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Original Articles

Sentinel node mapping with radiotracer alone in vulvar cancer: a five year single-centre experience and literature review

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Summary

Purpose of investigation: The pathologic status of lymph node represents the most important prognostic factor in vulvar cancer patients, but a complete groin dissection is associated with high post-operative morbidity. Sentinel lymph node (SLN) could be representative of the totality of regional lymph nodes and consequently its biopsy might have a significant impact on clinical management in vulvar cancer patients. *Materials and Methods:* From January 2006 to December 2010 45 patients with vulvar carcinoma are evaluated. Preoperative lymphatic mapping with technetium–99m–labeled nanocolloid was performed in all patients, followed by radioguided intraoperative detection. The detection rate is 100% of patients. All the SLNs were dissected separately for histopathological evaluation and a routine inguinofemoral lymphadenectomy was performed. *Results:* Nine patients had positive SLNs. In the remaining 36 patients with negative SLNs, one of them showed positive non-SLNs at histological examination. It was the only false negative case in the present series. *Conclusions:* Based on literature review, lymphoscintigraphy and sentinel node biopsy under gamma-detecting probe guidance offer a reliable and careful method to identify sentinel node in early vulvar cancer. Taking certain guidelines, SLN biopsy seems to be a safe alternative to inguinofemoral node dissection in order to reduce morbidity of surgical treatment.

Key words: Vulvar cancer; Sentinel-node biopsy; Squamous cells carcinoma; Radiotracer; Groin dissection.

Introduction

Vulvar cancer is a rare event in woman. It is responsible for three to four percent of all female neoplasia and for 0.3% of all female cancer deaths [1]. However in the United States 3,900 new cases and 920 deaths annually are reported [1, 2]. The mean age of presentation is about 70 years, affecting elderly women overall. The percentage of young patients is little in number, but has increased fourfold, caused by its association with human papillomavirus (HPV) infections [3]. An HPV association is present in about 40-60% of cases of vulvar cancer, and HPV 16 is the most frequent type of virus related to the tumoral lesions [4, 5]. Frequently the older patients have no evidence of precancerous lesion before diagnosis and they already have positive lymph node.

The majority of vulvar carcinomas are squamous cell carcinomas (SCC), which account for 90% of all vulvar carcinomas [6]. The pattern of spreading is principally via lymphatic channels to the inguinofemoral lymph nodes stations, especially when the depth of invasion exceeds one mm in the primary tumour [7]. The surgical approach with radical vulvectomy and bilateral inguinofemoral lymphadenectomy represent the standard treatment for patients affected by vulvar squamous cell carcinoma. While these techniques were performed, a long-term survival of approximately 70% resulted, with over 90% of Stage I patients alive in five years [8]. During lymphadenectomy, the Gynecologic Oncology Group (GOG) recommended a complete asportation of superficial and deep inguinal nodes, indeed the superficial inguinal node dissection alone is associated with a higher incidence of groin recurrence [9]. In effect, the rate of lymph node metastases in early-stage disease ranges from 25% to 35% of cases [10]. However, a large percentage of patients (ranging from 65% to 75%) will be at risk for significant morbidity when in absence of node metastasis. At short-term after vulvar radical surgery, the most important sequels remain infection and wound breakdown in 20% to 40% of patients, but at long-term the most significant consequence is lymphedema of the legs, present

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in 30% to 70% of patients [11]. Sentinel lymph node (SLN) biopsy seems to reduce incidence of long-time complication. In 1994 Levenback *et al.* were the first who applied SLN in squamous cell vulvar cancer [12-14].

The present study aims to assess the feasibility, efficacy, and accuracy of sentinel node detection with radiotracer alone, comparing it to the results reported in current literature.

Materials and Methods

From January 2006 to December 2010 a total of 48 women affected by tumour of the vulva were observed at the Department of Gynaecology of University of Genoa, Italy. Before initiation of the study, local Institutional Review Boards had approved the protocol. In 45 (94%) of these patients, the SLN technique was performed. Preoperatively, a diagnostic workup with complete anamnesis, physical and gynaecological examination, complete blood count and metabolic profile, electrocardiogram, thoracic x-ray, and abdomino-pelvic computer tomography were performed for all patients. The inclusion criteria were as follows: histologically confirmed diagnosis of invasive squamous vulvar cancer, no prior chemotherapy or radiotherapy, tumour lesions T1–T2, diameter of lesion ≤ four cm, clinically negative groins, absence of distant metastases, performance status < two according to the World Health Organization (WHO).

