SUBMITTED FOR EXAMINATION IN THE DEGREE

OF

DOCTOR OF MEDICINE

OF

GLASGOW UNIVERSITY

.....

A Study of the Histopathological changes in the Retina and late changes in the Visual Field in acute Methyl Alcohol Poisoning.

.....

Introduction

Literature

Histological Methods

Histological Findings in Retina

Field Changes in Methyl Alcohol Poisoning and Quinine Poisoning

Discussion

Bibliography

ProQuest LLC.
789 East Eisenhower Parkway
P.O. Box 1346
Ann Arbor, MI 48106 – 1346

Introduction

In April 1942 eighteen cases of poisoning by imbibition of methanol or methyl alcohol were treated in the Glasgow Western Infirmary.

Seven of those were gravely ill and died in hospital. We were able to secure the eyes of four of them, three men and one woman, for histological examination. In our investigation of the literature we were struck by the few histological examinations that have been made of the retinas of the victims of this poison. It is not common in this country, to have outbreaks of acute illness resulting in death from drinking methanol. Its use as an adjuvant, or fortification, to other drinks, is frequent on the Continent and was common in the days of prohibition in America. This may lead to gradual deterioration in vision. Similarly, it has been described as occurring in industries, such as varnish and shellac manufacture, where the fumes are absorbed from the lung. It may also be absorbed from the skin.

Throughout the literature the same difficulty is

encountered, namely, how much spirit did the patient have? The amount necessarily lethal is variable. Our fatal cases probably had several ounces of spirit over the forty-eight hours before admission to hospital. As nearly as we could estimate, one of the men who recovered and whose field of vision we discuss later, probably had six ounces at least, diluted in beer and coffee.

Wood-spirit drinking in sporadic cases may often be ignored as a cause of death. Jackson believes it to be one of the commonest causes of toxic blindness in America. But the incidence of several cases with a similar clinical picture from whom a history of a recent drinking bout can be obtained, should make one suspicious. Especially is this so if the potion has been retailed from one source mainly. It was sold clandestinely to our patients as gin, at a shilling a bottle, in a public house in the town. Where the history of drinking is not obtained, the picture may be puzzling. A practitioner who was called to see the husband and wife who died and whose retinas we report on, found them stuporose and cyanotic. He told us he could not make up his mind what was the

matter before he sent them to hospital.

It might be well here to sketch the clinical picture presented by those people on admission to hospital. The gravely ill were comatose, pale or cyanotic, perspiring and profoundly shocked. The pupils were widely dilated and immobile. As death approached cyanosis increased and the pulse became less and less perceptible. The four cases which came to post-mortem were in this category; one lived only two hours after admission. Sometimes convulsions preceded death. In the less severe type, the patient was confused, perspiring, pale, with the cramps and abdominal pains of acute irritant poisoning. Vision deteriorated in one case to the perception of hand movements within twelve hours of admission. This man, whose field of vision we shall later discuss, recovered useful vision. A common symptom is yellow vision. The patient may be comatose and recover, and he may be blind and recover vision, at least in large part. The milder cases have gastro-intestinal symptoms with transient dimness of vision.

Ophthalmoscopic signs are usually present in severe cases. Indistinctness of the disc edges and congestion

of the veins suggested oedema of the nerve head. Oedema of the retina around the disc was apparent in some. One man who was admitted in coma recovered rapidly and his visual acuity was $\frac{6}{9}$ Snellen, right and left, on recovery, so that gross visual loss does not necessarily accompany a degree of poisoning sufficient to produce coma.

The principles of treatment employed in the medical wards were elimination by maintained gastric lavage and the promotion of diuresis by intravenous glucose salines, lumbar puncture, cardiac and respiratory stimulants. The elimination of methyl alcohol is slow (Haggard and Greenberg).

Our interest in the incident of this outbreak was aroused because a scrutiny of the literature revealed only four papers that we could find on the histology of the retina and nerve in methanol poisoning in the human. These findings, as we shall show now from an examination of the literature, were the changes of oedema with changes in the nucleus of the ganglion cells, and little if any change in the nerve. We sought to use additional variations of technique by examining the retina in bulk, staining for lipoid change in addition to the routine methods. Then we examined those of the surviving patients who would submit, to estimate their visual impairment, if any.

Now that a year has elapsed we thought that a study of the visual field and ophthalmoscopic appearances would be useful in conjunction with the appearances and field changes which we have been observing over a period of two months in a man with quinine blindness.

The study of this last patient has provided us with so many clinical data that we propose to publish our findings in a separate communication.

The end results are very different after the action of these two producers of toxic blindness. As the mode of action of neither of them is certainly known, discussion on the facts of this investigation are probably justified.

Histological Methods.

Examination of Retina, in Bulk:

The retina which had been fixed in 8% formol-saline by injecting the globe shortly after death was taken from the eye intact and spread out flat on a slide. All debris and superfluous vitreous was removed with a camel-hair brush and the retina was washed in running water for ½ - 1 hour, to remove the fixative. After having removed a small piece for routine embedding and staining by Haematoxylin and Eosin, the remainder was stained by Spielmeyer's Scharlach Red for fat and mounted in Glycein. The details of this are as follows:-

Scharlach Red (Spielmeyer)

Abs. Alcohol	35 cc.
10% Caustic Soda	10 cc.
Aqua Dist	5 cc.
Scharlach Red to saturation	1 gm.

Leave in incubator for one hour.
Filter through three layers of filter paper into a closed dish.
Place sections in stain and heat gently until steam collects on underside of the glass dish cover.

Stain for a further twenty minutes.
Rinse in 50% Alcohol.
Rinse in water.
Mount in Glycerine.

After examination in bulk was complete, portions were taken for examination in section by washing in water for 10 - 20 minutes to remove the glycerine and embedded in gelatin. The details of embedding were as follows:-

Gelatin Embedding.

1. Transfer to 12.5% solution of Gelatin 3-6 hours.
2. Transfer to 25% solution of Gelatin 3-6 hours.
3. Embed in 25% solution of Gelatin and allow to set.
4. Immerse block in 5% Formalin overnight to fix Gelatin.
5. Freeze and section.
6. Transfer sections to water 10-20 minutes to remove Formalin.
7. Transfer to Mallory's instantaneous Haematoxylin 30-60 seconds.
8. Blue in tap water 5-10 minutes.
9. Mount in Glycerin jelly.

The procedures employed in investigation of the pathology of the nerve were Marchi's impregnation for degeneration in the myelin sheath and a rapid method of staining myelin described by Smith and Quigley instead of Weigart Pal, and then serial sections of the nerve were examined by staining with Haematoxylin and Eosin.

The steps as we applied them in Marchi's method were as follows:-

Marchi's Method.

1. **Fix** tissue not more than 2-3 mms. thick in the following fluid:

   Muller's Fluid ..... 2 parts
   1% Osmic Acid ..... 1 part

   7-21 days. Change fluid every 5 or 7 days.

   NOTE: Tissue may be transferred directly from Formalin to the Bichromate mixture but Formalin fixation should not be prolonged beyond a few days.

