Virchows Arch A (1987) 411:359–364

Virchows Archiv A

Hodgkin's disease in the spleen

A morphological study of 140 biopsy cases

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Summary. 140 spleens involved by untreated Hodgkin's disease were studied utilizing conventional histological methods. Regardless of the subtype of Hodgkin's disease, infiltrates of neoplastic cells were present either in the periarteriolar lymphoid sheath, the marginal zone or in both locations. Initially, infiltrates were confined to the splenic white pulp, later larger nodular foci of Hodgkin's disease developed by coalescence of several infiltrates. Neoplastic cells in Hodgkin's disease may reach the spleen by both retrograde lymphatic spread or the splenic artery; the presence of neoplastic cells in both T- and B-cell areas of the splenic white pulp implies a preference for Hodgkin's disease in the spleen with regard to a suitable microenvironment. This may be provided by certain macrophage subpopulations.

Key words: Hodgkin's disease – Histopathology – Spleen

Introduction

In untreated patients with Hodgkin's disease (HD) a staging laparotomy with splenectomy will demonstrate splenic involvement in 35% to 42% of the cases (Askergren et al. 1981; Kadin et al. 1971; Kaplan 1980a). Involvement of the spleen in HD is, among other factors, dependent on the histological subtype of the disease that is determined according to the Rye classification (Lukes et al. 1966). While lymphocyte predominance and nodular sclerosis subtypes exhibit splenic foci in 16% and 35%, respectively, mixed cellularity and lymphocyte depletion subtypes show splenic involvement in 59% and 83%, respectively (Kadin et al. 1971; Kaplan 1980b). The present study was conducted in order to determine whether the different morphological subtypes of HD apart from the frequency of splenic foci also influence the distribution pattern and the appearance of splenic infiltrates in HD.

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Material and methods

Spleens removed from 140 patients (76 females and 64 males aged five to 76 years with a median of 34 years) and showing HD at either macroscopic or microscopic examination were studied. In each case splenic tissue from three to five locations was fixed in 4% buffered formaldehyde, dehydrated in a graded alcohol series and embedded in paraffin. 2 μ to 4 μ thick sections cut semi-serially from each block were then stained with haematoxilin-eosin, periodic acid-Schiff, Giemsa and Gomori's silver stain. In all cases a detailed macroscopic evaluation of the splenectomy specimen was available; in addition in 46 cases lymph nodes from the splenic hilus were examined. In order to determine the histological subtype of HD the lymph node biopsies (76% cervical, 6% inguinal and 18% miscellaneous locations) leading to the diagnosis of HD were reviewed.

Hodgkin- and Sternberg-Reed cells were identified by their characteristic morphological appearance. Hodgkin cells were large mononuclear cells with a slightly basophilic cytoplasm and a single prominent nucleolus within a translucent karyoplasm, whereas Sternberg-Reed cells were considerably larger and possessed irregularly shaped nuclei again containing distinct nucleoli. As Hodgkin cells may occasionally be confused with immunoblasts or epitheloid cells, HD in the spleen was diagnosed only in the presence of pathognomonic Sternberg-Reed cells. Since we did not attempt to address the histogenesis of HD, immunohistochemical studies were not performed.

Results

In 10 cases the spleen showed HD of the lymphocyte predominance subtype, while in 57 cases of the nodular sclerosis and in 57 of the mixed cellularity subtype splenic involvement was noted. The lymphocyte depletion subtype was evident in 16 cases among which eight exhibited the features

of Hodgkin's sarcoma. The mean splenic weight in cases with infiltrates of the mixed cellularity and the lymphocyte depletion subtype significantly exceeded that of spleens infiltrated by lymphocyte predominance or nodular sclerosis subtypes (360 ± 92 g versus 240 ± 61 g; $p < 0.05$, Student's t-test). However, there existed a number of remarkable exceptions, since some spleens infiltrated by the nodular sclerosis subtype weighed more than 350 g, and some spleens exhibiting infiltration by the lymphocyte depletion subtype showed a weight of less than 300 g. As a rule, the size of the nodular infiltrates was also dependent on the subtype of HD. Lymphocyte predominance and nodular sclerosis subtypes caused nodules with a size of up to 1 cm in diameter in the majority of cases, while in mixed cellularity and lymphocyte depletion subtypes grossly visible lesions of up to 6 cm in diameter were evident in many instances. Regardless of the subtype the lesions were scattered throughout the parenchyma and appeared nodular and mostly well demarcated from non-involved splenic tissue. In a number of cases splenic involvement by HD was manifest only as a slightly irregular enlargement of Malpighian bodies, while in more advanced cases the lesions evolved into firm yellowish-white nodules that had the tendency to fuse into larger multilobated infiltrates sometimes showing areas of necrosis and in some instances causing infarction of adjacent splenic tissue.

