SUBVERSION OF HOST DEFENSE MECHANISMS BY MURINE TUMORS

I. A Circulating Factor that Suppresses Macrophage-Mediated Resistance to Infection*

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It has been hypothesized (1-3) that a major evolutionary function of the immune system of vertebrates is to recognize and destroy malignant neoplastic cells which are postulated to arise spontaneously with a high frequency as a result of somatic mutations. According to this hypothesis, then, those neoplasms that are seen as frank tumors must either have been naturally selected to avoid detection and destruction by immunosurveillance, or they were allowed to emerge because of some defect in the mechanism of immunosurveillance. It now seems quite clear, however, that presentation of an antigenically neutral surface is not a universal means that enables tumor cells to avoid detection by the immune system. On the contrary, most malignant tumors so far examined have been shown to possess tumor-specific antigens, and to be immunogenic to a larger or lesser degree (4-6). The fact that progressive tumor growth can occur in spite of an antitumor response on the part of the host is evidence by the numerous examples in tumor-bearing animals of concomitant immunity to a tumor cell challenge (reviewed in 7), and by the demonstrations (8) that tumor-bearing humans and animals can be shown to acquire leukocytes that are cytotoxic for tumor cells in vitro. Discovering why immunosurveillance fails to detect and destroy immunogenic tumor cells in the first place, and why an acquired mechanism of antitumor resistance fails to destroy a progressive tumor in the second place, remain major tasks for tumor immunologists (9).

In spite of the apparent wide acceptance of the idea of a mechanism of antitumor surveillance based on the discriminative powers of the lymphoid system, it is becoming increasingly evident that macrophages may also play an important role in protecting the host against colonization of its tissues by neoplastic cells. Indeed, it is no longer possible in formulating any hypothesis to explain antitumor surveillance, to ignore the flood of recent publications which show that normal (10) as well as activated macrophages (10-14) can recognize and inhibit the growth of neoplastic cells in a nonimmunological way in vitro. These publications, together with the knowledge that phagocytic cells, but not lymphocytes are possessed by all metazoan animals, allow the suggestion that phagocytic cells are the most likely candidates for serving in a universal native mechanism of defense against the emergence of neoplastic cells.

This paper is the first in a series that will examine the proposition that malignant neoplasms are those neoplasms that are naturally selected to avoid detection and destruction by mononuclear phagocytes. It tests the prediction

that any interference by neoplastic cells with the function of macrophages should be reflected in a reduced capacity on the part of the host to resist infection with bacterial parasites that are normally destroyed by macrophages. It will show that subcutaneous injection of murine tumor cells rapidly results in the liberation into the circulation of a factor(s) that strikingly impairs the capacity of the host to express macrophage-mediated resistance against experimental infection with the bacterial parasites, *Listeria monocytogenes* and *Yersinia enterocolitica*.

Materials and Methods

Mice. Breeding stock of all of the inbred strains employed was purchased from The Jackson Laboratory, Bar Harbor, Maine. Production of experimental inbred strains and F_1 hybrids was performed according to established breeding techniques. The F_1 hybrids used were (AB6)F_1 [A/J × C57BL/6], (CB6)F_1 [BALB/c × C57BL/6], (AD2)F_1 [A/J × DBA/2], and (B6D2)F_1 [C57BL/6 × DBA/2]. Mice were employed in experiments when they were between 8 and 12 wk of age.

Tumors. The SA1 spindle cell sarcoma syngeneic in the A/J strain, the Meth A fibrosarcoma syngeneic in BALB/c, the P-815 mastocytoma and CaD_2 mammary carcinoma syngeneic in DBA/2, and the EL4 lymphoma syngeneic in C57BL/6 were studied. All tumors except the CaD_2 mammary tumor can be grown in an ascites form. They were passaged weekly in the peritoneal cavity of syngeneic mice and harvested in Dulbecco's phosphate-buffered saline (PBS)[1] containing 1% fetal calf serum, and 10 U of heparin/ml. After two washes in PBS they were resuspended to an appropriate concentration in PBS for injection. The CaD_2 tumor was passaged subcutaneously by trocar. A single cell suspension of tumor cells was obtained by incubating finely diced pieces of the tumor at 37°C in modified Puck's saline containing 0.05% trypsin and 0.02% EDTA (Grand Island Biological Co., Grand Island, N. Y.). The digest was passed through six layers of sterile surgical gauze, and the resulting cell suspension was washed two times and the cells resuspended to an appropriate concentration in PBS. In those experiments that employed F_1 hybrid mice the tumors were first passaged twice in F_1 hybrids.

In most experiments tumor cells were injected subcutaneously in the right-hind foot pad in a vol of 0.05 ml of PBS. Tumor growth was followed by measuring changes in the dorso-ventral thickness of the foot pad with dial calipers.

