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VITAMIN A IN INFECTIVE HEPATITIS

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The Liver is the main storage place in the body for vitamin A. Liver diseases of various types may therefore be expected to interfere in one way or another with the metabolism and physiological activity of vitamin A. Xerosis in various forms, reduced vitamin A levels in the liver and blood plasma, and defective dark-adaptation have all been reported to result from deficiency of vitamin A secondary to damage to the liver.

Xerosis.—The early literature on the occurrence of xerosis in liver disease has been reviewed fully by Altschule (1935). Thus more than 50 years ago, before the discovery of vitamin A, Thompson (1894) described the development of keratomalacia in a 2-months-old infant suffering from jaundice. Later Bloch (1924) observed xerosis and arrested growth in a jaundiced child. The condition persisted even when cod-liver oil was administered, but cleared up when bile appeared in the faeces.

Vitamin A Levels.—Chemical estimations of vitamin A were first applied to subjects who had died of cirrhosis of the liver, and several investigators agreed that the vitamin A reserves of the liver were usually low and often undetectable (Wolff, 1932; Breusch and Scalabrino, 1934; Moore, 1937). In acute liver diseases, however, the hepatic reserves may not be depleted, although the level in the blood plasma is often low (Popper, Steigmann, and Zevin, 1943).

Dark-Adaptation.—With defective dark-adaptation as a criterion most workers have found that the vitamin A status is disturbed, although opinions differ as to the value of vitamin A therapy in correcting this abnormality (Jeghers, 1937; Haig, Hecht, and Patek, 1938; Patek and Haig, 1939; Ezickson, 1939; Wohl and Feldman, 1940, 1941).

The present paper deals with abnormalities in vitamin A metabolism in infective hepatitis only. In preliminary experiments (described below in an Addendum) defective dark-adaptation was used as an indication of vitamin A abnormality. It was found that the mean final rod threshold in patients tested soon after admission to hospital differed significantly from the mean value for the same patients after recovery, and also from that of normal young adults. In our main investigation, however, chemical methods were exclusively used. Vitamin A and carotenoids in the plasma were estimated in the early stages of the disease and during convalescence. Parallel examinations were made in some cases of the serum bilirubin, plasma prothrombin, and hippuric acid synthesis. Other patients were given massive doses of vitamin A, with a view to studying the efficiency of its absorption from the intestines and of

its transfer to the blood plasma. The vitamin A reserves of the liver were estimated in three fatal cases of hepatitis.

Material and Methods

The main investigation was made on 32 military patients admitted to an E.M.S. hospital under the M.R.C. Jaundice Research Team: 30 suffered from infective hepatitis and two from arsenotherapy jaundice, which presents a similar biochemical picture. None had a history of transfusion. The ages ranged from 18 to 41 (average 25). The patients received a normal diet, without restriction of fat and without extra protein or vitamins. A few cases were included in a clinical trial with methionine, but this substance did not appear to be beneficial (Wilson, Pollock, and Harris, 1945) or to affect the vitamin A status.

The technique used to estimate vitamin A and carotene was essentially that of Kimble (1939), as modified by Yudkin (1941) and by ourselves. The plasma was shaken with alcohol and petrol ether. Carotenoids were estimated in the extract by their yellow colour. The extract was then evaporated, and the vitamin A estimated by the blue colour produced on treatment with antimony trichloride, an allowance being made for the colour contributed by carotenoids to this reaction. Both yellow and blue colours were measured in a photoelectric absorptiometer provided with filters. In calibrating this instrument against vitamin A acetate a conversion factor of 1,600 between $E_{1\%}^{1\text{cm}}$ at 328 m μ . and i.u. of vitamin A per g. was assumed. The calibration for carotenoids was against the International Standard solution of β carotene, and for convenience results have been expressed as though all the carotenoids of the plasma were β carotene. Because of the presence of xanthophyll, α carotene, etc., the actual "biological value" of the carotenoid fractions was presumably somewhat less than the figures given. Values for both vitamin A and carotene are expressed in terms of i.u. per 100 ml. of plasma.

Serum bilirubin was estimated by the method of Mallory and Evelyn (1937). The prothrombin indices were determined by a method similar to that described by Harris (1943), using viper venom. Quick's (1939) modification of the hippuric acid synthesis test was employed, the results being expressed as grammes of sodium benzoate detoxicated in the first hour after administration.

Plasma Vitamin A and Carotene in Infective Hepatitis

Infection and Recovery.—Observations of plasma vitamin A and carotene at various stages of the disease

were made in 32 patients (Figs. 1 and 2) and the mean values are shown in Table I, grouped in five-day periods from the date of admission. The earliest group (0-4 days after admission) show a mean value of 54.6 i.u. per 100 ml., which may be compared with a mean value of 118 i.u. for a group of 41 healthy adults (Leitner and Moore, 1946).

FIG. 1.—Plasma vitamin A in 32 cases of infective hepatitis.

