Lymphoscintigraphy and sentinel lymph node biopsy, in cutaneous melanoma staging and treatment decisions

Ljiljana Jaukovic[1], MD, Phd
Goran Sijan[2], MD, MSc
Milica Rajović[2], MD
Lidija Kandolf Sekulovic[3], MD, Phd
Marija Radulovic[1], MD
Zeljko Mijuskovic[3], MD, Phd
Lidija Zolotarevski[4], MD, MSc
Boris Ajdinovic[1], MD, PhD
Marijan Novakovic[2], MD, Phd

1. Institute of Nuclear Medicine
2. Clinic for Plastic and Reconstructive Surgery
3. Clinic for Dermatology
4. Center of Pathology and Forensic Medicine
Military Medical Academy, Belgrade, Serbia

Keywords: Lymphoscintigraphy - Sentinel lymph node biopsy - Cutaneous melanoma

Correspondence address:
Dr Ljiljana Jaukovic,
Institute of Nuclear Medicine,
Military Medical Academy,
Crnotravska 17,
11 000 Belgrade, Serbia
Tel: +381638412795,
lilaj2805@gmail.com

Received:
5 March 2015
Accepted revised:
25 April 2015

Abstract

Objective: Sentinel lymph node biopsy (SLNB) is a widely accepted method in the management of clinically localized cutaneous melanomas. The aim of this study was to report the results on patients scheduled for preoperative lymphoscintigraphy and SLNB for staging and further treatment planning. **Subjects and Methods**: Two hundred and one patients (115 male and 86 female, median age 57 years, range 9-81) with cutaneous melanoma having undergone SLB at Military Medical Academy between November 2010 and October 2014, were recruited for retrospective study. Dual labeling method (Tc-99m Nanocolloid / blue dye) was used. In order to delineate the relation between patients' tumors and scintigraphic characteristics with positive SLN findings, we examined all variables by univariate logistic regression with odd ratios representing the size effect. **Results**: The overall identification rate of SLN was 98.5%. One or more positive SLN were seen in 47 (23.4%) of the patients. Drainage to one regional basin was noticed in 176 (88%) and multiple drainage regions, up to three, was noticed in 24 patients (12%). Transit lymph nodes were detected in 20 patients (10%). The characteristics that were assotiated significatly with sentinel lymph node metastases were Breslow thickness, nodular melanoma histological subtype and acral localization. **Conclusion**: Besides the well established primary tumor thickness being a predictor of SLN malignancy, we observed: acral body site location and nodular melanoma histological subtype to be significant independent factors in increasing the risk for regional metastases. Our results suport the clinical usefulness of SLNB within a multidisciplinary approach (dermatooncology, plastic/head and neck surgery, pathology, nuclear medicine), as a reliable method in staging and for treatment planning in melanoma patients.

Hell J Nucl Med 2015; 18(2): 146-151 *Epub ahead of print: 19 July 2015* *Published online: 5 August 2015*

Introduction

Cutaneous melanoma is the most rapidly increasing cancer in white population worldwide, responsible for approximately 75% of all deaths from skin cancer. Although clinical diagnosis of cutaneous melanoma is not difficult, there remain many controversies regarding treatment decision and the role of sentinel lymph node biopsy (SLNB). Sentinel lymph node is defined as the lymph node upon which a lymph vessel originating in the tumour drains directly. As the tumour may drain to more than one lymph node , term SLN encompass all nodes directly at risk of receiving tumour cells. Described by Morton et al. (1992) lymphatic mapping and SLN biopsy technique are used for accurate staging of cutaneous melanoma based on the assumption that the first node draining tumor area indicates tumor status of the related nodal basin [1]. Tumor negative SLN predicts that other nodes in the same drainage basin will also be tumor free, while tumor positive SLN predicts that nonsentinel nodes might also harbor metastasis [1, 2].

The most important prognostic factor in melanoma patients is the status of regional lymph nodes [3]. Lymph node metastases in concordance with Breslow thickness indicate a higher risk in cutaneous melanoma [4, 5].

Sentinel lymph node biopsy (SLNB) should be offered to patients with clinically localized disease and invasive melanoma, including intermediate thickness melanoma (1-4mm) and thick melanomas (thickness >4mm). Patients with melanomas thinner than ≤1mm are also candidates for SLNB in the presence of coexisting factors that may increase the risk of nodal metastases including ulceration, mitotic rate >0/mm^2, young age and male gender [6-10].

