

pH-Responsive and Thermoreversible Hydrogels of *N*-(2-hydroxyalkyl)-L-valine Amphiphiles[†]

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Gelation behavior of a series of low-molecular-weight-hydrogelators, *N*-(2-hydroxydodecyl)-L-amino acid, was studied in aqueous phosphate buffer (pH 12). The effect of head-group structure and chirality and hydrocarbon chain length on the gelation efficiency was investigated. Only *N*-(2-hydroxyalkyl)-L-valine (L-C_{*n*}HVal, *n* = 10, 12, 14, and 16) derivatives were found to form gel in aqueous buffer at pH 12. The increase of the chain length of the hydrocarbon tail enhances the ability to gelate buffered water up to C14 chain length. The L-C₁₆HVal amphiphile was found to have gelation ability lower than L-C₁₄HVal. The gelation number, mechanical strength, thermal stability, and morphology of the supramolecular aggregates were studied. The effect of salt concentrations on the gelation was investigated. Addition of NaCl increased gelation number but decreased melting temperature of the hydrogels slightly. The morphology of the hydrogels was characterized by electron microscopy and small-angle X-ray diffraction techniques. Rheology measurements were performed to examine the mechanical strength of the hydrogels. Both hydrogen-bonding and hydrophobic interactions were shown to be the driving forces for supramolecular aggregate formation.

Introduction

Low-molecular-weight gelators have received considerable attention in recent literature for their potential applications in industrial fields, such as cosmetics, foods, and tissue engineering.^{1–6} The low-molecular-weight hydrogelators (LMWHs) are also of important in the field of drug and gene delivery with respect to the entrapment in gels and their subsequent controlled release.^{7,8} The hydrogelators reported so far in the literature are mostly based on polymeric materials. However, as far as drug delivery is concerned, the disadvantage of polymer-based hydrogelators is that the three-dimensional (3-D) network structures being chemically cross-linked are often not biodegradable and hardly respond to biological stimuli such as heat, pH, and salt concentration. On the other hand, the network structure of hydrogels of LMWHs is formed either by physical entanglement and/or branching of polymer-like supramolecular aggregates. The supramolecular structures, such as nanofibers, nanoribbons, and nanosheets, are formed by the self-assembly of monomer units through hydrogen-bonding (H-bonding), van der Waals (hydrophobic), and π -stacking interactions. The solvent molecules are immobilized by the surface tension effect. Gelation occurs

when a delicate balance between solubility and precipitation of the gelator is maintained.⁹ In other words, a critical balance between the hydrophobicity and hydrophilicity of the gelator molecule is important for hydrogelation to occur. As a result, most gelators reported so far are rather serendipitous than by design. Therefore, a huge academic interest has grown in the recent past to understand the relationship between gelation and chemical structure of the LMWHs. Despite many reports on organogelators,¹⁰ there are only a limited number of hydrogels formed by low-molecular-weight amphiphiles.¹¹ Nonetheless, three-dimensional network approaches have been used to prepare hydrogels from low-molecular-weight compounds, such as simple amphiphiles,¹² bolaamphiphiles,¹³ gemini surfactants,¹⁴ and other hydrogelators.¹⁵ Dipeptide-based low-molecular-weight amphiphilic hydrogelators which have antimicrobial activities and have potentials in drug delivery have been reported.¹⁶ One of us

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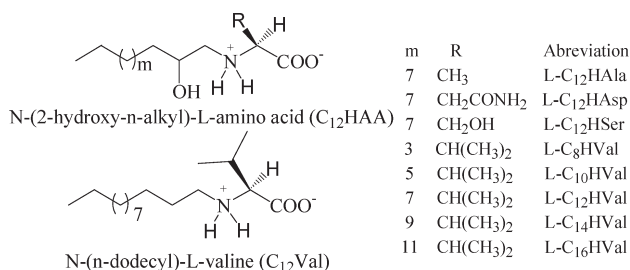
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Chart 1. Chemical Structures of *N*-(2-Hydroxy-*n*-alkyl)-L-amino Acids (L-C₁₂HAA) and *N*-(*n*-Dodecyl)-L-valine (L-C₁₂Val)