2. Wash in running water several days.

3. Dehydrate rapidly and embed in Paraffin.

4. Section.

5. Dissolve Paraffin in Xylol and mount in Balsaam.

The rapid staining of myelin sheaths was carried out as follows, after Smith and Quigley (quoted by McClung):-

Fix in Formalin, Alcohol Formalin and Bouin's Fluid.
Fixatives containing copper or chromium salts not applicable.
Paraffin, Celloidin or frozen sections may be used.

Directions.

1. Mordant sections for 15 minutes in a 4% aqueous solution of ferric ammonium sulphate ($FeNH4(SO_4)2$) prepared from violet crystals.

2. Rinse in 70% Alcohol to remove excess of mordant.

3. Place sections in a solution of 1% Haematoxylin containing 2-3% glacial acetic acid by volume. (The 1% Haematoxylin solution is made by the addition of distilled water to a 10% stock solution of Haematoxylin in absolute alcohol).

   The solution containing the sections is kept at or near 55 degrees centigrade. The sections at first stain deeply, then differentiate, and the depth of the staining of the myelin sheaths depends on the time the sections are left in the staining-differentiating solution. Usually between 30 and 60 minutes are required, but thin sections may require less time.

4. Place in a half saturated solution of Lithium Carbonate for 5-10 minutes.

5. Rinse in tap water.

6. Counterstain or stain for chromophilic substance if desired.

7. Dehydrate in Alcohol, clear in Xylol and mount in balsaam. Myelin is stained a deep blue and the background remains either colourless or slightly tinted, depending on the degree of differentiation.

Having in mind the pitfalls and artefacts that may occur in Marchi's method (Roussy and Lhermitte), we stained a normal nerve, one piece of which had been subjected only to the trauma of removing it from the skull and another portion of which was traumatised further by subjecting it to a weight of 14 lbs. for 24 hours.

The artefacts quoted are:-

1. Tissues must be carefully excised without squeezing, since degenerated myelin is easily displaced by pressure.

2. Over-fixation may produce black granules (Marchi's pseudo-granules).

3. The stain is liable to be extracted by prolonged dehydration.

4. The Lipochrome granules of the nerve cells may also be stained black and brown.

5. Previous prolonged ether anaesthesia is said to increase the artefact granules (Swank and Davenport).

Literature.

As we have stated, the communications dealing with the pathological changes in the retina in methanol poisoning are few. The work of Pick and Bielschowsky, McDonald and Menne are quoted in the text of the next section under "Histological Findings in the Retina." We have been unable to procure the paper of the Russian, Eleonskaja.

The literature shows two peaks of interest. On the Continent there was an outbreak of acute poisoning in 1911 in Berlin. In America during the years of prohibition there was a great increase in the number of addicts to denatured or "dehorn" alcohol.

The Berlin outbreak resulted from the drinking of cheap schnapps. There were 130 cases, and 58 died. The severe cases had marked gastro-intestinal symptoms with respiratory distress and cyanosis later. Onset of symptoms was 24-36 hours after drinking. The pupil was maximally dilated, not responding to light or accommodation. The vision failed and blindness was common. There was no paralysis of the extra-ocular muscles. Death was attributed to paralysis of the respiratory centre. Mild cases exhibited loss of vision with moderately

dilated but reacting pupils and sickness. Thirty of the cases were sick only, with no other features. In those who survived the vision tended to improve but it was impossible to give an accurate prognosis as sometimes the mild cases deteriorated in vision without any warning signs (Hirschberg).

Ophthalmoscopically where changes were noted they constituted a hyperaemia of the disc and congestion of the veins in the early stages. Of those who survived, 75% had some change in vision. They showed optic atrophy with a marked atrophic cup. It was emphasised that the large pupil was a sign that the danger of further loss of vision was not past. There is often a stage of improvement followed by deterioration. Lewy often found no change in the fundus, or at the most a slight opaqueness of the tissue of the disc. These had just as high an incidence of subsequent atrophy. On the other hand, he found that some cases of neuritis disappeared without any visual loss. Rost found that in convalescence the field was generally contracted and there was an absolute central scotoma. He believed that the diagnosis was easy, provided the condition was kept in mind. He believed that all methyl alcohol substances should be on the list of poisons. In therapy he em-

ployed gastric lavage and enemata, strychnine caffeine and hot packs.

In America the first reports of poisoning appear as early as 1879. By the end of the century there were many reports of blindness from this cause as the spirit, which had been treated to remove the bad taste, was put on the market. It was not until 1906 that Congress passed the Denatured Alcohol Bill. Between that time and the advent of prohibition in 1918 there were few cases reported. Cases of poisoning from inhalation and external application were reported about this time. Some people showed a special sensitiveness to the poison. In some, 10 cc. was enough to cause blindness, and 100 cc. had been known to cause death. A concentration of as little as 0.2% in the air had been known to cause poisoning.

The tendency to recover vision was considered poor. De Schweinitz (quoted Traquair) reckoned that 90% of cases of poisoning went on to blindness. Traquair, speaking of his experience in Edinburgh, thought that the tendency to recover was slight or absent. Other authors do not agree and state that the subsequent vision is always better than the vision existing when the patient is at his worst.

The symptomatology is divided into general and ocular manifestations in acute cases. Headache, dizziness, nausea and vomiting, abdominal pains, cardiac weakness, slow pulse, sighing respiration, marked prostration, weakness of the extremities, delirium, convulsions, stupor, and death are the general features. The visual symptoms are loss of vision, photophobia, pain on moving the eyes, and hemeralopia. The signs in the eye are lowered tension sometimes, dilated and fixed or sluggish pupils, normal or congested discs and central scotoma. The signs in chronic poisoning are the central scotoma with an atrophic nerve head, either complete atrophy or limited to the temporal side, with pallor.

The general pathological findings are acidosis with increased acid content of the aqueous; blood shows a reduced coagulation time, but increased viscosity with increase of erythrocytes and leucocytes and haemoconcentration. The lymphocytes however are decreased. There is a high output of ammonia in the urine in consequence of the acidosis. The gastro-intestinal tract shows mucosal haemorrhages. Menne writes of the complete pathological examination of twenty-two victims of methanol poisoning in Multnomah County, Ore., who died in 1934. He said that of the fifteen who were hospitalised, five

were comatose, five were conscious, and the remainder stuporose. Most of them were profoundly shocked, cold, clammy, and perspiring excessively.

The blood pressure of those in which it was recorded was, systolic 110 to 140, and diastolic 78 to 90. Most of them were too ill to comment on visual symptoms. They all had irregular, often laboured and spasmodic, respiration. The rate varied, and nearing the time of death was 2 or 3 per minute. Cyanosis was marked in the late stages. Death was from respiratory failure. Many were pulseless on arrival in hospital; in some others the heart rate was rapid but in most it was slowed with the respiration. Terminal convulsions were present in three cases. He expresses the usual doubt in assessing the amounts of alcohol taken. He could say that as much as 500 ccs. of spirit was drunk by one man. Similar difficulty is expressed in estimating the time elapsing between intake and onset of symptoms but in most it was from 16-24 hours. It was earlier in some. Six patients died within an hour of hospitalisation, three within five minutes, and the remainder within one to seven hours.

