

Communication

# Synthesis of 6-Methyluracilpentylviologen Resorcinarene Cavitand

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**Abstract:** Resorcinarenes, as macrocyclic compounds, are widely used to recognize substrates and create supramolecular assemblies. Their bowl-like form organizes functional groups at the upper and lower rims, which has a substantial impact on the molecular recognition of various substrates. As a result, resorcinarenes make good drug nanocarrier candidates. This paper presents the synthesis of a new resorcinarene cavitand functionalized along the upper rim with methyluracil and viologen fragments for its potential use in drug delivery. Methyluracils and viologens are well-known receptor-targeted compounds capable of facilitating the vector transfer of drugs and increasing the effectiveness of their action on cells. The paper describes the synthesis of resorcinarene modified with methyluracil and viologen groups along with its structure determined by <sup>1</sup>H-, <sup>13</sup>C-NMR and IR spectroscopy.

**Keywords:** resorcinarene; viologen; methyluracil; nanocarriers; targeted delivery



**Citation:** Ziganshina, A.Y.; Mansurova, E.E.; Shulaeva, M.M.; Syakaev, V.V.; Semenov, V.E.; Antipin, I.S. Synthesis of 6-Methyluracilpentylviologen Resorcinarene Cavitand. *Molbank* **2022**, *2022*, M1507. <https://doi.org/10.3390/M1507>

Academic Editor: R. Alan Aitken

Received: 29 October 2022

Accepted: 23 November 2022

Published: 25 November 2022

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## 1. Introduction

One of the emerging areas in medicinal chemistry is the development of vector nanocarriers for drug delivery [1,2]. The main feature of nanocarriers is their ability to encapsulate drugs for delivery to biotargets [3]. For this purpose, amphiphilic nanocarriers are being developed to increase the circulation time of drugs in the body, improve their penetration through tissues, and to accumulate in damaged areas [4,5].

There are several well-known types of macrocyclic compounds used as a platform for the synthesis of vector nanosystems [6–9]. The actively researched representatives in this field are resorcinarenes. Based on these macrocycles, various nanocontainers and nanocarriers have been created due to unique properties such as the presence of an aromatic cavity, the formation of “guest–host” complexes, as well as the possibility of modifying the upper and lower rims [10,11]. The use of specific functional groups makes it possible to change some properties, such as solubility in water, bioavailability, biological activity, and the ability to self-assemble [12].

Nanocarriers with viologen fragments have shown promise in early investigations for medication delivery. Dendrimers with viologens were shown to have an antiviral effect against the human immunodeficiency virus (HIV) [13]. These dendrimers function as polycationic ligands for the CXCR4 chemokine receptor and are capable of delivering genes while being simultaneously antagonistic to CXCR4 [14,15]. However, viologens cytotoxicity, interaction with the donor pieces of the cells, and formation of reactive oxygen species are their drawbacks. The introduction of additional receptor-directed groups into the nanocarrier base in addition to viologens could solve the toxicity problem.

