Polymer Protected Gold Nanoparticles

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Preface

The work for this thesis was carried out at the Laboratory of Polymer Chemistry, Department of Chemistry, University of Helsinki, during the years 2002-2006. This work was financially supported by the Finnish Technology Agency (TEKES), which is gratefully acknowledged.

I would like to express my greatest gratitude to my supervisor Professor Heikki Tenhu, for offering me the opportunity to set foot in the most intriguing field of nanoscience at present. It is a real challenge to me, but I am very happy that I have learnt many indispensable pieces of knowledge. I greatly appreciate his trust, encouragement, and his valuable scientific guidance throughout this research. Despite all his duties, he has always found time to discuss any possible problems in my work in his composed and amiable manner. Professor Tenhu is a kind and caring person. I am truly indebted to his support and friendship in everyday life.

This thesis is also a fruit of close cooperation among many scientists working in different fields. I am grateful to Professor Esko Kauppinen and Dr. Hua Jiang at the VTT Technical Research Center of Finland for their great help with HRTEM measurements of gold nanoparticles throughout this work during these years. I wish to express my sincere thanks to Dr. Tapani Viitala in the KSV Instruments Ltd., since I studied the Langmuir monolayers of amphiphilic gold nanoparticles in his laboratory under his supervision. I would also like to thank Dr. Jouko Peltonen in Åbo Akademi University, for kindly allowing me to investigate the in situ optical properties of the amphiphilic particles in his laboratory together with Dr. Tapani Viitala. My sincere thanks also go to Professor Anatoli Darinskii, Institute of Macromolecular Compounds, St. Petersburg, Russia, and Professor Françoise Winnik, Université de Montréal, for the useful discussions about the phase transitions of PNIPAM chains bound to gold nanoparticles.

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Finally, I would like to thank my dear wife Jie for her unfailing understanding with a big heart, as well as for her participation in a part of this work.

Helsinki, March 2006
Jun Shan
Abstract

Polymer protected gold nanoparticles have successfully been synthesized by both “grafting-from” and “grafting-to” techniques. The synthesis methods of the gold particles were systematically studied. Two chemically different homopolymers were used to protect gold particles: thermo-responsive poly(N-isopropylacrylamide), PNIPAM, and polystyrene, PS. Both polymers were synthesized by using a controlled/living radical polymerization process, reversible addition-fragmentation chain transfer (RAFT) polymerization, to obtain monodisperse polymers of various molar masses and carrying dithiobenzoate end groups. Hence, particles protected either with PNIPAM, PNIPAM-AuNPs, or with a mixture of two polymers, PNIPAM/PS-AuNPs (i.e., amphiphilic gold nanoparticles), were prepared. The particles contain monodisperse polymer shells, though the cores are somewhat polydisperse.

Aqueous PNIPAM-AuNPs prepared using a “grafting-from” technique, show thermo-responsive properties derived from the tethered PNIPAM chains. For PNIPAM-AuNPs prepared using a “grafting-to” technique, two-phase transitions of PNIPAM were observed in the microcalorimetric studies of the aqueous solutions. The first transition with a sharp and narrow endothermic peak occurs at lower temperature, and the second one with a broader peak at higher temperature. In the first transition PNIPAM segments show much higher cooperativity than in the second one. The observations are tentatively rationalized by assuming that the PNIPAM brush can be subdivided into two zones, an inner and an outer one. In the inner zone, the PNIPAM segments are close to the gold surface, densely packed, less hydrated, and undergo the first transition. In the outer zone, on the other hand, the PNIPAM segments are looser and more hydrated, adopt a restricted random coil conformation, and show a phase transition, which is dependent on both particle concentration and the chemical nature of the end groups of the PNIPAM chains.

Monolayers of the amphiphilic gold nanoparticles at the air-water interface show several characteristic regions upon compression in a Langmuir trough at room temperature. These can be attributed to the polymer conformational transitions from a pancake to a brush. Also, the compression isotherms show temperature dependence due to the thermo-responsive properties of the tethered PNIPAM chains. The films were successfully deposited on substrates by Langmuir-Blodgett technique. The sessile drop contact angle measurements conducted on both sides of the monolayer deposited at room temperature reveal two slightly different contact angles, that may indicate phase separation between the tethered PNIPAM and PS chains on the gold core.

The optical properties of amphiphilic gold nanoparticles were studied both in situ at the air-water interface and on the deposited films. The in situ SPR band of the monolayer shows a blue shift with compression, while a red shift with the deposition cycle occurs in the deposited films. The blue shift is compression-induced and closely related to the conformational change of the tethered PNIPAM chains, which may cause a decrease in the polarity of the local environment of the gold cores. The red shift in the deposited films is due to a weak interparticle coupling between adjacent particles. Temperature effects on the SPR band in both cases were also investigated. In the in situ case, at a
constant surface pressure, an increase in temperature leads to a red shift in the SPR, likely due to the shrinking of the tethered PNIPAM chains, as well as to a slight decrease of the distance between the adjacent particles resulting in an increase in the interparticle coupling. However, in the case of the deposited films, the SPR band red-shifts with the deposition cycles more at a high temperature than at a low temperature. This is because the compressibility of the polymer coated gold nanoparticles at a high temperature leads to a smaller interparticle distance, resulting in an increase of the interparticle coupling in the deposited multilayers.
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACPA</td>
<td>4,4’-Azobis(4-cyanopentanoic acid)</td>
</tr>
<tr>
<td>AIBN</td>
<td>2,2’-Azobis(isobutyronitrile)</td>
</tr>
<tr>
<td>ATRP</td>
<td>Atom transfer radical polymerization</td>
</tr>
<tr>
<td>AuNP</td>
<td>Gold nanoparticles</td>
</tr>
<tr>
<td>Cpa</td>
<td>4-Cyanopentanoic acid dithiobenzoate</td>
</tr>
<tr>
<td>CPADB</td>
<td>4-Cyanopentanoic acid dithiobenzoate</td>
</tr>
<tr>
<td>Cumyl</td>
<td>2-Phenylprop-2-yl dithiobenzoate</td>
</tr>
<tr>
<td>DCC</td>
<td>N,N-dicyclohexylcarbodiimide</td>
</tr>
<tr>
<td>DLS</td>
<td>Dynamic light scattering</td>
</tr>
<tr>
<td>4-DMAP</td>
<td>4-Dimethyl aminopyridine</td>
</tr>
<tr>
<td>DMF</td>
<td>N,N-dimethylformamide</td>
</tr>
<tr>
<td>DP</td>
<td>Degree of polymerization</td>
</tr>
<tr>
<td>EDAC</td>
<td>1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride</td>
</tr>
<tr>
<td>HS-DSC</td>
<td>High-sensitivity differential scanning calorimetry, or microDSC</td>
</tr>
<tr>
<td>HRTEM</td>
<td>High-resolution transmission electron microscopy</td>
</tr>
<tr>
<td>I-M</td>
<td>Insulator-to-metal</td>
</tr>
<tr>
<td>LB</td>
<td>Langmuir-Blodgett</td>
</tr>
<tr>
<td>LbL</td>
<td>Layer-by-layer</td>
</tr>
<tr>
<td>LCST</td>
<td>Lower critical solution temperature</td>
</tr>
<tr>
<td>LDI-MS</td>
<td>Laser desorption-ionisation mass spectrometer</td>
</tr>
<tr>
<td>MPC</td>
<td>Monolayer-protected clusters</td>
</tr>
<tr>
<td>MUD</td>
<td>11-Mercapto-1-undecanol</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear magnetic resonance</td>
</tr>
<tr>
<td>PDI</td>
<td>Polydispersity index</td>
</tr>
<tr>
<td>PEO</td>
<td>Poly(ethylene oxide)</td>
</tr>
<tr>
<td>PNIPAM</td>
<td>Poly(N-isopropylacrylamide)</td>
</tr>
<tr>
<td>PNIPAM-COOH</td>
<td>Carboxyl-terminated poly(N-isopropylacrylamide)</td>
</tr>
<tr>
<td>PNIPAM-SH</td>
<td>Thiolated poly(N-isopropylacrylamide)</td>
</tr>
<tr>
<td>PNIPAM-AuNP</td>
<td>Poly(N-isopropylacrylamide) protected gold nanoparticles</td>
</tr>
<tr>
<td>PNIPAM/PS-AuNP</td>
<td>Poly(N-isopropylacrylamide) and polystyrene protected gold nanoparticles</td>
</tr>
<tr>
<td>PS</td>
<td>Polystyrene</td>
</tr>
<tr>
<td>PVP</td>
<td>Polyvinylpyrrolidone</td>
</tr>
<tr>
<td>RAFT</td>
<td>Reversible addition-fragmentation chain transfer</td>
</tr>
<tr>
<td>RT</td>
<td>Room temperature</td>
</tr>
<tr>
<td>SAM</td>
<td>Self-assembled monolayers</td>
</tr>
<tr>
<td>SEC</td>
<td>Size exclusion chromatography</td>
</tr>
<tr>
<td>SPR</td>
<td>Surface plasmon resonance</td>
</tr>
<tr>
<td>THF</td>
<td>Tetrahydrofuran</td>
</tr>
<tr>
<td>TGA</td>
<td>Thermogravimetric analysis</td>
</tr>
<tr>
<td>XRD</td>
<td>X-ray diffraction</td>
</tr>
</tbody>
</table>
Symbols

\( \delta \) Chemical shift in NMR spectrum
\( \lambda_{\text{max}} \) Wavelength corresponding to absorption maximum of the SPR band
\( A_{\text{max}} \) Absorbance maximum of the SPR band
\( I_{\text{abs}} \) Absorbance at 650 nm in UV-vis spectrum
\( I_{\text{scattering}} \) Intensity of scattered light in dynamic light scattering
\( R_h \) Hydrodynamic radius
\( M_n \) Number average molecular weight
\( M_w \) Weight average molecular weight
\( \Delta H \) Enthalpy change
\( \Delta H^{\text{cal}} \) Calorimetric enthalpy change
\( \Delta H^{\text{v}} \) Van’t Hoff enthalpy change
\( R \) Ratio between the van’t Hoff enthalpy change and calorimetric enthalpy change, i.e., \( \Delta H^{\text{v}}/\Delta H^{\text{cal}} \)
\( T_m \) Temperature of maximum heat capacity
\( \pi-A \) Surface pressure vs. nanoparticle area compression isotherm
\( \gamma \) Surface tension
\( A_t \) Nanoparticle area at the onset of the pancake-to-brush transition in compression isotherm
\( \pi_t \) Surface pressure at the onset of the pancake-to-brush transition in compression isotherm
\( A_b \) Nanoparticle area at the brush stage in compression isotherm
\( \pi_b \) Surface pressure at the brush stage in compression isotherm
\( L_t \) Length of the PNIPAM chain at the onset of the pancake-to-brush transition in compression isotherm
\( L_b \) Length of the tethered polymer chain at the air-water interface at the brush stage
List of original papers

This thesis is based on the following five publications, which are referred to in the text by their Roman numerals I-V:


Author’s contribution to the publications:
Jun Shan was in close cooperation with Janne Raula to draw up the research plan and to prepare the manuscript for the publication I. For the other four publications, Jun Shan independently drew up the research plan and wrote the manuscripts.
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1. INTRODUCTION

1.1 Background

Nanoparticles in the 1-100 nm range can be said to bridge the gap between small molecules and bulk materials. As a class of materials, nanoparticles offer a variety of opportunities to investigate the evolution of material properties with particle dimensions. In fact, metal nanoparticles, especially gold, silver and copper nanoparticles, have been extensively investigated over the past decade due to their unique electronic, optical and catalytic properties. These properties are neither those of bulk metal nor those of molecular compounds as have been widely demonstrated in both experimental and theoretical investigations, but they strongly depend on the particle size, shape of the nanoparticles, and interparticle distance as well as the nature of the protecting organic shell.

