CEREAL FOOD POISONING AND ITS RELATION TO THE ETIOLOGY OF PELLAGRA.

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(With Plates III–V containing Figs. 1–18.)

Pellagra is a comparatively modern disease. The first account of it dates from 1735 when under the name *mal de la rosa* it was very fully described by Gaspar Casal who at that time practised at Oviedo and found it endemic among the Asturian peasantry. In Lombardy it was recognised about 1730 and only much later (1771) was systematically described by Frapolli as pellagra (rough skin), but Strambio, who was physician to a special hospital for pellagrins at Legnano, states (1784) that many of his patients told him that their fathers and grandfathers had suffered from the disease. Early in the nineteenth century it was comparatively common in Gascony and about 1836 it was beginning to be recognised in Roumania. In the United States sporadic cases seem to have been noticed in 1864, and since 1907 it has increased to an extent which has seriously disquieted the public health authorities. In Egypt, Turkey, Mexico, the Balkans, Brazil, the West Indies and other countries it is common and well known. Wherever it has appeared endemically it has followed on the introduction of maize as a field-crop and its adoption (by the rural population especially) as a chief and staple article of diet.

Maize is a cultivated grass, a native of America whence it was introduced into Spain about 1500 and it is now grown on a large scale in most countries where the climate and soil are suitable. How large a scale is evident from the following figures (for 1929–30). Taking the total area under cultivation of maize, wheat, oats and barley, maize accounts roughly for one-third of the whole in Hungary and the Argentine, one-fifth in Italy, rather under one-half in Roumania, Jugoslavia and the United States (where it is over one-third more than wheat), one-eighteenth in Russia, in Mexico six times more than wheat, and in South Africa four times more than these other cereals put together.

Already by certain of the earlier investigators pellagra was connected with the use of maize as an article of food and this view in one form or another has all along held its ground in spite of numerous dissentients and is strongly supported by its history which is the same for all the countries in which it is now endemic. Usually in about a generation after the introduction of maize into a district and after it has become largely grown and consumed sporadic cases of pellagra begin to appear and in a few years more the disease has assumed

much larger proportions, chiefly among the poorer agricultural and labouring classes. In Corfu in 1839 there was one case, in 1861 48 cases; in Northern Italy it was becoming common about 1730 and at the present day the number of pellagrins is estimated at 50,000 or more; in Roumania it made its appearance about 1833 and in 1898 there were 21,000 cases, in 1907 30,000, and they have gone on increasing; in the United States Roberts[1] estimates that from 1902 to 1920 there were 500,000 cases and 50,000 deaths; in South Africa three small outbreaks are recorded since 1906 and many sporadic cases. For Portugal, Egypt, Mexico and Turkey there are no available statistics but it is common enough in all. In Spain and Italy it is decreasing, but France is the only affected country in which it has disappeared; in 1844 there were some 3000 cases in the Landes and in 1880 none, the improvement being due to increased prosperity and a more varied and higher standard of food.

Pellagra is a chronic disease usually of many years' duration and characterised by gradually increasing physical and mental decadence. It may, however, be very acute or may be so mild as to be hardly noticeable and rarely there is no dermatitis. From the first the symptoms are referable to the nervous system and are motor, sensory and trophic. The early manifestations are a feeling of lassitude, weakness and incapacity for work, then vague nervous symptoms, stomatitis, salivation, gastro-intestinal disturbances and a skin rash. In progressive cases there follow signs of more marked involvement of the nervous system—pains in the head and body, optic nerve changes, vertigo, paresis (especially of the extensor muscles so that all the joints of the limbs tend to be semi-flexed), lessened or increased spinal reflexes, muscular tremors, tonic and clonic spasmodic attacks, serous diarrhoea, incontinence of bladder and bowel, and increase of the dermatitis. Many cases show marked evidence of mental decay ending in insanity, usually of a melancholic and suicidal type but sometimes maniacal. In Italy 4 per cent. of pellagrins require admission to asylums but the number of mentally affected is very much greater. In Egypt it has only been known for about 80 years but according to the Annual Reports of the Abassia Asylum "pellagra has now become the greatest cause of insanity in Egypt and of deaths among the insane," accounting for nearly one-sixth of the admissions and one-third of the deaths, while in the United States asylum cases are also numerous.

Slight and early cases recover readily if the diet is altered in time, a fact already noticed by Casal, but in all others the prognosis is absolutely bad.

The nervous system shows after death widely spread pathological changes which have been very thoroughly examined by many observers. There is scattered degeneration of nerve fibres in the spinal cord and peripheral nerves, and slight diffuse sclerosis in the system tracts of the cord and medulla. The cells of the cerebrum, cerebellum, pons, medulla, cord and posterior spinal ganglia all show chromatolysis, disappearance of Nissl's granules, and varying degrees of degeneration. The Betz cells of the cortex and Purkinje cells exhibit similar changes. The large viscera are more or less atrophied, pigmented and sclerotic, the muscles are wasted and the bones often thin and fragile. The skin is pigmented, rough and inflamed.

Although soon after the first appearance of the disease medical opinion had incriminated maize as its cause, an examination of the extensive literature reveals that at least twenty-five "theories" have been advanced to account for it. Among these are vitamin deficiency, fungus contamination, spoiled and infected maize, protein deficiency and infection, but none of them have so far stood the test of experimental and clinical criticism. In 1882 Antoniu[2] (Moldavia) settled the main question by feeding seven men on maize gruel

[1] *J. Amer. Med. Assoc.* (1920), p. 21.

[2] *Étiologie pellagreuse*, Borladu, 1882.

for a winter and all of them became pellagrins, while in 1920 Goldberger and Wheeler[1] (U.S.A.) kept eleven volunteers on a diet consisting largely of corn meal, wheat flour and potatoes and six of them developed slight symptoms of pellagra. The present investigation has been conducted on monkeys, rabbits, guinea-pigs and frogs with yellow and white maize, and shows:

I. That a maize diet can cause in monkeys characteristic nervous symptoms and death, and pathological changes in the nervous system identical with those found in pellagra.

II. That maize contains an acid which when administered to monkeys by the stomach or hypodermically as a sodium salt causes acutely the same symptoms and pathological changes as a maize diet.

III. That the acid is poisonous to other animals.

IV. That other cereals also contain the same or similarly acting acids and have actions on animals similar to those of maize.

Isolation of the acids.

In all the cereals examined the acids form insoluble lead salts and the problem is to obtain these as pure as possible and to purify further the acid after it has been detached from its combination with lead.

In the case of maize the ordinary meal may be employed but it is more convenient to use maize feeding-cake which consists of the residue of the whole grain after the starch (corn flour) and oil have been extracted. The cake was broken up and macerated in cold chloroform water for some days, the water expressed and the marc again macerated and expressed. The water was then filtered and boiled to precipitate albumen which is removed by filtration. The filtrate was concentrated *in vacuo* and precipitated by a saturated solution of lead acetate, the precipitate thoroughly washed with water, decomposed by H_2S and the lead sulphide filtered off. The filtrate was again concentrated by distillation *in vacuo* almost to a syrupy consistence and slowly added to a large excess of absolute alcohol when a white flocculent precipitate of acid was obtained mixed with impurities but free from lead and protein. This was washed with absolute alcohol, dried *in vacuo* in a desiccator, dissolved in water and neutralised with sodium bicarbonate. A varying amount of organic and inorganic impurities separates out and the filtrate from this after concentration *in vacuo* was slowly dropped into absolute alcohol which precipitates the sodium salt. On drying it forms a white tasteless powder, readily soluble in water and giving a clear neutral solution. Our experiments were made with this sodium salt. It is an active poison but was not chemically pure, containing inert inorganic bases (calcium and magnesium chiefly) and it did not crystallise.

