

Proceeding Paper

Synthesis and Cytotoxic Activity of Conjugates of Mitochondrial-Directed Cationic Compound F16 with Ursane Structure Triterpenic Acids Containing a Polyhydroxylated A-ring[†]

Anna Spivak^{*}, Darya Nedopekina^{*} and Eldar Davletshin

Institute of Petrochemistry and Catalysis, Russian Academy of Sciences, 141 Prospekt Oktyabrya, Ufa 450075, Russia; eldarik1996@mail.ru (E.D.)

^{*} Correspondence: spivak.ink@gmail.com (A.S.); rawbe2007@mail.ru (D.N.); Tel.: +7-9174217106 (A.S.); +7-9173565551 (D.N.)

[†] Presented at the 27th International Electronic Conference on Synthetic Organic Chemistry, 15–30 November 2023; Available online: <https://ecsoc-27.sciforum.net/>.

Abstract: We have chemically linked corosolic and asiatic acids and a synthetic polyoxygenated analogue of ursolic acid via an alkyl linker to the cationic mitochondrial targeting compound F16 (4-(1H-indole-3-ylvinyl)-N-methylpyridinium iodide). The conjugates were tested for cytotoxic activity against two human lung adenocarcinoma cell lines, H1299 and A549, and non-cancerous mouse embryonic fibroblast cells. The results showed that conjugation of polyoxygenated triterpene acids with the terminal cationic fragment F16 in the C-28 side chain enhanced cytotoxicity (30–35 fold) compared to the original natural ursolic acid. However, the presence of hydroxyl or acetyl functions in the A-ring of F16 conjugates of corosolic or asiatic acids resulted in a significant decrease in cytotoxicity compared to their structural analogue, the F16 derivative of ursolic acid.

Keywords: ursolic acid; conjugates; polyhydroxylated ring A; delocalized lipophilic cations; F16; anti-cancer agents

Citation: Spivak, A.; Nedopekina, D.; Davletshin, E. Synthesis and Cytotoxic Activity of Conjugates of Mitochondrial-Directed Cationic Compound F16 with Ursane Structure Triterpenic Acids Containing a Polyhydroxylated A-ring. *Chem. Proc.* **2023**, *14*, x. <https://doi.org/10.3390/xxxxx>

Academic Editor: Firstname Last-name

Published: 15 November 2023



Copyright: © 2023 by the authors. Submitted for possible open access publication under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Pentacyclic triterpenoids of the ursane series, including ursolic acid (3 β -hydroxy-urs-12-en-28-oic acid) and its oxygenated structural analogues, corosolic and asiatic acids, are found in many medicinal plants and various fruits, berries, and vegetables (Figure 1) [1–3].

These phytochemicals possess a broad spectrum of beneficial biological properties, particularly their multifunctional anti-cancer activity and their capacity to trigger the mitochondrial pathway of apoptosis in a range of tumor cell types [4–9]. Furthermore, as an integral component of the diet, these triterpenoids could offer preventive measures against human oncological diseases. However, their high hydrophobicity limits the penetration of triterpenic acids through cell membranes, inhibiting their ability to reach the intended target and exhibit the necessary therapeutic effects in animal models. To overcome this, extensive research has been conducted over the last several decades focusing on the chemical modification of triterpenic acids, which unfortunately, does not always yield the expected results [5,6]. For instance, their characteristic anti-tumor activity might disappear, or there might be a substantial increase in their toxicity to normal cells. Recently, to enhance the biological potential and bioavailability of pentacyclic triterpenes (such as betulin, betulinic and ursolic acids), we synthesized their derivatives that contain, at the end of the C-28 side chain, the fragment of the membranotropic delocalized cationic

