Cages, Cavities and Clefts

By building and setting molecular traps, chemists shed light on how one molecule recognizes another

By IVARS PETERSON

One molecule behaves like a Venus's-flytrap, enveloping smaller molecules that happen to infiltrate its open framework. Another dangles bits of molecular bait to entice certain molecules into its jaws. A third resembles an octopus, with an egg-shaped body and eight flailing arms. Others look like crinkly doughnuts and spiked crowns.

These curious constructions are examples of molecules recently synthesized by organic chemists interested in studying how molecules recognize one another. Such molecules are engineered to perform a variety of chemical functions similar to those that typically take place in biological systems.

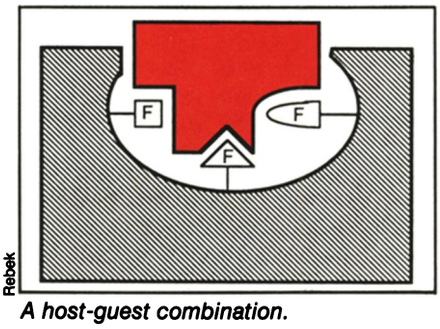
"Living systems provide organic chemists with a number of intriguing examples of molecular behavior," says Julius Rebek Jr. of the University of Pittsburgh. "My research group as well as others have been concerned with imitating this behavior using small, synthetically accessible molecules."

Biological systems accomplish a variety of complicated chemical tasks. Antibodies recognize and bind invading microorganisms. Enzymes collect raw materials and convert them into biologically useful products. Strands of DNA carry a chemical code to express genetic information and to provide for its replication. Ionophores trap and move ions across membranes. In these instances, huge, appropriately adorned molecules ensure that essential chemical ingredients are in the right orientation in the right place at the right time.

"New developments have led to a belief that such processes can occur in much smaller molecules," says Rebek. The challenge is to design and build compounds with structural features that give rise to a particular kind of chemical behavior.

To achieve recognition or catalysis, one molecule must fit into another as snugly as a hand in a glove. But it's not sufficient for the host (or receptor) molecule to have a cavity of just the right shape and size to receive a particular guest (or substrate) molecule. The host must be able to attract and hold its potential guest for as long as necessary.

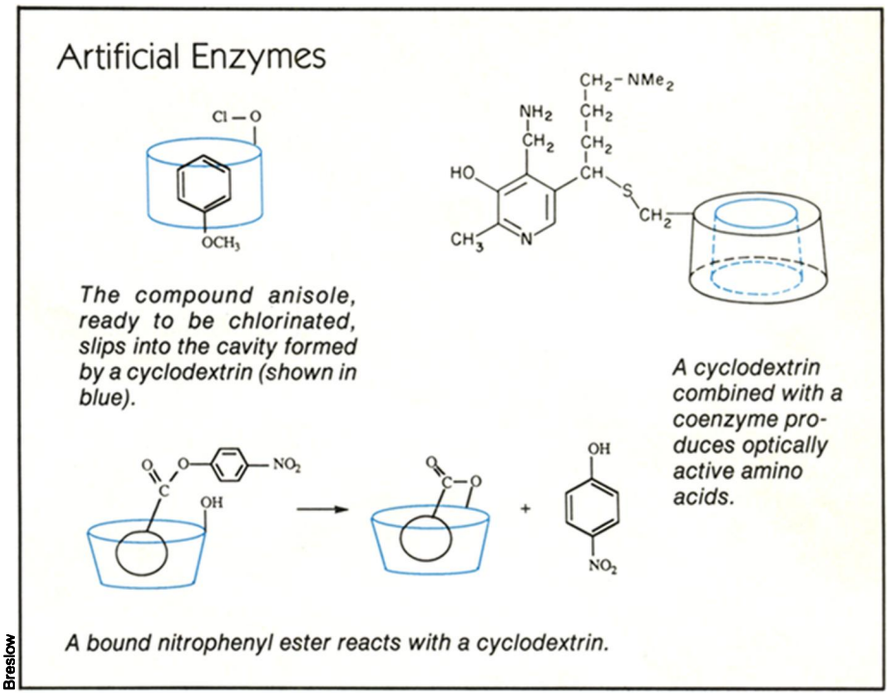
In general, a suitable host molecule has a bowl-shaped cavity, with binding sites arranged along the bowl's concave surface. These sites interact with matching positions on a guest molecule's convex surface.



Rebek

A host-guest combination.