Exclusion criteria included: tumour lesions T3–T4,diameter of lesion > four cm and clinically positive groin nodes or distant metastasis.

The median age was 75.5 (range 53-86 years). At the presentation the symptoms were: itch in vulvar region (39% of cases), bleeding (30% of cases), vulvar pain (27% of cases), and only two patients presented only a small mass in genital area without other symptoms. All patients were informed of sentinel node procedure and explained the procedure in detail before the acceptation. At the time of surgery, 34 patients presented with an intact primary lesion, while 11 patients had residual scarring from a previous excisional biopsy.

At the clinical FIGO staging, 14 patients were classified as T1 Stage and 31 as T2 Stage.

The lesion was located medially in 32 cases and laterally in 13 cases. For midline lesion the authors imply a tumour whose median margin was within one cm from the midline. Table 1 presents the patients' characteristics.

In all cases a lymphoscintigraphy was performed 24 hour before surgery: colloid particles of human albumin were labelled with Tc^{99} and injected near the tumour site. For every patient 40 MBq in a volume of 10 ml were injected subcutaneously by four perilesional injections, according to biopsy results. After 30-60 minutes, planar scans of the vulvar and inguinal areas in anterior and lateral projections were obtained. At the end of the last scan, a cutaneous marker with a suitable pen was signed in correspondence to the first lymph node chronologically revealed by the gamma-camera.

At the time of surgery, the SLN was detected before groin dissection, using a gamma probe, inserted in a sterile glove. The sentinel node was detected at the probe projection, then an inguinal incision was made to excise the lymph node labelled as SLN. The sentinel node was evaluated outside the lesion to confirm the radioactivity level. After sentinel node biopsy, the area was checked again with the gamma probe for residual radioactivity. The nodes radioactivity level was evaluated, when more radioactive nodes were detected in the same area.

Table 1. — *Patients' characteristics.*

Characteristic	N° of patients	%
Tumor stage		
Stage I	14	31
Stage II	31	69
Tumor grading		
Grade 1	14	32
Grade 2	22	48
Grade 3	9	20
Location of primary tumor		
Lateral	13	29
Midline	32	71
Treatment of vulvar tumor		
Hemivulvar excision	13	29
Radical vulvectomy	32	71
Lymphadenectomy		
Unilateral	13	29
Bilateral	32	71

The highest tracer uptake node was named as first sentinel node, all the others were considered second nodes. The first sentinel node and the second nodes were sent separately at histological evaluation. When no residual radioactivity was detected, the superficial and deep inguinal lymphadenectomy was performed as usual.

Surgical vulvar treatment consisted of a standard radical vulvectomy for 32 patients or hemivulvar excision in the remaining 13 patients.

Histological evaluation

The sentinel node and the other removal node were histologically examinated. Every node was cut to obtain three- to four-mm thick sections and then fixed with formalin. Every sections was coloured with haematoxylin and eosin and, if there was a doubt, they were coloured again with cytokeratin 1% AE1:AE3 antikeratin solution for immunohistochemical reaction.

Results

In all 45 patients, lymphoscintigraphy detected at least one sentinel node, identified as "hot" lesion by the gamma probe. A total number of 77 groin were investigated. The final number was 100 sentinel nodes among 669 nodes removed. The mean number of node per groin was 8.7, while the mean number of sentinel node was 1.3 per groin. The detection rate respectively "per" patient and "per" groin was 100% and 98.5%, respectively.

In every patient all the non-sentinel inguinofemoral nodes (NSLN) were removed, independently from the status of the sentinel node.

In all 13 patients with unilateral lesion, the sentinel node was always found ipsilateral to the primary lesion: only an ipsilateral lymphadenectomy was performed for these patients, because no contralateral lymphatic drainage was demonstrated.

