Copy number losses define subgroups of dedifferentiated liposarcoma with poor prognosis and genomic instability

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Translational relevance

Dedifferentiation of a well-differentiated liposarcoma (WDLS) is associated with the acquisition of metastatic potential and high rates of local recurrence. The genetic events that drive progression from WDLS to dedifferentiated liposarcoma (DDLS) remain undiscovered. Using comparative genomic hybridization, common, specific progression-associated copy number alterations (CNAs) were identified in 79% of DDLS but were rarely found in WDLS. These recurrent CNAs associate with tumor morphology, patient outcomes and expression of potential tumor suppressors/oncogenes. Our findings form the basis of a model describing liposarcoma progression and dedifferentiation. This model enables clinicians to better stratify DDLS patients for clinical trials and will improve outcome prediction for the individual patient.
Abstract

Purpose: Molecular events underlying progression of well-differentiated liposarcoma (WDLS) to dedifferentiated liposarcoma (DDLS) are poorly defined. This study sought to identify copy number alterations (CNAs) associated with dedifferentiation of WDLS, with DDLS morphology, and with patient outcomes.

Experimental Design: 55 WDLS and 52 DDLS were analyzed using Agilent 244K comparative genomic hybridization and Affymetrix U133A expression arrays. CNAs were identified by RAE analysis. Thirty-nine of the DDLS specimens were categorized morphologically by a single pathologist.

Results: Nine regions of CNA were identified as recurrent in DDLS but not WDLS; 79% of DDLS had at least one of these CNAs. Loss of the chromosome segment 11q23-24, the most common event, was observed only in DDLS that morphologically resembled the genomically complex sarcomas undifferentiated pleomorphic sarcoma and myxofibrosarcoma. 11q23-24 loss was itself associated with increased genomic complexity in DDLS. Loss of 19q13, but not 11q23-24, was associated with poor prognosis. Median disease-specific survival was shorter for patients with 19q13 loss (27 months) than for patients with diploid 19q13 (>90 months; p<0.0025), and 19q13 loss was associated with local recurrence (HR 2.86, p=0.013). Common copy number losses were associated with transcriptional downregulation of potential tumor suppressors and adipogenesis-related genes (e.g., EL24 and CEBPA).

Conclusions: Dedifferentiation of WDLS is associated with recurrent CNAs in 79% of tumors. In DDLS, loss of 11q23-24 is associated with genomic complexity and distinct morphology while loss of 19q13 predicts poor prognosis. CNAs in liposarcoma improve
risk stratification for patients and will help identify potential tumor suppressors driving liposarcoma progression.
Introduction

Liposarcoma is the most common type of soft tissue sarcoma, accounting for ~20% of the tumors in adults. Liposarcoma shows histologic and genetic variability, and has been subdivided into three main biological groups encompassing five subtypes. These are (1) well-differentiated/dedifferentiated (WDLS/DDLS), (2) myxoid/round cell and (3) pleomorphic liposarcoma. WDLS contains regions of mature adipose tissue, interspersed with lipoblasts and atypical stromal cells. DDLS always occurs in the context of a WDLS and represents progression to high-grade disease. Histologically, DDLS lacks mature adipose cells and can have variable microscopic appearance (1).

Although WDLS and DDLS are often surgically resectable, retroperitoneal tumors frequently recur, and 60% of patients eventually die of disease. Dedifferentiation is a poor prognostic feature, as DDLS has metastatic potential and a more aggressive local disease course; DDLS is 6 times as likely to recur as WDLS (2-4). Radiation and chemotherapy have a limited ability to control inoperable or metastatic disease. Therefore, it is essential that we improve our understanding of the molecular events underlying liposarcoma progression so as to develop novel targeted therapies.

Changes in genome structure resulting in copy number alterations (CNAs) occur commonly in human cancer and result in aberrant expression of putative oncogenes and tumor suppressors (5). Among WDLS and DDLS, over 90% of tumors have amplification of the chromosome segment 12q13-15 (1, 6, 7). The 12q13-15 amplicon carries supernumerary copies of the oncogenes CDK4 and MDM2 (8, 9). Expression of
CDK4 and MDM2 is essential for survival of liposarcoma cells in vitro, and therefore the 12q13-15 amplicon is thought to play a role in initiation of liposarcomagenesis. DDLS is genomically complex, however, and additional CNAs often affect multiple chromosomes (1). One such CNA affects 1p32 and is associated with JUN amplification, an event that may contribute to dedifferentiation, though it is not sufficient to induce progression (10, 11).

