ssPNA templated assembly of oligo(*p*-phenylenevinylene)s†

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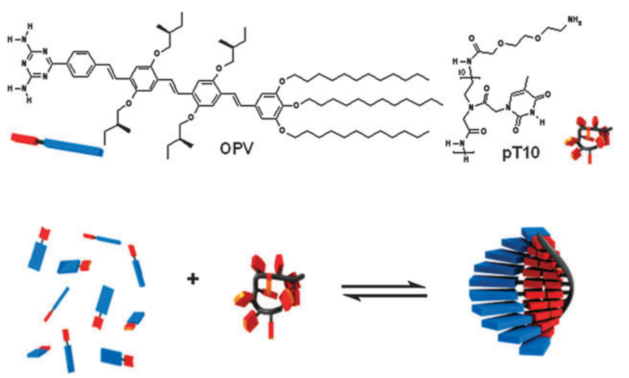
A single-stranded oligothymine peptide nucleic acid (PNA) was used as a template for the assembly of a chiral oligo-(*p*-phenylenevinylene) diaminotriazine derivative (OPV) in methylcyclohexane (MCH) revealing nanostructures in which the size is controlled by the template.

In nature, templates with specific binding sites are used to efficiently form assemblies and polymers with definite size or sequence.[1] This behavior has inspired many researchers to exploit templated polymerization[2,3] as a tool to control the size and sequence of synthetic polymers *via* a 'bottom-up' approach.[4–8] Especially oligonucleotides are interesting building-blocks, since they can be obtained monodisperse, functionalized and used to create predefined nanosized structures *via* sticky-end cohesion.[9]

In a previous study, we showed that the single-stranded desoxyribonucleic acid (ssDNA) oligothymine can act as a template for the assembly of complementary diaminotriazine equipped guest molecules in water.[7] In this construct, the single DNA strand templates a supramolecular strand of chromophores held together by π–π, hydrophobic and hydrogen bond interactions. The efficiency of this templated assembly depends on the host–guest and guest–guest interaction and can be described by a templated assembly model based on a one-dimensional Ising model.[7a] The use of DNA as template requires water as solvent[10] and therefore the variety of guest molecules is limited. In order to broaden the scope of this templated approach to organic solvents, we now report on the use of a single-stranded peptide nucleic acid (ssPNA), consisting of 10 thymine residues (**pT10**,‡Scheme 1), as a template for the assembly of a chiral π-conjugated oligo-(*p*-phenylenevinylene) diaminotriazine derivative[11] (**OPV**, Scheme 1) in MCH. PNA[12] is an achiral and uncharged analogue of DNA in which the phosphate backbone is replaced by an *N*-(2-aminoethyl)glycine backbone, making it soluble in a range of organic solvents. We have previously shown that **OPV** forms hydrogen bonded hexamers that subsequently self-assemble into helical fibers in heptane.[11a] Here, we describe the non-templated self-assembly and **pT10** templated assembly process of **OPV** studied by means of temperature-dependent UV-vis absorption and CD spectroscopy. The assemblies were visualized with atomic force microscopy (AFM).

The synthesis of **OPV**[11a] and **pT10**[13] were performed according to literature procedures. We have first investigated the non-templated self-assembly of **OPV**. In chloroform, **OPV** is molecularly dissolved and has an absorption maxima λ_{max} at 430 nm. In MCH[14] at 323 K, **OPV** (100 μM) is molecularly dissolved since a similar absorption maximum is found. Upon cooling to 263 K, hypochromicity, a red shift of the onset of the absorption, and an absorption maximum shifting from λ_{max} = 430 to 440 nm (Fig. 1a) are observed.[15] Simultaneously, at low temperatures, a positive Cotton effect is observed with a zero-crossing at λ_{z-c} = 434 nm,[16] indicating that **OPV** self-assembles into right-handed helical aggregates, similar as earlier observed in heptane.[11] The non-templated self-assembly process has been studied in more detail by monitoring the UV absorption at λ = 500 nm as a function of temperature at different concentrations. The self-assembly is fully reversible and the observed exponential transition is indicative of a cooperative non-templated self-assembly process.[11,17] By fitting both the concentration- and the temperature-dependent self-assembly data to the cooperative self-assembly model,[17] the enthalpy of binding ($\Delta H_e \approx -75 \pm 8$ kJ mol^{-1}) was determined (Fig. 1e and f).[17] To visualize the **OPV** assemblies, a MCH-solution has been drop-cast onto graphite (HOPG). AFM micrographs show the



Scheme 1 Molecular structures of the host template **pT10** and the guest **OPV** and a schematic representation of ssPNA templated self-assembly (in blue and red OPV, and in black and red the PNA template).

