[CANCER RESEARCH 31, 1794–1798, November 1971]

Clinical and Biological Significance of Vascular Invasion in Hodgkin's Disease[1]

Henry Rappaport, Stephen B. Strum, George Hutchison, and Lawrence W. Allen

Departments of Pathology [H. R., S. B. S.] and Medicine [L. W. A.], The School of Medicine of the University of Chicago, The Department of Radiation Therapy, Michael Reese Hospital [G. H.], and the Argonne Cancer Research Hospital [H. R.] (Operated by the University of Chicago for the United States Atomic Energy Commission), Chicago, Illinois 60637

Summary

Invasion of veins in diagnostic lymph node biopsies from patients with Hodgkin's disease occurs in 6 to 14% of cases studied in reported series. When both spleens and lymph node biopsies obtained at staging operations are examined for vascular invasion, the frequency of the invasion may increase to over 20%, being evident in the spleens of some patients in whom it was not demonstrable in the lymph node biopsies.

Vascular invasion in lymph nodes or spleens of patients with Hodgkin's disease is associated with a high incidence of extranodal organ involvement. Therefore, the detection of vascular invasion in a given patient could be an indication that the disease has spread by the hematogenous route. Our observations also suggest that hepatic involvement occurs by vascular spread via the splenic vein and the portal system.

Further studies are indicated to determine whether the histological search for vascular invasion should be considered an additional procedure in the assessment of the extent of Hodgkin's disease. Its presence in diagnostic lymph node biopsies, however, does suggest a more extensive involvement than initially evident even when the disease belongs to one of the more favorable histological types.

The present methods of staging of Hodgkin's disease are based on the concept that a truly localized form of the disease, amenable to eradication by local irradiation, does exist but that routine clinical and radiological examinations are inadequate to exclude involvement of lymph nodes and organs that are not enlarged, and that enlargement of lymph nodes is not necessarily due to Hodgkin's disease. This uncertainty could be only partly resolved by lymphangiographic studies, and even surgical staging procedures present sampling problems which may make it difficult to detect small microscopic foci of the disease in lymph nodes (11). Modern staging methods, however, have greatly contributed to our knowledge concerning the dissemination of the disease, which apparently spreads to contiguous lymph node groups in a "predictable" manner (7), probably via lymphatic channels. Invasion of nodal sinuses is readily demonstrable microscopically. Lymphatic spread, however, does not explain the occurrence of visceral and bone marrow involvement in the more advanced stages of the disease. The gross distribution of tumor nodules in the spleen and the occurrence of microscopic foci in organs and tissues to which the disease could not possibly have spread via the lymphatics suggest that the blood vessels may be another pathway of dissemination, and that blood-borne metastases do in fact occur. This was supported by the observation of Sternberg-Reed cells in the blood by Bouroncle (1), the recovery of Sternberg-Reed cells from the thoracic duct lymph by Engeset *et al.* (2), and our own studies of vascular invasion in diagnostic lymph node sections obtained prior to therapy (6, 9, 10).

The 1st study (6) was based on a survey of 499 lymph node biopsy specimens from 390 individuals with Hodgkin's disease. The occurrence of vascular invasion was noted incidentally during examination of tissue sections routinely stained with hematoxylin and eosin. Nine such instances were discovered in hematoxylin-and-eosin-stained sections for an incidence of 2.5%, which is very low indeed However, a subsequent systematic search for vascular invasion was made in 100 cases from the same series in which paraffin blocks for additional sections were available. Sections from each block were stained for elastica by Weigert's method. When vascular invasion was demonstrated, adjacent sections stained with hematoxylin and eosin were examined to confirm the presence of intraluminal Sternberg-Reed cells or other neoplastic histiocytes and to exclude nonspecific endophlebitis. Using this method, we found invasion of veins in 10 of 100 cases. We also concluded that the use of elastica stain is essential for such a survey, since the cellular proliferation of Hodgkin's disease may obscure or obliterate the vascular wall beyond recognition with hematoxylin-and-eosin-stained sections. When we classified the 10 cases of this survey according to the Rye modification (4) of the classification of Lukes and Butler (3), it was evident that vascular invasion was more common in Hodgkin's disease with lymphocytic depletion than in other histological variants. Six of the 10 cases were of the lymphocytic depletion type, 3 were Hodgkin's disease of the nodular sclerosing type, and 1 was Hodgkin's disease of the mixed cell type. The absence of vascular invasion in Hodgkin's disease with lymphocytic predominance and its relatively high incidence in Hodgkin's disease with lymphocytic depletion indicate that the general concept of degrees of aggressiveness and malignancy, which is being used for assessment of cancer in general, also applies to Hodgkin's disease. However. it was particularly significant that vascular invasion was not limited

