The effect of added gold nanoparticles on the volume phase transition behavior for PVCL-based microgels†

Lei Hou and Peiyi Wu*

The volume phase transition (VPT) behavior of poly(N-vinylcaprolactam-co-4-vinylpyridine) (PVCL-co-4VP) microgels and their hybrid counterparts containing Au nanoparticles (AuNPs) is explored and compared by FTIR measurements in combination with the perturbation correlation moving window (PCMW) technique and two-dimensional correlation spectroscopy (2Dcos) analysis. It is demonstrated that the incorporation of AuNPs inside the microgels cause a slight dehydration effect on the microgels in the swollen state and an increase of the VPT temperature. Moreover, the interaction between C=O and AuNPs can be confirmed by combining the conventional IR and 2DCos analysis. It is with the help of the additional cross-links between AuNPs and the polymer chains (C=O and pyridine groups) in the microgel network and the tendency of AuNPs to aggregate that the hybrid microgels exhibit a much greater dehydration during the VPT, as compared with the nonhybrid ones.

Introduction

Microgels are spherical comprising cross-linked polymeric networks in the colloidal range.1–3 Smart colloidal microgels, being able to respond to external stimuli such as temperature, pH, light, ionic strength, have been intensively explored in the past decades. Their high porosity, dehydrations in microreactors, sensors, drug/gene delivery, catalysts and chemical/biological separations. Of the two most extensively used building blocks for thermally responsive microgels, poly(N-isopropylacrylamide) (PNIPAM)4,5 and poly(N-vinylcaprolactam) (PVCL)6–8 exhibit lower critical temperatures (LCSTs) in aqueous media and go through coil-to-globule transitions upon heating. While, PVCL is regarded to have better biocompatibility than PNIPAM, since the hydrolysis of PVCL will not produce toxic low molecular weight amines. Moreover, it has been reported that the monomer N-vinylcaprolactam (VCL) is less cytotoxic than N-isopropylacrylamide (NIPAM), thus making PVCL more suitable for biomedical applications.9,10,11

In recent years, hybrid microgels, composed of polymer microgels and inorganic components like Au,12–14 Ag15,16 or magnetic nanoparticles17,18 and quantum dots,19–21 have witnessed increasing investigations.22,23 By combining the colloidal stability and responsiveness of microgels with optical, electronic, and magnetic features of the embedded inorganic nanoparticles, hybrid microgels have found their widespread applications in the functional materials arenas. To prepare hybrid microgels, in situ synthesis of inorganic nanoparticles within the microgel matrix is one of the most frequently used methods, which offers a two-fold advantage: the utilization of polymer microgels as microreactors and the fabrication of a material with structural hierarchy.24 Kumacheva et al.24 have presented comprehensively the successful synthesis of inorganic nanoparticles (CdS, Ag, and Fe3O4) with predetermined size, polydispersity, and optical and magnetic properties using P(N-isopropylacrylamide-co-acrylic acid-co-2-hydroxyethyl acrylate) (P[NIPAM-co-AA-co-HEA]) microgels as a template, providing a guidance for the in situ synthesis of functional components in the polymer microgel matrix. As for PVCL-based microgels, Pich et al.25 have described a method for targeted deposition of poly(3,4-ethylenedioxythiophene) nanorods and gold nanoparticles (AuNPs) on the P(N-vinylcaprolactam-co-acetoacetoxethyl methacrylate) (P[VCL-co-AAEM]) microgels via a two-step in situ reaction and demonstrated that the hybrid microgels exhibited extraordinary catalytic activity in aqueous media. More recently, they showed that a newly developed P[VCL-co-AAEM-co-AA] microgels could be used as a “catalyzing”-soft polymeric template for the forming of hybrid polymer–gold nanostructures at ambient temperature without additional reducing agents. In addition, the composite...
nanostructures could function as a tunable noble metal catalyst and be re-used with retention of the catalytic activity.26

The most frequently used method to characterize the structure of hybrid microgels is transmission electron microscopy (TEM). Due to the large difference in electron density of inorganic nanoparticles and polymer microgels, details on the shape, size and polydispersity of inorganic nanoparticles embedded in the microgel matrix can be directly observed. Dynamic light scattering (DLS) is another common technique to measure the size of hybrid microgels in solutions. It is especially helpful to monitor their size variations under external stimuli. Meanwhile, other methods have also been applied to study the internal structures or interactions in the hybrid microgels. Yao et al.27 have investigated the interactions between P(NIPAM-co-AA) microgels and incorporated silver nanoparticles (AgNPs) utilizing X-ray photoelectron spectroscopy (XPS) and showed that there was charge transfer from the carbonyl groups of the microgel particles to the AgNPs. Furthermore, they found that the charge-transfer efficiency increased as the diameter of the AgNPs was decreased. In Klitzing’s report,28 UV-Vis absorption spectroscopy was used to monitor the changes in the surface plasmon resonance of the AuNPs loaded within PNIPAM microgels. It was discovered that the optical properties of the colloids could be easily modulated by varying cross-linker density and temperature, which was directly related to the formation of dimers of AuNPs. Additionally, Suzuki et al.29 have studied the internal structures of P(NIPAM-co-3-(methacrylamino)propyltrimethylammonium chloride) microgels embedded with AuNPs by means of Small-Angle X-ray Scattering (SAXS). It was demonstrated that the hybrid microgels exhibited pronounced excess scattering caused by the AuNPs as compared with the nonhybrid microgels and the radii of the AuNPs evaluated from the SAXS data was broadly in accordance with those observed in TEM. However, to the best of our knowledge, neither the volume phase transition (VPT) of AuNPs–polymer hybrid microgels nor the influence of AuNPs on the VPT of microgels in aqueous media has ever been explored at the molecular level.

