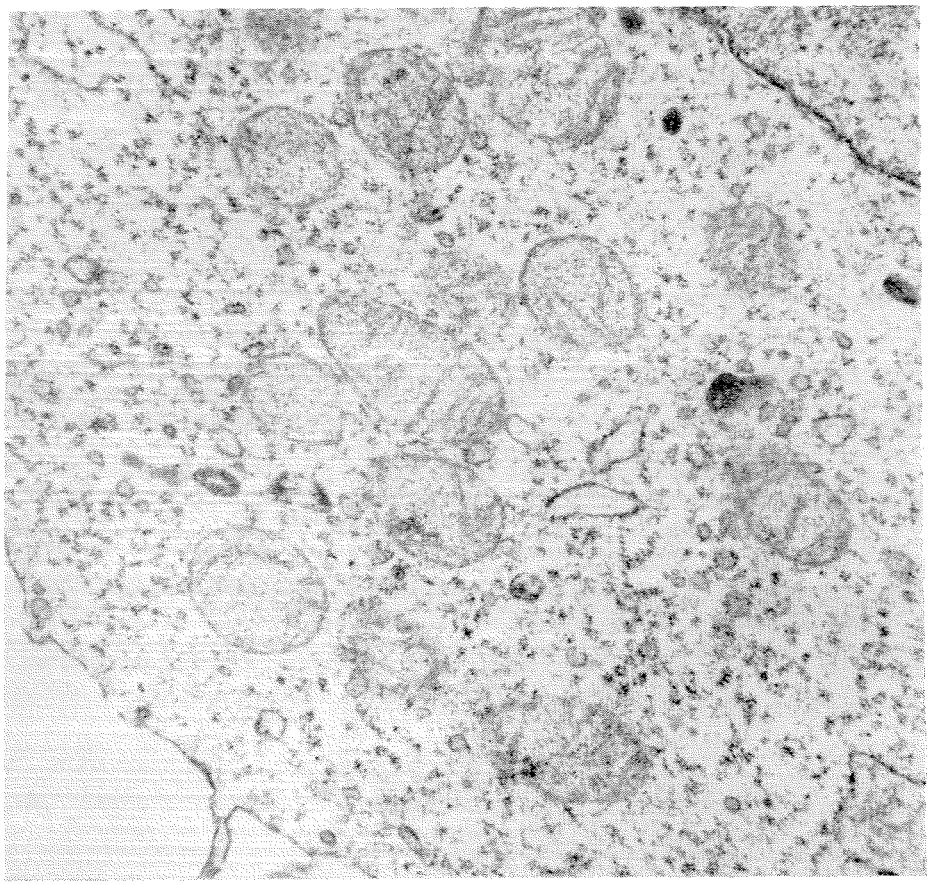

This electron micrograph of a portion of a HeLa cell shows part of the nucleus in the upper right corner. The large oval-shaped bodies are the mitochondria in the cell's cytoplasm. Messenger RNA was found in the free polysomes (the clusters of tiny black dots) and in the vesicles of rough endoplasmic reticulum (the two dark elongated rings to the right of center). (Magnification 34,500)

A Second Genetic System

In the long and compelling search for an understanding of how life perpetuates itself, deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) have been found to be at the basis of the processes involved. Cell growth and differentiation are known to be controlled by genes—represented by DNA segments—located in the nucleus of all animal and plant cells. A fundamental function of DNA is to serve as a template for the synthesis of messenger RNA, which carries coded instructions from the nucleus to the cytoplasm for the synthesis of proteins.

In addition to the nuclear genetic system, biologists have for some time had reason to believe that a second genetic system exists in the cytoplasm of molds, yeasts, protozoa, green algae, and higher plants. The presence of this second genetic system would account for certain phenomena of inheritance which do not follow the classical laws of heredity. This belief received scientific support recently when DNA was discovered in the cytoplasm of many kinds of organisms, in particular in the chloroplasts of plants and in the mitochondria (energy-producing organelles) of various organisms.

Direct evidence for the existence of this separate genetic system, which sends messages to the cell's protein-synthesizing machinery in the cytoplasm of animal cells, has now been obtained by Giuseppe Attardi, M.D., Caltech professor of biology, and his colleague (and wife) Barbara. They have found that a fraction of the messenger RNA, the primary product of the genes, is synthesized in the cytoplasm. This indicates that genes exist and are switched on outside the nucleus.

The Attardis made their discovery by using two kinds of cells cultivated *in vitro*—HeLa cells, derived from human tumor, and a strain of mouse cells. They found messenger RNA in two fractions of the cell's cytoplasm—in polysomes (the protein-synthesizing structures) suspended free in the cytoplasm and in polysomes associated with a membrane fraction of the cytoplasm.

In order to determine the site of synthesis of

A Caltech husband and wife research team finds that a fraction of messenger RNA is synthesized in the cytoplasm of animal cells.

Barbara and Giuseppe Attardi isolate DNA from the mitochondria of the cells of human tumor tissue (HeLa cells) by collecting fractions of a cesium chloride density gradient. This will be used for experiments of hybridization with fractions of messenger RNA also extracted from the cell's cytoplasm.

these two messenger RNA classes, the Attardis carried out two types of experiments. In one they added a radioactive precursor (one of the building blocks) of RNA to growing cell cultures. This tracer appeared rapidly in the membrane-associated RNA, but somewhat later in the free-polysome messenger RNA. This suggested that the free-polysome messenger RNA was first synthesized in the nucleus and then transported to the polysomes, while the membrane-associated messenger RNA was synthesized in the cytoplasm itself.

In order to obtain more direct evidence on this point, the Attardis extracted mitochondrial DNA and the two RNA fractions from HeLa cells. Each RNA fraction was incubated at high temperature in a salt solution with the DNA. The DNA formed molecular hybrids with the membrane RNA, but not with the free-polysome messenger RNA, indicating that the membrane messenger RNA is a "transcription" product (or complementary replica) of the mitochondrial DNA. Other evidence indicated that the major part of this RNA is associated with membrane structures outside the mitochondria, probably with the tubules and vesicles of the rough endoplasmic reticulum that electron microscopy has revealed in HeLa cells. The membrane messenger RNA would thus represent mitochondrial RNA which is exported to the rough endoplasmic reticulum. One fraction of it, however, presumably remains inside the mitochondria, since these organelles apparently are able to support protein synthesis.

The Attardis believe that the nuclear and cytoplasmic genetic systems are coordinated in some way. One possibility, suggested by other evidence, is that the cytoplasmic RNA carries instructions for the synthesis of structural protein components of mitochondria and other membranes of the cytoplasm. To test this hypothesis, the next step in their research will be to determine the nature of the proteins which are synthesized under the direction of this RNA.