The nature of the scrapie agent

TIKVAH ALPER

Medical Research Council Experimental Radiopathology Unit, Hammersmith Hospital, London

Our radiobiological and photobiological experiments on the scrapie agent (Alper, Haig, and Clarke, 1966; Alper, Cramp, Haig, and Clarke, 1967; Alper and Haig, 1968; Haig, Clarke, Blum, and Alper, 1969; Latarjet, Muel, Haig, Clarke, and Alper, 1970) were in the first instance aimed at determining its molecular weight, and in pilot runs we assumed that the agent was, basically, a 'conventional' virus despite its unusual properties. Thus the doses of ionizing radiation used were of a magnitude which would be appropriate for inactivating very small viruses.

Doubts about that assumption arose when we found that the inactivation doses of both ionizing radiation and ultraviolet at 254 nm were much larger for scrapie than for any known biologically active particle (including individual genetic markers) which depends for replication on the integrity of nucleic acid. This meant that the 'target molecular weight' (calculated from inactivation by ionizing radiation) was considerably less than the minimum required to code for replication of an infective agent; and suggested that the agent was transparent to ultraviolet at 'germicidal' wavelengths. Our later experiments have been aimed at testing the hypothesis that the scrapie agent does not depend on a nucleic acid moiety for replication. Alternative mechanisms for replication have been proposed (Gibbons and Hunter, 1967; Griffiths, 1967) within the framework of Crick's 'central dogma' (Crick, 1970).

The chemical nature of an unknown substance may often be inferred from its ultraviolet absorption spectrum. Those pertaining to nucleic acids will depend in detail on the relative proportion of the bases, but all are characterized by a broad maximum of absorption at 260 to 270 nm. The absorption spectrum of the scrapie agent cannot be determined, because preparations can be made only from the tissues of diseased animals and do not contain enough of the agent in relation to other components. An analogous method for inferring chemical constitution of a biological agent is to construct an 'action spectrum': the effectiveness of ultraviolet at a given wavelength in inactivating the particle will be proportional to absorption at that wavelength.

In collaboration with Drs Latarjet and Muel at the Institut du Radium we have examined the

Fig. The shaded area indicates the range of eight separate action spectra pertaining to viruses and genetic markers. The crosses show the relative effectiveness of five wavelengths in inactivating the scrapie agent.

relative effectiveness of ultraviolet in inactivating scrapie at five wavelengths, and so built up an outline action spectrum, as shown in Figure 1. This shows also the range of action spectra for viruses and genetic markers. These are similar to absorption spectra for nucleic acids, since the particles depend for replication on the integrity of nucleic acid. The most effective wavelengths are 260-270 nm; 280 nm is less effective; and 240 nm often of minimum effectiveness. In contrast, 250, 267, and 280 nm were equally effective in inactivating scrapie, but 237 nm was about four times as effective. These experiments have been done three times. The results support the inference that the replication of the scrapie agent does not depend on the integrity of a nucleic acid moiety.

This evidence on the 'nature of the scrapie agent' is negative. In collaboration with our French colleagues we have recently irradiated scrapie agent at other ultraviolet wavelengths and have also examined action spectra for several biological agents

including some which do not contain nucleic acid. We hope that something more positive about scrapie may then be inferred.

References

- Alper, T., Cramp, W. A., Haig, D. A., and Clarke, M. C. (1967). Does the agent of scrapie replicate without nucleic acid? *Nature (Lond.)*, **214**, 764-766.
- Alper, T., Haig, D. A., and Clarke, M. C. (1966). The exceptionally small size of the scrapie agent. *Biochem. biophys. Res. Commun.*, **22**, 278-284.
- Alper, T., and Haig, D. A. (1968). Protection by anoxia of the scrapie agent and some DNA and RNA viruses irradiated as dry preparations. *J. gen. Virol.*, **3**, 157-166.
- Crick, F. (1970). Central dogma of molecular biology. *Nature (Lond.)*, **227**, 561-563.
- Gibbons, R. A., and Hunter, G. D. (1967). Nature of scrapie agent. *Nature (Lond.)*, **215**, 1041-1043.
- Griffiths, J. S. (1967). Self-replication and scrapie. *Nature (Lond.)*, **215**, 1043-1044.
- Haig, D. A., Clarke, M. C., Blum, E., and Alper, T. (1969). Further studies on the inactivation of the scrapie agent by ultraviolet light. *J. gen. Virol.*, **5**, 455-457.
- Latarjet, R., Muel, B., Haig, D. A., Clarke, M. C., and Alper, T. (1970). Inactivation of the scrapie agent by near-monochromatic ultraviolet light. *Nature (Lond.)*, **227**, 1341-1343.