VIP POM–Peptide Hybrids

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Polyoxometalate Clusters Integrated into Peptide Chains and as Inorganic Amino Acids: Solution- and Solid-Phase Approaches**

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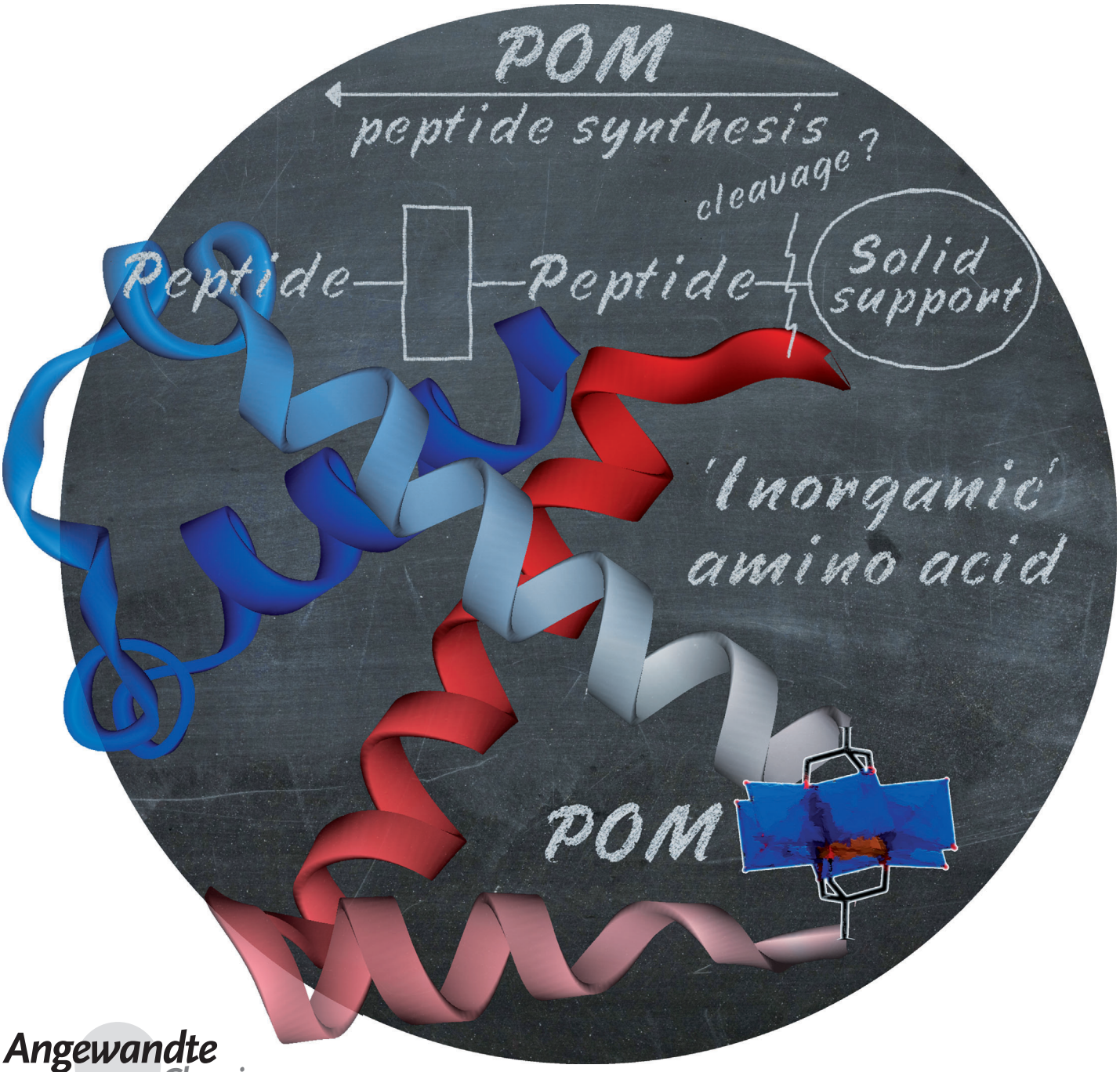
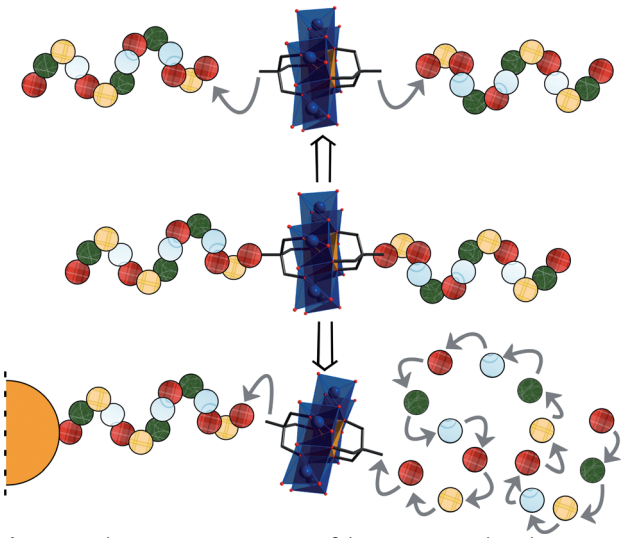


