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# New fluorescence active cellulosics prepared by a convenient acylation reaction

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## Summary

Novel, fluorescence active cellulosic polymers with high degree of functionalization of up to 1.0 were synthesized in homogeneous phase (cellulose in *N,N*-dimethylacetamide/LiCl and 6-*O*-thexyldimethylsilylcellulose in pyridine) with a mixed anthracene-9-carboxylic/*p*-toluenesulfonic acid anhydride. This novel and effective synthetic method yield the pure carboxylic acid esters of the polymers, which were characterized by means of FTIR- as well as $^{1}H$ and $^{13}C$ NMR-spectroscopy. The polymers show typical fluorescence spectra, which are identical with the non-bond anthracene-9-carboxylic acid.

## Introduction

Fluorescence spectroscopy is a very sensitive analytical technique to study the mobility and dynamic properties of fluorescent labeled molecules. Pyrene groups have been attached by the reaction of various cellulose ethers such as hydroxypropyl- and methylcellulose as well as tylose with 4-(1-pyrenyl)-butyl tosylate in the presence of sodium hydride [1,2]. By means of spectroscopic measurements it was shown that in aqueous solutions the fluorescent labeled cellulosics form interpolymeric aggregates via clusters of hydrophobic chromophores. However, the synthesis path used is limited to rather stable cellulose products like some ethers. In the case of esters or trialkylsilylethers a cleavage of the primary functional group may occur under these reaction conditions. An alternative synthetic approach to fluorescent labeled cellulosic polymers is the reaction with 5-dimethylamino-1-naphthalenesulfonyl chloride which can be carried out under comparatively mild condition (dipolar aprotic solvent, triethylamine, +10°C)[3]. A problem of sulfonyl esters may arise from the possibility to serve as a leaving group in nucleophilic displacement reactions and to be substituted by even mild nucleophils. Thereby, the molecular structure of the polymer is additionally altered in an uncontrolled way, which may influence the supramolecular architectures in solution as well [4].

In contrast to recent progress in cellulose chemistry, which is in particular stimulated by the use of non-aqueous solvents [5] and by regioselectively functionalized [6] or activated [7] derivatives, a fully satisfying solution of fluorescence labeling is still missing. In the present studies, we investigated the homogeneous acylation of cellulose in the solvent system *N,N*-dimethylacetamide/LiCl and of organo-soluble 6-*O*-thexyldimethylsilylcellulose with the fluorescent active anthracene-9-carboxylic acid by using a new and

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convenient acylation procedure. Both the new organo-soluble products and the additionally acetylated derivatives are characterized by means of FTIR- as well as $^{1}H$ and $^{13}C$ NMR spectroscopy. UV absorption and fluorescence of the cellulosics are discussed as well.

## Experimental

Cellulose (AVICEL PH 101, degree of polymerization 280) was dried in vacuum at 105°C. LiCl was dried in vacuum at 150°C. The solvents were purified using common methods and distilled prior use. Tosylchloride and anthracene-9-carboxylic acid were used without further purification.

All experiments were carried out in the darkness in order to prevent photochemically initiated reactions.

### *Cellulose anthracene-9-carboxylate 2*

In a typical procedure, 2.0 g (12.3 mmol) cellulose was suspended in 50 ml DMA. Under exclusion of moisture, the mixture was stirred for two hours at 120°C. After cooling down to 100°C, 3.0 g LiCl was added. The mixture was stirred at room temperature until a clear solution is obtained. 7.03 g (36.9 mmol, 3 mol/mol AGU) tosylchloride was dissolved and 8.20 g anthracene-9-carboxylic acid was suspended in the mixture. After stirring for 4 hours at 50°C it was precipitated in 400 ml ethanol. The separated polymer was carefully washed with ethanol and dried in vacuum with increasing temperature up to 40°C.

Yield: 4.0 g, DS: 0.52 (based on elemental analysis, 61.72% C, 5.76% H). The polymer is soluble in chloroform, tetrahydrofurane and methylenechloride.

