Microporous organic crystals: an unusual case for L-leucyl–L-serine†

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Cocrystallized acetonitrile solvent molecules located inside 5.2 Å channels in the crystal structure of L-leucyl–L-serine have been replaced by I_2 molecules with full retention of the peptide scaffold.

Efforts into crystal engineering have been driven by the desire to construct new solid-state structures with functional properties.[1] Much attention has been focused on materials for second-order nonlinear optical activity and for magnetism, but also on porous materials[2] for which we have adopted the use of dipeptides with two hydrophobic residues. These compounds have been established as useful building blocks due to their general inability to adopt the traditional crystal packing patterns of peptides, which are characterized by being divided into distinct hydrophobic and hydrophilic layers or by containing small hydrophobic columns.[3] A systematic survey of the crystal structures of hydrophobic dipeptides has revealed that compounds with L-valine, L-isoleucine (side chains branched at C^{β}) as well as L-alanine residues tend to form structures with hexagonally symmetric hydrophobic pores,[4–6] called the VA-class after its first member L-valyl–L-alanine.[6] Compounds with L-leucine and L-phenylalanine residues (branched at C^{γ}) can also give porous structures, but with hydrophilic rather than hydrophobic inner surfaces. This group has been called the FF-class after L-phenylalanyl–L-phenylalanine.[7]

As part of a recent project, several dipeptides with one hydrophilic and one hydrophobic residue were crystallized, some including L-serine and L-threonine. The structure determination for one of these compounds, L-alanyl–L-threonine (AT), led to the molecular structure shown in Fig. 1a.[8]

It was the unprecedented observation of an intramolecular hydrogen bond that made us realize that a *C*-terminal serine or threonine residue may behave like a hydrophobic residue as there is no absolute need to find an intermolecular acceptor for the alcoholic H atom. We thus decided to investigate more compounds in this subfamily of dipeptides in order to find recurring examples of intramolecular hydrogen bonds, and possibly new types of crystal packing patterns.

The first dipeptide studied after AT was L-valyl–L-serine (VS), which was crystallized as a trihydrate.[9] The structure turned out to be unusual in many respects, but did not contain intramolecular hydrogen bonds. The present communication deals with the next peptide, the title compound L-leucyl–L-serine (LS).

High-quality needles were obtained by diffusion of acetonitrile into an aqueous solution of the peptide. Upon data collection it was immediately confirmed that the LS crystals possessed equivalent hexagonal symmetry.‡ The molecular structure shown in Fig. 1b proved to be very similar to the structure of AT, including the intramolecular hydrogen bond. The elongated main chain conformation also mimics other 1–2 dipeptides where 1 is a hydrophobic residue or glycine and 2 is serine or threonine, Table 1, but the crystal packing pattern of LS depicted in Fig. 2 is completely unique.

For the first time for a small peptide with a leucine reside, wide hydrophobic channels run along the hexagonal axes. The 5.2 Å van der Waals diameter is approximately the same as for the largest VA-class channels [for VA and L-alanyl–L-valine (AV)], but there are several important differences between the structures: (1) LS has cell dimension $a = 18.1402(3)$ Å, $c = 6.1582(2)$ Å, while crystals in the VA-class typically have $a \approx 14.5$ Å and $c \approx 10.0$ Å. (2) The VA-class channels are lined with two different types of side chains (from both residues), while in LS columns are formed by leucine side chains only; the methylene groups of the serine side chains form independent, small hydrophobic columns. (3) Hydrogen bond types are different; the number of amino H atoms donated to carboxylate groups is 3 for LS, but only 2 for the

