VITAMIN A.

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Historical.

The existence of "fat soluble factor A" as opposed to "water soluble factor B" was first demonstrated by McCollum and Davies (1915). The story of how these two "factors" were found to be groups each containing several vitamins is common knowledge. The alphabetical priority of vitamin A as recognised to-day may be of some interest from the point of view of nomenclature, and is probably due to the ready appearance of the signs of deficiency in rats when restricted to the type of basal diet used in early nutritional research. Deficiency of the other prominent members of the fat-soluble group is more difficult to demonstrate in the rat. In the case of vitamin D a derangement of the calcium to phosphorus ratio of the diet is necessary, as well as a deficiency of the vitamin, for the development of rickets. With vitamin E the effects of deficiency are seen only after mating, or after very prolonged deprivation in the virgin animal.

For many years the investigation of vitamin A was complicated by misunderstandings due to the existence of the vitamin in two distinct forms. Steenbock (1919) in America, and others, found that a remarkable parallelism existed between the vitamin A activity of various foodstuffs, as measured by their power to promote growth in young rats, and yellow coloration. Thus, butter, egg yolk, carrots, yellow maize, and green vegetables containing carotenoid pigments were all found to be good sources of vitamin A. As a notable exception, however, cod-liver oil, an excellent source of vitamin A, was only faintly coloured. Tests with carotene, the pigment of carrots, were conflicting and unsatisfactory, but it was generally supposed that the pure substance was devoid of biological activity. This erroneous conclusion, which greatly delayed progress, can now be ascribed to inadequate precaution against the oxidation of the unstable pigment during the period of biological testing (Hume and Smedley-Maclean, 1930).

In the meantime patient research was undertaken on the concentration of the "colourless" form of vitamin A present in cod-liver oil. The vitamin was found to be stable to treatment with hot alcoholic potash, and to be concentrated in the non-crystalline fraction of the non-saponifiable residue. The Japanese worker Takahashi and his colleagues (1925) claimed to have isolated the vitamin in pure form, and described many of its properties for the first time. Thus, in a spectroscopic examination they correctly associated the vitamin with an intense absorption band in the ultra-violet region, which was subsequently rediscovered by Morton and Heilbron (1928), and used by them as the basis of an accurate method for the estimation of the vitamin. They also gave the first account of some of the ill effects of massive overdosing with the vitamin. In England, however, Drummond, Channon and Coward (1925) prepared concentrates of equal potency which they demonstrated were far from pure.

In both these investigations extensive use was made of colour reactions of the vitamin. It had long been known that cod-liver oil gives an intense purple colour on treatment with sulphuric acid, and through the fortunate inspiration of Rosenheim and Drummond (1920) this reaction was correctly attributed to vitamin A. Carr and Price (1926) devised an improved form of this test, in which sulphuric acid is replaced by a solution of antimony trichloride in chloroform, with a resulting change in the colour to bright blue. This reaction has been of immense service in facilitating research on the vitamin.

It had been well known that the carotenoid pigments as a class resembled cod-liver oil in producing blue or purple colorations on treatment with sulphuric acid. In 1928

Euler, Euler and Hellstrom applied the antimony trichloride reagent to a number of carotenoid pigments. Attracted by the similarity of the blue colours produced to that given by cod-liver oil they undertook biological tests on pure carotene, which they found to be active in doses as small as those found by other workers for the best concentrates of vitamin A prepared from cod-liver oil. This important discovery did not meet with immediate general acceptance. The activity of carotene was confirmed by some workers, disputed by others. It was plain that carotene and the vitamin A of cod-liver oil must be different substances. Thus carotene is a highly pigmented crystalline solid with a high melting point, while concentrates containing vitamin A were viscous fluids which were only slightly yellow. A suggestion that carotene might owe its activity to contamination with the colourless vitamin A was ruled out by a careful quantitative and spectroscopic comparison of the blue colours produced by the two substances.

The solution of the anomaly was found in experiments by Moore (1930). Rats were restricted to a diet deficient in vitamin A until signs of deficiency were evident. Some were then killed. The fat extracted from their livers gave no colour with the antimony trichloride reagent. Others were given large doses of carotene for several weeks before they were killed. The liver oils, although only faintly yellow, now gave intense blue colorations with the antimony trichloride reagent. Proof was thus obtained of the conversion of carotene into vitamin A in the animal body. The changes involved were summarised as follows :—

<i>Carotene</i>	<i>Vitamin A</i>
Synthesised in plant	Stored in animal
Intensely yellow	Almost colourless
328 m μ absorption band absent	328 m μ absorption band developed
Greenish blue SbCl ₃ reaction at 500 m μ .	Vivid blue SbCl ₃ reaction at 610-630 m μ .

