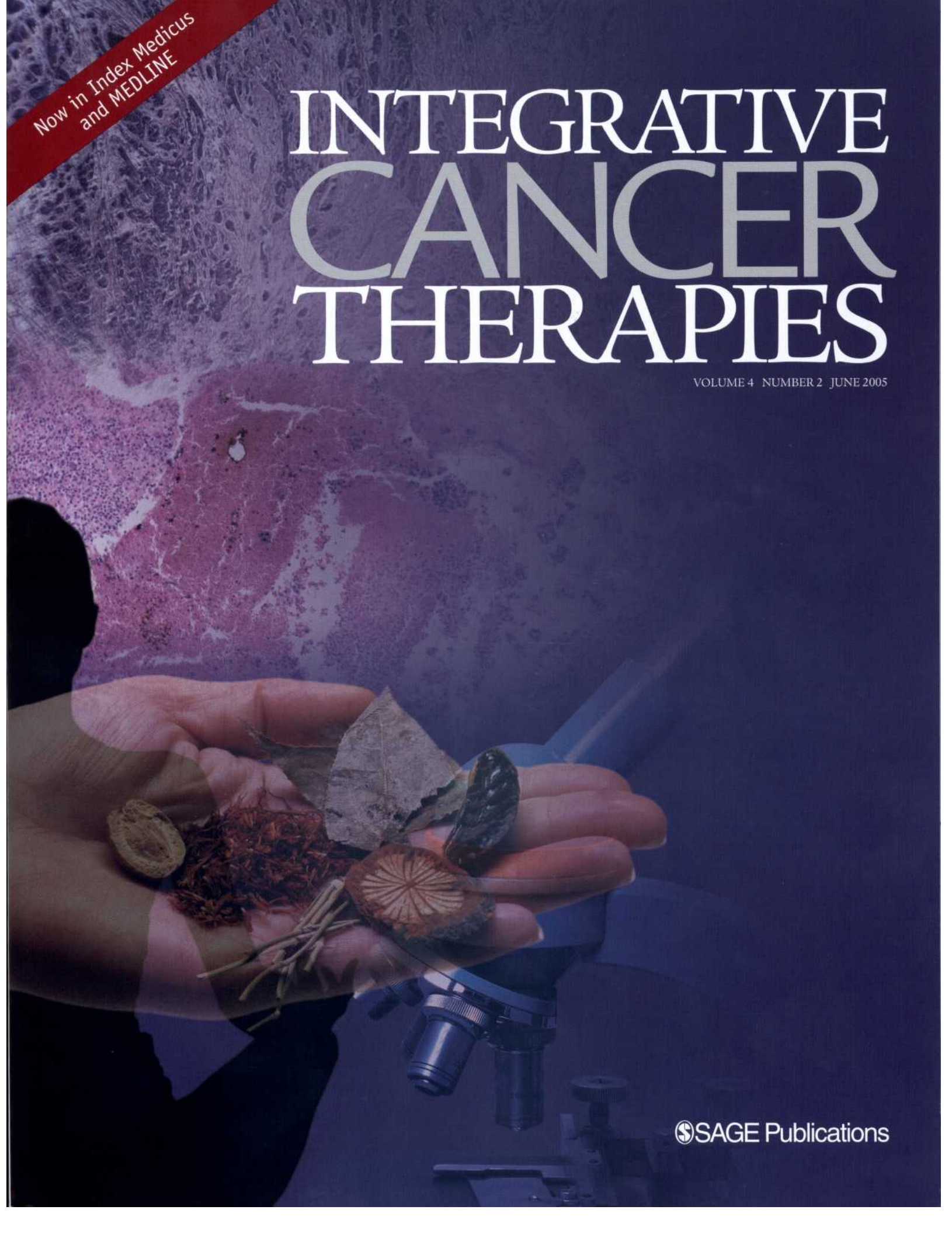


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Long-term Survival of High-Risk Pediatric Patients With Primitive Neuroectodermal Tumors Treated With Antineoplastons A10 and AS2-1

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Primitive neuroectodermal tumors (PNETs) are usually successfully treated with craniospinal radiation and chemotherapy; however, difficulties with standard treatment can be encountered in very young children, in adult patients at high risk of complication from standard treatment, and in patients with recurrent tumors. Thirteen children, either with recurrent disease or high risk, were treated in phase II studies with antineoplastons (ANP). The median age of patients was 5 years, 7 months (range, 1-11). Medulloblastoma was diagnosed in 8 patients, pineoblastoma in 3 patients, and other PNET in 2 patients. Previous treatments included surgery in 12 patients (1 had biopsy only, suboccipital craniotomy), chemotherapy in 6 patients, and radiation therapy in 6 patients. Six patients had not received prior chemotherapy or radiation. The treatment consisted of intravenous infusions of 2 formulations of ANP, A10 and AS2-1, and was administered for an average of 20 months. The average dosage of A10 was 10.3 g/kg/d and of AS2-1 was 0.38 g/kg/d. Complete response was accomplished in 23%, partial response in 8%, stable disease in 31%, and progressive disease in 38% of cases. Six patients (46%) survived more than 5 years from initiation of ANP; 5 were not treated earlier with radiation therapy or chemotherapy. The serious side effects included single occurrences of fever, granulocytopenia, and anemia. The study is ongoing and accruing additional patients. The percentage of patients' response is lower than for standard treatment of favorable PNET, but long-term survival in poor-risk cases and reduced toxicity makes ANP promising for very young children, patients at high risk of complication of standard therapy, and patients with recurrent tumors.

