and media in places, so that there was only thickened intima between the tuberculous tissue and the lumen of the aorta. In view of these findings we consider that this patient had a mycotic aneurysm of the abdominal aorta of the extra-vascular erosive type, the tuberculous disease in the lymph nodes having destroyed the adventitia and media of the aorta so that an aneurysm resulted.

Aortography performed one month after operation showed that there was good filling of the aorta, the orlon prosthesis, and the iliac arteries (Fig. 2). The patient has returned to work in his city office.

FIG. 2.—Post-operative aortogram showing the orlon cloth prosthesis in position.

Previous Reported Cases

In 1913 Haythorn reviewed the literature on tuberculosis of arteries and reported finding only one record of an aneurysm of the abdominal aorta due to erosion of the arterial wall by tuberculous granulation tissue. Since then occasional cases have been reported, and by 1933 Gellerstedt and Säfwenberg were able to collect 21 cases of aneurysm of the abdominal aorta due to erosion by tuberculous granulation tissue. The literature since 1933 contains reports of two more cases (Owens and Bass, 1944; Scott, Maxwell, and Grimes 1949), so it appears that, including our patient, only 24 cases of tuberculous mycotic aneurysm of the abdominal aorta have been recorded. Also, as the other reports were based upon material obtained at necropsy, ours is the first record of the diagnosis of this lesion with the subsequent recovery of the patient.

Summary

A case is reported of an aneurysm of the abdominal aorta due to erosion of the aortic wall by tuberculous granulation tissue from caseating abdominal lymph nodes. Treatment, which included resection of the aneurysm followed by reconstruction of the aorta with a prosthesis of orlon cloth, has enabled the patient to return to work.

Of 23 such cases previously reported in the literature, all were fatal.

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THE NUCLEIC ACID CONTENT OF BONE-MARROW CELLS IN PERNICIOUS ANAEMIA

BY

HUGH BREBNER, M.D., M.R.C.P.

Senior Registrar, Manchester Royal Infirmary

AND

JOHN F. WILKINSON, M.D., M.Sc., Ph.D.
F.R.C.P., F.R.I.C.

Physician and Director, Department of Haematology,
Manchester Royal Infirmary

The understanding of disordered processes of cell development has in the past depended mainly on the observation of morphological differences between cells at various stages of their life-cycle. By modern chemical and histochemical methods it is possible to analyse, in certain of the chemical constituents of cells, changes that are probably the underlying cause of alterations in morphology. The disordered cell development that occurs in pernicious anaemia is particularly worthy of study, for the cells are easily accessible, and, furthermore, the disease process is one that can be corrected by appropriate treatment. Of the therapeutic agents that restore the bone-marrow picture to normal, the most potent is vitamin B₁₂, which appears to be closely connected with the synthesis of nucleic acids that accompanies growth (Wright *et al.*, 1948; Rose and Schweigert, 1952). In spite of the fact that, from bacterial and animal experiments, much has been learned about the biological activity of vitamin B₁₂, very little is known of its mode of action in pernicious anaemia. There is as yet no real understanding of the nature of the cellular defect that is apparently remedied by vitamin B₁₂, but it seems possible that it may be a defect in nucleic acid metabolism.

It has been demonstrated that the deep basophilia of the cytoplasm of early megaloblasts is due to the presence of ribonucleic acid (Thorell, 1947; White, 1947); it has also been shown by chemical analysis that the mean amount of ribonucleic acid phosphorus (R.N.A.P.) per cell is much greater in megaloblastic marrows than in normal marrows (Davidson *et al.*, 1951). Accumulation of ribonucleic acid in the cell cytoplasm appears, therefore, to be a specific feature of megaloblastic haemopoiesis. That abnormalities may also occur in the cellular content of deoxyribonucleic acid (D.N.A.) was suggested by La Cour (1944), who, by histological methods, demonstrated chromosomal abnormalities in both the red-cell and the white-cell series in pernicious anaemia. The mean deoxyribonucleic acid phosphorus (D.N.A.P.) per cell is, in fact, greater in megaloblastic marrows than in normal marrows (Davidson *et al.*, 1951, 1953). On the other hand, it has been shown by microspectrophotometric techniques that the D.N.A. content of individual megaloblasts is no greater than that of normoblasts of comparable nuclear size (Reisner and Korson, 1951; Marinone, 1951).

In this paper we present the results of chemical estimations of the nucleic acid content of bone-marrow cells in pernicious anaemia, and make an attempt to assess the significance of the findings, particularly with regard to the cellular content of D.N.A.P. Quantitative changes in nucleic acids during *in vitro* culture will be described elsewhere.