For 32 patients with midline lesion, a "hot" sentinel node was identified in both groins only five times: in the first

Table 2. — *Site of SLN identification.*

Primary tumor site	SLN[a] identification site Right groin	Left groin	Bilateral	Total number
Right lateral	6	-	-	6(13%)
Left lateral	-	7	-	7(16%)
Midline	16	11	5	32(71%)
Total	22(49%)	18(40%)	5(11%)	45(100%)

SLN[a] = sentinel lymph node

Table 3. — *Patients with lymph node metastasis.*

Patient	FIGO Stage	SLN[a] Site	Number of positive SLN	NSLN[b] Histological result	Number of Metastatic NSLN
1	II	Right	1	negative	0/6
2	II	Right	2	negative	0/4
3	II	Bilateral	2	positive	3/14
4	II	Left	1	positive	3/4
5	II	Left	4/6	positive	6/11
6	II	Left	1	negative	0/7
7	II	Right	2	negative	0/6
8	II	Bilateral	3	positive	4/15
9	II	Left	1	positive	3/7
Total positive SLN: 17				Total positive NSLN: 19	

SLN[a] = sentinel lymph node; NSLN[b] = non-sentinel lymph node

one both the sentinel nodes were metastatic but not the other inguinal nodes; in the other four case the two sentinel nodes were negative as the remaining nodes. In the other 27 patients with midline lesion, the sentinel node was found for 16 times on the right side and for 11 times on the left (Table 2). In all cases of midline lesion the bilateral groins, lymphadenectomy was performed.

At pathologic examination, 17 out of 100 sentinel nodes were positive for metastases. These 17 nodes belonged to nine different patients (Table 3).

In patient n.1 of Table 3, the right sentinel node was the only positive one and it was affected by micro-metastases. In this patient, the inguinal femoral nodes of the right side were removed, according to intraoperative radioactivity detection. Right NSLN were all negative for metastases. In spite of this fact, the patient had a contralateral recurrence two years later, with left inguinal nodes positive for metastases. It is possible that left inguinal nodes were positive at first time of surgery, but the authors did not find signal of any left sentinel node during surgery.

In patient n.5 of Table 3, the authors found a group of nodes identified as SLN at time of surgery. During histologic evaluation, this group was composed by six different nodes and four of these were involved with metastatic spread. Also ipsilateral NSLN were interested by wide metastases.

In a single case out of 45, there was a false negative. This patient was treated with hemivulvectomy and bilateral inguinofemoral lymphadenectomy in January 2006: at gamma probe, a right SLN was found intraoperatively. At histological evaluation, this sentinel node was negative, but another superficial right NSLN was positive.

On the left side no SLN was detected during surgery; a complete groin dissection was performed and final histology found one involved node. After radiotherapy, a vulvar recurrence appeared six months later and further surgery was done with radical wide excision and pelvic lymphadenectomy. Because of distant metastases and neoplastic spread, this patient died six months later.

No difference in sentinel node detection was revealed between patients who have previous vulvar surgery and those with an intact lesion.

The sentinel node detection rate in all patients was 100%, with sensitivity of 90%, and negative predictive value of 97% (Table 4)

Discussion

SLN mapping offers a promising minimally invasive alternative for surgical staging of vulvar cancer. Inguinal node status is considered the most important prognostic factor: groin recurrences are fatal in the most part of women. Actually, radical inguinofemoral lymph node dissection may be accompanied by an impressive morbidity: early postoperative complication as wound infection and wound breakdown or long-term morbidity, as lymph cysts, groin swelling, edema of the mons pubis, and the lower limb are reported in up to 85% of patients [15]. Chronic lymphedema is reported to occur in up to 30% of the patients [16]. Preoperative non-invasive or minimal invasive methods such as palpation, ultrasound, and ultrasound-guided fine-needle aspiration, as well as magnetic resonance imaging (MRI) and computed tomography (CT) or positron emission tomography (PET) -CT are not sensitive enough to exclude micrometastases in lymph nodes [17].

