on such neglected tetraspanins will likely help to identify novel types of interaction partner, which may help us to understand a possible universal tetraspanin function. Another area that requires more study is structural research because a complete structural model is available for only one tetraspanin so far. We should generate more examples and additionally study tetraspanin complexes at atomic resolution. Finally, what is the role of the large intracellular domains that are present in a handful of tetraspanins, or of alternatively spliced forms that lack transmembrane segments? In fact, it appears as if the tetraspanin field is just at its beginning and many exciting discoveries are yet to be made.

Where can I find out more?

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Primer

Bird flocks

Steven J. Portugal

Birds often fly in aggregations and formations, sometimes gathering in vast and impressive numbers. The precise function of these flocks has been the topic of much debate. The many theories proposed to explain aerial flocking are probably not mutually exclusive. Such proposed functions include protection against predators, the sharing of orientation or information, and for certain species, energy conservation.

Flapping flight is the most energetically demanding mode of locomotion in vertebrates. How birds assemble and the dynamics within the group have significant implications for individual energy expenditure within the flock. Bird flocks are a particularly exciting study system due to their dynamic nature, the speed at which events and decision making need to occur, and the potential for collisions and injury. For example, herds of ungulates seemingly make group movement decisions through facing the same direction, or through slow directional movements aimed to illicit a response from other herd members. However, such signals pertaining to group movement and direction cannot be as subtle or slow for animal groupings moving in air, due to the risk of collision between flock members, or falling out of the sky. Birds can only fly so slow before they are no longer creating enough lift to keep themselves airborne. While this speed will vary greatly depending on the size of the bird, all birds will eventually stall below a certain minimum speed. Therefore, for birds in a flock, decisions need to be made literally on the wing, and at a certain speed. Likewise, for all animals travelling in groups collisions or indecisiveness may lead to individuals being more easily picked out by predators. Many studies have demonstrated that individuals who are less well aligned to the group, slower to respond or choose the wrong direction are more likely to be preyed upon. When birds flock in the air they form either clusters or V-formations. These two flock types are thought to provide different benefits and disadvantages.

V-formation flight

For centuries, people have been fascinated by V-formation flight. Pliny the Elder noted that flocks of geese flew 'like fast galleys, cleaving the air more easily than if they drove at it with a straight front'. Since then, numerous ideas have been proposed to explain the function of these V-shaped flocks that are a such a familiar sight. The notion that these distinctive formations provide energetic savings for those individuals not leading the formation was based on applying fixed-wing aerodynamic theory — typically applied to aircraft — to bird flight, even though many birds flap their wings between four and seven times a second. This notion was based on the potential positive aerodynamic interactions that may be taking place between members of the flock. When a bird flies, the lift required for flight is achieved through a pressure difference between the top and bottom of the bird's wing. This pressure difference cannot be maintained beyond the wingtips, as there is no longer a surface to create the pressure gradient. As a result, the high-pressure under the wing flows around the tip, and inwards across the dorsal surface of the wing. This in turn forms a stream of air trailing from the wingtip and behind into the birds wake, which is commonly referred to as upwash (Figure 2). This upwash flows outboard of the wing, while there is a region of downwash more centrally behind the main body of the bird. Theoretical studies have demonstrated that, if birds position themselves optimally in a V-formation, they can take advantage of the upwash from the preceding bird to contribute to lift and reduce power requirements. Simultaneously, such positioning also benefits the birds by avoiding the region of downwash, flying into which significantly increases the cost of flight.

The key to capturing upwash and avoiding downwash was believed to be due to appropriate wingtip spacing (Figure 2). Wingtip spacing is defined as the distance between the centers of two birds minus their mean maximum wingspan. In theory, as wingtip spacing decreases, the induced power required for flight also decreases as the bird is flying in stronger upwash. Hence, there is an optimal wingtip spacing that maximizes the reduction in induced power requirements through optimizing upwash capture. The first theoretical analyses in 1970 suggested that birds could reduce the power requirements for lift by a factor of 2.9 by flying in optimal positions, and that this reduction in power could equate to an increase in flight range of approximately 70%.

The first empirical evidence that flying in V-formation provided an energetic benefit to the group came in 2001 using free-flying great-white pelicans (*Pelecanus onocrotalus*). Birds flying out of formation had higher heart rates (expending more energy), and higher wingbeat frequencies — a proxy for work rate — in comparison to birds flying in V-formation. Birds flying in formation also glided more, contributing to the observed lower heart rates, saving between 11 and 14% of energy through V-formation flight.