Abstract: *General synthetic methods for the grafting of peptide chains onto polyoxometalate clusters by the use of general activated precursors have been developed. Using a solution-phase approach, pre-synthesized peptides can be grafted to a metal oxide cluster to produce hybrids of unprecedented scale (up to 30 residues). An adapted solid-phase method allows the incorporation of these clusters, which may be regarded as novel hybrid unnatural amino acids, during the peptide synthesis itself. These methods may open the way for the automated synthesis of peptides and perhaps even proteins that contain "inorganic" amino acids.*

Polyoxometalates (POMs) are a large family of metal–oxygen clusters, which mainly comprise tungsten, molybdenum, and vanadium oxo-based units, with properties suitable for diverse applications in catalysis, medicine, and nanotechnology.[1] Control of their properties and the development of robust synthetic methods for their integration into wider chemical systems represent key challenges that have to be overcome and limit their wider exploitation. Hybrid organic–inorganic POMs combine metal oxide clusters with organic ligands that are coordinated to the cluster surface.[2] Their importance stems from the potential of these organic ligands to provide a more accessible and general means to tune cluster properties for function or assembly.[3] Various hybrid POMs can be formed by the incorporation of alkoxide ligands into metal oxide frameworks,[4] among them the Mn-Anderson cluster, which was first reported in 2002 by Gouzerh et al.[5] Since then, many reports have focused on the formation[6] and functionalization[7] of this hybrid organic–inorganic POM.

Nature's building blocks, amino acids, can be linked to produce peptide and protein structures that can form modular functional structures with unparalleled characteristics and unmatched self-organization properties. Modern methods for peptide synthesis now allow the routine design and production of sequences that target specific structures and properties.[8] These may be exploited as subunits of target-specific recognition motifs,[9] interfaces with biological materials,[10] or nanostructured materials.[11] Significant progress has already been made in the establishment of methods that exploit small peptides as pendant ligands on hybrid POMs,[12] but it has not yet been possible to incorporate POMs in a head-to-tail fashion within a peptide chain as unnatural amino acids. We wanted to explore the addition of POMs as hybrid "inorganic" amino acids, as they promise to introduce higher charge densities, redox activity, as well as additional coordination/catalytic sites into the protein; this would lead to exciting new applications and tools for protein biochemistry and POM medicinal chemistry (as POMs have great potential in medicine[13]). To realize this concept, significant synthetic barriers to the insertion of hybrid POMs into peptide chains must be overcome. These include the compatibility of the POM coupling chemistry with the peptide and the stability of the POMs under peptide-synthesis conditions.

To address this problem, we decided to use the hybrid Anderson platform, as two divergent ligands can be grafted onto the otherwise symmetric metal oxide framework; recently, we demonstrated the use of chromatography to isolate "asymmetric" hybrid compounds (with two different ligands linked to the metal oxide).[14] Herein, we present a set of synthetic approaches to the insertion of these hybrid Mn-Anderson building blocks into peptide sequences, both by liquid-phase reactions with preformed peptides and by integration into solid-phase peptide synthesis (SPPS; see Scheme 1). We demonstrate that these methods produce a variety of peptide–POM hybrids under mild conditions and incorporate a range of peptide chains with up to 30 amino acid residues. These results represent the first reported incorporation of POM clusters into the main chain of a biopolymer (among very few examples that describe the incorporation of inorganic building blocks).[15] Furthermore, the approaches illustrated should find applications in the wider hybrid field; to the best of our knowledge, this is the first time that solid-supported synthesis techniques have been used to build, release, and isolate inorganic hybrids, such as the POM systems presented here.