The chemical stability of the particles is crucial to avoid degradation processes such as partial oxidation or undesired sintering of particles. The lack of sufficient stability of many nanoparticles has to some extent impeded the development of real world applications of nanomaterials. As we have been seeing, gold plays a special role in nanoscience and nanotechnology. This is due to, firstly, the fact that gold is the most stable noble metal at the nano-scale, while most less noble metals will be oxidised to a depth of a thousand nanometers or more, in many cases obliterating the nanoscale component. So the designers of any nano-device requiring metallic components are likely to consider gold favourably. Secondly, gold is a far better electron conductor than silicon, (whereas next to copper and silver). Thirdly, gold offers a unique surface chemistry that allows it to be used as a platform on which to self-assemble layers of organic molecules, usually bound to the gold by sulfur atoms. Such "self-assembled" structures may be used as sensitive biomedical or chemical sensors. Finally, gold is readily fabricated at the nanoscale by electrolytic or electroless deposition and may be further modified by straightforward extensions of existing lithographic technologies. Overall, better understanding and control of the essential properties of gold nanoparticles may lead to fascinating potential applications to areas such as outlined in Scheme 1.

1.2 Synthetic methods of monolayer-protected gold nanoparticles

To date, a number of procedures for synthesis of gold nanoparticles have been reported, of which the most widely used are wet chemistry techniques based on chemical reactions in solution that yield metal nanoparticle colloids with a wide range of sizes, shapes, and dielectric environments. Probably the most popular method of preparing Au nanospheres dispersed in water is the reduction of HAuCl\textsubscript{4} in a boiling sodium citrate solution. The average particle diameter can be tuned over a wide range (~ 10-100 nm) by varying the concentration ratio between HAuCl\textsubscript{4} and sodium citrate. Another impressive procedure that has become extremely popular for Au nanoparticle synthesis is the two-phase reduction method developed by Schiffrin and coworkers. As a result, alkanethiolate-protected gold nanoparticles (so called monolayer-protected clusters,
MPCs were produced with tunable particle size between ~ 1 and 10 nm depending on the ratio of the Au salt and the ligand (alkanethiol). Later on, Murry and coworkers enhanced the popularity of the method and explored routes to functionalized MPCs by ligand exchange reactions. Alkanethiolate MPCs can be repeatedly isolated from and redissolved in common organic solvents without irreversible aggregation or decomposition. The particles can also be kept in solid state under ambient conditions for long term without significant ageing effects.

The use of polymeric stabilizing species for metal nanoparticles has inspired the studies of various synthetic routes to link polymers to metal particles. Though an early report can be dated back to 1718, where starch was reported to stabilize water-soluble gold particles, much effort has been recently put to prepare polymer-stabilized gold nanoparticles (and also other metal particles) due to quantum-size-related interest. In these core-shell composite metal nanoparticles a polymer layer may be physically or chemically bound to a metal core. Gold nanoparticles have been generated in the presence of polyvinylpyrrolidone, PVP, or in polymeric micelles composed of amphiphilic block/star-block copolymers, where polymer chains were physically and randomly adsorbed on the surface of gold cores. The synthesis of gold nanoparticles with chemically bound polymer grafts can be carried out in two ways. In a “grafting-to” method, polymers end-capped with a thiol or dithioester, and polymers with a disulfide unit have been used. Monomers may also be polymerized from the particle surfaces carrying polymerization-initiating species, i.e. a “grafting-from” method.

Scheme 1. Outline of some potential areas of gold nanoparticle application.
Clearly, polymer-protected gold nanoparticles possess the functionalities and solubilities derived from the polymer shell. It is significant that both methods can be combined with controlled/living radical polymerization techniques, such as ATRP and RAFT, to obtain a monodisperse and dense polymeric shell around the gold cores. In addition, with “grafting-to” method, even chemically different polymers synthesized by RAFT technique and terminated with a dithioester group can be simultaneously introduced to a single gold core surface. This allows not only the adjustment of the solubilities of polymer-protected gold nanoparticles but also the buildup of ordered monolayers/multilayers using nanoparticles as building blocks.

1.3 Assembly of gold nanoparticles

Current nanotechnology research rests heavily on the “bottom-up” approach compared to the “top-down” approach. The “bottom-up” approach offers much better flexibility in terms of material design. However, a significant challenge of this approach is how to assemble the nano building blocks into predefined and sophisticated objects of precisely controlled geometry. The assembling of gold and other metal nanoparticles into two and three-dimensional ordered structures offers a special pathway for design of materials that have potential applications in optical and microelectronic devices, chemical and biosensor technology, optical energy transport, and in advanced spectroscopy (such as surface-enhanced Raman scattering). Self-assembling, templated-assembling and other techniques have extensively been used to achieve this goal. To date, many reports have been published on the use of, for example, Langmuir-Blodgett technique for deposition of monolayers and multilayers of gold and silver nanoparticles protected either by low molar mass organic substances or by polymers; electrostatic (or spin assisted) layer-by-layer (LbL) assembly to construct homogeneous films; and on the chemical and biological self-assembly. Langmuir trough enables a very high degree of control over the layer density and particle ordering, since the monolayer at the interface can be manipulated by compressing to a desired surface pressure prior to the transfer of the particles on a substrate. For the ligand-protected metal nanoparticles, the interparticle distance can be tuned in a Langmuir trough to be much less than that obtained by evaporating a nanoparticle solution on a suitable substrate. In the latter case, the separation between particles is controlled only by the length of the ligand chain. Therefore, Langmuir trough is especially useful in the construction of a closely packed monolayer of polymer-protected metal nanoparticles.

1.4 Optical properties of gold nanoparticles

The most interesting aspects of metal nanoparticles are their optical properties. The use of metal particles as decorative pigments may be dated back to the Roman times. Perhaps the most famous example is the Lycurgus Cup that was manufactured in the 5th to 4th century AD and possesses the unique feature of changing color depending upon the light in which it is viewed. It is ruby red in transmitted light and green in reflected light, due to the presence of a very small amount of tiny (70 nm) metal crystals containing Ag and
Au in an approximate molar ratio of 14:1. However, the scientific research on metallic colloids dates back to 1857 when Michael Faraday reported a systematic study of the synthesis and colors of colloidal gold.\textsuperscript{54} In 1908, Mie presented a solution to Maxwell’s equations that describe the extinction spectra (extinction = scattering + absorption) of spherical particles of arbitrary size.\textsuperscript{1,55} Mie’s solution is interesting still today, but the modern generation of metal nanoparticle science has provided new challenges for the theory.

An important characteristic of gold, silver and other noble metal nanoparticles is the surface plasmon resonance (SPR) in the visible spectrum, which gives rise to intense colors. SPR is due to the collective oscillation of the conduction band electrons on the particle surface induced by the interacting electromagnetic field. The SPR is absent for gold nanoparticles with core diameter less than 2 nm due to their conduction electrons being in discrete levels, while bulk gold has a continuous absorbance in the UV/Vis/IR region.\textsuperscript{3} In the construction of macrostructures using individual nanoparticles as elementary building blocks, one should realize that there are two factors that dominate the optical properties of the assembly. Firstly, the optical properties of small individual nanoparticles are strongly dependent upon the size and shape of the particles,\textsuperscript{56-59} and also upon their local environment such as the stabilizing ligand shell and the surrounding solvent\textsuperscript{60-66}, as well as the charging state of the metal cores.\textsuperscript{62,64,67} Secondly, when nanoparticles are sufficiently close to each other, the interparticle coupling begins to take effect and the interparticle distance becomes an important parameter to influence the optical properties. As reported,\textsuperscript{44-46,49} the SPR of the assemblies of metal nanoparticles can be significantly tuned by varying the interparticle distance.
1.5 Objectives of this study

The first objective of this study was to synthesize novel functionalized gold nanoparticles protected with different polymers. The polymer-protected gold nanoparticles were then expected to have different solubilities in water and in organic solvents. Accordingly, water-soluble and insoluble particles were obtained, depending on the polymers used in the synthesis. The water-insoluble particles were, however, amphiphilic.

To synthesize polymer-protected gold nanoparticles, synthesis methods were systematically studied using both “grafting-to” and “grafting-from” techniques. Controlled radical polymerization, RAFT, was used in the synthesis. In this way, monodisperse polymers with well-defined molar masses were obtained and bound to the gold cores. Gold nanoparticles with monodisperse shells were thus produced.

Two kinds of polymers with various molar masses, poly(N-isopropylacrylamide), PNIPAM, and polystyrene, PS, were chosen for this study and synthesized by a RAFT process. Accordingly, water-soluble gold nanoparticles protected only by PNIPAM chains, PNIPAM-AuNPs, were studied with respect to the thermal behavior. This was the second objective in the study.

The third objective was to build up 2D-nanostructures, monolayers and multilayers of the gold nanoparticles, by using the Langmuir-Blodgett technique. Amphiphilic gold nanoparticles protected with a mixture of two homopolymers, PNIPAM and PS, were employed. The compression isotherms of the monolayers were measured in a Langmuir trough both at room temperature and at elevated temperatures close to the lower critical solution temperature (LCST) of PNIPAM.

Because the separation between particles in a monolayer of gold nanoparticles can easily be tuned in a Langmuir trough, the optical properties of 2D-nanostructures were further investigated both in situ at the air-water interface upon compression and on the deposited multilayers upon deposition cycles. The temperature effects on the optical properties were also considered, because of the known thermo-responsive properties of PNIPAM tethered on the amphiphilic gold nanoparticles. This was the fourth objective of this study.
2. EXPERIMENTAL

This section summarizes the synthetic methods and the characterization of both the precursor polymers and the gold nanoparticles, AuNPs, protected with the polymers. Then, the studies of the thermal behaviors and the buildup of Langmuir monolayers and deposited films of AuNPs will be described. PNIPAM and PS were synthesized by RAFT process, and two kinds of gold nanoparticles were prepared, i.e., the water-soluble PNIPAM-AuNPs and the water-insoluble, amphiphilic PNIPAM/PS-AuNPs.

2.1 Polymer synthesis

2.1.1 PNIPAM and PS synthesized by RAFT polymerization

To obtain gold nanoparticles protected with a monodisperse polymer shell, monodisperse PNIPAM and PS with various molar masses were synthesized by a RAFT process where two types of RAFT agents, 2-phenylprop-2-yl dithiobenzoate (cumyl-RAFT agent) and 4-cyanopentanoic acid dithiobenzoate (cpa-RAFT agent), were employed for both monomers. Therefore, the resulting polymers are prefixed with cumyl- or cpa-, respectively. Scheme 2 illustrates the synthetic routes of both types of polymers. The details of the RAFT polymerization reactions are collected in Table 1. The RAFT-polymers were characterized by $^1$H NMR and size exclusion chromatography (SEC), and the results are summarized in Table 2.