The very variable number (between one and close to a hundred) in each macroscopic section and the diffuse distribution pattern of the lesions did not show any correlation with the histological subtype of HD.

Microscopically, the infiltrates consisted of Hodgkin- and Sternberg-Reed cells admixed with varying numbers of histiocytes, lymphocytes and eosinophilic granulocytes. In small infiltrates these were confined to the splenic white pulp.

Basically two types of infiltrates could be discerned at this early stage of splenic involvement by HD. In about 60% of the cases studied Hodgkin- and Sternberg-Reed cells were located focally in the periarteriolar lymphoid sheaths of the splenic white pulp (Fig. 1). From this site of early involvement the neoplastic cells invaded the splenic follicles, eventually replacing them with tumour tissue. In about 30% of the cases a different infiltration pattern was observed: Hodgkin- and Sternberg-Reed cells were first visible in the marginal zone of splenic follicles and within the immediately adjacent perifollicular region (Fig. 2). While in the first variant the neoplastic cells invaded the splenic follicles from within, marginal zone infiltrates had an obvious tendency to invade the follicles from without, eventually also replacing them with neoplastic tissue. In about 10% of the cases a combination of the two patterns was evident: Hodgkin- and Sternberg-Reed cells were present in the periarteriolar lymphoid sheaths as well as in the marginal zones of different Malpighian bodies of the same section (Fig. 3). Again, the pattern of infiltration could not be related to the histological subtype of HD present in the spleen.

In the early stages of splenic involvement by HD the nodular infiltrates of the white pulp led to disintegration of the zonal arrangement of the splenic follicles and to compression of adjacent sinus of the splenic red pulp (Fig. 4); in most cases, however, they appeared to be well demarcated and did not invade sinus or pulp cords. With continuing growth of the infiltrates the neoplastic tissue expanded and protruded into the red pulp; Hodgkin- and Sternberg-Reed cells became evident in the pulp cords and in the sinus. Eventually larger amounts of the red pulp were replaced by neoplastic tissue, and different infiltrates coalesced into a larger focus of HD. In the periphery of such expanding infiltrates an increase in epitheloid venules was noted. At this stage, several conspicuous features could be observed. The lymphocyte predominance and the nodular sclerosing subtype exhibited an expansion of the infiltrates by a blunt protrusion of neoplastic tissue from the Malpighian bodies replacing pulp cords and splenic sinus, the nodular sclerosis subtype exhibiting the characteristic zones of sclerosis at the periphery. Mixed cellularity and lymphocyte depletion subtypes, however, were noted to invade the splenic red pulp more diffusely and to invade larger splenic veins more readily. The latter phenomenon was observed

Fig. 1. Periarteriolar infiltrate composed predominantly of Hodgkin- and Sternberg-Reed cells. Note the circular shape of the infiltrate clustering around a follicular artery (*right*). Giemsa, original magnification ×160

Fig. 2. Numerous Hodgkin- and Sternberg-Reed cells within the marginal zone of a splenic follicle (Marginal zone infiltrate). Giemsa, original magnification ×160

Fig. 3. Infiltration of both the periarteriolar region (*right*) and the marginal zone (*left*) of a splenic follicle (Combination type infiltrate). The asterisk denotes the follicle artery. Giemsa, original magnification ×160