compound 4-(1-*H*-indol-3-ylvinyl)-*N*-methylpyridinium iodide (F16) [10]. This small cationic molecule is known for its ability to selectively accumulate in the mitochondrial matrix of various tumor cell lines [11]. These hybrid molecules demonstrated a significant synergistic enhancement of anti-tumor action against different human tumor cell lines (U937, Jurkat, K562). On the other hand, the data available in the literature today on the biological activity of oxygenated triterpenoids indicate that the configuration and quantity of hydroxyl or acetyl groups in the triterpenoid core of ursolic, corosolic and asiatic acid derivatives can significantly influence the biological activity and selectivity of these compounds concerning the tested cell lines [12–16]. For instance, natural triterpenoids with a polyhydroxylated A-ring, isolated from the plant *Euphorbia sieboldiana*, demonstrated potent anti-cancer effects on HeLa cells through the generation of active oxygen forms and blocking of the NF- κ B signaling pathway [12]. Introducing two hydroxyl groups at positions C-1 and C-2 of the ursolic acid triterpenoid core significantly amplified its antibacterial activity and altered the mechanism of antibacterial action of these triterpenoids [13]. Benzylamide derivatives of asiatic acid have demonstrated potent anticancer activity against the HL-60 cell line ($IC_{50} = 0.47 \mu\text{M}$) while exhibiting no cytotoxicity against normal human endothelial HUVEC cells [14]. Among a large series of conjugates of pentacyclic triterpenoids with rhodamine B, asiatic acid has emerged as a leading structure for the development of anticancer, mitochondria-targeted agents, displaying high cytotoxic activity coupled with high selectivity and the ability to overcome drug resistance. The potential of asiatic acid derivatives was further confirmed in preclinical models of human tumors [15,16].

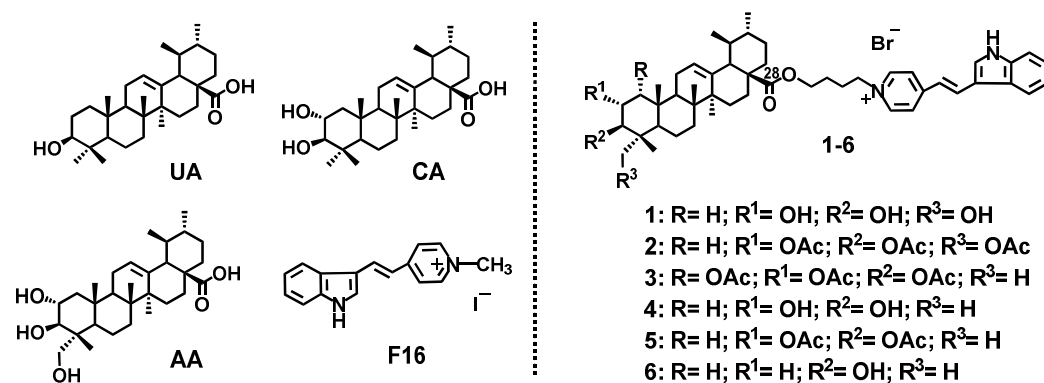


Figure 1. Molecular structures of ursolic acid (UA), corosolic acid (CA), asiatic acid (AA), F16 ([E-4-(1H-indol-3-ylvinyl)-*N*-methylpyridinium iodide]) and of F16-triterpenoid conjugates (1–6).

Given these promising results, it was of interest to synthesize and explore the anti-cancer effects of F16 derivatives of asiatic acid **1** and **2** and the tri-hydroxylated analog of ursolic acid **3**. The cytotoxicity of these hybrid compounds was tested by us on the lung adenocarcinoma cell lines A549 and H1299 and benign mouse embryonic fibroblasts MEFs, compared with the previously described F16 derivatives of ursolic and corosolic acids **4–6**. Ursolic acid was used as a reference compound.

2. Materials and Methods

2.1. Chemistry

Ursolic, oleanolic and asiatic acids were purchased from Acros Organics (Geel, Belgium). Corosolic acid was obtained from ursolic acid as previously described followed by chromatographic purification of crude product [17]. Acetates of asiatic and corosolic acids were easily synthesized employing the well-established method [18]. Polyhydroxy derivative of ursolic acid **12** was synthesized according to method [13].

