ABSTRACT

In the present paper we studied the immunological hyporesponsiveness and the suppressor activity in mice bearing a transplantable adenocarcinoma, ADK-11, that spontaneously arose in BALB/c mice. Phytohemagglutinin (PHA)- and lipopolysaccharide from Escherichia coli O1l1:BH-induced T- and B-cell proliferation and PHA-stimulated migration inhibitory factor (MIF) production were monitored. Mixing experiments were performed to detect the suppressor activity. The progressive growth of ADK-11 was accompanied by an increasing hyporesponsiveness of spleen cells to PHA-induced proliferation. The hyporesponsiveness was associated with concomitant suppressor activity by the spleen cells. Both hyporesponsiveness and suppression were abrogated by the following treatments of the spleen cells: passage over a nylon column; and pretreatment with carrageenan or carbonyl iron and magnet. The suppressor cells, moreover, were resistant to anti-Thy 1.2 plus complement treatment, and were adherent to plastic and nylon. Washing of the cultures before the addition of the tritiated thymidine or enrichment of the culture medium with glucose, arginine, or lysine did not affect the suppressor activity. We therefore concluded that macrophages or macrophage-like cells exerted an inhibitory effect on the PHA-induced proliferation. B-lymphocyte proliferation and MIF production were also inhibited by macrophages from the spleens of tumor-bearing mice. Macrophages from normal spleens demonstrated a significant but always smaller suppressive effect. The demonstration that macrophages suppress MIF production as well as proliferative responses provides further insight into the mechanism of the progressive depression of cell-mediated immunity that has been associated with tumor growth.

INTRODUCTION

The host immune reactivity against progressing tumors is a dynamic event in which multiple effector mechanisms are activated (8). A weak, but significant immune response against tumor antigens is often detected in early stages of tumor growth, followed by progressive decline and then total eclipse of reactivity, both in clinical neoplasia, as well as in experimental animal models (12, 20, 31). In the advanced stages of the host-tumor relationship, some of the loss of immune reactivity has been attributed to humoral blocking activities have been extensively studied, the suppressor cellular activities have been analyzed only in hosts bearing highly immunogenic tumors induced by viruses or chemicals or in hosts with primary tumors of undetermined immunogenicity (6, 11, 12, 16, 30).

Increasing evidence points to the importance of lymphokines (10) as antitumor effector mechanisms and as activators of cellular immunity (21, 26, 28). However, suppressor activity has been studied mostly with regard to lymphocyte proliferation, antibody production, and the generation of cytotoxic T-cells (24), while the cellular bases for the regulation of lymphokine production have been poorly characterized. Therefore, we thought that it was of interest to study the appearance of suppressor cells and to define their effect on mitogen-dependent lymphocyte proliferation and macrophage inhibitory factor (MIF3) production in mice bearing a spontaneous tumor that has been extensively characterized in our laboratory. The murine adenocarcinoma (ADK-11) used is a spontaneous transplantable tumor (1) that is poorly immunogenic in syngeneic hosts (4), but can elicit specific cellular and humoral reactivities (5) at early stages of the tumor growth. A detailed review of the dynamics of ADK-11 tumor-host relationship and of the characteristics of the tumor-associated antigens has been published (8).

In the present paper, we show that a macrophage-dependent suppression takes place during the progression of ADK-11 tumor, affecting the proliferation of B- and T-lymphocytes, and also inhibiting MIF production. Thus, even the earlier events of the in vitro lymphocyte activation, such as lymphokine production, may be suppressed by activated macrophages from tumor-bearing mice.

MATERIALS AND METHODS

Mice. BALB/c mice were bred in syngeneic conditions in our animal facilities from breeders originally obtained from the BALB/c colony of the Animal Production Branch of the NIH, Bethesda, Md. In the experiments reported here, 12-week-old animals were used.

Tumors. The mammary adenocarcinoma (ADK-11) used originated spontaneously in a BALB/c female of our colony (5, 7). The earlier transplant generations of the tumor were preserved by slow freezing and storage at −80°C. The tumor preparations used in these experiments had been transplanted in syngeneic recipients no more than 6 times. The animals were given s.c.

