had to be boiled off in all of the rocks at a fixed rate. This, says Silver, should not have happened, especially when one assumes that all the rocks would not have had the same ratios to start with. What was expected was that the ratios of lead would be spread out—say from 3.6 to 4.6 billion years, but they weren't. Why is not yet understood.

Another example is with sample 14163. This sample, says Silver, has already shown that some parts of the lead could not have formed more recently than 4 billion years ago, and it probably includes some components considerably older than 4.0 billion years. Silver heated the sample. At 550 degrees C. the lead that came off had very high lead 207 to 206 ratios. One would have expected to see a ratio of 0.6 lead 207 to 206 for lead that had been forming continuously since 4.5 billion years ago. But what he saw were ratios of 1.2 or 1.3. "This isotopic composition has never been observed anywhere in the material of the solar system," says Silver. If these lead ratios were interpreted as other ratios, the lead would have apparent ages as high as 5.5 billion years. But, says Silver, "We are probably looking at lead 207 made very early in the solar system before it could be diluted with lead 206, and this large amount of lead 207 has had more time to move around." Lead that is similarly bound comes off at the same temperatures. There is usually a correlation with the age of the lead, but the implications of this are not fully understood.

Tatsumoto and Doe have been working with lead at different temperatures (1,000 to 1,350 degrees C.), and they are getting similar results. The most significant has been isolating lead that consistently dates at 4.6 billion years old (SN: 12/18/71, p. 423).

The problem of how much lead was around to begin with still remains. This could be partially solved by dating all of the soil samples from the moon, determining the over-all effects on each soil sample and getting a convergence point.

The broader implications of the history of volatile metals are apparent even if not all of the results and answers are yet. Volatile metals such as mercury, lead, zinc, cadmium, bismuth, rubidium and potassium are important to man. If scientists could unlock the history of these chemical reservoirs—what the chemical pot started from, how it evolved and what makes it work—says Silver, and if they could understand these processes on the moon, they might know how to use them today on earth and predict for tomorrow. "We don't know the total chemistry of the earth, but our best chance of understanding it is on the moon." □

# Taking proteins apart

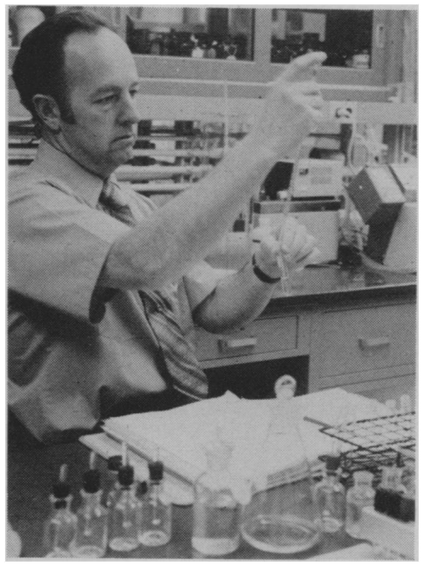
**Living cells take proteins apart and reuse their amino acids. Research is beginning to probe the mechanisms of the process.**

by Dietrick E. Thomsen

Living cells continually synthesize proteins from amino acids and continually take proteins apart. In healthy tissues the mass of protein inside the cells is kept in equilibrium by a balance between synthesis on the one hand and degradation and secretion on the other.

The synthesizing part of this cycle is a fashionable topic for biochemical investigation today, but, says J. Ken McDonald of the National Aeronautics and Space Administration's Ames Research Center, "the effort being expended on the investigation of intracellular protein degradation is barely perceptible by comparison."

Yet the degradation process is as important as synthesis. It involves, as McDonald puts it, the recycling of used proteins for the formation of new ones. One instance in which its role may even be predominant is the atrophy of tissue. The most visible example of atrophy is the shrinkage of muscles in a limb that is disused, because of paralysis or for some other possible reason. But there are other possibilities. The condition of weightlessness, to which astronauts are subject, may induce disuse atrophy in skeletal and heart muscles, bone and cartilage. Atrophy may result from a decreased rate of synthesis, an increased rate of degradation or a combination.



