.96 (q), 146.10 (q), 171.71 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_4$: calcd: 323.1037 $[M-H]^-$; found: 323.1035.

4.2.10. (1*R*,3*R*)-1-(3,4-Dihydroxyphenyl)-2,3,4,9-tetrahydro-1*H*-β-carboline-3-carboxylic acid (2j). Yield: 39%, brown solid.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 3.02 (m, 1H, CH_aH$_b$), 3.27 (m, 1H, CH$_a$$H_b$), 3.84 (m, 1H, C*H*COOH), 5.55 (s, 1H, C*H*Ph), 6.62–7.53 (m, 7H, ar), 10.58 (s, 1H, CN*H*C).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 22.82 (s), 56.02 (t), 57.59 (t), 104.66 (q), 111.24 (t), 114.66 (t), 115.09 (t), 117.98 (t), 118.69 (t), 118.78 (t), 121.22 (t), 125.88 (q), 131.37 (q), 135.36 (q), 136.58 (q), 144.25 (q), 144.87 (q), 171.66 (q).

HRMS (ESI, 70 eV): $C_{18}H_{16}N_2O_4$: calcd: 323.1037 $[M-H]^-$; found: 323.1033.

4.3. General procedure for the synthesis of the tetrahydro-β-carbolines

To a solution of the desired tetrahydro-β-carboline-3-carboxylic acid **2** (12 mmol) in a mixture of acetone (40 ml) and DMSO (17 ml), the corresponding isocyanate or isothiocyanate (12 mmol, 1 equiv) was added and heated to reflux. After 2 h, the progress of the reaction was monitored by TLC and prolonged for another 2 h if necessary. Finally, the mixture is hydrolyzed with water and extracted three times with dichloromethane. The combined organic layers were washed with water, dried, and concentrated in vacuo.

In case of the isothiocyanates, the product is *trans*. When using isocyanates, the result is either exclusively *cis*-product or a mixture of *cis* and *trans*. In the latter case, purification by column chromatography (silica gel, hexane/ethyl acetate 1:2) is necessary.

In case the sole product is *cis*-configured, the *trans*-product may be obtained as follows.

To a solution of *cis*-product (10.8 mmol) in acetonitrile (150 ml), potassium carbonate (14 mmol, 1.3 equiv) was added and stirred under reflux for 2 h. When the reaction was finished (monitored by TLC), the reaction mixture was filtered, the filtrate was concentrated in vacuo.

The Rf-values for the *trans*-products are generally lower than for the *cis*-products.

4.3.1. (5*S*,11a*S*)-2-Butyl-5-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-3. Yield: 63%, brown solid.

T_M = 195 °C.

^{1}H NMR (600 MHz, DMSO-d_6): δ = 0.92 (t, J = 7.5 Hz, 3H, CH_3), 1.33 (m, 2H, CH_2CH$_3$), 1.60 (m, 2H, CH_2CH$_2$CH$_3$), 3.07 (m, 1H, CH_aH$_b$), 3.48 (m, 2H, NCH_2), 3.53 (m, 1H, CH$_a$$H_b$), 4.39 (dd, J_1 = 4.6 Hz, J_2 = 11.4 Hz, 1H, C*H*C(O)N), 5.80 (s, 1H, C*H*Ph), 7.17–7.59 (m, 9H, ar), 10.77 (s, 1H, N*H*).

^{13}C NMR (151 MHz, DMSO-d_6): δ = 13.72 (p), 20.12 (s), 22.69 (s), 30.23 (s), 38.63 (s), 56.89 (t), 58.06 (t), 107.13 (q), 111.35 (t), 118.61 (t), 120.30 (t), 122.88 (t), 126.38 (q), 127.90 (2× t), 128.75 (t), 128.99 (2× t), 133.68 (q), 136.94 (q), 138.89 (q), 154.93 (q), 171.81 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2$: calcd: 374.1863 $[M+H]^+$; found: 374.1865.

4.3.2. (5*S*,11a*R*)-2-Butyl-5-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-3. Yield: 30%, brown solid.

T_M = 111 °C.

^{1}H NMR (600 MHz, DMSO-d_6): δ = 0.87 (t, J = 7.5 Hz, 3H, CH_3), 1.25 (m, 2H, CH_2CH$_3$), 1.51 (m, 2H, CH_2CH$_2$CH$_3$), 2.81 (m, 1H, CH_aH$_b$), 3.39 (m, 1H, CH$_a$$H_b$), 3.41 (m, 2H, NC$H_2$), 4.59 (dd, J_1 = 5.6 Hz, J_2 = 11.1 Hz, 1H, C*H*C(O)N), 6.23 (s, 1H, C*H*Ph), 7.03–7.54 (m, 9H, ar), 10.87 (s, 1H, N*H*).

^{13}C NMR (151 MHz, DMSO-d_6): δ = 13.46 (p), 19.36 (s), 22.72 (s), 29.64 (s), 37.67 (s), 51.53 (t), 52.88 (t), 105.98 (q), 111.37 (t), 118.20 (t), 118.84 (t), 121.68 (t), 125.77 (q), 127.78 (2× t), 128.11 (t), 128.72 (2× t), 131.23 (q), 136.70 (q), 140.13 (q), 154.38 (q), 172.65 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2$: calcd: 374.1863 $[M+H]^+$; found: 374.1860.

4.3.3. (5*R*,11a*R*)-2-Butyl-5-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-4. Yield: 73%, brown solid.

Overall yield for both enantiomers: 93%.

T_M = 201 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.3 Hz, 3H, CH_3), 1.29 (m, 2H, CH_2CH$_3$), 1.50 (m, 2H, CH_2CH$_2$CH$_3$), 3.03 (m, 1H, CH_aH$_b$), 3.30–3.38 (m, 3H, NCH_2, CH$_a$$H_b$), 4.58 (dd, J_1 = 4.3 Hz, J_2 = 11.3 Hz, 1H, C*H*C(O)N), 5.89 (s, 1H, C*H*Ph), 6.97–7.59 (m, 9H, ar), 10.77 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 14.22 (p), 20.07 (s), 22.56 (s), 30.40 (s), 38.14 (s), 56.42 (t), 58.26 (t), 105.74 (q), 112.02 (t), 118.90 (t), 119.54 (t), 122.13 (t), 126.62 (q), 128.08 (2× t), 128.73 (t), 128.94 (2× t), 135.51 (q), 137.42 (q), 141.58 (q), 154.91 (q), 172.44 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2$: calcd: 396.1683 $[M+Na]^+$; found: 374.1685.

4.3.4. (5*S*,11a*R*)-2-Butyl-5-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-4. Yield: 30%, brown solid.

Overall yield for both enantiomers: 93%.

T_M = 95 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.5 Hz, 3H, CH_3), 1.24 (m, 2H, CH_2CH$_3$), 1.52 (m, 2H, CH_2CH$_2$CH$_3$), 2.83 (m, 1H, CH_aH$_b$), 3.37–3.47 (m,

3H, CH_aH_b, NCH_2), 4.62 (dd, J_1 = 5.8 Hz, J_2 = 11.0 Hz, 1H, C*H*C(O)N), 6.25 (s, 1H, C*H*Ph), 7.02–7.59 (m, 9H, ar), 10.93 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 14.21 (p), 20.11 (s), 23.47 (s), 30.39 (s), 38.41 (s), 52.27 (t), 53.61 (t), 106.73 (q), 112.12 (t), 118.95 (t), 119.58 (t), 122.41 (t), 126.51 (q), 128.54 (2× t), 128.85 (t), 129.46 (2× t), 131.96 (q), 137.44 (q), 140.89 (q), 155.11 (q), 173.38 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2$: calcd: 396.1683 $[M+Na]^+$; found: 396.1681.

4.3.5. (5*S*,11a*S*)-2-Ethyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-5. Yield: 84%, brown solid.

T_M = 142 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.11 (t, J = 7.2 Hz, 3H, CH_3), 3.01 (m, 1H, CH_aH_b), 3.36 (m, 1H, CH_aH_b), 3.40 (m, 2H, NCH_2), 4.52 (dd, J_1 = 4.5 Hz, J_2 = 11.7 Hz, 1H, C*H*C(O)N), 5.78 (s, 1H, C*H*Ph), 6.62–7.58 (m, 8H, ar), 9.32 (s, 1H, OH), 10.75 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.37 (p), 21.80 (s), 32.75 (s), 55.69 (t), 57.63 (t), 104.90 (q), 111.32 (t), 114.20 (t), 114.45 (t), 118.13 (t), 118.21 (t), 118.82 (t), 121.39 (t), 125.88 (q), 129.18 (t), 134.85 (q), 136.66 (q), 142.14 (q), 153.96 (q), 157.20 (q), 171.47 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_3$: calcd: 362.1499 $[M+H]^+$; found: 362.1500.

4.3.6. (5*S*,11a*R*)-2-Ethyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-5. Yield: 84%, brown solid.

T_M = 165 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.13 (t, J = 7.2 Hz, 3H, CH_3), 2.83 (m, 1H, CH_aH_b), 3.35 (m, 1H, CH_aH_b), 3.45 (m, 2H, NCH_2), 4.47 (dd, J_1 = 5.6 Hz, J_2 = 10.9 Hz, 1H, C*H*C(O)N), 6.10 (s, 1H, C*H*Ph), 6.62–7.56 (m, 8H, ar), 10.96 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.27 (p), 22.62 (s), 35.73 (s), 51.52 (t), 52.74 (t), 105.70 (q), 111.32 (t), 115.32 (t), 115.67 (t), 118.04 (t), 118.69 (t), 121.51 (t), 125.75 (t), 129.38 (t), 131.48 (q), 136.61 (q), 141.03 (q), 153.97 (q), 160.09 (q), 162.24 (q), 172.37 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_3$: calcd: 400.1058 $[M+K]^+$; found: 400.1063.

4.3.7. (5*R*,11a*R*)-2-Ethyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-6. Yield: 87%, pale yellow solid.

