tract the information needed for their particular experiment. The tau was first noticed in this way by Martin Perl of Stanford and his collaborators from SLAC and the Lawrence Berkeley Laboratory in experiments with the Stanford storage ring.

It is an interesting coincidence that the signature for the tau, as for the upsilon, is two leptons, although in this case it is the combination of an electron and a muon rather than two muons. Perl's collaboration first reported the so-called e-μ events more than 2 years ago, but the heavy lepton hypothesis was only one of many possible explanations. A particle related to the J/psi could have been responsible, for example. Since then data gathered by Perl's and other groups at Stanford and by two groups using the DESY storage ring seem to have eliminated the possibility that particles containing charmed quarks are involved. The most widely accepted explanation is that two particles, the heavy lepton and its antiparticle, are produced in the collision between electrons and positrons and that these decay into the electron-muon pairs that are detected.

But the job of substantiating this hypothesis is likely to be a tougher task than that of tying down the upsilon. Part of the difficulty is that, as the heavy leptons decay into electrons and muons, neutrinos are also released. Since the neutrinos are not detected, not all the information investigators need to reconstruct the event is available. Elucidating the tau, then, is a matter of accumulating various, somewhat circumstantial data which, taken together, build up a strong case for the heavy lepton.

Some of this information is already available. Perl's collaboration at Stanford, for example, has acquired about 200 events over a 4-Gev-wide energy range; analysis of these events revealed the momentum distribution of the electron and muon, the angle between the two particles, and the probability of producing the electron-muon pair, all as a function of collision energy. This information points to a mass for the tau of about 1.9 Gev, making it 18 times as heavy as a muon. Similar data have been collected by a group at DESY using a detector called PLUTO, which is akin to the detector at Stanford. Although PLUTO does a better job of discriminating between events with three and with two particles and thus the data is "cleaner," only two dozen e-μ events have been found so far.

Besides searching for electron-muon pairs, Perl's collaboration and the PLUTO group have studied events, which are somewhat more numerous, consisting of a muon and any other charged particle; and other groups at Stanford and at DESY, which use detectors that are especially efficient at detecting electrons, found events consisting of an electron and any other charged particle. All results so far are consistent with the heavy lepton interpretation, but none are definitive.

Whatever the outcome of the investigations into the natures of the upsilon and the tau, physicists will likely remain as excited as they are now, for, if the new quarks and leptons fail to materialize, the particles will represent something even more novel and unexpected. New quarks and leptons will be interesting enough, however, because of the seeming proliferation of these most elementary particles. Moreover, the proliferation is further accelerated because physicists expect, for reasons having to do with the symmetries imbedded in the theories describing elementary particles, quarks and leptons to come in certain patterns. For example, in one version of the theory the particles come in pairs. Thus, in addition to the fifth quark and fifth lepton suggested by the experiments, there may be a sixth quark and a sixth lepton lurking in the vicinity and waiting to be found out.

The simplest symmetries are readily seen in the patterns of the four quarks and leptons known previously. Among the four quarks, the so-called up and down quarks and the strange and charm quarks seem to be connected in special ways that are manifested in the manner in which hadrons interact and decay. If this pattern were to be continued, notes theorist Fred Gilman of SLAC, then one would expect a sixth quark to be associated with the fifth, one being the top and the other the bottom quark. Gilman adds that two kinds of experiments would help sort out this pattern: ascertaining the electrical charge of the new quark and determining how particles related to the upsilon particle that contain this quark decay into other particles.

A parallel situation holds among the leptons, where the electron and the muon are each associated with neutrinos (the electron neutrino and the muon neutrino). The natural expectation is that there is a tau neutrino as well, although patterns other than the pairs are also conceivable and certainly not yet ruled out. Again, according to Gilman, the way to unravel this question is to make detailed observations of how the tau particle decays.

In their more expansive moments, physicists muse about the significance of an increasing number of elementary particles. At the moment there is no theory that predicts what and how many elementary particles there are. In the past, numerous physicists point out, the same sequence of events has been followed as the atom, the nucleus, and the hadron—each once thought to be elementary particles—have successively been shown to be composites of more elementary entities. A proliferation of particles accompanied by an underlying structure always seems to signal a new and more fundamental type of particle. If more and more quarks and leptons continue to be found, the question may well become: What are quarks and leptons made of?

