The value of bone marrow biopsy for staging of patients with primary CNS lymphoma

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Abstract

Background: In patients with presumed primary CNS lymphoma (PCNSL) a systemic manifestation is found only in a small minority. Although bone marrow biopsy (BMB) is recommended for staging, its diagnostic value is unclear.

Methods: A retrospective analysis of 392 patients with presumed PCNSL from three university hospitals and 33 patients with secondary CNS lymphoma (SCNSL) and initial CNS involvement from a multicentre Germany-wide prospective registry was performed.

Results: A BMB was performed and documented in 320/392 patients with presumed PCNSL; 23 had pathologic results. One harboured the same lymphoma in the brain and bone marrow (BM), 22 showed findings in BM discordant to the histology of brain lymphoma; n=12 harboured a low grade lymphoma in the bone marrow, the other showed B-cell proliferation but no proof of lymphoma (n=5), monoclonal B-cells (n=3) or abnormalities not B-cell associated (n=2). In the group of SCNSL with initial CNS manifestation 32/33 patients underwent BMB; seven were documented with bone marrow involvement (BMI); one had concordant results in the brain and bone marrow with no other systemic manifestation. Six had additional systemic lymphoma manifestations apart from the brain and bone marrow.

Conclusions: In only two out of 352 (0.6%) patients with CNS lymphoma (320 presumed PCNSL and 32 SCNSL) BMB had an impact on diagnosis and treatment. While collected in a selected cohort these findings challenge the value of BMB as part of routine staging in presumed PCNSL.

Keywords

primary central nervous system lymphoma – PCNSL – staging – bone marrow involvement – diffuse large B-cell lymphoma
**Keypoints**

- Bone marrow biopsy in staging of presumed PCNSL seems dispensable.
- Only in two out of 352 patients with CNS lymphoma bone marrow biopsy influenced diagnosis and treatment.

**Importance of the Study**

We report the results of a retrospective analysis on bone marrow biopsy as part of staging in CNS lymphoma. For correct diagnosis and appropriate treatment planning most centres perform bone marrow biopsy for histology, cytology and molecular genetics, although only few case reports on bone marrow involvement in presumed primary CNS lymphoma exist. Our data in a large cohort suggest that this invasive procedure may be dispensable.
**Introduction**

Primary CNS lymphoma (PCNSL) is a rare, aggressive disease which by definition affects the brain, leptomeninges, spinal cord and/or the vitreoretina of the eyes; it accounts for about 2% of all primary brain cancers and 7% of malignant primary brain tumours. Histopathologically, most PCNSL are diffuse large B-cell lymphomas (DLBCL). PCNSL has to be distinguished from secondary CNS lymphoma (SCNSL) – defined as CNS lymphoma occurring concomitantly with or as relapse of systemic lymphoma - since prognosis and treatment may significantly differ. In 4% to 12% of presumed PCNSL-patients a systemic manifestation was reported at first diagnosis, when a systematic thorough staging was performed.

To exclude systemic tumour manifestation in presumed PCNSL, the guidelines of the International PCNSL Collaborative Group (IPCG) and of the European Association of Neuro-Oncology (EANO) recommend a PET-CT (or chest and abdominal CT-scan with contrast medium) and a bone marrow biopsy (BMB). The analysis of bone marrow (BM) in PCNSL and SCNSL as in all other lymphomas comprises histopathology, cytology, flow cytometry and sometimes PCR based immunoglobulin heavy chain (IgH) analysis to proof B-cell clonality. The histopathology has the main impact on the interpretation of these findings.

While most of (neuro-) oncological centres, especially within clinical trials, perform BMB at diagnosis of presumed PCNSL, its necessity is questioned by some investigators. To our knowledge there is no data on the frequency of simultaneous lymphoma manifestation in CNS and bone marrow. Two single cases of concordant findings, with BM as the only systemic manifestation of the lymphoma had been reported. Some retrospective studies showed small numbers of discordant findings which did not influence treatment: One found two out of 86 patients from a clinical trial with low grade B-cell lymphoma in the BM and another detected monoclonal B-cells in the BMB flow cytometry in eight of 51 patients; four of those showed histological confirmation of a low grade B-cell lymphoma in BM.

For patients with systemic DLBCL concordant BMI was reported in 5% to 10% of patients and discordant findings in 5% to 12%. In systemic DLBCL, concordant BMI is an independent negative prognostic factor for progression-free survival (PFS) and overall survival (OS), while no prognostic impact of discordant BMI on OS has been found in most studies as compared to patients without BMI. However, some series showed a lower PFS or even a negative impact on OS in discordant BMI. The possibility of valuable diagnostic information has to be balanced against the invasiveness of a procedure. BMB may cause discomfort and pain. However, the risk for relevant complications like arterial bleeding during
BMB with is extremely low\(^2\). The objective of the present analysis was to evaluate, if BMB adds relevant information to staging of patients with CNS lymphoma.