T_M = 234 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 1.06 (t, J = 7.2 Hz, 3H, CH_3), 2.95 (m, 1H, CH_aH_b), 3.28 (m, 1H, CH_aH_b), 3.39 (m, 2H, NCH_2), 4.49 (dd, J_1 = 4.0 Hz, J_2 = 11.2 Hz, 1H, C*H*C(O)N), 5.73 (s, 1H, C*H*Ph), 6.59–7.54 (m, 8H, ar), 9.27 (s, 1H, OH), 10.70 (br, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.34 (p), 21.81 (s), 32.73 (s), 55.69 (t), 57.61 (t), 104.89 (q), 111.29 (t), 114.17 (t), 114.42 (t), 118.07 (t), 118.18 (t), 118.78 (t), 121.35 (t), 125.85 (q), 129.11 (t), 134.82 (q), 136.64 (q), 142.07 (q), 153.91 (q), 157.15 (q), 171.40 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_3$: calcd: 362.1499 $[M+H]^+$; found: 362.1502.

4.3.8. (5*R*,11a*S*)-2-Ethyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-6. Yield: 90%, brown solid.

T_M = 170 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 1.13 (t, J = 7.2 Hz, 3H, CH_3), 2.83 (m, 1H, CH_aH_b), 3.34 (m, 1H, CH_aH_b), 3.45 (m, 2H, NCH_2), 4.47 (dd, J_1 = 5.9 Hz, J_2 = 10.9 Hz, 1H, C*H*C(O)N), 6.10 (s, 1H, C*H*Ph), 6.58–7.56 (m, 8H, ar), 10.96 (br, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 14.00 (p), 23.33 (s), 33.68 (s), 52.29 (t), 53.47 (t), 106.37 (q), 112.07 (t), 116.22 (t), 116.57 (t), 116.68 (t), 118.77 (t), 119.43 (t), 122.22 (t), 126.48 (q), 130.05 (t), 132.31 (q), 137.34 (q), 141.71 (q), 154.70 (q), 161.49 (q), 173.16 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_3$: calcd: 400.1058 $[M+K]^+$; found: 400.1060.

4.3.9. (5*S*,11a*S*)-5-(3-Hydroxyphenyl)-2-propyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-7. Yield: 89%, yellow solid.

T_M = 92 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.81 (t, J = 7.8 Hz, 3H, CH_3), 1,49 (m, 2H, CH_2CH$_3$), 2.89 (m, 1H, CH_aH_b), 3.27 (m, 1H, CH_aH_b), 3.30 (m, 2H, NCH_2), 4.51 (dd, J_1 = 4.4 Hz, J_2 = 11.6 Hz, 1H, C*H*C(O)N), 5.74 (s, 1H, C*H*Ph), 6.59–7.54 (m, 8H, ar), 9.27 (s, 1H, OH), 10.72 (s, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 11.12 (p), 21.04 (s), 23.30 (s), 39.38 (s), 55.69 (t), 57.54 (t), 104.87 (q), 111.35 (t), 114.17 (t), 114.45 (t), 118.15 (2× t), 118.85 (t), 121.42 (t), 125.91 (q), 129.20 (t), 134.90 (q), 136.68 (q), 142.20 (q), 154.16 (q), 157.23 (q), 171.80 (q).

HRMS (ESI): $C_{22}H_{21}N_3O_3$: calcd: 398.1475 $[M+Na]^+$; found: 398.1478.

4.3.10. (5*S*,11a*R*)-5-(3-Hydroxyphenyl)-2-propyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-7. Yield: 87%, brown solid.

T_M = 120 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.85 (t, J = 8.2 Hz, 3H, CH_3), 1,55 (m, 2H, CH_2CH$_3$), 2.90 (m, 1H, CH$_a$H$_b$), 3.27 (m, 1H, CH$_a$H$_b$), 3.43 (m, 2H, NCH_2), 4.51 (dd, J_1 = 5.6 Hz, J_2 = 10.9 Hz, 1H, CHC(O)N), 6.16 (s, 1H, CHPh), 6.73–7.59 (m, 8H, ar), 9.50 (s, 1H, OH), 10.97 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 11.05 (p), 20.96 (s), 22.78 (s), 51.34 (s), 52.70 (t), 54.86 (t), 105.96 (q), 111.35 (t), 114.71 (t), 115.05 (t), 118.13 (t), 118.38 (t), 118.78 (t), 121.62 (t), 125.75 (q), 129.71 (t), 131.16 (q), 136.64 (q), 141.34 (q), 154.25 (q), 157.58 (q), 172.59 (q).

HRMS (ESI): $C_{22}H_{21}N_3O_3$: calcd: 376.1656 $[M+H]^+$; found: 376.1660.

4.3.11. (5*R*,11a*R*)-5-(3-Hydroxyphenyl)-2-propyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-8. Yield: 85%, yellow solid.

T_M = 110 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.81 (t, J = 7.8 Hz, 3H, CH_3), 1,50 (m, 2H, CH_2CH$_3$), 2.92 (m, 1H, CH$_a$H$_b$), 3.30 (m, 1H, CH$_a$H$_b$), 3.33 (m, 2H, NCH_2), 4.51 (dd, J_1 = 4.4 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.74 (s, 1H, CHPh), 6.58–7.54 (m, 8H, ar), 9.27 (s, 1H, OH), 10.71 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 11.12 (p), 21.04 (s), 23.30 (s), 38.95 (s), 55.68 (t), 57.53 (t), 104.86 (q), 111.34 (t), 114.16 (t), 114.45 (t), 118.15 (2× t), 118.84 (t), 121.42 (t), 125.90 (q), 129.20 (t), 134.89 (q), 136.67 (q), 142.21 (q), 154.15 (q), 157.23 (q), 171.80 (q).

HRMS (ESI): $C_{22}H_{21}N_3O_3$: calcd: 398.1475 $[M+Na]^+$; found: 398.1479.

4.3.12. (5*R*,11a*S*)-5-(3-Hydroxyphenyl)-2-propyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-8. Yield: 78%, brown solid.

T_M = 103 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.85 (t, J = 8.2 Hz, 3H, CH_3), 1.59 (m, 2H, CH_2CH$_3$), 2.89 (m, 1H, CH$_a$H$_b$), 3.30–3.45 (m, 3H, CH$_a$H$_b$, NCH_2), 4.51 (dd, J_1 = 5.5 Hz, J_2 = 10.6 Hz, 1H, CHC(O)N), 6.15 (s, 1H, CHPh), 6.70–7.58 (m, 8H, ar), 10.96 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 11.75 (p), 21.66 (s), 23.94 (s), 52.10 (s), 53.42 (t), 106.60 (q), 112.08 (t), 115.61 (t), 115.96 (t), 118.53 (t), 118.84 (t), 119.50 (t), 122.32 (t), 126.47 (q), 130.36 (t), 131.99 (q), 137.35 (q), 141.99 (q), 154.97 (q), 159.04 (q), 173.37 (q).

HRMS (ESI): $C_{22}H_{21}N_3O_3$: calcd: 376.1656 $[M+H]^+$; found: 376.1649.

4.3.13. (5*S*,11a*S*)-2-Butyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-9. Yield: 86%, brown solid.

T_M = 225 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.5 Hz, 3H, CH_3), 1.28 (m, 2H, CH_2CH$_3$), 1.52 (m, 2H, CH_2CH$_2$CH$_3$), 3.00 (m, 1H, CH$_a$H$_b$), 3.39 (m, 1H, CH$_a$H$_b$), 3.41 (m, 2H, NCH_2), 4.58 (dd, J_1 = 4.5 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.76 (s, 1H, CHPh), 6.70–7.58 (m, 8H, ar), 9.33 (s, 1H, OH), 10.87 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.46 (p), 19.36 (s), 22.78 (s), 29.64 (s), 37.49 (s), 56.42 (t), 58.06 (t), 107.98 (q), 111.37 (t), 115.26 (t), 115.35 (t), 118.20 (t), 118.84 (t), 118.86 (t), 121.72 (t), 125.54 (q), 129.54 (t), 131.11 (q), 136.68 (q), 139.98 (q), 154.08 (q), 157.65 (q), 172.62 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_3$: calcd: 412.1632 $[M+Na]^+$; found: 412.1635.

4.3.14. (5*S*,11a*R*)-2-Butyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-9. Yield: 84%, brown solid.

T_M = 118 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.84 (t, J = 6.8 Hz, 3H, CH_3), 1.25 (m, 2H, CH_2CH$_3$), 1.49 (m, 2H, CH_2CH$_2$CH$_3$), 2.75 (m, 1H, CH$_a$H$_b$), 3.30 (m, 1H, CH$_a$H$_b$), 3.42 (m, 2H, NCH_2), 4.41 (dd, J_1 = 5.7 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.04 (s, 1H, CHPh), 6.50–7.51 (m, 8H, ar), 9.33 (s, 1H, OH), 10.91 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.46 (p), 19.36 (s), 22.78 (s), 29.64 (s), 37.49 (s), 51.42 (t), 52.06 (t), 105.98 (q), 111.37 (t), 115.26 (t), 115.35 (t), 118.20 (t), 118.84 (t), 118.86 (t), 121.72 (t), 125.54 (q), 129.54 (t), 131.11 (q), 136.68 (q), 139.98 (q), 154.08 (q), 157.65 (q), 172.62 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_3$: calcd: 412.1632 $[M+Na]^+$; found: 412.1628.

4.3.15. (5*R*,11a*R*)-2-Butyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-10. Yield: 88%, yellow solid.