Advances in other directions followed. It was shown by Green and Mellanby (1930) that carotene possesses the same power as vitamin A in curing infections of the mucous membranes. Halibut liver oil, and several other fish-liver oils, were found to be many times richer than cod-liver oil in vitamin A (Schmidt-Nielsen and Schmidt-Nielsen, 1929), and progress towards the isolation of the vitamin advanced rapidly. The Swiss organic chemist Karrer (1931), who had already elucidated the structural formula of carotene, demonstrated by chemical means that the richest vitamin A concentrates then obtainable consisted mainly of the actual vitamin, with relatively small amounts of impurities. The molecule of vitamin A was found to be about half that of carotene in size. One of the ionone rings of carotene was retained intact, and the side chain of condensed isoprene units was terminated by an hydroxyl group. The following structural formulæ were given for β -carotene, the most important of the several isomeric forms of the pigment, and for vitamin A. The exact length of the side chain in vitamin A is still open to question.

B Carotene.

Vitamin A.

β -Carotene (p. 53) showing its relation to vitamin A, above.

In subsequent research vitamin A has been obtained in a virtually pure state by two methods. Holmes and Corbet (1937) in America succeeded in crystallising the vitamin at low temperatures from concentrates prepared from oils of very high original potency. In Japan, Hamano (1935, 1937) prepared various esters of the vitamin having relatively high melting points. Data observed upon preparations obtained by these alternative methods were in good agreement, and differed only slightly from values previously reported for non-crystalline concentrates prepared by Karrer and other workers. For pure vitamin A the spectroscopic absorption band in the ultra violet region has an intensity $\left(E \frac{\%}{\text{cm}} \right)$ of about 1800, which may be compared with a corresponding value of 40 for halibut-liver oil of average quality, or 1 for cod-liver oil. These values are proportional to the respective biological activities.

Properties and Distribution of Carotene.

Carotene is quite insoluble in water, and is soluble only to a limited extent in fats. The pure pigment is rapidly oxidised and bleached on exposure to air at room temperature. Thus low stability is to be expected in rancid fats, and in some simple esters such as ethyl oleate. In oily solution it is usually more stable, but much depends on the nature of oil and its state of preservation. In most vegetable tissues the pigment is remarkably stable, and withstands cooking, canning, or the drying of the vegetable under careful conditions.

The richest European source of carotene is the carrot, from which, of course, the pigment takes its name. Green vegetables also contain large amounts, together with the pigment xanthophyll, which differs from carotene in containing two hydroxyl groups and in being inactive as a source of vitamin A. Some yellow fruits, such as the apricot, owe their colour to carotene. The tomato is an excellent source, but the yellow colour of carotene in this fruit is masked by the presence of lycopene, an inactive red carotenoid. Red palm oil, an important commercial product of tropical Africa and Asia, contains carotene in high concentration. In addition to β -carotene several other carotenoids possess biological activity. All contain one-half of the β -carotene molecule intact. As examples α -carotene, present in red palm oil and mountain ash berries, kryptoxanthin, present in maize, and aphanin, present in algae may be mentioned.

Unchanged carotenoids are transferred in small amounts from the food to the fats of some animals, but remarkable differences are found between species. Thus, butter fat and human blood serum are coloured by carotene. The pigment of egg yolk is almost all xanthophyll. The fats of some other animals, such as the pig, sheep and rat, remain colourless even when large amounts of carotenoids are included in the diet.

Properties and Distribution of Vitamin A.

Like carotene, vitamin A is insoluble in water, but it differs in being miscible in all proportions with fats. In the absence of air the vitamin is stable to heat and may be readily distilled *in vacuo*. Decomposition occurs rapidly however under the combined

influence of heat and air (Hopkins, 1920) or on exposure to ultra-violet irradiation (Peacock and Wright, 1926). The vitamin A content of foodstuffs is not seriously reduced by ordinary processes of cooking and canning.