![Scheme 2. The schematic representation of reaction routes for RAFT-PNIPAM and -PS.](image-url)
Table 1. Summary of the RAFT polymerization conditions

<table>
<thead>
<tr>
<th>RAFT-polymer</th>
<th>[Monomer] (mol/L)</th>
<th>[RAFT] (mmol/L)</th>
<th>[Initiator] (mmol/L)</th>
<th>[RAFT]/[Initiator]</th>
<th>V (mL)</th>
<th>T (°C)</th>
<th>T (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>cumyl-PNIPAM-1</td>
<td>1.77</td>
<td>9.18</td>
<td>0.76</td>
<td>12.1</td>
<td>120</td>
<td>60</td>
<td>48</td>
</tr>
<tr>
<td>cpa-PNIPAM-2</td>
<td>1.77</td>
<td>11.8</td>
<td>0.75</td>
<td>15.7</td>
<td>60</td>
<td>60</td>
<td>28</td>
</tr>
<tr>
<td>cpa-PNIPAM-3</td>
<td>1.77</td>
<td>8.95</td>
<td>0.57</td>
<td>15.7</td>
<td>30</td>
<td>60</td>
<td>48</td>
</tr>
<tr>
<td>cpa-PNIPAM-4</td>
<td>1.77</td>
<td>8.95</td>
<td>0.65</td>
<td>13.8</td>
<td>60</td>
<td>60</td>
<td>48</td>
</tr>
<tr>
<td>cumyl-PS-1</td>
<td>8.73</td>
<td>24.8</td>
<td>2.10</td>
<td>12.0</td>
<td>12</td>
<td>60</td>
<td>48</td>
</tr>
<tr>
<td>cpa-PS-2</td>
<td>6.40</td>
<td>3.60</td>
<td>0.47</td>
<td>7.70</td>
<td>26</td>
<td>60</td>
<td>48</td>
</tr>
</tbody>
</table>

Notes: i) The initiator is AIBN in the syntheses of all RAFT-polymers. ii) The solvent used in the polymerization is dioxane for synthesis of all PNIPAM samples and cpa-PS-2. However, for cpa-PS-2, the total volume equals to the volume of dioxane (15 mL) plus styrene volume (11 mL). iii) An exception is cumyl-PS-1 which was synthesized in bulk with styrene volume of 12 mL. iv) All the reaction solutions were degassed by freeze-thaw cycles at least three times.

Table 2. The molecular characteristics of polymers

<table>
<thead>
<tr>
<th>RAFT-polymer</th>
<th>M_n (g/mol)</th>
<th>DP</th>
<th>PDI</th>
<th>used in article</th>
</tr>
</thead>
<tbody>
<tr>
<td>cumyl-PNIPAM-1</td>
<td>4722</td>
<td>40</td>
<td>1.10</td>
<td>II, III</td>
</tr>
<tr>
<td>cpa-PNIPAM-2</td>
<td>3000</td>
<td>24</td>
<td>1.16</td>
<td>V</td>
</tr>
<tr>
<td>cpa-PNIPAM-3</td>
<td>5418</td>
<td>46</td>
<td>1.13</td>
<td>II, III</td>
</tr>
<tr>
<td>cpa-PNIPAM-4</td>
<td>6450</td>
<td>55</td>
<td>1.11</td>
<td>IV</td>
</tr>
<tr>
<td>cumyl-PS-1</td>
<td>3200</td>
<td>28</td>
<td>1.07</td>
<td>V</td>
</tr>
<tr>
<td>cpa-PS-2</td>
<td>10000</td>
<td>94</td>
<td>1.07</td>
<td>IV</td>
</tr>
</tbody>
</table>

2.1.2 Synthesis and modification of carboxyl-terminated PNIPAM

Carboxyl-terminated PNIPAM, PNIPAM-COOH (M_n 10884 g/mol and PDI 1.52), was synthesized by conventional free radical polymerization using 4,4’-Azobis(4-cyanopentanoic acid), ACPA, as an initiator. PNIPAM-COOH was further modified into a thiolated PNIPAM, PNIPAM-SH, through the formation of an amide bond between the primary amino group of cysteamine and the carboxylic acid end group of PNIPAM.72-73 The carboxylic acid groups were activated by using 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride, EDAC. The thiol content of the PNIPAM-SH sample, determined by iodometric titration, was 17.6 µmol/g, i.e., 5 chains of PNIPAM only contain 1 SH group averagely. The reaction route is presented in Scheme 3 below.
Scheme 3. The schematic representation of reaction routes for PNIPAM-COOH and -SH.

2.2 Synthesis of polymer-protected gold nanoparticles

The synthesis of gold nanoparticles stabilised with polymers chemically bound through a sulfur atom can be carried out by two techniques, i.e., “grafting-from” and “grafting-to”.

2.2.1 PNIPAM-AuNPs by a “grafting-from” technique

The gold nanoparticles with PNIPAM grafts on the surface were prepared in three steps, as shown in Scheme 4. First, the functional gold particles protected with 11-mercapto-1-undecanol (MUD) were synthesized by the method of Yee et al. Secondly, the resulting gold particles were modified via connecting 4-cyanopentanoic acid dithiobenzoate (CPADB, i.e., cpa-RAFT agent) molecules to the MUD ligands by esterification. Thirdly, the CPADB modified gold particles were used as macroRAFT agents in the RAFT polymerisation of NIPAM. The feed for polymerisation included 2.1 mmol of NIPAM, 0.0004 mmol of AIBN as an initiator, and 70 mg of the CPADB modified gold particles, which contained 0.0054 mmol of CPADB. The molar ratios in the feed were, NIPAM:CPADB = 389:1 and CPADB:AIBN = 13.5:1, respectively. The polymerization
was conducted in 1.5 mL of DMF at 60 °C for 48 h. The molar mass and the polydispersity of the PNIPAM ligand was determined by SEC after the removal of PNIPAM from the gold surface, \( M_w = 20000-21000 \text{ g/mol} \) and PDI = 1.1-1.2.

**Scheme 4.** The synthetic steps for PNIPAM-AuNPs by a “grafting-from” technique.

### 2.2.2 PNIPAM-AuNPs by a “grafting-to” technique

Three types of PNIPAM products, cumyl-PNIPAM-1, cpa-PNIPAM-3 and thiolated PNIPAM-SH, described above (Table 2) were used to prepare gold nanoparticles by a “grafting-to” technique. Three synthetic routes were investigated as described in the article II, and are outlined in Scheme 5 below.
Because that the RAFT-polymers terminated with a dithioester group can readily be hydrolyzed using a base like hydrazine, the RAFT-polymers were directly employed in the synthesis of gold nanoparticles without prehydrolysis. The reductant, LiB(C₂H₅)₃H, also a strong base, can reduce HAuCl₄ to Au (0) and simultaneously hydrolyze the dithioester end group to a thiol which immediately protects the gold nanoparticles. This method is referred to as the one-step way described in the article II (see also (1) in Scheme 5). The route defined as the two-step way was also successfully used to prepare gold nanoparticles. This route was started with the pre-hydrolysis of RAFT-PNIPAM into a thiolated PNIPAM by hydrazine (see (2) in Scheme 5). But the three-step way failed in producing individual gold nanoparticles, instead, cross-linked particle aggregates were obtained.

As outlined in Table 3, the concentration of HAuCl₄ was always kept at 0.1 mmol, and the molar ratio of LiB(C₂H₅)₃H/HAuCl₄•xH₂O was over 10/1. The amount of polymer required in the syntheses was calculated taking into account the content of either dithioester or thiol in the polymers, which varied in the range from 1/1 to 1/12 of the amount of HAuCl₄. The gold core size was tuned mainly with the ratio between polymer and HAuCl₄. The resulting mixtures were purified through centrifugation at 15000 rpm to remove a small amount of big particles, followed by ultrafiltration with a membrane of the molar mass cutoff 10000 using deionized water as eluent. In the latter process, the filtrates were collected and checked by SEC in order to ensure that all the unreacted polymers were removed.

![Scheme 5](image)

**Scheme 5.** The synthetic routes for PNIPAM-AuNPs by a “grafting-to” technique.
### Table 3. Summary of the syntheses of PNIPAM-AuNPs

<table>
<thead>
<tr>
<th>PNIPAM-AuNPs</th>
<th>method</th>
<th>PNIPAM:HAuCl(_4)</th>
<th>HAuCl(_4) (mmole)</th>
<th>LiB(C(_2)H(_5))(_3)H:HAuCl(_4)</th>
<th>V(_{\text{THF}}) (mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/1)(^a)</td>
<td>two-step</td>
<td>1:1</td>
<td>0.1</td>
<td>10:1</td>
<td>20</td>
</tr>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/5)</td>
<td>two-step</td>
<td>1:5</td>
<td>0.1</td>
<td>10:1</td>
<td>20</td>
</tr>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/12)</td>
<td>one-step</td>
<td>1:12</td>
<td>0.1</td>
<td>10:1</td>
<td>20</td>
</tr>
<tr>
<td>cpa-PNIPAM-AuNPs (1/5)</td>
<td>one-step</td>
<td>1:5</td>
<td>0.1</td>
<td>10:1</td>
<td>20</td>
</tr>
<tr>
<td>cpa-PNIPAM-AuNPs (1/12)</td>
<td>one-step</td>
<td>1:12</td>
<td>0.1</td>
<td>10:1</td>
<td>20</td>
</tr>
</tbody>
</table>

\(^a\) The ratio in the bracket denotes the molar feed ratio of PNIPAM:HAuCl\(_4\) used in the synthesis of gold nanoparticles.

#### 2.2.3 Amphiphilic AuNPs by a "grafting-to" technique

Four types of amphiphilic gold nanoparticles, grafted with a mixture of PNIPAM and PS chains of different molar masses, were prepared through a one-step way in a homogeneous THF phase with a "grafting-to" technique.\(^{27,74}\) The synthetic method and the structure of amphiphilic gold nanoparticles are illustrated in Scheme 6. The recipes for the syntheses of amphiphilic gold particles, analogous to those above for the syntheses of PNIPAM-AuNPs, are summarized in Table 4. Both ultrafiltration and washing using appropriate solvents were applied to remove all unreacted polymers. None of the four products were water-soluble.

![Scheme 6](image)

**Scheme 6.** The synthetic method and structure of amphiphilic AuNPs by a “grafting-to” technique.

### Table 4. Summary of the syntheses of four types of amphiphilic gold nanoparticles

<table>
<thead>
<tr>
<th>AuNPs</th>
<th>Polymer used (M(_n))</th>
<th>PNIPAM:PS:HAuCl(_4)</th>
<th>LiB(C(_2)H(_5))(_3)H:HAuCl(_4)</th>
<th>V(_{\text{THF}}) (mL)</th>
<th>Used in article</th>
</tr>
</thead>
<tbody>
<tr>
<td>AuNP-1</td>
<td>cpa-PNIPAM-4 (6450)</td>
<td>1 : 1 : 10</td>
<td>10 : 1</td>
<td>20</td>
<td>IV</td>
</tr>
<tr>
<td></td>
<td>cpa-PS-2 (10000)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AuNP-2</td>
<td>(same as AuNP-1)</td>
<td>1 : 2 : 10</td>
<td>10 : 1</td>
<td>20</td>
<td>IV</td>
</tr>
<tr>
<td>AuNP-3</td>
<td>cpa-PNIPAM-2 (3000)</td>
<td>1 : 1 : 10</td>
<td>10 : 1</td>
<td>20</td>
<td>V</td>
</tr>
<tr>
<td></td>
<td>cumyl-PS-1 (3200)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AuNP-4</td>
<td>(same as AuNP-3)</td>
<td>1 : 2 : 10</td>
<td>10 : 1</td>
<td>20</td>
<td>V</td>
</tr>
</tbody>
</table>
2.3 Characterization

All the polymers utilized in this study synthesized via either RAFT polymerization or conventional free radical polymerization were characterized by FT-IR, $^1$H NMR and SEC with respect to the composition, the molar mass and polydispersity. For PNIPAM samples, $^1$H NMR was also applied to determine their molar masses. The SEC method used seems to be reliable for PNIPAM with low molar mass ($< 5 \times 10^4$ g/mol).$^{68}$

All the gold nanoparticles prepared in this study were subsequently characterized by

i) High-resolution transmission electron microscopy, HRTEM, for the size distribution and mean diameter of gold cores estimated from the enlarged micrographs. In the articles I-IV, the shape of the Au core was taken as the truncated octahedron.$^{75-76}$ Therefore, the number of Au atoms contained per core, the number of surface atoms, and the surface area for each sample were taken from refs. 77 and 78. However, for the two amphiphilic AuNPs employed in the article V, the approximate number of gold atoms of a single core was calculated using the density (59 atoms/nm$^3$) of bulk fcc-Au with a supposed spheric model.$^{56}$

ii) Thermogravimetric analysis, TGA, for the information of the total amount of the polymer grafts and the residual gold. Thus, the number of polymer chains grafted to a single core was calculated, and also the footprint as well as the density of polymer chains on the surface of a single core could be estimated.

iii) $^1$H NMR and FT-IR for the composition of the protective ligands, especially, for estimation of the molar ratio of PNIPAM/PS chains bound to the gold cores for amphiphilic gold nanoparticles employed in the articles IV and V.

iv) Dynamic light scattering, DLS, to determine the individual size of polymer protected gold nanoparticles dissolved in an appropriate solvent (in the articles I-IV) and to study the thermo-responsive properties of PNIPAM-AuNPs in water (in the articles I and III).

v) UV-vis spectroscopy to record the surface plasmon resonance (SPR) absorption spectra of both the gold nanoparticle solutions and the deposited films on substrates. Also, the in situ spectroscopic studies of the Langmuir monolayers of amphiphilic AuNPs at the air-water interface were carried out in the article V.

vi) X-ray diffraction, XRD, for the average crystallite size of MUD-AuNPs prepared in the article I.
2.4 Aqueous solutions of PNIPAM-AuNPs

The thermo-responsive properties of the aqueous solutions of PNIPAM-AuNPs were investigated by means of both UV-vis spectroscopy and dynamic light scattering upon heating (I). The structure of this type of PNIPAM-AuNPs has been shown in Scheme 4 above.