—ARTHUR L. ROBINSON

Neutron Scattering: New Look at Biological Molecules

Although biochemists have achieved a great deal of success in determining biological structures, there are still gaps in the accumulating body of knowledge that have been difficult to fill by conventional techniques. Now, however, advances in the biological applications of neutron scattering are providing structural information not previously obtainable. The techniques have proved especially valuable for elucidating the three-dimensional structure of ribosomes (small cellular particles where protein synthesis occurs) and chromatin (the complex of genetic material and protein in the nuclei of higher cells). Equally promising is the application of the techniques to the analysis of cell membrane structure. In a third area of investigation, neutron beams have been used to probe the arrangement of atoms in crystalline materials, including proteins.

In this country research into the biological application of neutron scattering is still a relatively small effort, partly because of the cost of running the experiments which require a nuclear reactor to

produce the neutron beam. At present, only two laboratories in the United States are equipped for biological research with neutron beams. These are Brookhaven National Laboratory, where biological research with a high-flux-beam reactor originated about 10 years ago, and the National Bureau of Standards (NBS) where this type of work is just getting under way. According to Benno Schoenborn of Brookhaven, who began the biological program there, the cost of the neutron beam alone amounts to $250,000 per year. But Schoenborn says that neutron diffraction is a powerful technique and well worth the money; it can reveal structural and functional details not accessible by either x-ray scattering or electron microscopy, two of the major tools with which biochemists explore the structures of large molecules and molecular aggregates. The techniques are not competitors, however; rather they are complementary methods that may reveal different facets of molecular structure.

Neutron and x-ray scattering techniques have much in common. When a beam of either kind of radiation strikes an object, some of the neutrons or x-rays are bent or diffracted out of the path of the incident beam to form a diffraction pattern that is determined by the object's structure. Information about the structure can then be derived from a mathematical analysis of the pattern.

The principal difference between neutrons and x-rays is that the latter interact with electrons whereas the former interact with atomic nuclei. As a result, hydrogen atoms, which have only one electron apiece, are invisible to x-rays but are detected by neutrons. In addition, hydrogen and deuterium scatter neutrons very differently; the two isotopes can be easily distinguished because the neutron scattering density of deuterium is much greater than that of hydrogen.

It is these two properties that biochemists are finding so useful. In neutron crystallographic studies of proteins, the investigators can for the first time unequivocally locate the positions of hydrogen atoms in addition to those of the other atoms. Hydrogen atoms are important in protein structure because of their ability to form bonds that help to hold the protein molecules in their characteristic three-dimensional shapes. Moreover, the mechanisms of catalysis of many enzymes involve the participation of hydrogen atoms, usually as protons. In an early study, Schoenborn used neutron crystallography to identify hydrogen atoms and bonds in the protein myoglobin. He says that the technique should be equally effective in spotting the specific hydrogen atoms that participate in catalytic mechanisms, although this has not yet been attempted.

For molecules that crystallize and consequently have highly ordered structures, neutron and x-ray crystallography can be used to determine the positions of individual atoms. This high degree of resolution is not possible for more complex materials, such as membranes and ribosomes, that do not crystallize, but low angle scattering studies can give a great deal of information about their overall shapes and the arrangement of their different components. (For low angle studies only beams diffracted less than 5 degrees out of the path of the incident beam are measured. Individual atoms do not contribute to this type of scattering which depends only on the size and shape of the whole molecules or particles.) Schoenborn says that neutrons are superior to x-rays for these studies because the neutron scattering densities of biological molecules vary much more than the comparable x-ray scattering densities. In addition, substances that can be labeled with deuterium will stand out even more. Deuterium is a good label for biological molecules because it does not distort their structures and the labeled molecules retain their activities.

Ribosome an Ideal Target

Donald Engelman and Peter Moore of Yale University are taking advantage of this phenomenon in their studies, performed with Schoenborn at Brookhaven, of ribosomal structure. They say that the ribosome is a perfect candidate for analysis by neutron scattering. It is too large to be studied by x-ray diffraction and too small for its features to be fully resolved by electron microscopy. And the ribosome, on whose surfaces amino acids are linked together to form proteins, occupies a central role in the flow of information from the DNA of the genes to protein structure.