As is known, Fourier transform infrared spectroscopy (FTIR) is rather sensitive to morphological or conformational variations since it reflects subtle information from the view of molecular level. It is a powerful method in recording the molecular motion and interactions between chemical groups and has been widely used in studying the VPTs30–32 or LCST transitions33–35 of thermally responsive systems. Nevertheless, none of those previous works have dealt with hybrid microgels. Herein, the VPTs of PVCL-based nonhybrid and hybrid microgels in water were investigated and compared by applying FTIR. In this way, details about the effect of AuNPs on the VPT of hybrid microgels corresponding to specific chemical groups were provided. Furthermore, with the aid of two spectral analysis techniques—perturbation correlation moving window (PCMWM) and 2D correlation spectroscopy (2Dcos), which have already been proved to be effective techniques in illustrating the phase transition behavior of thermally responsive polymers such as PNIPAM,36 PVCL,37 poly(vinyl methyl ether) (PVME),38 and copolymers based on oligo-(ethylene glycol) methacrylate (POEGMA),39 additional information on the specific order with regard to group motions during the VPT process of the hybrid microgels could be extracted, and thus, a more comprehensive understanding of the thermal-induced phase transition behavior of the organic–inorganic hybrid colloid would be obtained.

Experimental
Materials
4-Vinylpyridine (4VP), N,N'-methylenebisacrylamide (BIS), sodium dodecyl sulfate (SDS) and potassium peroxodisulfate (KPS) were purchased from Aladdin Reagent Co. N-Vinylcaprolactam (VCL) was obtained from Alfa Aesar Co. Chloroauric acid tetrahydrate (HAuCl4·4H2O) was supplied by Sinopharm Chemical Reagent Co. Ltd. Sodium borohydride (NaBH4) was purchased from Sigma-Aldrich. D2O was purchased from Cambridge Isotope Laboratories Inc. (D-99.9%). VCL and 4-VP were purified by passing through a short alumina column. KPS was recrystallized from deionized water and dried in vacuum. Other chemicals were used without further purification.

Preparation of P(VCL-co-4VP) microgels
The microgel particles were synthesized by aqueous free radical emulsion polymerization of VCL, 4VP and BIS (cross-linker), using SDS as stabilizer. KPS was used as the initiator.40 The chemical structures are presented in Fig. 1. Typically, 0.5 g VCL, 0.015 g 4VP, 0.01 g BIS and 0.02 g SDS were dissolved in 50 mL deionized water. The solution was placed into the reactor, equipped with a mechanical stirrer and reflux condenser, and stirred for 1 h at 70 °C under continuous purging with nitrogen. Then, 2 mL aqueous solution containing 0.01 g KPS was added to the reaction mixture. The polymerization reaction was allowed to continue under nitrogen atmosphere, with stirring at 400 rpm agitation rate for 4 h. After that, the reaction mixture was naturally cooling to room temperature, while maintaining...
the stirring. The obtained light-blue microgel dispersion was purified by dialysis with Millipore Dialysis System (cellulose membrane, MWCO 14 000) for a week. The determined solid content of the dispersion was 1 wt%.

Incorporation of AuNPs in P(VCL-co-4VP) microgels

The in situ synthesis of AuNPs inside P(VCL-co-4VP) microgels was performed according to some previous literatures. Densely, 20 mL of as-prepared microgel dispersion was placed into a glass flask and mixed with 4 mL 10 mM HAuCl₄ aqueous solution under continuous stirring at room temperature for 1 h. To initiate the aurate reduction process, 96 mL cold NaBH₄ aqueous solution (10 mM) was quickly added. Then the reaction was last for 30 min with vigorous stirring. The obtained hybrid colloidal was purified by dialysis with Millipore Dialysis System (cellulose membrane, MWCO 14 000) for a week.

**Instruments and measurements**

The volume phase transitions of the microgels (~0.02 wt% in H₂O) were monitored on a dynamic light scattering (DLS)-zetasizer nanosystem (Malvern) with the scattering angle of 173°. It was performed by following the reduction of the hydrodynamic diameter (Dh) of microgels upon heating with a heating rate of ca. 0.4 °C min and an interval of 2 °C. Each temperature point was held for 2 min before measurements for equilibration of the microgels. The transmission electron microscope (TEM) images of nonhybrid and hybrid microgels were taken on a carbon-coated copper grid (T11032) with a JEOL JEM2100F microscope (Japan) operated at 200 kV. Thermal gravimetric analysis (TGA) of the freeze-dried nonhybrid and hybrid microgels was performed on a Perkin-Elmer Pyris-1 at a heating rate of 10 °C min under nitrogen atmosphere with a flow rate of 40 cm³ min⁻¹. The temperature range was from 100 °C to 800 °C.

