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Inhibition of angiogenesis and growth of human nerve-sheath tumors by AGM-1470

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The effectiveness of AGM-1470, a potent, fungal-derived inhibitor of angiogenesis, in suppressing the neovascularization and growth of human Schwann cell tumors was tested in six schwannomas, seven neurofibromas, and one neurofibrosarcoma. Tumor fragments from surgical specimens were implanted into the subrenal capsule of 348 nude mice (nu/nu). Seven days after implantation, the tumors were measured and vascularity was graded. The animals were then randomly assigned to one of two groups, to receive either saline (control group) or systemic AGM-1470 treatment. After 2 to 6 weeks of treatment, tumor size and degree of vascularity were recorded. In the six different schwannomas implanted into 138 mice, the average vascular grade in the control group after 2 weeks of treatment increased from 2.2 to 3.2 (+1.0), while in the AGM-1470-treated group it decreased from 2.2 to 1.7 (−0.5) ($p < 0.01$). In the seven different neurofibromas implanted into 158 mice, the change in the average vascular grade in control and AGM-1470-treated animals was +0.5 and −1.0, respectively ($p < 0.01$). In the one neurofibrosarcoma implanted into 52 mice, the change in average vascular grade in each group during the 6-week treatment period was +1.9 and −1.0, respectively ($p < 0.01$). Neurofibrosarcoma growth after 6 weeks of AGM-1470 treatment was only 8.5% of the growth found in the control animals ($p < 0.01$). This study determined that AGM-1470 is effective in inhibiting angiogenesis and the growth of human nerve-sheath tumors.

KEY WORDS • **angiogenesis** • **AGM-1470** • **nerve-sheath tumor** • **schwannoma** • **neurofibromatosis** • **neurofibroma** • **neurofibrosarcoma**

ANGIOGENESIS, the formation of new blood vessels, is necessary for solid tumors to grow larger than a few cubic millimeters in size.[7,9,10] Angiogenesis inhibitors may provide a novel means of treating some tumors.[4,8,14] Schwannomas, like all solid tumors, depend on their blood supply to continue to grow and produce angiogenic growth factors, one of which is basic fibroblast growth factor.[24]

Neurofibromatosis comprises at least two autosomal dominant disorders, neurofibromatosis type 1 and type 2, affecting an estimated 100,000 Americans.[23] Patients with neurofibromatosis type 2 present with multiple nervous system tumors including bilateral acoustic neuromas, spinal schwannomas, neurofibromas, neurofibrosarcomas (malignant schwannomas), ependymomas, and meningiomas;[20,21,23] these patients are ideal subjects for the development of preventive therapeutic modalities because patients with this disease can often be identified prior to the onset of tumor growth.[27] Despite advances in understanding the pathogenicity of neurofibromatosis, no effective way of treating or preventing the development of these tumors exists, except for surgery. Moreover, surgical specimens of acoustic neuroma, neurofibroma, and neurofibrosarcoma may be very vascular,[13] and elicit a significant and consistent angiogenic response when implanted under the renal capsule of nude mice.[16] Neurofibrosarcomas are the most common malignancy in neurofibromatosis type 1 and are usually fatal despite aggressive surgery, chemotherapy, and radiotherapy.[32]

We previously reported the inhibition of growth and angiogenesis of neurofibrosarcomas by heparin and hydrocortisone therapy.[16] The purpose of the current study was to determine if one of the most potent angiogenesis inhibitors, AGM-1470 (O-chloroacetylcaramoyl fumagillol),[15] is effective in inhibiting the angiogenesis and growth of xenografts of human nerve-sheath tumors including schwannomas, neurofibromas, and neurofibrosarcomas. This agent is a novel, potent, fungal-derived, synthetic inhibitor of angiogen-

esis, which retards endothelial cell proliferation[15] and chemotaxis[2] in picogram amounts and also suppresses tumor growth;[12] AGM-1470 has also been shown to inhibit angiogenesis in multiple *in vivo* assays, including the chick chorioallantoic membrane,[12] rat and mouse subcutaneously implanted sponges, and the rat cornea.[15] We used the subrenal capsule assay to evaluate the inhibitory effect of AGM-1470 on human tumors of nerve-cell origin. This is the first report of the efficacy of angiogenic inhibition in a human tumor by AGM-1470, a compound planned for human therapeutic trials.

