PROTOCOL METADATA

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List of SI:

Supplementary video 1: A left-handed spring ($\phi = 45^\circ$) winds under irradiation with UV light.

Supplementary video 2: A right-handed spring ($\phi = 112^\circ$) unwinds under irradiation with UV light.

Supplementary video 3: A right handed-spring ($\phi = 158^\circ$) displays helix inversion under irradiation with UV light.

Supplementary video 4: Once illumination is stopped, a spring ($\phi = 0^\circ$) recovers its initial shape in ambient light conditions.

Supplementary Figure 1 Cross-section of a liquid crystal polymer film prepared in a commercially available twist cell of nominal thickness 50 μm, observed by scanning electron microscopy. The measured thickness of the film is 43 μm. Scale bar 10 μm.

Supplementary Figure 2 Differential scanning calorimetry of the liquid crystal mixture **3** without photoinitiator. The isotropic to nematic and nematic to crystalline transition are visible respectively at 65.72 and 16.56 ^{o}C.

Supplementary Figure 3 Photo of the dedicated set up for tensile strength measurements.

CFIs: (y/n) no

ALERT: Figure 1 contains stock images from Alamy. Apparently we have a mechanism for paying for these. Hopefully Sarah Bakewell will be able to help; otherwise contact the Editorial Assistant / Art Editor for Nature Chemistry, as they have some experience with this.

Web-only material (Accept)

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NPG ONTOLOGY TERMS

Subjects	Physical sciences/Materials science/Biomaterials/Bioinspired materials Physical sciences/Chemistry/Photochemistry Physical sciences / Chemistry / Materials chemistry / Soft materials / Liquid crystals Biological sciences / Biological techniques / Biophysical methods
Techniques	

PROTOCOLS KEYWORDS

Molecular motors and switches, Liquid crystal polymer networks, Smart materials, Soft robotics

EDITORIAL SUMMARY

Materials that change shape on illumination can be used in soft robotics and artificial muscles. This protocol describes how to make photoresponsive polymer springs using liquid crystals, a chiral dopant and a photo-switch derived from azobenzene.

TWEET

Preparation of biomimetic photo-responsive polymer springs

COVER TEASER

Photo-responsive polymer springs

Preparation of biomimetic photo-responsive polymer springs

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Abstract

This protocol describes the preparation of polymer springs that twist under irradiation with light, in a manner that mimics how plant tendrils twist and turn under the effect of differential expansion in different sections of the plant. The artificial springs are typically 1 mm in width, 50 µm in thickness and up to 10 ~~cm~~ mm in length, their length being limited by cell dimensions only. They are made from polymer networks that keep memory of a liquid crystalline order, and in which an azobenzene derivative is introduced covalently as a molecular photo-switch. This liquid crystal polymer is prepared by irradiation of a twist cell filled with a mixture of shape-persistent liquid crystals, liquid crystal having reactive end groups, molecular photo-switches, some chiral dopant and a small amount of photo-initiator. This cell is assembled out of two glass slides separated by a spacer and covered by a thin film of polyimide that was rubbed along the long axis of the cell for the bottom slide, and along the short axis of the

cell for the top slide. Once the cell is filled by capillarity, photo-polymerization takes place at 48 °C and takes approximately 1.5 h. The product is a photo-responsive liquid crystal polymer network that is characterised by optical microscopy, scanning electron microscopy and tensile strength measurements. The film is post-cured overnight at 60°C. Removing the resulting soft polymer film and cutting out the desired spring-like shape takes ~45 min. The springs operate at ambient temperature, by mimicking the orthogonal contraction mechanism that is at the origin of plant coiling. They are shape shifting under irradiation with ultraviolet light and can be pre-programmed to either wind or unwind, as encoded in their geometry. Once illumination is stopped, the springs return to their initial shape in ambient light conditions. ~~Irradiation with visible light accelerates the shape reversion.~~

INTRODUCTION

Incorporating molecular photo-switches in soft matter promises a route to smart materials that change their shape under illumination with light.[1,2,3] Engineering of large, complex and versatile shape transformations would allow the delivery of future materials that can produce work and eventually powerful movement, with potential applications in soft robotics and artificial muscles. The design, production and operation of functional smart photo-actuators thus constitute a major thrust of contemporary materials research, and biological materials have provided inspiration for relevant engineering paradigms. In nature, twisting and turning behaviours are widely used macroscale deformation modes that allow plants to achieve complex mechanical functions (Figure 1). Cucumber tendrils coil and wind *via* asymmetric contraction of an internal fibre ribbon of specialized cells,[4] seed pods open *via* a mechanism creating helical chirality,[5] stems twine by producing a helical tweezing force[6] and seeds bury into soil, as their helical bristles wind and unwind with daily cycles that change ambient humidity.[7] Understanding and learning how to engineer systems displaying these types of highly controlled helical shape transformations would provide the capacity and incentive to design active materials with specific helix-based functionality.

