Julia Langer

Post-Polymerization Functionalization of Poly(vinyl chloride) with Aromatic Thiols

A New Approach Leading to an Antimicrobial Polymer

MASTER THESIS

Masterarbeit

zur Erlangung des akademischen Grades einer

Diplom-Ingenieurin

der Studienrichtung Technische Chemie

erreicht an der Technischen Universität Graz

Betreuer: Assoc. Prof. Dipl. Ing. Dr. techn. Christian Slugovc

Institut für Chemische Technologie von Materialien

Technische Universität Graz

April 2011

Deutsche Fassung:

Beschluss der Curricula-Kommission für Bachelor-, Master- und Diplomstudien vom 10.11.2008

Genehmigung des Senates am 1.12.2008

EIDESSTATTLICHE ERKLÄRUNG

Ich erkläre an Eides statt, dass ich die vorliegende Arbeit selbstständig verfasst, andere als die angegebenen Quellen/Hilfsmittel nicht benutzt, und die den benutzten Quellen wörtlich und inhaltlich entnommene Stellen als solche kenntlich gemacht habe.

Graz, am …… ……..

(Unterschrift)

Englische Fassung:

STATUTORY DECLARATION

I declare that I have authored this thesis independently, that I have not used other than the declared sources / resources, and that I have explicitly marked all material which has been quoted either literally or by content from the used sources.

…… ……..

date (signature)

Acknowledgment

Herewith I'd like to express my sincere gratitude to everyone who has supported and helped me or contributed in any way to accomplish my work.

First of all I thank my supervisor Christian Slugovc for his guidance and mentoring and especially for all the helpful discussions and motivations whenever things didn't turn out the way as they were expected to.

Furthermore I am most grateful to Nadja Noormofidi for making it possible for me to work on this thesis in the first place, also for her constant inspirations and new ideas and in particular for the opportunity of taking part in the qualification programme "ErFIndERIn, which enabled amongst many other things my participation at international conferences.

My gratitude also goes to Karl Rametsteiner, representing KE KELIT Kunststoffwerk GmbH, for his constructive discussions and sincere interest in my work.

Special thanks goes to Julia Kienberger who always helped me with words and deeds; for sharing her know-how, helping out with practical tips and also for her optimism and encouraging words in difficult times.

Of course I'd also like to thank all my colleagues within our research group and at ICTM, for their helpfulness and support and for keeping the spirits high inside the lab.

Moreover I'm grateful to Petra Kaschnitz, Josefine Hobisch, Gregor Trimmel, Adriana Gregorova and Barbara Seibt for additional help concerning NMR, STA, DSC and elemental analysis measurements, as well as to the collaborators of TTZ Bremerhaven for the determination of antimicrobial activity.

Financial support by the Austrian Research Promotion Agency (FFG) in the framework of FEMtech Karrierewege (project ErFIndERIn), under contribution of KE KELIT Kunststoffwerke GmbH, is gratefully acknowledged.

Last but not least I'd like to express my deep gratitude to my family and friends for their financial help, their understanding and mental support and their constant faith in me. At this point I'd also like to thank Doris Höglinger for being a great friend during all the years since high school times.

ABSTRACT

Modification of PVC has been studied intensively over the past decades. It is an effective tool to alleviate inherent defects and thereof resulting limitations of this material. Especially dechlorination via nucleophilic substitution is a particularly feasible approach for the improvement of its properties which opens up new and attractive fields of applications for modified PVC as ion-selective electrode membranes or ion-exchange columns, biosensors or in biomedical devices.

This work presents the development of a PVC-based antimicrobial polymer. Functionalization with amine bearing aromatic thiols in solution and posterior transformations are delineated along with their means of characterization via NMR and IR as well as DSC and STA. A special focus was laid on the introduction of quaternary amino functionalities into the polymer and the testing of their microbiocidic activity.

4-Aminothiophenol and 2-mercaptopyridine proved to be suitable agents for partial nucleophilic substitution of PVC, concurrent with only low degrees of dehydrochlorination. However, these new species exhibited lowered thermal stability upon modification. The testing of PVC containing 2-mercaptomethylpyridinium iodide groups according to JIS Z2801:2000 suggests a strong antimicrobial activity against gram-positive and gram-negative bacterial strains of *S. aureus*, *L. monocytogenes*, *E. coli* and *P. fluorescens*.

KURZFASSUNG

Im Laufe der letzten Jahrzehnte wurde intensiv an verschiedenen Möglichkeiten der PVC-Modifikation geforscht. Sie stellt das Rüstzeug dar um die dem Material innewohnenden Defekte und die daraus resultierenden Limitierungen in der Verarbeitung und Anwendung zu beseitigen. Um diesen Zweck zu erfüllen bietet es sich vor allem an durch nucleophile Substitution der Chloratome an um gezielt andere funktionelle Gruppen in das Polymer einzuführen. Durch die verbesserten Eigenschaften des modifizierten Polymers ergeben sich hiermit neue Anwendungsgebiete, wie z.B. als Säulenmaterial für die Ionenaustauschchromatografie, in der Biosensorik oder als Material für Gebrauchsgegenstände im biomedizinischen Bereich.

Die vorliegende Arbeit beschäftigt sich mit der Entwicklung eines antimikrobiell wirksamen Polymers. Sie beschreibt sowohl die Modifizierung von PVC mit Amin-funktionalisierten Thioaromaten und die damit verbundene Folgereaktion, als auch die Charakterisierung der so modifizierten Polymere. Dies geschieht durch Anwendung verschiedenster Methoden wie NMR und IR Spektroskopie, sowie thermischer Analysen mittels DSC und STA. Hierbei wurde das Augenmerk besonders auf die Einführung von quaternären Aminogruppen in das Polymer gerichtet, und in weiterer Folge deren Einfluss auf die biozide Wirkung des Materials untersucht.

Als überaus geeignete Reagenzien für die nucleophile Modifikation erwiesen sich 4-Aminothiophenol und 2-Mercaptopyridin die eine teilweise Substitution, mit nur einem sehr geringen Anteil an HCl-Abspaltung, ermöglichten. Allerdings wurde in beiden Fällen eine verringerte thermische Stabilität des modifizierten PVCs festgestellt. Die Tests zur Abschätzung der antibakteriellen Wirksamkeit, durchgeführt in Anlehnung an JIS Z2801:200, ergaben eine Aktivität gegen die gram-positiven und gram-negativen Bakterienstämme *S. aureus*, *L. monocytogenes*, *E. coli* und *P. fluorescens*.

Table of Contents

1. Introduction

1.1. Chemical Modification of Poly(vinyl chloride)

Poly(vinyl chloride), commonly known as PVC, is one of the most widely used polymers. It is the second largest manufactured thermoplastic resin by volume with a worldwide capacity of more than 31 million tons.[1] Since its discovery in the early 19th century by Regnault[2] it has become a commodity with a great diversity of applications.

These applications range from piping and construction products, flooring and coverings, to flexible cable and wires, packaging and more recently biomedical materials and devices such as tubing, catheters, blood bags or medical gloves.[1, 3]

In this field the material has to meet high standards. A major problem in healthcare throughout the world are medical device-associated infections. Implants or catheters are particularly susceptible to infections. It is reported that in the United States alone 50000 to 200000 patients are effected per year, with a fatality rate of more than 20%.[4] The microorganism which is generally associated with with these nosocomial infections is the gram-positive bacterium *Staphylococcus aureus* and gram-negative *Escherichia coli*.

In order to avoid the formation of bacterial films numerous attempts exist to improve bacterial resistance.[4] Surface grafting techniques or covalent immobilization of biologically active agents onto the polymer should ensure contact activity without the release of the biocide.

For PVC these necessary modifications are basically achieved through dechlorination via nucleophilic or radical substitutions, dehydrochlorination and also grafting polymerizations (cationic and free radical pathways).[5] By far the most studied chemical route is the nucleophilic substitution reaction which was investigated under various conditions: both wet and dry processes, in organic or aqueous media, solvent / non-solvent systems or melts.[5]

So as to lower the risk of the competing dehydrochlorination one has to choose polar nucleophiles with a low basicity. Amongst others, aromatic thiols have proved to be appropriate

[1] Braun, D. *J. Polym. Sci. A Polym. Chem.* **2004**, 42, 578.

[2] Regnault, H. V. *Liebigs Ann*. **1835**, 14, 22.

[3] Wilkes, C. E. (ed.), with contributions by Berard, M. T. *PVC handbook*, Hanser, Munich, **2005**, chap. 17, p. 619-620.

[4] Lakshmi, S.; Kumar Pradeep, S. S.; Jayakrishnan, A. *J. Biomed. Mater. Res.* **2002**, 61, 26.

[5] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 303-331.

agents leading to a polymer with improved properties.[6] This group opens up the field for new attractive modifications of PVC, which might also cover biologically active formations with promising applications for biomedical purposes as well as for their use in packaging or piping.

1.2. Scope of this Work

The objective of this work was to determine an appropriate pathway of post-polymerization modification of PVC using thiol-based reagents, which enable a partial substitution of the chlorine atoms. In this context emphasis was laid on the introduction of amino functionalities that form the basis for another transformation reaction yielding an antimicrobial polymer.

In connection with that, different means of characterization such as NMR and IR spectroscopy, as well as calorimetric methods were applied to determine new polymeric features in addition to the testing of their antibacterial efficiency.

[6] Herrero M., Tiemblo, P.; Reyes-Labarta, J.; Mijangos, C.; Reinecke, H. *Polymer* **2002**, 42, 2631-2636.

2. General Aspects

2.1. Modification of PVC: Insights

One of the main goals of numerous research studies on poly(vinyl chloride) is the improvement of its low thermal stability and enhancement of mechanical properties. These peculiarities necessitate the usage of organic (cf. **Figure 1**) and inorganic thermal stabilizers, most importantly metal soap based Pb-, Ba-, Zn-, Ca- or Sn-stabilizers.[7]

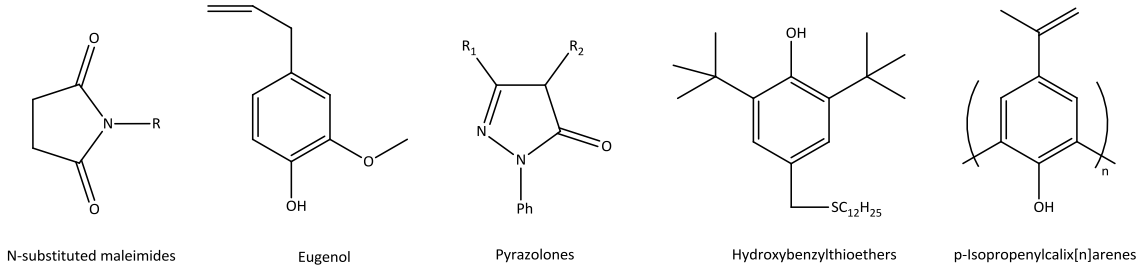


Figure 1: Novel organic PVC stabilizers[8]

Another possibility is the undertaking of chemical modifications of anomalous, that are, allylic and tertiary chlorine atoms, and the normal secondary ones. These anomalous, also referred to as labile chlorines are inherent structural defects which originate from PVC's classical synthesis via radical polymerization.

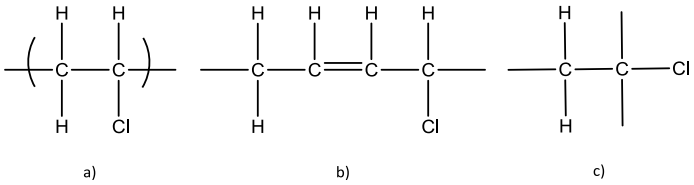


Figure 2: Normal *a)* and anomalous units *b), c)* of PVC

These units are believed to be the major source for PVC's poor thermal stabilities, as they are the starting points for the occurrence of the dehydrochlorination "zipper" reaction[9]. Evolving

[7] Röhrl, E. *PVC Taschenbuch*, Hanser, Munich, **2007**, chap. 3.1, p. 12.

[8] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 305.

[9] Starnes Jr, W. H. *J. Polym. Sci. A Polym. Chem.* **2005**, 43, 2452.

HCl catalyzes the elimination of HCl in the next monomeric unit and thus generates conjugated polyene sequences. Eventually these units undergo secondary degradation reactions like scission and crosslinking. As a result the color, crystallinity, processing and mechanical properties of the polymer are altered.[10]

Considering these adverse effects it is not surprising that a lot of chemical transformations have been performed, basically through dechlorination reactions. These processes include both substitution and elimination reactions, achieved via nucleophilic and radical pathways as well as grafting polymerizations, either via cationic or free radical types (ATRP, LCRP).

2.1.1. **Insights – Nucleophilic Substitution**

Nucleophilic modifications can be realized under different conditions: in organic media as well as in aqueous systems, in swollen states, phase transfer catalyses, solvent/non-solvent systems, UV irradiation, microwave plasma and by ultrasound means. The competing forces of substitution and elimination require carefully selected parameters in order to shift the equilibrium towards substitution products. Factors affecting the substitution in wet systems, apart from the type of nucleophile, are temperature or duration of the reaction and very importantly the solvent used.[11]

Kameda investigated the ratio of dechlorination / substitution for ethylene glycol (EG) and N,N-dimethylformamide (DMF) for five nucleophiles: OH^-, SCN^-, N_3^-, phtalimide anion and I^-.[12] In EG dechlorination could be achieved up to 98% but degrees of substitution were very low with 25, 21.5, 21, 6 and 1.5% (in the same order as listed above). For DMF the outcome was different: lower degrees of dechlorination (11-55%) but with a higher extent of substitution, as this solvent is known to be favoring S_N2 reactions. Moreover the order of preferential substitution is not corresponding to the Pearson nucleophilicity which appears in the order of $I^- > SCN^- > OH^- > N_3^-$ > phtalimide anion. The fact that a nucleophile like I^- is also a good leaving group, which leads to elimination of HI, can explain this effect.

[10] Endo, K. *Progr. Polym. Sci.* **2002**, 27, 2036.

[11] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 307.

[12] Kameda, T.; Ono, M.; Grause, G.; Mizoguchi, T.; Yoshioka, T. *Polym. Degrad. Stab.* **2009**, 94, 107-112.

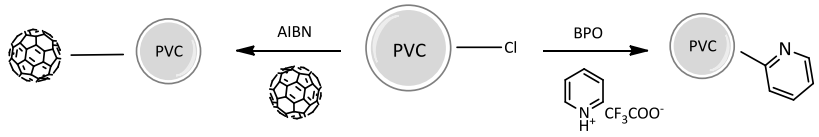
When basicity prevails over nucelophilicity a polyacetylenic (PA) material can be produced, where dehydrochlorination occurs almost exclusivelyto 98%.[13] After sulfurizing this polymer it exhibited electroconducting properties and could be tested as a cathode material for lithium batteries.

One interesting feature of PVC substitution is the regioselectivity of this reaction. There seems to be a predominant substitution of mainly isotactic and heterotactic regions, whereas syndiotactic are not so readily effected. Extensive work of Spanish researchers deals with the influence of the nucleophile's nature on the selectivity of the modification.[14] Accordingly they demonstrated a more pronounced stereospecific substitution with bulky and moderately reactive nucleophiles.

The change of the glass transition temperature, occurring after modification, can be attributed to these microstructural characteristics.

2.2. PVC-Modification: C_{PVC}-C

Some ways of attaching organic groups like pyridine, fullerenes, acrylates and allyl groups to PVC are described below. Reported degrees of modifications were in general below 10%. Pyridination[15] and substitution with fullerenes[16] could be achieved radically with BPO or AIBN acting as radical initiators. C_{60}-PVC revealed considerable electron acceptor abilities during electrochemical investigations of this material.



Scheme 1: Radical substitution of PVC

[13] Guo, L.; Shi, G.; Liang, Y. *Polymer* **2001**, 42, 5581-5587.

[14] Martínez, G.; Millán, J. L. *J. Polym. Sci. A Polym. Chem.* **2004**, 42, 1857-67.

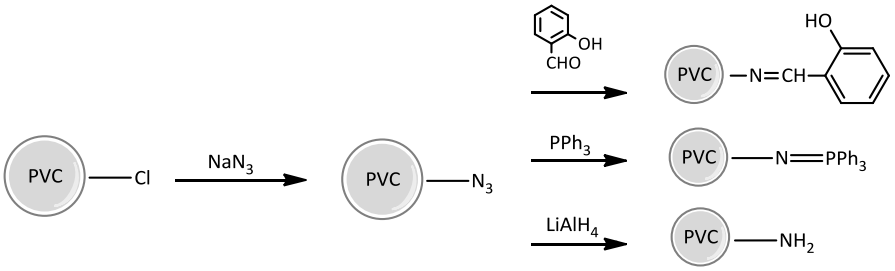
[15] Moulay, S.; Zeffouni, Z.; *Chin. J. Polym. Sci.* **2007**, 25, 297-302.

[16] Martínez, G.; Gòmez, M. A.; Gòmez, R.; Segura J. L. *J. Polym. Sci. A Polym. Chem.* **2007**, 45, 5408-5419.

