SUPPLEMENTARY MATERIAL

Synthesis of podophyllotoxin-glycosyl triazoles via click protocol mediated by Silver (I) N-heterocyclic carbenes and their anti-cancer evaluation as Topoisomerase-II inhibitors

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Abstract

Herein we report the regioselective synthesis of podophyllotoxin-Glycosyl triazole hybrids catalysed by Ag(I)- N-heterocyclic carbene (Ag(I)-NHC) in a short reaction time (~30 min) at ambient conditions. In principle, it is the first report of Click alkyne-azide cycloaddition catalysed by Ag(I)-NHC catalyst and moreover, this new methodology yielded good results when compared with traditional CuAAC in terms of reaction time and selectivity. The synthesised compounds were further explored for \textit{in vitro} anticancer activity against four human cancer cell lines Du145, HeLa, A-549, and MCF-7 and found that these synthesised compounds possess significant anticancer activity. Further, the compounds 5a and 5e were identified as promising leads due to their better activity across all cell lines than that of the standard drug etoposide. Molecular docking studies of 5a & 5e with DNA Topoisomerase-II were revealed that the free energy calculations of active compounds were in good agreement with observed IC\textsubscript{50} values.

Keywords: Podophyllotoxin; click chemistry; Ag (NHC) catalyst; podophyllotoxin-glycosyl triazoles; anticancer studies;
List of Contents

1. Figures and Tables cited in the Main Article 3-5

2. General 5

3. General Synthetic procedures 6-7
   3.1. General procedure for the synthesis of 4β-(prop-2-ynyloxy) epipodophyllotoxin (3)
   3.2. General procedure for the synthesis of Sugar azides (4a-l)
   3.3. General procedure for the synthesis of Podophyllotoxin-triazole hybrids (5a-l)

4. Spectral data of compounds (5a-l) 7-11

5. In vitro cytotoxicity assay 11

6. Molecular Docking Studies 11-13

7. Representative spectra for key compounds 14-20
1. Figures and Tables cited in the Main article

**Figure S1.** The Docking poses of 5a & 5e with Topoisomerase-II α enzyme (Homo sapiens) PBD ID 1ZXN

![Docking poses of 5a & 5e with Topoisomerase-II α enzyme](image)

**Table S1.** Optimization of reaction parameters for the synthesis of 5e

| Entry | Catalyst | Solvent | Yield (%) |
|-------|----------|---------|-----------|
| 1     | No Catalyst | EtOH     | --        |
| 2     | Cu       | EtOH     | Trace     |
| 3     | CuSO₄   | EtOH     | Trace     |
| 4     | Cu(OAc)₂| EtOH     | Trace     |
| 5     | AgOAc   | EtOH     | --        |
| 6     | AgOTF   | EtOH     | --        |
| 7     | Zn(OAc)₂| EtOH     | --        |
| 8     | Zn(OTf)₂| EtOH     | --        |
| 9     | Ag(I)-NHC | EtOH    | 94        |
| 10    | Ag(I)-NHC + Cu | EtOH | 88        |
| 11    | Ag(I)-NHC + Zn(OTf)₂ | EtOH | 85        |
| 12    | Organo-NHC | EtOH | 74        |
| 13    | Organo-NHC + AgOTf | EtOH | 91        |

*a* GC Yields; *b* After 24 h at reflux temperature; *c* After 24 h at RT
Table S2. Silver (I)-NHC catalyzed synthesis of podophyllotoxin-glycosyl triazoles (5a-l) \(^a\)

| Entry | R-N\(_3\) | Product | Reaction time (min.), (yield, %) \(^b\) |
|-------|----------|----------|--------------------------------------|
| 1     | \(\text{AcO-}\) | 5a       | 45 (92)                              |
| 2     | \(\text{AcO-}\) | 5b       | 40 (85)                              |
| 3     | \(\text{AcO-}\) | 5c       | 50 (82)                              |
| 4     | \(\text{AcO-}\) | 5d       | 50 (80)                              |
| 5     | \(\text{AcO-}\) | 5e       | 30 (94)                              |
| 6     | \(\text{AcO-}\) | 5f       | 35 (88)                              |
| 7     | \(\text{AcO-}\) | 5g       | 35 (90)                              |
| 8     | \(\text{AcO-}\) | 5h       | 30 (85)                              |
| 9     | \(\text{AcO-}\) | 5i       | 40 (92)                              |
| 10    | \(\text{AcO-}\) | 5j       | 40 (86)                              |
| 11    | \(\text{AcO-}\) | 5k       | 30 (92)                              |
| 12    | \(\text{AcO-}\) | 5l       | 30 (87)                              |