Various strategies have been developed to mimic the shape shifting of biological springs, primarily by transforming bi-dimensional polymer-based films into three-dimensional helix-based structures. Approaches based on polymer hydrogels provide springs that are pre-programmed to respond to changes in temperature or swell in response to an increase in humidity. Studart *et al.* have developed a strategy based on the temperature-dependent hydration of gelatine, alginate, or other polymers that were reinforced with inorganic platelets having varying orientations within the material.[8] The orientation of these reinforcing elements determines the directionality of swelling and thus promotes a winding motion. The strong reversible hygroscopicity of agarose-based hydrogels has been combined with anisotropic networks of glass fibers, and has shown potential for fast spiraling and twisting motion driven by changes in humidity, at room temperature.[9,10] Other hydrogel springs responding with deformation to changes in temperature were developed by implementing alternating bar-like regions of different hydrogels with differential swelling.[11] In humidity-responsive materials, asymmetry in swelling can also be programmed by introducing salts selectively, on one side of the film only.[12] Actuation mechanisms based on swelling make sense in terms of bio-mimetic character, because many plants move in response to the daily cycle of humidity. However the use of humidity as a trigger remains a limitation in terms of applications and de-swelling procedures can compromise the structural integrity of the material.

Light as a stimulus is compatible with a wide range of condensed phases, the kinetics of the photo-response can usually be controlled by light intensity and moreover light allows both spatial and temporal control over soft matter.[3] Here we report a bio-inspired design strategy based on the use of liquid crystal polymer networks that are doped with molecular photo-switches. Specifically, we encode orthogonal deformation modes in thin films of liquid crystal polymer networks, and combine this asymmetric deformation with a differential stiffness that runs through the thickness of the film, in order to form spring-like materials that respond to irradiation with light. We optimize the mechanical properties of the springs by adjusting the composition of the pre-polymer liquid crystalline mixture (Figure 2). The shape and photo-response of the springs depends on a large number of factors, including not only the composition of the polymer network but also the preparation procedure and the thickness of the film. This high sensitivity to variations in procedure can constitute a limitation that the present protocol is meant to address. We demonstrate that ultraviolet light induces structural changes at the molecular level, and how these molecular transformations are converted into large amplitude shape shifting. Once illumination with ultraviolet light is stopped, the springs return to their initial shape in ambient light conditions. Irradiation with visible light accelerates the shape reversion. This approach provides a versatile protocol as a large number of shapes can be formed in one single sheet of materials.

Development of the protocol

We have started a research program that aims to mimic the mechanical movements seen in living systems using artificial responsive materials. At the heart of this research is the idea that the helical designs seen in nature are ideal for translating and amplifying deformation modes in artificial systems as well. We recently reported synthetic systems capable of translating molecular movement across scale lengths into macroscopic rotational movements that could serve as the basis of light-driven functional materials.[13] We have demonstrated that a variety of complex deformations are possible including winding, unwinding and helix inversion and that devices can be constructed to move macroscopic objects. Our bio-inspired springs are based on liquid crystal polymer networks that incorporate azobenzene photo-switches covalently. When azobenzene photo-switches are in their stable *trans*-form, their rod-like shape does not disrupt the liquid crystalline order, the molecules forming the liquid crystal orient in one direction preferentially, and the polymer chains are elongated. When subjected to light, photo-isomerisation of the switches into their bent *cis*-form disrupts the liquid crystalline order, and the polymer chains recover a random coiled conformation in an entropically favored process. The individual changes in the shape of the polymer chains translate to the macroscopic level, and overall this mechanism induces an anisotropic deformation of the material: light-induced shrinkage occurs primarily along the direction of preferential alignment of the liquid crystal molecules (called the director).[14]

We have shown that the mechanical properties of light-responsive liquid crystal polymer springs can be engineered by adjusting the following parameters:

i) *Their aptitude to deform without breaking* relates to their cross-linking density, that itself relates to the ratios between acrylate and diacrylate liquid crystals forming the network,[15] and to the proportion of non-polymerisable molecules in the mixture also.

ii) *A gradient of stiffness through the thickness of the film* is introduced during photo-polymerisation. This results in the two surfaces of the film having different mechanical properties – the less deformable surface will be found on the outside of the helically shaped ribbon preferentially, once the polymer spring is formed;

iii) *The proportion of switches* in the liquid crystal polymer network must be sufficiently low for light to penetrate the thin film, but their concentration should be sufficient to efficiently disrupt the order of the polymeric material;

iv) *The cutting direction* fixes the molecular orientation within the ribbon. Thus, in combination with the materials chirality and stiffness across the sample's thickness, the cutting direction determines the geometry and the photo-actuation properties of the springs (Figure 3).