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1 The abbreviations used are: MIF, migration inhibitory factor; ADK-11, spontaneous adenocarcinoma; RPMI, Roswell Park Memorial Institute; FBS, fetal bovine serum; TBS, tumor-bearing mouse spleen cells; NS, normal spleen cells; PHA, phytohemagglutinin; LPS, lipopolysaccharide from Escherichia coli 01l1: BH; [3H]dThd, [3H]thymidine; d-NS, nylon wool-eluted normal spleen cells; a-TBS, plastic-adherent tumor-bearing mouse spleen cells; a-NS, plastic-adherent normal spleen cells.

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injections in the inguinal region of 0.1 ml of a single-cell suspension containing $5 \times 10^6$ ADK-11 cells. This dose induces 100% of tumor takes in recipient mice. After inoculation, mice were inspected twice weekly and neoplastic masses were measured with calipers.

**Cell Preparation.** Mice were sacrificed by decapitation. Sterile single-cell suspensions of spleen cells were prepared, and RBC were osmotically lysed. Unless otherwise specified, the cells were cultured in RPMI Medium 1640 supplemented with 5% FBS, 2 mM glutamine, 100 units penicillin per ml, 100 µg streptomycin per ml, 25 mM [4-(2-hydroxyethyl)-1-piperazine-ethanesulfonic acid] buffer, and 4 x $10^{-3}$ M 2-mercaptoethanol. All reagents were obtained from Grand Island Biological Co. (Grand Island, N.Y.). In some experiments, culture medium was enriched with glucose, arginine, and lysine (Sigma Chemical Co., St. Louis, Mo.), as specified in “Results.”

**Cell Separation.** Nylon wool purification was done as described by Julius et al. (15). Among different experiments, we obtained a yield of 20 to 30% with TBS, and 30 to 40% with NS. Removal of phagocytic cells by carbonyl iron and magnet treatment was performed as previously described (22). Splenic macrophages were obtained by plating $10^6$ spleen cells in 20 ml of RPMI Medium 1640 containing 10% FBS on 100- x 25-mm Falcon plastic culture dishes (Becton-Dickinson, Mountain View, Calif.) for 6 hr at 37°C. The nonadherent cells were vigorously washed away, and adherent populations were removed by scraping with a rubber policeman. When anti-Thy 1.2 serum treatment was required, $10^7$ spleen cells were preincubated with 0.1 ml of the antisera (generous gift of Dr. H. T. Holden, National Cancer Institute, Bethesda, Md.) (14) for 45 min at 4°C in 1 ml of medium, followed by incubation for 45 min at 37°C with a relatively nontoxic rabbit complement (less than 15% killing by complement treatment only). Anti-Thy 1.2 plus complement treatment resulted in a 40 to 60% cytotoxicity, completely abolished the PHA response of both TBS and NS and MIF production by PHA-stimulated NS. When carrageenan treatment was required, $10^7$ spleen cells were preincubated with 1 ml of medium containing $250 \mu g$ of carrageenan per ml (Sigma, type 1), for 4 hr at 37°C, and then washed 3 times.

**Mitogen Responses.** In lymphoproliferation, $4 \times 10^6$ of the various spleen cell preparations were distributed among the flat-bottomed wells of a microtiter plate (Cooke Engineering Co., Alexandria, Va.). Triplicate microcultures were incubated with or without PHA (Grand Island Biological Co.) 1:100 final dilution, or LPS (Difco Laboratories, Detroit, Mich.), $10 \mu g/ml$ final concentration, for 64 hr at 37°C in a humidified CO₂ atmosphere, before pulsing with $1 \mu Ci$ of $[^3H]$Tdr (specific activity, 6 Ci/mmmol; New England Nuclear, Boston, Mass.) for 8 hr. Cultures were harvested onto paper filters with an automatic suction device and counted by liquid scintillation spectrometry. Mitogen-induced DNA synthesis was expressed as the difference in incorporation of $[^3H]$t-Thd between control (unstimulated) and test lymphocytes (stimulated), as measured by cpm. One representative experiment of at least 4 is shown in the tables or figures. In these experiments, the S.E. of the mean of triplicate cultures did not exceed 10% and was not reported.