When a host and guest combine, the result is a chemical system known as a complex. Although the two components are bound together, the intermolecular forces holding them firmly in place are usually much weaker than the covalent bonds keeping a molecule's atoms together.



Breslow

The synthesis and study of enzyme mimics is one important example of efforts to imitate biological systems. "We tend to understand things by relating them to other, simpler things that we already believe we understand," says Ronald Breslow of Columbia University in New York City. By creating and manipulating simple artificial enzymes, chemists can test their theories about how more complicated natural enzymes speed up and regulate chemical reactions.

Furthermore, the properties and structures of enzymes themselves inspire the construction of novel chemical catalysts, says Breslow. "The aim is to produce new catalytic systems that might have some of the same high selectivities and high rates characteristic of enzyme processes, but that might have special advantages over natural enzymes." It's even possible to prepare artificial enzymes capable of catalyzing reactions for which no natural

enzymes exist.

Breslow and his colleagues have worked largely with naturally occurring compounds called cyclodextrins – rigid, doughnut-shaped molecules made up of glucose units. "The ready availability of cyclodextrins," says Breslow, "made them attractive in the first constructions of artificial enzymes."

Although this choice limited the study of catalytic interactions to molecules capable of binding with cyclodextrins, says Breslow, he and his group wanted to establish the potential of this approach for imitating enzymes. "You could always generalize it later," he says.

Cyclodextrins have several useful properties. The size and shape of a cyclodextrin's hole is determined by the number of glucose units that make up its ring. For example, cyclohexamylose, consisting of six glucose units, has a cylindrical cavity 7 angstroms deep and 5 angstroms across. Choosing among the different cyclodextrins for use in a given chemical system is not unlike selecting the appropriate wrench for a particular plumbing job.

Although cyclodextrins dissolve in water, their cavities repel water molecules. Such cavities are termed hydrophobic. By keeping water out, each cavity provides a sheltered spot in which small organic molecules can "keep dry" in an aqueous environment.

One of the simplest illustrations of how the formation of a complex can promote a specific chemical reaction is in the addition of a chlorine atom to the compound anisole. Anisole consists of a hexagonal ring of carbon atoms with a small molecular tail attached to one corner. Normally, when the compound reacts with chlorine, the chlorine atom ends up attached either to the corner next to the tail or to the corner opposite the tail. The result is a mixture of two types of chlorinated anisole.

However, in the presence of a cyclodextrin with six glucose members in its ring, an anisole molecule settles head first into the cyclodextrin's cavity. A molecular group attached to the cyclodextrin ring and capable of delivering a chlorine atom can then react with the anisole molecule. Because the anisole molecule is held fairly tightly in place, the chlorine atom always ends up in a position opposite the molecule's tail.

An enzyme called chlorinase is also able to chlorinate anisole, but it performs its function relatively randomly, producing a mixture of the two types of chlorinated anisole. The cyclodextrin system, says Breslow, "represents a simple illustration of the fact that selective chemical processes of interest to chemists may well be performed better by even simple enzyme mimics rather than by enzymes."

Breslow and his colleagues and a number of research groups throughout the world have tested a wide variety of cyclodextrins as enzyme mimics and potential catalysts. In most cases, a cyclodextrin does the binding and a ring-mounted molecular group mediates the chemical reaction.

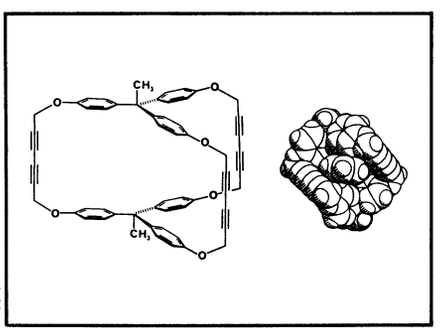
One of the toughest problems that researchers are now struggling with is the synthesis of enzyme mimics that produce amino acids. Because amino acids can come in left- and right-handed forms, depending on the arrangement of their constituent molecular groups, it's crucial to find a way for a host molecule to produce only one of the two forms. A sample consisting of only one of the two forms is said to be optically active.

"I think it's fair to say that nobody, including us, has anything that somebody can use to manufacture amino acids," says Breslow. His technique is to use a cyclodextrin to bind the aromatic part of an ingredient for making a particular amino acid. A natural coenzyme attached to the cyclodextrin ring performs the chemistry, while the structure of the ring itself and its attached molecular groups ensures that hydrogen atoms end up in the right places to turn the product into an optically active compound.

