SELF ORGANIZATION OF FLUORESCENT MOLECULAR NECKLACES IN AQUEOUS SOLUTION

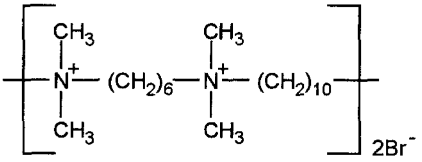
I. Kräuter, W. Herrmann, G. Wenz*
Polymer-Institut der Universität Karlsruhe,
Hertzstr. 16, D-76187 Karlsruhe, Germany

ABSTRACT

N-Fluoresceinyl-N'-(mono-6-desoxy-6-β-cyclodextrinyl)-thiourea **2** was synthesized from mono-6-amino-6-desoxy-β-CD **1** and fluorescein isothiocyanate. The fluorescent CD derivative **2** was threaded on a water soluble polymer, poly(N,N-dimethylammoniumhexamethylene-N',N'-dimethylammoniumdecamethylene dibromide) **3**. The existence of the molecular necklace was visualized by gel electrophoresis.

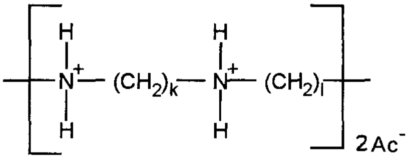
1. INTRODUCTION

Fluorescent CD derivatives are well known by the work of Ueno et al. for the detection of various guest molecules [1-4]. So, chromophores like naphthalene, pyrene, anthracene were attached to CDs. These fluorescent CDs were used to detect a great variety of guest molecules, e. g. steroids. Up to now, the detection of polymers by fluorescent CDs was never reported. Threading of unsubstituted CDs on polymer chains is well known since the year 1990 by the work of Harada and us. For example α-CD was threaded on poly(ethylene glycol), β-CD on poly(propylene glycol) and γ-CD on poly(methyl vinyl ether) or poly(isobutylene) to form polymer inclusion coumpounds which are insoluble in water [5-8]. We found that CDs form watersoluble polymer inclusion compounds with poly(N,N-dimethylammoniumoligomethylene)s **3** and poly(iminooligomethylene)s **4** if the alkyl segments are longer than eight methylene groups [9-13]. Here, we report of threading of a fluorescent β-CD derivative on a polymer chain to form watersoluble "shining" molecular necklaces.



$$\left[-N^{+}(CH_3)_2-(CH_2)_6-N^{+}(CH_3)_2-(CH_2)_{10}- \right] 2Br^-$$

3



$$\left[-N^{+}H_2-(CH_2)_k-N^{+}H_2-(CH_2)_l- \right] 2Ac^-$$

4

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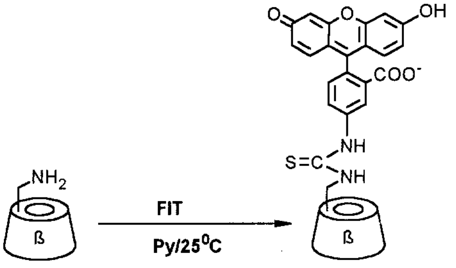
2. MATERIALS AND METHODS

Mono-6-amino-6-desoxy-β-CD **1** was synthesized starting from tosyl-β-CD via the mono-6-desoxy-6-azido-β-CD according to the literature [14]. The reduction of the azido derivative to the amine was performed by hydrogenation using the catalyst Pd/C in nearly quantitative yield. Fluorescein isothiocyanate (isomer I) was from *Aldrich* and β-CD from *Wacker AG*, München. Poly(N,N-dimethylammoniumhexamethylene-N',N'-dimethylammoniumdecamethylene) **3** was prepared by the *Menschutkin* analogous reaction of N,N,N',N'-tetramethyl-1,6-diaminohexane and 1,10-dibromodecane (both from *Aldrich*) in N-methylformamid / methanol 1:1 at 50°C for 14 days and purified by dialysis.