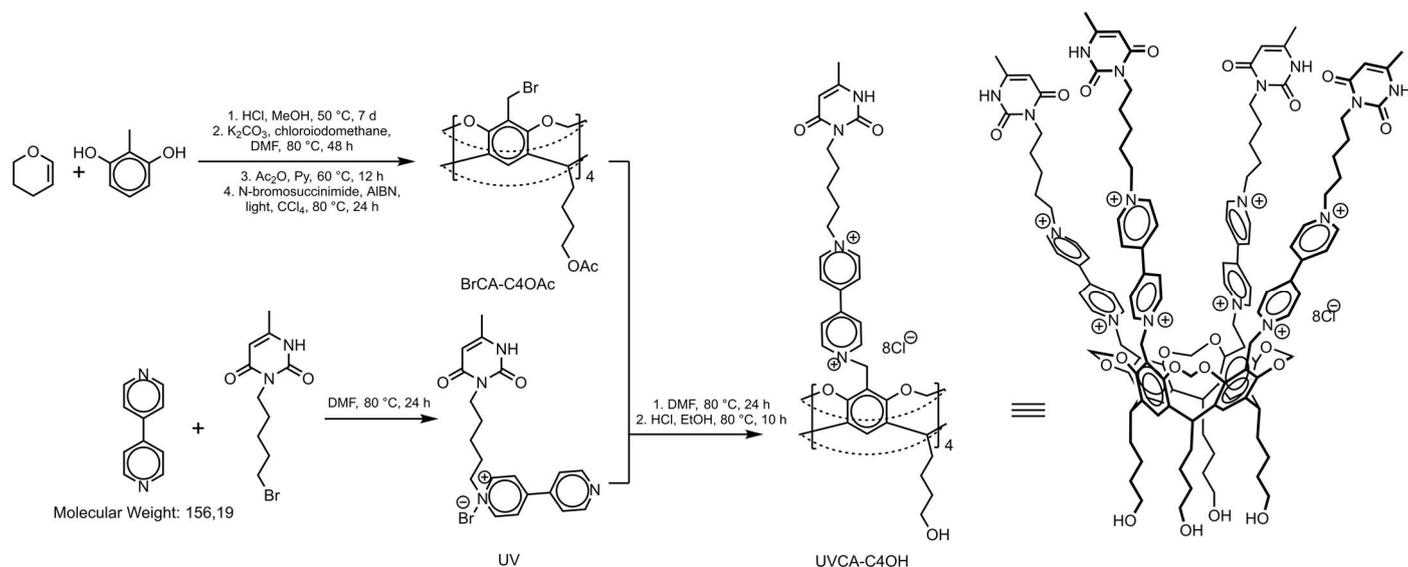
Uracils have a wide spectrum of biological activity, and they are employed to produce drugs with different pharmacological effects [16,17]. Derivatives of uracil have

anti-inflammatory, antiviral, and antioxidant properties and are used to treat herpes, hepatitis B and C, HIV, and other viruses. In vivo tests have demonstrated that 6-methyluracil derivatives can cross the blood–brain barrier and inhibit acetylcholinesterase in brain tissues [18]. Nanocarriers based on these compounds may also have anti-tumor activity and exhibit a synergistic effect with preparations.

The synthesis of resorcinarene cavitand with viologen and methyluracil groups on the upper rim is described in this study (UVCA-C4OH). Data from NMR, IR spectroscopy, and elemental analysis have all been used to confirm the structures of UVCA-C4OH and its intermediates. Future redox-controlled nanocarriers will be made from the resulting macrocycle. We are currently working on this.

## 2. Results

The synthesis of UVCA-C4OH was carried out as described in Scheme 1. First, bromomethylresorcinarene cavitand (BrCA-C4OAc) was synthesized starting from 3,4-dihydro-2H-pyran and 2-methylresorcine [19–21]. Simultaneously, an uracil-viologen derivative (UV) was obtained by reacting 3-(5-bromopentyl)-6-methyluracil with 4,4'-bipyridine in DMF. Then, by reacting UV with BrCA-C4OAc, followed by acidic hydrolysis to remove acetate groups, UVCA-C4OH was created with a total yield of 45% (Scheme 1).



**Scheme 1.** Synthesis of UVCA-C4OH.

To obtain the UV compound, 3-(5-bromopentyl)-6-methyluracil dissolved in DMF was added dropwise to a 4,4'-bipyridine solution at 80 °C and then it was mixed for two days. Then, the solvent was removed, and the remainder was washed with methylene chloride to eliminate the residual amount of bipyridine. The yield of the UV compound was 86.3%. The structure of the resulting compound was confirmed by <sup>1</sup>H and <sup>13</sup>C-NMR spectra, in which all the expected signals of bipyridine, methyluracil, and pentyl tail fragment atoms were observed (see Supplementary Materials (SM), Figures S1 and S2). A strong band of valence vibrations of the C-N<sup>+</sup> bond at 812 cm<sup>-1</sup> is fixed in the IR spectrum, confirming the formation of the pyridinium ion (Figure S3 in SM).

The UV compound was attached to the upper rim of the cavitand BrCA-C4OAc through the free nitrogen. The reaction was carried out in DMF at 80 °C for 24 h. Then DMF was removed and the hydrolysis with HCl was carried out in order to replace acetyl groups on the lower rim with hydroxyls. The unreacted reagents were removed by dialysis, which was carried out three times for 30 min. The yield of UVCA-C4OH was 62.8%.