The bromoalkyl esters **8**, **9**, **13**, **17**, **18** and conjugates **1–5** were synthesised as previously described [19,20]. The physico-chemical and spectral characteristics of the compounds were in full agreement with the data presented in the [20].

General Procedure for the Synthesis of the Conjugates **1–5**

A solution of the corresponding triterpenoic acid (488.7 mg, 1.0 mmol), K_2CO_3 (138 mg, 1.0 mmol) and 1,4-dibromobutane (0.48 mL, 4.0 mmol) in dry DMF- CH_3CN (3:1, 8 mL) was stirred at 50 °C for 3 h. The reaction mixture was poured into water (40 mL) and extracted with $CHCl_3$. The organic layer was washed with H_2O and dried over $MgSO_4$. The residue after evaporation of the solvent in vacuo was subjected to column chromatography (SiO_2 , hexane/EtOAc 30:1 \rightarrow 1:1) to yield bromoalkyl esters **8**, **9**, **13**, **17** and **18** each as a white powders. Yield: 68–88%.

The resulting compounds **8**, **9**, **13**, **17** and **18** were reacted with 4-(1H-indole-3-ylvinyl)-N-methylpyridine by refluxing in DMF at 85 °C in an argon atmosphere for 12 h. The mixture was cooled to room temperature and evaporated under reduced pressure. The residue was purified by column chromatography on silica gel, using $CH_2Cl_2/MeOH$ 30:1 \rightarrow 10:1, to give pure compounds **1–5** as a brown powders. Yield: 78–82%.

2.2. Biology

2.2.1. Cell Lines and Culture Conditions

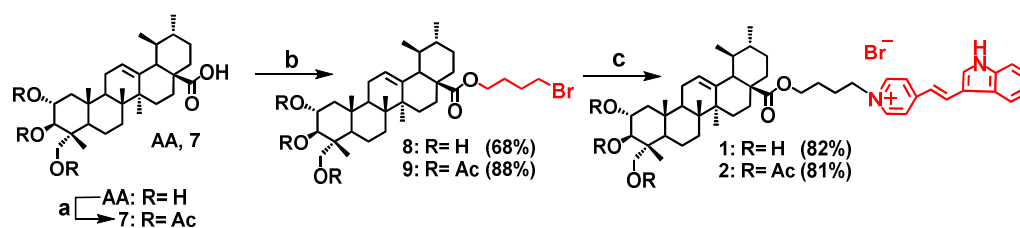
All cell lines used in this study (human non-small lung adenocarcinoma—H1299 and A549, mouse embryonic fibroblasts—MEFs) were purchased from ATCC. Cells were grown in DMEM media supplemented with 10% fetal bovine serum, 100 μ g/mL gentamycin, and 2 mM of glutamine at 37 °C in 5% CO_2 atmosphere.

2.2.2. Cytotoxic Assay of Conjugates (MTT Assay)

To carry out MTT assay, 3500 cells per well were planted overnight in 96-well plates. A day after the seeding, conjugates **1–6** dissolved in DMSO were added in the different concentrations. DMSO was used as a control. 48 h after, 10 μ L of 5 mg/mL Thiazolyl Blue (Paneko, Russia) solution was added to each well and cells were kept for 3 h at 37 °C in CO_2 incubator. After removing the thiazol-containing medium, 150 μ L of isopropyl alcohol (supplemented with 40 mM HCl and 0.1% NP-40) was added to dissolve the MTT-formazan salt. The absorbance at 570 and 630 nm (reference) was measured using BioRad iMark microplate reader (BioRad, United States). Three biological replicates were used for experiment. Results were processed by Excel software. IC_{50} values were calculated by using AAT Bioquest online calculator (<https://www.aatbio.com/tools/ic50-calculator>) and are represented as the mean \pm SD.

