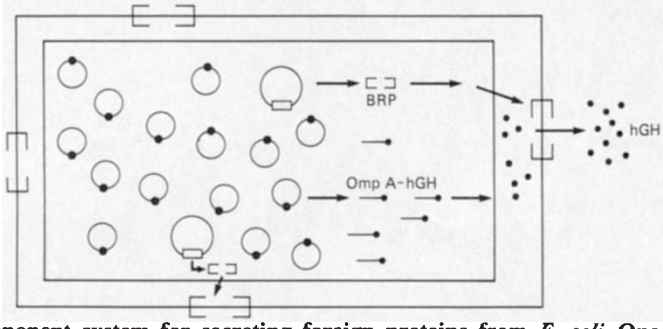
RESEARCH PAPER ANALYSIS

SUCCESS IN ENGINEERING *E. COLI* SECRETION



A bicomponent system for secreting foreign proteins from *E. coli*. One plasmid expresses bacteriocin release protein (BRP) that renders the outer membrane permeable. The other encodes the synthesis of human growth hormone (hGH) fused to a signal peptide (OmpA), allowing the hormone to enter the periplasm.

Manipulating *Escherichia coli* to secrete efficiently heterologous proteins into the culture medium has been a goal of genetic engineers since the first chimeric genes were expressed in this bacterium. In this issue of *Bio/Technology*, Hansen Hsiung and his colleagues at Eli Lilly Research Laboratories (Indianapolis, IN) and their collaborators at Vrije University (Amsterdam, Netherlands) describe a simple system to accomplish this hitherto elusive undertaking.

The key to their success is the use of a bicomponent vector system. One plasmid contains the gene of interest—in this case a human growth hormone-*ompA* signal peptide fusion construction. The other harbors a colicin plasmid-derived gene encoding bacteriocin release protein (BRP), which activates an outer membrane phospholipase that renders both the inner and outer bacterial membranes permeable. By regulating the copy number and expression of the genes, the researchers could prevent premature rupture of the bacteria, and allow the release of mature growth hormone into the culture medium at levels of 4–5 μg/ml.

A secretion system of this type offers a number of actual and potential advantages to cytoplasmic overproduction of foreign proteins. Purification is much simpler—a single reverse-phase chromatography step of the concentrated culture medium resulted in a 98 percent pure product—and proteins that tend to form insoluble inclusion bodies might be coaxed out of the cell before large amounts of improperly folded material accumulate. It should also be possible to design a continuous production system by using immobilized bacteria and controlling the amount of BRP synthesized so that cells remain viable for extended periods of time.

For scientists who wish to purify a few milligrams of a heterologous protein in their laboratories, the use of low copy number plasmids expressing BRP may well become a method of choice. **—Harvey Bialy**