For FTIR measurements, the freeze-dried microgels were dispersed in D₂O with concentration fixed to 10 wt% and placed at 4 °C for a week to ensure sufficient swelling. Herein, samples were prepared by sealing microgel D₂O solutions between two GaF₂ tablets, which had no absorption bands in the MIR region. All the temperature-dependent FTIR spectra were obtained on a Nicolet 6700 spectrometer equipped with a DTGS detector at a resolution of 4 cm⁻¹, and 32 scans were accumulated for an acceptable signal-to-noise ratio. With an electronic cell holder, a programmed method was used to control the temperature at a rate of ca. 1 °C/min with an interval of 0.5 °C (accuracy: 0.1 °C). The neat nonhybrid and hybrid microgel films for FTIR measurements were prepared by drop casting on the CaF₂ tablets from their aqueous dispersions. The baseline-correction process was carried out by the software of OMINIC 6.1a.

**Investigation method**

**Perturbation Correlation Moving Window (PCMW).** All the FTIR spectra recorded with an interval of 0.5 °C during the heating and cooling process were selected to perform PCMW analysis. Primary data processing was conducted with the method provided by Morita, and further correlation calculation was carried out by applying the software 2D Shige, ver. 1.3 (Shigeaki Morita, Kwansei-Gakuin University, Japan, 2004-2005) with an appropriate window size (2m + 1 = 11). The final contour maps were drawn in Origin Program ver. 8.0, with the red colors representing positive intensities and the green colors representing negative ones.

**Two-dimensional correlation spectroscopy (2Dcos).** 2Dcos analysis was conducted with all the temperature-resolved FTIR spectra collected during the heating process at an internal of 0.5 °C, using the same 2D Shige software, ver. 1.3 (Shigeaki Morita, Kwansei-Gakuin University, Japan, 2004-2005). Then, it was plotted into the contour maps with Origin Program ver. 8.0. In the contour maps, red colors represent positive correlation intensities, while the green colors represent negative correlation ones.

**Results and discussion**

**Preparation and characterization of hybrid microgels**

The P(VCL-co-4VP) microgels were successfully prepared via an emulsion polymerization procedure, using SDS as the stabilizer and BIS as the cross-linker. It is demonstrated in the TEM images that the synthesized microgel particles exhibit a spherical shape with high monodispersity (Fig. 1(a) and S1) (ESI†). Additionally, the incorporation of 4VP inside the microgels is confirmed in ¹H NMR spectra (Fig. S2) (ESI†) by the appearance of the peak at 8.5 ppm, which is related to the protons on the pyridine rings. The as-prepared microgels were then utilized as microreactors for the in situ synthesis of highly-dispersed AuNPs, using HAuCl₄ as a precursor and NaBH₄ as a reducing agent. It is believed that the addition of charged functional groups into the microgels can equip it with responsiveness towards pH or ionic strength, specific coordination sites and certain reactivity. Herein, 4VP is copolymerized with VCL in order to absorb the precursor ions of AuNPs. To be more specific, in the preparation of the hybrid microgels, the addition of HauCl₄ results in a decrease in pH and protonation of the pyridine rings. Consequently, the AuCl₄⁻ anions are drawn into the microgel network and localize themselves close to ionized pyridine rings by the electrostatic attraction. Once the reducing agent, NaBH₄, was added, AuNPs could be formed, using positive charged pyridine rings in the microgels as nucleation and growth sites. It is with the help of the network architecture of the microgels that aggregation of AuNPs is suppressed. The morphology of the P(VCL-co-4VP)/AuNPs hybrid microgels is investigated by TEM, as presented in Fig. 1(b) and S3(a) (ESI†). It is displayed that the synthesized AuNPs are spherical in structure and 2–5 nm in diameter. Moreover, almost all the AuNPs are well-distributed inside the microgels. To further confirm the importance of 4VP in the AuNPs deposition process, PVC microgels without 4VP as the comonomer were used as a control sample. In Fig. S3(b) (ESI†), the TEM image indicates that in the case of PVC microgels the AuNPs are partially localized around the microgel spheres but not concentrated inside them, and this might be just as a result of some physical adsorption.

Then, the nonhybrid and hybrid microgels were frozen-dried and investigated by TGA, as demonstrated in Fig. 2(c). It is
shown in the TGA curves that both microgel systems exhibit a good thermal property and keep stable under 400 °C. The weight loss from 400 to 500 °C is due to the thermal decomposition of the C–C backbone and the complete polymer degradation takes place in this temperature range, which can be derived from the TGA curve of the nonhybrid microgels. The residual mass remaining after heating of the samples above 500 °C can correspond to the AuNPs, since they do not degrade under such conditions. In this way, TGA measurements can clearly display the amount of AuNPs loaded inside the hybrid microgels, which is calculated to be around 30 wt%.