In this study, we aim to improve our understanding of the mechanisms underlying progression thorough systematic analysis of CNAs associated with dedifferentiation. A direct, unbiased comparison of CNAs in DDLS versus WDLS has not previously been performed. Using array comparative genomic hybridization (CGH), we identify specific CNAs that are common in DDLS but not in WDLS. We relate chromosome losses to (1) patient prognosis, (2) microscopic appearance of DDLS, (3) genomic complexity, and (4) expression of potential tumor suppressors. In doing so, we provide a framework for understanding progression in a representative cohort of WD/DDLS.

Methods and Materials

Patient population and sample acquisition

Fifty-five WDLS and 52 DDLS samples were collected from patients treated surgically from 2002 to 2009. Informed consent was obtained per a protocol approved by the institutional review board. Cryomolds were prepared from tumor samples, and a
dedicated sarcoma pathologist examined H&E sections from both cryomold surfaces to assess liposarcoma subtype.

**DNA/RNA isolation and array preparation**

For nucleic acid extraction, we selected cryomolds containing a single subtype with >75% cellularity. Cryomolds were macrodissected to remove normal, fibrotic, inflammatory, and necrotic tissue.

Genomic DNA and RNA were prepared as previously described using RNeasy Lipid Tissue Mini and DNeasy Blood and Tissue kits (QIAGEN) (6, 12). cDNA was synthesized in the presence of oligo(dT)$_{24}$-T7 from Genset, then cRNA was prepared using biotinylated UTP and CTP prior to hybridization to HG U133A arrays (Affymetrix) (12). Genomic DNA was analyzed with Agilent 244K (n=44 WDLS, 40 DDLS) or 1M (n=11 WDLS and 12 DDLS) oligonucleotide arrays according to manufacturer’s specifications. Competitive hybridization was performed in the presence of a DNA reference set (Human Genomic DNA from blood [buffy coat], Roche Applied Science).

**GC-normalization and RAE analysis of array CGH samples**

Analysis of array CGH data used a customized normalization method. Signals were expected (1) to be highly correlated between CGH probes targeting adjacent regions in the genome and (2) to show significant correlation with GC content. As no software was readily available to address these unique aspects of CGH data, novel algorithms for
normalization were developed; complete methods and code are available at http://cbio.mskcc.org/CGCC.

To identify CNAs, the normalized dataset was processed using a custom pipeline, which performs the standard circular binary segmentation from the R/bioconductor DNAcopy library, and then processes all samples using the RAE algorithm (6, 13). The gene-based and region of interest (ROI) copy number calls of -2, -1, 0, 1, or 2 (deletion, loss, copy-neutral, gain, amplification; as assessed by RAE) were computed on three samples cohorts (DDLS only, WDLS only, and all samples). Copy number changes for each gene and ROI were converted from the 5-level RAE scale to a 3-level scale: loss, neutral, or gain (-1, 0, 1). Fisher’s exact test was used to compare copy number between DDLS and WDLS for each ROI. In many cases, ROIs adjacent along the genome had nearly identical copy number calls; in such cases, ROIs were further collapsed into a single event for subsequent analysis. The ANOVA F-test was used to identify genes that were differentially expressed between copy number classes (i.e., -1, 0, or 1) and whose sign of change matched the copy number event. Genes of interest were defined as those having at least 2-fold change in expression for DDLS with a given CNA vs DDLS without the event or vs WDLS. Genomic complexity was calculated for each sample as the percentage of the genome altered (as scored by RAE).

Histology and survival analyses
In analyses of both histology and survival, CNAs were considered by chromosome arm. A given arm was scored as having a specific CNA if the three most common ROIs identified on that arm were all observed in the tumor.