In the case of the other grains the procedure followed differed somewhat in detail. After extracting with cold water and boiling to precipitate albumen the filtrate was further purified by precipitation with mercuric chloride, the precipitate filtered off and H_2S passed through the resulting filtrate to remove excess of mercury, the mercury sulphide being in turn removed by filtration. The filtrate was then concentrated at a gentle temperature and precipitated with a saturated solution of lead acetate which throws down the active acid and any other acids present which have insoluble lead salts. The lead precipitate after being thoroughly washed was decomposed by H_2S, filtered and the filtrate evaporated to a small bulk. It was then neutralised with sodium bicarbonate which throws out a mixture

[1] *Arch. Int. Med.* **25**, 450.

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of physiologically inactive inorganic and organic material and this was filtered off. The filtrate was next treated with a large quantity of absolute alcohol which precipitates the sodium salt of the active acid mixed with inorganic salts and colouring matter. When dried it forms a white powder or it may crystallise from a watery solution. The acid may be obtained from this in rosettes of pure needle-shaped crystals by dissolving in water, precipitating with lead acetate, decomposing the lead precipitate with H_2S and filtering. On evaporating the filtrate a thick syrupy liquid is obtained which on standing for some time usually yields a crop of crystals. The yield was always small and varied greatly in amount with different lots of the grains.

It was found that different samples of maize varied very greatly in toxicity and also that individual monkeys and rabbits exhibited very different degrees of susceptibility to maize feeding, complications which much prolonged our experimental work. When attempts were made to reinforce the daily ration by cooking it with water in which maize meal had been steeped the monkeys invariably refused to eat the porridge.

Experiment I.

A Bonnet monkey (1990 g.) was fed daily on 75 g. yellow maize meal made into porridge to which were added 10 g. butter, 60 c.c. milk, a little wheat bran and orange juice. It always got fresh fruit in addition. A week later it was somewhat less agile and in another week distinctly paretic, weight 1940 g. It became progressively more feeble and on the 30th day could only climb slowly and with difficulty, its muscles were tremulous on exertion, its back bent and its limbs flexed at the knees and elbows from weakness of the extensor muscles, weight 1760 g. Two days later its knees were so bent that it could only shuffle along on its hips with the support of its arms and its fingers were so flexed on the palms that it could not use them for grasping or climbing (Fig. 1). It was often drowsy, at other times mentally alert and ate all its food. By the 43rd day it was almost completely paralysed, its muscles were much wasted and its weight had fallen to 1505 g. (Fig. 2). It died on the 44th day from gradual paralysis.

There was marked loss of fat and wasting of the muscles. The large viscera were in appearance normal, and microscopic examination disclosed only a minor degree of congestion of the smaller blood vessels in the liver, spleen and kidney. The paracolic lymph glands were enlarged and hyperaemic and showed an enormous increase of the lymphocytes. As the changes in the nervous system were of the same nature with all the cereals investigated they will be considered together in a separate section.

Experiment II.

A *Rhesus* monkey was fed on 70 g. maize meal made into porridge, with 10 g. butter, 60 c.c. milk and a liberal supply of fresh fruit daily.

In 7 days the right arm was paretic and bent at the wrist and elbow but this passed off and it remained seemingly normal till the 22nd day when the arm again became very paretic. Otherwise it seemed well and active. Its

maize porridge was then cooked for 1 day with water in which 200 g. maize meal had been macerated. After this it was evidently more feeble. On the 34th day it was very feeble, sat with its back bent and its head down, its arms and legs semi-flexed at the joints, and when roused hardly able to climb. It had eaten well till now but had lost much flesh and a great deal of its hair. It gradually became more paralysed and died on the 36th day. There was much loss of fat and muscle, the large viscera were healthy and the nervous system showed widespread degeneration.

Experiment III.

A *Rhesus* monkey ate one full meal of maize porridge with butter and milk. Next morning it was found dead. On examination no gross lesion was discovered and it was evidently a case of acute maize poisoning. Similar acute poisoning has often occurred in farm animals with lathyrus and the bitter-vetch, the probable explanation being that the animal has had no opportunity to acquire tolerance and has been unable to detoxicate in its body the sudden dose of acid.

Experiment IV.

A *Rhesus* monkey fed for 41 days on a different lot of maize meal, with butter, milk and fruit, showed only slight weakness of its legs. Cold water extracts made from 700 and 1000 g. of the meal and given *per os* caused only slight general depression and some paresis. Several other lots of maize fed over longer periods gave somewhat similar results with other monkeys.

Experiment V.

A *Rhesus* monkey fed on fruit, bread, butter and milk was given by stomach tube on four successive days 1 g. of the sodium salt of the active acid dissolved in water. After the first dose it was very restless and uneasy for about 2 hours and scratched itself a great deal as if suffering from irritation of the skin. It then became listless and drowsy and remained so all day. Next morning it had apparently recovered. The same symptoms recurred after each dose but with more pronounced drowsiness and definite general paresis (Fig. 3). Owing to lack of material an interval of 11 days elapsed and by this time the animal appeared to be normal again. Doses of 2 g. were then given with an interval of 2 days between the first and second and of 3 days between the second and third. The first caused very marked paresis and depression which were still present 48 hours later when it received the second dose which greatly intensified the paralysis. Its joints were so flexed that it could hardly shuffle along, it could not use its hands to feed itself, and it sat with its back bent and its head down in a semi-comatose state. The third dose deepened the paralysis and coma and it died 26 hours later with all its limbs in marked flexion.

In this monkey there was acute poisoning and the post-mortem changes

were much more pronounced than in the prolonged feeding experiments with small daily doses. The protoplasm of the liver cells was granular and vacuolated, their nuclei stained poorly and in some areas there was complete loss of typical liver structure, the orderly columns of cells being replaced by irregular necrosed masses. In the malpighian areas of the spleen the lymphoid tissue was distinguishable only as a peripheral ring merging into a disorderly arrangement of strands of cells enclosing large irregular spaces. In the kidney there was congestion of the interstitial vessels, the cells of the convoluted tubules were slightly granular and in scattered areas their lumen was blocked with debris. The stomach and bowel showed no changes. On section the spinal cord and corpus striatum had a pinkish tinge due to distension of the small capillaries and minute extravasations of blood. Their nerve cells and those of the posterior root ganglia and sympathetic ganglia showed areas of pericellular oedema along with typical chromatolysis.

Experiment VI.

A *Rhesus* monkey fed on fruit, bread, butter and milk was given hypodermically 1 g. of the sodium salt. During the first hour it scratched itself restlessly as if the skin were itchy and then gradually became depressed and drowsy. The following morning there was some general paresis which lasted 3 days, passing off gradually. Seven days later 1 g. was again given, when the symptoms were the same as before but more marked (Fig. 4). It sat bent up with its head sunk on its chest and its movements were inco-ordinate. Next day it was generally paretic with flexion of its joints and took no food. It was killed by chloroform. Although the salt given in solution was neutral to litmus it had a very irritating action on the subcutaneous and underlying tissues. A phlegmon formed which, on the fifth day, became a clean cut ulcer with serosanguineous discharge, but did not appear to cause any pain. There was a patch of peritonitis underlying the ulcer.