Chemical studies have shown that ribosomes consist of two subunits. In *Escherichia coli* the larger subunit contains 34 proteins and two molecules of RNA; the smaller one consists of 21 proteins and one molecule of RNA. Engelman, Moore, and Schoenborn have been concentrating on the smaller subunit. They are now in the process of building a three-dimensional map of the location of all 21 proteins. They have now determined the relative positions of six of them.

Their approach is to use neutron scattering to measure the distances between the centers of mass of pairs of the proteins. As measurements of the pair distances accumulate, the positions of the proteins relative to one another can be determined by the process of triangulation. The investigators need to determine a minimum of 74 pair distances to locate the 21 proteins.

To measure the pair distances, Engelman, Moore, and Schoenborn first replace two of the proteins in the small subunit with the deuterated forms. This is possible because the ribosomal subunits can be disassembled into their component parts and then put back together without losing biological activity. If the labeled subunits were then suspended in pure water, the neutron beam would detect the contrast between the proteins containing deuterium and those containing hydrogen. But it would also detect an interfering signal produced by the contrast between the subunits as a whole and the water. However, a process called "contrast-matching" can eliminate the interfering signal by effectively making the nondeuterated components disappear.

Contrast matching is possible because the neutron scattering densities of most nondeuterated biological molecules lie between those of H_2O and D_2O, which are quite different. By mixing H_2O and D_2O in the appropriate proportions, investigators can produce a solvent with a scattering density equal to that of any of the molecules. When the scattering density of the solvent for the small ribosome subunits equals that of the average for the nondeuterated portion of the subunits, there is no contrast between them, and only the deuterated proteins remain visible to the neutron beam.

The two labeled proteins diffract the incident neutron beam. The distance between the particles can then be deduced from the interference pattern that results. The neutron diffraction data can also be used to derive information about the shapes of the ribosomal proteins. Moore points out that comparable structural information about the ribosome cannot be obtained by more conventional methods. Some of these are helpful in locating certain portions of the different protein molecules, but many of the proteins are large enough to extend for some distance through the ribosomal subunit particle and their shapes cannot be deduced by the other techniques.

The chromatin of nucleated cells is another complex of nucleic acid and protein whose structure is yielding to neutron scattering analysis. Here the nucleic acid is the DNA of the genes and the proteins are the histones that help to regulate gene expression. Investigators would like to know more about the relative arrangements of the DNA and the histones because such information

would help them to better understand both the mechanisms by which genes are turned on or off and also the manner in which the DNA itself is duplicated.

They already know that chromatin consists of repeating globular subunits called nucleosomes that are strung together like beads. Recently, E. Morton Bradbury and his colleagues at Portsmouth Polytechnic in Portsmouth, England, confirmed by neutron scattering that the beadlike nucleosomes consist of a histone core with the DNA on the outside where it can be readily recognized by enzymes that synthesize DNA or RNA. The nucleosomes are flat cylinders with overall dimensions of 11 by 11 by 5 nanometers. The Portsmouth group is now producing nucleosomes containing deuterium-labeled DNA or histones in order to determine the relative arrangement of the nucleosome components by a process analogous to that used by the Yale investigators to study ribosomal structure.

Investigators also use deuterium-labeled molecules and contrast-matching to aid in their neutron diffraction analyses of membranes. Cell membranes are complex structures that contain lipids and proteins arranged in layers. The nonpolar components are usually sandwiched between two layers bearing the polar or charged regions. Membranes participate in the regulation of such cell functions as the transport of materials into and out of cells. Properly functioning membranes are needed for nerve and muscle cells to respond to stimuli, for example. Although the investigation of membrane structure and function is one of the most active areas of biochemical and physiological research these days, interpretation of the data is often hampered by difficulties in determining how the various components are arranged. Proponents of neutron scattering think that the techniques can clear up some of the uncertainties.

