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NITRITE POISONING OF PIGS.

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SUMMARY.

Heavy mortality in pigs fed on soup prepared by cooking beef and offal in well water was investigated on two farms. The well water on each farm had a high nitrate content, equivalent to 2970 *and* 1740 *p.p.m. expressed as sodium nitrate. Batches of these soups which were analysed contained nitrites equivalent to* 1127 *and* 393·6 *p.p.m. sodium nitrite.*

It is considered that the mortality which occurred in pigs on these farms was due to nitrite poisoning and that the nitrite was derived from the nitrate in the well waters.

The toxicity of sodium nitrite for pigs was observed by dosing after 18 *hours fasting with a* 1 *per cent. aqueous solution. Doses of* 0·05, 0·06, 0·07, *and* 0·08 *gm. sodium nitrate per kgm. body weight produced moderate to severe non-fatal methaemoglobinaemia. Two pigs weighing* 61 *and* 80 *kgm. died two hours after they had been dosed at the rate of* 0·09 *gm. per kgm.*

Soups were prepared in the laboratory by boiling beef, bones and meat scraps in well water from one of the affected farms. The water contained nitrates equivalent to over 2300 *p.p.m. sodium nitrate and nitrites equivalent to less than* 10 *p.p.m. sodium nitrite. In two trials soups containing* 1430 *and* 1245 *p.p.m. sodium nitrite were obtained.*

INTRODUCTION.

The ability of ingested nitrite to reduce haemoglobin to methaemoglobin and thus to impair the oxygen capacity of the blood has long been known. Toxic symptoms appear when about 50 per cent. of the haemoglobin has been reduced to methaemoglobin (Nicholson, 1945).

Poisoning of cattle by oaten hay containing nitrate was described by Bradley, Eppson and Beath (1940) and Davidson, Doughty and Bolton (1941). Williams and Hines (1940) showed that the toxicity of the plant *Salvia reflexa* (mint weed) is due to nitrate. The nitrate is reduced by enzymes in the plant or in the rumen or liver of the animal to the more toxic nitrite. Gwatkin and Plummer (1940) showed that pigs dosed, after 24 hours' starvation, with potassium nitrate solution at the rate of 2·5 gm. of the salt per kgm. body weight suffered no ill effects, while 3·7 gm. per kgm. was fatal. Potassium nitrite given under similar conditions was much more toxic, as little as 0·25 gm. per kgm. being fatal.

McIntosh, Nielson and Robinson (1943) showed that the toxicity of cooked mangels fed to pigs was due to the presence of nitrite. They found that, to prevent the formation of toxic amounts of nitrite, it was necessary to cook the mangels at or near 100 deg. C. for at least two hours. McIntosh (1950) states that in this work mangels containing 6050-7360 p.p.m. sodium nitrate were cooked in a town water supply which, though not tested, probably contained minute traces only of either nitrate or nitrite.

In Queensland, at various times, heavy mortality has occurred in pigs fed cooked vegetables (most often pumpkin or mangels) or cooked meat or offal. None of the cases in which cooked vegetables were implicated has been investigated with nitrite poisoning in mind, but the New Zealand work of McIntosh *et al.* suggests that these Queensland outbreaks might have been due to nitrite poisoning.

The observations reported here relate to poisoning caused by soup made from boiled meat and offal. On two farms where detailed observations were made it was found that the water in which the offal was boiled had a high nitrate content. When offal was boiled in these waters nitrite was produced in amounts sufficient to render the food toxic to pigs.

Observations are recorded here on:—

(1) Natural outbreaks of nitrite poisoning in pigs fed on cooked offal.

(2) The toxicity of sodium nitrite for pigs.

(3) The toxicity of cooked offal prepared experimentally with well water which had a high nitrate content.

METHODS OF ANALYSIS.