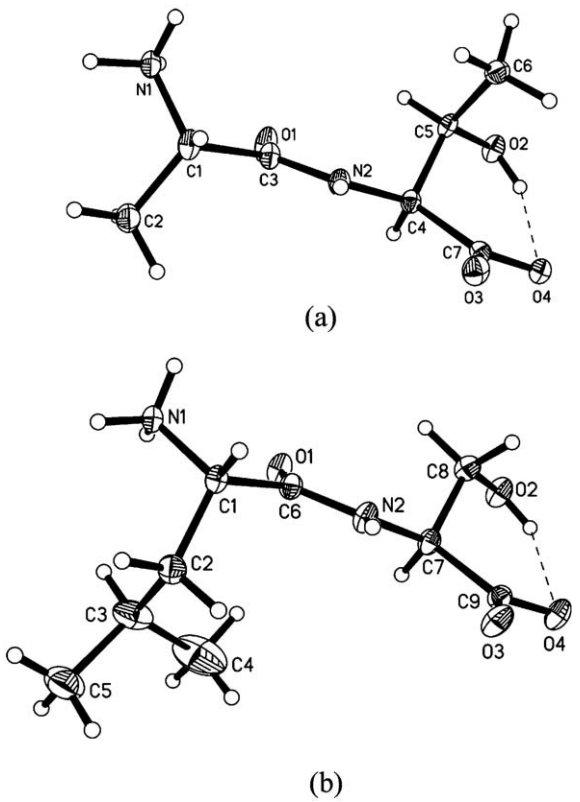

Fig. 1 The molecular structures of (a) AT and (b) LS. Displacement ellipsoids are shown at the 50% probability level.

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Table 1 Torsion angles (°) for selected dipeptides with *C*-terminal serine or threonine residues

	N_1–C_1^{α}–C'_1–N_2	C_1^{α}–C'_1–N_2–C_2^{α}	C'_1–N_2–C_2^{α}–C'_2	N_2–C_2^{α}–C'_2–O'_2	N_2–C_2^{α}–C_2^{β}–O_2^{γ}	C_2^{α}–C_2^{β}–O_2^{γ}–H
L-Leucyl–L-serine (LS)[a]	161.91(10)	167.73(10)	−165.10(10)	2.44(16)	−159.30(10)	−53.6(11)
Glycyl–L-serine[b]	165.1	172.0	−151.7	−21.8	61.1	−98.6
L-Alanyl–L-serine[c]	124.8	−178.0	−156.9	−1.7	62.9	−80.4
L-Valyl–L-serine (VS)[d]	134.2	174.7	−160.2	0.4	60.4	−79.3
Glycyl–L-threonine[e]	−169.4	−175.9	−123.4	−8.1	59.1	−116.3
L-Alanyl–L-threonine (AT)[f]	136.8	173.7	−166.2	−3.7	−164.3	−44.0

[a] Dry crystal, other torsion angles: N1–C1–C2–C3 58.15(15), C1–C2–C3–C4 –159.40(12), C1–C2–C3–C5 78.56(16). [b] ref. 10. [c] ref. 11. [d] ref. 9. [e] ref. 12. [f] ref. 7.

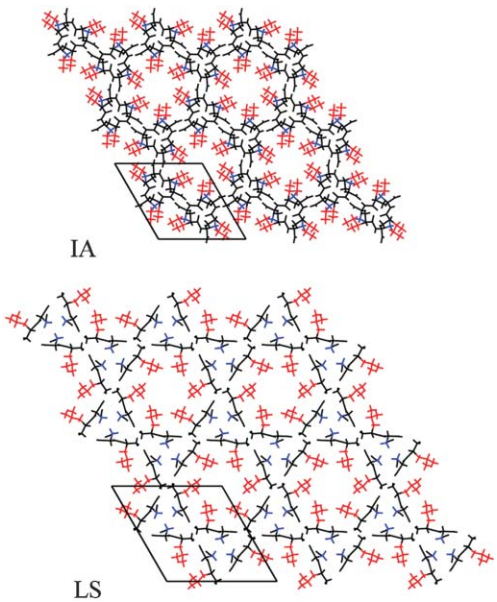

Fig. 2 Crystal packing of L-isoleucyl–L-alanine (IA, top),[4] an example of a member of the VA-class, compared with LS (bottom). Both structures are viewed along the hexagonal axes. Hydrophobic parts of the first and the second residue have been coloured red and blue, respectively.