However a more prevalently performed modification is grafting polymerization either via a cationic pathway or various techniques of living radical polymerizations (CM-LRP, CM-ATRP, SI-ATRP, SI-CLRP). An approach by Bicak et al.[17] observed an internal plasticization effect of the grafted polyacrylates, attached via copper-mediated atom transfer radical polymerization. This effect resulted in a decrease of the glass transition temperature of PVC. Surface-modified poly(hydroxyethyl acrylate) PVC has an interesting application as chelating resin for the extraction of heavy metal ions such as Cu(II), Hg(II), Zn(II) and Cd(II) in aqueous systems. After hydrolyzation the PAA-PVC exhibited a pH-dependent metal loading.[18]

2.3. PVC-Modification : C_{PVC}-N

The modification of PVC via nitrogen, sulfur or oxygen bonding can be achieved more easily than the formation of C_{PVC}-C. Especially the azidation of PVC has received much attention. First of all because PVC-N_3 can be subsequently transformed into a PVC-NH_2 species.



Scheme 2: Azidation of PVC and posterior transformation

PVC-N_3 itself can be synthesized through the nucleophilic substitution with sodium azide in DMF.[19] Either reduction with $LiAlH_4$ or the reaction of iminophosphorane-bearing PVC with ammonium hydroxide yields the corresponding PVC-NH_2.[20] As another example PVC-Schiff bases can be used for complexing heavy metal ions.[20]

[17] Bicak, N.; Ozlem M. J. *Polym. Sci. A Polym. Chem.* **2006**, 44, 1900-1907.

[18] Liu, P.; Liu, Y.; Su, Z. *Ind. Eng. Chem. Res.* **2006**, 45, 2255-2260.

[19] Rusen, E.; Marculescu, B.; Butac, L.; Preda, N.; Mihut, L. *Fuller Nanot. Carb. Nanostruct.* **2008**, 16, 178-185.

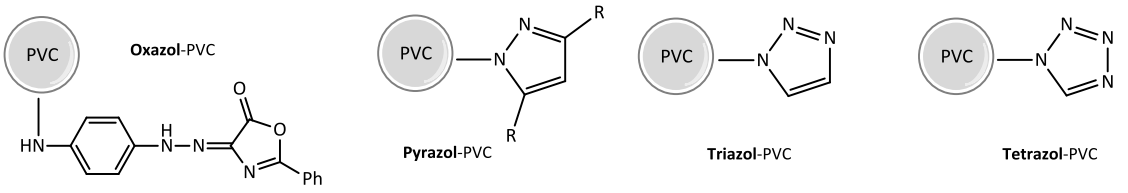
[20] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 308, 316.

In another pathway[21] the azidation of PVC-films could be achieved in a solvent/non-solvent mixture of H_2O/DMF with TBAB as a phase transfer catalyst. Resulting azidated surfaces were also subjected to bacterial adhesion tests. Earlier reports showed that gram-negative bacteria are inhibited in the presence of azide groups, whereas gram-positive develop normally.[22] In contrast azidated PVC-samples showed a reduced adhesion to both gram-negative *E. coli* and gram-positive *S. aureus* bacterial strains.

Several examples of N-modified PVC have also been reported as biocompatible materials, which can be applied for biomedical devices, such as prostheses, implants, orthopedic replacements, catheters or tubing.

For the aim of improving the blood compatibility of PVC a photochemical method for the surface modification with a biopolymer-derivative, namely O-butyrylchitosan, was successfully applied.[23] The surface of PVC was crosslinked with O-butyrylchitosan via an amino linkage. Increased hydrophilicity of the material circumvents the formation of clots within the blood as a result of anti-platelet adhesion. Also the immobilization of another protein and cell repelling agent, poly(ethylene glycol) induced hydrophilicity on the PVC surface, which affected antithrombogenicity in a positive extent.[24]

Nucleophilic substitution was performed as well with triazole and tetrazole metal salts, under optimum conditions with a degree of 72% for tetrazoles and remarkably 88% for methylated triazoles. Naturally the nature of the counterion, solvent and temperature affect reaction characteristics.[25] Other heterocyclic compounds which could be attached to PVC are oxazolinones and pyrazoles.



Scheme 3: PVC - nucleophilic modification with N-heterocyclic compounds

[21] Lakshmi, S.; Kumar Pradeep, S. S.; Jayakrishnan, A. *J. Biomed. Mater. Res.* **2002**, 61, 26-32.

[22] Snyder, M.L.; Lichstein, H.C. *J. Infect. Dis.* **1940**, 67, 113-115.

[23] Mao, C.; Zhao, W. B.; Zhu, A. P.; Shen, J.; Lin, *S. C. Process Biochem.* **2003**, 39, 1151-1157.

[24] Balakrishnan B.; Kumar, D. S. Yoshida Y.; Jayakrishnan, A. *Biomaterials* **2005**, 26, 3495-3502.

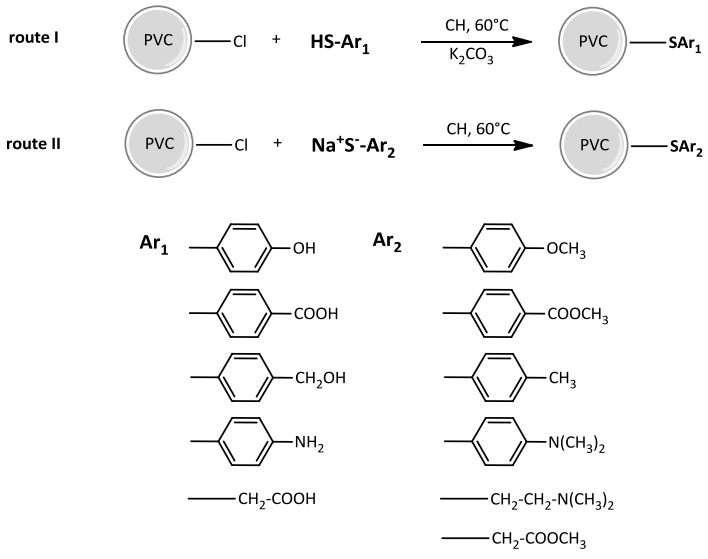
[25] Kruglova, V. A.; Dobrynina, L. M.; Vereshchagin, L. I. *Polym. Sci. Ser. A* **2007**, 49, 407-411.

PVC can also be used as a catalyst support. Tetraethylenepentamine-PVC, a cross-linked material could be used in Knoevenagel condensations of aldehydes or ketones with a C-H acidic species. The polymer-amine-complex was still catalytically active even after its fifth recovery.[26] Immobilization of nanopalladium on aminoethylated PVC served as a valuable catalyst for Heck reactions (aromatic vinylation). Selectively only trans-configurated products were obtained with yields between 62-90% and no loss of activity or selectivity could be observed during six cycles of re-use.

A novel concept of an ionic polymeric membrane electrode was developed by Varga and his co-workers.[27] Partial substitution of the chlorine atoms with triethylamine was achieved in a simple one-step procedure, producing a PVC with quaternized ammonium groups. This surfactant-selective polymer electrode exhibited a better long-time stability, selectivity with respect to the surfactant and shorter response time.

2.4. PVC-Modification: C_{PVC}-S

In the past years the Spanish group around Reinecke excelled in research of substitution with thiol modifiers. Preparation of the thiolate nucleophiles can be achieved either in a prior step through formation of a sodium salt, or in situ with K_2CO_3.



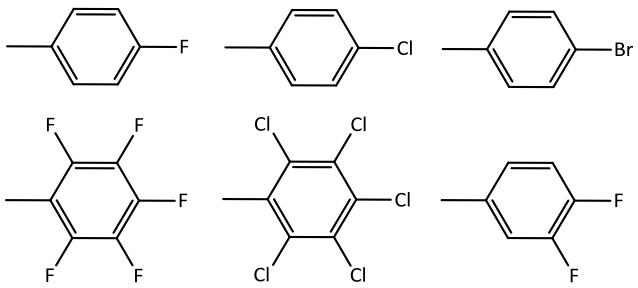
Scheme 4: PVC - nucleophilic modification with thiols

[26] Dong, F.; Li, Y. Q.; Dai, R. F. *Chin. Chem. Lett.* **2007**, 18, 266-268.

[27] Varga, I.; Mészáros, R.; Szakács, Z.; Gilányi, T. *Langmuir* **2005**, 21, 6154-6156.

A report which was published in 2002 by this group, investigated the influence of thioaliphatic and thioaromatic bifunctional modifiers on the final properties of the modified PVC.[28] Those nucleophiles that contained functionalities with mobile hydrogen (prepared according to route I) implied hydrogen bonding within the polymeric chains, whereas species without mobile hydrogen (prepared according to route II) consequently do not. It was confirmed that use of aliphatic thiols lead to much lower degrees of modification (<10%) than that of aromatic ones (32 – 68% under analogous conditions without elimination) and was accompanied by undesirable elimination of HCl. The degrees of substitution for those thiols with non-protic functional groups were twice as high as for those who carry a protic functionality. This was attributed to hydrogen bonding between the polymer chains. Hydrogen bonding is also responsible for a considerable increase of the glass transition temperatures, due to the newly introduced stiffness of the chains. Independent of the hydrogen bonding-capability bulky moieties resulted in larger free fractional volumes, a parameter influencing the gas transport characteristics of a polymeric material.

Employing the same substitution procedures for mono- and poly- halogenated thioaromatics the impact on glass transition temperature and thermal stability of the new compounds was studied.[29]



Scheme 5: PVC - nucleophilic modification with halogenated thioaromatics

Again aromatic thiols proved to be selective modifiers, under avoidance of elimination and with high degrees of modification up to 66%. The thermal stabilities of modified PVC containing chlorine or bromine atoms showed no significant improvement, compared to pure PVC. Degradation onset temperatures were determined between 256 – 274°C, for PVC it was 275°C.

[28] Herrero, M.; Tiemblo, P.; Reyes – Labarta, J.; Mijangos, C.; Reinecke, H. *Polymer* **2002**, 43, 2631-2636.

[29] Navarro, R.; Bierbrauer, C.; Mijangos, C.; Goiti, E.; Reinecke, H. *Polym. Deg. Stab.* **2008**, 93, 585-591.

Only when the modifier contains fluorine atoms decomposition starts not until a temperature point between 295 – 322°C is reached. All halogen modifiers obviously acted as internal plasticizers indicated by reduced values for the T_g. Fluorine containing polymers might be of practical interest because of their thermal stability and chemical resistance. PVC modified with pentachlorothiophenol might serve as efficient biocide. Low molecular pentachlorophenol is known as an effective pesticide and fungicide, thus already applied as an additive in PVC. Covalent immobilization onto the polymer would be welcome, as components of low molecular weight tend to migrate out of the polymer, which results on the one hand in the loss of its microbicidal properties and on the other hand it poses a danger for the environment or could lead to health problems.

Not only N-modified PVC polymers are feasible for application in the biomedical field. In a report of Grohens the adhesion properties of the two bacterial strains *Escherichia coli* and *Staphylococcus aureus* on S-modified PVC are examined.[30] Various types of modifiers were studied, namely 4-mercaptophenol, 4-mercaptobenzylalcohol, 4-aminothiophenol, 4-methoxybenzenethiol, 4-mercaptobenzoic acid methyl ester, 2-naphtalenethiol and 4-mercaptopyridine. Adhesion properties were determined by wettability measurements, captive bubble time-dependent measurements (both interfacial tension measurements) and static adhesion tests. However, the hydrophobic bacterium *S. aureus* showed no difference in its adhesion to pure or modified PVC, irrespective of the modifying group. In general the adhesion of *S. aureus* (θ_{water}=75°) was stronger than that of the hydrophilic *E. coli* species (θ_{water}=29°). Compared to pure PVC the adhesion of *E. coli* on all modified PVC films was remarkably reduced about 30-50%. These results were thought to be the result of an acid-base interactions between *E. coli* (γ_S^-= 89.1 mJ/m^2, with γ_S^- as the electron-donating solid-surface energy) as a base and the polymer samples, either acids or bases. The high retention of *S. aureus* was attributed to its low basicity (γ_S^-=15.8 mJ/m^2).

As already indicated above, the increase of the fractional free volume (inverse of the density or the specific volume) upon introduction of rather bulky groups changes the gas transport properties of the polymeric material in its use as gas permeation membrane. For PVC a higher degree of diffusion and permeation was determined after pyridination with 4-mercaptopyridine

[30] Herrero, M.; Quéméner, E.; Ulvé, S.; Reinecke, H.; Mijangos, C.; Grohens, Y. *J. Adhesion Sci. Technol.* **2006**, 20, 183-195.

for gases like O_2, N_2, CO_2.[31] A strong increase of these parameters due to the degree of modification could only be observed up to 21% pyridination and started to decrease at higher values of 46%. Also T_g values increased linearly with conversion up to 20% with almost no changes occurring between 30 - 70% modification.

2.5. PVC-Modification: C_{PVC}-O

Epoxy-PVC might be of a potential use as costabilizer or as a photocurable resin.[32] It can be obtained by epoxidation of dehydrochlorinated PVC (DPVC), as described by Szakákcs.[33]

Preparation of PVC-ethers with high yields between 50 – 83% could be achieved with mono- di- or aromatic alcohols.[34] This modification was performed without remarkable dehydrochlorination, which was attributed to the presence of the green bentonite-based catalyst “Maghnite-K”. Another group crosslinked PVC through reaction with sodium ethylene glycoxide, under avoidance of elimination.[35] This new material was then used to prepare an ion-exchange resin by encapsulation of silica gel particle and immersion in tetramethylammonium hydroxide.

2.6. PVC-Modification: C_{PVC}-Hal

Generally PVC halogenations were mainly performed via chlorination and iodination. Chlorinated PVC (CPVC) exhibits enhanced thermal properties, due to the alliviation of reactive structural defects. Its improved fire retardance efficiency and corrosion resistance can be explained by the higher chlorine content, which also increased the T_g remarkably (106 - 115°C). CPVC can be obtained by irradiation or a thermally induced free radical method.[36] Nevertheless degradation via dehydrochlorination still necessitates the incorporation of stabilizers into the polymer.

[31] Tiemblo, P.; Guzmán, J.; Riande, E.; Mijangos, C.; Reinecke, H. *Macromolecules* **2002**, 35, 420-425.

[32] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 308; 326.

[33] Szakáks, T.; Iván, B. *Polym. Degrad. Stab.* **2004**, 85, 1035-1039.

[34] Mekki, H.; Belbachir, M. *eXPRESS Polym. Lett.* **2007**, 1, 495-498.

[35] Bahaffi, S. O. S.; Abdelaal, M. Y.; Assirey, E. A. *Int. J. Polym. Mater.* **2006**, 55, 477-484.

[36] Moulay, S. *Progr. Polym. Sci.* **2010**, 35, 308; 327.

Green iodination with KI was reported in an aqueous/PTC system.[37] Substitution occurred to an extent of 27% (based on weight gain) and lead to lower thermal stability, as the decomposition onset was observed between 264 - 271°C that of PVC between 324 - 327°C. Iodinated PVC was not stable in aqueous environment, which was determined by UV spectroscopy after incubation in water for 27 days.

[37] Lakshmi, S.; Jayakrishnan, A. *J. Appl. Polym. Sci.* **2002**, 84, 493-499.

3. Results and Discussion

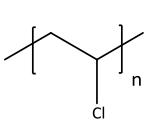
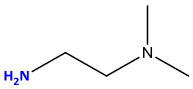
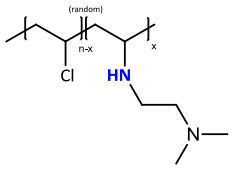
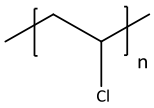
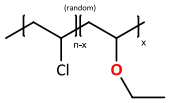
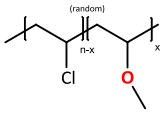
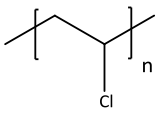
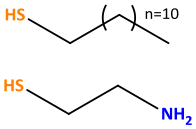
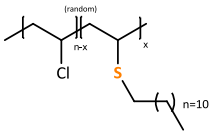
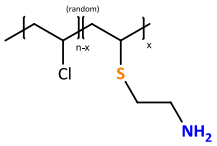
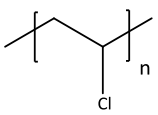
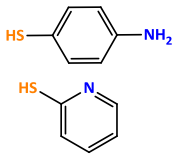
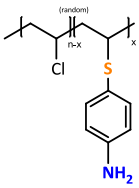
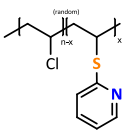
The general objective of the preparative work was to find a pathway for quantitative dechlorination of PVC via a nucleophilic substitution. Optimized reaction conditions should lead to selective modification with a preferably low degree of elimination side reactions and should ideally keep the input of solvents and reactants low. In a second step it was headed for the introduction of antimicrobial activity through quaternary amino functionalities.

In the following the syntheses of PVC containing amino functionalities are disclosed together with their various means of characterization via NMR and IR spectroscopy, DSC and STA for determination of their thermal properties and testing of microbicidal effects according to JIS Z2801:2000.

