tropical Atlantic, and so influence thermohaline circulation in the north of the ocean[11]. But these mechanisms will remain speculative as long as we lack adequate temperature reconstructions or other estimates of ENSO strength during deglaciation. All in all, the tropics are probably the biggest wild card left in the game of understanding abrupt climate change.

This point takes us back to a limitation of Knorr and Lohmann's study[2]. Because atmospheric dynamics are neglected, changes of ocean circulation patterns and their effect on atmosphere–ocean heat exchange and the freshwater balance through the hydrological cycle cannot be accounted for. So the next steps will be to perform experiments using more comprehensive models to test the importance of the processes that these authors have identified. ■

Thomas F. Stocker is in Climate and Environmental Physics, Physics Institute, University of Bern, 3012 Bern, Switzerland.
e-mail: stocker@climate.unibe.ch

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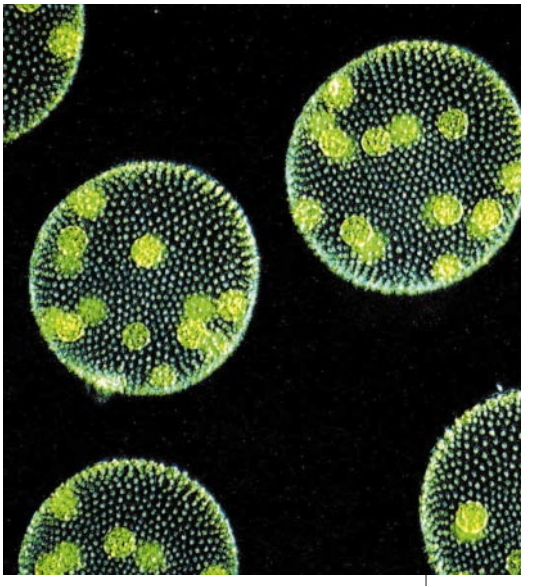

Figure 1 ***Volvox carteri*. The adult organism consists of some 2,000 small somatic cells located on the surface of a hollow sphere, and around 16 large reproductive cells just below, within a transparent extracellular matrix. By a series of predetermined cell divisions each reproductive cell will become an embryo that finally turns inside out by a process called inversion, to produce a juvenile with all its cells in the adult orientation.**

Developmental biology

How to turn inside out

Rüdiger Schmitt and Manfred Sumper

The discovery that a molecular motor of the kinesin family is involved in turning a multicellular green alga inside out might have implications for similar events in animal development.

Something quite remarkable happens to embryos of the multicellular green alga *Volvox carteri*: they turn completely inside out to establish the adult body plan. This inversion process closely resembles the initial stages of the more complex gastrulation that occurs in animal embryos — so *Volvox* could arguably be considered a simple model for analysing the principles that direct such changes in shape. As they describe in *Cell*, this idea led Nishii and colleagues[1] to re-examine and dissect the process of inversion in *Volvox*. Their concept helps to explain the biomechanics and the driving forces behind the curling of a cellular sheet.

In the embryos of most multicellular animals, sheets of cells invaginate during gastrulation, neurulation and organ formation. Gastrulation is the central process in early animal development, and occurs during the blastula stage — when the embryo consists simply of a hollow ball of cells. It involves complex movements that carry those cells whose descendants will form the future internal organs from their superficial position on the blastula to their definitive positions inside the embryo. Similarly, neurulation in vertebrates involves a complicated curling of a cellular sheet to form the neural tube, which in turn develops into the central nervous system.

Gastrulation has fascinated developmental biologists ever since it was recognized in 1874 (for a historical review, see ref. 2). Nearly 100 years ago, the Italian embryologist Angelo Ruffini first described the appearance of elongated cells — known as bottle or flask cells — at the onset of the process in amphibians. Then, in his classic papers on amphibian gastrulation, Johannes Holtfreter[3,4] claimed that the ability to invaginate is an innate property of flask cells. Modern investigations confirm that the number and arrangement of the flask cells are critical factors for proper initiation of invagination. But, as pointed out by Keller[5], what flask cells do — and how, in a biomechanical sense, they do it — remains to be elucidated. *Volvox carteri*, a much simpler organism, might be the Rosetta Stone that enables researchers to unlock the problem.