Keywords: PNET; medulloblastoma; pineoblastoma; brain tumor; long-term survival; antineoplastons; phase II study

Antineoplastons (ANP) A10 and AS2-1, which are synthetic analogs of naturally occurring derivatives of glutamine, isoglutamine, and phenylacetic acid, have shown an interesting spectrum of activity in primary brain tumors.¹ Published results of phase II studies

reported a significant percentage of objective responses.²⁻⁶ The ongoing clinical studies of ANP in primitive neuroectodermal tumors (PNETs) are not yet completed; however, interesting initial results and long-term survivals have prompted us to present these data.⁷

PNET was usually successfully treated with surgery and craniospinal radiation therapy. The addition of chemotherapy allowed a decrease in the total dosage of radiation and reduced debilitating chronic toxicity in young children.⁸⁻¹⁰ Despite these accomplishments, standard treatments are not as successful in high-risk patients.¹¹⁻¹³ Risk factors include age younger than 4 years, tumors in adults, subtotal resection, metastases, large cell anaplastic histology, and a number of genetic changes, including elevated expression of the genes related to cell proliferation and metabolism.^{14,15} High-risk patients were accrued to phase II studies with ANP.

Patients and Methods

Patients

A total of 13 evaluable pediatric patients diagnosed with poor-risk or recurrent PNET were treated under 2 protocols, CAN-01 and BT-12, supervised by the Food and Drug Administration (FDA) and the Institutional Review Board (IRB) of Burzynski Research Institute. The patients were children of median age of 5 years, 7 months (range, 1-11). Three patients were younger than 3 years. There were 10 males and 3 fe-

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Conflict of interest: SRB owns the Burzynski Clinic. RAW, TJ, BS, GJ, MK and VD are employees of the clinic.

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Table 1. Summary of Patient Characteristics (N = 13)

Characteristic	n	%
Gender		
Male	10	77
Female	3	23
Tumor type		
Medulloblastoma	8	61.5
Pinealoblastoma	3	23.1
PNET with neuronal and astrocytic differentiation	1	7.7
Supratentorial	1	7.7
Tumor spread		
Disseminated	8	60
Solitary ^a	5	40
Previous therapies		
SU/CH/RA	5	38
Bx/CH	1	8
SU/RA	1	8
SU	6	46
None	0	0
	<i>Range</i>	<i>Median</i>
Age, y	1-11	5.6
Karnofsky performance status	50-100	75

Patients were admitted between April 13, 1994, and December 4, 2001. Data are as of August 1, 2004. PNET = primitive neuroectodermal tumor; SU = surgery; CH = chemotherapy; RA = radiation; Bx = biopsy.

a. One patient had positive cerebrospinal fluid for neoplastic cells.

males. Average Karnofsky performance status (KPS) was 75 (from 50 to 100). Medulloblastoma was diagnosed in 8 patients, pineoblastoma in 3 patients, and other PNET in 2 patients. Involvement of the spinal cord was found in 1 patient and multiple cerebral metastases in 8 patients. Previous treatments included surgery in all patients (12 patients had resections and 1 had biopsy), chemotherapy in 6 patients, and radiation therapy in 6 patients. Among these high-risk patients, 6 had not received prior chemotherapy or radiation. In 12 patients, the tumor progressed prior to ANP, and the remaining patient had residual tumor after partial resection. Table 1 shows the patients characteristics.

Patient Assessment

For patients to be admitted to the studies, they had to have a KPS of 60 or higher, normal total serum bilirubin and creatinine levels, SGOT and SGPT levels not higher than 5 times the upper limit of normal, a leukocyte count of $2000 \times 10^3/\text{mm}^3$ or higher, and a platelet count greater than $50\,000 \times 10^3/\text{mm}^3$. Patients with a KPS of 50 or lower were treated under the FDA’s special exception. At least 8 weeks must have elapsed since the last dose of radiation therapy and at least 4 weeks since the last dose of chemotherapy (6 weeks for nitrosureas). Patients who were receiving corticosteroids were on a fixed dose for at least 1 week prior to baseline scan. Patients were excluded for the following reasons: serious active infections, fever or

any other serious disease that would interfere with evaluation of ANP, hypertension and congestive heart failure, and renal failure. Pretreatment evaluation was performed within 7 days before enrollment, including a complete medical history and physical examination, blood morphology and chemistry tests, urinalysis, and a baseline magnetic resonance image (MRI). Diagnosis was confirmed by pathologists not affiliated with the Burzynski Clinic.

Treatment

Protocol Design

Children diagnosed with PNET were treated in 2 different studies: CAN-01 and BT-12. The description and results of the CAN-01 study have already been published.² Three children diagnosed with PNET were treated in this study. The 2-stage phase II clinical study design proposed by Fleming was used in protocol BT-12.¹⁶ Initially, 20 adequately treated patients were to be assessed; if less than 1 objective response (complete or partial response) was observed, it would be concluded that there was less than desired activity and the study would be discontinued. If 1 or more objective responses were observed, 20 more patients would be accrued to the study. If 4 or more responses were observed among the 40 patients, the evidence would be considered sufficient to conclude that the treatment has the desired activity. The IRB supervised the study, and the membership of the IRB was in agreement with the FDA.

