

## A Novel Type of Small Organic Gelators: Bis(Amino Acid) Oxalyl Amides

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Bis(*S*)-leucine (**1**) and bis-(*R*)- $\alpha$ -phenylglycine (**2**) oxalyl amides are discovered to be efficient small organic gelling agents capable of gelling water while their stereoisomers *meso*-**1** and *meso*-**2** as well as their racemates are found to be non-gelling and poorly gelling, respectively; IR, fluorescence emission spectroscopy and TEM are used to study assemblies of (*S,S*)-**1** and (*R,R*)-**2** in gels.

There have been several recent reports describing the gelling properties of small organic molecules of diverse structures.<sup>1–7</sup>

Here we report on a novel type of bis(amino acid) gelators **1** and **2** whose gelation properties were discovered fortuitously during a variant synthesis of macrocycles **9**.<sup>8</sup> The gelling properties of **1** and **2** differ from those found for other amino acid type gelators,<sup>1,2,4,5</sup> **1** and **2** are able to gelate organic solvents and water, while the other gelators form gels only with organic solvents. This difference may be explained on the basis of the structural characteristics of **1** and **2**, such as the absence of long alkyl chains but the presence of carboxylic groups and an oxalyl amide fragment which gives them great hydrogen-bonding potential, with a total of eight hydrogen bond donor and acceptor sites in a molecule.

First, we investigated the gelling properties of single enantiomers (*S,S*)-**1** and (*R,R*)-**2**. These compounds are most efficient when gelling water (Table 1); the formed gels are stable at room temperature for weeks. Their thermal stability was determined by steel ball experiments,<sup>1,9</sup> which gave the gel-melting temperatures  $T_{gel}$  for (*S,S*)-**1** and (*R,R*)-**2** as 78 and 55 °C, respectively. The TEM image of the (*S,S*)-**1** aqueous gel (Fig. 1) shows a dense three-dimensional network of long fibres, the smallest being *ca.* 10–20 nm in diameter. The bis-(*R*)- $\alpha$ -phenylglycine derivative (*R,R*)-**2** also gelates dioxane with comparable efficiency but is unable to form gels with any other

solvent from Table 1. The bis(leucyl)derivative (*S,S*)-**1**, however, was found also to gelate solvents of medium and low polarity. The same compound formed gels with 1 : 2 and 1 : 15 mixtures of ethyl acetate with hexane and dichloromethane, respectively. The optically active analogues (*S,S*)-**3** and (*S,S*)-**4**, constructed from (*S*)-phenylalanine and (*S*)-valine, respectively, and possessing only one methylene substituent more or less on the chiral centre (in comparison with **2** and **1**) were unable to gelate any solvent from Table 1. Next, we tested optically inactive *meso*-diastereoisomers of **1** and **2** and found them unable to gelate any solvent from Table 1. The racemates of **1** and **2** were found to be poor gelators in comparison to single enantiomers. The above observations suggest that gelling properties of this type of gelator depend strongly on the structure of the substituent on the chiral centres as well as on the chirality of the gelator.

Compounds **5–8** were designed and synthesized specifically to contain selected structural fragments of gelators **1** and **2**.<sup>†</sup> Each of them, as well as the macrocycle **9**, has been tested as a gelling agent with the solvents used in the studies with **1** and **2**. None of compounds **5–9** were found to form gels with any of the solvents investigated. These experiments suggest that the presence of terminal free carboxylic groups together with oxalyl amide fragment are essential for aggregation, presumably through intermolecular hydrogen bonding. This was confirmed by IR spectroscopy of the gels formed with organic solvents. First, the IR spectrum of the xerogel obtained by slow evaporation of the solvents from the gel prepared from (*S,S*)-**4** and a 1 : 15 ethyl acetate–dichloromethane mixture was measured. It showed broad bands at 3260 (NH), 1730 (carboxylic C=O), 1668 (amide I) and 1515 (amide II)  $cm^{-1}$ , which are indicative of intermolecular hydrogen bonding.<sup>2,4</sup> The positions of the corresponding bands in the spectrum of a diluted dichloromethane solution of (*S,S*)-**4** were found at 3398, 1726, 1690 and 1505  $cm^{-1}$  due to non-hydrogen bonded functionalities. This was confirmed by taking the IR spectrum of the diluted gel formed by (*S,S*)-**4** and a 1 : 12 dioxane–dichloromethane mixture, where bands of free and hydrogen-bonded amide groups appeared simultaneously at 3395, 3250 (NH), 1731–1723 (COOH), 1690, 1670 (amide I) and 1515, 1505  $cm^{-1}$  (amide II).

