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REVIEW

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Current state of treatment for primary cutaneous melanoma

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Abstract The incidence of malignant melanoma has been rising steadily for the last 30 years. Through physician and patient education, surveillance of high-risk individuals, and biopsy of any suspicious lesions, more lesions are being diagnosed earlier, where there is a high cure rate. Unfortunately many patients will still present with thicker lesions or nodal involvement, which carries a significantly worse prognosis. Over the past decade, there have been several changes in the management of primary cutaneous melanoma. These have stemmed from novel surgical approaches, a new understanding of melanoma biology, and randomized clinical trials designed to improve outcome and decrease the morbidity of therapy. This article will review the clinical evidence behind the current treatment recommendations for primary cutaneous melanoma as well as some of the emerging data on innovative immunologic-approaches to melanoma treatment.

Key words Melanoma • Interferon • Sentinel lymph node biopsy • Vaccines

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Introduction

Melanoma is an increasingly prevalent disease, affecting tens of thousands of people of all ages. In 2004, it is estimated that there will be 55,100 new cases of melanoma and 7,910 deaths due to melanoma in the United States [1]. This will make melanoma the fifth most-common cancer in males and the seventh most-common cancer in females. Routine surveillance of high-risk individuals and biopsy of any suspicious lesion is critical for diagnosing melanoma at an early stage, as patients presenting with thicker lesions or regional nodal metastases have a significantly poorer prognosis. Once melanoma has spread to the lymph nodes, the survival drops off precipitously. Only 49% of all patients with nodal metastases survive 5 years (37% at 10 years), although the range is large: from 13% at 5 years for patients with the highest combination of risk factors (ulceration, high regional lymph node burden) to 69% at 5 years for the lowest combination of predictive factors [2]. This paper looks at the currently available treatment options for patients with primary cutaneous melanoma.

Treatment of the primary lesion

William Norris first described the concept of wide local excision of primary melanoma in 1857, and this remains the mainstay of therapy for localized melanoma nearly 150 years later [3]. The excision of the primary tumor with an adequate margin of normal-appearing skin down to the underlying fascia is curative for localized cutaneous melanoma. Limited excisions, such as excisional biopsies, are associated with local recurrence rates in the range of 30%–60% [4]. In 1907, Handley recommended a 2.5-cm margin after microscopic examination of strips of cutaneous melanoma and surrounding tissue from autopsy [5].

The recommended surgical margin was later increased to 5 cm to prevent local recurrence and to include possible microsatellites immediately adjacent to the excision site [6, 7]. As the incidence of melanoma increased, there was an emergence of interest in clinical trials to provide scientific data to support these recommendations.

Three prospective randomized studies compared these wide excisions (4- or 5-cm margins) with a more conservative 2-cm margin of resection. A multi-institutional prospective randomized trial from France compared a 5-cm margin with a 2-cm margin in 319 patients with melanomas ≤2 mm thick. There was no difference in local recurrence rate or survival [8]. The Intergroup Melanoma Committee conducted a randomized prospective study evaluating 2-cm versus 4-cm margins in 468 patients with intermediate thickness melanomas (1–4 mm) [9]. Again, there was no statistically significant difference in the local recurrence rate. Of note, there was a statistically significant difference in the need for skin grafts, with 46% of the 4-cm group requiring skin grafts and only 11% of the 2-cm group. More recently, the Intergroup Melanoma Surgical Trial has published their long-term 10-year follow-up results. The 10-year survival rates were not significantly different when comparing 2-cm versus 4-cm margins of excision [10]. Finally, the Swedish Melanoma Study Group compared 2-cm versus 5-cm margins of excision in 989 patients with melanomas between 0.8 and 2 mm thick. There were local recurrences in 1% of patients, equally distributed between the two groups, and there were no differences in recurrence-free or overall survival between the two study arms [11]. These data strongly demonstrate that a 2-cm margin for intermediate-thickness melanomas (1–4 mm) is not only safe and sufficient, but also significantly decreases the need for the extra expense and morbidity associated with a skin graft.

What about margins less than 2 cm? The World Health Organization (WHO) Melanoma Group prospectively randomized 612 patients with melanomas less than 2 mm thick to receive excision with either 1-cm (narrow) or 3-cm (wide) margins [12]. There were no local recurrences among patients with melanomas thinner than 1 mm, regardless of the excision margin. Of interest was the 2.7% risk of local recurrence in patients with melanomas 1.1–2 mm thick with 1-cm resection margins, whereas no local recurrences were seen in the same group of patients who received 3-cm margins. There was no difference between the two groups in the disease-free and overall survival rates after a median follow-up of 55 months. Updated data after 15 years again showed no difference in overall survival and disease-free survival for the two study arms [8]. The WHO Melanoma Group trial clearly demonstrated that a narrow excision margin for thin (i.e., less than 1 mm) melanomas is safe and provides excellent local control.