Volvox is a multicellular green alga (Fig. 1) that exhibits the simplest kind of differentiation — the division of labour between just two types of cell. The adult organism consists of about 2,000 mortal somatic cells, which make up the surface of a hollow sphere, and 16 larger, potentially immortal reproductive cells just below the surface. The development of *Volvox* starts with a single reproductive cell, which undergoes a patterned sequence of 11 cell divisions. The first five divisions are symmetrical, resulting in an embryo consisting of 32 cells of similar sizes. But the sixth division of the 16 most anterior cells is asymmetric, and results in the production of 16 cell pairs of unequal size. The larger cells will become the new reproductive cells.

As a result of geometrical constraints, at the end of the 11 cell divisions the embryo is inside out with respect to the adult configuration: the large reproductive cells protrude from the surface, and the bases of the developing flagella of all the somatic cells point to the interior of the hollow sphere. So cell division is followed by inversion, in which the curvature of the embryo is reversed to establish the adult configuration. Development of *Volvox* therefore resembles that of classic models of animal development, such as sea urchins and nematode worms, in that an important differentiating cell division is visibly asymmetric, and the adult configuration is attained by a gastrulation-like event.

How does inversion come about? First, the phialopore (a slit at the anterior end of the embryo) widens, and four lips of cells bend outwards and backwards over the adjacent cells — in other words, they curl outwards. The region of maximum curvature moves progressively towards the posterior pole, until the inverted region almost surrounds the posterior (non-inverted) hemisphere. At that point the posterior hemisphere 'snaps' through the opening at the equator, and the phialopore lips move to seal the gap at what is now the posterior pole.

Flask-shaped cells that are linked by a network of cytoplasmic bridges are the key

players in this dramatic shape change. Cells become flask-shaped — with a long stalk at their outer end — near the bend region, and they then move inwards relative to the network of cytoplasmic bridges, which runs through each cell and connects it to its neighbours. This is the key point in Kirk's model of inversion[6,7]: as flask cells proceed from being linked at their widest point to being linked at their thin outermost ends, the cell sheet is forced to curl sharply outwards (Fig. 2).

Nishii *et al.*[1] now propose that this displacement of flask cells relative to the cytoplasmic bridges is driven by a motor protein — a newly discovered kinesin denoted InvA.

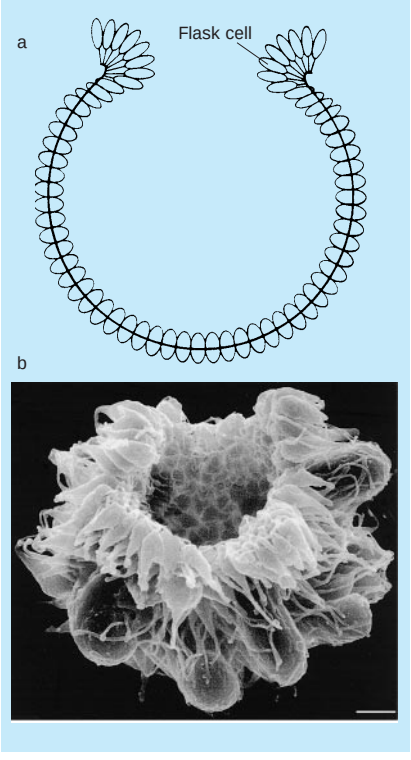

Figure 2 **Inverting *Volvox*.** a, **Cross-section through an inverting embryo. The continuous black line represents the system of cytoplasmic bridges that connects the cells and holds them together. The force necessary for inversion is generated by a change in cell shape, together with cell movement. First the cells become flask-shaped by forming long, narrow 'stalks' at their bottom (outward) ends; then they move inwards relative to the cytoplasmic bridges to a point where they are only linked at their narrow tips. These actions produce a bend region, where the cell sheet is folded back on itself. The results of Nishii *et al.*[1] suggest that a newly discovered kinesin, InvA, is located in the cytoplasmic bridges, and that, by moving along the microtubule filaments lining the flask cells, InvA produces the force that drives the cell body past the bridges to form the bend.** b, **An embryo with a mutation in InvA; it is unable to invert fully.** a **and** b **are reproduced from refs 9 and 1, respectively. Scale bar, 5** μ**m.**

The authors started by generating mutant *Volvox* embryos with defects in inversion. They then analysed one such mutant in depth, and found that although its cells became appropriately flask-shaped, they failed to move relative to the cytoplasmic bridges. Nishii *et al.* went on to show that the gene affected in this mutant encodes a kinesin, and that this molecular motor is located in the cytoplasmic bridges.