Successive estimations show a continuous rise until a mean value of 114.3 i.u. is reached for the group at 15-19 days after admission. Subsequently the observations are too few to be relied upon, and the readings from 20 to 29 days were combined, as were those for over 30 days from admission. There was, however, a suggestion in individual

TABLE I.—Plasma Vitamin A and Carotene in 32 Cases of Infective Hepatitis

Days after Admission	Vitamin A			Carotene		
	No. of Observations	Mean Plasma Vitamin A I.U. per 100 ml.	Standard "Error"	No. of Observations	Mean Plasma Carotene I.U. per 100 ml.	Standard "Error"
0-4 ..	27	54.6	± 3.7	27	67.2	± 6.1
5-9 ..	10	64.8	± 6.0	10	51.3	± 9.0
10-14 ..	9	80.4	± 12.0	10	78.9	± 10.3
15-19 ..	10	114.3	± 13.2	10	84.8	± 7.4
20-29 ..	12	124.3	± 11.9	11	118.2	± 17.3
Over 30 ..	15	123.9	± 12.2	15	114.8	± 9.8

The number of observations for vitamin A and carotene does not tally in every age group, because on two occasions a reading was obtained for one without a corresponding reading for the other.

patients that the plasma vitamin A may be unusually high during convalescence. This phenomenon has been noticed by Josephs (1943) in young children recovering from pneumonia. Plasma carotene values were also low in the early stages of the disease, the mean for the first period of five days being 67.2 i.u., as compared with 161 i.u. for healthy adults. During recovery the mean rose to 118.2 i.u. for 20-29 days after admission, but few cases reached the

normal mean even 30 days or more after admission. The correlation between corresponding readings for vitamin A and carotene was low ($r=0.2143$ for ungrouped data; $N=81$) and was barely significant.

Effect of Age, Fever, and Anorexia.—The effects of these factors were examined in 25 patients in whom blood

FIG. 2.—Plasma carotene in 32 cases of infective hepatitis.

vitamin A had been estimated within four days of admission to hospital. If there was more than one such reading the earlier value was used. In Table II these readings are grouped according to the age of the patient, the highest temperature recorded, and the duration of anorexia; the significance of the results is also estimated. The duration

TABLE II.—Relation of Plasma Vitamin A to Age, Fever, and Anorexia, 25 Cases With Values Obtained Within Four Days of Admission

	No. of Cases	Mean Plasma Vitamin A I.U. per 100 ml.	t*
Age in years:			
18-24 ..	13	59.1	2.070
25-34 ..	9	47.1	
Over 35 ..	3	38.7	
Maximum temperature:			
Under 99° ..	10	61.3	2.159
99°-100° ..	12	45.5	
100°-101° ..	3	51.0	
Anorexia:			
Nil ..	6	52.0	0.103
1-5 days ..	4	58.0	
6-10 ..	12	53.2	
Over 10 days ..	3	43.0	
Total ..	25	52.3	

* With 23° of freedom, t must equal or exceed 2.069 to give $p = 0.05$.

of anorexia did not appear to influence the plasma vitamin A. Higher mean values, however, were found for those under 25 years of age than for those over, and for those who showed no significant fever—under 99° F.

(37.2° C.)—than for those who did; both these differences were shown by the *t* test to be significant. It cannot be excluded that the effect of age was due to greater severity of the disease in the older men and to a higher incidence of fever in the more severe cases, and not to a direct relation between age or fever and plasma vitamin A. In fever other investigations have shown that there is a direct lowering of the plasma vitamin A, since this action is found in artificial pyrexia as well as infectious fevers (Clausen and McCoord, 1938). In any case this mechanism can only be of subsidiary importance, since instances of abnormally low plasma levels were found in the younger age groups and in the group without fever.

Relation to Bilirubinaemia and Liver Function Tests.—In nine cases in which serial estimations of plasma vitamin A were made, serial readings were also obtained of serum bilirubin, plasma prothrombin, and hippuric acid synthesis. The mean values for five-day groups are shown in Table III. No observations are available on the pre-

TABLE III.—*Relation of Plasma Vitamin A and Carotene to Serum Bilirubin, Prothrombin Index, and Hippuric Acid Synthesis. 9 Cases with 4 or more Serial Observations of Vitamin A and Carotene*

Days	Plasma Vitamin A I.U. per 100 ml.		Plasma Carotene I.U. per 100 ml.		Serum Bilirubin mg. per 100 ml.		Prothrombin Index		I.V. Hippuric Acid Synthesis	
	No. of Obs.	Mean	No. of Obs.	Mean	No. of Obs.	Mean	No. of Obs.	Mean %	No. of Obs.	Mean
0-4	11	53.1	11	57.7	13	6.97	11	75.7	7	0.56
5-9	9	65.9	9	46.7	12	8.66	5	72.0	7	0.60
10-14	5	91.0	6	80.5	7	7.56	5	90.3	3	0.92
15-19	7	113.7	7	84.9	6	2.37	4	94.2		
20-29	9	139.6	9	132.3	10	1.54	6	98.5	8	0.88
Over 30	6	147.0	6	126.0	13	1.11	4	98.0		

icteric stage of the disease, since the diagnosis of infective hepatitis is seldom made before the appearance of jaundice. However, most patients were admitted with a rising serum bilirubin, and the highest value was found in the 5-9-day group. It is notable that the plasma vitamin A was increasing from the time that the patients were first seen, and this improvement may be compared with the results of hippuric acid synthesis tests, which were also improving or stationary at a time when the serum bilirubin was still rising. Pollock (1945) has dealt with the improvement in hippuric acid synthesis in infective hepatitis and concludes that maximum liver damage in this disease occurs in the pre-icteric stage. In our investigation the parallelism between plasma vitamin A and hippuric acid synthesis was fairly close in individual cases. Curves for two illustrative cases are shown in Fig. 3.