\(^a\)All products were characterized by \(^1\)H, \(^13\)C NMR and mass spectral analysis. \(^b\)Isolated yields after column chromatography.
Table S3. *in vitro* cytotoxic activity of podophyllotoxin-glycosyl triazole hybrids (5a-l) on human cancer cell lines \(^a\) (IC\(_{50}\) \(\mu\)M)\(^b\)

| Entry | Compound | Du145 (Prostate) | HeLa (Cervical) | A-549 (Lung) | MCF-7 (Breast) |
|-------|----------|-----------------|----------------|-------------|---------------|
| 1     | 5a       | 1.02±0.02       | 2.82±0.07      | 2.01±0.12   | 1.51±0.06     |
| 2     | 5b       | 5.82±0.12       | 10.24±0.06     | 9.84±0.02   | 6.25±0.08     |
| 3     | 5c       | 14.12±0.09      | 13.06±0.18     | 9.20±0.02   | 14.05±0.02    |
| 4     | 5d       | 3.54±0.03       | 6.40±0.06      | 4.30±0.02   | 5.27±0.03     |
| 5     | 5e       | 1.94±0.02       | 3.76±0.01      | 1.98±0.10   | 1.25±0.02     |
| 6     | 5f       | 11.84±0.06      | 13.40±0.02     | 10.35±0.02  | 10.32±0.01    |
| 7     | 5g       | 14.07±0.02      | 12.04±0.25     | 10.05±0.01  | 12.13±0.05    |
| 8     | 5h       | 4.22±0.12       | 8.04±0.06      | 6.25±0.01   | 3.45±0.02     |
| 9     | 5i       | 12.52±0.02      | 10.28±0.11     | 13.25±0.05  | 9.70±0.02     |
| 10    | 5j       | 11.24±0.06      | 12.04±0.01     | 8.65±0.02   | 12.40±0.02    |
| 11    | 5k       | 9.35±0.01       | 11.20±0.05     | 10.35±0.20  | 8.75±0.25     |
| 12    | 5l       | 6.12±0.04       | 8.22±0.07      | 10.52±0.01  | 10.25±0.04    |
| 13    | Etoposide| 2.58±0.25       | 5.74±0.37      | 2.03±0.12   | 2.61±0.32     |

\(^a\) Data represent as mean ± SEM values. Cytotoxicity as IC\(_{50}\) for each cell line, is the concentration of compound which reduced by 50% the optical density of treated cell with respect to untreated cells using the MTT assay.

\(^b\) Data represent as mean ± SEM values of these independent determinations.

2. General

All commercially available reagents were used without further purification. Reaction solvents were dried by standard methods before use. Purity of the compounds was checked by TLC using Merck 60F254 silica gel plates. Elemental analyses were obtained with an Elemental Analyser Perkin-Elmer 240C apparatus. \(^1\)H and \(^{13}\)C NMR spectra were recorded with a Bruker 400 spectrometer (operating at 400 & 500MHz for \(^1\)H and 100 MHz for \(^{13}\)C); chemical shifts were referenced to TMS. ESI-MS spectra were determined on a LCQ ion trap mass spectrometer (Thermo Fisher, San Jose, CA, USA), equipped with an ESI source. Melting points were recorded on a hot stage melting point apparatus Ernst Leitz Wetzlar, Germany and were uncorrected.
3. General synthetic procedures

