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VOLUME 23 NUMBER 34 DECEMBER 1 2005

JOURNAL OF CLINICAL ONCOLOGY

ORIGINAL REPORT

Prospective, Randomized, Multicenter, Double-Blind Placebo-Controlled Trial Comparing Adjuvant Interferon Alfa and Isotretinoin With Interferon Alfa Alone in Stage IIA and IIB Melanoma: European Cooperative Adjuvant Melanoma Treatment Study Group

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Submitted December 2, 2004; accepted May 25, 2005.

Supported by Roche Austria GmbH, Vienna, Austria.

Authors' disclosures of potential conflicts of interest are found at the end of this article.

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0732-183X/05/2334-8655/$20.00

DOI: 10.1200/JCO.2004.00.8128

ABSTRACT

Purpose

The combination of interferon alfa (IFNα) and isotretinoin has shown a direct antiproliferative effect on human melanoma cell lines, but it remained unclear whether this combination is more effective than IFNα alone in patients with metastatic melanoma. We evaluated safety and efficacy of IFNα and isotretinoin compared with IFNα alone as adjuvant treatment in patients with primary malignant melanoma stage IIA and IIB.

Patients and Methods

In a prospective, randomized, double-blind, placebo-controlled trial, 407 melanoma patients in stage IIA (301 patients) and IIB (106 patients) were randomly assigned to either IFNα and isotretinoin (isotretinoin group; 206 patients) or IFNα and placebo (placebo group; 201 patients) after excision of the primary tumor. IFNα was administered three times a week at a dose of 3 million units subcutaneously for 24 months. Isotretinoin at a dose of 20 mg for patients $\leq$ 73 kg, 30 mg for patients greater than 73 kg, or placebo daily for 24 months.

Results

A scheduled interim analysis revealed no significant differences in survival rates, with the isotretinoin group and the placebo group showing 5-year disease-free survival rates of 55% (95% CI, 46% to 65%) and 67% (95% CI, 59% to 75%), respectively, and overall 5-year survival rates of 76% (95% CI, 67% to 84%) and 81% (95% CI, 74% to 88%), respectively. The trial was stopped for futility.

Conclusion

The addition of isotretinoin to an adjuvant treatment of low-dose IFNα in patients with stage IIA and IIB melanoma had no significant effect on disease-free or overall survival and is therefore not recommended.

J Clin Oncol 23:8655-8663. © 2005 by American Society of Clinical Oncology

INTRODUCTION

Interferons are a group of naturally occurring proteins with a large spectrum of biologic activities including antiviral, immunomodulatory, antiproliferative, and differentiation-inducing effects.[1-4] Treatment with interferon alfa (IFNα) has been shown to prolong disease-free survival in melanoma patients at moderate to high risk of developing metastatic disease after surgery (American Joint Committee on Cancer [AJCC] stage

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IIA to III)[5] and in some studies, to prolong overall survival in this patient population.[6-14]

As derivates of vitamin A, retinoids also show a wide spectrum of biologic activities, including immune modulation, sebosuppression, and effects on cell proliferation and cell differentiation in both melanoma and nonmelanoma cell lines.[15-19] In some clinical trials evaluating isotretinoin and etretinate for treatment of nonmelanoma skin cancers, tumor suppression was achieved,[18,20] although isotretinoin was ineffective in preventing basal cell carcinomas in a large study with 981 patients.[21] However, in an adjuvant treatment study,[22] vitamin A was not significantly better than observation alone in prolonging disease-free survival or overall survival in patients with primary melanomas thicker than 0.75 mm and with clinically negative lymph nodes.

Because the combination of IFNα and retinoic acid has shown a direct antiproliferative effect on malignant human cell lines, especially melanoma cell lines,[23-26] several phase II clinical trials have been performed to evaluate the combination therapy in patients with nonmelanoma skin cancers and in patients with metastatic melanoma.[27-36] In patients with metastatic melanoma disease, the effectiveness of the combination of IFNα and retinoic acid was unclear: some studies demonstrated efficacy, with total response from 20% to 30%,[27,32,33] whereas other studies reported no or extremely low response to the combination therapy.[30,35,36]

Motivated by the positive studies, we designed a prospective, randomized, multicenter, double-blind, placebo-controlled trial comparing adjuvant combination therapy of IFNα and isotretinoin with adjuvant therapy of IFNα alone in patients with primary malignant melanoma stage IIA and IIB AJCC/International Union Against Cancer [UICC] 1988[37] to investigate whether the addition of isotretinoin to IFNα resulted in greater treatment efficacy.