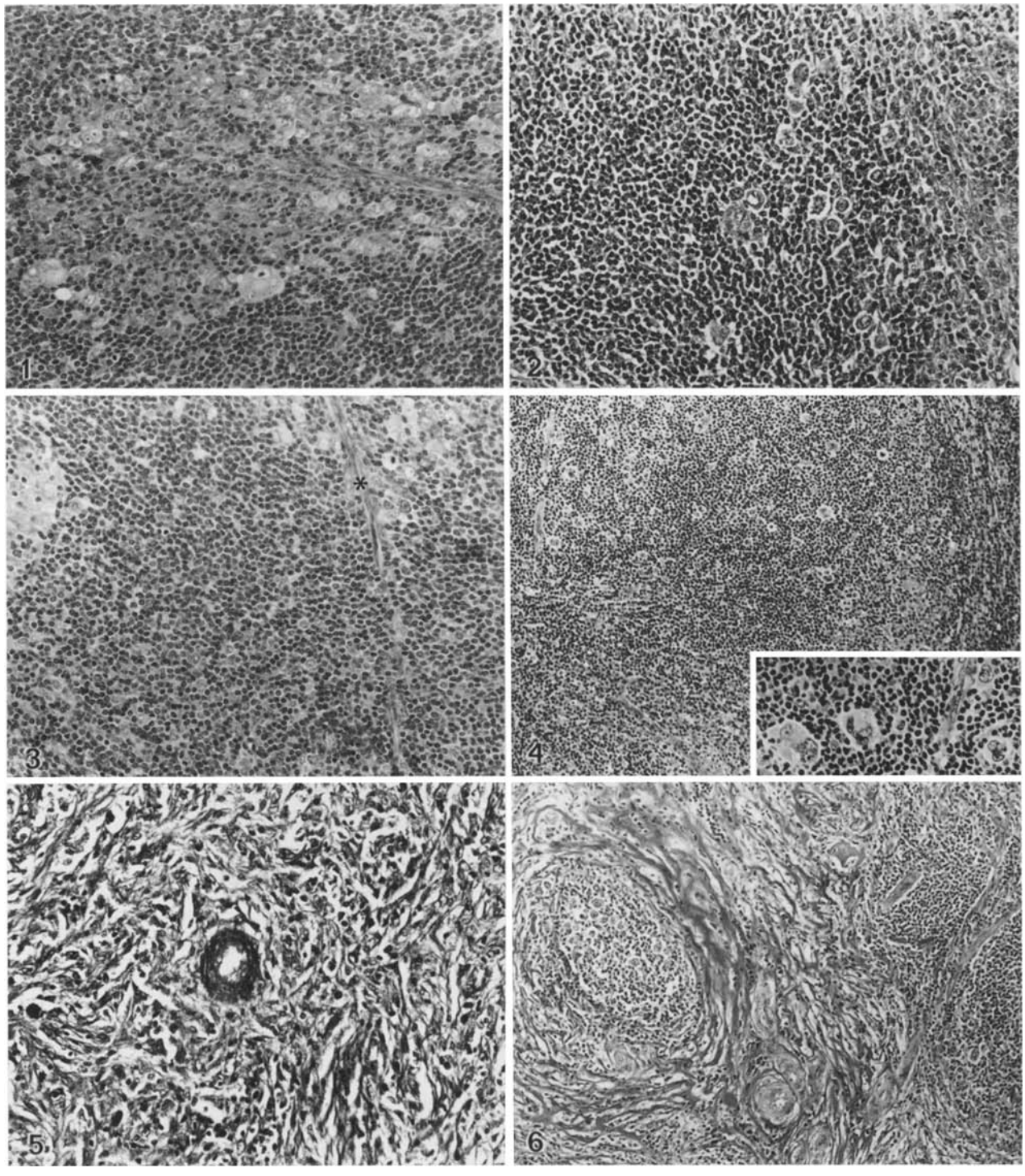

Fig. 4. Splenic follicle infiltrated by HD. The zonal arrangement has ceased to exist, neoplastic cells are visible in all portions of the follicle, and adjacent sinus are compressed (*upper right*). A higher magnification reveals typical Sternberg-Reed cells. Giemsa, original magnification ×80, inset ×160

Fig. 5. An erstwhile follicular artery within a neoplastic infiltrate with pronounced sclerosis. PAS reaction, original magnification ×160