3.1. General procedure for the synthesis of 4β-(prop-2-ynyloxy) epipodophyllotoxin (3)

Podophyllotoxin (1) (1 mmol) and propargyl alcohol (2) (1 mmol) was dissolved in dichloromethane (10 ml) and added slowly Boron trifloride etharate dropwise over a period of 10 minutes while maintaining the temperature at -20°C and stirring was continued for 2 hours at the same temperature. After conversion was complete, the mixture is quenched by addition of pyridine and diluting with dichloromethane (40 ml). The organic layer was separated and the aqueous layer extracted with dichloromethane (2 x 20 ml). The combined organic layers were washed consecutively with cold dilute HCl and brine solution. The extracted organic solution was dried (anhydrous Na₂SO₄) and evaporated under reduced pressure to afford a crude product which was subjected to column chromatography (silica gel, 60-120 mesh, eluent; n-hexane/EtOAc gradient) to afford pure product (3). Yield: 45%; White powder solid; mp: 152-154°C;

3.2. General procedure for the synthesis of Sugar Azides (4a-l)

Sugar azides were synthesised by the reported procedures in the literature based on Bertho’s method. Firstly, Sugars are acetylated adding an excess amount of acetic anhydride in anhydrous pyridine and the reaction mixture is allowed to stir overnight at room temperature. Concentrate the reaction mixture under reduced pressure and co-evaporate several times with Toluene to remove (traces of) Pyridine. Purify the residue with column chromatography with hexane:EtOAc (6:4). This should give you 95-97 % yield of your Alfa/beta product in 3:1 ratio. Pentacetylated sugars made to react with HBr in AcOH (33 %) at 0 °C. Then, stir the reaction mass at room temperature for 2 hours. The reaction mass was diluted with dichloromethane (40 ml) and water. The organic layer was separated and the aqueous layer extracted with dichloromethane (2 x 20 ml). The combined organic layers were dried (anhydrous Na₂SO₄) and evaporated under reduced pressure to afford a desired product glycosyl bromide in very high yield (~98 %). Pentacetylated glycosyl bromide was then refluxed for 2h at 100 °C with NaN₃ in the presence of PTC to yield corresponding glycosyl azides, which was subjected to column chromatography (silica gel, 60-120 mesh, eluent; n-hexane/EtOAc gradient) to afford pure products (4a-l).
3.3. General procedure for the synthesis of Podophyllotoxin-triazole hybrids (5a-l)

O-Propargylated podophyllotoxin (terminal alkyne) 3 (1 mmol) and Ag (I)-NHC (5 mol %) was dissolved in dry EtOH (10 ml) and added sugar azide (4a-l) (1 mmol). The reaction mass was stirred for 30 minutes. Complete consumption of starting material as judged by TLC and GC analysis. The reaction mass was evaporated under reduced pressure and diluting with dichloromethane (40 ml) and water. The organic layer was separated and the aqueous layer extracted with dichloromethane (2 x 20 ml). The combined organic layers were dried (anhydrous Na₂SO₄) and evaporated under reduced pressure to afford a crude product which was subjected to column chromatography (silica gel, 60-120 mesh, eluent; n-hexane/EtOAc gradient) to afford pure products (5a-l).

4. Spectral data of compound (5a-l).

4β-(1-Tetraacetyl-D-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5a)

Yield 94%; White solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.80 (s, 1H, CHN), 6.90 (s, 1H, B-ring-H), 6.58 (s, 1H, B-ring-H), 6.25 (s, 2H, E-ring-H), 6.02 (d, 1H, J=1.16Hz, anomerich-H), 6.00 (d, 1H, J=1.16Hz), 5.90 (dd, 1H, J=2.92 Hz, 3Hz), 5.44 (m, 2H, J=2.76 Hz, 3.24Hz), 5.26 (t, 1H, J=10.4Hz), 4.78 (d, 1H, J=10.48Hz), 4.73 (d, 1H, J=3.32Hz), 4.63 (d, 1H, J=5.36Hz), 4.37-4.27 (m, 3H), 4.20-4.16 (m, 1H), 4.05-4.01 (m, 1H), 3.81 (s, 3H), 3.74 (s, 6H), 3.42 (dd, 1H, J=5.34, 8.6Hz), 2.97 (s, 1H), 2.90 (s, 1H), 2.10 (s, 3H), 2.09 (s, 3H), 2.05 (s, 3H), 1.91 (s, 3H); 13C-NMR (CDCl₃, 100 MHz) δ 174.9, 170.5, 170, 169.4, 169 ,152.4, 148.5, 146.7, 145.3, 137, 135.3, 132.6, 121.1, 110.9, 109.6, 108, 101.5 (OCH2O), 77.3, 77, 76.7, 70.3, 67.5, 67.4, 62.4, 61.4, 60.7, 56.1, 43.9, 40.9, 38, 20.7, 20.6, 20.5, 20.1; HRMS (ESI) m/z calcd for C39H43N3NaO17[M+Na]⁺: 848.2490, found: 848.2488.