Scheme 1. Schematic representation of the two approaches that were considered for the integration of POM clusters into peptide chains: solution-phase reaction with pre-synthesized peptides (top) and incorporation of the POM building block as an unnatural amino acid during stepwise peptide synthesis (bottom).

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This new approach is based on the formation of *N*-hydroxysuccinimide (NHS) ester activated Mn-Anderson precursors (**1**–**2**; see Figure 1), which can be grafted at the N terminus of peptide chains. All precursors were synthesized

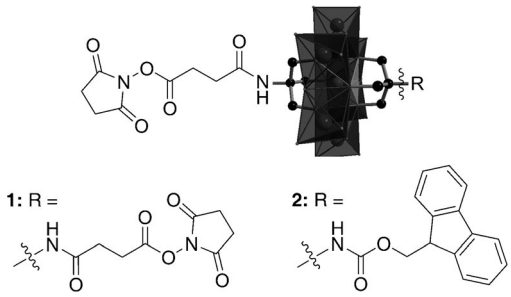

Figure 1. Mn-Anderson precursors **1**–**2** activated as NHS esters. Polyhedral/ball and stick representation of the Mn-Anderson $[MnMo_6O_{18}(O(CH_2)_3C)_2]^{3-}$ core; MnO_6 and MoO_6 are shown as light grey and black octahedra, respectively, and carbon atoms are depicted as black spheres; countercations were omitted for clarity.

by a two-step post-synthetic modification of previously reported Mn-Anderson clusters[6a,14] and isolated as pure solids. In each case, the free amine groups of the cluster were reacted with an excess of succinic anhydride, and the resulting carboxylic groups were then activated by treatment with *N*,*N*-dicyclohexylcarbodiimide (DCC) and NHS to form esters (see the Supporting Information for details). **1** is a symmetric cluster with two activated carboxylates, and **2** is an asymmetric compound with an activated "C terminus" and an Fmoc-protected "N terminus". By isolating the pure POM precursor, the carboxylic acid activation step (using DCC and NHS) is separated from peptide bond formation, which later allows the use of peptides in their native forms.

Initially, a liquid-phase approach was adopted, producing compounds **3**–**6** in good yields (>60%) and purity by the reaction of the symmetric precursor **1** with single amino acids or very short peptide sequences (Figure 2). All reactions were performed in *N*,*N*-dimethylformamide (DMF) at room temperature using equimolar amounts of the reactants and in the presence of *N*,*N*-diisopropylethylamine (DIPEA); products were isolated by precipitation with diethyl ether (Et_2O), with no further purification required. ESI-MS identification, along with the crystal structure that was obtained for **4**, confirmed the grafting by amidation of the very short peptides (or amino acids) onto the hybrid POM architecture. Despite their simple structure, very short oligopeptides are known to display interesting self-assembly behavior, especially di(phenylalanine) (Phe-Phe) and tri(phenylalanine) (Phe-Phe-Phe),[16] which have been employed as organizing motifs in many nanostructured materials;[17] as a result, the self-assembly properties of this series of hybrids are of considerable interest and will be extensively examined in a future publication.

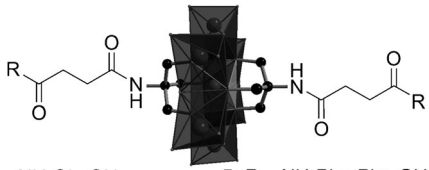

Figure 2. Compounds **3**–**6** were synthesized from precursor **1** using a liquid-phase approach.

To demonstrate that the broad applicability of this method reaches beyond small oligopeptides, a 15 amino acid peptide (H_2N-Ala-Asn-Thr-Leu-Ser-Ser-Thr-Ala-Ser-Thr-Leu-Glu-Ser-Tyr-Leu-OH; hereafter referred to as $\mathbf{P_1}$) was reacted in solution with precursor **1**. The resulting organic–inorganic hybrid **7** ($Na_{0.2}(C_{16}H_{36}N)_{2.8}[MnMo_6O_{24}(C_{74}H_{118}N_{17}O_{29})_2]$) was obtained in good yields (>80%) and fully characterized.

