See related commentary on pg 2466

Serial or Parallel Metastasis of Cutaneous Melanoma? A Study of the German Central Malignant Melanoma Registry

Maximilian Gassenmaier[1], Thomas Kurt Eigentler[1,2], Ulrike Keim[1,2], Matthias Goebeler[3], Eckhard Fiedler[4], Gerold Schuler[5], Ulrike Leiter[1,2], Benjamin Weide[1], Eva-Maria Grischke[6], Peter Martus[7] and Claus Garbe[1,2]

For more than a century the Halstedian hypothesis of contiguous metastasis from the primary tumor through the lymphatics to distant sites shaped lymph node surgery for melanoma. We challenge this dogma of serial metastatic dissemination. A single-center series of 2,299 patients with cutaneous metastatic melanoma was investigated to analyze overall survival and distant metastasis-free survival of stage IV patients with or without primary lymphatic metastasis. Results were then compared with those of 2,134 patients from three independent centers of the German Central Malignant Melanoma Registry. A multivariate binary logistic regression model was used to identify risk factors for the initial metastatic pathway. Distant metastasis-free survival (hazard ratio = 1.02; 95% confidence interval = 0.91−1.14; $P = 0.76$) and overall survival (HR = 1.09; 95% CI = 0.96−1.23; $P = 0.177$) did not differ between stage IV patients with primary hematogenous or primary lymphatic metastasis. Melanoma localization was the only significant risk factor for the initial metastatic pathway. These findings indicate that regional and distant metastases originate from the primary tumor itself in a rather parallel than serial fashion and could explain the lack of survival benefit associated with immediate complete lymph node dissection in sentinel lymph node-positive melanoma patients.

Journal of Investigative Dermatology (2017) **137,** 2570−2577; doi:10.1016/j.jid.2017.07.006

INTRODUCTION

For more than a century, lymph node surgery in breast cancer and melanoma followed the concept that regional lymph nodes are the first site of metastasis and that distant metastasis develops from metastatic lymph nodes.

In 1894 Halsted proposed a model of breast cancer progression in which the primary tumor spreads contiguously through the lymphatics to the regional lymph nodes and from there to distant sites (Halsted, 1907). Extensive surgery of the primary tumor, the lymphatics, and draining lymph nodes appeared to be necessary to control cancer. Shortly after, Handley and Pringle adopted this radical approach for the treatment of melanoma (Handley and Lond, 1907; Rebecca et al., 2012).

In 1968, Fisher contradicted Halsted's hypothesis and introduced a new concept of breast cancer metastasis, the so-called *alternative hypothesis* (Fisher, 1980; Fisher and Anderson, 2010). Fisher considered breast cancer as a systemic disease and positive lymph nodes as an indicator for distant disease rather than its instigator. He showed that tumor cells could transmigrate lymph nodes and refuted Virchow's assumption that lymph nodes are an effective barrier to cancer cell dissemination (Fisher and Fisher, 1966; Virchow and Chance, 1863). Thus, radical locoregional therapy is unlikely to improve overall survival (OS) (Fisher, 1980; Fisher and Anderson, 2010).

Several randomized landmark trials have tested the effects of complete lymph node dissection (CLND) on survival of breast cancer patients, and no improvement of survival has been observed (Fisher et al., 2002; Galimberti et al., 2013). However, treatment of stage III melanoma is still based on the Halstedian hypothesis (Balch, 1988). When melanoma patients present with solitary regional lymph node micro- or macrometastasis, it is state of the art to offer these patients CLND. CLND is supposed to prevent further metastasis and achieve local tumor control of the regional lymph node bed. Recently, the Multicenter Selective Lymphadenectomy Trial II comparing immediate CLND versus observation in patients who had melanoma with positive sentinel lymph node biopsy (SLNB) results showed that CLND did not increase melanoma-specific survival (MSS) (Faries et al., 2017) and confirmed the results of the DeCOG-SLT trial

[1]*Center for Dermatooncology, Department of Dermatology, Eberhard-Karls-University of Tuebingen, Tuebingen, Germany;* [2]*Central Malignant Melanoma Registry, Eberhard-Karls-University of Tuebingen, Tuebingen, Germany;* [3]*Department of Dermatology, Venereology and Allergology, University Hospital Wuerzburg, Wuerzburg, Germany;* [4]*Department of Dermatology and Venereology, Martin-Luther-University of Halle (Saale), Halle (Saale), Germany;* [5]*Department of Dermatology, University Hospital Erlangen, Erlangen, Germany;* [6]*Breast Cancer Center, Eberhard-Karls-University of Tuebingen, Tuebingen, Germany; and* [7]*Institute of Clinical Epidemiology and Applied Biostatistics, Eberhard-Karls-University of Tuebingen, Tuebingen, Germany*

Correspondence: Claus Garbe, Center for Dermatooncology, Department of Dermatology, University Medical Center Tuebingen, Liebermeisterstrasse 25 72076 Tuebingen, Germany. E-mail: claus.garbe@med.uni-tuebingen.de

Abbreviations: CI, confidence interval; CLND, complete lymph node dissection; DMFS, distant metastasis-free survival; HR, hazard ratio; IQR, interquartile range; MSS, melanoma-specific survival; OS, overall survival; SLNB, sentinel lymph node biopsy; TANS, thorax, upper arm, neck, scalp

Received 19 April 2017; revised 17 June 2017; accepted 12 July 2017; accepted manuscript published online 21 July 2017; corrected proof published online 14 October 2017

(Leiter et al., 2016). The consistency of these findings challenges the validity of the Halstedian hypothesis.