3.1. Substitution of PVC

As it was already presented in chapter 2.1.1, PVC can be modified with various types of modifiers. In this case the substitution via the formation of C_{PVC}-X with X= N, O, S was tested with selected reactants, adopting previously reported protocols.[38, 39, 40]

Table 1: Various nucleophilic substitution reactions tested on PVC

starting material	reactant	desired product	conditions	*conv.*
			*H_20, 80°C*	***no***
			H_20 / acetone, 70°C	***no***
			THF, r.t.	***no***
			*THF, r.t.*	***no***
			*K_2CO_3/18-crown-6-ether, 2-Butanone, 70°C*	***no***
			K_2CO_3, 2-Butanone, 70°C	***no***
			K_2CO_3, 2-Butanone, 70°C or Cyclohexanone, 60°C	***no***
			K_2CO_3, Cyclohexanone, 60°C	***no***
		 	K_2CO_3, Cyclohexanone, 60°C	***yes***

[38] Mekki, H.; Belbachir, M. *eXPRESS Polym. Lett.* **2007**, 1, 495-49

[39] Herrero, M. et al. *Polym. Degrad. Stab.* **2006**, 91, 1915-1918

[40] Navarro, R.; Pérez, M.; Rodriguez, G.; Reinecke, H. *Europ. Polym. J.* **2007**, 4516-4522

However most of the reactions did not lead to the desired products. Generally basicity prevailed over nucleophilicity and therefore causing dehydrochlorination instead of substitution.

Only the modification with thiolates could be achieved successfully with minimal elimination as the side reaction. Especially aromatic compounds exhibited favorable nucleophilicity.

3.2. Substitution of PVC with 4-Aminothiophenol

It has already been stated in the past that aromatic thiols turn out to be appropriate modifiers in order to achieve post–functionalization via nucleophilic substitution of the polymer.[41,42,43,44] Such modifications are reported to be extremely selective with respect to the mercapto group. Balanced nucleophilicity and basicity lead to high degrees of modifications without undesired dehydrochlorination.[45]

Since para – substituted agents exhibit favorable inductive and mesomeric effects, they are particularly appropriate modifiers for this type of reactions, which has been also reported previously by the same group.[44] Therefore 4-aminothiophenol was chosen as a modification reagent.

3.2.1. **Preparation of PVC–Thioaniline**

The procedures were carried out following a known protocol.[46] All chemicals were used as purchased without further purification. Commercially available PVC with average molecular weights of M_n = 22 000 / M_w = 43 000 was used in all reactions, unless it was stated otherwise.

As a "good" solvent for PVC cyclohexanone was used in all reactions. The attacking nucleophilic thiolate species was created in situ with potassium carbonate functioning as base.

[41] Reinecke H., Mijangos C. *Polym. Bull.* **1996**, 1, 13 - 18

[42] Navarro, R.; Bierbrauer, C.; Mijangos, C.; Goiti, E.; Reinecke, H. *Polym. Deg. Stab.* **2008**, 93, 585-591

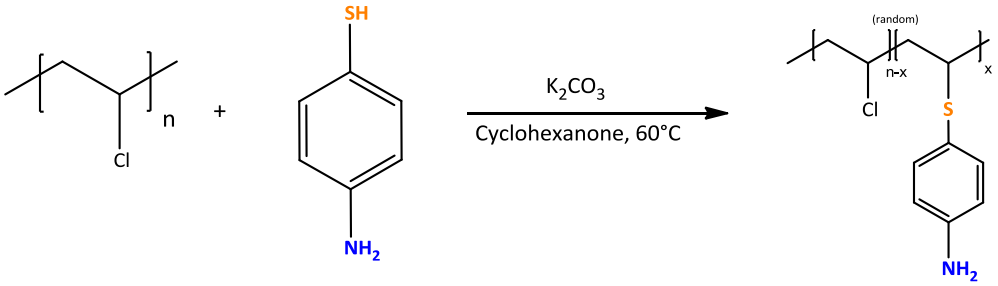
[43] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2199 - 2204

[44] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2291 – 2294

[45] Herrero, M.; Tiemblo, P.; Reyes – Labarta, J.; Mijangos, C.; Reinecke, H. *Polymer* **2002**, 43, 2631

[46] Navarro, R.; Bierbrauer, C.; Mijangos, C.; Goiti, E.; Reinecke, H. *Polym. Deg. Stab* **2008**, 93, 586

Hereby reaction conditions of 60°C of temperature and inert atmosphere of argon were employed. Reaction times range from 3 h to 68 h.



Scheme 6: Nucleophilic substitution of PVC with 4-Aminothiophenol

Precipitation in the polar solvent mixture of methanol and water (9:1 v/v) and vacuum drying yielded yellowish and rather brittle products.

A more detailed version of the procedure can be found in the experimental part of this work.

A crucial point in these modification reactions was the choice of solvent. Not all solvents favor nucleophilic substitution reactions and for PVC itself the choice of "good" solvents is limited. This fact is illustrated by several attempts using THF as the solvent. Both the syntheses under ambient conditions, as well as a microwave – assisted approach at higher temperature and pressure (120°C, 4bar, 30 min) were unsuccessful, as judged by interpreting the corresponding spectra.

As already mentioned above and described below, the usage of cyclohexanone enables a simple and successful reaction. Although the high affinity of the solvent's C=O group with the polar C-Cl unit is the main reason for PVC's good solubility[47], this might also be a disadvantage when it comes to purification. In order to ensure effective removal of solvent residues from the polymer, extensive drying steps are further necessary.

[47] Wilkes, C.E. (ed.), with contributions by Summers, J.W. *PVC handbook*, Hanser, Munich, **2005**, chapt. 1, p.6

3.2.2. **Characterization**

NMR spectroscopy

^{1}H NMR

NMR measurements provide information about a successful course of reaction and are a good tool for determining the extent of functionalization.

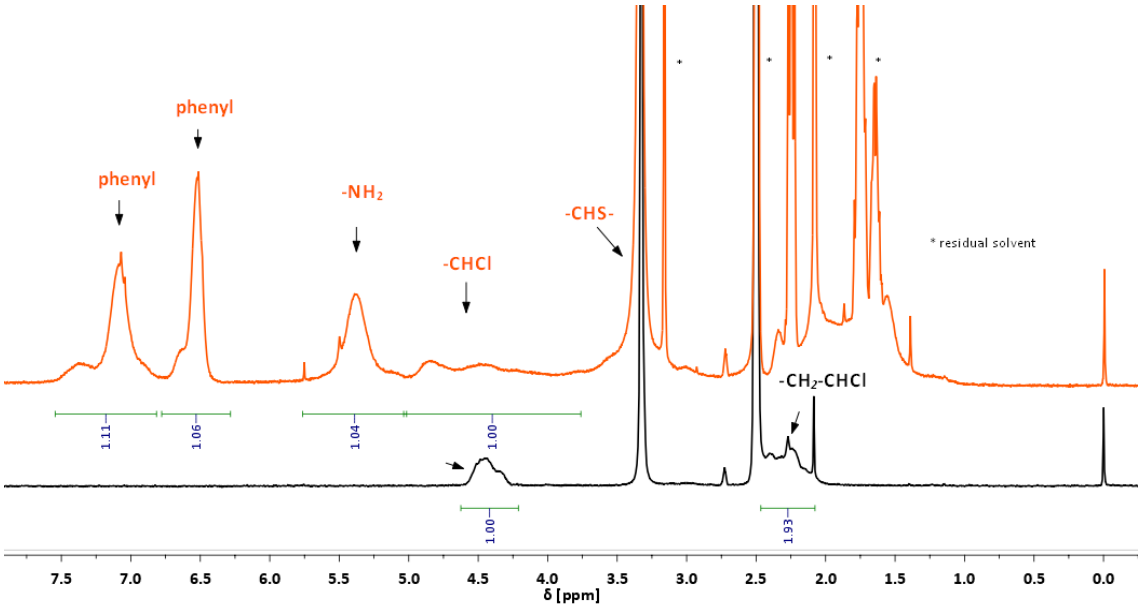


Figure 3: ^{1}H NMR spectra (300 MHz, DMSO-d_6) of PVC and PVC-thioaniline

Figure 3 shows a corresponding ^{1}H NMR spectrum of PVC before (black) and after the reaction with 4-aminothiophenol (orange). The characteristic bands representing the four aromatic protons arise in the region between 7.30 – 6.75 ppm as well as 6.70 – 6.35 ppm. In comparison with published data[48], the signal at 5.3 ppm can be assigned to the amino protons. This proposition is supported by the integral ratio of 2:1 of the aromatic to the amino protons. The original CH-Cl signal (4.55 – 4.25) is considerably broadened and shows an additional peak at 4.8 ppm. As stated elsewhere[49] a new resonance band of those protons, which are attached to

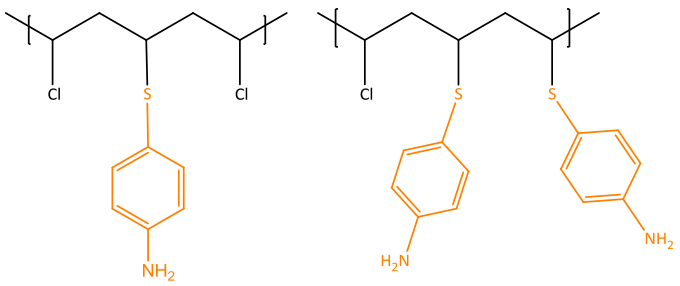
[48] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2292

[49] Mijangos, C.; López, D. *Macromolecules* **1995**, 28, 1365

thiophenol – substituted carbons, occur in the region between 4.0 and 2.8 ppm. This signal is unfortunately partially covered by the H_2O peak.

Additionally the region between 2.6 – 1.8 ppm, belonging to the methylene protons of the – CH_2- units, broadens upon modification, thus ranging from 2.6 – 1.0. Again solvent peaks of cyclohexanone superpose the signal.

Regarding the resonance bands of the aromatic protons additional shoulders appear beside the more pronounced peaks at 7.1 and 6.4 ppm. Published work[50] suggests two different interpretations for the occurrence of this second type of aromatic protons. Firstly it might originate from the distribution of the 4-aminothiophenolic units throughout the PVC chains. For instance, two vicinal positioned aminothiophenol units give rise to different proton signals than a thioaniline group, that finds two chlorines as a neighbor.



Scheme 7: Possible distribution of 4-aminothiophenol along the PVC chain

Another explanation can be the stereoselective nature of this nucleophilic substitution reaction.[51, 52] The formation of a second type of peaks might be related to the tacticity of the PVC chain, meaning that at first only isotactic and heterotactic chlorines react before the syndiotactic do.[53]

In the course of this work no such studies were performed in order to confirm these interpretations in the case of 4-aminothiophenol.

[50] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2201 - 2202

[51] Mijangos, C.; Hidalgo, M. *Polymer* **1994**, 35, 348

[52] Millán, J.; Martínez, G.; Mijangos, C. *J. Polym. Sci., Polym. Chem. Ed.* **1985**, 23, 1077

[53] Lopez, D.; Hidalgo, M., Reinecke, H.; Mijangos, *C. Polym. Int.* **1997**, 44, 1

The degrees of substitution, expressed as mole per cent, can be determined via the ratio of vinyl thioaniline / (vinyl thioaniline + vinyl chloride). Here the integrals of the thioaromatic protons and the C*H*-Cl protons respectively are the basis of these calculations.

Degrees of substitution were determined as follows:

- 15 – 20% for a reaction time of 18h using PVC of a higher average molecular weight (M_n= 67730, M_w = 182080)
- 30 – 35% for a reaction time of 18h using PVC of lower average molecular weight (as specified above)

IR spectroscopy

As a further characterization, FT-IR measurements were performed by emulsifying approx. 20 mg of the polymer sample in THF and drop casting it on CaF_2 plates. Characteristic peaks were assigned according to **Table 2** and **Table 3**.

Table 2: Characteristic IR vibrations of PVC-thioaniline *(cf. ref.[54])*

Functional group	v [cm-1]	Intensity	Comments
NH_2	3470, 3370, 3220	m	NH str
NH_2	1624	vs	NH def
C-C arom.	1600, 1500	vs	str
C-N	1180	m	str
C-H arom.	820	m	def, p-substituted aromatics

Table 3: Characteristic IR vibrations of PVC *(cf. ref.[55])*

Functional group	v [cm-1]	Intensity	Comments
CH_2 / CH	2974, 2910, 2866	s	str
CH_2	1435, 1426	s	def
CH	1330, 1252	m, s	CH in CHCl
C-C	1068	s	
CH_2	966, 910	m	str

[54] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2292

[55] Wilkes, C.E. (ed.), with contributions by Daniels, C.A., *PVC handbook*, Hanser, Munich, **2005**, chapt. 17, p.399

Exemplary spectra of a PVC-thioaniline sample (modification degree: 30 – 35%, determined by ^{1}H NMR spectroscopy) and pure PVC are depicted below.

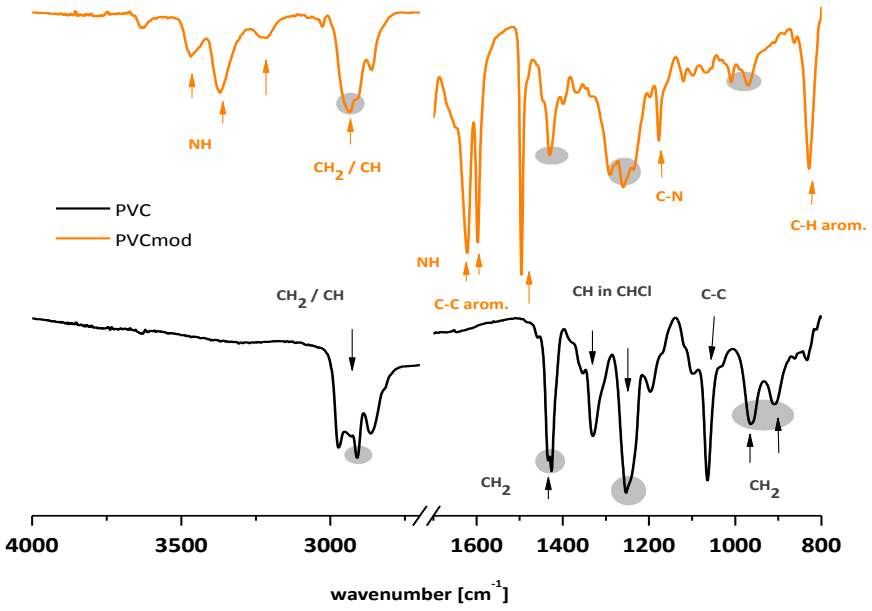


Figure 4: IR spectra of PVC and PVC-thioaniline

IR measurements indicate a selective reaction of the PVC chain with the thiol group of the modifier, since free amino groups are present after substitution. This observation is in accord with the interpretation drawn from the ^{1}H NMR spectra. The corresponding peaks appear between 3200 and 3500 cm^{-1}. Additional evidence for a successful modification are new vibrations of the aromatic ring structure that arise at lower wavenumbers and in the fingerprint region of the spectrum. Also, the original bands of the aliphatic CH_2 and CH stretching vibrations are shaped differently, which is due to an overlap with aromatic CH stretching vibrations of the thioaniline moiety. Comparison of the fingerprint regions below 1500 cm^{-1}, shows significant changes. Due to nucleophilic substitution of the chlorine, the relative intensity (if compared to the aliphatic vibrations between 3000 – 2800 cm^{-1}) of the CH (as CH in CHCl) vibration decreases and the peaks have slightly shifted. (see **Figure 4**)

DSC (Differential Scanning Calorimetry)

Calorimetric measurements were undertaken in order to investigate the influence of the modifier.

Table 4: Glass transition temperature as a function of substitution

Sample	T_g [°C]	DS [%]*[1]	DS [%]*[2]
PVC	**83**	0	0
PVC-thioaniline$_1$	**119**	15 - 20	35
PVC-thioaniline$_2$	**138**	30 - 35	50

*[1] DS - degree of substitution (as determined via ^{1}H NMR integral ratio)

*[2] DS – degree of substitution (as suggested by linear correlation of Tg and DS)[56]

Glass transition temperatures seem to increase with the degree of functionalization. Reinecke has determined a linear dependency and explains the new rigidity of the polymer system as a cause of chain-chain interaction via hydrogen bonding.[56]

Obtained values of 119°C and 138°C for the glass transition temperatures suggest an incorporation of 35% thioaniline in case of the sample exhibiting 15 – 20% according to NMR and a degree of 50% instead of 30 – 35%.

It is suggested that this deviation might be due to residual cyclohexanone enclosed in the polymer which would eventually result in a higher T_g as originally anticipated.

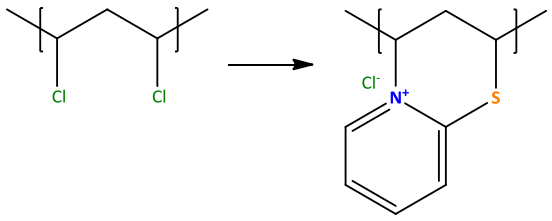
[56] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2293

3.3. Substitution of PVC with 2-Mercaptopyridine

In the further course of this work more focus was laid on the nucleophilic substitution reaction with 2-mercaptopyridine and its posterior transformation.

2-Mercaptopyridine exposes an aromatic thiol group, which should enable a quantitative substitution. Its tertiary amino functionality serves as a point of attack where biological activity could be introduced through the formation of quaternary ammonium groups.

A concurrent formation of an internally quaternized PVC-mercaptopyridinium species during substitution would be quite feasible as no second reaction step would be necessary. This might also have an economical benefit because less chemicals and solvents would be needed.