Its identity was confirmed primarily by ESI-MS, as all of the observed main peak envelopes are consistent with the proposed structure with various protonated and deprotonated forms of the peptide ligand and different countercations (Figure 3). The structure was corroborated by 2D NMR

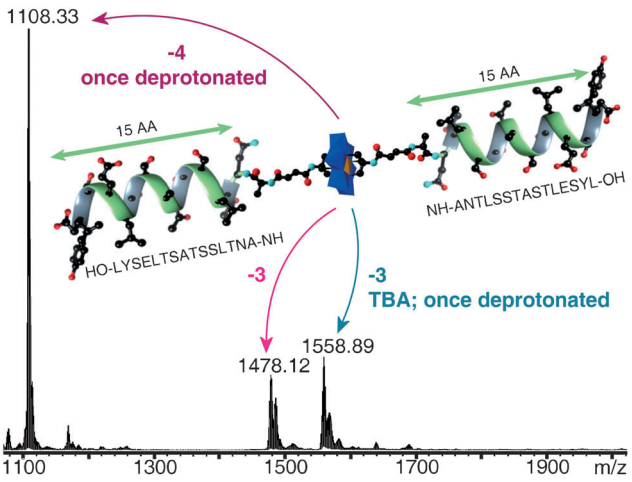

Figure 3. Mass spectrum of **7**; all major peaks are consistent with the desired product. For the representation of **7** it was assumed that the whole peptide chain adopts an α-helix arrangement.

spectroscopy with the identification of all of the components of the peptide chains, which confirmed that they had remained intact. An NH resonance, which results from the first alanine residue of $\mathbf{P_1}$, was also clearly observed, which confirmed that amide bond formation had occurred (for the free amine, no signal would have been detected).

Compound **7** is a well-defined covalent peptide–POM hybrid of unprecedented scale and can be seen as a POM cluster that has been incorporated as a linker of two peptide chains of 15 amino acids each. As the different structural motifs that are found in peptides exhibit distinct circular dichroism (CD) spectral features, and the overall secondary structure content of a protein can be assessed by a study of the absorption in the far-UV region,[18] we chose CD spectroscopy to investigate the secondary structure of this new hybrid. Considering its size and sequence, we did not expect $\mathbf{P_1}$ alone to manifest any regular secondary structure, and we did not know whether the presence of the charged metal oxide cluster would give rise to a specific folding of the peptide ligand. UV and CD spectra were acquired for both **7** and $\mathbf{P_1}$ in acetonitrile

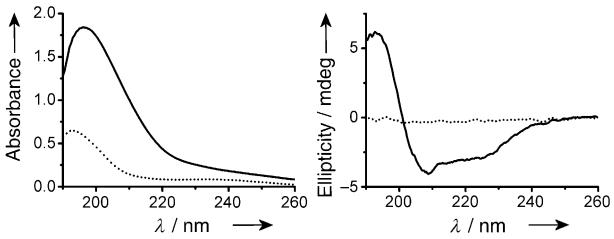

Figure 4. UV (left) and CD (right) spectra obtained for **7** (——) and $\mathbf{P_1}$ (•••••) in the far-UV region.

(MeCN; Figure 4). As expected, for each compound, a UV signal that is characteristic of the peptide bond adsorption (190–250 nm) was observed.

While the CD spectrum of $\mathbf{P_1}$ in this region is featureless as anticipated, that of **7** is dominated by a signal that is characteristic of an α-helix arrangement, with two troughs at 208 nm and 222 nm. It is not clear if this feature results from a regular secondary structure in only a part of the molecule, or if the cluster inclusion leads the entire peptide chain to adopt a helical arrangement. Unfortunately, the limited solubility of **7** in MeCN did not allow us to further investigate this structure by 2D NMR analysis; no peak characteristic of α-helices was observed during such a study in DMSO (which is unsurprising as DMSO is a more competitive solvent than MeCN). While further characterization of the origin of the CD signal observed for **7** is beyond the scope of this communication, its observation nonetheless highlights how the incorporation of a POM cluster into proteins or peptides can result in significant new structural features.