T_M = 134 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.90 (t, J = 7.5 Hz, 3H, CH_3), 1.30 (m, 2H, CH_2CH$_3$), 1.52 (m, 2H, CH_2CH$_2$CH$_3$), 2.99 (m, 1H, CH$_a$H$_b$), 3.39 (m, 1H, CH$_a$H$_b$), 3.41 (t, J = 6.9 Hz, 2H, NCH_2), 4.53 (dd, J_1 = 4.2 Hz, J_2 = 10.6 Hz, 1H, CHC(O)N), 5.79 (s, 1H, CHPh), 6.64–7.59 (m, 8H, ar), 9.33 (s, 1H, OH), 10.77 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.50 (p), 19.35 (s), 21.89 (s), 29.70 (s), 32.22 (t), 37.42 (s), 55.67 (t), 57.52 (t), 104.85 (q), 111.33 (t), 114.17 (t), 114.43 (t), 118.14 (t), 118.82 (t), 121.39 (t), 125.88 (q), 129.16 (t), 134.87 (q), 136.65 (q), 142.17 (q), 154.11 (q), 157.22 (q), 171.73 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_3$: calcd: 412.1632 $[M+Na]^+$; found: 412.1629.

4.3.16. (5*R*,11a*S*)-2-Butyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-10. Yield: quant., brown solid.

T_M = 107 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.9 Hz, 3H, CH_3), 1.22 (m, 2H, CH_2CH$_3$), 1.51 (m, 2H, CH_2CH$_2$CH$_3$), 2.83 (m, 1H, CH_aH$_b$), 3.30 (m, 1H, CH$_a$$H_b$), 3.42 (m, 2H, NC$H_2$), 4.41 (dd, J_1 = 5.8 Hz, J_2 = 10.9 Hz, 1H, CHC(O)N), 6.04 (s, 1H, CHPh), 6.50–7.51 (m, 8H, ar), 9.33 (s, 1H, OH), 10.91 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 14.21 (p), 20.13 (s), 23.51 (s), 32.94 (s), 38.42 (s), 52.13 (t), 53.45 (t), 106.66 (q), 112.12 (t), 115.62 (t), 115.96 (t), 118.74 (t), 118.89 (t), 119.53 (t), 122.36 (t), 126.51 (q), 130.41 (t), 132.01 (q), 137.40 (q), 142.06 (q), 154.99 (q), 158.92 (q), 173.35 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_3$: calcd: 412.1632 $[M+Na]^+$; found: 412.1632.

4.3.17. (5*S*,11a*S*)-5-(3-Hydroxyphenyl)-2-pentyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-11. Yield: 87%, pale yellow solid.

T_M = 201 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.87 (m, 3H, CH_3), 1.22–1.38 (m, 4H, CH_2CH_2CH$_3$), 1.53 (m, 2H, CH_2), 2.99 (m, 1H, CH_aH$_b$), 3.35 (m, 3H, NCH_2, CH$_a$$H_b$), 4.55 (dd, J_1 = 4.5 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.79 (s, 1H, CHPh), 6.64–7.58 (m, 8H, ar), 9.32 (s, 1H, OH), 10.77 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.83 (p), 21.66 (s), 21.93 (s), 27.24 (s), 28.27 (s), 37.49 (s), 55.65 (t), 57.50 (t), 104.82 (q), 111.32 (t), 114.17 (t), 114.42 (t), 118.11 (2× t), 118.81 (t), 121.38 (t), 125.38 (q), 129.14 (t), 134.87 (q), 136.66 (q), 142.18 (q), 154.09 (q), 157.22 (q), 171.71 (q).

HRMS (ESI): $C_{24}H_{25}N_3O_3$: calcd: 404.1969 $[M+H]^+$; found: 404.1973.

4.3.18. (5*S*,11a*R*)-5-(3-Hydroxyphenyl)-2-pentyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-11. Yield: 93%, brown solid.

T_M = 83 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.81 (m, 3H, CH_3), 1.22–1.36 (m, 4H, CH_2CH_2CH$_3$), 1.51 (m, 2H, CH_2), 2.78 (m, 1H, CH_aH$_b$), 2.95 (m, 1H, CH$_a$$H_b$), 3.39 (m, 2H, NC$H_2$), 4.46 (dd, J_1 = 5.8 Hz, J_2 = 11.0 Hz, 1H, CHC(O)N), 6.13 (s, 1H, CHPh), 6.70–7.54 (m, 8H, ar), 9.46 (s, 1H, OH), 10.93 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.79 (p), 21.64 (s), 21.92 (s), 22.78 (s), 37.95 (s), 51.36 (t), 52.71 (t), 106.00 (q), 111.40 (t), 114.75 (t), 115.08 (t), 118.18 (t), 118.42 (t), 118.82 (t), 121.66 (t), 125.78 (q), 129.75 (t), 131.20 (q), 136.68 (q), 141.38 (q), 154.26 (q), 157.63 (q), 172.58 (q).

HRMS (ESI): $C_{24}H_{25}N_3O_3$: calcd: 404.1969 $[M+H]^+$; found: 404.1972.

4.3.19. (5*R*,11a*R*)-5-(3-Hydroxyphenyl)-2-pentyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-12. Yield: 83%, pale yellow solid.

T_M = 200 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.85 (m, 3H, CH_3), 1.23–1.37 (m, 4H, CH_2CH_2CH$_3$), 1.53 (m, 2H, CH_2), 2.99 (m, 1H, CH_aH$_b$), 3.35 (m, 3H, NCH_2, CH$_a$$H_b$), 4.55 (dd, J_1 = 4.4 Hz, J_2 = 11.6 Hz, 1H, CHC(O)N), 5.78 (s, 1H, CHPh), 6.63–7.59 (m, 8H, ar), 9.31 (s, 1H, OH), 10.76 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.86 (p), 21.68 (s), 21.96 (s), 27.26 (s), 28.28 (s), 37.68 (s), 55.66 (t), 57.52 (t), 104.83 (q), 111.34 (t), 114.17 (t), 114.43 (t), 118.15 (2× t), 118.83 (t), 121.40 (t), 125.89 (q), 129.17 (t), 134.88 (q), 136.66 (q), 142.20 (q), 154.11 (q), 157.23 (q), 171.73 (q).

HRMS (ESI): $C_{24}H_{25}N_3O_3$: calcd: 426.1788 $[M+Na]^+$; found: 426.1784.

4.3.20. (5*R*,11a*S*)-5-(3-Hydroxyphenyl)-2-pentyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-12. Yield: 93%, pale yellow solid.

T_M = 81 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.86 (m, 3H, CH_3), 1.22–1.40 (m, 4H, CH_2CH_2CH$_3$), 1.58 (m, 2H, CH_2), 2.83 (m, 1H, CH_aH$_b$), 3.40 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.51 (dd, J_1 = 5.7 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.17 (s, 1H, CHPh), 6.74–7.58 (m, 8H, ar), 10.98 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.80 (p), 21.67 (s), 22.80 (s), 27.25 (s), 28.31 (s), 37.97 (s), 51.40 (t), 52.74 (t), 105.99 (q), 111.41 (t), 114.84 (t), 115.17 (t), 118.19 (t), 118.27 (t), 118.83 (t), 121.67 (t), 125.80 (q), 129.75 (t), 131.25 (q), 136.70 (q), 141.38 (q), 154.27 (q), 157.89 (q), 172.61 (q).

HRMS (ESI): $C_{24}H_{25}N_3O_3$: calcd: 426.1788 $[M+Na]^+$; found: 426.1784.

4.3.21. (5*R*,11a*S*)-2-Ethyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-13. Yield: 21%, yellow solid.

T_M = 91 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.10 (t, J = 7.2 Hz, 3H, CH_3), 3.17 (m, 1H, CH_aH$_b$), 3.45 (m, 1H, CH$_a$$H_b$),

3.49 (m, 2H, NCH_2), 4.60 (dd, J_1 = 5.2 Hz, J_2 = 11.0 Hz, 1H, CHC(O)N), 6.13 (s, 1H, CHPh), 7.03–8.12 (m, 8H, ar), 10.86 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.31 (p), 21.59 (s), 32.83 (s), 54.87 (t), 57.47 (t), 105.76 (q), 111.37 (t), 118.42 (t), 119.00 (t), 121.74 (t), 122.60 (t), 122.73 (t), 125.82 (q), 129.93 (t), 133.58 (t), 134.17 (q), 136.83 (q), 142.86 (q), 147.62 (q), 154.35 (q), 171.43 (q).

HRMS (ESI): $C_{21}H_{18}N_4O_4$: calcd: 391.1401 $[M+H]^+$; found: 391.1399.

4.3.22. (5*S*,11a*R*)-2-Ethyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-13. Yield: 21%, dark yellow solid.

T_M = 109 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.13 (t, J = 7.3 Hz, 3H, CH_3), 2.86 (m, 1H, CH_aH$_b$), 3.35 (m, 1H, CH$_a$$H_b$), 3.48 (m, 2H, NC$H_2$), 4.47 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.44 (s, 1H, CHPh), 7.04–7.96 (m, 8H, ar), 10.94 (s, 1H, NH).

^{13}C NMR (600 MHz, DMSO-d_6): δ = 13.18 (p), 22.36 (s), 33.00 (s), 50.87 (t), 53.14 (t), 106.47 (q), 111.38 (t), 118.31 (t), 118.93 (t), 121.86 (t), 122.63 (t), 123.08 (t), 125.66 (q), 130.22 (t), 130.34 (t), 134.44 (q), 136.73 (q), 142.05 (q), 147.85 (q), 154.42 (q), 172.27 (q).

HRMS (ESI): $C_{21}H_{18}N_4O_4$: calcd: 391.1401 $[M+H]^+$; found: 391.1404.

4.3.23. (5*R*,11a*R*)-2-Ethyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-14. Yield: 21%, yellow solid.

T_M = 92 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.11 (t, J = 7.3 Hz, 3H, CH_3), 3.12 (m, 1H, CH_aH$_b$), 3.38 (m, 1H, CH$_a$$H_b$), 3.43 (m, 2H, NC$H_2$), 4.60 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.12 (s, 1H, CHPh), 7.03–8.34 (m, 8H, ar), 10.84 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.26 (p), 21.62 (s), 32.81 (s), 54.89 (t), 57.45 (t), 105.74 (q), 111.33 (t), 114.58 (t), 118.35 (t), 118.95 (t), 121.70 (t), 122.53 (t), 122.70 (t), 125.79 (t), 129.84 (t), 130.87 (q), 133.53 (q), 136.82 (q), 142.81 (q), 147.60 (q), 154.30 (q), 171.34 (q).