Materials and Methods

Tumor Implant Preparation

Fourteen human tumors of nerve cell origin (six schwannomas, seven neurofibromas, and one neurofibrosarcoma) were obtained from surgery. After submission of tumor specimens for pathological studies, the remaining tissue was placed in vials containing nutrient medium (Dulbecco's minimum essential medium) supplemented with 10% fetal bovine serum, penicillin, amphotericin B (Fungizone), and streptomycin.* The tumor tissue was cut into approximately 0.5- to 1.5-cu mm fragments before implantation into the subrenal capsule of nude mice.

Surgical Procedure, Treatment Protocol, and Vascular Grading

Congenitally athymic female nude mice (NCr/Sed nu:nu),† each weighing 16 to 24 gm, were kept in groups of five or less, housed in sterile cages, and given free access to autoclaved food and water. Surgery was performed in a designated nude mouse facility under sterile conditions. Each mouse was anesthetized with an intraperitoneal injection of 0.25 ml Avertin (2,2,2-tribromoethanol + tert-amyl alcohol), supplemented with 0.03 to 0.15 ml Avertin when necessary. Implantation of tumor tissue beneath the subrenal capsule was carried out as described previously.[1,17]

A stereomicroscope, with an ocular micrometer accurate to 0.1 mm and × 60 amplification, and the same surgical and anesthetic techniques were used when measuring tumor volume and grading vascularity on Day 7 postimplant. Tumor dimensions were recorded by measuring two perpendicular diameters of the implant. Volume was estimated from the formula:[6] volume = length × width × width × 0.5. Tumor vascularity was graded as follows: Grade 0, no visible vessels; Grade 1, vessels present in one quadrant of the tumor surface or one vessel present in more than one quadrant; Grade 2, vessels present in two quadrants of the tumor surface, with a total of more than one vessel observed; Grade 3, vessels present in three quadrants of the tumor surface, with a total of more than two vessels observed, or vessels in all four quadrants, excluding the central area of the tumor; and Grade 4, vessels present in all four quadrants, including the central area. Grade 0 tumors were considered possibly not to have taken, and therefore were excluded from the study. Grade 4 tumors had already reached maximum angiogenic stimulation, and therefore new blood vessel growth would not be detected; thus, Grade 4 tumors were also excluded from the study. The remaining mice with Grade 1, 2, or 3 vascularity were randomly assigned to either the treatment or the control group.

Preparation of AGM-1470 and Control Solutions

The AGM-1470 was stored at −20°C and then dissolved and stored in a 10% (wt/vol) solution of 100% ethanol at 4°C. Immediately prior to treatment, the stock solution was diluted in 0.9% sodium chloride solution (20 μl stock solution/1 ml saline) in a polypropylene tube. A total of 30 mg/kg per treatment was injected intraperitoneally or subcutaneously. An equivalent volume of 2% ethanol solution in 0.9% saline was given to the control mice.

Subrenal Capsule Assay for Schwannomas and Neurofibromas

In order to determine the difference in effect and toxicity between the two administration routes of AGM-1470, the agent was injected subcutaneously in mice implanted with three schwannomas and three neurofibromas, and intraperitoneally in mice implanted with three schwannomas and four neurofibromas. The mice were injected every Monday, Wednesday, and Friday, starting on Day 7 postimplant and continuing through Day 21. On Day 19, the animals were reexplored, tumor dimensions were recorded, and vascularity was graded. The mice were weighed on Days 7 and 21. Deaths within 24 hours after the operation were ascribed to anesthesia or surgery; deaths after 24 hours were counted as treatment-related. The animals were humanely sacrificed after the last exploration with an overdose of Avertin.