Experimental design

Our design is based on the use of liquid crystal polymer networks that are characterised by their anisotropy, a feature that is relevant because in nature, plants use anisotropic expansion as a mechanism of movement. Specifically, plants expand perpendicularly to the orientation of fibrils when tissue swells in response to higher humidity.[16] Helix-based movement in plants, whether explosive release of strain or slow coiling motion, is a complex mechanism, the specifics are still under debate, but the deformation mechanism is usually based on a common principle: the orientation of cellulose microfibrils varies across the thickness of the plant, so when the tissue swells, it expands in different directions, creating a curvature.[17,18,19] In pinecones and in the seedpod of orchid trees, cellulose microfibrils are oriented perpendicularly at each side of the plant tissue.[8] The asymmetric contraction induced by differential orientation can be further influenced by differential lignification of the cells, that introduces gradients in stiffness, the stiffer region showing propensity to locate at the outer side of the curvature.[20]

A twist cell to encode orthogonal deformation modes We take advantage of the high sensitivity of liquid crystals to boundary conditions in order to reproduce differential deformation modes artificially. We use a cell constituted by two glass slides, each of them covered with a thin polymer film that promotes the alignment of the liquid crystal in a specific direction: at the top of the cell the rod-like molecules are preferentially oriented along the short axis of the cell, whereas at the bottom of the cell the molecules are preferentially oriented along its long axis. These boundary conditions promote the formation of a 90° twist through the thickness of the cell and the cell is referred to as a twist cell. The use of a twist cell to create a twist across the thickness of the cell is key in preparing spring-like actuators (Figure 4).[21,22,23] A small amount of chiral dopant is used to make sure that the twist has the same handedness over the whole sample and thus to secure cooperative effects in the film. In principle any chiral dopant can be used, provided it is miscible with the liquid crystal and preferably it should have a moderate helical twisting power (other groups have reported the use of dopants such as S-811[21] or R-1011[24]). The chiral dopant promotes a twist that is proportional to its concentration at low concentrations. By choosing the right concentration the twist is adjusted to match four times the thickness of the cell.

Origin of asymmetry Three sources of asymmetry contribute to shape generation and actuation in this experimental design: i) the angular offset that is introduced by cutting the ribbon at a specific angle ϕ ii) the handedness of the twist in the cell and iii) a gradient in cross-linking density of the liquid crystal network, that runs across the thickness of the film.

The gradient is formed during cross-polymerisation, by irradiating the liquid crystal in the twist cell, from one side of the cell exclusively.[25] After cross-polymerisation, the cell is opened. The thickness of these ribbons corresponds well to the nominal thickness of the cell, as estimated by using scanning electron microscopy images of their cross-section (Supplementary Figure 1). Shrinkage of about 15% in the thickness of the film was observed upon crosslinking. Once ribbons are cut out of the liquid crystal polymer film, they spontaneously curl into springs as a result of the orthogonal shrinking that occurs during cross-polymerisation.[26] In fact we can produce a variety of chiral and achiral shapes, depending on the offset angle ϕ, defined as the angle between the average orientation of the molecules at midplane, and the cutting direction (Figure 3).

Composition of the liquid crystal polymer network The composition of the liquid crystal must be adjusted to tune the properties of the springs in terms of their stiffness, shape and response to UV light (Table 1). The photo-switch Azo-**1** is incorporated into the liquid crystal polymer network in its *trans*-form, via two reactive end groups. The monomers used in polymerisation have either one reactive end group (C6BP and C6BPN), or they act as cross-linkers with two reactive end groups (C6M, Figure 2). It has been observed that incorporating a high ratio of cross-linking units in the network reduces the *trans* – to – *cis* photo-conversion and thus the amplitude of photo-induced deformation.[15] Moreover, a high ratio of cross-linkers promotes the formation of densely cross-linked polymer networks that are less deformable, and less prone to forming springs. Therefore we kept the proportion of cross-linker in the network as low as possible, typically lower than in previous works, and introduced a relatively long spacer in the structure of C6M (Figure 2). Some compositions provided fragile and breakable liquid crystal polymer films. We used E7, a well-known nematic liquid crystal that is a mixture of cyano-biphenyl derivatives (Figure 2), in order to promote the formation of a loosely-crosslinked and thus flexible polymer network. Eventually the optimal composition we designed is mixture **3**.[13] Preparation of cross-linkable liquid crystal **3** is described in the procedure section. Differential scanning calorimetry of mixture **3** (without photo-initiator) shows a classical liquid crystalline behaviour, with a nematic to isotropic transition occurring at 66 °C (Supplementary Figure 2).

In the absence of Azo-**1**, springs are formed but no photo-response was ever observed at room temperature. The springs obtained from mixture **2** are stiff, and also show no photo-actuation. Adding E7 and adjusting the ratio between these non-polymerizable molecules and the reactive monomers results in a dramatic improvement in the quality of the curled and twisted shapes obtained initially, and in their photo-responsive properties also (mixture **3**). Lowering the concentration of photo-switch decreases the photo-reactivity of the springs (mixture **4**). Increasing the concentration of Azo-**1** to values higher than 10 wt% does not improve the photo-response of the material because of the large absorption of light by the polymeric network (not shown here).

No.	**C6M (wt %)**	**C6BP (wt %)**	**C6BPN (wt %)**	**E7 (wt %)**	**S-811 (wt %)**	**Azo-1 (wt %)**	**Shape**	**UV actuation**
1	19.07	28.60	9.54	41.75	0.04	-	Curled	None
2	32.49	48.73	16.24	-	0.04	2.00	Flat	None
3	21.18	31.78	10.59	25.41	0.04	10.00	Curled	Yes
4	22.37	33.56	11.19	26.84	0.04	5.00	Flat	Low

Tab 1 | Shape and photo-response of liquid crystal polymer springs with different compositions. Each of these compositions also incorporates 1 wt% of the photo-initiator Irgacure 819.