The large viscera were healthy. In the cord certain of the nerve cells had lost their stellate shape and others showed early granular degeneration, there was chromatolysis in the nerve cells of the superior cervical ganglia and some diffuse myelin degeneration scattered throughout the medulla.

Rabbits. Numerous observations were made with rabbits fed on maize meal porridge and cabbage. Each animal was allowed 70 g. fresh cabbage daily and the maize porridge *ad libitum*. One half-grown rabbit (1035 g.) which was on this diet for 262 days grew uninterruptedly to full maturity (1895 g.), but none of the others throve well although they varied greatly in this respect. Some of them gained and others lost weight, but all of them shed their hair freely in large patches, looked in miserable condition and were weak on their legs. Fig. 5 shows an extreme effect in a rabbit after 70 days of maize feeding (with cabbage) compared with a control from the same litter. In some the legs became bent and X-ray examination showed the long bones to be mere shells, thinned and deprived of calcium salts (Fig. 6). One died on the 89th day in

clonic convulsions, another on the 101st day became very paretic, inco-ordinate and tremulous, some died apparently of exhaustion while others were alive after 7 months. If, during the experiment, wheat bran was substituted for the maize meal, the animal at once began to improve in health and appearance and to develop normally. Mature, fully-grown rabbits were not nearly so much affected as the immature.

On examining the nervous system by Marchi's method, little myelin degeneration of nerve fibres was found and the nerve cells of the cord and brain were apparently normal, but in some animals those of the root ganglia had undergone chromatolytic changes. Rabbits, however, are relatively insusceptible to the poisons of cereals or peas when these are given *per os* or hypodermically (but not per vein) and they thrive quite well when fed for long periods on lathyrus peas only.

All these animals received more than enough fresh cabbage to supply vitamins, and the explanation of their skin changes, malnutrition and nervous symptoms apparently lies in chronic intoxication by the acid present in the maize. The explanation of why one rabbit out of many should grow and thrive normally and why the others reacted so irregularly to the diet must lie in a varying power of detoxicating the acid.

When doses of 2 or 3 dg. of the sodium salt of the acid were injected into the ear vein the rabbit usually died almost at once but some of them survived much larger amounts.

The condition of the long bones in the rabbits which were kept on maize for some months is an interesting point. The shafts were thin, deprived largely of their inorganic constituents, very fragile and often bent. They differed from rickets in that the epiphyses were normal on X-ray and histological examination. The most probable explanation is that the prolonged daily ingestion of a strongly acid food such as maize disturbs the acid-base equilibrium and lessens the calcium concentration of the blood and to maintain these at their normal alkaline level (which is a necessity of existence) the large reserve store of alkali in the bones is gradually requisitioned and not replaced. As long ago as 1894 Weiske[1] observed that rabbits fed on oats lost mineral matter from their bones and this he attributed to the acid nature of their food and not to a deficiency of calcium, having also found that very dilute sulphuric acid fed to lambs had a similar effect. Mineral acids given *per os* to dogs and rabbits increase the output of urinary calcium and Bogert and Kirkpatrick[2] have shown that a diet of acid-forming foods has a similar effect. McCollum and others with young rats and Mellanby with young dogs in their investigations on rickets have used diets of maize and other cereals to prevent the normal deposition of inorganic salts in growing bone, their effects in this respect being attributed to the absence of vitamin D and not to chemical action of acid constituents in the food.

In our monkeys these bony changes were slight, possibly because the

[1] *J. Chem. Soc.* **66**, II, 286. [2] *J. Biol. Chem.* **54**, 375.

acuteness of the poisoning did not allow time for them to develop. They were, however, very marked in a young monkey which was fed chiefly on lathyrus peas for nearly two years.

Microscopically the skin showed keratosis, thickening of the epidermis due to proliferation of the deeper layers, degeneration of the cells lining the hair follicles and atrophy of the glands.

Guinea-pigs. (1) Guinea-pigs fed on maize meal porridge and fresh cabbage (as much of both as they cared to eat) throve and grew normally and one had healthy young. They differ, therefore, very markedly from rabbits on the same feeding.

(2) Four guinea-pigs about two-thirds grown were fed on maize meal porridge and orange juice. For 14 days they all gained weight, they then began to lose it somewhat rapidly, and after other 14 days two of them were just above and two just below their original weights, but all were in poor condition, depressed, inactive and with rough ill-nourished coats. Cabbage, 20 g. per day, improved their condition slightly and when it was given *ad libitum* they soon regained normal health and vitality.

(3) The preceding experiment was repeated with four other guinea-pigs of the same age, but vitamins A and D were supplied in the form of 1 c.c. cod-liver oil per animal per day, which was mixed with their maize porridge. They ate the mixture freely but the result was the same as in the preceding experiment, showing that the malnutrition was not due to lack of the cod-liver oil vitamins. When they were allowed a full supply of cabbage they ate less maize, and this may account for the improvement in their health and nutrition as less of the acid was ingested and more alkali furnished in their food.

Frogs. In frogs 2 or 3 cg. of the sodium salt given hypodermically caused depression or paralysis of the brain and spinal cord with fibrillary muscular twitchings lasting for 24 hours and gradually passing off to be succeeded by increased spinal reflexes. Larger doses were lethal, death usually occurring in a few hours from stoppage of the heart.

RICE.

Rice is the only other cereal the consumption of which is definitely known to cause pellagra and it has done so on a very insignificant scale. The evidence, however, is quite conclusive. In China, according to the *Tropical Diseases Bulletin*, some 12 cases have been reported in detail since 1919, all in persons of the poorer working class and whose staple diet was rice. Jefferys and Maxwell[1] state "All that can be said is that it exists as a definite disease seldom seen or else seldom recognised." In Japan, Takahashi (1929)[2] and others mention that over 70 cases have been reported, all in rice eaters. Sheppard[3] records that he met with cases in the Straits Settlements among

[1] *The Diseases of China*, 2nd ed. [2] *Japan J. of Dermat. and Urolog.* (1929), **29**, 65.
[3] *Brit. Med. J.* (1912), ii, 1773; (1908), ii, 1608.

the poverty-stricken class and that these people ate no maize. Stannus[1] has described an outbreak in Central Africa among men on a diet of very under-milled rice locally grown, and Viswalingam[2] one among Chinese coolies in the Malay States who were on a diet of polished rice, salted or dried fish, fat pork, sweet potatoes and some fresh leaves of plants. They ate no maize.

In making our experimental investigations on animals we were met with what at first seemed the insuperable difficulty of obtaining a poisonous rice. Monkeys were fed for 3 or 4 months at a time on samples of Patna, Rangoon, Siam and Carolina rice (all highly milled for the European market) with the result that they throve and remained in robust health. Watery extracts from the same rices given *per os* also proved innocuous, thus demonstrating that most samples of rice as met with in this country contain either no deleterious constituent or at most only a very small harmless amount. Finally, however, we happened on a Rangoon rice which was definitely poisonous and also on a much more active Carolina rice.

Experiment I.

A small *Rhesus* monkey to which was given *per os* a cold water extract of 2 kg. Rangoon rice was noticed next day to be slightly weak in the loins and legs and at the end of a fortnight the weakness was more pronounced, but it was otherwise well and active.