The temperature dependence of the hydrodynamic diameter ($D_h$) of the P(VCL-co-4VP) microgels and P(VCL-co-4VP)/AuNPs hybrid microgels in aqueous solutions were explored by DLS. As shown in Fig. 2(d), the nonhybrid microgels exhibit a typical thermal-responsive behavior, that is swelling at temperature below volume phase transition temperature (VPTT) and shrinking at above VPTT. It can be also observed that after the incorporation of AuNPs, the microgels retain their temperature-sensitive property. However, the presence of AuNPs could, in some extent, affect the swelling behavior of P(VCL-co-4VP) microgels. Firstly, for microgels in swollen status, a decrease in the $D_h$ of the microgel particles embedded with AuNPs is observed. It should be a normal phenomenon in hybrid microgel systems and could be due to the restricted polymer chain dynamics caused by strong interactions of both the pyridine and carbonyl groups along the polymer chains with AuNPs. In other words, additional physical cross-links are formed between pyridine/carbonyl groups and AuNPs within the hybrid microgels. These additional physical cross-links will play an important role in the VPT of hybrid microgels, which will be further discussed later. Secondly, the VPTT of microgels embedded with AuNPs seems to shift slightly to higher temperature as compared to pure P(VCL-co-4VP) microgels. This could be explained by the fact that the existence of AuNPs inside the thermosensitive microgels will partly restrict the mobility of the polymer network and increase its rigidity and hence higher temperature is necessary to achieve the microgels collapse.

Though DLS profiles describe the difference between the VPT behavior of the nonhybrid and hybrid microgels, they only provide general information and details with regard to chemical group motions within the microgels during the VPTs need farther investigations.

**Conventional FTIR analysis**

To understand the VPTs of nonhybrid and hybrid microgels in aqueous solutions and figure out the influence of AuNPs on the VPT from the molecular level, temperature-dependent FTIR spectra of P(VCL-co-4VP) microgels and P(VCL-co-4VP)/AuNPs hybrid microgels were collected and compared, as presented in Fig. 3. Herein, we selected D$_2$O, rather than H$_2$O, as the solvent in order to eliminate the overlap of the δ(OH) relating to H$_2$O at around 1640 cm$^{-1}$ with the amide I region of PVCL-based microgels, as well as reduce the interference of broad ν(OH) of H$_2$O in the C–H stretching region. It should be noticed that the amount of comonomer, 4VP, in the microgel network is so little that IR absorption relating to pyridine rings can't be observed. Though the interaction between AuNPs and pyridine groups can't be neglected, the analysis of the pyridine groups herein will not be discussed in detail. Then, we will mainly focus on two spectral regions: the C–H stretching vibration region (3010–2835 cm$^{-1}$) and C=O stretching vibration region (1650–1560 cm$^{-1}$), which reflect nearly all the group motions within the thermal-responsive part in the microgels. Through carefully examining the temperature-resolved FTIR spectra, we can find that, with temperature rising, all the C–H stretching bands exhibit red shift, while C=O presents a binary spectral intensity change in both nonhybrid and hybrid microgel systems. And the situation in the cooling process is just opposite to that in heating. The variations in the C–H stretching bands can be explained by the hydrophobic interaction of C–H groups with neighboring water molecules in the solution. It has been reported that higher number of water molecules surrounding C–H groups would result in higher vibrational frequency. On the basis of this point, it can be deduced that all the C–H groups in both microgel systems experience dehydration upon heating and hydration in cooling. As for Amide I region, it can be roughly divided into two parts: the lower wavenumber moiety and the higher one, corresponding to hydrogen bonded C=O with water and free C=O, respectively. Therefore, the heating-induced intensity changes in Amide I bands, namely, intensity of the lower wavenumber moiety decreasing and that of the higher one increasing, can be related to the transformation of hydrogen bonded C=O with water to free ones. From this point of view, the nonhybrid and hybrid microgel systems share the same variation tendency in the VPTs during the heating and cooling cycle, that is, chemical groups in the microgels undergoing dehydration process and the microgel network shrinking in heating and the case being inverse with cooling.

To further elucidate the difference related to chemical groups in the VPTs between nonhybrid and hybrid microgels, some contrast FTIR curves of P(VCL-co-4VP) microgels and P(VCL-co-4VP)/AuNPs hybrid microgels in D$_2$O at 25 °C and 48 °C during heating and the neat films of the microgels are
plotted in Fig. 4. For nonhybrid microgels, both C–H and C=O groups are free of hydrophobic hydration or hydrogen bonds with water in the neat film. Once the microgels are swollen with water, those groups will interact with water molecules, thus leading to the frequency shifts in IR absorption. Compared with the neat film, all the groups along the polymer chains in the microgels are in partial hydration at both 25 and 48 °C, which suggests that those groups only go through limited dehydration upon heating. While, a situation does not change too much except for that the IR intensity of C=O exhibits a slight decrease with the incorporation of AuNPs, the existence of AuNPs in the microgels might be originated from the changes found in the LCST-transition of linear PVCL aqueous solution. However, the changes here in the microgel systems are far more continuous than that in the linear chains. Taking the difference in the architecture into consideration, we suppose that the continuous transition in the microgel systems can be attributed to the confined network in which abrupt polymer chain collapse is inhibited and water molecules could be only gradually squeezed out. By looking into the curves more carefully, we can find that both the initial wavenumbers of νas(CH2) and νs(CH2) shift to lower positions after the deposition of AuNPs, indicating that the existence of AuNPs in the microgels could weaken the hydration of the microgels, which somehow is in accordance with the DLS curves that the beginning Dh of hybrid microgels is smaller than that of nonhybrid microgels and could be caused by those additional physical cross-links. Moreover, it can be intuitively discovered that C–H groups undergo much greater variation in the hybrid microgel systems compared with nonhybrid microgels and could be caused by those additional physical cross-links. Additionally, it is noticed for the neat films in the amide I region that the hybrid microgels exhibit a slight red shift as compared with the nonhybrid ones, which, from another perspective, indicates the interaction of C=O groups with AuNPs and will be further discussed in the 2Dcos analysis.

