Collective motion as a distinct behavioral state of the individual

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Abstract

Collective motion is an important biological phenomenon, ubiquitous among different organisms. Locusts are a quintessential example of collective motion: while displaying a minimal level of cooperation between individuals, swarms of millions are highly robust and persistent. Using desert locusts in a series of carefully controlled laboratory experiments, we show that locust coordinated marching induces a distinct behavioral mode in the individual that we term a "collective-motion-state". Importantly, this state is not induced by aggregation or crowding per-se, but only following collective motion. It is manifested in specific walking behavior kinematics and has a lasting effect. Simulations confirm that the collective-motion-state provides an individual-based mechanism, which increases the robustness and stability of swarms in the presence of fluctuations. Overall, our findings suggest that collective-motion is more than an emergent property of the group, but a specific behavioral mode that is rooted in endogenous biological mechanisms of the individual.

Introduction

The ability to form groups that move collectively is a key behavioral feature of many species [1,2], that increases the survival of both individuals and groups [3,4]. Collectively moving organisms, however, differ in the levels of peer-to-peer interactions, ranging from minimal cooperation to complex social behaviors [5,6]. Analysis of both experiments and theoretical modeling of collective motion suggests a highly stochastic (or chaotic) movement pattern, which makes the trajectory of individuals difficult to predict in practice [7–9]. Furthermore,

endogenous variability among individuals, heterogenic environments, and variability in the interactions between the individual and its direct environment, are all sources of randomness that may affect the coordinated behavior of the collective. Accordingly, it is not clear how synchronized collective motion constitutes such a robust phenomenon, maintaining its form across various group sizes and densities, and under heterogeneous and unpredictable environmental conditions.

One of the most interesting, albeit disastrous, examples of collective motion is that of the marching of locusts. These insects swarm in groups of millions, migrating in mass across large distances, devastating crops and agriculture [10–12]. In the context of social interactions, locust swarming is characterized by a minimal level of cooperation between individuals: collectivity is mostly manifested in alignment among neighboring individuals and in maintaining the overall movement in the same general direction. Nonetheless, the locust swarming phenomenon is extremely robust, with huge swarms demonstrating moderate to high collectivity on huge scales (up to 6-7 orders of magnitudes), in terms of both the number of animals and their spatio-temporal distribution [13,14] (see further references in [7]). Thus, locusts exhibit a considerable disparity between little local cooperation and large-scale collectivity.

What is the key to this ability of locust swarms to maintain their integrity? Here, we show by a series of carefully controlled behavioral experiments that collective movement induces an internal switch in individual gregarious locusts, activating a behavioral mode we refer to as a “collective-motion-state”. In this state, the kinematic behavior of individuals notably differs from that during the non-collective state. It is important to emphasize that

53 by “collective” and the “non-collective” states we are not referring to the well-known
54 solitarious-gregarious phase transition in locusts [10,11].

55 How does the collective-motion-state affect the formation and robustness of the
56 swarm? Interestingly, the switch into this state seems to occur rapidly, and in response to
57 coordinated walking. In particular, our experiments indicate that aggregation alone is not
58 sufficient. Switching out of the collective-motion-state occurs over a longer time scale –
59 significantly longer than the typical time scale of fluctuations in the swarm dynamics. Hence,
60 stochastic fluctuations are “smoothed-out”, leading to highly robust dynamics of the swarm
61 collective behavior.

62 This ability to switch between socio-behavioral states appears to be beneficial for
63 the swarm integrity. Using a simplified computer model, we simulated the swarming
64 properties of locust-like agents with different kinematic parameters, representing the
65 different behavioral states. The results demonstrate that the ability to switch between the
66 distinct behavioral states, as a consequence of both instantaneous and past stimuli, leads to
67 swarms with tighter groups, compared for example to fixed or continuously changing
68 dynamics, and without affecting the within-group alignment. Hence, we conclude that the
69 collective-motion-state provides an individual-based mechanism that increases the stability
70 of swarms in the presence of fluctuations, preventing the swarm from collapsing.

71

72 **Results**

73 The main objective of this report was to examine the behavior of individual animals
74 upon joining or leaving a group of conspecifics. Here we studied gregarious locusts, reared
75 in dense populations, one developmental stage before becoming adults and developing

76 functional wings (i.e. fifth larval instar). The experiments comprised three consecutive
77 stages, representing different conditions (Fig. 1 and Vid. S1): (1) Isolation stage: a single
78 animal was taken from its highly dense rearing cage, tagged with a barcode, and introduced
79 alone into a ring-shaped arena (outer and inner diameters: 60 cm and 30 cm, respectively);
80 (2) Grouping stage: nine other individually tagged animals were added to the arena; and (3)
81 Re-isolation stage: the nine added animals were removed from the arena, leaving the
82 original animal alone. The duration of each stage was one hour, during which the animals'
83 trajectories were fully reconstructed using a barcode tracking system. The middle 40
84 minutes of each stage were analyzed, as detailed in the Methods section. A range of
85 kinematic statistics was collected in order to classify and compare the locusts' behavior in
86 the different stages.

87 **Swarm formation – validation of collective motion**

88 In order to verify that our grouping conditions were indeed inducing collective
89 motion (swarming), we calculated the synchronization in movement of the grouped animals
90 (the order parameter, see Methods for definition). The median order parameter in the
91 grouping stage was found to be significantly higher than that obtained for computationally
92 randomized groups (as presented in our previous report, Knebel et al., 2019); medians:
93 0.348 and 0.239 respectively; Wilcoxon rank-sum test: $p < 0.01$). Consequently, we
94 conclude that the groups in our experimental setup indeed demonstrate swarming and
95 collective motion.