The changes found post-mortem in the central nervous system were sub-pial and moderate cortical and sub-cortical interstitial oedema. Only occasional focal haemorrhages

were seen. On the whole the cellular changes were not marked. "The extensive degeneration of the ganglion cells and the vascular endothelium with subsequent haemorrhage in the mid-brain, pons and medulla oblongata referred to by Weil was not observed, probably because of the acuteness of the intoxication", Menne says. We quote him again - "Geitler and St. George noted only pronounced cerebral congestion with an increase of spinal fluid and engorgement of blood vessels."

Detailed studies of brain histology were wanting, but he quotes the experimental work of Eisenberg who examined rabbits after a period of inhalation of 0.2% wood alcohol for periods of two, four, six, eight, and ten months. Eisenberg found degeneration of various degrees with an indefinite line of demarcation between the gray and the white matter, diminution of neurocytes, with spindling and disappearance of Nissl granules, and a scattering of brownish pigment. In the later stages of the severe intoxications in these rabbits there was a marked decrease in the size and number of parenchymal cells. The nuclear changes varied in degree up to complete karyolysis. We quote Menne again - "Scott and his associates exposed monkeys, rabbits, and rats to methyl alcohol by cutaneous absorption, inhalation and

ingestion. They found in their animals, capillary congestion, oedema and patchy degeneration of the neurons. These changes were more often found in the spinal cord than in the brain. These authors quote Rühle as having found in dogs scattered haemorrhages along the blood vessels of the pons, medulla and cord as well as large amounts of lipoid in the vascular endothelium and perivascular tissue. The deposition of lipoid often preceded the haemorrhage. Scott and his co-workers concluded that only parenchymal and neuronal tissues were affected. Such experimental evidence is probably of more value in depicting the injurious effect of methyl alcohol on the central nervous system than are the changes observed in such acute conditions as I have described in human beings, material which is not so accurately controlled. However, the susceptibility of the tissue of animals to wood alcohol must be considered in the evaluation, since there is such wide variation in the effects in different animals studied. The derivation of formic acid and the alcohol itself may becloud the real changes in the parenchyma of the central nervous system because of their fixative action."

Menne's examination of the eyes of which one was removed from each body with formalin fixative and routine

staining led him to state that "the most pronounced changes were in the ganglion cells of the retina which showed irregular staining, eccentric placement of nuclei, fraying of cycloplasmic outlines, vacuolation and autolysis. In many cases only about one in fifty of these cells approached normal, while in some of the eyes they were absent from wide areas. These changes in the ganglion cells were most marked near the disc. There were no marked changes in the glial cells, except oedema. The other layers of the retina were without notable alterations, with ordinary stains." No change was seen in the optic nerve.

He found no hypertensive changes in these people who were regular addicts to wood spirit, they even proferred wood spirit to any other, and were well known in the community for their habits. In only one was there cirrhosis of the liver, and that was of a minor degree. Many livers were enlarged, but there were no focal lesions such as are seen in experimental alcohol poisoning. The alimentary tract showed congestion of the mucosa and small haemorrhages and, in the stomach, areas of necrosis. The kidneys were enlarged slightly, engorged, and greyish, but the minute structure was remarkably preserved and there were no changes of note found in heart, spleen or

pancreas.

The remarkable fact was the good bodily preservation of these people who regularly consumed wood spirit in small quantities. There was nothing to show that it had produced an increased susceptibility to disease or had hastened the processes of degeneration and decay. The changes in the retina and brain were not regarded as selective, but merely the result of the greater vulnerability of specialised tissues to the poison, which in its clinical manifestations of disturbance of the respiratory centre, blindness, convulsions, shows toxic effects on the nervous system. "Experimentally Gradinesco and Degan showed that in weak concentrations (5% to 10%) methanol causes excitability and then diminution of response going on to complete paralysis. The effect undoubtedly varies with the rate of oxidation in different persons, hence the peculiar and varying clinical episode", Menne states. The accidental death of this series of cases of known addicts was due to the inadvertent sale of pure methanol. The normal industrial strength for most purposes in America has been reduced from 10% to 2%

Ethyl alcohol disappears from the body by two processes; elimination, largely through the lungs; and oxidation in the tissues. On the other hand, methyl

alcohol is also eliminated through the lungs, but it is not oxidised in the tissues to a great extent.

"The elimination of both alcohols is controlled by the same principle. It is the principle which has been described mathematically by Haggard for the elimination of ethyl ether, an entirely non-reactive substance. In the operation of this principle the coefficient of distribution of any volatile substance between the blood and air in the lungs plays an important part. For methyl alcohol the coefficient is high; that is, its solubility in the blood is high in relation to its vapour tension in the pulmonary air. The elimination of methyl alcohol through the lungs therefore is much slower than ethyl ether which has a lower coefficient of distribution." (Haggard and Greenberg). "Only about 10% of the total amount of ethyl alcohol which disappears from the body is eliminated in the expired air. For methyl alcohol on the contrary, Voltz and Dietrich have estimated that approximately 30% is eliminated; and Asser has estimated 53%. We find that more than 70% of the amount disappearing from the body appears in the expired air. We find further that the rate of elimination is not constant, but instead is a function of the concentrations in the blood and of the volume of the

pulmonary circulation; that the curve obtained by plotting the concentrations of methyl alcohol in the blood is exponential; and that the rate of loss of methyl alcohol by elimination through channels other than the lungs and by oxidation, is not constant, but is likewise a function of the concentration in the blood." (Haggard and Greenberg).

The only addition to the usual eliminative therapy with cardiac and respiratory stimulation that we could find was the suggested efficacy of thiamine hydrochloride in a case quoted by Simons which recovered vision. He states that he is aware that recovery might have taken place without thiamine, but that its use may be worth while. According to Norbury, "The chief physiologic role of vitamin B_1 in the body is regulation of cellular respiration. Nerve cells require relatively more oxygen than other types of cells and they are more susceptible to any lack of oxygen. If Friedenberg is correct when he says that in methyl alcohol poisoning, as in bee-stings, it is the formic acid that causes strangulation of the optic nerve fibres by the sudden tissue swelling, then it seems reasonable that an increased amount of thiamine would be of value in combating the anoxaemia.

Traquair described the field changes in methol

alcohol poisoning thus - "The clinical picture as described by Uhthoff, Goldflam and others is that of an acute intoxication. Usually two or three days after ingestion the visual loss commences, and complete or nearly complete blindness rapidly ensues. Both eyes are always affected though often unequally. After several days or even weeks, according to the severity of the case, vision begins to return at the field periphery, and a large absolute defect broken through at one side or the other becomes demonstrable. This breaking through may give the field an irregular crescentic, or sometimes pseudo-hemianopic appearance. Later the periphery may recover more or less all round, leaving a central scotoma.

The defect is peri-central rather than para-central or centro-caecal, though it often includes the blind spot and the intensity is usually high. According to the severity of the case, all degrees of size, intensity and permanence may result. In the worse cases little or no recovery may take place; in the milder ones normal vision may be restored. This stage is unfortunately frequently succeeded by a second and permanent loss of vision associated with pallor of the optic disc. The field becomes depressed and contracted; Goldflam noted

a return to blindness in a case which had reached normal vision. Only exceptionally does good vision return and remain when the field changes have been severe to begin with.

The field changes indicate the action of a violent poison on the retinal cells and optic nerve with special selectivity for the central elements. The ultimate deterioration of vision following primary recovery is probably due to a cicatricial process in the nerve depending on the violence of the reaction, leading to secondary atrophy.