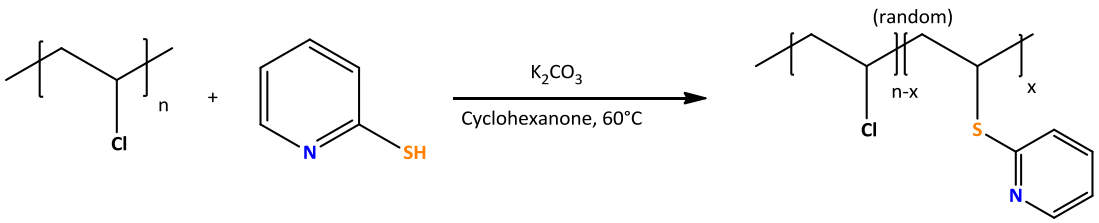
Scheme 8: Possible formation of an internal 6 –membered ring structure

The feasibility of formation of this structure is investigated through characterization of the nucleophilic substitution reaction products.

3.3.1. Preparation of PVC-Mercaptopyridine

Modification of PVC with 2-meraptopyridine was performed analogously to the procedure as described for 4-aminothiophenol.

Chemicals were again used as purchased without further purification. Commercially polymerized PVC with average molecular weights of M_n = 22 000 / M_w = 43 000, was used in all reactions.



Scheme 9: Nucleophilic substitution of PVC with 2-mercaptopyridine

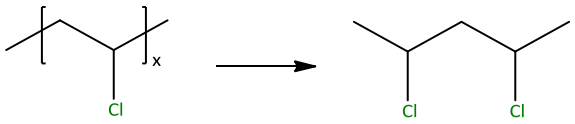
The purification procedure in order to remove residual cyclohexanone was improved by means of “extraction”. For this purpose the solid polymer species was stirred in diethylether several times and dried under vacuum.

A detailed version of the procedure is described in the experimental part.

3.3.2. **Reaction model**

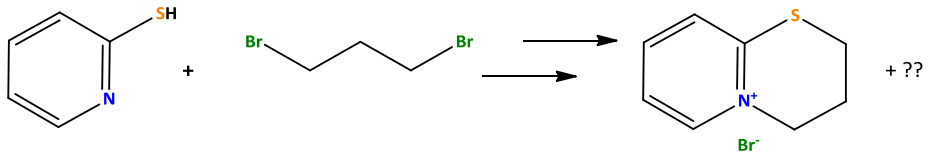
General aspects

Reaction behavior of 2-mercaptopyridine was studied on low – molecular basis in order to gain information about the formation of a 6-membered ring structure, leading to a positively charged molecule. Characteristic NMR signals of the reaction products are an additional help for interpreting polymer spectra.



Scheme 10: „Ideal“ PVC model substance

An “ ideal” PVC model substance to simulate “short-chain PVC” would be an internal alkyl dichloride with a unsubstituted $–CH_2-$ group in between. Instead, a dibromo alkane was used.

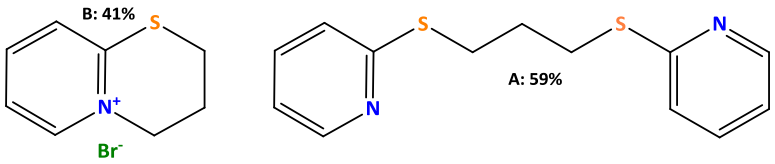


Scheme 11: Model reaction performed on 2-mercaptopyridine

The conversion of 2-mercaptopyridine with 1,3-dibromopropane or chlorocyclopentane was performed under the same experimental conditions as were applied for the nucleophilic substitution of the polymer, except the addition of K_2CO_3. These alkyl halides are more prone to nucleophilic substitution than a high-molecular polymer which makes the usage of base unnecessary. For a detailed description of the proceeding see chapter 5.2.4. Purification was unsuccessfully attempted by column chromatography in methanol / dichloromethane (1:1 v/v).

Characterization

Apparently the reaction of 2-mercaptopyridine with 1,3-dibromopropane leads to two products as indicated by the ^{1}H NMR spectrum.



Scheme 12: Reaction products

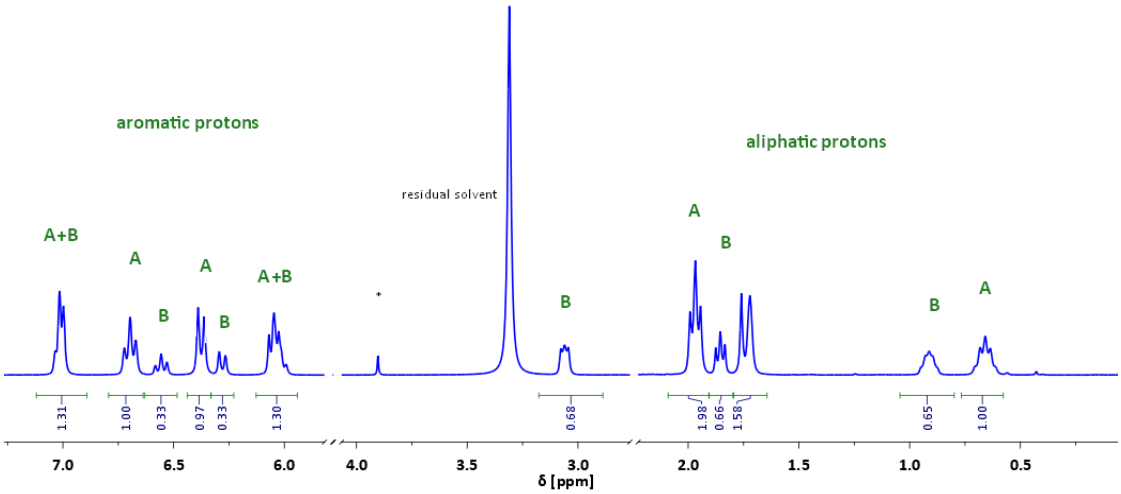


Figure 5: ^{1}H NMR spectrum (300 MHz, CD_3OD) of reaction product A /B

The formation of a bipyridine species confirms the highly nucleophilic character of the thiol moiety, as this product is favored in this reaction. A pyridinium species occurs nonetheless as the second product. A detailed listing of the corresponding peaks can be found in the experimental part.

Regarding the corresponding substitution of the polymer these observations on the low-molecular basis lead to the assumption that:

- Modification will be achieved preferentially via the thiol group
- The formation of a “self-quaternized” 6 – membered pyridinium ring is possible

3.3.3. **Characterization**

NMR spectroscopy

^{1}H NMR

In order to gain information about a successful modification and determine the degree of substitution with 2-Mercaptopyridine ^{1}H-NMR spectra were recorded. **Figure 6** compares both pure PVC and PVC upon conversion. A detailed listing of the signals can be found in the experimental part.

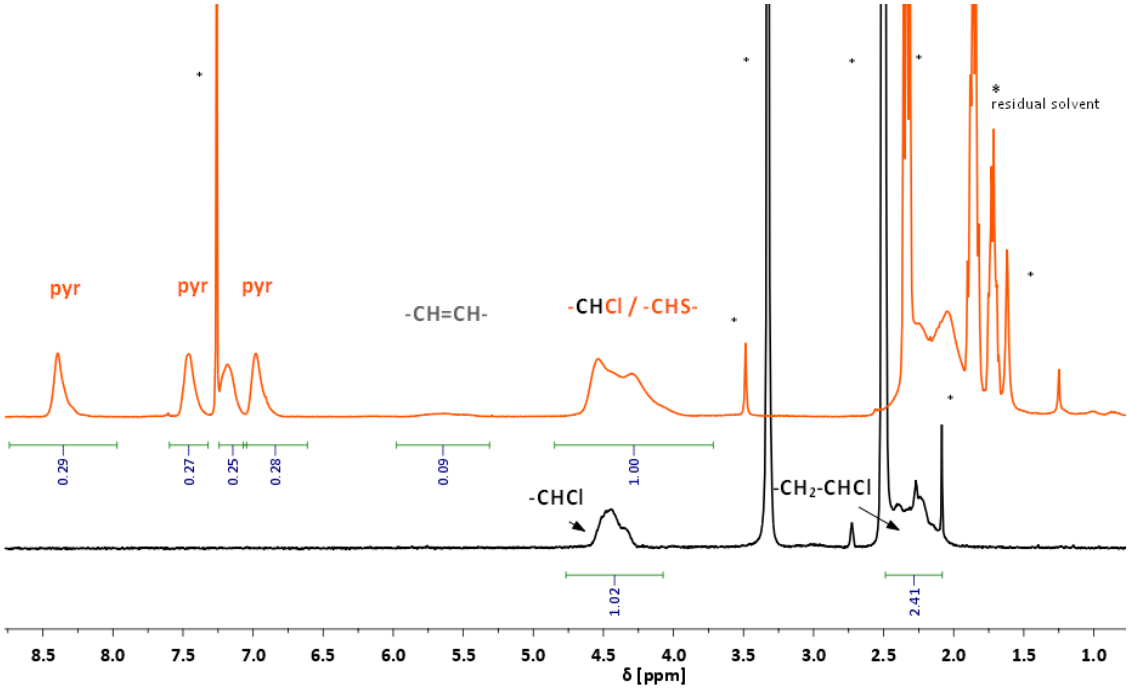


Figure 6: ^{1}H NMR spectra of PVC (300 MHz, DMSO-d_6) and PVC-mercaptopyridine (300 MHz, $CDCl_3$)

The four characteristic signals between 8.5 and 6.8 ppm can be attributed to the aromatic protons of the pyridine structure. According to spectroscopical investigations[57] and similar

[57] Mijangos, C.; López, D. *Macromolecules* **1995**, 28, 1365

reports[58] the thio-linkage accounts for the less deshielded protons of the –CH-S- fragment. The corresponding resonance band is therefore shifted highfield in comparison with the –CH-Cl protons. In this case these two signals seem to overlap, causing the original band at 4.6 – 4.25 ppm to broaden, wherefore the separate integration of each band is impossible.

At this point it has to be mentioned that solubilized mercaptopyridine structures also exist in a tautomeric 1*H*-pyridine-thion form.[59] These exhibit a rather basic character and consequently provoke dehydrochlorination within the polymer, which is proved by an evolving band between 5.85 – 5.30 ppm that can be assigned to the protons of –C*H*=C*H*-.

Regarding the possibility of an internal ring structure, as discussed in 3.3.2, these spectra do not indicate the occurrence of such formations. In this case one would anticipate an additional peak of the –CHN- protons, shifted to higher resonance frequencies, as it could be observed in the model reaction where the characteristic shift for $–CH_2$-N appears at 3.06 ppm and the corresponding $–CH_2$-S at 1.85 ppm. (see **Figure 5**).

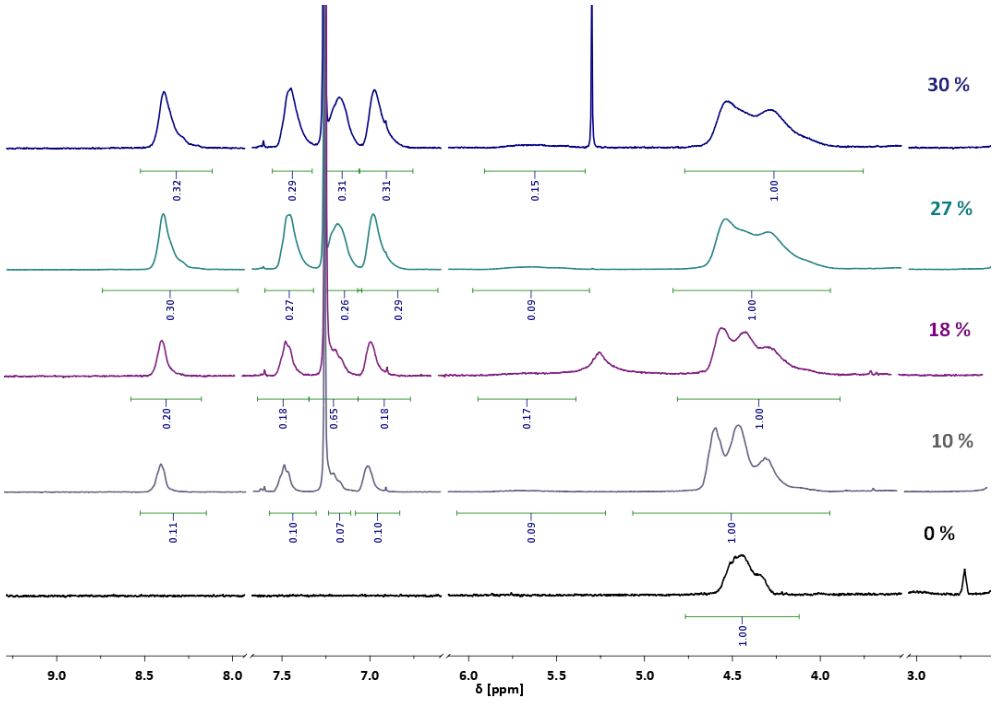


Figure 7: Evolution of ^{1}H NMR spectra (300 MHz, $CDCl_3$) of PVC upon conversion with 2-mercaptopyridine

[58] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2200

[59] Spinner, R. *J. Org. Chem.* **1958**, 23, 2037

With increasing degree of modification and accordingly reaction time not only the aromatic proton resonance bands are intensified, but also the alteration of the original -CHCl band can be demonstrated nicely. As already mentioned in the previous paragraph the CHS proton does not evoke a singular peak, as anticipated for the region between 4.0 – 3.4.[60,61] Instead the -CHCl and the –CHS- band overlap.

Degrees of substitution with 2-mercaptopyridine, expressed as mol per cent, were calculated via the ratio of (vinyl mercaptopyridine) / (vinyl mercaptopyridine + vinyl chloride) using the integral of the thioaromatic protons, the C*H*-Cl protons and taking the additional double bonds into account. The degree of substitution increases with reaction time as can be seen in **Figure 8**.

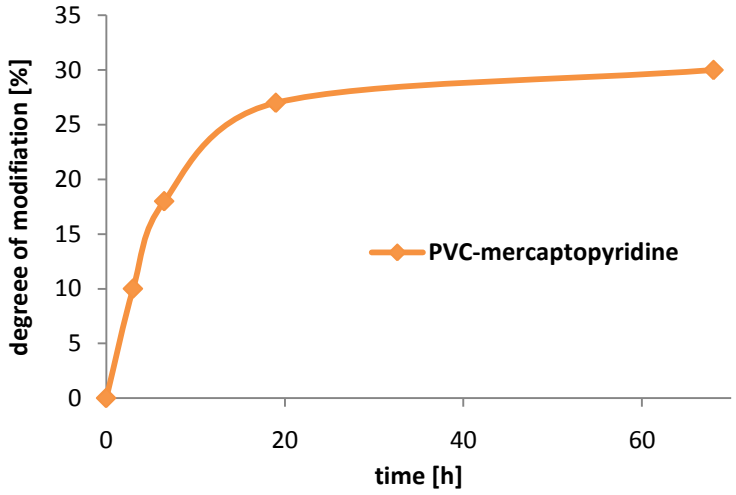


Figure 8: Dependence of degree of substitution on reaction time

Similar results have been published for PVC-thioaniline.[62] At the beginning the reaction proceeds quite fast and relatively high degrees of modification can be achieved after short reaction times. This also goes along with slightly increased degree of dehydrochlorination. By changing the molar ratio from 1:0.5 to 1:1 (PVC / PVC-mercaptopyridine) the reaction rate increases. (cf. **Table 5**)

[60] Mijangos, C.; López, D. *Macromolecules* **1995**, 28, 1365

[61] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2200

[62] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2293

Table 5: Overview – Degrees of substitution / elimination

Time [h]	DS [%]*	DE [%]**	Molar ratio (PVC / 2-mercaptopyridine)
3 - 6.5	10 - 18	5	1: 0.5
18 - 20	24 - 31	5	1: 0.5
68	30	7	1: 0.5
4 - 6.5	16 - 24	8	1:1

*DS – overall degree of substitution (as determined via ^{1}H NMR integral ratio)

**DE – overall degree of elimination (dehydrochlorination) (as determined via ^{1}H NMR integral ratio)

Obtained values for the degree of substitution were also compared with the results of elemental analysis.

Table 6: Comparison of the degree of substitution DS determined by elemental analysis and NMR

	Elemental analysis					NMR
Sample	**N [%]**	**C [%]**	**S [%]**	**H [%]**	DS[%]*	DS [%]*
A	1.24	36.3	3.92	4.42	9	10
B	4.11	49.9	9.96	5.24	23	27
C	4.73	49.7	11.3	4.97	29	30

*expressed as mole per cent

Thus the calculations based on the proton integrals of ^{1}H NMR spectra concur with the results determined by elemental analysis. The discrepancy of the results for sample B can be traced back to residual solvent, enclosed in the polymer which was submitted to elemental analysis.

^{13}C NMR

Successful nucleophilic substitution is also confirmed by the corresponding ^{13}C – NMR spectrum of PVC-mercaptopyridine (degree of substitution: 30%).