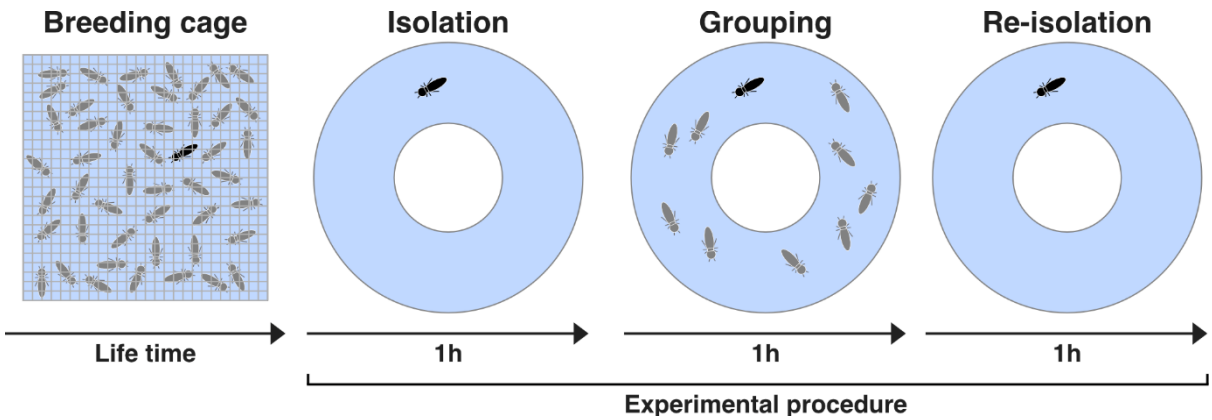

Figure 1. A schematic flow of the experimental procedure. Locusts were reared in high density conditions. The Experiments comprised the following consecutive stages: (1) isolation for one hour in the arena; (2) grouping for one hour; and (3) re-isolation for one hour.

Kinematic differences among the isolation, grouping, and re-isolation stages

Locusts walk in an intermittent motion pattern [16,17], i.e., movement occurs in sequences of alternating walking bouts and pauses. To characterize individual locust kinematics, we measured four parameters: (1) the fraction of time an animal spends walking; (2) the average speed while walking; (3) the average walking bout duration; and (4) the average pause duration. Comparing these values across the three experimental stages, we found several statistically significant differences (Fig. 2). In the following, *p*-values correspond to a Friedman test followed by a multiple comparison test using the Bonferroni method.

We found that when comparing between isolation and grouping, the fraction of time spent walking and the pause durations differed significantly ($p<0.01$ and $p<0.001$, respectively), showing a larger fraction of time walking and shorter pauses while grouping. These findings are in accordance with previous reports [15], and are consistent with the known propensity of locusts to walk more and rest for shorter times while in a swarm [15–18].

However, our experiments also revealed a new effect of swarming. Comparing the isolation and re-isolation stages, we found that all the studied parameters differed significantly. Specifically, the fraction of time walking, speed, and walking bout duration were all higher in the re-isolation stage, while the pause duration decreased ($p<0.001$, $p<0.001$, $p<0.05$ and $p<0.05$, respectively). Furthermore, comparing the grouping and re-isolation conditions revealed that the walking bout duration increased significantly following re-isolation ($p<0.01$). The data for all other comparison combinations were found not to differ significantly. The increase in activity following re-isolation is surprising, and suggests that the marching behavior of locusts is not dictated by instantaneous or immediate interactions among individuals per-se. Rather, our findings indicate that the interactions with other marching locusts induce a switch to a new internal behavioral state, which outlasts the presence of the swarm. In accordance with our results, we term this internal state the "collective-motion-state".

In order to verify that the observed behavioral changes indeed represent a transient state, rather than a permanent behavioral modulation, a simple control experiment was performed. Following the re-isolation stage, the locusts were returned to their rearing cage (in high crowding conditions without collective motion), and tested the next day again alone in the arena (isolation condition). We found no significant behavioral difference between this latter isolation and the first isolation stage of the previous day (n = 6). Hence, the collective-motion state is transient. A second series of control experiments was performed in order to exclude potential time effects on the locusts' behavior due to the duration of the experiments. To this end, locusts were tested in isolation for three consecutive hours. The

kinematic analysis procedure described above was then performed separately on three 40 min segments of the 3 h tests and no significant differences were found (n = 6).