**MIF Production.** MIF was produced by *in vitro* stimulation of NS from BALB/c mice. NS ($5 \times 10^6$) in 1 ml of RPMI Medium 1640 with 10% FBS were incubated with or without optimal concentration of PHA at 37°C for 4 hr in 5-ml Falcon No. 2063 tubes. Preliminary experiments had shown MIF production using PHA dilutions ranging from 1:250 to 1:25 with a peak at 1:100 dilution, a concentration that was routinely used. Cultures were spun down, and the supernatant was carefully aspirated. The pellet ($\leq 50 \mu l$) was resuspended in 4 ml of medium and centrifuged, and the supernatant was aspirated. The cells were then resuspended in 1 ml of RPMI Medium 1640 with 1% FBS and incubated at 37°C for an additional 24 hr. Together with the mitogen, different cell populations were added to both the stimulated and control cultures to test their effect on MIF synthesis.

**Agarose Droplet Assay.** Supernatants were harvested after 24 hr of culture, reconstituted to 10% FBS, and stored at -80°C until use. Control and stimulated supernatants were always tested simultaneously, using the same migrating populations. The migration inhibition assay was performed as previously described (22).

Percentage of migration inhibition was calculated by the formula:

$$\left( \frac{1 - \text{Migration area with supernatant from NS + PHA}}{\text{Migration area with supernatant from NS alone}} \right) \times 100$$

The effect of the addition of different populations of cells was calculated by the formula:

$$\left( \frac{1 - \text{Migration area with supernatant from NS + PHA + other cells}}{\text{Migration area with supernatant from NS + other cells}} \right) \times 100$$

The mean percentage of inhibition ± 2 S.E. from several controls of NS in the absence of mitogen, with or without different cells added, was $-2 \pm 15$. Therefore, any value greater than 13% of inhibition was considered positive. The S.E. of the mean percentage of inhibition among the replicates was 5% or less. All the values shown in “Results” were the means of at least 3 experiments.

**RESULTS**

**Kinetics of Lymphoproliferative Response to PHA during ADK-11 Growth.** BALB/c mice were given injections of $5 \times 10^6$ ADK-11 cells, a dose which kills 100% of the mice in 30 days. Kinetic studies on the PHA response during tumor growth showed a progressive hyporesponsiveness of the spleen cell proliferation parallel to the increasing tumor size and to the enlargement of the spleen (Chart 1). The experiments were performed using an optimal 1:100 final dilution of PHA, but the same pattern of hyporesponsiveness was observed when concentrations ranging from 1:35 to 1:200 were used (data not shown). The $[^3H]$Tdr incorporation of unstimulated cultures of spleen cells from TBS with tumor sizes of 3 to 8, 13 to 15, and $>25$ mm was, respectively, 2813, 4503, and 7115 cpm. Even though the background incorporation increased in parallel with the tumor size, the hyporesponsiveness of TBS was apparent and significant when these values were not subtracted from the incorporation of stimulated cultures.

By contrast, the passage of the spleen cells through a nylon wool column completely restored the PHA responsiveness of TBS as compared with BALB/c NS treated in the same way (Chart 1). Thus, hyporesponsiveness was not due to the lack of PHA-responsive lymphocytes in TBS. Moreover, the mac-
rophenage content of the TBS increased, reaching 15% with tumors larger than 25 mm, compared to the 6 to 8% of NS as measured both by counting of latex-ingesting cells and by differential counting of Giemsa-stained smears. Histological examination of the spleens of tumor-bearing mice always failed to demonstrate any metastases.

**Kinetics of the Suppressor Activity of TBS on the PHA Proliferative Response.** Studies on the PHA response of a mixture of 2.5 × 10^5 TBS at various stages of tumor growth with 2.5 × 10^5 d-NS were performed in order to detect any suppressor activity. Preliminary experiments had shown that the proliferative response to PHA was proportional to the cell number when they ranged from 2 to 8 × 10^5/well. Thus, the maximum of 5 × 10^5 cells/well, used in these experiments, was in the range where cell overcrowding does not affect the proliferation. For this reason, we expressed the response of the mixture of d-NS and TBS as the percentage of the expected response, calculated by adding the cpm of the [3H]dThd incorporated by d-NS and TBS when cultured separately.

