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Tumor Isoimmunity [1]

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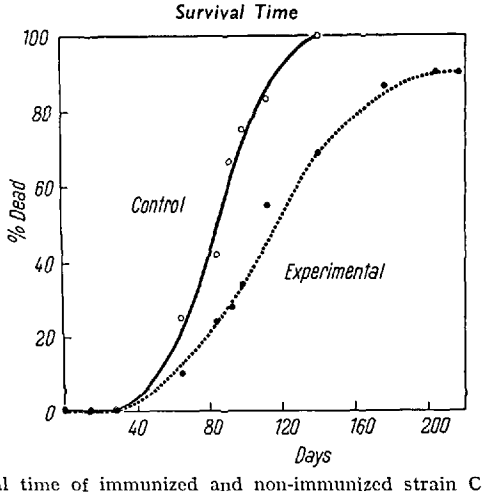
The problem of immunity in inbred animals against their own tumors has been a central one in experimental cancer research for many years. Its importance is due to the fact that the problem of tumor isoimmunity and tumor autoimmunity centers around the question of specific antigenic components present in tumor and absent in homologous normal tissue. A rational approach to tumor therapy by immunological means clearly hinges on this central point. Despite the many papers written on the subject, the desiderata for a critical attack on the problem have not always been realized[3], mainly because a clear distinction between isoimmunity, autoimmunity, and homograft immunity[4] has not frequently been made. Thus, as will be pointed out below, the relevance of much of the work involving long-transplanted or other unsuitable tumors, or unsuitable hosts, to the problem of isoimmunity, and of induced immunity against spontaneous tumor development, remains problematical.

No reference will be made here to the dilemmas which plagued the early workers in the field of tumor immunity and which we now know were clearly ascribable to phenomena involving homotransplantation reactions in genetically heterogeneous populations; this aspect of tumor immunity has been well summarized elsewhere (see, e.g., Medawar's Harvey Lecture[5]). We shall be dealing here, rather, with work which clearly lies or has been assumed to lie within the province of tumor isoimmunity.

Recent experiments by Hirsch *et al.*[3] have demonstrated a slight but definite amount of immunity in strain C (Bagg albino) mice against a homologous challenging tumor of recent spontaneous origin, following a course of immunizations with actively growing tumor of indigenous origin. No differences in number of tumors or in the time at which the tumors appeared were apparent, and immunity expressed itself only in a significantly longer survival time of the treated animals as compared with the control group. Some of the data obtained are summarized in the Figure.



Survival time of immunized and non-immunized strain C (Bagg albino) mice following challenge with strain C tumor. Tumor used for immunization: Strain C tumor in first transplant generation. Mean survival time for experimental group, 124 ± 8·7 days, for control group, 91 ± 7·3 days; P = < 0·01. For experimental details, see [6].

These results tend to indicate that an antigenic difference exists between host and tumor tissue. A number of definite reservations have been pointed out[6] which must be considered before this view can be fully accepted. Such a view is, however, in accordance with preliminary data published by Isojima *et al.*[7] and with findings obtained by methods employing anaphylactic phenomena by Grace[8], and by Zil'-

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[3] H. M. Hirsch, J. J. Bittner, H. Cole, and I. Iversen, Bact. Proc. *1958*, 60; Cancer Res. *18*, 344 (1958).

[4] For purposes of discussion, the terms autoimmunity, isoimmunity, and homograft immunity will be defined in this manner: autoimmunity—immunity elicited in an animal against his own normal or tumor tissue; isoimmunity—immunity elicited in a member of an inbred strain against normal or tumor tissue of another member of the same inbred strain; homograft immunity—immunity elicited in an individual against normal or tumor tissue of a genetically different individual of the same species, or that elicited in animals of one inbred strain against normal or tumor tissue of members of other inbred strains of the same species.

[5] P. B. Medawar, *The Harvey Lectures*, 1956–1957 (Academic Press, Inc., New York 1958), p. 144.

[6] H. M. Hirsch, J. J. Bittner, H. Cole, and I. Iversen, Cancer Res. *18*, 344 (1958).

[7] S. Isojima, R. M. Graham, and J. B. Graham, Proc. Amer. Ass. Cancer Res. *2*, 310 (1958).

[8] J. T. Grace, Jr., Personal communication.

BER[9] (see this paper for a review of pertinent earlier work).

A brief attempt will be made to correlate and interpret the facts described above as well as those found in the literature (for review see [10]), always keeping in mind the strictures alluded to above.

