

1 **A *Drosophila* larvae-inspired vacuum-actuated soft robot**

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14

15 **Abstract**

16 Peristalsis is one of the most common locomotion patterns in limbless animals. This
17 motion is generated by propagating muscular contraction and relaxation along the body axis.
18 While the kinematics of peristalsis has been examined intensively, the kinetics and
19 mechanical control of peristalsis remain unclear, partially due to the lack of suitable physical
20 models to analyse the force and temporal control in soft-bodied animals' locomotion. Here,
21 based on a soft-bodied animal, *Drosophila* larvae, we proposed a vacuum-actuated soft robot
22 replicating their crawling behaviour. The soft structure, made with hyperelastic silicon rubber,
23 was designed to mimic the larval hydrostatic structure. To estimate the adequate range of
24 pressures and time scales for control of the soft robots, a numerical simulation by the finite
25 element method was conducted. Pulse-Width-Modulation (PWM) was used to generate time-
26 series signals to control the vacuum pressure in each segment. Based on this control system,
27 the soft robots could exhibit the peristaltic pattern resembling fly larval crawling. The soft
28 robots reproduced two previous experimental results on fly larvae: slower crawling speed in
29 backward crawling than in forward crawling, and the involvement of segmental contraction
30 duration and intersegmental delay in crawling speed. Furthermore, the soft robot provided a
31 novel prediction that the larger the contraction force, the faster the crawling speed. These
32 observations indicate that the use of soft robots could serve to examine the kinetics and
33 mechanical regulation of crawling behaviour in soft-bodied animals.

34

35 **1. Introduction**

36 Over the past few decades, robotics researchers have been drawing inspiration
37 from diverse species of animals to design robots [1, 2]. A recent approach to building animal-
38 inspired robots is utilizing soft materials to construct flexible structures, mainly because the
39 flexibility enables adaptive motions [2, 3]. Furthermore, the development of soft robots has
40 provided insights into the biological mechanisms of animal motion [4, 5]. In particular, soft
41 robots are useful for understanding the kinematics of soft-bodied animals' behaviours since
42 their locomotion has a high degree of freedom. The use of physical simulation is
43 indispensable in examining the complicated dynamics [6]. The development of biomimetic
44 soft robots has provided valuable platforms for both robotics and neuroscience research
45 fields by referring to animals [7], including caterpillars [1], earthworms [8], and octopi [9].

46 Crawling behaviour is generated by the propagation of segmental contraction along the
47 body axis to move the body forward [10]. Crawling is one of the basic animal motions used
48 to move the body in one direction and has been mimicked by soft robots, including worm-like
49 robots [8], hornworm-like robots [1], snake-like robots [11], and multigait soft robots [12].
50 Previously, flexible braided mesh-tube structures, Meshworm and FabricWorm, were
51 designed based on the antagonistic muscular arrangement of earthworms [8, 13] and were
52 capable of exhibiting crawling. They used shape-memory alloys and linear springs as
53 actuators, respectively. Despite these recent advances in soft robots for studying crawling
54 behaviour, how crawling properties, including speed, are realized and regulated remains
55 unclear [14, 15].

56 Larvae of fruit flies, *Drosophila melanogaster*, have provided an excellent model of a
57 soft-bodied organism to investigate peristaltic mechanisms due to their relatively simple
58 structure, stereotyped behaviours, and accumulated knowledge of their neural circuits [16,
59 17]. The third instar fly larva is about 4 mm long and has a segmented body. The dominant
60 larval behaviour is forward crawling, which involves the sequential translation of body
61 segments via muscle contraction along the body axis. However, the larvae also exhibit
62 backward behaviour, with the same muscles activated in the opposite sequence [18]. One
63 soft maggot robot was previously designed to mimic larval muscular organization and
64 replicate larval crawling [19]. It consisted of a series of pneumatic chambers that enabled
65 body deformation by expansion. Although coordinated motions were generated, this previous
66 maggot robot did not realize crawling behaviour.

67 Pneumatic circuits have been used as an actuator in soft robots. By increasing and
68 decreasing internal pressure in chambers of soft material, adaptive and versatile motion can
69 be realized [14]. There have been various applications of pneumatic circuits to soft robots to
70 perform multiple gaits [14], a hybrid of hard and soft robots [20], and gloves for hand
71 rehabilitation [20].

72 In this work, we propose a new soft robot that can mimic larval crawling through
73 reference to the properties of fly larvae. To mimic the contraction of body segments, a vacuum
74 source and solenoid valves were used for actuation control. We implemented an asymmetric
75 interface between the robot and a ground substrate to enhance the forward crawling rather
76 than backward crawling. This larval robot successfully exhibited a crawling motion.

77 Perturbation experiments suggest that the contraction force and segmental phase delay are
78 critical for the crawling speed. This study indicates that our vacuum-based soft robots could
79 contribute to a better understanding of the mechanisms of crawling behaviour and the
80 development strategy of soft robots with faster crawling speeds.

81

82 **2. Methods**

83 By mimicking the biological properties of fly larvae, we proposed a soft robot
84 consisting of the following three components: A) a body structure with a chain of elliptical
85 cylinder-like segments, B) a vacuum-actuated control system, and C) software for controlling
86 and monitoring the motion of the maggot-like soft robot.

87

88 **2.1 Body structure**

89 When designing our soft larval robot, three structural properties in fly larvae were
90 taken into account: 1) hydrostatic structure, 2) repetitive muscular patterning and 3)
91 asymmetric substrate interaction via denticle bands.

92 First, the fly larval body is filled with body fluid, and the internal pressure and the
93 tension of the body wall play a role in supporting their body shape. To replicate the larval
94 hydrostatic properties, we adopted a pneumatic structure with silicone rubber among the
95 current soft actuator candidates [2, 20]. Second, the configuration of muscles in the body wall
96 is segmentally repeated in fly larvae. The larval body consists of 11 segments: three thoracic
97 segments and eight abdominal segments. Although the terminal segments (the first thoracic
98 (T1) and the last abdominal (A8) segments) have specialized structures, the other nine
99 segments have an almost consistent structure. The larval length and width were examined
100 as 3.69 ± 0.56 mm and 0.66 ± 0.09 mm [21]. The length-to-width ratio of a single segment in the
101 major middle segments is about 0.5 (Supplementary Figure 1). To mimic the larval shape,
102 the individual segment was designed as an elliptical cylinder chamber with a flat plane at the
103 bottom. We constructed two different soft robots with different segmental length-width ratios:
104 The width of all the segments was 30 mm, whereas the axial length of the segments of the
105 three robots was 20 mm and 30 mm, respectively. Accordingly, their length-to-width ratios of
106 them were 2/3 and 1, respectively. To simplify the robot while allowing us to analyze the
107 propagation of segmental contraction, we set the number of the segments in one robot as