FTIR (KBr): 3422 $cm^{-1}$ $\nu_{OH}$, 2882 $cm^{-1}$ $\nu_{CH2}$, 1790 $cm^{-1}$ and 1723 $cm^{-1}$ $\nu_{C=O}$ and the typical absorptions of the cellulose backbone

$^{1}H$ NMR (DMSO-$d_6$): 7.6-8.2 ppm $H_{aromatics}$

### *Acetylated cellulose anthracene-9-carboxylate 3*

1.6 g of polymer 2 was suspended in 20 ml dry pyridine and treated with 20 ml acetic acid anhydride. After stirring overnight at room temperature the mixture was stirred for 4.5 hours at 50°C. The product was carefully poured into a phosphate buffer solution (pH 7) containing nearly 10% $NaHCO_3$. The precipitated polymer was collected, washed with water and dried in vacuum at 40°C.

Yield: 1.82 g. The polymer is soluble in chloroform.

FTIR (KBr): no $\nu_{OH}$, 1754 $cm^{-1}$ $\nu_{C=O}$

$^{13}C$ NMR ($CDCl_3$): 169.1-170.1 ppm $C_{C=O}$, 124.6-130.9 ppm $C_{aromatics}$, 62.2-100.4 ppm $C_{cellulose\ backbone}$, 20.4 ppm $C_{CH3}$

$^{1}H$-NMR ($CDCl_3$): 7.3-8.6 ppm $H_{aromatics}$, 3.5-5.1 ppm cellulose backbone, 2 ppm $H_{CH3}$

### *6-O-Thexyldimethylsilylcellulose 1a*

50.0 g cellulose (0.309 mol) were suspended in 200 ml *N*-methyl pyrrolidone (NMP) and stirred at 80°C for 1 hour. After cooling down to -25°C 300 ml NMP, which was saturated with ammonia, were added. The mixture was stirred for 1 hour and a solution of 121.1 ml (0.617 mol; 2 mol/mol AGU) thexyldimethylchlorosilane in 120 ml NMP was

added. The precipitation of ammonium chloride occurs. After stirring for 1 hour at -25°C the mixture was slowly warmed to room temperature. It was allowed to stand overnight and stirred for 24 houres at 80°C. The highly viscous solution was poured into a buffer solution. The product was filtered off, washed and dried as described above. For purification, it was reprecipitated from NMP into water. After separation it was washed and dried in vacuum at 40°C.

Yield: 109.7 g (76%). $DS_{Si}$: 0.91 (based on the silicon content determined by elemental analysis, 8.75% Si).

FTIR (KBr): 3460 $cm^{-1}$ $\nu_{OH}$, 1253 $cm^{-1}$ $\delta_{Si\text{-}C}$, 833 $cm^{-1}$ $\nu_{Si\text{-}C}$, and the typical absorptions of the cellulose backbone.

***Thexyldimethylsilylcellulose anthracene-9-carboxylate 2a***

11.2 g (51.4 mmol, 3 mol/mol modified AGU) anthracene-9-carboxylic acid was suspended in 50 ml pyridine and treated with 9.9 g (51.4 mmol) tosylchloride. After stirring for 5 hours the mixed anhydride was added to a solution of 5.0 g 6-*O*-thexyldimethylsilylcellulose (17.1 mmol, DS 0.91) in 75 ml pyridine. The suspension was stirred at 50°C for 18 hours. After cooling down to room temperature the polymer was precipitated in 500 ml ethanol, collected and washed carefully with ethanol. The product was dried in vacuum at 40°C.

Yield: 7.25 g, $DS_{Si}$: 0.64; $DS_{acyl}$: 1.0 calculated on the basis of elemental analysis: 68,6% C, 6,8% H, 3,92% Si. The product is soluble in dimethylsulfoxide, *N,N*-dimethylformamide, and NMP.

***Measurements***

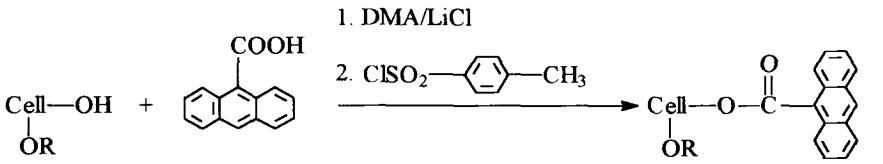
The NMR spectra were recorded on a Bruker DRX400 spectrometer. $^{13}C$ NMR spectra were measured in $CDCl_3$ (at 313 K) or DMSO-$d_6$ (at 333 K). 4700-18000 scans were accumulated. For the $^{1}H$-NMR spectra 16 scans were accumulated. FTIR-spectra were recorded on a BioRad FTS 25 spectrometer (KBr-technique).