The starting point of this study is the observation that primary hematogenous metastasis occurs in a subgroup of melanoma patients. If cutaneous melanoma progresses in a serial fashion, we would expect that stage IV patients with initial lymphatic metastasis have longer distant metastasis-free survival (DMFS) and OS than patients with initial hematogenous metastasis. We therefore investigated survival of patients with primary hematogenous or primary lymphatic spread in a large prospectively documented cohort of melanoma patients with distant metastases, and we assessed associated risk factors.

RESULTS

Metastatic pathways of the training cohort

Median follow-up of the whole training cohort was 57 months (interquartile range [IQR] = 29–107). Median follow-up of patients who did not undergo melanoma-specific death was 73 months (IQR = 34–129). Median time until occurrence of stage III was 23 months (IQR = 10–53) after exclusion of SLNB-positive patients (n = 482), and median time until occurrence of stage IV was 35 months (IQR = 18–68). Exclusive lymphatic metastasis was associated with excellent survival (5-year MSS = 98.4%; 1.6% of patients had melanoma-specific death without documentation of stage IV disease), whereas patients with distant metastases in their further course had a 5-year MSS of only 48.5%.

Metastatic pathways are summarized in Figure 1, and patients and tumor characteristics are shown in Table 1. Initial progression to stage III occurred in 74.6% (n = 1,715) and initial progression to stage IV in 25.4% (n = 584). One half (51.4%, n = 882) of the stage III patients stayed in their disease stage, and the other half (48.6%; n = 833) progressed to stage IV (Figure 1a). Regional lymph node metastasis was found in 79.3% (n = 1,360) of the stage III patients and locoregional metastasis (satellite and in-transit metastasis) in 49.6% (n = 851). Primary hematogenous metastasis occurred in 41.2% (n = 584) and primary lymphatic in 58.8% (n = 833) of the stage IV patients.

Most patients with primary hematogenous metastasis developed distant metastasis only. Of the 584 patients with direct progression to stage IV, 491 patients were followed up until death. Of these, 63.7% (n = 313) showed no evidence of regional lymph node or locoregional metastasis, 26.9% (n = 132) had a diagnosis of simultaneous regional lymphatic and distant hematogenous metastasis, and 9.4% (n = 46) had a diagnosis of regional lymphatic metastasis after distant hematogenous metastasis.

Regarding the metastatic pattern, 38.4% (n = 882) of all patients had lymphatic metastasis only, 16.2% (n = 373) had hematogenous metastasis only, and 45.4% (n = 1,044) had combined lymphatic and hematogenous spread (Figure 1b).

Median time until lymphatic metastasis only was 24 months (IQR = 12–56). Median time until hematogenous metastasis only was 43 months (IQR = 23–74). Lymphatic metastasis of patients with combined lymphatic and hematogenous metastasis occurred after a median of 23 months (IQR = 10.00–51.75), and hematogenous metastasis occurred after a median of 37 months (IQR = 19–71). SLNB-positive patients (n = 482) were excluded from this analysis.

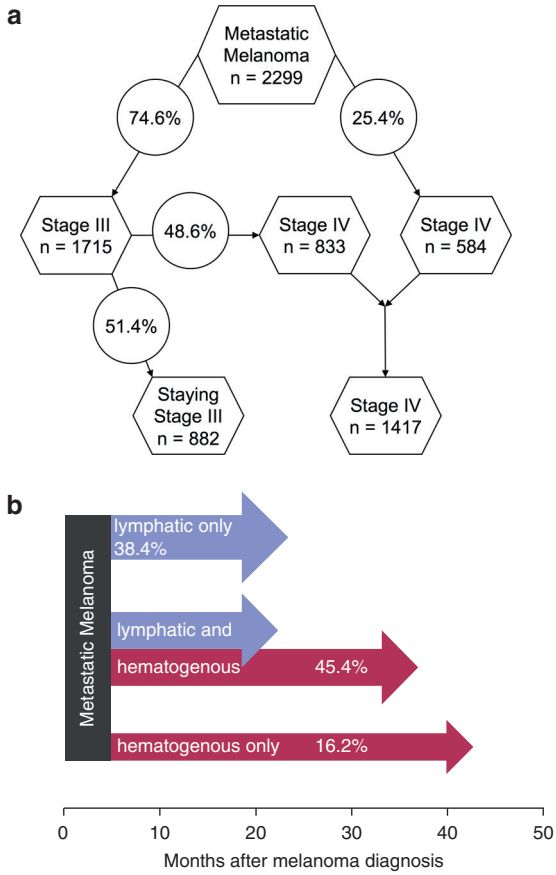

Figure 1. Metastatic pathways of the training cohort in Tuebingen. (**a**) Melanoma progression from the primary tumor to stage III and/or stage IV. (**b**) Distribution (percentage of total) of lymphatic and/or hematogenous metastasis. Arrowheads represent median time until occurrence of metastasis.

Initial lymphatic metastasis has no impact on survival of patients with distant disease

Table 2 shows patients and tumor characteristics of stage IV patients with and without prior stage III disease. Significant differences between both groups were found in melanoma localization (thorax, upper arm, neck, scalp [TANS] vs. non-TANS region) and tumor thickness. Sex, age, histopathological subtype, ulceration status, and tumor burden of distant disease did not differ between both groups.

