CHEMICAL ANALYSIS OF SOME NIGERIAN FOOD-STUFFS

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BY

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D E C L A R A T I O N

I hereby certify that the work herein described is the result of my own investigation. It is not being and has not been submitted elsewhere for the award of any other degree.

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SUPERVISOR

D E D I C A T I O N

TO MY WIFE AND CHILDREN

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ABSTRACT

Ten different types of bread samples were used in this study. The bread samples were obtained fresh from bakeries in Zaria (Nigeria) and some of its surrounding villages. Each bread sample was air-dried at room temperature. The temperature and relative humidity at this time averaged 21.5°C and 15% respectively (Samaru weather report, January, 1979). Duplicate samples of each bread type were used for each analysis for the following determinations:-
Water content, total solid, lipids, total and reducing sugar, ash, iron, calcium, sodium, potassium, chloride and phosphorus.

The fruits and vegatables were obtained as sold in the markets at Samaru (Zaria) and Sabon Gari (Zaria). They were analysed for minerals (metals) such as sodium, potassium, iron, calcium, manganese, zinc, magnesium and nickel.

Different brands of milk (twenty-seven in number) including fresh milk and yoghurts were analysed for the same metals as in the fruits and vegetables.

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OBJECTIVES OF THE ANALYSES

This work is intended to throw some more light into our knowledge of nutrients in a number of foods eaten in Nigeria as obtained locally in and around Zaria.

The art of bread making (mixing the basic ingredients of flour, water, butter and sugar in such proportion as to obtain bread with the right texture is known by any baker worth his salt. However, what makes a high quality bread-excellence of texture, high keeping quality, and smell, as well as nutritional value, depends on mixing the basic ingredient in the correct proportion as well as enrichment with such foods as egg, milk, meat, fruits etc.

It is known that most bread baked in Nigeria and in many other countries do not have any form of enrichment as mentioned above. Hence, there is good ground for comparison between the results of the present work and those for similar breads in other countries. The work is also aimed at finding how good bread is as a source of some important nutrients such as iron, calcium, phosphorus sodium and potassium.

For the fruits and vegetables, the number of works such as those of Oyenuga and Fetuga (1975), Bergman (1971),

Sai (1965) and Narayana et al., (1967) are appreciated. These not withstanding, the present work on the determination of metals in fruits and vegetables is worthwhile in view of the vast number of factors that affect the mineral constituents in plants as treated later. The results obtained by Oyenuga, V.A. and Fetuga, B.L. of the Department of Animal Science, University of Ibadan, Nigeria (in 1975) are given in Appendices 6 and 7 for comparison. Another good work of reference is the collection by Good'all and Gregory (1947).

A large number of brands of milk are available, being sold today in addition to fresh milk. The present work aims at establishing how some of these compare in their content of the same minerals (most of which are nutritionally important) studied under 'Analysis of fruits and vegetables' (Chapter Three). At the same time, it will show how milk and the plant foods studied compare in their ability to supply these selected minerals.

CHAPTER ONE

I N T R O D U C T I O N

I History of Bread

Bread is essentially a staple food and one of the major nutritional sources of ancient and modern man. There is evidence that the art of breadbaking started as far back as 4000 B.C. (Archeological revelations around Swiss Lake habitations), and even before that in Egypt. There are a number of biblical references to bread such as 'In the sweat of thy brow shall thou eat bread', a statement from God to Adam.

The Ancient Egyptian carried the rudimentary art to high perfection while Greece and Italy also continued the development of bread making and around 171-168 B.C., public bake houses came into being. The Romans however continued to regard breadmaking as a housewife's art (Spicer, 1975).

The early wheat was quite different from the ones we know today. It was actually Emmer or Spelt wheat with narrow pointed grains.

In London the bakers formed a brotherhood as early as 1155 A.D. and were incorporated in 1307 into two distinct bodies - The Company of White Bakers and the Company of Brown Bakers, according to the colours of their bread.

Mechanization gradually took over towards the end of the 19th century. The selection of wheats with the best baking qualities (Science of bread-making) started around 1950. Mixers had been developed some 25 years before that enabled production of quality bread without bulk fermentation. Inspite of technological help, baking is still largely an art craft with many uncertainties plaguing the baker till today.

II. Sociological aspects of Bread

Bread is a relatively cheap food item which most families in Nigeria can afford even if once in a while. It is a normal inclusion in the breakfast menu of an average Nigerian family. In hotels with standard menu bread is usually served as part of the first course. In many a gathering where food is

served, bread in various forms such as sandwich, toasted or fried bread is usually not left out.

Most people (the world over) prefer white to brown bread because the former tastes better and has more pleasant appearance although brown bread is known to contain higher proportion of nutrients (white bread is usually from 70% extraction flour while brown bread is from whole wheat or 95% extraction flour). In communities where the average per capita income is high; the importance of bread in their diets is diminishing because of ability to purchase more refined expensive wheat products such as biscuits and cakes. Fortification of flours with vitamins, fruits and proteins is now widely practised in the mere affluent societies to boost the bread appeal to people as well as increasing the nutrient value of bread, Aspects of bread fortification have been extensively treated by Chopra (1944).

III. Nutritional importance of Bread

The food constituents - water, carbohydrates, proteins, fats, mineral salts and vitamins are amply

present in bread. However, its content of phytic acid which is an anti-nutrient factor is relatively high. This acid combines with calcium and magnesium ions to form insoluble compounds. White bread contains phytic acid as much as 11.0 mg/100g, while 100% extraction flour (wholemeal) contains up to 225mg/100g. (McCance and Widdowson ., 1946).

The high carbohydrate content (energy source) makes bread a staple food in many parts of the world. Apart from carbohydrates, the other organic constituent of bread of considerable interest is protein, wheat endosperm contains a mixture of several proteins. The hard winter and hard spring wheats produced in northern parts of America have proteins averaging 12% or a little more which is ideal for breadmaking (Vogel et al, 1973; Akinkele and Shambe, 1974). It is known however, that carbohydrate and protein digestibility is impaired as the fibre (cellulose) content increases (Moran and Pace, 1942; Krebs and Mellanby, 1947; McCance and Widdowson, 1947). Approximate composition of wheat and products of milling are shown in

appendix 1, 2 and 3. The nutritive value of protein is also limited by its short supply of lysine which is an essential amino acid important for growth in animals as shown from experiments with rats (Bricker et al., 1945). Ordinary wheat rarely contains more than 3% lysine. Increase of lysine to more than 4% is required to bring it into reasonable balance with other essential amino acids (Johnson et al.,). Human tissues have a closely similar amino acid composition to rats and the same amino acids are nutritionally essential for each species (Allison, 1968). The slower rate of growth in humans makes children at all ages use very little protein for growth compared with maintenance (metabolic and endogenous loss of nitrogen, sweat and skin losses (Spicer, 1975).

The standard protein which is taken as whole egg protein contains 7% lysine. It has been found that human adults require not more than 2.2% lysine in their dietary protein (FAO/WHO review, 1973; Moran, 1959).

Wheat proteins are well digested (up to 90%) even by infants (Bressani et al., 1960; Graham et al., 1969). The net protein value (from wheats) which is taken as (quantity X quality) is similar to those for rice, maize and potatoes and much superior to cassava and plantain values (Spicer, 1975). None of the common methods for cooking or baking wheat products appears to cause a significant damage to the proteins. In natural mixed diets, the amino acid limitations of cereal proteins are compensated for to a large extent by the high lysine content of legumes, meat and milk proteins (Harper, 1965; Widdowmson and McCance, 1954; Moran, 1959).

Wheat protein is safe for almost all individuals except for a few who suffer from coeliac disease as a result of eating gluten (Dicke et al., 1953).

IV. Bleaching and Flour Improvers

Flours that have aged several weeks have been found to bake better and give bolder and more attractive (white) loaves than freshly milled ones. The colouring in freshly milled flour is pronounced

and is due to the yellow nature of the endosperm of wheat. To simulate aged flours, bleaching agents and flour improvers have been introduced. They are generally oxidants (France and Germany do not allow addition of chemicals to flours). These improvers include:-

Ascorbic acid $C_6H_7O_6$ (dehydro-form by enzyme action) potassium bromate $KBrO_3$

Ammonium Persulphate $(NH_4)_2S_2O_8$

Potassium Persulphate $K_2S_2O_8$

Chlorine dioxide ClO_2 (containing by volume less than 20% chlorine).

Azodicarbonamide $C_2H_4O_2N_4$.

They are believed to exert their effect by indirectly suppressing the proteolytic enzymes in flour (Balls and Hale 1935, 1936), and by oxidation of reducing substances (Shen and Geddes, 1942; Freilich and Frey, 1943).

The resultant effect of the bleach and improvers is in the improvement of the physical state of the dough (elasticity and stability) better oven development and improved crumb texture.

V . NUTRITIONAL IMPORTANCE OF FRUITS & VEGETABLES

All the chemical compounds and elements needed by humans to metabolise for healthy living are got directly and indirectly from plant sources which include fruits and seeds, roots, stem tubers and leaves.

Fruits are sources of accumulated sugars, vitamins and minerals; seeds such as beans are highly rich in proteins for body building, while groundnuts beni-seed, coconut, maize (corn) have abundant oil which is an important energy source. The yellow and green leafy vegetables have high amounts of carotene which is the precursor of vitamin A. Yam, potatoes (sweet and Irish types), guinea corn, millet, and maize which are used as vegatable foods, are very rich in carbohydrates (concentrated energy source). The vegetables also contain vitamins and minerals in varying amounts. Of the metals iron and calcium in varieties such as the spinach and rhubarb are practically or partially rendered unavailable due to the high Oxalate contents of these plants (Oyenuga and Fetuga, 1975; Oomen and Grubben, 1977).

In Nigeria, many people (especially those in the lower income group) take largely starchy foods.

To these group of people, economical source of these nutritional substances in fruits and vegetables is of critical importance to their well-being in augumenting their protein, vitamin and mineral intakes.

It has been suggested (Schmidt, 1975) that the present nutritional status of many people in tropical areas could be improved considerably by increased use of leafy vegetables. The use of mixtures of different species offers still greater advantages (Devades et al., 1964; Theophilus and Arulanantham, 1949). The human body requires seven mineral elements in macro amounts. These are Ca, Cl, Mg, K, Na, P & S and not less than eight in trace amounts - Co, Cu, Cr, Fe, Mn, Se, Zn and I (Sandstead, 1967). Calcium which is the most abundant mineral element in the human body is next only to patassium in order of abundance in leaf tissues (Schmidt 1971, Kefford, 1960). This is why green leafy vegetables are particularly important sources of calcium where dairy products are lacking in diets,

Magnesium, iron, copper, cobalt, manganese and zinc contents in leaves rank quite high among food substances (Schmidt, 1971; Watt and Merrill, 1963; Peterson and Elvehjem, 1928). The magnesium and cobalt contents of leaves are infact the highest of all known foods (Hurwitz and Beeson, 1944; Beeson, 1941).

Among the common fruits in Nigeria, Pineapple, banana, grapefruit and guava have higher postassium contents than other fruits, while pawpaw and lime contain the highest amounts of calcium. In general however, the amounts of nutritionally important minerals in fruits are much lower than those in leafy vegetables (Oyenuga and Fetuga, 1975). See appendix 2A.

VI. Factors Affecting Composition of Fruits and Vegetables

A number of factors influence the mineral composition of fruits and vegetables. These include availability of these minerals in the soil, use of fertilisers, degree of ripeness of fruits at time of harvest, place and time of cultivation, methods of

handling, transportation and marketing (when fresh items are being considered, as well as genotype (genetic factors) governing the ability of different species of the same plant to take up and concentrate minerals for use from the soil. In an experiment by E.L. Bergman of the Department of Horticulture, The Pennsylvania State University, under field conditions, six tomato cultivars were seeded, transplanted, grown side by side and sampled at the same time. Leaf analysis results (percentage of dry weight) for potassium and magnesium showed ranges of 2.39 - 3.74 and 0.55 - 0.89 respectively.

The presence of some ions such as potassium also affect uptake of other ions. Schmidt, D.R. (1971) showed that calcium and potassium ions are taken up by Amaranthus cruentus maximally at 240 ppm potassium ion concentration in the nutrient solution, while that for Mg^{2+} and Cl^{-} is maximal at K^{+} concentration of less than 60ppm in the nutrient solution. Different results were also recorded for other plants- Brassica Oleracea, Celosia argentea and Ipomea aquatica

that were also tested (Schmidt, 1971).

Age of plant also has influence on the composition of fruits and vegetables. Some elements decrease while others increase with age. Similarly, alkalinity and acidity of the soil affect plant composition (Schmidt, 1971; Doby, 1965).

VII. NUTRITIONAL IMPORTANCE OF MILK

Milk often referred to as nature's most perfect food because of its content of various nutrients in the right proportion as required by humans and other mammals, is usually the only source of food for most new born of these animals, The human infant depends solely on it for the first two or three months of life. It also constitutes a good proportion of the food of the growing child. At the old age, humans can derive immense nutritional benefit by taking adequate amounts of milk.

Milk and milk products provide some 35% of all animal protein and also 35% of all energy from animal sources. As a food 0.5 litre of milk could supply the entire daily protein and calcium needs for

children under 1 year and 49% of the protein and 71% of the calcium recommended for adults (Mc Dowell, 1977). A quart (1.141) of milk which is the quantity usually recommended per day, supplies all the protein requirements for children up to six years of age and more than 60% of the requirements for growing children up to 14 years of age, and as much as 44% of that for lactating humans (Schmidt and Van Vleck, 1974). Milk protein is especially valuable because it supplies many of the essential amino acids that are often lacking in cereal grains.

Calcium, the most abundant element in humans, is a major constituent of milk. Humans which do not consume milk or its products are prone to calcium defeciency diseases since adequate calcium can not be guaranteed in the human diet if milk is excluded. A quart (1.141) milk taken by pregnant or lactating women provides almost all of their calcium needs. The element (calcium) is particularly important in the diet of the elderly as well, since it diffuses out of the bones unless it is consumed daily. Failure to do

so leads to bone problems of the spine and hips.

Riboflavin and vitamin A which are the vitamins most likely to be defecient in human diet are abundant in milk. A quart supplies all of the riboflavin needs of growing children and adults except pregnant and lactating females and almost all of the vitamin A requirements of children less than one year old (Schmidt and Van Vleck, 1974).