Two types of gold nanoparticles, cumyl- and cpa-PNIPAM-AuNPs, prepared by a “grafting-to” technique, were employed (III). The structures and core sizes of the gold nanoparticles are shown in Scheme 8 in 3.2.2. The concentration and pH dependences of the phase transition of each PNIPAM brush were studied by high-sensitivity microcalorimetry.

2.5 Langmuir monolayers and deposited films of amphiphilic gold nanoparticles

Four types of amphiphilic gold nanoparticles (see Table 4) were employed for the assembly of nanoparticle monolayers in a Langmuir trough and for the deposition of nanoparticle films on substrates by Langmuir-Blodgett technique.

2.5.1 Surface isotherms

The surface pressure versus area compression isotherm (π-A curve) for each sample was obtained by using a KSV mini-trough with a constant barrier speed of 5 mm/min. The surface tension measurements were performed with the Wilhelmy plate technique. The particles were spread from dilute chloroform solutions (0.2 mg/mL). The subphase used was deionized water.

AuNPs-1 and -2 were employed and their surface isotherms were only conducted at 20 °C (IV). However, the surface isotherms of the other two samples in Table 4, AuNPs-3 and -4, where the gold cores were stabilized with both short PNIPAM and PS chains, were performed at 20, 30, and 35 °C, respectively (V). This was to observe the thermo-responsive properties in the compression isotherms that the amphiphilic AuNPs may have. Due to a slight difference in the isotherms between AuNPs-3 and -4, only the behaviour of AuNP-3 was presented.

2.5.2 Langmuir-Blodgett film deposition

The particle monolayers at the air-water interface were compressed at the desired temperatures to certain surface pressures with a constant barrier speed of 5 mm/min, and held over 10 min to allow for relaxation of the film before deposition. A selected substrate was lifted up and/or dipped into the water subphase through the particle monolayer with a constant deposition speed of 5 mm/min, meanwhile, keeping the surface pressure constant. The quality of the deposition was monitored by the transfer ratio.
Only AuNP-2 could be compressed to a densely packed monolayer. The monolayer was then deposited at 35 mN/m either on a hydrophilic SiO₂ substrate by vertically lifting up the substrate or on a hydrophobic substrate by dipping it into the subphase through the monolayer at the air-water interface. Therefore, both upper and lower sides of the AuNP-2 monolayer were obtained on which to conduct sessile drop contact angle measurements. Deposition of the monolayer on a copper grid for HRTEM measurement was achieved by the Schaefer transfer.

On the other hand, the depositions of the multilayers of AuNP-3 onto quartz slides at 30 mN/m were performed at 20 and 30 °C, respectively, by repeating up and down strokes. Later on, the quartz slides covered by deposited films on both sides were measured by UV-vis spectrophotometer at room temperature to get the absorption spectra. To study the structure by HRTEM, the particle monolayer was transferred onto a copper grid that was attached on a silicon slide by lifting up the slide with a speed of 5 mm/min.

### 2.5.3 In situ UV-vis spectroscopy of Langmuir monolayers

The in situ UV-vis spectroscopic studies of the monolayers of AuNP-3 at the air-water interface were carried out with a Photol MCPD-100 UV-visible spectrophotometer (Scheme 7). The visible light of the source was guided to the air-water interface with an optical fiber perpendicular to the interface. The signal reflected from the mirror on the bottom of Langmuir trough was detected by the same fiber. The particles were spread from a dilute chloroform solution (0.2 mg/mL) onto a pure water subphase in a KSV 3000 Langmuir trough at 20 °C. The trough was placed in a laminar flow cabin and was protected against natural daylight with an absorbing yellow semitransparent plastic sheet. The in situ UV-vis spectra were recorded at various surface pressures during the compression of the particle monolayer with a constant barrier speed of 5 mm/min at 20 °C and 30 °C, respectively.

![Scheme 7. Schematic representation of the measurement system for in situ UV-vis spectroscopic studies of Langmuir monolayer at the air-water interface.](image-url)
3. RESULTS AND DISCUSSION

3.1 Characterization of gold nanoparticles

3.1.1 PNIPAM-AuNPs synthesized by a “grafting-from” technique

PNIPAM-AuNPs were successfully synthesized in three steps by a “grafting-from” technique (see Scheme 4). The characteristics of the gold nanoparticles are summarised in Table 5. The sizes of gold nanoclusters were measured both by HRTEM and XRD. Whetten et al. systematically investigated the crystal structures of Au cores with different sizes, protected with small organic ligands such as alkanethiols, by using laser desorption-ionisation mass spectrometer (LDI-MS). They reported that the most likely packing structure of Au atoms is a truncated octahedron. This packing structure was also observed in this work, see Figure 1b. Comparison of the images of the MUD-AuNP (Figure 1a) and PNIPAM-AuNP (Figure 1b) shows the crystal structure and packing of the gold atoms in the clusters. The average radius of the Au core is about 1.6 nm. The molar mass and polydispersity of the PNIPAM ligand was determined by SEC after the removal of PNIPAM from the gold surface, see Table 5.

<table>
<thead>
<tr>
<th>Table 5. Characterization of gold nanoparticles.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>AuNPs</strong></td>
</tr>
<tr>
<td>-----------</td>
</tr>
<tr>
<td>MUD-AuNPs</td>
</tr>
<tr>
<td>PNIPAM-AuNPs</td>
</tr>
</tbody>
</table>

a) HRTEM; b) XRD; c) TGA; d) Assumed from the content of CPADB by ¹H NMR; e) SEC in THF with polystyrene standards.

Figure 1. TEM images of Au clusters. a) MUD-AuNP; b) PNIPAM-AuNP.

However, the amount of CPADB on the particle surface after esterification was only ca. 5-6 % of the total number of the surface ligands. It was expected that every CPADB group would be involved in the RAFT polymerisation and therefore, it can be estimated...
that there are about 13 PNIPAM chains grafted from the surface of each gold particle. The dispersity of the PNIPAM-AuNPs in water, acetone and THF were moderate, but excellent in DMF, ethanol and methanol. The low dispersity in the former solvents shows that the amount of PNIPAM grafts on the surface of the gold was not high enough to stabilise the particles in every solvent good for PNIPAM. However, $^1$H NMR and TEM investigations show that all the particles, the dispersed and non-dispersed, possess the polymer layer, and they are homogenous in structure.

### 3.1.2 PNIPAM-AuNPs synthesized by a “grafting-to” technique

PNIPAM homopolymer protected gold nanoparticles were successfully prepared via both the one-step way and the two-step ways (see (1) and (2) in Scheme 5), not in the three-step way (see the article II). The characteristics of the gold nanoparticles are outlined in Table 6. The first and second samples were prepared in the two-step way, whereas the rest three in the one-step way.

**Table 6. Outline of characteristics of PNIPAM-AuNPs**

<table>
<thead>
<tr>
<th>PNIPAM-AuNP</th>
<th>Diam. Au core (nm)</th>
<th>PNIPAM (wt%)</th>
<th>Formula of AuNP</th>
<th>Footprint/ density $^a$</th>
<th>Diam. AuNP (nm) $^b$</th>
<th>Shell thickness (nm) $^c$</th>
</tr>
</thead>
<tbody>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/1)</td>
<td>1.3 ± 0.4</td>
<td>90</td>
<td>PNIPAM$<em>{30}$Au$</em>{79}$</td>
<td>0.28 / 3.6</td>
<td>9.8</td>
<td>4.3</td>
</tr>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/5)</td>
<td>2.0 ± 0.8</td>
<td>78</td>
<td>PNIPAM$<em>{48}$Au$</em>{314}$</td>
<td>0.40 / 2.5</td>
<td>11.2</td>
<td>4.6</td>
</tr>
<tr>
<td>cumyl-PNIPAM-AuNPs (1/12)</td>
<td>2.2 ± 0.8</td>
<td>71</td>
<td>PNIPAM$<em>{53}$Au$</em>{459}$</td>
<td>0.45 / 2.2</td>
<td>11.4</td>
<td>4.5</td>
</tr>
<tr>
<td>cpa-PNIPAM-AuNPs (1/5)</td>
<td>1.5 ± 0.5</td>
<td>84</td>
<td>PNIPAM$<em>{26}$Au$</em>{116}$</td>
<td>0.42 / 2.4</td>
<td>10.0</td>
<td>4.3</td>
</tr>
<tr>
<td>cpa-PNIPAM-AuNPs (1/12)</td>
<td>2.3 ± 0.8</td>
<td>68</td>
<td>PNIPAM$<em>{44}$Au$</em>{459}$</td>
<td>0.54 / 1.8</td>
<td>12.3</td>
<td>5.0</td>
</tr>
</tbody>
</table>

$^a$ The footprint of a PNIPAm chain = the surface area of a gold core / the number of PNIPAM chains bound to the gold core, nm$^2$/chain. The surface density of PNIPAM chains is the reciprocal of the footprint, chain/nm$^2$; $^b$ The mean diameters of PNIPAM-AuNPs were determined by DLS in the dilute aqueous solutions; $^c$ The shell thickness of PNIPAM was calculated according to ½ (the mean diameter of PNIPAM-AuNP – the mean diameter of gold core).

It can be concluded that

1) the size of gold cores decreases with increasing the molar ratio of RAFT-PNIPAM/HAuCl$_4$ from 1/12 to 1/1 in the preparation, which is in good accordance with the previous studies of the alkanethiol monolayer-protected clusters.$^{77-79}$

2) PNIPAM is as good passivant as thiolated poly(ethylene glycol)$^{24}$ to protect the gold core from further growth when their molar masses are in the same order of magnitude. Polymers are more efficient ligands than small organic molecules such as alkanethiols, because the sizes of the gold cores protected with polymers are all smaller than those protected by alkanethiols as the similar ratios were used in the synthetich feeds in both cases.$^{77-79}$
3) two types of PNIPAMs with different end groups show a similar effectiveness to protect gold nanoparticles if using similar ratio (comparing cumyl-PNIPAM-AuNPs (1/12) with cpa-PNIPAM-AuNPs (1/12)).

4) by the three-step way, the gold nanoparticles mainly formed bundles and not the individual particles. This may be attributed to the existence of HS-PNIPAM-SH derived from the modification of HOOC-PNIPAM-COOH, which may bridge the gold clusters together through the telechelic thiol groups.