The metastatic pathway (directly stage IV vs. stage IV after stage III) had no impact on DMFS and OS according to univariate Kaplan-Meier method (Figure 2). Median DMFS was 35 months for patients with stage IV after stage III and 36 months for patients with direct progression to stage IV ($P = 0.68$) (Figure 2a). Median OS was 56 months for patients with stage IV after stage III and 59 months for patients with direct progression to stage IV ($P = 0.22$) (Figure 2b). Table 3 shows the multivariate Cox proportional hazards model for DMFS, MSS, and OS. We investigated the impact of primary lymphatic or hematogenous metastasis on survival of stage IV patients and found no differences in DMFS (hazard ratio [HR] = 1.02, 95%

Table 1. Characterization of the training cohort (Tuebingen)

Characteristics	n (%)[1]
Sex	
Female	1,038 (45.2)
Male	1,261 (54.8)
Body site	
TANS	1,412 (61.4)
Non-TANS	887 (38.6)
Tumor thickness in mm	
≤1.00	422 (18.4)
1.01–2.00	700 (30.4)
2.01–4.00	760 (33.1)
>4.00	417 (18.1)
Median (IQR)	2.10 (1.20–3.50)
Histopathological subtype	
SSM	1,052 (45.8)
NM	642 (27.9)
LMM	165 (7.2)
ALM	187 (8.1)
Others	249 (10.8)
Unknown	4 (0.2)
Ulceration	
Yes	681 (29.6)
No	1,127 (49.0)
Unknown	491 (21.4)
Age at melanoma diagnosis (years)	
>60 years	1,000 (43.5)
≤60 years	1,299 (56.5)
Mean ± SD	56.57 ± 15.54

Abbreviations: ALM, acral lentiginous melanoma; IQR, interquartile range; LMM, lentigo maligna melanoma; NM, nodular melanoma; SD, standard deviation; SSM, superficial spreading melanoma; TANS, thorax, upper arm, neck, scalp.
[1]The total number of patients was 2,299.

Table 2. Patient and tumor characteristics for patients with direct or indirect progression to stage IV (Tuebingen)[1]

Characteristics	Directly to Stage IV, n (%) (n = 584)	Stage IV after Stage III, n (%) (n = 833)	*P*-Value[2]
Sex			0.28
Female	241 (41.3)	368 (44.2)	
Male	343 (58.7)	465 (55.8)	
Body site			<0.001
TANS	435 (74.5)	466 (55.9)	
Non-TANS	149 (25.5)	367 (44.1)	
Tumor thickness (mm)			0.048
≤1.00	114 (19.5)	142 (17.0)	
1.01–2.00	176 (30.1)	225 (27.0)	
2.01–4.00	171 (29.3)	303 (36.4)	
>4.00	123 (21.1)	163 (19.6)	
Median (IQR)	2.07 (1.20 – 3.60)	2.35 (1.30 – 3.90)	
Histopathological subtype			0.052
SSM	291 (49.8)	367 (44.1)	
NM	159 (27.2)	266 (31.9)	
LMM	34 (5.8)	46 (5.5)	
ALM	33 (5.7)	74 (8.9)	
Others	66 (11.3)	79 (9.5)	
Unknown	1 (0.2)	1 (0.1)	
Ulceration			0.85
Yes	174 (29.8)	260 (31.2)	
No	267 (45.7)	375 (45.0)	
Unknown	143 (24.5)	198 (23.8)	
Age at melanoma diagnosis in years			0.31
>60 years	242 (41.4)	323 (38.8)	
≤60 years	342 (58.6)	510 (61.2)	
Mean ± SD	56 ± 15	55 ± 15	
Distant metastasis			0.064
Skin, SQ, or distant nodes	27 (4.6)	59 (7.1)	
Lung	61 (10.4)	104 (12.5)	
Nonpulmonary visceral	496 (84.9)	670 (80.4)	

Abbreviations: ALM, acral lentiginous melanoma; IQR, interquartile range; LMM, lentigo maligna melanoma; NM, nodular melanoma; SD, standard deviation; SQ, subcutaneous; SSM, superficial spreading melanoma; TANS, thorax, upper arm, neck, scalp.
[1]Total number of patients was 1,417.
[2]*P*-values refer to Pearson chi-square test.

confidence interval [CI] = 0.91–1.14, $P = 0.76$), MSS (HR = 1.08, 95% CI = 0.96–1.22, $P = 0.21$), and OS (HR = 1.09, 95% CI = 0.96–1.23, $P = 0.177$) irrespective of the initial metastatic pathway. These results could be independently confirmed from centers in Wuerzburg (DMFS: HR = 0.95, 95% CI = 0.80–1.14, P = 0.60; OS: HR = 0.86, 95% CI = 0.71–1.04, $P = 0.127$), Halle (DMFS: HR = 1.03, 95% CI = 0.83–1.29, $P = 0.79$; OS: HR = 0.80, 95% CI = 0.63–1.01, $P = 0.061$) and Erlangen (DMFS: HR = 1.25, 95% CI = 0.99–1.57, $P = 0.065$; OS: HR = 1.05, 95% CI = 0.75–1.45, $P = 0.79$) (see Supplementary Tables S1–S5 online).

The initial metastatic pathway depends on melanoma localization

By using a multivariate binary logistic regression model, we identified melanoma localization as the only significant risk factor associated with the initial metastatic pathway. The odds ratio for primary progression to stage IV was 2.18 (95% CI = 1.76–2.70, $P < 0.001$) for melanomas of the TANS region compared with melanomas of the non-TANS region (Figure 3). Tumor thickness, histopathological subtype, ulceration, age, and sex promoted neither initial lymphatic nor initial hematogenous metastasis.

DISCUSSION

Our understanding of melanoma metastasis and lymph node surgery is based on the idea that melanoma progresses from the primary tumor to the regional lymph nodes and from there to distant sites. This study shows that the Halstedian hypothesis cannot be confirmed for melanoma and that development of regional and distant metastasis is not a serial but probably a parallel process. These findings have clinically relevant implications for regional lymph node surgery.