VIII. FACTORS AFFECTING COMPOSITION OF MILK

The one fact that can be stated indisputably about milk composition is variation. Milks of different species of mammals vary greatly in composition. Within a given species, variations occur due to numerous factors which have been investigated by several workers which include Whittle (1943), Webb and Johnson (1965), and Armstrong (1959), who have shown the effect of breed on milk composition. The principal breeds of cow in U.S. that were used are - Ayrshire, Brown Swiss, Guernsey, Holstein, Jersey and Guernsy - Holstein. The effect of seasonal changes, lactation period,

temperature, and feed types have been demonstrated by Nickerson (1960), Macy et al., (1953), Cobble & Herman (1951).

Other factors that have been found to affect milk composition include - intervals of milking (Schmidt, 1960), milk from different quarters of the udder (Benton, 1929) and age of the cow (legates, 1960).

IX. MILK PRODUCTION IN DEVELOPING COUNTRIES

The ideal habitat for efficient and profitable industry for milk production is to be found in the humid and temperate countries. The tropical climate which is prevalent in many of the developing countries does not favour this type of animal husbandry and the associated fodder production. A lot of investments have to be made to overcome the limitations imposed by these adverse environments largely by changing traditional standards of animal feeding, management and disease control. This in turn means that farm workers in the tropicsneed higher level of knowledge and experience than is necessary among similar workers

in temperate zones, yet it is the reverse that obtains (Whyte, 1967).

It is to be noted however, that although the tropics or subtropics are not naturally suitable for milk production a lot of the concentrate feeds used by dairy farmers in temperate regions come from the tropical and subtropical countries. In the long run if the large scale practice of this animal husbandry still proves not viable in the tropics, then the choice will be to concentrate on production of feeds for cows in the tropics while the animals remain largely reared in the temperate countries. However, the suggestion that the traditional cattle belt in the northern states of Nigeria can be developed for breeding calves while the sub-humid zones with greater pasture potential can be utilized for fattening the cattle is also very relevant (Nuru, 1980).

X. BIOLOGICAL IMPORTANCE OF ALL MINERALS STUDIED

Many of the mineral elements determined in these samples are extremely important to animals and plants.

Sodium (Na^+) and potassium (K^+) are the main

extracellular and intracellular cations respectively. Both are involved in the active transport (Na^+-K^+ pump) of glucose and amino acid into cells against a concentration gradient. They are also involved in nerve impulse transmission in animals. Potassium, Calcium and Magnesium ions catalyse enzymes (ATPases) in muscular action (Oser, B.L.). Calcium in plants is essential for apical and root-tip development and is a principal element in skeleton of many animals (Allen, 1974; White, Handler and Smith (1968). Magnesium is needed by all organisms since it activates many enzyme systems. As Mg-ATPase chelate it catalyses all hexokinase reactions (ie. the phosphorylation of hexozes (6-Carbon sugars) in glucose metabolism) in all living cells. Chlorophyll which is necessary for photosynthesis in plants is a porphyrin chelate of magnesium (White, Handler and Smith; Vines & Rees, 1972). Iron as Fe^{2t} is linked to electron transport enzyymes involved in biological oxidations. Haemoglobin of blood is an iron-porphyrin chelate. Zinc is linked to the dehydro-

genases that catalyse many biological oxidations, while manganese is a cofactor for acetyl coenzyme A carboxylase which catalyses the first reaction in fatty acid synthesis. The significance of Nickel has however not yet been established. Phosphorus is essential in all living organisms especially as a constituent element of the ATP-ADP (Adenosine triphozphate - Adenozine diphosphate) system by means of which energy is made available for metabolic processes. It is a major element of bones and shells. As PO_4^{3-} (Phosphate) it is the major intracellular anion. Chlorine as Cl^- ion is important for ionic balance in cells and for osmosis. It is among the chief extracellular anions (vines and Rees, 1972).

CHAPTER TWO

ANALYSIS OF BREAD

2.1: NAMES AND ADDRESSES OF DELIVERY BREAD

SAMPLE NO.	NAME AND ADDRESS
1.	Agoro Bread(Slices), Tudun Wada, Zaria
2.	City Bread, No. 13, Ondo Street, Sabon Gari, Zaria.
3.	Omas United Bread, Sabon Gari, Zaria.
4.	Nasara Bread, Samaru, Zaria.
5.	Temidire Bread, Sabon Gari, Zaria.
6.	Yoka Bread, Funtua.
7.	Dan Mama Bread, Giwa.
8.	Manza Bread (rolls), Sabon Gari, Zaria.
9.	Vita Bread (rolls), 6, Ondo,Street, Sabon Gari, Zaria.
10.	Omas United Bread (rolls), Sabon Gari, Zaria.

2.2: DETERMINATION OF WATER CONTENT:

The water in cereals and their products can be taken as consisting of free and bound water. Since it is not possible to draw a line of demarcation between them, the determination is thus empirical, requiring strict control of condition which must be adhered to for purposes of comparing results. In this determination however, absolute moisture content is not sought for. The method employed here and those in subsequent determinations, except where otherwise indicated, are from 'Official Methods Of Analysis Of The Association Of Official Analytical Chemists' 11th edition, 1970. Editor - Horwitz, W.

Alternative methods, dependent on availability of instruments and reagents as well as convenience, are contained in works by Kent - Jones and Amos (1967), Hart and Fisher (1971), Pearson (1976), Fosnot and Haman (1945) and Kent(1975)

These include:-

(1) Direct distillation with reasonably volatile materials such as toluene, xylene, or from non-distilling mineral oils as in the Brown - Duvel Method.

(2) Vacuum ovens, air jacketed, De Bruyn type etc.

(3) Electrical (resistance or dielectric) method. This is very fast but is less accurate.

(4) Fosnot-Haman chemical method using the Karl-Fischer reagent.

(5) Infra red radiation technique.

METHOD

Apparatus:

Mettler balance (sensitive to 0.1g)- Gallenkamp Model P2010; knife (stainless steel) and large white sheets of paper.

Procedure:

Each loaf of bread was removed from the polythene containers and weighed immediately on delivery. It was cut into slices 2 - 3 mm thick and spread on paper to dry at room temperature for about 36 hours. All the slices for each bread type were then weighed. This procedure was repeated for a second set of bread and the average of the water content obtained from the two determinations.

Calculation:

Wt. of bread before drying = a g

Wt. of bread after drying = b g

Percentage water = $\frac{a - b}{a} \times 100$

2.3: DETERMINATION OF TOTAL SOLIDS

METHOD

Procedure:

The air-dried (crisp) slices of each bread type were ground well in mortar to pass No. 20 sieve and kept in air-tight containers.

About 2g of the powdered bread was accurately weighed in a dish with cover (previously) heated to 130°C and cooled). The sample was uncovered and the dish, cover and contents dried 1 hour in oven provided with opening for ventillation and maintained at 130° ± 3°C (1 hour drying period began when oven temperature was actually 130°C). The dish was covered while still in oven. It was transferred after the heating period to a desiccator and weighed soon after reaching room temperature.

Calculation:

(a) % Total solid in air-dried bread. $= \frac{\text{Wt. of bread before oven-drying}}{\text{Wt. of bread after oven-drying}} \times 100$

(b) % Total solid in bread as purchased = (a) x (100 - % water content in air-dried bread).

2.4: DETERMINATION OF LIPID CONTENT:

Fat in bread (and in other cereal products) exist in many forms - free, bound to proteins etc, When baked cereal products are being analysed, it is not possible to extract the fat by means of direct extraction using petroleum ether except for an approximate determination (Kent-Jones and Amos, 1967). Acid hydrolysis is best for total (free and bound) fat. Chloroform - methanol mixture in the ratio 2:1 is also recommended although the extract may contain phospholipids, lecithins, sterols, fatty acids & carotenoids (Hart and Fisher, 1971), Pearson, 1976), (Kent-Jones and Amos, 1967).

The method employed here is the direct but exhaustive Soxhlet Method of extraction using petroleum ether (Hart and Fisher, 1971; Pearson, 1976), (Litchfield, 1972).

Other methods that are applicable are - Ammonia in alcohol (Rose-Gottlieb Process) and the rapid volumetric method (Gerber Method) - (Pearson, 1976).

SOXHLET METHOD

Apparatus:

Soxhlet extractor, heating mantle, thimble, 500 ml round bottom distilling flask, Liebig's condenser.

REAGENT - Petroleum ether (analytical reagent grade, boiling range 80° - 100°C).

Procedure:

About 5g air-dried bread was weighed accurately in a thimble of known weight. About 300 ml petroleum ether was poured into the distilling flask. The soxhlet extractor into which the thimble with sample had been introduced was fitted into the flask and placed on the heating mantle.

Heating was started and increased slowly until the solvent boiled. The vapour of the boiling solvent condensed and collected in the thimble and extractor chamber where the lipid contained in the sample was dissolved. At a certain level, the solvent with the dissolved lipid ran back into the 500 ml round bottom flask. The heating and hence the extraction process was then continued for about 8 hours. The thimble with its contents was then removed, dried in an oven maintained at 50°C for 24 hours. It was transferred to a desiccator and weighed soon after reaching room tempera-ture. The amount of lipid extracted is obtained thus:-

Wt. of thimble = a g

Wt. of thimble + sample before extraction = b g

Wt. of thimble + sample after extraction = c g

Wt. of Lipid extracted = (b - c)g

$$\% \text{ Lipid} = \frac{(b - c)}{(b - a)} \times 100$$

2.5: DETERMINATION OF PROTEIN CONTENT

The amount of protein in a biological material is obtained by determining the amount of nitrogen present which is then multiplied by known and accepted factors (based on the proportion of nitrogen in the proteins). The factor of 5.7 is used for proteins in flour and in wheat.

In this analysis volumetric method of Kjeldahl (Horwitz, 1970) with semi-mirco modification (Pearson, 1976) is employed, and is relatively fast and most appropriate. The determination is made in two main stages - first, the sample is digested with conc. sulphuric acid and other additions that rapidly convert all the nitrogen into ammonium sulphate $(NH_4)_2SO_4$, and after making the mixture alkaline, the ammonia thus obtained is them determined by distillation and titration of the distillate.

Some workers have used modifications of the Kjeldahl method such as digesting with sulphuric acid - Hydrogen peroxide mixture (Allen, 1974). Also, Hashmi et al.,(1962) eliminated the distillation process by adding alkali to the digest followed by hypobromite solution. Potassium iodide is added and acidified. The liberated iodine is titrated with thiosulphate.

Other methods of protein determination include

the formol titration (Pearson, 1976), (Taylor, 1957), colorimetric (dyebinding) method of Udy (1954, 1956) and Turbidimetric method of Feinstein and Hart (1959) using sulphosalicylic acid.

KJELDAHL METHOD

Apparatus:

500 ml Kjelhahl flask

Kjeldahl distilling bulb

Straight - tube condenser

Heating mantle

REAGENTS

All the reagents used were of analytical reagent grade and free from nitrogen.

$CuSO_4.5H_2O$

K_2SO_4(anhydrous)

H_2SO_4(Sp.g 1.84)

Boric acid (2%m/v)

NaOH (40%m/v)

HCI (0.02M)

Mixed indicators - 0.2g Bromocresol green and 0.2 g methyl red ground together and dissolved in 200 ml ethanol.

Procedure:

About 2 g air-dried bread was weighed accurately

into Kjeldahl digestion flask. 10g powdered potassium sulphate, 0.5g copper sulphate and 25ml sulphuric acid were added. The flask was inclined and heated gently until frothing ceased. It was then boiled for four hours to obtain a clear solution which was allowed to cool.

The digested material was transferred into 250ml flask with about 100 ml water and made to mark, into the distilling flask 8ml of the 40% NaOH was placed and 10 ml of the sample solution added. 15ml of 2% boric acid was placed in the receiving flask with few drops of mixed indicator. With a current of steam, the liquid in the distilling flask was kept boiling throughout the distillation.

When the liquid in the receiving flask has reached the 75 ml mark, the distillation was stopped. The ammonia trapped in the boric and solution ~~is~~ was titrated against 0.02M HCl. Water was used as a blank in place of the sample solution.

% protein is given by -

$$\% \text{ protein} = \frac{M.V_1.V_2.1.4.5.7}{V_3C}$$

M = Molarity of HCl (millimoles).

V_1 = Volume (ml) of digested sample.

V_2 = Volume (ml) of HCl.

V_3 = Volume (ml) of sample solution distilled.

C = Weight of air-dried bread in gram.

1.4 = millimoles of N x 100.

5.7 = Factors to obtain protein from Nitrogen in bread.

2.6: DETERMINATION OF SUGAR CONTENT

Sugars are usually determined by their power to reduce copper ions in alkaline medium or by the ability of sugar solutions to rotate plane of polarised light. In the former, two methods of approach can be adopted. In one, the cuprous oxide produced by reduction of Fehling's solution under standardised conditions is weighed while in the other method, the volume of sugar solution required to reduce completely 10 or 25ml mixed Fehling solution using methylene blue as redox indicator is measured (Lane and Bynon, 1923).

Now, two main classes of sugar exist- reducing (e.g. dextrose, maltose, lactose) and non-reducing (e.g. sucrose). The latter after inversion (splitting of the complex non-reducing sugar by acid hydrolysis into simpler reducing ones), can be determined by the method for reducing sugars.

This analysis (for sugar in bread) is based on Luff-Schoorl indirect volumetric method. It is similar in principle to the method of Lane and Bynon (1923).

However, for rapid routine comparisons, a refractrometer can be used, or by methods based on specific gravities (though the latter are far less accurate) (Pearson, 1976).

LUFF-SCHOORL METHOD

Apparatus:

Mixer(magnetic stirrer)

REAGENTS - (All are of analytical reagent grade)- 40% ethanol.

Carrez solution I:21.9g zinc acetate and 3g glacial acetic acid in 100ml water.

Carrez solution II: 10.6g potassium ferrocyanide in 100ml water.

0.1% w/v methyl orange

4 M hydrochloric acid

0.1 M hydrochloric acid

0.1 M sodium hydroxide

Luff Schoorl reagent: a,b,c below used.

a. 25g Copper Sulphate in 100 ml water

b. 50g citric acid in 50 ml water

c. 143.8g anhydrous sodium carbonate dissolved in about 300 ml warm water, then cooled.

