

**Milk.**—Milks were titrated against a standard penicillin on a large double layer blood agar plate using a Group A streptococcus as test organism. The plate takes 48 tests in holes punched through the upper layer. The method is sensitive down to 0.07 unit and with care it will detect as little as 0.031 unit. The plate is easy to set up and a large number of estimations can be done quickly.

**Lady Florey:** Penicillin concentrations in serum after a single intramuscular injection in the cow as shown by Dr. Slavin follow much the same curve as has been found in man. A sharp initial fall is followed by a flattening indicating probably, that there is an optimum concentration in the blood-stream below which excretion is minimal. Relatively large doses producing initial high blood concentrations do not therefore produce correspondingly long periods of inhibition.

Two essentials for success in treatment are the full access of the drug to all infected parts and removal of all dead tissue. Local treatment through the udder may sometimes fail when all infected loculi are not reached by this route. Intramuscular treatment may then succeed, for by this means every part of the body is reached to which the circulation has access. Again a small piece of slough infected with *Staph. aureus* is sufficient to prevent complete sterilization of a wound even by extremely high concentrations of penicillin.

The "Drop in slide" method of Dr. N. G. Heatley is less tedious than the slide cell method of Wright for penicillin estimation in serum.

**Mr. J. Francis** asked whether milking was continued normally when two injections of penicillin were given on consecutive days and whether there was much difference in the susceptibility of different strains of *Str. agalactiæ* to penicillin. With regard to the treatment of *C. pyogenes* mastitis he pointed out that the failure of penicillin in this condition was in accord with experimental results with *C. diphtheriæ* infection, in which it was known that although penicillin exerted a striking effect on an acute bacteræmic infection it had little or no effect on a chronic local infection. He had found that penicillin had a marked effect on acute *C. pyogenes* infections in mice, but, like others, he had found it ineffective in the treatment of established *C. pyogenes* mastitis.

**Mr. K. D. Downham** stated that he had treated 50 cows suffering from streptococcal mastitis and that 75% of these animals had been freed from infection, as shown by four subsequent bacteriological examinations of milk samples. He had had no success in the treatment of mastitis due to *C. pyogenes* in dry cows. He described a cow which had suffered from mastitis due to infection with *Str. agalactiæ*, whose milk had contained these organisms for eighteen months, and which failed to respond to routine udder infusion with penicillin, but was successfully treated when penicillin was administered in a dosage of 30,000 units in each quarter for two successive days on several occasions with two days' interval after each two days' treatment. This method of intermittent treatment appeared to be of value in the treatment of cows which failed to respond to the routine method of infusion. The cow's milk was found to be free from *Str. agalactiæ* organisms when examined on four occasions after this method of treatment.

## An Outbreak of Industrial Fluorosis in Cattle

By H. H. GREEN, D.Sc.

It may be recalled that in the "Discussion on Fluorosis in Man and Animals" by this Section in February 1941<sup>1</sup> the occurrence of severe fluorosis in cattle was described on farms in the vicinity of brickworks in Bedfordshire. The purpose of the present communication is to report a similar occurrence associated with the calcining of ironstone in Lincolnshire. In this process the raw ironstone is mixed with about 7% of its weight of coal and ignited in the open, fresh layers being added to form a smouldering mound, the complete calcining of which occupies several weeks. The weight of the ironstone is so reduced for transport, and the ore rendered sufficiently porous for direct reduction in blast furnaces at distant smelting centres.

During the calcining process smoke drifts on to neighbouring farms, and on the nearest of these an obscure disease of cattle was reported by G. A. Moore, M.R.C.V.S. Samples of urine from affected cases, analysed at Weybridge, at once established a diagnosis by revealing 26 to 69 parts per million of fluorine. Analyses of materials collected during a visit to the area showed 1,200 p.p.m. fluorine in the ironstone itself and 100 to 180 p.p.m. in the various types of coal used. The calcined ore contained only 300 p.p.m., thus revealing a loss of three-quarters of the original fluorine or about four-fifths if loss of weight be allowed for. Imitation of the calcining process in the laboratory showed approximately 90% loss of fluorine at controlled muffle temperature of 850° C., but negligible loss at 600° C. It seems probable that the fluorine comes off as silicon fluoride which, reacting with atmospheric moisture and ammonia from the coal, condenses on the smoke particles as ammonium fluoride and silicofluoride and drifts on to the pastures. Samples of water from the affected farm showed only 0.5 p.p.m. fluorine but grass samples within a few hundred yards of the burning mounds showed over 2,000 p.p.m. on the dry matter. A straw stack about half a mile away showed 490 p.p.m. on the exterior and 70 p.p.m. in the interior. The danger zone has not yet been mapped out but it is not likely to exceed a radius of 2 miles.

<sup>1</sup>Proc. R. Soc. Med., 34, 391.