The cutting angle determines the shape and photo-mechanical properties Depending on the angle at which the ribbons are cut, different shapes are formed, including chiral shapes such as left-handed or right-handed springs, and for some cutting directions, non-chiral ribbons (flat ribbons or open rings) are formed also. When the chiral dopant S-811 is replaced by its enantiomer R-811, the shape of the ribbons are mirror images. For example the left-handed helix cut for S-811 with $\phi = 33^\circ$ is the enantiomer of a R-811 ribbon cut at $\phi = 147^\circ$ (right-handed helix with the same pitch as the precedent helix).[13] Upon irradiation with UV light three different actuation modes are observed: winding ($\phi = 45^\circ$, Supplementary Video 1), unwinding ($\phi = 112^\circ$ Supplementary Video 2) and helix inversion ($\phi = 158^\circ$, Supplementary Video 3). The actuation modes are encoded by design in the initial shape of each ribbon. For example, a ribbon cut at $\phi = 45^\mathrm{o}$ will undergo elongation of its outside surface and shrinkage of its inside surface, leading to an overall twisting motion upon appropriate irradiation with light (Figure 5).

Design of the molecular switch The best performing photo-switches will be those that absorb light efficiently, are fast, and undergo large geometrical changes that will efficiently generate disorder in the liquid crystal network.[27] Currently, azobenzene photo-switches are used primarily to actuate liquid crystal polymer networks because in their bent *cis*-form, they disrupt the liquid crystalline order effectively. Different variations in the connectivity of the photo-switches have been explored, and have suggested that switches used as cross-linkers are more efficient than switches that are incorporated as dangling bonds. The length of the spacers separating the rigid core of the liquid crystal molecules from the reactive end-groups, is known to influence the coupling between the photo-isomerisation of the switches and eventually the macroscopic deformation of the liquid crystal polymer network:[28] The mechanical response of the system is enhanced if the spacers are longer, up to a threshold length value. Further, the activation wavelength of the springs can be adjusted by molecular engineering of the azobenzene photo-switches.[29,30]

Mechanical properties of the biomimetic springs The stiffness of the springs was measured with a custom-made set-up, where the spring is clamped on one side and pulled on the other side to force its elongation (Supplementary Figure 3). Here we report the mechanical properties of a spring cut at $\phi = 45^\circ$. Drawing the pulling force with respect to elongation of the spring yields the curve shown Figure 6. For a specific elongation, the value of the stiffness corresponds to the tangent to this curve. Interestingly, our results show that the stiffness of the spring increases with the force that is applied to it, *i.e.* it behaves as a non-linear spring, as observed in vertebrate muscle fibers.[31] The use of non-linear springs is particularly attractive for robotic applications in human environments where complex stiffness adjustment allows for a variety of tasks that are hardly accessible to conventional linear springs. Flat ribbons proved to be extremely resilient, capable of holding up to 3000 times their weight and a stress up to 0.9 MPa.

Shape recovery of the biomimetic springs When illumination with ultraviolet light stops, the springs relax back to their original shape ($\phi = 0^\circ$**,** Movie 4). The kinetics of the shape recovery are determined by a balance between the elastic properties of the polymer network, the rate of relaxation of the *cis*- azobenzene, and the coupling of the photo-switch to its environment. Typically, the half lifetime for relaxation of Azo-**1** is about 15 min in ambient illumination conditions, and a few hours in the dark. This relatively slow process can be accelerated by irradiation with intense visible light. Up to ten cycles of alternate ultraviolet and visible irradiation can be performed, without evidence of degradation of the photo-mechanical properties.

MATERIALS

REAGENTS

- C6BP monomer : 4-methoxybenzoic acid 4-(6-acyloyloxy-hexyloxy)phenyl ester 97% (Synthon Chemicals cat. No. ST03866) (**Fig. 2**)
- C6BPN monomer : 4[4[6-acyloxyhex-1-yl)oxyphenyl]carboxy-benzonitril 97% (Synthon Chemicals cat. No. ST02760) (**Fig. 2**)
- C6M monomer : 1,4-bis[4-(6-acyloyloxyhexyloxylbenzoyloxy] 97% (Synthon Chemicals cat. No. ST00975) (**Fig. 2**)
- E7 liquid crystal used as received (Merck cat. T153-8927) (**Fig. 2**)
- S-811 chiral dopant : *S*-octan-2-yl 4-((4-(hexyloxy)benzoyl)oxy)benzoate (Merck cat. No. 111649) (**Fig. 2**)
- Azo-**1**. Photo-responsive Azo-**1** was synthesized by following reported procedures for the formation of an azobenzene[32] and its connection to acrylate linkers.[33] (**Fig. 2**).
- Photoinitiator: phenylbis(2,4,6-trimethylbenzoyl)phosphine oxide, 97.0% (Irgacure 819, Sigma-Aldrich cat. no. 511447) **! CAUTION** The reagent is sensitive to visible light and exposure to any light source should be avoided.
- Dichloromethane HPLC grade (DCM, Sigma-Aldrich, cat. no. 270997)
- Nitrogen gas
- Liquid nitrogen **! CAUTION** Liquid nitrogen should be handled with cryogenic gloves in a well-ventilated environment.