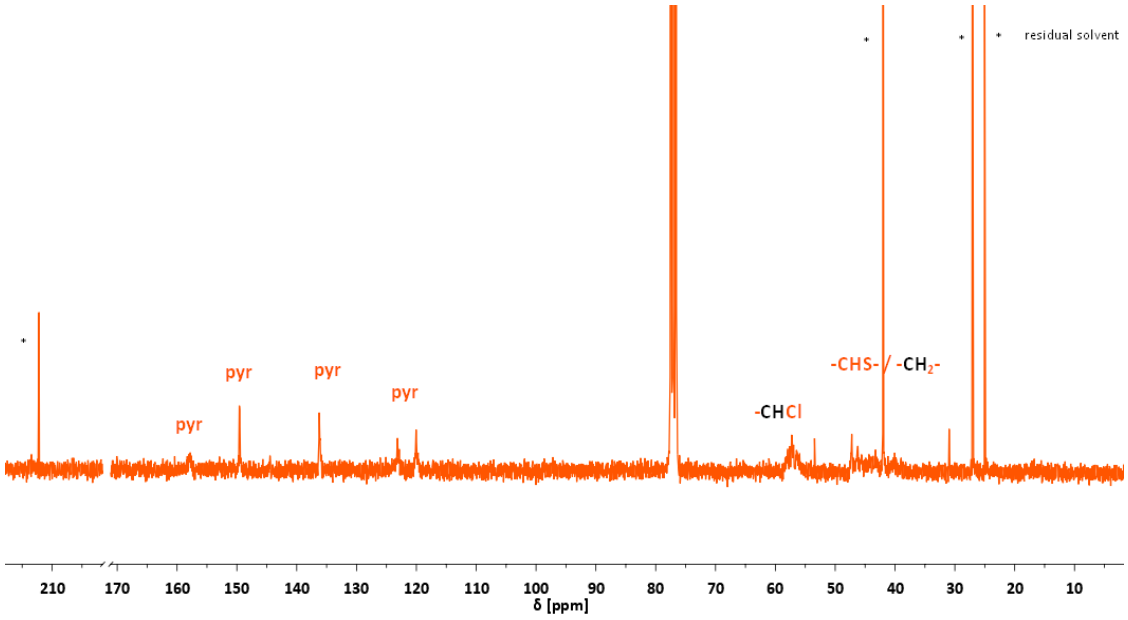


Figure 9: ^{13}C NMR spectrum (75MHz, $CDCl_3$) of PVC-mercaptopyridine

Peaks between 157.7 – 120.3 ppm represent the five mercaptopyridine carbons. The signal at 157.7 ppm is less intense, as it corresponds to the quaternary carbon, in *ortho* position to nitrogen. In accordance with experimental data[63] The peaks in the range of 59.0 – 55.5 ppm can be assigned to the methine carbons of PVC and the broader peaks from 48.5 – 42.5 ppm corresponds to –CHS- and –CH_2- carbons.

[63] Mijangos, C.; López, D. *Macromolecules* **1995**, 28, 1365

IR spectroscopy

FT-IR measurements were performed for qualitative characterization of the modified PVC. Again the samples were solubilized in THF (0.5 – 1% (w/v)) and dropcasted onto CaF_2 plates.

Table 7: Characteristic IR vibrations of PVC-mercaptopyridine *(cf. Ref.[64, 65, 66])*

Functional group	v [cm-1]	Intensity	Comments
=C-H	3075, 3046	w-m	str
C-C arom.	1580, 1450	vs	str
C-N	1557	s	str
C=S	1123	s	str

For characteristic IR vibrations of pure PVC see **Table 3.** A comparison of PVC before and after modification with 2-mercaptopyridine (modification degree: 24%) is shown below.

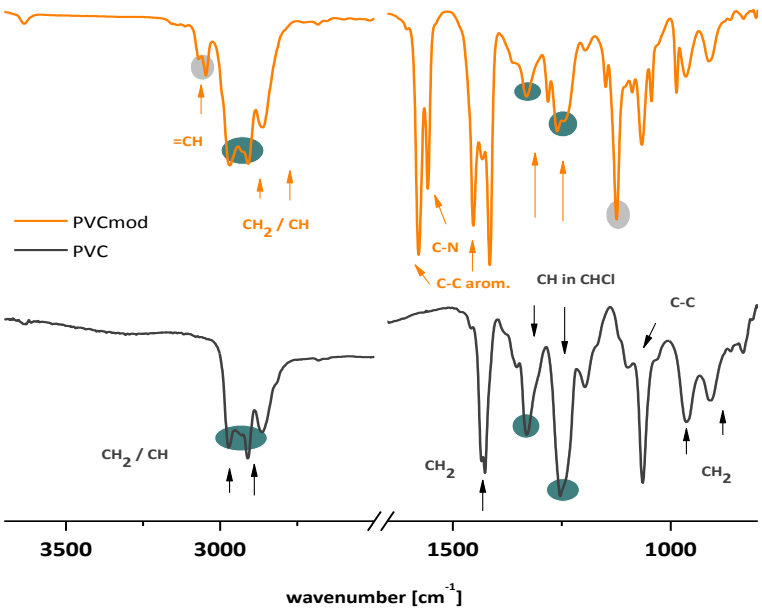


Figure 10: IR spectra of PVC and PVC-mercaptopyridine

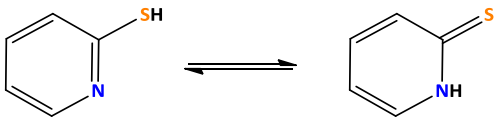
[64] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2202

[65] Socrates, G. (ed.) *Infrared Characteristic Group Frequencies* (2nd ed.), John Wiley & Sons, Chichester, chapt. 12, p. 133

[66] Spinner, E.; *J. Org. Chem.* **1958**, 23, 2038

IR measurements confirm that pyridine units have been introduced into the polymer. Characteristic peaks like the aromatic C-C vibrations or the band indicating C-N stretching can be observed (cf. **Figure 4)** Whereas the new peaks appearing at 3075 and 3046 cm^{-1}, as well as 1123 cm^{-1} (bright spots) are not clearly assignable. The former two are likely to stand for olefinic C-H stretching vibrations that occur due to the formation of double bonds after dehydrochlorination. The same signal could also be assigned to aromatic pyridine N-H vibrations or aromatic C-H vibrations. These bands usually appear in the same range as the aliphatic CH_2 and C-H vibrations of PVC (see **Table 3**).

The occurrence of a N-H peak, in combination with the peak at 1123 cm^{-1}, might indicate free 2-mercaptopyridine, because according to literature C=S stretching vibrations of 2-pyridthiones appear between 1145 and 1100 cm^{-1} [65]. 2-Mercaptopyridine also exists in its tautomeric form 1H-pyridine-2-thion. The equilibrium is influenced by solvent and temperature.



Scheme 13: Tautomeric equilibrium of 2-mercaptopyridine & 1*H*-pyridine-2-thion

Anyway the ^1H NMR spectrum showed no evidence of the existence of starting material. Alternatively to a selective modification with the thiol moiety, only the nitrogen could have been linked to the PVC chain instead, which would also result in a C=S vibration. Again this option seems rather unlikely. The basic character of the 2-pyridinethion rather favors elimination instead of a nucleophilic attack[67] which is also proved by ^1H NMR measurements. Additionally no significant evidence can be found regarding the formation of the internally quaternized ring structure.

Another proof for the partial substitution of the chlorine atoms are the weakened bands of the CH-Cl group (dark spots).

Additionally a comparative view of PVC obtained at different modification degrees, cannot be provided. This is due to bad solubility in adequate solvents with a low boiling point, which are suitable for dropcasting a polymer film. Corresponding ATR-IR spectra of selected samples are of low quality and accordingly not displayed here.

[67] Spinner, E.; *J. Org. Chem.* **1958**, 23, 2038

STA – Simultaneous Thermal Analysis

Thermal stability of PVC-mercaptopyridine was evaluated with simultaneous thermal analysis. These measurements were performed to judge its suitability for fabricating compounds.

For the sake of completeness another sample of PVC-thioaniline was measured too.

Usually the criterion for processing is a mass loss of less than 3%. But here the first weight loss can be attributed to the evaporation of trapped solvent hence the corresponding onset temperatures of degradation are determined at higher losses.

Table 8: Onset degradation temperatures T_D of modified PVC

Sample	T_D [°C]	Δm [%]
PVC	**270**	3
PVC-thioaniline$_{30\%}$	**140 - 158**	5
PVC-m.pyridine$_{27\%}$	**138 - 150**	5 -7

No exact temperature point, but rather a temperature range for the onset of degradation was determinable since this process seems to coincide with the evaporation of residual cyclohexanone. However, it can be concluded from this data that thermal stability was reduced considerably upon modification.

Processing temperatures for pure PVC are generally not too high in order to avoid dehydrochlorination which can be observed already at lower temperatures due to structural defects caused during the polymerization of vinyl chloride.[68] Working temperatures for extrusion of unplasticized PVC, used for rigid applications like profiles and piping, are between 170 – 190°C.[69]

Accordingly both polymer samples are not suitable for compounding.

Nevertheless this result is in not quite in accordance with the measured glass transition temperatures of 119°C and 138°C for PVC-thioaniline which would naturally imply a higher

[68] Starnes Jr, W.H. *J. Polym. Sci. A Polym. Chem.* **2005**, 43, 2451-2467

[69] Land, W. *Kunststoffpraxis:Eigenschaften*, Weka Media GmbH & Co. KG, Kissing, **2005**, volume 2, part 7, chapter 2.8.1., p. 7

temperature of decomposition. However, as already stated in chapter 3.2.2, measured T_g values are likewise deviating because of trapped solvent.

Figure 11 illustrates a 3 step degradation process for PVC-thioaniline, four steps for PVC-mercaptopyridine and two for pure PVC (all in consideration of trapped solvent).

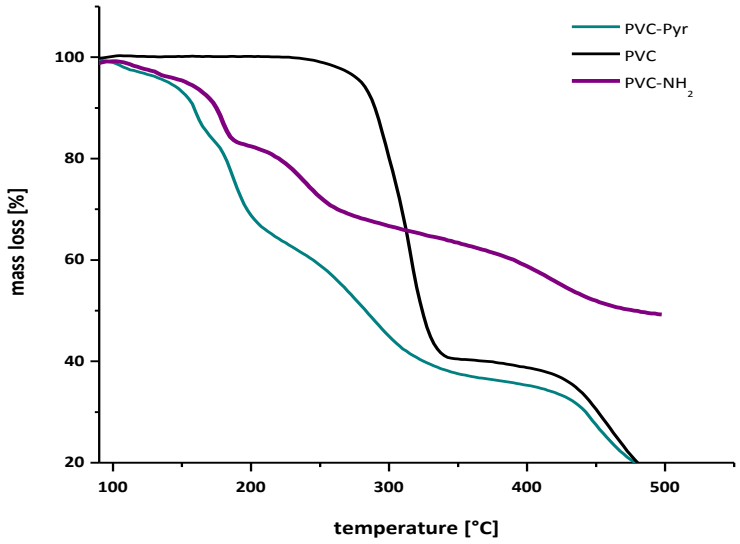


Figure 11: STA thermograms of pure and modified PVC

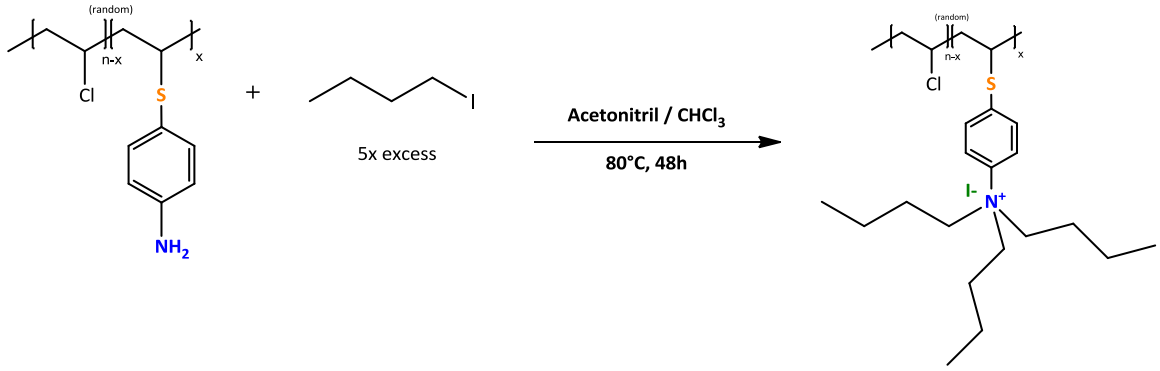
On the other hand it must be remarked that metrological fluctuations occurred in the temperature range up to approximately 200°C as a result of thermal inertia of the sample crucible (Al_2O_3) upon heating.

Therefore the estimation of the characteristic temperature points is already uncertain to some extent. Fluctuations could not be eliminated in a second test run, which lead to the same results.

3.4. Quaternization of PVC-Thioaniline

Cationic quaternary ammonium compounds are commonly known as a biologically active species, that are applied as antimicrobials and disinfectants.

Therefore it was aimed at the introduction of a positive charge to the thioaromatic groups of modified PVC. This can be achieved through alkylation of the amino functionality.



Scheme 14: Quaternization of PVC-thioaniline

Quaternization of the free amino group should be achieved with a 5 fold excess of iodobutane, with respect to thioaniline. These reaction conditions have already been successfully applied in order to yield quaternary polymer species.[70]

Nonetheless the resulting brownish product could not be solubilized both in polar and non-polar solvents. It was assumed that under the given conditions crosslinking was favored over quaternization.

[70] Doctoral thesis Seyfriedsberger, G.: *Kontaktbiozide auf Polymerbasis: Herstellung und Charakterisierung*, TU Graz, **2007**, 83-84

3.5. Quaternization of PVC-Mercaptopyridine

3.5.1. Preparation of PVC-Mercaptomethylpyridinium

In order to find suitable reaction conditions for the alkylation of PVC-mercaptopyridine, parameters like solvent and halide were varied and compared. Table 9 gives an overview.

Table 9: Reaction conditions for the alkylation of PVC-mercaptopyridine

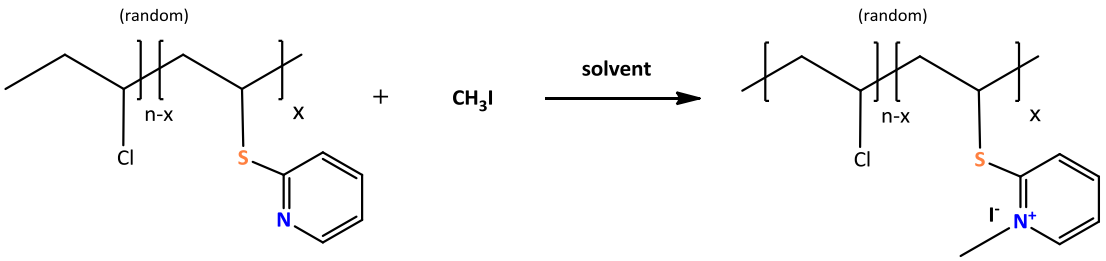
Alkylation agents (molar ratio)	Solvent	Reaction time	Alkylation
PVC-mpyr / C_4H_9I (1:3,5)	Acetonitrile / $CHCl_3$ (1:1.5 v/v)	20h	No
PVC-mpyr / CH_3I (1:3,5)	Acetonitrile / $CHCl_3$ (1:1.5 v/v)	24h	No
PVC-mpyr / CH_3I (1:20)	THF	6 days	Little
PVC-mpyr / CH_3I (1:5)	**$C_6H_5NO_2$**	**20h**	**Yes**
PVC-mpyr / CH_3I (1:10)	**CH_3NO_2**	**20h**	**Yes**

The transformation of PVC-mercaptopyridine into PVC-mercaptomethylpyridinium could not be achieved in a fairly polar acetonitrile / $CHCl_3$ solvent mixture, hereby adopting a previously tested system.[71] Also the usage of the more reactive alkyl halide CH_3I species did not lead to the desired outcome. As it was monitored via ^1H NMR spectroscopy partial alkylation could be achieved, leading to approx. 3% quaternization within the polymer. Nevertheless it was necessary to employ a large excess of halide and a long reaction time.

Nitrobenzene and nitromethane are both commonly used to perform quaternization reactions and an analogous application for a PVC modification can be found.[72] Both possibilities were examined more closely under the given conditions.

[71] Doctoral thesis Seyfriedsberger, G.: *Kontaktbiozide auf Polymerbasis: Herstellung und Charakterisierung*, TU Graz, **2007**, 83-84

[72] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2200



Scheme 15: Quaternization of PVC-mercaptopyridine

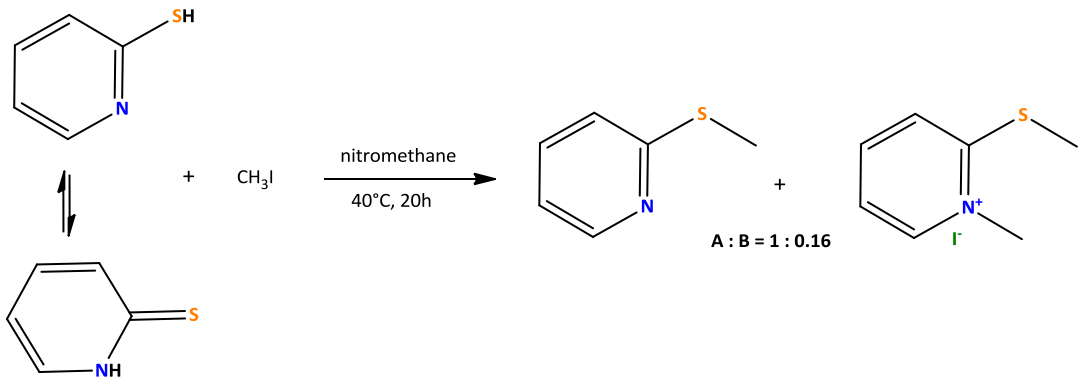
PVC-mercaptopyridine, synthesized and characterized as specified in chapter 3.3, was dissolved at first. Upon that a 5 – 10 fold excess of iodomethane was allowed to react for approx. 20 h at 40°C.