For more detailed comparison of group variations in the two microgel systems during the VPTs, temperature-dependent wavenumber shifts of ν(C–H) and quantitative analysis of the integral area changes relating to the C=O band vs. temperature have been performed, as presented in Fig. 5. For clarity, Boltzmann curve fitting is performed. In both microgel systems, the wavenumber shifts of νas(CH2) and νs(CH2) exhibit a relatively asymmetric sigmoid curve, namely a sharp change around VPT and a gradual change after VPT, which is consistent with the changes found in the LCST-transition of linear PVCL aqueous solution. Additionally, it is noticed for the neat films in the amide I region that the hybrid microgels exhibit a slight red shift as compared with the nonhybrid ones, which, from another perspective, indicates the interaction of C=O groups with AuNPs and will be further discussed in the 2Dcos analysis. For more detailed comparison of group variations in the two microgel systems during the VPTs, temperature-dependent wavenumber shifts of ν(C–H) and quantitative analysis of the integral area changes relating to the C=O band vs. temperature have been performed, as presented in Fig. 5. For clarity, Boltzmann curve fitting is performed. In both microgel systems, the wavenumber shifts of νas(CH2) and νs(CH2) exhibit a relatively asymmetric sigmoid curve, namely a sharp change around VPT and a gradual change after VPT, which is consistent with the changes found in the LCST-transition of linear PVCL aqueous solution. However, the changes here in the microgel systems are far more continuous than that in the linear chains. Taking the difference in the architecture into consideration, we suppose that the continuous transition in the microgel systems can be attributed to the confined network in which abrupt polymer chain collapse is inhibited and water molecules could be only gradually squeezed out. By looking into the curves more carefully, we can find that both the initial wavenumbers of νas(CH2) and νs(CH2) shift to lower positions after the deposition of AuNPs, indicating that the existence of AuNPs in the microgels could weaken the hydration of the microgels, which somehow is in accordance with the DLS curves that the beginning Dh of hybrid microgels is smaller than that of nonhybrid microgels and could be caused by those additional physical cross-links. Moreover, it can be intuitively discovered that C–H groups undergo much greater variation in the hybrid microgel system, which is almost twice as great as that in the nonhybrid one. In addition, as shown in the temperature dependence of the frequency shifts corresponding to C–H groups, it is obvious that after a heating-and-cooling cycle, those C–H groups in the nonhybrid microgels can exactly return back to the initial status and exhibit excellent reversibility. While for C–H groups in the
hybrid microgels, the situation is quite different. They present not only worse retrieval degree after cooling back to 25 °C but also a hysteresis during the cooling process. As is well-known that there also exists a hysteresis upon cooling in the LCST-transition of PNIPAM aqueous solution,\textsuperscript{35} and it is believed to arise from the inter-/intra-molecular hydrogen bonding C=O⋯H-N, acting as additional physical cross-links among the polymer chains and resulting in more entangled chains in the collapsed states, and such hydrogen bonds can't be easily removed in the cooling process. Deducing from this point, the appearance of hysteresis and insufficient reversibility in the cooling process of hybrid microgels could somehow confirm the formation of additional cross-links between AuNPs and the polymer chains. Furthermore, to study and compare the variations of the C=O groups, the initial integral areas in the amide I region of those two microgel systems are normalized and the shifts are drawn in Fig. 5(c), which shows the same trend as that observed in the C–H groups. What's more, we should note that an excess recovery phenomenon after a heating and cooling cycle occurs in the nonhybrid microgel system. It is presumed that such slight recovery difference between C=O and C–H groups could be derived from their distinct nature where C=O is hydrophilic while C–H is hydrophobic.

Perturbation correlation moving window (PCMW)

As for the asymmetric sigmoid curves in Fig. 5, it is difficult for us to accurately determine the transition points in the heating and cooling process. Therefore, a PCMW technique is employed here to ascertain the VPTTs regarding to various chemical groups in those two microgel systems. PCMW is a newly developed method, whose basic principles were firstly proposed by Thomas and Richardson.\textsuperscript{47} Through introducing a perturbation variable into the correlation equation, it was then improved for much wider applicability by Morita \textit{et al.} in 2006.\textsuperscript{48} Together with its ability to monitor the complicated spectral changes along the perturbation direction, PCMW can as well find the critical transition points with regard to specific chemical groups. Herein, only the synchronous maps of the PCMW are drawn for deeper investigation on the VPTTs of P(VCL-co-4VP) microgels and P(VCL-co-4VP)/AuNPs hybrid microgels during heating between 25 and 48 °C, as shown in Fig. 6.