EQUIPMENT

- Balances
- Utensils
- Hot plate Präzitherm, Harry Gestigkeit GMBH
- Magnetic stirring rod (7 mm x 2 mm)
- Metallic tip (lancet dissecting needle)
- Edmund MI-150 high-intensity illuminator (≈ 145 mW·cm^{-2})
- Cut-off filter (λ ≥ 420 nm) Edmund Optics, 425 nm OD 2 Longpass filter 50 mm diameter.
- Oven
- Dewar
- Hönle blue point light-emitting-diode (λ = 365 nm, 38.62 mW·cm^{-2} corresponding to 10 % of the maximal intensity was used for all experiments)
- Dino-Lite Pro AM4113T USB camera (x29 magnification)
- Glass slides
- Polarized optical microscope Olympus BX-51P coupled to a digital camera Olympus DP73.

Consumables

- Twist cell with a nominal 50 μm gap, ITO-covered, 10 x 5 mm, 90° (E.H.C. Co., Ltd. cat. No. KSRS-50/D607P1NSS). The cell walls are covered with a LX-1400 (Hitachi-Kasei) polyimide layer of thickness within 20 nm. This alignment layer is oriented by rubbing with the following specifications: a polyester fibre of 8mm length, with a 58 mm roll diameter, 600 rpm rotational speed, 30 $mm \cdot s^{-1}$ stage speed, 3 times round trip frequency.

- Planar cell with a nominal 50 μm gap, ITO-covered, 10 x 5 mm, 180° (E.H.C. Co., Ltd. cat. No. KSRP50/B107P1NSS). The cell walls are treated as for the twist cell.
- Glass vials
- Pasteur pipettes
- Aluminium paper

PROCEDURE

Preparation of liquid crystal polymer thin films TIMING ● 1 day

1. Eliminate possible dust in vials and Pasteur pipettes by flushing with a nitrogen stream before use, as dust may generate defects in the liquid crystal films.

2. Weigh the solid components that are not sensitive to irradiation with visible light: 25.42 mg of **C6M** (21.18 %wt), 38.13 mg of **C6BP** (31.78 %wt) and 12.71 mg of **C6BPN** (10.59 %wt), in three separate brown vials and 0.05 mg of chiral dopant S-811 (0.04 %wt) in another brown glass vial.

<CRITICAL STEP> To avoid composition errors involving quantities of chiral dopant S-811 in the transferring process, the vial containing S-811 is used as mixing vial, so that all the other components are transferred to this vial.

3. Weigh 30.50 mg of liquid crystal E7 using a pipette, in a separate vial.

4. Weigh photosensitive component: 12.00 mg of photo-switchable monomer Azo-**1** (10.00 %wt), in a separate brown glass vial to prevent photo-degradation.

5. Dissolve each component in a minimum volume of dichloromethane and transfer to the vial containing S-811 using pipettes. Rinse each vial three times with dichloromethane (the final volume is ≈ 3 mL.)

6. Evaporate the dichloromethane by using a steam of low-pressure nitrogen for one hour. During this step the vial is on a hotplate heated at 60°C and the mixture is protected from ambient light with aluminium foil. After all dichloromethane has evaporated, the components that remain in the flask compose a liquid crystal.

<CRITICAL STEP> Using a strong nitrogen stream tends to yield non-homogeneous liquid crystalline mixtures (possibly associated with the crystallization of some monomers).

7. Weigh 1.20 mg of photoinitiator (Irgacure 819, 1.00 %wt) in a separate brown glass vial.

<CRITICAL STEP> Manipulations involving the photo-initiator have to be performed in the dark.

8. Dissolve the photoinitiator in a minimum volume of dichloromethane and add to the liquid crystal prepared in step 6 (the final volume is ~1 mL).

<CRITICAL STEP> The photoinitiator is added at this later stage to reduce its exposure to heat so as to minimize thermal degradation.

9. Evaporate the dichloromethane using a low nitrogen stream at 60 ºC for one hour.

10. Stop the nitrogen stream and heat to 80 ºC for 30 minutes. At this point the mixture should be transparent to the eye, indicating that it is in the isotropic state). The liquid is then stirred with a magnetic rod for homogeneity. After mixing, the temperature is decreased to 48 ºC gradually by setting the hotplate at such temperature.

<CRITICAL STEP> Cooling down the temperature too quickly can cause inhomogeneity in the liquid crystal mixture.

11. Label one side of the cell by attaching a small adhesive paper to the edge. This is used to label which side of the cell promotes alignment along the long axis as this side is used as the reference to measure ϕ.

12. Wrap the cell and a metallic tip with aluminium foil and warm them up to 60 ºC by placing them in the oven for about one hour.

13. Unwrap the pre-warmed cell and place it on a hot plate preheated to 48 ºC. Place a drop of liquid crystal mixture on one of the two side openings of the cell with the aid of the pre-warmed metallic tip. The cell will be filled by capillarity action, but the hot tip helps to fill the cell efficiently. Proceed until the cell is filled completely (Figure 7). The time required to fill the cell with the mixture will depend on the nominal gap of the cell, the viscosity of the mixture as well as the filling temperature. In the case of the liquid crystal mixture described here, which contains azo switch-**1**, and a 12 mm x 19 mm x 50 µm cell, the filling time is approximately 15 min.