Table 4. — *Analysis of patients' data.*

Analysis by patients		TLN[a] status Positive	Negative	Total positive	Sensitivity (%)	NPV[b] 95%CI (%)	Detection Rate (%)
SLN status:	positive	9	0	9			
	negative	1	35	36			
Total		10	35	45	90	97 (±2.8)	100

SLN = sentinel lymph node; TLN[a] = Total lymph node; NPV[b] = negative predictive value

Table 5. — *Literature review.*

Author	Year	N° of patients	Detection techniques	Detection rate (%)	N° of false negatives
Levenback *et al.*[a]	1994-01	52	Blue dye	75	0
De Cesare *et al.* [19]	1997	10	Radioisotope	100	0
De Hullu *et al.*[b]	1998-04	59	Combined	100	0
Ansink *et al.* [22]	1999	51	Blue dye	82	2
De Cicco *et al.* [23]	2000	37	Radioisotope	100	0
Sideri *et al.* [24]	2000	44	Radioisotope	100	0
Sliutz et al. [25]	2002	26	Combined	100	0
Puig-Tintorè et al. [26]	2003	26	Combined	96	0
Moore *et al.* [27]	2003	21	Combined	100	0
Merisio *et al.* [28]	2005	20	Radioisotope	100	1
Nyberg *et al.* [29]	2007	47	Combined	98	1
Hampt *et al.*[30]	2008	127	Combined	98	3
Achimas-Cadariu *et al.* [31]	2009	59	Radioisotope	94	0
Levenback *et al.* [32]	2009	515	Blue dye vs Radiois	79 vs 96	-
Lindell *et al.* [33]	2010	77	Blue dye vs Radiois	94 vs 98	2
Radziszewski *et al.* [34]	2010	62	Blue dye vs Radiois.	76 vs 99	7
Ennik *et al.* [35]	2011	65	Combined	94	0
Levenback *et al.* [36]	2012	452	Combined	92.5	8
Zekan *et al.* [37]	2012	25	Radioisotope	100	1

[a] Combination of three reports [12, 13, 14]; [b] Combination of two reports [18, 20].

The SLN concept, validated in melanoma and breast cancer, affords the possibility of avoiding unnecessary lymphadenectomies. SLN technique could provide an opportunity to clearly identify the nodes most likely to harbour metastases. The best method to identify sentinel nodes seems the use of the hand-held gamma probe with preoperative lymphoscintigraphy, according to de Hullu *et al.* and De Cesare *et al.* [18, 19]. Using preoperative lymphoscintigraphy and intraoperative gamma-probe, the present authors reached a 100% identification rate in this series (Table 5). It seems clear that lymphoscintigraphy ahead of surgery aids in the process of node localization and significantly lowers the learning curve against dye alone technique [20]. Especially in the case of medially situated tumors, where the lymphatic drainage could be unilateral or bilateral, lymphoscintigraphy offers a reliable preoperative estimate of the location and number of the SLNs [18, 19, 21-29]. As seen in the present study, the primary site of the tumor affects the SLN identification rate: the only true false negative case was in a patient with a medial tumor. Levenback *et al.* demonstrated that the identification rate is higher for unilateral than for midline tumors (90% vs 69%, respectively) [14]. Hampl *et al.* reported three cases with false negative SLNs in tumors located in the midline and a single SLN detected, one on each side [30]. Nonetheless, the incidence of midline lesion in the present series points indicates the problem to treat this kind of lesion. Theoretically, surgeons have to investigate both groins, also when bilateral drainage is not demonstrated. Louise-Sylvestre *et al.* reported three of 13 patients with midline lesions and unilateral drainage; in fact with disease in the contralateral nodal basin [38]. Comparing to other studies, it appears that the further the lesion is localized from the midline, the less likely an SLN will be identified in the contralateral groin [39].

The same problem is faced with multifocal lesions: they must be considered as midline lesions and sentinel node identification must be performed in both groins, to avoid false negative of SLN biopsy [40].

According with Ennik *et al.,* as in the present series, there was no evident difference in sentinel node detection among patients who have previously vulvar surgery and patients with intact lesion [35].