For the mice implanted with the three schwannomas (SCH1, SCH2, SCH3) and three neurofibromas (NF1, NF2, NF3), the operation, random assignment, injection, and grading were performed by one person. To exclude any possibility of observer bias, the other experiments were carried out in a blinded manner as follows. Tumor implantation, treatment, and grading on Day 7 were done by one of the authors. The animals were blindly and randomly assigned to one of the two groups. The solutions (saline and AGM-1470) were prepared and code labeled "A" or "B" by a second investigator. After 2 weeks of treatment, the tumor vascularity was graded independently by two or three investigators in a blinded manner. Treatment codes were revealed only after the conclusion of each experiment.

Subrenal Capsule Assay for Neurofibrosarcoma

Treatment in the mice implanted with neurofibrosarcoma was continued for 6 weeks. The mice were treated in the same manner as those implanted with

* Nutrient medium, fetal bovine serum, and antibiotic agents supplied by GIBCO Laboratories Life Technologies, Inc., Grand Island, New York.

† Mice supplied by the Edwin L. Steele Laboratories for Radiation Biology, Massachusetts General Hospital, Boston, Massachusetts.

TABLE 1

*Mean vascular grades before and after treatment with saline or AGM-1470 in mice implanted with schwannomas and neurofibromas**

Tumor Type: Administration Method	Saline Control Group			Change in Vascular Grade	AGM-1470 Group			Change in Vascular Grade
	No. of Animals	Grade Before Treatment	Grade After Treatment		No. of Animals	Grade Before Treatment	Grade After Treatment	
schwannoma: subcutaneous injection								
SCH1	4	2.0 ± 0.8	3.2 ± 1.3	+1.2	5	2.0 ± 1.0	1.4 ± 1.7	−0.6
SCH2	6	1.5 ± 0.5	3.0 ± 1.1	+1.5	7	1.6 ± 0.5	1.3 ± 1.1	−0.3
SCH3	6	1.9 ± 0.9	3.0 ± 0.9	+1.1	8	2.0 ± 0.8	1.5 ± 1.1	−0.5
total	16	1.8 ± 0.8	3.0 ± 1.0	+1.2	20	1.9 ± 0.7	1.4 ± 1.2	−0.5
schwannoma: intraperitoneal injection								
SCH4	9	2.2 ± 0.8	3.3 ± 0.5	+1.1	7	2.2 ± 1.0	1.4 ± 1.3	−0.8
SCH5	6	2.4 ± 0.9	2.8 ± 0.8	+0.4	6	2.5 ± 0.8	1.3 ± 1.0	−1.2
SCH6	6	3.0 ± 0.0	3.8 ± 0.4	+0.8	10	2.9 ± 0.3	2.5 ± 1.0	−0.4
total	21	2.5 ± 0.7	3.4 ± 0.7	+0.9	23	2.6 ± 0.7	1.9 ± 1.2	−0.7
total	37	2.2 ± 0.8	3.2 ± 0.8	+1.0	43	2.2 ± 0.8	1.7 ± 1.2	−0.5
neurofibroma: subcutaneous injection								
NF1	5	2.0 ± 0.7	3.8 ± 0.4	+1.8	4	2.3 ± 0.5	1.8 ± 0.5	−0.5
NF2	8	1.4 ± 0.5	2.8 ± 0.9	+1.4	7	1.4 ± 0.5	1.0 ± 0.6	−0.4
NF3	9	2.6 ± 0.7	2.8 ± 1.1	+0.2	9	2.7 ± 0.7	1.1 ± 1.1	−1.6
total	22	2.0 ± 0.8	3.0 ± 1.0	+1.0	20	2.2 ± 0.8	1.2 ± 0.8	−1.0
neurofibroma: intraperitoneal injection								
NF4	8	2.5 ± 0.5	3.1 ± 0.9	+0.6	11	2.4 ± 0.7	1.7 ± 1.1	−0.7
NF5	8	2.6 ± 0.7	3.2 ± 0.7	+0.6	6	3.0 ± 0.0	1.1 ± 1.3	−1.9
NF6	11	2.6 ± 0.7	2.7 ± 1.6	+0.1	10	2.8 ± 0.4	2.2 ± 1.3	−0.6
NF7	10	2.6 ± 0.7	2.8 ± 1.1	+0.2	6	2.8 ± 0.4	0.9 ± 0.9	−1.9
total	37	2.6 ± 0.6	2.9 ± 1.2	+0.3	33	2.7 ± 0.5	1.6 ± 1.2	−1.1
total	59	2.4 ± 0.8	2.9 ± 1.1	+0.5	53	2.5 ± 0.7	1.5 ± 1.1	−1.0

* Mean values are expressed ± standard deviation.

schwannoma or neurofibroma. Animals were re-explored on Days 7, 21, 35, and 49 postimplant to determine vascular grade and tumor size.

