GENERAL SUMMARY AND CONCLUSIONS.
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In view of the importance of manganese in both plant and animal life and the resulting effects due to its deficiency well known in different parts of the world, and characterised by loss of appetite and body weight, thriftlessness and unproductive condition followed by poor general health the symptoms commonly observed in Indian cattle, 'studies on manganese in relation to cattle nutrition in India' were instituted in these laboratories in the year 1946.

A comprehensive review of the work done in various laboratories, leading to the development of concept of manganese as a factor in the nutrition of cattle, and covering the principle aspects of the subject from the initial stages to the present state of knowledge, has been presented.

Programme of Work.

To make a thorough survey of the problem of manganese nutrition under conditions of Indian dietetics and practical bearing on the stock raising industry, the following programme was chalked out:

1. Distribution of manganese in biological materials.
2. Manganese content of animal feeds.
3. Seasonal variation in the manganese content of some of the leafy fodders, the indigenous grasses and the pasture plants.
4. Variation in manganese content of some of the cultivated plants with progress of maturity.
5. Influence of irrigation, climate and other environmental conditions on the manganese content of common animal feeds.

II. Appetite, body weight and metabolism of major nutrients in relation to intake of manganese.

III. Metabolism of manganese and its minimum requirement for:

1. Adult kumauni bullocks.
Growing calves, and
lactating buffaloes.
Studies on the morphological and chemical composition of blood.
Elucidation of metabolic interrelationship between manganese and cobalt.

**Technique of the method.**

Before beginning the actual work the first essential requisite was an accurate, dependable and speedy method for the estimation of manganese in biological materials. A survey of the existing methods was made. After a survey of the existing methods and a comparison of the Cook's method with author's modified and improved technique for the estimation of manganese in biological materials, the interference due to various salts with particular reference to iron was studied and was found in amounts of as high as 10 mg. not interfering with the recovery of manganese contained in the sample. The recovery tests made on different samples of biological materials such as cakes, leafy fodders, grasses, straws, excretion, urine, blood and milk as well as synthetic solution demonstrated the reliability of the recommended procedure.

**Distribution of manganese in biological materials.**

Malnutrition in Cattle in India has very often been associated with the poverty of the feed in some of its major mineral nutrients like calcium and Phosphorus. There has been complete lack of information relating to the distribution of minor elements in nature under Indian conditions until the institution of trace element investigations in the Animal Nutrition Research Laboratories; at Izatnagar. Previous investigations from these laboratories have shown that a number of feed samples obtained from various localities were
were deficient in copper and cobalt. It is not unlikely, therefore, that a similar deficiency of manganese though not acute enough to develop clinical symptoms, may be traceable in feeds commonly fed to cattle in many tracts of this country.

**Manganese content of some of the common animal feeds.**

The amount of manganese in as many as 98 samples of common animal feeds was found to range between 2.5 and 210.5 p.p.m. of the dry matter and showed with the exception of paddy straw, significant variations from sample to sample.

The green feeds representing grasses, leafy fodders and cultivated plants showed a wider range of 12.35 in maize plant to 210.50 p.p.m. of the dry matter in sanjana leaves. The leafy fodders were found to be relatively richer in manganese. 30 concentrates including oil cakes grains, and feeds and their by products exhibited a range of 2.50 to 183.80 p.p.m. of manganese, maize cake being the poorest amongst the cakes and rice bran the richest in this respect.

When arranged in the ascending order of their manganese content, the straw (excluding paddy) and grains constitute the first group, concentrates and green feeds the second group and brans the third group. Wide variations were observed in the range of manganese concentration even in plants of the same order, differing, however, in age, suggesting the influence of progress of maturity on the manganese content of animal feeds.

**Seasonal variation in the manganese content of common animal feeds.**

(a) **Indigenous grasses:**

Seventeen pure species of grasses were examined for their
their manganese content in four cuts at monthly intervals, the first cut having been taken in early August. Almost majority of the samples showed continuous fall in their manganese content throughout the period of experimentation. The two samples of *Pennisetum arabicul* obtained from two different localities exhibited fall in the second and third stages of sampling followed subsequently by a rise in the 4th stage of sampling. The steady decline in the manganese content with increase of age of grasses suggests that either the rate of production of dry matter is such that the absorption of manganese lags behind or the plants possess an increased rate of absorption of manganese in the early stages of growth which subsequently declines.