In northern bald ibis (*Geronticus eremita*), the mechanism was revealed by which upwash could be captured. These birds have a 'V-favored position' where they spend most of their time when flying in formation. For the ibis, this was approximately 1.2 meters behind the preceding bird, at an angle of 45°. The center of the most populated spanwise region that birds inhabited during flight was 0.9 meters, resulting in a wingtip overlap of approximately 0.12 meters. Somewhat surprisingly, this positioning matched the predictions of fixed-wing theory for maximizing the benefits of upwash, despite the fact that the birds were continuously flapping their wings. What were the birds doing in this position to capture the beneficial upwash from the wingtips of the individual it was following? It turns out that the process of capturing upwash is not a passive process — birds do not simply sit in an approximate region and hope to encounter some upwash; rather, they use a phase-lock mechanism to actively track the upwash from the bird in front throughout the entire wingbeat cycle. This feat is achieved through a process known as 'wingtip path coherence', where the follower bird flaps its wing in spatial phase with that of the bird it is flying behind.

Figure 1. Different flocking types observed in birds.
Top: a V-formation of lesser flamingos (*Phoeniconaias minor*; photo: Gerard Glacz). Bottom: a cluster of homing pigeons (photo: Tommaso Rada).

Wingtip path coherence occurs when the wingtips of two different individuals follows the same pathway through the air, with the following bird matching the path that the wingtip of the bird in front took. The wing of the following bird goes up and down, tracking the path through the air that the preceding bird left behind. Thus, the wingtip of each bird is at the same point in the flap cycle (e.g. at the top of a wingbeat), in the same physical location as the bird it was following, just not at the same time. The upwash generated from the wingtip of the preceding bird trails behind, going up and down along with the wingtip as the bird beats it wings, and the following bird makes sure that its wingtip path matches that of the bird in front, to maximize upwash capture throughout the whole flap cycle. In essence, the following bird benefits from the leading bird leaving a pathway of least resistance through the air — upwash — which, if the bird can track it, will result in reduced energy expenditure. This effect persists for approximately four meters behind the bird, before the upwash dissipates or becomes less predictable. Impressively, the following birds adapt when and how they flap their wings depending on how far back they are from the bird in front, to ensure the phase shift mechanism is maintained.

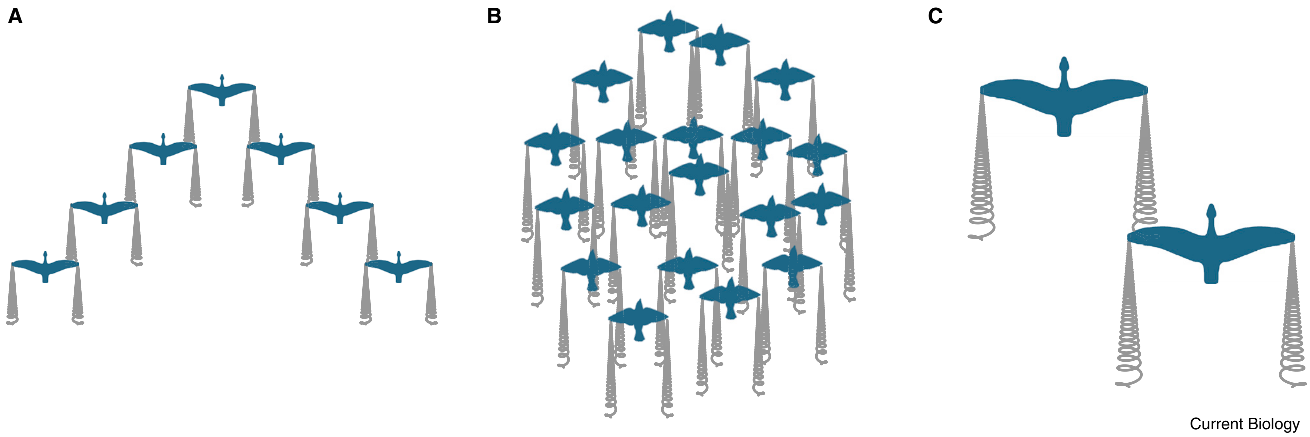

Figure 2. Simplified diagram of the theoretical positioning of wingtip vortices in two different flock types.
V-formation (left), cluster (center) and close-up of two birds within a V-formation (right). Grey lines depict crude positioning of wingtip vortices (not to scale), demonstrating the complexity of the local micro-scale aerodynamic environment in cluster flocks. The area behind each individual bird would be the region of downwash (not shown).