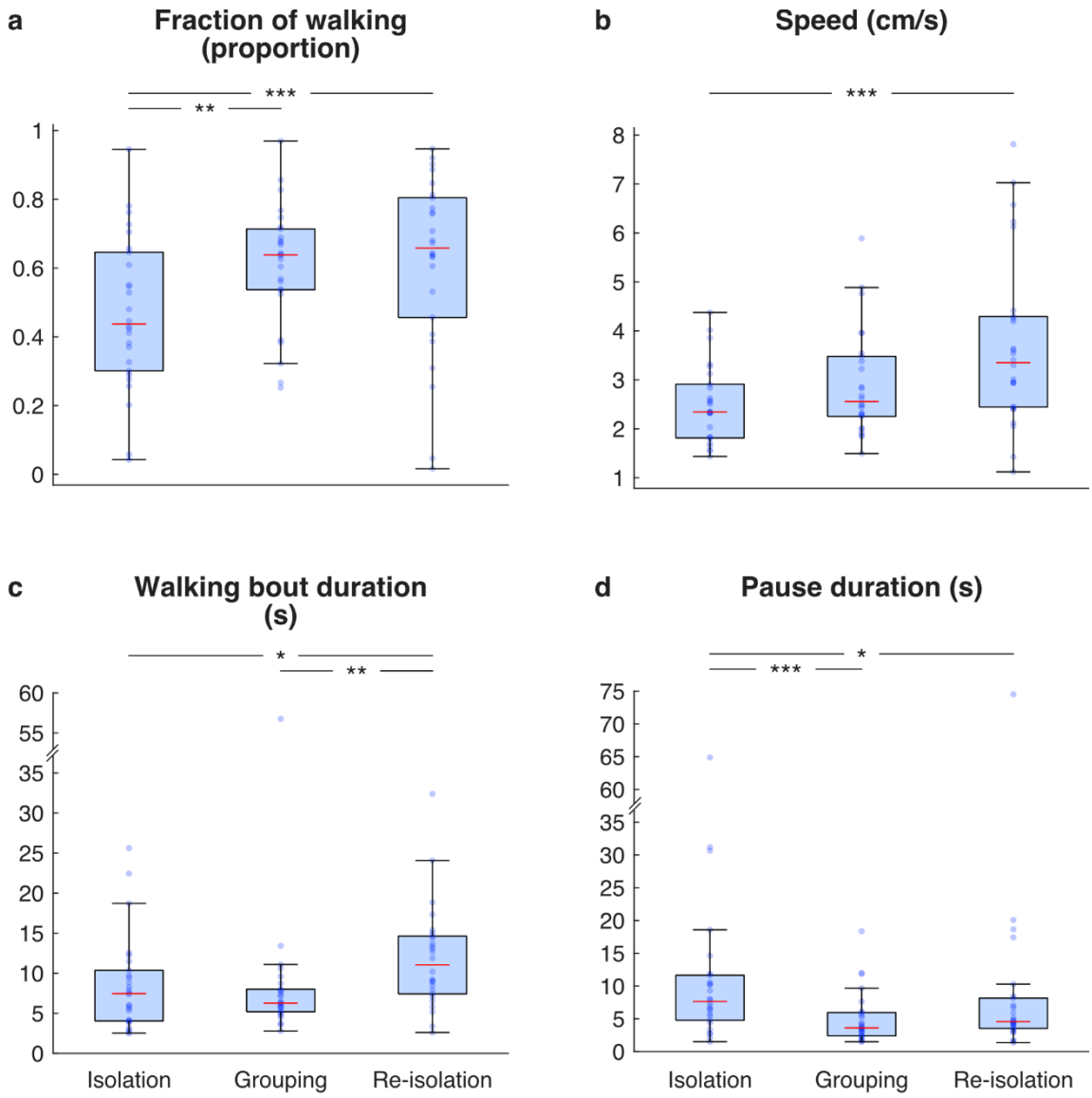

Figure 2. Kinematic changes throughout the three experimental conditions. (a) the fraction of walking, (b) the averaged walking speed, (c) the average duration of walking bouts, and (d) the average duration of pauses, of the traced animals in the isolation, grouping, and re-isolation stages. *p<0.05, **p<0.01, ***p<0.001.

145 **Consistency in individual behavioral tendencies**

146 Despite the major differences in behavioral kinematics among the three
147 experimental stages, we found that individual behavioral tendencies persisted. The fraction
148 of walking, speed, and pause durations all showed high within-individual correlation across
149 the three stages. The walking bout durations, however, were significantly correlated only
150 between the isolation and grouped stages (see Table 1 for numerical details).

151 The increased values of this latter parameter upon re-isolation may reflect a
152 mechanism that is activated when an animal finds itself away from the group, and overrides
153 its prior behavioral tendencies. To test this hypothesis, and further explore the effects of the
154 walking bout duration on the ability (and efficiency) of locusts to swarm, the effect of the
155 different measured kinematic parameters was further studied using a simplified computer
156 model.

157 **Modeling and simulations**

158 The experiments described above demonstrated that individual locusts introduced
159 into a collectively moving swarm undergo a switch into a distinct internal socio-behavioral
160 state. However, whether this change confers a benefit on the swarm formation and
161 maintenance, and if so, of what kind, remained unanswered. To explore this aspect of the
162 collective-motion-state, we developed a model that simulates locust swarms, in which
163 individual kinematics and population size could be manipulated. We focused ,in particular,
164 on the walking bout duration, as this specific parameter seems instrumental in
165 differentiating the locust behavior after re-isolation (Table 1).

166 To simulate swarms, we used a simplified agent-based model in a square domain
167 with periodic boundaries (size: 10 x 10). Agents were designed as rectangles with a circular

168 receptive field around their center. The agents' kinematics were programed to resemble
169 that of locusts, i.e. to move in an intermittent motion (pause-and-go) pattern: at every step
170 of the simulation, each agent made an individual decision whether to walk or stop, based on
171 its current state (walking or stopping) with predefined probabilities (p_W and p_S, respectively).
172 While moving, the speed was constant. The individual direction of movement was allowed
173 to change only when an agent changes its state from stopping to walking [16]. An agent's new
174 direction was a weighted sum of its own direction (inertia), the direction of other agents in
175 its visual field, with a short memory (see [19] for the importance of memory in pause-and-go
176 simulations), and noise. See Methods for details and Table S1 for parameter values.

177 The spatial and temporal scales of the model were set as follows: 1 cm was
178 considered as a distance of 0.1 in the simulated arena, and 1 sec corresponded to 1
179 simulation step. Thus, we fixed the model dimensions to correspond with real locusts' size
180 and movement parameters. The size of agents (0.1 x 0.4) maintains the proportions of 5th
181 larval instar locusts. Additionally, the visual range of 1 radius, ten times larger than the
182 agent's rectangle width, represents the proportional visual field of locusts (range just under
183 10 cm [16]). To adjust the temporal scale, an average stopping duration in the model was set
184 to be about 3 steps (by setting p_S to be 0.67; see Fig. S1), parallel to the median of 3 second
185 pause duration found in the grouping condition in the empirical experiment (Fig. 2d). The p_W
186 values were set to range between 0.6 and 0.92, making the walking bout durations
187 comparable with the empirical data (compare Fig. 2c and Fig. S1). Finally, the speed was set
188 to 0.25, corresponding to the typical swarming speed shown in Fig 2b.

189 In order to study the effect of individual bout duration on the swarm, we
190 concentrate on the effect of swarm size, N, and on p_W, describing the propensity of animals

to keep walking. We evaluated the effect of these parameters on four statistics that characterize collective motion in the swarm:

1. The order parameter (the size of the average direction vector).
2. Spread (the average distance between all pairs).
3. The number of neighbors (within the field of view).
4. Regrouping time (the average number of steps it takes an agent that has lost all other agents in its receptive field to re-obtain at least one neighbor).