PATIENTS AND METHODS

Study Design

The study was a prospective, 24-month, randomized, multicenter, double-blind, placebo-controlled, parallel-group trial evaluating the efficacy of adjuvant therapy of 3 million units (MU) of IFNα given subcutaneously three times a week in combination with daily oral isotretinoin (Roaccutan; Roche, Vienna, Austria; isotretinoin group) compared with IFNα alone (placebo group) for treatment of primary melanoma in stage IIA/IIB. The primary end point of the study was disease-free survival, and the secondary end points were overall survival and evaluation of quality of life during treatment. Disease-free survival was chosen as primary end point to get a sufficient number of events for the required power, with a limited sample size and duration of the trial. The study was sponsored by Roche Austria GmbH (Vienna, Austria). Twenty centers in Austria, Hungary, the Netherlands, and Greece participated. Patient recruitment was started on October 16, 1996, and stopped on December 31, 2002.

Four hundred seven melanoma patients who met the inclusion/exclusion criteria were randomly assigned to receive either IFNα and isotretinoin (isotretinoin group; 206 patients) or IFNα and placebo (placebo group; 201 patients). Treatment was started within 16 weeks after completion of all surgical interventions, including primary and secondary tumor excision.

IFNα was administered subcutaneously at a dose of 3 MU 3 times a week for 24 months. Isotretinoin was given daily for 24 months at a dose of 20 mg (two 10-mg capsules) for patients with weight $\leq$ 73 kg and at a dose of 30 mg (three 10-mg capsules) for patients with weight greater than 73 kg. Patients in the placebo group received an equivalent number of matching placebo capsules daily for 24 months.

Radiotherapy or concomitant medication with tetracyclines, other immunotherapy, chemotherapy, or any investigational drug was disallowed during the trial. Patients were permitted to use paracetamol/indomethacine to ameliorate the flu-like symptoms associated with IFNα therapy.

The following assessments were made at baseline after randomization, once a month during the first three months, then at 3-month intervals: CBC and blood chemistry (including leukocytes, hemoglobin, WBC differential count, platelets, total bilirubin, liver function enzymes, alkaline phosphatase, creatine phosphokinase, renal function, cholesterol, and triglycerides). Additionally, only at baseline, the following laboratory tests were performed: total protein, blood glucose, lactate dehydrogenase acid, high- and low-density lipoproteins, thyroid-stimulating hormone, serum electrolytes, and urine analysis. Chest x-ray, ultrasonography of the abdomen and the draining lymph nodes, and x-ray of the cervical spine, thoracic spine, lumbar spine, one knee, one ankle, and both forearms were required at baseline and at the end of treatment. Blood pressure and weight were measured at baseline and every 3 months. A visual analog scale was used to assess quality of life every 3 months during treatment and every 6 months after treatment cessation.

The trial protocol and five amendments were reviewed and approved by local ethics committees at the participating sites. The study was overseen by a Data Safety Monitoring Committee consisting of two clinical pharmacologists, a statistician, a dermatologist, and a retired expert on isotretinoin, none of whom was otherwise involved in the study. The Data Safety Monitoring Committee met regularly to review safety data, trial conduct, and protocol violations. The study was conducted in accordance with the Declaration of Helskinki and with Good Clinical Practice guidelines. All patients received a detailed explanation of the potential risks and benefits of the study and provided signed informed consent before participating in the study.

Patients

Initially all patients between 18 and 75 years of age with primary melanoma in stage IIA or IIB as defined by AJCC/UICC 1988 (IIA, localized melanoma with tumor thickness of 1.51 to 4.0 mm or Clark level IV; IIB, localized melanoma $>$ 4.0 mm tumor thickness or Clark level V)[37] were eligible for the trial. Because the performance of sentinel node biopsy in routine settings was steadily increasing after the start of the study, the protocol was amended (amendment 2; May 10, 1999) to permit patients to be enrolled after sentinel node biopsy, provided that they were sentinel node–negative or, in case of positivity, that positivity revealed only micrometastasis and that subsequent radical lymphadenectomy showed negative lymph nodes.