PAUSE POINT The rest of the liquid crystal mixture can be stored in the oven at 48 – 60 ºC for about a week without any noticeable changes in properties.

<CRITICAL STEP> The use of a cold metallic tip causes partial crystallization of the liquid crystal on the tip while filling the cell. The metallic tip should be pre-warmed before use.

14. Protect the cell from light by using aluminium foil and leave it at 48 ºC for 1 h. The liquid crystal will organize into the desired twist-configuration spontaneously.

15. Fix the cell to the top of a hot plate using scotch-tape. Note whether the marked face of the cell is on top (light-exposed face) or bottom (non-exposed face). The gradient formed during photo-polymerisation will influence the photo-actuation modes of the ribbons.

16. The entire set-up is covered with a box or with aluminium foil.

17. Perform photo-polymerisation by irradiating the top of the cell with visible light. Ensure that the light beam covers the entire surface of the cell. Illuminate for 90 min. (Figure 8)

<CRITICAL STEP> It is critical to place a UV-cut off filter (λ ≥ 420 nm) between the light source (Edmund MI-150 lamp high-intensity illuminator) and the cell as this minimizes *trans*-to-*cis* isomerisation of Azo-**1** during polymerisation.

! CAUTION Wear suitable eye-protection while performing the irradiation with light.

<CRITICAL STEP> If the light beam does not cover the entire surface of the film, an inhomogeneous film will be formed.

18. After photo-polymerisation, wrap the twist cell with aluminium foil and place it in an oven preheated to 60 ºC overnight for post-polymerisation curing.

Preparation of ribbons TIMING ●30-45 minutes

19. Fill a small Dewar with liquid nitrogen.

! CAUTION Wear suitable gloves and eye-protection when using liquid nitrogen. Use only approved unsealed containers.

20. Soak the four corners of the cell, where the cell is glued, in liquid nitrogen. Freezing disrupts the glue and facilitates opening the cell.

21. Dry the soaked cells with tissue paper and gently open the cell using a scalpel (Figure 9).

▲CRITICAL STEP The opening of the cell by using a scalpel can be potentially dangerous, use caution not to cut yourself, or to cause damage to the film.

<CRITICAL STEP> If you have difficulties opening the cell, refreeze the side you wish to open in liquid nitrogen, and try again.

22. After immersion into liquid nitrogen, the adhesive tape used to reference the alignment directions and mark which side of the film was exposed to visible light (the so-called "top-side"), often detach. Re-attach the markers now.

23. Repeat step ~~19-20~~20-22 on the other side of the cell to complete the procedure and achieve full opening of the cell (Figure 9, right). Once the cell is open fully, the film is sitting on one of the glass slides still.

24. Use a cutting board displaying different angles and fix the glass slide supporting the film onto the cutting board with scotch-tape. The cutting angle ϕ is defined here as the angle between the orientation of the molecules at mid-plane and the cutting direction, when the top of the film (its side that was exposed to irradiation with light), is facing up (Figure 3).

▲CRITICAL STEP If the liquid crystal polymer film sticks to the upper glass slide instead of sticking to the bottom glass slide, then instead of using the normal cutting angle ϕ, the "supplementary angle" $\phi_s = \pi - \phi$ should be used to predict the geometry and photo-response of the spring.

25. Place a metallic mask along the desired cutting direction and cut the ribbon using a razor blade (Figure 3). Typically the preparation of a ribbon involves four cuts, two for the length and two for the width of the ribbon. It is usually possible to cut multiple ribbons out of a single film.

26. Use thin tweezers to carefully remove the newly cut ribbon from the rest of the liquid crystal polymer film.

Photo-actuation of ribbons TIMING ●30-45 minutes

27. Prepare a box that is not transparent to UV light. Inside place the camera and the UV lamp. Use tweezers to hold the photo-responsive ribbon.

! CAUTION UV light is harmful to the eyes and skin, make sure that the box is sealed before UV irradiation starts.

28. Focus the camera on the ribbon.

29. Make sure that the light beam is placed so that the entire ribbon can be illuminated.

<CRITICAL STEP> use the same distance between the ribbon and the lamp for each experiment.

30. Set parameters of the UV irradiation such as power of the lamp and irradiation time (here we use 10% of the lamp's power and a 2 min irradiation time).

31. Record a video for the photo-actuation of the ribbon.

TROUBLESHOOTING

Occasionally the liquid crystal polymer films can display some low quality areas, where crystallisation occurs over time (Step 14). These defect zones are likely cause by dust particles or other impurities. Start the procedure again after double-checking that all consumables and chemicals are clean. If the polymer film undergoes complete crystallization, increase the polymerization temperature (50 °C). Crystallization can occasionally occur upon minor variations in the proportions of reagents.

The film is occasionally too fragile, making it difficult to remove the ribbon from the cell wall. This can originate in the polymerisation of the film being incomplete. The procedure can then be adapted by photo-polymerising with a stronger lamp or by irradiating for a few minutes longer (Step 17).