The results of a representative experiment (Chart 2) showed a decrease in the PHA response of d-NS when mixed with TBS, suggesting the presence of suppressor cells in the TBS population. The suppressor activity, moreover, increased in parallel with the tumor diameter. In 4 of 6 experiments, suppression occurred when the tumor was 3 to 8 mm in diameter and was always found to be associated with larger tumors. In some experiments, TBS from mice with tumor diameter of >25 mm completely abrogated the PHA response of d-NS. The suppressor activity was completely eliminated by removing the nylon-adherent cells from TBS (Chart 2).

**Characterization of the Suppressor Cells.** Spleen cells from mice bearing large tumors (>25 mm) were used in order to define the cell population responsible for the suppressor activity. The effects of different pretreatments of the TBS on their suppressor activity are listed in Table 1. These results showed that the suppression of the PHA response by TBS was abrogated by: (a) removal of nylon-adherent cells, (b) depletion of carbonyl iron-ingesting cells, and (c) pretreatment with 250 μg of carrageenan per ml. In contrast, the suppressor activity was retained in: (a) nylon-adherent TBS, (b) anti-Thy 1.2 plus complement-treated TBS, and (c) plastic-adherent TBS. We therefore conclude that the suppressor activity of TBS is dependent on splenic macrophages or macrophage-like cells.

**Suppressor Activity of Adherent Spleen Cells.** In order to characterize further the suppressor activity, a-TBS from mice with a tumor diameter of >25 mm were used as the source of suppressor cells. Their effect on the PHA response of d-NS was compared with that of a-NS. Populations of adherent cells, from both TBS and NS, recovered after 12 hr of incubation at 37° on plastic dishes, contained 95 to 99% of latex-ingesting cells and not more than 5% of polymorphonuclear cells as determined by differential counting of Giemsa-stained smears. The dose-response curves of the effect of increasing amount of a-TBS and a-NS on the PHA response of d-NS are shown in Chart 3. At all the concentrations tested, a-TBS showed more

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**Table 1**

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Cells added</th>
<th>Treatment</th>
<th>PHA stimulated</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>None</td>
<td>None</td>
<td>51</td>
<td>0.7</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>None</td>
<td>16</td>
<td>2.5</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Nylon wool</td>
<td>8</td>
<td>4.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-adherent</td>
<td>46</td>
<td>0.3</td>
</tr>
<tr>
<td>2</td>
<td>None</td>
<td>None</td>
<td>75</td>
<td>1.3</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>None</td>
<td>20</td>
<td>3.8</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Iron and magnet</td>
<td>67</td>
<td>4.8</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Carrageenan</td>
<td>74</td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Cells adherent to plastic</td>
<td>13</td>
<td>0.1</td>
</tr>
<tr>
<td>3</td>
<td>None</td>
<td>None</td>
<td>68</td>
<td>2.0</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>None</td>
<td>15</td>
<td>6.3</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Complement</td>
<td>18</td>
<td>6.5</td>
</tr>
<tr>
<td></td>
<td>TBS</td>
<td>Anti-Thy 1.2 + complement</td>
<td>13</td>
<td>5.1</td>
</tr>
</tbody>
</table>

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* TBS (2.5 × 10^5), treated as specified, were mixed with the same number of d-NS.
* The [3H]dThd incorporation in PHA-stimulated cultures is expressed as mean of cpm × 10^{-5} of triplicate cultures after subtracting that of unstimulated control cultures. S.E. did not exceed 10% of the mean for the experiments.
* [3H]dThd incorporation in nonstimulated cultures is expressed as mean of cpm × 10^{-5} of triplicate cultures. S.E. did not exceed 10% of the mean for these experiments.
potent suppressor activity than did a-NS. By calculating the ratio between the dose that inhibited 50% of the responses, the suppressor activity of the a-TBS was 6 times higher than that of the a-NS. However, a significant suppressive effect was evident also when a-NS were added. Among the experiments, the difference between the suppressor activity of a-TBS and a-NS ranged from 2- to 10-fold. We considered the possibility that exhaustion of the medium or cold thymidine release by macrophages might be trivial explanations for the observed inhibition of the response. However, as shown in Table 2, enrichment of the medium with asparagine or arginine, and/or glucose, did not influence the high suppressor activity of a-TBS or the lower one of the a-NS, nor did replacement of the medium before the pulse with [3H]dThd. Thus, macrophages induce a real inhibition of the proliferative response.