Relation to Prothrombin Index.—Prothrombin values were low in the early stages of the disease, though spontaneous haemorrhages were not observed. During recovery the plasma prothrombin returned to normal in general parallelism with the plasma vitamin A. Examination of individual case records, however, revealed no close or regular relationship. The prothrombin values exhibited a wider day-to-day variation, and tended to return to normal at about the same time or later than the plasma vitamin A. Plasma was also examined from five patients not included in this series who persistently had very low prothrombin values. The plasma vitamin A in these cases was not unusually depressed, and when the prothrombin index rose in response to parenteral administration of vitamin K analogues there was no parallel alteration of the plasma vitamin A. In four of the five patients, however, a rise in plasma vitamin A occurred some time after the prothrombin index had returned to normal.

Absorption of Massive Doses of Vitamin A

Several workers (Breese and McCoord, 1940; Ralli, Bauman, and Roberts, 1941; Breese, Watkins, and McCoord, 1942; Spector, McKhann, and Meserve, 1943; Popper, Steigmann, and Zevin, 1943; Steigmann and Popper, 1944; Aldersberg and Sobotka, 1944) have measured the plasma vitamin A at intervals after giving massive doses of the vitamin.* In normal subjects the

plasma level increases rapidly after dosing to a maximum in about five hours, and usually subsides to the original value within 24 hours. We have confirmed the finding that in infective hepatitis the plasma vitamin A sometimes fails to respond to dosing although our results for a series of cases have been erratic. Thus in preliminary tests two patients in the early stages of the disease were given 96,000 i.u. of vitamin A in the form of four "preparin" capsules. Subject C, whose resting plasma vitamin A of 84 i.u. per 100 ml. was but little below the normal mean, had a value of 77 i.u. after dosing. Even after four further doses there was still no significant increase in the plasma vitamin A. In contrast, patient D when first tested had a plasma vitamin A of only 50 i.u. Greater derangement of vitamin A metabolism than the

FIG. 3.—Relation of plasma vitamin A to serum bilirubin and hippuric acid synthesis. A, arsenotherapy jaundice. B, infective hepatitis. ●—●=Plasma vitamin A (i.u. per 100 ml.). ×—×=Serum bilirubin (mg. % × 10). □—□=Intravenous hippuric acid synthesis (g. sodium benzoate detoxicated in 1st hour × 100).

* Experiments by this procedure are called "tolerance tests" by some workers. The subject's resistance to toxic excess of vitamin A (Moore and Wang, 1945) is not, however, under consideration, and mention of "tolerance" is perhaps misleading: "absorption tests" seems a better description.

first subject might therefore have been expected. Nevertheless, after dosing, the plasma vitamin A responded with a rise to 299 i.u., which can be considered normal.

Reaction to Isolated Doses of Vitamin A.—In further experiments (Table IV) five patients were given single doses of 96,000

TABLE IV.—*Effect of Isolated Massive Doses (96,000 I.U.) of Vitamin A on the Vitamin A Content of the Blood Plasma, Faeces, and Urine*

Case	Days after Admin.	Plasma Vitamin A (I.U. per 100 ml.)			Vitamin A Excretion; Total Daily I.U.	
		Before Dose	After Dose	Difference	Faeces	Urine
<i>Infective Hepatitis</i>						
E	2	75	354	279	—	45
	34	109	258	149	—	45
F	2	67	130	63	—	0
	34	44	251	207	—	0
G	1	32	444	412	39,000	0
	34	82	171	89	0	0
H	4	35	67	32	58,000	56
	37	72	107	35	0	0
	73	78	242	164	0	0
I	2	33	186	133	1,000	0
	25	99	162	63	0	0
				(Mean 148)		
<i>Normal Subjects</i>						
J	—	103	309	206	0	0
K	—	128	932	804	0	0
L	—	179	403	224	0	0

i.u. of vitamin A at an early stage of the disease, and again on one or more occasions during the later stages. Estimations were made not only of plasma vitamin A but of the excretion of the vitamin in the urine, and in three cases in the faeces. For comparison the same procedure was followed in three normal subjects.

Plasma.—In the normal subjects the smallest increase in the plasma vitamin A after dosing was 206 i.u. In infective hepatitis the smallest increase was only 32 i.u. and the mean increase for all tests in the whole group was 148 i.u. It is worthy of remark that no consistent difference in the extent of the response after dosing was found between the early and late stages of the disease.

Faeces.—In normal subjects vitamin A is found in the faeces only after very heavy dosing. Even the massive doses given in the present experiments were not sufficient to cause faecal excretion of the vitamin in our control subjects. In contrast large quantities of vitamin A, amounting in one instance to more than half the dose given, were excreted in the early stages of the disease by all three patients with hepatitis whose faeces were examined. Even in the subject who excreted only 1,000 i.u. the presence of the vitamin was confirmed by the presence of an absorption band at 620 m μ . in the SbCl₃ test. No vitamin A was found in the faeces of the same subjects during later stages of their illness.