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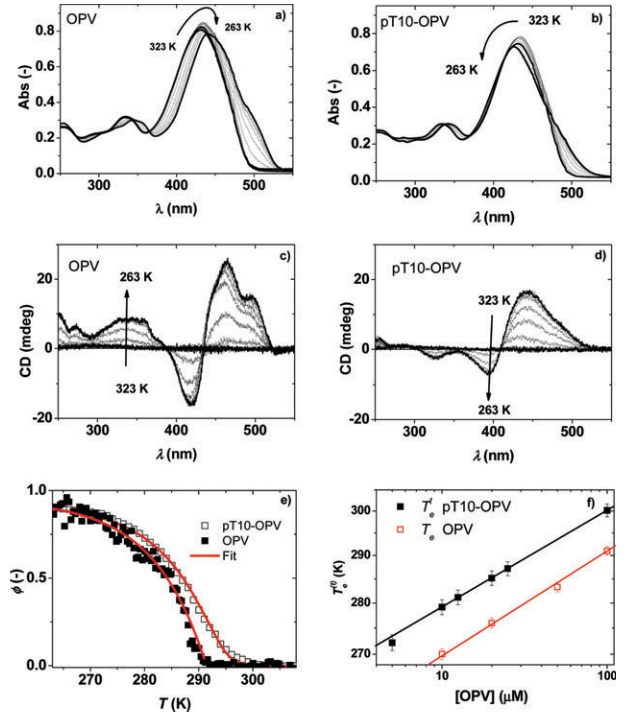

Fig. 1 Absorption and CD spectra at temperatures between 323 and 263 K for **OPV** (a and c, respectively) and **pT10–OPV** (1 : 10) (b and d, respectively) mixtures in MCH. (e) The self-assembled fraction upon cooling of **OPV** and **OPV–pT10** (10 : 1) mixtures and the fits to the cooperative self-assembly model[17] and templated assembly model,[7a] respectively. [**pT10**] = 10 μM, [**OPV**] = 100 μM. (f) T_e and T_e^t (inverted scale) as a function of [**OPV**] (logarithmic scale) for **OPV** and **pT10–OPV** (1 : 10) mixtures.

formation of fibers with a 4–6 nm height (Fig. 2a, c). This height corresponds well with the diameter of the fibers consisting of hexameric H bonded rosettes earlier reported for **OPV** in heptane.[11a]

To investigate the PNA templated assembly of **OPV**, a base-equivalent of **pT10** was added to a 200 μM solution of **OPV** in chloroform at 263 K. No Cotton effect and spectral changes were observed in the **OPV** absorption region indicating that there is no interaction between **OPV** and **pT10**. To increase the

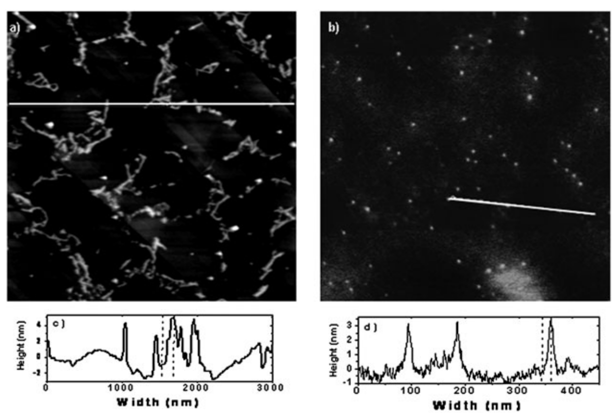

Fig. 2 Atomic force micrographs of (a) **OPV** (3 × 3 μm) and (b) **pT10–OPV** (1 × 1 μm) solutions drop-casted on HOPG at 273 K and the corresponding height cross-sections below. [**pT10**] = 10 μM, [**OPV**] = 100 μM.