Patients and methods

Data of 425 patients from two groups were retrieved: The PCNSL group (n=392) consisted of patients from the Department of Hemato-Oncology, Charité Berlin (n=138, between 2009 and 2018), from the Department of Neurology, Knappschaftskrankenhaus, University-Hospital, Ruhr-University Bochum (n=167, between 2004 and 2018) and from the Departments of Hemato-Oncology and Neurology at the University-Hospital Bonn (n=87, between 1995 and 2004). All those 392 patients had presented with initial CNS lymphoma and no history nor clinical signs or symptoms of systemic lymphoma.

The SCNSL group comprised 33 patients with initial CNS-involvement from a prospective registry with 200 patients from the Department of Haemato-Oncology, Charité Berlin, documented between 2011 and 2018.

We analysed the records of all 425 patients for information, whether BMB was performed, for its results and for demographic information (sex, age at diagnosis). Detailed original reports for histologic, cytologic and flow cytometry BM examination were evaluated. In case of insufficient data, we contacted the Department of Pathology, to which the biopsy had been sent to. If no written pathologic reports were accessible, we used comments in the medical records, as e.g. „no pathological findings in BM“. Only patients with information about the histology of BMB were included in the final analysis.

All pathology reports were anonymized and reviewed by a board certified Neurologist (U.S) and by a board certified Haemato-Oncologist (A.K.).

This analysis had been approved by the ethics committee of the University of Bochum. Data were analysed in Excel and SPSS-software.

Results

Patients characteristics

In the group of 392 patients with presumed PCNSL, 192 (49%) were male, the median age was 67 years (range 26-87 years); 96% of the patients harboured a DLBCL in the CNS (see table 1 for more details).

A BMB was performed in 330 cases (84%). The most important reason for not carrying out BMB was a low clinical performance status and the consecutive decision of starting immediate treatment (n=12) or symptomatic therapy only (n=2), in 30 patients there was no information why BMB was not done (n=30). For 18 patients it remained unclear, if BMB was performed.
Results of the BMB were available in 320/330 patients (97%), mostly as written original reports (see table 2). Additional PCR analyses were performed in 26 cases (8%), when the report of histopathology and cytopathology stated “unclear findings/significance”. In synopsis of histology, cytology and flow cytometry 23/320 cases were classified as abnormal (see figure 1 and figure 2); one represented concordant lymphoma in BM with regard to the brain lymphoma, and 22 were discordant (see figure 3 and table 1 for details). Out of the 320 patients evaluated, brain lymphoma had been diagnosed by biopsy of the brain lesion in 313, in the CSF in 2 and in the vitreous fluid in 5.

The median age of the cohort of 33 SCNSL patients with CNS involvement at initial diagnosis was 64 years (range: 35-86 years), 20 (61%) were male. A BMB was performed in 32 cases with all results available. Seven patients showed pathologic results with two concordant and three discordant results in histology, for two patients there was no further specification of “BMI”. Six of these seven patients with pathologic findings in BM had other systemic lymphoma manifestation in addition to BMI. In only 11 of the 32 SCNSL patients with BMB a biopsy of the brain lymphoma was performed: Ten had a DLBCL, one had a low-grade malignant B-cell non-Hodgkin lymphoma. The others were diagnosed by biopsy of the systemic lymphoma and typical lesions in the brain, CSF or both (see supplementary table S1 for details).

Concordant findings

Two patients with concordant findings in CNS and BM with BM representing the only systemic/extra-CNS manifestation of a DLBCL were identified – one in the „presumed PCNSL“ group and one in the SCNSL group. Both were diagnosed and treated as SCNSL with R-CHOP (rituximab, cyclophosphamide, hydroxydaunorubicine, vincristine, prednisone) according to the results of BMB. These two patients were 72 and 74 years old and had a grievous course of disease with an early progression and an OS of only seven months each.

Discordant findings in patients with presumed PCNSL

Discordant pathological findings in BM were reported in 7 % (22 of 320 BMBs). In 12 of these a low-grade lymphoma was found in BM, five showed B-cell proliferations without proof of lymphoma and three monoclonal B-cells. Only two patients had BM findings not related to lymphoma or B-cells (one myelodysplastic syndrome, one chronic myeloproliferative neoplasm (see figure 3 and table 1).
Compared to the patients with normal results in the BM patients with discordant findings in the BM were slightly older at the time of diagnosis with a median of 70 versus 66 years (range: 47-83 years versus 26-84 years).