108 five (Figure 1). Third, spike-like structures align segmentally at the bottom of the fly larvae,
109 named denticle bands. Denticle bands act as anchorage points to the ground during the
110 propagation of segmental deformation [22]. Each denticle band normally consists of six rows
111 of denticles in a larva. The hooked tips of four out of the six rows point posteriorly while the
112 remaining two rows point anteriorly [22], suggesting that the friction between the ventral body
113 surface and the ground substrate should be different between forward and backward motions.
114 To mimic this asymmetric denticle structure between anterior and posterior directions, we
115 implemented an asymmetric friction structure by glueing a piece of paper (Whatman paper
116 1001-917) at the anterior side of the segment boundary (shown as the thick lines in
117 Supplementary Figure 1). Silicone rubber is stickier than paper; hence the physical contact
118 between the silicone rubber and a ground substrate would generate larger friction than the
119 one between paper and the ground substrate. By virtue of this property and the geometric
120 fact that the angle of the segment boundary depends on the direction of the propagation of
121 segment contraction, asymmetric friction during locomotion in different directions could be
122 realized.

123 We fabricated soft robots based on the design shown above. First, the moulds for
124 the soft robots were printed with Acrylonitrile Butadiene Styrene (ABS) material with a 3D
125 printer (WANHAO Duplicator 4S). Then, the moulds were integrated (Figure 1A), and silicone
126 rubber (Ecoflex 00-30) was poured into the space between the inner and the outer moulds.
127 After the gap was filled up with silicone rubber, a piece of paper (Whatman paper 1001-917)
128 was inserted between the anterior surface of the liquid rubber and the mould (Figure 1B).
129 The rubber was cured in a drying oven (at 80°C for about 20 minutes) and cooled down at
130 room temperature. Then, the single segment structure was demolded, as shown in Figure
131 1C. Finally, five segmental structures were glued together using the Ecoflex mixture to build
132 the final structure (Figures 1D and 1E). An example is shown in Figure 1F.

133

134 **2.2 Vacuum control system**

135 In order to enable the segmental chambers to contract like body segments in fly
136 larvae, we took advantage of a vacuum-based actuator. We built a system consisting of
137 pneumatic circuits, electrical circuits (dashed and solid lines respectively in Figure 2), and a
138 graphical user interface software (implemented on "Computer" in Figure 2).

139 The pneumatic circuits were established to control the pressure in each segment
140 chamber. Different from previous larvae-like robots based on segmental expansion [19], in
141 this study, we adopted contraction as a driving force for larval locomotion to mimic muscular
142 contraction in fly larvae. Pneumatic pathways linked a vacuum source (TAITEC VC-15s, with
143 a pressure range from -110kPa to 0kPa), three-way solenoid valves (ZHV 0519), vacuum
144 pressure sensors (MPXV6115V), and robotic chambers (see section 2.1). Each chamber and
145 the vacuum source were connected through a vinyl tube and a solenoid valve. The tube was
146 flexible and lightweight (1.4 g) compared with the soft robots (16.9 – 29.9 g). Even when we
147 held the tubes, the motion of the soft robots was not disturbed, showing that the tube has
148 little effect on the kinematics of the soft robot. The pressure within each chamber was
149 regulated by gating the solenoid valves.

150 Electrical circuits were built to control the pressure valves for individual chambers.
151 A microcontroller (Arduino Mega) was selected considering the number of PWM pins and the
152 output voltage. The microcontroller was connected to solenoid valves and vacuum pressure
153 sensors to regulate and monitor robot deformation. The microcontroller's detailed electrical
154 circuits are presented in Supplementary Figure 2, including modules for the solenoid valves
155 and vacuum sensors. Here, an NPN transistor (TIP120) modulates an external high-power
156 source to drive the solenoid valves based on the PWM signal from the microcontroller. The
157 6V external power, provided via a DC-DC power supply regulator (ARD-PWR), was the
158 maximal operational voltage for the solenoid valves. Since the valves possessed non-
159 negligible inductance, diodes (1N4007) are used in a flyback configuration to prevent a large
160 back-electromotive force (back emf), which could damage these valves. These diodes could
161 dissipate the remaining energy. The vacuum pressure, in the range of -115 to 0 kPa relative
162 to the standard atmosphere, was measured by the output of the voltage-based vacuum
163 sensor based on the following equation:

$$164 \quad P = \frac{V_{OUT} - 0.92 \cdot V_S}{0.007652 \cdot V_S} \pm c_{Temp} \cdot P_{error} \quad (1)$$

165 where P is the vacuum pressure, and V_{OUT} and V_S represent the output voltage and
166 voltage supply, respectively, in the pressure sensor. P_{error} is the pressure error, which
167 indicates the measurement error in the pressure at a standard temperature (the dimension
168 of P_{error} is pressure). c_{Temp} is a temperature factor for the pressure error, which is 1 within
169 the temperature ranges from 0 to 85°C (c_{Temp} is dimensionless). According to the sensor

170 datasheet, the maximum value of P_{error} is 1.725 kPa within the range of our experiments.
171 Low-pass filters were applied to pressure signals from vacuum sensors (Supplementary
172 Figure 2).

173

174 **2.3 Software for controlling and monitoring soft robot locomotion**

175 To monitor the position of the segment boundaries and their segmental pressures
176 in operation in real-time, we designed a graphical user interface (GUI) (Supplementary Figure
177 3) based on Python libraries (Tkinter and pySerial). The USB camera (HOZAN camera with
178 12mm lens) is attached to a bracket right above the soft robot. Its frame rate is up to 40
179 frames per second, used in the following experiments. The images were sent to the computer
180 via a USB connector, and the robotic motion could be observed on the GUI (Figure 3 right,
181 Supplementary Figure 3). The position of the front end (“head”) and rear end (“tail”) was
182 measured by a ruler shown in Figure 3. Meanwhile, pressure signals of robot segments were
183 delivered from the Arduino board to the computer. Example locomotion and pressure signals
184 are shown in Supplementary Figure 3.