Experimental Work

Estimations of nucleic acids were carried out on suspensions of blood or bone marrow containing a known number of nucleated cells per cubic millimetre, thus enabling the mean amount of nucleic acid per cell to be calculated. Sternal marrow samples were taken from 18 patients suffering from classical pernicious anaemia, and the series of observations has been compared, not with normal marrows, but with observations on nine bone marrows showing normoblastic hyperplasia. Cases in the latter group were selected because the clinical and bone-marrow findings suggested that the erythroid elements of the marrow might be functioning near their maximum capacity, which would cause the marrow aspirates to contain a high proportion of "active" cells. The use of such cases was thought to afford a better comparison between megaloblastic and normoblastic haemopoiesis than the use of normal subjects, from whom marrow aspirates contain a high proportion of mature leucocytes. In addition, seven samples of normal blood were examined.

Technique

Blood samples of approximately 3 ml., obtained by venepuncture, were collected in bijou bottles containing as an anticoagulant the standard mixture of ammonium and potassium oxalate. Sternal marrow puncture was performed by the standard technique, employing a Salah sternal puncture needle and an all-glass syringe, both silicined. When approximately 1 ml. of the blood-and-marrow mixture had been aspirated, the syringe was quickly detached from the needle and its contents were ejected into 20 ml. of normal saline in a universal container. Although anticoagulants were not used, it was found that if the mixture of marrow and saline was shaken for 20 minutes clotting did not occur. After centrifugation for five minutes, the fatty debris was taken from the top of the liquid and the supernatant fluid removed by Pasteur pipette until about 2 ml. remained. Agitation then produced an even cellular suspension, on which three independent nucleated cell counts were made.

Nucleic acids were estimated as nucleic acid phosphorus (N.A.P.) by the technique described by Davidson *et al.* (1949), in which the D.N.A.P. and R.N.A.P. fractions are separated by the method of Schmidt and Thannhauser (1945). The amount of inorganic phosphorus present in the two fractions was measured by the colorimetric method of Berenblum and Chain (1938). Readings were taken on a calibrated Unicam spectrophotometer, and by this technique satisfactory measurement was obtained of quantities of phosphorus between 0.5 and 4 μg .

Results

In Table I, II, and III are recorded the mean values of nucleic acid phosphorus per cell found in normal leucocytes, in cells from bone marrows undergoing active normoblastic hyperplasia, and in cells from the bone marrow of patients suffering from pernicious anaemia. No figures are recorded for the mean amount of R.N.A.P. per leucocyte, because difficulty was experienced in extracting all the acid-soluble phosphorus compounds from whole blood, thus making the R.N.A.P. figures unreliable.

The mean value for the cellular content of R.N.A.P. is higher in the pernicious anaemia series than in the normoblastic ($t=2.548$; $n=19$; $P<0.02$); this finding is consistent with the observations that the early megaloblasts pre-

sent in the marrow of pernicious anaemia are unusually rich in R.N.A. (White, 1947; Thorell, 1947). The distribution of the individual D.N.A.P. results in each series is depicted in the Chart, and the grand means are compared in Table IV. The lack of a significant difference between the marrow cells

TABLE II.—Mean Cellular Content of D.N.A.P. and R.N.A.P. in the Bone Marrow of Patients in whom Clinical and Bone-marrow Findings indicated the Presence of Active Blood Regeneration

Case No.	Age and Sex	Diagnosis	Hb g./ml.	R.B.C. in mills./c.mm.	Retic. %	W.B.C. per c.mm.	D.N.A.P.	R.N.A.P.
							$\mu\text{g.} \times 10^{-7}$ per cell	$\mu\text{g.} \times 10^{-7}$ per cell
2	18 M	Acholuric jaundice	12.9	4.2	10.3	8,900	7.0	15.5
8	65 M	Paroxysmal nocturnal haemoglobinuria	6.2	1.3	11.4	9,000	6.2	—
17	23 M	Familial haemolytic anaemia	10.8	3.3	13.1	5,400	9.8	4.8
19	73 F	Cancer of bladder	7.6	2.4	—	6,050	8.8	5.4
25	61 F	Haematuria + Acholuric jaundice	8.6	3.0	30.4	4,200	6.2	14.5
27	35 F	" "	11.3	4.5	10.4	16,200	8.9	16.0
34	51 M	Acquired haemolytic anaemia	3.5	0.8	60.6	11,350	5.1	—
41	51 M	Acholuric jaundice	12.9	4.2	4.4	10,050	8.6	7.3
22	48 F	Chronic haemolytic anaemia	10.1	3.5	6.2	7,600	16.0	11.3

Grand mean D.N.A.P. per cell = 8.51×10^{-7} μg . Grand mean R.N.A.P. per cell = 10.69×10^{-7} μg .