Statistics were averaged over all agents and simulation steps. In order to allow comparison with our experimental findings, we focus on relatively small swarms (up to 40 agents). Fig. 3 shows the median value over 40 independent repetitions for each parameter set. We found that the order parameter increases both with p_W and with the number of agents, N (Fig. 3a). The spread shows a more complex dependence on p_W, depending on N (Fig. 3b). For small swarms, large p_W leads to a smaller spread, but the trend is reversed for large swarms, showing a smaller spread with small p_W. The number of neighbors (NN) is decreases with p_W but increases with N (Fig. 3c), and the slope is much larger for large swarms. For small swarms, the NN is small and practically independent of p_W. The regrouping time varies significantly with swarm size (increasing by a factor of up to 9) and is mostly independent of p_W for large groups (Fig. 3d). However, small groups (up to 20) reduce their time to regroup with higher p_W. This result makes sense assuming that isolated individuals perform a random search until they join a new cluster. Because agents only change direction when starting to move, the length of walking segments between rotations increases with p_W.

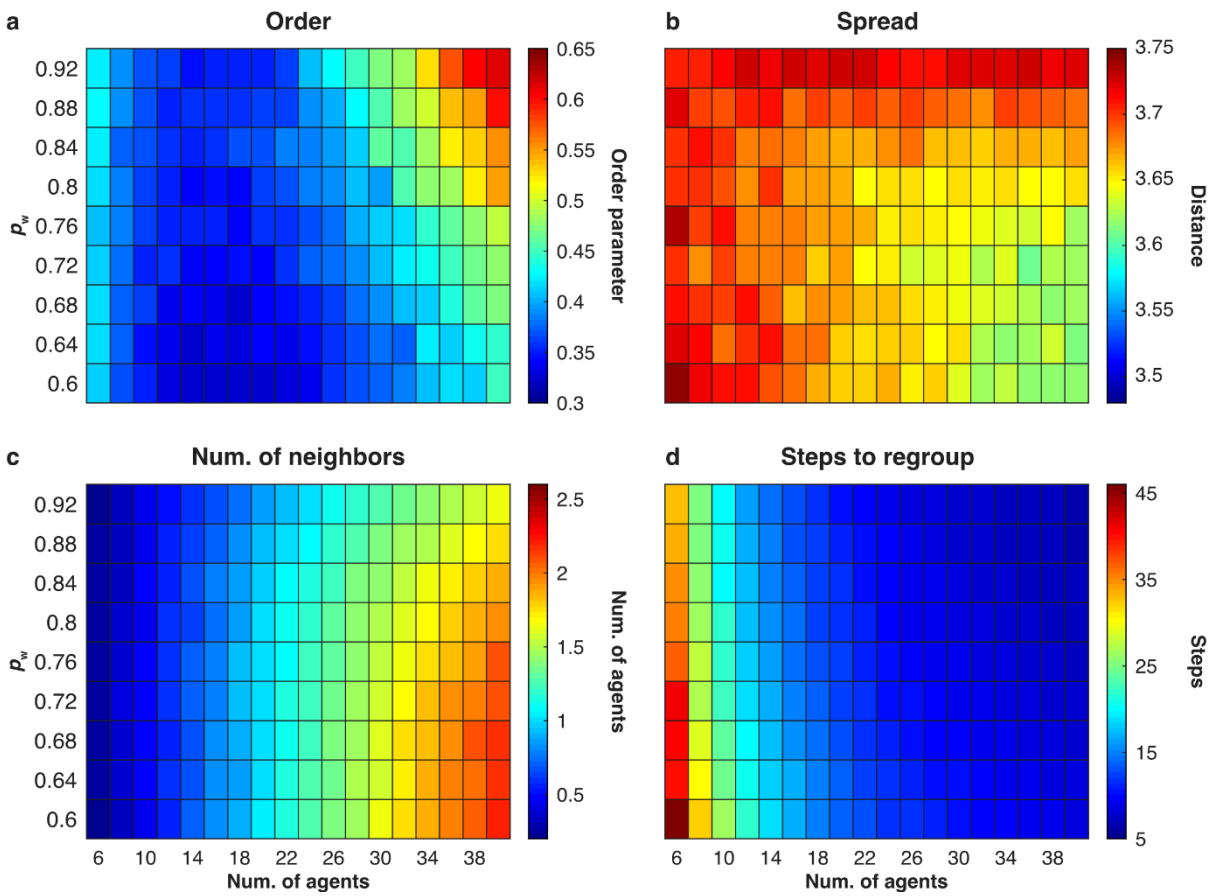

Figure 3. The influence of different p_W values on the collectivity parameters of simulated swarms. (a) The order parameters, (b) spread , (c) the average number of agents within each agent's visual field , and (d) the average number of steps to regroup, for different p_W values (rows), and different group sizes (columns). Each colored box represents the median of 40 simulations.

Fig. S2-S6 show a detailed sensitivity analysis, demonstrating the differences obtained when changing individual parameters. This analysis shows that, despite some quantitative differences, our qualitative observation that the propensity of animals to walk (modelled by the parameter p_W) has a different effect on small and large groups, holds in a wide range of parameters. However, some parameters do show a notable effect on the collective dynamics: increasing the noise level substantially (Fig. S3) may completely destroy collectivity, leading to a small order parameter. This observation is related to the well-known phase transition in Vicsek-like models [20]. In addition, several physical parameters,

227 such as the visual range (Fig. S4), speed (Fig. S5), and the size of agents (Fig. S6) have a
228 partial influence on some of the statistics. It is therefore important that such parameters are
229 assigned realistic values, as measured from experiments.

230 Overall, these findings indicate that large p_W values, namely, longer walking bouts,
231 increase the chances that low- density groups will stay together (smaller spreads, shorter
232 time to regroup). However, for larger swarms, smaller p_W – shorter walking bouts – is
233 favorable (more neighbors, smaller spread and a marginal cost in the order parameter and
234 regrouping time). Therefore, if aggregation is essential to individuals, our results suggest
235 that it might be beneficial for an agent to change its p_W as a function of the group size.