185

186 **3. Simulation**

187 To determine the suitable ranges of the pressure and time scale for robot
188 deformation, we applied the finite element method (FEM) to simulate our soft robot via the
189 commercial FEM software (Abaqus). To achieve a reliable simulation, suitable viscoelastic
190 physical models were required. In our project, the main soft body was constructed using
191 hyperelastic material (Ecoflex 00-30). In line with previous modelling and validation work [6],
192 the Ogden model was adopted to model this material. The strain energy potential function U ,
193 which describes the elastic properties of the material, is defined as:

194
$$U(\lambda_1, \lambda_2, \lambda_3, J) = \sum_{i=1}^N \frac{2\mu_i}{\alpha_i^2} (\lambda_1^{\alpha_i} + \lambda_2^{\alpha_i} + \lambda_3^{\alpha_i} - 3) + \sum_{j=1}^N \frac{1}{D_j} (J - 1)^{2j} \quad (2)$$

195 where μ_i and α_i ($i = 1, 2, 3$) are the primary fitting parameters, and λ_j ($j = 1, 2, 3$) are the
196 stretches along the x, y, and z axes. The material constant N was set as three. The second
197 summation term contains fitting parameters D_j ($j = 1, 2, 3$) to the volumetric deformation and
198 the material Jacobian matrix J . We referred to the previous parameters for this hyperelastic
199 material [6]. Meanwhile, regarding the paper (Whatman paper) on the intersection of soft
200 robots, the material density is $4.83e-10 t/mm^3$, and its young’s module is 1.71GPa and

201 poisson ratio is -0.3 [23]. Considering the segmental deformation under vacuum pressure,
202 we also configured the self-contact condition for the interaction step.

203 We calculated the responses of the soft robot under distinct pressures using the
204 FEM software Abaqus (Figure 4). With respect to the nonlinear dynamics within our model,
205 we configured parameters for incrementation to ensure a successful simulation: the
206 maximum number of increments as 100 and the minimum increment size as 1e-15. Here, the
207 example soft robot with five 20 mm segments was indicated as 20mmx5-SR (20 mm, five
208 segments, and Soft Robot), and a similar shorthand notation was used for 30 mm segments
209 30mmx5-SR. Two points at the bottom of the terminal segment (marked by red points in
210 Figure 4A) were used to monitor the head and tail segmental position in the longitudinal axis
211 to record their asymmetric deformation. When the negative pressure of -10kPa was applied
212 to the most anterior chamber (head chamber) of 20mmx5-SR, the head marker exhibited a
213 negative displacement (Figure 4C), which indicated that the head segment was contracted
214 and the end of the head moved backwards. The head marker moved faster when a larger
215 absolute value of the negative pressure (from -20kPa to -70kPa) was applied to the head
216 chamber. This observation indicated that the kinematics of the segment dynamics could be
217 regulated by the pressure within the soft robot chambers. Since there was no asymmetry
218 along the body axis in the FEM simulation, when negative pressures were applied at the
219 most posterior chamber (tail chamber), the tail marker showed similar displacements but in
220 the opposite direction (Figure 4F). The larger soft robot (30mmx5-SR) exhibited faster
221 segment contraction (Figure 4D and 4G). In either case, the movement was carried out in
222 less than 1 second, comparable to the stride duration in fly larval locomotion.

223

224 **4. Results**

225

226 **4.1 Test for control signal and Comparison with simulation responses**

227 In this study, we analyzed the locomotion of the soft robots under conditions with
228 varied contraction forces and distinct intersegmental phases. Since the pressure of the
229 vacuum source was constant, we tried to realise varied pressures by controlling the duty
230 cycle of the gating of solenoid valves. To this aim, we adopted the Pulse-Width-Modulation
231 (PWM) method [11] to control the temporal patterns of the chamber pressures. In this method,

232 a series of pulses are generated, and the frequency and the duty cycle of the pulses can be
233 tuned. We set the frequency as 1000 Hz because it was fast enough to reproduce fly larval
234 crawling that occurred on the scale of 100 ms. The frequency of 1000 Hz was realized by
235 setting a parameter OCRnA for the Arduino Mega board as 249 (= 16 MHz / 1000 Hz / 64 –
236 1). On the other hand, the duty cycle could be adjusted by OCRnB/OCRnC, another
237 parameter for the board. Both OCRnA and OCRnB/OCRnC are in the range from 0 to 255
238 (Figure 5A). OCRnB/OCRnC set the value thresholding of a sawtooth timer signal to produce
239 PWM signals with varied duty cycles (Figure 5A). We then used the resulting PWM signal to
240 control segmental deformation via vacuum pressure. The deformation of the head segment
241 was monitored under different duty cycles (Figure 5B). The results showed that the
242 segmental deformation couldn't be observed when the OCRnB is smaller than 190, which
243 means that the solenoid valve doesn't work in this case. On the other hand, when OCRnB
244 was 195 or more, the contraction of the head segment was observed. In particular, as OCRnB
245 increased, the speed of the deformation increased, which suggested that the contraction
246 force within the head segment chamber was higher when the duty cycle was larger.
247 Accordingly, we succeeded in temporally controlling the pressures within the segments of the
248 soft robot. Even when we changed the waveform from the square to others, such as
249 sinusoidal and saw-like waveforms, the temporal profiles of chamber pressure were similar
250 to that with a square waveform input (Supplementary Figure 4). Then we decided to use the
251 square waveform to control the chamber pressure in the following analyses.

252 Referring to the results from the FEM simulation (see section 3), we analyzed the
253 response of the soft robots to the pneumatic operation (Figure 6). When negative pressure
254 was applied to the head chamber, the head marker on the 20mmx5-SR exhibited negative
255 displacements, which was consistent with the FEM simulation results (Figure 6A). The
256 displacement gradually reached a stable value in less than 1 second. A similar tendency was
257 observed when negative pressures were applied to the tail chamber (Figure 6B). The larger
258 soft robot (30mmx5-SR) showed weaker displacement than 20mmx5-SR (Figures 6C and
259 6D). All of these observations in the measurement in the soft robots were consistent with the
260 FEM results. These analyses indicated the larger mass of soft robots, such as 30mmx5-SR,
261 would make the robot harder to deform and move. Hence, we described experimental results
262 for the 20mmx5-SR soft robot in the following sections.

263 One striking difference between the simulation and the soft robot experiment was
264 the existence of asymmetric friction. As described in Supplementary Figure 1C, the friction
265 at the interface between the soft robot and the ground substrate was asymmetric. Due to this
266 effect, the tail marker showed a larger displacement upon applying negative pressure to the
267 tail chamber than the head marker did upon the negative pressure applied to the head
268 chamber. (Figure 6).

269 In summary, we implemented and optimized three properties in the soft robot to
270 mimic fly larvae: The contraction of chambers instead of the expansion, which was used
271 previously [19], the asymmetric feature of the interface between the soft robot and the ground
272 substrate, and optimal ranges in pressure, time scale, and the size of the chambers.

