Simple sugars are key to complex sex

It is common knowledge that fertilization in higher animals results from a fusing of egg and sperm. But only recently have scientists begun to describe the molecular mechanisms that govern the binding and interaction between these two unique cell types.

New research reported in Boston two weeks ago provides some of the finest details yet of the mechanics of fertilization, with potential implications for the fields of contraceptive technology and *in vitro* fertilization. Presented at the annual meeting of the American Association for the Advancement of Science, the research takes a bit of the mystery out of the miracle of conception, and suggests that fertilization may hinge on the mere presence or absence of a couple of hydrogen atoms on the human egg.

The study, led by Paul M. Wassarman of the Roche Institute of Molecular Biology in Nutley, N.J., focused on the insoluble outer coat, or zona pellucida, of the mammalian egg. Previous research had shown that the zona contains three varieties of glycoproteins (protein chains with carbohydrate branches), called ZP1, ZP2 and ZP3, and that ZP3 contains the specific receptor to which a sperm must bind to induce fertilization. But little was known about which part of the ZP3 molecule is critical to this binding, and even less was known about the "hardening" of the zona that follows within moments of fertilization. Zona hardening prevents subsequent sperm from also fertilizing the egg – a phenomenon called polyspermy – which would result in an overabundance of DNA in the developing egg and in most cases cell death.

Working with mouse eggs, which are very similar to human eggs, Wassarman focused on the so-called O-linked oligosaccharides, complex sugar chains that grow like branches off ZP3's protein "trunk." He induced minor changes in the branches to see which part of the oligosaccharide molecule actually binds to sperm. He reports that the six-carbon sugar alpha galactose – a common sugar that, when bound to glucose, makes the milk sugar lactose – appears to be the critical link to sperm recognition. Moreover, he found, if the No. 6 carbon on alpha galactose is oxidized to its aldehyde form, C-H = O, it fails to recognize sperm. In its reduced form, CH_2-OH, the molecular complex binds to sperm, triggering a cascade of reactions that leads to the dissolving of cell membranes and the fusing of genetic material from the sperm and egg.

Although the simple loss of two hydrogens can thus make ZP3-bound galactose unrecognizable to sperm, Wassarman says that zona hardening following fertilization is probably a more complicated process than simply this. He hypothesizes that conformational changes in ZP3 chains following fertilization may activate specific enzymes to clip ZP2 chains into smaller pieces. Its structure thus disturbed, the molecular mesh surrounding the egg may collapse a bit, squeezing out water and making the zona impenetrable to late-arriving sperm.

It's possible, say Wassarman and others, that scientists may develop molecular biological techniques to induce zona hardening before fertilization, resulting in a very natural sort of contraception. Similarly, it may be possible to either produce antibodies against the sperm-binding oligosaccharides or mass-produce synthetic oligosaccharide to coat approaching sperm, thus blocking fertilization (SN: 7/21/84, p.38).

Alternatively, in the field of *in vitro* fertilization, a better understanding of zona hardening may make it easier to tell if a retrieved egg is still capable of being fertilized. Currently, scientists must look at retrieved eggs under a light microscope to determine an egg's viability and susceptibility to fertilization. That technique is less than perfect, however, and can result in expensive, futile attempts at fertilization. *– R. Weiss*

Identity crises in AIDS virus studies?

With so much still unknown about AIDS, each additional piece of information is welcome. But data must be gathered carefully and critically, according to recent reports illustrating the potential pitfalls of virus identification. In two studies released last week, one group of researchers suggests that the most virulent immunodeficiency viruses may escape isolation and detection by current laboratory methods, while another presents evidence that a virus isolate thought to be HIV-2 is actually a contaminant from monkeys.

By studying a virus responsible for feline leukemia, researchers at the Harvard School of Public Health in Boston and at Colorado State University in Fort Collins found that procedures used to separate viruses may fail to isolate "acutely pathogenic" (disease-producing) virus strains while selecting for less virulent viruses. The scientists, hoping to develop an animal model for AIDS, have been focusing on a naturally occurring virus strain they call FeLV-FAIDS, which also causes a fatal feline immunodeficiency syndrome.

In 1986, when the scientists announced that they had found the feline immunodeficiency virus, they also reported that there were several types of FeLV in the tissues of affected cats. Now, after cloning and DNA sequencing these different viruses, they write in the Feb. 19 SCIENCE that the feline viruses causing the immunodeficiency syndrome are "replication defective," or unable to multiply well *in vitro* – suggesting that researchers must take the viruses directly from tissues in order to identify all involved. The authors say that this diversity also occurs among viruses linked to human and monkey AIDS, and that the current approach of growing viruses *in vitro* before study may not isolate the more important strains.

In the second study, it apparently was a problem of too many virus isolates rather than too few. Reporting in the Feb. 18 NATURE, researchers at the New England Regional Primate Research Center in Southborough, Mass., say they have "strong evidence" to confirm scientists' suspicions that a virus used at the Harvard School of Public Health as an HIV-2 isolate is actually a simian immunodeficiency virus (SIV) from rhesus macaque monkeys.

Thought to be associated with an AIDS-like condition primarily found in West Africa, HIV-2 apparently is less virulent than HIV-1, but is considered an important key to understanding the spread of AIDS. Trying to sort out the various viruses somehow affiliated with AIDS, however, can be confusing, as the current report suggests.

The Southborough group isolated the virus the scientists called SIV_{MAC} from captive macaques in 1985. Within two years, the Harvard group and a group of French scientists had reported the isolation from humans of HTLV-IV and LAV-2 respectively, both later renamed HIV-2. The Harvard researchers had also isolated a virus from green monkeys that they called $STLV\text{-}III_{AGM}$, and both the green monkey virus and HTLV-IV were used in subsequent studies in Africa. But the Southborough group's comparison of DNA sequences from HTLV-IV and $STLV\text{-}III_{AGM}$ now shows that the two viruses are actually a variant of SIV_{MAC} that probably contaminated Harvard's cultures.

This mistaken identity is readily acknowledged in an accompanying reply from Harvard's Myron Essex and Phyllis Kanki. They say, however, that – because SIV and HIV-2 can be used "interchangeably" in screening tests – results from epidemiology studies based on what have proved to be lab contaminants "remain as originally stated." Commenting in the same issue of NATURE, Carel Mulder of the University of Massachusetts Medical School in Worcester says that "this episode should serve as a strong warning for all virologists working with multiple isolates." *– D.D. Edwards*