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The patients had to meet also the following inclusion criteria: baseline laboratory results including leukocyte count (WBC) $\geq 3.0 \times 10^9$/L, platelets count $\geq 100 \times 10^9$/L, hemoglobin $\geq$ 10 g/100 mL (1 dL), cholesterol $\leq$ 300 mg/dL, and triglycerides $\leq$ 250 mg/dL. Women of child-bearing potential were informed orally about the teratogenic effect of retinoids and given a patient information sheet. Women of child-bearing potential were required to use an accepted method of birth control for at least 1 month before beginning therapy, during treatment, and for a minimum of 3 months after discontinuation of therapy. Women of child-bearing potential were also required to have a serum pregnancy test performed by a physician within 2 days of treatment initiation and monthly until 3 months after treatment cessation.

Pregnant or lactating women, individuals younger than 18 years, and individuals with psychiatric disorders, depression, or seizure disorders or compromised CNS function were excluded. Persons with history or presence of autoimmune disease, with serious infections within previous 28 days, and with severe systemic disease (such as cardiac disease, severe liver disease, severe renal disease, or myeloid dysfunction) were also excluded. Prior immunotherapy, chemotherapy, or therapy with an investigational drug within 3 months of study initiation was not permitted. Presence of neoplastic disease within the previous 5 years was an exclusion criterion, except for basal cell carcinoma of the skin, actinic keratoses, and carcinoma-in-situ of the skin or cervix, provided that these had been cured by surgery.

Randomization

Patients were centrally randomized by the Department of Medical Statistics at the University of Vienna. Major inclusion criteria were checked before randomization. A biased coin randomization was applied to balance treatment groups for sex, stage (IIA *v* IIB), age (≤ 50 *v* > 50 years), weight (≤ 73 *v* > 73 kg) and study center. The allocation probabilities to the two treatments were calculated on the basis of an imbalance score defined as a weighted sum of the differences of the treatment allocation numbers in the strata defined by the five risk factors. Unconstrained randomization was used in the beginning.[38]

Blinded medication was prepared by the galenic division of Hoffmann-La Roche, Basel, Switzerland, and stored at Roche (where there was no access to the code). The randomization process generated a patch number of the patient's blinded medication, which was printed and faxed to Roche for distribution to the recruiting center. In this way, all patients and investigators were blinded regarding treatment allocation. Each center received a sealed envelope containing patient number and treatment allocation, which could be opened only in case of a serious adverse event that necessitated disclose of the treatment. A total of 20 envelopes were opened before the end of the study or were missing when the study was terminated.

Compliance

IFNα was given monthly under the trade name used in the participating countries and injected by the patients themselves. Study participants were instructed to store IFNα in the refrigerator from 2°C to 8°C. Patients were supplied with oral study medication (isotretinoin or placebo) monthly, and instructed to store the medication at room temperature. Any unused capsules were returned at each visit. Compliance was calculated at the end of the trial from pill counts and was defined on the basis of the intake of blinded study medication (isotretinoin/placebo). For each patient, a compliance ratio (actual cumulative medication intake/scheduled cumulative medication intake) was calculated. In the case of missing data, the medication intake was set to zero. Patients with a compliance ratio less than 70% were excluded from the per-protocol population.

Follow-Up Evaluation

After end of treatment patients were evaluated monthly for 3 months and than every 3 months until the third year after study initiation. Afterward, visits took place every 6 months until 5 years after study initiation.

Disease-free status was maintained when there was no hint of disease progression by either clinical assessment or imaging procedures.

Statistics

The main analysis was performed for all randomized patients. Survival and disease-free survival were compared between the treatment groups with the log-rank test (SAS procedure LIFETEST; SAS Institute, Cary, NC) at the two-sided level .05. Disease-free survival was defined as the time from start of treatment to the time of progression or death, whichever occurred first.

Patients who were lost to follow-up were censored at the time of the last observation. Kaplan-Meier curves and 5-year survival rates with 95% CIs including CIs for the differences in the 5-year survival rates were calculated. In addition, a multivariate analysis was performed based on the proportional hazards model, with stratification by center and using as covariables melanoma stage, sex, age, and weight. Centers that recruited 20 or fewer patients were combined (summarized) to a group (small centers) per country. Patients from Greece and the Netherlands were combined in a single group. For the subgroup of patients for whom sentinel lymph node biopsy results were available, an additional analysis was performed using a proportional hazards model with the additional factor sentinel lymph-node-positive/negative. Differences between the treatment groups in quality of life during treatment were tested with an independent samples *t* test, comparing the area under the curve relative to the baseline value of the quality-of-life measurements on a visual analog scale. The rates of patients experiencing adverse events were compared with χ^2 tests. Missing values were imputed according to the last observation carried forward principle. In the interim analysis a significance level of .01 (two-sided) was applied. Additionally, it was laid down to stop the study for futility if the conditional power (assuming a difference of 10% in the proportion of disease-free patients after 5 years) was less than 20%. The level for the final analysis was set to .05 (two-sided).