UV absorption, fluorescence and fluorescence excitation spectra were measured on Perkin Elmer spectrometers (Lambda 16 and LS 50, respectively). The fluorescence quantum yield was obtained using quinine bisulfate in 1m $H_2SO_4$ ($\Phi_f$=0.55) as a standard (including refractive index correction). The fluorescence decay time measurements were carried out by a single photon counting technique (FL900CDT, Edinburgh Analytical Instruments).

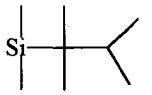
## Results and Discussion

The solvent system *N,N*-dimethylacetamide(DMA)/LiCl is an appropriate reaction medium for the homogeneous acylation of cellulose using reactive carbonic acid derivatives like chlorides or anhydrides [5]. The carbonic acid itself, on the other hand, does not yield cellulose esters. An acidic catalysis, which is useful for esterification of low-molecular alcohols and for acetylation of cellulose, is not very suitable for the introduction of bulky carbonic acids caused by promotion of chain degradation due to long reaction times. A new interesting method for increasing the carbonyl activity consists in the in situ formation of mixed acid anhydrides [8]. It is realized by the treatment of the dissolved cellulose with carbonic acid in the presence of an equimolar amount of *p*-toluenesulfonyl(tosyl)chloride forming the mixed acid anhydride, which is the reactive

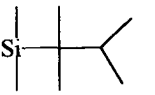
acylating agent. However, it may also be synthesized in a first step and subsequently allowed to react with the polymer.

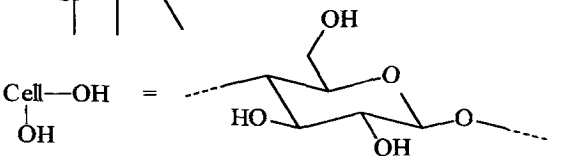



**1** R= H
**1a** R= H or according to DS $_{Si}$ 0.91




**2** R= H
**2a** R= H or




DMA= *N,N*-dimethylacetamide

In a first series of experiments, solutions of cellulose **1** in DMA/LiCl were allowed to react with anthracene-9-carboxylic acid in the presence of tosylchloride. At a molar ratio of 3 mol anthracene-9-carboxylic acid per mol anhydroglucose unit (AGU), a degree of substitution ($DS_{anth}$) of 0.52 (calculated on the basis of elemental analysis) was reached within a reaction time of 4 h at 50°C (**2**). In the FTIR spectra of **2** the typical absorption bands of the cellulose backbone were found and additionally a signal at 1723 $cm^{-1}$ $\nu$ $C{=}O_{ester}$ indicating the presence of the ester moiety. Due to the steric hindrance of the bulky anthracenoyl group, the $DS_{anth}$ 0.52 reached is comparatively high.

The treatment of **2** with acetic acid anhydride in pyridine gave the mixed ester **3** without any free hydroxyl functions. The polymers **3** are readily soluble in $CHCl_3$, e.g.

All polymers were characterized by means of NMR spectroscopy. Due to the remaining OH-groups in **2**, the resolution of the spectra is rather bad as a result of intra- and intermolecular hydrogen bonds forming aggregates having long relaxation lime ($T_1$). The complete acetylation of all OH-groups, as described, yields products, which gave well resolved $^{13}C$ NMR spectra. Fig. 1 shows a typical $^{13}C$ NMR spectrum of the acetylated cellulose anthracene-9-carboxylate **3**. The resonances of the AGU carbon atoms were observed between 62.16 and 100.36 ppm, however, with a rather low intensity. The signals of the aromatic carbons appear between 124.68 and 130.91 ppm and their chemical shift values agree very well with those of a model compound anthracene-9-carboxylic acid methyl ester [9]. The two signals at 169.16 and 169.65 ppm result from the carbonyl C-atom of the acetyl group and the signal at 170.10 ppm is assigned to the carbonyl carbon resonance of the anthracene-9-carboxylate. That means the analytical data obtained are sufficient to prove the molecular structure of the polymers.