The solution-phase peptide addition approach presented herein is suitable for the grafting of POMs to pre-synthesized peptide chains of different lengths. It demonstrates many positive aspects of our activated precursor approach: the reaction of the NHS ester and the amine is stoichiometric; the precursor can be produced and isolated in large batches; peptides are used in their native form; the reaction conditions are mild and compatible with biological building blocks; little variation in conditions should be required, apart from for the solubility demands of the different peptides. However, much of the power of peptide synthesis is derived from the stepwise modular nature of the synthetic procedure; to be able to incorporate the metal oxide building block directly during the synthesis of a peptide, as conveniently as any other amino acid, would open a range of possibilities, particularly for combinatorial synthesis.

Precursor **2** can be viewed as an unnatural amino acid with an activated C terminus and an Fmoc-protected N terminus, and as such it is a potential building block for SPPS. Clearly, some aspects of the SPPS method required careful consideration, given the sensitivity of the POM to acids, strong bases, and oxidants. The Fmoc strategy was favored for the very mild conditions that can be used for the removal of this protecting group (20% piperidine in DMF), which we recently demonstrated to be compatible with these clusters.[14] Activation of the amino acids by the reaction with a carbodiimide was considered as many POMs are stable to this reagent type,[19] and amidations were recently performed on the TRIS-Mn-Anderson cluster ($[MnMo_6O_{18}((OCH_2)_3CNH_2)_2]^{3-}$) through reactions with anhydrides.[7d] As a solid support, we selected a polyethylene glycol (PEG) grafted polystyrene (PS) resin, which is suitable for a broader range of reactions than standard PS resins because of reduced steric hindrance and its amphiphilic character.[20] Furthermore, previous efforts in our group had suggested that standard PS resins may not be compatible with hybrid clusters. The key point was to select a linker that could be cleaved under appropriate conditions; if this step was unsuccessful, any synthetic attempts would have been pointless. Of the somewhat narrow range of linkers that meet these requirements, we chose trityl linkers, which are easy to handle and can be cleaved under very mild conditions (solution of 1,1,1,3,3,3-hexafluoro-2-propanol (HFIP, 20%) in CH_2Cl_2).[21] A test reaction was carried out to see whether the Mn-Anderson cluster would be stable under these cleavage conditions by treating the TRIS-Mn-Anderson compound with the cleavage solution for one hour. The mass spectrum of the isolated product that was recorded after this treatment did not show any signs of degradation of the metal oxide cluster (by comparison with the spectrum recorded before the treatment; see Figure S25).

Having established that the common cluster core is stable under the SPPS conditions in isolation, we set out to manually synthesize a simple sequence, namely compound **8**, which can be described as H_2N-Val-Leu-**Hyb**-Ala-Val-Leu-Ala-OH, where **Hyb** denotes the hybrid amino acid residue that results from the incorporation of **2** (Figure 5).

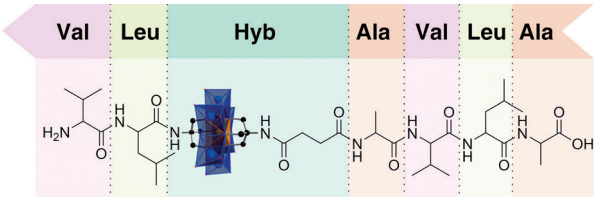

Figure 5. Representation of **8** (H_2N-Val-Leu-**Hyb**-Ala-Val-Leu-Ala-OH) with **Hyb** being the hybrid amino acid resulting from incorporation of **2**. The arrow indicates the order of addition for the amino acids during the Fmoc-based solid-phase synthesis.

For convenience, a Tentagel S resin (based on polymer-bound 4-[poly(oxyethylene)carbamoyl]trityl ester) preloaded with one alanine residue was used; however, this is not necessary as the use of this and similar resins is well-established.[22] For the section that has to be synthesized prior to incorporation of **2** (Ala-Leu-Val), we employed a standard SPPS protocol (5 equiv of each amino acid and double couplings of 30 min each) using *N*,*N*′-diisopropylcarbodiimide (DIC) as the activating agent. The hybrid amino acid **2** was then incorporated by a double coupling, with a first reaction allowed to proceed for one hour, and the second overnight (each using 3 equiv of **2** in the presence of DIPEA). The excess of **2** was collected and reactivated to be re-used in the synthesis of other peptides. In initial tests, the same standard SPPS protocol was then used for the addition of the remaining amino acids; then, the resulting peptide was deprotected (with a 20% piperidine solution in DMF),