Nitrates were determined by a modification of Ulsch's method in which nitrate was converted to ammonia with sulphuric acid and reduced iron, the ammonia being distilled by the usual Kjeldahl method, using magnesium oxide instead of sodium hydroxide to liberate ammonia. Devarda's alloy was not available for the reduction.

Nitrites were estimated by 1940 AOAC methods of analysis (Association of Official Agricultural Chemists, 1940).

Nitrate and nitrite levels are expressed throughout as the equivalents in parts per million of sodium nitrate and sodium nitrite respectively.

Haemoglobin determinations were made by J. M. Harvey in the Department's Chemical Laboratory by measuring the oxygen capacity of the blood in a Van Slyke manometric gas analysis apparatus.

CASE REPORTS.

Farm A (Beaudesert District).

This herd was fed on boiled slaughteryard offal, bones and meat scraps (mostly beef). Water was obtained from two sources—an earthen tank (or dam) and a well 135 feet deep. There was a local belief that pigs could not be fattened at this piggery owing to the toxicity of the well water. Consequently the owner, a butcher, used the piggery only when he had an excess of offal. Further, he did not use the well water unless the dam was dry. The offal was cooked principally to secure the tallow.

Information from the owner showed that in 1945 the dam dried up and offal was boiled in well water and fed to the pigs. Six pigs died during the following week and pig fattening was then abandoned for some months.

About April, 1946, he again penned pigs for fattening and used water from the dam to boil the offal. No sickness or deaths occurred. By July 1 the dam was empty and well water was brought into use. The herd appeared healthy at feeding time on the evening of July 6, but on the following morning 16 of a herd of about 30 were dead. Analyses of samples submitted by the local Inspector of Stock showed that no arsenic or heavy metals were present in the stomach contents and that the waters were low in total chlorides.

As butcher's offal was used it was suspected that beef pickled with nitrate may have been included in the offal. Analysis confirmed nitrite in all specimens, as follows:—

Well water—8·9 p.p.m. sodium nitrite.

Well water stored in iron tank—69·0 p.p.m. sodium nitrite.

Stomach contents of dead pig—3·0 p.p.m. sodium nitrite.

The water samples were received in 1 lb. glass jars with metal screw caps. The nitrites in the sample from the iron tank rose in a few days from 69 p.p.m. to 189 p.p.m., and later to 234 p.p.m. Nitrates were present in large quantities, viz., 2888 and 2324 p.p.m. sodium nitrate in the well and tank waters respectively.

The piggery was visited on July 19, 1946, to observe the boiling processes and to examine the possibility of the nitrate in the well water being derived from contamination of the well by refuse. It was found that the position of the well precluded external contamination. Field tests showed that the nitrites were higher in well water stored in a 400-gallon iron tank than in the water as it was pumped from the well. The dam water was free from nitrite. Laboratory analyses (Table 1) showed that the total chlorides calculated as sodium chloride were the same in both waters (viz., 761 p.p.m.), indicating that the increase in nitrite was not due to evaporation. Two batches of soup composed of offal, bones

Table 1.

CHEMICAL ANALYSES OF SAMPLES COLLECTED AT FARM A.

Date.	Sample.	$NaNO_2$ p.p.m.	$NaNO_3$ p.p.m.	NaCl p.p.m.
July, 1946 ..	Well water direct from delivery pipe of pump*	6·1	2970	761
	Well water stored in iron tank	41·6	2890	761
	Soup during boiling	1127	..	..
	Soup cooled and tallow removed ready for feeding	1225	..	..
July, 1947 ..	Well water direct from delivery pipe of pump	5·0	1970	569
	Well water stored in iron tank	100	2110	627
	Soup ready for feeding	2·5	trace	..
	Dam water being used for preparation of soup	nil	nil	..

* All samples were taken in glass bottles with cork stoppers and were filled to leave no air-space.

and beef scraps were also high in nitrite. A batch which had been boiling for several hours and was still boiling when sampled contained 1127 p.p.m. sodium nitrite. A soup which had cooled and from which the tallow had been skimmed contained 1225 p.p.m. sodium nitrite.