In the <sup>1</sup>H-NMR spectrum, the proton peak of the methylene bridge between the cavitand and viologen is fixed at 5.87 ppm, while the proton signals of methylene bridges

connecting resorcinol rings appear at 5.63 and 6.39 ppm (Figure S4 in SM). The proton signals of the UV groups are shifted to the low-field compared with the initial UV compound. The signals in the range of 1.30–2.50 ppm and at 3.55, 3.82 ppm were assigned to the hydroxybutylic groups of the lower rim and pentyl linkers. The structure of the UVCA-C4OH was also confirmed by the  $^{13}\text{C}$ -NMR spectrum, in which the signals of all carbon atoms are fixed (Figure S4 in SM). The signals were assigned using 2D-NMR spectroscopy (HSQC, COSY, HMBC, Figures S5–S7 in SM). The IR spectrum shows the broadened O-H and N-H stretching bands at  $3393\text{ cm}^{-1}$  and a vibration band of the C-N+ bond at  $826\text{ cm}^{-1}$  (Figure S3 in SM).

### 3. Materials and Methods

NMR spectra were recorded on a Bruker Avance 600 MHz spectrometer. IR spectra were recorded using a Vector-27 FTIR spectrometer (Bruker, Germany) in the  $400\text{--}4000\text{ cm}^{-1}$  range. The samples were prepared as KBr pellets. The elemental analysis was carried out on a CHNS analyzer Vario Macro cube (Elementar Analysensysteme GmbH, Langenselbold, Germany). The samples were weighed on Sartorius Cubis II (Goettingen, Germany) microbalance in tin capsules. VarioMacro Software V4.0.11 (Langenselbold, Germany) was used to perform quantitative measurements and evaluate the data received.

BrCA-C4OAc was synthesized as described in [19–21] from 2-methylresorcinol, 3,4-dihydro-2H-pyran, bromochloromethane, acetic anhydride, and *N*-bromosuccinimide. 3-bromopentyl-6-methyluracil was synthesized as described in [22]. Commercially available 4,4'-bipyridine (Alfa Aesar, 98%) was used without purification.

#### 3.1. Synthesis of UV

3-(5-Bromopentyl)-6-methyluracil (0.810 g, 2.9 mmol) in 20 mL of DMF was added dropwise to a solution of 4,4'-bipyridine (1.811 g, 11.6 mmol) in 20 mL of DMF at  $80\text{ }^{\circ}\text{C}$ . Then, the mixture was heated at  $80\text{ }^{\circ}\text{C}$  for 48 h and DMF was removed at reduced pressure. The resulting oil was washed with methylene chloride to remove the residual amount of bipyridine. The precipitate was separated by centrifugation and dried to yield UV as a brown solid. The yield was 1.08 g (2.5 mmol, 85.3%).  $^1\text{H}$ -NMR  $\delta_{\text{H}}$  (600 MHz,  $\text{D}_2\text{O}$ ): 8.91 (2H, d,  $J = 6.4\text{ Hz}$ , C-H<sub>viologen</sub>), 8.74 (2H, d,  $J = 6.3\text{ Hz}$ , C-H<sub>viologen</sub>), 8.37 (2H, d,  $J = 6.4\text{ Hz}$ , C-H<sub>viologen</sub>), 7.87 (2H, d,  $J = 6.4\text{ Hz}$ , C-H<sub>viologen</sub>), 5.58 (1H, s, CH<sub>uracil</sub>), 4.63 (2H, t,  $J = 7.2\text{ Hz}$ , CH<sub>2</sub>CH<sub>2</sub>N<sup>+</sup>), 3.78 (2H, t,  $J = 7.2\text{ Hz}$ , CH<sub>2</sub>CH<sub>2</sub>N), 2.07 (2H + 3H, m CH<sub>2</sub>CH<sub>2</sub>N<sup>+</sup> and CH<sub>3</sub>), 1.61 (2H, p,  $J = 7.5\text{ Hz}$ , CH<sub>2</sub>CH<sub>2</sub>N), 1.32 (2H, p,  $J = 7.9\text{ Hz}$ , CH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>).  $^{13}\text{C}$ -NMR  $\delta_{\text{C}}$  (600 MHz,  $\text{D}_2\text{O}$ ): 166.3, 153.9, 153.6, 153.1, 150.2, 144.9, 142.7, 126.1, 122.6, 99.3, 61.5, 40.1, 30.0, 26.2, 22.5, 17.8. IR (KBr,  $\text{cm}^{-1}$ ): 3400 br (N-H), 3140–2800 m (C-H), 1706 s (C=O), 1640 s (C=C), 1665 s (C-N), 1599, 1544 s (C-C<sub>Ar</sub>), 812 m (C-N<sup>+</sup>). Anal. Calcd. for  $\text{C}_{20}\text{H}_{23}\text{N}_4\text{O}_2^+ \times \text{Br}^-$ , %: C 55.69; H 5.37; N 12.99; Br 18.53. Found C 55.14; H 5.44; N 13.28; Br 18.85.