A false negative case is present in this series: Raspagliesi *et al.* were the first reporting a false negative case using lymphoscintigraphy [41]. The present authors' failure could be given to different reasons:

- at histological evaluation metastases have been found in bilateral inguinofemoral nodes. It indicates that metastatic spread could be change the anatomic conformation of lymphatic channels; superficial nodes could be totally replaced by metastases and a lymphatic stasis - that did not allow radiotracer spread-could be created;
- histological evaluation demonstrated a premature lymphovascular spreading;
- tumour characteristics, as midline position, large dimension (three cm of highest diameter) associated to high body mass index (BMI = 42,2) of the patient may have affected SLN detection.

As reported by other authors in breast cancer [42] or in vulvar cancer patients [28, 38], these features may render more difficult the detection of SLN.

In this time the present authors are in accordance to the recently published statement of the expert-panel which has been formed during the 6th Biennial International Sentinel Node Society Meeting in February 2008. SLN biopsy could be performed only in patients with tumours smaller than four cm and with no clinical suspicious nodes. In case of a midline tumor, a SLN should be identified in both groins. If the SLN is not identified or any doubt exists regarding the correlation between a preoperative lymphoscintigram and the operative findings, then the SLN biopsy procedure should be abandoned and lymphadenectomy performed.

Gynecologic oncologists should perform a certain learning curve (at least ten groin dissections), in a contest of multidisciplinary group, with successful identification of the SLN and no false negative, before recommending SLN biopsy alone [34,40].

In the end, ultrastaging may be recommended for all removed SLNs, especially to identify micrometastases; it has been shown that when SLN harbor only micrometastases (< two mm focus of metastatic cells), non-sentinel nodes from the same inguinal basin will be negative for metastatic disease [43]. When a micrometastases is detected in a SLN, a complementary groin dissection or postoperative inguinal radiotherapy must be performed.

The recent creation of multicentric trials gave us more reliable results. In this regard, a very important European study was conducted in different centres of Netherlands, Belgium, Italy, and Germany: it suggested that sentinel node dissection, performed by a quality-controlled multidisciplinary team, should be part of the standard treatment in selected patients with early-stage vulvar cancer [40]. Indeed this observational study (GROINSS-V) prove that in a manner of 403 patients with FIGO I and II Stages with squamous cell carcinomas of the vulva ≤ four cm in size, in case of a negative sentinel node, a complete inguinofemoral lymphadenectomy can be omitted. The data analysis showed that, in a follow-up after two years, in the presence of unifocal tumor, the percentage of inguinal relapse was 2.3% (six groin recurrences in 259 patients) and a three-year survival rate of 97% [40]. However, Oonk *et al.* showed that the prognosis for patients with sentinel node metastasis larger than two mm is poor and they suggested new treatment regimens for these patients [44].

The result of GOG protocol 173, begun in 2000, were presented by Levenback *et al.* at the annual meeting of the American Society of Clinical Oncology (ASCO) in 2009 [32]. In 411 patients with vulvar cancer and clinically negative groin status, 282 patients had negative nodes and 129 patients had positive nodes. The study showed a sensitivity of 89.9%, a negative predictive value of 95.6% and a false-negative predictive value of 4.4%.

In terms of morbidity related to surgical procedure, two studies showed that the SLN biopsy was associated with significantly less short-term and long-term morbidity, in comparation to complete lymph node dissection.

At the analysis of short-term morbidity, Van der Zee *et al.* reported that wound breakdown occurred in 11.7% and cellulitis in 4.5% in SLN patients vs respectively, 34% and 21.3% in inguinal lymphadenectomy patients ($p < 0.001$) [40]. Also, the time of hospitalisation is significantly shorter in event of SLN biopsy when compared to inguinal lymphadenectomy (8.4 days vs 13.7 days ($p < 0.001$) [40].

At the analysis of long-term morbidity, Van der Zee *et al.* showed that the incidence of lymphedema and recurrent erysipelas decreased significantly from 25.2% to 1.9% and from 16.2% to 0.4%, respectively, in SLN biopsy patients vs inguinal lymphadenectomy ($p < 0.001$) [40]. Relative to rate of lymphedema, Johann *et al.* also reported that this rate decreased from 39% to 13% in SLN biopsy patients [45].