Bacteria. A log phase culture of *L. monocytogenes* (strain EGD) seeded from an infected spleen, was grown in trypticase-soy broth, and stored in small aliquots at −70°C. Before each experiment an aliquot was quickly thawed and diluted in a standard fashion in 0.9% sodium chloride solution for intravenous injection. The organism had a mean lethal dose (LD_{50}) of 5×10^3 by the intravenous route.

Y. enterocolitica (strain WA) was obtained from Dr. P. B. Carter of the Trudeau Institute, Inc. It has an LD_{50} of 2×10^2 by the intravenous route. A log phase culture, seeded from an infected spleen, was grown in trypticase-soy broth and stored in small aliquots at −70°C. It was prepared for intravenous injection in the same way as *Listeria*. Bacterial growth in the livers was enumerated by plating 10-fold serial dilutions of whole organ homogenates on trypticase-soy agar.

Gamma Irradiation. Ascites tumor cells were harvested in heparinized PBS, washed two times, and suspended at a concentration of 10^7/ml in Fischer's medium (Grand Island Biological Co.) containing 10% fetal calf serum. The cell suspension was then placed in large plastic flasks and subjected to 3,000 rad of gamma irradiation generated by a cesium-137 irradiator with a midphantom dose rate of 35.5 rads/min. The cells were then washed and resuspended at an appropriate concentration in PBS.

Results

Rapid Systemic Suppression of Antibacterial Resistance after Subcutaneous Injection of Tumor Cells. It was discovered during a study of nonspecific

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[1] *Abbreviation used in this paper:* PBS, phosphate-buffered saline.

antitumor resistance generated during experimental bacterial infection, that a subcutaneous injection of syngeneic P-815 mastocytoma cells given 1 h after a sublethal intravenous infection with *L. monocytogenes* resulted in early death from overwhelming bacterial growth. To determine whether the capacity to rapidly suppress resistance to bacterial infection was a property shared by other murine tumors, syngeneic, semisyngeneic, and allogeneic mice were injected in the right-hind foot pad with 10^6 cells of the SA1 spindle cell sarcoma, the Meth A fibrosarcoma, the P-815 mastocytoma, the CaD_2 mammary carcinoma, or the EL4 lymphoma. 1 h later the mice were injected intravenously with a sublethal inoculum of *Listeria,* and the growth of the organism in their livers was compared with its growth in the livers of control mice.

The results in Figs. 1–3 show that subcutaneous injection of 10^6 cells of any one of the tumors tested resulted in a striking suppression of the capacity of syngeneic, semisyngeneic, and allogeneic mice to resist sublethal *Listeria* infection as evidenced by an enormous increase in bacterial growth in their livers. All of these mice eventually died from overwhelming infection, in contrast to control mice which easily checked the growth of the sublethal inoculum.

Suppression of Antibacterial Resistance by Lethally Irradiated Tumor Cells. Fig. 4 shows the results of an experiment which investigated whether a foot pad injection of 10^6 lethally irradiated SA1 cells can also suppress the capacity of $(AB6)F_1$ mice to resist an intravenous sublethal inoculum of *Listeria*. It can be seen from the liver growth curves that lethally irradiated tumor cells were just as effective as nonirradiated tumor cells in causing a severe impairment of the capacity of mice to resist sublethal infection. It was also determined that these same irradiated cells were incapable of giving rise to a subcutaneous tumor.