The herd was being fed entirely on cooked offal diluted with an equal part of dam water, so that the final nitrite content would be 500-600 p.p.m. sodium nitrite (500 p.p.m. = 35 grains per gallon).

There had been no deaths from July 7 until the afternoon before the visit, when one pig died. A post-mortem examination was done on this animal about 18 hours after death, but the carcase was so decomposed as to be of little value. The liver contained 2·9 p.p.m. and the stomach content 7·8 p.p.m. sodium nitrite.

Experiments reported below showed that 0·09 gm. sodium nitrite per kgm. body weight may kill pigs. The pigs on farm A weighed about 45 kgm., hence 8·1 litres of a soup containing 500 p.p.m. sodium nitrite could be fatal to such animals. As no other feed was given it is likely that this amount of soup would be consumed.

The nitrite in the cooked offal was evidently derived from the nitrate in the well water. It will be noticed that some nitrite was produced during the storage in the iron tank and that a great increase in nitrite occurred when the water was boiled with meat in large iron vessels over wood fires.

Heavy rain (18·17 inches) fell between January 18 and February 1, 1947. On February 10 the well water contained 9·5 p.p.m. sodium nitrite and 2433 p.p.m. sodium nitrate, indicating very little dilution by the heavy rain. On February 26 the well water contained 8·4 p.p.m. sodium nitrite and 2331 p.p.m. sodium nitrate. The well was still high in nitrate (1970 p.p.m.) in July, 1947, and soup being prepared at that time with dam water contained only negligible nitrite, viz., 2·5 p.p.m. (Table 1).

Farm B (Pittsworth District).

This herd consisted of 80 mixed pigs fed on whey, crushed sorghum, wheat, barley and wheaten or lucerne hay. On August 9, 1946, beef scraps were boiled in water pumped from a deep bore and fed to pigs. Each day thereafter this beef-soup was reboiled in well water and some was fed. On August 12, 14 store pigs 3-5 months old died within $2\frac{1}{2}$ hours of feeding. Analyses of samples collected on the day of this mortality by the local Inspector of Stock resulted as follows:—

Bore water—1740 p.p.m. sodium nitrate;
0·37 p.p.m. sodium nitrite.

Beef soup—1690 p.p.m. sodium nitrate;
393·6 p.p.m. sodium nitrite.

Stomach contents of dead pig—4·9 p.p.m. sodium nitrite.

Inflammation of the gastric mucosa, with many haemorrhages from pin-point up to 2 cm. in diameter, was found in a dead pig examined by the Inspector of Stock. No other lesions were noticed. The characteristic feature of nitrite poisoning is the brown colour imparted to the blood by methaemoglobin, but this may disappear a few hours after death.

It will be noted that the beef soup was boiled three times before deaths occurred. The final product contained 393·6 p.p.m. (28 grains per gallon) sodium nitrite. Assuming that these pigs weighed 25 kgm. and that 0·09 gm. of sodium nitrite per kgm. body weight constitutes a fatal dose, then about $5\frac{1}{2}$ litres of this beef soup would be fatal. McIntosh *et al.* (1943) stated that boiled mangels containing 20 grains sodium nitrite per gallon (i.e. 286 p.p.m.) are dangerous to small pigs.

THE TOXICITY OF SODIUM NITRITE.

Pigs varying in weight from 48 to 80 kgm. were dosed experimentally with sodium nitrite. The experimental animals were in good condition and had been fed in pens on crushed grain containing about 9 per cent. meatmeal and 10 per cent. lucerne chaff for several weeks before they were used. The morning feed was omitted on the day the pigs were dosed. A small feed of crushed grain was given about two hours after treatment. The results are summarized in Table 2.

Table 2.

EFFECT ON PIGS OF 1% SOLUTION OF SODIUM NITRITE GIVEN BY STOMACH TUBE.