Sample Size

A total sample size of 400 patients was planned, which would give a power of approximately 80% for detecting a 10% difference in 5-year disease-free survival rate, assuming a 75% disease-free survival rate in the placebo group, a recruitment period of 4 years, a total study period of 9 years, and exponential disease-free survival.

Collection of Data and Data Management

Regular monitoring of the investigational sites was performed. The data were stored in a relational database (Oracle) located in the Statistical Center at the Department of Medical Statistics at the University of Vienna. The database was closed on February 29, 2004.

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RESULTS

Patients

A total of 407 patients were recruited, with 206 patients randomly assigned to the isotretinoin group and 201 patients randomly assigned to the placebo group. Baseline characteristics of the patients are listed in Table 1.

The 407 patients were recruited in 20 centers as follows: 368 patients from 11 centers in Austria, 34 patients from seven centers in Hungary, three patients from one center in Greece, and two patients from one center in the Netherlands. The median number of recruited patients of the six largest centers was 40 patients, and six centers recruited three patients or fewer.

Interim Analysis

A scheduled interim analysis performed 9 months after the end of the recruitment period showed a conditional power of only 1.3%, far below the threshold 20% laid down in the protocol. Consequently, a decision was made to stop the trial immediately for futility, to examine the patients currently enrolled, and to perform a final analysis within the next 3 months.

Final Analysis

Disease-free survival. Disease progression was observed in 67 patients in the isotretinoin group and in 53 patients in the placebo group, and two patients in each group died without progression. There was no significant difference in disease-free survival between the treatment groups ($P = .25$, log-rank test, two sided). The 5-year disease-free survival rates were 55% (95% CI, 46% to 65%) in the isotretinoin group and 67% (95% CI, 59% to 75%) in the placebo group (Fig 1). The rounded difference in the 5-year disease-free survival rates was −11 percentage points (95% CI, −24% to 1%).

Table 1. Baseline Characteristics of the Patients Enrolled in the Study

Characteristic	Placebo		Isotretinoin		Total	
	No.	%	No.	%	No.	%
No. of patients	201	49.4	206	50.6	407	100
Sex						
Male	98	48.8	117	56.8	215	52.8
Female	103	51.2	89	43.2	192	47.2
Stage						
IIA	155	77.1	146	70.9	301	74.0
IIB	46	22.9	60	29.1	106	26.0
Clark level						
Not available	21	10.5	21	10.2	42	10.3
III	30	14.9	39	18.9	69	17.0
IV	139	69.2	134	65.1	273	67.1
V	11	5.5	12	5.8	23	5.7
Breslow, mm						
Not available	17	8.5	20	9.7	37	9.1
1.5-2.0	70	34.8	63	30.6	133	32.7
2.01-3.0	49	24.4	50	24.3	99	24.32
3.01-4.0	28	13.9	28	13.6	56	13.8
> 4.0	37	18.4	45	21.8	82	20.2
Localization						
Not available	19	9.5	18	8.7	37	9.1
Head and neck	13	6.5	22	10.7	35	8.6
Extremities	73	36.3	69	33.5	142	34.9
Trunk	96	47.8	97	47.1	193	47.4
Sentinel biopsy						
Not available	121	60.2	128	62.1	249	61.2
Negative	59	29.4	57	27.7	116	28.5
Positive	21	10.5	21	10.2	42	10.3
Age, years						
Mean	51.9		53.3		52.6	
SD	13.1		14.4		13.8	
Range	18-74		19-75		18-75	
Weight, kg						
Mean	76.8		77.7		77.3	
SD	15.9		14.9		15.4	
Range	46-127		47-134		46-134	

Abbreviation: SD, standard deviation.