Urine.—After dosing, traces of vitamin A were present in the urine of two patients with hepatitis. Although the urinary loss was quantitatively insignificant it must be emphasized that in health the urine contains no vitamin A even after heavy dosing. Vitamin A is, however, excreted in the urine in certain diseases, and its appearance always has pathological significance (Lawrie, Moore, and Rajagopal, 1941).

An interesting abnormality was shown by subject H. For about a week after the first dose of vitamin A had been given the colour produced by the plasma extract with the SbCl₃ reagent was a faint orange in place of the usual blue. We were unable to ascertain whether vitamin A, or its oxidation products, played any part in producing this abnormal colour.

Frequent Dosing with Vitamin A.—Two subjects were given massive doses of vitamin A frequently over a period of about four weeks. The total amount received was nearly 3,000,000 i.u., which may be compared with the median total liver reserve of 500,000 i.u. (Moore, T., unpublished data). The course of dosing was therefore expected to alter the liver reserves substantially. The effect of the first dose on the plasma vitamin A was examined five hours after its administration. After the

penultimate dose had been given a period of 36 hours was allowed to elapse so that the plasma vitamin A might reach a resting level before giving the final dose. A specimen of blood was then taken again five hours after the last dose. It will be seen from Table V that in both cases the increase in the

TABLE V.—*Effect of the Frequent Massive Doses (96,000 I.U.) of Vitamin A on the Plasma Vitamin A*

Case	Days after Admin.	Plasma Vitamin A			
		No. of Doses Received	Before Dosing (I.U.)	After Dosing	Difference
M	1	1	80	108	28
	32	31	227	473	246
N	1	1	70	68	-2
	32	31	150	406	256

plasma vitamin A was much greater after the last dose than after the first. These results, taken in conjunction with those for subjects F and H (Table IV) agree with the conclusion of Breese and McCoord (1940) that in those subjects in which defective absorption is found in the early stages of the disease improved absorption may be expected during recovery.

The Liver Reserves of Vitamin A in Infective Hepatitis

There were no fatal cases in the present series, but necropsy material was obtained from other sources. Specimens of liver from two cases of acute yellow atrophy following infective hepatitis, or homologous serum jaundice, were kindly supplied by Capt. J. A. R. Miles, R.A.M.C. Values of 225 and 500 i.u. per g. were found. Another specimen was kindly supplied by Dr. J. F. Gaskell, of Addenbrooke's Hospital, and in this instance the evidence showed that the low blood vitamin A in infective hepatitis is not necessarily associated with a depleted liver reserve. Although shortly before death the plasma vitamin A was only 19 i.u. per 100 ml., with carotene 68 i.u., the liver contained 900 i.u. per g. None of the above patients had been specially dosed with vitamin A.

In a recent survey of the vitamin A reserves of the population of this country in cases of accidental death (Moore, T., unpublished data) a wide range of values was found, with a median of about 300 i.u. per g. of liver. So far as can be judged from the small number of subjects, therefore, there is in acute liver disease no severe depletion of the pre-existing vitamin reserves such as commonly occurs, for example, in cirrhosis of the liver or in chronic nephritis, for which diseases median liver reserves of 6 and 25 i.u. per g. respectively have been reported (Moore, 1937).

Discussion

Both acute febrile illnesses and diseases of the liver affect the metabolism of vitamin A. Since infective hepatitis presents a combination of these lesions the disturbances in vitamin A metabolism will obviously be complex. In the present investigation we have observed in the acute stage of the disease: (1) reduced plasma vitamin A and carotene; (2) failure of the plasma vitamin A to respond to massive dosing with vitamin A; and (3) the excretion of large amounts of vitamin A in the faeces after massive dosing, with traces also in the urine.

In experiments at Sheffield with human volunteers the plasma vitamin A decreased only slightly after restriction for some months to a diet virtually devoid of carotene and vitamin A (Med. Res. Cncl. Vitamin A Subcommittee, 1945). Levels of the order of 30 i.u. per 100 ml. of plasma, at which defective dark adaptation was observed, occurred only in subjects deprived of the vitamin for over a year. Since in our experiments equally low values were often found in patients with infective hepatitis in the first few days of their illness, the brief initial period of anorexia clearly cannot in itself account for the decrease in plasma

vitamin A. Neither can a full explanation be found in the defective intestinal absorption of vitamin A and other fatty substances which is characteristic of hepatitis. In fatal cases the liver was found to contain adequate reserves of vitamin A, which in healthy subjects would have ensured a normal plasma level even if dietary supplies of the vitamin had been completely withdrawn. Failure in the mechanism for the release of vitamin A from the liver to the plasma must therefore be strongly suspected. One possibility, suggested by the recent experiments of Glover, Goodwin, and Morton (1946), is that there may be interference in the hydrolysis of vitamin A esters stored in the liver to liberate free vitamin A alcohol for mobilization into the blood stream. Any deficiency of the liver in this direction, however, may be intensified during fever by an increased expenditure of vitamin A in the plasma and tissues.