Treatment According to Protocol BT-12

The first 3 weeks of the treatment were administered at the Burzynski Clinic. All patients’ guardians signed informed consent forms that were reviewed by the FDA and approved by the IRB. Treatment consisted of daily intravenous (IV) injections of ANP A10 (A10 I; 300 mg/mL) and AS2-1 (80 mg/mL). On the first day of treatment, the patient received 10 mL of each formulation, A10 I and AS2-1, every 4 hours (6 times a day). Beginning the second day of treatment, the dose of each injection was increased daily in increments of 10 mL until the highest tolerated dosage (or effective dosage) was reached, not exceeding 25.0 g/kg/d of A10 and 0.6 g/kg/d of AS2-1. For children between 12 and 16 years of age, the dose of A10 was escalated daily in increments of 20 mL and in increments of 40 mL daily in patients older than 16 years. Patients were monitored daily for compliance and toxicity. Toxicity was graded based on the National Cancer Institute (NCI) common toxicity criteria, and the dose was modified as follows: (1) leukocyte count of 1.0 to $2.0 \times 10^3/\text{mm}^3$ or platelet count of 25 to $50 \times 10^3/\text{mm}^3$ = de-

crease both agents by 50%, (2) leukocyte count less than $1.0 \times 10^3/\text{mm}^3$ and platelet count less than $25 \times 10^3/\text{mm}^3$ = discontinue treatment, (3) nausea or vomiting of grade 3 or higher = decrease ANP by 50%, (4) increased sleepiness and weakness = decrease ANP by 25%, (5) transaminases SGOT and SGPT 10 times the upper normal limit and total bilirubin level 3 times the upper normal limit = discontinue treatment for 2 days and restart at 50% of the previous dose, (6) allergic skin reaction = discontinue treatment, (7) hypokalemia = replace with potassium chloride as required, (8) hyperuricemia greater than 12 mg/dL = add allopurinol, (9) hypernatremia = discontinue treatment until serum sodium concentration is below 148 mEq/L and provide hydration as needed, and (10) any other grade 3 or 4 toxicity = withhold treatment until toxicity has reduced to grade 1 or less and then restart with the dose decreased by 25%.

Family members received training in standard catheter care and programming of the intravenous pump. Patients and families were allowed to return home only after they were sufficiently trained. Patients were required to have a history and physical examination monthly, complete blood count with differential, urinalysis, and serum biochemistry profile once a week and electrolytes every other day during escalation of the doses and 3 times a week thereafter.

Evaluation of Treatment Efficacy

NCI criteria were used for evaluation of responses based on MRIs for both protocols. Some patients also had confirmation of results by positron emission tomography (PET) scans. Response criteria were as follows:

- complete response: complete disappearance of all contrast-enhanced tumor(s) on imaging studies for at least 4 weeks and receiving no corticosteroids;
- partial response: more than 50% reduction in the sum of the products of the greatest perpendicular diameters of the contrast-enhanced tumor(s) for at least 4 weeks with no appearance of new lesions on a decreasing or stable dose of corticosteroids;
- stable disease: less than 50% change (either greater or smaller) in the sum of the products of the greatest perpendicular diameters of the contrast-enhanced tumor(s) for a minimum of 12 weeks on a stable or decreasing dose of corticosteroids;
- progressive disease (PD): greater than 50% increase in the sum of the products of the greatest perpendicular diameters of the contrast-enhanced tumor(s) compared with nadir evaluation or the appearance of new lesions;
- progression-free survival (PFS): time from initiation of ANP to the date of PD, death, or the last contact with the patient; and

- overall survival: from diagnosis (OSD) and from ANP start (OSS).

Statistics

The studies under FDA-reviewed protocols were considered as the source of all cases described in this article. Response rates, survival statistics, incidences of adverse events, maximum dosages, average dosages, duration of treatment, and summary of patients' statistics were obtained using Microsoft Excel 97 and Access 02. A biostatistician reviewed the survival data.

Results

This article describes the results of treatment with IV ANP of children diagnosed with PNET in phase II studies, with special attention to the patients who survived longer than 5 years from the start of treatment. Study CAN-01 is completed and closed for admission, but there is ongoing accrual of new patients to the BT-12 study. Table 2 lists individual patient characteristics.

Efficacy

ANP IV was administered for an average duration of 20 months. The average dosage of A10 was 10.30 g/kg/d and of AS2-1 was 0.38 g/kg/d. Table 3 shows a summary of treatment data.

Complete responses were observed in 3 cases (23%), a partial response in 1 case (8%), stable disease in 4 cases (31%), and progressive disease in 5 cases (38%). Six patients (46%) survived more than 5 years from initiation of ANP. Table 4 summarizes ANP responses, baseline and current status, OSD, OSS, and PFS.

Long-term Survival

All children had 1 or more risk factors, including age younger than 4 years, subtotal resection, metastases, or large-cell anaplastic histology. The group of long-term survivors (over 5 years) includes 6 children; 5 males and 1 female, diagnosed from the ages of 1 to 9 years. Among these were 3 cases of medulloblastoma, 1 pineoblastoma, and 2 of other PNET. Three of these children had disseminated disease, and another 1 had involvement of the brainstem. Five of them had prior subtotal tumor resection. One patient did not have tumor resection (biopsy only) and was treated with combination chemotherapy. None of these patients received radiation therapy. Three of these patients obtained complete responses; 2 had stable disease and 1 developed progressive disease and subsequently received standard radiation therapy. One patient passed away after 6 years, 10 months from the start of the treatment (3 years after discontinuation of ANP). The cause of death was recurrent pneumonia, possibly due