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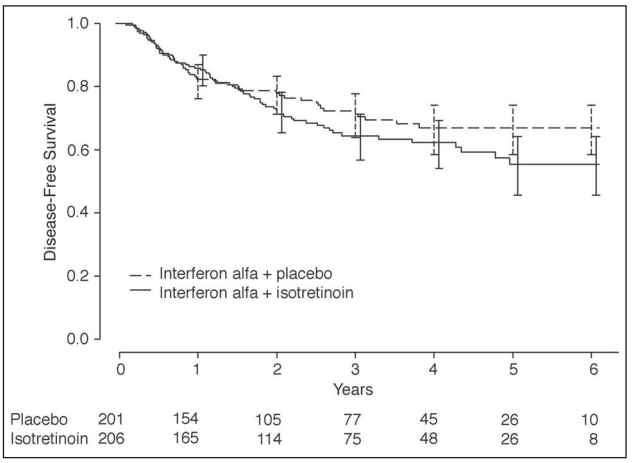

Fig 1. Kaplan-Meier estimate of disease-free survival in all 407 randomly assigned patients ($P = .25$, log-rank test, two sided). The Nos. above the time axis indicate the patients still at risk. The bars give pointwise 95% CIs.

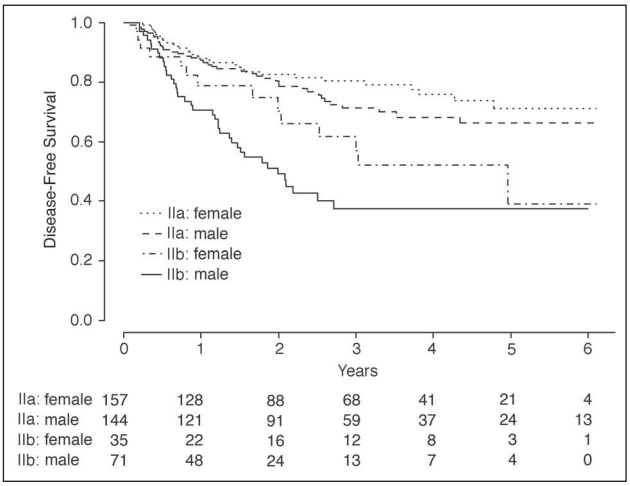

Fig 2. Kaplan-Meier estimate of disease-free survival in all 407 randomly assigned patients by stage and sex ($P = .001$, log-rank test, two sided). The Nos. above the time axis indicate the patients still at risk.

Overall survival. Survival rates did not differ significantly between the treatment groups ($P = .8$). During the study period, a total of 57 patients (30 in the isotretinoin group and 27 in the placebo group) died, 53 of these patients (28 in the isotretinoin group and 25 in the placebo group) from disseminated melanoma. The 5-year survival rates were 76% in the isotretinoin group (95% CI, 67% to 84%) and 81% in the placebo group (95% CI 74% to 88%). The difference in the 5-year survival rates was −5 percentage points (95% CI, −16% to 6%).

Prognostic factors. In the multivariate analysis, the only significant prognostic factors were stage ($P < .0001$; hazard ratio for stages IIB/IIA = 2.33; 95% CI, 1.60 to 3.39) and sex ($P = .027$; hazard ratio for male/female = 1.62; 95% CI, 1.06 to 2.50). The multivariate analysis showed no significant differences between the treatment groups ($P = .81$; hazard ratio for isotretinoin/placebo = 1.05; 95% CI, 0.72 to 1.52).

Figure 2 shows the Kaplan-Meier estimates by stage (IIA *v* IIB) and sex. Clearly there was a worse prognosis for stage IIB. Sex seemed to be an independent prognostic factor, with men showing an inferior outcome compared with women.

The analysis for the 158 patients for whom sentinel lymph node biopsy results were available showed no significant differences between treatment groups ($P = .39$; hazard ratio for isotretinoin/placebo = 1.41; 95% CI, 0.65 to 3.08). Stage was again a significant prognostic factor ($P = .02$; hazard ratio for stage IIB/IIA = 2.52; 95% CI, 1.16 to 5.48), and significantly worse prognosis was seen for patients with positive sentinel lymph node ($P = .02$; hazard ratio for sentinel lymph node–positive/negative = 2.31; 95% CI, 1.15 to 4.67).

Per-Protocol Analysis

As specified in the analysis plan, the data were also analyzed in the per protocol population. This analysis confirms the findings from the all randomized patient population. Reasons for exclusion from the per-protocol population were laid down in a blinded review. A total of 173 patients (isotretinoin group, n = 90; placebo group, n = 83) were excluded from the per-protocol population, with no substantial difference in the exclusion pattern under isotretinoin and placebo. A detailed flow chart of the reasons of exclusion from the per-protocol population is shown in Figure 3.