Management of the regional lymph nodes

The regional lymph nodes are the most-common site of metastases and the likelihood of finding metastatic disease is highly dependent on the depth of the primary lesion. In patients with no palpable nodal disease, primary lesions less than 1 mm thick have a <10% likelihood of having nodal metastases. This number increases to 20% for lesions 1.01–2.00 mm thick, 33% for lesions 2.01–4.0 mm thick, and 40% for lesions >4.0 mm thick [13].

Approximately 5% of patients present with clinically apparent regional lymph node involvement at the time of diagnosis [14]. Any palpable nodes that are enlarged (generally ≥1–1.5 cm in maximum diameter) or very hard or fixed to adjacent structures must be considered suspicious for metastatic involvement. Metastatic nodal involvement can usually be verified with a fine needle aspiration biopsy. Patients with biopsy proven palpable nodal involvement should undergo wide local excision of the primary tumor and complete lymph node dissection (LND), as surgical excision may be curative. For patients with gross axillary disease, the axillary lymphadenectomy should include levels I, II, and III nodes to provide the best regional control. For patients with inguinal disease, the extent of lymphadenectomy is more controversial, given the high rate of complications involved with deep inguinal LND. In one study, deep inguinal lymph nodes were involved in 43% of patients with palpable inguinal nodes, advocating complete superficial and deep inguinal LND in these patients [15]. Others reserve deep inguinal node dissection for the patients with a positive Cloquet's node or multiple involved nodes.

Given the high incidence of metastasis to the regional lymph nodes in patients without palpable disease, it is logical to believe that prophylactic or elective lymphadenectomy may be beneficial with clinically negative nodes. Since Snow first recommended complete lymph node dissection in patients without nodal disease in 1892 [3], advocates have claimed that resection of occult metastases in the regional nodes could prevent disseminated disease and therefore lead to improved disease-free survival as well as overall survival. A retrospective review compared 10-year survival statistics for patients with localized melanomas (stage I and II) who underwent wide excision alone with those who underwent wide excision plus elective lymph node dissection (ELND) [16]. Patients with intermediate-thickness melanomas (0.76–4 mm) who underwent wide excision plus ELND had a significantly higher survival rate than those who had wide excision alone, even after the analysis was stratified for tumor sites. In contrast, there was no survival benefit for ELND in patients with either thin (≤0.75 mm) or thick (≥4 mm) lesions. Similarly, a second retrospective review in 1988 again suggested a survival advantage for ELND compared with clinical staging

with subsequent therapeutic dissection at the time of nodal recurrence [17].

Critics counter that an ELND exposes many node-negative patients to the morbidity of a LND. Any number of unaccounted variables may have played a role in the choice between ELND and therapeutic LND, thus calling the conclusions of the retrospective data into question [18]. In contrast to the data derived from retrospective reviews, a number of prospective studies addressing ELND have found no survival benefit for patients treated with ELND [19, 20]. A small study conducted by the Mayo Clinic found no disease-free or overall survival advantage for ELND [20]. The WHO Melanoma Group randomized two groups of patients to receive either wide excision plus ELND (n=267) or wide excision with subsequent therapeutic lymphadenectomy if clinically indicated (n=286) [19]. Analysis of these data revealed no difference in survival between the two groups. With follow-up now at greater than 20 years, the WHO Trial 1 comparing wide local excision alone versus wide local excision combined with ELND still shows no statistical improvement in either survival or disease-free interval [8]. At 8 years of follow-up, the WHO Trial 14 which compared excision only with excision plus ELND in patients with melanoma of the trunk showed a borderline difference in survival, benefiting patients undergoing ELND [8, 21].

The largest trial to examine the issue was the Intergroup Melanoma Surgical Program, randomizing 740 stage I and II melanoma patients to ELND or observation [22]. Overall, there was no significant difference between the two groups. However, a significant improvement in survival with ELND was seen in certain subsets, including patients younger than 60 years or patients with non-ulcerated primaries. Long-term results confirmed no significant 10-year survival difference between ELND or observation (77% vs. 73%, P=0.12) [23]. However, a significant reduction in mortality with ELND was seen for patients with non-ulcerated melanomas, tumors between 1.0 and 2.0 mm, and limb melanomas. While it is possible that ELND benefits a subset of melanoma patients, it does not appear to improve survival for the majority of patients with clinically negative nodes.