236 **Sociality-dependent p_W**

237 Several previous theoretical models of collective motion have found that adaptable
238 interactions, i.e., interactions that can change according to the local environment, can
239 improve the cohesiveness and performance of swarms [21–23]. Accordingly, in light of our
240 experimental results, we sought to study the effect of the individual's ability to change its
241 walking properties. To this end, we introduced an experience-dependent p_W to the
242 simulated agents (Fig. 4). The p_W could change during a simulation as a result of the agent's
243 receptive field of view in the last simulation steps. For simplicity, we assume linear
244 dependence (Ariel et al., 2014b). Biologically, such a dependence may be due to the way
245 locusts perceive their surrounding: many locusts walking in the same direction around one
246 locust would affect it more than only a few. To be precise, denote by $\bar{n}_i(t)$ the average
247 number of neighbors of a focal animal i in the previous k (memory length) steps before time
248 t, the probability that animal i keeps walking, $p_W(i,t)$ was given by

$$p_w(i,t) = \begin{cases} 0.92 - \alpha\bar{n}_i(t), & 0.92 - \alpha\bar{n}_i(t) \geq 0.6 \\ 0.6, & 0.92 - \alpha\bar{n}_i(t) < 0.6 \end{cases}$$

where α is a constant determining the slope of the linear effect ($\alpha = 0.1$ in Fig. 4). As can be seen, p_W was ranged between 0.6 and 0.92, in order to avoid phlegmatic behavior or unnatural hyperactive of the agent (respectively). See Fig. S7 for the obtained p_W values and walking bout durations in those simulations.

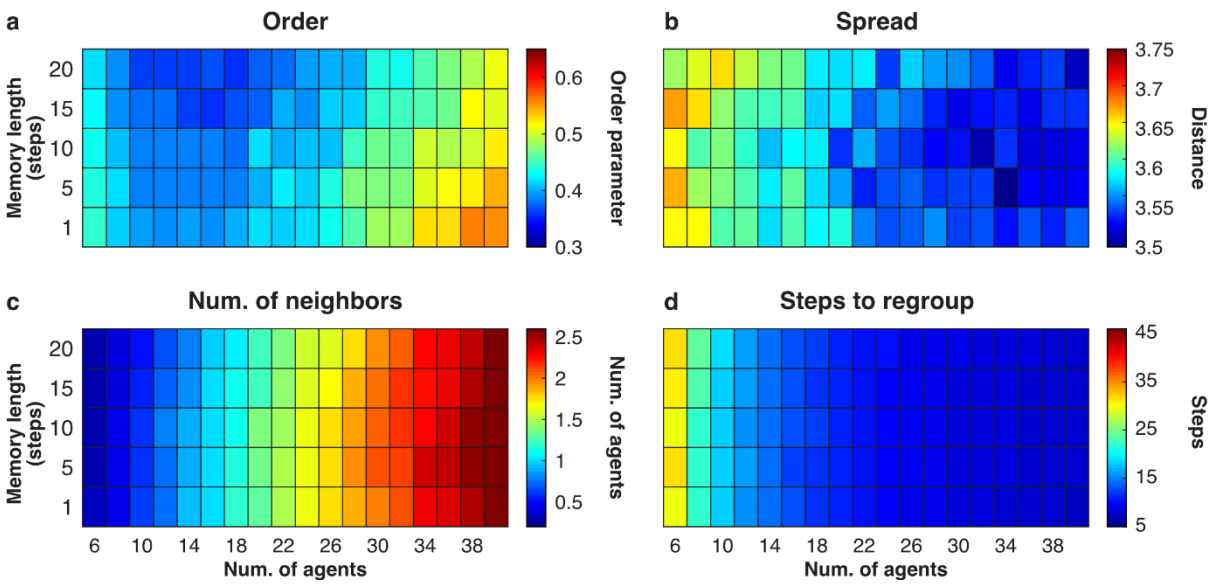

Figure 4. The influence of adaptive p_W values with different memory lengths on the collectivity parameters of simulated swarms. (a) The order parameters, (b) spread , (c) the average number of steps to regroup, and (d) the average number of agents within each agent's visual field, for different memory length (rows) and group sizes (columns). Each colored box represents the median of 40 simulations.

This conditional individual behavior was found to minimize the spread and regrouping time, while maximizing the agent's number of neighbors for almost all swarm sizes (Compare Fig. 3 and 4). This conditional behavior, however, did not increase the order parameter above that in the fixed p_W case (Fig. 4a).

We tested different memory lengths that affected the adaptive p_W. The average p_W did not change dramatically (Fig. S7b) but did result in higher variance across the simulation

267 steps – the shorter the memory was, the higher the step-to-step p_W fluctuations were (Fig.
268 S7c). This behavior is expected, as the sensitivity to the current situation of the agent
269 decreased with memory length. Fig. 4 shows that while the order and spread favors short
270 memory, the NN and steps to regroup were largely indifferent to it.

271 Overall, our simulations show that increasing the walking bout duration when an
272 agent finds itself alone has a beneficial impact on its chances of meeting other agents and
273 maintaining high group density. In regard to the empirical results, our simulations suggest
274 an instrumental role for the increased bout duration in regrouping when an animal is
275 isolated after swarming.

276

277 **Discussion**

278 Our findings reported here suggest that, in locusts, the sensory-motor act of
279 collective motion is accompanied by an internal state of the individual locust – a collective-
280 motion-state that is manifested in specific behavioral kinematics. This state is induced by
281 the experience of synchronous, collective marching. In turn, it has an important role in
282 maintaining the integrity and consistency of the swarm. Below we discuss several key
283 aspects and implications of this finding.

284 In a recent study [15], we have introduced a comparison between the walking behavior
285 kinematics of individual gregarious (crowded-reared) locusts in different social (density)
286 contexts. Our reported findings are reconfirmed and further elucidated here by the results
287 of the initial isolation and grouping stages in our experiments. The novel idea posited here is
288 that these differences represent and are induced by not only the spatially and temporally
289 immediate social environment, and the instantaneous local interactions among locusts, but

290 also by the effects of an internal state induced by the general experience of collective
291 motion. A fundamental aspect of the concept of the collective-motion-state arises from our
292 findings related to its persistent effect in time: Upon re-isolation, the individual locust
293 adopted behavioral kinematics that critically differed from that in the first experimental
294 stage (initial isolation). We also showed that, as expected, the collective-motion-state is
295 transient. If the locust does not experience collective motion for some time, and is then
296 isolated once more, it loses the unique walking-related kinematics it previously adopted in
297 response to the collective motion, i.e. the internal collective-motion-state.