has also recently reported hydrogels of mixed surfactant systems.¹⁷

As part of our ongoing research on the self-assembly formation of amphiphilic molecules with amino acid as polar head group,^{18–22} we have designed and synthesized (see Supporting Information for details) a series of new amphiphiles, *N*-(2-hydroxyalkyl)-L-amino acid (see Chart 1 for structures), and have studied their aggregation behavior in water. The self-assembly and physicochemical properties of *N*-(2-hydroxydodecyl)-L-alanine (L-C₁₂HAla)²³ and *N*-(2-hydroxydodecyl)-L-valine (L-C₁₂HVal)²⁴ amphiphiles in water have been reported in our recent publications. It has been shown that one of these amphiphiles in dilute solutions forms large tubular aggregates in water at alkaline pH. It was demonstrated that intermolecular hydrogen bonding through the OH group in the hydrocarbon chain is responsible for the formation of supramolecular aggregates. In the course of our study of the aggregation behavior of L-C₁₂HVal in water at pH 12, it was accidentally discovered that water gelled at higher concentrations (>20 mM). In this report, we describe gelation ability of the amino acid-derived LMWHs, *N*-(2-hydroxy-*n*-alkyl)-L-valine (L-C_{*n*}HVal, *n* = 8, 10, 12, 14, and 16), *N*-(2-hydroxy-*n*-dodecyl)-L-alanine (L-C₁₂HAla), *N*-(2-hydroxy-*n*-dodecyl)-L-serine (L-C₁₂HSer), *N*-(2-hydroxy-*n*-dodecyl)-L-aspartic acid (L-C₁₂HAsp), *N*-(2-hydroxy-*n*-dodecyl)-DL-valine (DL-C₁₂HVal), and *N*-(*n*-dodecyl)-L-valine (L-C₁₂Val) in water. The objectives are (i) to investigate the effect of head-group structure and chain length, (ii) to examine the role of stereogenic center on the gelation ability and supramolecular structure formation, and (iii) to study how the hydrogels respond to change in pH, heat, and salt concentration.

Results and Discussion

Gelation Behavior. As already reported in our earlier works, the amphiphiles are soluble in water only at pH > 11.5.^{23,24} Therefore, the gelation studies were carried out in aqueous phosphate buffer of pH 12. In a typical gelation test, 10 mg of L-C₁₂HVal (33 mM) contained in a screw-capped vial was dispersed in 1 mL of buffer by sonicating for 10 min in a bath-type sonicator and then heated to 343 K in a water bath. The vial was then left to cool at room temperature. The heating–cooling cycle was repeated three times and finally left to cool at 298 K in a thermostating

water bath. The gelation occurred after 10–12 h. Interestingly, gelation could also be induced by gentle shaking of the vial after 2 h of cooling at 298 K. The gel formation of the amphiphile in water was tested by the “stable-to-inversion of the test tube” method (Figure 1). The hydrogel appeared optically opaque. However, under similar conditions, other structurally similar amphiphiles, L-C₁₂HAla, L-C₁₂HSer, and L-C₁₂HAsp, failed to gelate water. We tried also with higher concentration and/or lower pH. Depending upon solubility, they either formed a clear solution or precipitate even at higher concentration (50 mM) and/or lower pH. The results have been summarized in Table 1. The data suggest that a precise balance between hydrophilicity and hydrophobicity of the amphiphilic molecule is important for gelation. It is important to note that the corresponding DL-valine derivative was not observed to gelate water, which means enantiomeric purity is necessary for the gelation to occur. It should be noted that the amphiphile, L-C₁₂Val, which lacks the OH group in the hydrocarbon chain, also failed to exhibit gelation of water. The gelation was thermoreversible and could be achieved even after 10 heating–cooling cycles. The hydrogel of L-C₁₂HVal was found to be stable for months when left undisturbed under a condition of constant temperature (~298 K) and pH.

In order to examine the effect of hydrocarbon chain length on the hydrogel formation, the same study was performed with the amphiphiles L-C₁₆HVal, L-C₁₄HVal, L-C₁₀HVal, and L-C₈HVal. It was observed that while L-C₁₄HVal, L-C₁₂HVal, and L-C₁₀HVal produced hard gel, L-C₁₆HVal produced a weaker hydrogel, which started to flow upon gentle shaking. This means that weak gels of L-C₁₆HVal have yield stress much lower than that of the hydrogels of the other gelators. This has been discussed further below. On the other hand, L-C₈HVal produced precipitate (see Figure 1A). It should be noted that the gelation time decreases with the increase of chain length. The gelation ability of the amphiphiles was quantitatively evaluated by determining “critical gelator concentration” (CGC), which is defined as the minimum amount of gelator required to gelate 1 mL of solvent at a give temperature. The gelation ability can also be expressed in terms of “gelation number”, which is defined as the mole ratio of entrapped water and the gelator. In other words, gelation number is the maximum number of solvent molecules that are immobilized per molecule of the gelator. The CGC values as well as gelation numbers have been included in Table 1. It can be observed that the amphiphiles have reasonably high gelation numbers (i.e., low CGC values) and are comparable to those reported for other amino acid-derived hydrogelators.^{4,25} The data in Table 1 show that the gelation number is maximum for L-C₁₄HVal (*n* = 14). This means that an optimum lipophilicity of the hydrocarbon chain is needed for efficient gelation of water by the amphiphile. It is well-known that during gelation amphiphilic gelators self-assemble into fibrous aggregates, which cross-link or entangle during the aggregation process, resulting in a rigid 3-D network structure that accommodates the solvent by interfacial tension and supramolecular interactions. It can be found that the C8 amphiphile does not form gel. Since C8 and C14 amphiphiles are structurally similar