5) The surface density of the polymer bound to the gold cores is much higher than those observed for self-assembled polymer monolayers on the 2D gold surfaces.28

6) from the estimation of the PNIPAM shell thickness, an extended conic conformation may be assumed to be adopted by the polymer chains bound to the surface of gold cores, different from a random coil of a polymer dissolved in a good solvent.24

3.1.3 Amphiphilic PNIPAM/PS-AuNPs

Four types of amphiphilic gold nanoparticles prepared in the one-step way were characterized and the results are given in Table 7.

<table>
<thead>
<tr>
<th>Amphiphilic AuNPs</th>
<th>Mean diam. Au core (nm)</th>
<th>PNIPAM/PS (molar ratio)b</th>
<th>PNIPAM/PS/Au (wt%)c</th>
<th>Formula Used in article</th>
</tr>
</thead>
<tbody>
<tr>
<td>AuNP-1 (1/1/10)a</td>
<td>2.5 ± 0.7</td>
<td>1.0/0.18</td>
<td>57.5/17.8/24.7</td>
<td>PN33PS6Au459 IV</td>
</tr>
<tr>
<td>AuNP-2 (1/2/10)</td>
<td>3.0 ± 1.0</td>
<td>1.0/0.48</td>
<td>34.4/26.6/39.0</td>
<td>PN27PS13Au976 IV</td>
</tr>
<tr>
<td>AuNP-3 (1/1/10)</td>
<td>4.2 ± 0.9</td>
<td>1.0/1.4</td>
<td>11.4/17.2/71.4</td>
<td>PN24PS34Au2289 V</td>
</tr>
<tr>
<td>AuNP-4 (1/2/10)</td>
<td>3.8 ± 0.8</td>
<td>1.0/2.5</td>
<td>8.70/23.2/68.1</td>
<td>PN14PS35Au1695 V</td>
</tr>
</tbody>
</table>

a) The ratio in the bracket denotes the molar feed ratio of PNIPAM:PS:HAuCl₄ used in the synthesis of gold nanoparticles (see Table 4). b) The molar ratios between PNIPAM and PS chains covalently bound to the gold cores were determined by ¹H NMR. c) The mass losses were obtained by TGA measurement, in combination with the molar ratio of PNIPAM/PS from ¹H NMR (see the description in the following text).

In the amphiphilic gold nanoparticles, both PNIPAM and PS chains with a certain molar ratio are together bound to the gold cores. The absorption bands typical for each polymer can be found in their FT-IR spectra, and the grafting ratio between PNIPAM and PS can also be observed qualitatively. However, the molar grafting ratio can quantitatively be obtained by ¹H NMR from the integrals of the typical absorption peaks of each polymer. Figure 2 shows the ¹H NMR spectra of AuNP-1 and AuNP-2 studied in the article IV, where the resonance labeled a (δ = 3.99 ppm) represents the lone proton of the N-isopropyl groups of the PNIPAM chains, the resonances labeled b, c, and d (δ ~ 6.3 – 7.4 ppm) represent the five aromatic protons of the phenyl groups in the PS chains. In the spectrum (A), the integral of the resonance labeled a is 35.8%, and the total integral of the resonances labeled b, c, and d is 64.2%. In the spectrum (B), the integrals are 16.7% and 83.3%, respectively. Based on the spectra as well as the known degrees of polymerization of PNIPAM and PS, the important parameter, the molar grafting ratio of
PNIPAM/PS bound to the gold nanoparticles can eventually be calculated to be 5/1 and 2/1 for AuNP-1 and -2, respectively.

Figure 2. $^1$H NMR spectra of AuNP-1 (A) and AuNP-2 (B).

The mass loss due to the thermal degradation of polymer bound to the gold cores can be determined by TGA, but, the respective mass loss fractions of the PNIPAM and the PS chains cannot separately be recognized from the thermogravimetric curve. This is because both PNIPAM and PS show thermal decomposition in the same temperature range of 220-460°C. However, the respective mass loss fractions of the PNIPAM and the PS chains can be calculated by combining the molar grafting ratio obtained by $^1$H NMR, the total mass loss fraction of the polymers obtained from TGA, as well as the molar masses of both PNIPAM and PS. The respective numbers of the PNIPAM and the PS chains bound to a single gold core can hence be figured out by using the molar grafting ratio and the number of the gold cores derived from the observed gold residue in TGA.

In the samples listed in Table 7, the PS content and thus, the hydrophobicity increases steadily from AuNP-1 to AuNP-4. In fact, AuNP-1 and AuNP-2 both contain longer polymer chains than AuNP-3 and AuNP-4. The former two samples were used to build up Langmuir monolayers at the air-water interface and deposit the monolayer films on substrates (IV). The latter two were employed in studies on the thermally responsive character and optical properties of both the Langmuir monolayers and the deposited films.
3.2 Thermal behaviors of PNIPAM-AuNPs in water

3.2.1 Thermo-responsive properties in aqueous dispersions

For PNIPAM-AuNPs prepared by a “grafting-from” technique, the thermo-responsive properties of the PNIPAM graft with a molar mass of ca. 20000 g/mol was studied in a dilute solution by DLS. An aqueous PNIPAM-AuNP solution with the concentration of 0.29 g/l was slowly heated from 20 °C to 45 °C (Figure 3). At 20 °C the particles form aggregates with average hydrodynamic radius, \(<R_h>\), of 83 nm. As the solution is heated the aggregate size increases, the increase being most pronounced around the LCST of PNIPAM at the observed critical temperature of 34 °C. At 45 °C PNIPAM ligands are fully collapsed resulting in the aggregate radius of about 114 nm. The aggregates dissociated as temperature was decreased back to 20 °C. The reason why the PNIPAM-AuNPs form aggregates in water at room temperature may be attributed to the low number of PNIPAM ligands anchored on the AuNPs (see the discussion in 3.1.1). However, the aggregation degree at room temperature decreases with dilution. The dissociation of aggregates with dilution was observed in UV-vis measurement at room temperature as a blue-shift of the maximum of the surface plasmon band of the gold nanoparticles. Figure 4 shows the dependence of the \(\lambda_{\text{max}}\) of the surface plasmon band and \(I_{\text{abs}}\) at 650 nm on the particle concentration in water. The \(\lambda_{\text{max}}\) decreased from 535 nm with decreasing concentration, but levelled off at 525 nm. This is due to the breakage of the aggregates with dilution resulting in a decrease in the interparticle coupling.\(^2\)\(^-\)\(^3\)

![Figure 3](image1.png)

**Figure 3.** Average hydrodynamic radius, \(<R_h>\), of the aqueous PNIPAM-AuNP solution with increasing temperature. Particle concentration is 0.293 g/l.

![Figure 4](image2.png)

**Figure 4.** the \(\lambda_{\text{max}}\) of the surface plasmon band and the \(I_{\text{abs}}\) at 650 nm of the aqueous PNIPAM-AuNP solution as a function of particle concentration.
3.2.2 Two-phase transitions of tethered PNIPAM chains

High-sensitivity microcalorimetry was employed to study the thermal properties of the aqueous solutions of two types of PNIPAM-AuNPs, synthesized by a “grafting-to” technique. The diameters of the gold cores are similar in both cases, as is the number of NIPAM repeating unit around the cores. The difference is in the end groups of the PNIPAM chains (see Scheme 8).

As has been already discussed in the subsection 3.2.1, aqueous PNIPAM-AuNPs show thermo-responsive properties which originate from the tethered PNIPAM chains. This can be observed by DSC. It is known that aqueous solutions of linear PNIPAM with high molar mass (>15000 g/mol)\textsuperscript{80-82} reveal a sharp endothermic peak within a narrow temperature range centered at $T_m = 33-34^\circ C$. Microcalorimetry detects the disruption of the hydrogen bonds among the amide groups and water molecules in the course of the phase transition. The enthalpy change in the phase transition, $\Delta H$, is 5.5-7.5 kJ/mol per repeating unit,\textsuperscript{80-81,83-86} consistent with the loss of ca. one hydrogen bond per repeating unit upon phase transition.\textsuperscript{81,86-87}

\begin{align*}
\text{cpa-PNIPAM-AuNPs (1/5)} & \quad \text{cumyl-PNIPAM-AuNPs (1/1)} \\
\begin{array}{c}
\begin{tikzpicture}
\node at (0,0) {HOOC-CH$_3$-CN-HN-0} node at (1.5,0) {Au$_{116}$} ;
\end{tikzpicture}
\end{array} & \begin{array}{c}
\begin{tikzpicture}
\node at (0,0) {\text{CH$_3$-HN-0}} node at (1.5,0) {Au$_{79}$} ;
\end{tikzpicture}
\end{array}
\end{align*}

\text{Diameter of Au core, D = 1.5 ± 0.5 nm} \quad \text{D = 1.3 ± 0.4 nm}

\begin{align*}
\text{cpa-PNIPAM-3, } M_n = 5418 \text{ g/mol} & \quad \text{cumyl-PNIPAM-1, } M_n = 4722 \text{ g/mol}
\end{align*}

Scheme 8. The structures of two types of PNIPAM-AuNPs and the corresponding core sizes.

The tethered PNIPAM chains of both AuNPs exhibit two separate transition endotherms, i.e., the first transition with a sharp and narrow endothermic peak occurs at lower temperature, and the second one with a broader peak at higher temperature. The peak temperatures are shown in Figure 5. The endotherms for the free PNIPAM, cpa-PNIPAM-3 and cumyl-PNIPAM-SH (obtained from hydrolysis of cumyl-PNIPAM-1), as shown in Figure 6, are broad and asymmetric,\textsuperscript{80-82} The $\Delta H$ values for the free PNIPAMs are 6.70 kJ/mol and 6.28 kJ/mol per repeating unit, respectively, in good agreement with values reported in literature. Comparing both the peak temperatures and the peak shapes in Figure 5 to those in Figure 6, it can be concluded that neither of the peaks in Figure 5 are due to the corresponding free PNIPAMs.

The transition temperature of aqueous linear PNIPAM depends on the polymer concentration.\textsuperscript{82,88} The effect of the particle concentration on the phase transitions was subsequently studied for cpa-PNIPAM-AuNPs, see Figure 7. The thermograms show that
with decreasing concentration the first peak practically does not change in shape and the corresponding $T_{m1}$, the temperature of maximum heat capacity, only slightly shifts from 30.7 to 32.4°C, while the second transition shifts considerably and $T_{m2}$ changes from 35.2 to 43.1°C. The aqueous solutions of cumyl-PNIPAM-AuNP show a similar variation in their endotherms with dilution (not shown here, see the article III). Thermodynamic parameters calculated from the thermograms for cpa-PNIPAM-AuNPs are listed in Table 8.

![Figure 5](image1.png)  
**Figure 5.** Microcalorimetric endotherms for aqueous solutions of the cumyl-PNIPAM-AuNPs (0.19 mmol/L, solid line) and the cpa-PNIPAM-AuNPs (0.16 mmol/L, dotted line).

![Figure 6](image2.png)  
**Figure 6.** Microcalorimetric endotherms for aqueous solutions of the cumyl-PNIPAM-SH (0.21 mmol/L, solid line) and the cpa-PNIPAM-3 (0.18 mmol/L, dashed line).