To evaluate whether the synthesis path is also suitable for cellulose derivatives, which are rather instable under alkaline conditions, the acylation of thexyldimethylsilylcellulose was studied. The regioselectively substituted 6-*O*-thexyldimethylsilylcellulose **1a** (DS 0.91, details of this protected cellulose product are described in ref. [10]) was allowed to react with 3 mols mixed carboxylic sulfonic acid anhydride per mol modified AGU. In this case, the acylation agent was pre-formed. After a reaction time of 18 hours at 50°C a product (**2a**) of $DS_{Si}$= 0.64 and $DS_{anth}$= 1.0 was obtained. Obviously, the *p*-

toluenesulfonic acid formed leads to a partial desilylation of the starting polymer. On the other hand, the etherification of **1a** under typical conditions of the Williamson ethersynthesis (catalyzed with NaOH) results in a complete removal of all silyl functions. Furthermore, no remarkable formation of cellulose tosylate occurs, as concluded from elemental analysis and FTIR spectroscopy. The polymer **2a** is readily soluble in common organic solvents as well.

By means of $^{13}C$ NMR spectroscopy in DMSO-$d_6$, no signals of the modified AGU can be recognized. This is due to the high molecular mass of both functional groups compared with the AGU as well as due to large difference in relaxation behavior ($T_1$) of the polymer chain and the functional groups. The peak of the carbonyl carbon atom of the anthracene-9-carboxylate group can be observed at 170.0 ppm. Further signals are found between 126-130 ppm characteristic for the aromatic carbons. The typical resonances for the carbon atoms of the thexyldimethylsilyl groups appear in the range from -3 to 20 ppm.

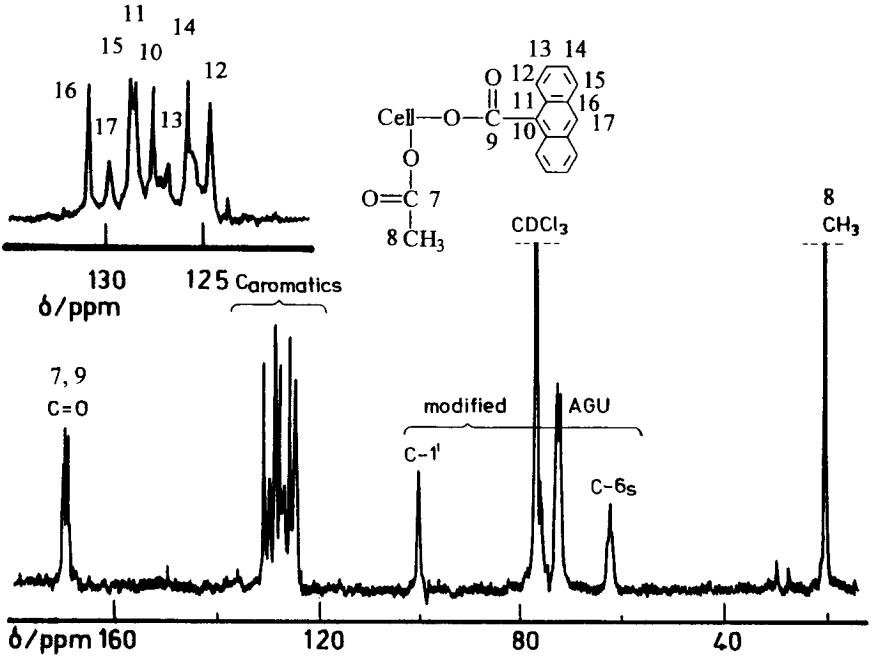



Fig. 1. $^{13}C$ NMR spectrum of the fully acetylated cellulose anthracene-9-carboxylate **3** recorded in $CDCl_3$ at 313 K (4742 scans were accumulated)

The absorption spectra of anthracene, anthracene-9-carboxylic acid and the labeled cellulose derivatives (**2, 3**) are almost identical. However, drastic changes occur in the fluorescence spectra. Whereas anthracene shows a good mirror image relationship between fluorescence and fluorescence excitation spectrum with a very small Stokes shift, the fluorescence spectrum of the anthracene-9-carboxylic acid shows a broad structureless band with a large Stokes shift (Fig. 2).