Wingbeat phasing had previously been identified in tethered flying locusts, where distance manipulations between a leading locust and a follower altered the phase patterns of their wingbeats. In locusts at least, the airflow is sensed by the animal's cephalic hairs, as covering these hairs resulted in the disappearance of any wingbeat coupling. In birds, the precise mechanism for how they sense the upwash and subsequently track it is not fully understood. Possible explanations include a positive feedback loop between flock positioning and work rate, or via filoplumes, mechano-sensitive feathers that might be able to sense changes in air pressure.

In contrast to the spatial phasing by birds in their V-favored positions, they display spatial anti-phasing of their wingbeats when they find themselves flying behind another individual. Spatial antiphasing results in the wingtip paths of the following bird no longer matching that of the preceding bird, and this change in timing and location of wingbeats could be interpreted as the birds evading the adverse effects of downwash.

If flying behind an individual is considered bad due to the detrimental effects of downwash, why do you sometimes see birds flying in a line, immediately behind one another? Such line formations are typically observed when larger bird species such as pelicans are flying low over the water or close to the ground; they are taking advantage of what is known as 'ground effect'. The airflow around the wing is modified by the ground/water, increasing lift and decreasing drag. Birds flying in ground effect can experience reduced costs of transport of around 15%, and a reduction of up to 35% in the mechanical power required for flight. Birds flying in ground effect are more commonly observed over water than land. To benefit from the aerodynamic interactions with the surface, the distance between said surface and the bird needs to be equal to or smaller than the bird's wingspan; flying so low over land would entail risk of collision. Airplane pilots have noted that flying close to a fixed surface and experiencing ground effect feels to them as though the aircraft is floating.

Cluster flocking

Cluster flocking is a loose term used to describe bird flocks that are somewhat of an indiscriminate blob with no apparent structure, at least to the human eye. In contrast to the largely positive aerodynamic interactions between members within a V-formation flock, flying in a cluster comes at a cost for most individuals. Homing pigeons, for example, increase their wingbeat frequency when flying close to or behind another bird, suggesting being at the front of the flock may be aerodynamically optimal. In general, higher wingbeat frequency offers better flight control and greater maneuverability, which in turn can reduce risk of collision. Birds are likely to constantly adjust to their flight course to contend with the airflow coming from the surrounding birds in front. With a variety of individual flight trajectories and wingbeat frequencies, airflow may be unpredictable and extensive (Figure 1,2). Thus, this increase in wingbeat frequency is likely to be a direct result of flying in a cluster and will come at an energetic cost.

Indeed, rather than an increase in work rate only observed when birds fly in a cluster, the simple act of flying with one other individual results in an increased wingbeat frequency of approximately 18% in pigeons. Birds are able to increase this frequency through a quicker upstroke, yet this amplified wingbeat frequency only results in a 3% increase in airspeed, suggesting a real cost to flying in a pair. Accompanying this surge in wingbeat frequency is a reduction in the oscillatory displacement (oscillation around a mean position) of the body of around 22%; the increased wingbeat frequency boosts the stability of the body and the head. This reduction in body and head movements might be needed in a cluster due to the increased requirement for visual stability. Increased visual stability will help to avoid collisions, maintain flock positioning, and keep an eye on flock mates. Interestingly, there is no evidence of wingtip path coherence in cluster flocking birds. This is likely to be due to the unpredictability of the upwash being created by the bird in front, coupled with the comparatively smaller size, meaning the effort of tracking the upwash is not counteracted by any potential energetic gains.

Additional costs to flying in a flock may come in the form of individuals compromising on their preferred flight speed to remain part of the group. Optimal solo flight speeds in birds — where mechanical power is minimal — is typically determined by body mass, with heavier birds having a faster optimal flight speed than lighter individuals. When groups of pigeons fly in a cluster flock, they are compromising on their preferred solo flight speed by up to 6% to remain part of the group; indeed, pigeon flocks fly at an intermediate of the solo speeds of the group members, suggesting speed averaging. Taken together, this suggests that, unlike in V-formation flocks, birds in cluster flocks do not gain aerodynamic benefit from flocking, and instead pay an energetic cost. Evidence for the impact of these increased flight costs comes from long-duration flights where, over time, individuals within a cluster begin to spread out. This suggests that the ultimate benefits afforded by tight clusters, such as increased vigilance, dilution effects, predator confusion and shared navigation, become outweighed by the proximate costs of flying close to a neighbor, necessitating greater distances between individuals.