Table 2. Individual Patient Characteristics

<i>Case</i>	<i>Protocol</i>	<i>Gender</i>	<i>Age at Admission, y</i>	<i>Ethnicity</i>	<i>Date of Initial Diagnosis</i>	<i>Tumor Type</i>	<i>Tumor Dissemination</i>	<i>KPS Baseline</i>	<i>SU</i>	<i>CH</i>	<i>RA</i>
1	BT-12	M	1	W	Jan 5, 1996	PNET with neuronal and astrocytic differentiation	Solitary	60	Bx	Yes	No
2	BT-12	M	4	W	May 28, 1996	Medulloblastoma	Disseminated	80	Yes	No	No
3	BT-12	M	4	W	Feb 9, 1996	Medulloblastoma	Disseminated	70	Yes	Yes	Yes
4	BT-12	F	1	W	Dec 18, 1996	Pinealoblastoma	Solitary	60	Yes	No	No
5	BT-12	M	9	W	Mar 18, 1997	Supratentorial	Disseminated	70	Yes	No	No
6	BT-12	M	6	W	Aug 11, 1997	Pinealoblastoma	Solitary	60	Yes	No	Yes
7	BT-12	F	5	W	Jan 8, 1998	Pinealoblastoma	Solitary	90	Yes	No	No
8	BT-12	M	9	W	Oct 16, 1997	Medulloblastoma	Disseminated	100	Yes	No	No
9	BT-12	M	5	W	May 10, 2000	Medulloblastoma	Disseminated	80	Yes	Yes	Yes
10	BT-12	F	9	W	Apr 16, 2001	Medulloblastoma	Disseminated	90	Yes	Yes	Yes
11	CAN-01	M	2	W	Mar 1, 1994	Medulloblastoma	Solitary	100	Yes	No	No
12	CAN-01	M	11	W	May 2, 1994	Medulloblastoma	Disseminated	90	Yes	Yes	Yes
13	CAN-01	M	6	W	Oct 17, 1994	Medulloblastoma	Disseminated	50	Yes	Yes	Yes

Patients admitted between April 13, 1994, and December 4, 2001. Data are as of August 1, 2004. KPS = Karnofsky performance status; SU = surgery; CH = chemotherapy; RA = radiation therapy; M = male; W = white; PNET = primitive neuroectodermal tumor; Bx = biopsy.

Table 3. Summary of Treatment Data With Antineoplastons

Case	Start Date	Stop Date	Days on Treatment ^a	Average Dosage, g/kg/d	
				A10	AS2-1
1	Apr 9, 1996	Mar 4, 2000	1173	5.65	0.34
2	Jul 8, 1996	Mar 25, 1997	256	13.31	0.47
3	Dec 18, 1996	Jan 24, 1997	37	7.63	0.40
4	Feb 27, 1997	Mar 6, 2003	2012	11.69	0.38
5	May 1, 1997	Jan 27, 1998	163	9.45	0.24
6	Dec 13, 1997	Dec 25, 2000	1044	10.04	0.34
7	Apr 1, 1998	May 14, 1998	43	11.79	0.35
8	May 8, 1998	Nov 3, 1998	157	12.66	0.40
9	Dec 4, 2001	Apr 18, 2002	133	21.60	0.54
10	Aug 29, 2003	Oct 21, 2003	42	9.21	0.29
11	Apr 13, 1994	Jun 16, 1999	952	2.95	0.43
12	Jan 24, 1996	Jul 23, 1996	131	8.55	0.39
13	Feb 9, 1996	May 9, 1996	90	9.39	0.37
Average			479	10.30	0.38
Median			157		

Patients admitted between April 13, 1994, and December 4, 2001. Data are as of August 1, 2004.

a. Indicates actual days on treatment. The time from start to stop date is not necessarily the same as the days on treatment, due to the short periods of discontinuations.

Table 4. Summary of Results of Treatment With Antineoplastons

Case	Response	Radiological PD	PFS, mo	Status	KPS Baseline	KPS Follow-up	Reason for Withdrawal	OSD, mo	OSS, mo
1	CR	NR	82.1	F	60	80	CR	85.3	82.1
2	SD	Dec 19, 1996	5.4	A	80	100	PD	78.8	77.5
3	PD	Jan 21, 1997	1.1	E	70	70	PD	12.4	2.1
4	CR	NR	89.0	A	60	90	CR	91.4	89.0
5	SD	NR	86.9	A	70	70	N	88.4	86.9
6	PR	NR	37.8	F	60	100	N	41.8	37.8
7	PD	May 6, 1998	1.2	F	90	90	PD	5.5	2.8
8	PD	Jun 15, 1998	1.3	A	100	90	PD	62.2	55.5
9	SD	Apr 15, 2002	4.3	F	80	80	PD	36.5	17.6
10	PD	Oct 16, 2003	1.6	E	90	90	PD	30.9	2.4
11	CR	NR	123.6	A	100	100	CR	125.0	123.6
12	SD	NR	16.3	F	90	90	N	37.1	16.3
13	PD	Mar 27, 1996	1.6	D	50	50	D	18.8	3.0
Median			5.4		80	90		41.8	37.8

Patients admitted between April 13, 1994, and December 4, 2001. Data are as of August 1, 2004. PFS = progression-free survival; KPS = Karnofsky performance status; OSD = overall survival from diagnosis; OSS = overall survival from start; CR = complete response; NR = not recurrent; F = died more than 30 days after treatment; SD = stable disease; A = alive; PD = progressive disease; E = died within 30 days after treatment; N = patient request; D = died on treatment.

to chronic immunosuppression from chemotherapy administered prior to ANP (patient 1). Two additional patients were lost to follow-up after 6 years, 3 months (patient 2) and approximately 5 years (patient 8), but they are believed to be alive since we do not have confirmation of their death. The 3 remaining patients are now alive (as of July 27, 2004) from more than 7 years to more than 10 years from the beginning of treatment (patients 4, 5, 11). These 3 patients underwent partial tumor resection, but none were treated with chemotherapy and radiation therapy prior to ANP. Patient 5 obtained stabilization of the disease as a result of ANP and thereafter underwent radiation therapy. Patient 4 was diagnosed with pineoblastoma, and patient 11 was diagnosed with medulloblastoma. They obtained complete responses as the result of ANP, and

they are off ANP at present, live normal lives, and were never treated with radiation therapy or chemotherapy.

Following is a short description of these 2 cases.