In the animal vitamin A may be absorbed from the alimentary tract in large amounts much more rapidly than carotene. The main site of storage is the liver, which has a remarkable pre-eminence over the other organs in its power to absorb and retain the vitamin. Thus in the rat the amount accumulated may be enough theoretically to suffice normal requirements for a century, although the life span is only three years. The blood serum carries small amounts of vitamin A, together with carotene, but none is found in the corpuscles. The vitamin is also found in the kidneys, lungs, storage fats and suprarenals; elsewhere it is unusual for more than traces to be present.

Research by Dann (1932) and others has shown that the amount of vitamin A which passes through the placenta to the foetal liver is usually very small, even when large amounts of vitamin are included in the maternal diet. On the other hand, a considerable concentration of vitamin A is found in colostrum, which is much richer than the subsequent milk (Dann, 1933). The importance of the colostrum to newly-born calves in preventing "white scour" has been indicated by Stewart and McCollum (1938).

Sources of preformed vitamin A may be grouped under three main headings. In the first, and most important group, we have the dairy products, milk, butter, cheese and eggs. Although of essentially different origin vitaminised margarine may conveniently be included in this group. Secondly, we have liver, sold as butcher's meat. Large amounts of vitamin A may be obtained from this source by habitual consumers, but the amounts eaten must vary widely according to individual taste. Thirdly, we have the group of fish liver oils, chiefly cod-liver oil and halibut-liver oil, which may perhaps be considered as therapeutic agents rather than as ordinary foodstuffs. In its natural sources vitamin A is almost invariably present in esterified form in combination with fatty acids.

Lesions in Vitamin A Deficiency.

When experimental animals are restricted to diets deficient in vitamin A a wide variety of lesions may result. In epithelial tissues there is widespread keratinisation or xerosis, with the replacement of columnar by stratified formations (Wolbach and Howe, 1925). In the conjunctiva this often leads to the well-known xerophthalmia. The power of the diseased membranes to resist invasion by saprophytic bacteria is reduced, and localised pyogenic infections are common. Defective tooth formation often results from injury to the enamel-forming cells. Damage to the epithelium of the kidneys and bladder may sometimes lead to the formation of stones (Osborne, Mendel and Ferry, 1917).

Epithelial lesions, however, although so important as to justify the description of vitamin A as the "anti-keratinising factor," or "anti-xeratic factor" (Harris, Innes and Griffith, 1932), are not the only ill-effects of deficiency. The well-known night blindness, discussed in more detail below, results from a deficiency of vitamin A in the retina. Nerve degeneration (Hart and McCollum, 1914, Mellanby, 1926) and abnormalities in bone (Mellanby, 1938), blood (Frank, 1934), and the reticulo-endothelial system (De Ruyter, 1934) have also been reported.

In man vitamin A deficiency is known to lead to night-blindness and later to xerophthalmia, but there is as yet little conclusive evidence to include the other diseases found in experimental deficiency in animals among the effects of clinical deficiency in man. Some support may be found for the view that vitamin A deficiency causes urolithiasis in man (McCarrison and Ranganathan, 1931) and alterations in the blood picture also probably occur (Wagner, 1940). There have been many indications that in man vitamin A deficiency may be of importance in the etiology of skin diseases

(Loewenthal, 1933). Thus the tropical disease of "phrynoderma" or "toad skin" has been attributed to deficiency of vitamin A, although it is possible that deficiency of other vitamins is involved (Nicholls, 1933). In this country Mackay (1934) has found that vitamin A, as cod-liver oil, reduces the incidence of minor skin ailments, such as napkin-rash, in infants.

Vitamin A and Dark Adaptation.

Defective dark adaptation is of particular interest as the earliest sign of vitamin A deficiency. It might therefore be expected in subjects receiving diets low in vitamin A, but not so devoid in the vitamin as to cause xerophthalmia and other more serious lesions. The cure of night blindness by liver was known by Hippocrates, and modern research has amply confirmed the influence of vitamin A deficiency in the etiology of this abnormality. Jeans and Zentmire (1933) suggested that efficiency of dark adaptation, as measured by the Birch-Hirschfeld photometer, might be used for the clinical detection of vitamin A subnormality. Theoretical support for this possibility was afforded by the experiments of Wald (1934), who demonstrated that visual purple consists essentially of vitamin A combined with protein.