298 The individual locusts in our experiments retained the variability observed among
299 them (see also Knebel et al., 2019), while demonstrating a second layer of variability or
300 plasticity upon experiencing collective motion, when entering the collective-motion-state.
301 Considerable research has been devoted in recent years to understanding the effect of
302 variability among individuals on the group's collective behavior, both experimentally—
303 ranging from bacteria to primates [24–33]—and theoretically [34–40]; (see [41–43] for recent reviews).
304 The interactions between variability in specific aspects of the individuals' behavior and
305 group-level processes were found to be complex and, moreover, bidirectional (e.g. [15]).
306 Variability among individual animals was found to have important consequences for the
307 collective behavior of the group (e.g. [44,45]).

308 However, beyond the variability among the individuals composing a group,
309 variability is also expected in the behavior of the individual animal over time, as it
310 experiences changes in environmental and social conditions. The swarm (or flock, shoal,
311 herd, etc.) is a heterogeneous entity, moving in a heterogeneous environment. The
312 individual is bound on occasion to find itself in different locations within the swarm (leading

edge, at the outskirts, trailing), and it may also find itself separated from the group by natural obstacles (vegetation, rocks and boulders). It is essential for the robustness and consistency of the swarm that throughout these changing conditions, the behavior of the individual will adapt accordingly, such as to be appropriate for the changing context. For example, if temporarily separated from the core of the swarm, a locust's walking kinematics should change to support rapid reunion with the group, as reported in both our experimental and simulation findings (e.g. increased fraction of walking and duration of walking bouts). If previously naïve to collective motion, that individual's kinematics would however be disadvantageous, or even hinder the formation of a swarm.

In [17], the authors suggest that behavioral variability can be explained by the existence of two internal states. Studying single locusts in isolation for eight consecutive hours, they have observed changes in behavioral kinematics that were suggested to result from "internal state behavioral modulation". The observed variations, however, were merely attributed to changes in "starvation/satiation state", i.e. as the locust becomes starved, it changes its walking behavior, searching more vigorously for food. Moreover, they conclude that animals continually switch between the two states on a scale of minutes. The collective-motion-state reported here is, of course, a very different type of internal behavioral state, which is strongly involved with the locust past and current social environment. It may be viewed as a form, or a manifestation of a social carry-over effect [46], where a social environment experienced by a focal individual affects aspects of its locomotion behavior at a later, non-social context. As noted, however, the change in behavioral state described here is induced by collective marching, i.e., a particular mode of social interaction, rather than by aggregation or being around other conspecifics per-se.

336 Moreover, as our simulations show, the enhanced marching displayed in the re-isolation
337 stage is advantageous for maintaining collective swarming - it is still much related to the
338 social context rather than carried-over to a non-social one.

339 It should be stressed once again that the current study focused on gregarious,
340 crowded-reared locusts only. The described behavioral states should not be confused,
341 therefore, with the well-known and much researched locust density-dependent phase
342 polyphenism [10,11]. Collective motion is limited to the gregarious, swarming and migrating
343 phase. Accordingly, all our experimental animals were taken from our gregarious breeding
344 colony, maintained in crowded conditions for many consecutive generations. In their
345 breeding cages, mostly due to the physical constraints and abundance of food, despite
346 experiencing high density the locusts very rarely, if at all, demonstrate collective motion.
347 Thus, they adopted the collective-motion-state only upon experiencing and taking part in
348 collective marching within the experimental arena.

349 The reported collective-motion-state is also in accord with the overall daily
350 behavioral changes of marching locust swarms. The swarm will spend the night (as well as
351 times of low temperature or other unfavorable climatic conditions) roosting among the
352 vegetation. Upon suitable conditions, after a period of feeding, the locusts will initiate
353 marching – highly synchronized, collective motion. Frequently, when temperature becomes
354 too high around noon, or when dusk arrives, the swarm will again switch to feeding and
355 roosting. These daily patterns call for corresponding changes in the internal behavioral
356 states of the individual locusts and mostly a dedicated collective-motion-state.

357 In the current work we are cautious in talking about *behavioral* states. Although this
358 is beyond the scope of this study, it is clear that these behavioral states represent

359 physiological states. With some confidence, we can speculate about the nature or the
360 physiological mechanisms involved in the demonstrated behavioral states. Behavioral
361 plasticity in locust behavior has been attributed to various second messengers or
362 neuromodulators, or to the balance among them. Most notable are the biogenic amines
363 (e.g. Serotonin, a prominent bio-amine, was recently reported to inhibit walking behavior in
364 *Drosophila* [47]). Hence, it may well be that the (spatial and temporal) immediate social
365 environment affects biogenic amine levels, and these in turn modulate the walking-related
366 behavioral kinematics manifested in the different behavioral states.

367 Another candidate that may be involved in the collective-motion-state is the locust
368 adipokinetic hormone (AKH). AKH is a metabolic neuropeptide principally known for its
369 mobilization of energy substrates, notably lipid and trehalose, during energy-requiring
370 activities such as flight and locomotion, but also during stress (e.g. [48]). It is well accepted
371 that the metabolic state affects the level of general activity of an organism, and AKHs are
372 reported to stimulate locomotor activity, either directly by way of their activity within the
373 central nervous system (e.g. [49]), or via octopamine – a biogenic amine with ample
374 behavioral effects [50,51].

375 Furthermore, as noted, we have demonstrated here an extended effect of the
376 experience of collective motion. Hence, learning and memory-related mechanisms would
377 also seem to be involved. Again, previous work may suggest some candidate molecules and
378 pathways, including cGMP-dependent protein kinase (PKG), and Protein Kinase A [52–54].

379 Last, as noted, solitarious phase locusts lack the capacity to demonstrate collective
380 motion, and thus also the collective-motion-state. Accordingly, they differ from gregarious

381 locusts in all the above physiological pathways (bioamines: e.g. [10,55,56]; AKH: [57,58]; PKG: [53];
382 PKA: [54]. An in-depth investigation of the development of gregarious-like states in solitary
383 locusts should prove to be very enlightening.