Statistical Analysis

The exact Kruskal-Wallis test was used to compare changes in vascularity in the treatment and control animals implanted with a single tumor specimen. Data pooled from the mice implanted with different specimens of the same tumor type and treated with the same drug were analyzed by the Pearson's p-lambda test.[28] The results of each group regarding tumor volume and body weight were evaluated using the one-factor analysis of variance test. For analysis of the death rate in each group, the chi-squared test and Fisher's exact test were used. Statistical significance was set at $p < 0.05$.

Results

Survival Characteristics

Tumor fragments from six acoustic neuromas were implanted in a total of 138 mice. Histologically, all tumors were schwannomas. Three mice died perioperatively. At first grading, 22 (16.3%) and 10 (7.4%) of the 135 tumors were Graded 0 and 4, respectively, and therefore were excluded from the experiment, leaving 103 mice for analysis. Tumor fragments from seven neurofibromas were implanted in a total of 158 mice. Four mice died perioperatively. At first grading, seven (4.5%) and six (3.9%) of the 154 tumors were Graded 0 and 4, respectively; these were excluded from the study, leaving 141 mice for analysis. In the 52 mice implanted with the neurofibrosarcoma, six (11.5%) of the tumors were Graded 0 at the first grading but no tumor was Graded 4, leaving 46 mice for analysis.

Angiogenesis in Schwannomas and Neurofibromas

Subcutaneous Administration. In the initial set of studies, the mice implanted with three different schwannomas (SCH1, SCH2, SCH3) and three different neurofibromas (NF1, NF2, NF3) were treated subcutaneously with saline or AGM-1470. For the mice implanted with schwannomas surviving through the treatment period, the average vascular grade for the control group treated with saline (16 mice) increased from 1.8 to 3.0 (+1.2), and for the group treated with AGM-1470 (20 animals) decreased from 1.9 to 1.4 (−0.5) ($p < 0.01$) (Table 1). For the mice implanted with neurofibromas surviving through the treatment period, the average vascular grade for the control group (22 animals) increased from 2.0 to 3.0 (+1.0), and for the AGM-1470-treated group (20 mice) decreased from 2.2 to 1.2 (−1.0) ($p < 0.01$) (Table 1).

Intraperitoneal Administration. The mice implanted with three different schwannomas (SCH4, SCH5, SCH6) and four different neurofibromas (NF4, NF5, NF6, NF7) were treated intraperitoneally with saline or AGM-1470. For the mice implanted with schwannomas surviving through the treatment period, the average vascular grade for the control group (21 mice) increased from 2.5 to 3.4 (+0.9), and for the AGM-

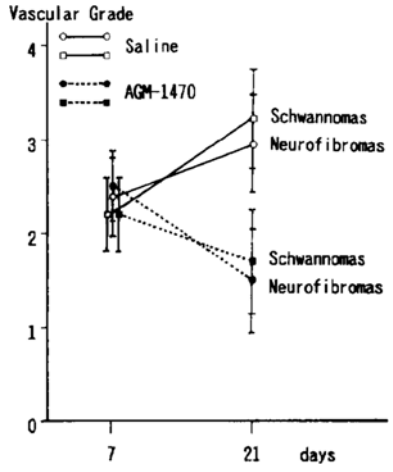

FIG. 1. Graph showing changes in vascular grade in xenografts of human schwannomas and neurofibromas. Following 2 weeks of treatment, the schwannomas in mice treated with AGM-1470 *(closed squares)* had significantly less vascularization than the schwannomas in the saline-treated control animals *(open squares)* ($p < 0.01$), and the neurofibromas in AGM-1470-treated animals *(closed circles)* had less vascularization than the neurofibromas in the saline-treated control mice *(open circles)* ($p < 0.01$). *Vertical bars* indicate standard deviation from the mean.