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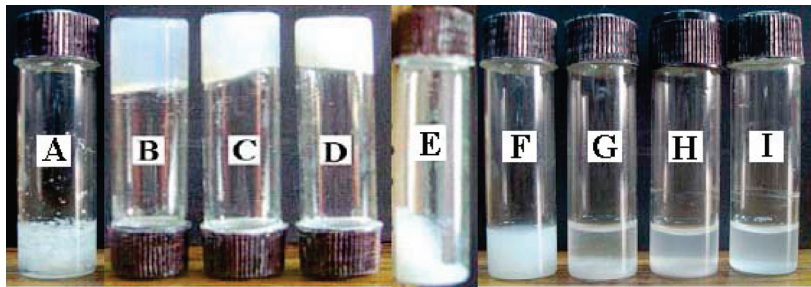


Figure 1. Photographs of the vials showing hydrogel formation: (A) 12 mg of C₈HVal, (B) 6 mg of C₁₀HVal, (C) 6 mg of C₁₂HVal, (D) 5 mg of C₁₄HVal, (E) 6 mg of C₁₆HVal, (F) 15 mg of (±)-C₁₂HVal, (G) 14 mg of C₁₂HAla, (H) 15 mg of C₁₂Hser, and (I) 16 mg of C₁₂HAsp in 1 mL of pH 12 buffer.

Table 1. Gelation Properties of *N*-(2-Hydroxyalkyl)-L-amino Acids in Phosphate Buffer, pH 12, at 298 K^a

compound	mol wt	state	CGC (mg/mL)	gelation no. (±52)	$T_{gs} \pm 1$ K (20 mM)
L-C ₁₂ HAla	273.50	P			
L-C ₁₂ HAsp	317.50	P			
L-C ₁₂ HSer	289.60	P			
L-C ₈ HVal	245.39	P			
(±)-C ₁₂ HVal	301.45	P			
L-C ₁₀ HVal	273.42	G	4.1 (3.9)	3688 (3926)	314 (311)
L-C ₁₂ HVal	301.45	G	3.6 (3.0)	4679 (5510)	323 (318)
L-C ₁₄ HVal	329.95	G	3.3 (2.9)	5581 (6248)	326 (324)
L-C ₁₆ HVal	357.53	WG	3.7 (3.4)	5398 (5790)	300 (298)

^a P: precipitate at 50 mM; G: gel; WG: weak gel; the quantities within the parentheses represent values obtained in the presence of 100 mM NaCl.

except the hydrocarbon chain length, the greater gelation ability of C14 must be due to the greater hydrophobicity of the alkyl chain. In other words, hydrophobic interaction seems to be the dominant driving force for gelation by the amphiphilic molecules. However, despite having higher hydrophobicity of the alkyl chain, the C16 gelator has gelation number lower than that of C14 amphiphile. This is perhaps because of the formation of relatively less rigid 3-D network as a consequence of either reduction in flexibility due to shortening of the length of the fibers or change in morphology of the supramolecular aggregates. This is also evidenced by the lower gel melting temperature (T_{gs}) in the case of C16 amphiphile (see discussion below) compared to L-C₁₄HVal.

Gelation of Salt Solutions. Since the gelation occurred in buffer solution, it can be concluded that perhaps the prior presence of different ions in water does not affect gel formation by the amphiphiles. Indeed, gelation occurred in the presence of salts such as NaCl. However, the presence of different ions in water (pH 12) led to an increase in time required for the onset of gelation. For example, while 20 mM phosphate buffer could be gelated by L-C₁₂HVal in 2 h, with 100 mM NaCl solution the gelation by the same gelator occurred after 6 h at 298 K. This means that inclusion of salts affected the kinetics of the gel formation. The gelation behavior of L-C₁₂HVal was also studied in phosphate buffer (pH 12) containing different NaCl concentration. Depending upon the salt concentration it took about 2–12 h to gelate the buffer. The variation of gelation number with NaCl concentration is shown by the plot in Figure 2. It is observed that gelation number increases linearly with the increase of salt concentration, reaching a plateau at ~100 mM, within the experimental error limit. To compare the gelation behavior of the gelators in the presence of salt, the gelation number was also determined in the presence of 100 mM NaCl. The CGC and corresponding gelation number values have been included in Table 1. With all the gelators