Table 1: UV absorption and fluorescence data of anthracene, anthracene-9-carboxylic acid, cellulose anthracene-9-carboxylate **2**, and thexyldimethylsilylcellulose anthracene-9-carboxylate **2a**

| Compound | Solvent | $\lambda_a$ (nm)[a] | $\lambda_f$ (nm)[b] | $\phi_f$[c] | $\tau_f$ (ns)[d] |
|---|---|---|---|---|---|
| Anthracene | CH[e] | 376 | 378 | | |
| | TCE[f] | 380 | 384 | | |
| | $CH_2Cl_2$ | 378 | 382 | 0.25 | 2.1 |
| | $CHCl_3$ | 378 | 383 | | |
| | $C_2H_5OH$ | 375 | 379 | | |
| | | (342)[g] | (385)[g] | | |
| Anthracene-9- | CH[e] | 380 | 456 | | |
| carbonic acid | TCE[f] | 385 | 478 | | |
| | $CH_2Cl_2$ | 383 | 769 | 0.35 | 7.3 |
| | | (357)[g] | (421)[g] | | |
| Cellulose anthracene-9- | TCE | 387 | 472 | | |
| carboxylate **2** | $CH_2Cl_2$ | 384 | 469 | 0.35 | 7.1 |
| Thexyldimethyl | CH[e] | 384 | 459 | | |
| silylcellulose | TCE[f] | 387 | 464 | | |
| anthracene-9- | $CH_2Cl_2$ | 386 | 463 | 0.032 | 8.2 |
| carboxylate **2a** | $CHCl_3$ | 387 | 463 | | |
| | $C_2H_5OH$ | 383 | 460 | | |

[a] Position of the longest wavelength absorption band, [b] Position of the fluorescence band, [c] Fluorescence quantum yield, [d] Fluorescence decay time, [e] Cyclohexane, [f] 1,1',2,2'-Tetrachloroethane, [g] Calculated [11]

Quantum chemical calculations (MNDO-PM3 [11]) reveal that due to the carboxylic acid substituent at 9-position a charge transfer occurs, which causes a considerable change of the bonding order (large $\pi$ bond delocalization in the equilibrated $S_1$ state) in the electronically excited state. This gives rise to a decrease of the energy gap between the $S_1$ and the $S_0$ state at the geometry of the excited state resulting in a large Stokes shift.

Position and shape of the fluorescence band is practically not altered by the formation of the cellulosic esters as well as by changing the solvent polarity (Fig. 3, Tab. 1). That means it is not possible to distinguish between bond and not-bond fluorophore.

Surprisingly the anthracene-9-carboxylic ester of thexyldimethylsilylcellulose **2a** shows a comparatively low fluorescence quantum yield caused by a quenching of the excited state of the anthracene label by the silicone, which acts as a strong electron donor towards the excited state of anthracene-9-carboxylic acid ester (Tab. 1). Since the fluorescence decay time of **2a** is practically identical with that of **2**, it can be concluded that the observed fluorescence results from those modified AGUs which do not contain silyl substituents. This effect might be used to analyze the distribution of silylether groups in the important class of trialkylsilylcellulose derivatives and will be studied in detail in another paper.