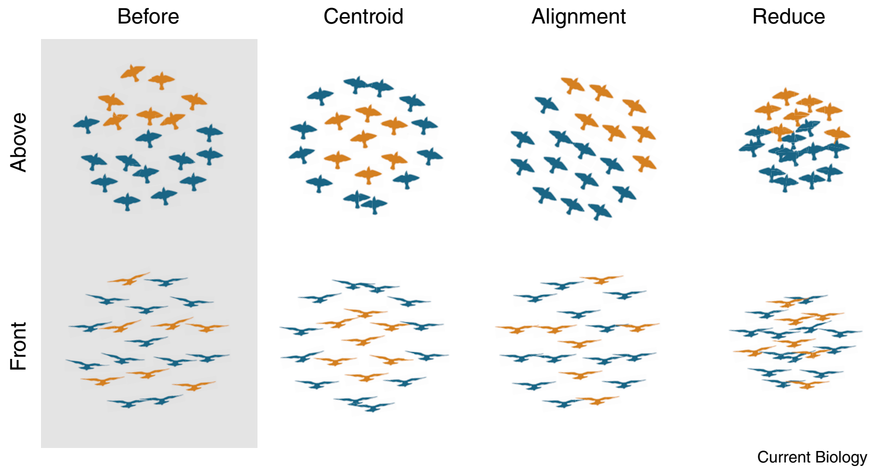

Figure 3. The role of individual phenotype in flock positioning within cluster groups.
Dominant birds (orange) may position themselves in the aerodynamical optimal position at the front of the cluster flock during periods of perceived 'safe' flight. When a threat presents itself, the flock can respond potentially in three different ways; head for the centroid, increase alignment, or decrease inter-individual distances. Dominant birds may manipulate positioning within a flock to their advantage.

Leadership in bird flocks

There are navigational leadership hierarchies in bird flocks, such that hierarchal positions are reflected in the weight of each individual's contribution to a decision-making process. Leadership can be based on a variety of factors, typically related to experience and knowledge, for example of a foraging location, a suitable roost site or a migration route. Leaders can be better, more dominant or older, more motivated due to hunger or chick-feeding requirements, quicker, or simply more 'stubborn'. It is thought that birds flying in a cluster flock typically employ the three interaction rules described in collective behavior to remain a coherent flock: move away from nearby neighbors that are getting too close; adopt the same direction as the individuals immediately around you; and avoid ending up being alone. Therefore, from the individual's perspective, there are three zones around them; the zones of repulsion, alignment and attraction. It is likely that the risk of collision is greater in cluster flocks in comparison to V-formations due to the characteristic higher flight speeds, greater maneuverability and smaller distances between individuals. The precise rules used by birds in V-formations to maintain structure, however, have not been studied in detail. More recently, a hybrid form of formation flight has been described in mixed-species flocks of shorebirds, termed a 'compound-V'. Compound-V formations are defined as an intermediate between cluster flocking and the classic V-formation shape. The result is a flock-type that casually resembles a cluster flock, but the aerodynamic interactions taking place between members are more akin to a V-formation flock type. It is likely that, as more flocking species are studied in detail, a greater spectrum of flock modes will emerge, as a binary categorization of cluster or V-formation is an oversimplification of a complex process.

Individuals in bird flocks

While many forms of group locomotion do not necessarily require a detailed level of individual recognition to successfully travel in a group, other varieties do. In V-formation flight, for example, differences in age and experience mean that specific individuals may preferentially lead, and these are typically adults, rather than juveniles. How do birds recognize each other during flocking events, and over what time periods can individuals remember one another? These questions will be particularly pertinent for groups in which reciprocal cooperation is prevalent, such as that seen during V-formation flight. In juvenile northern bald ibis, the amount of time a bird spends leading a formation is strongly linked to the time that individual itself can profit from flying in the wake of another bird. Moreover, birds match the time they spend in the wake of each other by frequent pairwise switches of the leading position, suggesting that birds in a V-formation are cooperating through using a form of direct reciprocity.