The diagnosis depends upon the clinical picture as a whole, which usually suggests this kind of poisoning: the prognosis should be guarded at first, even although the field defects have greatly improved, as secondary atrophy may supervene. The fields should therefore be watched for some time after apparent recovery."

There is a very large literature on amblyopia following quinine poisoning and we do not propose to go into that in detail. We shall show and discuss the fields of a man who was under our care for many weeks because they make a helpful contrast to the field changes in methanol poisoning. Again the question of field changes in quinine is best summarised by quoting Traquair.

He says — "In the second group of toxic amblyopias, quinine poisoning occupies the most important position. In mild cases a temporary dimness of vision may occur; where the poisoning is more severe, rapid and complete blindness results. After a few hours or days, sometimes weeks, vision begins to return, recovering centrally much more than peripherally, and the fields show depression, especially peripherally and contractively. The light sense is greatly reduced. At this stage the edge of the field is extremely steep and disproportion between the fields for colour and white is present. Though often more or less reduced, central vision by Snellen's types may be nearly or quite restored, while the peripheral field remains depressed or constricted. The loss of field may be extreme, the part remaining, extending only a few degrees round the fixation point; in such cases the patients exhibit great orientation difficulty.

In shape the outline of the field is often laterally extended and vertically compressed following a somewhat horizontal elliptical figure. This is due to an apparently greater tendency of the temporal and nasal parts, than of the upper and lower, to recover. The restoration may be irregular, some peripheral parts recovering while others remain blind. In this way various forms of

sector-like defect may be produced, and when the lost areas are symmetrical in the two fields a superficial resemblance to hemianopia may arise. In other cases a more regular concentric contraction leading to an approximately circular field occurs. Much depends on the care with which the fields are taken, as especially in the early stages of recovery, discrepant and inconsistent responses are apt to be obtained and stringent precautions to check the patients' replies are necessary.

Several months may elapse before the fields become stationary, and in mild or favourable cases they may recover almost completely, but some peripheral depression persists; in more severe cases a more or less high degree of depression and contraction remains permanently. Central scotoma has been recorded but it is very rare, and possibly really a complication rather than a true manifestation of quinine poisoning. Colour disproportion disappears as the condition becomes stationary."

The question as to whether the field changes are due to the poisoning of the ganglion cells or whether they result from the ischaemia due to the marked narrowing of the arteries with consequent atrophy has often been discussed in the literature. Ballantyne has emphasised for us what may be called the paradox of qhinine blindness,

i.e. the onset of narrowing of the arteries after the vision has begun to improve: this narrowing continuing while vision returns. There may be nothing of note in the fundus in the early stages, or there may be a well marked "milky" retina with cherry spot and hazy disc margins.

In Ballantyne's case he found the first fundal change apart from what he describes as "perhaps a little haziness of the nasal border of each disc, and some streakiness of the retina along the course of the vessels, leaving the temporal side of the discs", on the 5th day of examination and nine days after the ingestion of quinine when "the discs were found to be pale and all the vessels abnormally pale and narrow. There was still no vision, and the light-reaction of the pupil was very doubtful even with strong focal illumination. Twenty-four hours later vision began to return. There was steady improvement in vision from day to day and a gradual expansion of the field, but at the same time an increasing pallor of the disc and narrowing of the vessels."

Ballantyne concluded that the blindness and loss of field was due to an alteration of the retinal elements of an invisible nature not as a sequel to the ophthalmoscopically observed changes in the vessels with ischaemia.

He was of the opinion that the toxic action was directly on the retina and that "the ultimate recovery of central vision with loss of peripheral vision and failure of vision in twilight, suggests a selective action of the poison upon the rods." Experimental work on quinine poisoning done on dogs and rabbits with a view to determining the histological changes after poisoning, is summarised thus, "changes have been observed within ten hours of administration of the drug, a chromatolysis and degeneration of the ganglion cells, degeneration in the nerve fibres, and, in the later stages, thickening of the walls of the blood vessels, with thrombosis and obliteration of their lumen.

Oguchi (1932) found that the metabolism of the retina was considerably reduced by an amount varying from 20% to 30%. Pathological examinations of human eyes have been few (Fortunati, 1905; Gianini, 1934) but have shown the same changes." (Duke Elder).

Finally, the literature on the late ophthalmoscopic signs in quinine poisoning describes pallor of the nerve head with extreme narrowing of the arteries, and in methyl alcohol poisoning, pallor of the disc with cupping, the changes in the vessels not being very marked.

Histological Findings in the Retina.

Four retinas were available for histological study. The retina was first examined in bulk, stained with Scharlach Red, and mounted in Glycein (Lowenstein). By this means it is possible to examine the whole retina at once and to examine it in layers for fatty changes. With high power, and with the diaphragm cut down, it is possible to examine the ganglion cells minutely, and by continuous movement of the focussing adjustment of the binocular microscope, details of contour are enhanced by the stereoscopic effect. This is the dynamic microscopy technique (Lowenstein).

In all cases the ganglion cells contained fatty droplets to such a degree as to be apparent in (Fig.1, x 50) with low power. This appears as in (Fig.11, x 300) with higher power. The ganglion cells are full of dark red droplets and the big ganglion cells are almost exclusively affected. The droplets are of different size, the smallest scarcely recognisable with the (x 600) power, and the largest are about one-tenth of the size of an erythrocyte. With the method of staining retina in bulk with scarlet red, we recognise that the capillaries show, quite markedly, lipoid droplets in the walls,

(Fig. 3 A and B).

In a common investigation with Lowenstein (yet unpublished) we have studied these changes in the retinal ganglion cells in different decades of life. We found these fatty changes in the ganglion cells of the aged but they were very obvious even in the middle-aged. Only the retina of youth is free of them. This corresponds with the findings of Obermeyer, quoted secundum Spielmeyer. In many papers on brain histology this author found that senile ganglion cells, especially in the anterior horn, and Clark's columns, contained fatty corpuscles regularly increasing in incidence as age advanced. In the brain, however, these lipoid droplets are recognisable from the early age of eight years, until in extreme age the cells are filled with droplets. In our paper we prove that the condition in the retina is analogous. We can say that the lipoid changes we have found in these cases of the fourth decade suffering from methyl alcohol poisoning are no more marked than those we find in normal retinas of the same age, nor yet are the fatty infiltrates in the capillary wall more pronounced than those in the normal, similar age group.

In each retina there was an area of gross droplet deposition as in (Fig.4) situated about half way between

the disc and the periphery, but in no case at the macula. This area was trephined, mounted in Gelatine, and sectioned. The fatty droplets were large and extracellular. They were situated mostly in the ganglion cell layer but some were in the outer molecular layer (Figs. 7 and 8). They frequently showed a biphasic staining reaction with Scharlach Red, part of the droplet being dark purple and the remainder bright red as in (Fig. 5). The significance of this we do not yet know. It would be unwise to attach importance to this appearance of gross extracellular droplet formation. One would expect a change consequent on a diffusible circulating poison to be diffuse, not localised and intense. We feel that it is best regarded as post-mortem change.

The choroid was examined in bulk and fatty changes were found in the pigment cells with changes also in the capillary walls.