"It's really quite a remarkable object," says Breslow. "It does everything the natural enzyme does, although it doesn't work as well as an enzyme yet. It's a little floppy, and we're trying to tighten it up."

Breslow has also synthesized cage-like molecules that, like the cavities in cyclodextrins, are hydrophobic. One such molecular cage has a three-dimensional, open structure that allows a guest molecule such as benzene to enter. The self-adjusting cage then closes up around the guest, clamping it tightly in place.

"You want to have as much rigidity as you can get away with," says Breslow, "but you've got to have enough flexibility to let things in, to be grabbed and operated on, and to open up to spit them out again."



Breslow

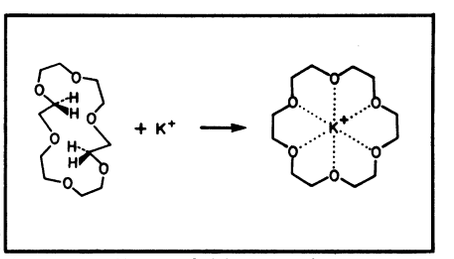
Breslow's cage molecule (left) can trap a benzene molecule. The resulting benzene-cage complex is shown at right.

Synthetic cage molecules have the advantage that they can be tailored for specific purposes. Breslow and his colleagues are exploring the possibility of hanging various molecular groups from a cage molecule. Such groups would allow the compound to dissolve in water and to catalyze certain chemical reactions.

Frequently, the construction of a mimic is just the first step along a path that can take an organic chemist far afield. "Once you model a biological system, you can go off on your own," says Donald J. Cram of the University of California at Los Angeles. "You're inspired by your results to go further."

While Breslow and other researchers have focused largely on modifying cyclodextrins, Cram and his colleagues have spent the last two decades constructing large organic molecules specifically designed to bind certain types of guest molecules. "We set out to design and make a whole series of cavitands – rigid compounds with rigidly formed concave surfaces of molecular dimensions," says Cram.

The initial inspiration for Cram's effort was a group of compounds known as crown ethers. A typical crown ether consists of a carbon-atom ring with oxygen atoms strategically placed at certain positions. These molecules were among the first cavity-containing compounds to be synthesized rather than extracted from natural sources. Crown ethers act almost exclusively on positively charged ions, readily unfolding and stretching to engulf their unwitting guests.



Cram

A crown ether unfolds to grab a potassium ion.

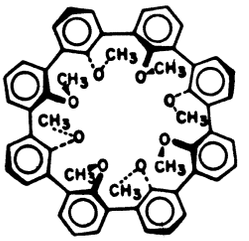
Cram and his group have concentrated on building rigid molecules that, unlike crown ethers, have a permanent cavity. Cram believes that a host molecule must be ready to accept a particular guest without having to change its form to accommodate the guest. "If you have flexible molecules," says Cram, "they tend to fill their own cavities. They bunch up."

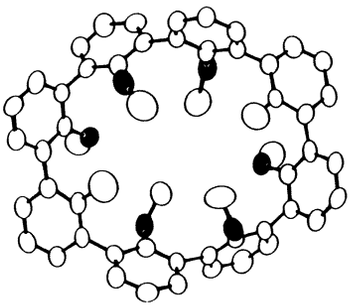
As a result, Cram's synthesized molecules have a permanent cavity of the right size and shape to attract and hold a specific guest. Additional groups of atoms hang from strategic locations near the cavity so that they can act cooperatively to bind guest molecules and take part in various chemical reactions.

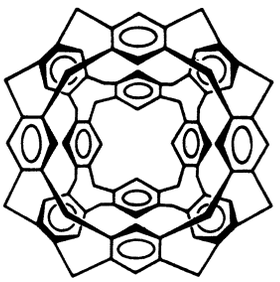
Guided by plastic molecular models,

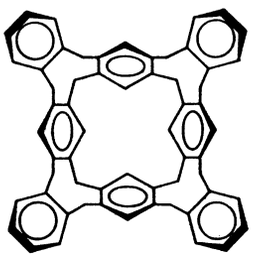
Cavitands

This "spherand," depicted as a structural formula (near right) and as it would appear as a molecule (far right), has a cylindrical cavity with a diameter of 3 angstroms and a length of 8 angstroms.