HRMS (ESI): $C_{21}H_{18}N_4O_4$: calcd: 413.1220 $[M+Na]^+$; found: 413.1218.

4.3.24. (5*R*,11a*S*)-2-Ethyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-14. Yield: 21%, yellow solid.

T_M = 86 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.14 (t, J = 7.3 Hz, 3H, CH_3), 2.86 (m, 1H, CH_aH$_b$), 3.35 (m, 1H, CH$_a$$H_b$), 3.48 (m, 2H, NC$H_2$), 4.70 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.45 (s, 1H, CHPh), 7.07–8.26 (m, 8H, ar), 10.93 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.25 (p), 22.42 (s), 33.08 (s), 50.95 (t), 53.23 (t), 106.55 (q), 111.46 (t), 118.37 (t), 118.99 (t), 121.92 (t), 122.73 (t), 123.14 (t), 125.73 (t), 130.29 (t), 130.40 (q), 134.52 (q), 136.81 (q), 142.14 (q), 147.92 (q), 154.50 (q), 172.35 (q).

HRMS (ESI): $C_{21}H_{18}N_4O_4$: calcd: 413.1220 $[M+Na]^+$; found: 413.1219.

4.3.25. (5*S*,11a*S*)-5-(3-Bromophenyl)-2-ethyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-15. Yield: 22%, brown solid.

T_M = 137 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.10 (t, J = 7.3 Hz, 3H, CH_3), 3.05 (m, 1H, CH_aH$_b$), 3.30–4.51 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.55 (m, 1H, C$H$C(O)N), 5.90 (s, 1H, C$H$Ph), 6.90–7.61 (m, 8H, ar), 10.81 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.46 (p), 22.59 (s), 37.49 (s), 56.42 (t), 58.06 (t), 107.13 (q), 111.27 (t), 115.36 (t), 115.35 (t), 118.17 (t), 118.84 (t), 118.68 (t), 121.72 (t), 125.54 (q), 129.54 (t), 131.11 (q), 136.68 (q), 141.88 (q), 154.08 (q), 154.57 (q), 172.62 (q).

HRMS (ESI): $C_{21}H_{18}BrN_3O_2$: calcd: 424.0655 $[M+H]^+$; found: 424.0663.

4.3.26. (5*S*,11a*R*)-5-(3-Bromophenyl)-2-ethyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-15. Yield: 35%, brown solid.

T_M = 116 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.13 (t, J = 7.3 Hz, 3H, CH_3), 2.81 (m, 1H, CH_aH$_b$), 3.24 (m, 1H, CH$_a$$H_b$), 3.50 (m, 2H, NC$H_2$), 4.67 (dd, J_1 = 4.4 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 6.24 (s, 1H, CHPh), 7.01–7.96 (m, 8H, ar), 10.91 (s, 1H, NH).

^{13}C NMR (600 MHz, DMSO-d_6): δ = 13.46 (p), 22.59 (s), 37.49 (s), 51.42 (t), 52.06 (t), 105.98 (q), 111.27 (t), 115.36 (t), 115.35 (t), 118.17 (t), 118.84 (t), 118.68 (t), 121.72 (t), 125.54 (q), 129.54 (t), 131.11 (q), 136.68 (q), 139.98 (q), 154.08 (q), 157.47 (q), 172.62 (q).

HRMS (ESI): $C_{21}H_{18}BrN_3O_2$ H^+: calcd: 424.0655 $[M+H]^+$; found: 424.0663.

4.3.27. (5*R*,11a*R*)-5-(3-Bromophenyl)-2-ethyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-16. Yield: 8%, brown solid.

T_M = 120 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.11 (t, J = 7.3 Hz, 3H, CH_3), 3.05 (m, 1H, CH_aH$_b$), 3.30–3.44 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.57 (dd, J_1 = 4.0 Hz, J_2 = 10.8 Hz,

1H, C*H*C(O)N), 5.91 (s, 1H, C*H*Ph), 6.97–7.62 (m, 8H, ar), 10.81 (s, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.30 (p), 21.60 (s), 32.78 (s), 55.04 (t), 57.54 (t), 105.45 (q), 111.32 (t), 118.26 (t), 118.88 (t), 121.34 (t), 121.56 (t), 125.80 (t), 126.39 (t), 130.30 (q), 130.35 (t), 130.43 (q), 133.97 (q), 136.73 (q), 143.28 (q), 154.12 (q), 171.36 (q).

HRMS (ESI): $C_{21}H_{18}BrN_3O_2$: calcd: 424.0655 $[M+H]^+$; found: 424.0661.

4.3.28. (5*R*,11a*S*)-5-(3-Bromophenyl)-2-ethyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-16. Yield: 95%, brown solid.

T_M = 225 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 1.08 (t, J = 7.0 Hz, 3H, CH_3), 2.79 (m, 1H, CH_aH$_b$), 3.38–3.48 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.63 (dd, J_1 = 5.6 Hz, J_2 = 10.6 Hz, 1H, C*H*C(O)N), 6.20 (s, 1H, C*H*Ph), 6.97–7.55 (m, 8H, ar), 10.87 (br, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 14.00 (p), 23.19 (s), 33.77 (s), 51.80 (t), 53.82 (t), 107.01 (q), 112.19 (t), 119.03 (t), 119.65 (t), 122.55 (t), 122.71 (t), 126.47 (t), 127.71 (t), 131.32 (q), 131.38 (t), 131.69 (q), 131.81 (q), 137.50 (q), 143.44 (q), 155.11 (q), 173.15 (q).

HRMS (ESI): $C_{21}H_{18}BrN_3O_2$: calcd: 446.0475 $[M+Na]^+$; found: 446.0480.

4.3.29. (5*S*,11a*S*)-2-Butyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-17. Yield: 23%, dark yellow solid.

T_M = 97 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.83 (t, J = 7 Hz, 3H, CH_3), 1.24 (m, 2H, CH_2CH$_3$), 1.49 (m, 2H, CH_2CH$_2$CH$_3$), 3.09 (m, 1H, CH_aH$_b$), 3.32 (m, 1H, CH$_a$$H_b$), 3.41 (t, J = 6.9 Hz, 2H, NCH_2), 4.56 (dd, J_1 = 4.4 Hz, J_2 = 11.4 Hz, 1H, C*H*C(O)N), 6.08 (s, 1H, C*H*Ph), 6.98–8.13 (m, 8H, ar), 10.82 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.46 (p), 19.30 (s), 21.68 (s), 29.60 (s), 37.48 (s), 54.82 (t), 57.33 (t), 105.67 (q), 111.36 (t), 118.41 (t), 118.97 (t), 121.72 (t), 122.56 (t), 122.64 (t), 125.80 (t), 129.90 (t), 133.56 (q), 134.10 (q), 136.81 (q), 142.87 (q), 147.59 (q), 154.48 (q), 171.66 (q).

HRMS (ESI): $C_{23}H_{22}N_4O_4$: calcd: 419.4602 $[M+H]^+$; found: 419.4598.

4.3.30. (5*S*,11a*R*)-2-Butyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-17. Yield: 13%, yellow solid.

T_M = 51 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.88 (t, J = 7 Hz, 3H, CH_3), 1.28 (m, 2H, CH_2CH$_3$), 1.54 (m, 2H, CH_2CH$_2$CH$_3$), 2.85 (m, 1H, CH_aH$_b$), 3.40–3.46 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.73 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, C*H*C(O)N), 6.45 (s, 1H, C*H*Ph), 7.04–8.25 (m, 8H, ar), 10.95 (s, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.42 (p), 19.31 (s), 22.53 (s), 29.57 (s), 37.69 (s), 50.89 (t), 53.15 (t), 106.50 (q), 111.43 (t), 118.37 (t), 118.97 (t), 121.91 (t), 122.66 (t), 123.12 (t), 125.70 (t), 130.28 (t), 134.44 (q), 134.85 (q), 136.77 (q), 142.10 (q), 147.89 (q), 154.65 (q), 172.57 (q).

HRMS (ESI): $C_{23}H_{22}N_4O_4$: calcd: 419.4602 $[M+H]^+$; found: 419.4597.

4.3.31. (5*R*,11a*R*)-2-Butyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-18. Yield: 23%, yellow solid.

T_M = 79 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.83 (t, J = 7 Hz, 3H, CH_3), 1.24 (m, 2H, CH_2CH$_3$), 1.46 (m, 2H, CH_2CH$_2$CH$_3$), 3.07 (m, 1H, CH_aH$_b$), 3.20–3.41 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.58 (dd, J_1 = 4.2 Hz, J_2 = 11.2 Hz, 1H, C*H*C(O)N), 6.09 (s, 1H, C*H*Ph), 6.97–8.28 (m, 8H, ar), 10.82 (br, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.42 (p), 19.28 (s), 21.68 (s), 29.58 (s), 37.46 (s), 54.82 (t), 57.32 (t), 105.68 (q), 111.33 (t), 118.38 (t), 118.96 (t), 121.71 (t), 122.53 (t), 122.61 (t), 125.79 (t), 129.87 (t), 133.53 (q), 134.08 (q), 136.81 (q), 142.84 (q), 147.60 (q), 154.46 (q), 171.63 (q).

HRMS (ESI): $C_{23}H_{22}N_4O_4$: calcd: 419.4602 $[M+H]^+$; found: 419.4598.

4.3.32. (5*R*,11a*S*)-2-Butyl-5-(3-nitrophenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-18. Yield: 20%, pale brown solid.