Paraffin blocks from the original surgical specimens were re-reviewed; presence of necrosis and tumor grade (regions of low-grade tumor, high-grade or both) were documented. Tumors with greater than 5 mitoses per 10 high-power field were characterized as high grade. The percentage of each lesion consisting of DDLS histology was recorded. Size of the patient’s primary tumor was obtained from a prospectively collected database of sarcoma patients treated surgically at our institution. DDLS were characterized as resembling undifferentiated pleomorphic sarcoma (UPS, also referred to as malignant fibrous histiocytoma or MFH), myxofibrosarcoma, spindle cell sarcoma not otherwise specified (resembling adult fibrosarcoma), small blue round cell tumors, or solitary fibrous tumor. Areas of divergent morphology (osteosarcoma or rhabdomyosarcoma) and inflammatory infiltrate were noted. Groups defined by individual histologic characteristics or morphology were tested for differences in CNAs using Fisher’s exact test, and tested for differences in the percentage of the genome altered using the Kruskal-Wallis test for non-parametric data. In a subset of analyses, UPS and myxofibrosarcoma were considered as one morphologic subgroup since these tumors can represent a spectrum of disease and no consensus has been reached on definitions of the two histologies.

Patient outcomes were prospectively collected from the time of surgical resection and were retrospectively reviewed. Local-recurrence–free survival (LRFS), distant-
recurrence–free survival (DRFS), and disease-specific survival (DSS) were calculated from the date of surgical resection. Effect of CNA and clinical characteristics on DSS was evaluated using the Cox regression model and Kaplan-Meier analyses. For analysis of LRFS and DRFS, patients were stratified by the presence of individual CNAs and analyzed by the competing risk survival analysis methods (Gray’s test); the competing event was death (14). For multivariate analyses, competing risk regression models were used (15).

Results

Common chromosomal gains and losses characterize the transition from WDLS to DDLS

CNAs were analyzed in 55 WDLS and 52 DDLS samples (Tables 1, S1, S2). RAE analysis identified CNAs on 11 chromosomes in WDLS (Figure 1a; Table S3). These included the classic 12q13-15 amplification encompassing CDK4, MDM2, and HMGA2. These genes were amplified in 95%, 87%, and 76% of WDLS, respectively. Only one WDLS had no 12q amplification. Less common CNAs in WDLS included gains at 1q21-25 and 7q34 and copy number losses at 1q25-44, 8q24, 9p24, 10q26, 11p13-15, 13q14, 13q31, 18q23, and 22q11-13. Copy number alterations observed in specimens of WDLS regions within tumors classified histologically as DDLS (27% of WDLS specimens) were compared to those observed in samples from tumors classified histologically as WDLS. Copy number alterations in these subsets were not significantly different.
As a group, DDLS tumors had more complex genomic alterations than did WDLS. CNAs affected on average 5.7% of the genome in WDLS (±7.0% [standard deviation]; range 0.1% to 36%), versus 21% (±16%; range 1% to 62%; p<0.001) in DDLS (Figures 1b, S1). In DDLS, CNAs were identified on 21 chromosomes (Table S4). To assess which CNAs may be important in progression of WDLS to DDLS, the two tumor types were directly compared. CNAs with at least 20 percentage points higher frequency in DDLS vs WDLS and found to be significantly different between the two groups (FDR<0.001) were defined as progression-associated CNAs. Nine such CNAs were observed: losses centered at 3p14-21, 3q29, 9p22-24, 10p15, 11q23-24, 17q21, and 19q13 and gains at 17p11 and 20q11 (Tables 2, S5). Seventy-nine percent of DDLS tumors had at least one progression-associated CNA. In nine cases, WDLS and DDLS samples from the same tumor or serial resections from the same patient were available for analysis (Table S1); five of the DDLS but none of the WDLS in this subset had at least one of these CNAs. Of the progression-associated CNA, 11q23-24 loss was the most commonly observed. This loss was noted in 44% of DDLS compared to 4% of WDLS (FDR=0.0002). The previously observed amplification of 1p32, which includes the JUN locus, was observed in 15 DDLS (28%) but also in 5 WDLS (9%; FDR = 0.03) so did not meet our criteria for a progression-associated CNA.

**Copy number alterations correlate with tumor histology and genomic instability**

The microscopic appearance of DDLS exhibited significant heterogeneity. Thirty-nine DDLS samples were available for pathologic re-review. The morphology most
commonly appeared similar to UPS, myxofibrosarcoma, spindle cell sarcoma not otherwise specified (similar to adult fibrosarcoma), or small blue round cell tumors (Figure S2). One tumor appeared similar to solitary fibrous tumor and one had areas morphologically reminiscent of desmoid-type fibromatosis. Prominent inflammatory components and divergent morphologies (osteosarcoma, rhabdomyosarcoma) were each observed in 4 tumors. In 23 tumors (59%), the DDLS component had a variegated morphology (Table S6).