TABLE III.—Mean Cellular Content of D.N.A.P. and R.N.A.P. in the Bone-marrow of Pernicious Anaemia

Case No.	Age and Sex	Hb g./ml.	R.B.C. mills./c.mm.	D.N.A.P. $\mu\text{g.} \times 10^{-7}$ per Cell	R.N.A.P. $\mu\text{g.} \times 10^{-7}$ per Cell
1	41 M	7.7	1.93	5.8	10.8
9	61 F	9.2	2.05	6.25	—
10	58 F	7.7	1.77	7.5	—
14	38 F	3.7	1.10	6.65	5.6
				(7.2, 6.1)	
15	65 M	5.0	1.10	5.7	28.2
				(6.7, 6.5, 4.0)	(32.8, 40.2, 11.5)
18	76 M	5.9	1.36	6.7	20.0
20	58 M	8.6	2.18	10.2	22.1
				(10.6, 8.6, 11.4)	(23.0, 17.3, 26.0)
23	54 F	7.4	1.63	10.45	20.3
				(11.2, 9.7)	(20.0, 20.6)
24	62 F	8.3	2.11	10.5	12.2
				(8.6, 11.1, 11.8)	(14.0, 11.1, 11.6)
26	44 F	6.8	1.51	13.0	27.3
				(12.1, 13.9)	(27.2, 27.5)
28	69 M	8.0	1.82	16.1	28.6
29	64 M	4.7	1.07	8.3	24.3
				(10.9, 5.7)	(25.6, 23.0)
35	67 F	7.0	1.59	6.1	11.7
36	41 M	5.3	1.40	14.6	—
38	63 F	6.8	1.41	4.5	12.7
				(5.9, 3.0)	(16.2, 9.2)
39	68 F	8.4	1.94	7.1	—
42	42 F	5.0	1.07	12.9	14.7
43	38 F	6.7	1.56	6.9	20.9
				(6.9, 7.1, 6.8)	(21.5, 19.2, 22.0)

Figures in parentheses are individual results in cases in which estimations were carried out on more than one marrow suspension from the same patient. Clearly, the wide range of nucleic acids in pernicious anaemia is due, not entirely to experimental error, but mainly to a real case-to-case difference. Grand mean D.N.A.P. per cell = 8.85×10^{-7} μg . Grand mean R.N.A.P. per cell = 18.53×10^{-7} μg .

TABLE IV.—Comparison of the Mean Nucleic Acid Phosphorus Per Cell in Blood and Bone-marrow Cells (Numbers of Cases in Parentheses)

	A	B	C
	Blood Leucocytes	Active Normoblastic Marrow	Pernicious Anaemia Marrow
D.N.A.P. as $\mu\text{g.} \times 10^{-7}$ /cell	5.89 (7)	8.51 (9)	8.85 (18)
R.N.A.P. as $\mu\text{g.} \times 10^{-7}$ /cell	—	10.69 (7)	18.53 (14)

The difference between the D.N.A.P. means in A and B is not significant ($t=2.116$, $n=14$, $P=0.053$); that between the means in A and C is of borderline significance ($t=2.258$, $n=23$, $P=0.05$). The R.N.A.P. mean of C is greater than that of B ($t=2.548$, $n=19$, $P=0.02$).

TABLE I.—Mean D.N.A.P. of Normal Blood Leucocytes

Case No.	Mean D.N.A.P. per cell in $\mu\text{g.} \times 10^{-7}$
30	5.67
31	5.37
32	5.39
37	5.70
44	6.92
45	5.90
46	6.31

Mean = 5.89×10^{-7} μg .

as compared with leucocytes is due to the relatively small numbers in two of the three series, but the present object is not to demonstrate such a difference but rather to compare the bone marrow of pernicious anaemia with that of active normoblastic hyperplasia. The results indicate that the cellular content of D.N.A.P. is quantitatively similar in these two types of bone marrow.

Chart showing the mean cellular content of D.N.A.P.

Discussion

Appreciation of the significance of the results depends on a knowledge of the role of D.N.A. in the life of the cell, in which it forms an essential part of nuclear chromatin (Brachet, 1933). A considerable amount of evidence points towards the idea that, in any given genus, the mean content of D.N.A. per mature diploid cell is constant, being twice that of the haploid sperm (Boivin *et al.*, 1948; Vendrely and Vendrely, 1948, 1949; Mandel *et al.*, 1950; Davidson *et al.*, 1951).