Evidence that Tumor Cells Interfere with the Antibacterial Function of a Normal and an Activated Macrophage System. The only cell that is known to possess the capacity to destroy *Listeria* in the mouse is the macrophage (15). It follows, therefore, that the tumor-induced suppression of anti-*Listeria* resistance was caused by interference with the antibacterial function of these phagocytic cells. It can be suggested moreover, that the increase in bacterial growth that resulted from subcutaneous injection of tumor cells occurred too early to have resulted from a block in the development and expression of T-cell-mediated anti-*Listeria* immunity (16). The increased bacterial growth must have resulted, instead, from suppression of some pre-existing mechanism of antibacterial resistance that is expressed very early in infection. That such a mechanism of native antibacterial resistance is possessed by mice was revealed by the results of a previous study (17), which showed that macrophages in the livers of normal mice are capable of rapidly destroying over 50% of the *Listeria* load ingested by this organ during the first 8 h of infection. The same study also showed that a much greater rate of destruction of *Listeria* occurs during the same early time period in the livers of mice whose macrophage systems have been activated by BCG infection. This knowledge made it possible, therefore, to test whether subcutaneous injection of tumor cells suppress the early expression of antibacterial activity of a normal and an activated liver macrophage system. Thus, the suppressive effect of a subcutaneous injection of 10^6 SA1 cells on the fate over the
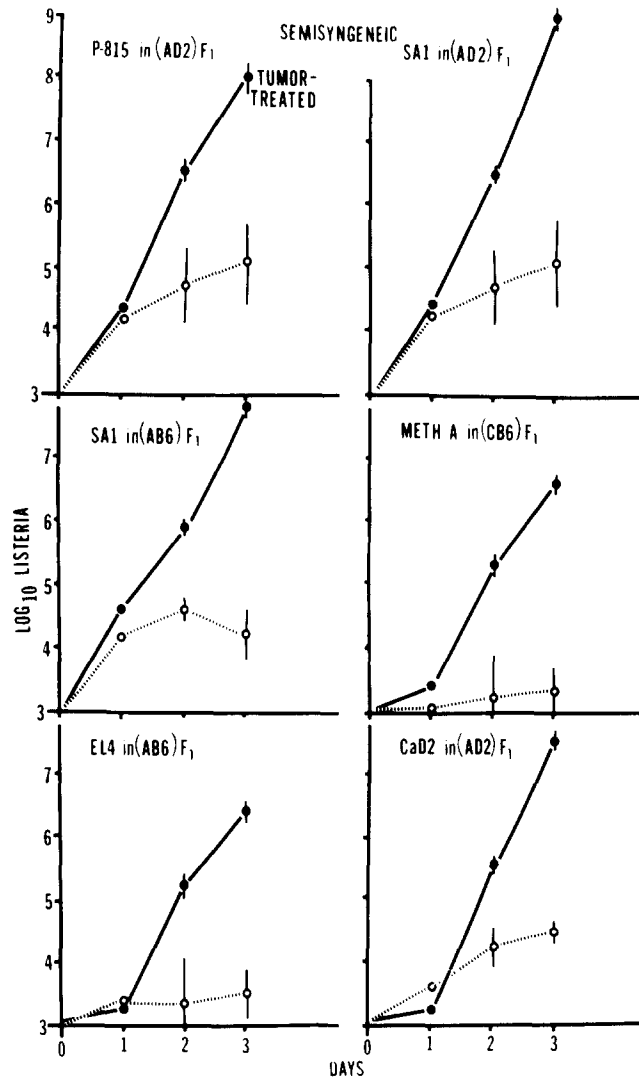
FIG. 1. Suppressive effect of a subcutaneous injection of 10^6 tumor cells on the capacity of semisyngeneic mice to resist sublethal intravenous *Listeria* infection as determined by growth of the parasite in their livers. Subcutaneous injection of cells of any one of five tumors given 1 h before intravenous inoculation with *Listeria* resulted in an enormous increase in bacterial growth in the liver (solid lines), in contrast to control mice which easily checked bacterial growth (broken lines). Means ± 2 SE of five mice per time point.

first 24 h of an intravenous *Listeria* challenge infection was measured in the livers of normal (AB6)F₁ mice, and in the livers of (AB6)F₁ mice whose macrophage systems had been activated as a result of a 12 day BCG infection (17), a 6 day *Listeria* infection (18), or a subcutaneous injection of *Corynebacterium parvum* given 7 days previously (19). In these experiments the SA1 cells were injected 24 h before the *Listeria* challenge.

The effect of subcutaneous SA1 cells on the early destruction of a sublethal
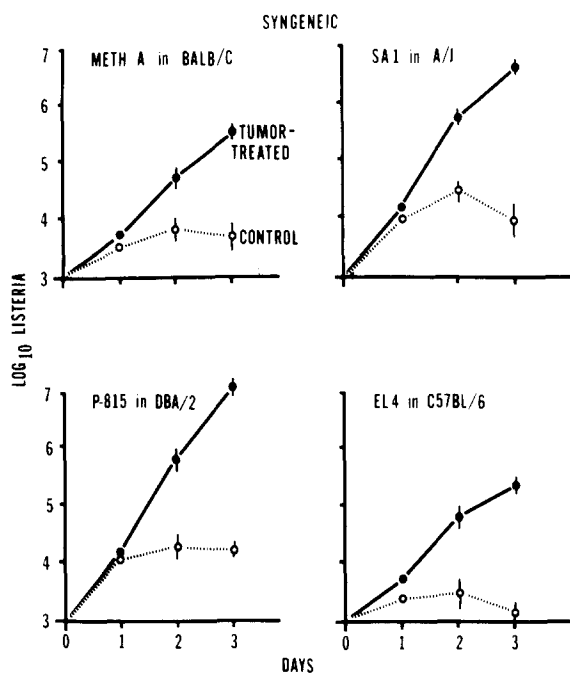
FIG. 2.   Same experiment as described for FIG. 1 except that syngeneic mice were employed.
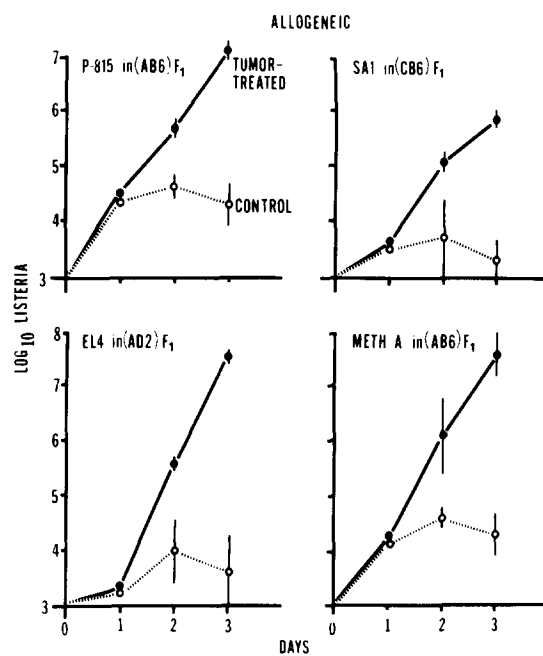