The position in regard to the carotenoid level is different. In the Sheffield experiment the plasma carotenoids, in contrast to vitamin A, fell to a low level soon after restriction to the deficient diet. The effect of anorexia in hepatitis cannot therefore be discounted. The stores of carotenoids from which the plasma may be replenished are, moreover, much smaller than those of vitamin A. The decrease in plasma carotenoids may be, due to defective absorption and a decreased dietary intake, combined possibly with an increased rate of degradation during fever.

The complexity of the lesions in infective hepatitis, however, becomes most apparent when we consider the reaction to massive doses of vitamin A. We have seen that during the early stages of the disease the plasma vitamin A often fails to respond to dosing, and, moreover, that vitamin A is excreted in the faeces and sometimes in the urine. It might be expected that in individual cases these abnormalities would be interrelated and would all occur with about equal severity. Thus the loss of vitamin A in the faeces would at first sight afford a ready explanation of its failure to increase in the blood plasma. In our experiments, however, we have found no consistent correlation between the various disabilities involved in each case. For example when patient G was given his first massive dose of vitamin A about 40% was excreted in the faeces; the plasma vitamin A, nevertheless, increased by 412 i.u. per 100 ml. In contrast, when the same patient was later given a second dose there was no faecal excretion, but the plasma level was increased by only 89 i.u. In patient E abnormality in vitamin A metabolism was clearly indicated by urinary excretion, but after the first massive dose the plasma level increased by 279 i.u. We have already referred to the superiority of the response to massive dosing shown by patient D as compared with patient C, whose plasma had a higher resting level of vitamin A.

The most plausible explanation of these apparent anomalies is that the various mechanisms concerned with the metabolism of vitamin A are not always equally affected. Thus the absorption of vitamin A from the intestines, its deposition in the liver, its release from the liver to the plasma, and possibly its stability when present in the plasma may suffer to varying extents in different individuals. On the basis of this assumption patient C may have been unable to absorb vitamin A from his intestines but capable of mobilizing reserves from his liver into his blood stream. Patient G, on the evidence of faecal excretion in the early stage of his illness, could obviously absorb only part of his dose of vitamin A; but possibly this abnormality was complicated by an even more severe disability in the liver's power to remove excess of vitamin from the blood stream. In that event the plasma vitamin A, denied its usual outlet, would "pile up" to a high level, and would thus convey a false impression of the efficiency of intestinal absorption.

The possibility of differences in the degree of impairment in the functions of the liver and the intestines, and of complications arising from pyrexia *per se*, must raise doubts as to the value of "vitamin A absorption tests" as a measure of the injury sustained by the liver. Repeated failure of the plasma vitamin A to respond to dosing even after pyrexia has subsided must strongly suggest damage to the liver; a good response to dosing, however, may not necessarily imply that liver function is normal. Moreover, while we have found that the increase in plasma vitamin A during convalescence moves parallel with the improved values in prothrombin and liver function tests, and conversely to the plasma bilirubin, the interrelationships between these four criteria are not consistent enough to allow inferences as to the general condition of the patient to be drawn from estimation of the plasma vitamin A alone.

Summary

In the early stages of infective hepatitis the vitamin A content of the plasma was often very low. In estimations by the antimony trichloride method values of 30 i.u. per 100 ml. or less were sometimes observed, as compared with an average of 118 i.u. previously found for normal subjects. The low plasma vitamin A seems an adequate cause of the impairment of dark-adaptation in this disease.

The age of the patient and the degree of pyrexia appeared to influence the extent of the fall in plasma vitamin A, but no correlation was found with the duration of anorexia.

During convalescence the plasma vitamin A increased until the mean for all cases equalled or exceeded the mean for normal subjects. There was no evidence that the vitamin A reserves of the liver were seriously reduced.

Variations in the carotenoid content of the plasma were similar to those found for vitamin A, but the return to normal was less rapid.

The changes in plasma vitamin A and carotenoids went roughly parallel with the prothrombin value and with hippuric acid tolerance, and moved in an opposite direction to the plasma bilirubin. There was perhaps a tendency for the recovery in vitamin A to begin slightly before bilirubin had started to decline.

In "absorption tests" with massive doses of vitamin A it was found that in the early stages of infective hepatitis substantial amounts of vitamin A were excreted in the faeces, and sometimes there were traces in the urine. Excretion was not found in normal subjects given the same doses or in the same patients when dosed during convalescence.

In some cases the plasma vitamin A failed to show the usual rapid increase after massive dosing. No consistent correlation was found, however, between this defect and either faecal excretion of the vitamin or a low resting value in the plasma.

It appears that in infective hepatitis the mechanisms responsible for the absorption of vitamin A from the intestines, for its deposition in the liver, and for its release into the blood stream are not always affected to the same degree. The interpretation of the results of "absorption tests" as a measure of the degree of injury to the liver therefore seems uncertain.