The management of clinically node-negative melanoma patients changed considerably with the advent of the sentinel lymph node biopsy (SLNB), a technique based on the anatomical concept that lymphatic fluid from defined regions of skin drains specifically to an initial node or nodes ("sentinel nodes") prior to disseminating to other nodes in the same or nearby basins. Morton et al. [24] described this anatomical concept and described a reliable method for surgical identification and removal of the sentinel node draining the site of a cutaneous melanoma. More importantly, this same group showed that the pathological status of the sentinel node accurately determined whether melanoma cells have metastasized to that specific lymph node basin [25]. Other institutions have recapitulated these results, confirming that melanoma patients with pathologically negative sentinel nodes have detectable metastases in non-sentinel nodes less than 5% of the time [26, 27]. Regional recurrence after sentinel node biopsy is infrequent [28, 29].

An important aspect of SLNB is the ability to perform a more-detailed histological examination of the sentinel lymph nodes. Identification of micrometastases in sentinel nodes is enhanced by careful sectioning of the node (step-sectioning) as well as the use of immunohistochemical staining with anti-S-100, anti-MART-1, or HMB-45 (anti-gp100) antibodies [30]. Even microscopic foci of melanoma detected only by immunohistochemical staining are clinically significant. With this increased sensitivity, sentinel lymph node status is the most-important predictor of survival for patients with melanoma. Patients with a negative sentinel node are over 6 times more likely to survive than those with a positive sentinel lymph node, making the predictive impact of sentinel node status much greater than any other prognostic factor [31].

Given the demonstrated prognostic power of SLNB, elucidating the indications for the use of SLNB becomes central. Several clinical factors correlate with the likelihood of lymph node involvement and thus influence the decision regarding SLNB. Cascinelli et al. [32] reported sentinel lymph node positivity rates of 16% in lesions thicker than 1.0 mm. Among the 829 patients in a WHO study, positivity rates of 2% (<1.0 mm), 7% (1.0–1.99 mm), 13% (2.0–2.99 mm), and 31% (≥3.0 mm) were reported. In addition to tumor thickness, other factors such as tumor ulceration, young age, and mitotic rate have been shown to be associated with sentinel lymph node positivity [31, 33, 34]. Based on these data, as well as additional corroborating studies, the SLNB procedure should be routinely considered for primary melanomas deeper than 1.0 mm and selectively applied for tumors 1.0 mm or less, when other worrisome features are present.

SLNB plays a central role in staging the regional lymph nodes and is the standard of care in many major melanoma centers [32]. With the widespread use of SLNB, the range of survival rates among various subgroups of pathological stage III (node-positive) patients is enormous, because of "upstaging" based on a direct examination of the sentinel lymph nodes by histopathological examination [35]. Furthermore, melanoma patients who were clinically staged compared with those whose nodal disease was staged pathologically differ significantly in their survival rates [36]. Currently, the compelling prognostic value of knowing the nodal status makes SLNB indispensable for accurate staging, and thus a key component of future studies examining adjuvant therapy.

Patients who have a positive sentinel lymph node should undergo a completion LND [37]. This "selective" approach to node dissection spares patients with negative nodes the morbidity of the procedure, while offering

improved regional control and whatever survival benefit there may be from elective dissection to the node-positive patient. Additional positive non-sentinel lymph nodes (NSN) will be found in 7%–33% of patients with a positive sentinel node [24, 27, 31, 38–42], but predicting which patients will have residual disease in the NSN is not currently possible [40, 43]. Even patients with the most-favorable primary melanomas have a substantial risk of additional disease in the basin. While still unproven, it is possible that early complete node dissection may impact survival in patients with microscopic disease in the sentinel node. Although not a direct analogy to the sentinel node biopsy situation, when the survival of patients in the WHO Program 14 Trial with microscopic disease at ELND was compared with those who had regional recurrences during observation, the survival was significantly improved in the former group (48.2% vs. 26.6%, P=0.04) [21, 43]. The Multicenter Selective Lymphadenectomy Trial I has recently completed accrual, and will hopefully define the therapeutic benefit of SLNB and selective LND [44].

The role of adjuvant radiation therapy to the nodal basin after resection of regionally advanced melanoma has not been clearly defined, although there is a growing body of literature to support its use [45]. There is a high risk of regional failure-up to 50%-after therapeutic cervical LND, especially in the presence of large-sized nodes, multiple involved nodes, or extracapsular extension [45]. Several non-randomized studies have suggested that postoperative radiation to the neck or axilla after radical LND decreases regional recurrence rates in node-positive patients [45–48]. Only one small randomized trial of adjuvant radiotherapy has been completed in cutaneous melanoma, and this yielded a negative result [49]. This study utilized conventional radiation fractions and a treatment break, both currently felt to be suboptimal for melanoma. The Radiation Therapy Oncology Group initiated a randomized trial of postoperative radiation using larger treatment fractions in patients undergoing neck dissections for melanoma. Accrual to this trial was poor, however, and the trial ultimately failed to meet its accrual goal [50]. Another randomized trial is presently underway in Australia. Until larger randomized trials are conducted, it is reasonable to consider postoperative radiation therapy in patients with gross extracapsular extension or multiple involved lymph nodes.