273

274 **4.2 Robotic locomotion and its quantification**

275 We tested the crawling ability of the soft robot. Using the PWM method, we
276 generated temporal patterns coding that negative pressure was sequentially applied from the
277 posterior to anterior chambers (Figure 7). As an initial case, we set the vacuum pressure as
278 -10 kPa, segmental contraction duration as one second, and overlap of the contraction time
279 between neighbouring segments as zero (Figure 7A). Under this condition, the soft robot
280 exhibited a crawling motion and could move forward (Figure 7B). Accordingly, this result
281 suggests that our soft robot could provide a physical model to analyze the crawling behaviour.
282 In the following sections, we attempted to reproduce two previous experimental results and
283 make one new prediction on fly larval locomotion with the soft robot.

284

285 **4.3 Asymmetric speed between forward and backward crawling**

286 As the first experimental observation in fly larvae, we focused on the difference
287 between forward and backward crawling speed. A previous study showed that fly larvae move
288 faster by forward crawling than backward [24]. However, the possible contribution of the
289 difference in the body-ground friction during forward and backward motion to the different
290 speeds between forward and backward crawling has not been tested. By using our soft robot,
291 we tested this possibility. The displacements of the soft robot were calculated based on the
292 position of its front end (the head marker in Figure 8). The speed of robot crawling was
293 measured using five successive strides. To generate backward crawling, we controlled the

294 spatiotemporal pattern of the chamber pressures in a similar way to that of forward crawling
295 but reverse order (Figure 8). We scanned the maximum pressures, stride durations, and
296 intersegmental delays and compared the speed of crawling between backward and forward
297 locomotion. In all the cases, backward crawling was slower than forward crawling (Figure
298 8C). This observation implies that the asymmetric properties of the interface between the
299 larval body and the ground substrate could be a critical factor in the asymmetric crawling
300 speed between forward and backward movements.

301

302 ***4.4 Involvement of segmental contraction duration and intersegmental phase delay in***
303 ***locomotion speed***

304 We next analyzed the relationship between the segmental kinematics and crawling
305 speed. A previous study reported that a class of inhibitory interneurons (PMSIs, period-
306 positive median segmental interneurons) was involved in crawling speed in fly larvae [25].
307 When blocking the activity of PMSIs, the segmental contraction duration and the delay in
308 contraction between neighbouring segments were elongated. Furthermore, the larvae with
309 reduced PMSI activity exhibited slower crawling. These observations suggested that either
310 the segmental contraction duration or the intersegmental phase delay should be involved in
311 crawling speed, but this hypothesis has not been tested. Taking advantage of our soft robot,
312 we investigated this possibility.

313 First, we changed the segmental contraction duration while keeping the pressure
314 constant and no overlapping between neighbouring segments (Figures 9A and 9B). The
315 result showed that the soft robot with a shorter segmental contraction duration (up to 0.2
316 seconds) exhibited faster crawling (Figure 9C). This observation was consistent with the
317 observation in the loss of function experiment of PMSIs.

318 Next, we perturbed the intersegmental phase delay while keeping other conditions.
319 We defined intersegmental phase delay as the ratio of an intersegmental time delay to
320 segmental contraction duration (Figures 9D and 9E). In fly larvae, the contraction of
321 neighbouring segments overlaps during larval crawling [26]. The stride duration we measured
322 from fly larvae is about one second, and the average segmental contraction duration is
323 around 0.5 seconds. Intersegmental delay is obtained by dividing the stride duration (1
324 second) by the number of segments (10), giving a 0.1-second intersegmental delay.

325 Accordingly, the phase delay of neighbouring segments in fly larvae is 20% (= 0.1 sec / 0.5
326 sec). We investigated the effects of segmental phase delay on crawling speed, which has
327 not been examined before.

328 We measured robot locomotion with various segmental phase delays. For our five-
329 segment robot, 20% and 40% phase delays were too short to generate stable locomotion
330 because all the segments were shrunk under these conditions. To make time for segments
331 relaxed, we tested segmental phase delays of 60%, 80%, and 100% while keeping
332 segmental contraction time constant (from 0.2 to 1 second) (Figure 9F). We found that as the
333 segmental phase delay increased, the crawling speed became slower (Figure 9F). A similar
334 observation was obtained in operation with different segmental contraction durations
335 (Supplementary Figure 6). These phenomena might be because the smaller segmental
336 phase delay promoted cooperative contraction of neighbouring segments, leading to larger
337 contraction of segments and faster crawling speed (Figure 9). To sum, the perturbation
338 experiments with our soft robot showed that both the segmental contraction duration and the
339 intersegmental phase delay could be involved in crawling speed, consistent with the previous
340 experimental observation.

341

342 **4.5 Prediction of the relationship between the maximum contraction force and
343 locomotion speed**

344 Finally, we analyzed the relationship between segmental contraction force and the
345 crawling speed, which has not been examined in fly larvae yet. We systematically changed
346 the vacuum pressure from -10 kPa to -70 kPa while keeping the segmental contraction
347 duration constant (one second corresponding to the segmental contraction time of 0.2
348 seconds for a five-segment soft robot) and no overlapping between neighbouring segmental
349 contractions. As a result, the soft robot operated with larger absolute values of the negative
350 pressure and exhibited a faster crawling speed (Figure 10). This tendency could be observed
351 in different segmental contraction durations. This observation could provide a prediction that
352 a larger muscular force could generate faster crawling in fly larvae.

353

354 **5. Discussion**

355 In this work, we developed a new vacuum-actuated soft robot designed to mimic fly larval

356 locomotion. Our soft robot can show peristaltic locomotion patterns produced by the
357 propagation of segmental contraction waves. Two points are crucial to realizing effective
358 peristaltic locomotion in our soft robots: First, pieces of paper inserted in the cross-section
359 help generate asymmetric friction between the soft robot and ground substrate, resulting in
360 different locomotion speeds in different directions. Currently, designs for asymmetric friction
361 in soft robots are still paid little attention. The simple but effective implementation using paper
362 in our robot implies the importance of friction asymmetry. Second, to determine the control
363 signals for the soft robot, we used the properties in fly larval crawling and configured various
364 parameters, including pressures, stride durations, and intersegmental phase delays via PWM
365 control. Based on the robotic locomotion, we found that the contraction force, intersegmental
366 phase delay, and stride duration all contribute to the regulation of crawling speed.