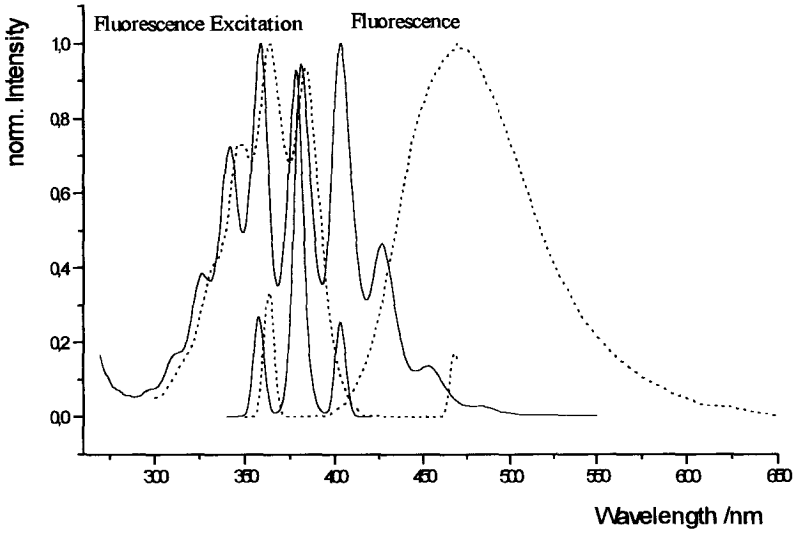



Fig. 2. Fluorescence and fluorescence excitation spectra of anthracene (solid line) and anthracene-9-carboxylic acid (dotted line) in $CH_2Cl_2$

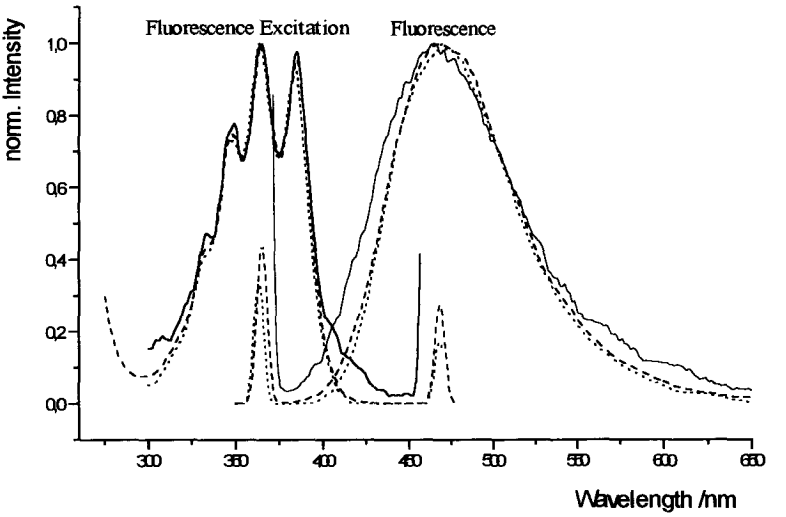



Fig. 3. Fluorescence and fluorescence excitation spectra of anthracene-9-carboxylic acid (dotted line), cellulose anthracene-9-carboxylate **2** (dashed line), and 6-*O*-thexyldimethylsilylcellulose anthracene-9-carboxylate **2a** (solid line) in $CH_2Cl_2$

**References and notes**


1 Winnik F M, Sudarshi T A R, Desmond Goddard E (1997) Langmuir 13: 111
2 Winnik F M, Winnik M A, Tazuke S, Ober C K (1987) Macromolecules 20: 38
3 Heinze T, Camacho Gomez J A, Haucke G (1996) 37: 743
4 Heinze T, Rahn K (1996) Papier (Darmstadt) 50: 721
5 For review see: Dawsey T R (1994) Application and Limitation of LiCl/N,N-Dimethylacetamide in the Homogeneous Derivatization of Cellulose. In: Gilbert R D (ed) Cellulosic Polymers, Blends and Composites. Hauser Publ. Munich Vienna New York pp 157-171
6 Klemm D, Stein A, Heinze T, Philipp B, Wagenknecht W (1996) Cellulose (Regioselectively Substituted Esters and Ethers). In: Salamone J C (ed) Polymeric Materials Encyclopedia: Synthesis, Properties and Application. CRC Press Boca Raton New York London Tokyo vol 2, pp 1043-1054
7 Heinze T, Liebert T, Cellul Chem Technol, in press
8 Sealey J E, Samaranayake G, Todd J G, Glasser W G (1996) J Appl Polym Sci Part B Polym Phys 34: 1613
9 SpecInfo (1993) Reg. No. STCC-93851-833, $^{13}C$ NMR data, spectrum number CNCC-95868-601
10 Koschella A, Klemm D (1997) Macromol Symp, in press
11 Rauhut G, Alex A, Chandrasekhar J, Steinke T, Sauer W, Beck B, Hutter M, Gedeck P, Clark T, VAMP V6.0 Oxford Molecular Ltd. Oxford England