Data on OS was available for 192 patients with PCNSL who had undergone BMB, 10 of those had discordant findings in BM. There was no significant difference in OS for patients with discordant and normal findings in BM (26 months [95%-CI 3-49 months] and 35 months [95%-CI 20-50 months]; p=0.55, figure 4).

Discussion

Bone marrow biopsy (BMB) is a recommended part of systemic staging in presumed PCNSL. The aim of this study was to investigate, if BMB is of diagnostic value in patients with lymphoma of the brain and no other systemic lymphoma manifestation. In other words, how often has the diagnosis of presumed PCNSL to be changed by BMB alone to SCNSL due to detection of concordant lymphoma in bone marrow and brain with no other sign of systemic lymphoma? In the literature only rare reports document bone marrow involvement as the only systemic manifestation in brain lymphoma. Is it such a rare event, that we can therefore spare patients this invasive procedure?

While the present study could address this question only in pre-selected cohorts, all those patients with presumed PCNSL, i.e. patients presenting with initial CNS lymphoma and no history of pre-existing systemic lymphoma, were systematically analysed: The reports on bone marrow histology, cytology, flow cytometry and optional PCR analysis in 320 “PCNSL” patients from different institutions and in 32 SCNSL patients with initial brain involvement from a registry were evaluated. Our data show that in only two of 352 patients with evaluable reports on BMB results concordant findings of a diffuse large B-cell non-Hodgkin lymphoma in the brain and in the bone marrow as the only systemic manifestation were documented. In these two cases (one retrieved from a SCNSL registry, one from a cohort of presumed PCNSL patients) diagnosis was changed from presumed PCNSL to SCNSL by BMB alone. We conclude from these numbers - in line with sparse reports in the literature - that the frequency of concordant CNS and bone marrow lymphoma with no other systemic manifestation is exceedingly low.

Concordant bone marrow infiltration is associated with poor prognosis in systemic DLBCL. We encountered only two such patients in this series, but both suffered from an early progression and showed an OS of only seven months each.
In addition to these two patients with concordant findings we found 22 others with suspected PCNSL and abnormal, but discordant findings in bone marrow; 12 of them harboured a low grade lymphoma, eight another B-cell associated pathology (B-cell proliferation without proof of lymphoma n=5; monoclonal B-cells n=3) and only two had a not B-cell related pathology (myelodysplastic syndrome n=1, chronic myeloproliferative neoplasm n=1). In accordance to these findings Wong and colleagues reported 2/86 patients with PCNSL and a low grade B-cell lymphoma in BM\textsuperscript{11}, while Brandt and colleagues reported on 8/51 PCNSL patients with monoclonal B-cells in BM and evidence of low grade lymphoma in four of these\textsuperscript{12}. Compared to systemic DLBCL the rates of discordant findings in BM (5-12\% in the literature\textsuperscript{13-17}) are similar to those in PCNSL including the present series with 7\%.

Interestingly the finding of monoclonal B-cells in BM disappeared in 3/3 PCNSL patients from the cohort investigated by Brandt and colleagues who underwent a second BMB after treatment\textsuperscript{12}. The same group proved clonal relation between DLBCL in the brain and monoclonal B-cells in BM via immunoglobulin heavy chain variable (IGHV) sequencing in one of two cases, both of them showed histopathological signs of a low grade lymphoma in BM\textsuperscript{12}. With the same method Malecka and colleagues reported clonal relation between CNS lymphoma and monoclonal B-cells in BM for three of six PCNSL patients\textsuperscript{21}. In relation to that Kremer and colleagues report a common clonal origin (detection of clonal IgH or bcl-2 rearrangement) in distinct patients with systemic DLBCL and discordant BMI (8/12), while the other four of those seemed to harbour two clonally unrelated neoplasms, leading to the hypothesis that patients with discordant findings in BM may not be a homogeneous group\textsuperscript{22}.

Also using IGHV- and immunoglobulin variable analysis, respectively, two groups reported tumour related B-cell clones in BM in 4/7 and 2/3 PCNSL-patients\textsuperscript{23,24}, which they considered as subclinical systemic disease. McCann and colleagues proved unique extracerebral variants “as a sign of separate development without a re-entry in the brain”\textsuperscript{24}. On the other hand, IGHV-gene and gene expression profiling results pointed to the possibility, that lymphoma precursor cells might develop outside the CNS and give rise to PCNSL by malignant transformation in the microenvironment of the brain.