TIMING

Steps 1-17: Preparation of liquid crystal polymer thin films TIMING ● 1 day

Steps 18-25: Preparation of ribbons TIMING ●30-45 minutes

Steps 26-30: Photo-actuation of ribbons TIMING ●30-45 minutes

Anticipated results

The reported procedure yields distinct morphology and photo-actuation of geometrical ribbons as reported in Figure 10. Supplementary videos 1-4 show winding of a polymer spring cut at $\phi = 45^\circ$ (Supplementary Video 1), unwinding of a ribbon cut at $\phi = 112^\circ$ (Supplementary Video 2) and helix inversion of a ribbon cut at $\phi = 158^\circ$ (Supplementary Video 3) upon irradiation with UV light.

Note : Any Supplementary Information and Source Data files are available in the online version of the paper.

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AUTHOR CONTRIBUTION N.K. and S.P.F initiated the project and designed the research. S.I., E.V., F.L. and S.J.A. conducted the experiments and analysed the data. F.L. conducted the tensile strength measurements. N.K., S.P.F., E.V. and F.L. wrote the manuscript and all authors contributed to discussing the results and the manuscript at all stages.

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References

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1 Finkelmann, H., Nishikawa, E., Pereira, G. G. & Warner, M. A. New opto-mechanical effect in solids. *Phys. Rev. Lett*. **87**, 015501 (2001).

2 Yu, Y., Nakano, M. & Ikeda, T. Directed bending of a polymer film by light. *Nature* **425**, 145 (2003).

3 White, T. & Broer, D. J. Programmable and adaptive mechanics with liquid crystal polymer networks and elastomers. *Nature Mater.* **14**, 1087 – 1098 (2015)

4 Wang, J.-S *et al*. Hierarchical chirality transfer in the growth of the *Towel Gourd* tendrils. *Sci. Rep.* **3**, 3102 (2013).

5 Forterre, Y. & Dumais, J. Generating helices in nature. *Science* **333**, 1715 - 1716 (2011).

6 Isnard, S. & Silk, W. K. Moving with climbing plants from Charles Darwin’s time into the 21st century. *Am. J. Bot*. **96**, 1205 – 1221 (2009).

7 Evangelista, D., Hotton, S. & Dumais, J. The mechanics of explosive dispersal and self-burial in the seeds of filaree erodium cicutarium. *J. Exp. Biol.* **214**, 521 – 529 (2011).

8 Erb, R. M., Sander, J. S., Grisch, R. & Studart, A. R. Self-shaping composites with programmable bioinspired microstructures. *Nat. Commun.* 4:1712 (2013).

9 Zhang, L. & Naumov, P. Light- and humidity-induced motion of an acidochromic film. *Angew. Chem. Int. Ed.* **54**, 8642 – 8647 (2015).

10 Zhang, L., Chizhik, S., Wen, Y. & Naumov, P. Directed motility of hygroresponsive biomimetic actuators *Adv. Funct. Mater.* **26**, 1040-1053 (2016).

11 Wu, Z. L. *et al.* Three-dimensional shape transformations of hydrogel sheets induced by small-scale modulation of internal stresses. *Nat. Commun.* **4**, 4:1586 (2013).

12 de Haan, L.T *et al.* Humidity-responsive liquid crystalline polymer actuators with an asymmetry in the molecular trigger that bend, fold, and curl. *J. Am. Chem. Soc.* **136**, 10585-10588 (2014).

13 Iamsaard, S. *et al.* Conversion of light into macroscopic helical motion. *Nature Chem.* **6**, 229 – 235 (2014)

14 Harris, K. D. *et al.* Large amplitude light-induced motion in high elastic modulus polymer actuators. *J. Mater. Chem.* **15**, 5043 - 5048 (2005).

15 Yu, Y., Nakano, M, Shishido, A., Shiono, T. & Ikeda, T. Effect of cross-linking density on photoinduced bending behavior of oriented liquid-crystalline network films containing azobenzene. *Chem. Mater.* **16**, 1637-1643 (2004).

16 Dumais, J. & Forterre, Y. Vegetable dynamics: the role of water in plant movements. *Annu. Rev. Fluid Mech.* **44**, 453 – 478 (2012).

17 Witztum, A. & Schulgasser, K. The mechanics of seed expulsion in acanthaceae. *J. Theor. Biol.* **176**, 531 – 542 (1995)

18 Dawson, C., Vincent, J. F. V. & Roca, A. M. How pine cone opens. *Nature* **390**, 668 (1997)

19 Elbaum, R., Zaltzman, L., Burgert, I. & Fratzl, P. The role of wheat awns in the seed dispersal unit. *Science* **316**, 884 – 886 (2007)

20 Studart, A. R. & Erb, R. M. Bioinspired materials that self-shape through programmed microstructures. *Soft Matter* **10**, 1284 – 1294 (2014).

21 Sawa, Y. *et al.* Shape selection of twist-nematic-elastomer ribbons. *Proc. Natl. Acad. Sci.* USA **108**, 6364 - 6368 (2011).

22 Sawa, Y. *et al.* Shape and chirality transitions in off-axis twist nematic elastomer ribbons, *Phys. Rev. E* **88**, 022502 (2013).