We believe that the technique of examination of the retina in bulk allows us to appreciate the size and shape of the ganglion cells and their minute structure better than by routine methods. There is no distortion from fixation and embedding. One can examine the cell in depth and get a stereoscopic picture.

Some of the giant cells have a diameter of 30 U..

The smaller ones are little larger than the rod bipolar. After examining hundreds of ganglion cells in our cases of alcohol poisoning we found most of them to have a clear regular cell outline. The cytoplasm was granular. This granular appearance was most striking in the giant cells. The nucleus was clearly defined, and frequently, the nucleolus. A proportion had not a clear cell outline and in some no nucleus could be seen. These same variations we found in our scrutiny of normal retinas of the same age.

Pick and Bielschowsky found the large ganglion cells showed a change in the arrangement of Nissl granules in the nucleus in their routine Haemalum and Eosin sections. They found that the granules were normally arranged coronally in the nucleus, while in their poisoning cases they were arranged in ill-defined clumps. They found that the fibre layer was fragmented and disintegrated. They thought that the inner nuclear layer stained more intensely, and changes in the nerve were inconsiderable. McDonald found that the ganglion cells were swollen and the fibre layer was thickened. Menne made similar observations also on routine Haemalum and Eosin sections.

Clinically, as we have said above, the fundal changes in life are oedema of the nerve head and of

the retina. Where this occurs one would expect the fibre layer to be thickened and less compact. But clinically there may be no fundal changes. It is our routine, when circumstances permit, to inject a few minims of 8% Formol-saline into the vitreous of the cadaver as soon after death as possible. We were not able to do this in our alcohol cases. The fibre layer consequently does not cut so well and there are interstices in the layer and the fibres are fragmented. With this in mind then, we would not venture to dogmatise on the question of thickness of the fibre layer or to say that the ante-mortem integrity of the fibres was in doubt. We think that the omission of early intraocular fixation may account for the fact that in our routine Haemalum and Eosin sections the fibre layer is fragmented and not compact. As Eugene Wolff so rightly stresses, post-mortem autolysis in the retina is so frequent and so rapid. We have not had the privilege of seeing McDonald's or Menne's sections and we certainly do not wish to dispute their findings in their examination of routine Haemalum and Eosin sections in methanol poisoning of the retina on the evidence of their photomicrographs.

We would like to show a photomicrograph of our routine sections in which so many shapes and sizes of

ganglion cells appear. Some are swollen and some are shrunken. In other slides some ganglion cells have no nucleus. But we have seen all those variations in retinas other than in alcohol poisoning cases, even in normal ones. The slight change in compact structure of the fibre layer was adequately explained by the absence of pre-fixation (Fig.6).

We have attached more significance to the conclusions we have been able to draw from examination of the ganglion cells in retina in bulk than to our routine sections.

Finally the nerve was stained in serial section, every fortieth section, by Weigert's Haematoxylin and nothing of note was found. This was followed by Marchi's technique which gave a slight Marchi positive of an unconvincing appearance. We stained a control nerve with Marchi and another piece of the same control which, in addition to the trauma of removal from the skull, was traumatised by leaving it under a weight of 14 lbs. for 24 hours. The nerve from the spirit poisoning patient and the normal control were identical. The traumatised control nerve showed some Marchi positive staining at the periphery. We did not think there was any evidence of degenerative changes in the myelin content of the nerve in our cases of methyl alcohol poisoning.

Fig. 1a (×50)

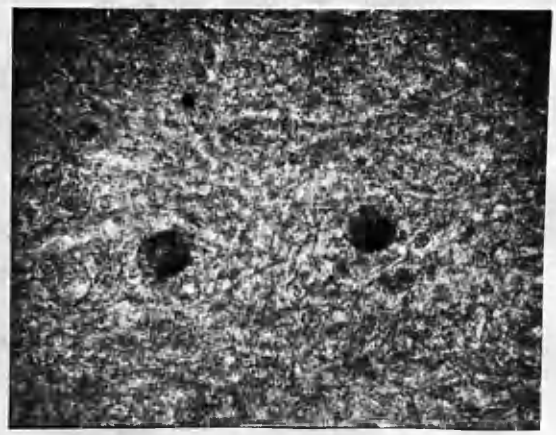


Fig. 1b (×150)

Photomicrograph of retina in bulk stained with Scharlach Red. Note the large ganglion cells standing out in the picture with the droplet staining.

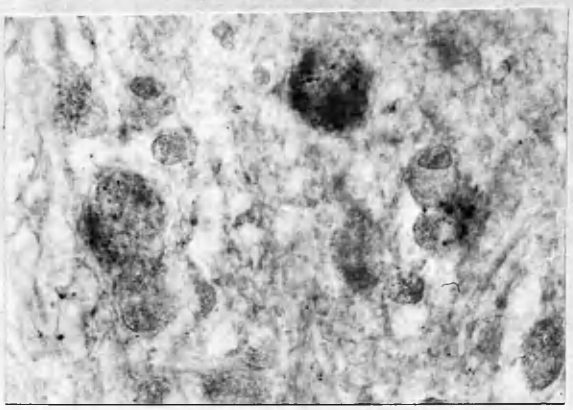


Fig. 2.

Stained with Scharlach Red in bulk without nuclear staining. Diaphragm very much cut down. Different sizes of ganglion cells are seen. Smaller ones show nucleus and nucleolus. The fatty droplets are of different density and size.

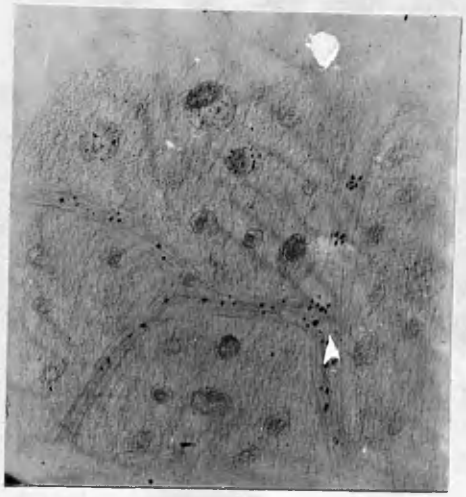


Fig. 3a.

Drawing showing droplet staining with Scharlach Red of the capillary walls.

Fig. 3b (×300).

Photomicrograph of retina in bulk stained with Scharlach Red, showing the droplet staining in the wall, the vessel being empty.

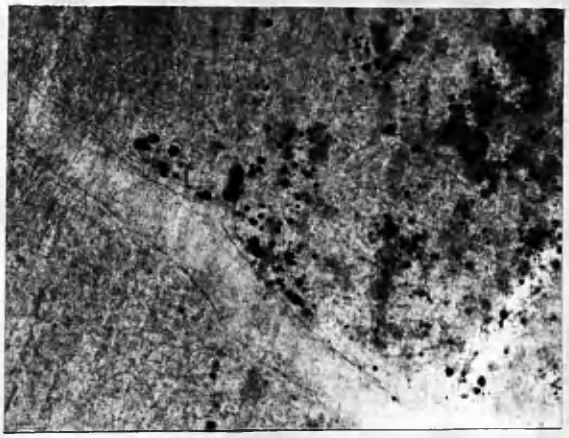


Fig. 4 (×50).