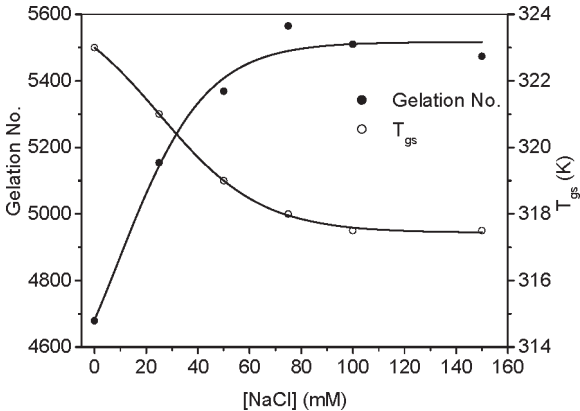


Figure 2. Plot of gelation number and T_{gs} of the hydrogel of C₁₂HVal in phosphate buffer, pH 12, as a function of [NaCl].

the gelation number increased in the presence of NaCl. It is well-known that salt promotes growth of micelles of ionic surfactants. This is because the presence of NaCl decreases the electrostatic repulsion and as a consequence increases the hydrophobic interaction, leading to tight packing of the hydrocarbon chains. Such aggregate growth results in more noncovalent cross-linking and/or mechanical entanglement among the fibers to create a more rigid 3-D network and thus enhances the gelators ability of solvent entrapment. If this was the case, then one would expect an increase of gel melting temperature in the presence of NaCl. However, as discussed below, T_{gs} decreased in the presence of salt. Therefore, it appears that increase of gelation number is due to change in morphology of the supramolecular aggregates. The excess water gelled, however, might be partly due to water molecules required for hydration of the ions of the added salt.

Thermal Stability of the Hydrogels. Thermal stability of the hydrogels was investigated by the inverted-tube method. To compare the thermal stability, we have measured the gel melting temperature (T_{gs}) for a known concentration

(20 mM) of each of the gelators. The results have been included in Table 1. The hydrogels indicated melting in the temperature range 300–326 K. Above this temperature the gelled mass started to flow and changed to sol form. The gel melting temperature is observed to increase with the chain length, reaching a maximum at C₁₄ and then drops down. Thus, L-C₁₆HVal has lower T_{gs} value compared to that of L-C₁₄HVal. This is consistent with the variation of gelation number with the chain length (see data in Table 1) and therefore can be explained on the same line. The large difference between T_{gs} values of C14 and C16 amphiphiles is perhaps due to partial bending (or tilting) of the hydrocarbon tail of C16 amphiphile, which causes the packing of the hydrocarbon tails less tight. This reduces the growth of the supramolecular aggregates. That is the length of the fibers is decreased, resulting in less cross-linking or entanglement. This means formation of less rigid 3-D network of the fibers, which is manifested by the decrease of melting temperature. This as discussed below is further supported by the lower yield stress value of the hydrogel of C16 amphiphile compared to that of C14 amphiphile.

Increasing the concentration of the gelator is known to increase the thermal stability of the gel, which means an increase of the T_{gs} value.²⁶ In the present study, we also found a similar trend in which the melting temperature of the gel was observed to increase with increasing gelator concentration. As a representative example, the effect of concentration of L-C₁₂HVal on the thermal stability of the resultant gel is shown in Figure 3. The higher thermal stability at higher concentration might be because of tight packing of the gelator molecules due to enhanced H-bonding and hydrophobic interactions as a result of incorporation of larger number of gelator molecules in the aggregate. This means that the increase of T_{gs} is linked to growth of aggregates. The growth of the one-dimensional aggregates makes them more flexible, causing more entanglement of the fibers and thereby increasing the T_{gs} value.

The thermal stability of the hydrogels in the presence of salt was also investigated. Figure 2 shows the plot of T_{gs} for L-C₁₂HVal gelator as a function of [NaCl]. As seen, the T_{gs} value linearly decreases initially with [NaCl] and then levels off at concentration ~100 mM. The T_{gs} values of the gelators measured in the presence 100 mM NaCl have been listed in Table 1. The data show that T_{gs} value decreased by 2–5 K in the presence of salt. That is the hydrogels become thermally less stable upon addition of salt. This could be, for the reasons described in the preceding section, due to change in the microstructure of the supramolecular aggregates. In the presence of salt tight packing of the hydrocarbon chains might weaken or disrupt the intermolecular hydrogen bonds, thus transforming ribbonlike aggregates into long flexible rodlike micelles.

Rheology. To examine the mechanical strength, the hydrogels were characterized by rheological measurements. The storage modulus (G') is an important parameter which characterizes the strength of gels, which can estimate the degree of resistance against mechanical disturbance. On the other hand, the loss modulus (G'') measures the tendency of a material to flow under stress. Plots G' and G'' vs the imposed stress for the hydrogels of L-C₁₂HVal and L-C₁₄HVal and L-C₁₆HVal amphiphiles have been depicted in Figure 1S of