![Figure 7](image3.png)  
**Figure 7.** Microcalorimetric endotherms for aqueous solutions of cpa-PNIPAM-AuNPs with varying concentration of PNIPAM brush (as denoted by arrow, concentrations 1.24, 0.62, 0.31, and 0.16 mmol/L).
The calorimetric enthalpy change ($\Delta H^{\text{cal}}$) is considered as a measure of the energy needed for the disruption of the hydrogen bonds and was found for linear PNIPAM to be about 5.5-7.5 kJ/mol per NIPAM unit as mentioned above. The van’t Hoff enthalpy change ($\Delta H^v$) derived from the consideration of the temperature dependence of the equilibrium process,\(^{89}\) is the enthalpy change of a chain segment which participates independently in the phase transition, and is shape-dependent, related to the width of the endotherm at its half-height.\(^{80,85}\) It can be seen in Table 8 that, 1) $\Delta H_1^{\text{cal}}$ is always lower than $\Delta H_2^{\text{cal}}$, implying that two distinct zones may exist in the tethered PNIPAM brush, i.e., a less hydrated zone and a more hydrated one; 2) the sum of $\Delta H_1^{\text{cal}}$ and $\Delta H_2^{\text{cal}}$ for each PNIPAM-AuNP is close to the enthalpy change for linear PNIPAM mentioned above; 3) the ratio of $R = \Delta H^v/\Delta H^{\text{cal}}$ may give information on the cooperativity of structural units of the polymers that cooperate with each other in the transition.\(^{89}\) In this study, $R_1 \gg R_2 > 1$ indicates that the tethered PNIPAM chains cooperate more in the less hydrated zone than in the more hydrated zone in the course of the phase transition.

### Table 8. Thermodynamic Parameters of cpa-PNIPAM-AuNPs

<table>
<thead>
<tr>
<th>C (mmol/L)</th>
<th>$T_{m1}$ (°C)</th>
<th>$T_{m2}$ (°C)</th>
<th>$\Delta H_1^{\text{cal}}$ (kJ/mol)</th>
<th>$\Delta H_2^{\text{cal}}$ (kJ/mol)</th>
<th>$(\Delta H_1^{\text{cal}} + \Delta H_2^{\text{cal}})$ (kJ/mol)</th>
<th>$\Delta H_1^v$ (kJ/mol)</th>
<th>$\Delta H_2^v$ (kJ/mol)</th>
<th>$R_1$</th>
<th>$R_2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.16</td>
<td>32.4</td>
<td>43.1</td>
<td>78.2 (&lt;1.70)&lt;sup&gt;a&lt;/sup&gt; 191&lt;sup&gt;b&lt;/sup&gt; (4.16)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.86</td>
<td>1421</td>
<td>376</td>
<td>18.2</td>
<td>1.97</td>
<td></td>
</tr>
<tr>
<td>0.31</td>
<td>32.0</td>
<td>41.6</td>
<td>84.0 (1.83)</td>
<td>5.41</td>
<td>1409</td>
<td>435</td>
<td>16.8</td>
<td>2.64</td>
<td></td>
</tr>
<tr>
<td>0.62</td>
<td>31.6</td>
<td>40.2</td>
<td>88.2 (1.92)</td>
<td>5.48</td>
<td>1375</td>
<td>460</td>
<td>15.6</td>
<td>2.80</td>
<td></td>
</tr>
<tr>
<td>1.24</td>
<td>30.7</td>
<td>35.2</td>
<td>82.8 (1.80)</td>
<td>5.71</td>
<td>1400</td>
<td>343</td>
<td>16.9</td>
<td>1.91</td>
<td></td>
</tr>
</tbody>
</table>

\(^{a}\) The calorimetric enthalpy change per mole of the tethered PNIPAM chains. \(^{b}\) In the bracket, the calorimetric enthalpy change per mole of the NIPAM units. \(^{c}\) The sum of the calorimetric enthalpy change per mole of the NIPAM units = $(\Delta H_1^{\text{cal}} + \Delta H_2^{\text{cal}})$. \(^{d}\) The van’t Hoff enthalpy change. \(^{e}\) The ratios of $R_1 = \Delta H_1^{v}/\Delta H_1^{\text{cal}}$ and $R_2 = \Delta H_2^{v}/\Delta H_2^{\text{cal}}$.

Each PNIPAM chain tethered to the gold cores is supposed to adopt a stretched conic conformation\(^{24}\) with a high surface density of ca. 2.4 PNIPAM chains/nm\(^2\) for both studied particles. The density of the PNIPAM shell increases from the outer surface towards the gold core. A model is therefore put forward to explain the two phase transitions, in which the tethered PNIPAM chains can be subdivided into two zones, i.e., the inner zone and the outer zone, as illustrated in Scheme 9. In the inner zone, the PNIPAM segments are less hydrated and densely packed, whereas in the outer zone the PNIPAM segments are more hydrated and looser, and may adopt a restricted coil-like conformation. As a result, that the inner zone collapses at lower temperature than the outer one may be understood, since the inner zone is denser than the outer one.\(^{88}\) A sharp peak derived from the inner zone in the thermograms may be due to the existence of a higher interchain cooperativity as the ratio $R_1 \gg 1$ indicated in Table 8. The concentration dependence of the two endotherms also supports the suggested model, i.e., with increasing concentration, the outer shell, not the inner one, should be most affected.
Scheme 9. The tethered PNIPAM chains composed of a less hydrated inner zone and a more hydrated outer zone.

Just recently, Liu and coworkers have observed a similar phenomenon, i.e., a double phase transition of the PNIPAM shell \( (M_w = 7.60 \times 10^5 \text{ g/mol}) \) grafted from a hydrophobic dendritic core.\(^9\) The peak temperatures were at 26 and 30 °C, respectively. It was postulated that the inner layer of the PNIPAM brush collapses first, followed by the collapse of the outer layer of the brush.

3.3 Langmuir monolayers of amphiphilic gold nanoparticles

3.3.1 Surface isotherms

Two types of amphiphilic gold nanoparticles, AuNP-1 and AuNP-2, were first employed to build up Langmuir monolayers at the air-water interface at room temperature by using a Langmuir trough. In these two samples, the polymers used are all longer than those in AuNP-3 and AuNP-4, see Tables 4 and 7.

Many amphiphilic linear and star block copolymers can self-assemble at the air-water interface into various microphase-separated structures, which are called surface-micelles.\(^91-9\) At low surface pressures, the hydrophobic blocks aggregate to form isolated structures of different shapes mainly depending on the block copolymer composition. The aggregates are stabilized by surface-active hydrophilic blocks spread on the water surface. The surface micelles exhibit various shapes from circular micelles to large planar aggregates via rod-like micelles, depending on the relative sizes of the two blocks. As the film is compressed, several regions that can be assigned to different polymer conformations, i.e., pancake, pancake-to-brush, and brush, can be recognized in the compression isotherms.

The amphiphilic AuNPs are expected to spread into a film at the air-water interface in a way analogous to these amphiphilic block copolymers, due to the similarities in their structures and properties. The PNIPAM chains tethered on the surface of a gold core via a
sulphur bridge preferentially adsorb on the water surface at the initial stage after spreading from a chloroform solution, whereas PS chains collapse towards the surface of a gold core during evaporation of chloroform, and consequently the gold-PS entity acts as a water-insoluble buoy at the air-water interface. The tethered PNIPAM chains at the air-water interface have an extended conformation, due to the strong anisotropic interactions among polymer chains arising from the high surface density and a repulsive effect of the collapsed PS on PNIPAM.

The compression isotherms of both AuNPs are shown in Figure 8. We can see that for both AuNPs, at large areas the tethered PNIPAM chains adsorbed at the air-water interface may be assumed to adopt a two-dimensional flattened conformation — a pancake-like structure. The surface pressure increases steadily with film compression as the surface density of the particles increases until a pseudoplateau occurs correspondingly. The pseudoplateau for the AuNP-1 monolayer is much more extended compared to that for the AuNP-2 monolayer, and it never reaches a densely packing region as occurred for the AuNP-2 monolayer. This was the case even if the AuNP-1 monolayer compression was started at a high initial surface pressure by spreading a relatively large amount of the chloroform solution of AuNP-1 onto water surface. A probable explanation for this phenomenon is that AuNP-1 dissolves into the water subphase at higher surface pressures due to its more hydrophilic character.

Figure 8. Compression isotherms of surface pressure versus area per nanoparticle for AuNP-1 and AuNP-2, conducted at 20 °C.

The pseudoplateau can be associated with the dissolution of PNIPAM chains into the water subphase when the PNIPAM surface density exceeds a critical concentration, indicative of the onset of the transition from two to three dimensions, i.e., the pancake to brush transition. Table 9 lists the isotherm parameters for both AuNPs, such as the surface pressures and the nanoparticle areas at both the onset of the pancake-to-brush transition.
transition ($A_t$ and $\pi_t$) and the brush stage ($A_b$ and $\pi_b$), respectively. These parameters were determined from the intersections of the linear fits to the corresponding adjacent regions (pancake, pancake-to-brush, and brush), as indicated in Figure 8.

The PNIPAM length ($L_t$) at the onset of the pancake-to-brush transition can be estimated from the nanoparticle area at the transition point $A_t$ by $L_t = (A_t/\pi)^{1/2} - D/2$, where $D$ is the diameter of the gold core, listed in Table 9 for both AuNPs. It is evident that the PNIPAM chains in the AuNP-1 are 2 nm longer than those in the AuNP-2, indicating that the PNIPAM chains in the AuNP-1 monolayer are more stretched at the air-water interface than those in the AuNP-2 monolayer. Comparing with the length of a fully stretched PNIPAM chain, ca. 13.8 nm, assuming that the PNIPAM backbone adopts an all trans conformation, we can conclude that the PNIPAM chains for both AuNPs adopt an extended conformation at the air-water interface at the transition stage, and the PNIPAM chain lengths are as large as about the half-length of the fully stretched PNIPAM.

### Table 9. Overview of parameters measured with compression isotherms for both AuNPs

<table>
<thead>
<tr>
<th>Amphiphilic AuNP</th>
<th>$\pi_t$ (mN/m)</th>
<th>$A_t$ (nm$^2$)</th>
<th>$\pi_b$ (mN/m)</th>
<th>$A_b$ (nm$^2$)</th>
<th>$L_t$ (nm)</th>
<th>$L_b$ (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AuNP-1</td>
<td>20.5</td>
<td>313.0</td>
<td>-</td>
<td>-</td>
<td>8.7</td>
<td>-</td>
</tr>
<tr>
<td>AuNP-2</td>
<td>25.3</td>
<td>212.1</td>
<td>28.0</td>
<td>80.0</td>
<td>6.7</td>
<td>3.54</td>
</tr>
</tbody>
</table>

By analysis of the limiting area of nanoparticles in the brush stage (i.e., the brush area) from the isotherm of AuNP-2, $A_b = 80.0$ nm$^2$ (at $\pi_b = 28.0$ mN/m), the diameter ($D$) of these particles can be calculated to be $D = 10.1$ nm according to $A_b = \pi(D/2)^2$, much larger than the diameter of the gold cores of 3.0 ± 1.0 nm. The length of the tethered polymer ($L_b$) can further be obtained by $L_b = (D - 3.0)/2$, to be 3.54 nm, about a half of the PNIPAM chain length ($L_t$) of 6.7 nm at the pancake-to-brush transition. This means that about half length of each PNIPAM chain dives into the water subphase, avoiding overlapping with each other,92-93,96 and the other half is still at the air-water interface, during compression from the onset of the pancake-to-brush transition to the initial brushing stage.

![HRTEM image of Langmuir monolayer of AuNP-2 transferred at $\pi = 35$ mN/m by Schaefer deposition at 20 °C.](image)
To illustrate the formation of a monolayer, the AuNP-2 film was transferred onto a carbon-coated copper grid by Schaefer deposition at $\pi = 35$ mN/m in the brush region (see the arrow in Figure 8). The HRTEM image is shown in Figure 9. We can see that the gold cores are well separated in the film with no overlapping, indicating that a monolayer of AuNP-2 forms at the air-water interface.

3.3.2 Thermo-responsive properties at the air-water interface

AuNP-3 and AuNP-4, both protected with short PNIPAM and PS chains (Tables 4 and 7), were parallelly studied with respect to the surface isotherms, in situ optical properties at the air-water interface and optical properties of the transferred films. However, only a slight difference in the properties between both amphiphilic AuNPs was found. Therefore, we only present the behavior of AuNP-3 in the article V.