Several investigators, including J. Kent Blasie of the Johnson Foundation of the University of Pennsylvania, Donald Caspar of Brandeis University, and Schoenborn have demonstrated the feasibility of examining the structure of both artificial and natural membranes with neutrons. The most ambitious membrane study in the Unites States, however, has been carried out by Mark Yeager of Yale on the rods of intact frog retinas. The rods contain the visual pigment rhodopsin and are the light-detecting cells of the retina. Much is known about the chemical changes induced by light striking rhodopsin; far less is known about how this chemical change is converted into a nerve impulse.

It is known that each rod contains a stack of approximately 1500 discs and and that each disc consists of two membrane layers in which the rhodopsin is thought to be imbedded. There is some controversy about just how the rhodopsin is oriented in the discs. But Yeager now says that his neutron diffraction studies indicate that the pigment is located on the outer faces of the discs from which it extends into the cytoplasm between them. He thinks that it should be possible to observe the structural changes that occur in the disc membranes as a result of illumination by comparing neutron diffraction patterns from specimens in the dark with those from specimens in light.

Low Fluxes Slow Research

When asked what are the greatest problems hindering the biological applications of neutron scattering, the investigators uniformly cite the low flux of the neutron beams and the high cost. Alexander Wlodawer of the NBS says that the neutron flux in the best beams is 6 orders of magnitude lower than the available x-ray fluxes. In order to get measurable quantities of diffracted neutrons, investigators have to use large samples and expose them to neutrons for long periods of time. Whereas the data for an x-ray crystallographic experiment can be acquired in a few days, that for neutron crystallography may require a few months to collect.

Unlike x-rays, however, neutrons do not cause the samples to deteriorate. A protein crystal may be used repeatedly for neutron crystallography studies without being damaged. Membranes, however, are not as stable as crystals and the long times (hours are needed for the low angle scattering experiments) required for the studies can be a handicap.

Both the Brookhaven and NBS investigators are trying to improve the efficiency of the data collection and decrease the time needed for it. One way to tackle this problem is to increase the number of diffracted waves that can be collected at one time. Linear detectors capable of simultaneously measuring the neutron flux at many points on a plane have already been installed at both Brookhaven and NBS, and, in addition, the former now has an area-wide detector that can be used for low angle scattering studies.

Another obvious approach is to try to increase the neutron fluxes. Room temperature neutrons are currently used for all biological experiments in this country. These are relatively slow neutrons, most of them having wavelengths of about 1.5 angstroms. Neutrons at this wavelength are very good for crystallographic work but less suitable for the analysis of larger structures. Cooling the neutron beam would slow the neutrons and thus increase the number having higher wavelengths that are better for low angle scattering studies of materials like ribosomes and membranes. Devices to do this, which are called cold moderators, are being installed at Brookhaven and NBS, but are not yet ready for use.

Meanwhile, the reactor of the Institut Max von Laue-Paul Langevin in Grenoble, France, already has a working cold moderator. In fact, most observers agree that this reactor now has the best facilities in the world for exploring the biological applications of neutron scattering. The Europeans generally have been mounting a much larger effort in this research area and are currently involved in a variety of studies on ribosomes, membranes, chromatin, and other biological structures. According to a report by the National Academy of Sciences, no more than 22 scientists in the United States in 1976 participated in neutron scattering studies of biological systems, but at least 80 scientists did so in Europe.

The technical problems encountered in neutron scattering analysis may ultimately prove easier to resolve than the financial ones, however. Biologists are not as accustomed as physicists are to the kinds of expenses incurred in doing experiments involving nuclear reactors. The costs are sufficiently high to make the programs very visible at a time when many research facilities have to cut back to fit a tight budget. The budget for the biological neutron scattering program at Brookhaven has not been decreased, but neither has it increased during an inflationary period. Schoenborn says that he is particularly concerned because he was not able to replace staff members who have left.

The situation is further complicated by the fact that neither the Department of Energy, which is taking over Brookhaven, nor the Department of Commerce, which runs NBS, is primarily concerned with biological research. Wlodawer points out that nuclear reactors are built and run by physicists; not surprisingly, they do not always relish the prospect of giving up a beam or two to the biologists.

The situation may be alleviated by a new facility for low angle neutron scattering now being planned by the National Science Foundation (NSF). It will include provisions for biological research. Lewis Nosanow of NSF says that he expects a decision about where the facility will be located no later than the beginning of the new year.—JEAN L. MARX