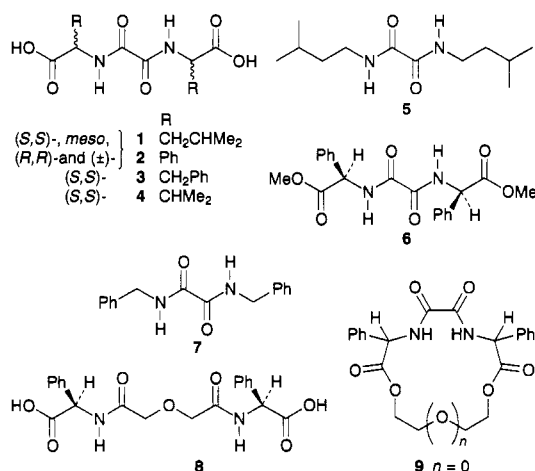


Table 1 Gelation ratios of **1**, **2** and various solvents

Solvent	Molar ratio solvent : gelator <sup>a</sup>			
	( <i>S,S</i> )- <b>1</b>	( $\pm$ )- <b>1</b>	( <i>R,R</i> )- <b>2</b>	( $\pm$ )- <b>2</b>
Water	1760	880	1786	ng <sup>b</sup>
Ethanol	556	305	ng	ng
Dioxane	279	74	1678	84
Tetrahydrofuran	156	ng	ng	ng
Ethyl acetate	113	ng	ng	ng
Acetone	150	ng	ng	ng

<sup>a</sup> Solvent added onto compound (10 mg) under investigation dissolved in 1–2 drops of Me<sub>2</sub>SO. <sup>b</sup> ng = no gelation occurred.

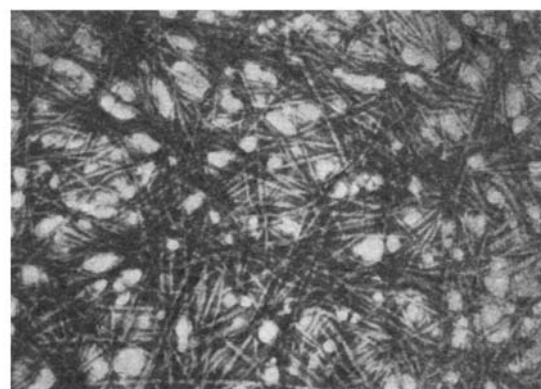


Fig. 1 TEM image of (*S,S*)-**1** aqueous gel negatively stained by osmic acid

The gelation of water by **1** and **2**, however, deserves special attention. As is well known, the formation of hydrogen bonds between solutes in water is highly disfavoured and occurs only by a cooperative process between sufficiently large aggregates or polymers. On the other hand, hydrophobic effects are known to induce formation of initial superstructures from amphiphiles or bola-amphiphile gelators in aqueous media.<sup>10</sup> On this basis, it can be assumed that the first aggregates of **1** or **2** in water are formed by hydrophobic interactions of Bu<sup>i</sup> or Ph groups which, in a second step, leads toward cooperative formation of intermolecularly hydrogen-bonded aggregates. In order to find proof for the possible hydrophobic interactions between the phenyl groups of (*R,R*)-**2**, we investigated its fluorescence properties in aqueous solution and in the aqueous gel. The fluorescence emission intensity ( $\lambda_{\text{ex}} = 290$ ;  $\lambda_{\text{em}} = 376$  nm) dependence on the concentration of (*R,R*)-**2** in water is shown in Fig. 2. The measurements showed clear drop in intensity on going from a  $10^{-4}$  to a  $10^{-3}$  mol dm<sup>-3</sup> solution. At a concentration of  $10^{-2}$  mol dm<sup>-3</sup> a gel was formed, resulting in a broad, low intensity emission spectrum. The spectrum showed, in addition to the 370 nm maximum, a new longer wavelength maximum at 425 nm. Heating the gel produced an increase in the emission intensity. The observed concentration and temperature effects on the fluorescence spectra of gel-forming (*R,R*)-**2** suggest the formation of aggregates with phenyl groups in close proximity, resulting in their mutual quenching, and in the gel, most probably the formation of excimers which give rise to the 425 nm band. It should be noted that the formation of excimers from poly-(*S*)-tyrosine with  $\lambda_{\text{ex}} = 420$  nm has been reported and taken as evidence for stacking interactions of *p*-hydroxyphenyl rings.<sup>11</sup> The concentration and temperature effects on the fluorescence emission of the non-gel-

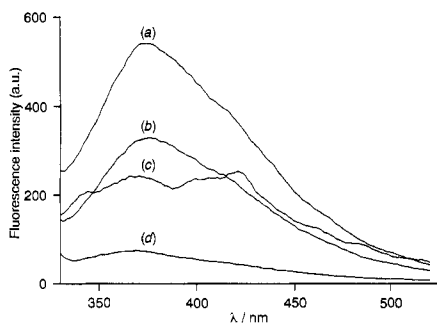


Fig. 2 The fluorescence emission spectra of (*R,R*)-**2** in water at concentrations of (a)  $10^{-4}$ ; (b)  $10^{-3}$ ; (c)  $10^{-2}$  (gel); and (d)  $10^{-5}$  mol dm<sup>-3</sup>