Selective biopsy of sentinel nodes can distinguish patients who have occult regional

metastases and may benefit from complete lymph node dissection (CLND) from those with no metastases. Despite the prevailing opinion that the main goals of SLNB still remain staging and local tumor control with no overall survival benefit, the very recent study of Morton et al (2014) showed that biopsy-based management improved the 10-year rate of distant disease-free survival and the 10-year rate of melanoma-specific survival for patients with nodal metastases from intermediate-thickness melanomas [11]. Several clinical and histological characteristics of primary melanoma have been examined as predictors of SLN positivity including Breslow thickness [10, 12-20], Clark level [10, 12, 14, 17] tumor mitotic rate [13, 14], histological type [15-17], ulceration [12-14, 17] and regression [18-20]. Regarding the patients' characteristics, SLNB positivity was also linked to age [4-21], sex [4-21], tumor location [21-24], number of draining basins [25, 26] etc. Only a few of these variables, like Breslow thickness and Clark level were found to be reproducibly statistical predictors. This could be linked to the lack of standardised approach to the use of SLNB procedure in various institutions, to variation of study design and of data collected. In this paper, we reviewed our experience with SLNB in cutaneous melanoma in our population, aiming to evaluate factors related to patients' and tumors' characteristics, as their scintigraphic pattern, which may be related with SLN metastatic involvment.

Patients and Methods

Patients

During the period November 2010 to September 2014, lymphoscintigraphy and SLNB were performed at our Institution in 250 patients with cutaneous melanoma. We report the results on 201 patients who met the inclusion criteria. Two hundred and one patients (115 men and 86 women) constituted our study sample for statistical analysis. The mean patient age was 53.6±1.1 years (median 57.0, range 8-91). The indication for SLNB and the procedure was made according to international guidelines [7, 27].

Criteria for inclusion into the study were: (a) Patients with cutaneous melanoma >1mm thick with no evidence of distant metastases or clinical lymphadenopathy, (b) Patients with melanomas of less than 1mm in Breslow thickness with risk factors for metastases (ulceration or mitotic rate >1/mm^2), (c) sufficient data obtained on histopatology after surgical extirpation of SLN.

Methods

Demographic data and clinicopathologic characteristics of our patients included: age, sex, site of primary tumor, Breslow thickness, Clark level, T classification, histological type and mitotic tumor rate. Data related to the scintigraphic pattern included the number of SLN detected, the number of draining basins and the presence of in transit nodes.

Dynamic lymphoscintigraphy which was used to visualize the lymph flow, was initiated immediately after 3-4 intradermal injections of 15MBq of ^{99m}Tc-nanocolloid around the scar. Early and delayed lymphoscintigraphy images were obtained over the regional basins of drainage (Figure 1). Once the SLN was

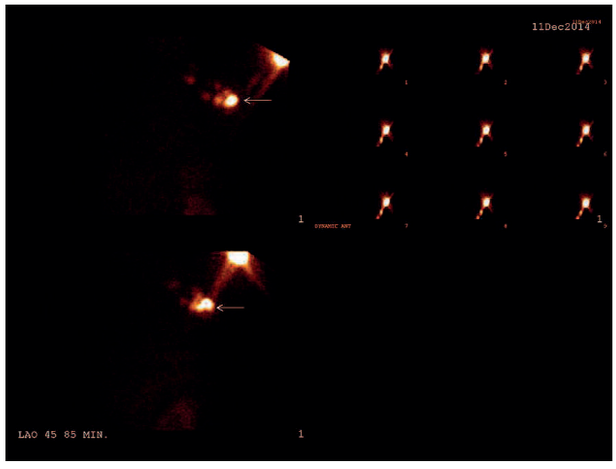

Figure 1. Dynamic images recorded immediately after i.v. injection of ^{99m}Tc-nanocolloid in left upper arm showing more lymph nodes in the left axillary region (on the right side of the picture). Scintigraphic image 45min after injection, in anterior, posterior and oblique positions showing SLN in left axilla on the left side of the picture.

Figure 2. Methylene blue dye (1%) was injected prior to operation, around the scar.