Cooperation in V-formation is likely explained through the stability of certain groupings (e.g. family groups during migration) and the immediacy of reciprocation opportunities. In cluster flocks, their more dynamic nature — a system more akin to a fission–fusion society — means that such opportunities for immediate reciprocation might be sparse: an individual you just helped out might be never seen again. Nevertheless, if specific keystone individuals have more weight in decision-making processes, it is likely that some form of individual recognition is a prerequisite, although other factors such as flight speed, trajectory and positioning may be used instead. It is known that sheep can remember the faces of other sheep for years. This would suggest that both recognizing and remembering individuals are strong selective pressures for other flocking species. Understanding how, and the duration of which, individuals in bird flocks recognize each other would be a fruitful avenue for further exploration.

Bird flocks present an exciting opportunity to study social interactions in a dynamic moving system, yet integrating individual-based personality traits with physiology, energy expenditure and aerodynamic trade-offs has been relatively understudied. In cluster flocks, flying at the front with large distances between flock members is the optimal aerodynamic situation. However, during predator attacks such a flock structure may be far from ideal. In response to a potential threat, birds may cluster more, increase alignment to maximize predator confusion, or adopt a 'head to the flock centroid' approach — resembling the selfish herd theory — as has been observed in terrestrial mammals (Figure 3). Heading to the centroid can mean better protection from attacking predators, using your flock mates as a barrier between oneself and the predator. Which individuals would potentially move to position themselves within the safety of the cluster center is not clear, and it is possible that individual personality traits may play a role in such decisions and movements. Dominant birds, for example, may take advantage of the aerodynamically optimal frontal positions within the cluster when not feeling vulnerable, but then move themselves towards the center of the flock when under threat, thus essentially dictating the structure of the flock (Figure 3).

A further component of flock dynamics is the impact of social networks, and how they govern individual positioning within flocks, and influence decision-making processes. In corvid flocks, close bonds between pairs typically result in individuals flying closer to one other flock member. Such a preference results in an internal flock micro-structure, whereby paired birds interact less with other flock members. Interestingly, paired birds also had lower wingbeat frequencies than unpaired birds, suggesting that flying with a well-known individual can mitigate some of the energetic costs of flying in clusters. This could potentially be linked to predictability of flight styles and trajectories between familiar individuals well-known to each other, which results in fewer minor adjustments required during flight. Flocks of Eurasian jackdaws (*Corvus monedula*) behave differently under different conditions and use distinctive context-dependent rules to move together. When travelling to roosts, jackdaws interact with a fixed number of individual neighbors, but when they are mobbing a potential predator, the birds coordinate with neighbors based on distance.

The study of bird flocks

Historically, flocking has been a tricky subject to study as the birds fly high in the sky, often at high speed. Researchers would traditionally use a sequence of photographs, video footage and occasionally radar to study the positioning of individuals within a flock. The main caveat of using photographs and video images is that they cannot always accurately capture the difference in height between individuals. When flying in V-formation in particular, birds can adopt three approaches to formation flight, with respect to vertical spacing between individuals: ascending, descending, or level (planar). The different approaches to vertical spacing within the flock have substantial implications for the aerodynamic interactions between individuals. Thus, incorrect assumptions about the positioning of birds within a V-formation, particularly assuming they are flying in planar flight when they are not, will have significant consequences for any calculations of potential energetic savings taking place within the group.

A step-change in how flocking behavior is studied has occurred with the advent of biologging. Biologging involves the deployment of miniaturized devices on the animal that can record an array of behavioral, physiological and location-based variables. Such variables can include heart rate, body temperature, heading (via a magnetometer), GPS (the location of the animal) and acceleration (speed, and what the animal is doing). In particular, the fusion of GPS and accelerometry-based devices, coupled with increases in their sampling frequency and accuracy, allows for the in-depth study of flocking dynamics in free-flying birds in a manner that was not previously feasible. Data from accelerometer loggers can record every single wingbeat a bird undertakes during a flocking event, providing information on wingbeat frequency and wingbeat amplitude, two important proxies for calculating work rate. Combined with GPS data, it is then not only possible to pinpoint the location of every bird within a flock, but the location of every wingbeat that took place. In addition to lifting the lid on how bird flocks operate, these data offer practical applications in the fields of conservation, biomimetics, artificial intelligence, robotics and in managing human–wildlife conflicts.

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