Given the variability of both CNAs and morphology observed in DDLS, we hypothesized that histologic morphology may reflect the presence of specific CNAs. No CNAs were significantly associated with primary tumor size, tumor grade, or presence of necrosis in the specimen (as documented in Table S6), but 17p11 gain in DDLS was associated with an inflammatory UPS-like morphology (p=0.028). In addition, progression-associated CNAs affecting 11q23-24, 9p22-24, 10p15, 17p11, 17q21, and 20q11 were identified only in DDLS with regions appearing similar to UPS and myxofibrosarcoma, not in DDLS that had only spindle cell not otherwise specified, small blue round cell, or solitary fibrous tumor-like morphologies. This association was significant in the case of 11q23-24 loss (Tables 3, S6; p<0.02).

UPS and myxofibrosarcoma are genomically complex tumors (1), and analysis of DDLS that contained a component resembling UPS or myxofibrosarcoma showed that these tumors tended to have CNAs in a higher percentage of the genome (23±16%) than did other subtypes (11±5.6%), though this finding did not reach statistical significance.
Loss of 11q23-24, which, as noted above, was found only in this subset of DDLS, was also associated with increased genomic complexity; CNAs affected 29±15% vs 15±14% of the genome in DDLS with vs without 11q23-24 loss (p=0.002; Figure S3b). Loss of 11q23-24 was specifically associated with other progression-associated CNAs on 9p22-24, 10p15, and 17q21 (all p<0.03).

CNAs at 3p14-21, 3q29, and 19q13 were not associated with any histologic correlate or with genomic complexity (Table S6).

**Survival analysis**

We examined survival following R0 or R1 resection of primary DDLS or first local recurrence of DDLS. Median follow-up for this cohort of 40 patients was 2.3 years. Clinical variables (margin, gender, age, and presentation status) did not associate with DSS; however, loss of 19q13 was associated with significant reduction in DSS (HR 7.36, p=0.0025, Table 4, Figure 2). Loss of 19q13 was also associated with reduced LRFS (HR 2.86), as were 9p22-24 loss (HR 2.35), 17q21 loss (HR 2.75), R1 resection, and young age. On multivariate analysis of LRFS, loss of 19q13 and young age were independently predictive of shorter LRFS (HR 2.99 and 0.36, respectively; both p=0.01; Table S7). Because of the limited number of events in this cohort, only three clinicopathologic characteristics could be evaluated in a single analysis. When multivariate analyses including 19q13 loss and 17q21 or 9p22-24 loss were performed, the effect of 19q13 loss remained significant (HR 2.62, p=0.025 and HR 2.74, p=0.019, respectively).
For DRFS, R1 resection and 3q29 loss were associated with better outcomes. Strikingly, no patient with loss of 3q29 was found to develop distal metastases (n=16).

**Possible tumor suppressors on chromosomes 11 and 19**

To identify possible tumor suppressors affected by progression-associated CNAs in DDLS, we integrated CNA and gene expression data for 36 DDLS and 33 WDLS samples. 430 genes located within a CNA had a substantial expression change associated with dedifferentiation and/or with presence of the CNA (Table S8). The 11q23-24 loss spanned multiple genes that were underexpressed in DDLS compared to WDLS and are therefore potential tumor suppressors. Three of these (ZBTB16, PPP2R1B, and EI24) are known tumor suppressors in other cancers. Among DDLS samples, a CNA affecting EI24 was specifically associated with reduced EI24 expression; EI24 expression tended to be lower in DDLS tumors with 11q23-24 loss versus those with diploid 11q23-24 (Figure S4a).

The transition from WDLS to DDLS is characterized by downregulation of the normal adipogenesis program, and several genes that regulate this process were identified in regions of copy number loss. ASAM (CLMP, which encodes an adhesion protein implicated in adipocyte maturation) was the gene most commonly lost on 11q23-24, though loss of gene expression was not clearly associated with dedifferentiation or CNA. PLIN2 (perilipin 2, on 9p22), LIPE (hormone-sensitive lipase, on 19q13) and CEBPA (19q13) are also located within regions of chromosome loss and were under-expressed...
in DDLS compared to WDLS. The magnitude of CEBPA downregulation tended to be greater in tumors with CEBPA loss than in those without CEBPA loss (Figure S4b). DLAT and ACAD8, both genes involved in fatty acid metabolism, are located within the 11q23-24 region of loss and were downregulated in concordance with this CNA (i.e., more significantly in tumors with 11q23 loss versus those without the event; Table S8).