cleaved (20% HFIP in CH_2Cl_2), and isolated. Unfortunately, whereas ESI-MS analysis showed that the Mn-Anderson amino acid had been successfully coupled to the growing peptide sequence, only fragments that are consistent with the formation of H_2N-**Hyb**-Ala-Val-Leu-Ala-OH were observed (Figure S26); this implies that the amino acids that had been added subsequently had failed to react with the amine of the **Hyb** residue. It was clear that, following addition of the **Hyb** residue, the coupling procedure needed to be altered to take into account the unusual reactivity of the new residue, which presumably stems from the hindered nature of the TRIS $-NH_2$ group. To overcome this, six times as many equivalents of amino acid were used, and the reactions were left for three hours. The product resulting from this improved method was deprotected, cleaved, isolated, and analyzed by mass spectrometry (Figure S24); the success of the synthesis was confirmed by all major peak envelopes being consistent with the proposed structure of **8**. Further characterization by elemental analysis confirmed the composition of compound **8**.

In summary, we have presented robust complementary synthetic methods for the incorporation of hybrid POMs as inorganic amino acids into a peptide based on the use of NHS ester activated precursors. Furthermore, we have demonstrated that the incorporation of clusters into the main chains of biopolymers, hitherto unprecedented for inorganic clusters, can be achieved both by conjugation with pre-synthesized peptide chains, and by integration into Fmoc solid-phase synthesis as a unique unnatural amino acid. Regardless of the route taken, preliminary studies indicate that the incorporation of POMs into peptide chains can result in structural modifications, suggesting that the nascent field of hybrid peptides may prove valuable in both biomedical and nanoscience research. Furthermore, we believe that the approach outlined for this first solid-supported synthesis of POM hybrids should find considerable application in the wider hybrid-cluster field, laying the foundations for the combinatorial synthesis of hybrids. In further work, we will incorporate this strategy into automated peptide synthesis to systematically explore the structure and function of this new class of hybrid peptides. In particular, we aim to investigate molecular recognition motifs and catalytically active or structurally important proteins in which key amino acids are replaced by POM amino acids.

Experimental Section

For detailed experimental conditions, including SPPS experiments, extensive NMR and MS analysis, elemental analysis, and crystallographic data (compounds **1** and **4**), see the Supplementary Information.

$(C_{16}H_{36}N)_3[MnMo_6O_{24}(C_{12}H_{15}N_2O_5)_2]$ (**1**): 75% yield.
$(C_{16}H_{36}N)_3[MnMo_6O_{24}(C_{12}H_{15}N_2O_5)(C_{19}H_{18}NO_2)]$ (**2**): 88% yield.
$(C_{16}H_{36}N)_3[MnMo_6O_{24}(C_{10}H_{15}N_2O_4)_2]$ (**3**): 63% yield.
$(C_{16}H_{36}N)_3[Mn\ Mo_6O_{24}(C_{17}H_{21}N_2O_4)_2]$ (**4**): 78% yield.
$(C_{16}H_{36}N)_3[MnMo_6O_{24}(C_{26}H_{30}N_3O_5)_2]$ (**5**): 68% yield.
$(C_{16}H_{36}N)_3[MnMo_6O_{24}(C_{35}H_{39}N_4O_6)_2]$ (**6**): 82% yield.
$Na_{0.2}(C_{16}H_{36}N)_{2.8}[MnMo_6O_{24}(C_{74}H_{118}N_{17}O_{29})_2]$ (**7**): 80% yield.
$(C_{16}H_{36}N)_{1.4}H_{1.6}[MnMo_6O_{24}(C_{25}H_{42}N_5O_7)(C_{15}H_{28}N_3O_2)]$ (**8**): 70% yield.