Adjuvant therapy for high-risk melanoma

The poor prognosis once melanoma has metastasized has generated significant interest in reducing recurrence rates through adjuvant therapies. Many of these trials were performed prior to a full understanding of recognized prognostic factors, and hence risked potential imbalances between the treatment and control arms. Compounding these problems were small sample sizes and the inclusion of heterogeneous groups of at-risk patients in many of the adjuvant trials conducted to date. It is important to bear in mind that these methodological problems make comparisons of trials difficult, even when the trials are purporting to study similar interventions, and they preclude lending serious weight to the results of subset analyses. A large variety of different interventions have been tested in the randomized trials conducted to date.

Retinoids and hormones

The use of oral vitamin A for the adjuvant therapy of melanoma in humans was examined in a randomized, controlled trial conducted by the Southwest Oncology Group (SWOG) beginning in 1981, based on preclinical studies in the 1970s demonstrating the efficacy of retinoids against melanoma both in vitro and in vivo. SWOG 8049 investigated the use of oral vitamin A in 240 eligible melanoma patients with tumors ≥0.75 mm in depth and clinically negative lymph nodes, and found no disease-free or overall survival benefit [51]. Megestrol acetate (Megace), a progesterone analogue, has also been studied, based on evidence that the hormonal milieu may play a role in the clinical course of melanoma. A small randomized clinical trial of 67 eligible patients showed a survival benefit for megestrol [52]. This study prompted the performance of a larger randomized, double-blind, placebo-controlled phase III trial of megestrol in the treatment of high-risk melanoma in the adjuvant setting. In this study of 262 eligible patients, no relapse-free or overall survival benefit was seen in patients receiving megestrol versus placebo [53].

Cytotoxic chemotherapy

Despite multiple trials involving a variety of cytotoxic drugs available to clinical investigators throughout the years, no confirmed studies have demonstrated a benefit of adjuvant chemotherapy in melanoma patients at high risk for relapse. Single agents with recognized but minimal activity against advanced disease include dacarbazine (DTIC), the nitrosoureas, the vinca alkaloids, cisplatin, paclitaxel, and bleomycin. Although DTIC results in objective responses in up to 18%–22% of patients with metastatic disease, numerous studies have failed to show a benefit as postsurgical adjuvant [8, 54–56]. Multiagent cytotoxic therapy has similarly been unfruitful in the adjuvant setting. Karakousis and Emrich [57] found no benefit for the combination of DTIC and estramustine in a small, three-arm study comparing multiagent chemotherapy or BCG with a no-treatment control group. Four two-arm, randomized

controlled trials of multiagent chemotherapy have been performed. A trial using BCNU, dactinomycin, and vincristine [58] and a small trial of DTIC, CCNU, and vincristine [59] both suggested a benefit for multiagent chemotherapy, while two other trials did not [60, 61]. Several small trials have also explored postoperative adjuvant therapy with DTIC plus BCG, with [55, 62] or without a no-treatment arm [63–67]. DTIC plus BCG does not appear to have any efficacy in the adjuvant setting, which is hardly surprising in view of the lack of activity of the individual agents and the absence of any suggestion of synergy between them. Tamoxifen and cisplatin has been shown to exhibit cytotoxic synergy in vitro. An initial phase II trial treating 153 patients with four cycles of tamoxifen and cisplatin for 28 days revealed a disease-free survival of 68.4% and an overall survival of 84.5% at 36 months follow-up. The authors argue that the projected 5-year disease-free survival and overall survival were comparable to the results with interferon in the ECOG 1684 trial, and therefore warranted further investigation of cisplatin and tamoxifen [68]. A second phase II study utilizing higher doses of tamoxifen and weekly cisplatin, however, demonstrated only modest activity (32% overall response rate) with only 1 patient achieving a complete remission that lasted 22 months. Furthermore, while the combination of tamoxifen and cisplatin demonstrated some activity, the toxicity was substantial and the authors did not recommend its clinical use at the higher dosages used [69]. Until and unless randomized trials in advanced disease confirm the efficacy of cytotoxic chemotherapy, adjuvant systemic therapy should be considered only in the context of a clinical trial.

Immunostimulants

Bacillus Calmette-Guerin (BCG) is a mycobacterial agent that was shown by Morton in 1970 to elicit an antitumor response when injected into cutaneous metastatic melanoma nodules [70]. Interestingly, regression of non-injected tumor nodules was also noted in some patients receiving this treatment, suggesting that this effect was immune mediated [70, 71]. Based on these results, several randomized studies of BCG therapy versus observation were conducted in patients with resected primary melanoma greater than 1.5 mm in thickness. None demonstrated a significant survival advantage to BCG therapy in this setting [72]. The only study to demonstrate a disease-free survival advantage of BCG therapy over observation was a small trial of 20 eligible patients with stage III disease [73].