Dose Rate gm./kgm.	Animal No.	Weight kgm.	Haemoglobin (gm. per 100 ml.)			Results.
			Pre-treatment.	3 hr.	5 hr.	
0·04	487	73	..	..	..	Non - fatal methaemoglobinaemia 2–4 hours after treatment. Recovered in 6–7 hours
0·045	581 (capsule)	73	..	..	..	
0·045	485	66	..	..	..	
	486 (capsule)	73	14·6	9·4	13·0	
0·05	484	53	14·2	8·7	11·3	
0·06	488	48	..	..	..	
0·07	488	51	..	..	..	Severe non-fatal methaemoglobinaemia 2–4 hours after treatment. Recovery in 7 hours
0·08	487	75	..	..	..	
0·09	484	61	..	..	..	Dead in 2 hours
	486	80	..	..	..	

The effect of sodium nitrite given in the solid form in gelatin capsules was compared with the effect of a 1 per cent. aqueous solution given by stomach tube. A pig dosed by capsule developed similar, but slightly less intense, symptoms to those shown by a pig dosed with a 1 per cent. solution at the same dose rate—viz., 0·04 gm. sodium nitrite per kgm. body weight. At a dose rate of 0·045 gm. per kgm., administration by capsule and stomach tube produced symptoms which were similar in intensity and in the time of their development and eventual disappearance. Subsequent observations were made by dosing by stomach tube with the 1 per cent. solution.

The lowest dose rate used—viz., 0·04 gm. per kgm.—produced definite symptoms of methaemoglobinaemia. About two hours after treatment the skin over the whole body was blanched. The pigs became docile and lethargic but they ran normally when chased. The conjunctival vessels were a definite nut-brown colour. About five hours after treatment these symptoms had disappeared and the animals appeared normal.

Single pigs were then dosed with 0·05, 0·06, 0·07, and 0·08 gm. per kgm. Symptoms of methaemoglobinaemia developed in all cases and became more severe as the dose rate was increased, but none of these doses was fatal. Symptoms were at their height three to four hours after treatment. Blanching of the skin was the first visible symptom, and it was easily seen when there were treated and untreated pigs in the same pen. (All the experimental pigs were Large Whites.) The higher dose rates produced methaemoglobinaemia, which was so marked that distinctly coffee-coloured blood was visible in the conjunctival vessels. Respiration became rapid and shallow and the pigs were very disinclined to exert themselves.

Two pigs weighing 61 and 80 kgm. were then dosed at the rate of 0·09 gm. per kgm. Both pigs developed severe methamoglobinaemia and died two hours after treatment. There was no struggling until about five minutes before death.

Autopsy immediately after death of the two fatal cases revealed similar lesions in each animal. The subcutaneous blood vessels were engorged with chocolate-coloured blood. The heart was in diastole. The lungs were pale and contracted and approximately 20 sub-pleural petechiae were found about the hilus of each organ. Congestion and petechiation was noted on the glandular mucosa of each stomach. The duodenum was catarrhal but the remainder of the small and large intestines of each pig appeared normal. The livers and kidneys were greatly engorged with chocolate-coloured blood and the spleens were contracted. Liver, kidneys, urine and contents of the alimentary tract of these two pigs were analysed for nitrite. The results of these analyses are shown in Table 3.

Table 3.

PIGS DOSED WITH 1% SOLUTION SODIUM NITRITE: SODIUM NITRITE CONTENT OF ORGANS FROM FATAL CASES.

Pig No.	Specimen.	Total Weight gm.	Weight of Sample gm.	$NaNO_2$ p.p.m.
484	Stomach content	566	566	887
	Small intestinal content	336	336	2·96
	Large intestinal content	not weighed	762	3·45
	Liver..	1816	681	less than 1
	Two kidneys	227	227	less than 1
	Urine	about 100	..	less than 1
486	Stomach content	1589	1589	250
	Small intestinal content	364	364	4·0
	Large intestinal content	3458	790	3·2
	Liver..	2000	454	less than 1
	Two kidneys	252	252	less than 1
	Urine	nil	nil	..