CCDC 976413 (**1**) and 976414 (**4**) contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

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[1] a) M. T. Pope, A. Müller, *Angew. Chem.* **1991**, *103*, 56–70; *Angew. Chem. Int. Ed. Engl.* **1991**, *30*, 34–48; b) D.-L. Long, E. Burkholder, L. Cronin, *Chem. Soc. Rev.* **2007**, *36*, 105–121; c) D.-L. Long, R. Tsunashima, L. Cronin, *Angew. Chem.* **2010**, *122*, 1780–1803; *Angew. Chem. Int. Ed.* **2010**, *49*, 1736–1758.
[2] a) P. Gouzerh, A. Proust, *Chem. Rev.* **1998**, *98*, 77–112; b) A. Proust, R. Thouvenot, P. Gouzerh, *Chem. Commun.* **2008**, 1837–1852; c) A. Dolbecq, E. Dumas, C. R. Mayer, P. Mialane, *Chem. Rev.* **2010**, *110*, 6009–6048.
[3] a) A. Proust, B. Matt, R. Villanneau, G. Guillemot, P. Gouzerh, G. Izzet, *Chem. Soc. Rev.* **2012**, *41*, 7605–7622; b) Y.-F. Song, R. Tsunashima, *Chem. Soc. Rev.* **2012**, *41*, 7384–7402.
[4] a) M. I. Khan, Q. Chen, D. P. Goshorn, H. Hope, S. Parkin, J. Zubieta, *J. Am. Chem. Soc.* **1992**, *114*, 3341–3346; b) A. Müller, J. Meyer, H. Bögge, A. Stammler, A. Botar, *Chem. Eur. J.* **1998**, *4*, 1388–1397; c) H. Zeng, G. R. Newkome, C. L. Hill, *Angew. Chem.* **2000**, *112*, 1841–1844; *Angew. Chem. Int. Ed.* **2000**, *39*, 1771–1774.
[5] B. Hasenknopf, R. Delmont, P. Herson, P. Gouzerh, *Eur. J. Inorg. Chem.* **2002**, 1081–1087.
[6] a) P. R. Marcoux, B. Hasenknopf, J. Vaissermann, P. Gouzerh, *Eur. J. Inorg. Chem.* **2003**, 2406–2412; b) E. F. Wilson, H. N. Miras, M. H. Rosnes, L. Cronin, *Angew. Chem.* **2011**, *123*, 3804–3808; *Angew. Chem. Int. Ed.* **2011**, *50*, 3720–3724; c) M. H. Rosnes, C. Yvon, D.-L. Long, L. Cronin, *Dalton Trans.* **2012**, *41*, 10071–10079.
[7] a) Y.-F. Song, D.-L. Long, L. Cronin, *Angew. Chem.* **2007**, *119*, 3974–3978; *Angew. Chem. Int. Ed.* **2007**, *46*, 3900–3904; b) Y.-F. Song, N. McMillan, D.-L. Long, J. Thiel, Y. Ding, H. Chen, N. Gadegaard, L. Cronin, *Chem. Eur. J.* **2008**, *14*, 2349–2354; c) H.-K. Yang, M.-M. Su, L.-J. Ren, J. Tang, Y.-K. Yan, W.-K. Miao, P. Zheng, W. Wang, *Eur. J. Inorg. Chem.* **2013**, 1381–1389; d) M. Hutin, C. Yvon, J. Yan, A. Macdonell, D.-L. Long, L. Cronin, *CrystEngComm* **2013**, *15*, 4422–4430; e) M. H. Rosnes, C. Musumeci, C. Yvon, A. Macdonell, C. P. Pradeep, C. Sartorio, D.-L. Long, B. Pignataro, L. Cronin, *Small* **2013**, *9*, 2316–2324.
[8] A. L. Boyle, D. N. Woolfson, *Chem. Soc. Rev.* **2011**, *40*, 4295–4306.
[9] T. R. Pearce, K. Shroff, E. Kokkoli, *Adv. Mater.* **2012**, *24*, 3803–3822.
[10] a) H. Shin, S. Jo, A. G. Mikos, *Biomaterials* **2003**, *24*, 4353–4364; b) K. M. Stewart, K. L. Horton, S. O. Kelley, *Org. Biomol. Chem.* **2008**, *6*, 2242–2255.
[11] a) D. N. Woolfson, M. G. Ryadnov, *Curr. Opin. Chem. Biol.* **2006**, *10*, 559–567; b) E. Gazit, *Chem. Soc. Rev.* **2007**, *36*, 1263–1269; c) R. V. Ulijn, A. M. Smith, *Chem. Soc. Rev.* **2008**, *37*, 664–675.
[12] a) S. Bareyt, S. Piligkos, B. Hasenknopf, P. Gouzerh, E. Lacôte, S. Thorimbert, M. Malacria, *Angew. Chem.* **2003**, *115*, 3526–3528; *Angew. Chem. Int. Ed.* **2003**, *42*, 3404–3406; b) S. Bareyt, S. Piligkos, B. Hasenknopf, P. Gouzerh, E. Lacôte, S. Thorimbert, M. Malacria, *J. Am. Chem. Soc.* **2005**, *127*, 6788–6794; c) C. Boglio, K. Micoine, É. Derat, R. Thouvenot, B. Hasenknopf, S. Thorimbert, E. Lacôte, M. Malacria, *J. Am. Chem. Soc.* **2008**, *130*, 4553–4561; d) K. Micoine, B. Hasenknopf, S. Thorimbert,