Patient 4

The patient was a 1-year-old female who, in December 1996, was diagnosed with a brain tumor and underwent partial resection on December 18, 1996. The pathology diagnosis was pineoblastoma. The patient did not receive any further treatment, and her follow-up MRIs of January 7, 1997, and February 26, 1997, revealed a recurrent and progressive tumor. She began ANP on February 27, 1997. Follow-up MRIs have shown a decrease of the tumor size, and PET scans of the brain on September 16, 2002; September 25, 2003;

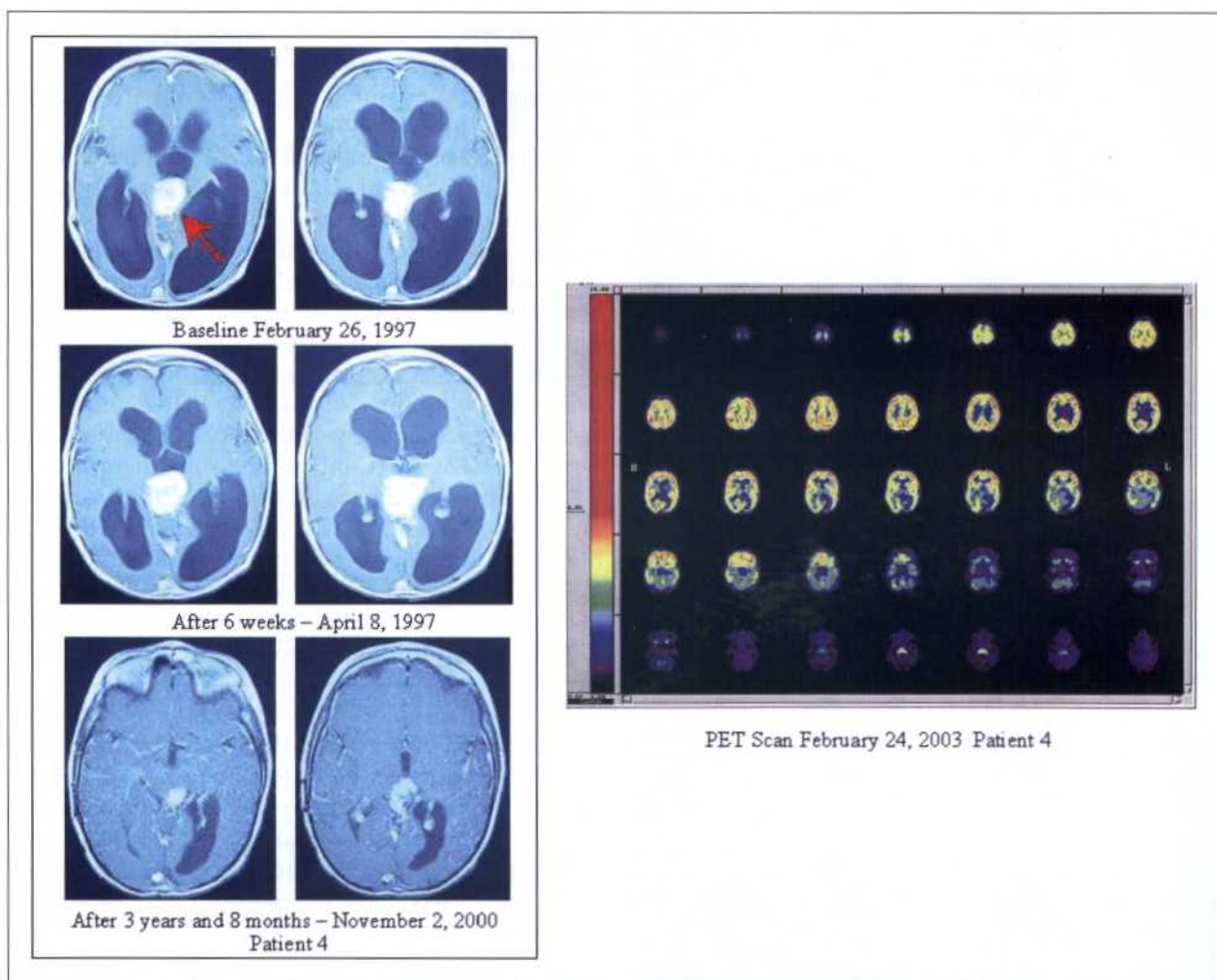


Figure 1 Treatment of a 1-year-old girl with pineoblastoma (patient 4). The patient's tumor progressed after partial resection and was not treated with radiation and chemotherapy. T1-weighted gadolinium magnetic resonance imaging of the head shows decrease of the tumor size (left). Positron emission tomography (PET) scan confirms complete response (right). The patient has been living a normal life for 7½ years since initiation of antineoplastons.

and December 4, 2003, did not reveal any hypermetabolic lesion, confirming a complete response. The patient has recovered completely from her symptoms, including blindness, and has been living a normal life for more than 7½ years since ANP was started (Figure 1).

Patient 11

The patient was a 2-year-old white male who, on February 13, 1994, underwent a subtotal tumor resection and was diagnosed with medulloblastoma of the cerebellum and brainstem. He did not receive any further conventional treatment. On April 11, 1994, the patient began ANP. The dosage of A10 was 0.77 g/kg/d and the dosage of AS2-1 was 0.38 g/kg/d. The pre-treatment MRI of the head on March 8, 1994, showed residual enhancing tumor measuring 1.8 × 0.8 cm. After 6 weeks of treatment, the tumor was no longer visible, indicating complete response. On March 15,

1997, the treatment with IV ANP was discontinued. The patient now lives a normal life, more than 10 years, 4 months since the start of treatment (Figure 2).

Toxicity

There were no grade 4 toxicities. Grade 3 toxicities included a single occurrence of fever and reversible granulocytopenia and anemia.