#### 3.2. Synthesis of UVCA-C4OAc

1.08 g (2.5 mmol) of UV and 0.854 g (0.63 mmol) of BrCA-C4OAc were dissolved in 10 mL of DMF, and the mixture was stirred in an argon atmosphere at  $80\text{ }^{\circ}\text{C}$  for 24 h. Then, DMF was removed at reduced pressure to give an oily brown substance.

#### 3.3. Synthesis of UVCA-C4OH

The oil obtained in the previous step was dissolved in 20 mL of ethanol, and 2 mL of the concentrated hydrochloric acid was added. The mixture was stirred at  $80\text{ }^{\circ}\text{C}$  in an argon atmosphere overnight. The solvent was removed at reduced pressure and the resulting solid was treated with acetonitrile. The solid was then dissolved in 5 mL of water, and the resulting solution underwent three dialyses for 30 min each (versus 800 mL of water). After the solvent was removed at reduced pressure, the remaining material was treated with acetonitrile and dried to produce UVCA-C4OH as a brown solid. The yield was 1.015 g (0.40 mmol, 62.8%).  $^1\text{H}$ -NMR  $\delta_{\text{H}}$  (600 MHz,  $\text{D}_2\text{O}$ ): 9.08 (16H, m, C-

H<sub>viologen</sub>), 8.51 (16H, m, C-H<sub>viologen</sub>), 7.79 (4H, s, H<sub>Ar</sub>), 6.39 (4H, m, OCH), 5.87 (8H, s, CH<sub>2</sub>), 5.64 (4H + 4H, m, CH<sub>uracil</sub> and OCH), 3.82 (8H, m, CH<sub>2</sub>CH<sub>2</sub>N), 3.55 (8H, m, CH<sub>2</sub>OH), 2.47 (8H, m, CH<sub>2</sub>), 2.13 (12H + 8H, m, CH<sub>2</sub> and CH<sub>3</sub>), 1.64 (8H + 8H, m, CH<sub>2</sub>CH<sub>2</sub>N and CH<sub>2</sub>CH<sub>2</sub>OH), 1.38 (8H + 8H, m, CH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub> and CH<sub>2</sub>CH<sub>2</sub>CH). <sup>13</sup>C-NMR δ<sub>C</sub> (600 MHz, D<sub>2</sub>O): 166.4, 153.6, 153.5, 153.1, 150.6, 149.9, 145.8, 145.6, 139.2, 127.1, 126.1, 124.7, 119.9, 100.4, 99.3, 62.1, 61.7, 55.2, 40.1, 37.4, 31.2, 30.1, 28.9, 26.2, 23.6, 22.7, 17.9. IR (KBr, cm<sup>-1</sup>): 3393 br (O-H), 3118–2859 m (C-H), 1711 s (C=O), 1636 s (C=C, C-N), 1557, 1505 s (C-C<sub>Ar</sub>), 826 w (C-N<sup>+</sup>). Anal. Calcd. for C<sub>132</sub>H<sub>152</sub>N<sub>16</sub>O<sub>20</sub><sup>8+</sup> × 8Cl<sup>-</sup>, %: C 61.78; H 5.97; N 8.73; Cl 11.05. Found C 61.39; H 6.23; N 8.95; Cl 10.78.