In the per-protocol analysis with the remaining 234 patients, no significant differences were detected in either disease-free survival or overall survival ($P = .61$ and $P = .25$, respectively, log-rank test, two sided). The 5-year disease-free survival rates were 65% (95% CI, 54% to 76%) in the isotretinoin group and 71% (95% CI, 62% to 80%) in the placebo group. The difference in the 5-year disease-free survival rates was −6 percentage points (95% CI, −20% to 8%).

The 5-year survival rates were 83% (95% CI, 74% to 92%) in the isotretinoin group and 77% (95% CI, 66% to 88%) in the placebo group. The difference in the 5-year survival rates was 7 percentage points (95% CI, −8% to 21%).

Adverse Events

A higher number of adverse events were reported in the isotretinoin group (889 events) than in the placebo group (740 events). This difference was due to higher incidences in the isotretinoin group versus the placebo group of both cheilitis/xerosis (175 *v* 44 events, respectively) and hyperlipidemia (56 *v* 34 events, respectively). 84% percent of the patients in the placebo group and 91% of the patients in the isotretinoin group observed at least one adverse event ($P = .03$).

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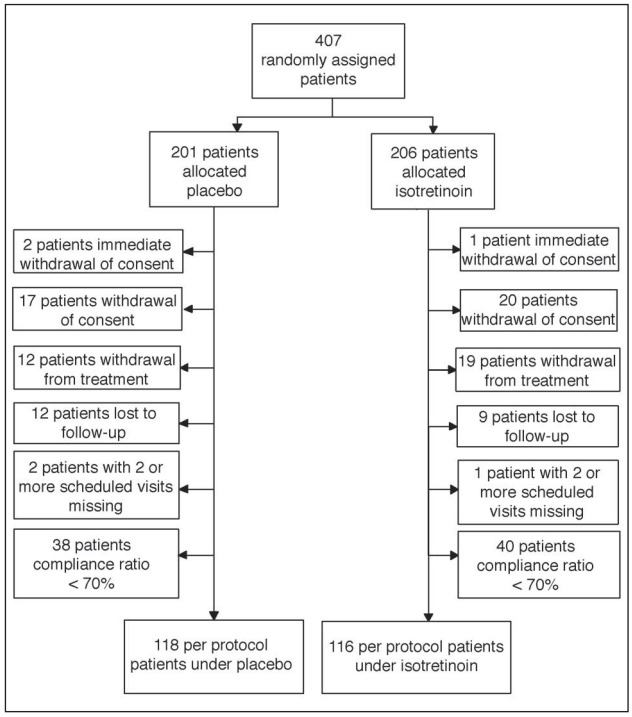

Fig 3. Flow chart showing reasons of exclusion from the per-protocol analysis.

Table 2. Adverse Events Assessed As WHO Grade 3 or 4

Adverse Event (WHO grade 3 or 4)	No. of Events: Placebo Group (n = 44)	No. of Events: Isotretinoin Group (n = 45)
WHO grade 4		
Total	2	1
Ischemic brain stroke	1	0
Dyspnea	1	0
Depression	0	1
WHO grade 3		
General disorders (total)	17	10
Flu-like symptoms	8	8
Tooth pain	4	0
Sleeping disorders	3	0
Others (taste alteration, hearing loss, weight loss, hair loss)	2	2
Skin and tissue disorders (total)	4	13
Cheilitis and xerosis	2	12
Others (exanthema, aggravation of psoriasis)	2	1
Laboratory disorders (total)	10	17
Leucopenia	1	3
Elevation of liver enzymes	3	0
Hyperlipidemia	4	12
Others (anemia, thrombopenia, increase of creatinin phosphokinase)	2	2
Inflammation and infection (total)	10	7
Inflammation	3	4
Infection	6	3
Others (polyarthritis)	1	0
Gastrointestinal and renal disorders (total)	5	2
Pancreatitis, diarrhea, cholecystolithiasis, nephrolithiasis, gastritis	5	2
Hormonal disorders, total	10	3
Menstrual disorders	9	1
Others (impotence, hypothyreosis)	1	2
Neurological disorders, total	13	9
Depression	1	3
Migraine	10	3
Others (paresis, neuritis, Grand Mal epilepsy)	2	3
Cardiac disorders, total	5	2
Angina pectoris	4	1
Other (hypertension)	1	1
Circulatory disorders, total	4	2
Thrombosis	2	2
Other (bleeding)	2	0
Musculoskeletal disorders, total	2	1
Hernia inguinalis, herniated disk	1	0
Ophthalmologic disorders, total	0	1
Vitreous floaters	0	1
Total	80	67