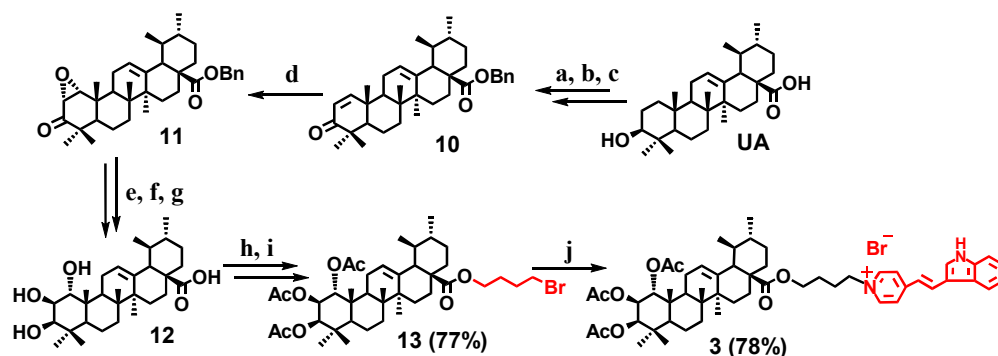
3. Results and Discussion

F16 conjugates with asiatic acid **1** and **2** were synthesized from available asiatic acid, which was transformed into acetate **7**. Next, triacetate **7** and the starting asiatic acid were converted into bromoalkyl esters **8** and **9** in an alkaline medium using a fourfold molar excess of 1,4-dibromobutane. The reaction of esters **8** and **9** with (E)-4-(1H-indol-3-ylvinyl)pyridine in DMF at 85 °C gave conjugates **1** and **2** in 81–82% yield after purification by column chromatography on silica gel (Scheme 1).



Scheme 1. Synthesis of conjugates **1** and **2**: **a** AcCl, THF, pyridine, DMAP, rt; **b** 1,4-dibromobutane, K₂CO₃, CH₃CN, DMF, 50 °C; **c** (E)-4-(2-(1*H*-indol-3-yl)vinyl)pyridine, DMF, 85 °C, 12 h.

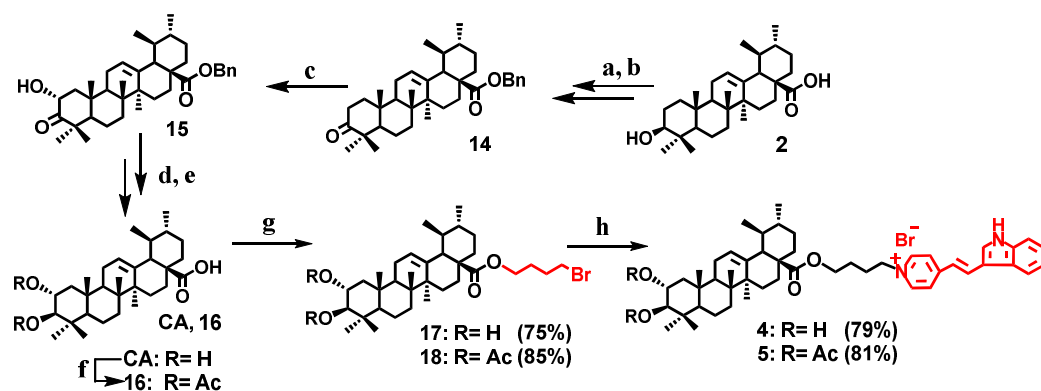
The starting compound for the synthesis of hybrid molecule **3** was triterpenic acid **12** with a tri-hydroxylated A ring, which was obtained from available ursolic acid according to a previously developed multistage scheme (Scheme 2) [13].