Nitrobenzene solubilizes PVC-mercaptopyridine species quite easily, independently of the degree of 2-mercaptopyridine substitution. Anyhow the final product requires an extensive purification procedure in order to obtain a solvent-free polymer sample. (A detailed description of this procedure is listed in the experimental part.)

Such products of high purity are absolutely necessary when it comes to the testing of antimicrobial activity. Residues of a highly toxic solvent like nitrobenzene would falsify the results. However nitromethane is rather low-boiling, compared to the nitro aromatic solvent. Hence it can be removed more easily by means of vacuum drying under ambient temperature.

3.5.2. **Reaction Model**

Performing the alkylation of 2-mercaptopyridine under the same experimental conditions (10 fold excess of alkyl halide) as for PVC-mercaptopyridine should give information about product distribution and characteristics in ^{1}H NMR spectra. (For the detailed proceeding see chapter 5.2.4)



Scheme 16: Reaction model - Quaternization of 2-mercaptopyridine

NMR spectroscopy indicates a favored alkylation of the thiol. Quaternization of the tertiary amino functionality only occurs to a small degree. However exact product ratios were not determined due to the occurrence of a third species that could not be clearly assigned but is likely to correspond to an oxidized side product. The synthesis was not performed under inert conditions, which supports the previous assumption.

Regarding the polymer, these results now could lead to the conclusion, that quaternization of mercaptopyridiyl groups of PVC-mercaptopyridine might not occur to the full extent.

3.5.3. **Characterization**

NMR spectroscopy

New characteristic alkyl proton peaks (unfortunately overlapping with the CHCl / CHS band) can be observed after the reaction with iodomethane, and additionally new resonances in the aromatic region, as marked in blue in the figure below. Thus it can be concluded that an alkylation of the pyridine nitrogen took place.

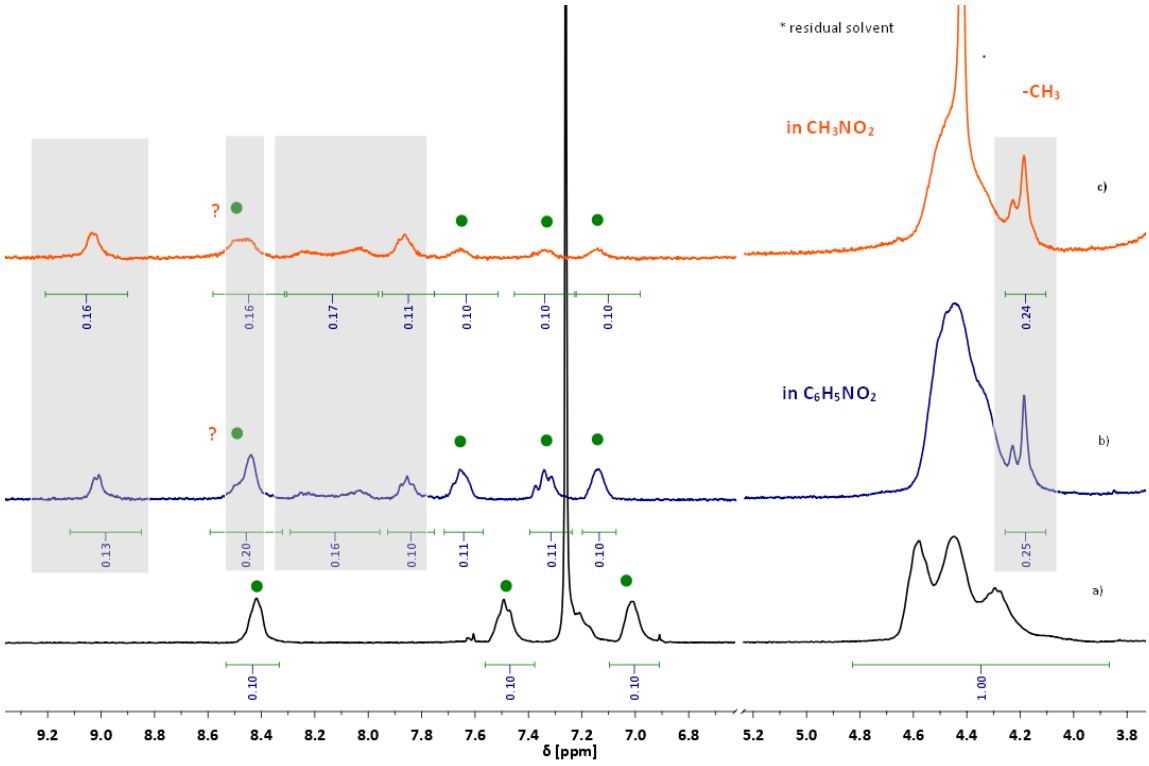
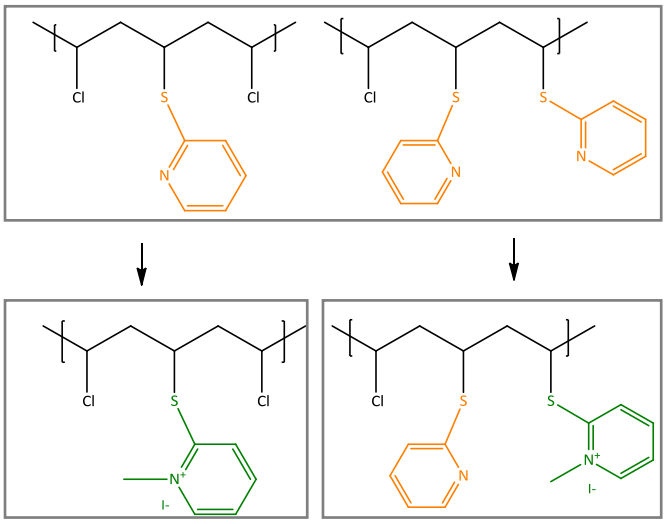


Figure 12: ^{1}H NMR spectra (300MHz, $CDCl_3$ for a), DMSO-d_6 for b) / c)) of PVC-mercaptopyridine before *a)* and after quaternization *b), c)*

Apparently the mercaptopyridyl groups were only partially quaternized. Original bands of the PVC-mercaptopyridine educt are still present, but shifted towards a lower field which might be due to their new chemical environment or /and the usage of DMSO-d_6 instead of $CDCl_3$. However, the exact integration of the signals is not so easy because of the bad signal/noise ratio of the recorded spectra which can be traced back to the limited solubility behavior of the samples in DMSO-d_6. It makes the determination of the degree of quaternization, as well of the assignment of peaks more difficult.

There seems to be a third type of aromatic protons, indicated on the one hand by the integral ratio and other hand by the broad and humpy shape of the new bands. One interpretation might be again the influence of the distribution of the mercaptopyridyl and mercaptomethylpyridinium groups.



Scheme 17: Possible distribution of 2-mercaptopyridine /2-mercaptomethylpyridinium along the PVC chain

Partial quaternization might lead to different kinds of interactions, namely singular pyridinium units, pyridine-pyridinium and pyridine-pyridine sequences, as suggested by this scheme. This interpretation could also explain the occurrence of two alkyl proton peaks at 4.20 ppm and 4.23 ppm.

However degrees of quaternization, determined via the ratio of (methyl*0.3) / (methyl*0.3 + mercaptopyridine) are listed below.

Table 10: Degrees of quaternization of PVC-mercaptopyridine

	DS [%]*	Reaction time [h]	DQ [%]**
Experiment 1	10	20	4 -5
Experiment 2	30	20	5 -6
Experiment 3	10	20	4 -5

*DS – overall degree of substitution (as determined via ^{1}H NMR integral ratio)
**DQ – overall degree of quaternization (as determined via ^{1}H NMR integral ratio)

Moreover usage of nitromethane (experiment 3) or nitrobenzene (experiment 1 / 2) does not seem to influence the outcome of the reaction according to their ^{1}H NMR spectra.

IR spectroscopy

FT-IR measurements could only be achieved for one PVC species. Again the samples were solubilized in THF and dropcasted onto CaF_2 plates. ATR-IR spectra were recorded for those that were insoluble.

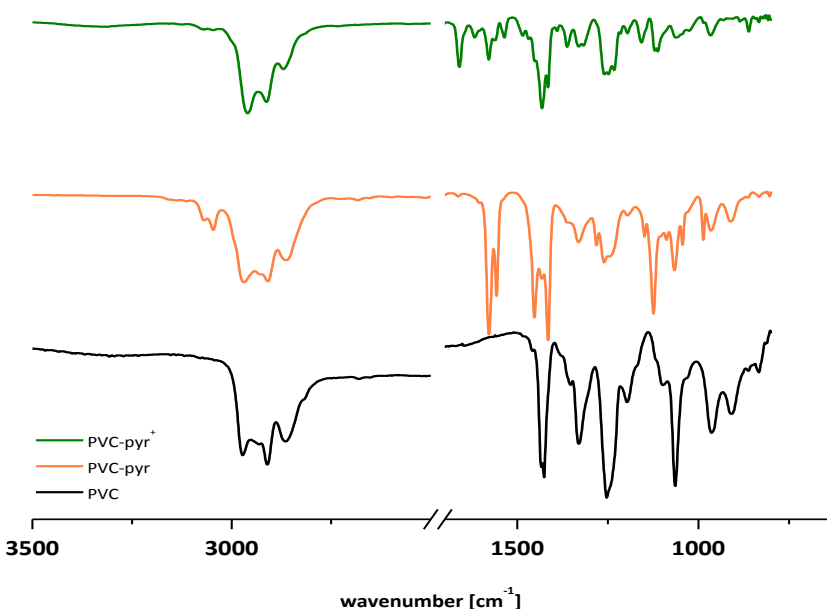


Figure 13: IR spectra of PVC, PVC–mercaptopyridine (DS: 28%), PVC-mercaptomethylpyridinium (DS: 10%, DQ: 4-5%) samples

It can be seen that the bands corresponding to the olefinic protons of pyridine (3075 cm^{-1}, 3046 cm^{-1}) have almost disappeared, and also other characteristic bands between 1600 and 1400 cm^{-1} are considerably weakened or not assignable. Considering the fact that the PVC-mercaptomethylpyridinium was not wholly soluble in THF it could have occurred that only the less substituted/quaternized polymer chains dissolve in THF. Therefore some characteristic bands are not so prominent.

The extent of quaternization cannot be estimated with IR spectroscopy.

Unfortunately the quality of the ATR-IR spectra is very low and no new information could be obtained.

3.5.4. **Antimicrobial Activity of Modified PVC**

The antimicrobial activity and efficacy of selected PVC samples was tested according to the Japanese industrial standard (JIS Z2801:2000). This method is designed to quantitatively test the ability of plastics to inhibit the growth of microorganisms or kill them, over a 24 h period of contact.

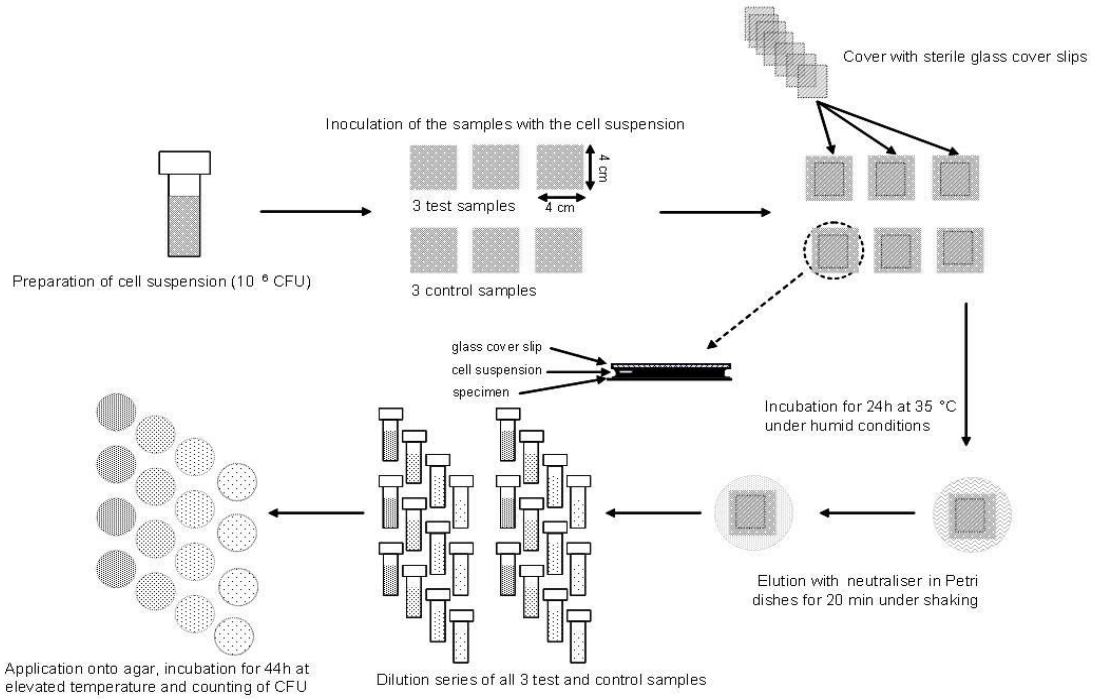


Figure 14: Illustration of the JIS Z2801 method

Sample Preparation:

A solution of 1% (w/v) of polymer was prepared in THF and then spincoated on glass substrates (dimension: 40 x 40mm). These substrated were cleaned previously with acetone or 2-propanol in an ultrasonic bath

Spincoating parameters:

- Rotational speed: 1000 rpm
- Acceleration: 1000 rpm/s
- Period of rotation: 20 s

Testing of the samples was performed by TTZ Bremerhaven, as follows:[73, 74]

Beforehand test microorganism are prepared by growth in a liquid culture medium (LB – lysogeny broth for *E. coli* and *S. aureus*, BHI – brain heart infusion for *L. monocytogenes*, *P. fluorescens*). A standardized dilution of the cell suspension of 10^6 cells /mL was used to inoculate the control and test surfaces in duplicate. These were then covered with sterile glass covers to ensure close contact. Microbial concentrations, expressed as CFU (colony forming units) were determined at "time zero" (after 15 min) and after 24 h. Reference and test samples were incubated at 36±1°C for 24 h. Afterwards elution and neutralization was performed in Petri dishes, containing a CSL (casein peptone soybean flour peptone) – THSC (Tween – Saponine – Histidine – Cysteine) broth. Dilution series of 10^{-1} and 10^{-2} were plated onto CSA (casein peptone soybean flour peptone agar) and incubated for 44 h at 36±1°C.

Tested organisms were the gram – positive *S. aureus* and *L. monocytogenes* as well as gram – negative *E. coli* and *P. fluorescens* bacteria.

Results

Antibacterial efficacy is quantified via a reduction factor R. It expresses the degree of bacterial cell reduction on the potential antimicrobial surface with respect to the control sample.

$$\mathbf{R = \log(CFU_{control}) - \log(CFU_{sample})}$$

CFU….colony forming units (corresponds to bacterial cell concentration)

The higher factor R the more antimicrobial activity can be assigned to the polymer sample. In respect to that, a negative factor confirms cell growth instead of lysis.

[73] Figure and procedure: Doctoral thesis Kreutzwiesner, E. *Entwicklung von compoundierbaren Kontaktbioziden auf Basis aminfunktionalisierter Polymere,* TU Graz **2010,** 57; 66

[74] Procedure: http://www.antimicrobialtestlaboratories.com/JIS_Z_2801_Antimicrobial_Surface_Test.htm

Table 11: Antimicrobial testing of PVC-mercaptopyridine$_{24\%}$

Sample	*L. monocytogenes*	
	log(CFU)*	R
Control: *PVC*	5.44 ± 0.04	-
1: *PVC-m.pyridine$_{24\%}$*	6.76 ± 0.05	**-1.32 ± 0.01**

*CFU determined after 24h

Previously to the quaternization reactions an original PVC-mercaptopyridine sample with an estimated degree of substitution of 24% was tested against the gram-positive test organism *L. monocytogenes.* In this case the negative reduction factor expresses adverse activity: Modification with mercaptopyridyl goups lead to cell proliferation, compared to pure PVC. It is remarkable that this control sample even caused a slight cell reduction of 0.24 logarithmic units.

For the reason of a better comparability normalized log – values of control and sample are used in the following figures.

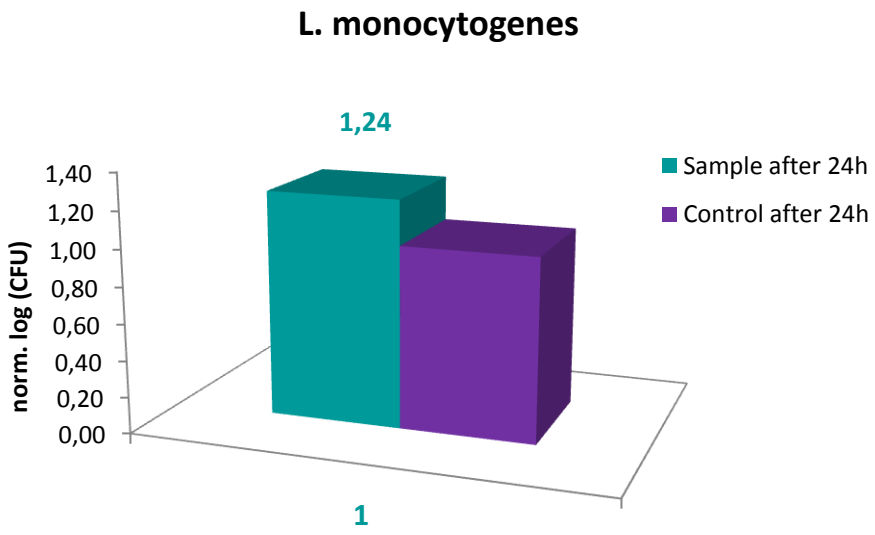


Figure 15: Biocidal activity of PVC-m.pyridine$_{24\%}$ (1) versus PVC

Accordingly the influence of quaternization on microbicidal efficacy was analyzed. Samples were tested both against gram-positive (*S. aureus, L. monocytogenes)* as well as gram-negative (*E. coli*, *P. fluorescens*) microorganisms.