(b) *Leafy fodders.*

Three successive bi-monthly loppings, from November to March, of 17 trees were analysed for their manganese content. 27.8 percent of the samples show continuous fall, 33.3 percent exhibit rise in the second lopping stage with a decline in the third stage of lopping while the remaining 38.9 percent of the samples show continuous rise from the first stage to the third stage of lopping. The Concentration of manganese varied significantly with advance in season from November to March. The range of manganese at the second stage of lopping which closely corresponds to spring (end of January) is definitely superior to the first stage of lopping but equally good as the third stage of lopping (end of March). The absence of a regular trend in tree leaves has, however, been ascribed to difference in the rate of production of dry matter and the attainment of maturity in different cases.
(c) **Cultivated fodder plants.**

Maize, Jowar, cowpeas and bajra comprising the Kharif crop and wheat, oat, gram and barley the Rabi crop were analysed for their manganese and crude protein contents with progress of maturity. Like grasses, the manganese content showed continuous fall. The protein content also gradually decreased. The concentration of manganese ranged from 21.23 for Barley to 462.50 p.p.m. for Bengal gram. on the whole the 'Rabi' crops were superior to 'Kharif crops' in this respect.

**Influence of irrigation on the manganese and crude protein contents of maize plant and its parts.**

The manganese content of the maize plant and its various parts shows continuous fall with age with the exception of stems. The crude protein content also behaves in the same way in both irrigated and non-irrigated conditions, other factors remaining the same. Comparatively the plants grown in the irrigated plots show higher manganese content than the non-irrigated ones both in plants and other parts. This high absorption of manganese can probably be explained on the assumption that the water supply keeps the manganese salts in solution and ready for absorption.

**Influence of seasonal variation of the manganese content of pasture grasses.**

A study of the manganese content of grass samples from four pastures, consisting essentially of *Cynodon dactylon* revealed that the manganese content suffered during reise, on an average in 75% of the samples. The fall in the manganese content may be attributed to either the leaching action of rains on the assimilable manganese salts present in the soil or to an increased rate of growth of grasses during the rainy season or both. The effects of growth and monsoon having died out with the onset of winter, the rate of assimil-
assimilation of manganese exceeded the rate of production of dry matter, with the result that an increased recovery of manganese was observed during the winter months which was maintained during the spring season.

Influence of soil, manurial and other environmental conditions on the manganese content of straw and grasses.

The manganese content of as many as 60 samples of straw mainly wheat, paddy and jowar collected from various parts of the country is found to range between 14.76 and 786.11 p.p.m. Practically 50 percent of the samples contained less than 50 p.p.m. of manganese on the dry matter basis. It was observed that in the United Provinces, the average manganese content of wheat straw is minimum for the Eastern zone, maximum for the central zone while that of the western zone lay in between.

The manganese content of paddy straw samples from Madras is higher than that of the United Provinces. Grasses exhibited a narrower range of manganese than the straws, all the 35 samples examined lying within (22.33 - 197.69 p.p.m.) on dry weight basis. 33 percent of the samples fell below 50 p.p.m., the critical concentration of manganese recommended by Wisconsin workers where as in two third of the total samples it ranged between 50-197.69 p.p.m. There are variations in the manganese content of different grasses from the same locality wide enough to suggest the possibility of the botanical composition of the samples being responsible for it. Although the number of samples are too small to permit any definite conclusions being arrived at, it appears that apart from species, soil and other environmental conditions also influence the manganese content. The usefulness of a comprehensive work in this line has been suggested to demarcate zones deficient in manganese.
Manganese content of tissues and blood of normal animals.

A few more important organs of cattle were examined four times in the season from June to November, every time three animals of uniform build having been selected. The concentration of manganese ranged from 0.0180 to 0.0192 mg/100 ml. in blood; 20.31 to 18.09 p.p.m. in liver; 4.26 to 6.59 p.p.m. in kidneys; 4.76 to 5.30 p.p.m. in bone marrow; 5.22 to 7.15 p.p.m. in pancreas; 2.10 to 2.92 p.p.m. in testes and 1.20 to 1.36 p.p.m. in brain on dry basis. Among the different organs, liver, kidneys and bone marrow showed consistently high values for manganese content throughout the period of experimentation the rest in order being pancreas, testes and brain and blood, the last two exhibiting only very slight variations.

Manganese metabolism in ruminants.