Discussion
While 12q13-15 amplification is associated with the initiation of WDLS, the molecular events that characterize progression to the DDLS histology were previously uncharacterized. In this study, we have assembled the largest data set describing genomic characteristics of WDLS and DDLS. Using array CGH, we demonstrated increased genomic complexity in DDLS versus WDLS and identified nine common progression-associated CNAs that are nearly specific to DDLS (versus WDLS). Our results correlated one of these events, 11q23-24 loss, with variations in the genomic complexity and in the morphologic phenotype of DDLS, while demonstrating that 19q13 loss is associated with LRFS and DSS. Each of these represents a novel finding in the description of liposarcomagenesis.

The most common progression-associated CNA involved variable lengths of 11q (centered at 11q23-24). Similar losses have been associated with melanoma and neuroblastoma, though interestingly gains, not losses, on 11q correlate with glioma progression (16-18). In DDLS, 11q23-24 loss was associated with greater genomic complexity. Tumors with 11q23-24 loss, compared to those without this loss, have
higher percentages of their genomes altered by CNAs and higher frequencies of other progression-associated CNAs. The genomic complexity associated with 11q23-24 was of particular interest in the context of our examination of tumor histology. The UPS and myxofibrosarcoma subtypes of soft tissue sarcoma are genomically complex. In DDLS, tumors that had components resembling UPS and myxofibrosarcoma tended to have increased genomic complexity compared to those that did not. Moreover, 11q23-24 loss was identified only in DDLS with UPS or myxofibrosarcoma-like components.

Chromosome region 11q23-24 carries several genes that were downregulated during dedifferentiation (e.g., ZBTB16, PPP2R1B, and EI24) and a subset involved in fatty acid metabolism (e.g., DLAT, ACAD8). It remains unclear, however, whether the loss of 11q23-24 is a result of genomic instability, or if a gene on the chromosome modulates genomic stability. The 11q23 gene H2AX is thought to play a role in regulation of genomic stability in neuroblastoma (17), but we were unable to detect downregulation of H2AX in DDLS with the 11q23-24 loss either by U133A array or quantitative RT-PCR (not shown). Many of the genes on 11q23-24 whose expression is downregulated in conjunction with loss of copy number are poorly characterized, and could modulate chromosome integrity.

Loss of 19q13, although associated with progression of WDLS, was not associated with an overall increase in genomic complexity, with the other progression-associated CNAs, or with DDLS histology. However, 19q13 loss was closely associated with patient outcomes. In this relatively homogenous group of tumors (in terms of size, site and
margin status), outcome could not be clearly stratified based on clinical factors, but loss of 19q13 was associated with both LRFS and DSS. Though 3q29, 9p22-24, and 17q21 loss were associated with LRFS or DRFS, these events did not seem to alter DSS; this finding was specific for 19q13 loss.

Tumors with 19q13 loss (and concomitant CEBPA loss) tended to have lower expression of CEBPA, a gene recently identified as a tumor suppressor in DDLS (Wu YV et al, manuscript submitted). Lower expression of CEBPA is also associated with worse outcomes among patients with liposarcoma of any subtype, and this gene contributes to a multigene predictor designed to identify patients with high-risk liposarcoma (19). The association we observed between CEBPA loss and CEBPA levels suggests that expression of this gene may be a marker of 19q13 loss or, given its tumor suppressor effects in DDLS in vitro, may partially mediate the survival effects seen in this subgroup of patients.

Taken together, our results suggest a model in which 12q13-15 amplification causes initiation of liposarcomagenesis and formation of WDLS (Figure S5). Tumors that dedifferentiate develop any of several divergent morphologies in response to varied genomic events. Loss of 11q23-24, either in response to an unknown event or as an independent event, is associated with tumors that are chromosomally complex and are morphologically similar to myxofibrosarcomas or UPS. For tumors without 11q23-24 loss (tumors of other morphologies and a subset of myxofibrosarcoma-like and UPS-like lesions), progression to DDLS is caused by unknown events. Loss of 19q13 is
associated with an aggressive phenotype, perhaps as a result of reduced expression of 
CEBPA and is independent of morphology.