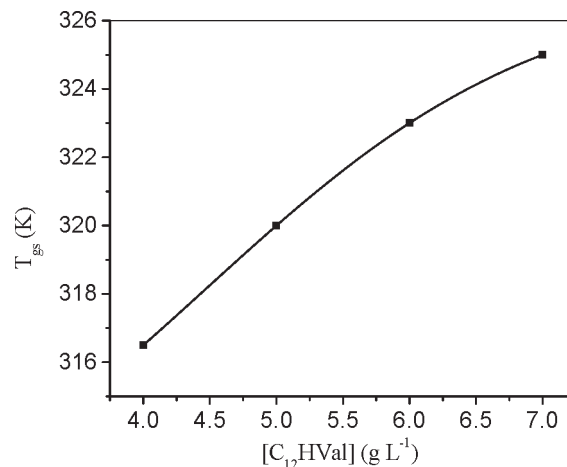


Figure 3. Effect of concentration of L-C₁₂HVal on T_{gs} .

the Supporting Information. The so-called “yield stress” (designated as σ_y) at which the gel breaks under the applied force and begins to flow was recorded as 360, 600, and 28 Pa for L-C₁₂HVal, L-C₁₄HVal, and L-C₁₆HVal gelators, respectively. Thus, the rigidity of the hydrogels is in the order L-C₁₄HVal > L-C₁₂HVal > L-C₁₆HVal. Three typical rheograms of the oscillatory shear for three hydrogel samples having same concentration of L-C₁₂HVal and L-C₁₄HVal and L-C₁₆HVal at 298 K have been shown in Figure 4. It can be found that the hydrogel samples showed similar rheological behavior. Both G' and G'' , within the limit of experimental error, show very little dependence of frequency over the range below 100 rad/s, which is characteristic of gels. In the case of L-C₁₄HVal (Figure 4b), G' is almost an order of magnitude greater than G'' , demonstrating the dominant elastic behavior of the system. Similar behavior can also be observed with L-C₁₂HVal and L-C₁₆HVal (Figure 4a,c). However, in the case of L-C₁₄HVal, the magnitudes of G' and G'' are much higher than the corresponding quantities of the L-C₁₂HVal and L-C₁₆HVal gelators. This suggests that the mechanical strength of the hydrogel of L-C₁₄HVal is much higher than that of L-C₁₂HVal and L-C₁₆HVal. Such a significant difference in the viscoelastic properties of the hydrogels can be attributed to the variation of supramolecular interactions and 3-D network structure in the gels of the amphiphiles. The order of stability/rigidity of the hydrogels is consistent with the values of melting temperatures. The plots of complex viscosity (η^*) as a function of frequency show a continuous nonlinear decrease, suggesting non-Newtonian behavior. Thus, it can be concluded that the rheological behavior of the hydrogels is characteristic of soft viscoelastic solids.

Morphology of the Hydrogels. To investigate the nature of microstructures and morphologies, a thin film of the hydrogels was dried under vacuum desiccators for 24 h, and then the specimens were examined under FE-SEM. The micrographs (see Figure 5) of the hydrogels exhibit 3-D network formed by cross-linking and/or physical entanglement of ribbonlike micelles of high aspect ratio. The average diameter of the ribbons is about 80–120 nm and a few micrometers long, which accounts for the opacity of the hydrogels (Figure 1). The water molecules are immobilized in such a network of fibers through surface tension resulting in gel formation.^{1b} It should be noted that gels prepared from the same amphiphile exhibit same morphology after 12 h aging and 2 h aging followed by gentle shaking at 298 K.

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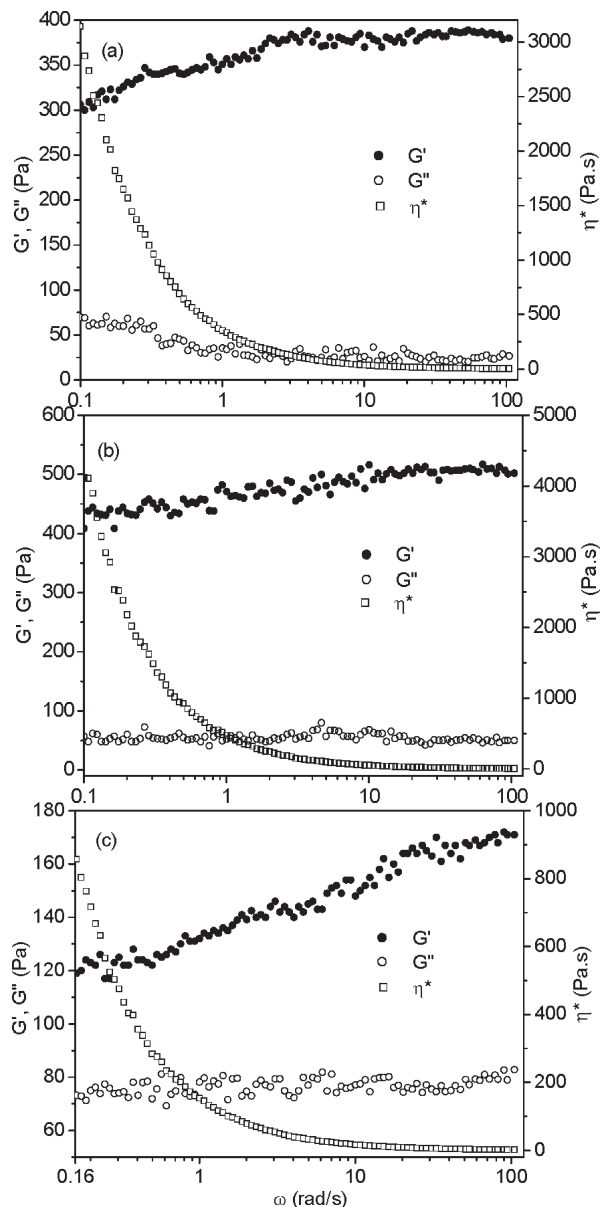