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REFERENCES

- Aldersberg, D., and Sobotka, H. (1944). *Bull. N.Y. Acad. Med.*, **20**, 411.
 Altschule, M. D. (1935). *Arch. Path.*, **20**, 845.
 Bloch, C. E. (1924). *Amer. J. Dis. Child.*, **28**, 659.
 Breese, B. B., and McCoord, A. B. (1940). *J. Pediat.*, **16**, 139.
 — Watkins, E., and McCoord, A. B. (1942). *J. Amer. med. Ass.*, **119**, 3.
 Breusch, F., and Scalabrino, R. (1934). *Z. ges. exp. Med.*, **94**, 569.
 Clausen, S. W., and McCoord, A. B. (1938). *J. Pediat.*, **13**, 635.
 Ezickson, W. J. (1939). *J. Lab. clin. Med.*, **24**, 836.
 Glover, J., Goodwin, T. W., and Morton, R. A. (1946). *Biochem. J.* (in press).
 Haig, C., Hecht, S., and Patek, A. J. (1938). *Science*, **87**, 534.
 Harris, H. J. (1943). *Proc. roy. Soc. Med.*, **11**, 5.
 Jeghers, H. (1937). *Ann. intern. Med.*, **10**, 1304.

- Josephs, H. W. (1943). *Amer. J. Dis. Child.*, **65**, 712.
 Kimble, M. S. (1939). *J. Lab. clin. Med.*, **24**, 1055.
 Lawrie, N. R., Moore, T., and Rajagopal, K. R. (1941). *Biochem. J.*, **35**, 825.
 Leitner, Z. A., and Moore, T. (1946). *Lancet*, **2**, 262.
 Mallory, H. T., and Evelyn, K. A. (1937). *J. biol. Chem.*, **119**, 481.
 Med. Res. Cncl. Vitamin A Subcommittee (1945). *Nature*, **156**, 11.
 Moore, T. (1937). *Biochem. J.*, **31**, 155.
 — and Wang, Y. L. (1945). *Ibid.*, **39**, 222.
 Patek, A. J., and Haig, C. (1939). *J. clin. Invest.*, **18**, 609.
 Pollock, M. R. (1945). *British Medical Journal*, **2**, 878.
 Popper, H., Steigmann, F., and Zevin, S. (1943). *J. clin. Invest.*, **22**, 775.
 Quick, A. J. (1939). *Amer. J. digest. Dis.*, **6**, 716.
 Ralli, E. P., Bauman, E., and Roberts, L. B. (1941). *J. clin. Invest.*, **20**, 709.
 Spector, S., McKhann, C. F., and Meserve, E. R. (1943). *Amer. J. Dis. Child.*, **66**, 376.
 Steigmann, F., and Popper, H. (1944). *Amer. J. med. Sci.*, **207**, 468.
 Thompson, J. T. (1894). *British Medical Journal*, **2**, 597.
 Wilson, C., Pollock, M. R., and Harris, A. D. (1945). *Ibid.*, **1**, 399.
 Wohl, M. G., and Feldman, J. B. (1940). *J. Lab. clin. med.*, **25**, 485.
 — (1941). *Amer. J. digest. Dis.*, **8**, 464.
 Wolff, L. K. (1932). *Lancet*, **2**, 617.
 Yudkin, S. (1941). *Biochem. J.*, **35**, 551.

ADDENDUM ON DARK-ADAPTATION

In these experiments, which preceded and prompted the biochemical investigations described above, the course of dark-adaptation was determined by means of the Craik adaptometer in a group of 37 Service men with infective hepatitis. They were all tested within one week of admission, and serial tests were performed on a few patients throughout their stay in hospital, at intervals varying from a day to a week. An attempt was made to test all patients again one month after discharge, on their return visit from the convalescent depot; for administrative reasons this was possible in only 14 cases.

Apparatus and Method.—A pea-lamp run at 2,360 K. illuminates a flashed opal screen subtending 7° at the subject's eye—at a distance of 2 ft. 6 in. (0.76 m.)—through an opal window and a neutral wedge, which the subject moves by a long shaft. The position of the wedge is recorded automatically by a pencil on a rotating drum, plotting the logarithm of the threshold brightness against time. A celluloid disk with a "V" can also be placed in front of the screen to test form perception, and a red fixation point of variable brightness is provided. Vision is binocular with natural pupils. The brightness range of the instrument is from 0.0000001 to 0.01 equivalent foot-candles. Tests are carried out in a dark room. The subject is first bright-adapted for five minutes by looking at a surface of 750 equivalent foot-candles brightness. The light is then turned off, and he is instructed to turn to the adaptometer and to keep the test light just visible by moving the neutral wedge, looking in the general direction of the fixation spot but not staring rigidly at it, as this causes local fatigue of the retina. In this way a continuous record is obtained in the form in which such results are usually plotted—namely logarithm of threshold against linear time. Thresholds of light perception are expressed as the negative logarithms of the brightness in equivalent foot-candles. A rise in the logarithm implies that a dimmer light can be perceived and that the efficiency of dark-adaptation has therefore increased. The final rod threshold is reached when the logarithm has reached a steady value consistent with maximum dark-adaptation.