367 Analyses using our soft robots are consistent and give novel interpretations to previous
368 works. The mechanism of PMSI neurons indicates that shorter intersegmental phase delays
369 promote faster speed. Assays using soft robots provided evidence (Figure 10D-G) consistent
370 with this. Furthermore, the observation that our soft robot crawls faster in the forward than
371 backward direction is consistent with the previous kinematic study [24]. According to the
372 present study, the difference in speed between forward and backward crawling is partially
373 attributed to the asymmetric friction property between forward and backward directions.
374 Since two separate neural circuits are involved in forward and backward crawling in the
375 central nervous system [18], the different forward and backward crawling speeds might be
376 realized by two parallel mechanisms: friction asymmetry and circuit specialization.

377 There are several ways to improve the performance of robotic crawling in future studies.
378 First, the physical properties of soft materials are critical for the dynamics of soft robots. The
379 crawling speed should be improved by utilizing soft materials with larger frictional coefficients
380 for the soft robot. Second, a better actuation source, which can make the soft robot
381 untethered, would broaden its applications in practical scenarios. Third, implementing soft
382 sensors on the robotic body can make it more bionic. It has been reported that the
383 proprioception of the body wall is key to generating an innate crawling speed in *Drosophila*
384 larvae [27]. By monitoring the deformation on the surface of the soft robot and using this
385 information to control the crawling behaviour, the soft robot could exhibit flexible and adaptive
386 locomotion in various complicated environments. Last but not least, although we tested three

387 different sizes of robots, the scale of the robot can still be modified. By changing the number
388 and size of the segments, locomotion ability could be changed. By optimizing these
389 conditions, the application range of our soft larval robots would be broadened.

390 The kinematic results in our soft robot system have the potential to inspire further study
391 of larval motor outputs, including the mechanisms and kinematic effects of segmental
392 contraction force and phase delay. And our attempts to better understand the mechanisms
393 in soft-bodied animals will contribute to designing adaptive and robust soft robots.

394

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399

400 **Conflict of interest**

401 We have no conflict of interest with respect to the work.

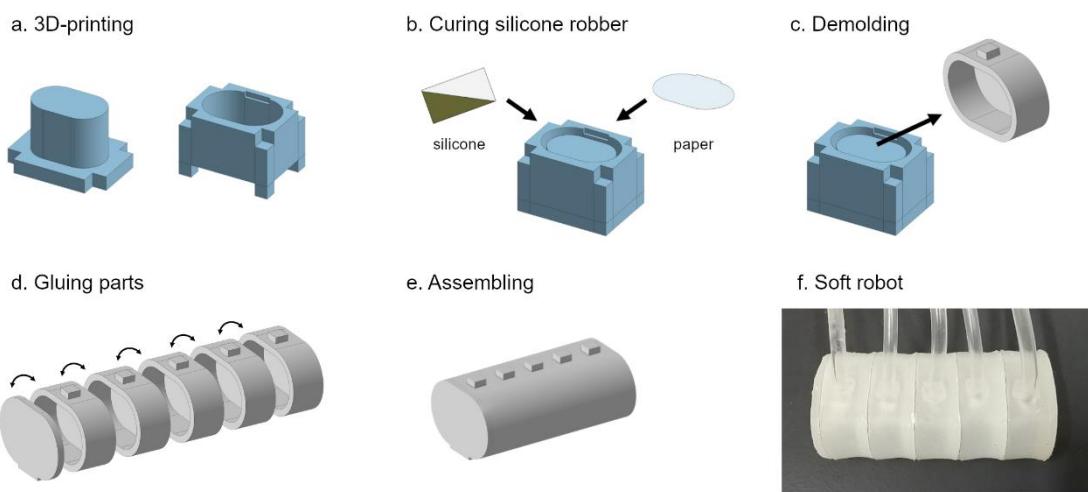
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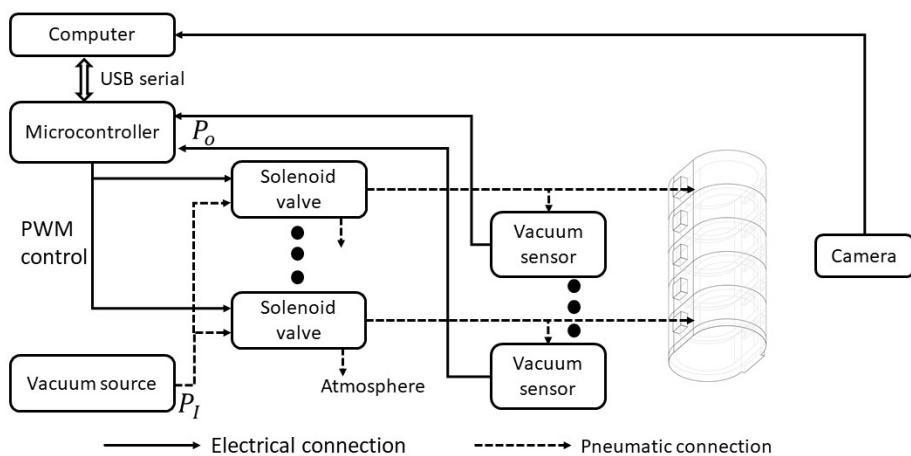
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468 **Figure 1. The fabrication process for the soft robot**

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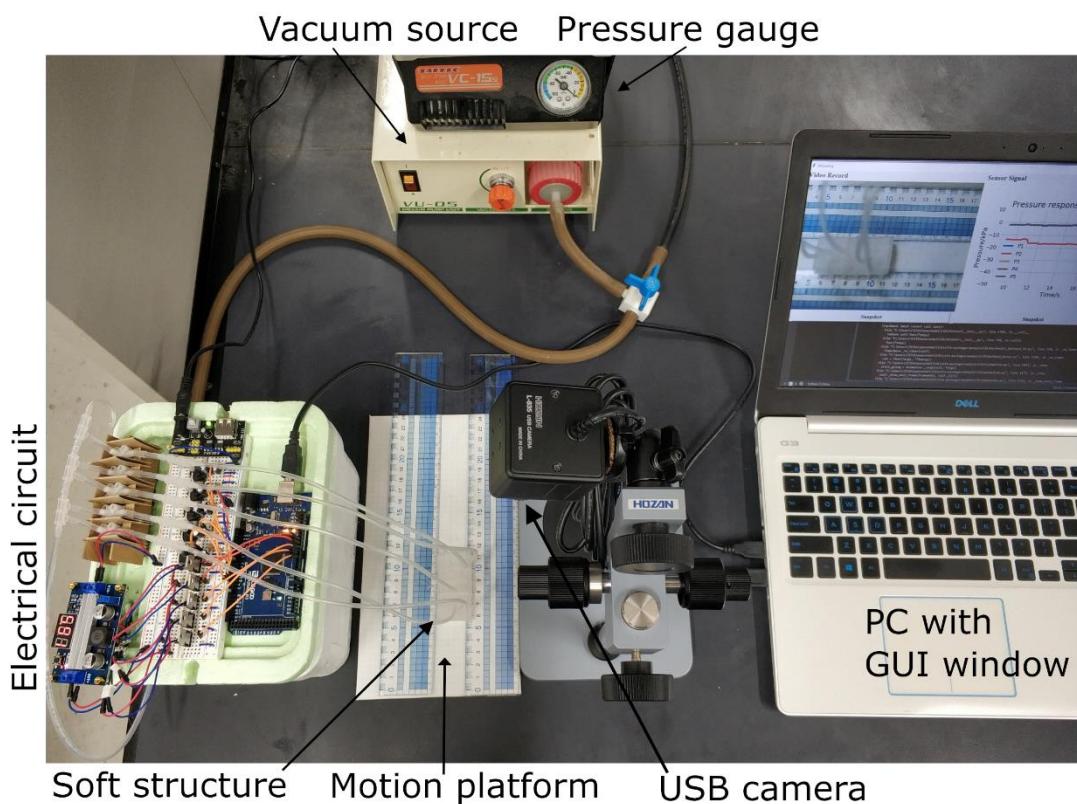