T_M = 67 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.2 Hz, 3H, CH_3), 1.28 (m, 2H, CH_2CH$_3$), 1.54 (m, 2H, CH_2CH$_2$CH$_3$), 2.80 (m, 1H, CH_aH$_b$), 3.40–3.46 (m, 3H, CH$_a$$H_b$, NC$H_2$), 4.72 (dd, J_1 = 5.4 Hz, J_2 = 10.5 Hz, 1H, C*H*C(O)N), 6.45 (s, 1H, C*H*Ph), 7.04–8.58 (m, 8H, ar), 10.94 (br, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.42 (p), 19.31 (s), 22.53 (s), 29.57 (s), 37.69 (s), 50.89 (t), 53.15 (t), 106.50 (q), 111.43 (t), 118.37 (t), 118.97 (t), 121.91 (t), 122.66 (t), 123.12 (t), 125.70 (t), 130.28 (t), 134.44 (q), 134.85 (q), 136.77 (q), 142.10 (q), 147.89 (q), 154.65 (q), 172.57 (q).

HRMS (ESI): $C_{23}H_{22}N_4O_4$: calcd: 441.1533 $[M+Na]^+$; found: 441.1530.

4.3.33. (5*S*,11a*S*)-2-Butyl-5-(2,3-dihydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-19. Yield: 25%, purple solid.

T_M = 176 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.85 (t, J = 7.0 Hz, 3H, CH_3), 1.24 (m, 2H, CH_2CH$_3$), 1.46 (m, 2H, CH_2CH$_2$CH$_3$), 2.88 (m, 1H, CH_aH$_b$), 3.28 (m, 1H, CH$_a$$H_b$), 3.33 (m, 2H, NC$H_2$), 4.48 (dd, J_1 = 4.4 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.66 (s, 1H, CHPh), 6.62–7.53 (m, 7H, ar), 8.72 (br, 1H, OH), 8.78 (br, 1H, OH), 10.67 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.45 (p), 19.32 (s), 21.94 (s), 29.65 (s), 37.34 (s), 55.51 (t), 57.59 (t), 104.65 (q), 111.25 (t), 114.67 (t), 115.10 (t), 118.00 (t), 118.70 (t), 118.79 (t), 121.23 (t), 125.89 (q), 131.39 (q), 135.38 (q), 136.60 (q), 144.78 (q), 144.89 (q), 154.00 (q), 171.69 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_4$: calcd: 406.1761 $[M+H]^+$; found: 406.1763.

4.3.34. (5*S*,11a*R*)-2-Butyl-5-(2,3-dihydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-19. Yield: 40%, purple solid.

T_M = 128 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.89 (t, J = 7.2 Hz, 3H, CH_3), 1.25 (m, 2H, CH_2CH$_3$), 1.53 (m, 2H, CH_2CH$_2$CH$_3$), 2.80 (m, 1H, CH_aH$_b$), 3.40 (m, 1H, CH$_a$$H_b$), 3.42 (m, 2H, NC$H_2$), 4.45 (dd, J_1 = 5.8 Hz, J_2 = 10.9 Hz, 1H, CHC(O)N), 6.10 (s, 1H, CHPh), 6.62–7.54 (m, 7H, ar), 8.99 (br, 2H, OH), 10.94 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.44 (p), 19.36 (s), 22.80 (s), 29.66 (s), 37.63 (s), 51.05 (t), 52.42 (t), 105.84 (q), 111.29 (t), 115.23 (t), 115.52 (t), 118.06 (t), 118.70 (t), 118.86 (t), 121.51 (t), 125.76 (q), 130.94 (q), 131.67 (q), 136.56 (q), 145.21 (q), 145.35 (q), 154.03 (q), 172.55 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_4$: calcd: 406.1761 $[M+H]^+$; found: 406.1764.

4.3.35. (5*R*,11a*R*)-2-Butyl-5-(2,3-dihydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-20. Yield: 45%, purple solid.

T_M = 154 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.90 (t, J = 7.2 Hz, 3H, CH_3), 1.26 (m, 2H, CH_2CH$_3$), 1.49 (m, 2H, CH_2CH$_2$CH$_3$), 2.93 (m, 1H, CH_aH$_b$), 3.33 (m, 1H, CH$_a$$H_b$), 3.42 (m, 2H, NC$H_2$), 4.52 (dd, J_1 = 4.5 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.70 (s, 1H, CHPh), 6.64–7.57 (m, 7H, ar), 8.80 (br, 2H, 2× OH), 10.67 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.47 (p), 19.34 (s), 21.97 (s), 29.67 (s), 37.35 (s), 55.53 (t), 57.59 (t), 104.66 (q), 111.24 (t), 114.66 (t), 115.09 (t), 117.98 (t), 118.69 (t), 118.78 (t), 121.22 (t), 125.88 (q), 131.37 (q), 135.36 (q), 136.58 (q), 144.77 (q), 144.87 (q), 153.97 (q), 171.66 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_4$: calcd: 406.1761 $[M+H]^+$; found: 406.1759.

4.3.36. (5*R*,11a*S*)-2-Butyl-5-(2,3-dihydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-20. Yield: 45%, purple solid.

T_M = 134 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 0.85 (t, J = 7.3 Hz, 3H, CH_3), 1.19 (m, 2H, CH_2CH$_3$), 1.52 (m, 2H, CH_2CH$_2$CH$_3$), 2.82 (m, 1H, CH_aH$_b$), 3.40 (m, 1H, CH$_a$$H_b$), 3.42 (m, 2H, NC$H_2$), 4.46 (dd, J_1 = 5.5 Hz, J_2 = 10.7 Hz, 1H, CHC(O)N), 6.08 (s, 1H, CHPh), 6.57–7.57 (m, 7H, ar), 10.94 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.47 (p), 19.37 (s), 22.82 (s), 29.82 (s), 37.64 (s), 51.07 (t), 52.44 (t), 105.87 (q), 111.33 (t), 115.26 (t), 115.55 (t), 118.12 (t), 118.74 (t), 118.90 (t), 121.56 (t), 125.80 (q), 130.98 (q), 131.71 (q), 136.60 (q), 145.25 (q), 145.40 (q), 154.08 (q), 172.62 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_4$: calcd: 406.1761 $[M+H]^+$; found: 406.1758.

4.3.37. (5*S*,11a*S*)-5-(3-Hydroxyphenyl)-2-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-21. Yield: 35%, brown solid.

T_M = 170 °C (decomp.).

^{1}H NMR (200 MHz, DMSO-d_6): δ = 3.15 (m, 1H, CH_aH$_b$), 3.34 (m, 1H, CH$_a$$H_b$), 4.69 (dd, J_1 = 4.4 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.81 (s, 1H, CHPh), 6.59–7.57 (m, 13H, ar), 9.29 (s, 1H, OH), 10.75 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 21.84 (s), 55.90 (t), 57.63 (t), 104.89 (q), 111.28 (t), 114.19 (t), 114.38 (t), 114.44 (t), 118.11 (t), 118.78 (t), 121.37 (t), 121.80 (t), 125.83 (q), 126.44 (t), 127.69 (t), 128.64 (t), 129.10 (t), 131.89 (q), 134.60 (q), 136.64 (t), 139.70 (q), 141.92 (q), 153.03 (q), 157.13 (q), 170.58 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_3$: calcd: 410.1499 $[M+H]^+$; found: 410.1500.

4.3.38. (5*S*,11a*R*)-5-(3-Hydroxyphenyl)-2-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-21. Yield: 35%, pale yellow solid.

T_M = 161 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 3.10 (m, 1H, CH_aH$_b$), 3.48 (m, 1H, CH$_a$$H_b$), 4.70 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.24 (s, 1H, CHPh), 6.78–7.61 (m, 13H, ar), 11.12 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 22.65 (s), 51.66 (t), 52.80 (t), 106.09 (q), 111.42 (t), 114.99 (t), 115.25 (t), 118.15 (t), 118.23 (t), 118.39 (t), 118.86 (t), 121.71 (t), 125.82 (q), 126.93 (t), 127.95 (t), 128.74 (t), 129.77 (t), 131.11 (q), 131.95 (q), 136.69 (t), 141.17 (2× q), 153.27 (q), 157.96 (q), 171.61 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_3$: calcd: 410.1499 $[M+H]^+$; found: 410.1496.

4.3.39. (5*R*,11a*R*)-5-(3-Hydroxyphenyl)-2-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-22. Yield: 61%, yellow solid.

T_M = 171 °C (decomp.).

^{1}H NMR (300 MHz, DMSO-d_6): δ = 3.09 (m, 1H, CH_aH$_b$), 3.45 (m, 1H, CH$_a$$H_b$), 4.73 (dd, J_1 = 4.2 Hz, J_2 = 11.4 Hz, 1H, C*H*C(O)N), 5.87 (s, 1H, C*H*Ph), 6.65–7.61 (m, 13H, ar), 9.35 (s, 1H, OH), 10.81 (br, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 21.86 (s), 55.93 (t), 57.66 (t), 104.93 (q), 111.34 (t), 113.86 (t), 114.22 (t), 114.44 (t), 118.18 (t), 118.85 (t), 121.45 (t), 121.80 (t), 125.89 (q), 126.51 (t), 127.77 (t), 128.77 (t), 129.19 (t), 131.93 (q), 134.66 (q), 136.69 (t), 139.70 (q), 142.00 (q), 153.10 (q), 157.19 (q), 170.67 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_3$: calcd: 410.1499 $[M+H]^+$; found: 410.1501.

4.3.40. (5*R*,11a*S*)-5-(3-Hydroxyphenyl)-2-phenyl-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-22. Yield: 97%, dark brown solid.