Discussion

PNETs, which are the most common malignant brain tumors in children, include medulloblastoma, pineoblastoma, supratentorial PNET, and ependymoblastoma. The exact mechanisms leading to these tumors are not known, but a number of genetic changes have already been described. Among common abnormalities is overexpression of proto-oncogene *MYCC*, *ERBB2*, and germ-line mutation of

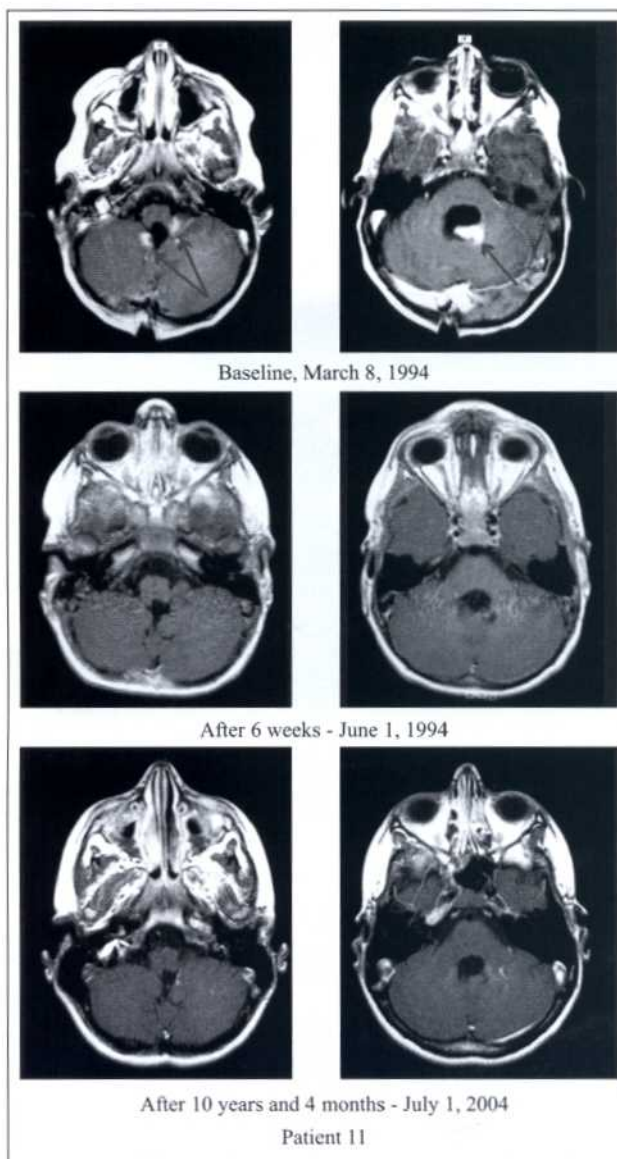


Figure 2 Successful treatment of a 2-year-old male diagnosed with medulloblastoma of the cerebellum and brainstem on February 13, 1994 (patient 11). After subtotal resection, the patient did not receive radiation therapy or chemotherapy. T1-weighted gadolinium magnetic resonance imaging of the head shows disappearance of the tumor size after treatment with antineoplastons. The patient is now alive and well more than 10 years, 4 months since the start of treatment.

the sonic hedgehog (*SHH*) receptor patched (*PTCH*).^{14,17,18} This last change is more typical for the less-malignant desmoplastic variant of medulloblastoma, which occurs in approximately 15% of cases. In contrast, *MYCC* is overexpressed or amplified in most cases of high-risk medulloblastoma.¹⁷ *MYCC* promotes cell growth and proliferation, inhibits terminal differentiation, and sensitizes cells to apoptosis, if the proliferative pathway or antiapoptotic proteins are blocked.¹⁹ *MYCC*,

through *MYC*-*MAX* dimers, promotes G1-S phase progression by activating genes *CCND2* (encoding cyclin D2) and *CDK4* (which encodes cyclin-dependent kinase 4) (Figure 3).^{20,21}

This leads to sequestration of KIP1 (or p27), which is an inhibitor of CDK2.²² KIP1 undergoes ubiquitination promoted by *CUL-1* and *CKS*, which are the genes targeted by *MYC*.²³ Under such conditions, KIP1 is no longer inhibiting the cyclin E-CDK2 complex, which permits phosphorylation of this complex by cyclin-activating kinase and entry to the G2 phase.²⁴ *MYC*-*MAX* represses *INK4B* (or p15) and p21, which are inhibitors of cyclin-E/CDK2 and which arrest cell cycle progression.²⁵ *MYC*-*MAX*, in association with MIZ1 (*MYC*-interacting zinc finger protein 1), prevents the transactivation of *CDKN1A*, which encodes p21, and *CDKN2B*, which encodes *INK4B*.^{19,26} Such growth-promoting activity of *MYC* is inhibited by PN, which activates *CDKN1A* through demethylation of its promoter.²⁷ The *MYC*-*MAX* complex also inhibits differentiation through chromatin remodeling (Figure 4).^{19,28}

Such action is blocked by the *MAD* protein, which replaces *MYC* in the *MAD*-*MAX* complex and binds *MYC*-*MAX* target genes.²⁹ Induction of *MAD* is facilitated by *MEFD2D* (*MAD*S box transcription enhancer factor). *PG* increases the expression of *MEFD2D* in a dose-dependent manner and causes terminal differentiation.^{30,31} *MYC*-induced apoptosis is an independent process from *MYC*-promoted cell proliferation (Figure 5).¹⁹

Protection of neoplastic cells from apoptosis occurs through mutation or silencing of tumor-suppressor genes and overexpression of antiapoptotic proteins. The 17p deletion, hypermethylation of tumor-suppressor gene *HIC-1* (hypermethylated in cancer-1), and activation of the downstream *RAS*/*MAPK* pathway was found in poor-risk medulloblastomas.^{32,34} *PN* interrupts signal transmission through the *RAS* pathway by decreasing farnesylation of the *RAS* protein, and *PG* decreases expression of *AKT2* in a dose-dependent fashion.^{30,31,35} This removes inhibitory effects of *AKT2* on the proapoptotic protein *BAD*. *PN* downregulates antiapoptotic *BCL2*, and *PG* may also inactivate antiapoptotic protein *BCL-X_L* through deamidation.^{36,37} *BCL2* and *BCL-X_L* reside in the outer mitochondrial membrane and suppress apoptosis by blocking mitochondrial outer membrane permeabilization³⁸ through the sequestration of activated *BAX*.³⁸ Inactivation of *BCL-X_L* allows *MYC* to release cytochrome c from mitochondria and trigger apoptosis.