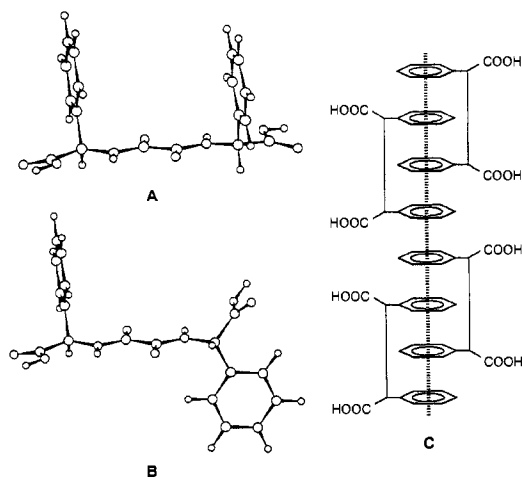


Fig. 3 The low energy conformations of A, (*R,R*)-**2** and B *meso*-**2**. C, Schematic presentation of the primary hydrophobically driven aggregation model for (*R,R*)-**2** in water.

forming *meso*-**2** are opposite to those observed for gelator (*R,R*)-**2**. The emission intensity increases with increasing concentration of *meso*-**2**, while increasing the temperature causes a lowering of the intensity as expected, due to non-radiative decay by enhanced collisions. These observations support the conclusion that the effects found for (*R,R*)-**2** originate from aggregation. They also strongly suggest that the primary aggregation of (*R,R*)-**2** and similarly that of (*S,S*)-**1** in aqueous media is governed by hydrophobic interactions between Ph and Bu<sup>i</sup> groups, respectively.

To shed more light on the intriguing fact that optically inactive *meso*-**1** and -**2** are not able to form gels, in contrast to single enantiomers of **1** and **2**, we used molecular modelling to identify the low energy conformations of the diastereoisomers.† A search of the conformational space of (*R,R*)-**2** gave the low energy conformation A (Fig. 3) with both phenyl rings in *syn* positions, face-to-face oriented, and at a distance of 3.6 Å which corresponds to the van der Waals thickness of a phenyl ring. On the contrary, the low energy conformation B of *meso*-**2** has its phenyl rings in almost the *anti* positions. The calculations showed that conformation A of *meso*-**2** should be ca. 8 kcal mol<sup>-1</sup> less stable than B. Combination of the observations from fluorescence measurements with the molecular modelling results provides a plausible explanation for primary (hydrophobically driven) aggregation of (*R,R*)-**2** in its stable conformation A by a zipper-like assembly (Fig. 3). C In such a model the phenyls are favourably stacked and shielded from hydration, while the more polar carboxylic groups are oriented toward bulk water. It is obvious that *meso*-**2** in the stable conformation B can not form such aggregates. Further work on the synthesis of novel gelators of this type and investigation of their aggregations are in progress.

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## Footnotes

† Compounds **1**–**8** were fully characterized by spectroscopy (IR, NMR) and elemental analyses

‡ A search of the conformational space for (*R,R*)-**2** and *meso*-**2** was performed by a standard procedure provided by the SYBYL software package of TRIPOS Assoc.

## References

- 1 E. J. de Vries and R. M. Kellogg, *J. Chem. Soc., Chem. Commun.*, 1993, 238.
- 2 K. Hanabusa, Y. Matsumoto, T. Miki, T. Koyama and H. Shirai, *J. Chem. Soc., Chem. Commun.*, 1994, 1401.
- 3 K. Hanabusa, T. Miki, Y. Taguchi, T. Koyama and H. Shirai, *J. Chem. Soc., Chem. Commun.*, 1993, 1382.
- 4 K. Hanabusa, K. Okui, K. Karaki, T. Koyama and H. Shirai, *J. Chem. Soc., Chem. Commun.*, 1992, 1371.
- 5 K. Hanabusa, J. Tonge, Y. Taguchi, T. Koyama and H. Shirai, *J. Chem. Soc., Chem. Commun.*, 1993, 390.
- 6 T. Brotin, R. Untermohlen, F. Fages, H. Bouas-Laurent and J.-P. Desvergne, *J. Chem. Soc., Chem. Commun.*, 1991, 416.
- 7 J.-H. Fuhrhop, P. Schneider, J. Rosenberg and E. Boekema, *J. Am. Chem. Soc.*, 1987, **109**, 3387.
- 8 M. Žinić, B. Bosnić-Kašnar and D. Kolbah, *Tetrahedron Lett.*, 1980, **21**, 1365.
- 9 A. Takahashi, M. Sakai and T. Kato, *Polym. J.*, 1980, **12**, 335.
- 10 J.-H. Fuhrhop and M. Krol, *Self-Assembling Lipid Membranes—From Planar Bilayer Sheets to Cloth-like Aggregates of Micellar Fibers in Frontiers in Supramolecular Organic Chemistry and Photochemistry*, ed. H.-J. Schneider and H. Durr, VCH, Weinheim, 1991.
- 11 S. S. Lehrer and G. D. Fasman, *Biopolymers*, 1964, **2**, 199.