In an endeavour to elucidate the physiological and the metabolic behaviour of cattle in response to long term feeding of manganese deficient diet and to work out their minimum requirement, and experiment extending over a period of eight months was instituted with a uniform set of Kumauni bullocks. The basal diet consisted of washed wheat straw as the roughage component of the diet and maize cake as the concentrate.

General behaviour of Kumauni bullocks in relation to manganese intake.

The animals belonging to the control group began to show the deficiency symptoms as judged by the gradual loss in vigour, body weight and appetite almost five months after they have been on the basal-diet. Towards the end of the experiment, the coat became rough and the animals became unthrifty.
Supplementation of the basal ration with manganese at a level very close to the minimum requirement successfully prevented these gross symptoms to develop, the appetite and the utilization of the feed improved around improvement in these animals. Through out the period of experimentation, they maintained their normal thrift and vigour, the coat remaining soft and shining.

**Metabolic behaviour of kumauni bullocks in relation to intake of manganese.**

With continued feeding of manganese deficient diet, the digestibility of the whole ration and that of the crude protein was adversely affected. Consequently the availability of the various nutrients from the feed became restricted and the retention fell. The order of the magnitude of manganese deficiency as judged by metabolic behaviour became increasingly pronounced with the duration of the experiment.

Manganese was mainly excreted through the faeces and only about one percent through the urine.

The retention of manganese by kumauni bullocks increase with levels of intake and the animals on manganese deficient diet were excreting more than the intake. The excretion through the gut alone exceeded the intake suggesting that the alimentary tract of this species is permeable to manganese.

**Minimum requirement of manganese for cattle.**

The minimum requirement of manganese for the adult kumauni bullocks when at rest has been found to be 37.64 mg. per 100 lb. body weight and p.p.m. of the dry matter consumption as well. The estimated values for buffaloes in milk per 100 lb. body weight is 72.99 mg. per 100 lb. body weight and 48.32 p.p.m. of the dry matter consumption. However, the requirement as measured in terms of p.p.m. of the dry...
dry matter consumption in these groups of animals roughly varies with
in 39.08 to 48.32 and any ration having a lower level would be consi-
dered deficient in this respect for cattle.

**Blood composition in relation to manganese intake.**

In view of the importance of the physiological approach to the
problem as to evolve out a suitable diagnostic procedure for the
detection of manganese deficiency, and to study to changes in blood
composition consequent on manganese deficiency in the ration and
manganese therapy, a systematic study of the various morphological
and chemical constituents of blood of six kumauni bullocks was made.

**Morphological constituents of blood in relation to manganese intake.**

Departure in the blood picture of the kumauni bullocks from the
normal in response to manganese deficiency in the ration could not be
reflected until after the 5th month of the institution of the
experiment. However, by the end of the experimental period of eight
months, the red cell count, the haemoglobin concentration and the
corpuscular volume were finally reduced by 11.58, 5.30 and 3.80
percent respectively in the control group.

Supplementation of the ration with manganese, raising the
manganese status of the diet to 37.54 p.p.m. of the dry matter,
which had proved to be preventive against an allround deterioration,
was also effective in respect of blood picture. It was only at the
second level of manganese ingestion, equivalent 39.08 p.p.m. of the
dry matter, that the blood picture improved.
Chemical constituents of blood in relation to manganese intake.

The chemical constituents of blood, serum plasma such as calcium, inorganic phosphorus, magnesium, total protein, and phosphatase exhibited normal range throughout the experiment irrespective of manganese concentration in the feed while the vitamin C concentration fell in the control group and remained unaffected in the supplemented group.

The first visible symptoms of manganese deficiency in the ration as judged in terms of manganese concentration in the blood appeared during the 5th month of the experimentation and continued with increasing rapidity with prolonged manganese starvation. Supplementation of the ration with manganese immediately returned its concentration in the blood to its normal level, which however, remained unaltered though the level of intake was raised to nearly double.

The fall in the manganese content of blood, which appeared towards the 5th month, preceded the development of the gross symptoms as also the haematological deterioration, and, thus may serve as a guide in locating manganese deficiency in cattle.

Metabolism of manganese in relation to cobalt intake.

The importance of manganese in the cellular oxidation of Vitamin B1 and the suggested role of cobalt in the synthesis of vitamin B1 a possibility of the metabolic interdependence between manganese and cobalt was suspected. However, it was shown that the intake of cobalt within the limits of the experiment, failed to alter the metabolism of manganese in kumauni bullocks. It may be that large amounts of cobalt and manganese if offered simultaneously, may result in a better utilization of these essential ingredients of the diet.