This model can act as a scaffold for our understanding of liposarcomagenesis. Data 
regarding tumors that do not have common CNAs can easily be integrated into this 
outline, and genomic and histologic subtype data can be used as the basis for 
correlative studies in upcoming clinical trials. The tumor subgroups defined by this 
model may respond very differently to therapies, whether novel or traditional. For 
example, 11q23-24 carries the gene E124, implicated in regulation of p53 and 
representing a potential tumor suppressor in DDLS, and 11q23-24 loss is associated 
with resistance to etoposide-based chemotherapies in other diseases (20, 21). 
Therefore, 11q23-24 copy number (and E124 expression) may identify patients more or 
less likely to respond to etoposide-containing combination therapies. Similarly, high-risk 
patients stratified based on 19q13 loss may be more likely to benefit from neoadjuvant 
regimens or combination therapies at the time of disease recurrence.

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Figure Legends

Figure 1. Statistically significant genomic alterations in well-differentiated (A) and dedifferentiated (B) liposarcoma. Plots indicate the false discovery rate (q-value, y-axis), with red peaks representing amplifications and blue representing deletions. The most common CNAs associated with dedifferentiation were on 11q23-24, 19q13, 3p14-21, and 3q29.

Figure 2. (A) Recurrence-free survival (including both local and distant recurrence) and (B) disease-specific survival among 40 patients with DDLS stratified based on 19q13 copy number.
References


**Table 1. Patient and tumor characteristics**

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<th>Sample Type</th>
<th>WDLS (n=55)</th>
<th>DDLS (n=52)</th>
</tr>
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<tr>
<td><strong>Age, median (range)</strong></td>
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<td>60 (41-90)</td>
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<tr>
<td><strong>Gender</strong></td>
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<td></td>
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<tr>
<td>Male</td>
<td>28 (51%)</td>
<td>32 (62%)</td>
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<tr>
<td>Female</td>
<td>27 (49%)</td>
<td>20 (38%)</td>
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<tr>
<td><strong>Tumor histology</strong>*</td>
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<td></td>
</tr>
<tr>
<td>Well-differentiated</td>
<td>40 (73%)</td>
<td>3 (5.8%)</td>
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<tr>
<td>Dedifferentiated</td>
<td>15 (27%)</td>
<td>49 (94%)</td>
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<td><strong>Primary vs. recurrent</strong></td>
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<tr>
<td>Primary</td>
<td>44 (80%)</td>
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<td>First recurrence</td>
<td>9 (16%)</td>
<td>12 (23%)</td>
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<td>Second or later recurrence</td>
<td>2 (3.6%)</td>
<td>4 (7.7%)</td>
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<td><strong>Tumor location</strong></td>
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<td>Retroperitoneal/ Intraabdominal</td>
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<td>49 (94%)</td>
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<tr>
<td>Chest</td>
<td>2 (3.6%)</td>
<td>3 (5.8%)</td>
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<tr>
<td>Extremity</td>
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<td>0 (0%)</td>
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<tr>
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<td><strong>Primary tumor size</strong></td>
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<tr>
<td>5-10 cm</td>
<td>6 (11%)</td>
<td>3 (5.8%)</td>
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<tr>
<td>&gt;10 cm</td>
<td>46 (84%)</td>
<td>48 (92%)</td>
</tr>
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</table>

* Tumor defined as DDLS if DDLS comprises >20% of tumor volume.
Figure 1

A.

![WDLS](image)

B.

![DDLS](image)

- **WDLS**
  - Chromosome 1
  - q-value range: $10^{-8}$ to $10^{-4}$

- **DDLS**
  - Chromosome 1
  - q-value range: $10^{-8}$ to $10^{-4}$

**Key:**
- **3p14-21 & 3q29**
- **11q23-24**
- **19q13**
<table>
<thead>
<tr>
<th>Copy Number Alteration</th>
<th>DDLS n (%)</th>
<th>WDLS n (%)</th>
<th>FDR</th>
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<tr>
<td>3p14-21 loss</td>
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<td>9p22-24 loss</td>
<td>14 (27)</td>
<td>2 (4)</td>
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<td>10p15 loss</td>
<td>12 (23)</td>
<td>0 (0)</td>
<td>0.0006</td>
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<td>22 (42)</td>
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<td>17p11 gain</td>
<td>13 (25)</td>
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</tr>
<tr>
<td>17q21 loss</td>
<td>14 (27)</td>
<td>2 (4)</td>
<td>0.0006</td>
</tr>
<tr>
<td>19q13 loss</td>
<td>17 (32)</td>
<td>0 (0)</td>
<td>0.0003</td>
</tr>
<tr>
<td>20q11 gain</td>
<td>14 (27)</td>
<td>0 (0)</td>
<td>0.001</td>
</tr>
</tbody>
</table>