Gross fatty droplets in the fibre and inner molecular layer. This was present in each one of the four examined retinas;we do not exclude post-mortem change.

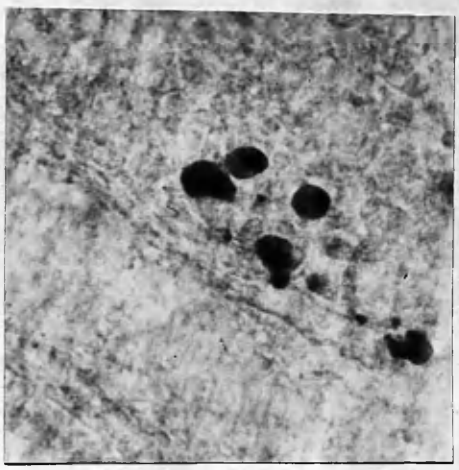


Fig. 5 (×300).

Higher power of fig. 4 showing the biphasic staining of fat, the nature of which is unknown.

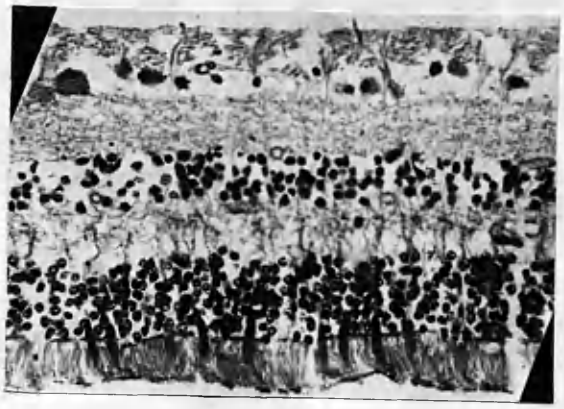


Fig. 6 (×300).

Paraffin section of retina stained with H and E. Various shape and size of ganglion cells.

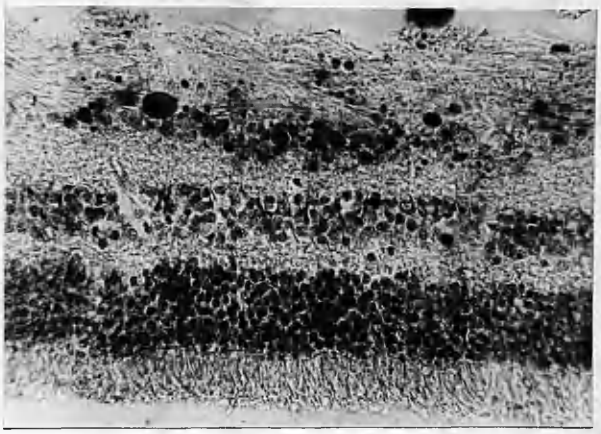
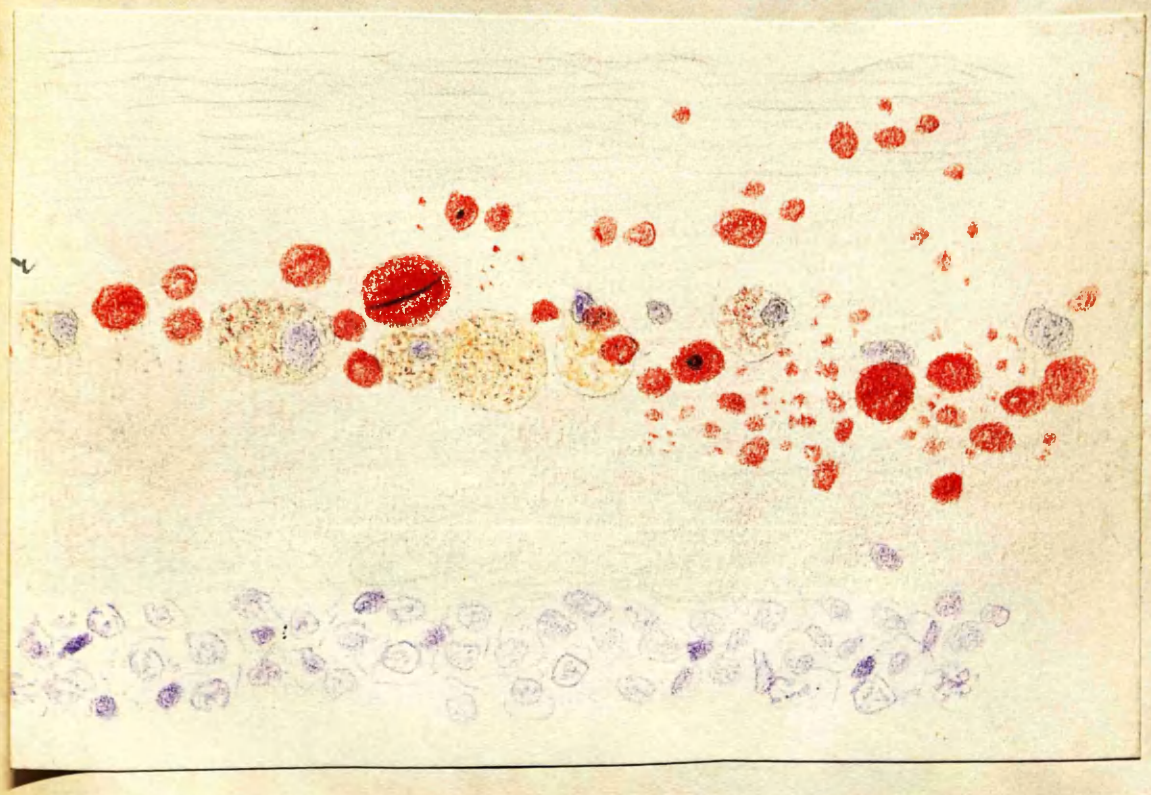


Fig. 7 (×300).

Stained retina in bulk with Scharlach Red embedded in gelatin, cut with the freezing microtome, counter-stained with Haematoxylin. Fatty droplets of various size and intensity appearing shining red in the nerve fibre and inner molecular layer.

Fig. 8 (×600).

Drawing of a slide. Technique as in fig. 7. Brown ganglion cells in section and dark purple droplets. Ganglion cells of different size with and without nuclei. The fatty droplets are mostly in the ganglion cell layer but are between the cells.



Changes in the Field of Vision in Methyl Alcohol and Quinine Poisoning.

After the lapse of a year we considered that the damage done to the eyes of those people who survived the poisoning would possibly be stable. Accordingly we summoned all of them several times to be examined. The hours of examination were arranged after working but the demands of war industries made it impossible for us to review more than four cases. They were seen on two occasions. We give the history and findings in each case.

CASE I. Mr. P.M., aged 33, had at least two ounces of spirit, well diluted. Legs were weak, no abdominal pains, haze over the eyes for a day. On this day his discs were described as reddish with blurred margin. On 1/5/42 they are described as normal and vision on the optometer was, right eye - $\frac{6}{12}$ and left eye - $\frac{6}{12}$ On 12/4/43 his fields of vision were perfectly full with two isopters, $\frac{7}{330}$ and $\frac{3}{330}$ white. His colour field was normal. There was no relative scotoma. His corrected vision was, right eye - $\frac{6}{9}$ and left eye - $\frac{6}{6}$. His fundi were normal.