The host does develop immunity against tumors indigenous to his own strain, and possibly even against his own tumor[11], but this immunity is slight and may be overcome by the independent and aggressive behaviour of the tumor. It would seem that HAALAND's dictum 'an animal cannot be immunized against its own neoplastic cells' may, like most dicta, have been too categorical.

Host-tumor interplay must thus be viewed as a phenomenon whose outcome hangs in the balance, but one which is prejudiced in favor of the tumor. Immunization will work the better, the further removed the tumor is from the original host[12], a condition which is facilitated by continued transfer of the tumor, and repeated immunization bolsters the host's immune response[13]. Conversely, the closer the tumor is to the host, genetically, the less the immunity[6]. In fact, as far as transplantability in the original host is concerned, work involving long-transplanted tumors can essentially be considered a problem in homotransplantation rather than isotransplantation. Prolonged serial transplantation of the tumor is not always a necessary prerequisite for such changes in the tumor to occur, and changes in transplantability characteristics may be accompanied by changes in the ploidy of the tumor[14]. On the other hand, the host's immune response, even if once well established, can be overcome by too great a challenge dose[15].

Additional evidence for the view that use of long-transplanted tumors elicits homograft, rather than isograft, reactions with respect to the original host comes from work comparing immune responses of inbred animals against indigenous tumors and against tumors induced by carcinogenic agents. FOLEY[16] has shown that, following removal of transplantable tumors of recent spontaneous origin (mammary adenocarcinomas), no immunity was found to be present under his experimental conditions following challenge with the same tumor (but see [6]), while removal of transplanted tumors recently induced with carcinogens consistently was found to result in immunity against subsequent challenge with the same tumor. FOLEY's work has recently been confirmed by BALDWIN[17] and by PREHN and MAIN[18]. These data are well accounted for on the basis that the carcinogen-induced tumors have undergone more mutational changes than the spontaneous tumors[19] and are thus immunogenetically further removed from the host tissue than are spontaneous tumors. Antigens not present in normal host tissues have been reported in carcinogen-induced tumors by ZIL'BER *et al.*[20], NARTSISSOV and ZIL'BER[21], and WEILER[22].

If a mutated tumor, regardless of its mode of origin, represents a problem in homograft immunity rather than isoimmunity with respect to the reaction elicited following implantation in the original host strain, why is it that such tumors frequently grow well in their hosts of origin? The answer to this apparent contradiction is this: the mutated tumor grows in its strain of origin, and the so-called 'market' tumors even grow easily in any number of strains despite the fact that they have become quite different immunogenetically from their hosts because, during the long-extended periods of transfer usually characteristic of such tumors, cell lines have been selected out which have very great invasive power and growth potential and which are thus able to establish themselves before the immune response of the host comes into play. That this accords with the facts is borne out by observations that it actually is easy to immunize against these tumors by appropriate methods which prevent the initial overgrowth of the host by the tumor[23]; this explanation avoids ascribing to these tumors such attributes as 'reduced specificity' or 'antigenic simplification'.

MARTINEZ *et al.*[24] have recently described conditions under which certain mice proved immune to reinoculation of a transplantable tumor although they were already succumbing to lung metastases formed from the originally inoculated tumor. This phenomenon can be explained on a similar basis; i.e., the metastases were able to establish themselves at a time when no immunity had yet been produced and when, in fact, conditions of enhancement were present. A later reinoculation of tumor was unsuccessful due to establishment of immunity, although this immunity was not

[9] L. A. ZIL'BER, Uspekhi Sovremennoi Biologii *30*, 188 (1950). Translated at the National Institutes of Health.

[10] H. M. HIRSCH, J. J. BITTNER, H. COLE, and I. IVERSEN, Cancer Res. *18*, 344 (1958). – T. S. HAUSCHKA, Cancer Res. *12*, 615 (1952). – A. GOLDFEDER, Brit. J. Cancer *8*, 320 (1954). – E. J. FOLEY, Cancer Res. *13*, 578, 835 (1953).

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[15] L. GROSS, Cancer Res. *3*, 770 (1943).

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[17] R. W. BALDWIN, Brit. J. Cancer *9*, 652 (1955).

[18] R. T. PREHN and J. M. MAIN, J. nat. Cancer Inst. *18*, 769 (1957).

[19] L. C. STRONG, Genetics *11*, 294 (1926); Brit. J. Cancer *3*, 97 (1949).

[20] L. ZIL'BER, N. NARTSISSOV, T. RIVKIND, and Z. BAIDAKOVA, Vest. A.M.N. SSSR. *3*, 36 (1948). – L. ZIL'BER, V. FREIMAN, I. ZBARSKII, and S. DEBOV, Dokl. Akad. Nauk SSSR. *65*, 97 (1949).