23 Teresi, L. & Varano, V. Modeling helicoid to spiral-ribbon transitions of twist-nematic elastomers. *Soft Mater* **9**, 3081 - 3088 (2013).

24 Wie, J. J. *et al.* Torsional mechanical responses in azobenzene functionalized liquid crystalline polymer networks. *Soft Matter* **9**, 9303 - 9310 (2013).

25 van Oosten, C. L. *et al.* Bending dynamics and directionality reversal in liquid crystal network photoactuators. *Macromolecules* **41**, 8592 - 8596 (2008).

26 Liu, D. & Broer, D. J. Liquid crystal polymer networks: preparation, properties, and applications of films with patterned molecular alignment. *Langmuir* **30**, 13499 - 13509 (2014).

27 Liu, D. & Broer, D. J. New insights into photoactivated volume generation boost surface morphing in liquid crystal coatings. *Nat. Commun*. **6**, 8334.

28 Sanchez-Ferrer, A., Merekalov, A. & Finkelmann, H. Opto-mechanical effect in photoactive nematic side-chain liquid-crystalline elastomers. *Macromol. Rapid Commun*. **32**, 671–678 (2011).

29 Zeng, H. *et al.* High-resolution 3D direct laser writing for liquid-crystalline elastomer microstructures. *Adv. Mater*. **26**, 2319–2322 (2014).

30 Min Lee, K., Lynch, B. M., Luchette, P. & White, T. J. Photomechanical effects in liquid crystal polymer networks prepared with m-fluoroazobenzene. *J. Polym. Sci. A* **52**, 876 – 882 (2014).

31 Shadmehr, R. & Arbib, A. M. A mathematical analysis of the force-stiffness characteristics of muscles in control of a single joint system. *Biol. Cybern*. **66**, 463-477 (1992).

32 Mossety-Leszczak, B., Wlodarska, M., Galina, H. & Bak, G. W. Comparing liquid crystalline properties of two epoxy compounds based on the same azoxy group. *Mol. Cryst. Liq. Cryst*. **490**, 52–66 (2008).

33 Li, C. *et al.* Synthesis of a photoresponsive liquid-crystalline polymer containing azobenzene. *Macromol. Rapid. Commun*. **30**, 1928–1935 (2009).

Figure Captions

Figure 1|Helix-based motion and mirror-image helices in biological systems. a) Passion flower tendril. Their twisting appears to originate in the asymmetric contraction of cellulose micro-fibrils. b) Common broom (Sarothamnus scoparius) seedpods. Seedpods open via a mechanism creating helical chirality. c) Seed and awn of *Erodium.* The awns display humidity-driven motion. Reproduced with permission from [7]. d) Twining Fringe Lily.

Figure 2|Components of the materials used in this experimental design. Complex behaviour emerges from the engineering of these relatively simple molecules.

Figure 3|A variety of springs can be prepared by cutting a homogeneous thin film of liquid crystal network (top panel). The shape of the springs is reported as a function of the cutting angle ϕ (bottom and right panels).

Figure 4|A specific liquid crystal cell is required to prepare liquid crystal polymer springs. **(a)** shows a single twist cell filled with a liquid crystal of composition 3 Table **1.** The green rods represent the liquid crystal molecules schematically, and the distribution of their preferential orientation through the thickness of the film. The red rectangle shows the cutting edges for the ribbon shown in **c**. **(b)** shows a side representation of a planar cell. **(c)** The ribbon cut at ϕ = 45° twists under irradiation with light. **(d)** shows a planar cell filled with a liquid crystal of composition 3 Table **1**. **(e)** shows a side representation of a planar cell. All liquid crystal molecules are preferentially oriented in the same direction. **(f)** When a planar cell is used, for the same offset angle ϕ = 45° a flat ribbon bends under irradiation with light, but it does not twist.

Figure 5|Photo-actuated twisting and/or untwisting motion is encoded via orthogonal deformation modes: in all cases, the outside of the ribbons deforms perpendicularly to the inside of the ribbons. Specifically, in the example of a ribbon cut at ϕ = 45°, the outside of the ribbon elongates, whereas its inside layer contracts primarily, which overall leads to twisting upon irradiation with light at λ = 365 nm. The green rods schematically represent the preferential alignment of the liquid crystal molecules. The composition of this specific ribbon is described as composition **3** in Table **1**.

Figure 6 | Non-linear mechanical character of a biomimetic photo-responsive polymer spring ($\phi = 45^{\circ}$). The graph displays the pulling force that is applied to the spring, with respect to its elongation.

Figure 7 | Filling of the cell with the liquid crystal pre-polymer mixture. a) The liquid crystal is inserted by capillarity action into the cell, by using a metal tip. b) The step is repeated until the cell is filled completely.

Figure 8 | Photo-polymerisation. The cell is irradiated ($\lambda \geq 420$ nm) at 48 ºC and post-cured overnight at 60 ºC in the oven.

Figure 9 | Opening of the cell. a) The cell is opened with a surgical knife and b) the liquid crystal polymer film is removed by hand from one of the cell walls.

Figure 10 | The procedure allows producing a variety of photo-responsive polymer springs. Their photo-response is encoded into their structure.