Each flask cell has cytoskeletal filaments of microtubules that run along its length, just inside the plasma membrane, and Nishii *et al.* propose that InvA molecules attempt to move downwards on these filaments. But, because the InvA molecules are anchored to the bridges, and the bridges themselves are fixed in place, InvA cannot move significantly. Instead the force produced by this motor causes the microtubules — and consequently the whole cell — to move past the cytoplasmic bridges. This perpendicular movement eventually connects the cells at their thin outermost ends and so creates a sharp bend.

The discovery of a special kinesin as part of the machinery that causes the curling of a cellular sheet in *Volvox* will stimulate a search for a related motor protein with the same function in animals. Might such a motor exist? It seems plausible: molecules such as cytoskeletal proteins are found in most organisms, from unicellular yeasts to humans, and were probably present in the unicellular common ancestor of plants and animals. Their existence there would predetermine similar solutions to given morphogenetic problems. Moreover, if the displacement of asymmetrically shaped cells with respect to a fixed framework of cell–cell connections is the key concept underlying curling, then the cytoskeleton and cell–cell adhesion systems would have to play a key part. This would mean that similar mechanisms operate even in evolutionarily very distant[8] organisms, from multicellular algae to animals. ■

Rüdiger Schmitt and Manfred Sumper are at the Institut für Biochemie, Genetik und Mikrobiologie, Universität Regensburg, 93040 Regensburg, Germany.
e-mails: rudy.schmitt@biologie.uni-regensburg.de manfred.sumper@vkl.uni-regensburg.de

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Materials science

The road to diamond wafers

S. T. Lee and Y. Lifshitz

Diamond could rival silicon as the material of choice for the electronics industry, but has been held back by the difficulty of growing large enough wafers. This problem may now be solved.

Diamond is the king of gemstones. Less well known is that it could also be an outstanding semiconductor material, superior in many ways to silicon, which is currently the most widely used electronic material. Diamond devices could operate at higher temperatures (more than 400 °C) and higher power than those of silicon, as well as being faster, denser and more resistant to radiation. But practical diamond electronics will need large-area, single-crystal diamond wafers to be fabricated, analogous to the 6–12-inch silicon wafers commonly used in the semiconductor industry. Two papers from Golding and colleagues, in *Applied Physics Letters*[1] and *Diamond and Related Materials*[2], now show that this may be possible if sapphire wafers are used as substrates on which to grow the diamond.

Diamond can be grown on diamond ('homoepitaxy') by chemical vapour deposition: a diamond substrate, at a temperature of 600–800 °C, is exposed to an ionized mixture of roughly 1% hydrocarbon and 99% hydrogen. Electronics-grade diamond has been made in this way[3]. The newly grown diamond wafer can be cut from the diamond substrate, and the process can be repeated many times, reusing the substrate. But single-crystal diamond substrates are small and expensive, so this is not a viable way to fabricate large-diameter diamond wafers.

The alternative is to grow diamond on a foreign (non-diamond) single-crystal wafer — this is 'heteroepitaxy', the oriented growth of one crystal on another. Heteroepitaxy is readily achieved if the atomic spacings in the foreign substrate match those in diamond crystals. Of the various substrates tested — such as silicon, silicon carbide, nickel, cubic boron nitride and platinum — iridium is the best found so far[4]. The quality of epitaxial growth is measured by the average angular spread of the diamond-crystal orientation along a certain direction (also called mosaicity). The minimum angle achieved is 3.9° for silicon, less than 2° for platinum, 1.5° for