The proposed mechanisms of actions of ANP are of a hypothetical nature, and they need to be confirmed in additional studies in medulloblastoma cells since

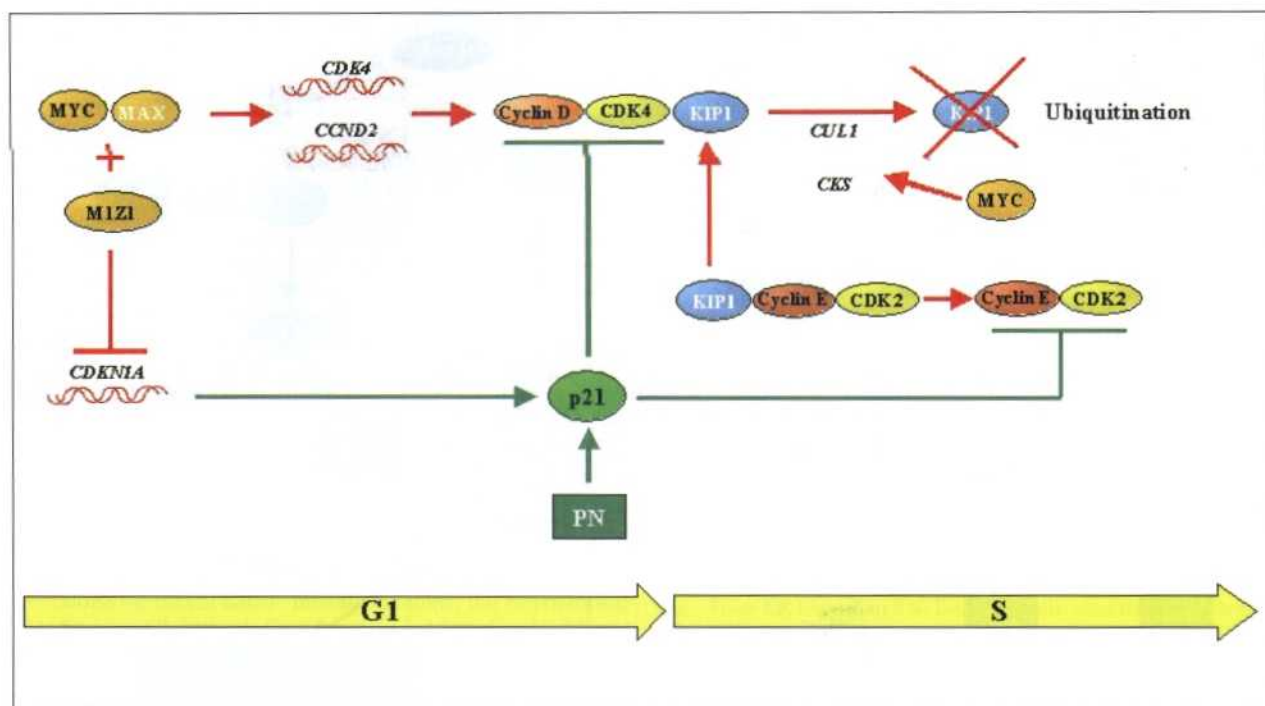


Figure 3 Inhibition of G1-S progression in medulloblastoma by PN (ingredient of antineoplastons).

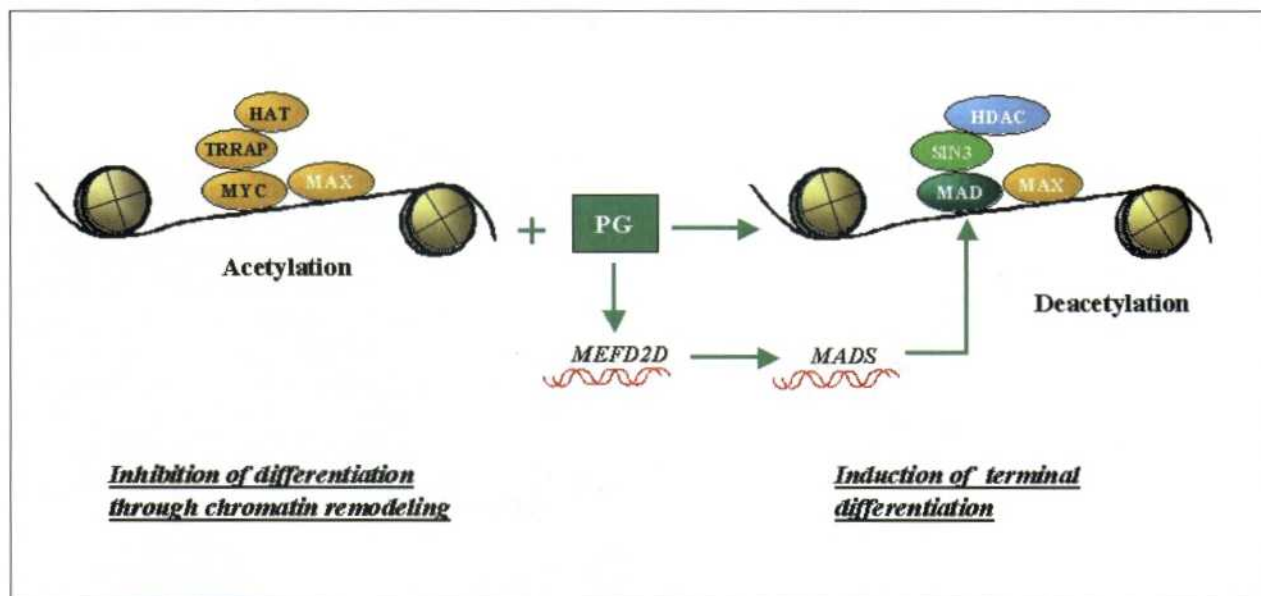


Figure 4 Induction of terminal differentiation in medulloblastoma by PG (ingredient of antineoplastons).