The citric acid solution (b) was poured into the sodium carbonate solution (c) with stirring. Copper Sulphate solution (a) was added and made up to 1 litre

with water (PH of solution was approx. 9.4)

9. Sodium thiosulphate solution 0.1 M
10. Starch solution: 1.0g soluble starch in 5ml water was poured into 200ml boiling water boiled for 3 minutes and allowed to cool (2mg mercuric iodide may be added as preservative).
11. Potassium iodide (30% w/v)
12. Sulphuric acid 3 M

Procedure:

EXTRACTION OF SAMPLE

2.5g of the sample was accurately weighed and placed in a 250 ml volumetric flask. 200 ml 40% v/v ethanol is added and mixed in a tumbler for 1 hour 5 ml Carrez Solution I was added, stirred for a minute, followed by 5 ml Carrez solution II and stirred again for one minute. It was made up to volume with 40% ethanol, homogenised and fitered. 200 ml of the filterate was removed, evaporated to approximately half volume in order to remove most of the ethanol. The residue was transferred quantitatively to a 200 ml volumetric flask with warm water, cooled and made up to mark. This solution used to determine the amount of reducing sugar and after inversion, of total sugar.

DETERMINATION OF REDUCING SUGAR

Using a pippette, 25 ml Luff-Schoorl reagent was

transferred to a 300 ml Erlenmeyer flask and 25 ml of the sugar solution added together with some boiling chips. It was heated over a flame, stirring by hand, and bringing the lquid to the boil in about 2 minutes. A reflux condenser was fixed to the Erlenmeyer flask and the liquid was again boiled for exactly 10 minutes. It was cooled immediately in cold water and after approximately 5 minutes, it was titrated as follows:-

10 ml potassium iodide solution was added carefully, followed by 25 ml 3M sulphuric acid. It was titrated with 0.1M sodium thiosulphate until a dull yellow colour. Five drops of the starch solution was added and the titration continued to completion.

The same titration was carried out on a 25 ml Luff-Schoorl reagent, 25 ml water, 10 ml patassium iodide and 25 ml 3M sulphuric acid as blank.

Calculation:

Using the Table under Appendix 5, the amount of glucose in mg which corresponds to the difference between the values of the two titrations is obtained, and the result expressed as percentage of the sample weight.

DETERMINATION OF TOTAL SUGAR AFTER INVERSION

50 ml of the clarified sugar solution was taken with pippette into a 100 ml volumetric flask. A few drops methyl orange solution was added. 4M hydrochloric acid

solution was also added carefully with stirring until the liquid turned a definite red. 15 ml 0.1M hydrochloric acid was added and the flask immersed in a fast boiling water bath and kept there for thiry minutes.

It was then cooled rapidly to about 20°C. 15 ml 0.1M sodium hydroxide was added and made to 100 ml with water. 25 ml of the solution was removed and the content of sugar (as reducing sugar) was determined by the Luff-Schoorl method.

2.7a: ASH CONTENT (JAOAC)[9]

5 g air-dried bread was accurately weighed in shallow, relatively broad silica or porcelain dishes which had been ignited previously, cooled in a desiccator and weighed soon after reaching room temperature.

A few drops olive oil was added to sample to prevent spattering and was ignited in furnace gradually to about 550°C (dull red). A light gray or white ash resulted (ashing overnight or for 24 hours). The ash was cooled in desiccator and weighed soon after reaching room temperature. The percentage ash is given by -

$$\frac{\text{Weight of Ash}}{\text{Weight of air-dried bread}} \times 100$$

2.7b: PREPARATION OF ASH SOLUTION:

The weighed ash was dissolved in HCl(1+2 V:V) and evaporated to dryness. The residue was re-dissolved in HCl(1+4V:V) and filtered into volumetric flask.

The filter was washed thoroughly and diluted to volume.

2.8: DETERMINATION OF IRON CONTENT:

In this analysis the total iron (Fe^{2+}&Fe^{3+}) in the various bread types was determined from the solution of their ash by spectrophotometric o - phenanthroline method at about 510 nm after reduction with hydroxylanime hydrochloride (J.A.O.C)[27,28].

Other methods are available based on gravimetry by precipitation with bulky organic precipitants such as cupferron (the ammonium salt of nitrosophenylhydroxylamine), hexamine (hexamethylenetetramine) and 8 - hydroxyquinolinate (oxinate) but are slower, cumbersome and are interferred by numerous other metal ions (Vogel, 1961). A polarographic method is also suitable for the determination since iron forms a complex with solochrome violet which is readily reduced at dropping mercury electrode to give a sensitive wave at - 0.80V (Allen et al., 1974), (Willard and Dean, 1960).

METHOD

Apparatus:

Spectronic 20(BAUSCH & LOMB - model 33:31:72)

REAGENTS:

a. Iron Standard Solution:

0.01 mg Fe/ml

3.512g ferrous ammonium sulphate

$Fe(NH_4)_2.(SO_4)_2.6H_2O$ dissolved in water, two drops of HCl added and diluted to 500 ml. 5ml of this solution was further diluted to 500 ml in volumetric flask.

b. Orthophenanthroline Solution:

0.1g of the compound ($C_{12}H_8N_2$) dissolved in 80 ml water at 80°C, cooled and diluted to 100 ml.

c. Hydroxylamine hydrochloride solution:

10g $NH_2OH.HCl$ was dissolved in water and diluted to 100 ml.

d. Magnesium nitrate solution:

50g $Mg(NO_3)_2.6H_2O$ dissolved in water and diluted to 100 ml.

e. Acetate buffer

8.6 anhydrous sodium acatate (previously dried at 100°C is dissolved in water and 12 ml acetic acid added then diluted to 1000 ml.

Procedure:

Into a silica dish, 10g dried bread was accurately weighed and ignited in a furnace at about 550°C until light gray ash got and was then cooled. The ash was moistened with 0.5 - 1.0 ml magnesium nitrate solution or with redistilled nitric acid. It was evaporated to dryness on water bath and carefully ignited in furnace (raising the temperature gradually until at 550°C). On

cooling, 5 ml HCl was added letting acid rinse upper portion of dish. It was evaporated to dryness on steam bath and the residue dissolved by addition of 2 ml HCl accurately measured. It was heated for 5 minutes on steam bath, covered with watch glass. The watch glass was rinsed with water into dish, filtered into 100 ml volumetric flask, cooled and diluted to volume.

10 ml aliquot of the solution was added with pippette into 25 ml volumetric flask and 1 ml hydroxylamine hydrochloride solution added. After a few minutes, 5 ml buffer solution and 1 ml O - phenanthroline solution were added, diluted to mark with water and shaken.

The intensity of colour was read in spectrophotometer (spectronic 20) at 510 nm. From the graph of standard curve the maount of iron in each sample was calculated as mg/100g.

PREPARATION OF STANDARD CURVE

Ten solutions containing 0.0, 2.0, 5.0, 10.0, 15.0, 20.0, 25.0, 30.0, 35.0 and 40.0 ml respectively of the final diluted standard solution (a) plus 2 ml HCl were prepared in 100 ml volumetric flasks.

10 ml of each of these solutions was used and proceeding as above beginning 'ml hydroxylamine hydrochloride solution was added .....' Scale readings were

plotted against concentration of iron which were 2.0, 5.0, 10.0, 15.0, 20.0, 25.0, 30.0, 35.0 and 40.0 g/25ml.

2.9: DETERMINATION OF CALCIUM CONTENT:

In this study calcium was precipitated as calcium oxalate, redissolved in dilute suphuric acid and titrated against standard potassium permanganate solution (JAOAC)[28]; Vogel, 1961). This method is suitable even in presence of large amounts of magnesium because the latter is not precipitated but form a magnesium-oxalate complex in solution. To check co - and post precipitation of magnesium, the precipitation of the calcium oxalate was repeated and the solutions in all cases were made dilute. Alexeyev(1969) treated this in detail. Pearson (1976) has also described a suitable volumetric method using (Ammonium purpurate) as indicator while Moss(1961) has discussed interference by ions such as Al^{3+}, Mg^{2+}&PO_4^{3-} at various levels. Basir and Melnoo (1970) have also discussed methods for determination of calcium in general.

METHOD

REAGENTS (All reagents used were of analytical grade).

0.01 M potassium permanganate.

Bromocresol green indicator 0.04% (0.1g indicator and 14.3 ml 0.01M sodium hydroxide were diluted to

250 ml in volumetric flask).

20% w/v oxalic acid solution.

Ammonium hydroxide solution (1+50 V:V).

Sulphuric acid solution (125 ml H_2O + 5 ml H_2SO_4).

Procedure:

10g dried bread was placed in a silica dish and ashed proceeding as in the determination of iron through "The watch glass was rinsed with water.....", then filtered into 400 ml beaker (50 ml from the solution used in the determination of iron may be transferred to 400 ml beaker and diluted to approximately 150 ml). 8-10 drops bromocresol green indicator were added and enough 20% sodium acetate solution to change the pH to 4.8 - 5.0 (blue). It was covered with watch glass and heated to boiling. The calcium was precipitated slowly by adding 3% oxalic acid solution, one drop every 3 - 5 seconds until the pH was between 4.4-4.6 (which is the optimum for calcium precipitation), as indicated by distinct green shade, and avoiding excess of oxalic acid.

It was boiled one to two minutes and the mixture allowed to settle until clear (or overnight). The supernatant was filtered and the beaker and precipitate washed with 50 ml ammonium hydroxide (1 + 5) in small portions using wash bottle to deliver very small stream

of solution. The point of the filter paper was broken and the precipitate and paper washed with mixture of 125 ml water and 5 ml suphuric acid at 80 - 90°C. The solution obtained was titrated at 70 - 90°C. with 0.01 M $KMnO_4$ until slight pink was obtained. The filter paper was added and the titration continued as necessary. A reagent blank titration was made as well and after correcting for the blank, the calcium content was calculated as mg/100g bread.

1 ml 0.01M $KMnO_4$ ≡ 1 mg Ca.

2.10: DETERMINATION OF CHLORIDE CONTENT

A relatively simple method of determining chlordie as precipitate of silver chloride was employed in this analysis. It is also described in (JAOAC)[6,11] and in the Textbook of Inorganic Analysis by A.1 Vogel (1961).

With careful handling, quite accurate results are obtainable with this method. The aqueous chloride solution is acidified with dilute nitric acid (HNO_3) in order to prevent precipitation of other silver salts such as phosphate and carbonate which might form in neutral solution and also to produce a more readily filterable precipitate. The precipitation action is done in as subdued light as possible with occasional stirring, Anions which give silver salts which are insoluble in dilute nitric acid must be absent. These

include Br^-, I^-, SCN^-, S^{--}, $S_2O_3^{--}$, $Fe(CN)_6^{4-}$ and $Fe(CN)_6^{3-}$ (Vogel, 1961).

Volumetric methods which are equally applicable include -

(a) Volhard's indirect titration method where excess standard silver nitrate employed, is back-titrated with standard ammonium or potassium thiocyanate (nitrobenzene added to prevent silver chloride going into solution).

(b) Mohr's direct titration using standard silver nitrate and potassium chromate as indicators.

(c) Mercuric nitrate method using diphenl carbazone or the mixture with bromophenol blue as indicator.

(d) Two spectrophotometric methods for trace amounts of chlorde ions are - mercuric chloroanilate and mercuric thiocyanate methods.

METHOD

Apparatus: Gooch Sintered crucible

REAGENTS- 10% w/v $AgNO_3$; HNO_3(1 + 9 V:V) Analytical grade reagents

Procedure:

5g dried bread was ashed and dissolved in HNO_3 (1+9 V/V). The solution was filtered off any residue which was then washed a few times with hot water and/ chloride

determined in the combined filterate and washings.

To the combined solution 10% silver nitrate solution was added avoiding more than slight excess. It was then heated to boiling, while being protected from light and allowed to stand until precipitate coagulated. This was filtered on a weighed gooch (sintered) crucible previously heated to 140° - 150°C and washed with hot water. The washing was tested to prove excess of $AgNO_3$.

The percentage NaCl in sample is given by:

% chloride in ash X % ash in sample $\times \frac{58.5}{35.5}$.

2.11: DETERMINATION OF PHOSPHORUS CONTENT (AS P_2O_5)

The method employed here is the relatively simple but sensitive spectrophotometric molybdovanadate one (Kitson and Mellon, 1944).

Other methods (Vogel, 1961). That are useful depending on whether the phosphate concentrations are high or low, include:-

i) Gravimetry: Phosphate is determined as Magnesium ammonium phosphate ($MgNH_4PO_4.6H_2O$) or as the pyrophosphate ($Mg_2P_2O_7$). When other ions are present which might be co-precipitated, it is determined as Ammonium phosphomolybdate in warm nitric acid solution.

ii) EDTA Titration: Phosphate is first precipitated as $Mg(NH_4)PO_4.6H_2O$ in the presence of excess

EDTA (Ethylene diamine tetra-acetic acid as a sodium salt) so as to complex with all polyvalent metal cations. The precipitate is redissolved in HCl (dilute), excess standard EDTA added, and back-titrated with standard magnesium salt (Erio-T as indicator).

iii) Spectrophotometric molybdenum blue method except phosphate levels are very low and IV, Nephelometric method (strychnine phosphomolybdate suspension) most sensitive where phosphate concentration is not more than 100 mg/litre (Allen el al., 1974).

METHOD

Apparatus: Spectrophotometer (SP 1750 Ultraviolet spectrophotometer, PYE UNICAM)

REAGENTS:

1. MOLYBDOVANADATE Reagent.

In hot distilled water (900 ml), 60g ammonium molybdate was dissolved, cooled and diluted to 100 ml. 1.5g metavanadate was also dissolved in 690 ml hot water and 300 ml nitric acid added. It was cooled and diluted to 1 litre. The molybdate solution was gradually added to vanadate solution with stirring. It was stored in polythene bottle at room temperature (the solution is discarded if precipitate forms).

2. Phosphate Standard Solution

a) Stock Solution: 0.5 mg P_2O_5/ml

0.2397g pure and dried (2hr. at 105°C) primary standard KH_2PO_4 (Potassium dihydrogen orthophosphate) was dissolved in water and diluted to 250 ml.

b) Working Solution:

0, 1, 2, 3, 4, 5, 6 and 7 ml stock was diluted to 100 ml to obtain 0,0.05, 0.10, 0.15, 0.20, 0.25, 0.30 and 0.35 mg P_2O_5/10 ml respectively.