1470-treated group (23 animals) decreased from 2.6 to 1.9 (−0.7) ($p < 0.01$) (Table 1). For the mice implanted with neurofibromas, the average vascular grade for the control group (37 animals) increased from 2.6 to 2.9 (+0.3), and for the AGM-1470-treated group (33 mice) decreased from 2.7 to 1.6 (−1.1) ($p < 0.01$) (Table 1).

Comparison of Treatment Methods. The pooled data from the mice implanted with six schwannomas and seven neurofibromas showed statistically significant inhibition of angiogenesis by AGM-1470 treatment ($p < 0.01$) (Fig. 1). There were no significant differences between the two treatment methods, subcutaneous versus intraperitoneal, in either the mice implanted with schwannomas or those with neurofibromas ($p > 0.05$). These results indicate that AGM-1470 is effective in inhibiting angiogenesis in the subrenal capsule model of human xenografts of schwannomas and neurofibromas through either intraperitoneal or subcutaneous injection.

Effect of AGM-1470 on Animal Weight

Among the schwannoma-implanted animals, the 21 mice in the control group treated intraperitoneally gained a mean of 0.5 gm during the treatment period, whereas the 23 mice in the AGM-1470 group treated intraperitoneally lost a mean of 1.6 gm. The 16 mice in the control group treated subcutaneously gained a mean of 1.2 gm, whereas the 20 mice in the AGM-1470 group treated subcutaneously had no mean weight change. There were statistically significant differences in the two groups: the intraperitoneal control group versus the intraperitoneal AGM-1470-treated group

TABLE 2

*Weight changes in mice implanted with schwannomas or neurofibromas and treated with saline or AGM-1470**

Tumor Type & Treatment Group	IP Injection		SC Injection		p Value
	No. of Mice	Mean Weight (gm)	No. of Mice	Mean Weight (gm)	
schwannoma					
control	21	+0.5 ± 1.9	16	+1.2 ± 1.8	NS
AGM-1470	23	−1.6 ± 2.7	20	0.0 ± 1.2	$p < 0.02$
p value		$p < 0.01$		$p < 0.01$	
neurofibroma					
control	37	+1.7 ± 2.3	22	+1.9 ± 1.3	NS
AGM-1470	33	−0.7 ± 2.6	20	+0.3 ± 0.8	$p < 0.02$
p value		$p < 0.01$		$p < 0.01$	

* Mean values are expressed ± standard deviation. IP = intraperitoneal; SC = subcutaneous; NS = not significant.

($p < 0.01$), and the subcutaneous control group versus the subcutaneous AGM-1470-treated group ($p < 0.02$) (Table 2).

In neurofibroma-implanted animals, although the mice treated subcutaneously with AGM-1470 gained weight, there were statistically significant differences in the two groups: the intraperitoneal control group versus the intraperitoneal AGM-1470-treated group ($p < 0.01$), and in the subcutaneous control group versus the subcutaneous AGM-1470-treated group ($p < 0.01$).

For both types of tumor implant, the mice in the control and AGM-1470 groups treated subcutaneously gained more weight than those in the control and AGM-1470 groups treated intraperitoneally. The animals undergoing subcutaneous administration of AGM-1470 showed no weight loss at a dose of 30 mg/kg. However, intraperitoneal administration at this same dose resulted in a 4% weight loss.

Effect of AGM-1470 on Death Rates

For the mice implanted with neurofibromas, the death rates in the AGM-1470 and control groups treated intraperitoneally were 32.7% (16 of 49) and 21.3% (10 of 47), respectively. There were no statistically significant differences between the death rate in control groups and that in AGM-1470-treated groups. However, there were statistically significant differences between the mice treated intraperitoneally and the mice treated subcutaneously ($p < 0.02$), with more deaths in the intraperitoneally treated group.