It was then given *per os* a water extract of 200 g. Carolina rice of the best quality, highly milled and having the pericarp and aleurone layer completely removed. In an hour there was increased weakness of the legs and this continued throughout the day. A similar dose was given daily for the next 4 days and very decided general muscular weakness developed. The legs, arms and loins were especially affected, the fingers were so flexed on the palms that its hands were almost useless, its legs were bent at all the joints and it either shuffled along on its hips or using its arms as support swung its body forward as if on crutches. If it attempted to jump it landed heavily in a heap. It was lethargic and drowsy. It was then given at short intervals extracts from 400, 800, 1000 and 2000 g. of the same rice and became extremely feeble and debilitated, only able to shuffle about. It was not affected in proportion to what one would have expected from the increased dosage, and hence it is probable that a tolerance to the poison is established or that it becomes in some way partially detoxicated in the body. It ate well all the time and did not lose weight. As it did not die it was killed by chloroform on the 30th day after starting the Carolina rice extracts and its organs were examined at once. The viscera were all healthy and so to outward appearance was the nervous system. Microscopic examination showed no myelin degeneration with Marchi's stain, but there were marked degenerative changes in the cells of the spinal cord and root ganglia.

[1] *Trans. Soc. Trop. Med.* (1913–14), **7**, 47; *Trans. Roy. Soc. Med.* (1929–30), **23**, 740.

[2] *J. Trop. Med. and Hyg.* (1918), **21**, 153; (1920), **23**, 46.

Chemical examination of many samples of rice was usually successful in isolating very small amounts of an acid which proved to be poisonous when tested on frogs. When 2 or 3 cg. neutralised with sodium bicarbonate were given hypodermically the animal in a few minutes became more or less deeply paralysed from an action on the brain and spinal cord and, if it survived, this was succeeded by a period of greatly increased reflexes.

We have been unable so far to obtain from the rice at our disposal a sufficient amount of the active acid to conduct a satisfactory investigation of its action on monkeys and rabbits. The foregoing experiments, however, seem to furnish conclusive proof that rice contains an acid having an action on animals similar to that of the acid in maize, while the clinical records show that, like maize, rice is capable of producing the disease pellagra in man. There is, however, this important practical difference that whereas maize often contains a relatively large amount of acid and if consumed in sufficient quantity is certain to produce pellagra, rice on the other hand only very exceptionally does so and this owing to the relatively and usually very small quantity of the poison present in it. It is not a question of vitamins or deficiency, but a question of an active substance present in the grain and which is capable of causing acute poisoning. The monkey in our experiment was being fed on an abundance of fruit with bread, milk and butter and got the rice as extracts in single doses and not continuously over a long period as food.

OTHER CEREALS HAVE A POISONOUS ACTION ON ANIMALS SIMILAR TO THAT OF MAIZE.

An argument which has been much insisted on as telling against the etiological importance of maize as *the* factor in causing pellagra is that the disease has frequently occurred in persons who had eaten little or no maize and in countries and institutions where it is sparingly used as a regular article of food. It is unnecessary to labour this point as the matter is discussed and instances cited in many contributions to the subject of pellagra, but we would like to refer very shortly to certain cases which have arisen in Great Britain. From 1913 to 1924, 54 cases occurred in Rainhill Asylum (Liverpool)[1] of which 47 ended fatally, and previously and since a number of sporadic cases have been reported in which maize could be definitely excluded as the causative factor. But unfortunately in all these little or no detailed account is given of the dietary as regards other cereals nor of the consumption of proteins, milk and vegetables. As a consequence we have been able to pick out from the fairly numerous case reports only three in which other cereals can be definitely presumed as the cause. These are:

(1) A woman, 21, who lived largely on bread and tea and who died of pellagra[2].

[1] Watson, *Seventh Annual Report of the Board of Control*, for 1921, and *Eleventh Annual Report*, for 1924.

[2] Brown and Low, *Edin. Med. J.* (1909), **3**, 197.

(2) A woman, 40, who since an early age had been in the habit of eating raw oatmeal, became mental and died of pellagra[1].

(3) A woman, 50, who for 2 years lived chiefly on bread and butter and tea, recovered[2].

The following experimental investigation of oats, rye and wheat may suffice to explain such cases, and also why a diet consisting chiefly of cereals or in which maize has been replaced by another cereal may not satisfactorily cure or abolish pellagra. It has often done so, but that is because these cereals are usually much less deleterious than maize and are sometimes, as we repeatedly found, practically innocuous. It may also provide a different interpretation of some of the results of the numerous laboratory experiments on feeding animals with cereals, results which have usually been attributed to a deficiency of vitamins or inorganic salts.

Rye.

Two separate lots of rye grain (free from ergot and other grains) were fully investigated. One of these was innocuous and quantities as large as 12 kg. yielded merely a very small amount of a poisonous acid. The other proved to be poisonous by feeding to a monkey and from it a certain amount of the sodium salt of the acid was obtained crystalline.

The action on animals of rye grain and its active acid suggests very strongly that the "spasmodic" or "nervous" form of "ergotism" is not due to any poison formed in the grain by the *Claviceps purpurea* but on the contrary is due to a poison in the grain itself and results from an excessive consumption of rye bread. This opinion is strengthened if one compares the pictures[3] of human cases of spasmodic ergotism with those of monkeys, pigeons, hens and ducks subjected to feeding experiments with other cereals and various kinds of peas[4]. With all of them, when small doses of the poison are ingested daily in the food over long periods, spasms, flexor contractions and paresis are the prominent symptoms, whereas when a large dose of acid is given at once the paralysing action is the more prominent. In man the onset and course of the clinical nervous symptoms in convulsive ergotism, even to the occurrence of insanity, bear a close resemblance to those of pellagra[5] and suggest that the cause is fundamentally similar in both.

The rye of one harvest and in one district may be innocuous in the quantity ordinarily consumed and usually is so, while in another year or in another place it may develop an unusual quantity of its active substance and become poisonous. But always previous to an epidemic there has been local failure

[1] Sambon and Chalmers, *Brit. Med. J.* (1912), ii, 1093.

[2] Henderson, *Rev. of Neurol.* (1916), **13**, 579.

[3] Heusinger (1856) *Studien über den Ergotismus*, Marburg. Thieme (1930), *Veröffentl. a. d. Geb. d. Medicinalverwaltung*, **32**, 5.

[4] Stockman (1917), *Edin. Med. J.* (Lathyrus peas); (1931), *J. Hygiene*, **37** (various peas); Anderson, Howard and Simonsen (1924–5), *Ind. J. Med. Res.* **12** (*Vicia angustifolia*).

[5] *See* Thieme, *loc. cit.*

of crops with resulting scarcity of food or actual famine and in consequence the cheap rye bread has come to take an unduly large place in the daily dietary. The same conditions have been seen and noted time and again in connection with outbreaks of lathyrism.

Experiment I.

A *Rhesus* monkey was fed on 100 g. rye grain (grown in Scotland) coarsely ground and made into porridge, *plus* 10 g. butter, 60 c.c. milk and some fruit daily. For 50 days there was no very perceptible change except some paresis of the left arm. To increase the dose 200 g. of rye were soaked in water and the water used to make the porridge. It ate the first lot and was slightly affected, but in spite of several endeavours refused to eat any more made in this way. It was kept therefore on its usual ration, and on the 65th day the left arm was very distinctly weak, flexed at the wrist and elbow and useless. From now on the paresis spread to its other limbs but varying greatly in degree from day to day (Fig. 7). Sometimes the monkey was fairly active, at other times it was unable to jump, could only climb laboriously, could not use its hands properly and walked with its hips and knees flexed. On the 93rd day it suddenly became more paralysed and next day was much worse, hardly able to walk and sat huddled up or lay on its side (Fig. 8). It recovered to a slight extent but all its joints remained flexed, its muscles very weak and limp and at times it had coarse muscular twitchings and tremors. As it had become very thin and showed no tendency to recover, it was chloroformed on the 102nd day.