EXPERIMENTS WITH COOKED OFFAL.

Observations were made on the nitrite and nitrate content of beef soups prepared experimentally with well water from farm A. The toxicity of the soup was tested by feeding it to pigs.

The soup was made by boiling beef scraps (30 lb.), usually rib and shin bones and a head, for four to five hours in well water. The vessels used were an iron petrol drum (44 gallons) and a kerosene tin. The latter was a 4-gallon iron vessel coated with tin by dip galvanizing.

Table 4.

NITRITE CONTENT OF SOUPS PREPARED BY BOILING BONES AND OFFAL IN WELL WATER FROM FARM A.

Trial.	Treatment.	Vessel.	$NaNO_2$ p.p.m.
1	First boiling	Iron drum ..	..
	Second boiling	Iron drum ..	..
	Third boiling	Iron drum ..	..
	Fourth boiling	Kerosene tin	720
2	First boiling	Iron drum ..	258·8
	Second boiling	Iron drum ..	241·6
	Third boiling	Iron drum ..	156·0
	Fourth boiling	Kerosene tin	1060·0
	Fifth boiling	Kerosene tin	..
	After cooling overnight	Kerosene tin	1430·0
3	First boiling	Iron drum ..	162·6
	Second boiling	Glass flask ..	101·0
	Growth of air-borne micro-organisms, 7 days at room temperature	..	9·9
4	First boiling	Iron drum ..	15·5
	Second boiling	Iron drum ..	23
	Third boiling	Kerosene tin	1070
	After cooling overnight	Kerosene tin	1245

The nitrite content of the soup was estimated after each boiling (Table 4). When the content had reached a level considered likely to be toxic, pigs which had been fasted overnight were dosed by stomach tube. The amount of soup administered was determined by its nitrite content and the body weight of the pig. It was intended to dose at the rate of 0·09 gm. per kgm., but in both trials where death resulted the nitrite content of the soups increased during the interval between the last estimation and drenching so that the doses given exceeded 0·09 gm. per kgm. (Table 5). The usual morning feed was omitted prior to drenching.

Blood haemoglobin estimations were made before and at intervals after dosing. Pigs which died were autopsied immediately after death.

Trial 1.

The water used in this trial had been collected at farm A six months previously. It contained at the commencement of the trial 2·06 p.p.m. sodium nitrite and 2783 p.p.m. sodium nitrate.

The soup was boiled in an iron drum, but the nitrite content did not rise to a level considered toxic for pigs. After three boilings the volume was considerably reduced. The soup was then transferred to a kerosene tin and again boiled. This fourth boiling increased the nitrite content to 720 p.p.m. Three litres of this soup were given to a pig (no. 492) weighing 37 kgm. The dose rate was 0·058 gm. sodium nitrite per kgm., but no adverse effects were noted.

Table 5.

EFFECT OF EXPERIMENTALLY PREPARED SOUPS (TABLE 4) ON PIGS.

Trial.	Pig No.	Weight of Pig kgm.	Soup.		Dose Rate of $NaNO_2$ gm./kgm.	Haemoglobin (gm./100 ml.)		Result.
			Nitrite Content p.p.m.	Vol. Dosed ml.		Pre-treatment.	Just before Death.	
1	492	37	720	3000	0·058	..	..	No adverse effects
2	493	46	1430	4000	0·124	12·1	1·5*	Death in 51 minutes
2	496	38	1430	3200	0·120	10·7†	2·6	Death in 40 minutes
4	495	82·1	1245	6900	0·105	17·8	5·3	Death in 63 minutes
4	492	76·2	1245	3500	0·059	13·5	10·3‡	Non-fatal methaemo-globinaemia

* Sample taken just prior to death was showing a tendency to clot, so result is not accurate.