Figure 4. Rheological measurement of hydrogels after 48 h of aging; 10 mg of each amphiphiles in 1 mL pH 12 phosphate buffer: (a) L-C₁₂HVal, (b) L-C₁₄HVal, (c) L-C₁₆HVal.

Nature of Supramolecular Aggregation in Hydrogels. It has been well established that the self-aggregate structure of amphiphile in the gel is reflected by the dry gel state, and much structural information was obtained from the XRD spectrum of the dry gel in the small angle region.²⁷ In fact, XRD is a powerful tool in elucidating long-range structures of molecular ordered assemblages. Therefore, in order to obtain the organized structure of the self-assembly, we have performed XRD measurements with the dry gel of L-C₁₂HVal as representative of the series. The XRD pattern is illustrated in Figure 6. The figure reveals three sharp reflection peaks indicating that the amphiphile assembles into an ordered structure. The long d spacing (2.0, 0.98, and 0.65 nm) of the aggregates are almost at a ratio of 1:1/2:1/3 and correspond to the 100, 200, and 300 planes of the

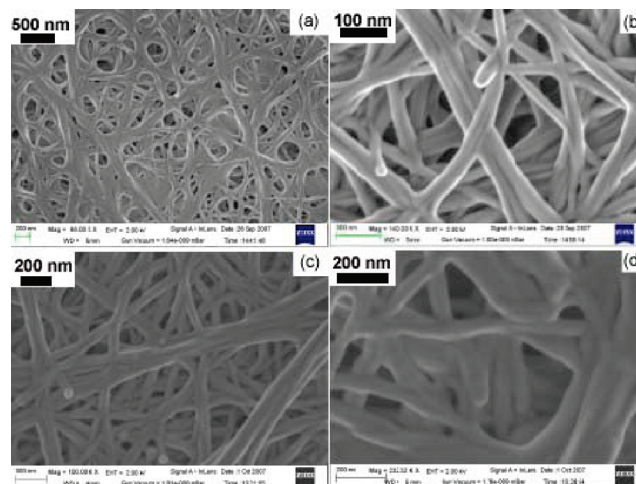


Figure 5. FESEM micrograph of the air-dried gels of (a) 15 mg of L-C₁₀HVal, (b) 10 mg of L-C₁₂HVal, (c) 10 mg of L-C₁₄HVal, and (d) 8 mg of L-C₁₆HVal in 1 mL of pH 12 buffer.

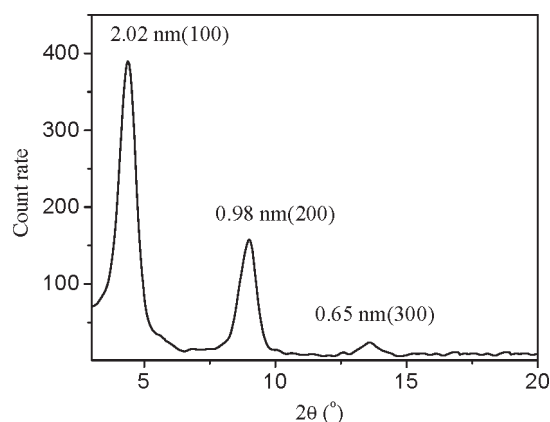


Figure 6. Powder XRD of the dry gel prepared from L-C₁₂HVal in pH 12.

lamellar structure. In fact, studies on the aggregation behavior of the amphiphiles in water have also demonstrated formation of bilayer structures above a critical aggregation concentration.^{23,24} The bilayer thickness (d) of the (100) plane as calculated from the position of the intense peak is about 2.0 nm, which is smaller than twice the extended hydrocarbon chain length (1.40 nm) of L-C₁₂HVal. To obtain the hydrocarbon chain length, the structure of L-C₁₂HVal was drawn first using a Chem Draw (Chem3D Std 4.0) software, and then from the software, the energy was minimized using the MM2 program. From this energy-minimized structure the hydrocarbon tail length was calculated as the linear distance between the carbon atoms attached to the NH group and the CH₃ carbon of the hydrocarbon chain. This means that the hydrocarbon chains of the two adjacent layers are interdigitated. The specific structure giving the gel properties can be understood from the FE-SEM pictures. A two-layer tail-to-tail arrangement of amphiphiles (see Figure 7) has been confirmed by the XRD spectrum of the dry gel of L-C₁₂HVal. The fact that the increase of the chain length of the hydrocarbon tail enhances the ability to form gel is consistent with the mechanism proposed for the formation of hydrogel as the formation of bilayer lamellar self-assemblies in water, which depends upon the chain length of the hydrocarbon tail of amphiphiles.