Defective Dark-Adaptation in Infective Hepatitis.—The results of testing an unselected group of 37 patients with hepatitis and 20 normal men of comparable age are shown in Table VI. The final rod threshold was impaired in the hepatitis

TABLE VI.—Final Rod Threshold in 37 Subjects With Infective Hepatitis and 20 Controls: Mean Values

Days after Admission	No. of Observations	Final Rod Threshold
0-4	30	5.315
5-9	10	5.350
10-19	10	5.315
20-39	7	5.686
40-59	6	5.642
Over 60	5	5.710
Under 20	50	5.332
Over 20	18	5.678
Controls	20	5.758

patients, and no improvement in mean value was shown until 20 days after admission. The t test of significance was applied to the difference between the means in the following paired groups:

A. Under 20 days from admission	...	5.332 log. units
Over 20 days	" "	5.678 " "
B. Under 20 days	" "	5.332 " "
Controls	...	5.758 " "
C. Over 20 days from admission	...	5.678 " "
Controls	...	5.758 " "

The comparisons A and B showed significant differences ($t = 2.695$ with $n = 66$ and $t = 5.926$ with $n = 68$ respectively, both giving $P = 0.05$). In the comparison C the difference is insignificant ($t = 0.920$ with $n = 36$).

The Improvement in Dark-Adaptation during Convalescence.—The same conclusion may be reached from Table VII, where

TABLE VII.—Final Rod Threshold in 14 Subjects With Infective Hepatitis: Retested in Convalescence

Case	First Test	Days after Admission	Second Test	Days after Admission
1	5.30	2	5.45	27
2	4.95	4	5.60	53
3	5.10	5	5.45	47
4	5.50	8	5.80	82
5	5.15	1	5.60	71
6	5.20	9	5.65	51
7	5.30	3	5.95	66
8	5.00	10	5.30	101
9	5.80	2	5.60	58
10	5.50	3	6.00	52
11	5.20	1	5.90	69
12	5.40	4	6.00	27
13	5.40	2	5.75	35
14	5.35	7	5.80	35
Mean:	5.297		5.704	

the results for the 14 men who were retested during convalescence are shown independently. The mean values for final threshold were 5.297 during the active stage of the disease and 5.704 during convalescence; and again the difference is significant ($t = 4.892$. With 26° of freedom t need equal only 2.056 to give $P = 0.05$). The individual readings all improved except for one man whose threshold was initially within normal limits. An examination of the superimposed tracings of the curves provided by these 14 patients showed slight shortening of the rod-cone transition time with convalescence. Although the earlier curves lay within the normal range the shortening in transition time during convalescence occurred in 11 out of 14, which suggests that the earlier times were in fact abnormally long for these individuals. The range of the observed alterations was from 1 to 4½ minutes, with a mean of 2½ minutes. No alteration in cone threshold was found.

Dosing with Vitamin A.—Large doses of vitamin A (96,000 units) daily by mouth were given to four subjects, not included in the above tables, to see whether the final rod threshold could be restored to normal. The results shown in Table VIII are

TABLE VIII.—Effects of Repeated Doses of Vitamin A (96,000 Units Daily) On Final Rod Threshold In Patients With Infective Hepatitis

Test Subjects				Controls		
Case	Days after Admission	Total Vitamin A Received	Final Rod Threshold	Case	Days after Admission	Final Rod Threshold
A	1	0	5.40	1	4	5.40
	2	96,000	5.70		13	5.60
	5	384,000	5.80		20	5.50
	8	672,000	5.90		27	6.00
B	1	0	5.20	2	2	5.40
	3	192,000	5.50		9	5.50
	6	480,000	5.50		16	5.65
	9	768,000	5.60		23	5.85
C	1	0	5.10	3	36	5.75
	3	192,000	5.55		2	5.30
	5	384,000	5.75		11	5.50
	8	672,000	5.90		15	5.35
D	1	0	5.40		27	5.45
	2	96,000	5.60			
	5	384,000	5.95			
	8	672,000	6.00			

suggestive but not conclusive. Three patients returned to normal after eight days and the fourth improved considerably.

This may be contrasted with the slow recovery in the other patients in the series and in the three controls shown in the table. There are two reasons, however, for caution in interpreting these results: (1) Final rod threshold is not a very satisfactory measurement to apply to small groups because of the range of individual variation, though it is believed that the differences shown here are too great to appear by chance. Again, it cannot be excluded that as a result of the greater frequency of testing in these men facilitation of response may have contributed to the improvement. (2) The plasma vitamin A was estimated chemically in three subjects, and in two of them the rise produced by 96,000 units of vitamin A was so slight that it is difficult to see how it could have improved their dark-adaptation.

Discussion

Vitamin A abnormality in infective hepatitis is associated with a rise of the final rod threshold and only slight alteration in the rod-cone transition time. This is the type of change that has been found in most experiments on simple acute dietary avitaminosis A. In contrast Haig, Hecht, and Patek (1938) found that in chronic liver disease the lengthening of the rod-cone transition time was as marked as the impairment of the final rod threshold. Similarly in the recent Sheffield experiments (Med. Res. Cncl., 1945), in which volunteers were kept on a vitamin-A-free diet for long periods, the rod-cone transition time was affected as well as the final rod threshold. These differences may be explained by the suggestion made by Hecht and Mandelbaum (1939) that "it may well be that this difference in behaviour of the rod-cone break is purely a matter of short-period avitaminosis versus prolonged deprivation."