Figure 3. Intraoperative identification of SLN using gamma probe.

visualized, the surface location of the SLN was marked on the skin to assist the surgeon to localize the SLN nodes. Biopsy of the SLN was performed the day after lymphoscintigraphy. In selected cases, methylene blue dye (1%) in a volume of 1-2mL was injected intradermaly 15min before surgery (Figure 2). Dissection was guided by a hand-held gamma detection probe and by visual identification of the blue staining (Figure 3). The lymph node with the highest radioactivity compared to the neighbouring lymph nodes was regarded as the SLN. After each node was dissected, its count rate was recorded and the operated site was checked for any remaining activity.

Histopathology of the SLN was done using 2x10 full face serial sections stained with hematoxylin-eosin and S-100 and additionally with melan-A and Human Melanoma Black (HMB-45) staining. The primary outcome of this study was to find positive SLNB, which was defined as one or more positive SLN indicating metastatic disease. The study protocol was approved by the Institution's Medical Ethics Committee.

Statistics

Statistical analyses were performed using SPSS software, version 20.0.

Descriptive statistics, Mann-Whitney test for continuous variables and χ^2 test for analysis of categorical data were used for statistical analysis between the SLNB positive and SLNB negative group. A significance level of $P<0.05$ was used. Breslow thickness was also examined as a categorical variable: less than 1mm, 1.01-4.00mm and >4mm based on American Joint Committee on Cancer (AJCC) staging system [2]. Mitotic count was classified as <1; 1–5 or >5 per square millimeter. Tumor sites were grouped into five as: head and neck, upper extremities, trunk, lower extremities and acral. Acral melanoma was defined by anatomic location as melanoma on palmar, plantar, or subungual sites.

Variables analyzed for association with a positive SLN included patients' age, sex, site of primary tumor, Breslow thickness, clinicopathological type, mitotic count, presence of in transit nodes, the number of draining basins and body localization of primary melanoma. We also used univariate logistic regression with odds ratios representing the effective size.

Results

On lymphoscintigraphy, the SLN were detected in all patients but one. In two patients, in the operation room, the SLN were not identified, resulting in an overall identification rate of 98.5%. A total of 416 lymph nodes were excised. Among them were 1-6 SNL per patient (mean range 2.07). The most frequent site of primary melanomas was in the trunk (88/ 201) followed by lower extremities, head and neck, upper extremities and acral area in 39, 30, 29 and 15 patients respectively. Nodal basins included unilateral axilla in 45.7%, groin in 26.9%, neck region in 15.4%, bilateral axillae in 6.5% and in other sites in 5.5%. Drainage to one regional basin was seen in 176 patients (88%). Multiple drainage sites, were detected in 24 patients (12%). In-transit lymph nodes were detected in 20 patients in the popliteal region, thoracic wall and cubital region, in 8, 10 and 2 patients, respectively. One or more positive metastatic SLN were seen in 47/201 patients (23.4%). The mean Breslow was 3.37mm (median 2.25). Forty eight patients (24.4%) had a thin lesion ≤1mm. Breslow thickness significantly differed between the groups of positive and of negative SLN (median thickness 4.40mm vs 1.80mm). No significant difference was found in age and/or gender between the groups with positive or negative SLN results. Metastatic rate significantly differed between the groups of patients with acral and nonacral location of metastases (P=0.04). In addition, mitotic count rate did not significantly differ between groups. The clinical and pathologic characteristics of patients and tumors related to SLN status are shown in Table 1.

Univariate regression analysis with 201 cases revealed: Breslow thickness, nodular melanoma histological subtype and acral localization to be significant independent predictors of SLN status ($P<0.05$) (Table 2). The other variables: Clark level of invasion, mitotic rate, number of SLN draining basins and presence of interval nodes were not significantly correlated with SLN status. The odds of a positive SLN biopsy result were more than two-fold higher among patients with nodular melanoma histology subtype than among those with other kinds of melanoma subtypes. Nodular histology and acral location were correlated with higher rates of SLN metastases even after adjustment for tumor thickness (Figures 4, 5).