Preparation of Sample Solution:

Ash from 10g dried bread was dissolved in 10 ml HCl (1+3 V:V) and evaporated to dryness on steam bath. The residue was redissolved in 10 ml HCl (1 + 9 V:V) on steam bath and was transferred to 100 ml volumetric flask. It was cooled, diluted to mark and mixed (any insoluble, residue was filtered through dry filter paper). Duplicate sample weights were taken for the analysis.

Preparation of Standard curve:

From each working solution 10 ml was pippetted into a conical flask and stoppered. As rapidly as possible for the entire series, 5.0 ml molybdovanadate reagent was pippetted into each flask, stoppered and mixed.

The solutions were set aside for 10 minutes for colour development and the absorbance read for each solution within 1 hour at 400 nm (nonameter) using reagent blank.

Determination:

Into separate 25 ml conical flasks 10 ml aliquots of standard solutions containing 0.00 and 0.20 mg P_2O_5 per 10 ml are taken.

The colour is developed as for standard curve. The instrument is adjusted to 0% for 0.00 mg P_2O_5/10ml and the absorbance (A) of 0.20 mg P_2O_5/10 ml solution taken. (Absorbance of this solution should be within ± 1% of absorbance of standard curve, if not new standard curve is prepared). The colour of the sample ash solutions are similarly developed concurrently as the standard solutions.

Calculation from Standard Curve

$$\text{mg } P_2O_5/100\text{g sample} = 100 \times \frac{(\text{mg } P_2O_5/10\text{ml from curve})}{\text{g sample in 10 ml solution.}}$$

2.12, 2.13 DETERMINATION OF SODIUM & POTASSIUM

The alkali metals are easily and accurately determined by flame photometry, which is simple and specific as well as being highly sensitive (concentrations as low as 0.0005 microgram per millilitre can be detected). Although interfernces due to radiation effects from band spectra of other elements or molecules occurs, the concentrations at which these take place differ with each type of instrument. Mutaul interference between sodium and potassium is negligible in organic materials.

However, interference due to high levels of calcium can be corrected for by including corresponding levels in the standards (Allen et al., 1974; Dean, 1960). A gravimetric method based on the insoluble sodium zinc uranyl acetate is also applicable when very high levels of sodium (uncommon though in organic materials) are expected (Vogel,1961; Horwitz, 1970).

In this analysis the sodium and potassium contents of bread are determined by **flame** photometry.

PRINCIPLE:

A few atoms of an element when heated in a flame, absorb some of the heat energy and one or more electrons are promoted to higher energy levels. On reverting to the groundstate, the electrons lose this energy as light energy. For each element there are certain permitted electronic transitions giving rise to a series of lines, each series being characteristic of the element. If operating conditions are kept constant the intensity of the radiation is proportional to the number of atoms present at any given time, i.e. to the concentration of ions.

METHOD

Apparatus:

Gallenkamp flame analyser, FH-500.

STOCK SOLUTIONS

1000 ppm sodium 2.542 g NaCl (Analytical reagent grade) dried overnight at 100°C) was dissolved in water and made to 1 litre.

1000 ppm potassium 1.907 g KCl (Analytical reagent grade) dried as above was dissolved in water and made to 1 litre.

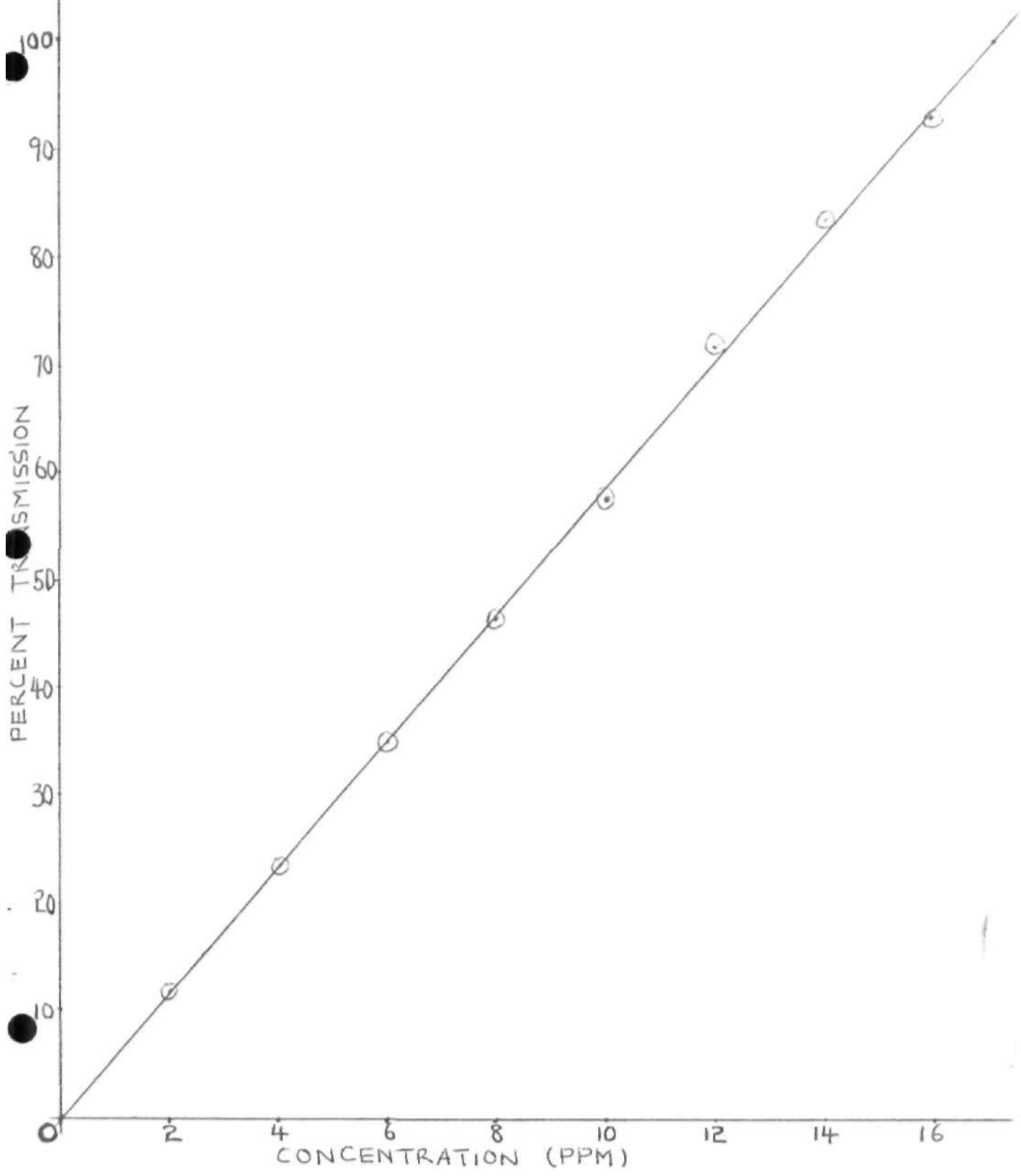
WORKING SOLUTIONS AND STANDARD CURVE

0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 ppm solutions were prepared from the potassium stock while 0 - 20 ppm were prepared from the sodium stock (10 ppm give only about 50% full scale deflection on flame photometer). A curve was prepared for the set of standards from readings on the photometer).

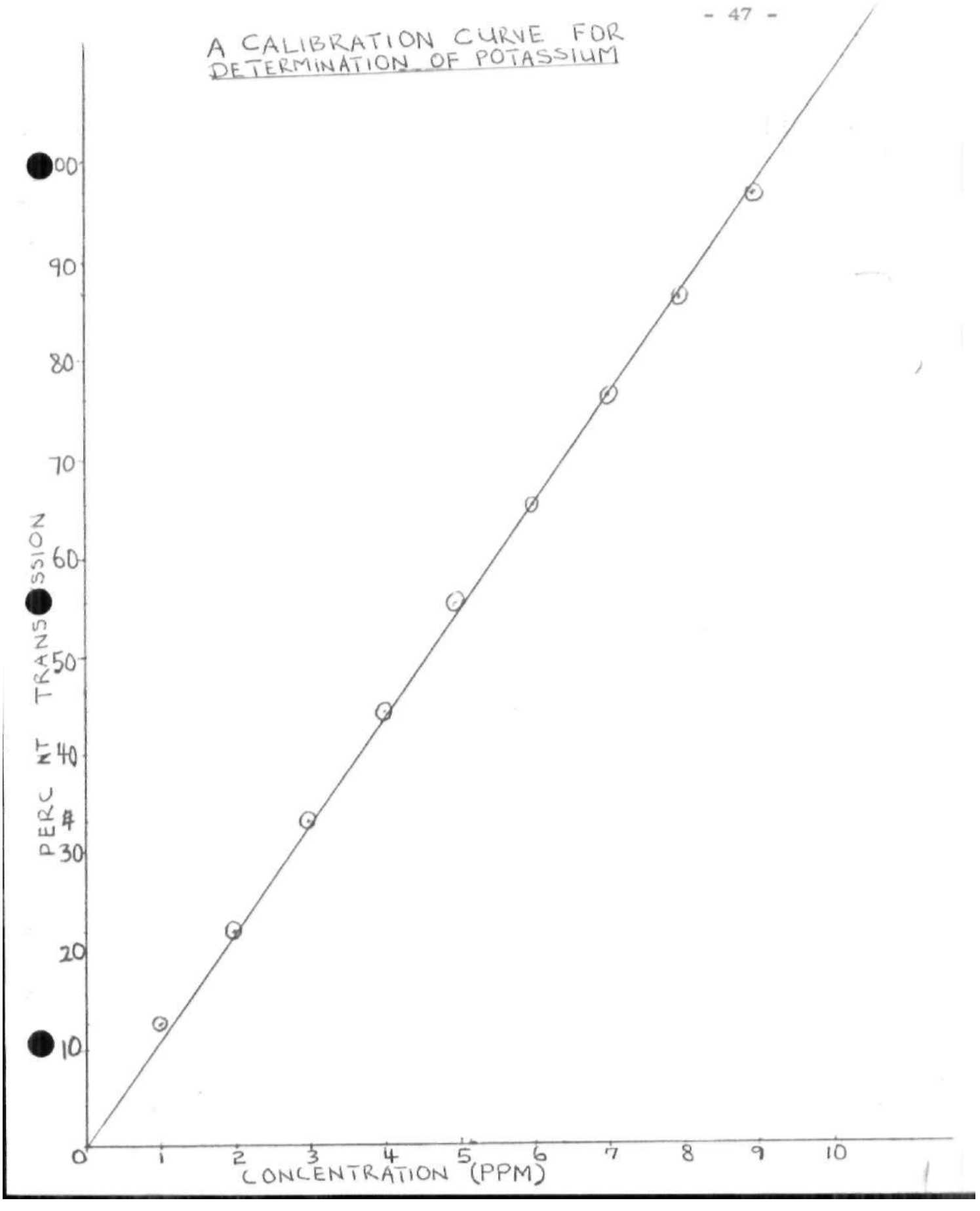
Determination:

Ash from 10g dried bread was dissolved as for phosphate above and made up to 100 ml. The sample solutions were further diluted 10 times for the sodium determination while for potassium determination they were diluted 100 times, and readings taken on the flame photometer.

A CALIBRATION CURVE FOR DETERMINATION OF SODIUM



A CALIBRATION CURVE FOR DETERMINATION OF POTASSIUM



R E S U L T S

The results for all food items analysed are expressed either as percentages, mg/100g or mg/100ml of sample as purchased.

All results are averages of at least three samples taken for analysis.

BREAD CONSTITUENTS

TABLE 1.1: SAMPLE NO	% WATER CONTENT(AIR-DRIED)
1	26.92
2	21.65
3.	28.00
4	22.90
5	27.40
6	23.20
7	23.85
8	27.10
9	21.90
10	28.30

TABLE 1.2 SAMPLE NO	% TOTAL SOLID CONTENT
1	53.90
2	60.60
3	52.85
4	58.80
5	52.40
6	58.42
7	57.91
8	53.23
9	60.42
10	52.15

TABLE 1.3 SAMPLE NO	% LIPID CONTENT
1	3.66
2	3.72
3	3.30
4	3.77
5	3.53
6	3.81
7	3.80
8	3.29
9	3.60
10	3.55

TABLE 1.4 SAMPLE NO	% PROTEIN CONTENT
1	8.73
2	8.65
3	8.45
4	8.30
5	8.25
6	9.16
7	9.00
8	8.47
9	8.85
10	8.30

TABLE 1.5: SAMPLE NO	% REDUCING SUGAR CONTENT
1	3.00
2	7.90
3	6.75
4	8.52
5	6.88
6	4.00
7	4.32
8	4.22
9	8.80
10	3.67

TABLE 1.6: SAMPLE NO	% TOTAL SUGAR CONTENT
1	3.25
2	8.08
3	6.80
4	8.67
5	6.90
6	4.08
7	4.48
8	4.25
9	8.86
10	3.84

TABLE 1.7: SAMPLE NO	% ASH CONTENT
1	1.29
2	1.72
3	1.17
4	1.19
5	0.83
6	1.13
7	1.75
8	1.52
9	1.45
10	1.35

TABLE 1.8: SAMPLE NO	IRON CONTENT (mg/100g)
1	4.4
2	2.9
3	2.2
4	2.4
5	2.8
6	4.2
7	3.1
8	2.8
9	3.7
10	2.3

TABLE 1.9: SAMPLE NO	CALCIUM CONTENT(mg/100g)
1	20.5
2	23.5
3	21.0
4	19.4
5	15.5
6	22.5
7	23.0
8	23.3
9	15.7
10	20.2

TABLE 1.10: SAMPLE NO	% CHLORIDE (AS NaCl)
1	0.41
2	0.94
3	0.52
4	0.42
5	0.29
6	0.61
7	0.79
8	0.50
9	0.80
10	0.52

TABLE 1.11: SAMPLE NO	PHOSPHORUS CONTENT (AS P_2O_5) mg/100g
1	1,461
2	1,520
3	1,804
4	1,705
5	1,891
6	1,468
7	1,384
8	1,720
9	1,368
10	1,835

TABLE 1.12 SAMPLE NO	SODIUM CONTENT (mg/100g)
1	1,279
2	2,064
3	1,858
4	1,864
5	1,757
6	1,596
7	1,633
8	1,760
9	1,520
10	1,856

TABLE 1.13: SAMPLE NO	POTASSIUM CONTENT (mg/100g)
1	174
2	252
3	174
4	182
5	188
6	192
7	128
8	179
9	278
10	182

TABLE 1.12 SAMPLE NO	SODIUM CONTENT (mg/100g)
1	1,279
2	2,064
3	1,858
4	1,864
5	1,757
6	1,596
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TABLE 1.13: SAMPLE NO	POTASSIUM CONTENT (mg/100g)
1	174
2	252
3	174
4	182
5	188
6	192
7	128
8	179
9	278
10	182

2.15 DISCUSSION

The results of the various analyses show similarity in composition of nutrients in most of the bread samples studies. This is not unexpected since all the bread types analysed, fall in the group of 'unenriched bread'. However, the bakers have exhibited the know-how of bread-making to some degree as it concerns mixing the basic ingredients in the right proportion to give bread of acceptable texture.