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472 **Figure 2. The framework of the whole system**

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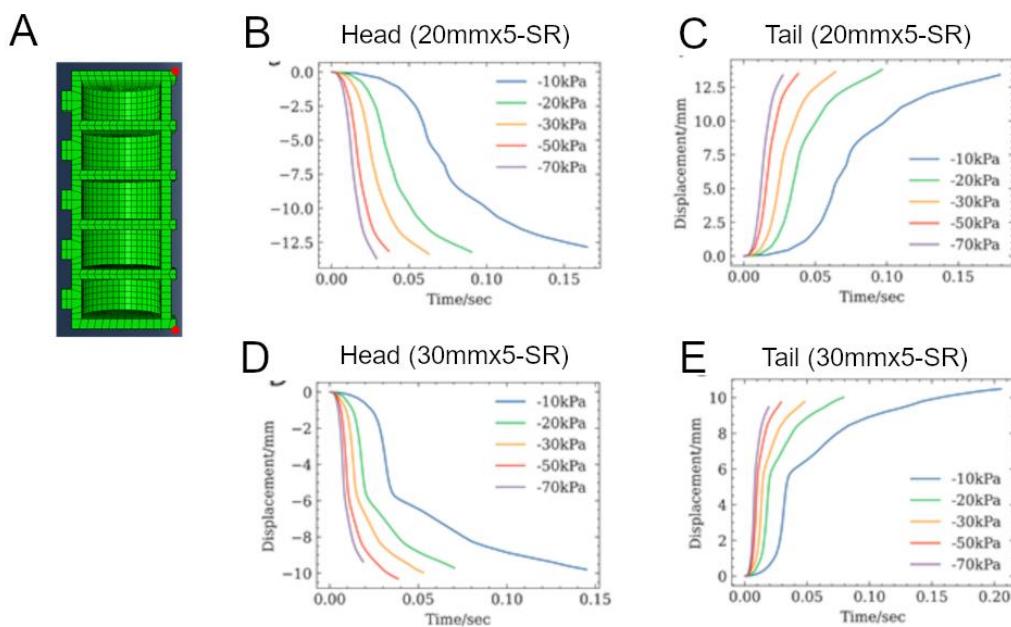
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476 **Figure 3. System overview**

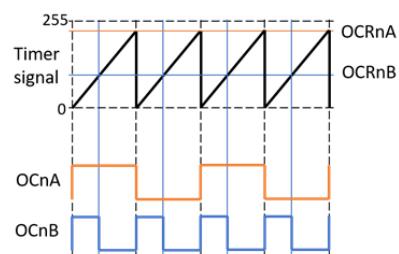
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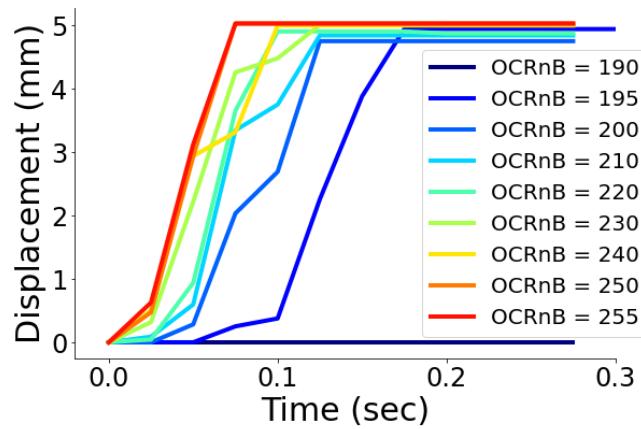
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479 **Figure 4. Simulation of the soft robots by FEM**