Table 1. *Patients' characteristics and sentinel lymph node status*

Variable	SLN positive	SLN negative	N
Age (years)			201
30	4	14	18
30-49	15	38	53
50-69	22	73	95
70	6	29	35
Sex			201
Male	31	84	115
Female	16	70	86
Tumor location			201
Head/neck	6	24	30
Upper extremities	3	26	29
Trunk	17	71	88
Lower extremities	13	26	39
Acral	8	7	15
Breslow thickness (mm)			197
≤ 1	6	42	48
1.1-4.0	14	72	86
≥ 4	25	38	63
Clark level			190
I	0	1	1
II	2	19	21
III	8	49	57
IV	29	70	99
V	5	7	12
No of basins			200
1	41	135	176
2	6	18	24
Tumor mitotic rate			179
< 1	9	22	31
1-5	20	90	110
>5	12	26	38

Table 2. *Variables significantly associated with SLNB positivity on the univariate regression analysis*

Variable	Positive SLN status, no (%)	OR	95% CI	P value
Breslow thickness	8 (53.3)	1.115	1.021-1.217	0.015
Acral location	13 (32.5)	4.308	1.472-12.610	0.008
Nodular histology type		2.214	1.021-4.800	0.04

SLN- sentinel lymph node; OR-odds ratio, CI- confidence interval

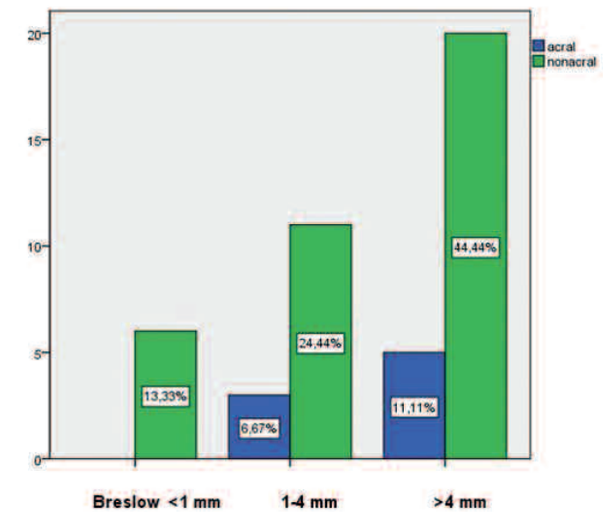

Figure 4. Association between acral location and positive SLN stratified by Breslow thickness.

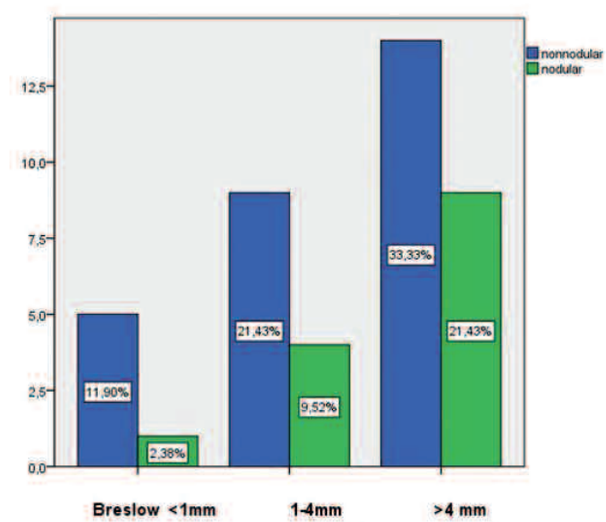

Figure 5. Association between nodular histology type and positive SLN stratified by Breslow thickness.

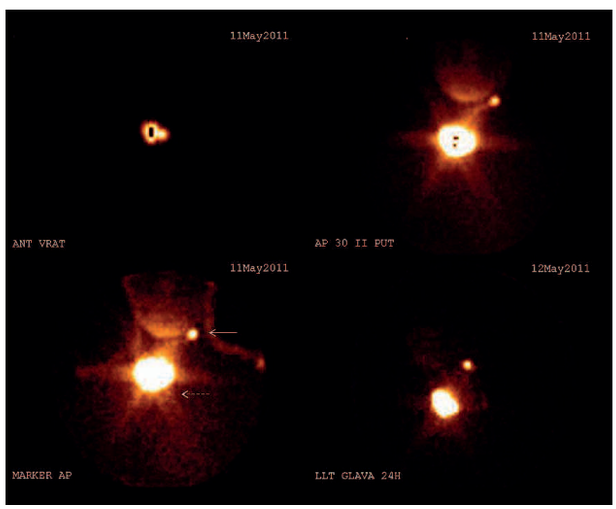

Figure 6. Sentinel lymph nodes in the submandibular region on the side opposite to the site of primary melanoma.