There were granular changes in the liver cells and the liver, spleen and kidney were congested with deposits of blood pigment in the capillaries. The nervous system showed the usual degenerations.

Experiment II.

A large *Rhesus* monkey was given hypodermically 3 g. of the sodium salt. In 20 minutes it was much quieter and looked very drowsy. In an hour this had increased and it was given a further 2·5 g. During the next hour it became paretic and lay down frequently on its side, gradually becoming worse (Fig. 9). For the rest of the day it was hardly able to walk, it moved its limbs stiffly, propped itself up with its arms or the wall when sitting, and was very indifferent and depressed. For the next two days it moved about very feebly and then gradually began to recover. The injections caused very considerable local irritation.

Experiment III.

A rabbit which got hypodermically 1 g. of the sodium salt showed no definite effects.

Frogs. With 0·035 g. of the sodium salt given hypodermically a large frog lay as if dead in 10 minutes. Next day it was very paretic but with some

increase of its spinal reflexes, and this lasted 4 days, the paresis gradually lessening and the reflexes increasing. It was then killed and the muscles at the place of injection did not react to the faradic current. Doses of 1 to 3 cg. of the acid (neutralised with sodium bicarbonate) cause depression of the brain and spinal cord followed or accompanied by increased reflexes. Several frogs had violent tetanus. With larger doses the heart stopped in diastole but only after some hours.

Wheat.

Only one lot of wheat was examined. It was grown in Scotland and proved to be highly poisonous. From 15 kg. there was obtained 5·5 g. of the sodium salt and smaller quantities yielded somewhat similar proportionate amounts. Hart, Miller and McCollum[1] had previously found that pigs fed chiefly on wheat grain for several months developed paralytic symptoms due to changes in the spinal cord, and they attributed the results partly to a toxicity of the wheat and partly to a deficient supply of inorganic salts. The explanation, however, lies in the presence of an acid having an action similar to the acid found in other cereals and peas and which can be extracted by similar chemical methods.

Experiment I.

A *Rhesus* monkey was fed on 120 g. wheat grains cooked whole by steaming *plus* 10 g. butter, 60 c.c. milk and fruit daily. In 12 days the left arm was paretic, held up flexed at the elbow and adducted, and it sat at times with its back much bent and its head on its chest. After 30 days' feeding it was much in the same condition, some days apparently well, other days very distinctly affected. The experiment was then stopped and the animal recovered completely in a few days.

Experiment II.

A *Rhesus* monkey was fed for 10 days on the same diet as above, when it showed signs of being very slightly affected. It was then given by stomach tube a cold water extract of 400 g. ground wheat. In an hour it had gradually become very apathetic and drowsy, sat with its head down between its legs and was difficult to rouse (Fig. 10). It became more helpless and collapsed and died suddenly 6 hours after the administration. It was examined at once and most of the wheat extract was found to have left the stomach.

This monkey evidently failed to detoxicate the poison, and was poisoned acutely by a massive dose. The microscopic examination of its nervous system and viscera revealed general destructive changes which may fairly be described as a "pathological explosion." The liver, spleen and kidney were intensely congested and showed widespread cell damage. The lungs were crepitant and showed no congestion. In the brain and spinal cord the pial vessels were engorged, there were numerous minute haemorrhages into the nerve tissue,

[1] *J. Biol. Chem.* (1916), **25**, 239.

and there was widespread cellular damage throughout. There was some hyperaemia of the lesser curvature of the stomach but no special signs of irritation.

Experiment III.

A small *Rhesus* monkey was given hypodermically 2 g. of the sodium salt of the active acid. In 5 minutes it appeared depressed and began to lie down at short intervals. After an hour it was either sitting with bent back supporting itself by the help of its arms, or lying down and very paretic (Fig. 11). It remained so all day, but in addition developed very marked flexure of the fingers and wrists so that it could not use its hands and walked on the back of its wrist joints. Its gait was stiff and very slow, with an evident want of co-ordination of the flexor and extensor muscles. Next day it had recovered to a very great extent and on the third day seemed normally active.

Experiment IV.

A rabbit (1155 g.) received hypodermically 1 g. of the sodium salt. In 15 minutes it had a tonic spasm of the hind legs and for the rest of the day all four legs remained very weak and it sat quiet, very depressed and unwilling to move. Next day it had quite recovered.

Experiment V.

Frogs. Doses of 1 to 3 cg. of the acid neutralised with sodium bicarbonate and given hypodermically caused more or less paralysis of the brain and cord, with succeeding marked increase of reflexes lasting for some days if the animal remained alive.

OATS.

Compared with other cereals oats contain a large amount of acids. The sodium salt of its poisonous acid is neutral but, when injected subcutaneously dissolved in water, it causes very great local irritation and marked lesions in the nervous system. Prolonged feeding also affects the nervous system and, in addition, deprives the bones of their inorganic salts, its action in this latter respect being more virulent, according to Mellanby, than that of other cereals and due to the absence of vitamin D. He also observed that his experimental dogs developed degenerative changes in the spinal cord which he states could be prevented by giving vitamin A (butter or cabbage) but not by giving vitamin D[1]. In our feeding experiments the monkeys got relatively large amounts of butter, milk and fresh fruit in addition to the cereal, and yet they showed the characteristic symptoms of nerve degeneration. Further, administration of the active acid hypodermically produced the same nerve lesions as feeding, and here there could be no question of a dietetic or vitamin deficiency.

In this country there can be very few individuals, if any, who consume oatmeal in such quantity as to be a menace to health or a cause of rickets,

[1] *Brain* (1931), **54**, 347.

and the age-long experience of mankind all over the world conclusively demonstrates that cereals can and do furnish a large part, and very often the larger part, of the food of normally healthy and robust populations. Gross over-consumption of cereals and peas, especially if combined with a very limited variety and supply of other foods, is however a very different matter, and is undoubtedly capable of causing poor health and organic disease. These ill-effects fall mainly, but not solely, on members of the poverty-stricken classes in certain countries, whose food consists almost exclusively of maize or rice consumed in such quantity that their acids cannot be dealt with and rendered harmless to the body. Even then it is only a small minority who suffer in consequence from pronounced diseases such as beri-beri or pellagra.

Experiment I.

A *Rhesus* monkey was fed on 100 g. Scotch oats coarsely ground and made into porridge *plus* 60 c.c. milk, 10 g. butter and a liberal supply of fruit. During the first 45 days little effect was noticeable, but sometimes for a day or two its arms were slightly paretic and its joints semi-flexed. Its oats were then cooked for 5 days with water in which oatmeal had been soaked, but it then refused to eat more than a small amount and remained fairly active although definitely less agile. On the 58th day it was given hypodermically 0·25 g. of the active acid (neutralised with sodium bicarbonate) and in a few minutes became very paretic, and either supported itself by leaning against the wall or lay down (Figs. 12, 13), but if roused could run about and even jump, although rather inco-ordinately. For some days the paresis continued, its joints were flexed, there were fine muscular tremors on exertion, and it climbed and jumped poorly. It never came back quite to normal, and it was evident that its nervous system had been slightly damaged. A phlegmon formed at the area of the subcutaneous injection, followed by necrobiosis of the skin with no suppuration. It healed satisfactorily. On the 92nd day it was given hypodermically 1 g. of the active acid as a sodium salt, and in 3 minutes lay down and became very helpless, somnolent and drowsy, with muscular twitchings at intervals. During the next 4 days it was semi-paralysed with all its joints flexed and, as it showed no signs of recovery, it was killed by chloroform. There was much local irritation round the area of the subcutaneous injection with some necrosis. The viscera were healthy, and microscopic examination of the nervous system showed the same degenerative changes as with other cereals.