† The pre-treatment sample of approximately 5 ml. of blood was added to 0·5 ml. of aqueous sodium citrate, so the figure given is about 10% low. Dried sodium citrate was used for all other blood samples.

‡ Sample taken 2 hours after dosing.

Trial 2.

The water used in this trial (and in trials 3 and 4) was also from farm A and contained 8·4 p.p.m. sodium nitrite and 2331 p.p.m. sodium nitrate. Soup was prepared as in trial 1 and boiled for four hours in the iron drum. The first boiling increased the nitrite content to 258·8 p.p.m., but further boiling decreased the concentration, so that after the third boiling it was down to 156 p.p.m. The soup remaining in the drum was stored in glass jars for seven days, during which time there was a profuse bacterial growth. The soup was then transferred to a kerosene tin and boiled for the fourth time for four hours. The nitrite content rose to 1060 p.p.m. The soup was boiled a fifth time and then two pigs were each dosed with a quantity estimated to contain 0·09 gm. sodium nitrite per kgm. Subsequent analyses showed that the last boiling and cooling had increased the concentration to 1430 p.p.m. sodium nitrite. The pigs therefore received a dose of 0·12 gm. per kgm. Both pigs developed severe methaemoglobinaemia. Death occurred 40 minutes (pig. no. 496) and 51 minutes (pig no. 493) after dosing.

Post-mortem examinations done immediately after death showed similar lesions in both pigs. The subcutaneous vessels were engorged with chocolate-coloured blood. The heart was in diastole but showed no macroscopic lesions. The lungs were pale and contracted, with a few subpleural petechiae. The stomach of one pig showed a marked catarrhal inflammation. The glandular mucosa was congested and seemed to strip rather readily. Inflammation of the duodenum was most marked and extended for several feet. The liver and kidneys were engorged with coffee-coloured blood. Samples of liver, spleen, kidney, stomach and intestinal contents were collected for chemical analysis (Table 6).

Table 6.

PIGS DOSED WITH EXPERIMENTAL COOKED OFFALS: SODIUM NITRITE CONTENT OF ORGANS FROM FATAL CASES.

—	Pig No.	Volume of Soup and Nitrite Content.	Weight of Pig kgm.	Dose Rate gm./kgm.	Specimen.	$NaNO_2$ p.p.m.
Trial 2	493	4000 ml. containing 1430 p.p.m. $NaNO_2$	46	0·124	Stomach content ..	7·6
..	..	..	..	..	Intestinal content ..	6·6
..	..	..	..	..	Liver..	less than 1
..	..	..	..	..	Kidney	1·75
..	..	..	..	..	Spleen	1·6
Trial 2	496	3200 ml. containing 1430 p.p.m. $NaNO_2$	38	0·120	Stomach content ..	101
..	..	..	..	..	Intestinal content ..	5·7
..	..	..	..	..	Liver..	less than 1
..	..	..	..	..	Kidney	2·75
..	..	..	..	..	Spleen	3·5
Trial 4	495	6900 ml. containing 1245 p.p.m. $NaNO_2$	82·1	0·105	Stomach content ..	478
..	..	..	..	..	Small intestinal content	514·5
..	..	..	..	..	Large intestinal content	less than 1
..	..	..	..	..	Liver..	less than 1
..	..	..	..	..	Kidney	less than 1
..	..	..	..	..	Spleen	less than 1

Trial 3.

As trials 1 and 2 indicated that repeated boilings in the iron drum did not raise the nitrite content to a toxic level, it was thought that perhaps bacteria may have been responsible for the reduction of nitrate to nitrite, so the following experiment was undertaken.

A sample (about 2 litres) was taken from a batch of soup which had been boiled once for four hours in the iron drum. The nitrite content was 162·6 p.p.m. The sample was boiled for one hour in a flask and then exposed to contamination by air-borne micro-organisms for seven days. At this time the nitrite content was 9·9 p.p.m., compared with 101·0 p.p.m. immediately after boiling.

This experiment indicates that the increase in nitrite was not due to micro-organisms.