Scheme 2. Synthesis of conjugate **3**: **a** CrO₃, H₂SO₄, acetone, 0 °C; **b** BnCl, K₂CO₃, DMF, 50 °C, 2 h; **c** PhSeCl, EtOAc, *m*CPBA, pyridine, rt; **d** 30 % H₂O₂, 10% NaOH, MeOH, rt; **e** NaBH₄, MeOH–THF, rt; **f** HClO₄, H₂O, acetone, rt; **g** Pd–C/10%, H₂, MeOH–THF, rt; **h** AcCl, THF, pyridine, DMAP, rt; **i** 1,4-dibromobutane, K₂CO₃, CH₃CN, DMF, 50 °C; **j** (E)-4-(2-(1*H*-indol-3-yl)vinyl)pyridine, DMF, 85 °C, 12 h.

The synthesis involved the production of ursolic acid derivative **10**, containing an enone fragment in the A ring, and stereoselective epoxidation of the enone double bond using 30% H₂O₂ in an alkaline medium, resulting in 1 α ,2 α -epoxyketone **11** as the only product. Interaction of epoxyketone **11** with NaBH₄, stereoselective opening of the epoxy ring using HClO₄ in acetone, and subsequent removal of the benzyl protection yielded a polyoxygenated analog of ursolic acid **12** with a 1 α ,2 β ,3 β -trihydroxylated A ring. Further modification of acid **12** involved acetylation of hydroxyl groups, etherification of the carboxyl function using an excess of 1,4-dibromobutane in a K₂CO₃/DMF solution, and interaction of the bromoalkyl ether **13** with the neutral **F16** precursor (Scheme 2).

The corosolic acid used in the synthesis of conjugates **4** and **5** was derived from ursolic acid using a method based on the stereoselective electrophilic attack of *m*-chloroperbenzoic acid in the presence of H₂SO₄ on the C-2 atom of the benzyl ester of ursonic acid **14** (Scheme 3) [17].



Scheme 3. Synthesis of conjugates **4** and **5**: **a** CrO₃, H₂SO₄, acetone, 0 °C; **b** BnCl, K₂CO₃, DMF, 50 °C, 2 h; **c** *m*CPBA, H₂SO₄, MeOH–CH₂Cl₂, 0 °C; **d** NaBH₄, MeOH–THF, rt; **e** Pd–C/10%, H₂, MeOH–THF, rt; **f** AcCl, THF, pyridine, DMAP, rt; **g** 1,4-dibromobutane, K₂CO₃, CH₃CN, DMF, 50 °C; **h** (E)-4-(2-(1*H*-indol-3-yl)vinyl)pyridine, DMF, 85 °C, 12 h.

The reduction of the 3-keto function in 2 α -hydroxyketone **15** with NaBH₄ predominantly resulted in the desired 2 α ,3 β -diol, which, after chromatographic isolation on a column with SiO₂ and the removal of benzyl protection, yielded the required corosolic acid. Further transformations of corosolic acid and its acyl derivative **16** into bromoalkyl esters **17** and **18** and the synthesis of conjugates **4** and **5** were conducted under conditions previously described for compounds **1–3**.

The structures of all compounds were determined using IR, ¹H NMR, ¹³C NMR, and MS spectra. The physicochemical and spectral characteristics of asiatic, corosolic, and polyhydroxylated analogues of ursolic acid, as well as their acetates, conformed to literature data [13,17,18,20]. Moreover, the relative configuration of the epoxy ring and the orientation of hydroxyl functions in ring A in compounds **10–13** and **15–18** were conformed using DEPT, HMBC, HSQC, COSY and NOESY experiments.

The synthesized conjugates **1–5** were examined for cytotoxic activity against two human lung adenocarcinoma cell lines H1299 and A549 and non-cancerous MEFs (mouse embryonic fibroblasts). The cytotoxicity of the previously synthesized ursolic acid **F16**-derivative **6** was also examined on these cell lines for comparison (Table 1) [19].

Table 1. Cytotoxic activity of **F16** conjugates with triterpenoids **1–6** against human lung adenocarcinoma H1299 and A549 and non-cancerous mouse fibroblasts MEFs: IC₅₀ values from MTT tests after 48-h treatment are given in μ M.