During cell proliferation, changes must of course occur, and these have, in recent years, been studied intensively by a number of workers using quantitative spectrophotometric techniques that enable the D.N.A. content of individual cells to be estimated. It has been shown that D.N.A. increases during interphase until at prophase twice as much is present as in the non-dividing cell; hence daughter nuclei have the normal complement of D.N.A. (Swift, 1950; Pasteels and Lison, 1950; Walker and Yates, 1952). Thus if the mean amount of D.N.A. per cell in a population is high it may be presumed that a high proportion of cells are of actively dividing type, or that mitotic abnormalities have occurred, resulting in polyploid cells, or that some fundamental abnormality exists in the chromosomal content of D.N.A.

The blood leucocyte is a good example of a mature non-dividing diploid cell, and the results recorded in Table I indicate that in normal subjects the mean amount of D.N.A.P. per leucocyte is remarkably constant. Moreover, as the results are in agreement with published figures for the D.N.A.P. content of other mammalian cells (Table V), they lend support to the theory of constant D.N.A. per cell.

Tables II and III demonstrate the wide variability that occurs between different bone marrows as regards the mean

TABLE V.—Mean D.N.A.P. Content of Various Mammalian Cells. D.N.A.P. as $\mu\text{g.} \times 10^{-7}$ Per Cell. (Figures Published as D.N.A. Have Been Multiplied by 0.099.)

Author	Normal Sperms	Normal Leuco-cytes	Other Mature Cells*	Normal Marrow	Active Normoblastic Marrow	Megaloblastic Marrow
Boivin <i>et al.</i> (1948)	3.37	—	6.43	—	—	—
Mirsky and Ris (1949) ..	2.79	—	6.14 to 8.37	—	—	—
Mandel <i>et al.</i> (1950)	—	6.78	—	—	—	—
Davidson <i>et al.</i> (1951)	3.14	7.04	8.3 to 10.0	8.69	—	12.7
Present ..	—	5.89	—	—	8.51	8.85

* Cells from liver, kidney, etc. (may include some dividing cells).

content of D.N.A.P. per cell, a finding that has also been observed by Davidson *et al.* (1951) and by Menten and Willms (1952). In a series of 31 cases showing a preponderance of erythroblasts in bone-marrow smears, the latter authors found values of D.N.A.P. per cell ranging from 4.0 to $26.3 \times 10^{-7} \mu\text{g.}$ Included in their series were six cases of pernicious anaemia, the mean value for those being $16.0 \times 10^{-7} \mu\text{g./cell.}$ Values higher than the present figure of 8.81 were also found by Davidson *et al.*, who in 17 cases of megaloblastic marrows reported a mean value of $12.7 \times 10^{-7} \mu\text{g. D.N.A.P. per cell.}$ However, the latter figure represents approximately 180% of their mean value of D.N.A.P. per leucocyte (7.04), whereas the mean of the present series represents 150% of the present leucocyte value, so that differences between the present results and those of Davidson *et al.* probably depend on technique and may not be of great significance. Other important findings of Davidson *et al.* were that the mean content of D.N.A.P. per cell in megaloblastic marrows was higher than in normal marrows, marrows from cases of leukaemia, or marrows of iron-deficiency anaemia. Those authors considered that the high values in their megaloblastic series indicated the presence of nuclear abnormality, either polyploidy or polyteny. Such a theory is in agreement with the frequent finding of abnormal mitotic figures in megaloblastic marrow smears and with the suggestion, made by La Cour (1944), that tetraploid cells are common in the bone marrow of pernicious anaemia.

The present results, although they do not in any way invalidate the theory of nuclear abnormalities, are more in favour of the alternative hypothesis that the high cellular D.N.A.P. of the pernicious anaemia marrow is due to the presence of proliferating cells. In the hyperplastic normoblastic marrows a considerable amount of cellular multiplication was going on, and the figures for the mean cellular D.N.A.P. must depend on the fact that there was a mixed population of cells, some containing the diploid quantity, others containing an excess of D.N.A.P. in preparation for mitosis. The mean content of D.N.A.P. per cell for the whole series of pernicious-anaemia marrows is similar to that of the normoblastic series, which suggests that the same factors may have operated in each case.

Thus it would appear that the high cellular content of D.N.A.P. encountered in the bone marrow in pernicious anaemia may be due, not to some specific abnormality of mitoses, but to the prevalence in the marrow of cells of proliferative type.

Summary

Chemical estimations of nucleic acids have been carried out on suspensions of blood and bone-marrow cells.