NASA Ames Research Center

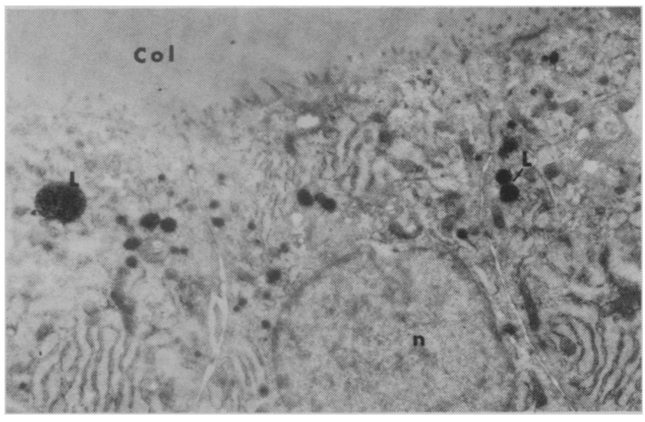
*McDonald: Recycling used proteins.*

Proteins and polypeptides consist of long chains in which the individual units are various amino acids. The route of degradation from protein to single amino acids involves a series of steps in which the compounds are broken into smaller and smaller pieces. The breaking occurs under the chemical influence of certain enzymes, and study of the action of these proteolytic enzymes is expected to lead to an understanding of the intracellular degradation process.

Proteolytic enzymes not only accomplish the recycling of proteins, they also produce substances with important physiological effects.

For example, in certain cases of physiological stress a particular proteolytic enzyme normally present in blood plasma in an inactive state releases a peptide called bradykinin from a plasma globulin. Bradykinin is one of the most potent agents known for dilating blood vessels and lowering blood pressure; it is believed to be instrumental in producing shock. A converse to bradykinin is angiotensin, which raises the blood pressure. Angiotensin is also formed in the blood, and requires the action of proteolytic enzymes from both kidney and lung. Once their intended physiological effects have been produced, these polypeptides, in their turn, are degraded to inactive substances by yet other proteolytic enzymes.

A third reason to study proteolytic enzymes is that they can degrade hormones such as growth hormone, adrenocorticotrophic hormone (ACTH), parathyroid hormone, thyrocalcitonin, vasopressin and insulin into inactive substances. All these are polypeptides, or small proteins. Certain of these hormones—for example growth hormone and thyrocalcitonin—may perform significant actions offsetting detrimental atrophic changes that occur during weightlessness. If they do, it would be important to know how their activities could be regulated.




*Electron micrograph of a thyroid cell (15,000x): The dark bodies are lysosomes that show the presence of DAP II.*

Ken McDonald

The properties of particular proteolytic enzymes and the specific substances that each will work on are areas in which there is still a great deal to be learned. Says McDonald: " . . . it is not possible at the present state of our knowledge to describe the metabolic course of degradation in terms of specific enzymes of even one intracellular polypeptide or protein."

One part of the action that is becoming known, through the work of McDonald and his chief, Stanley Ellis, is the chemistry of a particular class of proteolytic enzymes they have discovered. These enzymes go by the name dipeptidyl aminopeptidase (DAP), and their characteristic property is that they remove dipeptides (groups of two amino acids) in sequence from longer polypeptide chains. Initially, other enzymes break large proteins into polypeptide chunks, and finally still others separate dipeptides into single amino acids. The single amino acids then go back into the pool for reutilization in synthesis.

There are four DAP enzymes now known. The one that has undergone the most study is DAP I, which is especially interesting because of the wide variety of polypeptides it can break. The others are more selective about the substances they will work on. (DAP I was formerly known under the name cathepsin C, but its activator requirements and its broad range of action on polypeptides were not noticed for a long time. McDonald and his collaborators have suggested the change in nomenclature to emphasize what they consider to be its most important physiological activity.)

McDonald says that he and his collaborators first came to appreciate the properties of DAP I when they encountered the enzyme in extracts of pituitary glands, whereupon they purified it and showed that it could inactivate and degrade ACTH. They found that the degradation of the hormone by DAP I proceeded in an orderly fashion, starting from one end of the polypeptide chain and breaking every second bond. There are only two kinds of bonds that it will not break, and when it reaches them, it stops. But even in cases of partial breaking, important physiological consequences may follow. For instance angiotensin II is a polypeptide composed of eight amino acids. The first pair of amino acids on the chain, aspartic acid and arginine, are essential for the hypertensive activity. When they are cut off—and DAP I will do the job—angiotensin II is rendered inactive. In fact all the hormones shown in the chart are inactivated with the very first bond cleaved by DAP I.