Table 2 lists serious adverse events (WHO grades 3 or 4) reported in the study. Twenty-one percent of the patients in both the placebo and the isotretinoin group experienced at least one adverse event of grade 3 or 4 (P close to 1.00). Three life-threatening adverse events (WHO grade 4) were observed during the study. In the placebo group, one patient experienced an ischemic brain stroke after 1 month of treatment, and one patient experienced dyspnea at the start of treatment. In both patients treatment was stopped immediately, and both patients showed complete recovery. In the isotretinoin group, one patient experienced severe depression after 6 months of treatment. Treatment was continued along with antidepressant medication and the patient recovered. Similar numbers of WHO grade 3 serious adverse events were reported in the isotretinoin group (67 events) and in the placebo group (80 events).

Minimal to moderate adverse events (WHO grades 1 to 2), including laboratory disorders, general disorders, skin and tissue disorders, infection, inflammation, gastrointestinal disorders, hormonal disorders, neurologic disorders, cardiac disorders, circulatory disorders, and ophthalmologic disorders were seen in 821 reports in the isotretinoin group and in 658 reports in the placebo group. All disorders were distributed equally within both groups with the exception of cheilitis/xerosis and hyperlipidemia more often seen in the isotretinoin group.

One patient became pregnant during the study. Treatment was immediately stopped, and the treatment assignment was unblinded by the center, revealing that the patient had been in the placebo group. This patient had a spontaneous abortion after 4 months. Another patient, this time in the isotretinoin group, became pregnant 6 months after treatment cessation and delivered a healthy boy.

Four patients (two in each group) died of causes unrelated to melanoma. One patient in the isotretinoin group

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died of cardiac decompensation 33 months after the end of treatment. A second patient in the isotretinoin group died of myocardial infarction 13 months after start of treatment. In the placebo group, one patient died in a car accident and one patient died of cholangiocellular carcinoma. None of the four deaths were considered drug-related.

Quality of Life

Quality-of-life measurements at baseline, 6 months, and 2 years were available for 68%, 63%, and 31% of the patients with only marginal differences between treatment groups. The area under the baseline values for the visual analog scale to evaluate quality of life showed no significant difference between treatment groups ($P = .94$, two-sided t test).

Secondary Malignancies

Secondary malignancies reported in the isotretinoin group (five patients) included secondary melanoma (three patients), basal cell carcinoma (one patient), and gastric cancer (one patient). Secondary malignancies reported in the placebo group (10 patients) included basal cell carcinoma (four patients), squamous cell carcinoma (one patient), breast cancer (one patient), cholangiocellular carcinoma (one patient), mixed carcinoma (one patient), multiple myeloma (one patient), and cervical dysplasia (one patient).

DISCUSSION

The optimal care for melanoma patients with moderate and high risk to develop metastasis (stage IIA and IIB AJCC/UICC 1988)[37] and for patients with micrometastasis in the sentinel lymph node (stage IIIA AJCC)[5] has been a matter of debate.[6,10,11,14,39,40] IFNα is the drug most frequently prescribed for adjuvant treatment of melanoma, but currently there is no consensus on the optimal regimen. Prolongation of overall survival in patients with moderate and high risk has been demonstrated in studies evaluating high-dose IFNα treatment.[6,12-14,41] However, although high-dose IFNα treatment regimens are widely used in the United States and, according to some, should be the gold standard of adjuvant therapy,[4,40] the high cost of high-dose IFNα treatment, the toxicity profile (leading in some cases to the death of disease-free patients),[42,43] and conflicting efficacy results have created a dilemma for dermatologists and oncologists practicing in Europe. Consequently, low- and intermediate-dose IFNα regimens are still under consideration in Europe.[14] Some studies evaluating treatment of this patient population with low-dose IFNα showed prolongation of disease-free survival,[7-9] but others did not.[13,14,44] The question "high-dose, low-dose, no dose, which dose?" for the adjuvant treatment[39] has not yet been resolved, although low-dose IFNα at a dose of 3 MU 3 times a week is registered for adjuvant treatment in the participating countries.