BCG monotherapy was also compared with DTIC and the combination DTIC and BCG [64]. Combination therapy was found to be better than either modality alone, but BCG therapy was not found to have efficacy greater than DTIC alone. As there was no observation or placebo arm in this trial, conclusions as to the efficacy of BCG alone could not be drawn. Considering all of the data obtained from these small trials together, there is no suggestion that BCG therapy provides any meaningful benefit in the adjuvant setting. Heat-killed *Corynebacterium parvum* is another bacterial agent that is thought to be immunostimulatory. Along the same lines as BCG, the efficacy of *C. parvum* against high risk melanoma in the adjuvant setting has been tested in several clinical trials. Studies of *C. parvum* versus observation [74] and placebo [75] have failed to demonstrate any benefit. When compared with BCG therapy, however, two studies, [76, 77] have shown *C. parvum* therapy to confer a statistically significant survival advantage, although these studies did not have a placebo arm. Although a benefit of BCG and *C. parvum* single-agent therapy over and above observation alone has not been definitively demonstrated, BCG continues to play a significant role as a control arm, as well as an immune adjuvant in combination with vaccine therapy in ongoing clinical trials.

Levamisole is an antihelminthic drug that has been found to have immunostimulatory properties. While the mechanism of immunopotentiation of this agent is not well understood, it has nevertheless been investigated as an anticancer agent.

Of the four randomized, controlled studies of levamisole in the treatment of high-risk melanoma [55, 78–80], three failed to show a statistically significant benefit from levamisole therapy as far as disease-free or overall survival. The fourth study, by Quirt et al. [78], did demonstrate a trend towards a disease-free and overall survival benefit of levamisole, although it was of borderline statistical significance. Levamisole has also been tested in combination with interleukin-2 in two randomized studies of patients with advanced melanoma, but no benefit was obtained [81, 82]. No further investigations are planned for this agent.

Transfer factor is an extract obtained from disrupted leukocytes that was first described by Lawrence in 1955 [83]. When prepared from leukocytes from an antigen-sensitized donor and then administered to a naïve recipient, transfer factor has been shown to elicit delayed-type hypersensitivity responses. Early applications of transfer factor therapy were based on the hypothesis that its mechanism of action was antigen specific. To this end, transfer factor used in preliminary investigations of its role in the treatment of melanoma was obtained from donors whose lymphocytes demonstrated reactivity to melanoma antigens (typically melanoma patients who had undergone disease regression or surgical cure, and family members and close contacts of the study subjects) [84, 85]. Based on encouraging results obtained in these early studies in patients with metastatic disease [84, 85], two small clinical trials were performed to test the efficacy of transfer factor on melanoma in the adjuvant setting [86, 87]. The material used for these studies was obtained from normal healthy donors, based on the

knowledge that transfer factor exerts both specific and non-specific effects on cellular immunity. Neither of these trials demonstrated a disease-free or overall survival benefit due to transfer factor therapy. There is currently no role for transfer factor in the adjuvant therapy of melanoma.

Granulocyte macrophage colony stimulating factor

Cytokines with roles in the growth and maturation of hematopoietic and dendritic cells, such as granulocyte/macrophage colony stimulating factor (GM-CSF), have been incompletely studied in the adjuvant setting. No prospective, randomized, multicenter trials have yet been completed with these agents to support their use in the adjuvant therapy of high-risk melanoma outside clinical investigations. In one phase II trial investigating GM-CSF as surgical adjuvant therapy in patients with stage III or IV melanoma, Spitler et al. [88] showed that GM-CSF prolonged overall and disease-free survival compared with matched historical controls. This has prompted a number of prospective intergroup trials that are currently investigating the potential function of GM-CSF. One such trial is the ECOG trial E4697, which is evaluating GM-CSF (alone or administered with vaccination against a multi-epitope peptide vaccine) among patients with resectable disease, advanced regional nodal disease with extracapsular extension, or failure after adjuvant interferon therapy [89]. Definitive recommendations regarding the use of GM-CSF in the adjuvant setting for melanoma must await the results of these trials.

High dose interferon-α2b

In 1980, interferon (INF) was shown to inhibit the growth of B16 melanoma in vitro and in vivo [90]. Since then, multiple trials involving adjuvant IFN-α have involved a wide range of dosing regimens [91]. Early trials utilized low-or intermediate-dose regimens, and while there was a suggestion of some effectiveness [92, 93], re-analysis and additional trials have demonstrated no benefit to survival [94, 95]. Although there was no overall survival advantage, the disease-free survival advantage to low-dose IFN seen in the French and Austrian trials has led to its approval in Europe for thick primary melanomas [66, 67] Unfortunately, the recently published results of the AIM-HIGH study in the United Kingdom of low-dose extended-duration IFN-α2a demonstrated no clear differences between IFN and observation in either overall or relapse-free survival for any subset of patients [98].