T_M = 168 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 3.08 (m, 1H, CH_aH$_b$), 3.44 (m, 1H, CH$_a$$H_b$), 4.70 (dd, J_1 = 5.4 Hz, J_2 = 10.8 Hz, 1H, C*H*C(O)N), 6.24 (s, 1H, C*H*Ph), 6.75–7.62 (m, 13H, ar), 9.52 (s, 1H, OH), 11.00 (br, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 22.65 (s), 51.63 (t), 52.80 (t), 106.12 (q), 111.43 (t), 114.91 (t), 115.18 (t), 118.17 (t), 118.64 (t), 118.88 (t), 121.73 (t), 121.81 (t), 125.82 (q), 126.93 (t), 127.96 (t), 128.75 (t), 128.80 (t), 129.81 (q), 131.07 (q), 131.95 (t), 136.70 (q), 141.21 (q), 153.29 (q), 157.66 (q), 171.61 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_3$: calcd: 410.1499 $[M+H]^+$; found: 410.1494.

4.3.41. (5*S*,11a*S*)-2-Benzyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-23. Yield: 94%, pale yellow solid.

T_M = 131 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 3.00 (m, 1H, CH_aH$_b$), 3.35 (m, 1H, CH$_a$$H_b$), 4.53–4.65 (m, 3H, C*H*C(O)N, CH_2Ph), 5.76 (s, 1H, C*H*Ph), 6.59–7.55 (m, 13H, ar), 9.27 (s, 1H, OH), 10.71 (s, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 22.58 (s), 41.81 (s), 56.45 (t), 58.41 (t), 105.49 (q), 112.01 (t), 114.92 (t), 115.16 (t), 118.82 (t), 118.88 (t), 119.51 (t), 122.09 (t), 126.55 (q), 127.23 (q), 127.68 (t), 128.06 (t), 128.89 (t), 129.19 (t), 129.84 (t), 135.47 (q), 137.28 (t), 137.33 (q), 142.71 (q), 154.57 (q), 157.88 (q), 172.27 (q).

HRMS (ESI): $C_{26}H_{21}N_3O_3$: calcd: 424.1656 $[M+H]^+$; found: 424.1656.

4.3.42. (5*S*,11a*R*)-2-Benzyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-23. Yield: 79%, brown solid.

T_M = 151 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.82 (m, 1H, CH_aH$_b$), 3.38 (m, 1H, CH$_a$$H_b$), 4.55–4.62 (m, 3H, C*H*C(O)N, CH_2Ph), 6.16 (s, 1H, C*H*Ph), 6.70–7.56 (m, 13H, ar), 9.47 (s, 1H, OH), 10.93 (s, 1H, N*H*).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 23.51 (s), 42.16 (s), 52.24 (t), 53.68 (t), 106.68 (q), 112.15 (t), 115.55 (t), 115.89 (t), 118.95 (t), 119.21 (t), 119.56 (t), 122.44 (t), 126.51 (q), 128.11 (2× t), 128.18 (t), 129.31 (2× t), 130.54 (q), 131.87 (q), 137.32 (q), 137.42 (t), 142.05 (q), 154.81 (q), 158.38 (q), 173.22 (q).

HRMS (ESI): $C_{26}H_{21}N_3O_3$: calcd: 424.1656 $[M+H]^+$; found: 424.1647.

4.3.43. (5*R*,11a*R*)-2-Benzyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-24. Yield: 86%, yellow solid.

T_M = 204 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 3.04 (m, 1H, CH_aH$_b$), 3.38 (m, 1H, CH$_a$$H_b$), 4.58–4.68 (m, 3H, C*H*C(O)N, CH_2Ph), 5.82 (s, 1H, C*H*Ph), 6.64–7.59 (m, 13H, ar), 9.34 (s, 1 H, OH), 10.78 (s, 1 H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 22.62 (s), 41.88 (s), 56.49 (t), 58.45 (t), 105.55 (q), 112.06 (t), 114.96 (t), 115.21 (t), 118.87 (t), 118.94 (t), 119.57 (t), 122.13 (t), 126.59 (q), 128.14 (2× t), 129.23 (2× t), 129.90 (q), 128.89 (t), 135.52 (q), 137.34 (t), 137.38 (q), 142.76 (q), 154.61 (q), 157.94 (q), 172.36 (q).

HRMS (ESI): $C_{26}H_{21}N_3O_3$: calcd: 424.1656 $[M+H]^+$; found: 424.1653.

4.3.44. (5*R*,11a*S*)-2-Benzyl-5-(3-hydroxyphenyl)-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-24. Yield: 79%, brown solid.

T_M = 148 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.86 (m, 1H, CH_aH$_b$), 3.40 (m, 1H, CH$_a$$H_b$), 4.59–4.65 (m, 3H, C*H*C(O)N, CH_2Ph), 6.19 (s, 1H, C*H*Ph), 6.74–7.59 (m, 13H, ar), 9.51 (s, 1H, OH), 10.98 (s, 1H, N*H*).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 23.50 (s), 42.14 (s), 52.23 (t), 53.66 (t), 106.65 (q), 112.11 (t), 115.52 (t), 115.87 (t), 118.93 (t), 119.19 (t), 119.57 (t), 122.42 (t), 126.49 (q), 128.08 (t), 128.17 (2× t), 129.30 (2× t), 130.52 (q), 131.86 (t), 137.29 (q), 137.38 (q), 142.02 (q), 154.79 (q), 158.34 (q), 173.21 (q).

HRMS (ESI): $C_{26}H_{21}N_3O_3$: calcd: 424.1656 $[M+H]^+$; found: 424.1656.

4.3.45. Methyl(2*S*)-4-methyl-2-(5*S*,11a*S*)-5-(3-hydroxyphenyl)-1,3-dioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo-[1,5-*b*]-β-carboline-pentanoate *cis*-25. Yield: 13%, brown solid.

T_M = 123 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.90 (m, 6H, CH(CH_3)$_2$), 1.56 (m, 1H, CH(CH$_3$)$_2$), 1.77 (m, 1H, CHCH_aH$_b$CH), 2.06 (m, 1H, CHCH$_a$$H_b$CH), 2.92 (m, 1H, CC$H_aH_b$), 3.41 (m, 1H, CCH$_a$$H_b$), 3.63 (m, 3H, OC$H_3$), 4.69 (m, 2H, C$H$C(O)N, NC$H$C(O)OMe), 5.85 (s, 1H, C$H$Ph), 6.61–7.59 (m, 8H, ar), 9.34 (s, 1H, OH), 10.80 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 20.88 (p), 22.22 (s), 23.03 (p), 24.33 (t), 36.23 (s), 50.12 (t), 52.57 (p), 55.71 (t), 57.16 (t), 104.40 (q), 111.31 (t), 114.04 (t), 114.44 (t), 117.90 (t), 118.07 (t), 118.81 (t), 121.40 (t), 125.84 (q), 129.11 (t), 134.68 (q), 136.61 (q), 141.85 (q), 153.02 (q), 157.21 (q), 169.58 (q), 171.47 (q).

HRMS (ESI): $C_{26}H_{27}N_3O_5$: calcd: 462.2024 $[M+H]^+$; found: 462.2026.

4.3.46. Methyl(2*S*)-4-methyl-2-(5*S*,11a*R*)-5-(3-hydroxyphenyl)-1,3-dioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo-[1,5-*b*]-β-carboline-pentanoate *trans*-25. Yield: 65%, brown solid.

T_M = 102 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.84 (m, 6H, CH(CH_3)$_2$), 1.39 (m, 1H, CH(CH$_3$)$_2$), 1.76 (m, 1H, CHCH_aH$_b$CH), 2.05 (m, 1H, CHCH$_a$$H_b$CH), 2.71 (m, 1H, CC$H_aH_b$), 3.43 (m, 1H, CCH$_a$$H_b$), 3.61 (m, 3H, OC$H_3$), 4.57–4.72 (m, 2H, C$H$C(O)N, NC$H$C(O)OMe), 6.14 (s, 1H, C$H$Ph), 6.70–7.56 (m, 8H, ar), 9.49 (s, 1H, OH), 10.95 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 20.83 (p), 22.96 (s), 23.10 (p), 24.45 (t), 36.42 (s), 50.36 (t), 51.39 (p), 52.57 (t), 52.64 (t), 105.68 (q), 111.41 (t), 114.56 (t), 114.67 (t), 118.24 (t), 118.35 (t), 118.89 (t), 121.77 (t), 125.68 (q), 129.81 (t), 130.96 (q), 136.62 (q), 141.12 (q), 153.49 (q), 157.62 (q), 169.63 (q), 172.39 (q).

HRMS (ESI): $C_{26}H_{27}N_3O_5$: calcd: 484.1843 $[M+Na]^+$; found: 484.1843.

4.3.47. Methyl(2*S*)-4-methyl-2-(5*R*,11a*R*)-5-(3-hydroxyphenyl)-1,3-dioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo-[1,5-*b*]- β-carboline-pentanoate *cis*-26. Yield: 15%, yellow solid.

T_M = 127 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.89 (m, 6H, CH(CH_3)$_2$), 1.55 (m, 1H, CH(CH$_3$)$_2$), 1.76 (m, 1H, CHCH_aH$_b$CH), 2.08 (m, 1H, CHCH$_a$$H_b$CH), 2.95 (m, 1H, CC$H_aH_b$), 3.38 (m, 1H, CCH$_a$$H_b$), 3.63 (m, 3H, OC$H_3$), 4.69 (m, 2H, C$H$C(O)N, NC$H$C(O)OMe), 5.79 (s, 1H, C$H$Ph), 6.63–7.60 (m, 8H, ar), 9.31 (s, 1H, OH), 10.82 (br, 1H, NH).

^{13}C NMR (600 MHz, DMSO-d_6): δ = 20.82 (p), 22.08 (s), 23.09 (p), 24.31 (t), 36.43 (s), 49.89 (t), 52.37 (p), 55.47 (t), 57.14 (t), 104.18 (q), 111.32 (t), 113.77 (t), 114.38 (t), 117.67 (t), 118.14 (t), 118.83 (t), 121.43 (t), 125.85 (q), 129.10 (t), 134.72 (q), 136.61 (q), 142.09 (q), 153.05 (q), 157.25 (q), 169.66 (q), 171.52 (q).