Fig. 6. Coalescence of numerous infiltrates showing remnants of follicles and periarteriolar lymphoid sheaths with interspersed areas of hyalinization. Giemsa, original magnification ×160

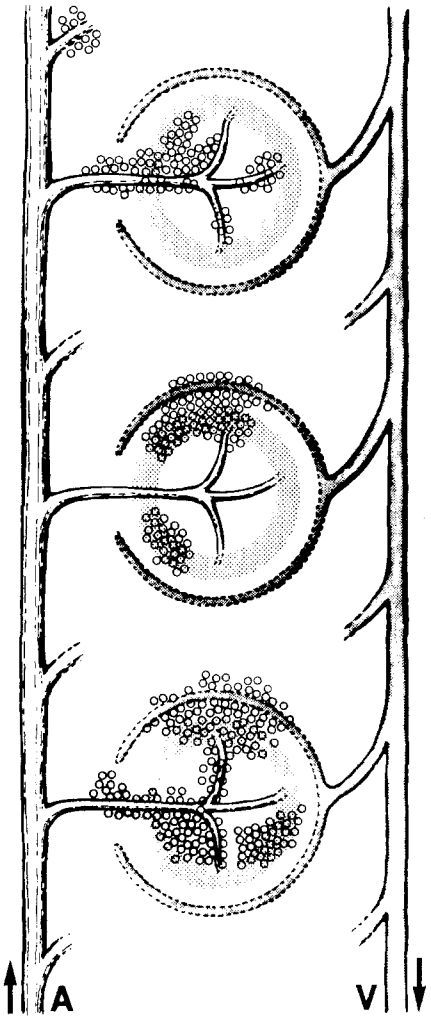

Fig. 7. Infiltration patterns of HD in the spleen. Top: Periarteriolar infiltrate within the periarteriolar lymphoid sheath (cf Fig. 1) Middle: Infiltration of the marginal zone and adjacent perifollicular regions (cf Fig. 2). Bottom: Combination type with neoplastic cells both within the periarteriolar region and the marginal zone (cf Fig. 3)

in fourteen cases (3 nodular sclerosis, 7 mixed cellularity and 4 lymphocyte depletion subtypes). In addition, in 75% of lymphocyte predominance and nodular sclerosis subtypes small arteries surrounded by neoplastic tissue could still be observed (Fig. 5), while in the other subtypes significantly fewer remnants of splenic tissue were noted. Whereas the modes of infiltration differed, all subtypes of HD in the spleen eventually led to the coalescence of several infiltrates into larger nodules containing, apart from neoplastic tissue, numerous periarteriolar lymphoid sheaths and splenic follicles (Fig. 6). This morphological entity represented the final stage of splenic infiltraton by HD. Fig. 7 summarizes the infiltration patterns of HD in the spleen encountered in the present study.

Discussion

A representative sample of spleens involved by all four major histological subtypes of HD (Lukes et al. 1966) was investigated by conventional morphological methods. In the spleen the individual subtype present was easily recognizable only in cases exhibiting the typical morphology of the nodular sclerosis and the lymphocyte depletion subtype, while the distinction between a mixed cellularity and a nodular sclerosis or even a lymphocyte predominance subtype seemed problematical in many cases (Kadin et al. 1971). Therefore, splenic infiltrates were classified according to the results of the preceeding lymph node biopsy. As in earlier studies (Farrer-Brown et al. 1972; Fischer et al. 1974), we observed major differences between the putative histological classification of HD in the spleen and lymph node biopsy of individual patients, with the same subtype being diagnosed in 75% of the cases. In the spleen foci of HD tended to assume the appearance of the nodular sclerosis subtype – save for the presence of lacunar cells – more readily. This may be due partly to the compression and subsequent hyalinization of sinus and pulp cords adjacent to HD of the white pulp.