Structural differences of gram – positive and gram – negative bacterial cells often account for diverse killing behavior of the antimicrobial agent.[75] To be exact, gram - positive organisms possess a thicker and thus more stable outer cell wall than gram – negative and thus cannot be destroyed that easily.[76]

Table 12: Antimicrobial testing of PVC-mercaptomethylpyridinium$_{4\text{-}5\%}$

Sample	*S. aureus*		*E.coli*	
	log(CFU)	R	log(CFU)	R
Control: *PVC*	4.43 ± 0.57	-	**5.81 ± 1.10**	-
2: *PVC-m.pyridinium$_{4\text{-}5\%}$*	3.05 ± 0.35	**1.38 ± 0.17**	3.69 ± 0.26	**2.12 ± 0.28**
3: *PVC-m.pyridinium$_{4\text{-}5\%}$*	0	**4.43 ± 0.00**	0	**5.81 ± 1.10**

Sample	*L. monocytogenes*		*P. fluorescens*	
	log(CFU)*	R	log(CFU)*	R
Control: *PVC*	6.26 ± 0.08	-	4.29 ± 0.25	-
2: *PVC-m.pyridinium$_{4\text{-}5\%}$*	6.01 ± 0.14	**0.25 ± 0.01**	3.00 ± 0.26	**1.29 ± 0.10**
3: *PVC-m.pyridinium$_{4\text{-}5\%}$*	**4.52 ± 0.83**	***1.74 ± 0.31***	**-**	**-**

*CFU - averaged (determined after 24h)

Reduction factors between 0.25 and 2.12 indicate only **weak** antibacterial activity for **polymer 2**, whereas it has to be noted that experimental results for the *E. coli* control sample show a broad distribution. Furthermore no significant differences between gram-positive or gram-negative organisms could be observed. The same polymer did not show a biocidic effect against *L. monocytogenes*.

In the tests against *S. aureus* and *E. coli* polymer 3 clearly exhibited a **strong activity**. However the tests results for *P. fluorescens* showed an extremely broad distribution of three log units. No average or reduction factor was therefore calculated. By trend a moderate antibacterial effect could be observed. Accordingly the experimental error for *L. monocytogenes* is also considerably high. The calculated reduction factor of 1.74 attributes only a weak activity against this organism.

[75] Doctoral thesis Kreutzwiesner, E. *Entwicklung von compoundierbaren Kontaktbioziden auf Basis aminfunktionalisierter Polymere,* TU Graz **2010,** 58;59

[76] Franklin, T. J.; Snow, G. A. *Biochemistry of Antimicobial Action*, Chapman and Hall, **1981**

A reason for this might be physical aging or low stability of the polymer film. Tests against *L. monocytogenes* and *P. fluorescens* were performed one month later. During storage the plastic film could have possibly degraded and /or become brittle which lead to inhomogeneous films on the glass substrate. In this context it has to be mentioned that the films of polymer 3 on glass are much thinner than those of polymer 2, because of its limited solubility in THF, which was needed for spincoating. Such thin films are thus more likely to be instable and become inhomogeneous and do not guarantee optimal testing conditions.

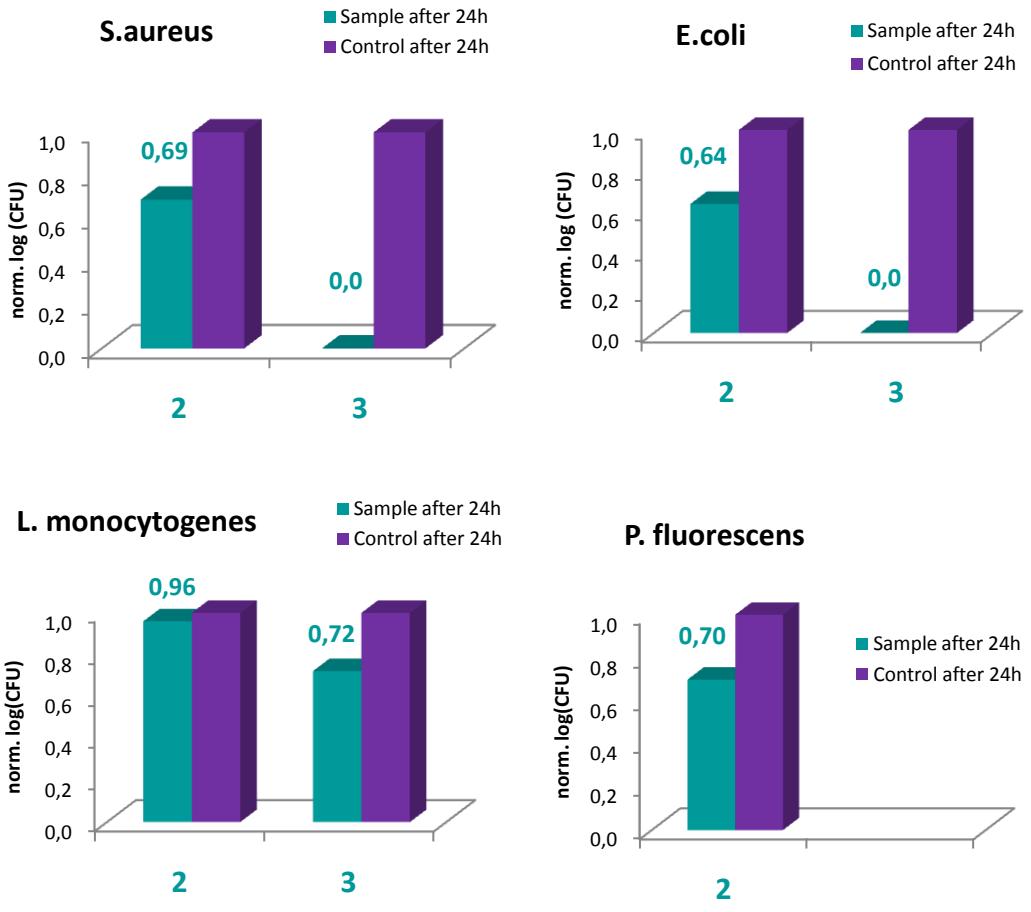


Figure 16: Biocidal activity of PVC-m.pyridinium$_{4-5\%}$ (2), (3) versus PVC (control) against gram-positive (*S. aureus, L. monocytogenes*) and gram-negative (*E. coli, P. fluorescens*)

Moreover the quite different behavior of polymer 2 and 3 is in general very surprising, because the overall degree of substitution (10%) and quaternization (5%) is the same for both, as determined by ^{1}H NMR. So far their differing activity against microbes might be caused by other factors.

Table 13: Polymer properties potenially influencing microbiocidic performance

Sample	DS [%]	DQ [%]	Reaction solvent	Solubility in THF	Biocidic
1: *PVC-m.pyridine*$_{24\%}$	24	0	Cyclohexanone	**Yes**	**No**
2: *PVC-m.pyridinium*$_{4\text{-}5\%}$	10	4 - 5	$C_6H_5NO_2$	**Partly**	**Weak**
3: *PVC-m.pyridinium*$_{4\text{-}5\%}$	10	4 - 5	CH_3NO_2	**Bad**	**Strong**
4: *PVC-m.pyridinium*$_{5\text{-}6\%}$	28	5 - 6	$C_6H_5NO_2$	**Not at all**	**n.d.**

n.d. not determined

A recurring problem in handling modified PVC polymers is their solubility. Regarding **Table 13** it is obvious that the existence of pyridinium units worsens solubility in THF due to the higher polarity of the polymer. Polymer 4, containing 5 – 6% pyridinium groups and exhibiting an overall substitution degree of 28%, could therefore not be solubilized at all. Solubility was tested in more (THF + EtOH or MeOH) or less polar (THF + $CHCl_3$) mixtures without any striking improvement.

Polymer 2 could be dissolved to a large extent whereas only a small amount of polymer 3 was dissolvable. Although great efforts were taken to purify the samples, minimal residues of nitrobenzene enclosed in polymer 2 might have improved its solubility compared to polymer 3

So only polymer surface come in close contact with microorganisms the mere thickness of these films should not influence the activity of the plastic in a direct way.

On the other hand, if the biocidic effect was caused by a decomposition product of the polymer it could rather explain why polymer 3 exhibited a stronger activity. The thinner polymer layer might be more prone to environmental attack because most likely the existence of solid polymer particles also prevented the formation of homogenous films during the process of spincoating.

Such a decomposition product, probably a pyridinium compound, could therefore act as a water-soluble biocide. This interpretation is speculative and cannot be supported by experimental evidence.

Eventually more information can be gained by monitoring the chemical stability and aging of the polymer, including IR characterization or contact angle measurements, which investigates the polarity of the surface. The existence of a water soluble biocidic agent also has to be determined.

4. Conclusion and Outlook

This work presents a post-polymerization modification pathway for PVC, which was achieved by nucleophilic substitution of the chloride moieties with aromatic thiols in solution, namely 4-aminothiophenol and 2-mercaptopyridine. The resulting polymer was subjected to further modifications, by quaternization of amino functionalities, which were previously introduced into the polymer by these bifunctional thio-based modifiers. Herewith it was aimed at the development of an antimicrobial polymer.

At first different N-, O- and S-based modification reagents were tested, whereupon aromatic thiols proved to be the most promising candidates to perform a selective substitution under avoidance of dehydrochlorination. It was possible to achieve high degrees of substitution within relatively short reaction times for both 4-aminothiophenol and 2-mercaptopyridine. Dehydrochlorination was just observed in the case of the pyridine compound, but only to a small extent. Thermogravimetric measurements indicate an unfavorably low thermal stability of PVC after modification. However these results do not match with the significant increase of the glass transition temperatures. A crucial point in this context, but valid for all means of characterization, was the effective removal of the solvent residues. Otherwise this leads to falsified and contradicting results or impedes exact characterization. In the further course of this work more stress was laid on modification with 2-mercaptopyridine, as a potential candidate for a concurrent formation of an internally quaternized PVC species through an additional linkage of the pyridine nitrogen. On the basis of a low-molecular based reaction model the feasibility of this structure was proved, but could not be determined in the case of the PVC polymer.

Therefore PVC-mercaptopyridine was alkylated in a second reaction step, yielding a partially quaternized PVC-mercaptomethylpyridinium compound. Testing of its microbicidic activity against selected gram-positive and gram-negative test organisms showed that an efficient performance generally necessitates the presence of such charged groups which influence bacterial growth already at low degrees of incorporation in PVC. However quite differing results for polymer 2 and 3, both of the same degree of quaternization, were observed, as well as a partly broad distribution of the test results. In conclusion several factors might have contributed to this outcome. Physical aging or low stability of the polymer might have lead to inhomogeneous films which do not guarantee optimum testing conditions. Furthermore it has to be determined if the biocidic effect was probably caused by a decomposition product of the polymer. No obvious experimental evidence could explain the different behavior of two

“identical” polymers but in consideration of this previous context it is suggested that a thinner film of polymer 3 (exhibiting a strong activity) degraded more rapidly, thus causing a higher activity.

In conclusion these results require closer investigation of the chemical and physical stability of the modified polymers and films, respectively. Regarding the cause of the biocidic effect it has to be determined if it is related to the decomposition of the polymer, whereupon potentially water-soluble microbiocidic agents might be released. Based on this information optimization of the existing concept and examination of other PVC-modifications is worth being pursued more closely.

5. Experimental Part

5.1. Instruments and Materials

All chemicals and solvents required in the course of this work were acquired from commercial sources (Alfa Aesar, Fluka or Sigma Aldrich) and used as received without further purification. Unless otherwise stated commercial bulk polymerized PVC with average molecular weights of M_n= 22000 and M_w = 43000, obtained from Sigma Aldrich, was used as purchased in all reactions.

Thin layer chromatography was performed on sheets from Merck (silica gel 60 on aluminium).

^{1}H and ^{13}C NMR spectra were recorded on a Bruker Avance III 300 MHz spectrometer, referenced to $SiMe_4$ and deuterated solvents ($CDCl_3$, CD_3OD, DMSO-d_6).[77] All deuterated solvents were purchased from Cambridge Isotope Laboratories Inc. Peaks were characterized as follows: s (singlet), d (doublet), t (triplet), q (quadruplet), m (multiplet), b (broad), bs (broad singlet).

IR measurements were performed on a Perkin Elmer Spectrum One spectrophotometer with a DTGS detector, operating in transmission mode. Thin films of the samples were solvent cast on CaF_2 plates. Corresponding bands were characterized as very strong (vs), strong (s), medium (m) and weak (w).

Elemental analysis of the elements C, H, N, and S was carried out on a Universal Elemental Analyzer.

Thermal characterization involved DSC and STA measurements. DSC data were collected with a Perkin Elmer Pyris Diamond. Samples were heated up to 250°C and quenched under a nitrogen atmosphere at a rate of 20°C / min. Reported T_g values were taken from the second heat run.

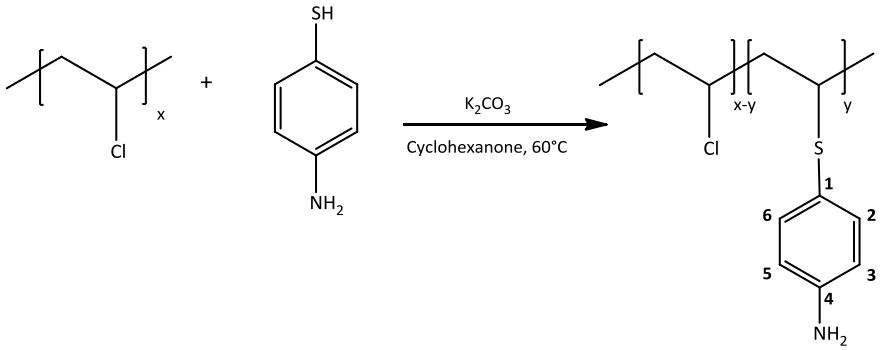
STA was accomplished on a Netzsch 449C apparatus, using He as purge gas. Thermogravimetric losses were monitored up to a temperature of 550°C applying a heating rate of 10°C / min.

[77] Gottlieb, H.E.; Kotlyar, V.; Nudelman, A. *J. Org. Chem.* **1997**, 62, 7512-7515

Studies on the antimicrobial efficacy of selected polymer samples were performed externally by Technologie Transfer Zentrum TTZ Bremerhaven according to a modified protocol of the Japanese Industrial Standard JIS Z2801:2000. An illustration and a detailed description of this method can be found in chapter 3.5.4.

5.2. Preparations

5.2.1. **Substitution of PVC with 4-Aminothiophenol**



Scheme 18: Modification of PVC with 4-aminothiophenol

Based on the approach of Reinecke[78] 200 mg (3.2 mmol, 1 eq., referring to one monomer unit) of PVC was dissolved in 20 mL of cyclohexanone and stirred for 30 min. 660 mg (4.8 mmol, 1.5 eq.) of potassium carbonate, as well as 400 mg of 4-aminothiophenol (3.2 mmol, 1 eq.) were added to the solution. The reaction was allowed to stir over night (18h) at 60°C under an inert atmosphere of Ar. Precipitation was achieved using a cold methanol / water (9:1 v/v) mixture. Removal of residual solvent involved several purification steps: At first the product was dried under reduced pressure in a drying oven at 40°C for 18 - 20 h. The solid was then extracted in 5 – 10 mL of diethylether for 2 – 3 times, altogether for at least 24 h. As a final step it was dried again under vacuum.