Conclusion

The authors believe that sentinel node procedure in the management of early-stage vulvar cancer could decrease morbidity without compromising groin recurrence or survival rates, while taking certain guidelines into account.

References

[1] Tropè C.G., Makar A.P.H.: "Epidemiology, etiology, screening, prevention and diagnosis in female genital cancer. Review". *Curr. Opin. Oncol.*, 1991, *3*, 908.

[2] American Cancer Society: "Cancer Facts & Figures 2010". Atlanta: American Cancer Society, 2010. Available at: http://www.cancer.org/acs/groups/content/@nho/documents/document/acspc-024113.pdf

[3] Hampl M., Deckers-Figiel S., Hampl J.A., Rein D., Bender H.G. : "New aspects of vulvar cancer: changes in localization and age of onset." Gynecol Oncol, 2008, 109, (3),340.

[4] Hampl M., Sarajuuri H., Wentzensen N., Bender H.G., Kueppers V.: "Effect of human papillomavirus vaccines on vulvar, vaginal and anal intraepithelial lesions and vulvar cancer." Obstet Gynecol, 2006, 108,1361.

[5] De Vuyst H., Clifford G.M., Nascimento M.C., Madeleine M.M., Franceschi S.: "Prevalence and type distribution of human papillomavirus in carcinoma and intraepithelial neoplasia of the vulva, vagina and anus: a meta-analysis". *Int. J. Cancer*, 2009, *124*, 1626.

[6] Hacker N.F.: "Vulvar cancer." *In:* Berek J.S., Hacker N.F. (eds). *Practical gynecologic oncology*. 3rd ed. Baltimore, MD: Williams & Wilkins, 2000, 553.

[7] Parry-Jones E.: "Lymphatics of the vulva." *J. Obstet. Gynaecol. Br. Emp.*, 1963, *70*, 751.

[8] Van der Velden K.: "Some aspects of the management of squamous cell carcinoma of the vulva". *In:* Shaw R.W., Luesley D., Monga A.K. (eds). *Gynaecology*, 4th ed. Edinburgh: Churchill Livingstone, 2011.

[9] Stehman F., Bundy B., Dvoretsky P.M., Creasman W.T.: "Early stage I carcinoma of the vulva treated with ipsilateral superficial inguinal lymphadenectomy and modified radical hemivulvectomy: a prospective study of the Gynecologic Oncology Group". *Obstet. Gynecol.*, 1992, *79*, 490.

[10] De Hullu J.A., Hollema H., Lolkema S., Boezen M., Boonstra H., Burger M.P.: "Vulvar carcinoma: the price of less radical surgery". *Cancer*, 2002, *95*, 2231.

[11] Gaarenstroom K.N., Kenter G.G., Trimbos J.B., Agous I., Amant F., Peters A.A., Vergote I.: "Postoperative complication after vulvectomy and inguinofemoral lymphadenectomy using separate groin incision". *Int. J. Gynecol. Cancer*, 2003, *13*, 522.