#### 4. Conclusions

In this work, 6-methyluracilpentylviologen resorcinarene cavitand was synthesized for potential application as a nanocarrier for drug delivery. The cavitand and intermediate products are characterized by a set of physicochemical methods (<sup>1</sup>H-NMR, <sup>13</sup>C-NMR, and IR spectroscopy).

**Supplementary Materials:** Figure S1: <sup>1</sup>H-NMR spectrum of UV; Figure S2: <sup>13</sup>C-NMR spectrum of UV; Figure S3: IR spectra of UV and UVCA-C4OH; Figure S4: <sup>1</sup>H- and <sup>13</sup>C-NMR spectra of UVCA-C4OH; Figure S5: HSQC NMR spectrum of UVCA-C4OH; Figure S6: COSY NMR spectrum of UVCA-C4OH; Figure S7: HMBC NMR spectrum of UVCA-C4OH.

**Author Contributions:** Conceptualization, V.E.S.; methodology, A.Y.Z.; investigation, E.E.M., M.M.S. and V.V.S.; data curation, A.Y.Z. and I.S.A.; writing—original draft preparation, E.E.M.; writing—review and editing, A.Y.Z. and I.S.A. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by the government assignment for FRC Kazan Scientific Center of RAS.

**Data Availability Statement:** Not applicable.

**Acknowledgments:** The authors gratefully acknowledge the Assigned Spectral-Analytical Center of the FRC Kazan Scientific Center of RAS for spectral and analytical measurements.

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

#### References

1. Peer, D.; Karp, J.M.; Hong, S.; Farokhzad, O.C.; Margalit, R.; Langer, R. Nanocarriers as an emerging platform for cancer therapy. *Nat. Nanotechnol.* **2007**, *2*, 751–760. [[CrossRef](#)] [[PubMed](#)]
2. Kenchegowda, M.; Rahamathulla, M.; Hani, U.; Begum, M.Y.; Guruswamy, S.; Osmani, R.; Ali, M.; Gowrav, M.P.; Alshehri, S.; Ghoneim, M.M.; et al. Smart nanocarriers as an emerging platform for cancer therapy: A review. *Molecules* **2022**, *27*, 146. [[CrossRef](#)] [[PubMed](#)]
3. Cai, H.; Dai, X.; Wang, X.; Tan, P.; Gu, L.; Luo, Q.; Zheng, X.; Li, Z.; Zhu, H.; Zhang, H. A nanostrategy for efficient imaging-guided antitumor therapy through a stimuli-responsive branched polymeric prodrug. *Adv. Sci.* **2020**, *7*, 1903243. [[CrossRef](#)]
4. Perrault, S.D.; Walkey, C.; Jennings, T.; Fischer, H.C.; Chan, W.C. Mediating tumor targeting efficiency of nanoparticles through design. *Nano Lett.* **2009**, *9*, 1909–1915. [[CrossRef](#)]
5. Allen, T.M. Ligand-targeted therapeutics in anticancer therapy. *Nat. Rev. Cancer* **2002**, *2*, 750–763. [[CrossRef](#)]
6. Bai, H.; Wang, J.; Li, Z.; Tang, G. Macrocyclic compounds for drug and gene delivery in immune-modulating Therapy. *Int. J. Mol. Sci.* **2019**, *20*, 2097. [[CrossRef](#)]
7. Zheng, Z.; Geng, W.-C.; Xu, Z.; Guo, D.-S. Macrocyclic amphiphiles for drug delivery. *Isr. J. Chem.* **2019**, *59*, 913–927. [[CrossRef](#)]
8. Chen, J.; Zhang, Y.; Zhao, L.; Zhang, Y.; Chen, L.; Ma, M.; Du, X.; Meng, Z.; Li, C.; Meng, Q. Supramolecular drug delivery system from macrocycle-based self-assembled amphiphiles for effective tumor therapy. *ACS Appl. Mater. Interfaces* **2021**, *13*, 53564–53573. [[CrossRef](#)]
9. Yu, J.; Qi, D.; Li, J. Design, synthesis and applications of responsive macrocycles. *Commun. Chem.* **2020**, *3*, 189. [[CrossRef](#)]
10. Fan, X.; Guo, X. Development of calixarene-based drug nanocarriers. *J. Mol. Liq.* **2021**, *325*, 115246. [[CrossRef](#)]
11. Hoskins, C.; Curtis, A.D.M. Simple calix[n]arenes and calix[4]resorcinarenes as drug solubilizing agents. *J. Nanomed. Res.* **2015**, *2*, 65–71.
12. Shetty, D.; Skorjanc, T.; Olson, M.A.; Trabolsi, A. Self-assembly of stimuli-responsive imine-linked calix[4]arene nanocapsules for targeted camptothecin delivery. *Chem. Commun.* **2019**, *55*, 8876–8879. [[CrossRef](#)] [[PubMed](#)]