Most patients with direct progression to stage IV had no evidence of regional lymph node or locoregional metastasis until death. These results indicate that the primary tumor itself must seed distant disease in at least this subgroup of melanoma patients. The fact that DMFS and OS did not differ

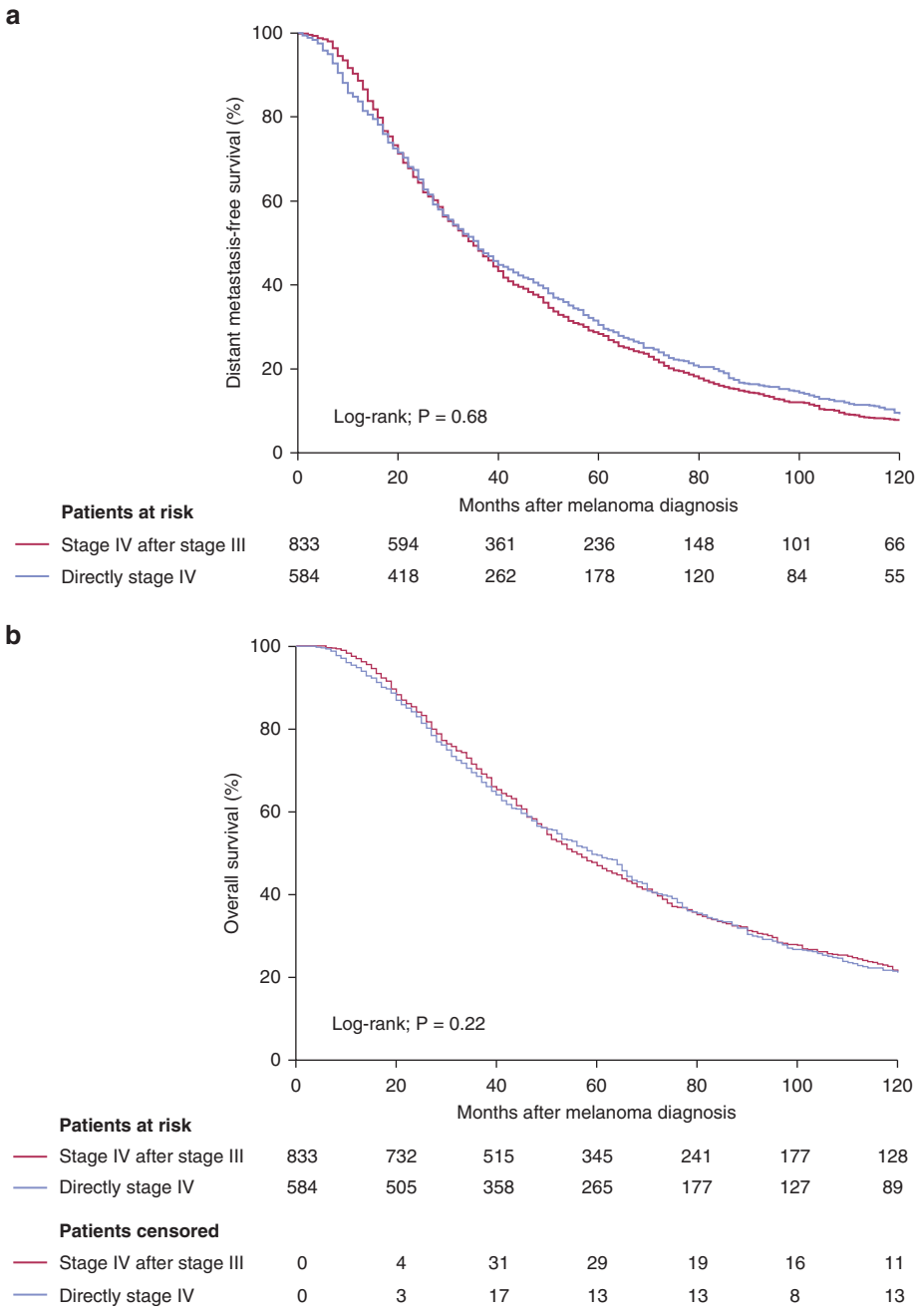

Figure 2. Survival of stage IV patients with respect to the primary metastatic pathway. (**a**) Distant metastasis-free survival and (**b**) overall survival were compared using the log-rank test. Total number of patients was 1,417.

between stage IV patients with and without prior stage III disease further supports the idea that primary lymphatic metastasis does not lead to secondary distant metastasis and that distant disease has the same origin in patients with and without primary lymphatic metastasis, namely the primary tumor.

These findings lead to a different understanding of the types of metastatic spread in melanoma, and probably also in many other types of solid cancers: The first type of metastatic spread is lymphatic only, with in-transit, satellite, or lymph node metastasis without subsequent distant metastasis. This occurred in 38.4% of our cohort, and patients are cured with surgery. It remains unknown whether simple metastasectomy could be substituted for CLND in these patients. The second type of metastatic spread is combined lymphatic and hematogenous spread originating from the primary tumor, which was found in 45.4% of our cohort. The third type is pure hematogenous spread from the primary tumor, found in 16.2% of our cohort (Figure 1b).