FIG. 3.   Same experiment as in Figs. 1 and 2 except that allogeneic mice were employed.
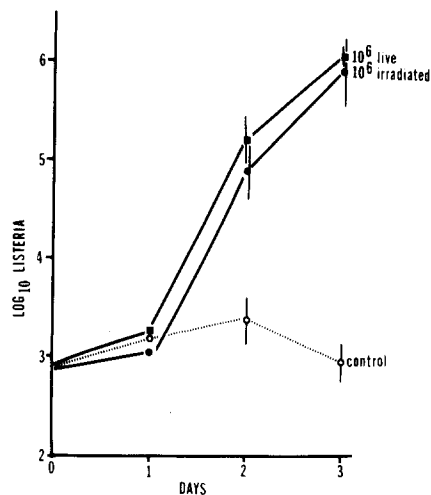
FIG. 4. Evidence that subcutaneous injection of 10^6 lethally irradiated SA1 cells was just as effective as the same number of nonirradiated SA1 cells in suppressing resistance to *Listeria* infection as measured by bacterial growth in the liver. Means ± 2 SE of five mice per time point.

Listeria inoculum in the livers of normal mice is shown in Fig. 5, where it can be seen that the subcutaneous presence of tumor cells greatly impaired the capacity of normal liver macrophages to reduce the *Listeria* load in the liver during the first 8 h of infection. Whereas control mice reduced the bacterial load by 0.75 logs in 8 h, tumor-treated mice achieved only 0.2 log bacterial destruction in the same time period. This resulted in a much larger base for subsequent bacterial multiplication, and consequently in larger numbers of bacteria in the livers at later times.

The suppressive effect of subcutaneous injection of tumor cells on the expression of antibacterial capacity of activated liver macrophages is shown in Fig. 6 where it can be seen that the greatly increased nonspecific antibacterial capacity expressed over the first 24 h by BCG-, *Listeria*-, and *C. parvum*-activated macrophage systems were severely impaired. For instance, the capacity of a BCG-activated liver macrophage system to kill *Listeria* over the first 12 h was reduced by 1.5 logs. It will be noted, however, that the bactericidal capacity of activated macrophages was not suppressed below that of normal controls. An above normal antibacterial capacity survived, therefore, in spite of the suppressive influence of tumor cells.

Additional Events in the Host's Antibacterial Response are Suppressed by Tumor Cells. The foregoing results indicate that one way that tumor cells can suppress the capacity of mice to resist *Listeria* infection is by suppressing the capacity of resident macrophages of the liver to rapidly kill a significant proportion of the bacterial inoculum. To determine whether this was the only reason for the increased bacterial growth that follows tumor cell injection, an experiment was designed to discover whether increased bacterial growth occurs when 10^6 SA1 cells are injected after the initial phase of destruction of *Listeria* has been achieved.
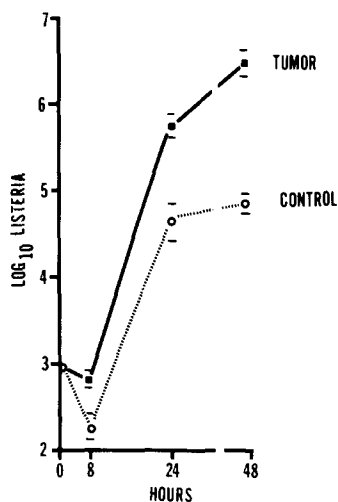
FIG. 5.   Evidence that suppressed anti-*Listeria* resistance that results from subcutaneous injection of tumor cells can be caused partly by a suppressed capacity of resident macrophages of the liver to reduce the bacterial load in this organ during the first 8 h or so of infection. Means ± 2 SE of five mice per time point.
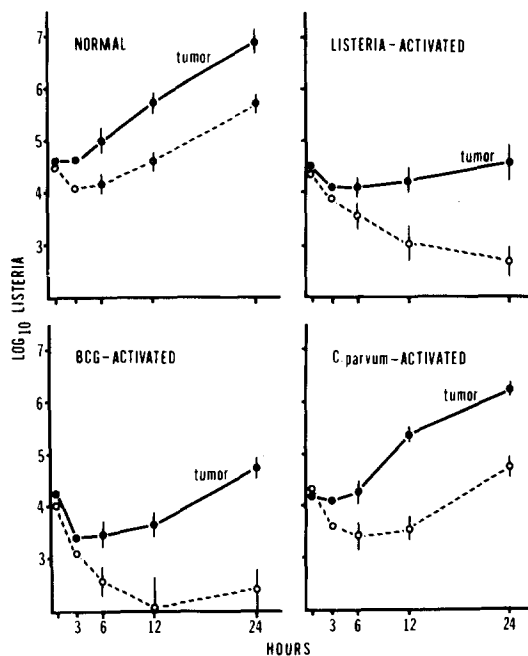