Most authors did not observe systemic relapse of PCNSL in patients with discordant findings in BM or tumour related B-cells outside the CNS\textsuperscript{12,23,24}. This is in contrast to our observation in one patient of this series with a low grade lymphoma in BM at initial staging and relapse of a DLBCL in a cervical lymph node. Provencher and colleagues described two PCNSL patients among 209 with a systemic relapse of a DLBCL in BM /soft tissue,
who had lymphoid small cells in BM at initial staging\textsuperscript{25}. However, it cannot be concluded from these rare observations, that patients with PCNSL and discordant findings in BM have a higher risk for systemic relapse.

The question if there is a specific subgroup among patients with presumed PCNSL for which BMB should be considered cannot be answered based on our findings, because of the small number of only two patients with concordant findings in BM. Unfortunately, there are no comprehensive molecular data on these specimens.

For systemic DLBCL, \textsuperscript{18}FDG-PET/CT has a high negative predictive value for detection of bone marrow involvement\textsuperscript{26,27} and there is also one study that suggests that this might also be true for PCNSL\textsuperscript{28}. The question if \textsuperscript{18}FDG-PET/CT can be used to identify a subgroup of patients for which BMB should be performed might be the subject of future investigations.

A limitation of our study is that in 104/320 of patients flow cytometry had not been performed or results of it were not longer available anymore. This was the case for many patients who were diagnosed with PCNSL prior to 2004. Further, emerging technologies like circulating tumor DNA (ctDNA) in blood or flow cytometry in blood have not been performed in this patient cohort. For systemic DLBCL the concentration of circulating tumor DNA in blood correlates with tumor burden and shows a significant correlation to the international prognostic index (IPI) and with lactate dehydrogenase (LDH) levels\textsuperscript{29,30}. Corresponding data concerning ctDNA in blood of PCNSL patients has not yet been published. As peripheral blood involvement in DLBCL is very rare and infrequent in PCNSL, flow cytometry in blood is no part of diagnostic work up in PCNSL yet.

Conclusion

According to the results of the present series BMB is not essential for staging of patients with presumed PCNSL.

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Figures:

Figure 1: Results of bone marrow biopsy in patients with CNS lymphoma

Legend:

PCNSL = primary central nerve system lymphoma, SCNSL = secondary central nerve system lymphoma, BMB = bone marrow biopsy

*n=2 concordant, n=3 discordant, n=2 “BMI” not further specified

Figure 2: Distribution of pathological findings in bone marrow of patients with CNS-lymphoma and no signs of systemic lymphoma (n=24)

Legend:

None

Figure 3: 22 patients with presumed PCNSL and discordant results in bone marrow biopsy

Legend:

NHL = non-Hodgkin’s lymphoma, MDS = myelodysplastic syndrome, CMN = chronic myeloproliferative neoplasm

Figure 4: Kaplan-Meier-Curve of patients with PCNSL and discordant (n=10) and normal findings (n=182) in bone marrow biopsy

Legend:

None
Table 1 Characteristics of CNS-lymphoma patients with no signs of systemic lymphoma, but findings in the bone marrow