It has been reported that in the PCMW contour maps, the positive synchronous correlation can indicate an increase in the spectral intensity, while a negative one can indicate a decrease in the spectral intensity.\textsuperscript{46} Thus it can be directly inferred from the synchronous maps that intensity related to hydrated groups decreases while intensity related to dehydrated ones increases with temperature rising in the FTIR spectra. At the same time, we can also find that almost all the intensity changes spread over the whole temperature change, demonstrating relatively continuous VPTTs of both microgel systems, which parallels with the observation in the conventional IR analysis.

In addition to monitor the spectral variations along temperature perturbation, PCMW synchronous maps are also helpful to determine the transition points. Thus the VPTTs with regard to C–H and C=O groups located around 34 °C in the nonhybrid microgels and 36.5 °C in the hybrid ones, respectively, which accords well with DLS measurements that the incorporation of AuNPs in the microgels will bring about higher VPTT. Meanwhile, it is noticed that the transition temperatures determined in the PCMW may differ from those observed in the DLS curves. This dissimilar should be understandable that the chemical group variations might be not exactly the same with $D_h$ changes. Besides, the temperature measured in the IR and DLS instruments might also have some difference. In spite of this, the variation tendency of the VPTs in the two microgel systems maintains. With more careful inspection on the PCMW maps, we are able to uncover another fine distinction between those two microgel systems in which different groups in the nonhybrid microgels display different sensitivity to temperature variation while groups in the hybrid microgels respond to temperature change simultaneously. In particular, the intensity changes in the C–H stretching region reveal that the dehydrated C–H groups have an obviously earlier response than the hydrated ones during the heating process of the nonhybrid microgel system. It is widely believed that PVCL microgels possess an inhomogeneous network structure\textsuperscript{49} with a densely cross-linked core and sparsely cross-linked shell due to the different reaction kinetics of the cross-linker BIS and the monomer VCL. Under such circumstance, it can be assumed that the PVCL-based microgels in this work also hold a relatively dehydrated core and a relatively hydrated shell. Thus the
observation of earlier response of the dehydrated C–H groups in nonhybrid microgels infers that C–H groups located in the relatively dehydrated core are somewhat more sensitive to temperature rising. As for the hybrid microgels, the inhomogeneous network structure is not that obvious with the emergence of additional physical cross-links between AuNPs and active groups on the polymer chains. Consequently, all the groups in the hybrid microgels do not exhibit obvious differences in responding temperatures upon heating.

Two-dimensional correlation spectroscopy (2Dcos)

2Dcos is a mathematical method, whose basic rules were originally proposed by Noda.49,50 Up to date, it has been widely utilized to explore the spectroscopic intensity fluctuations of different chemical species under various external perturbations, such as temperature, time, pH, pressure, concentration, etc.51 By spreading the original spectra along a second dimension, 2Dcos can sort out complex or overlapped features, which are not visible in the conventional analysis, and accordingly achieve an enhanced spectral resolution. Additionally, the specific event sequence in the system during the perturbing process can be extracted in 2Dcos analysis too, thus a more comprehensive understanding of the changes in different chemical species under a certain external variable is available. To obtain the 2Dcos spectra of nonhybrid microgel system, all the FTIR spectra between 25 and 48 °C are employed and the generated synchronous and asynchronous spectra are displayed in Fig. 7. The 2D synchronous spectra can reflect simultaneous variations between two given wavenumbers. The bands at 2951, 2862 and 1608 cm⁻¹ all hold positive cross-peaks in the synchronous maps, revealing their similar response of spectral intensities to the temperature perturbation. Referring to the original spectra, we are able to find that all the intensities of these bands decrease with heating. Similarly, it can be deduced that the changes at bands 2918, 2852 and 1628 cm⁻¹ occur in the other direction—that is, increase upon heating. With the help of 2D asynchronous
Table 1  Tentative band assignments of P(VCL-co-4VP) microgels in D2O according to 2Dcos results5,30,37

<table>
<thead>
<tr>
<th>Wavenumber (cm⁻¹)</th>
<th>Tentative assignments</th>
</tr>
</thead>
<tbody>
<tr>
<td>2951</td>
<td>νas(CH₂) (hydrated)</td>
</tr>
<tr>
<td>2918</td>
<td>νas(CH₂) (dehydrated)</td>
</tr>
<tr>
<td>2862</td>
<td>νs(CH₂) (hydrated)</td>
</tr>
<tr>
<td>2852</td>
<td>νs(CH₂) (dehydrated)</td>
</tr>
<tr>
<td>1628</td>
<td>ν(C=O)</td>
</tr>
<tr>
<td>1608</td>
<td>ν(C=O···D-O-D)</td>
</tr>
<tr>
<td>1587</td>
<td>ν(C=O···2D-O-D)</td>
</tr>
</tbody>
</table>

spectra, significantly enhanced spectral resolution is obtainable. For example, splitting bands located at 1628, 1608 and 1587 cm⁻¹, corresponding to C=O stretching vibration in various hydrated states can be recognized. For the convenience of discussion, all the bands identified in the 2D asynchronous spectra and related assignments are listed in Table 1.