host–guest and guest–guest interaction in the templated PNA assembly, MCH was used as a solvent. In this solvent **OPV** itself already forms self-assembled fibers (*vide supra*) which have to be less stable than the proposed **OPV–pT10** constructs. When **OPV** is mixed with a base-equivalent of **pT10** in MCH at 323 K and cooled down to 263 K, hypochromicity is accompanied by a blue shift of λ_{max} to 425 nm and a red shift of the onset (Fig. 1b). Compared to the non-templated **OPV** self-assembly, the **OPV–pT10** mixture has a lower intensity of the Cotton effect (Fig. 1d). Furthermore, the zero-crossing of the Cotton effect λ_{z-c} is 410 nm[16] for the **pT10–OPV** mixture, while for the **OPV**, λ_{z-c} is 434 nm (Fig. 1b, c). This indicates that **OPV** is differently organized when the template **pT10** is present.[18]

The templated assembly process has been studied in more detail by monitoring the UV-vis absorption at λ = 500 nm as a function of temperature at different concentrations and compared to the non-templated self-assembly. The transition temperatures, below which the two types of self-assembly set in, are defined as the *elongation temperature* T_e[17] for non-templated self-assembly and as the *apparent elongation temperature* T_e^t for templated assembly.[7a] For a similar concentration, the T_e^t of the **pT10–OPV** mixtures is higher than the T_e of **OPV** (Fig. 1e and f), showing that the **pT10–OPV** assemblies are more stable than the non-templated self-assemblies of **OPV**. When fitting the temperature-dependent data to the templated self-assembly model as described previously,[7a] an enthalpy of $\Delta H_e^t \approx -90 \pm 10$ kJ mol^{-1}, a guest–guest interaction energy of $\varepsilon = -6.2 \pm 0.5\ kT_p$ was obtained.[19]

The enthalpy values extracted for the templated and non-templated assembly processes suggest that the higher stability of the templated assembly as indicated by the higher melting temperature is due to its larger enthalpy gain. A necessary condition for the predominance of templated assembly over self-assembly is that the free-energy change resulting from the combined effects of host–guest and templated guest–guest interaction molecules is larger than the free-energy gain from the stacking of guest molecules in self-assembly. As a consequence, the presence of the PNA template effectively suppresses the self-assembly of **OPV** unless a large excess of **OPV** is present in the solution and only then when the **pT10** templates are filled.

To visualize the **pT10–OPV** assemblies, an MCH-solution was drop-casted on graphite (HOPG). In contrast to the sample containing only **OPV** (Fig. 2a), the AFM micrographs of the **pT10–OPV** mixture show uniform small particles with a height of 3–4 nm and a deconvoluted width[20] of 5–10 nm (Fig. 2b, d).[7a] The expected dimensions of the **pT10–OPV** complexes are ~4 × 4 × 4 nm (the length of **pT10** and **OPV** are 3.6 and ~4 nm, respectively) and correspond to the size of the objects observed, revealing that the PNA-template controls the size of the **OPV** assemblies.

In conclusion, PNA-templated assemblies have been constructed in MCH of which the size is controlled by the PNA template. This PNA-templated approach can in principle be applied to any functional molecule and makes it possible to construct size-controlled functional nanostructures in organic media.

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Notes and references

‡ **pT10**:[13] MALDI-TOF MS (M = 2823.5) m/z = 2824.6 [M + H$^+$]. For experimental details and general methods see ref. 7*a*. Sample preparation: **pT10** and **OPV** were dissolved in chloroform. After solvent removal and intensive drying, MCH was added to obtain the appropriate concentration and the solution was heated to 333 K to dissolve both components and slowly cooled. For atomic force microscopy, 2 μl of the solution at 273 K was drop-cast on freshly cleaved HOPG and allowed to dry in air.

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