Effect of AGM-1470 on Vascular Grade and Growth of Neurofibrosarcoma

Because benign schwannomas and neurofibromas grow so slowly, the effect of AGM-1470 on Schwann cell tumor growth could not be determined. Therefore, we also studied a malignant Schwann cell tumor from a patient with neurofibromatosis type 1. To test the effect of AGM-1470 on the neovascularization and growth of the neurofibrosarcoma, tumor specimens were implanted in the subrenal capsule of 52 nude mice, graded on Day 7 after implantation, randomly

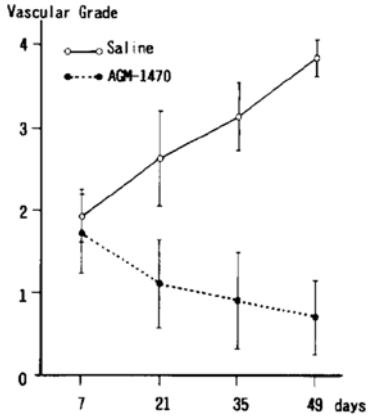

FIG. 2. Graph demonstrating that neovascularity in xenografts of human neurofibrosarcoma progressively increased over 6 weeks in animals treated with saline *(open circles)*. In contrast, animals treated with AGM-1470 showed a decrease in vascularity *(closed circles)* ($p < 0.01$ vs. controls). *Vertical bars* indicate standard deviation from the mean.

assigned to a control or treatment group, and treated with saline or AGM-1470 for 6 weeks thereafter.

On Day 7, the average vascular grades in the control and AGM-1470-treated groups were 2.0 and 1.8, respectively, and there was no statistically significant difference between the two groups. The average vascular grade of the tumors in the control mice increased to 3.9 at 6 weeks after implantation, in contrast to that in the AGM-1470-treated group which decreased to 0.8 (Fig. 2).

Tumor volume was also analyzed. In the control group, tumors increased in volume from 0.86 to 2.23 cu mm, a statistically significant increase ($p < 0.005$ vs. control). In contrast, the tumor volume in the AGM-1470-treated group decreased with time from 0.69 to 0.19 cu mm ($p < 0.02$ vs. control) (Fig. 3). There were two deaths in the control group and no deaths in the AGM-1470-treated group.

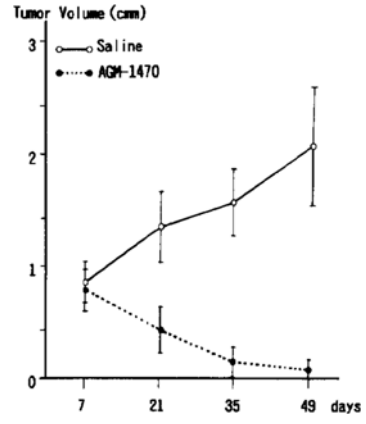

FIG. 3. Graph showing progressive enlargement in human neurofibrosarcoma xenografts in saline-treated animals *(open circles)*. Treatment with AGM-1470 caused a reduction in tumor volume *(closed circles)* ($p < 0.02$ vs. controls). *Vertical bars* indicate standard deviation from the mean.

Discussion

Angiogenesis and Tumor Growth

This study demonstrates that AGM-1470 inhibits both tumor-induced angiogenesis and growth of human nerve-sheath tumors. Folkman[8] hypothesized that the growth of solid tumors depends on continued angiogenesis in the surrounding area and that inhibitors of neovascularization might be used to suppress tumor growth. Prior studies have focused only on malignant tumors. However, angiogenesis can also be induced by benign tumors,[17] and the amount of angiogenesis induced by these tumors may correlate with their growth.[17] Such angiogenic tumors are potential targets of angiogenesis inhibitors.[4,5,7,9]

Angiogenic stimulation is essential for the growth of solid tumors.[7,9] Therefore, agents that block neovascularization might be able to stop or retard the growth of these tumors. The agent AMG-1470 is a potent angiogenesis inhibitor, which retards endothelial cell proliferation[12] and its chemotaxis in picogram amounts.[2] When given systemically, AGM-1470 results in the suppression of tumor growth[12] and metastasis.[2]