4β-(1-D-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5b)

Yield 85%; White solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.82 (s, 1H), 6.85 (s, 1H), 6.71 (s, 1H), 6.20 (s, 2H), 6.12 (d, 1H, J=1.16Hz), 6.00 (d, 1H, J=1.16Hz), 5.95 (dd, 1H, J=2.92 Hz, 3Hz), 5.40 (m, 2H, J=2.76 Hz, 3.24Hz), 5.30 (t, 1H, J=10.4Hz), 4.72 (d, 1H, J=10.48Hz), 4.68 (d, 1H, J=3.32Hz), 4.62 (d, 1H, J=5.36Hz), 4.40-4.28 (m, 3H), 4.21-4.14 (m, 1H), 4.08-4.02 (m, 1H), 3.75 (s, 3H), 3.80 (s, 6H), 3.40 (dd, 1H, J=5.34, 8.6Hz), 3.00 (s, 1H), 2.92 (s, 1H); 13C-
NMR (CDCl₃, 100 MHz) δ 174.9 (-CO-), 152.4, 148.5, 146.7, 145.3, 137, 135.3, 132.6, 121.1, 110.9, 109.6, 108, 101.5 (OCH₂O), 77.3, 77, 76.7, 70.3, 67.5, 67.4, 62.4, 61.4, 60.7, 56.1, 43.9, 40.9, 38; HRMS (ESI) m/z calcd for C₃₁H₃₅N₃NaO₁₃ [M+Na]+: 680.2068, found: 680.2059.

4β-(1-Tetraacetyl-D-C₂-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5c)

Yield 94%; White solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.82 (s, 1H), 6.91 (s, 1H), 6.58 (s, 1H), 6.38 (s, 2H), 6.10 (d, 1H, J=3.46 Hz), 6.00 (d, 1H, J=8 Hz), 5.94 (dd, 1H, J=2.92 Hz, 3.62 Hz), 5.61 (m, 2H, J=2.82 Hz, 8.13 Hz), 5.48 (t, 1H, J=10.1 Hz), 4.76 (d, 1H, J=8.48 Hz), 4.71 (d, 1H, J=4.34 Hz), 4.60 (d, 1H, J=5.36 Hz), 4.37-4.30 (m, 3H), 4.20-4.16 (m, 1H), 4.08-4.06 (m, 1H), 3.90 (s, 3H), 3.81 (s, 6H), 3.56 (dd, 1H, J=3.56, 8 Hz), 3.01 (s, 1H), 2.94 (s, 1H), 2.12 (s, 3H), 2.10 (s, 3H), 2.08 (s, 3H), 1.91 (s, 3H); 13C-NMR (CDCl₃, 100 MHz) δ 175.8, 170.3, 170, 169.1, 152.7, 148.4, 146.6, 145.6, 137.1, 135.8, 132.2, 121.4, 110.9, 109.8, 108.4, 101.6 (OCH₂O), 77.4, 77.3, 76.5, 70.3, 67.5, 67.1, 62.6, 61, 60.7, 56.4, 44, 41.2, 38.2, 20.7, 20.6, 20.4, 20.1; HRMS (ESI) m/z calcd for C₃₉H₄₃N₃NaO₁₇ [M+Na]+: 848.2490, found: 848.2489.