Our proposed model of metastatic spread is corroborated by several clinical studies. Four randomized controlled trials comparing immediate elective lymph node dissection in patients with localized melanoma versus observation and delayed surgery in case of nodal recurrence were conducted in the pre-SNLB era (Balch et al., 1996; Cascinelli et al., 1998; Sim et al., 1978; Veronesi et al., 1977). In none of these studies did prophylactic surgery improve survival. Additionally, the Multicenter Selective

Table 3. Prognostic factors of melanoma patients with distant metastasis in their further course[1]

	DMFS (n = 1,417 events)		OS (n = 1,154 events)		MSS (n = 1,128 events)	
Characteristics	**HR (95% CI)**	***P*-Value**	**HR (95% CI)**	***P*-Value**	**HR (95% CI)**	***P*-Value**
Metastatic pathway						
Directly to stage IV vs. stage IV after stage III	1.02 (0.91–1.14)	0.76	1.09 (0.96–1.23)	0.177	1.08 (0.96–1.22)	0.21
Sex						
Male vs. female	1.15 (1.03–1.29)	0.012	1.24 (1.10–1.40)	0.001	1.25 (1.10–1.41)	<0.001
Body site						
TANS vs. Non-TANS	1.07 (0.95–1.20)	0.29	1.11 (0.98–1.27)	0.101	1.11 (0.98–1.27)	0.113
Tumor thickness						
pT3 and pT4 vs. pT1 and pT2	1.41 (1.21–1.65)	<0.001	1.62 (1.44–1.82)	<0.001	1.63 (1.44–1.84)	<0.001
Histopathological subtype						
NM vs. SSM	1.13 (1.00–1.28)	0.056	1.02 (0.89–1.17)	0.82	1.00 (0.87–1.15)	0.95
LMM vs. SSM	1.02 (0.81–1.30)	0.85	1.01 (0.78–1.30)	0.97	0.97 (0.74–1.26)	0.80
ALM vs. SSM	1.17 (0.95–1.44)	0.142	1.16 (0.92–1.46)	0.21	1.18 (0.94–1.48)	0.161
Others vs. SSM	1.09 (0.92–1.29)	0.35	1.09 (0.90–1.31)	0.39	1.07 (0.89–1.29)	0.48
Ulceration						
Yes vs. no	1.47 (1.32–1.65)	<0.001	1.42 (1.26–1.61)	<0.001	1.39 (1.23–1.57)	<0.001
Age at diagnosis						
>60 years vs. ≤60 years	1.51 (1.35–1.69)	<0.001	1.41 (1.25–1.59)	<0.001	1.36 (1.20–1.54)	<0.001

Abbreviations: ALM, acral lentiginous melanoma; CI, confidence interval; DMFS, distant metastasis-free survival; HR, hazard ratio; LMM, lentigo maligna melanoma; MSS, melanoma-specific survival; NM, nodular melanoma; OS, overall survival; SSM, superficial spreading melanoma; TANS, thorax, upper arm, neck, scalp.

[1]Results of multivariate analysis using Cox proportional hazards model (Tuebingen). Total number of patients was 1,417.

Lymphadenectomy Trial I, comparing SLNB with immediate CLND in cases of positive sentinel lymph node and nodal observation and delayed CLND in case of nodal recurrence, showed no difference in the 10-year melanoma-specific survival (Morton et al., 2014). Recently, the Multicenter Selective Lymphadenectomy Trial II (Faries et al., 2017) confirmed the results of the previous DeCOG-SLT trial, showing that immediate CLND in SLNB-positive melanomas was not superior to observation (Leiter et al., 2016). These results are in line with several retrospective trials comparing immediate CLND versus observation that found no improvement of MSS (Kingham et al., 2010; Wong et al., 2006). Clinical trials in numerous other cancer entities further corroborate the idea that prophylactic regional lymph node surgery has no impact on survival (Galimberti et al., 2013; Gervasoni et al., 2007; Klein, 2009).

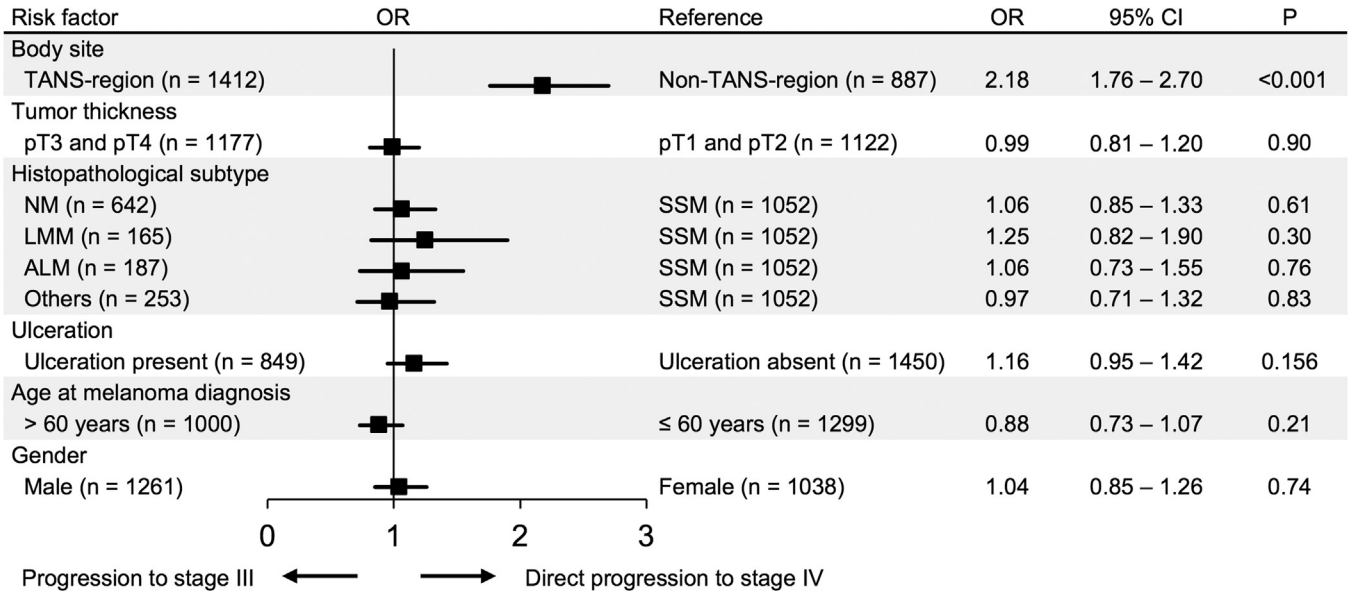

Figure 3. Risk factors for the primary metastatic pathway (Tuebingen). Results of multivariate analysis using binary logistic regression (progression to stage III vs. direct progression to stage IV) adjusted for localization, tumor thickness, histopathological subtype, ulceration, age, and sex. Total number of patients was 2,299. ALM, acral lentiginous melanoma; CI, confidence interval; LMM, lentigo maligna melanoma; NM, nodular melanoma; OR, odds ratio; SSM, superficial spreading melanoma; TANS, thorax, upper arm, neck, scalp.