2.15.1 Water Content:

The water content in bread studied ranges from about 22% to 28% (Table 1.1). This represents the equilibrium moisture content for the samples under the same conditons of temperature and relative humidity. The analysis was carried out in January 1979 in Zaria (at A.B.U. Main Campus). The weather at this time of the year is usually quite dry due to harmattan winds. The average temperature and relative humidity for the month (January, 1979) was 21.5^{o}C and 15% respectively (obtained from Ahmadu Bello University (A.B.U.) Weather Station).

The value for the water content are lower than those shown in Appendix 3. This can be explained by the fact that to attain absolute water removal a much higher temperature than the room temperature (other

conditions being constant) is required. This temperature is usually taken as 130 $\pm$ 3^{o}C, which is used in determing the total solid in the bread samples (Table 1.2), which show a range of about 52 - 61% for the total solid. Allowing for the lipid content which is about 4% in all the samples, since most lipids would have been volatalised at 130^{o}C as well as volatile acids, esters, carbonyls and alcohols, the total solid would be in the range 56 - 65%. Thus the water content (absolute) in the different bread samples ranges between 35 - 44%. This range is about 5% higher than the range 32 - 36% which obtains in the bread baked in other countries shown in Appendix 3. This difference in desirable in view of the drying effects of the weather in the tropical regions.

The maximum permitted moisture content in some countries with legal standard for moisture in bread, is 38% in America and 45% in Australia (Kent, 1975).

It is to be noted however that the water requirement for a good quality bread is governed by protein content of the baking flour as well as degree of damaged starch granules during milling. The protein and damaged starch granules absorb relatively more water than do the undamaged starch granules. Hence flour from strong wheat (with higher protein content) and flour from hard

wheat (with higher damaged starch content) require more water than is needed by flour from weak (lower protein) wheat or soft (less damaged starch) wheat to make a dough of standard consistency (Kent, 1975).

The variation of water content in the day to day making of bread in a given bakery would not affect drastically the bread quality as long as it is within a narrow limit.

Although heating in a ventilated oven at 130°C for 24 hours is generally adopted for true moisture determination, a method by Sair and Fetzer involves drying in a vacuum the material for 340 hours at 40°C. This permitted the sample to retain about 1% of moisture and eliminates any chance of decomposition or loss of volatile components.

2.15.2 Total Solid

This consists of all the solid matter in bread which include carbohydrate (in the main), proteins, non volatile lipids and mineral salts. Table 1.2 shows a range of approximately 52 - 60%. Now, allowing about 13% for proteins and lipids, the carbohydrate content would be about 43 - 45% which is quite high. The food energy of bread is about 300 Kcal. per 100g. This is close to that of other staples such as corn, rice, semolina, sorghum or even whole wheat which is slightly

above 350 Kcal, in all cases (Mc Cance and Widdowson, 1946).

2.15.3 Lipid Content

The result for lipid content (Table 1.3), more appropriately petroleum - ether' extract, show very close values. This is in conformity with the small amount of lipid (margarine) needed for obtaining quality bread. The values are similar to those in Appendix 3 except for the Italian and American rye bread which have very low fat content (0.8 - 1%). This suggests similar methods of lipid extraction may have been used in the present work and in that of the other workers. Ethyl-ether' extract would give slightly higher values.

It should be noted however that petroleum - ethers' extract does not include bound lipids so that the results in Table 1.3 are quite lower than true fat content since a lot of fat is often bound to proteins which give strength to bread - by their power to absorb large amount of water to give erect loaves of desirable elasticity and good texture. The extent of bound fat may vary from one bread sample to another depending on the type and grade of flour used in the different bakeries. In Nigeria many bakers use the Golden Penny Flour from Flour Mills of Nigeria Ltd.

Total (actual or true) fat can be determined by acid hydrolysis using 70% hydrochloric acid to break down the organic matrix and free all fat which are then separated by liquid-liquid partition chromatography (Kent-Jones and Amos, 1967). Herd, C.W. and Amos A.J. (1930) have also compared lipid content of flours by employing various methods of extraction.

2.15.4 Protein Content

The protein content of bread as shown in Table 1.4 is about 8.5 - 9.0%. The closeness of the values points to the fact that none of the samples has enrichment in form of protein. Ordinary flour itself has protein content of about 11.5 - 12.5% or a little higher in some cases. The result of this analysis which is in line with those of other workers (Appendix 3), shows bread as a good source of proteins. Bread has about the same protein percentage content as rice, sorghum, corn, and barley which are all cereals. It surpasses such vegetables as broad and green beans (Vicia faba, and Phaseolus vulgaris respectively), white and sweet potato (Solanum tuberosum and Ipomea batatas respectively, as well as lettuce (Lactuca sativa) which have protein contents of between 1.0 - 2%. Many other members of the legume family seeds such as French bean (Phaseolus vulgaris), scarlet bean (Phaseolus coccineus) and

cowpea (Vigna unguiculata) are much more superior than bread with respect to protein. They have about 21 - 23% protein (Mc Cance and Widdowson, 1946).

The protein in bread derived from flour plays a very important role. It has to be right if good quality bread with correct strength and texture is to be obtained from the flour. It is the proteins which absorb water molecules to form gluten which gives bread its elasticity and yet strong enough to prevent gas escaping too readily which would lead to collapse of bread (Kent, 1975). Also, interaction of added fat with the proteins enhance this adequate gas retention.

2.15.5 Reducing and Total Sugar Content

These results are shown in Tables 1 5 and 1.6. The differences in reducing and total sugars are quite small. The bulk of the sugar in bread is that added during baking (commercial non-reducing sugar sucrose, converted to reducing fractions by hydrolysis in the baking processes). Of all the constituents analysed, the sugar is one of those that show widest variations. This is due to different bakers using amounts of sugar of their choice to make their products sweet since the sugar content would not drastically affect the bread texture. The bread with the lowest sugar content is Agoro bread (3.25%), while Vita bread has the highest

(8.86%). The variations cut across the desires of most of the consumers - there are those who prefer very sweet bread and some others who go for the not too sweet bread. The sugar composition of the various bread samples can be considered as fair and in general not detrimental to health.

Analysis of sugar calls for strict adherence to all the conditions of the experiment to obtain reproducible and reliable results since the extent of reduction of the ions in solution varies with the experimental conditions. The Luff-Schoorl method (which involves back titration against iodine liberated by reduction of remaining copper ions unreduced by sugar), is favoured since no requirement for a minute excess of reducing sugar solution is necessary to reduce indicator as in Lane and Eynon's

2.15.6: Ash Content

The result in Table 1.7 shows bread to have percentage ash content of higher than 1% except in a single case where it is 0.83%. The ash content is indicative of the total mineral component of food when water is eliminated and all organic matter burnt off. Hence it is one parameter the value of which can be considered constant in any given flour or bread since others such as sugar, fat water and protein can be altered by environmental conditions or enzyme activities.

The lower ash value in Temidire bread (0.83%) was reinvestigated by ashing samples of all the bread types baked on a different day. All the others except Temidire gave more or less the same value as in the previous determination while a value of 1.55% was obtained for Temidire (nearly twice the previous value). This may have been due to an error in the course of preparing the bread sample (instrumental etc) when it was obtained for analysis, or losses due to sudden drought in furnace used for the ashing. Non-flour (e.g. other cereal) addition by the baker may not change the ash content drastically. Another probable reason is addition of smaller quantity of salt by the baker since the ash content of bread is determined largely by the extraneous salts added rather than the mineral content of wheat (which is very low for white bread - Appendix IB).

The value of ash content (about 1.2-1.8) some what lower than those in Appendix 3 (1.7 - 2.1). This is due to the higher moisture content of the Nigerian bread (freshly baked) though the result of analysis of sodium (Table 1.12) showed high amounts of sodium salts added to the Nigerian bread. This is however made for by the much higher content of Calcium in the bread from European countries (Appendix 3).

Ash determination is affected by many factors which include types of crucibles (silica or platinum), sample size and nature (acid or alkaline), temperature and time of ashing. All the bread samples were ashed in shallow silica dishes of either 50 ml or 100 ml capacity, at a temperature not exceeding 550°C usually for 24 hours.

Wichmann in his classical series of papers on ashing techniques has made decomposition curves of ash of several agricultural products by plotting weight loss against temperatures. Potassium carbonate which predominate in plant ash is known to volatilize considerably at 700°C and almost completely at 900°C. Klemm who has surveyed up to 50 ashing procedures (AOAC, 9th ed. 1960), concluded that it is virtually impossible to in-corporate every applicable factor governing ashing of different types of food into one general method.

2.15.7: Mineral Composition

The most abundant minerals in bread as shown in Tables 1.8 - 1.3 are sodium, potassium and phosphorus. Of the other minerals analysed, iron is the least abundant. The values for iron are however, higher than those in Appendix 3. This is mainly due to the higher concentration of iron in Nigerian tap waters compared

to that of waters in European countries.

2.15.7.1: Iron Content

The iron content of 100g bread (2.2 - 1.4mg) shown in Table 1.8 makes bread a chief source of this mineral. Many food substances including milk, fruits and vegetables (see tables on pages (82-84and 98-100), are deficient in iron. The table under Appendix 3 shows the comparative value of enriched bread as supplier of certain essential nutrients (including iron). Thus apart from the natural mineral constituent of bread, it can be further enriched to meet nutritional requirements as compared to foods eaten in their natural form or cooked on harvest.

The values in Table 1.8 show a few of the bread samples having quite higher percentages of iron than the rest. This bound to happen in view of contamination from metal containers during baking and handling of bread or slicing (Agoro bread is machine sliced while some others are sliced manually).

2.15.7.2: Calcium Content

Table 1.9 shows the calcium content in bread. These values are about 25% of those shown for calcium in Appen-dix 3. This can be accounted for by the fact that except for deionised waters - the calcium content of waters in the European countries are of much higher

calcium concentration than Nigerian waters.

Two bread samples (Temidire and Vita breads) have quite low calcium content compared to the others. This could result from occasional use of water from drinking wells having low calcium concentration or use of certain grades of flours. One of them - Temidire bread was noted for its low ash content previously.

It is to be observed that adequate intake of bread can provide to some extent the requirements (for proper function of the human body) of the two elements calcium and iron discussed above, the requirements of which are (for normal adults) 800mg and 10 - 15 mg respectively - as recommended by the American National Research Council. The actual body requirement (losses due to metabolism apart) may be much less than these figures. Moore has reported 0.5 - 1mg iron as required by normal adults.

2.15.7.3: Chloride Content

This has been estimated as sodium chloride since all the chloride present may be taken as virtually having come from added sodium chloride. The values range from about 0.4 - 0.9% (Table 1.10). This represents a quantity of about $\frac{3}{4}$ -1$\frac{1}{2}$ unheaped tea spoonfuls per loaf of about 450g weight.

The wide difference in the sodium chloride content is (like in the sugar content) due to the discretion of

the different bakers to add quantities of sodium chloride as they chose as long as the taste of the bread remains pleasant. Also, in Nigeria there is no enforced legal limitation to the salt content in bread (nor the sugar content).

2.15.7.4: Phosphorus Content

The values of phosphorus content in the bread samples have been expressed as mg P_2O_5 per 100g sample (as purchased). This gives values that are not too different from actual phosphate content of the samples. The values shown in Table 1.11 indicate the high content of phosphorus in bread. The element together with calcium are the main constituents of bone and teeth, and phosphorus is abundant in many of the softer tissues of the body playing many important roles in life processes (Oser, 1965).

2.15.7.5: Sodium and Potassium Content

Apart from sodium derived from added sodium chloride, a large proportion also comes from added baking powder (which contains up to 20% or slightly more sodium bicarbonate). Tables 1.12 and 1.13 show the values for the sodium and potassium contents of bread respectively. The potassium content is similar to that in Appendix 3, while that for sodium in Table 1.12 is about twice that in Appendix 3. This can be accounted for by the use of

greater amount of table salt (sodium chloride) by the bakers here (in Nigeria), which they might have found adequate.

In plant tissues generally, it is the potassium concentration that is much higher than that of sodium. The trend in bread can easily be understood by the extraneous additions of sodium salts in the process of bread-making. Self-raising flours (which contain sodium bicarbonate incorporated are also quite popular. Since many food substances contain adequate sodium and potassium salts, deficiency in diets is uncommon. However, prolonged consumption of potassium-deficient diet could result in weakness and paralysis of muscles. Thus its presence in diet is highly desired.

CHAPTER THREE

ANALYSIS OF FRUITS AND VEGETABLES

3.1 COMMON AND BIOLOGICAL NAMES OF FRUITS AND VEGETABLES STUDIED

	COMMON NAME	BIOLOGICAL NAME
	FRUITS	
1.	Banana	Musa sapientum
2.	Grape	Vitis vinifera
3.	Guava	Psidium guajava
4.	Mango	Mangifera indica
5.	Orange	Citrus sinensis
6.	Pawpaw	Carica papaya
7.	Pineapple	Ananas comosus
8.	Plantain	Musa paradisiaca
	VEGETABLES	
1.	Baobab leaves	Adansonia digitata
2.	Beans (cowpea)	Vigna unguiculata
3.	Beniseed	Sesamum radiatum
4.	Brown beans	Vigna unguiculata
5.	Cabbage	Brassica oleracea
6.	Cucumber	Cucumis sativus
7.	Garden egg(white)	Solanum melongena
8.	Garden egg(violet)	Solanum
9.	Green beans	Phaseolus vulgaris
10.	Groundnuts	Arachis hypogaea

	COMMON NAME	BIOLOGICAL NAME
	VEGETABLES	
11.	Lettuce	Lactuca sativa
12.	Locust bean	Parkia filcoidea
13.	Okra	Hibiscus esculentus
14.	Onion	Allium cepa
15.	Sorrel (green)	
16.	Spinach	Spinacia oleracea
17.	Tomato	Lycopersicum esculentum
18.	Yam	Dioscorea rotundatum
19.	Waterleaf	Talinum triangulare
20.	'Agbono'	Irvingia gabonensis
21.	'Ayoyo'	Cochorus olitorius

3.2 MATERIALS AND METHODS

All the plant materials were bought from Samaru or Sabon Gari markets (fresh or dry) and the analyses based on weights of these as at time of purchase.