4β-(1-D-C₂-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5d)

Yield 85%; White solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.78 (s, 1H), 6.80 (s, 1H), 6.75 (s, 1H), 6.17 (s, 2H), 6.11 (d, 1H, J=2.16 Hz), 6.00 (d, 1H, J=5.24 Hz), 5.88 (dd, 1H, J=2.78 Hz, 4.63 Hz), 5.37 (m, 2H, J=2.23 Hz, 3.26 Hz), 5.27 (t, 1H, J=10 Hz), 4.68 (d, 1H, J=10.18 Hz), 4.66 (d, 1H, J=3.24 Hz), 4.60 (d, 1H, J=2.79 Hz), 4.40-4.30 (m, 3H), 4.18-4.13 (m, 1H), 4.08-4.00 (m, 1H), 3.73 (s, 3H), 3.70 (s, 6H), 3.41 (dd, 1H, J=5.31, 8.2 Hz), 3.02 (s, 1H), 2.90 (s, 1H); 13C-NMR (CDCl₃, 100 MHz) δ 175.3, 170.0, 169.1, 152.2, 148.5, 147, 145.1, 138, 135.2, 132.3, 121.3, 110.2, 109.2, 108.4, 101.3, 77, 76.9, 76.7, 70.3, 67.3, 67.2, 62, 61.4, 60.8, 56, 44.2, 41.2, 38.5; HRMS (ESI) m/z calcd for C₃₁H₃₅N₃NaO₁₃ [M+Na]+: 680.2068, found: 680.2059.

4β-(1-Tetraacetyl-D-galactosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5e)

Yield 91%; Grey solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.85 (s, 1H), 6.91 (s, 1H), 6.57 (s, 1H), 6.25 (s, 2H), 6.00 (d, 2H, J=12.52 Hz), 5.87 (d, 1H, J=9.24 Hz), 5.54 (dd, 2H, J=3.12, 11.84 Hz), 5.27 (dd, 1H, J=3.2 Hz, 7.08 Hz), 4.82-4.73 (m, 3H), 4.62 (d, 1H, J=5.24 Hz), 4.33-4.17 (m, 5H), 3.80-3.74 (m, 10H), 3.42 (dd, 1H, J=5.32, 8.6 Hz), 2.95-2.89 (m, 1H), 2.05 (s, 4H), 2.02 (s, 3H), 1.91 (s, 3H), 1.26 (s, 1H); 13C-NMR (CDCl₃, 100 MHz) δ 177.6, 175.9, 175.1, 170.0, 169.1, 152.5, 148.5, 135.3, 132.6, 121.2, 111, 109.6, 108.2, 101.5, 86.4, 77.2, 77, 76.7,
HRMS (ESI) m/z calcd for C39H43N3NaO17[M+Na]+: 848.2490, found: 848.2489.

4β-(1-D-galactosyl-4-methyl-1, 2, 3-triazolyl)epipodophyllotoxin (5f)
Yield 91%; Grey solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.80 (s, 1H), 6.89 (s, 1H), 6.61 (s, 1H), 6.15 (s, 2H), 6.05 (d, 2H, J=12.5 Hz), 5.90 (d, 1H, J=9.2 Hz), 5.55 (dd, 2H, J=11.8 Hz), 5.27 (dd, 1H, J=5.2 Hz), 4.62 (d, 1H, J=5.2 Hz), 4.37-4.25 (m, 5H), 3.80-3.74 (m, 10H), 3.42 (dd, 1H, J=5.3, 8.6 Hz); 13C-NMR (CDCl₃, 100 MHz) δ 177.6, 152.5, 148.5, 135.3, 132.6, 121.2, 111, 109.6, 108.2, 101.5, 86.4, 77.2, 77, 76.7, 74.2, 73.7, 70.6, 68.0, 67.4, 66.8, 61.1, 60.7, 56.2, 50.8, 43.9, 41.0, 38.1; HRMS (ESI) m/z calcd for C31H35N3NaO13 [M+Na]+: 680.2068, found: 680.2059.