[1] This investigation was supported in part by Grants CA-05183, CA-8970-04, and CA-8971-04 from the National Cancer Institute, NIH, USPHS.

to that type of Hodgkin's disease in which neoplastic cells predominated, *i.e.*, in the lymphocytic depletion type, but was also observed in instances in which inflammatory cells were abundant. This seemed to indicate that the characteristic histological pattern, consisting of a proliferation of Sternberg-Reed cells and other neoplastic histiocytes and a host reaction of inflammatory cells, is maintained even after the vessel walls are penetrated and tumor thrombi are formed. Correlation between vascular invasion and survival was of limited value in this series. In a view of the overall picture, the mean duration of the disease from the time of biopsy showing vascular invasion was 13 months in the patients with vascular invasion as compared to 3.6 years in the total series. Because of the preponderance of Hodgkin's disease with lymphocytic depletion, it was difficult to establish to what extent the vascular invasion and the unfavorable histological type contributed to the low survival rate. The vascular invasion was, however, a contributory factor in low survival as suggested by the finding that in 6 patients with Hodgkin's disease with lymphocytic depletion who had vascular invasion the mean survival was 3.9 months, and in 8 cases of the same type without vascular invasion it was 8.3 months.

The 2nd report (10) was based upon a prospective study in which diagnostic lymph node biopsy sections obtained prior to therapy in 153 patients enrolled in a Hodgkin's disease collaborative clinical trial were examined. This study includes only patients who were clinical Stage I or II (5), and was designed to evaluate the relative merits of involved *versus* extended field radiotherapy in patients with Hodgkin's disease of lymph nodes either above or below the diaphragm. As a prerequisite of entry into the study, the extent of the disease was determined by procedures previously published (10). Of 191 patients enrolled, vascular invasion could be properly evaluated in 153 because of the availability of sections for elastica stains. Invasion of veins was detected in 9 of these 153 cases, or 5.9%. This figure is lower than that of 10% in the 1st series, and is readily explainable by the fact that clinical Stages III and IV were excluded from the study. Each biopsy was again classified according to the Rye modification (4) of the classification of Lukes and Butler (3). In addition, immediately adjacent sections stained with hematoxylin and eosin and Weigert's elastica stain, respectively, were examined. The presence or absence of vascular invasion was recorded without prior knowledge of the course of the patient's illness.

Table 1

Histopathological classification of biopsy sections of patients enrolled in the collaborative clinical trial[a]

Histological type	Group I: All cases accepted into collaborative clinical trial		Group II: Cases in Group I in which vascular invasion could be evaluated	
	No.	%	No.	%
Lymphocytic predominance	16	8.3	13	8.5
Nodular sclerosis	106	55.5	84	54.9
Mixed cellularity	63	32.9	54	35.3
Lymphocytic depletion	3	1.6	1	0.6
Unclassifiable	3	1.6	1	0.6
Totals	191		153	

[a] Tables 1 to 5 are from the article of Strum *et al.* (10).

Table 2

Prevalence of vascular invasion and relation to histological type in cases enrolled in collaborative clinical trial (elastica-stained sections available)

Histological type	Vascular invasion	
	No.	%
Lymphocytic predominance	0/13	0
Nodular sclerosis	4/84	4.9
Mixed cellularity	4/54	7.4
Lymphocytic depletion	1/1	
Unclassifiable	0/1	0
Totals	9/153	5.9

Table 3

Time under observation (mo.)