No	H1299	A549	MEF	SI ¹	SI ²
1	13.51 \pm 1.81	9.03 \pm 0.59	20.04 \pm 6.73	1.48	2.21
2	8.51 \pm 2.05	3.80 \pm 1.86	5.85 \pm 0.22	0.68	1.54
3	3.57 \pm 0.20	4.81 \pm 2.15	3.79 \pm 0.32	1.06	0.79
4	4.01 \pm 1.69	1.87 \pm 0.14	6.76 \pm 1.58	1.69	3.62
5	3.58 \pm 0.30	3.04 \pm 0.34	5.79 \pm 0.49	1.62	1.91
6	2.80 \pm 0.25	2.40 \pm 0.30	n.d.	–	–
UA	97.60 \pm 5.63	68.93 \pm 19.63	50.39 \pm 16.92	0.51	0.74

^{1,2}The selectivity indices (SI) are defined as: SI = IC₅₀ (MEF)/IC₅₀ H1299 or SI = IC₅₀ (MEF)/IC₅₀ A549.

From the results presented in the table, it is evident that introducing a terminal cationic fragment into the C-28 side chain of the examined triterpenic acids resulted in significant enhancement of cytotoxicity compared to ursolic acid, regardless of the presence of acetylated or free hydroxyl groups in ring A. In particular, the **F16** conjugate with corosolic acid **4** was 24 and 36 times more effective than the original ursolic acid against the A549 and H1299 tumor cell lines, respectively, while the **F16**-derivative of acetylated corosolic acid **5** exceeded the anti-tumor effect of ursolic acid by 23–27 times. However, the additional presence of hydroxyl or acetyl functions in ring A led to some reduction in the cytotoxic effect compared to the **F16** derivative of ursolic acid, contrary to known literature facts [14–16]. For instance, regarding the H1299 tumor cell line, the cytotoxicity IC₅₀ values for conjugates **1**, **4**, and **6** were respectively 13.51 μ M, 4.01 μ M, and 2.80 μ M. Natural ursolic acid and the studied conjugates did not show selectivity against tumor cells and mouse fibroblasts except for the **F16** derivative of corosolic acid **4**, which demonstrated a selectivity difference between tumor and healthy cells with a selectivity index in the range of 1.7–3.6.

4. Conclusions

In the present work, we have demonstrated the cytotoxic effects of conjugates of the membrane-penetrating cation **F16** with several polyoxygenated triterpenoids of the ursane structure on lung adenocarcinoma tumor lines A549 and H1299 and on healthy mouse fibroblast cells MEF. All synthesized conjugates surpassed natural ursolic acid in cytotoxic action by many times. Among the hybrid compounds, the **F16**-derivative of

corosolic acid, along with a high antitumor effect, showed a selectivity index between tumor and healthy cells in the range of 1.7–3.6.

Supplementary Materials: The following are available online at www.mdpi.com/xxx/s1.

Author Contributions: Validation and writing—review and editing, A.S.; performing the chemistry experiments, D.N. and E.D. The manuscript was prepared through the contributions of D.N. and E.D. All authors have read and agreed to the published version of the manuscript.