4β-(1-Tetraacetyl-D-mannosyl-4-methyl-1, 2, 3-triazolyl)epipodophyllotoxin (5g)
Yield 94%; white solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.86 (s, 1H), 6.92 (s, 1H), 6.53 (s, 1H), 6.23 (s, 2H), 6.05 (d, 1H, J=2.14 Hz), 6.02 (d, 1H, J=3.27 Hz), 5.95 (dd, 1H, J=4.96 Hz), 5.48 (m, 2H, J=2.67 Hz), 5.37 (t, 1H, J=8.34 Hz), 4.86 (d, 1H, J=10.48 Hz), 4.80 (d, 1H, J=6.12 Hz), 4.56 (d, 1H, J=5.32 Hz), 4.35-4.27 (m, 3H), 4.20-4.18 (m, 1H), 4.05-4.03 (m, 1H), 3.86 (s, 3H), 3.62 (s, 6H), 3.38 (dd, 1H, J=8.62 Hz), 2.92 (s, 1H), 2.90 (s, 1H), 2.11 (s, 3H), 2.09 (s, 3H), 2.06 (s, 3H), 1.92 (s, 3H); 13C-NMR (CDCl₃, 100 MHz) δ 175.2, 172.5, 171, 168.8, 169.4, 152.4, 148.4, 146.8, 145.6, 137, 135.2, 132.8, 121.1, 110.9, 109.8, 108.6, 101.8, 77.3, 77, 76.8, 70.6, 67.5, 67.4, 63.8, 61.6, 60.8, 56.1, 44.1, 41.2, 38.8, 20.7, 20.6, 20.5, 20.3; HRMS (ESI) m/z calcd for C31H35N3NaO13 [M+Na]+: 680.2068, found: 680.2059.

4β-(1-D-mannosyl-4-methyl-1, 2, 3-triazolyl)epipodophyllotoxin (5h)
Yield 85%; White solid; 1H-NMR (CDCl₃, 400 MHz) δ 7.85 (s, 1H), 6.86 (s, 1H), 6.70 (s, 1H), 6.18 (s, 2H), 6.16 (d, 1H, J=1.12 Hz), 6.00 (d, 1H, J=3.24 Hz), 5.93 (dd, 1H, J=11.4 Hz), 3.16 Hz), 5.38 (m, 2H, J=2.82 Hz), 5.32 (t, 1H, J=6.20 Hz), 4.70 (d, 1H, J=6.84 Hz), 4.66 (d, 1H, J=11.4 Hz), 4.64 (d, 1H, J=5.36 Hz), 4.40-4.30 (m, 3H), 4.20-4.16 (m, 1H), 4.08-4.02 (m, 1H), 3.72 (s, 3H), 3.82 (s, 6H), 3.46 (dd, 1H, J=6.34, 2.46 Hz), 3.04 (s, 1H), 2.90 (s, 1H); 13C-NMR (CDCl₃, 100 MHz) δ 174.6, 152.2, 148.7, 147, 144.3, 137, 135.3, 133.4, 121.1, 110.9, 110.6, 108.6, 101.5, 77.3, 77, 76.4, 70.3, 67.8, 67.4, 62.4, 61.6, 60.8, 55.8, 43.7, 41.6, 38.6; HRMS (ESI) m/z calcd for C31H35N3NaO13 [M+Na]+: 680.2068, found: 680.2059.
$4\beta$-(1-triacetyl-D-arabinosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin(5i)

Yield 94%; Grey solid; 1H-NMR (CDCl$_3$, 400 MHz) $\delta$ 7.86 (s, 1H), 6.85 (s, 1H), 6.57 (s, 1H), 6.25 (s, 2H), 6.01-5.97 (m, 2H), 5.78 (d, 1H, $J$=9.12 Hz), 5.57 (t, 1H, $J$=9.28 Hz, 3Hz), 5.47 (s, 1H), 5.28 (dd, 1H, $J$=3.4, 6.76 Hz), 4.82 (d, 2H, $J$=11.8 Hz), 4.70 (d, 1H, $J$=3Hz), 4.63 (d, 1H, $J$=5.12Hz), 4.31 (d, 1H, $J$=6.96 Hz), 3.98 (d, 1H, $J$=13.44 Hz), 3.81 (d, 3H $J$=2.72 Hz), 3.75 (d, 5H $J$=4.84Hz), 3.47-3.42(m, 1H), 2.97-2.88(m, 1H), 2.24 (s, 3H), 2.06 (s, 3H), 1.92 (s, 3H), 1.27(s, 3H); 13C-NMR (CDCl$_3$, 100 MHz) $\delta$ 175.2, 170.4, 169.5, 169.2 , 151.6, 147.5, 145.7, 143.8, 137, 135.3, 133.3, 121.1, 111, 109.4, 108.2, 101.6, 78.3, 77.5, 76.7, 74.3, 68.4, 64.2, 61.9, 61.7, 55.1, 44.8, 42.2, 38.5, 20.8, 20.7, 20.5; HRMS (ESI) $m/z$ calcd for C36H40N3O15 [M+H]+: 754.2459, found: 754.2451.