Elevated levels of basic FGF messenger ribonucleic acid transcripts in human schwannomas (acoustic neuromas and spinal schwannoma)[24] may account for some of the angiogenic activity of these tumors, since basic fibroblast growth factor is one of the most potent angiogenic proteins.[3] Extracts from nerve-sheath tumors, such as neurofibromas, also express basic fibroblast growth factor activity.[29] Furthermore, in the chick chorioallantoic membrane and the subrenal capsule assays, tumors of Schwann cell origin elicited a strong angiogenic response.[17,31] Therefore, from a biological basis, patients with neurofibromatosis would serve as good candidates for therapy with angiogenic inhibitors. In addition, we reported that heparin and hydrocortisone therapy inhibited angiogenesis and growth of human neurofibrosarcoma[16] (the most common malignancy in neurofibromatosis type 1), suggesting that malignant human nerve-sheath tumors may be treated by angiogenesis inhibitors. In the current study, we showed *in vivo* blocking of neovascularization by AGM-1470 in benign nerve-sheath tumor xenografts and inhibition of both vascularization and growth of a malignant nerve-sheath tumor.

Study Analysis

In our experiments, we used the subrenal capsule assay in athymic nude mice. This assay allows precise measurement (accurate to 0.1 mm) of change in tumor size. We graded the vascularity in a blinded manner by two or three observers. There was a high level of consistency in grading between the blinded observers (coincidence rate of 79.3%); this method is simple and reflects accurately the degree of neovascularization.

We demonstrated that AGM-1470 suppressed the neovascularization in schwannomas, neurofibromas, and neurofibrosarcoma. Furthermore, inhibition of neovascularization in neurofibrosarcoma during 6 weeks of treatment was also correlated with the inhibition of growth. These findings provide further evidence that

these tumors must stimulate blood vessels in order to grow.

No effective therapy currently exists for neurofibrosarcoma. Storm, *et al.*,[32] reported that, even with complete local disease control, 44% of patients died of metastasis, and a projected 5-year survival rate was only 40%. Our data show that AGM-1470 effectively inhibits not only angiogenesis, but also the growth of this often fatal tumor.

There was no significant difference in the death rate between the mice treated with saline and those treated with AGM-1470. However, the mice treated with AGM-1470 gained less weight than those treated with saline. Ingber, *et al.*[12] reported that animals treated with AGM-1470 gained weight slowly but consistently.

Clinical Relevance

This is the first study of angiogenic inhibition by AGM-1470 in human tumors. While most prior studies have emphasized the use of angiogenic modulators for malignant tumors, this study also suggests for the first time the possible use of angiogenic modulation for histologically benign tumors, such as those that occur in patients with neurofibromatosis type 1 or type 2. Since such patients may be identified while Schwann cell tumors are very small, this form of therapy could be used to inhibit the early stages of tumor formation or growth. By inhibiting tumor growth, multiple operations could be prevented and patients could live a relatively normal life. Additionally, growth inhibition could decrease the incidence of further tumor cell mutation and neoplasia. Since neurofibrosarcoma is the most common malignancy in neurofibromatosis type 1 and is often fatal despite surgery, irradiation, and chemotherapy, this mode of therapy may decrease the incidence of sarcoma formation and, for those sarcomas that do form, it may inhibit local tumor growth as well as metastasis, and thus prolong life.

Other angiogenesis inhibitors, such as all transretinoic acids,[11] cartilage-derived inhibitor,[22] Herbimycin A,[26] 22-oxa-1A, 25-dihydroxy vitamin D_3,[25] copper depletion and penicillamine,[5] platelet factor 4,[19] laminin peptide,[30] minocycline,[33] and a bacteria-derived sulfated polysaccharide-peptidoglycan complex,[34] have not been tested in this system and may also be proven to have therapeutic potential. Additionally, since angiogenic modulators may not kill tumor cells themselves, this new strategy could be combined with other treatment modalities. Studies have shown that antiangiogenic compounds may enhance the antitumor efficacy of conventional chemotherapeutic agents.[18] Thus, AGM-1470 may provide a new therapeutic modality for neurofibrosarcoma. If safety and efficacy are demonstrated in patients with malignant Schwann cell tumors, AGM-1470 may then be considered as a therapeutic agent for neurofibromatosis type 1 and type 2 wherein benign tumors may progressively grow and cause neurological deterioration and death.

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