A



B

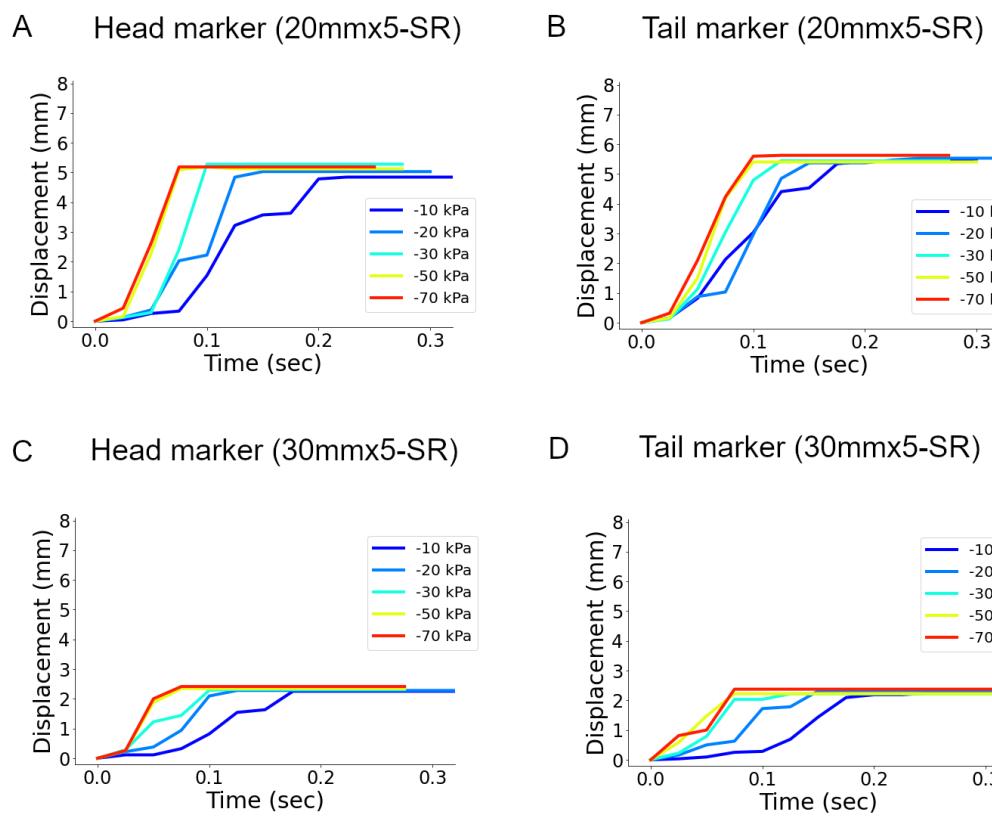


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481 **Figure 5. Test for a suitable range in the PWM control**

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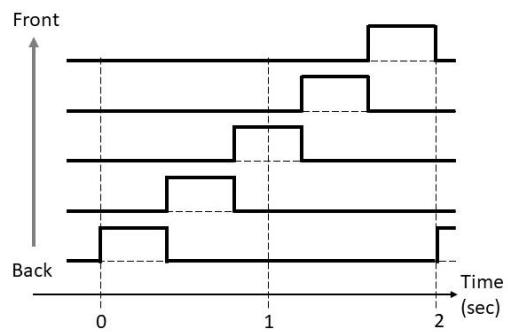
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486 **Figure 6. Segmental deformation in the soft robots**

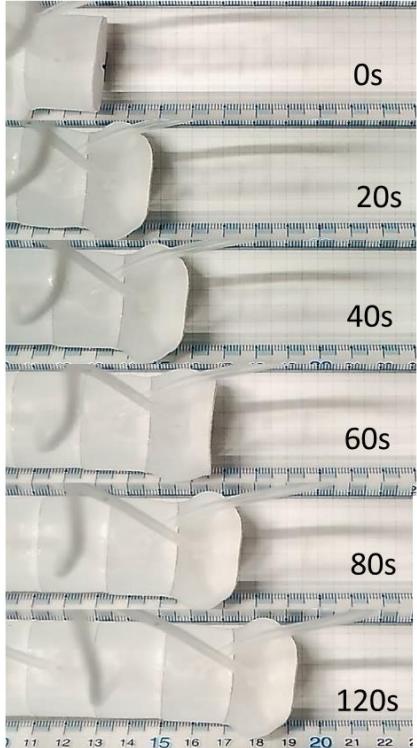
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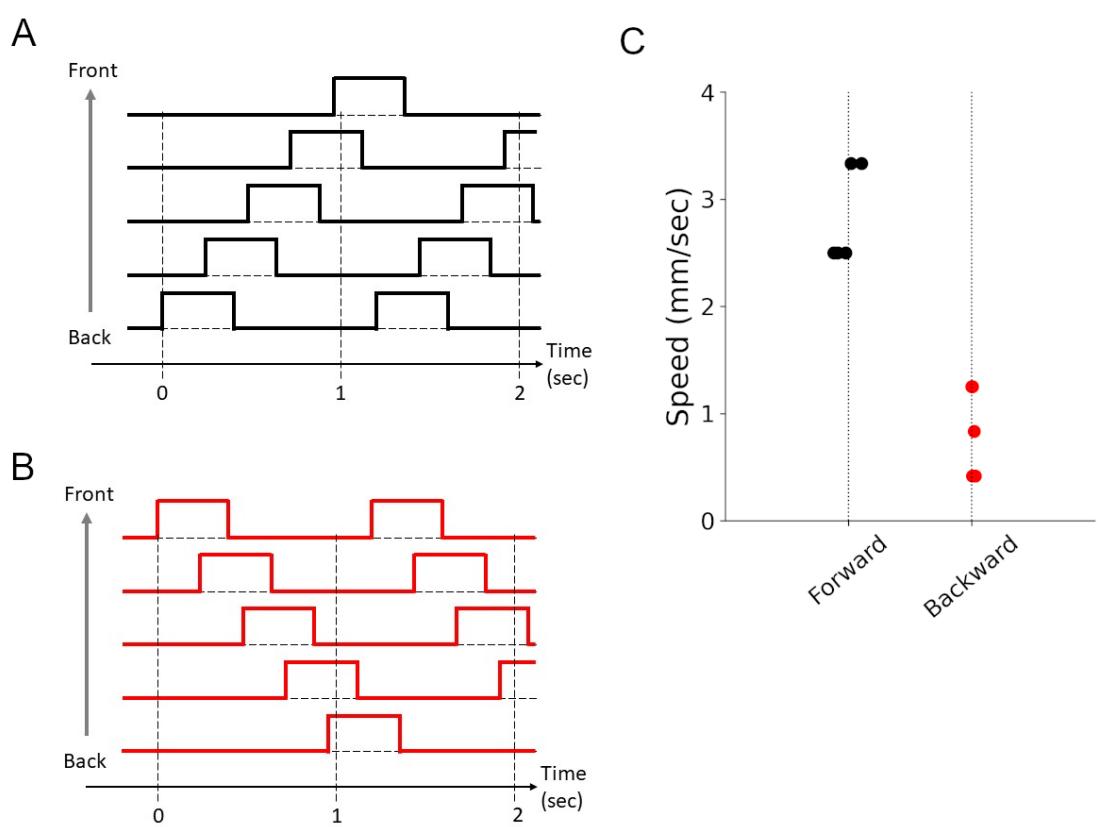
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491 **Figure 7. Performance of the soft robots**

