In a backward stepwise regression analysis among all predictor variables, only DPG contributed significantly to the model; that is, vasodilation of distal skin regions was the best predictor of the body's readiness for sleep (Fig. 1). Because interventions such as light or large carbohydrate-rich meals differentially manipulated the independent variables, the effect on the dependent variable showed that the link between distal vasodilation and the ability to fall asleep is functional, not just correlative.

The circadian clock prepares the thermoregulatory system for vasodilation to begin in the early evening as sleepiness increases, followed by a drop in core body temperature. Even lying down increases sleepiness by redistributing heat in the body from the core to the periphery. Turning out the light is a complex cognitive and physiological signal that also leads to vasodilation. There is a tight correlation between the timing of the endogenous increase in melatonin in the evening and vasodilation, an effect that is mimicked by pharmacological doses of melatonin. Before bedtime, then, many overlapping events orchestrate the thermoregulatory overture.

We would predict that classical hypnagogic and somatic illness may be secondary to an inability to vasodilate and prepare the body for sleep. Kurt Kräuchi, Christian Cajochen, Esther Werth, Anna Wirz-Justice

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**Connecting stalks in V-type ATPase**

In all organisms, adenosine triphosphate (ATP) provides metabolic energy for driving energy-dependent processes. It is synthesized and/or utilized by enzymes known as F-type and V-type ATPases, which are small rotary motors. Both types consist of a headpiece, F₁ or V₁, respectively, which is membrane-bound part, F₀ or V₀. Electron microscopic analysis of negatively stained particles has revealed a peripheral stalk, or stator, between V₁ and V₀ of the V-type (Na⁺) ATPase of the thermophilic bacterium *Clostridium fervidus*. We have analysed many more particles and now present a more complete structure of the V-type ATPase stator moiety.

A central stalk in the ATPase rotates within a ring of three α- and three β-subunits in F₁ (refs 7, 8), or three A and three B subunits in V₁, in discrete steps of 120°. At the F₀/V₀ end, the central stalk is connected to a ring of α subunits in the membrane. These subunits rotate against other subunits of F₀ and V₀ allowing ion translocation at the interface. A stator structure in the form of additional connections between F₁/V₁ and F₀/V₀ must presumably be present to prevent futile rotation of the α3β3 and A3B3 headpiece.

We classified a set of 7,500 molecular projections of detergent-solubilized, negatively stained V₁/V₀, and found a few preferential orientations in which V₁ has either two or three lobes. In about 28% of all views, there are three connections between V₁ and V₀, but in the rest either one (33%) or both (39%) of the peripheral connections appear to be missing. The three connections are well separated in the bilobed views (Fig. 1a). In the classes with trilobed views, the left connection is less well resolved because it is closer to the central stalk and overlaps more with V₁ (Fig. 1b). The two peripheral connections are each attached to an oval-shaped area of density on top of V₁. In some other classes (not shown), one of the oval densities is absent, which correlates with the loss of a peripheral connection. We conclude that the intact V-type ATPase has a central stalk and two stator connections (Fig. 1c, d). Likely candidates for the stators are the bacterial V-type subunits I, E and F, several copies of which are present.

The discovery of the second stator raises several points. First, although it has been suggested that there may be two stators, this feature has not been observed before. This could be because the intact structure is easily damaged on preparation, which can be detected only by classifying large sets of projections. The two stators may be unique for V-type ATPases because, in the stalk region of F-type ATPases, only the b subunit is present in two copies, as a dimer, participating in one stator. It is harder to visualize the stator(s) in F-type ATPase by electron microscopy because the stalk region is much shorter than in V-type ATPase.

Second, there could be more than two stalks: three would match the three-fold symmetrical ring of the six large subunits in F₁ (refs 7, 8), or three A and three B subunits in V₁, in discrete steps of 120°. At the F₀/V₀ end, the central stalk is connected to a ring of α subunits in the membrane. These subunits rotate against other subunits of F₀ and V₀ allowing ion translocation at the interface. A stator structure in the form of additional connections between F₁/V₁ and F₀/V₀ must presumably be present to prevent futile rotation of the α3β3 and A3B3 headpiece.