Experiment II.

A *Rhesus* monkey was fed on the same diet as the preceding. In 66 days it was definitely paretic, its hands were weak and flexed and its knees and back bent, but it was fairly active and quite alert. It was then put on a diet of bread, butter, milk and a large amount of fruit, and in 3 weeks had quite recovered except for a slight degree of wrist-drop.

It was then given hypodermically 1·4 g. of the active acid (not well purified), neutralised with sodium bicarbonate. During the first day it was very paretic and lay down a great deal, and on the second day was much worse. There was much local irritation round the injection and it was killed with chloroform. The nervous system showed the usual degenerative changes.

Experiment III.

Frogs. Doses of 1 to 3 cg. neutralised with sodium bicarbonate and given hypodermically caused more or less marked paralysis of the brain and spinal cord lasting for some hours to a day, and followed by increased reflexes for several days if the animal survived.

When 5 cg. were given the frog in a few minutes lay as if dead, but it sometimes recovered and had its reflexes increased almost to tetanus. The muscles at the place of injection were inexcitable to the faradic current, but there was not the same violent local irritation as in monkeys.

PATHOLOGY.

The pathological changes in the nervous system resulting from poisoning by maize and other cereals or by their toxic constituents were fully investigated in the monkeys, and were found to be always the same in kind, namely, a widely-spread degeneration of nerve fibres and injury or destruction of nerve cells in both the cerebro-spinal and sympathetic systems. In the nerve fibres the myelin sheath is the structure attacked. The earliest changes are not detectable by our present methods of investigation and probably may give rise only to functional disorder, but when the process is well advanced the myelin breaks up and collects into droplets, which show up black with Marchi's stain. Many of the nerve cells remain apparently unaffected, while others are more or less damaged. The axis cylinders showed no change and this may account for the rapid recoveries sometimes seen in our experimental monkeys. These appearances are not peculiar to maize or pellagra but constitute the reaction of the nervous system to many and very different kinds of poison, and if the damage has not been too severe the nerve fibres regain their normal structure, sometimes in a comparatively short time.

The nerve fibres.

Although the degeneration of the nerve fibres can be traced from the peripheral nerves up to the cerebral cortex it is not confined to any anatomical division of the spinal cord or brain stem but is scattered very irregularly and the number of affected fibres varies greatly. When few they appear isolated amid areas of healthy tissue, or at places they may greatly outnumber the normally myelinated fibres (Fig. 14). The fibres in the cauda equina (Fig. 15), the cervical and lumbar enlargements and the posterior columns of the cord are invariably most damaged. The lateral areas, although affected, seldom

show much change except at the periphery. The degeneration may be traced from the peripheral nerve trunks via the nerve roots into the grey matter of the spinal cord.

On the *sensory side* the afferent and efferent fibres of the posterior root ganglia are degenerated in proportion to the involvement of the posterior columns of the cord, the point of entry of the roots into the posterior cornua being most markedly affected (Fig. 14). Strands of medullated fibres passing from the region of the posterior columns into the grey matter of the posterior horns, the motor nerve roots, and the fibres issuing from the anterior cornua are similarly involved. The ground bundles adjacent to the grey matter show no visible change. The ascending spino-cerebellar areas (Flechsig and Gower), the spino-thalamic and the spino-tectal tracts are also generally damaged, but the descending bundles not to any extent.

In the *medulla oblongata* degeneration occurred in the region of the restiform body, the mesial fillet and the peripheral areas. In one case the fibres of the accessory and hypoglossal nerves took on the Marchi staining very deeply.

In the *mesencephalon* there was slight involvement of the crus and the antero-lateral region but more in the mesial area of the fillet. In one monkey (maize) the oculo-motor fibres were degenerated (Fig. 16). In the *cerebrum* degenerated fibres were fairly numerous, running in definite strands in the internal capsule.

The amount of degeneration in the white fibres of the *cerebellum* varied a good deal.

In the *peripheral nerves* examination of the sciatic, internal popliteal and musculo-spiral revealed more or less destruction of the myelin sheath which was broken up and collected into droplets.

The nerve cells.

The cells of the *posterior root ganglia* were involved in chromatolytic changes which varied greatly in degree and intensity. The earliest stage consists in swelling with disappearance of the chromophile (Nissl's) granules from an outlying portion of the cell and the perinuclear region, and ultimately the granules are no longer distinguishable, the cell groundwork staining faintly and appearing as if studded with fine dust-like material. As the degeneration spreads towards the centre the nucleus becomes displaced to one side and in extreme cases lies at the periphery as if being extruded (Fig. 17). The final stage appears to be one of removal of the cell débris by phagocytes as in some preparations the remains of the disintegrated cells were seen in the midst of thick clusters of other cells derived by proliferation from the surrounding neuroglia (Fig. 17).

A relatively acute phase was seen when the toxic acids (as sodium salts) were given hypodermically or by the mouth. This was characterised by cell

death, evidenced either by swelling or shrinkage of the cell, chromatolysis, and nuclear disintegration.

The superior cervical, stellate and lumbar *sympathetic ganglia* were examined. In the first two chromatolysis and eccentricity of the nucleus were well marked with occasional vacuolation. The lumbar ganglia were not much affected.

In one (oats) monkey the superior cervical and stellate ganglia showed a marked increase of neuroglia cells with a suggestion of commencing fibrosis. *The nerve cells of the grey matter of the spinal cord* were deeply affected. In acute cases some of the large multipolar cells of the anterior horn showed partial or complete chromatolysis with displacement and blurring of the nucleus as if it were being extruded (Fig. 18), the lumbar and to a less extent the cervical enlargements being the sites of greatest damage. In the feeding experiments where the paralysis was of gradual onset a series of changes occurred analogous to those seen in the root ganglion cells. The column of Clarke was always widely implicated, the appearances being striking and constant and first evidenced in the perinuclear region as a pale annular area surrounded by fragmentary granules. All gradations of degeneration are met with, the extreme being a swollen pale-staining cell with an ill-defined displaced nucleus and vacuolated cytoplasm. In *the brain* similar degenerative changes were found in the medullary and thalamic nuclei. In the acutely poisoned animals the pyramidal cells of the motor cortex were involved. The Purkinje cells of the cerebellum in the cases examined were apparently intact.

In a previous paper by one of us on poisoning by leguminous seeds it was stated that a detailed description of the pathological changes found in monkeys after poisoning by lathyrus, peas and lentils would be published later. A separate publication is, however, rendered unnecessary by the fact that the pathological findings in these cases have proved to be similar to those above detailed produced by cereals.

COMMENTARY.

All the cereal grains examined proved to be poisonous to monkeys when they constituted the bulk of their food even when relatively large amounts of fruit, milk and butter were consumed along with them. So far as tested they were also poisonous to rabbits and guinea-pigs.

Acids isolated from the various grains and given as sodium salts hypodermically and by the mouth to animals on their ordinary diet and in good health were also poisonous and produced similar symptoms to feeding.