E. Lacôte, M. Malacria, *Angew. Chem.* **2009**, *121*, 3518 – 3520; *Angew. Chem. Int. Ed.* **2009**, *48*, 3466 – 3468.
[13] a) J. T. Rhule, C. L. Hill, D. A. Judd, R. F. Schinazi, *Chem. Rev.* **1998**, *98*, 327 – 358; b) S. Vanhaecht, G. Absillis, T. N. Parac-Vogt, *Dalton Trans.* **2012**, *41*, 10028 – 10034; c) K. Stroobants, E. Moelants, H. G. T. Ly, P. Proost, K. Bartik, T. N. Parac-Vogt, *Chem. Eur. J.* **2013**, *19*, 2848 – 2858.
[14] C. Yvon, A. Macdonell, S. Buchwald, A. J. Surman, N. Follet, J. Alex, D.-L. Long, L. Cronin, *Chem. Sci.* **2013**, *4*, 3810 – 3817.
[15] a) K. J. Kise, B. E. Bowler, *Inorg. Chem.* **2002**, *41*, 379 – 386; b) J. F. Valliant, K. J. Guenther, A. S. King, P. Morel, P. Schaffer, O. O. Sogbein, K. A. Stephenson, *Coord. Chem. Rev.* **2002**, *232*, 173 – 230; c) L. Barišić, V. Rapić, N. Metzler-Nolte, *Eur. J. Inorg. Chem.* **2006**, 4019 – 4021; d) H. Ishida, Y. Maruyama, M. Kyakuno, Y. Kodera, T. Maeda, S. Oishi, *ChemBioChem* **2006**, *7*, 1567 – 1570.
[16] a) M. Reches, E. Gazit, *Science* **2003**, *300*, 625 – 627; b) P. Tamamis, L. Adler-Abramovich, M. Reches, K. Marshall, P. Sikorski, L. Serpell, E. Gazit, G. Archontis, *Biophys. J.* **2009**, *96*, 5020 – 5029.
[17] a) X. Yan, P. Zhu, J. Li, *Chem. Soc. Rev.* **2010**, *39*, 1877 – 1890; b) A. M. Smith, R. J. Williams, C. Tang, P. Coppo, R. F. Collins, M. L. Turner, A. Saiani, R. V. Ulijn, *Adv. Mater.* **2008**, *20*, 37 – 41.
[18] S. M. Kelly, N. C. Price, *Biochim. Biophys. Acta Protein Struct. Mol. Enzymol.* **1997**, *1338*, 161 – 185.
[19] a) Y.-F. Song, N. McMillan, D.-L. Long, S. Kane, J. Malm, M. O. Riehle, C. P. Pradeep, N. Gadegaard, L. Cronin, *J. Am. Chem. Soc.* **2009**, *131*, 1340 – 1341; b) J. Zhang, P. Yin, J. Hao, F. Xiao, L. Chen, Y. Wei, *Chem. Eur. J.* **2012**, *18*, 13596 – 13599.
[20] H. Becker, H.-W. Lucas, J. Maul, V. N. R. Pillai, H. Anzinger, M. Mutter, *Makromol. Chem. Rapid Commun.* **1982**, *3*, 217 – 223.
[21] R. Bollhagen, M. Schmiedberger, K. Barlos, E. Grell, *J. Chem. Soc. Chem. Commun.* **1994**, 2559 – 2560.
[22] M. Amblard, J.-A. Fehrentz, J. Martinez, G. Subra, *Mol. Biotechnol.* **2006**, *33*, 239 – 254.