Together with enhancing spectral resolution, 2Dcos can as well detect the specific order occurring along external perturbations. The judging rule can be outlined as Noda’s rule53—namely, when the cross-peaks (ν₁, ν₂, assuming ν₁ > ν₂) in synchronous and asynchronous spectra hold the same sigh, then the change at band ν₁ may take place earlier than or prior to that at band ν₂, and vice versa. We have described a simplified method for the determination of the sequence order in our previous work,53 and herein we only present the final sequence order in the heating-induced VPT of the nonhybrid microgels, that is [→ means prior to or earlier than] 1608 → 2918 → 1587 → 2852 → 2951 → 2862 → 1628 cm⁻¹ or ν(C=O···D-O-D) → νas(CH₂) (dehydrated) → ν(C=O···2D-O-D) → νas(CH₂) (dehydrated) → νs(CH₂) (hydrated) → νs(CH₂) (hydrated) → νs(CH₂) (hydrated) → νas(CH₂) (hydrated) → νas(CH₂) (hydrated) → νas(CH₂) (hydrated) → νas(CH₂) (hydrated) → νas(CH₂) (hydrated). Without considering the differences in stretching modes, we have C=O → C–H, revealing that hydrogen bonding transformation of amide groups is the driving force of the heating-induced VPT. Moreover, it is also observed that the dehydrated C–H groups have an earlier response than the hydrated ones upon heating, which is in consistency with the above PCMW analysis. From the overall sequence order, we can additionally find that the change of C=O and C–H groups take place alternately, indicating that in the VPT of nonhybrid microgels C=O and C–H groups work cooperatively to make the polymer chains collapse and gradually expel water molecules out of the microgel network. On the basis of the above analysis upon heating, we are able to propose a chain collapse mechanism of the nonhybrid PVCL-based microgels. At temperatures below the VPTT, the microgels are well swollen in the aqueous solution with both C–H and C=O groups associated with water molecules. When the temperature increases above the VPTT, the hydrogen bonds between C=O groups and water molecules start to break, which begins from the relatively less hydrated inner core. With the help of cooperative dehydration among the C=O and C–H groups and the resulting adjustment of the polymer chains in the microgels, water molecules are finally squeezed out from inside the microgels step by step.

As for the P(VCL-co-4VP)/AuNPs hybrid microgels, 2Dcos analysis is also applied using all the FTIR spectra in heating from 25 to 48 °C with an increment of 0.5 °C and the related synchronous and asynchronous spectra are shown in Fig. 8. Thanks to the enhanced spectral resolution of the 2Dcos, hydrated and dehydrated states corresponding to the C–H groups which are overlapped in the conventional IR spectra can be easily identified. In the amide I region, three different kinds of C=O groups with bands at 1632, 1608 and 1587 cm⁻¹, respectively, can also be differentiated, which is in accordance with the bands in the nonhybrid microgel system. In addition to that, a new band, located at 1616 cm⁻¹ comes into being in the

Fig. 8  2D synchronous and asynchronous spectra of 10 wt% P(VCL-co-4VP)/AuNPs hybrid microgels in D2O obtained from all spectra between 25 and 48 °C during heating. Herein, warm colors (red) indicate positive intensities, while cool colors (green) indicate negative ones.
2D asynchronous map. It is widely known that lower frequency implies stronger interaction in the Amide I region of the IR spectra. Then, we can conclude that the appearance of band at 1616 cm\(^{-1}\) means that there exists a kind of interaction, which is weaker than hydrogen bonding between C=O and one water molecule, within the C=O groups. By considering the presence of AuNPs embedded in the hybrid microgels and other literatures\(^6\),\(^{12,14}\) on the interaction between AuNPs and C=O groups, we attribute the new band at 1616 cm\(^{-1}\) to the C=O groups absorbed on the AuNPs, which hence, provides a direct evidence of the formation of additional physical cross-links in the hybrid microgel networks. Now, when we look back at Fig. 4, we can confirm that the red shift of amide I region in the neat film of hybrid microgels is caused by the interaction of C=O groups with AuNPs. For clarity, Table 2 lists all the bands detected from 2Dcos asynchronous spectra in the heating process and their tentative assignments.