Numerous investigations have been undertaken both to test the reliability of dark adaptation methods and to apply them to estimations of the incidence of vitamin A subnormality in various groups of subjects. The evidence of different workers has been somewhat divergent, but this is understandable in view of the high degree of skill which is necessary in planning and performing the experiments. In this country Harris and Maitra (1937) found that up to 36 per cent. of poorly fed children from elementary schools had definitely subnormal dark adaptation, as compared with only one instance of subnormality among forty well-nourished boys from a public school, where fish-liver oil was given as a prophylactic. The reliability of the method as a measure of vitamin A deficiency was checked by observing the improvement in the subjects showing subnormal dark adaptations when vitamin A was given.

There can be no doubt that vitamin A plays an important role in dark adaptation, and in subjects receiving inadequate amounts of the vitamin measurable deterioration in dark adaptation must inevitably take place. It seems dangerous, however, to accept inferior dark adaptation as a certain indication of deficiency of the vitamin without adequate control tests. The formation of visual purple must be a complicated process, depending not only on the amount of vitamin ingested, but on the ability of the retina to absorb the vitamin, and the efficiency of the blood stream in carrying vitamin A from the intestines and liver to the retina. McFarland and Forbes (1940) have recently claimed that the efficiency of dark adaptation is reduced by anoxia or hypoglycemia. In the other direction it has been suggested that the ingestion of alcohol may improve dark adaptation by facilitating the mobilisation of vitamin A from the liver reserves (Pett, 1940). Habitual alcoholism sufficiently severe to cause cirrhosis of the liver has, as will be seen below, an adverse effect on dark adaptation. Before defective dark adaptation in specific individuals or groups of subjects can be definitely ascribed to vitamin A deficiency, therefore, evidence must be obtained of consistent improvement after the administration of the vitamin. All effects due to increased familiarity with the method of testing, to the presence or absence of fatigue, or to other minor alterations in the experimental conditions must be carefully ruled out.

Secondary ("conditioned") Deficiency of Vitamin A.

Evidence of the existence of secondary deficiency was first obtained during investigations of the vitamin A contents of specimens of human livers taken at autopsy (Wolff, 1929, 1932, Moore, 1932, 1937). Moore, who examined over 1,000 specimens, found that in almost all diseases the ranges of vitamin A reserves were lower than in accidental

death, but that in some diseases the depression was much greater than in others. Thus in accidental death the medium reserve was found to be 220 I.U. per g., in hernia 160, in pneumonia 63, in chronic nephritis 25 I.U. per g. Recent experiments have shown that in rats deficiency of vitamin E leads to a great reduction in the ability of the liver to store vitamin A (Moore, 1940).

Further evidence suggesting secondary deficiency of vitamin A has been obtained by dark adaptation tests on diseased subjects. Thus in urolithiasis (Ezickson and Feldman, 1937), cirrhosis of the liver (Haig, Hecht and Patek, 1938) thyroid diseases (Wohl and Feldman, 1939) and diabetes (Brazer and Curtis, 1939) inferior dark adaptation fails to respond, or responds but slowly, to treatment with vitamin A.

All workers are agreed that in cirrhosis of the liver the reserves of vitamin A in the liver at autopsy are often extremely low. In this instance therefore the conclusions reached by the alternative methods of dark adaptation and liver assay are in agreement.

Diabetes, however, presents an interesting problem. In this disease the vitamin A reserves of the liver are usually abnormally high. The association of increased reserves of vitamin A in the liver with the carotenemia common in diabetes can hardly find sufficient explanation in an increased consumption of vegetables. The explanation is probably to be found in a retardation in the metabolism of both carotene and vitamin A. Carotenemia is also found in hypothyroidism, and the limited evidence available suggests that the vitamin A reserves of the liver are also raised in this disease. There is some evidence that abnormalities both in dark adaptation and the metabolism of carotene and vitamin A also occur in hyperthyroidism. It seems probable, therefore, that in all these diseases faulty dark adaptation is associated with an abnormality in the metabolism of vitamin A, rather than with simple dietary deficiency or inadequacy of the reserves present in the liver. While thyroid diseases certainly affect the carotene-vitamin A system the evidence for a specific antagonism between thyroxine and vitamin A seems inadequate (Baumann and Moore, 1939).

The Excretion of Vitamin A in Pathological Urine.