| No | Sex | Age | Histology | Cytology | Flow-Cytometry | PCR | Diagnosed as CNS lymphoma | Biopsy of CNS lymphoma |
|----|-----|-----|-----------|----------|----------------|-----|--------------------------|-----------------------|
| 1  | F   | 72  | Normal    | CLL      | CLL            | Not performed | Low grade NHL             | B-cell lymphoma*      |
| 2  | F   | 72  | Hypercellular BM, 3 paratrabecular B-cell infiltrates | Normal | Normal | Normal | Low grade NHL | DLBCL |
| 3  | F   | 70  | Paratrabecular B-cell infiltrates | Normal | Normal | Normal | Low grade NHL | DLBCL |
| 4  | F   | 76  | Normal | Proliferation of lymphocytes | Light chain restriction Possible B-cell NHL | Not performed | B-cell proliferation, no proof of lymphoma | DLBCL |
| 5  | M   | 65  | Normal | Normal | Normal | Monoclonal B-cells | Monoclonal B-cells in PCR | DLBCL |
| 6  | M   | 68  | Normal | Normal | Normal | Monoclonal B-cells | Monoclonal B-cells in PCR | DLBCL |
| 7  | F   | 71  | Paratrabecular B-cell infiltrates | Highly elevated number of adult lymphocytes, possible infiltration | Normal | Not performed | Low grade NHL | Not performed** |
| No. | Gender | Age | diagnosis            | BM findings                                                                 | Immunophenotypic findings | Histopathology findings                                                                 | Final diagnosis        |
|-----|--------|-----|----------------------|------------------------------------------------------------------------------|---------------------------|----------------------------------------------------------------------------------------|------------------------|
| 8   | F      | 71  | Normal               | Normal                                                                       | Normal                    | Proliferation of adult lymphocytes                                                       | DLBCL                  |
| 9   | M      | 54  | Normal               | Normal                                                                       | Normal                    | Proliferation of adult lymphocytes B-cell proliferation dominate lambda light chain expression | DLBCL                  |
| 10  | M      | 68  | Hypercellular BM,  | No result available                                                          | No result available       | B-cell proliferation, no proof of lymphoma                                               | B-cell lymphoma        |
|     |        |     | B- and t-cell        |                                                                             |                           | High malignant lymphoma                                                                   |                        |
| 11  | F      | 52  | Normal               | Normal                                                                       | Immunocytoma              | Not performed                                                                           | Low grade NHL          |
| 12  | F      | 78  | Infiltration by B-cell lymphoma possible mantle cell lymphoma | Normal                     | Normal                   | Monoclonal B-cells                                                                      | DLBCL                  |
| 13  | M      | 47  | MDS                  | Normal                                                                       | Normal                    | Not performed                                                                           | MDS                   |
| 14  | M      | 75  | Medium-sized peritrabecular lymphoid infiltrates: possible NHL or reactive changes | Normal                     | Normal                   | Monoclonal B-cells                                                                      | DLBCL                  |
| 15  | M      | 83  | CMN                  | Normal                                                                       | Not available             | Not performed                                                                           | Other                 |
| 16  | M      | 70  | Pathological         | Normal                                                                       | Normal                    | B-cell proliferation, no proof of lymphoma                                               | DLBCL                  |
|     |        |     | immunohistology, possible low grad NHL |                                                                             |                           |                                                                                         |                        |
| 17  | F      | 76  | Normal               | Normal                                                                       | Pathological light chain restriction | Not performed                                                                           | Low grade NHL          |
| 18  | F      | 61  | Normal               | Normal                                                                       | Normal                    | CLL                                                                                     | Diagnosis by CSF***    |
| 19  | F      | 58  | CLL                  | Normal                                                                       | Normal                    | Not performed                                                                           | Low grade NHL          |
| 20  | M      | 68  | Normal               | Normal                                                                       | Normal                    | B-CLL                                                                                   | DLBCL                  |
| 21  | F      | 70  | Normal               | Normal                                                                       | Pathological plasma cell infiltration (40%) | Not performed                                                                           | Low grade NHL          |
| 22  | F      | 61  | Hypercellular BM, small-medium-sized B-cell infiltrates | Normal                     | Normal                   | Not performed                                                                           | Low grade NHL          |
|   |   |   | Infiltration by DLBCL | Infiltration by lymphoma | Normal | Not performed | Concordant findings | DLBCL |
|---|---|---|-----------------------|--------------------------|--------|--------------|---------------------|-------|
| 23 | M | 74 | Infiltration by DLBCL | Normal | Not performed | Concordant findings | DLBCL |
| 24 | M | 72 | Infiltration by adult cell NHL-lymphoma | Infiltration by B-NHL | Not performed | Concordant findings | DLBCL |

M = Male, F = Female, CMN (chronic myeloproliferative neoplasm), MDS = myelodysplastic syndrome, DLBCL = diffuse large B-cell-lymphoma, NHL = non Hodgkin’s lymphoma

*Unspecific result, because of biopsy under steroid therapy

**Typical brain lesions, malignant large B-cells in vitreous aspiration

***Typical brain lesions, B-cell-lymphoma in CSF
Table 2: Availability of bone marrow biopsy results

|                           | Patients with presumed PCNSL (n=320) | Patients with SCNSL (n=32) |
|---------------------------|--------------------------------------|----------------------------|
|                           | Histology | Cytology | Flow cytometry | PCR analysis | Histology | Cytology | Flow cytometry | PCR analysis |
| Available results         | 320       | 297      | 216            | 26           | 32        | 30       | 27            | not assessed |
| Available as original reports | 277       | 265      | 214            | 26           | 20        | 18       | 15            | not assessed |
| Available as cited in medical reports | 43        | 32       | 2              | 0            | 12        | 12       | 12            | not assessed |
Figure 2

Histology

Cytology

Flow-cytometry

PCR

8

2

2

2

1

1

4
Figure 3

- MDS n=1
- CMN n=1
- Monoclonal B cells in PCR n=3
- B cell proliferation without proof of lymphoma n=5
- Low grade NHL n=12