The surface isotherms of AuNP-3 were measured at 20, 30, and 35 °C. It is known that the surface tension of water as a subphase in Langmuir trough decreases with increasing temperature, resulting in an increment of surface pressure of water itself (e.g., $\pi = \gamma_{20} - \gamma_{30}$, $\gamma$ is the surface tension of water at 20 and 30 °C, respectively). To clearly compare the three isotherms, two of them measured at 30 and 35 °C were normalized according to that at 20 °C by subtracting a corresponding increment of surface pressure, respectively. The isotherm measured at 20 °C and the normalized isotherms are shown in Figure 10.

![Figure 10](image-url)

**Figure 10.** The isotherm measured at 20 °C and the normalized isotherms measured at 30 and 35 °C, respectively.

The shapes of all isotherms are very similar to those for both linear and star block PS-b-PEO copolymers that have short PEO blocks and a low content of PEO. All isotherms in Figure 10 can be divided into three regions. At a large nanoparticle area, the surface pressure increases very slowly if at all ($\pi \approx 0$ mN/m) with compression (region I). This region is typically known as the “pancake” region due to the pancake-like structure that the hydrophilic block (e.g., PEO block for PS-b-PEO or PNIPAM in the present case) is...
assumed to adopt when adsorbed at the air-water interface in the initial stage. As the compression continues, the surface pressure, passing through the first critical area $A_1$ (defined by extrapolating the linear portion of the isotherm to $\pi = 0 \text{ mN/m}$), goes into the first rapid rise region II. A common feature for all isotherms is that no pseudoplateau appears. This is because the short PNIPAM chains tethered on the gold cores would be forced to submerge into water when compressing close to $A_1$ in the region II, as observed for the short PEO blocks at lower surface pressures.\textsuperscript{92-97} Upon further compression, after passing through an inflection point where $\pi \approx 19 \text{ mN/m}$, a second steep rise region III of the surface pressure starts and continues until the monolayer collapses at $\pi > \sim 35 \text{ mN/m}$. In this region, the particles are closely packed, and a brush conformation of the PNIPAM chains is expected. A significant feature in this region (III) is that both of the isotherms measured at 30 and 35 °C are more or less shifted to lower values of the nanoparticle area relative to that at 20 °C, i.e., a slight shift for that measured at 30°C but a pronounced shift for that at 35 °C. This is due to the thermal transition of PNIPAM. At higher temperatures close to the LCST of PNIPAM, the PNIPAM chains get dehydrated and shrink towards the gold cores, causing a decrease in the dimension of the coated particles. This leads to the shift of the isotherms (at 30 and 35 °C) observed in the regions II and III. In other words, the monolayers at higher temperatures can be compressed to smaller nanoparticle areas.

As a result, it can be concluded that the amphiphilic AuNPs show a thermally responsive feature in the compression isotherms. This type of amphiphilic AuNPs may have a high potential in building up ordered arrays/superlattices of metal nanoparticles for electronic and optoelectronic applications, because a key parameter — the interparticle distance — can be manipulated in a Langmuir monolayer by varying both the surface pressure and temperature.

3.3.3 Characterization of deposited monolayers

3.3.3.1 Interparticle distance in deposited monolayers

To observe the monolayer structure and estimate the distance between adjacent particles, the Langmuir monolayers of AuNP-3 compressed at 20 and 30 °C, respectively, were transferred onto grids at the desired surface pressures, corresponding to the regions (I), (II) and (III) in the isotherms. The micrographs are shown in Figure 11. The particles show no evident overlap in the monolayers during the whole compression. The difference between them is the interparticle distance in terms of the edge-to-edge distance between adjacent particles $(d-2r)$ and the normalized distance $(d/2r)$, calculated from micrographs and listed in Table 10.

It can be seen that the particles become closer to each other in a monolayer compressed at 30 °C than in the one at 20 °C. However, comparing to the alkanethiol protected AgNPs and AuNPs reported by Heath et al.\textsuperscript{39,100} and by Quinn et al.,\textsuperscript{42} one can observe that the polymer protected AuNPs could not be compressed as close to each others as the alkanethiol protected metal particles. The length of a fully stretched PNIPAM chain used
in this study is ca. 8.0 nm, assuming an all trans conformation for PNIPAM, while that of dodecanethiol (C_{12}SH) is ca. 1.5 – 2.0 nm. As can be seen from Figures 9 and 11 the polymer protected AuNPs in the compressed monolayer could not form closest packing like a hexagonal structure as has been the case with some alkanethiol protected metal nanoparticles.\textsuperscript{38,42}

Figure 11. HRTEM images of the transferred monolayers for AuNP-3 at 20 and 30 °C, and at the desired surface pressures, $\pi = 0$, 5 and 30 mN/m, respectively.

Table 10. The interparticle distance in terms of both edge-to-edge distance between adjacent gold cores, $d - 2r$, and the center-to-center distance normalized by the diameter of the gold cores, $d/2r$, estimated from HRTEM micrographs for AuNP-3.

<table>
<thead>
<tr>
<th>$\pi$ (mN/m)</th>
<th>$d - 2r$ (nm)</th>
<th>$d/2r$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>3.1</td>
<td>1.7</td>
</tr>
<tr>
<td>5</td>
<td>2.6</td>
<td>1.6</td>
</tr>
<tr>
<td>30</td>
<td>2.4</td>
<td>1.6</td>
</tr>
</tbody>
</table>

Note: $d$ is the center-to-center distance between adjacent gold cores, and $r$ is the radius of the gold core.

The interparticle distance in the assembly of metal nanoparticles is an important factor to impact optical properties. The optical properties of AuNP-3 monolayer will be investigated and compared with those of alkanethiol protected metal nanoparticles.
3.3.3.2 Contact angle measurements on deposited monolayers

Although the preparation of the amphiphilic gold nanoparticles takes place in a homogeneous THF phase, it may be assumed that the chemically different PS and PNIPAM chains tend to be unevenly distributed on the surface of the gold cores but form phase separated regions. In fact, this is extremely difficult to verify experimentally for nanoparticles. An evidence of the phase separation has recently been given in the mixed self-assembled monolayers (SAM) formed by the coadsorption of hydroxyl- and methyl-terminated alkanethiols with similar chain length on a bulk gold surface by means of friction force microscopy and chemical force microscopy.\(^{101}\)

To investigate the possible phase separation between the tethered PS and PNIPAM chains, the monolayers of AuNP-2 were deposited from the air-water interface to either a hydrophilic SiO\(_2\) substrate or a pretreated hydrophobic substrate at \(\pi = 35\) mN/m. Therefore, two sides, i.e., both upper and lower surfaces, of the AuNP-2 monolayer were obtained with LB transfer according to either lifting up or dipping in a substrate (corresponding to hydrophilic or hydrophobic transfer), and were then denoted as the surface-L and the surface-D, respectively. Later on, the sessile drop contact angle measurements were conducted on both surfaces.

One may expect that if there is any phase separation between the tethered PNIPAM and PS on the gold core, the contact angles on surface-L and surface-D should be different. Thus, a hydrophilic transfer would lead to a more hydrophobic surface (i.e., the surface-L) and a hydrophobic transfer to a more hydrophilic one (i.e., the surface-D), following the general rules of LB transfer. Table 11 lists the sessile drop contact angles of water measured on both surface-L and surface-D, respectively. It can be seen that the contact angle on the surface-L is ca. \(5^\circ\) higher than that on the surface-D. Thus, the surface-L shows a more hydrophobic character than the surface-D.

<table>
<thead>
<tr>
<th>Surface</th>
<th>Contact angle / °</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Literature data</td>
</tr>
<tr>
<td>PS film [102](^a)</td>
<td>90.93 ± 0.32(^b)</td>
</tr>
<tr>
<td>PS film [103]</td>
<td>88.42 ± 0.28(^b)</td>
</tr>
<tr>
<td>PS film [104]</td>
<td>86 ± 1(^b)</td>
</tr>
<tr>
<td>PNIPAM brush [105]</td>
<td>84 ± 5(^b)</td>
</tr>
<tr>
<td>PNIPAM brush [106]</td>
<td>66(^b)</td>
</tr>
<tr>
<td>Surface-L</td>
<td>82 ± 2(^c)</td>
</tr>
<tr>
<td>Surface-D</td>
<td>77 ± 2(^c)</td>
</tr>
</tbody>
</table>

\(^a\) the number in the bracket is the corresponding reference. \(^b\) advancing contact angle. \(^c\) sessile contact angle.

The contact angle can be affected by many factors, such as interfacial tension, chemical heterogeneity, surface roughness, reorganization, and the procedure of the film
The techniques applied to measure the contact angle, e.g., the sessile drop contact angle or the dynamic contact angle (advancing and receding contact angles) techniques, may give different values. It is known that the dynamic contact angle at low advancing speed is often, but not always, identical to the static contact angle. Table 11 also lists some literature data of advancing contact angles measured at low advancing speed for PS film and PNIPAM brush. By comparing the advancing contact angles of water on the PS films with the measured sessile drop contact angle of $82 \pm 2^\circ$ on the surface-L, it is evident that the surface-L is not as hydrophobic as the pure PS film, but shows a slightly hydrophilic nature. This may be speculated to be due to the coexistence of both the PNIPAM and PS chains on the surface-L. As discussed in 3.3.1, about half length of PNIPAM chains for AuNP-2 is still at the air-water interface in the brush region and thus coexists with the collapsed PS chains on the surface-L, after the hydrophilic transfer is performed. This part of the PNIPAM chains starts to reorganize and dissolves rapidly into a water drop when the contact angle measurement is conducted on the surface-L at room temperature, which results in a slightly hydrophilic character of the surface-L. On the other hand, by reference to the literature data of advancing contact angles of water on the PNIPAM brushes in Table 11, measured at room temperature, the sessile contact angle of $77 \pm 2^\circ$ on the surface-D is just the average of the literature data.

As a result of the contact angle measurements, it may be concluded that the phase separation between the tethered PNIPAM and PS on the surface of the gold cores may occur during the binding the polymer chains to the metal surface. However, this needs to be proved in the future studies.

3.4 Optical properties of amphiphilic gold nanoparticles

Gold nanoparticles have a feature of SPR in the visible spectrum, which makes gold nanoparticles colourful mainly dependent upon the size evolution. However, this feature is also dependent on the nature of protecting ligands. The amphiphilic gold particles, show thermo-responsive properties derived from the tethered PNIPAM chains, as has been shown above. Hence, it is of interest to show 1) how the in situ SPR band of the amphiphilic particles at the air-water interface behaves upon the monolayer compression, and how temperature does affect the SPR; 2) how the SPR bands do shift along the deposition cycles in multilayers.

3.4.1 In situ SPR bands at the air-water interface

The in situ UV-vis absorption spectra of AuNP-3 monolayers are demonstrated in Figure 12 as a function of surface pressure ($\pi$) during the compressions at 20 and 30 °C, respectively. During the whole compression from the initial stage ($\pi \approx 0$ mN/m) to the monolayer collapse, the SPR bands for AuNP-3 at 20 and 30 °C steadily shift to higher energies, i.e., blue shift. The observation was contrary to expectation that a red shift in the SPR would be observed when the particles are getting closer to each other with the monolayer compression. The red shift in the SPR has been reported by Heath and coworkers for alkyl thiol protected Ag nanoparticles and by Quinn and co-
workers\textsuperscript{42} for alkanethiol protected Au nanoparticles. Heath and coworkers\textsuperscript{39,100,107} observed an insulator-to-metal (I−M) transition by in situ tuning a Langmuir monolayer of thiol-passivated silver nanocrystals with recording the UV-vis reflectance spectra as the monolayer was compressed. They found that the interparticle distance $d/2r = 1.2$ can be referred to as a critical value at which a strong interparticle coupling occurs and causes a red shift in the SPR. In our case, however, a strong interparticle coupling is absent due to larger values of $d/2r$ as shown in Table 10, estimated from HRTEM micrographs for AuNP-3. Therefore, a question arose, what factor causes the blue shift in the SPR band during the monolayer compression of AuNP-3.