Since vitamin A is strictly fat-soluble its presence in urine would not be expected. In agreement with this supposition the vitamin is invariably absent from normal human urine. Boller and Brunner (1936), however, found considerable amounts of vitamin A in urine excreted in some types of diseases, and their findings have been confirmed and amplified by many other workers. Excretion is most commonly found in pneumonia and other acute infections, in chronic nephritis and some skin diseases.

In pneumonia the vitamin, which is excreted in an extremely concentrated form, in relation to the lipoid content of the urine, may exceed the daily intake. The excretion ceases abruptly after the crisis has been passed. In chronic nephritis the rate of excretion is usually lower, but the loss continues as a permanent abnormality. The presence of vitamin A in the urine in kidney disease suggests a grave prognosis.

It seems significant that some of the diseases showing urinary excretion of vitamin A are among those in which low ranges of the vitamin have been found in the liver at autopsy. The extent of urinary excretion of vitamin A however does not appear sufficient to account quantitatively for the depression of the liver reserves.

The Daily Requirement of Vitamin A.

In 1937 a Technical Commission of the League of Nations assessed the vitamin A requirement of the human adult at about 3,000 I.U. per day (3,000 I.U.=1.8 milligrams of β -carotene). A recent estimate by Wagner (1940) in Germany, based on careful experiments with volunteers, places the requirement at 2,500 I.U. of preformed vitamin A, or 5000 I.U. of carotene in oily solution. This difference is presumably due to the inferior

ability of man to use carotene as opposed to the rats employed in calibrating vitamin A against the carotene standard. In practice the vitamin A requirement will not be obtained exclusively as either preformed vitamin A or carotene, but as a mixture in which the former will be represented mainly by milk, butter, eggs and liver, and the latter by green vegetables, carrots, and some yellow fruits such as the apricot. As a rough estimate 1000 I.U. daily of vitamin A may be taken by the average adult as foodstuffs of the first group, leaving at least 2000 I.U. to be provided as vegetables. Since the absorption of carotene from vegetables may be very inefficient it is desirable that carotene should actually be available at three or four times this level. This may readily be achieved by including a generous allowance of carrots.

TABLE 1.
SOURCES OF VITAMIN A OR CAROTENE.

		<i>I.U. per g.</i>	<i>Price per 100 g.</i>	<i>Daily Requirement (g).</i>	<i>Price of 3,000 I.U.</i>
Halibut-liver oil		60,000	32/-	0.05	0.21 d.
Cod-liver oil		1,500	1/24	2	0.28
Butter		50	44	60	2.6
Milk		2	1/2	1500	9.2
Carrots		200	1/2	15	0.082
Red palm oil		1,000	1/4	3	0.0086

The typical vitamin A or carotene contents of some of the more important sources are given in Table 1, together with the approximate retail prices at the beginning of the war. It will be seen that the concentration of vitamin A varies over a very wide range, as also does the cost of the daily requirement of 3,000 I.U. Thus sufficient halibut-liver oil to meet the daily requirement costs less than a farthing. With milk, in which vitamin A is only a minor component, the cost is over ninepence. It must be emphasised that an attempt to satisfy the whole national requirement of vitamin A with halibut- or cod-liver oil would probably lead to an acute shortage of these commodities. The supply of carrots, on the other hand, is sufficient to meet all demands at an even cheaper cost. Red palm oil is an extremely rich and cheap source of carotene, but, although it is used in the tropics as a cooking fat its taste might not be generally approved in this country (Fitzgerald Moore, 1940).

The Value of Vitamin A Therapy.

In tropical countries where xerophthalmia is endemic the value of vitamin A as the specific remedy for this disease is beyond question. The same may be said in regard to the treatment of overt deficiency occurring in temperate zones as a result of economic stress. The outbreak of xerophthalmia which occurred among young children in Denmark during the last war as the result of the excessive export of butter to Germany may be recalled as a situation leaving no doubt as to the necessity for vitamin A therapy (Bloch, 1921).

In normal medical practice in temperate countries, however, the indications for vitamin A therapy are less clearly defined. The dark adaptation test, which might be expected to allow the detection of early or partial deficiency, is of recent introduction and is reliable only in expert hands. In the absence of definite indications of deficiency the vitamin is often given to undersized children, to subjects particularly prone to colds or to convalescent patients in place of a tonic. Large amounts must also be taken by healthy subjects through the appeal of advertisements in the press. Questions arise as to how far such indiscriminate dosing with vitamin A can be justified, and to what purposes our available supplies of the vitamin can most profitably be applied.