CASE II.—Mr. D.M., aged 64, drank three ounces of industrial alcohol. Soon after he had pains and cramps and yellow vision. Semi-comatose on admission. Vision gradually got worse. In was, right eye - hand movements; and left eye - fingers at 2 feet; on the day after admission, i.e., 27/4/42. The fundi were normal. The following day the discs had blurred margins and there was oedema of the retina spreading down from the disc. The right pupil responded slightly to light and the left was dilated and inactive.

On 28/4/42 there was a central scotoma in both eyes, worse in the left eye. Vision was, right eye - hand movements; and left eye - perception of light. On 29/4/42, central scotoma was complete for 10 mm. white spots. Vision, right eye - hand movements, and left eye - fingers at 2 feet. On 30/4/42, vision, right eye - fingers at 2 feet, and left eye - fingers at 5 feet. On 2/5/42, vision, right eye - $\frac{4}{9}$ and left eye - $\frac{4}{18}$. When When seen a year later his vision was, right eye - $\frac{6}{60}$ and left eye - $\frac{6}{60}$ correctable to right eye - $\frac{6}{24}$ and left eye $\frac{6}{36}$. Pupils were equal but reacted sluggishly to light and briskly to near. His Wassermann was negative. The fundi showed some slight narrowing of the arteries, the preponderant ratio being A:V = 1:2. Some nipping

and bending of veins. Both discs showed bluish pallor and well marked cup. His intraocular tension was normal. No sheathing of vessels at the disc. Pallor was greatest temporally and the choroid was thinned and had a moth-eaten or nibbled appearance on the temporal side of the disc. Pigment was scattered round the temporal side of the disc. The left macula showed pigmentary change and the retina showed a curious patchy irregular reflex, best seen with the Mercury Vapour lamp. In the periphery of both fundi there was a fine pigmentary deposit with occasional white spots. He states that in 1939 he volunteered to serve in his third war and his eyesight was satisfactory, but they found out his age. He has tried several opticians for glasses since he left hospital last year. He cannot read any number on the Isihara plates.

His peripheral field is shown in Fig. 1, with red - green scotoma in hatching the right eye, and the only remaining island of red - green vision outlined in the left eye. In (Fig.2) the relative scotoma was recorded on the screen showing in the right eye central scotoma for $\frac{20}{1000}$ red and blue, breaking through to the nasal side; in the left eye (b) is the area of scotoma for $\frac{20}{1000}$ blue and (a) enclosed by interrupted line, is the remaining island

of vision for $\frac{20}{1000}$ red - green.

CASE III. Mr. B.M., aged 46. This patient was in hospital for a cement burn of the left eye in 1939. His vision then on dismissal was, right eye - $\frac{6}{9}$ and left eye - $\frac{6}{9}$. He had about an ounce and a half of methanol in a pint of water. He had pains in the abdomen and mistiness of vision. His vision was reduced to perception of light. He vomited shortly after drinking the spirit. On 28/4/42. Small corneal opacity, left eye, not interfering with vision. Both pupils active and fundi within normal limits. On 29/4/42, vision was, right eye - $\frac{6}{9}$, and left eye - $\frac{6}{60}$.

Central scotoma left eye. Right eye - Isihara normal; left eye - no letters seen entirely, upper part seen sometimes. Large scotoma in lower half of field. On 15/5/42, vision, right eye - $\frac{6}{9}$, and left eye - $\frac{1}{60}$. The left disc shows temporal pallor. There is a large scotoma to $\frac{5}{1000}$ white (Fig.3).

On 12/4/43, a year later, vision was, right eye - $\frac{6}{9}$, and left eye - $\frac{1}{60}$. Both pupils reacted normally. The right fundus was normal. The left disc was pale and cupped, the vessels showed no sheathing, and there was pigmentary deposit on the outer side with choroidal thinning similar to that seen in the preceding case. His

Discussion.

We have presented our case of blindness from quinine poisoning because the appearances of the fundus in the acute stage differ from the appearances in methanol poisoning, and because the late changes in the visual field resulting from damage by quinine are peripheral, contrasting so markedly with the central field defect of methanol poisoning. Our case showed a "milky" retina and "cherry" spot at the macula. Only several days later did the vessels begin to become narrow and the disc become pale. This milky fundus has been often described in the literature. In Ballantyne's case as we quoted above, the early fundal changes were insignificant. The inference is that the poison may act, producing blindness by changes invisible to the ophthalmoscope. Perhaps the opaque appearance of the retina so often seen in these cases is just a more profound cellular alteration of the retina by the poison. There is no coincident change of the nerve head. In the early days the disc colour is unaltered, the margin clearly defined, and the vessels unchanged. The opinion of most writers is that the retinal elements are poisoned directly by quinine, and not the nerve. We think that the early appearances of

the fundus support this conception. We find the question constantly recurring in the literature, Whether does the poison or toxin act on the nerve cells of the retina or nerve primarily, or does it act on the vessels to produce damage to the nerve elements by ischaemia? Ballantyne has emphasised for us, as we quoted above, the paradox of quinine blindness, namely, the fact that as the vision improved the vessels become narrow. Ischaemia has nothing to do with the initial blindness, though it may be the cause of the deterioration after recovery which occurs unfortunately in some cases.

Most cases of quinine blindness recover good central vision and any permanent loss is at the periphery of the field. There is often a coincident night blindness. From these facts Ballantyne further localises the action of the poison to the rods.

We should like to mention a few points of interest in our case before passing to the principal discussion of the action of methanol. This man did not recover well. The extent of the field recorded soon after vision began to return was very similar to the extent of the field recorded about six weeks later. The sides of the two and three isopter white fields were consistently steep. These are facts of poor prognostic significance.

The field was vertically compressed as Traquair describes, but was approximately 30°-35° horizontally from fixation, and 25° vertically. He had good central vision and a reasonable field. Yet he behaved like a blind man, finding his way very uncertainly about the hospital corridors. When we examined his field further on the screen it was apparent that the field was normal for only a few degrees around the fixation point. Outside this isopter the target disappeared and reappeared with great frequency. It presented multiple minute scotomata; we have called this the "sieve-like" field. We have not been able to find this type of field described in the literature.

We should now like to discuss the methanol poisoning group. Fundal changes are not always present in the early stages, but they are of a consistent nature. Haziness of the disc margins, engorgement of the veins, and in more marked cases, oedema of the retina spreading from the disc, is the picture presented in the literature. Of the four cases whose fields we discuss the first had slight changes in the disc and two others had changes in the disc and oedema of the retina. Nowhere in the literature have we found a description of oedema of the retina and a normal disc. The fundal changes centre

round the nerve head. The first case had changes in the disc and impaired vision. He recovered. The second was blind and had a bilateral central scotoma in the early days of recovery. He had marked fundal changes. A year later he had a relative central scotoma with impaired vision in both eyes. The third case had no record of fundal changes and a central scotoma in the left eye during the early days of recovery. A year later his right eye was normal and the left eye showed an absolute central scotoma with vision grossly reduced. All three had fields of normal peripheral extent.

The last case had marked bilateral fundal changes, with scotomata in both eyes in the early stages of recovery and a year later the visual acuity and field of vision were normal in one eye and in the other there was some nasal peripheral loss; indeed the inner isopter had an approximately hemianopic appearance, with a relative central colour scotoma for red and green. His visual acuity was a little impaired in this eye.