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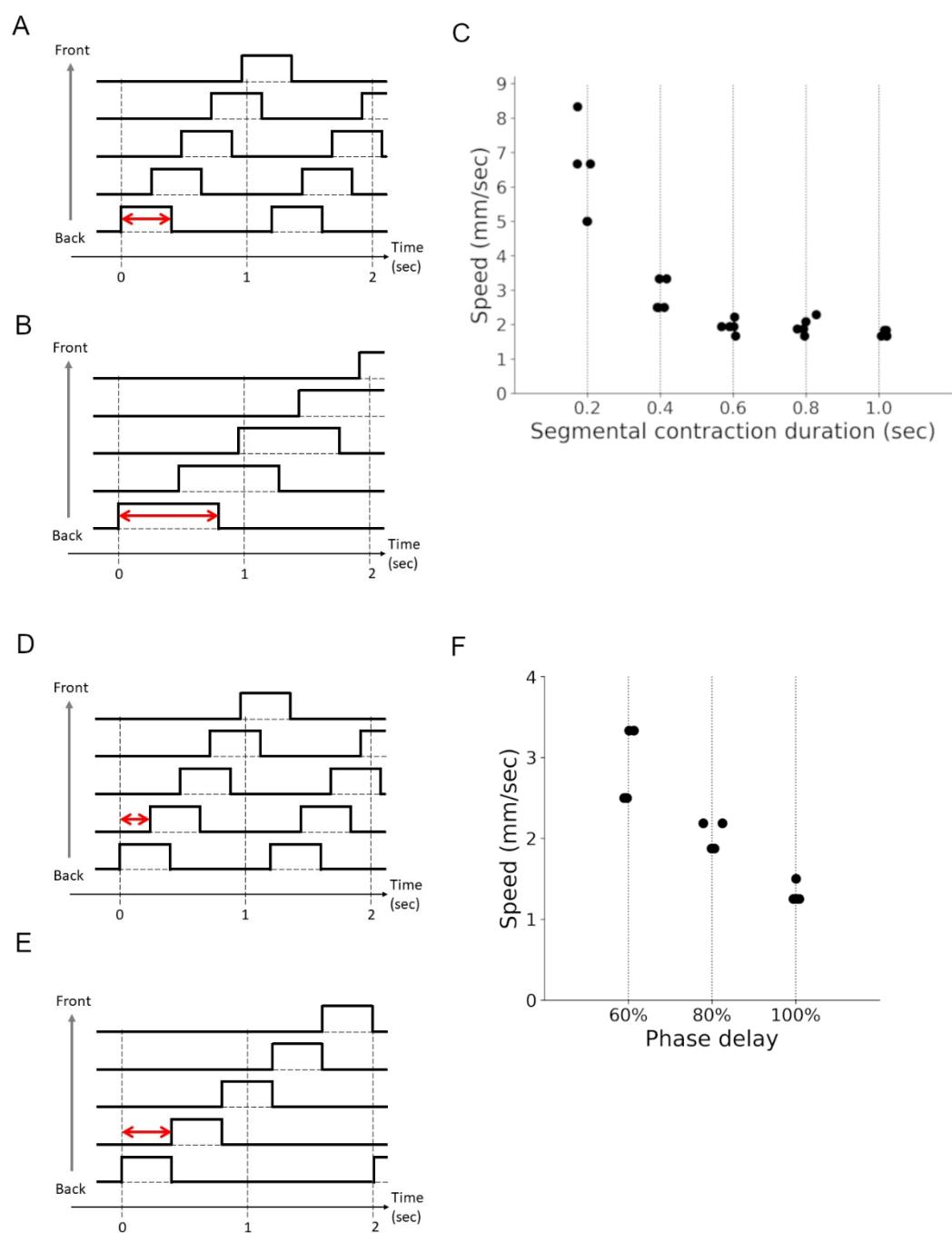
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496 **Figure 8. Forward and backward crawling in the soft robot**

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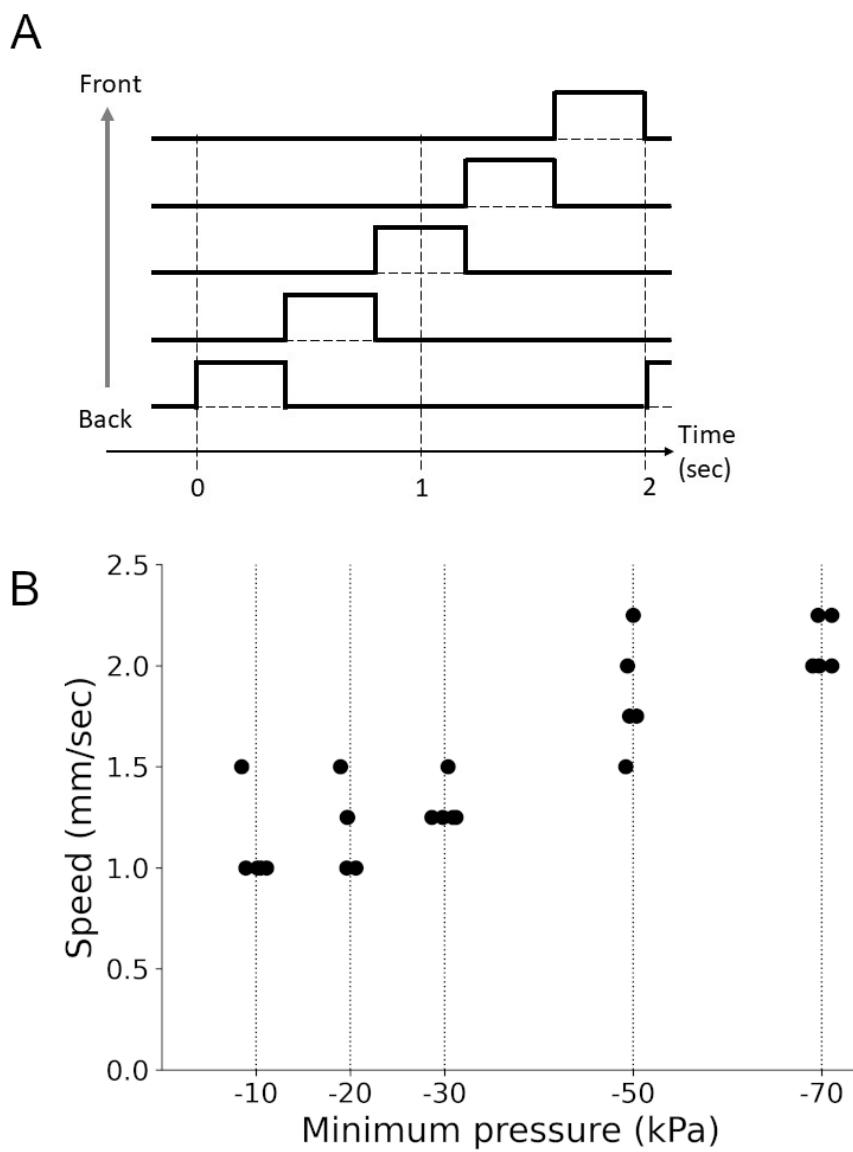
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501 **Figure 9. Involvement of intersegmental phase delay and segmental contraction**
502 **duration in crawling speed**

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506 **Figure 10. Relationship between the contraction force and crawling speed**