At the present stage of research the following factors would appear to be of importance in deciding the desirability of vitamin A therapy in any particular case: (1) The special requirement as influenced by growth, pregnancy or lactation. (2) The existence of any abnormality, such as steatorrhoea or gastro-intestinal obstruction, which would be expected to interfere with the absorption of carotene. (3) Pneumonia, or some other disease causing a rapid wastage of the vitamin A reserves. (4) Economic or political circumstances which seriously depress the consumption of dairy products.

Although in pregnancy there is little apparent increase in the demand for vitamin A it is advisable that the reserves should be built up to provide for the considerable excretion during lactation. The total loss until weaning has been calculated by Ellison and Moore (1937) to be roughly equivalent to the entire maternal reserve at the commencement of lactation. Both for the mother and child therefore an adequate supply of vitamin A is essential. When the mother is unable or unwilling to include in her diet large amounts of dairy products, or adequate allowances of carrots and green vegetables, the administration of a vitamin A preparation is to be recommended. A daily dose of 5,000 I.U., equal to 5 drops of standardised halibut-liver oil, should be sufficient. As an additional precaution a small dose, of say 2 drops per day, may be given to the baby. This provision appears well-advised even when the mother's diet is rich in vitamin A, and in failure of lactation it becomes more urgent. Since the available data suggests that the vitamin A reserves, which are very small in the new-born infant, do not usually reach the adult level until adolescence it is important that a generous allowance should be given during childhood. Here again the necessity for vitamin A therapy will be decided by the adequacy of the diet and the willingness of the child to eat carrots and green vegetables. For backward children with poor or perverted appetites vitamin A therapy should be employed.

In steatorrhoea it seems reasonable to give large amounts of vitamin A to compensate for the poor absorption of the fats. In pneumonia vitamin A therapy may be advised for two distinct reasons. In the first place determinations of the liver reserves in cases of accidental death have shown that in any large group of subjects a few will have low, or even negligible, reserves of vitamin A. Pneumonia in experimental animals with equally low reserves would almost inevitably be fatal. Vitamin A may therefore be given to the many in the hope of helping the few. Secondly, it is well established that in almost all cases of pneumonia a heavy loss of vitamin A occurs in the urine, which partially accounts for the lowered reserves of vitamin A generally found in the livers of fatal cases. Vitamin A should therefore be given to permit the restoration of the reserves to the level prevailing before the disease. Since a loss of about half the liver reserves may be suspected intensive therapy seems advisable. A dose of 60,000 I.U. per day, or about 50 drops of halibut-liver oil, does not seem excessive, and should be continued during convalescence. The same procedure may be advised in other acute infections, and in most serious acute illnesses the administration of the vitamin during convalescence will probably be helpful. It is doubtful whether the secondary deficiencies of vitamin A often found in chronic nephritis, cirrhosis of the liver, and some other chronic diseases will be alleviated by dosing with the vitamin, but a moderate daily allowance might serve to supplement the action of the defective liver in maintaining the level of vitamin A in the tissues. Much research remains to be done on this very difficult problem.

An instance of large scale vitamin A therapy in times of scarcity of dairy products may be found in the production of the new national vitaminised margarine. Vitamin A from marine sources is supplied in solution in hardened fat much more cheaply than when purchased as butter. Similarly, as indicated above, vitamin A may be supplied for more intensive therapy much more cheaply in the form of cod-liver oil, halibut-liver oil or a

concentrated preparation than as butter, milk or eggs, although, of course, vitamin A represents only a small fraction of the value of these foodstuffs.

In wartime vitamin A therapy finds a particular use in attempts to maintain dark adaptation at the utmost limit of efficiency. To the civilian the motive may be merely a more comfortable passage through the "black-out," but to the soldier, sailor or airman the ability to see may be of vital importance. The ingestion of a moderate dose of vitamin A some hours before night duty can be recommended as a step in the right direction, although it must not be forgotten that other factors, such as the absence of fatigue and practice in interpreting the significance of what little can be seen, must also be of importance in deciding the degree of efficiency in action at night.

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