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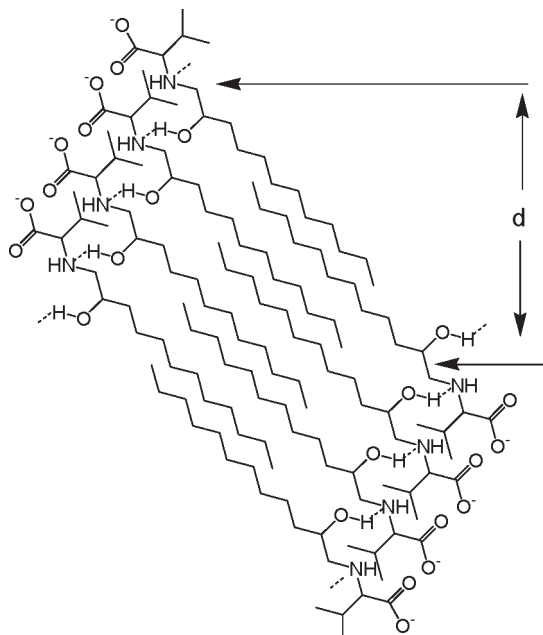


Figure 7. Bilayer arrangements of the L-C₁₂HVal amphiphilic molecules in the gel fibers.

Driving Force for Supramolecular Aggregation. It has been reported in the literature that carboxylic acid group can promote gelation by intermolecular hydrogen bonding.²⁸ However, this opportunity is lost when the acid is ionized. Each of the amphiphiles described above has an acidic (COOH) and a basic (NH) functional group. Therefore, the ionization state of the amphiphiles may have an effect on the gelation behavior in a given solvent. In order to examine this, the effect of pH on the gelation behavior of the amphiphiles was investigated. The hydrogels could not be prepared when pH of water was even slightly greater than 12. On the other hand, the gelators remained insoluble at pH < 11.5. Since the pK_a of the carboxylic acid and secondary amine group is around 5 and 10, respectively, the amphiphiles are expected to be present mostly in the anionic form at pH > 11.5. Thus, it is clear that hydrogen bonding among carboxylic acid groups is not responsible for gelation. Since the amphiphiles have a secondary amine group and a secondary alcohol group in the hydrocarbon tail, perhaps intermolecular hydrogen-bonding interaction between these groups might be a factor for strong hydrophobic interaction among gelator molecules. The necessity of hydrogen bond formation through the OH group is confirmed by the failure of hydrogel formation by the structurally similar amphiphile, L-C₁₂Val, which does not have OH group in the hydrocarbon chain. In fact, such a conclusion was also made in our earlier work on the self-assembly behavior of structurally similar amphiphile L-C₁₂HAla.²³ This is a clear indication that the OH proton is hydrogen bonded. Though N—H—O hydrogen bonding, in principle, is possible, can be neglected because it is much weaker compared to the O—H—N hydrogen bond. Thus, on the basis of above results, a two-layer tail-to-tail arrangement of amphiphiles (Figure 7) in which the OH proton is hydrogen bonded to the N atom of the secondary amine group of neighboring molecule is proposed.

Conclusions

In conclusion, we have demonstrated that *N*-(2-hydroxyalkyl)-L-valine amphiphiles at a fairly low concentration (~0.4% w/v) form thermoreversible hydrogels in buffered water at pH 12. The gelation ability was found to be highest with L-C₁₄HVal amphiphile due to optimum lipophilicity of the hydrocarbon chain. The enantiomeric purity is important for gelation to occur. Structurally similar amphiphiles with other L-amino acids (alanine, serine, aspartic acid) as head group failed to gel water under similar conditions. The gelation number of the L-C₁₄HVal gelator is quite high compared to L-C₁₄HAla. The gelation was found to be very sensitive to pH. No gelation could be observed below pH 11.5 or at pH > 12. The hydrogels are reasonably stable with respect to temperature and salt concentration. Among the hydrogelators, L-C₁₄HVal has the highest gel melting temperature (326 K). The melting temperature was observed to increase with the increase of gelator concentration. Addition of NaCl made the gel slightly less stable. The gel melting temperature decreased continuously with the increase of NaCl concentration. The supramolecular aggregates of the amphiphiles have ribbonlike structures, which are formed through intermolecular hydrogen-bonding interaction between the OH and NH groups of the hydrocarbon chain of adjacent molecules. However, the hydrophilic and lipophilic balance is very important for the gelation.