For the electrophoresis gel plates of a size of 10x10 cm^2 were casted from a 1.2 % solution of agarose (15510-019, *Gibco* BRL) in 50 mL 1 x TAE buffer at pH = 8.3. The electrophoresis was run at 90 V and 70 mA using a standard equipment.

3. RESULTS AND DISCUSSION

The fluorescent β-CD derivative **2** was synthesized by addition of mono-6-amino-6-desoxy-β-CD **1** to fluorescein isothiocyanate in 85 % yield. The addition reaction was followed by IR spectroscopy by means of the rising thiourea band at 1614 cm^{-1} and the decaying isothiocyanate band at 2041 cm^{-1} The structure of **2** was proved by NMR spectroscopy. The product shows narrow bands in both the absorption spectrum at $\lambda = 493$ nm and the emission spectrum at $\lambda = 519$ nm.



1 **2**

In contrast to previous fluorescent CD derivatives, **2** is quite well soluble in water at pH > 9. The inclusion of a guest causes only very small changes of the UV/VIS spectra (fig. 1). Consequently, the hydrophilic fluorescein substituent seems not to interact strongly with the β-CD cavity.

Fluorescent β-CD **2** can be threaded on the polymer **3** in aqueous solution. To prevent the labeled rings **2** from sliding off the polymer chain, α-CD was threaded afterwards. From earlier studies we know that α-CD propagates very slowly along the polymer chain. Therefore it can be used as an efficient blocking entity. The resulting structure is an example of a *pseudopolyrotaxane* (fig.2).

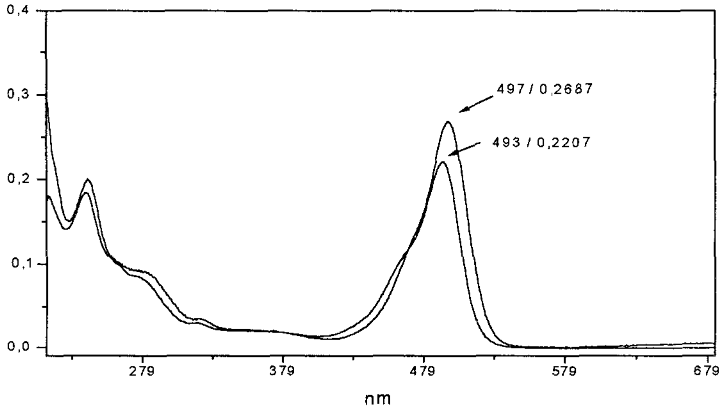


Fig. 1 UV/VIS spectra of a 3 * 10^{-6} M solution of the fluorescent ß-CD **2**, free (493 nm) and threaded on the ionene **3** (497 nm).

The existence of this supramolecular structure was proved by horizontal gel electrophoresis in a straightforeward way. This proof was based on the fact that free fluorescent β-CD **2** has a negative charge, while threaded fluorescent β-CD **2•3** has a net positive charge due to the excess positive charge of the polymer **3**. Therefore free compound **2** should migrate to the anode and threaded **2•3** to the cathode. This was really the case. To exclude a loose association of fluorescent β-CD **2** at the polymer **3**, we also performed a control experiment: α-CD was threaded first on the polymer chain **3** to block the chain ends. Indeed nearly no fluorescent β-CD **2** could be threaded afterwards. Consequently, the formation of fluorescent pseudopolyrotaxanes (fig. 2) was proved unambiguously.

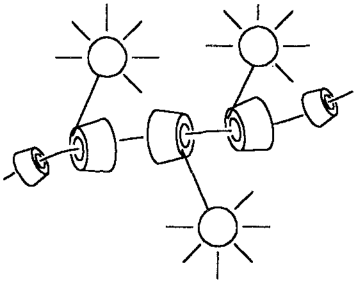


Fig. 2 Schematic drawing of the fluorescent pseudopolyrotaxane

4. CONCLUSION

A new watersoluble fluorescent CD **2** derivative was synthesized. Polymer chains can be made visible by inclusion in fluorescent CD **2**. Gel electrophoresis is well suited in general to detect the inclusion of polymer chains.

5. ACKNOWLEDGEMENTS

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