Regardless of the histological subtype, however, foci of HD in the spleen exhibited a common pattern with respect to their manifestation and growth. Early infiltrates were localized either in the periarteriolar lymphoid sheath (60%) or in the marginal zone at the periphery of the follicles (30%), a combination pattern being present in 10% (Fig. 7). In progressive disease first the white pulp and then the red pulp were replaced by bluntly invading neoplastic tissue. The findings confirm the results of earlier studies (Burke 1981; Farrer-Brown et al. 1972; Halie et al. 1978; Yam and Li 1976), but also allow us to draw some conclusions on the development of HD in the spleen. First of all, the neoplastic cells of HD in the spleen are localized in T-lymphocyte- (periarteriolar lymphoid sheath) as well as in B-lymphocyte-associated (marginal zone) regions (Müller-Hermelink et al. 1974; Van Krieken and te Velde 1986). Despite evidence that Hodgkin- and Sternberg-Reed cells may be histogenetically related to B- and/or T-lymphocytes (Stein et al. 1985) our results do not support a 'homing mechanism' (Halie et al. 1978) restricted to B- or T-cell areas that might be responsible for the particular growth patterns of HD in the spleen. Earlier studies have shown that certain monocyte-macrophage subpopulations in spleens with HD exhibit distinct changes with regard to their composition and distribution

pattern. This is predominantly reflected by a decrease of cells of the monocyte-macrophage lineage stained by the monoclonal antibodies KiM 2, KiM 3 and KiM 5 (Radzun and Parwaresch 1983; Falk et al. 1986 and in preparation). Since these macrophages are localized in the periateriolar lymphoid sheath as well as in the marginal zone and the perifollicular area, these results point to the importance of a suitable microenvironment provided by different macrophage subpopulations that transcends the concept of B- and T-lymphocyte associated regions (Grogan et al. 1983) and that may play an important role in the localization and spread of HD in the spleen.

Splenic involvement by HD may develop as a consequence of three different mechanisms. Since the spleen lacks afferent lymphatic vessels (Heusermann and Stutte 1977; Kellner 1962; Vaclav and Weiss 1972), neoplastic cells may reach the organ via the hilar lymph nodes, by retrograde lymphatic spread leading to the presence of Hodgkin- and Sternberg-Reed cells within periarteriolar lymph vessels and the development of neoplastic foci within the periarteriolar lymphoid sheath. We have observed, albeit only in rare instances, Hodgkin- and Sternberg-Reed cells within perivascular lymph vessels in the spleen.

Since we could not demonstrate that an involvement of hilar lymph nodes leads to either a constant presence of neoplastic cells in lymph vessels or to an infiltration pattern restricted to periarteriolar regions, the contribution of this mechanism to the spread of HD to the spleen remains to be determined.

Moreover, as periarteriolar infiltrates were also present in spleens with negative hilar lymph nodes, retrograde lymphatic spread cannot account for all periarteriolar infiltrates. Colonisation of the spleen by neoplastic cells via the splenic artery has to be assumed. This mechanism may be held responsible for periarteriolar infiltrates with negative hilar lymph nodes as well as for foci within the marginal zone where most afferent blood vessels terminate (Saitoh et al. 1982). The adjacent perifollicular region with its retarded blood flow and its filtering capacity may also be affected (Van Krieken et al. 1985). In these particular areas an increase in the number of epitheloid venules also points to a relationship between the neoplastic infiltrates and vascular structures that is reminiscent of the situation in lymph nodes affected by HD (Falk et al. In preparation). Obviously, there are strong associations between either splenic lymphatic or blood vessels and infiltrates of HD. A third, more remote mechanism consists of the de novo development of HD in the spleen (Halie et al. 1978); however, the clinical course of HD (Kaplan 1980a) and the lack of morphological evidence render this assumption highly unlikely.

In summary, several conclusions may be drawn from the results of the present study: Regardless of the histological subtype, HD in the spleen shows a uniform behaviour with respect to the localisation of the infiltrates and their growth pattern. HD reaches the spleen either from affected hilar lymph nodes via retrograde spread along efferent lymphatic vessels or by hematogenous dissemination. The first mechanism causes periarteriolar infiltrates, the second gives rise to foci within the marginal zone and/or the periarteriolar region. The presence of HD in both B- and T-lymphocyte-associated regions of the spleen and accompanying changes of macrophage subpopulatons localized in these regions rule out simple B- or T-lymphocyte-associated homing mechanisms, but point to more complicated preferences of the neoplastic cells with regard to a suitable microenvironment for splenic foci in HD.

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Accepted May 11, 1987