*AN UNIDENTIFIED, FILTRABLE AGENT ISOLATED FROM TRANSPLANTED HUMAN TUMORS**

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Cell-free fractions of a number of transplanted human tumors induce a deformity in newborn hamsters characterized by small size, flattened foreface or microcephalic, domed head, protruding eyes and tongue, absent or abnormal teeth, and fragile bone structure.[1] The effect is now thought to be the late result of infection by a filtrable agent readily demonstrable in the tissues of the test animals as well as the tumors. The titer of the agent, presumably a virus, and the virulence of the infection increase rapidly with passage and induce severe morphologic changes and death before the deformity is manifest.

The agent, which is not dialyzable, has been found in all 8 of the transplanted human tumors tested. It has been found also in tissues of patients having cancer. It has not been obtained, with one possible exception, from preparations of rat, mouse, or normal hamster tissues, nor in a variety of transplantable animal tumors. Infant mice and rats are completely resistant. It is neutralized by the sera of normal rats and mice, and, to some degree, by sera from normal rabbits, guinea pigs, horses, calves, and chickens, but not by the sera of healthy persons and normal hamsters. Heating sera at 56° C for 1 hr does not destroy their neutralizing power.

While infectivity is rapidly destroyed by ultraviolet irradiation (the simultaneous application of both long (3660A) and short (2537A) waves at a distance of 17 cm for 15 min at 3°C) and by heat, it is remarkably stable in the cold. Preparations stored at 4°C have remained infectious for over a year as have tumors frozen at −79°C for 2 years. The infectious filtrates contain a preponderance of particles of approximately 700A diameter. Particles of similar size have been seen in the HEp 1 transplanted tumor, where they are individual but congregated in vesicles of light-electron density. They characteristically contain a nucleoid 180 to 250A in diameter and are unlike those tumor viruses known to us.

The disease has also been induced in hamsters inoculated *in utero*. In these experiments, the inoculum was injected subcutaneously, usually in the dorsal region, in 0.01 or 0.02 ml amounts. The abnormality produced was the same whether the injections were made at periods varying from 1–7 days prior to birth or within 48 hr after birth. Two of 36 mothers injected intraperitoneally, 1 to 14 days before delivery, gave birth to young that later became deformed. In each instance only a single animal of each litter was abnormal. Usually the mothers appeared quite healthy after such injections, although once when the embryos were injected 5 days before their delivery, the babies were stillborn, and the mother's liver was enlarged and hemorrhagic. Filtrates of the livers of the babies and that of the mother were both active.

Within a few days following inoculation of potent preparations, lesions may appear in the Kupffer cells of the liver. These enlarge, deteriorate, and may

contain fragments of red blood corpuscles, or, later, hemosiderin. At this stage, subcutaneous extravasations of blood are not uncommon, and at times subcutaneous edema and peritoneal transudate have been seen. Skeletal lesions appear within 10 days. A variety of morbid changes have been seen in the bones, including the formation of peculiar osteoid tissue and, at the growing ends of long bones, a zone that superficially resembles the "gerustmark" seen in scorbutus. Weakening of the bones is evident by fractures associated with imperfect repair.

The common pattern of adaptation of the virus to newborn hamsters has been characterized by initial deformities, followed in subsequent passages by deaths at progressively earlier times with rapidly increasing titers of infectivity. In an extensive series begun with a filtrate of the A-42 tumor, only deformities were encountered in the first passage. The titer rose from roughly 10^{-2} (0.02 ml of a filtrate of a 1 per cent suspension of pooled liver, spleen, and kidneys) in the second passage to 10^{-4} in the third and at least 10^{-6} in the fourth passage. In another isolation (HEp 3 tumor) the first passage was blind. No deaths or deformities occurred. However, a suspension of their pooled livers and spleens collected on the 7th day caused the death of all the test animals from the 8th to 11th days. The liver of the conditioned rat that bore the tumor was infectious and induced deformities in the first generation. There were no deaths.

Filtrates of the livers of the rats bearing all the 8 transplanted human tumors have likewise been infectious, while preparations from normal or conditioned rats without tumors have not been active. This is of particular interest, since the livers and spleens of cancer patients but not their tumors have induced abnormalities to date.

Inocula prepared from 4 human tumors that have been propagated *in vitro* for many years (HEp 1, HS 1, HEp 2, and Hela), have not produced abnormalities in newborn hamsters. These were cultivated in a medium containing horse or calf sera. The spleen of a 14-day-old infected baby hamster was grown in media containing human serum. The medium was changed weekly. The supernatant was highly active throughout the 5 weeks the culture was tested.

Isolations have been made repeatedly from the following transplantable human tumors carried *in vivo*: HEp 1, Hep 3, HEp 4, Hep 5, HS 1, HEmbRh 1, HAd 1, and A–42.[2–4] A number of transplantable mouse tumors, namely Crocker sarcoma 180, Ehrlich carcinoma (both of these in the ascitic form), the Bashford carcinoma, and Glioma 26, as well as 3 spontaneous mammary tumors of random-bred Swiss mice, failed to yield the agent. In these cases 2 blind passages were made, not only from the tumors, but also from the pooled livers and spleens and the brains of the mouse hosts. In addition, the Jensen rat sarcoma and the Walker rat carcinosarcoma 256 have been tested with negative results. Further testing of specimens from patients is under way.

It seems evident that the newborn hamster is suitable for the isolation of a virus commonly associated with transplantable human tumors. The virus causes unique lesions followed by a characteristic deformity. While the significance of the agent in oncology is unknown, it is assumed that others will wish to know of these observations.

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INVARIANT IMBEDDING, CONSERVATION RELATIONS, AND NONLINEAR EQUATIONS WITH TWO-POINT BOUNDARY VALUES

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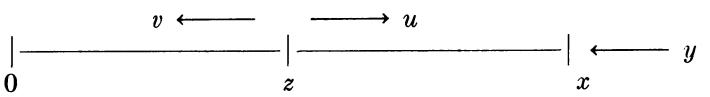
1. *Introduction.*—The mathematical description of physical processes along conventional lines yields linear and nonlinear functional equations with boundary value and initial value conditions. The boundary value aspects render the analytical discussion of existence and uniqueness of solutions rather complex and the computational solution even more difficult. In our survey paper[1] may be found an outline of the way in which many boundary value problems can be reduced to initial value problems. In a recent paper,[2] we have shown how the theory of invariant imbedding can be used to provide conservation relations which enable us to bypass spectral theory in the establishment of existence and uniqueness theorems. In this paper we wish to consider transport processes in which collision effects introduce nonlinearities and indicate how conservation relations permit us to obtain existence theorems for nonlinear differential equations with two-point boundary values.

2. *Classical Description.*—Consider a transport process in a one-dimensional rod, $0 \leq z \leq x$ in which as a result of interaction with the medium particles are absorbed and scattered. Furthermore, collision between particles traveling in opposite directions results in further absorption. Let y represent the intensity of flux per unit time incident at x, and $u = u(z)$ denote the flux per unit time passing z to the right and $v = v(z)$ the corresponding flux per unit time to the left.



The usual reasoning[1] yields the equations

$$\begin{aligned} u'(z) &= -\sigma u + \sigma F u + \sigma B v - \phi(u,v), \\ -v'(z) &= -\sigma v + \sigma F v + \sigma B u - \phi(u,v), \end{aligned} \tag{1}$$

with the boundary conditions

$$v(x) = y, \; u(0) = 0, \tag{2}$$

a two-point boundary condition.

The parameters B and F are nonnegative quantities representing the effects of