Discussion

Sentinel node biopsy is accepted worldwide as an important part of the management of clinically localized cutaneous melanoma. The identification rate of SLN in our melanoma patients was very high (98.5%). Dual labeling method performed in 79/201 patients showed that radiocolloid identified more nodes per patient than the blue dye which is in concordance with literature data [28]. Using the gamma camera we detected SLN in all patients except one. This patient had undergone a month before wide local excision of primary melanoma in the trunk. On the contrary, in another patient who had previously undergone wide excision of melanoma of the frontal region of the head, SLN was successfully detected in the neck region. According to the international guidelines [27] a contraindication for SLN biopsy is previous wide excision of the lymph nodes of the area because lymphatic pathways may be excessively disrupted, although there are some objections to that. [29]. In one of our patients lymphoscintigraphy failed to identify the drainage of the lymph from one SLN to the contralateral site SLN (Figure 6). In patients with head and neck melanoma difficulties in SLN detection are mainly due to small distance between injection site and draining nodal basin, to difficulty to identify locations of the SLN like located in the parotid gland, or to discordant lymphatic drainage contrary to what was clinically expected. However, a recent study of Erman et al. (2012) showed that the accuracy and prognostic value of SLNB in the head and neck region was comparable to that of other regions [30].

Drainage to more than one regional basin was shown in 24 patients (12%). Nearly half of them had the trunk primary draining to both axilla. .Metastatic involvement of SLN was found in 6/24 patients (25%). This rate did not significantly dif-

fer from the same rate in patients with drainage to single lymph basin (P=0.80). Lymphatic nodes in the area between the primary melanoma and the regional basin are called: "in-transit nodes" or "interval nodes" and are considered SLN. We detected interval nodes in 20 patients (10%). This indicates the importance of preoperative lymphoscintigraphy. In these interval nodes metastatic cell deposits were identified in two of ten patients who had popliteal and epitrochlear nodes. Other studies with a larger number of patients also often showed metastatic cells in in-transit SLN [31, 32]. The European Association of Nuclear Medicine recommends that interval nodes visualized by lymphoscintigraphy should be removed along with the SLN when found in standard node basins, since they may be the only metastatic lymph nodes [27]. The prognostic significance of a positive SLN does not change if this node is found in a major lymphatic basin or is detected as in-transit node.

Out of the 201 patients in our study 23.4% had one or more positive SLN. The slightly greater positivity rate in our study compared to other studies may be due to the rather small number of our patients, as well as to the different study population, but nevertheless is in agreement with other larger studies [14, 20, 24]. Most studies reported increasing Breslow thickness to be predictive of positive SLN status in melanoma patients [3-20, 23]. Results for other variables have been conflicting. [3-23].

The mean Breslow thickness in our study was 3.37mm (median 2.25) and was significantly different in patients with positive SLN versus those with negative SLN (P<0.001). The estimated risk for SLN metastases in Breslow 2-4mm raised from 1.361 (0.486-3.810; CI 95%) to 4.605 (1.706-12.434, CI 95%) for Breslow more than 4mm. Hinz et al. (2012) showed that in high risk melanomas patients because of the high lymph node metastatic rate of 27.4 %, SLNB had to be recommended even after exclusion of distant metastases [33].

Anatomic location of the primary tumor generally plays a minor prognostic role compared to other factors. The acral localization of the primary melanoma in our study was a significant predictor of SLN positivity in univariate model. Acral localization enhanced the odds of SLN positivity by 4.3 folds (versus non acral localization). Although the cohort of patients with acral melanoma in our study was very small, in this group there was high incidence of positive SLN, consistent with other studies [24]. This observation might be due to the difference in Breslow depth which was statistically significant (P=0.02) between acral and nonacral groups, in line with studies showing that acral melanoma exhibits aggressive histopathologic features and poorer survival [22, 34, 35]. An alternative, hypothesis is that cutaneous melanomas in acral sites, regardless of histology, tend to be diagnosed at an advanced stage probably owing to older patient age and difficult-to-see sites [24].