13. Ciepluch, K.; Katir, N.; El Kadib, A.; Felczak, A.; Zawadzka, K.; Weber, M.; Klajnert, B.; Lisowska, K.; Caminade, A.-M.; Bousmina, M.; et al. Biological properties of new viologen-phosphorus dendrimers. *Mol. Pharm.* **2012**, *9*, 448–457. [[CrossRef](#)] [[PubMed](#)]
14. Bongard, D.; Bohr, W.; Swierczek, M.; Degefa, T.H.; Walder, L.; Brandt, R. Alkylene-bridged viologen dendrimers: Versatile cell delivery tools with biosensing properties. *Org. Biomol. Chem.* **2014**, *12*, 9583–9591. [[CrossRef](#)] [[PubMed](#)]
15. Li, J.; Lepadatu, A.-M.; Zhu, Y.; Ciobanu, M.; Wang, Y.; Asaftei, S.C.; Oupický, D. Examination of structure–activity relationship of viologen-based dendrimers as CXCR4 antagonists and gene carriers. *Bioconjug. Chem.* **2014**, *25*, 907–917. [[CrossRef](#)]
16. Pałasz, A.; Cież, D. In search of uracil derivatives as bioactive agents. Uracils and fused uracils: Synthesis, biological activity and applications. *Eur. J. Med. Chem.* **2015**, *97*, 582–611. [[CrossRef](#)] [[PubMed](#)]
17. Ramesh, D.; Vijayakumar, B.G.; Kannan, T. Therapeutic potential of uracil and its derivatives in countering pathogenic and physiological disorders. *Eur. J. Med. Chem.* **2020**, *207*, 112801. [[CrossRef](#)] [[PubMed](#)]
18. Semenov, V.E.; Zueva, I.V.; Mukhamedyarov, M.A.; Lushchekina, S.V.; Kharlamova, A.D.; Petukhova, E.O.; Mikhailov, A.S.; Podyachev, S.N.; Saifina, L.F.; Petrov, K.A.; et al. 6-Methyluracil derivatives as bifunctional acetylcholinesterase inhibitors for the treatment of Alzheimer’s disease. *ChemMedChem* **2015**, *10*, 1863–1874. [[CrossRef](#)]
19. Bossmann, S.; Leaym, X.; Kraft, S. Synthesis of Water-soluble highly charged and methylene-bridged resorcin[4]arenes. *Synthesis* **2008**, *6*, 932–942. [[CrossRef](#)]
20. Gibb, B.C.; Chapman, R.G.; Sherman, J.C. Synthesis of hydroxyl-footed cavitands. *J. Org. Chem.* **1996**, *61*, 1505–1509. [[CrossRef](#)]
21. Sultanova, E.D.; Mukhitova, R.K.; Ziganshina, A.Y.; Konovalov, A.I.; Krasnova, E.G.; Kharlamov, S.V.; Nasybullina, G.R.; Yanilkin, V.V.; Nizameev, I.R.; Kadirov, M.K.; et al. Thermoresponsive polymer nanoparticles based on viologen cavitands. *ChemPlusChem* **2015**, *80*, 217–222. [[CrossRef](#)]
22. Semenov, V.E.; Akamsin, V.D.; Reznik, V.S. Synthesis of acyclic and macrocyclic analogs of di-, tri-, and tetranucleotides. *Russ. J. Gen. Chem.* **2007**, *77*, 1430–1440. [[CrossRef](#)]