FDR, false discovery rate
Table 3. DDLS Morphology (n=39)

<table>
<thead>
<tr>
<th>Morphology</th>
<th>n  (%)</th>
<th>Tumors with chromosome 11q loss (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UPS only</td>
<td>6 (15)</td>
<td>3</td>
</tr>
<tr>
<td>Mixed UPS + other morphology</td>
<td>7 (18)</td>
<td>4</td>
</tr>
<tr>
<td>Myxofibrosarcoma only</td>
<td>5 (13)</td>
<td>1</td>
</tr>
<tr>
<td>Mixed myxofibrosarcoma + other morphology</td>
<td>10 (26)</td>
<td>6</td>
</tr>
<tr>
<td>Spindle cell sarcoma, NOS only*</td>
<td>3 (8)</td>
<td>0</td>
</tr>
<tr>
<td>Solitary fibrous tumor only</td>
<td>1 (3)</td>
<td>0</td>
</tr>
<tr>
<td>Mixed small blue round cell + other morphology</td>
<td>3 (8)</td>
<td>0</td>
</tr>
<tr>
<td>Divergent only</td>
<td>1 (3)</td>
<td>0</td>
</tr>
</tbody>
</table>
| Mixed divergent + other morphology             | 3 (8)  | 2**                                 

* Resembling adult fibrosarcoma
** Both with myxofibrosarcoma component
UPS, undifferentiated pleomorphic sarcoma
NOS, not otherwise specified
Table 4. Factors associated with patient outcome

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>LRFS</th>
<th>DRFS</th>
<th>DSS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HR</td>
<td>p-value</td>
<td>HR</td>
</tr>
<tr>
<td>Surgical margin (R1 vs. R0)</td>
<td>2.92</td>
<td>0.035</td>
<td>0.12</td>
</tr>
<tr>
<td>Gender (male vs. female)</td>
<td>0.46</td>
<td>0.084</td>
<td>0.53</td>
</tr>
<tr>
<td>Age (&gt;60 vs ≤60 years)</td>
<td>0.32</td>
<td>0.011</td>
<td>0.66</td>
</tr>
<tr>
<td>Presentation status</td>
<td>1.4</td>
<td>0.43</td>
<td>1.13</td>
</tr>
<tr>
<td>Copy number alteration</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3p14-21 loss</td>
<td>1.94</td>
<td>0.16</td>
<td>0.33</td>
</tr>
<tr>
<td>3q29 loss</td>
<td>1.57</td>
<td>0.32</td>
<td>0.00</td>
</tr>
<tr>
<td>9p13 loss</td>
<td>2.35</td>
<td>0.037</td>
<td>1.28</td>
</tr>
<tr>
<td>10p15 loss</td>
<td>1.05</td>
<td>0.92</td>
<td>0.48</td>
</tr>
<tr>
<td>11q23-24 loss</td>
<td>1.84</td>
<td>0.16</td>
<td>1.80</td>
</tr>
<tr>
<td>17p11 gain</td>
<td>2.08</td>
<td>0.13</td>
<td>1.06</td>
</tr>
<tr>
<td>17q21 loss</td>
<td>2.75</td>
<td>0.021</td>
<td>1.65</td>
</tr>
<tr>
<td>19q13 loss</td>
<td>2.86</td>
<td>0.013</td>
<td>1.51</td>
</tr>
<tr>
<td>20q11 gain</td>
<td>0.97</td>
<td>0.96</td>
<td>1.40</td>
</tr>
</tbody>
</table>

DRFS, distant-recurrence–free survival; DSS, disease-specific survival; HR, hazard ratio; LRFS, local-recurrence–free survival.
Figure 2

A. Recurrence-free survival — no 19q13 loss — 19q13 loss

B. Disease-specific survival — 19q13 loss

p=0.0051

p=0.0025
Copy number losses define subgroups of dedifferentiated liposarcoma with poor prognosis and genomic instability

Aimee Crago, Nicholas D. Socci, Penelope L. DeCarolis, et al.

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