FIG. 6.   Additional evidence to support the interpretation that subcutaneous injection of tumor cells suppresses the capacity of phagocytes in the liver to accomplish appreciable bacterial destruction during the early stages of infection. The greatly increased anti-*Listeria* resistance expressed nonspecifically by liver macrophages activated as a result of either BCG, or *Listeria* infection, or by treatment with *C. parvum* was greatly suppressed by subcutaneous injection of 10^6 SA1 cells. Means ± SE of five mice per time point.
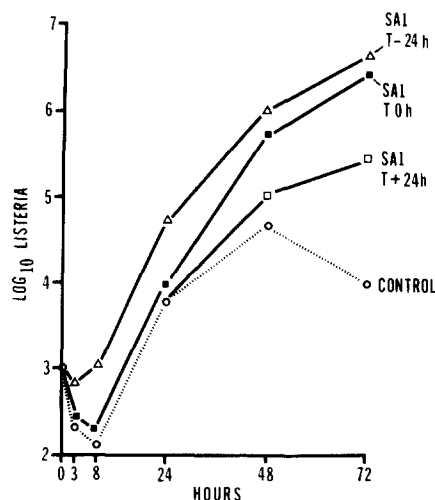
Fig. 7.   Effect on bacterial growth in the liver of subcutaneous injection of SA1 cells given at the time of, 24 h before, or 24 h after initiating *Listeria* infection. Only SA1 cells injected 24 h before caused suppression of early antibacterial events in the liver. Suppression of antibacterial resistance that occurred when tumor cells were given at later times, therefore, was caused by interference with additional antibacterial mechanisms. Means of five mice per time point.

The results in Fig. 7 show that suppression of the early phase of destruction of *Listeria* in the liver only resulted when tumor cells were injected 24 h before infection was initiated. It can be seen that although an injection of tumor cells given either at the time of initiating infection, or 24 h later had no effect on bacterial destruction during the first 8 h, it did cause a large increase in bacterial multiplication after about a 24 h delay. It is apparent, therefore, that tumor cells can suppress antibacterial mechanisms in addition to those that are expressed by resident macrophages during the first 8 h or so of infection. It is also apparent that it takes about 24 h for the subcutaneous deposit of tumor cells to cause systemic suppression of antibacterial resistance. It should be realized that it is not until after 24 h of infection that infective foci in the liver become heavily populated with monocyte-derived macrophages (20).

Suppression of Resistance to Infection with Y. Enterocolitica.   To determine whether a subcutaneous injection of tumor cells can suppress resistance to infection with bacterial parasites other than *Listeria*, an experiment was performed to determine whether resistance to an intravenous *Yersinia* infection was also suppressed by the injection of tumor cells. *Yersinia* was chosen for this study because of the published knowledge (21) that a large proportion of an intravenous inoculum of this organism is destroyed by normal macrophages of the liver during the first 12 h or so of infection.

It can be seen in Fig. 8 that subcutaneous injection of 10^6 SA1 cells into normal mice and mice whose macrophage systems had been activated by a 12 day BCG infection resulted in a striking reduction in their capacity to destroy *Yersinia* in their livers during the first 8–12 h of infection. Suppression of the greatly enhanced anti-*Yersinia* activity of the BCG-treated mice was particularly se-
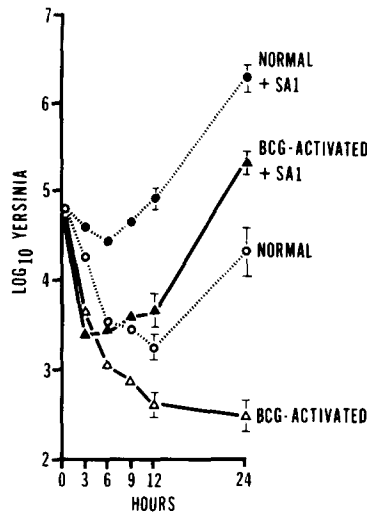
FIG. 8. Evidence that subcutaneous injection of tumor cells suppresses resistance to intravenous infection with *Y. enterocolitica*. Subcutaneous injection of 10^6 SA1 cells 24 h before intravenous *Yersinia* challenge greatly decreased the capacity of both a normal and a BCG-activated liver macrophage system to destroy the challenge organisms during the first 12 h or so of infection. Means ± SE of five mice per time point.