$4\beta$-(1-D-arabinosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin(5j)

Yield 94%; Grey solid; 1H-NMR (CDCl$_3$, 400 MHz) $\delta$ 7.82 (s, 1H), 6.90 (s, 1H), 6.58 (s, 1H), 6.24 (s, 2H), 6.00-5.84 (m, 2H), 5.78 (d, 1H, $J$=9.12 Hz), 5.47 (t, 1H, $J$=4.2 Hz, 3Hz), 5.37 (s, 1H), 5.28 (dd, 1H, $J$=3.15, 6.74 Hz), 4.81 (d, 2H, $J$=9.2 Hz), 4.68 (d, 1H, $J$=3.12Hz), 4.63 (d, 1H, $J$=5.12Hz), 4.33 (d, 1H, $J$=6.00 Hz), 4.12 (d, 1H, $J$=11.27 Hz), 3.80 (d, 3H, $J$=2.34 Hz), 3.72(d, 5H $J$=4.82Hz), 3.32-3.23 (m, 1H), 3.02-2.88 (m, 1H), 2.23 (s, 3H); 13C-NMR (CDCl$_3$, 100 MHz) $\delta$ 174.8, 151.1, 146.3, 144.7, 142.2, 136.1, 135.3, 132.7, 120.3, 111.4, 109.5, 108.5, 101.9 , 77.8, 77.5, 76.9, 75, 69.3, 63.9, 61.9, 61.3, 53.7, 43.7, 41.5, 37.6, 36.5; HRMS (ESI) $m/z$ calcd for C30H33N3NaO12 [M+Na]+: 650.1962, found: 650.1954.

$4\beta$-(1-triacetyl-D-xylosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin(5k)

Yield 94%; White solid;1H-NMR (CDCl$_3$, 400 MHz) $\delta$ 7.82(s, 1H), 6.74 (s, 1H), 6.35 (s, 1H), 6.20 (s, 2H), 6.00-5.95 (m, 2H), 5.62 (d, 1H, $J$=5.14 Hz), 5.43 (t, 1H, $J$=7.12 Hz, 4.14Hz), 5.47 (s, 1H), 5.16 (dd, 1H, $J$=9.12, 6.58 Hz), 4.75 (d, 2H, $J$=11.8 Hz), 4.65 (d, 1H, $J$=9.12Hz), 4.53 (d, 1H, $J$=5.05 Hz), 4.25 (d, 1H, $J$=6.94 Hz), 4.24 (d, 1H $J$=8.62 Hz), 3.92 (d, 3H, $J$=5.14 Hz), 3.62 (d, 5H, $J$=3.14Hz), 3.52-3.44 (m, 1H), 3.27-2.95 (m, 1H), 2.25 (s, 3H), 2.07 (s, 3H), 1.98 (s, 3H), 1.26(s, 3H); 13C-NMR (CDCl$_3$, 100 MHz) $\delta$ 176.5, 171.4, 169.8, 169.4, 151.6, 148.8, 146.1, 144.3, 137.7, 135.5, 133.6, 122.2, 112, 110.7, 109.4, 102.2 , 79.1, 77.5, 76.5, 75.8, 69.8, 65.3, 62, 61.8, 56.4, 43.8, 42.5, 39.5, 20.9, 20.8, 20.6; HRMS (ESI) $m/z$ calcd for C36H40N3O15 [M+H]+: 754.2459, found: 754.2451.
**4β-(1-D-xylosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin (5l)**