Histological subtype of nodular melanoma, in our study was associated with positive SLN in 31.8% of patients vs. 18.0% of patients with non nodular subtype (P=0.048). This difference was not related to the difference in Breslow tumor thickness but was significantly associated with nodular histology in patients with Breslow up to 4mm. Thus, nodular histology was a significant independent predictor of SLN positivity in an univariate model. Nodular histology enhanced the odds of SLN positivity by 2.21 fold (1.021- 4.800; CI 95 %). These findings are in accordance with the data of other researchers [16, 17].

In conclusion, our results confirm previous studies in melanoma patients and show SLN identification rate of 98.5% and percentage of SLN positive patients 23.4%. In our study the detection of SLN was independent of the site of primary melanoma. In-transit lymph nodes detected by lymphoscintigraphy should be removed. We also observed a significant association between positive SLN and primary tumor thickness, acral body site location and nodular histology. The influence of SLN status on prognosis should be defined through further follow-up studies.

Conflict of interest- nothing to declare.

Bibliography

1. Morton DL, Wen DR, Wong JH et al. Technical details of intraoperative lymphatic mapping for early stage melanoma. *Arch Surg.* 1992; 127: 392-9.
2. Morton DL, Cochran AJ, Thompson JF et al. Sentinel node biopsy for early-stage melanoma: accuracy and morbidity in MSLT-I, an international multicenter trial. *Ann Surg*. 2005; 242: 302-11.
3. White RL Jr, Ayers GD, Stell VH, Ding S et al Sentinel Lymph Node Working Group. Factors predictive of the status of sentinel lymph-nodes in melanoma patients from a large multicenter database. *Ann Surg Oncol.* 2011;18(13): 3593-600.
4. Osella-Abate S, Ribero S, Sanlorenzo M et al. Risk factors related to late metastases in 1,372 melanoma patients disease free more than 10 years. *Int J Cancer* 2015; 136(11): 2453-7.
5. Bartlett EK, Karakousis GC. Current Staging and Prognostic Factors in Melanoma. *Surg Oncol Clin N Am*. 2015; 24(2): 215-27.
6. Hieken TJ, Grotz TE, Comfere NI et al. The effect of the AJCC 7th edition change in T1 melanoma substaging on national utilization and out comes of sentinel lymph node biopsy for thin melanoma. *Melanoma Res*. 2015; 25(2): 157-63.
7. Wong SL, Balch CM, Hurley P et al. Sentinel lymph node biopsy for melanoma: American Society of Clinical Oncology and Society of Surgical Oncology joint clinical practice guideline. *J Clin Oncol*. 2012; 30: 2912-8.
8. Han D, Zager JS, Shyr Y, Chen H et al.. Clinicopathologic predictors of sentinel lymph node metastasis in thin melanoma. *J Clin Oncol.* 2013; 31(35): 4387-93.
9. Faries MB, Wanek LA, Elashoff D et al. Predictors of occult nodal metastasis in patients with thin melanoma. *Arch Surg*. 2010; 145: 137-42.
10. Doumas A, Dionyssopoulos A, Christoforidis T et al. Is 0.75mm Breslow thickness the correct cut-off point for performing sentinel node biopsy in patients with melanoma? *Hell J Nucl Med*. 2010; 13(3): 253-6.
11. Morton DL, Thompson JF, Cochran AJ et al. Final trial report of sentinel-node biopsy versus nodal observation in melanoma. *N Engl J Med*. 2014; 370: 599-609.
12. Balch CM, Thompson JF, Gershenwald JE et al. Age as a predictor of sentinel node metastasis among patients with localized melanoma: an inverse correlation of melanoma mortality and incidence of sentinel node metastasis among young and old patients. *Ann Surg Oncol*. 2014; 21: 1075-81.
13. Paek SC, Griffith KA, Johnson TM et al. The impact of factors beyond Breslow depth on predicting sentinel lymph node positivity in melanoma. *Cancer* 2007; 109: 100-8.
14. Han D, Zager JS, Shyr Y et al. Clinicopathologic predictors of sentinel lymph node metastasis in thin melanoma. *J Clin Oncol*. 2013; 31: 4387-93.
15. Ito T, Moroi Y, Oba J et al. The prognostic value of a reverse transcriptase-PCR assay of sentinel lymph node biopsy for patients with cutaneous melanoma: a single-center analysis in Japan. *Melanoma*