VA-class. The VA-class carboxylate groups instead accept the amide >N–H of the peptide bond, a H atom that is donated to the side-chain –OH group in LS. (4) Model studies indicate that the crystal packing of LS is incompatible with branching at C^{β}.

The refinement of the LS structure obtained form the acetonitrile-wet crystal revealed diffuse electron density inside the channels. We interpret this as coming from acetonitrile, since its use as the precipitating agent had a profound effect on the size of the needles formed compared to experiments with 2-propanol or with slow evaporation. The sum of the occupancies for two acetonitrile positions (rigid refinement) was 0.084(5), or about 0.504 for one unit cell. Integration of the pore electron density using the SQUEEZE function in PLATON[13] was hampered (for all data sets) by the missing 010/100 reflections, which were behind the beam stop, but using the F_c-value as an estimate for F_o gives 10.2 electrons in a 147 Å^3 channel, while 11.1 electrons were expected from the X-ray refinement. Combining the length of the hexagonal axis, ~6.16 Å, with the effective van der Waals length of 4.9 Å for acetonitrile (sum of bonds 2.2 Å, sum of van der Waals radii for N and H 2.7 Å), we find that a head-to-tail stacking would give 1.26 solvent molecules for a unit cell. This shows that acetonitrile escapes the crystal easily; within less than a minute (the time taken from removing the crystal from the mother liqueur to mounting in the cold stream of the diffractometer) the solvent content was reduced to about 40% of the theoretical maximum. By comparison AV retained a 100% acetonitrile content for several minutes (and possibly for hours) at room temperature, while VA lost all alcohol solvent in a few seconds.

After drying (323 K for two days), a new data set was collected. Subsequent structure refinement showed no trace of electron density inside the channels (SQEEZE result 2.5 electrons). Loss of solvent for LS proceeds without visible changes to the crystal, a rare property shared with some coordination compounds[2] and nanotubular structures generated from stacking of large molecules such as bis-urea macrocycles[14] or cyclic oligopeptides,[15] but among small organic molecules probably only with the dipeptides of the VA-class. Modifications of the cell dimensions and the molecular geometry are negligible, Table 2. The calculated density of the dry crystal, 1.241 g cm^{-3}, is significantly higher than for VA and AV (1.041 and 1.069 g cm^{-3}, respectively), although the channels are of the same dimensions. This indicates that apart from the channels the close-packing of LS molecules is very efficient and accounts for the high stability of the system.

The specimen was finally soaked in toluene saturated with I_2 for three days. Toluene is too large to enter the channels, but I_2 does, whereupon the colour of the crystal changes from colourless to dark brown, Fig. 3. The transition from the acetonitrile solvate to the dry crystal and further to the I_2 complex is shown in Fig. 4. Two I-positions inside the channels were refined, with a combined

Table 2 Crystal data for the LS structures

Compound	LS acetonitrile solvate	LS (dry crystal)	LS I_2 complex
Chemical formula	$C_9H_{18}N_2O_4$·0.084CH_3CN	$C_9H_{18}N_2O_4$	$C_9H_{18}N_2O_4$·0.034I_2
Formula weight	221.4	218.3	226.9
Crystal system	Hexagonal	Hexagonal	Hexagonal
Space group	$P6_5$	$P6_5$	$P6_5$
a/Å	18.1402(3)	18.1253(3)	18.1470(3)
c/Å	6.1582(2)	6.1497(2)	6.1536(2)
V/Å^3	1754.96(7)	1749.66(7)	1754.97(7)
Z	6	6	6
μ/mm^{-1}	0.098	0.097	0.282
$N_{measured}$	11812	12017	27814
N_{unique}	1554	1581	6208
$N_{observed}$ $[F^2 > 2\sigma(F^2)]$	1482	1542	5153
R_{int}	0.020	0.019	0.050
R $[F^2 > 2\sigma(F^2)]$	0.025	0.025	0.054
w$R(F^2)$	0.068	0.069	0.173

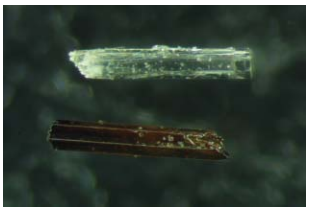

Fig. 3 Dry LS crystal (top) and soaked in I_2-solution (bottom).