vere. As with *Listeria*, this resulted in a larger base for subsequent bacterial multiplication and consequently in a higher level of infection at later times. It should be realized, however, that the 10^5 *Yersinia* inoculum employed was lethal for control mice as well as tumor-treated mice.

Suppression of Antibacterial Immunity is Mediated by a Factor in Circulation. The foregoing results showed that a subcutaneous injection of tumor cells in the hind foot pad of mice suppressed their capacity to express antibacterial resistance at a remote site. This implied that the effect was caused by a factor in circulation. This possibility was investigated by determining whether an infusion of serum from tumor-implanted donor mice was capable of suppressing the capacity of normal recipient mice to resist *Listeria* infection. Syngeneic donor mice were bled for serum 24 h after being injected subcutaneously with 10^6 SA1 cells.

Fig. 9 shows that an intraperitoneal injection of 0.5 ml of serum from tumor-treated donors caused a striking suppression of the capacity of normal syngeneic recipients to resist sublethal *Listeria* infection as evidenced by increased bacterial growth in their livers. Serum from Meth A- and mastocytoma-bearing mice was equally active.

The Serum Suppressor Factor is Rapidly Generated and Possesses Potent Physiological Activity. The speed at which the suppressor factor appeared in the circulation of (AB6)F₁ mice after injecting them subcutaneously with tumor cells was determined by measuring the capacity of serum collected from them at progressive times to suppress antibacterial resistance in normal recipients. Thus, (AB6)F₁ mice were injected with either 10^5 or 10^6 SA1 cells and a sample of the populations bled for serum at progressive times over 24 h. In all cases, 0.5 ml of tumor-bearer serum was infused intraperitoneally into test recipients 2 h
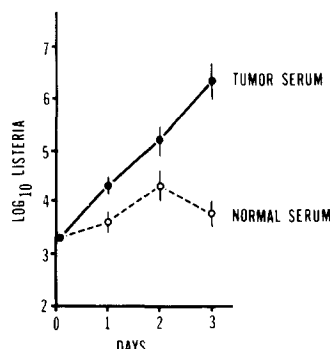
FIG. 9. Evidence that tumor-induced suppression of antibacterial resistance is mediated by a factor in circulation. An intraperitoneal infusion of 0.5 ml of serum from 24-h SA1-bearing donors caused a striking suppression of the capacity of normal recipients to control the growth of a *Listeria* infection in their livers. Means ± SE of five mice per time point.

before injecting them with 2×10^3 *Listeria*. The increase in bacterial growth that resulted in each case was determined by subtracting the 24-h growth of *Listeria* in the livers of recipients of normal serum from its 24-h growth in the livers of recipients of tumor-bearer serum.

Fig. 10 shows that physiologically active concentrations of suppressor factor were present in the circulation within 8 h of injecting tumor cells subcutaneously, and that the factor progressively increased in concentration until the experiment was terminated at 24 h. It will be noted that there was very little difference between the speed at which the suppressor factor appeared in circulation of mice injected with 10^5 and 10^6 tumor cells.

A similar assay procedure was used to determine the rate of decay in normal recipients of the suppressive effect of an intraperitoneal infusion of 0.5 ml of tumor-bearer serum. Normal recipients were infused intraperitoneally with 0.5 ml of tumor-bearer serum and changes against time in their capacity to resist a standard *Listeria* challenge were followed.

It can be seen in Fig. 11 that the suppressive effect of an infusion of tumor-bearer serum progressively decayed over a 48 h period, and showed an apparent half-life of 24 h. It should be realized that this experiment does not distinguish between the decay of the factor as such, and the decay of its effect.

The potency of serum collected from 24-h tumor-bearing donors was investigated by measuring the capacity of limiting dilutions of it to suppress antibacterial resistance in normal recipients. Thus, serum obtained from donor mice 24 h after injecting them subcutaneously with 10^6 SA1 cells was subjected to twofold serial dilution in PBS. Each dilution was injected intraperitoneally into normal recipients and their ability to resist a *Listeria* challenge was then determined. It can be seen in Fig. 12 that as little as 0.015 ml of tumor-bearer serum caused a significant reduction in the capacity of normal recipients to resist infection with a standard sublethal *Listeria* inoculum.