384 A central question is whether a collective (herd, flock, or swarm) is merely a sum of
385 its parts, or a new entity. Most related studies have perceived collectivity as a self-emergent
386 phenomenon, suggesting that new dynamics and behavior are the result of intricate, multi-
387 body, typically non-linear interactions (e.g. [20,59]). One hidden assumption underlying this
388 perception, is that individuals remains inherently unchanged when isolated or in a crowd.
389 Even studies of heterogeneous swarms, in which conspecifics may differ from each other,
390 still assume consistency in the properties of the individual over time. This is essentially a
391 physical point of view, in the sense that agents/individuals possess certain properties that
392 determine their behavior across a range of situations. Thus, the collective motion is an
393 emergent property that builds up in particular contexts, such as a sufficiently high local
394 density of animals. This point of view allows, among others, extrapolation from experiments
395 with one, two, or a few animals to large swarms (e.g. [30]).

396 Our findings reported here suggest a fundamentally different point of view. We
397 perceive the sensory-motor act of collective motion as accompanied by an internal state – a
398 collective-motion-state that is manifested in specific behavioral kinematics. This state is
399 induced by the experience of synchronous, collective motion. Most importantly, it is *not*
400 induced by spatial aggregation alone. Collectivity, therefore, is not just self-emerging.
401 Rather, the collective-motion-state has an important role in maintaining the integrity and
402 consistency of the swarm. The robustness of the swarm is also a major challenge and

403 requirement in swarming robotics, making the current novel insights applicable and even
404 important also to this emerging field.

405 In the case of locusts, our far from complete understanding to date of this
406 phenomenon is also proving crucial for human well-being and even survival, as evident from
407 the current devastating locust situation in large parts of Africa and Asia [60]. Much scientific
408 attention has been dedicated to the perception, decision-making, and individual kinematics
409 of locusts in a swarm. These efforts have led to various models that attempt to explain the
410 collective behavior on the basis of local interactions among the individual locusts (see [7] for
411 review). The current study is, to the best of our knowledge, the first to provide a more in-
412 depth focus, by including the internal state of the individual locust as an important factor in
413 dictating its behavior, and in turn affecting the maintenance and the properties of the
414 swarm.

415

416 ## Methods

417 **Animals**

418 The empirical experiments were conducted on gregarious desert locusts,
419 *Schistocerca gregaria* (Forsskål), which are bred at Tel Aviv University, School of Zoology.
420 The animals are kept at high density: over 100 animals in cages of 60 l, under a 12 h:12 h
421 light/dark regime, a temperature of 30℃, and 30-65 % humidity. Animals are fed daily with
422 wheat seedlings and dry oats. All experimental locusts were the offspring of many
423 generations of gregarious locusts reared in these conditions.

424 **Experimental setup**

425 The ring-shaped arena comprised blue plastic walls (60 cm diameter and 55 cm high)
426 with an inner central cone dome (30 cm diameter). The bottom of the structure was
427 covered with a thin layer of Fluon (Whitford Plastics Ltd., Runcorn, UK), in order to prevent
428 locusts from climbing. A white paper covered the floor of the arena and was changed before
429 each experiment. Light sources around the arena lit its floor evenly. A video camera (Sony
430 FDR-AXP35: 4K Ultra HD) was positioned above the arena and filmed the locusts throughout
431 the experiment.

432 **Monitoring the locust movements**

433 Prior to the experiments, each of the experimented locusts was tagged with a
434 unique barcode glued to the dorsal side of its prothorax using a drop of Epoxy glue. Using
435 the video footage at 25/3 frames per second, the trajectory of each animal was
436 reconstructed offline using BugTag software (Robiotec Ltd., Israel). To this we added a
437 custom-designed multiple-target tracking and a trajectory-smoothing method (for details:
438 Ariel et al., 2014b). briefly, segments in which tags were not identified (less than 5 cm or 25
439 s) by the system positions were interpolated. We thus obtained the positions of the tags'
440 center of mass relative to the arena center, at a final resolution of 2-3 pixels (ca. 0.5 mm),
441 for about 99% of the video frames. In order to reach 100% recognition, we completed the
442 analysis manually for the remaining frames. In three out of the 26 experiment the tracking
443 system failed to recognise 1-2 animals in the grouping stage. Therefore, the order
444 parameter was calculated based on the recognized animals only. These miss-identifications,
445 which only slightly affect the order parameter calculated in the grouping stage, have no

446 influence on any of the kinematic statistics in any of the stages (that only describe the focal
447 animal).

448 **Experimental procedure**

449 In the morning of each experiment day, 10 locusts were taken from their cage and
450 individually tagged. Thereafter, one locust was randomly chosen and placed in the arena
451 alone for one hour. The other 9 locusts were then added for an additional hour; after which
452 they were removed. The remaining locust was then filmed for another hour.

453 **Analyzed parameters**

454 Following the recognition process, we analyzed the middle 40 min of each of the 3 hours
455 of the experiment using MATLAB (MathWorks, Natick, MA, USA). The following parameters
456 were calculated:

457 1. Order parameter – the absolute value of the average direction of movement of the
458 walking animals (clockwise denoted as -1 and anticlockwise as +1), averaged for all
459 the frames.
460 2. Fraction of walking – the number of frames an animal walked divided by the total
461 number of analyzed frames, averaged over all experimental animals.
462 3. Detection of walking bouts – a walking period is defined as walking at a speed above
463 0.25 cm/sec for more than one third of a second.
464 4. Speed – averaged for all experimental animals and analyzed frames when the animal
465 walked and the speed differed from zero.
466 5. Walking bout duration – the time an animal moved continuously between one pause
467 to the next, averaged for all walking bouts and experimental animals.

6. Pause duration – the time an animal did not walk between one walking bout and the next, averaged for all pauses and experimental animals.