	Vascular invasion (n = 9)	No vascular invasion (n = 144)
a. Mean	17.8	15.6
b. Median	18	15
c. Range	4–28	2–36

Distribution of patients with respect to months under observation

Mo.	No.	%	No.	%
2–5	1	11	17	12
6–11	1	11	26	18
12–17	2	22	50	36
18–23	3	33	23	16
Over 23	2	22	28	19
Total	9	99	144	101

Table 1 shows the histopathological classification of biopsy sections in patients enrolled in the collaborative study, and Table 2 shows the prevalence of vascular invasion in relation to histological types. Table 3 presents the length of reported follow-ups of cases with vascular invasion in comparison with that of the remaining enrolled patients. The differences were not considered to be significant. Table 4 indicates that over one-half (56%) of the vascular invasion cases have demonstrated extension to nonadjacent areas while only one-fifth (21%) of the patients without vascular invasion have had such advanced disease or have died. Table 5 indicates the life table survival free of extension for each group. This method of analysis takes into account any differences in lengths of follow-up between 2 series. The survival for patients with vascular invasion are generally below the percentages for the remaining patients. By 2 years the vascular invasion survival has fallen to 32% as compared to 57% without demonstrable vascular invasion. At this time only 2 patients in the vascular invasion group have been observed beyond 2 years; one of these showed extension of disease at 14 months and was free of evident extension at 28 months. Thus, the finding of vascular invasion in lymph node biopsy sections can

Table 4
Degree of extension of disease

	Vascular invasion		No vascular invasion	
	No.	%	No.	%
No extension	4	44	107	74
Adjacent nodes only	0	0	7	5
Nonadjacent nodes on same side of diaphragm only	1	11	2	1
Nodes on opposite side of diaphragm	1 >56%	11	12 >21%	8
Extranodal	2	22	11	8
Death	1	11	5	3
Total	9	99	144	99

Table 5
Survival free of extension

Mo.	Vascular invasion (%)	No vascular invasion (%)
0	100	100
3	78	94
6	78	89
12	64	78
18	32	70
24	32	57

be correlated with an increased incidence of nonadjacent and extranodal disease.

In a 3rd study (11), we attempted to correlate the relationship between vascular invasion and involvement of the spleen and other extranodal sites. A series was selected for study to include all admissions from January 1969 to December 1970 to the University of Chicago Hospitals and Clinics of patients with Hodgkin's disease confined to lymph nodes and spleen, or to these sites plus lung in cases in which lung was involved by direct extension from mediastinal nodes. Thirty-three cases satisfying the above description prior to laparotomy were identified, and all of these subsequently underwent laparotomy. A major interest of the study was to compare staging information available before laparotomy with that available at the completion of laparotomy. For this purpose it was desirable to have laparotomy performed at a short time interval after diagnostic biopsy. Somewhat arbitrarily, the limit of this time interval was taken to be 1 year, and 4 cases were eliminated because more than 1 year had passed between diagnosis and laparotomy. The series for study therefore consisted of 29 patients whose preoperative stages were I, II, or III, or one of these stages plus lung involvement. The same method for the demonstration of vascular invasion was used as in the preceding 2 studies. These observations concern 29 patients with Stage I, II, and III disease from a series of 33 patients representing consecutive admissions to the University of Chicago Hospitals and Clinics. Only patients who were Stage I, II, or III prior to laparotomy were accepted in the study, while patients with Stage IV disease were excluded. Two patients with proven lung involvement by direct extensions from the mediastinal lymph nodes in whom laparotomy was performed prior to irradiation, however, remained in the study. For inclusion of patients in this study, an arbitrary limit of 1 year between biopsy and laparotomy was set. Four cases in which this limit was exceeded were eliminated. Data from the remaining 29 patients formed the basis of this report.