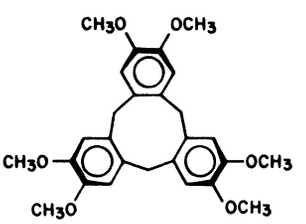


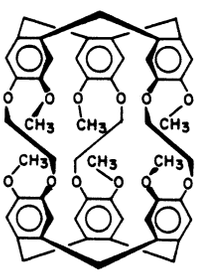




This roughly spherical molecule, seen from above, can imprison small organic compounds (far left). If the whole molecule were the size of a melon, its cavity would be roughly the size of a large orange. Using saucer-shaped building blocks, chemists may also be able to construct similar molecules in the shape of pots, collars, bowls and vases. A vase-shaped form is shown (near left).

Starting with saucer-shaped units of cyclotriveratrylene (near right), chemists can synthesize cavitands such as the one illustrated (far right). According to experiments with molecular models, this particular cavitand can hold up to six water molecules.





Cram

Cram and his collaborators have gambled on syntheses that take as many as 30 steps to get the requisite forms. "Most of the complementary relationships between hosts and guests could not have been anticipated without use of [molecular] models," says Cram.

Using his techniques, Cram has developed molecular systems that selectively remove potassium ions from mixtures that also contain sodium ions. One compound even changes color if sodium and lithium are present but not if potassium is also there. That test may soon be used for biological fluid assays.

"High selectivity factors for sodium, lithium and potassium ions are particularly important to analytical methods for measuring concentrations of these ions in serum and urine," says Cram. "The wide distribution of sodium chloride in the environment and its ability to harm circuit boards of computers and increase corrosion rates make sensitive and simple methods for its detection important."

Cram can also combine a pair of his cavitands to form a closed, hollow shell. He calls the resulting molecule a "carcerand," from the Latin word for a prison. It's like immersing two bowls in a stew, Cram says, then fitting the rims against each other and trapping some of the stew inside. Such a structure allows chemists to study the differences in properties between a small molecule in solution and the same molecule imprisoned within another molecule where its movement would be restricted.

Carcerands themselves turn out to be highly insoluble. To increase their solubility in organic solvents, Cram has added as many as eight long chains of carbon atoms to each shell, giving it the appearance of an octopus. Further alterations make carcerands soluble in water.

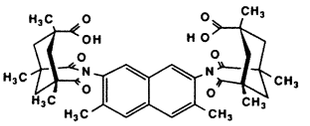
"Molecular motion itself will separate ordinary host-guest combinations, given time, because they are not chemically bonded together," says Cram. "But the space inside a carcerand is like no other chemical environment, and the guest molecule finds itself locked up for life."

Cram suggests that similar molecular shells may eventually be useful as delivery systems for active chemicals such as drugs or pesticides. The degradation of these shells within a living organism would slowly release any compounds held within the shells. Strong acids, for example, tend to break down the bonds of a carcerand cage, releasing any trapped molecules.

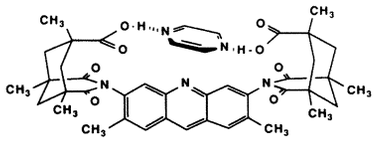
Rebek and his research group have tried to avoid complexity. "Our approach has been a minimalist one," Rebek says, "with concentration on design and testing with incisive experiments. We attempt to build the smallest system capable of the target behavior."

Rebek's principal tool for studying mo-

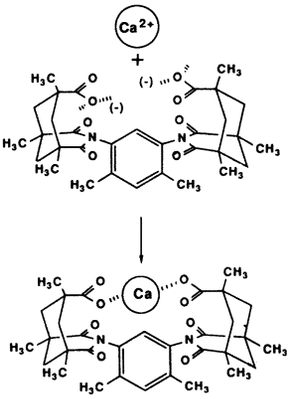
Molecular Clefts



A typical molecular cleft consists of two carboxyl groups separated by a spacer unit such as naphthalene.



The compound pyrazine is caught in the jaws of a particular molecular cleft.



Even spherical ions, such as calcium ions, respond to the vise-like shape of certain molecular clefts.

Rebek

lecular recognition is a special set of synthesized molecules, each roughly shaped like the letter C. This rigid but adjustable scaffolding provides a convenient basis for pinpointing how one molecule responds to another.

A typical C-shaped "molecular cleft" consists of two specially modified carboxyl groups separated by a rigid molecular spacer. The carboxyl groups act somewhat like sentries at a gateway, screening visiting molecules and allowing only a select few into the cleft. The spacers, made up of naphthalene-like molecular segments, keep the carboxyl groups separated from each other.