HRMS (ESI): $C_{26}H_{27}N_3O_5$: calcd: 462.2024 $[M+H]^+$; found: 462.2012.

4.3.48. Methyl(2*S*)-4-methyl-2-(5*R*,11a*S*)-5-(3-hydroxyphenyl)-1,3-dioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-pentanoate *trans*-26. Yield: 61%, brown solid.

T_M = 70 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.82–0.90 (m, 6H, CH(CH_3)$_2$), 1.47 (m, 1H, CH(CH$_3$)$_2$), 1.81 (m, 1H, CHCH_aH$_b$CH), 2.09 (m, 1H, CHCH$_a$$H_b$CH), 2.77 (m, 1H, CC$H_aH_b$), 3.49 (m, 1H, CCH$_a$$H_b$), 3.66 (m, 3H, OC$H_3$), 4.63–4.78 (m, 2H, C$H$C(O)N, NC$H$C(O)OMe), 6.20 (s, 1H, C$H$Ph), 6.75–7.61 (m, 8H, ar), 9.59 (s, 1H, OH), 11.02 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 20.63 (p), 22.76 (s), 23.02 (p), 24.67 (t), 36.29 (s), 50.20 (t), 51.81 (p), 52.78 (t), 53.04 (t), 105.72 (q), 111.43 (t), 114.50 (t), 114.87 (t), 117.93 (t), 118.36 (t), 118.99 (t), 121.71 (t), 125.55 (q), 129.67 (t), 130.86 (q), 136.69 (q), 141.22 (q), 153.67 (q), 157.22 (q), 169.43 (q), 171.96 (q).

HRMS (ESI): $C_{26}H_{27}N_3O_5$: calcd: 462.2024 $[M+H]^+$; found: 462.2021.

4.3.49. (5*S*, 11a*R*)-2-Butyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 27. Yield: 84%, yellow-greenish solid.

T_M = 52 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.90 (t, J = 6.8 Hz, 3H, CH_3), 1.28 (m, 2H, CH_2CH$_3$), 1.61 (m, 2H, CH_2CH$_2$CH$_3$), 2.93 (m, 1H, CH_aH$_b$), 3.49 (m, 1H, CH$_a$$H_b$), 3.76 (m, 2H, NC$H_2$), 4.79 (dd, J_1 = 6.2 Hz, J_2 = 10.9 Hz, 1H, CHC(O)N), 6.73–6.78 (m, 1H, ar), 6.84 (s, 1H, CHPh), 6.89–7.59 (m, 7H, ar), 9.51 (s, 1H, OH), 11.05 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 13.62 (p), 19.47 (s), 23.05 (s), 29.35 (s), 40.41 (s), 54.94 (t), 55.32 (t), 105.61 (q), 111.44 (t), 115.36 (t), 115.45 (t), 118.30 (t), 118.96 (2× t), 121.84 (t), 125.62 (q), 129.69 (t), 131.22 (q), 136.76 (q), 140.10 (q), 157.54 (q), 173.19 (q), 179.78 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2S$: calcd: 406.1584 $[M+H]^+$; found: 406.1587.

4.3.50. (5*R*,11a*S*)-2-Butyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 28. Yield: 92%, yellow solid.

T_M = 78 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.91 (t, J = 6.8 Hz, 3H, CH_3), 1.28 (m, 2H, CH_2CH$_3$), 1.61 (m, 2H, CH_2CH$_2$CH$_3$), 2.93 (m, 1H, CH_aH$_b$), 3.49 (m, 1H, CH$_a$$H_b$), 3.76 (m, 2H, NC$H_2$), 4.79 (dd, J_1 = 6.2 Hz, J_2 = 10.9 Hz, 1H, CHC(O)N), 6.73–6.78 (m, 1H, ar), 6.84 (s, 1H, CHPh), 6.89–7.59 (m, 7H, ar), 9.51 (s, 1H, OH), 11.05 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 13.62 (p), 19.47 (s), 23.05 (s), 29.35 (s), 40.41 (s), 54.94 (t), 55.32 (t), 105.61 (q), 111.44 (t), 115.36 (t), 115.45 (t), 118.30 (t), 118.96 (2× t), 121.84 (t), 125.62 (q), 129.69 (t), 131.22 (q), 136.76 (q), 140.11 (q), 157.54 (q), 173.20 (q), 179.79 (q).

HRMS (ESI): $C_{23}H_{23}N_3O_2S$: calcd: 406.1584 $[M+H]^+$; found: 406.1585.

4.3.51. (5*S*,11a*R*)-2-Allyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 29. Yield: 98%, yellow solid.

T_M = 116 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.97 (m, 1H, CH_aH$_b$), 3.49 (m, 1H, CH$_a$$H_b$), 4.38 (m, 2H, NC$H_2$), 4.84 (dd, J_1 = 6.1 Hz, J_2 = 10.7 Hz, 1H, CHC(O)N), 5.13 (m, 2H, CHCH_2), 5.85 (m, 1H, CHCH$_2$), 6.73–6.78 (m, 1H, ar), 6.85 (s, 1H, CHPh), 6.90–7.60 (m, 7H, ar), 9.52 (s, 1H, OH), 11.06 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 23.03 (s), 42.88 (s), 55.04 (t), 55.40 (t), 105.57 (q), 111.41 (t), 115.38 (t), 116.76 (t), 116.97 (t), 118.25 (s), 118.89 (t), 121.81 (t), 125.59 (q), 129.63 (t), 131.13 (t), 131.39 (q), 131.65 (t), 136.72 (q), 140.01 (q), 157.49 (q), 172.71 (q), 179.46 (q).

HRMS (ESI): $C_{22}H_{20}N_3O_2S$: calcd: 390.1271$[M+H]^+$; found: 390.1273.

4.3.52. (5*R*,11a*S*)-2-Allyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 30. Yield: 98%, yellow solid.

T_M = 70 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.96 (m, 1H, CH_aH$_b$), 3.50 (m, 1H, CH$_a$$H_b$), 4.35 (m, 2H, NC$H_2$), 4.84 (dd, J_1 = 6.2 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 5.07 (m, 2H, CHCH_2), 5.82 (m, 1H, CHCH$_2$), 6.75–6.78 (m, 1H, ar), 6.85 (s, 1H, CHPh), 6.91–7.60 (m, 7H, ar), 9.53 (s, 1H, OH), 11.01 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 23.74 (s), 43.61 (s), 55.77 (t), 56.15 (t), 106.32 (q), 112.16 (t), 116.01 (t), 116.14 (t), 117.70 (s), 119.05 (t), 119.65 (t), 119.70 (t), 122.58 (t), 126.32 (q), 130.41 (t), 131.89 (q), 132.42 (t), 137.46 (q), 140.77 (q), 158.24 (q), 173.50 (q), 180.21 (q).

HRMS (ESI): $C_{22}H_{20}N_3O_2S$: calcd: 390.1271$[M+H]^+$; found: 390.1272.

4.3.53. (5*S*,11a*S*)-5-(3-Hydroxyphenyl)-2-[2-(trifluormethoxy)phenyl]-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-31. Yield: 44%, pale yellow solid.

T_M = 159 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.99 (m, 1H, CH_a H$_b$), 3.52 (m, 1H, CH$_a$$H_b$), 4.82 (dd, J_1 = 3.9 Hz, J_2 = 11.4 Hz, 1H, CHC(O)N), 5.89 (s, 1H, CHPh), 6.67–7.64 (m, 12H, ar), 9.36 (s, 1H, OH), 10.86 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 22.40 (s), 55.97 (t), 57.75 (t), 104.50 (q), 111.38 (t), 114.38 (t), 114.51 (t), 117.65 (t), 118.19 (t), 118.91 (t), 121.51 (t), 121,84 (t), 124.61 (q), 125.88 (t), 127.83 (t), 128.10 (q), 129.10 (t), 130.94 (t), 134.69 (q), 136.75 (q), 141.78 (q), 144.46 (q), 152.13 (q), 157.28 (q), 165.02 (q), 170.23 (q).

HRMS (ESI): $C_{26}H_{18}F_3N_3O_4$: calcd: 494.1322 $[M+H]^+$; found: 494.1319.

4.3.54. (5*S*,11a*R*)-5-(3-Hydroxyphenyl)-2-[2-(trifluormethoxy)phenyl]-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-31. Yield: 90%, brown solid.

T_M = 158 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.79 (m, 1H, CH_aH$_b$), 3.54 (m, 1H, CH$_a$$H_b$), 4.78 (dd, J_1 = 5.1 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.29 (s, 1H, CHPh), 6.77–7.67 (m, 12H, ar), 9.54 (s, 1H, OH), 11.04 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 22.52 (s), 51.80 (t), 53.26 (t), 105.62 (q), 111.46 (t), 115.08 (t), 115.19 (t), 115.28 (t), 118.29 (t), 118.60 (t), 118.92 (t), 121.84 (t), 124.43 (q), 125.79 (t), 125.79 (t), 128.42 (q), 129.78 (t), 131.04 (q), 131.23 (q), 136.69 (t), 140.95 (q), 144.55 (q), 152.42 (q), 157.65 (q), 171.02 (q).

HRMS (ESI): $C_{26}H_{18}F_3N_3O_4$: calcd: 494.1322 $[M+H]^+$; found: 494.1312.

4.3.55. (5*R*,11a*R*)-5-(3-Hydroxyphenyl)-2-[2-(trifluormethoxy)phenyl]-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *cis*-32. Yield: 44%, yellow solid.

T_M = 175 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.93 (m, 1H, CH_aH$_b$), 3.45 (m, 1H, CH$_a$$H_b$), 4.80 (dd, J_1 = 3.9 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 5.87 (s, 1H, CHPh), 6.65–7.63 (m, 12H, ar), 9.35 (s, 1H, OH), 10.84 (s, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 23.09 (s), 56.67 (t), 58.67 (t), 105.18 (q), 112.10 (t), 115.06 (t), 115.19 (t), 118.33 (t), 118.96 (t), 119.64 (t), 122.25 (t), 122.67 (t), 125.33 (q), 126.58 (q), 128.58 (q), 128.92 (t), 129.84 (t), 131.61 (t), 131.73 (t), 135.42 (q), 136.43 (q), 142.50 (q), 145.14 (q), 152.86 (q), 158.01 (q), 171.00 (q).