The lack of a continuous basement membrane and tight inter-endothelial junctions makes lymphatic vessels relatively leaky and easy for cancer cells to enter compared with blood vessels (Alitalo and Carmeliet, 2002). This would explain why lymphatic metastasis is seen more often than hematogenous metastasis. Shain and Bastian (2016) explained the head start of regional metastases with their direct connection to the primary tumor through the lymphatics. Repeated seeding over time could thereby increase the number of founding cells at the same localization and increase the probability of successful metastasis formation compared with distant metastasis (Shain and Bastian, 2016).

Moreover, experimental studies have refuted Virchow's assumption that lymph nodes are an effective barrier to cancer cell dissemination and have shown that lymphatic and lymphatic-venous shunts facilitate cancer cells bypassing lymph nodes and allow both lymphatic and hematogenous spread (Cady, 1984; Fisher and Fisher, 1966; Virchow and Chance, 1863). These observations suggest a parallel progression model, whereby the primary tumor seeds regional lymph node and distant metastasis (Klein, 2009). Recently, whole-exome sequencing of matched primary melanomas and their regional and distant metastases has facilitated a better understanding of metastatic pathways (Sanborn et al., 2015). Delineation of the phylogenetic relationship between tumor populations at different sites provided evidence that genetically distinct subpopulations of the primary tumor found both regional and distant metastases and intratumoral heterogeneity may explain why metastatic patterns vary widely among melanoma patients. Moreover, sites of metastatic lesions seem to be determined by the organ microenvironment, indicating that the "seed" has to be compatible with the "soil" (Fidler, 2003; Paget, 1989). Additionally, active recruitment of cancer cells via chemokine gradients could explain the tissue tropism observed in melanoma and other cancers (Roussos et al., 2011). Experimental studies have shown that lymphatic endothelial cells are able to recruit melanoma cells toward the draining lymph node via release of the chemokine CC-ligand 21 (Takeuchi et al., 2004; Wiley et al., 2001). High expression of the corresponding CC-chemokine receptor 7 on melanoma cells could thereby promote lymphatic metastasis in the absence of hematogenous metastasis. Our findings further show that primary lymphatic or hematogenous spread depends on melanoma localization. Lymphatic metastasis is obviously facilitated by localizations with well-defined lymph drain such as the extremities and aggravated by body sites with ambiguous lymph drain such as the head, neck, and trunk (TANS region). Drainage to a single sentinel node field is mostly encountered in patients with melanomas of the upper or lower limb, whereas drainage to multiple node fields and across the midline is often seen in patients with melanomas on the head, neck, and trunk (Uren et al., 2003). Sugarbaker and McBride (1976) and Norman et al. (1991) pointed out that lymphatic drainage up to 11 cm from the midline is highly variable and unpredictable.

Although the primary metastatic pathway of stage IV patients had no impact on survival, several other prognostic factors were in line with previous publications and could be confirmed in our multivariate analysis (age, sex, tumor thickness, and ulceration) (Table 3) (Balch et al., 2001).

A limitation of this study is that the 10-year follow-up of patients with melanoma diagnosis from 2006 onward was not finished at the time of the analysis, resulting in an underestimation of stage IV disease in this group. However, this did not bias the conclusions of the study, because DMFS did not differ between patients with direct or indirect progression to stage IV. Further, synchronous regional and distant metastasis was detected in 26.0% of patients with direct progression to stage IV and follow-up until death. We cannot exclude that some of these patients might in truth have had unnoticed primary progression to stage III.

The study is strengthened by the prospective documentation, the use of multiple independent centers as confirmation cohorts, and strict inclusion criteria. We included only patients who initially presented with localized cutaneous melanoma and absence of regional and distant metastasis. This allowed us to accurately estimate the DMFS in patients with direct or indirect progression to stage IV. This study dissects biologically distinct progression patterns from the endpoint "distant metastasis" and uses a multivariate binary logistic regression model to identify risk factors for the initial metastatic pathway. We compared the DMFS and OS of stage IV patients with and without prior lymphatic metastasis, whereas earlier reports and clinical trials primarily focused on the impact of regional lymph node surgery (SLNB or CLND) on survival of stage III patients (Kingham et al., 2010; Leiter et al., 2016; Morton et al., 2014; Wong and Hynes, 2006).

In conclusion, our findings improve the understanding of melanoma progression and add to the growing body of evidence that distant metastases originate from the primary melanoma itself and not from regional metastases. We showed that metastatic dissemination is orchestrated in a rather parallel than serial fashion and that the Halstedian hypothesis should be rejected. These insights could explain why immediate CLND in SLNB-positive patients fails to improve survival and is not superior to observation.