Yield: 84 – 87% (197 – 231 mg) of yellowish to brownish, brittle solid

Table 14: Overview - Reaction conditions and corresponding degrees of substitution

	PVC	Reaction time [h]	DS [%]*	yield
Experiment 1	M_n = 67730 M_w = 182080	18	15 - 20	197 mg (84%)
Experiment 2	M_n = 22000 M_w = 43000	18	30 - 35	231 mg (87%)

*DS - degree of substitution (as determined via ^{1}H NMR integral ratio, see 3.2.2)

[78] Reinecke, H.; Mijangos, C. *Polymer* **1997**, 38, 2291

^{1}H NMR (δ, 20°C, DMSO-d_6, 300 MHz): 7.30 – 6.75 (m, 2H, $ph^{2,6}$), 6.70 – 6.35 (m, 2H, $ph^{3,5}$), 5.00 – 3.70 (m, 1H, CH_2=C*H*-Cl), 3.70 – 3.20 (m, 1H, CH_2=C*H*S) superposed by water peak of solvent

^{13}C NMR (δ, 20°C, DMSO-d_6, 75 MHz)**:** not measured because of poor solubility

FT-IR (cm^{-1}, film on CaF_2): 3470 (m), 3370 (m), 3220 (m), 2970 (s), 2910 (s), 2866 (s), 1624 (vs), 1600 (vs), 1500 (vs), 1180 (m), 820 (m)

DSC:

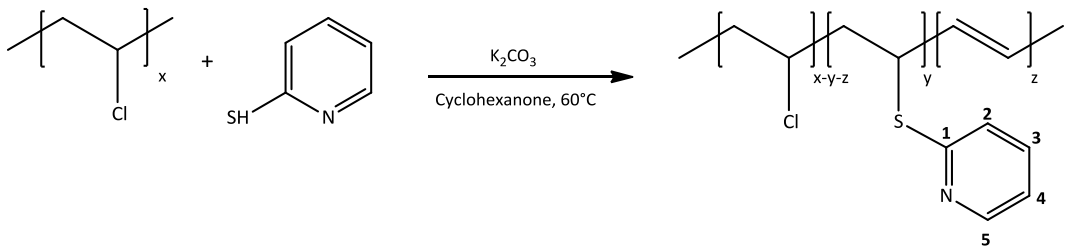
Table 15: Overview - Glass transition temperatures (T_g)

	PVC	DS [%]	Tg [°C]
Experiment 1	M_n = 67730 / M_w = 182080	15 - 20	119
Experiment 2	M_n = 22000 / M_w = 43000	30 - 35	138

*values might differ because of residual solvent (see 3.3.3)

STA: onset degradation temperature for DS = 30%: T_D = 140-158°C (at 5% mass loss)

5.2.2. **Substitution of PVC with 2-Mercaptopyridine**



Scheme 19: Modification of PVC with 2-mercaptopyridine

On the basis of the procedure of Reinecke[79] 200 mg (3.2 mmol, 1 eq., referring to one monomer unit) of PVC was dissolved in 10 mL of cyclohexanone and stirred for 30 min. 330 mg (2.4 mmol, 0.75 eq.) of potassium carbonate, as well as 178 mg of 2-mercaptopyridine (1.6 mmol, 0.5 eq.) were added and allowed to react at 60°C under Ar atmosphere. In order to obtain modified PVC of various degrees of substitution, the reaction was carried out in different time intervals. The reaction was terminated by precipitating the mixture in methanol / water (2:1 v/v) and the pre-dried at 40°C under reduced pressure for 18 – 20 h. A following purification step involved extraction with 5 – 10 mL diethylether for 2 – 3 times over a 24 h period of time and eventually drying under vacuum.

Yield: 81% - 94% (183 mg – 207 mg) of yellowish or brownish, brittle solid

Table 16: Overview - Reaction conditions and corresponding degrees of substitution

	Reaction time [h]	DS [%]*	yield
Experiment 1	3	10	192 mg (91%)
Experiment 2	6.5	18	207 mg (94%)
Experiment 3	18	27	183 mg (81%)
Experiment 4	68	30	195 mg (82%)

*DS – overall degree of substitution (as determined via ^{1}H NMR integral ratio, see 3.3.3)

[79] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2199

Elemental analysis:

Table 17: Overview - Elemental analysis and calculated degrees of substitution

	N [%]	C [%]	S [%]	H [%]	DS [%]*
Experiment 1	1.24	36.3	3.92	4.42	9
Experiment 3	4.11	49.9	9.96	5.24	23
Experiment 4	4.73	49.7	11.3	4.97	29

*DS – overall degree of substitution (based on elemental analysis)

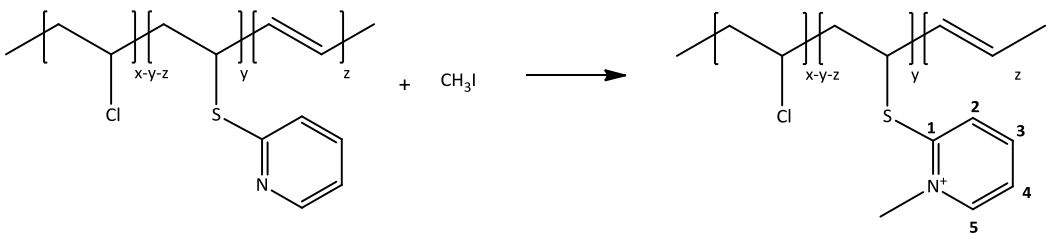
^{1}H NMR (δ, 20°C, $CDCl_3$, 300 MHz): 8.50 - 8.20 (bs, 1H, pyr^5), 7.55 – 7.35 (bs, 1H, pyr^3), 7.30 – 7.05 (bs, 1H, pyr^2), 7.08 – 6.82 (bs, 1H, pyr^4), 5.85 – 5.30 (m, 2H, C*H*=C*H*), 4.75 – 3.90 (m, 2H, -CH_2-C*H*-Cl / -CH_2-C*H*-S)

^{13}C NMR (δ, 20°C, $CDCl_3$, 75 MHz): 157.7 (1C, Cq, pyr^1), 149.7 (1C, pyr^5), 136.4 (1C, pyr^3), 123.3 (1C, pyr^2), 120.3 (1C, pyr^4), 59.0 - 55.5 (1C,-C*H*Cl-), 48.5 – 42.5 (2C, C*H*S-pyr; -CH_2-)

FT-IR (cm^{-1}, film on CaF_2): 3075 (w), 3046 (m), 2970 (s), 2910 (s), 2860 (s), 1580 (vs), 1557 (s), 1450 (vs), 1123 (s)

STA: onset degradation temperature for DS = 27%: T_D = 138-150°C (at 5-7% mass loss)

5.2.3. **Quaternization of PVC–Mercaptopyridine**



Scheme 20: Alkylation of PVC-mercaptopyridine

Version A:

According to the proceeding of Reinecke[80] further modification of the pyridyl group was accomplished as follows: 400 mg of PVC-mercaptopyridine (3.0 mmol, 1eq., based on one vinylthiopyridine unit (M = 137.2 g/mol)) were solubilized in nitrobenzene. The volume of the solvent was varied, depending on degrees of substitution of PVC-mercaptopyridine (DS - 10%: 35 mL; DS - 30%: 65 mL). After 1 h a 5 fold molar excess of iodomethane (with respect to vinylpyridine) was added to the solution and stirred over night (20 h) at 40°C. The final product was precipitated in diethyl ether, then re-precipitated in diethylether / pentane (2:1 v/v) (DS: 30%) or ethanol (DS: 10%) from a solution in nitrobenzene.

In order remove residual nitrobenzene the orange-brownish colored polymer was at first dried in the vaccum drying oven at 40°C for 2 - 3 days and then extracted in diethylether for 2 - 3 times, for altogether 24 h. Removal of nitrobenzene was monitored via TLC (toluene). As a final step the product was vacuum-dried again for 24 h.

Yield: 10 – 20% (45 – 95 mg) of orange-brownish, brittle solid

[80] Reinecke, H.; Mijangos, C. *Macromol. Chem. Phys.* **1998**, 199, 2200

Table 18: Overview - Reaction conditions and corresponding degrees of overall quaternization

	DS [%]*	Reaction time [h]	DQ [%]**	Yield
Experiment 1	10	20	4 -5	95 mg (20%)
Experiment 2	30	20	5 -6	45 mg (10%)

*DS – overall degree of substitution (as determined via ^{1}H NMR integral ratio, see 3.5.3)

**DQ – overall degree of quaternization (as determined via ^{1}H NMR integral ratio, see 3.5.3)

^{1}H NMR (δ, 20°C, DMSO-d_6, 300 MHz): 9.16 – 8.91 (bs, 1H, pyr^{6}), 8.66 – 8.34 (bs, 1H, pyr^{3}) 8.34 – 7.96 (bs, 2H, pyr^{2,4}), 4.80 – 3.90 (m, 2H, CH_2=C*H*-Cl / CH_2=C*H*-S), 4.20 (bs, 3H, N^+-CH_3)

^{13}C NMR: not determined (poor solubility)

FT-IR (cm^{-1}, film on CaF_2): 3075 (w), 3046 (w), 2970 (s), 2910 (s), 2860 (s), 1662 (m), 1580 (m), 1557 (w), 1430 (s)

Version B:

In analogy to version A: 400 mg of PVC-mercaptopyridine with a DS of 10% (3.0 mmol, 1eq., based on one vinylthiopyridine unit (M = 137.2 g/mol)) were dissolved in 50 mL nitromethane for about 1 h. Thereupon a 10 fold molar excess of the alkyl halide (with respect to vinylpyridine) was added to the orange and viscous solution and allowed to react over night (20 h) at 40°C. Then a volume of 50 mL diethylether was added to precipitate the orange product. Purification was achieved by re-dissolving in nitromethane, re-precipitation in diethylether respectively and removal of residual solvent under vacuum.

Yield: 40% (178 mg) of orange solid

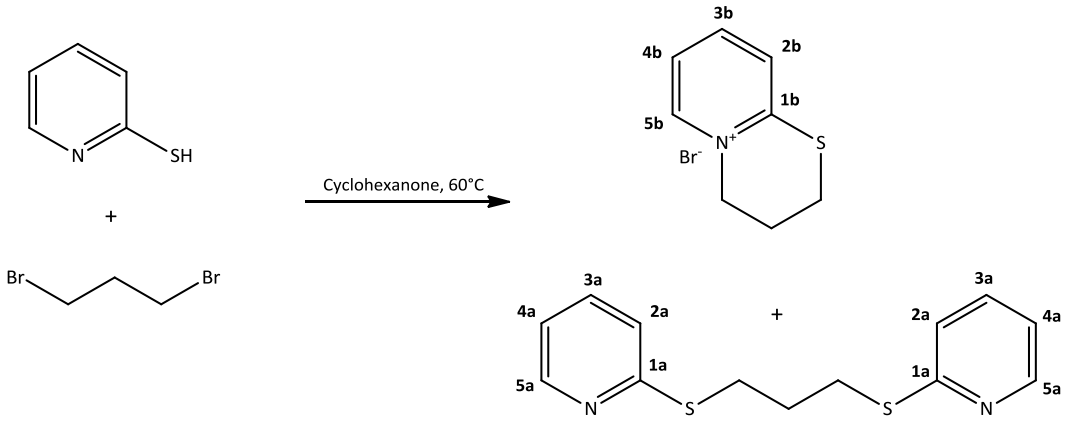
Overall degree of quaternization: (as determined via ^{1}H NMR integral ratio) 4 -5%

^{1}H NMR (δ, 20°C, DMSO-d_6, 300 MHz): signals equal to Version A

^{13}C NMR: not determined (poor solubility)

5.2.4. **Reaction Models**

Substitution with 2-Mercaptopyridine



Scheme 21: Reaction of 2-mercaptopyridine with 1,3-dibromopropane

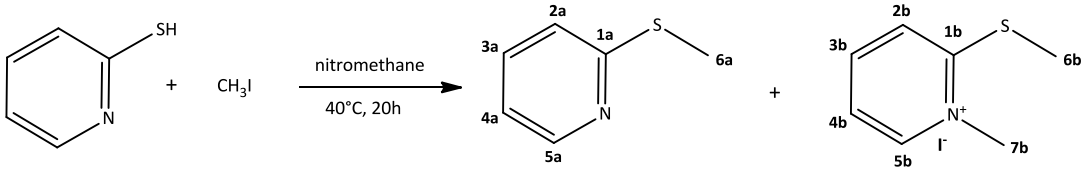
506 mg (4.6 mmol, 1 eq.) of 2-mercaptopyridine were dissolved in 15 mL of cyclohexanone under an inert atmosphere of N_2. In the next step 0.458 mL (4.6 mmol, 1 eq.) of 1,3-dibromopropane were added to the solution and stirred at 60°C under inert conditions of an Ar atmosphere. After 0.5 h precipitation of a white salt could be observed. Reaction progress was monitored via TLC with cyclohexane / ethyl acetate (5:1 v/v). The white salt was filtered off after 3h with a glass frit and washed with 15 mL of pentane. Solvent residues were removed under vacuum.

Yield: 650 mg (53%) of white solid, whereas 59% were found to be the 1,3-bis(2-thiopyridinyl)propane and 41% the 2-mercaptomethylpyridinium bromide salt (as determined by ^{1}H NMR integral ratio).

^{1}H NMR (δ, 20°C, CD_3OD, 300 MHz): 7.08 – 6.95 (m, 3H, pyr$^{5a, 5b}$); 6.78 – 6.63 (t, 2H, pyr^{3a}), 6.61 – 6.50 (t, 1H, pyr^{3b}), 6.45 – 6.32 (d, 2H, pyr^{2a}), 6.31 – 6.22 (d, 1H, pyr^{2b}), 6.12 – 5.94 (m, 3H, pyr$^{4a, 4b}$), 3.12 – 3.00 (t, 2H, N^+-C*H*$_2$-), 2.05 – 1.90 (t, 4H, -S-C*H*$_2$-CH$_2$-C*H*$_2$-S-), 1.91 – 1.81 (t, 2H, -S-C*H*$_2$-(CH$_2$)$_2$-N^+-), 0.96 – 0.84 (m, 2H, -S-CH$_2$-C*H*$_2$-CH$_2$-N^+-), 0.73 – 0.58 (m, 2H, -S-CH$_2$-C*H*$_2$-CH$_2$-S-)

^{13}C NMR (δ, 20°C, CD_3OD, 75 MHz): 158.15 (3C, Cq, pyr^{1a,1b}), 147.18 (1C, pyr^{5b}), 146.66 (2C, pyr^{5a}), 143.74 (2C, pyr^{3a}), 143.47 (1C, pyr^{3b}), 128.94 (1C, pyr^{2b}), 126.73 (2C, pyr^{2a}), 123.75 (2C, pyr^{4a}), 123.25 (1C, pyr^{4b}), 57.14 (1C, N^+-*C*H_2-), 31.48 (2C, -S-*C*H_2-CH_2-*C*H_2-S-), 28.76 (1C, -S-CH_2-*C*H_2-CH_2-N^+-), 28.04 (1C, -S-CH_2-*C*H_2-*C*H_2-S-), 22.40 (1C, -S-*C*H_2-$(CH_2)_2$-N^+-)

Quaternization of 2-Mercaptopyridine



Scheme 22: Reaction of 2-mercaptopyridine with 1,3-dibromopropane

In 10 ml of nitromethane 136 mg (1.2 mmol, 1 eq.) of 2-mercaptopyridine were dissolved, which resulted in a clear and yellow solution. Upon addition of 0,774 mL (12.2 mmol, 10 eq.) iodomethane the reaction mixture was heated up to 40°C and after 0.5 h the color of the solution had turned orange. Under these conditions the solution was allowed to stir for 20 h, thereafter exhibiting a deep red color. Evaporation of the solvent was achieved under reduced pressure, yielding a yellow-brownish powder, which was further dried under vacuum.

Yield: n.d., yellow-brownish solid; with a ratio of 2-(methylthio)pyridine : 1-methyl-2-(methylthio)pyridine-1-ium iodide equating to **1:0,16** (as determined by ^{1}H NMR integral ratio) and a third unassigned species.

^{1}H NMR (δ, 20°C, DMSO-d_6, 300 MHz): 8.87 – 8.78 (d, 1H, pyr^{5b}), 8.47 – 8.36 (d, 1H, pyr^{5a}), 8.33 – 8.23 (m, 1H, pyr^{3b}), 7.91 – 7.78 (m, 2H, pyr^{2a,2b}), 7.70 – 7.62 (m, 1H, pyr^{4b}), 7.48 – 7.40 (d, 1H, pyr^{3a}), 7.28 – 7.18 (m, 1H, pyr^{4a}), 4.04 (s, 3H, -N^+-C*H*$_3$), 2.76 (s, 6H, -S-C*H*$_3$)

Elemental analysis:

N [%]	C [%]	S [%]	H [%]
5.00	25.9	10.9	2.70

6. Appendix

6.1. List of Abbreviations

AIBN	Azo*bis*isobutyronitrile
ATRP	Atom transfer radical polymerization
BPO	Benzoylperoxide
CD_3OD	Methanol (deuterated)
$CDCl_3$	Chloroform (deuterated)
$CHCl_3$	Chloroform
CM	Copper mediated
CPVC	Chlorinated poly(vinyl chloride)
DE	Degree of overall elimination
def	Deformation
DMSO-d_6	Dimethylsulfoxide (deuterated)
DPVC	Dehydrochlorinated poly(vinyl chloride)
DQ	Degree of overall quaternization
DS	Degree of overall substitution
DSC	Differential scanning calorimetry
E. coli	Escherichia coli
FT-IR	Fourier-transform infrared
HCl	Hydrochlorid acid
L. monocytogenes	Listeria monocytogenes
LCRP	Living/controlled radical polymerization
M_n	Average-number molecular weight
M_w	Average-weight molecular weight

n.d.	Not determined
NMR	Nuclear magnetic resonance
PAA	Poly(acrylic acid)
P. fluorescens	Pseudomonas fluorescens
ppm	Parts per million
PTC	Phase transfer catalysis
PVC	Poly(vinyl chloride)
r.t.	Room temperature
S. aureus	Staphylococcus aureus
SI	Surface initiating
STA	Simultaneous thermal analysis
str	Stretching
TBAB	Tetrabutylammonium bromide
T_D	Decomposition temperature
T_g	Glass transition temperature
THF	Tetrahydrofurane
UV	Ultraviolet

6.2. List of Figures

6.3. List of Tables

6.4. List of Schemes