Figure 12. In situ UV-vis absorption spectra of the Langmuir monolayer of AuNP-3 at the air-water interface with the surface pressures ($\pi$). The arrow indicates the rise of the surface pressure from the initial compression stage to the monolayer collapse. (a) compressed at 20 °C, $\pi = 1.0 \rightarrow 45$ mN/m. (b) compressed at 30 °C, $\pi = 0 \rightarrow 44$ mN/m.

Figure 13 shows the peak position of the in situ SPR band as a function of the surface pressure for the AuNP-3 monolayer compressed at 20 and 30 °C, respectively. It can be seen that the peak of the SPR band drastically shifts to a lower wavelength during the initial increase of the surface pressure. This corresponds to the onset of the first rapid rise region II of the surface pressure in the isotherms for AuNP-3 (see Figure 10), where the initially flattened PNIPAM chains on the water surface start to submerge into the water subphase forming a brush-like structure. This implies that the conformational change of the PNIPAM chains at the air-water interface may significantly affect the optical properties of the amphiphilic AuNPs. We later conducted an experiment, in which the amphiphilic AuNPs spread on the water surface floated over two hours with no
compression of monolayer, and the UV-vis absorption spectra were recorded in situ in every 5 min. The absorption spectra did not show any changes. This experiment confirms that the blue shift in the in situ SPR band is induced by the monolayer compression. Moreover, we can see from Figure 13 that, with a further rise in the surface pressure, the peak position of the SPR band steadily decreases until levelling off, that is, no more drastic decrease is observed in the peak position in a broad range of the surface pressure spanning through the regions II and III in the isotherms (see Figure 10). In conclusion, we can see that the blue shift in the in situ SPR is compression-induced and is closely related to the conformational change of the PNIPAM chains, especially at the onset of the region II.

\[ \text{Figure 13. The peak position of in situ UV-vis absorption spectrum as a function of surface pressure for the AuNP-3 monolayers compressed at 20 and 30 °C, respectively.} \]

Mie–Drude theory,\textsuperscript{44,60-67} outlined in the article V as Supporting Information, shows how the SPR of metal nanoparticles shifts with factors such as the dielectric function of the surrounding medium, the thickness of the coating/shell layer, or the volume fraction \( g \) of the shell layer in a coated particle. The theory predicts that a red shift in the SPR band occurs i) with an increase of the solvent refractive index\textsuperscript{63}, ii) with an increase of the refractive index of the shell layer,\textsuperscript{66} \( \varepsilon_s = n_s^2 \), and iii) with increasing the chain length of ligands since \( g \) increases with the chain length.\textsuperscript{62,64} In the present case, we can surely assume that the polymer shell layer is much thicker than that in any alkanethiol protected metal nanoparticles. However, this does not help to explain the blue shift in the SPR band during compression. Also, a blue shift of the SPR band promoted by light scattering, observed for the gold particles coated with a silica shell over 40 nm in thickness,\textsuperscript{62} should be negligible, due to the small size of the amphiphilic AuNP-3.

To rationalize the experimental observations, it is significant to think how the properties of the local environment around the gold cores vary with the conformational change of the PNIPAM chains at the onset of the region II (Scheme 10). At the initial compression of the monolayer (\( \pi \approx 0 \text{ mN/m} \)), most of the tethered PNIPAM chains adsorbed on the
water surface presumably adopted an extended and flattened conformation. Meanwhile, the downward part of the gold core with a relatively loose polymer coating may be greatly exposed to water molecules. With the monolayer compression into the onset of the region II, the PNIPAM chains are forced to submerge into the water phase and to form a brush-like structure around the gold core, in a way that a large portion of the water molecules previously surrounding the downward part of the gold core are squeezed further out from the metal surface. This may result in a decrease in the polarity of the local environment of the gold core, leading to a notable blue shift at the onset of the region II as presented in Figure 13. Subsequently, the peak position of the SPR band only slowly blue-shifts with further compression (increasing the surface pressure) due to the fact that a further packing of the PNIPAM chains in water only slightly decreases the dielectricity of the particle surroundings.

Scheme 10. Schematic representation of the conformational change of tethered PNIPAM chains (blue ones) from the flattened and stretched structure at the air-water surface to a brush-like structure in the water subphase upon the monolayer compression. The collapsed PS chains are red.

Up to now, the blue shift in the in situ SPR band has been rationalized. However, in Figure 13, a noticeable effect of temperature on the in situ SPR band during compression may be observed, as well. At a constant surface pressure, an increase in temperature leads to a red shift in the SPR. As has been mentioned in the introduction, the interparticle distance is a key parameter to affect optical properties due to the coupling effect between adjacent nanoparticles when they are sufficiently close to each other in the assemblies. In the present study, however, due to higher values of $d/2r$ shown in Table 10 and the slight difference between the values at 20 and 30°C, only a weak interparticle coupling may take place during the compression conducted at 30 °C. Considering the factors which cause a red shift, based on Mie-Drude theory, it seems reasonable to conclude that the red shift observed with increasing temperature is likely due to the shrinking of the tethered PNIPAM chains at a higher temperature and therefore forming a thicker shell, as well as to the weaker interparticle coupling.
The blue shift was not observed in the in situ SPR band of alkanethiol protected metal nanoparticles during the monolayer compression in a Langmuir trough.\textsuperscript{39} What is the difference between our amphiphilic AuNPs and alkanethiol-protected metal nanoparticles? Alkanethiol molecules as protective ligands with a short chain length (e.g., 1.5-2.0 nm for dodecanethiol C\textsubscript{12}SH) are usually considered as fully stretched rigid chains covalently bound to the surface of the gold cores via a sulfur atom. In marked contrast, each PNIPAM chain (molar mass 3000 g/mol) used in this study possesses about 24 repeating units, i.e., 48C a linear main chain, with a length of ca. 8 nm for a fully stretched chain. Hence, the long PNIPAM chains covalently bound to the gold cores are more flexible and are readily forced to rearrange their conformation during the monolayer compression. This most probably causes the blue shift in the in situ SPR band. The rigid alkanethiol chains do not change their conformation during compression and thus, the interparticle coupling becomes a dominant factor to influence the SPR and leads to a red shift as $d/2r$ approaches the critical value of 1.2 with the monolayer compression.

### 3.4.2 SPR bands of deposited films

The AuNP-3 monolayer and subsequent multilayer films were deposited onto quartz slides under the conditions of $\pi = 30$ mN/m and 20 °C and 30 °C, respectively. In the deposited films the particles are wrapped by the solid polymer shell, different from that discussed above. The UV-vis absorption spectra, measured at room temperature, of the deposited films as a function of deposition cycle are shown in Figure 14 (same as that in the Supporting Information of the article V). We first compare the absorption spectra of the deposited AuNP-3 monolayers. The SPR peak positions of the monolayers are at 529 nm (transferred at 20 °C) and 533 nm (transferred at 30 °C), respectively. It can be seen that the peak position of the SPR band slightly shifts to longer wavelengths, i.e., a slight red-shift, with a rise of the transfer temperature. This may be due to a weak interparticle coupling.

![Figure 14. UV-vis absorption spectra of the deposited AuNP-3 multilayers with the deposition cycle at 20 °C (a) and at 30 °C (b). Deposition at a constant surface pressure of 30 mN/m. The number above the spectrum is the deposition cycle.](image-url)
Interestingly, comparing to the peak positions of the in situ SPR bands discussed above (see Figure 13), we find that the peak positions of the in situ SPR bands are situated in the range of 550 – 560 nm, longer than those for the deposited monolayers. The difference may imply that the deposition process of the monolayer onto a slide may somehow distort the in situ monolayer structure.

For the AuNP-3 multilayers deposited at 20 and 30 °C, respectively, the maximum absorbance ($A_{\text{max}}$) against the deposition cycle is plotted in Figure 15. The linear increase in $A_{\text{max}}$ with increasing the deposition cycle clearly suggests that the film deposition is successful and the particle coverage is homogeneous.$^{44,108}$

Figure 16 shows the plot of the SPR peak position ($\lambda_{\text{max}}$) of the transferred AuNP multilayer as a function of the deposition cycle. A gradual red shift over 10 nm in the SPR after several deposition cycles is observed. Also, the $\lambda_{\text{max}}$ of the multilayer deposited at 30 °C red-shifts more with the deposition cycles than that deposited at 20 °C. As discussed above, the compressibility of polymer-coated AuNPs at a high temperature leads to a smaller interparticle distance, resulting in an increase of the interparticle coupling in the multilayer.
4. CONCLUSIONS

In this work two types of gold nanoparticles, those protected only with PNIPAM and amphiphilic AuNPs protected with both of PNIPAM and PS chains, were studied. The research proceeded from the synthesis of the polymer-protected gold nanoparticles to the investigation of thermal behavior of the PNIPAM shell, and further to the studies of the Langmuir monolayers and optical properties of amphiphilic gold nanoparticles.

Gold nanoparticles protected either with PNIPAM or with a mixture of PNIPAM and PS have been successfully prepared. The sizes of the gold cores varied between 1 nm and 5 nm. The synthesis methods were systematically studied. RAFT process offers an opportunity to prepare gold nanoparticles protected with a monodisperse polymer shell. PNIPAM-AuNPs prepared by the “grafting-to” technique showed a much higher polymer density surrounding the gold cores than those prepared by the “grafting-from” technique. The latter nanoparticles contain a lower amount of polymer chains due to the low number of RAFT agents bound to the precursor gold nanoparticles through esterification. However, this does not mean that the “grafting-from” technique is a less favorable way, since the number of the functionalities created on the particle surface can be increased.

Two separate endothermic peaks were observed in the microcalorimetric studies of thermally induced phase transitions of the PNIPAM-AuNPs in aqueous solutions/dispersions. A model was proposed to describe the structure of the tethered PNIPAM chains according to which the polymer layer is composed of an inner and an outer zone. The narrow endothermic peak occurring at a lower temperature originates from the inner zone of the PNIPAM chains with high cooperativity, while the broad one observed at a higher temperature is due to the outer zone with low cooperativity.

Langmuir monolayers of the amphiphilic gold nanoparticles have successfully been built up in a Langmuir trough. The amphiphilic particles show thermo-responsive properties at the air-water interface. The monolayers can be compressed to smaller nanoparticle areas at elevated temperatures close to the LCST of PNIPAM. This is possible because the shrinkage of the tethered PNIPAM causes a decrease in the dimension of the coated particles. The sessile drop contact angle measurements conducted on both sides of the monolayer deposited at room temperature reveal two slightly different contact angles. This may indicate that the phase separation of the tethered PNIPAM and PS chains has taken place on the gold core.

The optical properties of the amphiphilic gold particles have been studied both in situ at the air-water interface and on the deposited films, respectively. The in situ SPR band of the monolayer shows a blue shift with compression. The blue shift is related to a conformational change of the tethered PNIPAM chains, which may cause a decrease in the polarity of the local environment around the gold cores. A red shift in the SPR, getting more pronounced with repeating deposition cycles was observed on the deposited films. The red shift is due to a weak interparticle coupling between adjacent particles. Temperature effects on the SPR bands were also investigated both in situ and on the deposited films. In the in situ case, at a constant surface pressure, an increase in
temperature leads to a red shift in the SPR, due to the shrinking of the tethered PNIPAM chains at elevated temperatures and the weak interparticle coupling. However, in the case of the deposited films, the SPR band red-shifts more with the deposition cycles at a high temperature than at a low temperature. This is because the compressibility of polymer-coated nanoparticles at high temperatures leads to a smaller interparticle distance, resulting in an increase of the interparticle coupling in the deposited multilayers.
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