It has previously been pointed out that dark-adaptation is less satisfactory as a measure of vitamin A abnormality than chemical estimation. In this trial, for instance, there was no diagnostic impairment in individual patients, and statistical analysis was necessary to show the effect, whereas using chemical estimation of the plasma vitamin A as an index a definitely abnormal result was obtained in most patients. The insensitivity in measurement of dark-adaptation arises from two factors: the range of normal variation from individual to individual and the wide margin of error in the same individual found by repeated testing on the same day.

A second disadvantage is that dark-adaptation may be affected by other factors than vitamin A lack. Malingering can generally be detected with the Craik adaptometer, since a self-inconsistent record is produced, but some psychoneurosis can cause poor dark-adaptation (Wittkower, Rodger, Scott, and Semeonoff, 1941). Pyrexia is a cause which can be disregarded in the present series, since most of the cases had no significant fever. It has been suggested that raised blood bile salts might play a part, as bile salt is a solvent for visual purple *in vitro*. No evidence was obtained for this when one of us (K.J.W.C.), after dark-adaptation was complete, was given an intravenous injection of bile salts. The injection was given by a dim light while the subject wore dark goggles and the record continued immediately. No alteration in threshold resulted either from bile salts or from the control injection of sterile water.

Summary

The course of dark-adaptation was investigated by means of the Craik adaptometer in patients with infective hepatitis.

The mean final rod threshold for 37 men tested within one week of admission to hospital was significantly impaired in comparison with that of 20 normal adults of comparable age.

In 14 men retested after an interval of one month or more the mean final rod threshold was significantly improved.

No alteration was apparent in cone threshold, and the rod-cone transition time was only slightly prolonged.

Dark-adaptation under the conditions of this experiment was a less sensitive measure of vitamin abnormality than chemical estimation of plasma vitamin A.

REFERENCES

- Haig, C., Hecht, S., and Patek, A. J. (1938). *Science*, **87**, 534.
Hecht, S., and Mandelbaum, J. (1939). *J. Amer. med. Ass.*, **112**, 1910.
Med. Res. Cncl. Vitamin A Subcommittee (1945). *Nature*, **156**, 11.
Wittkower, E., Rodger, T. E., Scott, G. I., and Semeonoff, B. (1941). *British Medical Journal*, **2**, 571.

TUBERCULOUS PERICARDITIS

BY

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Pericardial effusion caused by tuberculosis has always been considered a rare condition, carrying with it a high mortality. Of recent years, however, more cases have been diagnosed as being due to tuberculosis, and doubt has been cast upon other cases which had previously been labelled rheumatic. Some of the many cases published have been proved to be tuberculous either during life or at necropsy, but in many a diagnosis has been presumed only by the presence of other tuberculous manifestations or by the absence of signs which have been taken to be rheumatic in origin. Thus in Harvey and Whitehill's (1937) collection of 95 cases taken from 45 years of records of the Johns Hopkins Hospital there are 37 proved cases, 34 unproved, and 24 proved in which the pericardial lesion was of no clinical importance. The mortality varied greatly in this series; in proved cases it was 83%, whereas in the unproved cases it was only 6%. There was little difference in the characteristic signs and symptoms in the unproved group from those in the proved group, and it was on these signs that the clinical diagnosis was achieved. These groups were further subdivided into those with large pericardial effusion and those without effusion, and while in the proved group of 37 cases 20 had effusion, in the unproved group 31 of the 34 cases had large effusions. In Ellman's (1945) excellent paper on this subject he quotes six cases, and, again, although the clinical picture and progress are strongly suggestive of tuberculous pericarditis, in three cases the diagnosis was not proved.

In all these series of cases in which the diagnosis has been tuberculous pericarditis with effusion the following features have been taken to be characteristic of the clinical picture: (1) no enlargement of the heart; (2) no gross murmurs or murmurs indicative of valvular involvement; (3) no rheumatic manifestation; and (4) no past or family history of rheumatism. In addition, there was no marked anaemia, an insidious onset, no leucocytosis, and, although the effusion might be large, the symptoms, especially precordial pain, were not marked.

Massie and Levine (1939) investigated 135 cases of rheumatic pericardial disease diagnosed by the presence of pericardial rub or pericardial effusion. They found that 110 cases had definite arthritis or arthralgia, one case had chorea, and 17 of the others had a past history of one or more attacks of rheumatic fever, sore throat, or a family history of rheumatism. On the other hand, of the 82 cases followed up for a period of seven years one-third had no evidence of any organic heart disease. Although Sheldon (1946) has recently published six cases demonstrating tuberculous arthralgia, acute rheumatism with serositis, and chronic rheumatism, this should not confuse the diagnosis with that of rheumatism if the criteria of the small heart, etc., are taken into consideration. While dealing with the differential diagnosis of tuberculous and rheumatic pericardial effusion it is also of interest to note that in Harvey and Whitehill's (1937) series several cases developed auricular fibrillation when the effusion was maximal, and that there was no evidence of myocardial involvement clinically or in those cases which came to post-mortem examination. In addition, these cases appeared particularly susceptible to digitalis, heart-block occurring with relatively small doses.