Kirkwood et al. [99] dramatically increased the dose of INF to the maximally tolerated dose in a randomized trial for ECOG, E1684. This regimen involved an "induction" phase of IFNα-2b 20 MU/m^2 IV 5 days a week for 4 weeks, followed by a "maintenance" phase of 10MU/m^2 Sq 3 days a week for the remainder of a year. While this regimen was more toxic, the results were positive. Patients randomized to the treatment group had a significant improvement in disease-free and overall survival compared with the control group. IFN-α2b therapy increased the median relapse-free survival by 9 months (1.72 years for IFN-α2b patients versus 0.98 years for observation patients) and produced a 42% improvement in the 5-year relapse-free survival rate [46% for IFN-α2b patients (95% confidence interval 19%–34%) versus 37% for observation patients (95% confidence interval 30% to 46%)]. Based on these results, IFN-α2b was approved by the`FDA foz the adjuvant treatment of high-risk melanoma.

In order to verify the results of E1684, as well as better define the benefit of adjuvant therapy with melanoma, an ECOG-coordinated Intergroup trial was initiated as a follow-up to E1684. E1690 compared the high-dose IFN-α2b and a 2-year low-dose IFN-α2b with to observation after complete resection of all known disease [100]. While the results of this trial confirmed the disease-free survival advantage for high-dose IFN-α seen in E1684, there was no overall survival advantage.

A third trial, Intergroup E1694, compared 1 year of high-dose IFN-α2b with 2 years of a ganglioside vaccine called GMK. Gangliosides are carbohydrate antigens found on the surface of melanoma cells, as well as normal cells of neural crest origin and tumor cells of other types. A randomized trial suggested a disease-free survival benefit in patients who were treated with GM2 plus BCG compared with those treated with BCG alone following resection of stage III disease [101]. In May 2000, the E1694 Trial's Independent Data Safety Monitoring Committee concluded that the high-dose IFN arm was associated with significantly improved relapse-free and overall survival, and mandated that the study be terminated early and the results disclosed [102].

Advocates of IFN point out that of the three high-dose trials described above, all three demonstrated improvement in relapse-free survival, and two of the three demonstrated an improvement in overall survival. In addition, advocates of adjuvant IFN therapy point out that the reason that E1690 failed to demonstrate a survival advantage is likely due to differences in eligibility criteria and, more importantly, the subsequent availability of post-relapse IFN-α2b crossover therapy in the E1690 trial compared with E1684 [100].

However, several questions still remain as to whether these trials support a role for IFN in the adjuvant setting. Despite the enthusiasm and multiple large controlled studies, there has been no demonstrable rationale of the mechanisms of action of this biological response modifier [8]. ECOG 1684, the trial that established the adjuvant use of IFN-α2b is not without concerns. Patients were not strati-

fied by the number of positive lymph nodes; a recognized prognostic factor. Therefore it is possible that there may have been an unrecognized imbalance between the treatment and control groups that influenced the outcome. In addition, longer follow-up data on ECOG 1684 show that at a median follow-up of 12.6 years there is a continued disease-free survival advantage for high-dose IFN, but the benefit in overall survival loses statistical significance [103]. A pooled analysis of updated data from E1684 and E1690 demonstrates that relapse-free survival, but not overall survival, was significantly prolonged for patients treated with high-dose IFN versus observation [103]. Therefore, while high-dose IFN clearly improves disease-free survival, the question of overall survival remains controversial [91].

The concerns about E1684, specifically the possible imbalance between the treatment and control groups, were supposed to be addressed in E1690. However, while there was an improvement in disease-free survival for high-dose IFN-α2b, the study failed to demonstrate an improvement in overall survival for either high- or low-dose IFN-α2b. The crossover data from 1690, which advocates of IFN therapy use as the reason E1690 failed to validate E1684, are from a retrospective analysis and must be considered unproven. Finally, E1694 appears to confirm the disease-free and overall survival benefit of high-dose IFN-α2b demonstrated in ECOG 1684. However, this study did not have an observation arm. Therefore, a deleterious effect of the GMK vaccine cannot be ruled out. While this appears unlikely, such an effect would lead to the appearance of a "benefit" for IFN-α2b in that trial when none existed. With so many differences between the trials, it is difficult to directly compare the results.