Invasion of veins by Hodgkin's tissue was noted in the lymph node biopsies in 4 of the 29, or 13.8%, of the cases. These 4 patients had Hodgkin's disease of the nodular sclerosing type. All 29 patients had splenectomy, and 14 of these were found to have histological evidence of splenic Hodgkin's disease. Unequivocal histological classification of the splenic lesion was possible in 12 of the 14 cases; in 2 cases, histological classification was not attempted because only single, small foci of splenic Hodgkin's disease were evident. These were both classified as nodular sclerosis on the basis of the diagnostic lymph node biopsy. Of the remaining 12 cases. nodular sclerosis was observed in the spleen in 10, lymphocytic predominance was seen in 1, and mixed cellularity was found in 1. In all 12 cases, the histological classifications in the spleen and in the corresponding diagnostic lymph node biopsies were identical. Three of the 4 patients with vascular invasion in the lymph node biopsy showed Hodgkin's disease in the spleen. The patient in whom splenic Hodgkin's disease was not found had, at thoractomy, pulmonary involvement which included, in addition to direct extension of the disease from the mediastinal lymph nodes into the upper lobe of the right lung, a small, noncontiguous focus of Hodgkin's disease in the right middle lobe. In 13 of the 14 cases with splenic Hodgkin's disease, the presence or absence of vascular invasion could be evaluated by appropriate elastica stains. In 3 of these, vascular invasion was evident in the spleen. In 1 of the 3, it had also been previously observed in sections of the diagnostic lymph node biopsy.

Thus, of a total of 29 patients in whom biopsy was followed by laparotomy and splenectomy within 12 months, vascular invasion was found in diagnostic lymph nodes biopsies (3 patients), or the spleen (2 patients), or both (1 patient) in 6 patients, bringing the frequency of this phenomenon to 20.7%. The frequency of vascular invasion in relation to histological type is recorded in Table 6. Table 7 shows the relationship between vascular invasion and stage as determined at laparotomy, and Table 8 shows the relationship between vascular invasion and visceral involvement at the time of surgical exploration or later in the course of the disease.

The data demonstrated that the patients in whom vascular invasion was found in either lymph nodes or spleen or both

had a higher frequency of the advanced stages than those lacking these features. All 4 patients with nodal vascular invasion were Stage III or IV and 1 of the 3 with splenic vascular invasion was Stage IV after laparotomy, the remaining 2 being Stage III by definition. In contrast, when vascular invasion was not demonstrable, only 10 out of 23 or 43% were either Stage III or IV after surgical staging.

More striking is the difference in the frequency of liver, lung, or bone marrow involvement in the 2 groups. When vascular invasion was demonstrable, the liver, lung, or bone marrow was involved either initially or within 1 year in 5 of 6 cases. In contrast, only 1 of 23 patients in whom vascular invasion was not demonstrable had visceral involvement. These data suggest that vascular invasion in an original or secondary site of involvement may be an indicator of hematogenous dissemination.

Table 6

Incidence of vascular invasion in relation to histological type[a]

Histological type	Lymph node	Spleen	Lymph node and spleen	
			No.	%
Lymphocytic predominance	0/1	0/1	0/1	
Nodular sclerosis	4/23	2/11	5[b]/23	21.7
Mixed cellularity	0/5	1/1	1/5	20.0
Totals	4/49	3/13	6[b]/29	20.7

[a] Tables 6 to 8 are from the article by Strum *et al.* (9).
[b] In 1 case, vascular invasion was demonstrated in both lymph node and spleen.

Table 7

Relationship between vascular invasion in diagnostic lymph node or spleen or both and stage as determined by laparotomy

	Stage							
	I		II		III		IV	
	A	B	A	B	A	B	A	B
Vascular invasion present (6 cases)					2	1	2	1
Vascular invasion not demonstrable (23 cases)	5	1	6	1	6	3	1[a]	

[a] In this case, a splenic trabecula was infiltrated by Hodgkin's disease but invasion of a trabecular vein was not demonstrable.

Table 8

Vascular invasion in relation to stage and extranodal involvement

		Vascular invasion			Visceral involvement		
	Histological type	Lymph node	Spleen[a]	Operative stage	Liver	Lung	Bone
1. K. M.	Nodular sclerosis	+	–	IIIA	–	–	–
2. M. D.	Nodular sclerosis	+	–	IVA[b]	–	+ initial	–
3. D. C.	Nodular sclerosis	+	Not involved	IVB[b]	– Suspected 12 mo.	+ initial	+ 12 mo.
4. J. A.	Mixed cellularity	–	+	IIIB	+ 8 mo.	+ 12 mo. autopsy	+ 12 mo. autopsy
5. B. V.	Nodular sclerosis	–	+	IVA	+ initial	–	–
6. P. A.	Nodular sclerosis	+	+	IIIA	–	+ 12 mo.	+ 1 mo.