Cattle on the nearest farm were stunted and lame and the owner described some of the earlier cases as having to be destroyed after "crawling on their knees". Two cases have been purchased for post-mortem study by F. Blakemore at Cambridge and will doubtless show lesions similar to those described in 1941.

The human family on the same farm is a large one and urine samples of 9 people showed from 1.3 p.p.m. to 4.2 p.p.m. fluorine, averaging less than one-tenth that of the cattle urines but nevertheless being above normal and far above the value in the drinking water. Surface contamination of green vegetables is suggested as the source. Dr. Dagmar Wilson has been asked to consult with the local M.O.H. with regard to clinical symptoms in the children.

**Dr. Dagmar Wilson:** Our knowledge of human fluorosis has been considerably extended since fluorine was the subject of discussion by this Section in 1941. Fluorine is now generally recognized as an important trace element in human nutrition which exerts influence at non-toxic as well as at toxic levels.

The incidence of fluorotic lesions probably bears a definite relation to the economic and nutritional status of a community. A small amount of fluorine—under 1.0 p.p.m.—is present in many British water supplies and a lowered incidence of dental caries is found amongst people who have used a drinking water containing 0.5 to 1.0 p.p.m. fluorine for at least the first eight years of life. In the United States fluorination of water supplies is being considered for the partial control of dental caries. Human balance experiments show a close correlation between fluorine in the drinking water and in urine, and the elimination of fluorine is practically complete when the quantity absorbed does not exceed 5 mg. daily.

In human toxic fluorosis acute generalized and local effects are usually accidental. Chronic poisoning in man occurs in two ways, as chronic endemic fluorosis and as an occupational disease. Bone abnormalities and systemic illness, of which gastric derangement is an early sign, may occur when fluorine tolerance levels are exceeded.

Fluorides are being increasingly used in industry and large amounts of fluorine are associated with recent industrial development in this country. Methods for the control of the fluorine evolved are known and are not difficult of application, but the fluorine hazards for factory workers, for their families living near-by, and for other people resident or employed in the neighbourhood, as well as for the animal population, are not sufficiently appreciated.

**Dr. Margaret M. Murray:** In 1937 when Roholm's book "Fluorine Intoxication" was published and reviewed, my attempts to interest appropriate persons in the possibility of a fluorine hazard to human beings in Great Britain, particularly in the Scottish Highlands, failed. Since then we know of three outbreaks of fluorosis in farm animals and it is significant that in each case it was the effects on grazing animals which brought to light the existence of the fluorine hazard; consideration of the effects on human beings has in each case been secondary.

We owe it to Dr. H. H. Green and his colleagues that the cause of the outbreak in the Marston Valley was discovered and the plans for elimination of the danger were successfully worked out.

At Fort William it was because sheep in the vicinity of the aluminium works did not thrive that an inquiry was made into the question of a hazard to human beings. Examining school children at Inverloch, which has only 0.2 p.p.m. fluorine in the drinking water and which is situated in the direction of the prevailing wind carrying the fumes from the aluminium works, Dr. Wilson and I found the white mottling of dental fluorosis present even in the temporary teeth. We were, and still are, at a loss to explain what was the source of the fluorine which caused this mottling, particularly of temporary teeth.

Dr. Green's investigations show the fundamental importance and usefulness of the determination of fluorine in urine in the study of fluorine intoxication. These outbreaks of fluorosis in farm animals have also shown that not all animals are similarly affected by the ingestion of toxic amounts of fluorine. In cows the long bones are severely affected, while in the sheep at Fort William, investigated by Professor Boddie of the Royal Dick Veterinary College, Edinburgh, it was the jaw bones which were grossly abnormal and because of this the animals could not obtain food. This is the condition seen in the sheep, goats, camels and horses of the "darmous" region of Morocco. In man it is usually the vertebral column which is first affected and this effect may not be detectable until there has been a long period of exposure. Therefore, in the absence of positive X-ray findings, the only means of testing for the ingestion of fluorine at levels which will ultimately cause these bone changes, is by the determination of fluorine in the urine.

#### REFERENCE

BODDIE, G. F. (1945) Chronic Fluorine Intoxication in Sheep and its Effect upon the Teeth, *Proc. Nutr. Soc.*, 3, 94.

## The Chick-Embryo in Chemotherapeutic Research. [Summary]

By JOHN FRANCIS, B.Sc., M.R.C.V.S.

OGSTON (1881) was probably the first to cultivate bacteria in the incubating egg, but with the development of satisfactory culture media his work was forgotten. Goodpasture (1938) reviewed other early work with the chick-embryo and Landauer (1941) published an extensive monograph on the factors which influence the hatchability of eggs. Buddingh and Polk (1939*a* and *b*) found that by using embryos of suitable ages and inoculating meningococci by various routes they could produce lesions of septicæmia, sinusitis, pneumonia and meningitis, similar to those found in the natural disease; in addition they showed that immune serum had a protective effect. Following this work a considerable number