"The molecules are quickly assembled in the laboratory, they can be adjusted for size and shape, and their chemical linings can be altered," says Rebek. "This high degree of control is a dramatic improvement in exploring the rules of recognition."

By changing the nature of the carboxyl groups at the molecule's tips and by altering the spacing between the groups, Rebek can tailor his compounds to recognize a wide variety of specific substrates, including acids, metal ions and certain neutral compounds. "These concave clefts can be lined with all sorts of functional groups," says Rebek. "You can make whatever microenvironment you want." The trick is to find complementary groups that will attract the appropriate substrate to make a perfect match.

Like the work of Cram and Breslow, Rebek's research also leads to an improved understanding of how large molecules found in biological systems function. Rebek's approach is unique because his molecular mimics are so small. Both the binding process and the chemical reaction take place close together inside the structure. "Rebek's systems have the elegance of simplicity," says Cram.

"It seems reasonable that clefts could now be designed and efficiently assembled to recognize almost any small molecule or ion," Rebek writes in the March 20 SCIENCE. "Larger substrates such as carbohydrates, peptides and nucleotides are likely future targets for recognition." One group of molecular clefts is so good at trapping calcium ions that Rebek has applied for a patent, and the process is being tested for medical and industrial use.

"The opportunities for fruitful research in the host-guest complexation field are boundless," writes Cram in the December 1986 issue of ANGEWANDTE CHEMIE (International Edition in English). Host molecules that specifically bind any ion of the periodic table remain to be synthesized and tested. Practical methods for extracting gold or uranium from seawater still elude researchers. A variety of organic guests such as carbohydrates and fats have yet to be complexed.

"One of the beauties of this field," says Cram, "is that it's big enough to embrace a tremendous number of different approaches." He adds, "Research in this field is particularly rewarding because scientific and aesthetic content merge and become visible in the structures of the many complexes." □

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Birding Around the World: A Guide to Observing Birds Everywhere You Travel — Aileen R. Lotz. The objective of this book, according to the preface, is to encourage the pursuit of birdwatching in fascinating places around the world or anywhere you travel. Includes information to help the local birdwatcher "graduate into the world birding class." Suggests localities around the world for birdwatching, and lists birding tour operators worldwide and selected "birder-friendly accommodations" in North America. Dodd, 1987, 266 p., illus., $18.95, paper, $10.95.

Brown Bear Summer: Life Among Alaska's Giants — Thomas Bledsoe. At McNeil Falls in Alaska, brown bears fish, fight, mate and play with little or no regard for their human observers, according to the author. This wildlife biologist spent three summers on a Utah State University research project studying the brown bear. The book is a fascinating account of this bear's behavior. Dutton, 1987, 249 p., line drawings by Elizabeth Mills and photographs, $18.95.

Doing Things: A Guide to Programming Activities for Persons with Alzheimer's Disease and Related Disorders — Jitka M. Zgola. Although the book is written for adult day-care programs, its insights translate easily into care for persons living at home. Provides an understanding of the working of the impaired mind and of effective approaches to use with patients with dementing illnesses. Johns Hopkins, 1987, 149 p., illus., $20, paper, $8.95.

The Gardener's Illustrated Encyclopedia of Trees & Shrubs: A Guide to More Than 2,000 Varieties — Brian Davis. Offers information on trees and shrubs together with color photographs illustrating many of these plants and highlighting their outstanding seasonal features. Entries include details on plants' origins, their use for particular sites and descriptions of foliage, flowers, fruit and bark. Practical details about soil conditions for growing, hardiness, height over a period of years, sun/shade and pruning requirements and propagation are also included. Both the botanical and common names are given and indexed for each tree and shrub. Rodale Pr, 1987, 256 p., color illus., $24.95.

Heroes in Space: From Gagarin to Challenger — Peter Bond. Tells in detail the fascinating story of the first 25 years of manned spaceflight in the United States and the Soviet Union. Focuses on the human aspects of space travel rather than the technology. Basil Blackwell, 1987, 467 p., illus., $24.95.

Scientific Genius and Creativity: Readings from SCIENTIFIC AMERICAN — Introduction by Owen Gingerich. Biographies of 10 great scientists representing science from the Renaissance into the Atomic Age. Each biography shows how a major breakthrough in science was achieved. Together these biographies raise a provocative question, says the introduction, about the role of scientific genius in the shaping of science. Includes articles by Jacob Bronowski and Gunther Stent that probe the nature of scientific creativity and discovery. W H Freeman, 1987, 110 p., illus., paper, $11.95.