Similarly, the sequence order of different chemical groups occurring during heating of the hybrid microgels in D\(_2\)O can be also deduced based on the Noda’s rule (\(\rightarrow\) means prior to or earlier than): 1608 \(\rightarrow\) 1587 \(\rightarrow\) 1616 \(\rightarrow\) 2951 \(\rightarrow\) 2867 \(\rightarrow\) 1632 \(\rightarrow\) 2858 \(\rightarrow\) \(\nu(C=O\cdots D-O-D)\) \(\rightarrow\) \(\nu(C=O\cdots 2D-O-D)\) \(\rightarrow\) \(\nu(C=O\text{ absorbed on AuNPs})\) \(\rightarrow\) \(\nu_{as}(CH_2)\) (hydrated) \(\rightarrow\) \(\nu_{as}(CH_2)\) (hydrated) \(\rightarrow\) \(\nu_{s}(free\ C=O)\) \(\rightarrow\) \(\nu_{as}(CH_2)\) (dehydrated) \(\rightarrow\) \(\nu_{s}(CH_2)\) (dehydrated). Obviously, the C=O groups exhibit an earlier change than the C-H groups, behaving in the same way as the nonhybrid microgels demonstrate. Thus, the VPT of P(VCL-co-4VP)/AuNPs hybrid microgels upon heating is also driven by the hydrogen bonding transformation of C=O groups. Nevertheless, unlike the cooperative dehydration between C=O and C-H groups in the nonhybrid microgel system, the change of C=O groups in the hybrid microgels seems to be in an absolute predominance during the VPT since almost all kinds of C=O groups change prior to the C-H groups, as observed in the overall sequence order. And such phenomenon might be ascribed to that due to the attractive interactions between the AuNPs and C=O groups, C=O groups prefer to be absorbed on the AuNPs before the whole polymer chains’ conformational adjustment when the hydrogen bonds between C=O and water break. According to the previous discuss, the VPT process of the hybrid microgels can be summarized as follows: the hybrid microgels are in swollen state through the hydrogen bonding between hydrophilic C=O groups with water molecules and hydrophobic hydration of C-H groups at temperatures below the VPTT. While, due to the presence of AuNPs, additional physical cross-links between AuNPs and C=O/pyridine groups exist in the hybrid microgels, thus resulting in a slight contraction of the network and relatively dehydrated microgels as compared with the nonhybrid ones. As heating the solution above the VPTT, C=O groups gradually break the association with water molecules and adsorb onto the AuNPs, which produces more cross-links within the microgels. Under such circumstance, it will be much easier for C-H groups to dehydrate and water molecules to be squeezed out. Together with the tendency of AuNPs incorporated in the microgels to aggregate to reduce the total surface area, a much more dehydrated or collapsed microgel is obtained after the VPT, compared with the nonhybrid microgel system. For limpidity of demonstration, the VPTs of the nonhybrid and hybrid microgels in heating are illustrated in Fig. 9.

![Fig. 9 Schematic illustration and comparison of the VPTs of P(VCL-co-4VP) nonhybrid microgels and P(VCL-co-4VP)/AuNPs hybrid microgels in water. The light blue color represents the distribution density of water molecules.](image)

**Table 2** Tentative band assignments of P(VCL-co-4VP)/AuNPs hybrid microgels in D\(_2\)O according to 2Dcos results\(^6\),\(^{30,37}\)

<table>
<thead>
<tr>
<th>Wavenumber (cm(^{-1}))</th>
<th>Tentative assignments</th>
</tr>
</thead>
<tbody>
<tr>
<td>2951</td>
<td>(\nu_{as}(CH_2)) (hydrated)</td>
</tr>
<tr>
<td>2929</td>
<td>(\nu_{as}(CH_2)) (dehydrated)</td>
</tr>
<tr>
<td>2867</td>
<td>(\nu_s(CH_2)) (hydrated)</td>
</tr>
<tr>
<td>2858</td>
<td>(\nu_s(CH_2)) (dehydrated)</td>
</tr>
<tr>
<td>1632</td>
<td>(\nu(C=O))</td>
</tr>
<tr>
<td>1616</td>
<td>(\nu(C=O\text{ absorbed on AuNPs}))</td>
</tr>
<tr>
<td>1608</td>
<td>(\nu(C=O\cdots D-O-D))</td>
</tr>
<tr>
<td>1587</td>
<td>(\nu(C=O\cdots 2D-O-D))</td>
</tr>
</tbody>
</table>

**Conclusions**

In this work, we mainly employed DLS and FTIR measurements, as well as perturbation correlation moving window (PCMW) technique and 2D correlation spectroscopy (2Dcos) analysis, to explore and compare the thermally induced VPT behavior of the P(VCL-co-4VP) nonhybrid and P(VCL-co-4VP)/AuNPs hybrid microgels from the molecular level. Herein, the nonhybrid microgels are prepared via an emulsion polymerization procedure using VCL and 4VP as monomers and AuNPs are deposited inside the microgels through in situ reduction utilizing the nonhybrid microgels as templates. The hybrid microgel structure is revealed by TEM images, in which almost all the AuNPs are well-distributed inside the microgels. In addition,
calculated from the TGA curves, the amount of AuNPs loaded inside the microgels is around 30 wt%. Then, DLS measurements are performed to give a macroscopic view of the heating-induced VPTs of those two microgel systems, where we can find that the incorporation of AuNPs inside the microgels results in a decrease of \( D_h \) of the microgels in swollen status and an increase of the VPTT. Such phenomena can be also observed in the quantitative analysis of the temperature-dependent FTIR spectra and the PCMW analysis, respectively. It is believed that the former could be attributed to additional cross-links formed between AuNPs and pyridine/carbonyl groups and the latter is related to the increase in the rigidity of the polymer network with the presence of AuNPs. By combining the conventional IR and 2Dcos analysis, the interaction between the C═O and AuNPs can be further confirmed, which will consequently create more cross-links in the microgel network upon heating. Along with the tendency of AuNPs to aggregate, the hybrid microgels exhibit a much greater dehydration during the VPT as compared with the nonhybrid ones. We believe that the details about the effect of gold nanoparticles on the volume phase transition of the microgels revealed by FTIR will help to better understand the properties of the hybrid microgels in a wide range of applications.

Acknowledgements

We are very grateful for the financial support of the National Natural Science Foundation of China (NSFC) (no. 21274030).

Notes and references