The cellular content of R.N.A.P. is higher in pernicious-anaemia marrows than in hyperactive normoblastic marrows; accumulation of R.N.A. in the cell is therefore a specific feature of megaloblastic haemopoiesis.

The cells of pernicious-anaemia marrows and those of hyperactive normoblastic marrows contain approximately the same amount of D.N.A.P., the figures in each

case being about 150% of values for blood leucocytes. This finding suggests that many of the cells of the pernicious-anaemia marrow are of proliferative type and are engaged in building up D.N.A. in preparation for mitosis.

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CEREBROVASCULAR ACCIDENTS IN HOSPITALS FOR ACUTE CASES

BY

P. E. BROWN, M.B., M.R.C.P.Ed., D.P.H.

(From the Department of Public Health and Social Medicine, University of Edinburgh)

Patients with intracranial vascular lesions, especially those over 70, are a serious problem to the general practitioner called upon to arrange suitable care and accommodation. Hospitals are unwilling to admit cases likely to remain as chronic hemiplegics occupying acute medical beds for long periods. Those who do get admitted are often accepted with reluctance after tedious negotiations which are irritating to both house officers and practitioners. The attitude of hospital staffs would be easier to defend if it were not based as much on traditional prejudice as on an appreciation of what actually happens to these cases after admission.

This paper reports an analysis of 239 cases admitted after cerebrovascular accidents in which the length of stay in hospital and the eventual disposal have been investigated.

Material and Methods

The in-patient records of two hospitals were examined for the two-year period 1948-9. Extracts of age, sex, length of stay, and disposal were made of all cases admitted with the diagnoses of cerebral haemorrhage, thrombosis and embolism, subarachnoid haemorrhage, hemiplegia, and coma of unknown cause. The 239 cases collected were analysed with respect to length of stay in hospital by age and sex. The duration of symptoms before admission was not studied in detail, but only three of the cases were admitted as long-standing hemiplegias and the majority were victims of recent cerebrovascular accidents.

The two hospitals selected are situated in a large industrial county town in the midlands of England. Hospital A is an ex-voluntary hospital of over 600 beds with a casualty department. Hospital B is an ex-municipal hospital of 400 beds with no casualty department. Between them the two hospitals accommodate most of the acute medical admissions from a population of 600,000.

Results

Table I summarizes the patients with intracranial vascular lesions admitted to the two hospitals. Of the 239 patients 165 (69%) died in hospital and 74 (31%) were discharged. Of the discharges 61 (25.5% of admissions) were sent home, and 13 (5.4% of admissions) were transferred to other hospitals.

TABLE I.—Summary of 239 Cases with Intracranial Vascular Lesions Admitted to Hospital in 1948-9

	Hospital A			Hospital B			Total		
	Men	Women	Total	Men	Women	Total	Men	Women	Total
Discharged home	10	17	27	12	22	34	22	39	61
Transferred	6	3	9	4	—	4	10	3	13
Died	34	55	89	27	49	76	61	104	165
Total	50	75	125	43	71	114	93	146	239

Cumulative frequency diagram showing the percentage of patients remaining in hospital, discharged, or dead after a given period of time.

The length of stay in hospital is shown in the form of a cumulative frequency diagram. Over 30% of admissions died in the first two days. By the end of 10 days 54.2% had died and 5% had been discharged, leaving 40.8% still in hospital. After six weeks 67.6% had died, 21.8% had been discharged, and 10.6% remained. Only 3.8% stayed in hospital more than 12 weeks.

Table II gives the length of stay in hospital according to age. Of those over 60 years old 10 out of 141 (7.1%) stayed in more than six weeks, compared with 14 out of 98 (14.3%) of those under 60. The difference between these percentages is not significant. The number of deaths in the higher

TABLE II.—Length of Stay in Hospital According to Age. Discharges by Death are Given in Parentheses

Week of Discharge	Age Group						
	20-29	30-39	40-49	50-59	60-69	70-79	80-89
1-2	3 (2)	3 (3)	16 (13)	35 (28)	58 (54)	40 (37)	8 (8)
3-4	—	1	7 (2)	10 (3)	8 (3)	6 (4)	2 (2)
5-6	—	1	6	2	5 (2)	4	—
7-8	—	—	1	3	1	—	—
9-10	—	1 (1)	1	3 (1)	3 (1)	—	1
11-12	—	—	—	1 (1)	1 (1)	—	—
Over 12	—	1	—	4	3	1	—
Total	3 (2)	7 (4)	31 (15)	57 (32)	79 (61)	51 (41)	11 (10)