The aim of our study was not to attempt to resolve the issue of optimal IFNα dosage, but to look carefully at the question of whether the addition of another active antitumor agent to an adjuvant low-dose IFNα regimen widely used in Europe could significantly improve both disease-free survival and overall survival in patients with moderate to high risk of developing metastatic disease. We chose to add isotretinoin, a derivate of retinoic acid, with sebosuppressive, anti-inflammatory, cell differentiation, and immunomodulatory functions, which has been shown to be effective in the treatment of severe acne and other inflammatory diseases.[16,18] In both laboratory studies and clinical trials, retinoids have been shown to have antineoplastic activity, inhibiting growth of tumor cells directly or by induction of differentiation, inhibiting cell invasion of melanoma cells, influencing the expression of tumor cell surface markers, and inducing apoptosis in leukemic and solid tumor cells.[16-20,45]

Our patient population consisted of patients in stage IIA and stage IIB, based on the definition of localized melanoma with tumor thickness from 1.51 to 4.0 mm or Clark level IV (IIA) or localized melanoma with tumor thickness greater than 4.0 mm or Clark level V (IIB) without nodal metastases (AJCC/UICC 1988).[37] A total of 301 patients (74%) were in stage IIA and 106 patients (26%) were in stage IIB, a percentage also comparable to both other large studies in those stages of the Austrian and the French groups.[7,8] During the course of the study, the definition was modified to include also patients with micrometastasis in the sentinel lymph node and the study protocol was amended to permit the enrollment of those patients. Similar numbers of men and women were enrolled onto the study, with a slight predominance of men (53%), as also seen in other studies.[7,8]

Patients with stage IIA to IIB disease have an estimated 5-year disease-free survival rate between 45% and 50% without any treatment and between 57% and 66% with low-dose IFN treatment.[7,8] Disease-free survival of our study population overall was within the expected range at 61% (95% CI, 55% to 67%), with no significant difference between the isotretinoin and placebo groups (55% and 67%, respectively; $P = .25$). Similarly, estimated 5-year survival of our study population was within the expected range and was equivalent in the two treatment groups (76%, isotretinoin group; 81%, placebo group; $P = .8$). The lack of efficacy of isotretinoin in this regimen might reflect the heterogeneous response of melanoma cells to isotretinoin treatment, as shown in variable response rates of melanoma cell lines in laboratory studies[24,25] and in conflicting efficacy results seen in clinical trials in patients with metastatic melanoma.[27,30,32,33,35,36] Response proportion and survival was also not improved in a study in patients with advanced renal cell carcinoma.[46]

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Sentinel node positivity is found in up to 30% of patients with stage IIA and IIB melanoma.[47-49] In our study population, sentinel positivity was detected in 42 (27%) of 158 patients for whom sentinel node biopsy results were available. The multivariate analysis accounting for positive sentinel biopsy confirmed the overall negative study result. Consistent with results reported earlier,[50,51] our analysis showed melanoma stage and patient sex to be the only significant prognostic factors. In the subgroup where sentinel node biopsy results were available, sentinel node positivity was an independent negative prognostic factor for disease-free survival.

A limitation of the study is that disease-free survival instead of overall survival has been used as the primary end point. However, when looking at our data, we see that from 120 patients with progression as defined in the study, approximately 44% died in the observation period. Of the 287 patients who had no progression, only four patients died. Hence, our data show a close relationship between progression and survival.

In conclusion, in our study, the addition of isotretinoin to a 2-year low-dose IFNα treatment did not have any impact on either disease-free survival or on overall survival in patients with malignant melanoma stage IIA and IIB AJCC/UICC 1988.

Acknowledgment

We thank Barbara J. Rutledge, PhD, for editing assistance.

Appendix

The Appendix is included in the full-text version of this article, available online at www.jco.org. It is not included in the PDF (via Adobe® Acrobat Reader®) version.

Authors' Disclosures of Potential Conflicts of Interest

Although all authors completed the disclosure declaration, the following author or immediate family members indicated a financial interest. No conflict exists for drugs or devices used in a study if they are not being evaluated as part of the investigation. For a detailed description of the disclosure categories, or for more information about ASCO's conflict of interest policy, please refer to the Author Disclosure Declaration and the Disclosures of Potential Conflicts of Interest section in Information for Contributors.

Authors	Employment	Leadership	Consultant	Stock	Honoraria	Research Funds	Testimony	Other
Peter Pilarski	Roche Austria GmbH (N/R)							
Dollar Amount Codes	(A) < $10,000	(B) $10,000-99,999	(C) ≥ $100,000	(N/R) Not Required				

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