Experimental Section

Materials. Deuterium oxide (99.99 atom % D), NaOD (40 wt % in D₂O, 99.5 atom % D), CDCl₃ (99.96 atom % D), 1,2-epoxyalkanes (85–96%) (Aldrich), 1-bromododecane (98%), L-alanine (99%), L-valine (99%), L-serine (99%), L-aspartic acid (99%), and DL-valine (99%), (SRL, Mumbai) were used without further purification. All solvents used were obtained locally and were distilled and dried whenever required. Analytical grade disodium hydrogen phosphate, sodium hydroxide, and hydrochloric acid were procured locally and were used directly from the bottle. The amphiphiles, *N*-(2-hydroxy-*n*-alkyl)-L-amino acids, were prepared according to the procedure described elsewhere²⁹ and was purified by recrystallization from ethanol or 50–75 v/v% ethanol–water mixture. The chemical structure of the amphiphiles was identified and checked for purity by usual methods (IR, ¹H NMR, TLC). The details are available in the Supporting Information.

General Methods. The ¹H and ¹³C NMR spectra were recorded on a Bruker 400 MHz instrument. IR spectra were recorded with a PerkinElmer model 883 IR spectrometer. The specific rotations were measured with a Jasco P-1020 digital polarimeter. Melting point was determined by use of an Instind (Kolkata) melting point apparatus in open capillaries. A Thermo Orion (model 710 A+) pH meter fitted with a combined glass electrode was used for all pH measurements. Temperature-controlled measurements were carried out by use of a Thermo Neslab RTE-7 circulating bath.

Gelation Test of Water. Required amount (weight) of the amphiphiles was taken in a 4 mL glass vial, and a known volume of pH 12 buffer solution was then added. The solid was dispersed by sonication for 10 min and heated to 343 K and then cooled to 298 K. The heating–cooling cycle was performed at least three times, and the mixture was kept at 298 K in a thermostating water bath. The gelation was confirmed by inversion test of the vial. The gel–sol transition temperature (*T*_{gs}) was determined by checking the flow behavior at different

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temperature. The temperature at which the gels started to flow upon inverting the vial was recorded as T_{gs} .

Field Emission Scanning Electron Microscopy (FE-SEM).

For FE-SEM measurements, a thin layer of the sample was cast on a thoroughly cleaned copper foil and dried in vacuum desiccators for 24 h. A layer of gold was spluttered on the sample by vacuum spray to make conducting surface and finally transferred into the field emission scanning electron microscope (FESEM, Zeiss, Supra-40) operating at 5–10 kV.

X-ray Diffraction (XRD). Small-angle X-ray diffraction measurements were performed using cast films of the gel. Spreading a few drops of the dispersion on a glass slide and slowly drying in atmosphere at room temperature prepared the films. The XRD patterns of the cast films were measured on a PAN Analytical (PW 3040/60), X' pert PRO (Netherland) diffractometer using Cu K α (154.05 pm) radiation (40 kV, 30 mA). The spectrum was measured at room temperature between 2° and 40° in the 2 θ scan mode (2.5° min⁻¹) in steps of 0.02° in 2 θ .

Rheological Measurements. The viscoelastic properties of the samples were investigated using an ADS rheometer (Bohlin Instruments, Malvern, UK). Parallel plate (25 mm diameter) geometry was employed with a gap of 0.08 mm. The plate temperature was controlled by a water-circulating thermostating bath (MC, Julabo, Germany) with a temperature stability of ± 0.1 K. All measurements were taken on matured gels after 48 h of cooling. An amount sufficient to fill completely the gap between the plates was carefully scooped out and spread over

the lower plate. An equilibration time of 10 min was allowed before taking measurements for each sample. Solvent evaporation was minimized by covering the rheology plates with a solvent-soaked sponge chamber. The hydrogels were subjected to an oscillatory strain amplitude sweep measurement at a constant frequency of 10 rad/s for the strain range 0.001–0.2% at 298 K to determine the linear viscoelastic region. Oscillatory frequency sweep measurements were performed at a constant strain of 0.05% in the frequency range 0.1–100 rad/s. The ADS-100 software converted the torque measurements into either G' or G'' with angular frequency (ω). Dynamic viscosity (η^*) was calculated using G' and G'' by the following equation:³⁰

$$\eta^* = \frac{[G'^2(\omega) + G''^2(\omega)]^{0.5}}{\omega} \quad (1)$$

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Supporting Information Available: Synthetic procedure, elemental analysis, and chemical identification of all the amphiphiles; determination of yield stress from the plot of storage modulus (G') and loss modulus (G'') vs stress. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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