**Suppression of the LPS Response.** Since we observed an inhibitory activity of macrophages on the PHA-induced proliferation of T-cells, we were interested in looking at their effect on LPS-induced B-cell proliferation. At all concentrations tested, a-TBS showed an inhibitory activity on the LPS response of NS (Chart 4). In the majority of the experiments, 100% of inhibition was observed when 12% of a-TBS was added. Also, in this case, a-NS showed some suppressive activity, but of lower intensity.

**Suppression of the MIF Response.** We were interested in looking at the effect of suppressor macrophages on MIF production, since it is an early expression of the activation of lymphocytes by PHA, compared to the proliferative response. MIF was produced after a 4-hr pulse with 1:100 final dilution of PHA, followed by extensive washing of the stimulated cells and incubation of the spleen cells for 24 hr. The addition of increasing amounts of a-TBS inhibited MIF production, as reflected by decreased migration inhibition (Chart 5). Significant suppression by a-NS was achieved only with high concentrations (20%). Thus, a-TBS, and a-NS to a considerably lower extent, were able to suppress MIF production.

**DISCUSSION**

In the present paper we have demonstrated immunological hyporesponsiveness in the spleen cells of mice bearing the spontaneous tumor, ADK-1t. The decreased proliferative re-

---

Table 2

<table>
<thead>
<tr>
<th>Culture conditions</th>
<th>PHA response ([3H]dThd incorporation in cpm x 10^3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>No addition</td>
<td>a-TBS added^a</td>
</tr>
<tr>
<td>None</td>
<td>60 22 40</td>
</tr>
<tr>
<td>Glucose (100 mg/ml) + arginine (100 mM) + lysine (100 mM)</td>
<td>72 14 32</td>
</tr>
<tr>
<td>Glucose (100 mg/ml) + arginine (100 mM) + lysine (100 mM)</td>
<td>81 20 58</td>
</tr>
<tr>
<td>Medium replaced before [3H]dThd addition</td>
<td></td>
</tr>
</tbody>
</table>

^a Ten % of macrophages, either from TBS or NS, were added to 2.5 x 10^5 d-NS, and the PHA response was assayed in the specified culture conditions.

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Chart 3. Effect of increasing concentrations of normal and TBS macrophages on PHA response of NS. d-NS (2 x 10^6) were mixed with a-NS (C) or a-TBS (B). The percentage of inhibition is relative to that of 2.5 x 10^6 d-NS alone (75,215 cpm). The [3H]dThd of the macrophages alone was always <1,000 cpm.

---

Chart 4. Effect of increasing concentrations of normal and TBS macrophages on LPS response of NS. NS (2 x 10^6) were mixed with a-NS (C) or a-TBS (B). The percentage of inhibition is relative to that of 2.5 x 10^6 NS alone (48,370 cpm). The [3H]dThd of the macrophages alone was always <1,000 cpm.

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Chart 5. Suppressor activity of macrophages on MIF production. Increasing concentrations of macrophages either from TBS (B) or NS (C) were added to 5 x 10^6 NS, and the MIF production after PHA stimulation was evaluated.
immunogenicity (24). Suppressor macrophages have been found in mice bearing Moloney sarcoma virus-induced tumors (16, 24), plasmacytomas (18), and methylcholanthrene-induced sarcomas (27). Moreover, they have been described in rats (23) and humans (12). We were interested in defining the immune depression produced by the growth of ADK-1t, since this tumor, as well as other spontaneous tumors (13), has poor immunogenicity (4), and induces a very poor and transient specific response in the syngeneic host (8).