Acknowledgments: The researches was carried out in accordance with federal program №FMRS-2022-0081.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. He, X.; Liu, R.H. Triterpenoids isolated from apple peels have potent antiproliferative activity and may be partially responsible for apple's anticancer activity. *J. Agric. Food Chem.* **2007**, *55*, 4366–4370. <https://doi.org/10.1021/jf063563o>.
2. Woźniak, L.; Szakiel, A.; Glowacka, A.; Rozpara, E.; Marszałek, K.; Skapska, S. Triterpenoids of three apple cultivars—biosynthesis, antioxidative and anti-inflammatory properties, and fate during processing. *Molecules* **2023**, *28*, 2584. <https://doi.org/10.3390/molecules28062584>.
3. Neto, C.C.; Ursolic acid and other pentacyclic triterpenoids: Anticancer activities and occurrence in berries. In *Berries and Cancer Prevention*; Stoner, G.D., Seeram, N.P., Eds.; Springer Science+Business Media: New York, NY, USA, 2011; pp. 41–49. https://doi.org/10.1007/978-1-4419-7554-6_2.
4. Dzubak, P.; Hajduch, M.; Vydra, D.; Hustova, A.; Kvasnica, M.; Biedermann, D.; Markova, L.; Urbanc, M.; Sarek, J. Pharmacological activities of natural triterpenoids and their therapeutic implications. *Nat. Prod. Rep.* **2006**, *23*, 394–411. <https://doi.org/10.1039/b515312n>.
5. Hussain, H.; Green, I.R.; Ali, I.; Khan, I.A.; Ali, Z.; Al-Sadi, A.M.; Ahmed, I. Ursolic acid derivatives for pharmaceutical use: A patent review (2012–2016). *Expert. Opin. Ther. Pat.* **2017**, *27*, 1061–1072. <https://doi.org/10.1080/13543776.2017.1344219>.
6. Chen, H.; Gao, Y.; Wang, A.; Zhou, X.; Zheng, Y.; Zhou, J. Evolution in medicinal chemistry of ursolic acid derivatives as anti-cancer agents. *Eur. J. Med. Chem.* **2015**, *92*, 648–655. <https://doi.org/10.1016/j.ejmech.2015.01.031>.
7. Shanmugam, M.K.; Dai, X.; Kumar, A.P.; Tan, B.K.; Sethi, G.; Bishayee, A. Ursolic acid in cancer prevention and treatment: Molecular targets, pharmacokinetics and clinical studies. *Biochem. Pharmacol.* **2013**, *85*, 1579–1587. <https://doi.org/10.1016/j.bcp.2013.03.006>.
8. Loc, T.V.; Quynh, N.V.T.; Chien, T.V.; Phuong, T.T.T.; Ninh, P.T.; Thanh, N.T.; Thu, H.L.T.; Nga, T.N.; Thao, D.T.; Sung, T.V. Synthesis of asiatic acid derivatives and their cytotoxic activity. *Med. Chem. Res.* **2018**, *27*, 1609–1023. <https://doi.org/10.1007/s00044-018-2176-y>.
9. Xu, Y.; Ge, R.; Du, J.; Xin, H.; Yi, T.; Sheng, J.; Wang, Y.; Ling, C. Corosolic acid induces apoptosis through mitochondrial pathway and caspases activation in human cervix adenocarcinoma HeLa cells. *Cancer Lett.* **2009**, *284*, 229–237. <https://doi.org/10.1016/j.canlet.2009.04.028>.
10. Spivak, A.Yu.; Nedopekina, D.A.; Gubaidullin, R.R.; Davletshin, E.V.; Tukhbatullin, A.A.; D'yakonov, V.A.; Yunusbaeva, M.M.; Dzhemileva, L.U.; Dzhemilev, U.M. Pentacyclic triterpene acid conjugated with mitochondria-targeting cation F16: Synthesis and evaluation of cytotoxic activities. *Med. Chem. Res.* **2021**, *30*, 940–951. <https://doi.org/10.1007/s00044-021-02702-z>.
11. Fantin, V.R.; Berardi, M.J.; Scorrano, L.; Korsmeyer, S.J.; Leder, P. A novel mitochondriotoxic small molecule that selectively inhibits tumor cell growth. *Cancer Cell* **2002**, *2*, 29–42. [https://doi.org/10.1016/s1535-6108\(02\)00082-x](https://doi.org/10.1016/s1535-6108(02)00082-x).
12. Huang, R.-Z.; Jin, L.; Wang, C.-G.; Xu, X.-J.; Du, Y.; Liao, N.; Ji, M.; Liao, Z.X.; Wang, H.-S. A pentacyclic triterpene derivative possessing polyhydroxyl ring A suppresses growth of HeLa cells by reactive oxygen species-dependent NF- κ B pathway. *Eur. J. Pharmacol.* **2018**, *838*, 157–169. <https://doi.org/10.1016/j.ejphar.2018.08.032>.
13. Huang, L.; Luo, H.; Li, Q.; Wang, D.; Zhang, J.; Hao, X.; Yang, X. Pentacyclic triterpene derivatives possessing polyhydroxyl ring A inhibit Gram-positive bacteria growth by regulating metabolism and virulence genes expression. *Eur. J. Med. Chem.* **2015**, *95*, 64–75. <https://doi.org/10.1016/j.ejmech.2015.01.015>.
14. Lu, Y.-H.; Chen, M.-C.; Liu, F.; Xu, Z.; Tian, X.-T.; Xie, Y.; Huang, C.-G. Synthesis and cytotoxic activity of novel C-23-modified asiatic acid derivatives. *Molecules* **2020**, *25*, 3709. <https://doi.org/10.3390/molecules25163709>.
15. Kraft, O.; Hartmann, A.-K.; Brandt, S.; Hoenke, S.; Heise, N.V.; Csuk, R.; Mueller, T. Asiatic acid as a leading structure for derivatives combining sub-nanomolar cytotoxicity, high selectivity, and the ability to overcome drug resistance in human pre-clinical tumor models. *Eur. J. Med. Chem.* **2023**, *250*, 115189. <https://doi.org/10.1016/j.ejmech.2023.115189>.
16. Heise, N.; Becker, S.; Mueller, T.; Bache, M.; Csuk, R.; Guttler, A. Mitochondria-targeting 1,5-diazacyclooctane-spacer triterpene rhodamine conjugates exhibit cytotoxicity at sub-nanomolar concentration against breast cancer cells. *Int. J. Mol. Sci.* **2023**, *24*, 10695. <https://doi.org/10.3390/ijms241310695>.
17. Nelson, A.T.; Camelio, A.M.; Claussen, K.R.; Cho, J.; Tremmel, L.; DiGiovanni, J.; Siegel, D. Synthesis of oxygenated oleanolic and ursolic acid derivatives with anti-inflammatory properties. *Bioorg Med. Chem. Lett.* **2015**, *25*, 4342–2346. <https://doi.org/10.1016/j.bmcl.2015.07.029>.