[12] Levenback C., Burke T.W., Gershenson D.M., Morris M., Malpica A., Ross M.I.: "Intraoperative lymphatic mapping for vulvar cancer". *Obstet. Gynecol.*, 1994, *84*, 163.
[13] Burke T.W., Levenback C., Coleman R.L., Malpica A., Lucas K.R., Gershenson D.M.: "Potential application of intraoperative lymphatic mapping in vulvar cancer". *Gynecol. Oncol.*, 1995, 59, 216.
[14] Levenback C., Coleman R.L., Burke. T.W., Bodurka-Bevers D., Wolf J.K., Gershenson DM: "Intraoperative lymphatic mapping and sentinel node identification with blue dye in patients with vulvar cancer." *Gynecol. Oncol.*, 2001, *84*, 449.
[15] Rouzier R., Haddad B., Dubernard G.: " Inguinofemoral dissection for carcinoma of the vulva: effect of modifications of extent and technique on morbidity and survival". *J. Am. Coll. Surg.*, 2003, *196*, 442.
[16] Gould N., Kamelle S., Tillmanns T.: "Predictors of complications after inguinal lymphadenectomy". *Gynecol. Oncol.*, 2001, *82*, 329.
[17] Oonk M.H., Hollema H., De Hullu J.A., van der Zee A.G.: "Prediction of lymph node metastases in vulvar cancer: a review". *Int. J. Gynecol. Cancer*, 2006, *16*, 963.
[18] De Hullu J.A., Doting E., Piers D.A., Hollema H., Aalders J.G., Koops H.S. et al.: "Sentinel lymph node identification with technetium-99m-labeled nanocolloid in squamous cell cancer of the vulva". *J. Nucl. Med.*, 1998, *39*, 1381.
[19] De Cesare S.L., Fiorica J.V., Roberts W.S., Reintgen D., Arango H., Hoffman M.S., *et al.*: "A pilot study utilizing intraoperative lymphoscintigraphy for identification of the sentinel lymph nodes in vulvar cancer". *Gynecol. Oncol.*, 1997, *66*, 425.
[20] Coleman R.L.: "Vulvar Lymphatic Mapping: Coming of Age? Editorial". *Ann. Surg. Oncol.*, 2002, *9*, 823.
[21] De Hullu J.A., Oonk M.H.M., Ansink A.C., Hollema H., Jager P.L., van der Zee A.G.: "Pitfalls in the sentinel lymph node procedure in vulvar cancer". *Gynecol. Oncol.*, 2004, 94, 10.
[22] Ansink A.C., Sie-Go D.M., van der Velden J., Sijmons E.A., de Barros Lopes A., Monaghan J.M., et al.: "Identification of sentinel lymph nodes in vulvar carcinoma patients with the aid of a patent blue V injection: a multicenter study." Cancer, 1999, 86, 652.
[23] De Cicco C., Sideri M., Bartolomei M., Grana C., Cremonesi M., Fiorenza M., *et al.*: "Sentinel node biopsy in early vulvar cancer". *Br. J. Cancer*, 2000, *82*, 295.
[24] Sideri M., De Cicco C., Maggioni A., Colombo N., Bocciolone L., Trifirò G. et al.: " Detection of sentinel nodes by lymphoscintigraphy and gamma probe guided surgery in vulvar neoplasia." *Tumori*, 2000, *86*, 359.
[25] Sliutz G., Reinthaller A., Lantzsch T., Mende T., Sinzinger H., Kainz C., Koelbl H.: "Lymphatic mapping of sentinel nodes in early vulvar cancer". *Gynecol. Oncol.*, 2002, *84*, 449.
[26] Puig-Tintoré L.M., Ordi J., Vidal-Sicart S., Torné A., Pahisa J., Iglesias X.: "Further data on the usefullness of sentinel lymph node identification and ultrastaging in vulvar squamous cell carcinoma". *Gynecol. Oncol.*, 2003, *88*, 29.
[27] Moore R.G., DePasquale S.E., Steinhoff M.M., Gajewski W., Steller M., Noto R., Falkenberry S. "Sentinel node identification and the ability to detect metastatic tumor to inguinal lymph nodes in squamous cell cancer of the vulva." *Gynecol. Oncol.*, 2003, *89*, 475.
[28] Merisio C., Berretta R., Gualdi M., Pultrone D.C., Anfuso S., Agnese G., *et al.*: "Radioguided sentinel lymph node detection in vulvar cancer". *Int. J. Gynecol. Cancer*, 2005, *15*, 493.
[29] Nyberg R.H., Iivonen M., Parkkinen J., Kuoppala T., Mäenpää J.U.: "Sentinel node and vulvar cancer: a series of 47 patients". *Acta Obstet. Gyn. Scan*, 2007, *86*, 615.