Some authors have found that the outlook was almost uniformly bad as far as useful central vision was concerned. Our follow-up series is very short, but as far as we see from a year's observation, and perhaps even this

is not long enough, the outlook is not altogether bad. Only one man has an absolute scotoma in one eye, the other being normal; another, a relative colour scotoma with peripheral loss in one eye, and the other eye normal; another, no defect that we could discover; and another, a bilateral relative scotoma. They all had a considerable amount of spirit, as can be seen from the case histories. As we mention above, however, the estimation of how much spirit was drunk is only approximate in most papers.

The ophthalmoscopic appearances in those four men were interesting. Where vision was impaired and the field affected, the disc was pale and cupped. Cupping was the more marked where the visual loss was greatest. The man with the complete scotoma had a very deep cup. His intraocular tension was normal. Where the nerve was profoundly affected, it was uniformly pale; where there was slight loss, the pallor was marked on the temporal side. It is interesting to note here that in the last case where the right eye had normal vision and field, and the left a relative colour scotoma with peripheral nasal loss, temporal pallor was quite marked in both, although more so in the left eye.

We appreciate that it is difficult to be dogmatic

always about temporal pallor, but we thought it was quite appreciable in this case. It interested us that it should be present in the normal eye. It called to mind those cases of retrobulbar neuritis in which the vision returns but the pallor remains. The facts of the late atrophy and cupping have often been described in these cases. The atrophy was simple; there was no glial proliferation.

Another point interested us in the examination of the discs. In the eye which had suffered impairment of vision or field, the choroid on the temporal side of the disc looked thin and moth-eaten or nibbled, with pigment scatter. This appearance was seen in every eye which was impaired but not in the normal eyes. Bearing in mind the frequent appearance of pigment at the ~~other~~ outer side of the disc as a simple structural variation without pathological significance, we thought that this was rather different. It is not a finding of great significance, and we have no histological control to interpret the appearance, but it was sufficiently characteristic for us to be on the lookout for it in cases of damage to the papillo-macular bundle. We have not found it described in the literature.

Most of the cases that we see of retrobulbar neuritis

are of an evanescent nature, tending to recover quickly. The chronic ones originating in diabetes, the anaemias, the acute infection, lactation, avitaminosis, and so on, while they are rarer, have admitted of histology. The papillo-macular bundle is affected while the rest of the nerve escapes and the defect is a central scotoma. As we said above when speaking of quinine blindness, the question arises as to whether this is produced by an affect on the blood vessels with thrombosis or ischaemia, or primarily on the neurone. The centre of the nerve is said to be less well supplied with blood vessels from the pial network than the periphery, and this is quoted as a factor in the frequent affection of the central bundle in disease. Alcohol is a vasodilator. Methol alcohol is very slowly eliminated. The vasodilator action is therefore prolonged. No appreciable change was noted in the retinal arteries.

Menne in his paper on the complete post-mortem examination of 22 cases of death from methanol poisoning speaks of the remarkably negative findings in the central nervous system, apart from oedema. He suggests that this is really not surprising when death occurs so soon after poisoning, and directs our attention to the experimental work in which animals have been poisoned over

periods up to a year and the central nervous system examined. He quotes Scott and his co-workers who conclude that vascular damage was slight, and parenchymal and neuronal tissue were affected by the poison. It may well be that the oedema which we see in the nerve head is the effect of the poison on the conducting neurone of the papillo-macular bundle.

The oedema which Menne describes is probably not inflammatory, but passive. The difficult point is the occurrence of simple atrophy without sheathing of the vessels. The common passive oedema of the disc is plerocephalic oedema and this is a much more sustained process than the comparatively transient oedema that we are considering. It is much more understandable that papilloedema should provoke glial formation because it is invariably present for weeks, or even months perhaps, before it is diagnosed.

However, when we come in conclusion to consider the histo-pathological findings in the cases who died of acute poisoning, there is a further possibility to be considered. By the use of routine staining methods and by examination of the retina in bulk, we were of the opinion that in our cases there were no changes characteristic of methanol poisoning. From this and collateral

studies with Lowenstein (yet unpublished, quoted above), of the ganglion cells of the retina in health and disease, we would not like to say that they are altered to any great extent, either in shape or in size, cytoplasmic structure, nuclear content, or structure, or in the lipoid change which they exhibit. We have examined many hundreds of ganglion cells in our cases of spirit poisoning and in normal retinas of most decades and we find no substantial difference between the normal retinal ganglion cells of the fourth decade and our methanol poisoning cases which were in the fourth decade. It may be that the thickening of the fibre layer described by other authors was due to the oedema that we see clinically. Where the vitreous is not injected with formol-saline soon after death the fibre layer tends to be fragmented and loose. Post-mortem autolysis in the retina is very rapid. We would like to say, as Menne said of his findings generally, that it would perhaps be unwise to put too much stress on negative histological findings in such an acute poisoning. Had the patients lived some weeks instead of some hours, we might be able to say histologically that the poison attacked either the ganglion cell, or the neurone in the nerve primarily. By that time the evidences of decay would be there for us to see.

The fact that the ganglion cells looked normal is no proof that their physiology was not impaired and that the impairment might later lead to death of the cell eventually, with an atrophy in continuity of the neurone in the nerve.

Bibliography.

BALLANTYNE, A.J.: B.J.O., p.153, vol.1, 1917.

ELDER, DUKE: Text Book of Ophth., vol.111, p.3032.

ELEONSKAJA: Russ. O.J., IV, 40, 1925.

HAGGARD, H.W., and GREENBERG, L.A.: Jour.of Pharmacol. and Exper.Therapeut., pp.479-496, vol.66, 1939.

HIRSCHBERG: Centralblatt f.prakt Augenheilk., seite 44, 1912.

JACKSON: Amer.J.Ophth., p.150, vol.111, 1920.

LOWENSTEIN, ARNOLD: Arch.of Ophth., pp.73-90, vol.27, 1942.

McCLUNG; Microscopical Methods, p.473, 1937.

McDONALD: XIII Internat.cong., Amsterdam, p.440, 1929.

MENNE, FRANK R.: Arch.of Path., pp 79-92, vol.26, 1938.

PICK und BIELSCHOWSKY: Klin.Wochen.Berl., seite 888, 1912.

ROST: Med.Klin., Berl., seite 129, 1912.

ROUSSY and LHERMITTE: quoted by Carleton's Histol.Tech., 1938.

SIMONS: Amer.J.Ophth., pp.446-451, vol 22 1942.

SMITH, HAROLD: T.O.S., p.310, vol.39, 1919.

SMITH and QUIGLEY: Amer.Jour.of Path. quoted McClung, 1937.

STADELMAN und MAGNUS LEWY: Med.Klin., Berl., seite 127, 1912.

SWANK and DAVENPORT: quoted by Carleton's Histol.Tech., 1938.

TRAQUAIR, H.M.: An Introduction to Clinical Perimetry, p.153, 1938.

TRAQUAIR, H.M.: Trans.Ophth.Soc., U.K., vol.50, p.351, 1930.

WOLFF, EUGENE: Personal Communication.

-------oOo-------