Given the significant side effects of high dose IFN, there is considerable interest in realizing the potential benefit of interferon without the toxicities associated with the regimen. ECOG 1697 is currently examining whether patients with stage II or stage III (one lymph node positive only) melanoma benefit from just the induction phase of high-dose IFN-α2b. EORTC study 18991 is investigating the use of low-dose pegylated IFN-α2b over 5 years in high-risk patients. EORTC 18-952 has enrolled more than 1,400 node-positive patients in a three-armed study of adjuvant intermediate dose IFN-α2b. In this study, the efficacy of two intermediate-dosing regimens of IFN-α2b, 10 MU SC 3 times a week for 4 weeks, followed by either 10 MU SC 3 times a week for 1 year, or 5 MU SC 3 times a week for 2 years are compared with observation. The Scandinavian Melanoma Cooperative Group is randomizing patients between observation or one of two intermediate dose regimens (10 MU SC 5 times a week for 1 month followed by 10 MU SC 3 times a week for 1 or 2 years). Until the results of these studies show that an intermediate dose of IFN is as effective, high-dose IFN-α2b remains the standard therapy in the adjuvant setting.

In-transit metastasis

Approximately 2%–3% of melanoma patients will develop in-transit metastasis, which is the appearance of metastasis along the path from the primary tumor to its regional nodal basin, and which is lymphatic in nature. The management of local or in-transit metastasis is dictated by the number and the size of the lesions. With isolated local recurrence that is few in number, surgical excision is the best option. Surgery consists of excision of metastases with a margin of surrounding normal cutaneous and subcutaneous tissue. When there are multiple local recurrences, excision becomes an unlikely option and the more reasonable options include intralesional therapy, hyperthermic isolated limb perfusion (ILP), and radiation therapy.

Intralesional therapy is desirable in that it does not have the systemic toxicity as many other therapeutic modalities. Local injection of multiple agents such of fotemusine [104], bleomycin [105], cisplatin [106], IL-2 [107], and IFN-α [108], has been investigated as a method of controlling cutaneous and subcutaneous metastatic melanoma. Another commonly used intralesional therapy GM-CSF, which can result in significant regression of melanoma deposits [109, 110].

Although melanoma is relatively radiation resistant, it can provide palliation in unresectable lesions in approximately two-thirds of cases [45, 111]. The response of dermal, subcutaneous, or lymph node metastases to the radiation therapy depends mainly on their size. Complete response was seen in 71% of patients with lesions less than 3 cm, but only in 20% in patients with lesions >5 cm in size [112]. Therefore, radiation therapy should be considered in those patients with smaller volume of cutaneous or subcutaneous metastases.

Hyperthermic isolated limb perfusion is a way of isolating the blood circuit to the extremity and administering chemotherapeutic agents regionally at a concentration 15–25 times higher without resulting in systemic side effects [113]. Melphalan has been used as a standard drug for ILP secondary to its efficacy and low regional toxicity [114, 115]. While this has not been shown to improve survival, the use of hyperthermic provides a significant palliation of locoregional symptoms when other options are not available. Isolated limb infusion (ILI) is a technically less-complex variation of ILP, in which a low-flow perfusion is performed via percutaneously inserted catheters, but without oxygenation [116]. The early results seem to be similar to those obtained by ILP, although further evaluation is needed before definitive recommendation can be made [117].

On the horizon – vaccine therapy

Based on evidence that the immune system plays a natural role in melanoma progression, there has been hope that the power of the immune system could be harnessed through

the use of melanoma vaccines. While multiple trials have demonstrated the ability to generate an anti-tumor immune response, to date no large, randomized trial has demonstrated an impact on survival with any melanoma vaccine. As opposed to other adjuvant therapies where we accept small gains in exchange for significant toxicity, the side effects of vaccines are limited, confined mostly to mild flu-like symptoms and reactions at the injection site. Therefore, there remains considerable interested in developing improved vaccine strategies that will make a significant clinical impact.

One common approach has been to vaccinate patients with antigens present on melanoma cells. Bystryn et al. [118] used antigens shed into culture from three human and one hamster melanoma cell lines to create a polyvalent melanoma antigen vaccine capable of generating both a humoral and cellular response in a subset of patients. Recently, this vaccine was evaluated in a randomized phase II study involving a high-risk stage III patient population, and demonstrated a statistically significant disease-free survival advantage for the vaccine arm [119]. A statistically non-significant overall survival advantage was seen in the vaccine arm. Gangliosides have also been found to be effective targets for active immunotherapy [119–122]. Gangliosides are glycosphingolipids present on melanoma cells, as well as some non-neoplastic cells, with carbohydrate moieties expressed on the cell surface available for antibody recognition [123]. Vaccinating patients with the GM2 ganglioside along with the adjuvant BCG had promising results [101], which were improved by conjugating GM2 to keyhole limpet hemocyanin (KLH) and replacing the BCG with the adjuvant QS-21 [124, 125]. In a randomized trial, this vaccine was shown to be less effective than high-dose IFN in patients with resected stage IIB-III melanoma [102]. It is currently being studied in a European (EORTC) trial in stage II melanoma patients comparing the same vaccine with observation after surgery.