The Tumor Suppressor Factor is Dialyzable.   To obtain an initial idea of the size of the suppressor molecule, its ability to pass through a conventional dialysis membrane was investigated. This involved dialyzing one volume of 24-h tumor-bearer serum against one volume of PBS for 48 h, and comparing the
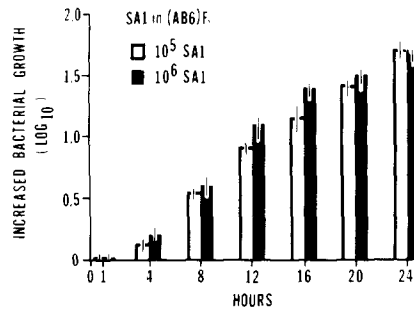
FIG. 10. Speed of appearance of suppressor factor in circulation after injecting either 10^5 or 10^6 SA1 cells subcutaneously. Shown is the increase in 24-h *Listeria* growth in normal recipients that resulted from infusion of 0.5 ml of serum collected from tumor-treated donors at the times indicated after injecting tumor cells. Appreciable suppressor activity was present in donor serum within 8 h of injection tumor cells. Means of five mice ± SD.
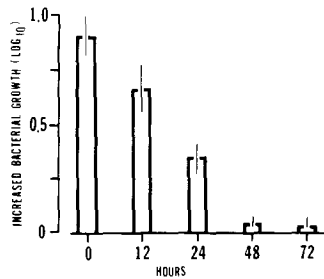

FIG. 11. Speed of decay in normal recipients of suppressor action of an intraperitoneal infusion of 0.5 ml of serum from 24-h tumor-treated donors. Shown are the increases in 24-h *Listeria* growth in the recipients' livers that occurred when a standard *Listeria* inoculum was injected at the times indicated after infusing them with tumor-bearer serum. Means ± SD of five mice per time point.

ability of an intraperitoneal injection of the dialysate with the ability of an injection of the parent serum to suppress resistance to *Listeria* infection.

It can be seen from the liver growth curves in Fig. 13 that the serum dialysate was just as effective as the parent serum in suppressing resistance to a sublethal infection. Therefore, at least part of the suppressive activity of serum is associated with a molecule with a mol wt of 12,000 or less.

Discussion

This study shows that subcutaneous injection of cells of any one of five unselected murine tumors resulted very rapidly in the liberation into the circulation of a factor(s) that severely suppressed the capacity of the host to resist intravenous infection with the bacterial parasites, *L. monocytogenes* and *Y. enterocolitica*. It is reasonable to suggest, therefore, that the capacity to impair resistance to infection with a spectrum of microorganisms will be found to be a property shared by many other tumors. The possibility that a similar mechanism is responsible for the reported (22) high incidence of death from natural infection of terminal cancer patients must therefore be considered. It should be realized, however, that death of experimental animals from acute
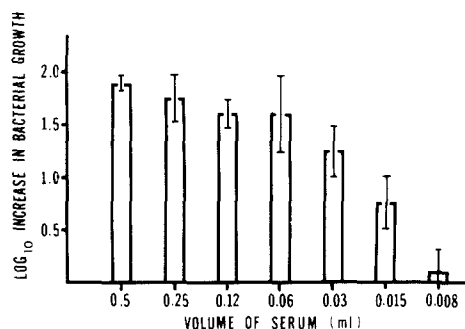
FIG. 12. Evidence that serum suppressor factor possesses potent physiological activity. Shown are increases in 24-h *Listeria* growth in the livers of recipients that received limiting dilutions of serum from 24-h tumor-bearing donors. In this experiment as little as 0.015 ml of tumor-bearers' serum caused significant suppression of antibacterial resistance. Means ± SE of five mice per time point.
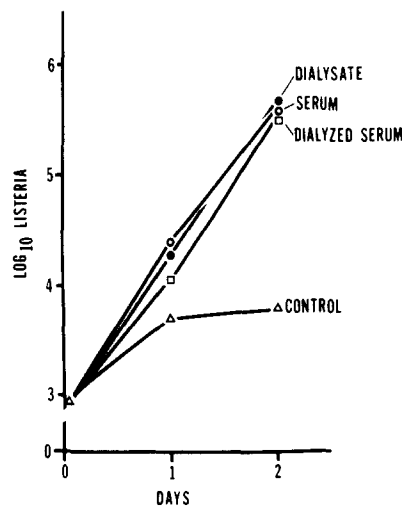

FIG. 13. Evidence that tumor-suppressor factor is small enough to pass through a dialysis membrane. 0.5 ml of serum dialysate was just as effective as 0.5 ml of the parent serum or dialyzed serum in suppressing anti-*Listeria* resistance.

natural infections is not a commonly reported consequence of tumor cell injection. The reason for this will be revealed in the paper which follows.