M E T H O D S

The method employed for the analysis of all the metal ions other than sodium and potassium is the Atomic Absorption method. The latter two are determined by flame photometry.

Atomic Absorption is a convenient, rapid and highly sensitive method of detecting and measuring metal ions in solution. The limits of detectability by Absorption Flame Photometry are shown in Appendix 8. The method is based on the principle that atoms whose electrons are in the ground state can absorb radiation of certain (specific) wavelengths.

A solution of an element is sprayed into a relatively cool flame in which the atoms tend to remain in the ground state: Radiation of a characteristic wavelength from a hollow cathode discharge lamp is passed through the fflame and the decrease in intensity is measured using a monochromator and detector system. This decrease is proportional to the element in solution.

After elimination of flame and chemical interferences, other important sources of error to guide against are nebuliser blockage, changes in air and fuel (acetylene gas) flow rate wavelength drifting off peak or very low acetylene cylinder pressure. Otherwise the method is extremely reliable giving reproducible reaults. The instrument used is PYE UNICAM SP 1900 ABSORPTION SPECTROPHOTOMETER.

REAGENTS

All the salts used were of Analytical reagent grade.

Iron wire was obtained from M & B Laboratory Chemicals, May and Baker Ltd., Dagenham, England. Assay not less than 99.3%.

SOLUTIONS

1. SODIUM AND POTASSIUM STANDARDS

Stock and working standard solutions of Sodium and Potassium were made as in the determination of these elements in bread, Apropriate acid extractant (1+4V:V) HCl as in all subsequent preparations were added to the standard solutions so as to mathh the samples.

2. IRON STANDARDS

Stock Solution (100 ppm Fe): 0.1g clean untarnished iron (sand paper previously used to remove tarn) was dissolved in about 10ml hot (not boiling) 10% H_2SO_4.

On cooling the solution was made to 1 litre.

Working Standards

0,0.5, 1,1.5, 2,3,4,5ml of the stock solutions were separately added into 100ml volumetric flasks and made to volume to give 0, 0.5, 1, 1.5, 2,3,4 and 5ppm solutions of the metal ion. About 1 ml H_2SO_4 was added to sample solution (to give approx. 1%^{v}/v H_2SO_4) to match standards.

3. CALCIUM STANDARDS

Stock solution (100ppm Ca): 0.2497g $CaCO_3$ (dried at 80^{o}C overnight) was dissolved in 100ml HCl (2.5%^{v}/v), heated to drive off CO_2 and made to 1 litre after cooling.

Working Standards

A range of 0-10ppm was prepared. About 0.1g (few grains) lantuanum chloride ($LaCl_3.7H_2O$) was added to the sample extracts and standard solutions to bring lanthanum concentration to about 400 ppm (this removes interference effects due to AL, P and Fe) before the solutions are aspirated for determining calcium, and later magnesium after the other metals had been determined.

4. MAGNESIUM STANDARDS

Stock solution (100ppm Mg): 1.0548g $Mg(NO_3)_2.6H_2O$ was dissolved in water containing about 1 ml H_2SO_4 and diluted to 1 litre. La $Cl_3.7H_2O$ was added to the standard solutions as above.

Working Standards

0,0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.8, 1.0ppm solutions were prepared from a 10ppm stock prepared from the original stock solution above.

5. ZINC STANDARDS

Stock Solution (100ppm Zn): 0.2087 of $ZnCl_2$ was dissolved in water and made to 1 litre.

Working Standards

0-5 ppm solutions were prepared.

6. MANGANESE STANDARDS

Stock solution (100ppm Mn): 0.3794g $MnCl_2.4H_2O$ was dissolved in water and made to 1 litre.

Working Standards

0-3 ppm solutions were prepared.

7. NICKEL STANDARDS

Stock Solution (100ppm Ni): 0.4049g$NiCl_2.6H_2O$ was dissolved in water and made to 1 litre.

Working Standards

0-3 ppm solutions were prepared.

APPARATUS

1. Thermostated ovens (Gallenkamp size one, model OV-010)
2. Muffle furnace - Gallenkamp FR-604, FR-614(0-1100^{o}C) and FR-645(0-1500^{o}C).

3. Atomic Absorption Spectrophotometer (Pye Unican SP 1900).

4. Steam bath.

PROCEDURE

About 20g dry and 50g fresh plants materials were weighed in porcelain crucibles and driedkovernight at 100^{oC} in ovens. They were then burnt over a bunsen flame (few drops olive oil added to prevent spattering) until well charred and whitish ash partly appearing. This was followed by asking in a furnace started from cold and maintained at 525^{o}C for 24 hours. Samples which ashed with difficulty (appearance of residual black particules) were treated with distilled water, dried and reashed. On complete ashing, the curcibles and ash were transferred from furnace to desiccators and weighed on cooling to room temperature.

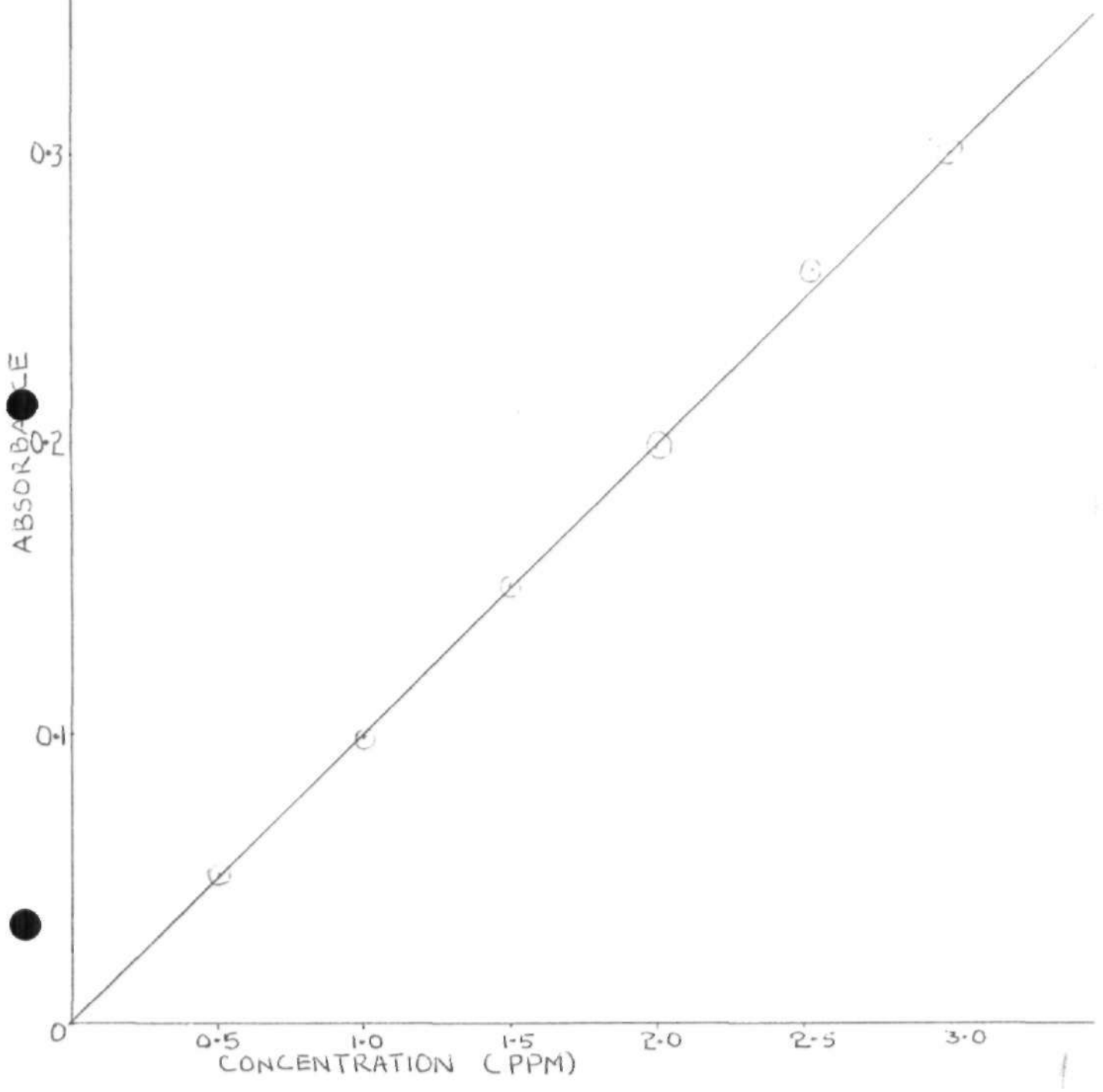
0.15g ash representing about 20g fresh sample was weighed from each sample and extracted first with (1+2V:V) HCl and evaporated to dryness over steam bath. Each is then reextracted with (1+4V:V)HCl and filtered off any residue using whatman no.2 (qualitative) filter paper and made to 100ml after addition of any reagents as indicated under solution preparations above.

TECHNIQUE OF ATOMIC ABSORPTION SPECTROMETRY

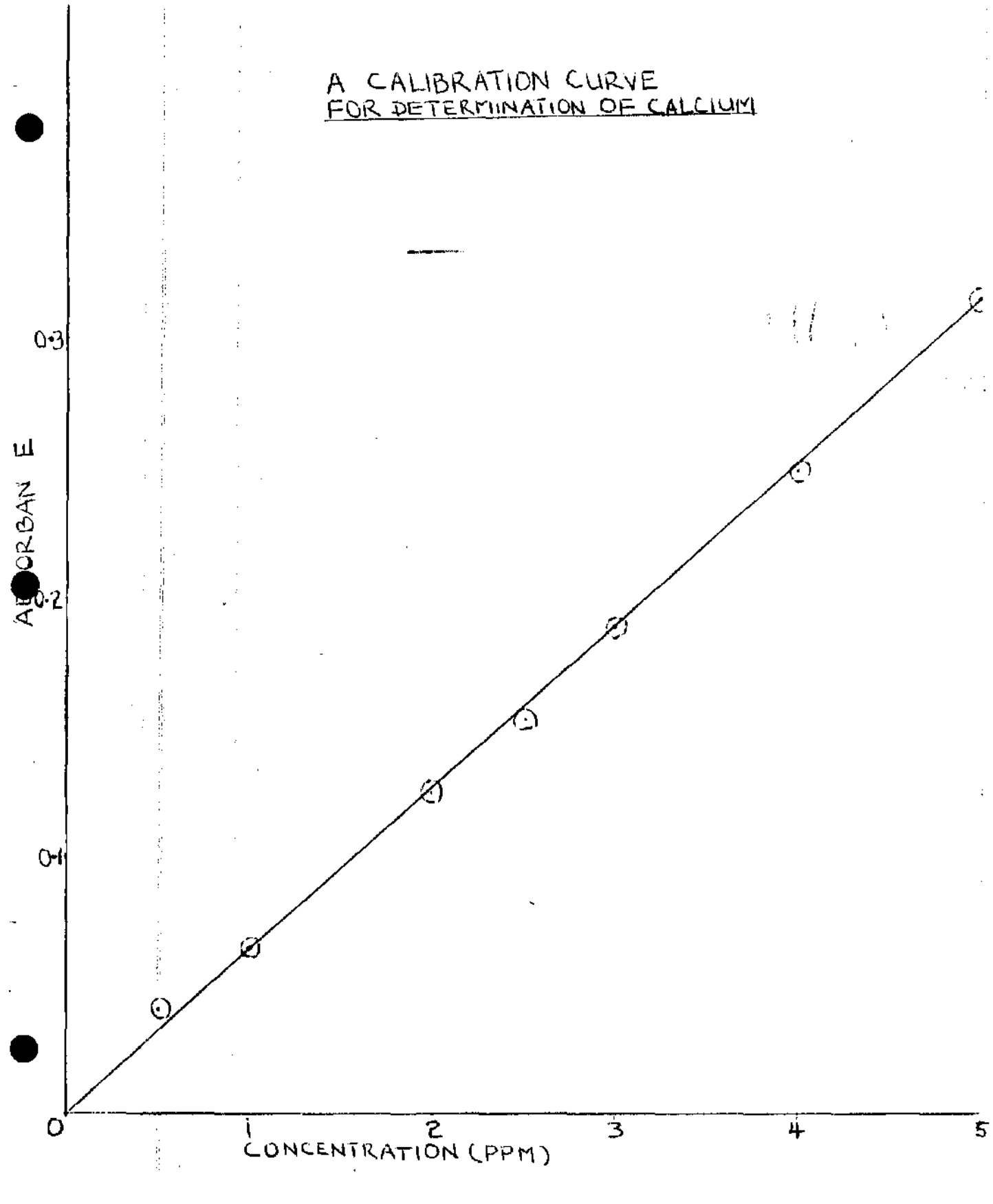
The atomic absorption instrument was switched on for some time to warm up. The fuel cylinder was opened and the air compressor switched on, while the fuel switch on the instrument remained put off. After warming up, the cathode lamp for the metal to be determined was placed in position and the correct current (midway between 0 and maximum current shown on lamp) was set. The gas switch on the instrument was sitched on and the burner lit. Each solution of standards for the element of interest was aspirated into the flame after ensuring that the energy indicator (pointer) was in a balanced position (more or less vertical). The reading for each standard solution was recorded (that is the most constant figures which appeared on the spectrophotometer). The sample solutions were similarly aspirated and readings recorded. A calibration curve was plotted for each set of standards for the different elements determined at the same time. A new calibration curve was plotted each time for any one element being determined at a different time or day from the previous determinations.