Yield 94%; Grey solid; 1H-NMR (CDCl$_3$, 400 MHz) δ 7.80 (s, 1H), 6.87 (s, 1H), 6.46 (s, 1H), 6.18 (s, 2H), 6.10-5.92 (m, 2H), 5.70 (d, 1H, $J$=3.15 Hz), 5.46 (t, 1H, $J$=4.20 Hz, 3.12Hz), 5.28 (s, 1H), 5.20 (dd, 1H, $J$=3.12, 6.54 Hz), 4.84 (d, 2H, $J$=9.22 Hz), 4.62 (d, 1H, $J$=3.27 Hz), 4.58 (d, 1H, $J$=8.14 Hz), 4.33 (d, 1H, $J$=9.24 Hz), 4.25 (d, 1H, $J$=11 Hz), 3.78 (d, 3H, $J$=2.54 Hz), 3.72(d, 5H $J$=5 Hz), 3.30-3.20 (m, 1H), 3.00-2.86 (m, 1H), 2.43 (s, 3H); 13C-NMR (CDCl$_3$, 100 MHz) δ 174.2, 150.7, 146.5, 144.4, 141.8, 136, 135.6, 132.2, 120, 111.2, 109.4, 108.4, 102.3, 77.3, 77.5, 76.4, 75.2, 69.4, 63.6, 61.7, 61, 53.5, 43.6, 41.2, 37.2, 36.4; HRMS (ESI) m/z calcd for C$_{30}$H$_{33}$N$_3$NaO$_{12}$ [M+Na]$^+$: 650.1962, found: 650.1954.

5. **in vitro Cytotoxicity assay**

An in vitro cytotoxicity study was performed by adopting MTT assay. In this assay the yellow tetrazolium MTT (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) is reduced by metabolically active cells by the action of dehydrogenase enzymes. The resulting intracellular purple formazan can be solubilised and quantified by spectrophotometric means. The MTT reagent yields low background absorbance values in the absence of cells. For each cell type the linear relationship between cell number and signal produced is established, thus allowing an accurate quantification of changes in the rate of cell proliferation.

6. **Molecular Docking Studies**

Auto Dock software is used to dock the 5a & 5e with DNA Topo-II isomerase enzyme (PBD ID 1ZXN). The 3D structures of the compounds were optimised prior to docking with the enzyme and obtained significant docking scores with free energy -9.2 & -9.3 for the compounds respectively.

*Figure S2. 2D-Docking poses of 5a with Topoisomerase II enzyme*
Figure S3. 3D-Docking poses of 5a with Topoisomerase II enzyme

Figure S4. 3D-Ligand (5a) interactions with receptor

Figure S5. 2D-Docking poses of 5e with Topoisomerase II enzyme
Figure S6. 3D-Docking poses of 5e with Topoisomerase II enzyme

Figure S7. 3D-Ligand (5e) interactions with receptor
7. Representative spectra of key compounds:

*Figure S8. $^1$H-NMR spectra of 4β-(prop-2-ynyloxy) epipodophyllotoxin (3)*

*Figure S9. $^{13}$C-NMR spectra of 4β-(prop-2-ynyloxy) epipodophyllotoxin (3)*
Figure S10. $^1$H-NMR spectra of 4β-(1-Tetraacetyl-D-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5a)

Figure S11. $^{13}$C-NMR spectra of 4β-(1-Tetraacetyl-D-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5a)
Figure S12. $^1$H-NMR spectra of $4\beta$-(1-Tetraacetyl-D-galactosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5e)

Figure S13. $^{13}$C-NMR spectra of $4\beta$-(1-Tetraacetyl-D-galactosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5e)
Figure S14. $^1$HNMR spectra of 4β-(1-triacetyl-D-arabinosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin(5i)
Figure S15. HRMS spectra of 4β-(1-Tetraacetyl-D-glucosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5a)
Figure S16. HRMS spectra of 4β-(1-Tetraacetyl-D-galactosyl-4-methyl-1, 2, 3-triazolyl) epipodophyllotoxin (5e)
Figure S17. HRMS spectra of 4β-(1-triacetyl-D-arabinosyl-4-methyl-1,2,3-triazolyl)epipodophyllotoxin(5i)