Rather than using single or multiple antigens, several researchers have attempted to use entire cells to stimulate an immune response. Autologous cellular vaccines require the surgical resection of the melanoma, which, after being irradiated or lyzed, are given back to the patient along with an adjuvant to promote immune recognition. Berd et al. [126] tested a novel vaccine in which the hapten ditnitrophenyl was conjugated to proteins on autologous tumor cells in order to increase the immunogenicity. In a total 77 patients with clinically evident nodal metastases were given the vaccine with BCG, now called "M-Vax," in the adjuvant setting after lymphadenectomy. The authors reported a more favorable than expected 5-year relapse-free and overall survival rate (45% and 58%, respectively) [127, 128]. A multi center randomized controlled trial of M-Vax as adjuvant therapy for resected AJCC stage III melanoma had significant difficulty with specimen transportation, illustrating the difficulties of performing large trials with autologous vaccines. The use of autologous tumor cell vaccines is limited to individuals with palpable nodal disease or resectable metastatic disease so that sufficient tumor can be obtained to prepare a vaccine, and even then there is only enough tumor to provide a limited number of vaccinations. In addition, such patients have a poor overall prognosis and are likely to have significant residual tumor burden, making them less-than-ideal candidates for any immunotherapeutic approach.

Allogeneic cellular vaccines take advantage of the fact that melanoma-associated antigens are shared among a large number of patients. There is well-documented evidence that this type of vaccination can induce immune responses to several melanoma antigens [129, 130]. Allogeneic vaccines are readily available, even for patients who lack sufficient tumor to produce an autologous tumor cell vaccine, and can be standardized, preserved and distributed in a manner akin to any other therapeutic agent. Because of this, they are more readily available for evaluation in large prospective, randomized trials. Presently, there are two major allogeneic vaccines being evaluated as an adjuvant therapy for melanoma.

Canvaxin is an allogeneic vaccine composed of three viable irradiated melanoma cell lines, chosen for their high content of immunogenic melanoma- and tumor-associated antigens [131, 132]. This vaccine has been shown to enhance the immune response to melanoma, and this response correlates with outcome [129, 133–137]. In a phase II trial, 935 patients with AJCC stage III melanoma who underwent complete lymphadenectomy were treated with the vaccine. Compared with a historical cohort of 1,677 similar patients who did not receive the vaccine, median overall survival and 5-year overal survival were significantly higher [138]. Currently a multicenter, randomized, controlled trial is underway comparing Canvaxin plus BCG with placebo plus BCG in both stage III and stage IV melanoma status post surgical resection.

Melacine consists of a lysate of two homogenized melanoma cell lines that are combined with the adjuvant DETOX ("detoxified Freund's adjuvant," composed of monophosphoryl lipid A and a purified mycobacterial cell-wall skeleton) [139, 140]. In stage IV melanoma, vaccination with Melacine induced an objective response in 19% of patients [141], and resulted in a median survival comparable to combination chemotherapy (dacarbazine, cisplatin, BCNU, and tamoxifen) with significantly less toxicity [142, 143]. Based on these results, Melacine was approved in Canada in May 2000 for the treatment of advanced melanoma.

The Southwest Oncology Group (SWOG) completed a randomized trial comparing adjuvant Melacine with observation for patients with intermediate thickness (1.5–4.0 mm), node-negative melanoma [144]. Although there was no significant advantage of the vaccine compared with observation, it had previously been reported that melanoma

patients who expressed at least two of the following five alleles: HLA-A2, A28, B44, B45, and C3, had a significant response when given the vaccine [142]. Patients in the SWOG trial in the vaccine arm who expressed two or more of these alleles had a superior disease-free survival than the corresponding patients in the observation arm (4-year disease-free survival 87% vs. 64%, P=0.0001). The specific alleles contributing the major component of this effect were HLA-A2 and C3. A2+ and/or C3+ vaccinated patients [178 patients of 294 total vaccine arm patients (61%)] had an 82% 4-year disease-free survival (P=0.001 compared with observation arm or A2/C3- vaccine arm patients) [145]. A follow-up trial of adjuvant Melacine limited to patients with HLA A2 or C3 is presently being planned.

Conclusions

Early detection, and ultimately prevention, of melanoma remains the best way to minimize morbidity and mortality from this increasingly common form of cancer. Aggressive surgical approaches can salvage some patients with even advanced disease, while the use of sentinel node biopsy has allowed the identification and early surgical treatment of patients with occult nodal metastases. Adjuvant therapy is a logical way to impact the natural history of high-risk melanoma; to date only high-dose IFN has been shown in clinical trials to improve relapse-free survival and possibly overall survival. New approaches continue to be needed to treat metastatic melanoma and to prevent metastases from occurring.

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