Sample solutions were further diluted as necessary eg. the solutions for determination of magnesium were diluted 100 times and further 5 times in volumetric flasks.

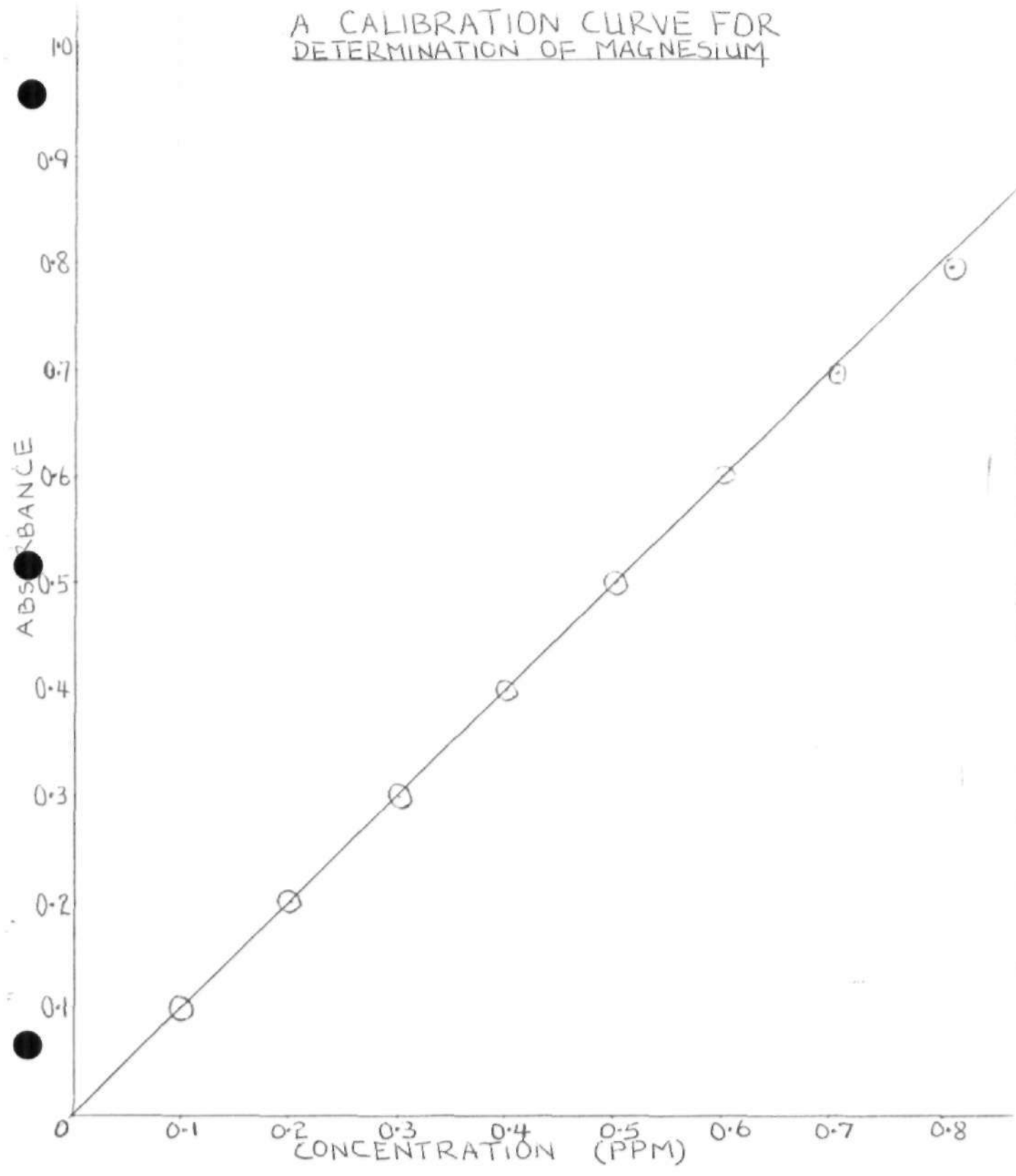
A CALIBRATION CURVE FOR DETERMINATION OF IRON



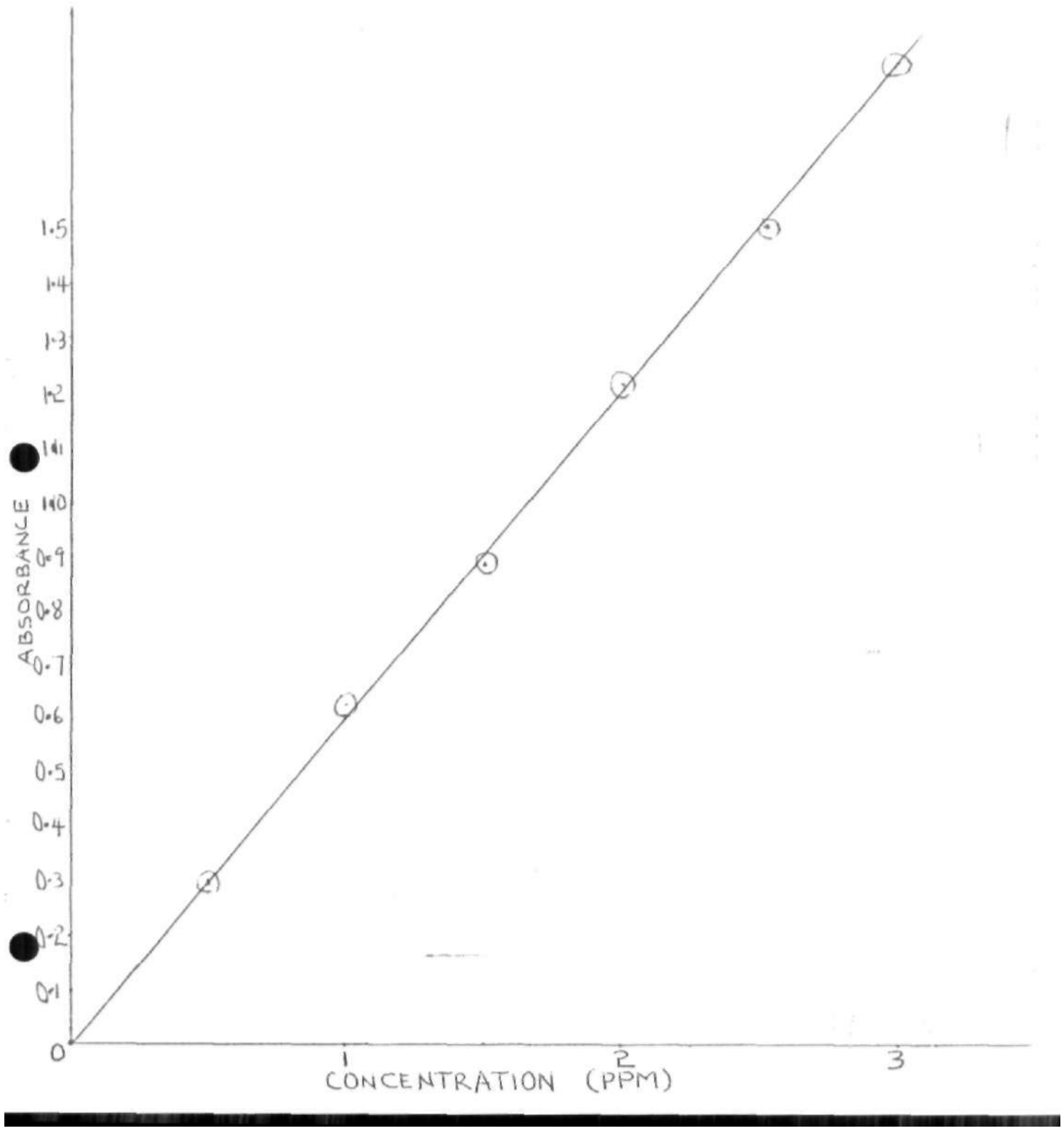
A CALIBRATION CURVE
FOR DETERMINATION OF CALCIUM



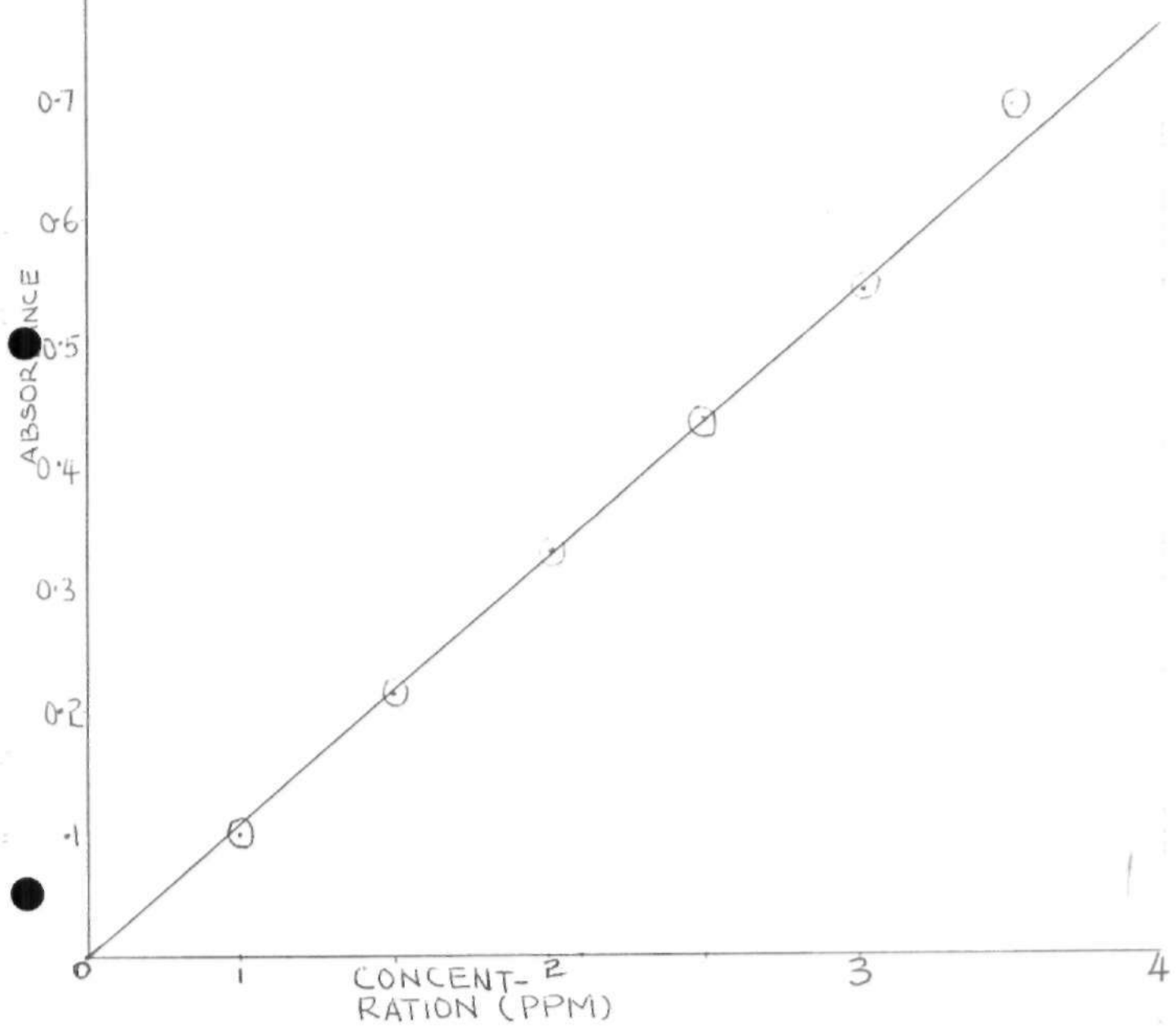
A CALIBRATION CURVE FOR
DETERMINATION OF MAGNESIUM



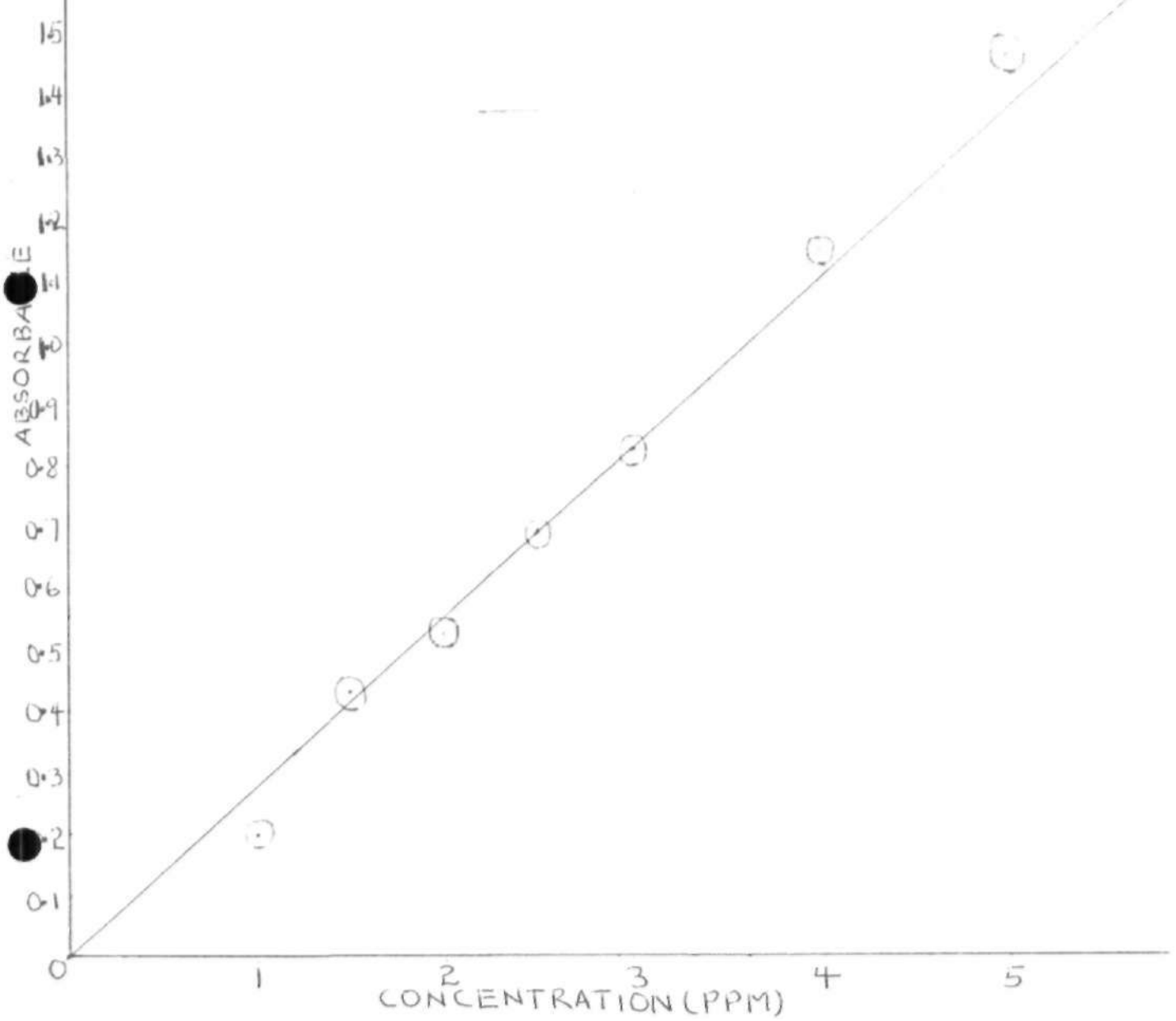
A CALIBRATION CURVE FOR
DETERMINATION OF ZINC