MATERIALS AND METHODS

Patient population and follow-up

Between 1976 and 2015, 127,262 melanoma patients were documented in the nationwide German Central Malignant Melanoma Registry. More than 10% of these patients were diagnosed and treated at the Department of Dermatology of the University Hospital Tuebingen (n = 13,994). Only patients with invasive cutaneous melanoma presenting with stage IA–IIC at primary diagnosis and developing stage III or stage IV disease in the further course were included into our analysis. Patients with unknown primary melanoma, known metastasis at the time of the initial diagnosis, and follow-up of less than 3 months were excluded. Of 12,493 patients with invasive cutaneous melanoma and sufficient follow-up, 9,582 (76.7%) remained in stage IA–IIC, and 2,911 (23.3%) developed metastases. A total of 612 patients (4.9%) had already-disseminated disease at primary melanoma diagnosis. Inclusion and exclusion criteria resulted in selection of 2,299 patients of the clinical center Tuebingen. This was the training cohort of this work. The three next-largest centers were Wuerzburg (n = 946), Halle (n = 683), and Erlangen (n = 505), which were referred to as confirmation cohorts.

Follow-up was performed according to the current valid version of the guidelines of the German Society of Dermatology, which remained rather constant over time. Physical examination, consisting of a thorough inspection of the entire skin and palpation of the excision site, in-transit route, and regional lymph nodes, was performed every 3−12 months over 10 years. Furthermore, ultrasonographic scans of regional lymph nodes and the upper abdomen, chest X-ray, and blood testing including lactate dehydrogenase and tumor marker S100B were performed every 3−12 months. Computed tomography scans of the whole body and head and magnetic resonance imaging, respectively, were carried out in high-risk melanoma patients (stage IIC−IV) and if metastasis was suspected.

After obtaining informed written consent from the patients, personal and initial staging data were collected and submitted to the German Central Malignant Melanoma Registry. The study was approved by the ethics board of the University Hospital Tuebingen.

At documentation, localization was subdivided into face, other parts of the head, neck, breast/upper abdomen, back, lower abdomen, buttocks, external genitalia, upper arm including elbow, forearm, hand, thigh including knee, lower leg, and foot. To generate fewer subgroups for statistical analysis, melanoma localization was summarized according to TANS region (thorax, upper arm, neck and scalp) and non-TANS region (all other localizations) (Garbe et al., 1995). Patients were further split into different age groups: those 60 years or younger and those older than 60 years.

Histological report included tumor thickness in millimeters (Breslow), level of invasion (Clark), histopathological subtype, and presence or absence of ulceration. Unchanged status or progression of the disease was recorded at follow-up visits. Progression was documented in detail and differentiated between locoregional metastases (satellite and in-transit metastases), regional lymph node metastases, and distant metastases (skin, distant lymph node, visceral, lung, bone, central nervous system, liver, intestine, and others). SLNB was routinely performed at the Department of Dermatology of the University Hospital Tuebingen from 1996 onward.

Statistical analysis

Statistical calculations were performed with IBM SPSS Statistics version 23.0 (IBM SPSS, Chicago, IL). Numerical variables were described by mean value and standard deviation if approximately normally distributed or median value and IQR if skewed. Proportions were presented with 95% CI.

Only deaths due to cutaneous melanoma were considered events for the calculation of MSS, and deaths from all causes were considered events for the calculation of OS. In patients for whom mortality and cause of death were not directly reported, registration offices were systematically addressed. Survival rates were estimated according to Kaplan-Meier and compared with the log-rank test.

Association between type of metastasis (lymphatic vs. hematogenous) and patients/tumor characteristics was assessed with the univariate chi-square test. Further analysis was performed with multivariate binary logistic regression models using progression to stage IV as the dependent variable (*yes* or *no*) and patients/tumor characteristics as covariates. We estimated the HRs for death and occurrence of distant metastasis by initial or delayed progression to stage IV by using multivariate Cox proportional hazards models adjusted for sex, localization, tumor thickness, histology, ulceration, and age. Results of Cox proportional hazards modeling were described as HRs together with 95% CIs and *P*-values. Missing values were imputed using multivariate imputation by chained equations strategies (van Buuren and Groothuis-Oudshoorn, 2011). Throughout the analysis, *P*-values less than 0.05 were considered statistically significant.

CONFLICT OF INTEREST

The authors state no conflict of interest.

ACKNOWLEDGMENTS

We thank the patients participating in the German Central Malignant Melanoma Registry, and everyone who has been involved in data collection, documentation, and maintenance of the database.

SUPPLEMENTARY MATERIAL

Supplementary material is linked to the online version of the paper at www.jidonline.org, and at http://dx.doi.org/10.1016/j.jid.2017.07.006.

REFERENCES

Alitalo K, Carmeliet P. Molecular mechanisms of lymphangiogenesis in health and disease. Cancer Cell 2002;1:219−27.

Balch CM. The role of elective lymph node dissection in melanoma: rationale, results, and controversies. J Clin Oncol 1988;6:163−72.

Balch CM, Soong SJ, Bartolucci AA, Urist MM, Karakousis CP, Smith TJ, et al. Efficacy of an elective regional lymph node dissection of 1 to 4 mm thick melanomas for patients 60 years of age and younger. Ann Surg 1996;224:255−63.

Balch CM, Soong SJ, Gershenwald JE, Thompson JF, Reintgen DS, Cascinelli N, et al. Prognostic factors analysis of 17,600 melanoma patients: validation of the American Joint Committee on Cancer melanoma staging system. J Clin Oncol 2001;19:3622−34.

Cady B. Lymph node metastases. Indicators, but not governors of survival. Arch Surg 1984;119:1067−72.

Cascinelli N, Morabito A, Santinami M, MacKie RM, Belli F. Immediate or delayed dissection of regional nodes in patients with melanoma of the trunk: a randomised trial. WHO Melanoma Programme. Lancet 1998;351(9105):793−6.

Faries MB, Thompson JF, Cochran AJ, Andtbacka RH, Mozzillo N, Zager JS, et al. Completion Dissection or Observation for Sentinel-Node Metastasis in Melanoma. N Engl J Med 2017;376:2211−22.