[21] N. NARTSISSOV and L. ZIL'BER, Dokl. Akad. Nauk SSSR. *65*, 229 (1949).

[22] E. WEILER, Z. Naturforsch. *11b*, 31 (1956).

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[24] C. MARTINEZ, J. B. AUST, and J. J. BITTNER, Cancer Res. *16*, 1023 (1956).

sufficient, as has frequently been shown before, to interfere with already established tumor growth.

Exactly the same holds true of tumors that have arisen from normal tissues of the mouse *in vitro* in tissue culture. SANFORD *et al.*[25], in appraising the antigenic changes that may occur during this process, have shown that certain cell lines, in contrast to others, can establish tumors in the host precisely because their growth rate is such that they can keep ahead of the host's immune response.

It does not seem impossible, in view of these considerations, that tumor cells originate in the body by mutation or otherwise *all the time*, but that those most removed from the host tissue are destroyed by the body's immunological defense mechanisms; only those tumor cells which are *very similar* to the host tissue thus would have a chance to develop into a tumor because the immune response of the host directed against such cells would be very weak. At variance with this is the finding, referred to above, that carcinogen-induced tumors are immunogenetically further removed from the host than are spontaneous tumors. The question can be asked: If such tumors are antigenically different from their hosts, how can they establish themselves in the first place? The answer may well lie in the finding that a general decline in antibody production occurs soon after the application of carcinogens[26].

The possibility cannot, therefore, be excluded that the cancerous state may be accompanied by a lessened plasticity or lowered response of the reticulo-endothelial system. Carcinogenic and mutagenic agents, such as ionizing radiations, radiomimetic chemicals, and several carcinogenic substances are known to lower immune responsiveness; their use thus might result in both the induction of tumor cells as well as in increased opportunities for such cell variants to establish themselves. The demonstration of a reduced homograft response in cancer patients[27] and of anergic phenomena and altered skin homograft rejections in patients with Hodgkin's disease[28] lends some support to the possibility that this may be a factor also in some spontaneous cancers.

One piece of evidence in support of the hypothesis of spontaneous tumor formation proposed comes from the well-established fact[29] that normal tissue kept in tissue culture, where it escapes the immunological and other restraining influences of normal body control, frequently undergoes cancerous changes. Of course, other causes or trigger mechanisms are by no means excluded for this effect to take place. That such tumorous tissues are, however, different from true spontaneous tumors is shown by the observation of SALK and WARD[30] (see also[25]) that on reimplantation of the tissue-culture-derived tumors into the host of origin, cytotoxic antibodies against the tumors are formed; this has so far not been convincingly demonstrated with tumors of spontaneous origin. In this respect, tumors that have arisen *in vitro* resemble carcinogen-induced tumors which also, in contrast to spontaneous tumors, serve as good immunizing agents in the host of origin[31]. The conclusion seems inescapable that both the '*in vitro* tumors' and the carcinogen-induced tumors are immunogenetically further removed from the host than are tumors of spontaneous origin. In this connection it should be noted that recent experiments demonstrating successful immunization of animals against homologous, strainspecific tumors have, without exception, been based on work employing long-transplanted tumors, carcinogen-induced tumors, hybrid though compatible recipient hosts, or a combination of these[32].

In view of this it would seem that the time is at hand to use, in the study of problems of tumor isoimmunity, spontaneous tumors rather than long-transplanted tumors, tissue culture-derived tumors, or carcinogen-induced tumors. MCDOWELL[33] has pointed out that long-established transplanted tumors offer opportunity for analytical experimentation to discover mechanisms working in these tissues and in their hosts; knowledge of these mechanisms underlying specific phenomena may have general implications. However, in searching for specific means of inducing resistance to cells of the type found in spontaneous cases, cells of this type should be used in the tests'. These remarks are as apropos today as they were 20 years ago. They apply both to the type of cell used to induce immunity and to the type of tumor to be immunized against.

Zusammenfassung

Einige neue Ergebnisse auf dem Gebiete der Tumor-Immunität werden mit der Literatur in Einklang gebracht, und eine Hypothese zur spontanen Tumorbildung wird vorgeschlagen. Ferner wird auf die Notwendigkeit eingegangen, bei Tumor-Isoimmunitäts-Experimenten Spontantumoren zu verwenden an Stelle von lange transplantierten, von *in vitro* entstandenen oder durch Karzinogene erzeugten Tumoren.

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