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**brief communications**

in V. There is no evidence for this, but the presence of another stator connection is a possibility if there is overlap in projection.

Finally, it is not clear why the ATPase motor has such a complicated stator moiety, with an overall U-shaped form that must avoid friction with the central rotating stalk, although it is likely to be related to its mechanical stability.

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**Evolutionary origins of bee dances**

Although bumble-bees are highly social insects, their foraging has been considered to be managed as an individual initiative,1–4, in which each bumble-bee visits flowers not only to collect food, but also to gather information about other potential food sources5. Here we show that bumble-bees instead use a primitive, but surprisingly efficient, recruitment system: by performing extended excitation runs in the nest, a single successful forager can alert the entire foraging force of a bumble-bee colony. But in contrast to what happens in other social bees, such as honeybees, the recruits are not informed about the location of the food. Instead, the successful forager brings home the odour of the newly discovered food source, conveying to the recruits information about the species of flower. These findings about bumble-bee communication shed new light on the early evolutionary origins of the elaborate dance language of the honeybee.

To investigate whether bumble-bees (Bombus terrestris) can communicate information about the discovery of a food source, we connected a nest box with a bipartite flight arena. A single forager was allowed to collect sucrose solution from an artificial flower in one half of the arena, whereas all other bees had access only to the other half of the arena, which did not contain food. This procedure ensured that interactions between bees could take place only in the nest, not at the food source. The number of bees entering the empty flight arena to search for food was counted during 12 periods of 1 hour each, when foragers were rewarded.

When compared with unrewarded control periods, the searching activity strongly increased when a single bee foraged (P < 0.01, Wilcoxon test; Fig. 1a). Successful foragers in the nest, instead of just emptying their crop load and continuing to forage, would typically spend several minutes running across the nest, frequently bumping into nestmates and occasionally buzzing their wings.

Odour-preference tests indicated that bees leaving the hive strongly preferred the odour that was brought home by the forager (n = 90, P < 0.001, x^2 test; Fig. 1b). Because floral scents are species specific, the odour helps recruits to find the food source used by the successful forager. To test whether positional information is also conveyed, we trained three foragers to collect sucrose from a feeder positioned 100 m west of the hive in an open field. Recruits had the choice between that feeder and two additional feeders placed 100 m north and south of the nest. New recruits distributed themselves randomly at these three feeders (P = 0.21; x^2 test). This failure of bumble-bees to recruit to specific points in space may explain why their communication has previously been overlooked. Bumble-bees therefore have a recruitment system that is specific for a flower species but independent of its location. This system for transmitting information resembles the round dance used by honeybees when food is found in the immediate vicinity of the hive6–8.

But why have bumble-bees not evolved a communication system that includes information about where to forage? Such a system can be very costly. Honeybee recruits may take more than an hour to decide where to go, even if only two different locations are advertised7. They also take a long time to find a food source after receiving information about its location.

Bumble-bees live in small colonies, mainly in temperate habitats where floral food is less clumped than in the tropical habitats where the communication of honeybees and stingless bees evolved9,6.10.6. The advantage of communicating location might therefore not offset the cost. It may be sufficient to specify flower species to recruits, who can then rely on their own memory or searching ability to find flowers but still know when and for what to forage.

Understanding the communication of bumble-bees is central to reconstructing the evolutionary origins of the honeybees’ waggle dance, which is one of the most complex systems in animal communication. Such a reconstruction requires us to compare the honeybees’ behaviour with that of their extant relatives, the bumble-bees and stingless bees, which have excitory motor patterns that serve to recruit nestmates to food sources10. This behaviour may be derived from a social facilitation of activity at the nest entrance, which is widespread among social insects10. Bumble-bees also share with stingless bees and honeybees6,10 the ability to learn the floral odours brought back by returning foragers. The bumble-bee recruitment system might therefore resemble that of the last common ancestor of the eusocial bees.

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**Insect behaviour**

**Figure 1 Recruitment in bumble-bees.** a, The number of bees that leave the hive in a 5-minute period increases dramatically when one bee forages successfully (brown bar). b, Most bees choose the odour that is brought into the nest by a forager.**