Fidler IJ. The pathogenesis of cancer metastasis: the 'seed and soil' hypothesis revisited. Nat Rev Cancer 2003;3:453−8.

Fisher B. Laboratory and clinical research in breast cancer—a personal adventure: the David A. Karnofsky memorial lecture. Cancer Res 1980;40:3863−74.

Fisher B, Anderson SJ. The breast cancer alternative hypothesis: is there evidence to justify replacing it? J Clin Oncol 2010;28:366−74.

Fisher B, Fisher ER. Transmigration of lymph nodes by tumor cells. Science 1966;152(3727):1397−8.

Fisher B, Jeong JH, Anderson S, Bryant J, Fisher ER, Wolmark N. Twenty-five-year follow-up of a randomized trial comparing radical mastectomy, total mastectomy, and total mastectomy followed by irradiation. N Engl J Med 2002;347:567−75.

Galimberti V, Cole BF, Zurrida S, Viale G, Luini A, Veronesi P, et al. Axillary dissection versus no axillary dissection in patients with sentinel-node micrometastases (IBCSG 23-01): a phase 3 randomised controlled trial. Lancet Oncol 2013;14:297−305.

Garbe C, Buttner P, Bertz J, Burg G, d'Hoedt B, Drepper H, et al. Primary cutaneous melanoma. Prognostic classification of anatomic location. Cancer 1995;75:2492−8.

Gervasoni JE Jr, Sbayi S, Cady B. Role of lymphadenectomy in surgical treatment of solid tumors: an update on the clinical data. Ann Surg Oncol 2007;14:2443−62.

Halsted WS. I. The results of radical operations for the cure of carcinoma of the breast. Ann Surg 1907;46:1−19.

Handley W, Lond M. The pathology of melanotic growths in relation to their operative treatment. Lancet 1907;1(4363):996−1003.

Kingham TP, Panageas KS, Ariyan CE, Busam KJ, Brady MS, Coit DG. Outcome of patients with a positive sentinel lymph node who do not undergo completion lymphadenectomy. Ann Surg Oncol 2010;17:514−20.

Klein CA. Parallel progression of primary tumours and metastases. Nat Rev Cancer 2009;9:302–12.

Leiter U, Stadler R, Mauch C, Hohenberger W, Brockmeyer N, Berking C, et al. Complete lymph node dissection versus no dissection in patients with sentinel lymph node biopsy positive melanoma (DeCOG-SLT): a multicentre, randomised, phase 3 trial. Lancet Oncol 2016;17:757–67.

Morton DL, Thompson JF, Cochran AJ, Mozzillo N, Nieweg OE, Roses DF, et al. Final trial report of sentinel-node biopsy versus nodal observation in melanoma. N Engl J Med 2014;370:599–609.

Norman J, Cruse CW, Espinosa C, Cox C, Berman C, Clark R, et al. Redefinition of cutaneous lymphatic drainage with the use of lymphoscintigraphy for malignant melanoma. Am J Surg 1991;162:432–7.

Paget S. The distribution of secondary growths in cancer of the breast. 1889. Cancer Metastasis Rev 1989;8:98–101.

Rebecca VW, Sondak VK, Smalley KS. A brief history of melanoma: from mummies to mutations. Melanoma Res 2012;22:114–22.

Roussos ET, Condeelis JS, Patsialou A. Chemotaxis in cancer. Nat Rev Cancer 2011;11:573–87.

Sanborn JZ, Chung J, Purdom E, Wang NJ, Kakavand H, Wilmott JS, et al. Phylogenetic analyses of melanoma reveal complex patterns of metastatic dissemination. Proc Natl Acad Sci USA 2015;112:10995–1000.

Shain AH, Bastian BC. From melanocytes to melanomas. Nat Rev Cancer 2016;16:345–58.

Sim FH, Taylor WF, Ivins JC, Pritchard DJ, Soule EH. A prospective randomized study of the efficacy of routine elective lymphadenectomy in management of malignant melanoma. Preliminary results. Cancer 1978;41:948–56.

Sugarbaker EV, McBride CM. Melanoma of the trunk: the results of surgical excision and anatomic guidelines for predicting nodal metastasis. Surgery 1976;80:22–30.

Takeuchi H, Fujimoto A, Tanaka M, Yamano T, Hsueh E, Hoon DS. CCL21 chemokine regulates chemokine receptor CCR7 bearing malignant melanoma cells. Clin Cancer Res 2004;10:2351–8.

Uren RF, Howman-Giles R, Thompson JF. Patterns of lymphatic drainage from the skin in patients with melanoma. J Nucl Med 2003;44:570–82.

van Buuren S, Groothuis-Oudshoorn K. mice: multivariate imputation by chained equations in R. J Stat Softw 2011;45:1–67.

Veronesi U, Adamus J, Bandiera DC, Brennhovd IO, Caceres E, Cascinelli N, et al. Inefficacy of immediate node dissection in stage 1 melanoma of the limbs. N Engl J Med 1977;297:627–30.

Virchow RLK, Chance FN. Cellular pathology. Philadelphia: JB Lippincott & Co.; 1863.

Wiley HE, Gonzalez EB, Maki W, Wu MT, Hwang ST. Expression of CC chemokine receptor-7 and regional lymph node metastasis of B16 murine melanoma. J Natl Cancer Inst 2001;93:1638–43.

Wong SL, Morton DL, Thompson JF, Gershenwald JE, Leong SP, Reintgen DS, et al. Melanoma patients with positive sentinel nodes who did not undergo completion lymphadenectomy: a multi-institutional study. Ann Surg Oncol 2006;13:809–16.

Wong SY, Hynes RO. Lymphatic or hematogenous dissemination: how does a metastatic tumor cell decide? Cell Cycle 2006;5:812–7.