Res. 2012; 22: 38-44.
16. Kunte C, Geimer T, Baumert J et al. Prognostic factors associated with sentinel lymph node positivity and effect of sentinel status on survival: an analysis of 1049 patients with cutaneous melanoma. *Melanoma Res*. 2010; 20: 330-7.
17. Cadili A, Dabbs K. Predictors of sentinel lymph node metastasis in melanoma. *Can J Surg*. 2010; 53: 32-6.
18. Testori A, De Salvo GL, Montesco MC et al. Clinical considerations on sentinel node biopsy in melanoma from an Italian multicentric study on 1,313 patients (SOLISM-IMI). *Ann Surg Oncol* 2009; 16: 2018-27.
19. Kaur C, Thomas RJ, Desai N et al. The correlation of regression in pri mary melanoma with sentinel lymph node status. *J Clin Pathol* 2008; 61: 297-300.
20. Morris KT, Busam KJ, Bero S et al. Primary cutaneous melanoma with regression does not require a lower threshold for sentinel lymph-node biopsy. *Ann Surg Oncol* 2008; 15: 316-22.
21. Gillgren P, Brattström G, Frisell J et al. Effect of primary site on prog nosis in patients with cutaneous malignant melanoma. A study using a new model to analyse anatomical locations. *Melanoma Res* 2005; 15: 125-32.
22. Callender GG, Egger ME, Burton AL et al. Prognostic implications of anatomic location of primary cutaneous melanoma of 1mm or thicker. *Am J Surg* 2011; 202: 659-64.
23. Fadaki N, Li R, Parrett B, Sanders G et al. Is head and neck melanoma different from trunk and extremity melanomas with respect to sen tinel lymph node status and clinical outcome? *Ann Surg Oncol* 2013; 20: 3089-97.
24. Soudry E, Gutman H, Feinmesser M et al. "Gloves-and-socks" melanoma: does histology make a difference? *Dermatol Surg* 2008; 34: 1372-8.
25. Navalkissoor S, Bailey PS, Quigley AM et al. Sentinel node studies in truncal melanoma: does an increased number of draining basins correlate with an increased. risk of lymph metastasis? *Cancer Imaging*. 2012;12: 279-82.
26. Piñero A, de Torre C, Martínez-Escribano J et al. Multiple lymphatic basin drainage from cutaneous melanoma as a prognostic factor. *World J Surg*. 2012; 36(3): 579-85.
27. Chakera AH, Hesse B, Burak Z et al. EANM-EORTC general recommendations for sentinel node diagnostics in melanoma. *Eur J Nucl Med Mol Imaging* 2009; 36: 1713-42.
28. Sadeghi R, Forghani MN, Memar B et al. Comparison of pre-operative lymphoscintigraphy with inter-operative gamma probe and dye technique regarding the number of detected sentinel lymph nodes. *Hell J Nucl Med* 2009; 12(1): 30-2.
29. Mills JK, White I, Diggs B et al. Effect of biopsy type on outcomes in the treatment of primary cutaneous melanoma. *Am J Surg*. 2013; 205(5): 585-90.
30. Erman AB, Collar RM, Griffith KA et al. Sentinel lymph node biopsy is accurate and prognostic in head and neck melanoma. *Cancer* 2012; 118: 1040-7.
31. Caracò C, Marone U, Di Monta G et al. Surgical management of sentinel lymph node biopsy outside major nodal basin in patients with cutaneous melanoma. *Ann Surg Oncol* 2014; 21: 300-5.
32. Kidner TB, Yoon JL, Faries MB et al. Epitrochlear sentinel lymph nodes in melanoma: interval or independent? *Am Surg* 2012; 78: 702-5.
33. Hinz T, Ahmadzadehfar H, Wierzbicki A et al. Sentinel lymph node status as most important prognostic factor in patients with high-risk cutaneous melanomas (tumour thickness >4.00mm): outcome analysis from a single institution. *Eur J Nucl Med Mol Imaging* 2012; 39: 1316-25.
34. Bello DM, Chou JF, Panageas KS et al. Prognosis of acral melanoma: a series of 281 patients. *Ann Surg Oncol* 2013; 20: 3618-25.
35. Egger ME, McMasters KM, Callender GG et al. Unique prognostic factors in acral lentiginous melanoma. *Am J Surg* 2012; 204: 874-9.