[a] Involved by Hodgkin's disease unless otherwise specified.
[b] According to the staging classification proposed by Rosenberg, Kaplan (8), Case 2 would be classified as III-$_{SE}$A and Case 3 would be II-$_{E}$B.

Because of the rigid criteria that we used for the diagnosis of vascular invasion, the figure of 20.7% of the total number of cases is probably a conservative one. It is anticipated, therefore, that the presence of vascular invasion may be more significant in the assessment of a given case than is its absence.

All of the patients with proven liver disease had definite or equivocal vascular invasion of the spleen. One showed definite invasion of the splenic vein, and another showed massive invasion of a relatively large splenic trabecula in which a lumen could not be demonstrated. In the 3rd case, the liver biopsy taken at laparotomy showed only nonspecific portal infiltrates at a time when invasion of intrasplenic veins was present. Definite Hodgkin's disease was found in a repeat liver biopsy 7 months later.

The observations of vascular invasion in the spleen removed during a laparotomy for staging of Hodgkin's may enhance the ability to predict hepatic involvement. This observation also suggests that hematogenous dissemination of Hodgkin's disease via the portal system may be an important pathway for the spread of disease to the liver.

Our studies of the frequency, the biological and clinical significance of vascular invasion in surgical material of lymph nodes, spleen, or both of patients with Hodgkin's disease suggest that the search for this feature in histological material obtained prior to therapy should be recommended as an additional procedure in the assessment of the extent of the disease. This recommendation is made because the presence of vascular invasion suggests, even in the more favorable histological types, a more extensive involvement than initially evident, particularly with respect to the presence of extranodal disease. The demonstration of vascular invasion in diagnostic lymph node biopsies, spleen, or both should be investigated in appropriate studies as a possible indication for systemic therapy in place of or in addition to therapy directed solely against lymph nodes.

References

1. Bouroncle, B. A. Sternberg-Reed Cells in the Peripheral Blood of Patients with Hodgkin's Disease. Blood, *27:* 544–555, 1966.
2. Engeset, A., Brennhovd, I. O., Christensen, I., Hagen, S., Hoeg, K., Høst, H., Liverud, K., and Nesheim, A. Sternberg-Reed Cells in the Thoracic Duct Lymph of Patients with Hodgkin's Disease. Blood, *31:* 99–103, 1968.
3. Lukes, R. J., and Butler, J. J. The Pathology and Nomenclature of Hodgkin's Disease. Cancer Res., *26:* 1063–1081, 1966.
4. Lukes, R. J., Carver, L., Hall, T., Rappaport, H., and Rubin, P. Report of Nomenclature Committee. Cancer Res., *26* (Part 1)*:* 1311, 1966.
5. Nickson, J. J. Hodgkin's Disease Clinical Trial. Cancer Res., *26* (Part 1)*:* 1279–1283, 1966.
6. Rappaport, H., and Strum, S. B. Vascular Invasion in Hodgkin's Disease: Its Incidence and Relationship to the Spread of the Disease. Cancer, *25:* 1304–1313, 1970.
7. Rosenberg, S. A., and Kaplan, H. S. Evidence for an Orderly Progression in the Spread of Hodgkin's Disease. Cancer Res., *26* (Part 1)*:* 1225–1230, 1966.
8. Rosenberg, S. A., and Kaplan, A. Hodgkin's Disease and Other Malignant Lymphomas. Calif. Med., *113:* 23–38, 1970.
9. Strum, S. B., Allen, L. W., and Rappaport, H. Vascular Invasion in Hodgkin's Disease: Its Relationship to Involvement of the Spleen and Other Extra-Nodal Sites. Cancer, in press.
10. Strum, S. B., Hutchison, G. B., Park J. K., and Rappaport, H. Further Observations on the Biologic Significance of Vascular Invasion in Hodgkin's Disease. Cancer, *27:* 1–6, 1971.
11. Strum, S. B., and Rappaport, H. The Significance of Focal Involvement of Lymph Nodes for the Diagnosis and Staging of Hodgkin's Disease. Cancer, *25:* 1312–1317, 1970.