[30] Hampl M., Hantschmann P., Michels W., Hillemanns P. German Multicenter Study Group.: "Validation of the accuracy of the sentinel lymph node procedure in patients with vulvar cancer: results of a multicenter study in Germany". *Gynecol. Oncol.*, 2008, *111*, 282.
[31] Achimas-Cadariu P., Harter P., Fisseler-Eckhoff A., Beutel B., Traut A., Du Bois A.: "Assessment of the sentinel lymph node in patients with invasive squamous carcinoma of the vulva". *Acta Obstet. Gynecol. Scand.*, 2009, *88*, 1209.
[32] Levenback C.F.: "Sentinel node (SN) biopsy in patients with vulvar cancer: a Gynecologic Oncology Group (GOG) study". *J. Clin. Oncol.*, 2009, *27*, 15s.
[33] Lindell G., Jonsson C., Ehrsson R.J., Jacobsson H., Danielsson K.G., Källström B.N., Larson B.: "Evaluation of preoperative lymphoscintigraphy and sentinel node procedure in vulvar cancer". *Eur. J. Obstet. Gynecol. Reprod. Biol.*, 2010, *152*, 91.
[34] Radziszewski J., Kowalewska M., Jedrzejczak T., Kozlowicz-Gudzinska I., Nasierowska-Guttmejer A., Bidzinski M., *et al.*: "The accuracy of the sentinel lymph node concept in early stage squamous cell vulvar carcinoma". *Gynecol. Oncol.*, 2010, *116*, 473.
[35] Ennik T.A., Allen D.G., Bekkers R.L., Hyde S.E., Grant P.T.: "Effect of previous surgery on the detection of sentinel node in women with vulvar cancer". *Int. J. Gynecol. Cancer*, 2011, *21*, 1679.
[36] Levenback C.F., Ali S., Coleman R.L., Gold M.A., Fowler J.M., Judson P.L., *et al.*: "Lymphatic mapping and sentinel lymph node biopsy in women with squamous cell carcinoma of the vulva: a Gynecologic Oncology Group study". *J. Clin. Oncol.*, 2012, *30*, 3786.
[37] Zekan J., Mutvar A., Huic D.,Petrovic D., Karelovic D., Mitrovic L.: "Reliability of sentinel node assay in vulvar cancer: the first Croatian validation trial". *Gynecol. Oncol.*, 2012, *126*, 99.
[38] Louise-Sylvestre C., Evangelista E., Leonard F., Itti E., Meignan M., Paniel B.J.: "Sentinel node localization should be interpreted with caution in midline vulvar cancer". *Gynecol. Oncol.*, 2005, *97*, 151.
[39] Hauspy J., Beiner M., Harley I., Ehrlich L., Rasty G., Covens A.: "Sentinel lymph node in vulvar cancer". *Cancer*, 2007, *5*, 1015.
[40] Van der Zee A.G., Oonk M.H., De Hullu J.A., Ansink A.C., Vergote I., Verheijen R.H., *et al:*. "Sentinel node dissection is safe in the treatment of early-stage vulvar cancer". *J. Clin. Oncol.*, 2008, *6*, 884
[41] Raspagliesi F., Ditto A., Fontanelli R., Maccauro M., Carcangiu M.L., Parazzini F., *et al.*: "False-negative sentinel node in patients with vulvar cancer: a case study". *Int. J. Gynecol. Cancer*, 2003, *13*, 361.
[42] Cox C.E., Dupont E., Whitehead G.F., Ebert M.D., Nguyen K., Peltz E.S., *et al.*: "Age and body mass index may increase the chance of failure in sentinel lymphnode biopsy for women with breast cancer". *Breast J.*, 2002, *8*, 88.
[43] Robison K., Steinhoff M.M., Granai C.O., Brard L., Gajewski W., Moore R.G.: "Inguinal sentinel node dissection versus standard inguinal node dissection in patients with vulvar cancer: a comparison of the size of metastases detected in inguinal lymph nodes". *Gynecol. Oncol.*, 2006, *101*, 24.
[44] Oonk M.H., van Hemel B.M., Hollema H., de Hullu J.A., Ansink A.C., Vergote I., *et al.*:" Size of sentinel node metastasis and chances of non-sentinel-node involvement and survival in early stage vulvar cancer: results from GROINSS-V, a multicentre observational study". *Lancet Oncol.*, 2010, *11*, 646.
[45] Johann S., Klaeser B., Krause T., Mueller M.D..: "Comparison of outcome and recurrence-free survival after sentinel lymph node biobsy and lymphadenectomy in vulvar cancer". *Gynecol. Oncol.*, 2008, *110*, 324.

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