Exactly how DAP I cleaves the bonds is not yet known, but McDonald has shown that the action depends on the presence of negative chlorine ions. The chlorine ions may help by making some essential contribution of electric charge at the point where the action takes place, or they may be involved in some interaction at a number of points on the enzyme molecule, by which the chlorine ion acts as a so-called effector, helping the substance that is being worked on to bring the enzyme into a catalytically active state.

The intracellular locations where the DAP enzymes do their work appear to be particular organelles. DAP I and II occur within structures called lysosomes, whereas DAP III is freely soluble in the cytoplasm. Researchers in Finland have located DAP IV in microsomes. The DAP enzymes are found in most tissues, but their proportions vary. DAP I is richest in liver; DAP II is richest in thyroid.

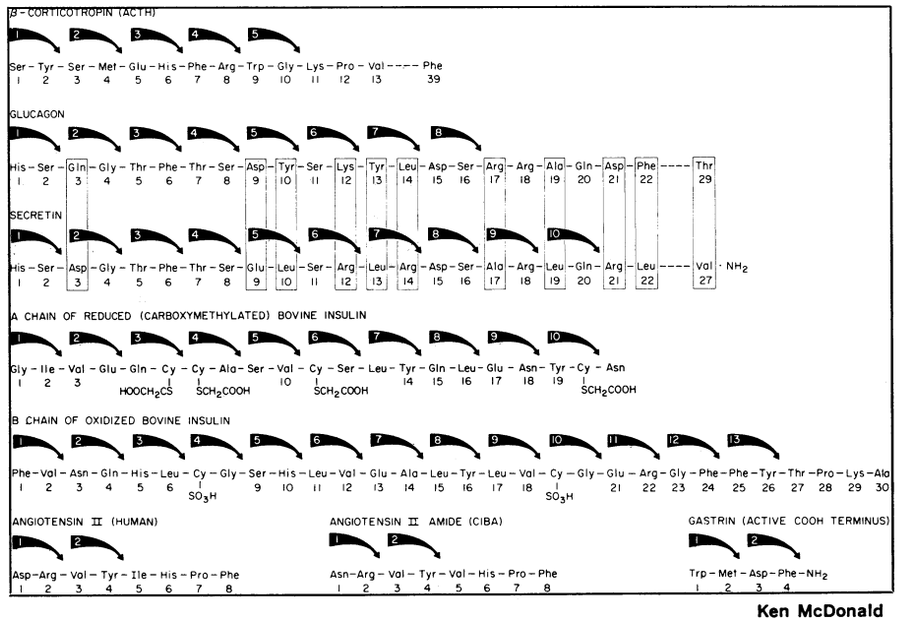
But it is one thing to know that these enzymes can enter into chemical reactions that result in breaking certain polypeptides into dipeptides; it is another thing to prove that they play such a role in living tissue. Experiments in living tissue designed to check their action are necessary.

There are of course indications. McDonald asks the rhetorical question: "What is the role of DAP I in regulating blood levels of angiotensin II?" He proceeds to answer that the liver and kidneys are two of the important sites where angiotensin II is rendered inactive. DAP I is rich in liver and kidney lysosomes. The combination gives a certain circumstantial evidence.

Similarly the DAP enzymes are suspected of a role in tissue atrophy. Proving it will require experiments in which atrophy is induced in animal muscles by limb fixation or severing nerves so that what happens can be followed in a controlled way.

The ultimate hope of this and other work on proteolytic enzymes is that some therapeutic procedures may eventually come out of it. It may someday be possible, speculates McDonald, to use hormones or inhibitors to retard or control enzymes responsible for atrophy.

But it will be a long time before any of these promises are near fulfillment. The work is still mostly in a very basic biochemical stage; it is a long way from clinical application. □




Ken McDonald

*The sequence in which DAP I breaks bonds in various polypeptides.*