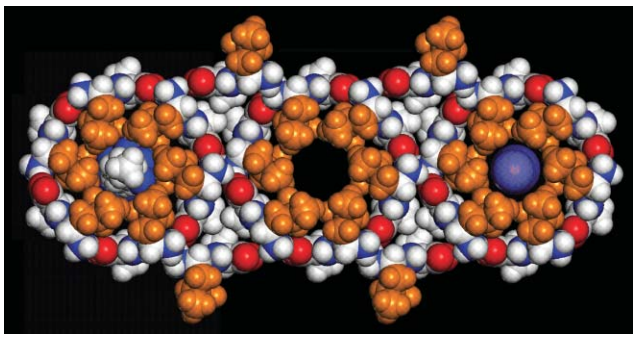

Fig. 4 Part of the LS crystal structure with, from left to right, channels filled with acetonitrile, empty and I_2. Colour coding is by atom type, except atoms in the leucine side chain which have been depicted in orange.

occupancy of 0.068(2), or 0.408 for one unit cell corresponding to 22 electrons; the SQUEEZE result in this case is 24 electrons within the same void volume as before, 147 Å^3. The effective van der Waals length of I_2 is 7.05 Å (2 × van der Waals radius 2.15 Å + I–I bond length 2.75Å). A head-to-tail stacking of I_2 molecules would thus give 0.87 I_2 molecules for each unit cell, or 1.75 I atoms. The refined occupancies thus indicate a degree of filling around 24%. Crystallographic data collected for a crystal soaked for 15 min (not presented in detail here) gave a filling degree of about 11%. The soaking time is thus obviously of importance, but a filling degree significantly lower than 100% may also reflect local adaptations of the channel structure to accommodate I_2 molecules, which make head-to-tail stacking impossible. Structure modifications furthermore lead to a higher final *R*-factor for the structure refinement compared to the acetonitrile-wet and the dry crystal, Table 2. Even though I_2 and acetonitrile are of approximately the same size as far as the van der Waals diameters are concerned (4.3 Å for I and 4.0 Å for methyl, respectively), van der Waals interactions are evidently much stronger for I_2 than for acetonitrile as crystals were stored for months at room temperature without any significant change in colour. A TGA/DSC analysis of a 1.1 mg sample of I_2-loaded crystals was carried out on a Rheometric Scientific STA 1500 with a supply of dry nitrogen set to 19 ml min^{-1}. The temperature was ramped at 5 °C min^{-1} from 20 to 300 °C. The weight remained constant to 120 °C, and from 150 to 300 °C an almost linear reduction in weight to 0 was observed. Thus, there are no indications that all I_2 leaves the host before it starts breaking down. The dry crystal melts/decomposes at 250 °C.

It can be seen from Table 2 that cell dimensions for the process illustrated in Fig. 4 remain fairly constant; the cell volume is only marginally smaller for the dry crystal than for complexes. This observation includes also the 11% loaded structure which has a = 18.1434(3) Å, c = 6.1597(2) Å and V = 1756.01(7) Å^3.

In summary, an uptake of guests has previously been associated primarily with metal–organic frameworks.[2,16] We have demonstrated, for the first time for a crystal of a small organic molecule, complete loss of cocrystallized solvent (acetonitrile) and re-uptake of an inorganic molecule (I_2) with full retention of the supramolecular host network (peptide).

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Notes and references

‡ Crystallographic data are reported in Table 2. Siemens SMART 1000 CCD-diffractometer, $Mo_{K\alpha}$ radiation (λ = 0.71069 Å), data collection on a 0.85 × 0.15 × 0.15 mm needle at 105 K with SMART,[17] data integration and cell refinement with SAINT,[18] absorption correction by SADABS,[19] structure solution by and least-squares refinement on F^2 with SHELXTL.[20] CCDC 266867–266869. See http://dx.doi.org/10.1039/b504976h for crystallographic data in CIF format.

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