The pathological changes were the same in both cases and implicate chiefly the nervous system, but the bones and viscera were also more or less involved. The pathological changes account fully for the various symptoms.

The poisonous substance is in all an acid or acids, but possibly not the same in all. The most probable explanation of their action is that they are

not only irritant and poisonous in themselves, but that they have a secondary effect of gradually or suddenly withdrawing alkalis and especially calcium from the blood and tissues.

It is not, therefore, a question of vitamins or of any kind of deficiency in the grains, for all the animals were getting vitamin-containing foods in abundance. The larger the quantity of cereal food consumed and the larger the dose of acid given by stomach or hypodermically, the more severe were the symptoms—facts which militate very strongly against any idea of a deficiency.

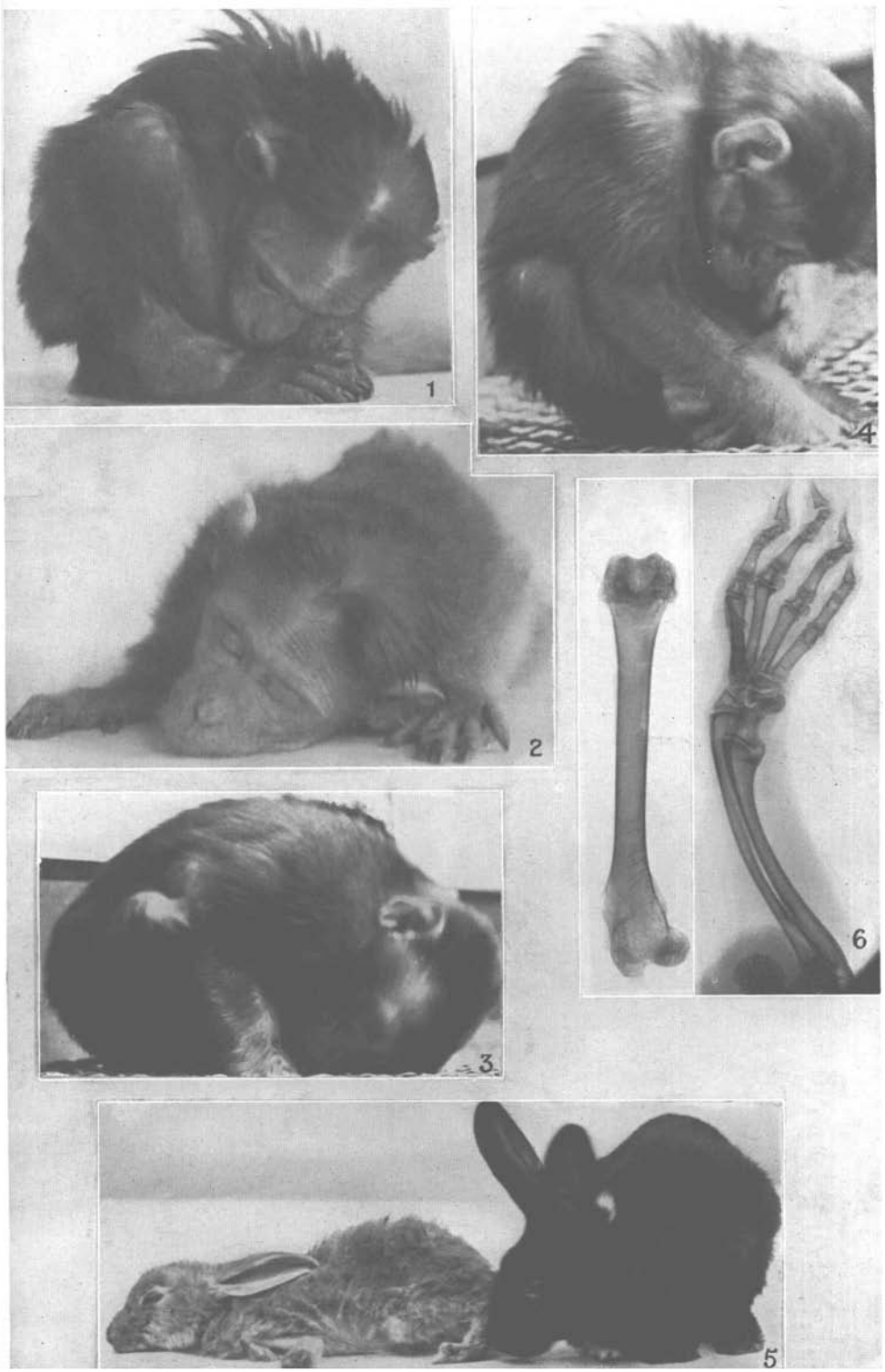
As regards pellagra there is evidence that it is occasionally caused by cereals and diets other than maize, and therefore there must be a factor common to them all. This, we think, is explained by the presence of a positive poison and by their acidity, with consequent injury to the nervous and other tissues and disturbance of the calcium and alkali balance in the body. Practically, however, pellagra exists as a maize disease with its symptoms dependent on functional disturbances and organic lesions of the nervous system. The prominent skin lesion is probably due to a tropho-neurosis which renders the skin unduly sensitive to any irritant such as sun rays, or merely to pressure as it occurs in persons lying in bed and not exposed to the sun. Our monkeys and rabbits reproduced, probably as nearly as they can be reproduced in animals, the symptoms and lesions of pellagra, and the pathological findings in the nervous system were identical with those described by numerous investigators as occurring in pellagra. The rabbits showed very marked skin lesions and thinning of bones.

The incidence of pellagra in maize eaters is essentially a question of the quantity eaten. It occurs only when maize bulks preponderatingly in the dietary and it has been demonstrated repeatedly and convincingly that the disease can be abolished or prevented by lessening the quantity of maize and substituting other articles of food. This is certainly the best method of prevention but unfortunately is not always practicable with a poverty-stricken and backward population often living on the verge of starvation and with no other food to turn to.

EXPLANATION OF PLATES III–V.

PLATE III.

Fig. 1. Maize: feeding 25th day.
Fig. 2. Maize: feeding 43rd day.
Fig. 3. Maize: sodium salt *per os* 2nd day.
Fig. 4. Maize: sodium salt hypodermically.
Fig. 5. Maize: after 70 days' feeding and control.
Fig. 6. Maize: bones of rabbit thinned and bent.