Statistical analysis

All statistical tests were conducted with MATLAB. To compare the order parameters, Wilcoxon rank-sum test was used. To compare among the three stages, Friedman Test was used. Since all comparisons were significant according to the latter test, Bonferroni multiple comparison post-hoc test accompanied each Friedman Test, in order to reveal the significantly different groups. The p values noted are of the Bonferroni multiple comparison post-hoc test. Correlations were conducted using Spearman's Rank Correlation test.

Modeling

We employed a two dimensional model comprising N agents moving at a fixed speed v in a square domain of linear size L and periodic boundaries. Position and heading (direction) of each rectangle center are denoted by $x_i(t)$ and $\hat{v}_i(t)$, respectively, where $x_i(t) \in [0, L]^2$ and $\|\hat{v}_i(t)\| = 1$. In addition, $w_i(t)$ is a Boolean variable that denotes whether at time t, agent i is moving ($w_i(t) = 1$) or pausing ($w_i(t) = 0$). In order to take into account the physical size of animals, we assume that each agent is a rectangle with sides $a_1 \times a_2$.

In each simulation step, the position of moving agents changes by amount v in the direction $\hat{v}_i(t)$,

$$x_i(t+1) = x_i(t) + v w_i(t) \hat{v}_i(t) \quad (mod\ L).$$

The new velocity of agent i depends on three terms: i. Inertia (its own direction); ii. the direction of other agents in its receptive field (see below), possibly with a memory; and iii. a random vector with unit norm $\xi_i(t)$. The receptive field view is a circle around $x_i(t)$. Unlike previous models, we assume that an agent sees all other agents whose rectangle is within a distance *r* from $x_i(t)$, and not merely the center of the rectangle. Denoting the four corners of the rectangle describing agent *j* as indexes $c_i^1(t), \dots, c_i^4(t)$, we define the set of observable neighbors of agent *i* at time *t* as

$$I_i(t) \coloneqq \{j: \left\|x_i(t) - c_j^k(t)\right\| \leq r \text{ for some } k = 1 \dots 4\},$$

the average direction in the receptive field at time t, is $\hat{u}_i(t) = u_i(t)/\|u_i(t)\|$, where

$$u_i(t) = \left(\frac{1}{n_i(t)}\sum\nolimits_{j\in I_i(t)} \sin(\hat{v}_j), \frac{1}{n_i(t)}\sum\nolimits_{j\in I_i(t)} \cos(\hat{v}_j)\right).$$

Here, $n_i(t)$ is the number of elements in $I_i(t)$.

Overall, the new heading $\hat{v}_i(t+1)$ is the weighted average of four terms: inertia, direction of moving intersecting rectangles at time t, the average heading of moving intersecting rectangles at time $[t-5 \dots t-1]$, and independent random vector with unit norm $\xi_i(t)$. Let the set of the weights be $[q_1 \dots q_4]$.The updated weighted averaged direction, is, therefore, $\hat{v}_i(t) = v_i(t)/\|v_i(t)\|$, where

$$v_i(t+1) = q_1\hat{v}_i(t) + q_2\hat{u}_i(t) + q_3\frac{1}{5}\sum_{s=1}^{5}\hat{u}(t-s) + q_4\xi_i(t)$$

In Fig. 3 and 4, the weights were set to $Q = [0.2, 0.3, 0.2, 0.3]$.

Switching between walking and pausing is determined as follows. Assuming exponential distributions of moving and pausing durations, the probability to proceed moving is given by a single parameter, p_w, describing the probability per-step of an agent to continue walking, $p_w = P(w_i(t+1) = 1 | w_i(t) = 1)$. Similarly, $p_s = P(w_i(t+1) = 0 | w_i(t) = 0)$ is the probability per-step that a stopping agent will continue stopping. In the simulations which introduce adaptive p_w, the latter was updated each time "walking" started, and ranged between 0.4 and 0.8, as a linear function of the average number of neighbors , $\bar{n}_i(t)$ in the last k. steps.

The following parameters were calculated over 200-1000 stimulation steps. Brackets denote averaging over all analyzed frames.

1. Order: $\theta(t) = \frac{1}{N} \langle \left\| \sum_{i=1}^{N} \hat{v}_i \right\| \rangle$.
2. Spread: $S(t) = \frac{1}{N(N-1)} \langle \sum_{i=1}^{N} \sum_{j \neq i} \left| x_i(t) - x_j(t) \right| \rangle$
3. Number of neighbors: $n_i(t) = \frac{1}{N} \langle \sum_{i=1}^{N} I_i(t) \rangle$
4. Steps to regroup: the number of steps that passed from the first time an agent is alone ($n_i(t) = 0$) until it has a first neighbor ($n_i(t+s) > 0$).

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Author contributions

D.K., G.A., and A.A. designed the study. D.K. and C.S. performed the experiments. D.K. and C.S. analyzed the data. D.K. N.A. and G.A constructed the model. D.K. wrote the code, performed simulations and analyzed the data. D.K., G.A., and A.A. wrote the manuscript. All authors reviewed and approved the paper.

Competing interests

The authors declare that they have no competing interests.

Data availability

The data is available upon request.

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681

682

683 # Tables

	p-Value	***rho*-Value**
Fraction of Walking		
Isolation - Grouping	0.000	0.706
Isolation - Re-isolation	0.001	0.679
Grouping - Re-isolation	0.002	0.638
Speed		
Isolation - Grouping	0.001	0.679
Isolation - Re-isolation	0.010	0.561
Grouping - Re-isolation	0.002	0.636
Walking bout duration		
Isolation - Grouping	*0.002*	0.628
Isolation - Re-isolation	*0.148*	0.391
Grouping - Re-isolation	*0.354*	0.314
Puase duration		
Isolation - Grouping	0.007	0.580
Isolation - Re-isolation	0.003	0.612
Grouping - Re-isolation	0.000	0.708

684

685 **Table 1. Correlation values between kinematic parameters throughout the three experimental conditions.** The
686 fraction of walking, the averaged walking speed, the average duration of walking bouts, and the average
687 duration of pauses were tested for correlation across the three experimental conditions: (1) isolation, (2)
688 grouping, and (3) re-isolation. The underlined values mark significant correlations.

689