TICKER TAPE BACTERIA

Scientists are working to understand, and to harness, the unusual ability of one type of microorganism to produce long, strong cellulose ribbons

BY JULIE ANN MILLER

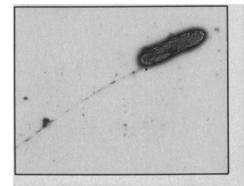
Genetic engineering has opened new vistas for microbiology — both practical and whimsical. And sometimes it is hard to tell the two apart. With cartoonists sketching bacteria constructed to convert polyester into prune yogurt, can a microorganism that turns sunlight into high-strength textile fiber be a serious goal? It is for R. Malcolm Brown and colleagues at the University of North Carolina in Chapel Hill. They are hot on the trail of a Rumpelstilskin "bug," a bacterium or alga that would spin from inexpensive chemicals a fiber as good as gold.

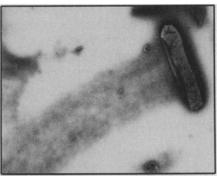
Twisted ribbons of pure cellulose may be the thread of future textiles. A polymer of glucose units, cellulose is most commonly found as the skeletal structure of plant cell walls. But a variety of microorganisms also make the polysaccharide. Cellulose producers include rhizobia, the bacteria that fix nitrogen in root nodules of legumes, and agrobacteria, which cause plant tumors (SN: 7/15/78, p. 45). Brown suggests that the cellulose helps bacteria attach to their hosts.

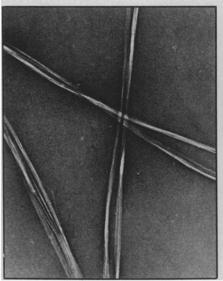
As far back as the mid nineteenth century scientists such as Louis Pasteur recognized that *Acetobacter xylinum* makes cellulose. The bacterium, which is found in fermenting wine and on rotting, sugary fruits and vegetables, synthesizes a twisted cellulose ribbon. Recently, scientists have been working out the details of the operation, both to obtain information about cellulose production in higher plants and to put the bacteria to use.

When A. xylinum grows in liquid culture, a tough skin of intertwined cellulose ribbons forms across the liquid's surface. If that membrane is dried, it resembles a piece of thin, white translucent paper. A mutant bacterium with altered cellulose production makes a thicker film. Brown said at the recent meeting in Cincinnati of the American Society for Cell Biology that he expects the bacterium, and its genetic variants, to be valuable in producing paper-like specialty products for industry and medicine. Brown predicts also that fabric made of such cellulose fibers could be less expensive than cotton and, because of the long length of the fibers, could be exceptionally strong.

Bacteria make their cellulose ribbons by a sequential clustering of bundles,





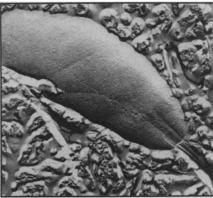




Acetobacter xylinum normally produces a narrow ribbon of cellulose (left top and, at greater magnification, bottom). In presence of the dye Calcofluor, cellulose production is dramatically altered (right top and, at greater magnification, bottom).

Brown and colleagues find. In the first step, sugar units are added to simple chains within the outer envelope that encloses the cell. Pores, lined up in groups of three along the envelope, each extrude 12 to 15 chains of cellulose that become stably bonded together. Outside the cell, three of the aggregates assemble into a "microfibril" 35 angstroms wide. Then, neighboring microfibrils come together into a "bundle," and the 10 to 12 bundles (50 to 80 microfibrils) produced by a bacterium associate into a ribbon.

Each bacterium adds 100 million sugar units per hour, producing 3 to 4 millimeters of ribbon every day. The lifetime of the bacterium and the investigators' patience are the only limits on the ultimate length. The ribbon even bridges the bacterial generation gap. As the cell elongates before cell division, it adds sets of extrusion sites, so each daughter cell receives a complete complement. When the cell divides, the daughter cell with the older set of pores continues adding to the parental ribbon, while the other daughter starts a new thread. Occasionally, the new set of pores begins operation before the parent



"Synthesis-extrusion" sites make a line on this bacterium. Freeze-fracture photo also shows ribbon print at lower right.

cell divides. The scientists then observe a single cell making two ribbons at once.

This orderly assembly of cellulose ribbon can be disrupted with a variety of dyes. Candace H. Haigler, Moshe Benziman and Brown reported at the Cincinnati meeting (and in the Nov. 21 SCIENCE) that the production becomes disordered, for

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example, in the presence of Calcofluor White ST, a fluorescent brightener used commercially on textiles and paper.

Crystallization of sugar chains into cellulose microfibrils is the step disrupted by the dye, the scientists find. By forming hydrogen bonds with sugar chains, the dye molecules prevent the chains from bonding to each other to form the normal crystalline lattice of the microfibril.

In the presence of dye, a broad band of cellulose, instead of the normal narrow ribbon, extends from the bacteria, and a soft, filmy mass of altered cellulose accumulates at the bottom of the culture vessel. If this noncrystalline cellulose is dried, however, the Calcofluor is displaced and, surprisingly, the cellulose lattices form. When free Calcofluor is removed from the medium, the bacteria immediately begin to synthesize the normal ribbons.

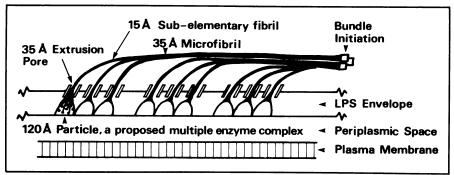
From their observations of the Calcofluor-disrupted bacteria making broad bands of cellulose, Brown and colleagues conclude that the linking of sugar units into simple chains and the crystallization of bundles of chains into microfibrils are separate, although coordinated, processes. They find that the rate of production of the chains is limited by the rate of crystallization: When Calcofluor uncouples the processes, the chains are produced at four times the normal rate.

The scientists now envision cellulose being formed in a "cell-directed" self-assembly process. Complexes of enzymes associated with the pores determine the number and orientation of sugar chains. As the chains are extruded, they aggregate by hydrogen bonding. The precise arrangement of the "synthesis-extrusion" sites on the cell envelope facilitates self-assembly of the aggregates into microfibrils, then bundles and ribbons.

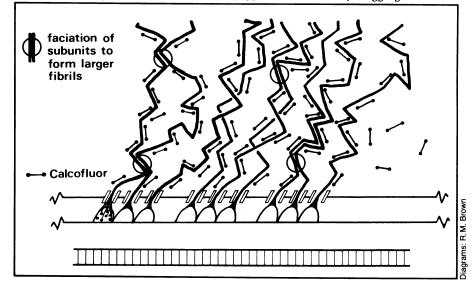
Currently the major drawback of the bacterium as a fiber producer is its rather expensive raw material — glucose. Although using bacteria to make fibers would save on agricultural and processing expenses, A. xylinum just cannot compete economically with cotton plants that get energy directly from the sun.

The ultimate answer, Brown says, is to have the cellulose-making ability in a photosynthetic microorganism. Techniques for genetic manipulation may allow scientists to transfer genes required for cellulose production into a suitable host. Already there have been reports that some blue-green algae naturally make cellulose, and Brown and colleagues are investigating likely algal candidates.

Obtaining fibers suitable for highstrength fabrics from microorganisms may be the new biotechnology's way of making a silk purse from a sow's ear. And the future promises other practical microbiological projects that, at present, seem as whimsical as converting polyester into prune yogurt.



Bacteria normally form cellulose bundles (top); Calcofluor disrupts aggregation.



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