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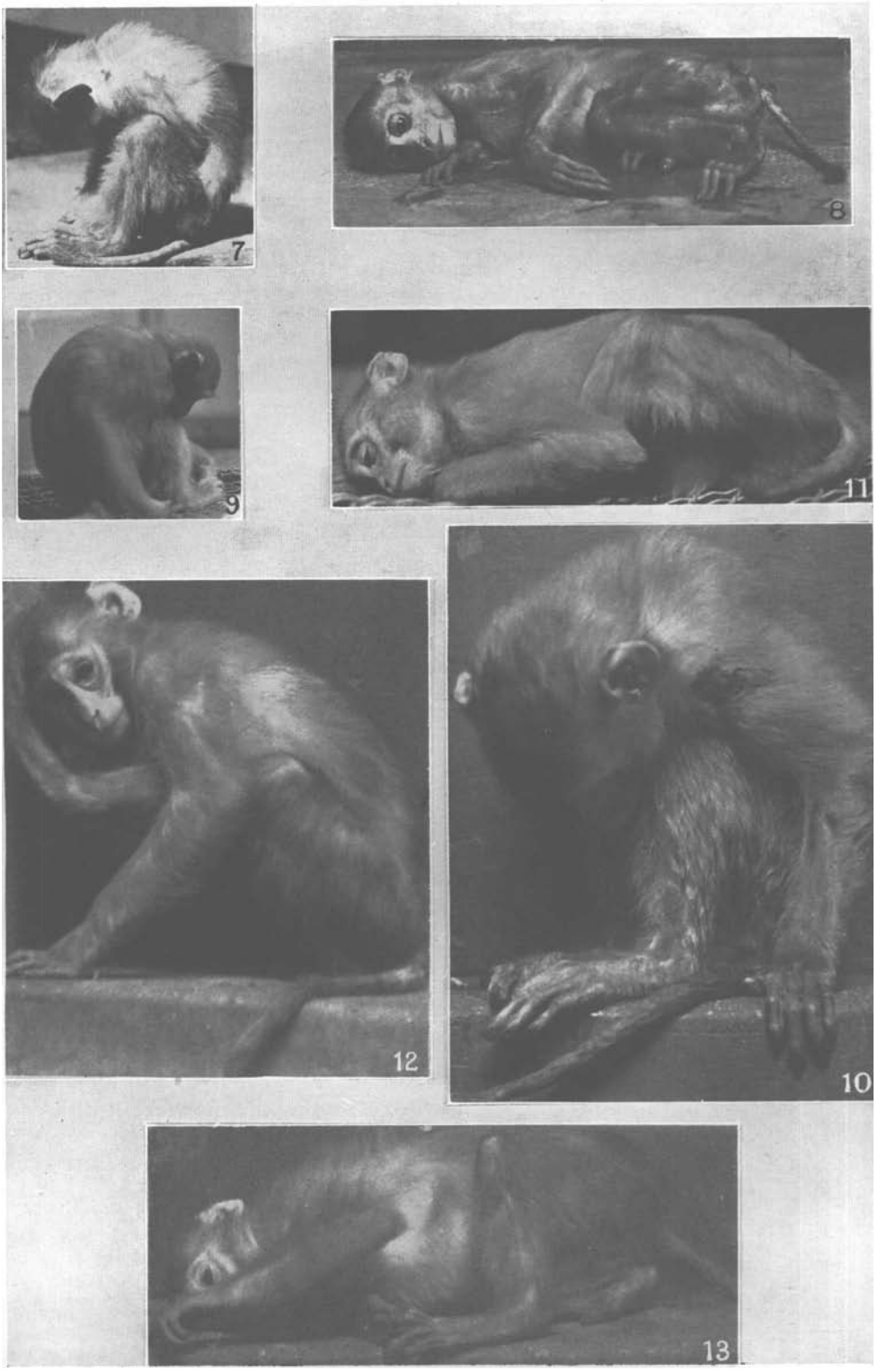

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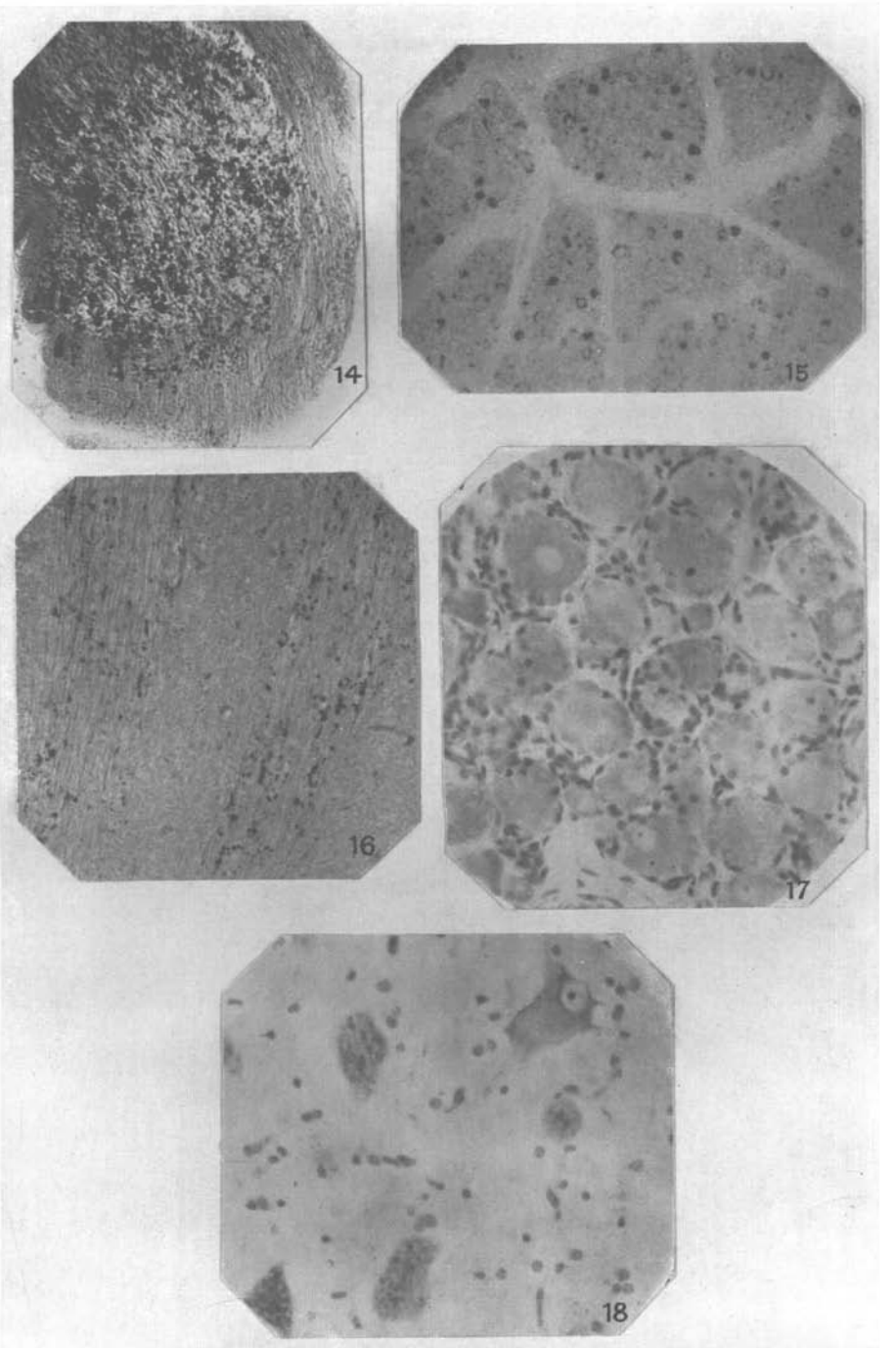

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PLATE IV.

Fig. 7. Rye: feeding.
Fig. 8. Rye: feeding 94th day.
Fig. 9. Rye: sodium salt hypodermically.
Fig. 10. Wheat: extract *per os* (acute poisoning).
Fig. 11. Wheat: sodium salt hypodermically.
Fig. 12. Oats: sodium salt hypodermically.
Fig. 13. Oats: sodium salt hypodermically.

PLATE V.

Fig. 14. Entrance of posterior root to spinal cord (maize).
Fig. 15. Cauda equina (oats) low power.
Fig. 16. Oculo-motor fibres in brain (maize).
Fig. 17. Posterior root ganglion (maize sodium salt).
Fig. 18. Cells of anterior horn of spinal cord (maize) shows displacement of nucleus and chromatolysis.

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