The progressive growth of ADK-1t in BALB/c mice was accompanied by an increase in the size of the spleen and in its percentage of macrophages. Similar changes have been described in spleens of mice bearing murine sarcoma virus-induced regressing sarcomas (16). In mice bearing ADK-1t, irrespective of the time after inoculation and of spleen size, the response to PHA could be completely restored by removal of nylon-adherent cells from the spleen. This demonstrated that hyporesponsiveness in tumor-bearing mice was not an intrinsic defect of the PHA-responsive cells. In addition, by mixing experiments with NS, the TBS were shown to have strong suppressor activity. Both hyporesponsiveness and suppressor activity were abrogated by the same treatments of the TBS: (a) passage over a nylon column; (b) treatment with carrageenan, and (c) carbonyl iron and magnet treatment. These data ruled out the possibility that the hyporesponsiveness was due to a dilution of PHA-sensitive cells with nonresponder cells in vivo. Carrageenan which has specific effects in macrophages (19) and iron and magnet treatment do not affect the relative proportion of nonphagocytic cells. Furthermore, a small number of a-TBS or nylon-adherent TBS were sufficient to inhibit the response of d-NS. The above-mentioned properties of the suppressor cells, taken together with their insensitivity to anti-

The suppressor activity exerted by macrophages of ADK-1t-bearing mice was not restricted to inhibition of lymphoproliferative responses to T-cell mitogen, PHA, since the response to LPS was also strongly inhibited. In addition, preliminary experiments indicate that they can inhibit responses in mixed-leukocyte cultures. Macrophages present in the spleens of mice bearing large ADK-1t tumors were suppressive at a very low concentration (0.5 to 2.5%). The dose-response curve of the inhibition of the proliferative response was nearly linear but rather resembled a logarithmic function. This implies that a given number of macrophages was more suppressive at a low concentration than at the higher concentrations.

The issue is frequently raised as to whether apparent suppressor activity by macrophages could be attributed to culture artifacts, particularly inhibition of [3H]dThd uptake by release of thymidine from the macrophages (25). In our studies, no differences in suppressor activity were observed when the culture medium had been enriched in amino acids and glucose or when it was replaced before pulsing of the cells with [3H] dThd.

Interestingly, we saw that normal splenic BALB/c macrophages are able, to some extent, to suppress the lymphocyte proliferation induced either by PHA or by LPS. Suppressor activity by macrophages has generally been considered to be associated with a state of activation. However, Oehler et al. (23) showed that normal rat spleen cells were suppressive in a mixed-lymphocyte-tumor interaction, although not as sup-

pressive as spleen cells from tumor-bearing rats. Elgert et al. (6) found that macrophages of normal BALB/c mice were as suppressive as those from mice bearing a chemically induced fibrosarcoma. Baird and Kaplan (1) also demonstrated that normal, as well as activated, macrophages could suppress PHA responsiveness through a contact-dependent process.

In our system, the macrophages from ADK-1t-bearing mice were always more suppressive than were the normal at the same concentration. This was also true for the suppression of MIF production. Macrophages from TBS completely inhibited MIF production by PHA-stimulated lymphocytes at a concentration of 10%, while a-NS had the same effect only at a concentration of 20%. MIF synthesis appears to be independent of proliferation (3, 29). The demonstration of suppression of lymphokine production by activated macrophages supports the recent findings (17, 29) that proliferation-associated immunoresponses are not the only targets of macrophage-dependent suppression. Presently, it is not clear whether the suppression of lymphokine production reflects inhibition of an early metabolic step leading to proliferation or whether suppression of lymphokine production and lymphoproliferation are unrelated parallel phenomena.

MIF production has been observed as part of the specific immune response to different tumors (21). The ability of macrophages from ADK-1t-bearing mice to suppress MIF production may be an additional reason for the progressive debilitation of the cell-mediated immunity connected with tumor growth.

It is of interest that a strong macrophage-dependent suppressor activity has been observed in tumors with a wide range of immunogenicity, from the poorly immunogenic ADK-1t to the highly immunogenic murine sarcoma virus-induced regressor tumors (24, 29). It seems that the appearance of suppressor cells is unrelated to the antigenicity of the tumor. We may speculate that the activation of suppressor cells is not a homostatic phenomenon regulating the antitumor response but rather is related to tumor growth. Suppressor cells could be generated by products released by the tumor or by the general inflammation caused by the growing neoplastic mass. In order to better understand the factors involved in induction of suppressor activity by macrophages, it will be helpful to carefully compare the events occurring in poorly immunogenic tumors with those occurring with the more widely studied and strongly immunogenic tumors.

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Suppression of Proliferative Response and Lymphokine Production during the Progression of a Spontaneous Tumor

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