Trial 4.

Two boilings in the iron drum reduced the volume of soup from 11 gallons to 2¾ gallons. The nitrite content increased from 9 p.p.m. in the original water to 20 p.p.m.

The soup was then transferred to a kerosene tin and boiled for the third time. The nitrite content rose to 1070 p.p.m. After the soup had stood overnight in the tin, the nitrite content increased further to 1245 p.p.m.

This soup administered to pig no. 495 at the rate of 0·105 gm. per kgm. produced methaemoglobinaemia and death in 63 minutes. Observations on this animal were:—

Pig. No. 495. Weight 181 lb. (82·1 kgm.).

10.0 ..	Dosed by stomach tube with 6900 ml. of soup containing 1245 p.p.m. sodium nitrite.
10.5 ..	Urinated.
10.28 ..	Urinated.
10.29 ..	Paddling gait, respiratory rate 68 per minute.
10.35 ..	Respiratory rate 80 per minute.
10.37 ..	Animal lay down.
10.39 ..	Animal rose and defaecated.
10.40 ..	Restless; skin blanched.
10.47 ..	Vomited twice; quantity (estimated) 300-400 ml.
10.49 ..	Vomited about 200 ml.
10.50 ..	Marked ataxia.
10.51 ..	Went down and struggled.
10.53 ..	Buccal mucosa very cyanotic; marked respiratory distress.
10.55 ..	Animal lying on its side paddling; dyspnoeic.
11.03 ..	Death.

A post-mortem examination done within 30 minutes of death presented lesions identical with those described in trial 2. Liver, kidneys, spleen and contents of the alimentary tract were analysed for nitrites; the results are shown in Table 6. The remainder of this batch of soup (3,500 ml.) was administered to pig no. 492. The animal weighed 76·2 kgm. so the dose rate was 0·059 gm. sodium nitrite per kgm. Non-fatal symptoms of methaemoglobinaemia were exhibited.

The Reduction of Nitrate to Nitrite in Well Water.

The original well water from farm A contained approximately 10 p.p.m. sodium nitrite and 2800 p.p.m. sodium nitrate. It was thought at first that boiling with offal was sufficient to reduce the nitrate, but this was not substantiated by the experiments described above. Certain bacteria are capable of reducing nitrate to nitrite. It was considered that the increase in nitrite concentration in trial 2 may have been brought about in this manner, but this was not confirmed in trial 3.

Boiling in the iron drum resulted in an initial increase in nitrite followed by a decrease (trial 2). Concentration by boiling in this vessel did not increase the nitrite content. However, when the mixture was transferred to a kerosene tin and boiled there was, on each occasion (i.e., in trials 2 and 4), a marked increase in nitrite. It was felt that the tin coating of the kerosene tin may have been responsible for the conversion of nitrate to nitrite either by acting as a catalyst or by preventing the complete reduction of nitrate to ammonia.

DISCUSSION.

The evidence presented indicates that the field outbreaks were due to nitrite poisoning resulting from feeding offal cooked in water with a high nitrate content.

The mechanism of the reduction of nitrate to nitrite is still obscure. Suckling (1944) stated:—"Many waters containing nitrates, if allowed to stand in contact with iron, zinc or lead in pipes or cisterns, act upon the metal, the nitrates being in part reduced to nitrites and ammonia." This might explain the increase in nitrite in the water samples kept in the glass jars with metal screw caps and also in the water stored in the iron tank at farm A.

The experiments reported here in which offal was boiled in an iron drum and in a kerosene tin do not duplicate the conditions at farm A, where the boiling was done in a cast iron vat. Also, in the field excess soup was retained and boiled with the next batch of offal. Further chemical experiments are being done on the factors concerned in the production of nitrite from these waters.

The excess nitrate in the soup did not appear sufficient to be toxic, because in trial 1 pig no. 492 received 0·058 gm. per kgm. sodium nitrite plus the unreduced nitrate and suffered no ill effects.