18. Van, L.T.; Thi, Q.N.V.; Van, C.T.; Thi, P.T.T.; Thi, N.P.; Tuan, T.N.; Thi, T.H.L.; Thi, N.N.; Thi, T.D.; Van, S.T. Synthesis of asiatic acid derivatives and their cytotoxic activity. *Med. Chem. Res.* **2018**, *27*, 1609–1623. <https://doi.org/10.1007/s00044-018-2176-y>.
19. Dubinin, M.V.; Nedopekina, D.A.; Ilzorkina, A.I.; Semenova, A.A.; Sharapov, V.A.; Davletshin, E.V.; Mikina, N.V.; Belsky, Y.P.; Spivak, A.Y.; Akatov, V.S.; et al. Conjugation of triterpenic acids of ursane and oleanane types with mitochondria-targeting cation F16 synergistically enhanced their cytotoxicity against tumor cells. *Membranes* **2023**, *13*, 563. <https://doi.org/10.3390/membranes13060563>.
20. Spivak, A.Y.; Nedopekina, D.A.; Davletshin, E.V.; Shuvalov, O.I.; Kirdeeva, Y.N.; Belsky, Y.P. Synthesis and cytotoxic evaluation of F16 derivatives pentacyclic triterpenoid acids possessing a polyhydroxylated ring A. *Preprint* **2022**. <https://doi.org/10.21203/rs.3.rs-1827300/v1>.

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.