The major aim of the experiments reported in this paper was to examine the proposition that malignant neoplasms are those neoplasms that are naturally selected to avoid destruction by the antitumor activity of the host's macrophage system. This proposition was suggested on the basis of a large body of published experimental evidence (10–14) which shows that these mononuclear phagocytic cells can display potent antitumor activity in vitro. The experiments tested the prediction that any interference with the normal function of macrophages by neoplastic cells should be reflected in a reduced capacity on the part of the host to resist experimental infection with those bacteria that are destroyed by macrophages. Since there is convincing evidence (15) that the only cells in the

mouse with the capacity to destroy *Listeria* are macrophages, it follows that the systemic suppression of the capacity of mice to resist *Listeria* infection by tumor cells must have been caused by the liberation into the circulation of a factor that interfered with the function of these phagocytic cells. Indeed, it was shown that subcutaneous injection of tumor cells not only severely suppressed the expression of the anti-*Listeria* activity of macrophages in the livers of normal mice, but also suppressed the enhanced, nonspecific, anti-*Listeria* and anti-*Yersinia* activity of liver macrophage systems that were highly activated as a result of BCG infection, *Listeria* infection, or by treatment with *C. parvum*.

The way in which the tumor-suppressor factor interferes with the function of macrophages is not yet known. It either directly suppresses the phagocytic and bactericidal activity of these cells, or it indirectly interferes with their ability to express these functions. Both possibilities are presently under investigation. The finding that the factor severely suppressed the substantial destruction of *Listeria* that occurs in the liver during the first few hours of infection indicates that it can directly interfere with the antibacterial function of resident macrophages of the liver. The additional finding, however, that the factor can cause increased growth of *Listeria* when injected at later stages of infection could mean that it also interferes with the function of monocyte-derived effector macrophages which are known (20) to populate infective foci in the liver in large numbers after 24 h of infection. The possibility that the factor interferes with the focusing of blood monocytes at infective foci can be suggested on the basis of results of another study (to be published) which show that subcutaneous injection of the same tumor cells severely reduces the rate at which blood leukocytes emigrate into sterile inflammatory peritoneal exudates. That murine tumors produce an anti-inflammatory factor has been reported by others (23).

The origin of the suppressor factor is not known. It is either a direct product of tumor cells, or tumor cells trigger its synthesis by the host. The findings that subcutaneous injection with as few as 10^5 or 10^6 tumor cells can result in suppressive concentrations of the factor in blood within 8 h, and that an infusion of as little as 0.015 ml of serum from 24-h tumor-bearing donors severely suppressed the antibacterial capacity of normal recipients, indicate that the factor is very rapidly synthesized, and that it possesses potent physiological activity. It should be realized in this connection that an infusion of 0.015 ml of tumor-bearer serum into a normal recipient mouse represents a dilution of about 1 in 300 of the factor in the recipient's blood.

The chemical identity of the factor is currently being investigated. All that can be stated at this time is that the suppressive activity is associated with a molecule that is small enough to pass through a cellophane dialysis membrane. The possibility exists that the factor is a prostaglandin, particularly since these compounds can be present in above normal concentrations in tumor-bearing individuals (24). It is unlikely, on the other hand, that the factor is a corticosteroid. This can be tentatively concluded from the knowledge (25) that relatively large quantities of corticosteroids are required to suppress anti-*Listeria* resistance; quantities too large to be accommodated in as little as 0.015 ml of serum. Again, whereas the amount of corticosteriod required to suppress anti-*Listeria* resistance results in pronounced atrophy of lymphoid tissue (26) no such atrophy

results from infusions of factor-containing serum (unpublished observations). It should be pointed out that injections of normal liver and kidney cells had no effect on the capacity of mice to resist *Listeria* infection.

Summary

The subcutaneous injection of cells of any one of five unselected murine tumors resulted very rapidly in the liberation into the circulation of a small molecular weight factor that severely impaired the capacity of the host to resist experimental infection with *Listeria monocytogenes* and *Yersinia enterocolitica*. It was found that the factor appeared in blood within 8 h of injecting tumor cells subcutaneously. That it possessed potent physiological activity was evidenced by the demonstration that an infusion of as little as 0.015 ml of tumor-bearer serum strikingly suppressed the capacity of normal recipients to resist bacterial infection. It was reasoned on the basis of the knowledge that the only cells in mice with the capacity to destroy *Listeria* are macrophages, that suppression of antibacterial resistance was caused by the ability of the tumor-suppressor factor to interfere, either directly or indirectly, with the antibacterial functions of these mononuclear phagocytic cells. The results are consistent with the hypothesis that at least some malignant neoplastic cells are naturally selected to avoid destruction by native and acquired antitumor mechanisms of mononuclear phagocytes.

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