From their work on nitrite poisoning in pigs fed on cooked mangels, McIntosh *et al.* (1943) concluded:—"Mangels cooked at temperatures below boiling point, or for too short a time at boiling point, were shown to be dangerous to pigs, owing to the amount of nitrite formed during cooling. . . . Production of nitrite on cooling may be avoided by boiling mangels for at least two hours, at or near boiling point." McIntosh (1950) points out that in their experiments no nitrite was present at the completion of the cooking. All the nitrite was produced during cooling and its presence or absence depended solely on the temperature during cooking. In cooked offal, on the other hand, toxic levels of nitrite are not avoided by cooking at 100 deg. C., because a vat at farm A which had been boiling for several hours and was still boiling when sampled, contained 1127 p.p.m. sodium nitrite (Table 1).

In Queensland, four natural well waters, besides those from farms A and B described above, have been encountered with nitrate contents ranging from 270 to 4000 p.p.m. There is a record also of a sub-artesian water analysed in the Department's Chemical Laboratory in 1930 containing the equivalent of 2380 p.p.m. sodium nitrate. This water was submitted as the suspected cause of deaths in sheep, but the particulars of the outbreak could not be found. The observations reported above indicate the danger of using such waters to prepare cooked offal for feeding to pigs. Such waters may be dangerous also to human beings, particularly infants. Faucett and Miller (1946) record three cases of methaemoglobinaemia in infants fed milk mixtures diluted with well waters high in nitrate. The water used by two patients contained 70 mgm. nitrate nitrogen per litre (i.e., 425 p.p.m. sodium nitrate) and a trace of nitrite nitrogen per litre. In the other case the well water contained 300 mgm. nitrate nitrogen per litre (i.e., 1821 p.p.m. sodium nitrate) and a trace of nitrite nitrogen per litre. Ferrant (1946) reported two cases of methaemoglobinaemia in newborn infants fed a powdered milk formula made up with boiled well water. In the first case the water contained 497 p.p.m. nitrate (i.e., 681 p.p.m. sodium nitrate) and 0·114 p.p.m. nitrite (i.e., 0·171 p.p.m. sodium nitrite), and in the second case 180 p.p.m. nitrate (i.e., 247 p.p.m. sodium nitrate) and a trace of nitrite.

Gwatkin and Plummer (1946) showed that pigs dosed with 10 per cent. solution of potassium nitrite after 24 hours fasting died of methaemoglobinaemia, whereas pigs given a similar dose rate without previous fasting suffered only slight ill effects. The presence of food in the stomach probably prevents fatal anoxaemia from sudden conversion of haemoglobin to methaemoglobin by delaying absorption of nitrite. It seems, therefore, that in the field mortality from nitrite poisoning may be greatly influenced by the amount of ingesta in the stomach when the nitrite-containing feed is given.

Nitrite poisoning can be diagnosed by the distinct brown colour imparted to the blood by methaemoglobin. The brown colour is readily detected in sick or freshly dead animals but it may not be detected if the autopsy is done several hours after death, because the methaemoglobin is converted by bacteria back to haemoglobin (Williams and Hines, 1940).

The nitrite content of specimens from pigs which died of experimental nitrite poisoning (Tables 3 and 6) indicates that the stomach content is the most suitable specimen for diagnosis, though there is a wide variation in the amount of nitrite recovered. The stomach contents of three pigs which died after dosing with experimental cooked offals contained 7·6, 101 and 478 p.p.m. sodium nitrite (Table 6). The variation is presumably due to the action on the nitrite of enzymes and bacteria. Thus the presence of only a small amount of nitrite in the stomach or other organs does not necessarily exclude a diagnosis of nitrite poisoning.

Symptoms and lesions of fatal nitrite poisoning were produced in three pigs dosed with these soups in amounts corresponding to 0·105, 0·120 and 0·124 gm. sodium nitrite per kgm. body weight.

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