most researchers used different types of neoplastic cells. Most recent data, however, indicate without any doubt that PN inhibits proliferation and induces differentiation in human medulloblastoma cells.³⁹ The studies show that most patients responded to ANP and only 38% developed progressive disease. Close to 50% of patients survived more than 5 years from treatment start. It is important to notice that of these 6 long-term

survivors, none received radiation therapy and only 1 was treated with combination chemotherapy prior to ANP. It is well known that most patients diagnosed with PNET die within 1 year from diagnosis, if their treatment is limited to tumor resection and not followed by radiation therapy or chemotherapy. Contrary to a good prognosis and high response rate for children with favorable- or average-risk PNET, the

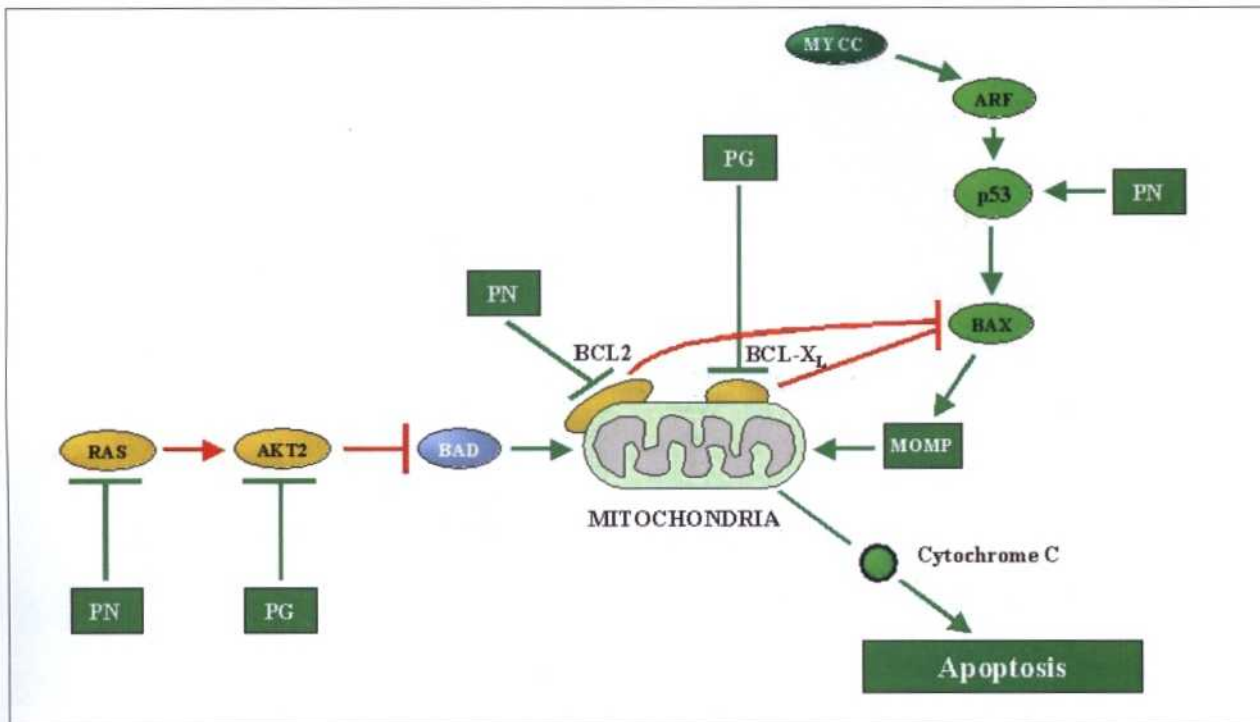


Figure 5 Induction of apoptosis in medulloblastoma by PN and PG.

children with poor-risk or recurrent tumors have substantially shorter survival. Children treated with radiation therapy who survive PNET suffer chronic toxicity, including hormonal deficiencies, intellectual decline, and the risk for secondary malignancies. The percentage of patient response and the long-term survival rate for patients treated for PNET with ANP are lower than in standard therapies used in children with favorable tumor characteristics, but on the other hand, these high-risk patients are very difficult to treat. Long-term survivals after ANP do not show any signs of chronic toxicity and are able to live normal lives.

Conclusion

In a small group of 13 children with high-risk PNET, the majority responded to treatment with ANP, and only 38% of the patients developed progressive disease. Six patients (46%) survived more than 5 years. Five patients in this group were not treated with radiation or chemotherapy prior to ANP. The treatment was well tolerated with minimum toxicity. The possible mechanism of action of ANP in PNET consists of inhibition of *MYCC* promotion of growth and proliferation and induction of terminal differentiation and apoptosis. The results are promising, taking into consideration reduced survival and response rates for children with poor-risk and recurrent PNET undergoing standard treatment and suggest possible new treatment for such patients. Continuation of the studies,

with accrual of additional patients, is necessary to prove the efficacy of the treatment of high-risk PNET patients.

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