HRMS (ESI): $C_{26}H_{18}F_3N_3O_4$: calcd: 494.1322 $[M+H]^+$; found: 494.1313.

4.3.56. (5*R*,11a*S*)-5-(3-Hydroxyphenyl)-2-[2-(trifluormethoxy)phenyl]-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1,3-dione *trans*-32. Yield: 44%, brown solid.

T_M = 160 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.80 (m, 1H, CH_aH_b), 3.55 (m, 1H, CH$_a$$H_b$), 4.79 (m, 1H, C$H$C(O)N), 6.30 (s, 1H, C$H$Ph), 6.79–7.68 (m, 12H, ar), 9.57 (s, 1H, OH), 11.08 (s, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 22.62 (s), 51.80 (t), 53.26 (t), 105.62 (q), 111.45 (t), 114.98 (t), 115.13 (t), 115.26 (t), 118.09 (t), 118.40 (t), 118.92 (t), 121.78 (t), 124.22 (q), 125.71 (q), 125.79 (q), 128.14 (t), 129.70 (t), 129.77 (t), 131.07 (t), 131.23 (q), 136.67 (q), 140.94 (q), 144.51 (q), 152.41 (q), 157.62 (q), 171.01 (q).

HRMS (ESI): $C_{26}H_{18}F_3N_3O_4$: calcd: 494.1322 $[M+H]^+$; found: 494.1324.

4.3.57. (5*S*,11a*R*)-2-Ethyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 33. Yield: 24%, brown solid.

T_M = 118 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.17 (t, J = 7.0 Hz, 3H, CH_3), 2.94 (m, 1H, CH_aH_b), 3.48 (m, 1H, CH$_a$$H_b$), 3.80 (m, 2H, NC$H_2$), 4.79 (dd, J_1 = 6.2 Hz, J_2 = 10.6 Hz, 1H, CHC(O)N), 6.84 (s, 1H, CHPh), 6.69–7.54 (m, 8H, ar), 9.46 (s, 1H, OH), 11.00 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 12.73 (p), 22.83 (s), 35.88 (s), 54.81 (t), 56.26 (t), 105.52 (q), 111.33 (t), 115.26 (t), 115.35 (t), 118.16 (t), 118.86 (2× t), 121.73 (t), 125.54 (q), 129.54 (t), 131.11 (q), 136.68 (q), 139.98 (q), 157.43 (q), 172.76 (q), 179.43 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_2S$ H^+: calcd: 378.1271 $[M+H]^+$; found: 378.1266.

4.3.58. (5*R*,11a*S*)-2-Ethyl-5-(3-hydroxyphenyl)-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 34. Yield: quant., pale yellow solid.

T_M = 132 °C.

^{1}H NMR (200 MHz, DMSO-d_6): δ = 1.16 (t, J = 7.3 Hz, 3H, CH_3), 2.97 (m, 1H, CH_aH_b), 3.46 (m, 1H, CH$_a$$H_b$), 3.75 (m, 2H, NC$H_2$), 4.78 (dd, J_1 = 6.2 Hz, J_2 = 10.7 Hz, 1H, CHC(O)N), 6.84 (s, 1H, CHPh), 6.77–7.55 (m, 8H, ar), 9.51 (s, 1H, OH), 11.05 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 12.78 (p), 22.84 (s), 35.90 (s), 54.83 (t), 55.30 (t), 105.57 (q), 111.38 (t), 115.31 (t), 115.40 (t), 118.23 (t), 118.92 (2× t), 121.79 (t), 125.58 (q), 129.62 (t), 131.15 (q), 136.72 (q), 140.05 (q), 157.48 (q), 172.84 (q), 179.47 (q).

HRMS (ESI): $C_{21}H_{19}N_3O_2S$ H^+: calcd: 378.1271 $[M+H]^+$; found: 378.1270.

4.3.59. (5*S*,11a*R*)-5-(3-Hydroxyphenyl)-2-phenyl-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 35. Yield: quant., auburn solid.

T_M = 118 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 3.19– 3.59 (m, 2H, CH_aH_b), 4.98 (dd, J_1 = 6.2 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.78 (m, 1H, ar), 6.94 (s, 1H, CHPh), 6.96–7.63 (m, 12H, ar), 9.57 (s, 1H, OH), 11.10 (br, 1H, NH).

^{13}C NMR (75 MHz, DMSO-d_6): δ = 23.04 (s), 55.18 (t), 55.77 (t), 105.73 (q), 111.49 (t), 115.46 (t), 115.64 (t), 118.33 (t), 119.03 (t), 119.21 (t), 121.89 (t), 125.69 (q), 125.97 (t), 128.01 (t), 128.76 (t), 129.07 (t), 129.74 (t), 129.93 (t), 131.24 (q), 133.56 (q), 136.79 (q), 140.04 (q), 157.60 (q), 172.67 (q), 179.82 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_2S$: calcd: 426.1271 $[M+H]^+$; found: 426.1266.

4.3.60. (5*R*,11a*S*)-5-(3-Hydroxyphenyl)-2-phenyl-3-thioxo-6*H*-1,2,3,5,11,11a-hexahydro-imidazo[1,5-*b*]-β-carboline-1-on 36. Yield: quant., auburn solid.

T_M = 107 °C.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 3.23 (m, 1H, CH_aH_b), 3.56 (m, 1H, CH$_a$$H_b$), 4.99 (dd, J_1 = 6.3 Hz, J_2 = 10.8 Hz, 1H, CHC(O)N), 6.78 (m, 1H, ar), 6.95 (s, 1H, CHPh), 6.97–7.63 (m, 12H, ar), 9.58 (s, 1H, OH), 11.11 (br, 1H, NH).

^{13}C NMR (50 MHz, DMSO-d_6): δ = 23.03 (s), 55.13 (t), 55.71 (t), 105.67 (q), 111.41 (t), 115.40 (t), 115.58 (t), 118.24 (t), 118.95 (t), 119.13 (t), 121.81 (t), 125.63 (q), 125.85 (t), 127.90 (t), 128.66 (t), 128.97 (t), 129.64 (t), 129.83 (q), 131.16 (q), 133.48 (t), 136.73 (q), 139.67 (q), 157.53 (q), 172.55 (q), 179.74 (q).

HRMS (ESI): $C_{25}H_{19}N_3O_2S$: calcd: 426.1271 $[M+H]^+$; found: 426.1267.

4.3.61. Methyl *N*-(oxomethylen)-L-leucinate 37. *Caution! The evolving phosgene is a very toxic gas!.*

To a solution of L-leucine methylester (1.18 g, 6.5 mmol) in a mixture of dichloromethane (25 ml) and sat. NaHCO$_3$-solution (25 ml), triphosgen (0.65 g, 2.2 mmol, 0.34 equiv) was added at 0 °C under vigorous stirring. After 15 min, the reaction was warmed to rt and extracted three times with ethylacetate. The combined organic layers were dried over Na_2SO_4 and concentrated in vacuo.

Yield: 945 mg, 5.5 mmol, 85%, colorless oil.

^{1}H NMR (300 MHz, DMSO-d_6): δ = 0.91 (d, 6H, 2× CH_3), 1.50–1.74 (m, 3H, CH_2CH(CH$_3$)$_2$), 3.77 (s, 3H, OCH_3), 4.40 (dd, J_1 = 3.6 Hz, J_2 = 5.8 Hz, 1H, NCH).

4.4. Biology/biochemistry

4.4.1. Proteins. A sequence of *Xenopus laevis* KSP (Eg5) corresponding to amino acids 1–487 ($KSP_{487}H$) was expressed and purified as described.[22] Tubulin was purified as described.[24] Microtubules were polymerized in vitro according to a standard procedure.[22]

4.4.2. Compound library screen. The measurements were performed in 384 well plates and every compound was measured in duplicate. The compound solutions in DMSO (final concentration 46 μM) were added to a solution of Eg5 (48 nM) and microtubules (80 nM) in assay buffer (20 mM pipes, pH 6.8 1 mM EGTA, 1 mM $mgCl_2$, 0.1 mg ml^{-1} BSA, 1 μM Taxol). After 10 min, a solution of ATP (50 μM) was added to start the reaction. The incubation period of 15 min is followed by addition of malachite green (33 μM), ammonium molybdate tetrahydrate (775 μM) and Perchloric acid (425 mM), and incubation of another 20 min. Finally, the absorption at 610 nM is measured.

4.4.3. IC_{50} determination. To determine the inhibitor concentration that inhibits the microtubule-stimulated ATPase activity of Eg5 by 50% (IC_{50}) we used two different assays: the malachite green assay (see above) and the EnzChek® Phosphate Assay (Molecular Probes). In both cases, 1% (v/v) inhibitor in DMSO at different concentrations (or 1% (v/v) DMSO as a control) was added at the start of the experiment. At least four measurements were performed per condition and the results were then averaged.

For the EnzCheck® Phosphate Assay final concentrations were 40 nM $Eg5_{487}H$, 4 μM microtubules (tubulin dimer concentration as determined by Bradford using BSA as standard), 1 mM MgATP, and 46 μM inhibitor. The ATPase activity was measured as an increase of absorbance at 365 nm within 2 h at room temperature. The microtubule-stimulated activity of $Eg5_{487}H$ was obtained by subtracting background activity (measured with microtubules only in the absence of $Eg5_{487}H$) from measurements containing $Eg5_{487}H$ and microtubules. The IC_{50} was determined by assuming non-competitive enzyme inhibition and fitting the theoretically expected dependence of the enzyme activity on the inhibitor concentration to the experimental microtubule-stimulated ATPase activities.

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