A CALIBRATION CURVE FOR DETERMINATION OF MANGANESE



A CALIBRATION CURVE FOR
DETERMINATION OF NICKEL



R E S U L T S

3.3.1 MINERAL CONSTITUENTS OF FRUITS (mg/100g) EDIBLE PORTION)

TABLE 2

FRUIT	ASH %	Na	K	Fe	Ca	Mg	Zn	Mn	Ni
1. Banana	0.76	18.2	294	0. 1	15.2	28.0	0.6	0. 4.	0
2.Grape	1.15	7.5	391	0. 2	48.3	42.2	0.9	Trace	0
3. Guava	0.60	2.1	276	0. 1	3.4	18.0	0.2	0. 4	0
4. Mango (1)	0.36	1.1	117	Trace	13.4	13.0	0.3	0. 2	0
5. Mango (11)	0.21	1.4	50.0	Trace	11.2	10.8	0.3	0. 2	0
6. Orange	0.45	2.5	150	Trace	50.0	14.3	0.3	Trace	0
7. Pawpaw (yellow)	0.38	12.4	106	Trace	20.8	12.0	0.3	Trace	0
8. Pawpaw (pink)	0.36	22.2	80	Trace	20.6	15.6	0.5	Trace	0
9. Plantain	WET DIGEST	5.3	426	0.1	Trace	42.0	0.3	0.5	0
10. Pineapple(s	0.41	1.5	87.5	Trace	66.7	41.0	0.2	2.2	0
11. Pineapple(m	0.34	1.9	66	Trace	27.2	39.0	0.3	1.5	0
12. Pineapple(L	0.43	2.4	106	0.1	57.3	20.0	0.8	0.4	0

Note* Mango (1) = medium (7-8 cm.long). Mango (ii) = Large (10-12cm long).

S = Small m = medium L = Large

3.3.2 MINERAL CONSTITUENTS OF VEGETABLES(Mg/100g) EDIBLE PORTION

TABLE 3

VEGETABLE	ASH %	Na	K	Fe	Ca	Mg	Zn	Mn	Ni
1. Baobab leaves (d)	10.54	88.4	773	0	562	425	5.8	0.6	0
2. Beans (Sokoto Type (d)	3.10	22.7	1240	2.0	11.0	204	3. 5	1.6	1.0
3. Beans (Zaria Type) (d)	3.07	22.5	1350	0	9.2	200	3.5	0.7	0
3. Beniseed (d)	4.4	29.3	1760	2.9	73.6	325	4.8	1.1	0
5. Brown beans (d)	3.2	20.2	1280	1.5	166	192	4.8	2.4	0.6
6. Cabbage	WET DIGEST	14.6	347	0.4	85.0	24.5	0.3	1.6	0
7. Cucumber	0.33	4.0	123	0.1	1.6	13.5	0.2	0.4	0
8. Garden egg (white)	0.35	3.0	99.2	0.1	12.6	15.2	0.4	0.2	0
9. Garden egg (violet)	0.53	4.8	219	0	1.4	22.0	0.2	0.4	0
10.Green beans	0.80	2.7	331	0	5.3	26.7	0.4	0.3	0
11.Groundnuts(d)	2.22	67.6	873	0.8	5.6	178	2.3	0.7	0
12.Lettuce	0.90	25.5	294	1.0	11.5	24.4	0.4	0.3	0
13.Locust bean(d)	6.46	99.0	775	8.2	65.5	410	8.3	3.3	0
14. Melon (d)	3.65	3.4	612	[illegible]	[illegible]	[illegible]	6.8	2.9	

NOTE: (d) = dry
Sokoto type beans are greyish and tiny.
Zaria type beans are white and big.

TABLE 3 CONTD. (mg/100g EDIBLE PORTION)

	VEGETABLE	ASH %	Na	K	Fe	Ca	Mg	Zn	Mn	Ni
15.	Okra	0.94	3.0	144	0.3	86.5	106.3	1.2	0.8	0
16.	Onion	WET DIGEST	5.3	158	0.2	0.2	11.5	0.3	0.2	0
17.	Sorrel (Green)	0.96	11.5	128	1.1	57.6	69.0	0.5	2.5	0
18.	Spinach	2.50	2.8	285	2.3	400	166	2.4	4.6	0
19.	Tomato (oblong)	0.30	5.1	77.6	0.9	8.0	15.0	0.4	0.2	0
20.	Tomato (round)	0.40	5.7	130	0.9	3.5	17.3	0.3	0.2	0
21.	Yam (ph)	0.84	3.6	358	0.1	15.7	32.2	0.7	0	0
22.	Yam (rh)	0.77	3.5	347	0.1	18.5	25.7	0.5	0	0
23.	Water leaf	1.47	21.6	490	0.5	21.0	103	0.4	2.0	0
24.	'Agbono'(d)	1.81	12.1	543	0.7	11.6	158	2.0	2.2	0
25.	'Ayoyo'	1.17	9.7	328	1.0	15.8	70.0	0.7	0.6	0

NOTE* ph = pointed head
rh = round head

3.4 DISCUSSION

In Tables 2 and 3 the results obtained for the constituents of various fruits and vegetables respectively are shown. Variations (minor or significant) in content would obtain in nature for any one sample type depending on the numerous factors elucidated under introduction. To be able to compare these samples fairly, the nature of the sample (wet or dry) used for analysis is taken cognisance of in the discussions below. The dry samples (nutrients concentrated) have higher percentage values, as would be expected. The elements are discussed under macro - and micro constituents depending on the relative abundance in the tissues analysed. The result shows all of the food substances to have some measure of mineral salts which varies over wide ranges in different plant types.

3.4.1 ASH CONTENT

The fruits are fairly rich in mineral content. Their mineral concentration is generally lower than that in vegetables because of the relatively higher water content in most fruits. Grape is praticularly rich in minerals. Its ash content (1.15%) is the highest among the fruits studied. Its contents of the individual minerals (Table 2) rank highest in most cases.

Among the dry vegetables, baobab leaves (Adansonia digitata) have the highest total mineral content. Ash value is 10.54% compared to the least 1.81% in 'Agbono' (Table 3). The others which rank high in mineral content include locust bean (Parkia filcoidea), beniseed (Sesamum radiatum) and melon (Cucumis melo). As for the fresh vegetables, spinach (Spinacea oleracea) has the highest total mineral content (2.5%) followed by water leaf (Talinum triangulare) and Cochorus olitorius.

The three samples - cabbage, onion and plantain whose ash contents are not shown were wet digested by perchloric nitric-sulphuric acid mixture (after Allen, 1974).

3.4.2 MACRO MINERAL CONSTITUENTS

Potassium, Magnesium, Sodium and Calcium are the more abundant minerals in the plant tissues (Table 2 & 3). The first two are the most abundant however, this being linked to the important roles the two play in plant life. While magnesium is constituent of chlorophyll, potassium is involved in catalysing reactions that lead to chlorophyll synthesis. The abundance of potassium in plants thus rules out the possibility of deficiency in normal mixed diets.

Sodium, though fairly abundant in plant materials, its concentration is much lower than that of potassium. Since animals require sodium in higher amounts than can be supplied by plants, this has necessitated addition of sodium salts to animal foods (man and domesticated animals).

Herbivores may travel long distances to salt licks. This is all the more necessary because large potassium intake through plant tissues result in increased excretion of sodium (Oser, 1965). Banana and pawpaw (pink type) are noted for their high sodium content (Table 2). This element is again very abundant in baobab leaves, groundnuts and locust bean (Table 3).

Calcium in plants occur in such tissues as cell walls deposited as calcium pectate , as well as crystals of calcium oxalate. Baobab leaves (dry) and spinach have quite high concentrations of calcium (562 and 400 mg/100g respectively) Table 3. Okoh (1973) reported much higher value for calcium in baobab leaves (2,700mg/100g). The difference may be due to tree location, time of harvest of leaves (young or older leaves and season), as well as possible mixing of other leaves by sellers to increase bulk of their materials. Although the amounts of calcium found in baobab and spinach leaves are quite high, the availability of the mineral to animals from these plant sources is negligible because of their high oxalate contents (Chakraborty, 1977; Okoh, 1973 and Oke, 1965). More valuable(utilisable) sources of calcium would be from juices of fruits especially grape, orange, pawpaw and pineapples (see Table 2 & Appendix 7).

3.4.3 MICRO MINERAL CONSTITUENTS

Iron, zinc and manganese are present in minute quantities in plant tissues (Tables 2 & 3). Nickel is almost absent in all the plant samples studied and infact when present, may be a pollutant and toxic to plants in moderate concentrations.

Iron which is so important to animals is unfortunately deficient in plants occuring in traces or completely absent (as the level of detection in this analysis permits). However, discrete amounts of iron is inevitable even in plant lives since it is linked to porphyrin enzymes involved in respiration. In Table 3 it is shown that one of the varieties of beans (Sokoto type) is quite rich in iron. The highest concentration is in the locust bean (as much as 8.2 mg/100g of dry matter). Such vegetable is invaluable to pregnant women or those in menopause. The other vegetables, fairly rich in iron are beniseed, brown beans and melon. The fruits are generally poor as sources of iron. It is in response to this natural deficiency that animals have evolved efficient system of conserving iron once ingested from foods.

Zinc is fairly distributed amongst plant tissues. It is an extremely important micro-nutrient (see page 17) The essence of it is in continuity of the species (ribonucleic and synthesis). Except in refined flour and cereals

(see Appendix 2) defeciency is not common in normal mixed diets. Kapu (1979) established that zinc defeciency is not a common feature in Nigeria (from studies on various Nigerian subjects).

Manganese is distributed among plant materials in a similar fashion to iron. Its non detectability in yam may be due to its low concentration relative to the mass of the yam tissue cells, since it can not be completely absent in a living cell in view of its role in life processes (see page 18).

Nickel is virtually absent in all plant tissues studied. The only sources of the metal being beans(Sokoto type and brown beans). The metal is of unknown function in both plants and animals, and may have been present in these samples as a result of pollution from soil.

CHAPTER FOUR

ANALYSIS OF MILK AND MILK PRODUCTS

4.1.1 DESCRIPTION AND SOURCES OF THE VARIOUS SAMPLES OF MILK ANALYSED;

POWDERED MILK SAMPLES

	NAME OF MILK	DESCRIPTION AND SOURCE
1.	COAST	INSTANT Full cream milk powder 28% butterfat. Made in Holland.
2.	DANO	Whole milk powder. Made in Denmark.
3.	FRISIANA	INSTANT Full cream powder. Made in Holland.
4.	LACTOGEN	Full-protein with iron &vitamins 19% butter fat, 6.0mg iron and 5.0mg calcium pantothenate per 100g. Made in Denmark for NESTLÉ.
5.	Mix Me	INSTANT milk powder, vitamin enriched. Made in Republic of Ireland by LIPTON.
6.	NAN	Infant food formula in powder form. Made in France by NESTLÉ.
7.	NIDO	INSTANT Full cream milk, Enriched with vitamins A and D. Contain 28% butterfat and 0.2% lecithin Made in France by Sopad.

DISCRIPTION AND SOURCES OF THE VARIOUS SAMPLES OF MILK ANALYSED (CONTD.) - LIQUIDS.

NAME OF MILK	DESCRIPTION AND SOURCE
1. BLACK & WHITE	Full cream evaporated milk. 9% butter fat, Made in Nigeria.
2. DIAMOND BRAND	Full cream evaporated milk 9% butter fat, Made in Canada.
3. FARM CHOCOLATE MILK	Sterilised and homogenised. 3.25% butter fat. Made in Rotterdam, Holland by the United Dairymen.
4. FARM	Sterilised homogenised full cream milk. 3.25% butter fat. Made in Rotterdam, Holland by the United Dairymen.
5. FRISIAN FLAG CHOCOLATE MILK	Made of partly skimmed sterilised milk. Made in Holland by Condensed Milk Factory 'Friesland'-Leeuwarden,
6. GREENLAND	Full cream unsweetened evaporated milk, 9% butter fat. Contains no preservative or adulterant and is guaranteed sterile. Produced in Canada for the Canadian Dairy Commission.

NAME OF MILK	DESCRIPTION AND SOURCE
7. LIBBY'S	Evaporated full-cream milk 9% butter fat. Made in (W) Germany.
8. Little Mary	Pure natural milk. 3.1% butter fat. Sterilised and homogenised. Made in Holland by Holland Canned Milk Veghel.
9. N.N.S.C.	(Nigeria National Supply Co. Milk). Whole, Unsweetened, condensed milk 9% butter fat. Made in Canada by the Canadian Dairy Commission.
10. PEAK BRAND	Unsweetened full-cream condensed milk. Enriched with vitamin D_3. 9% butter fat. Made in Nigeria by The West Africa Milk Co., Ikeja.
11 UNIFOOD	Pure cow's milk. Natural, homogenised, sterilized. Made in Holland by Van-heel's Condensed Milk Coy. B.V. Leeuwarden.

DESCRIPTION AND SOURCES OF THE FRESH MILK ANALYSED - (FACTORY PRODUCTS):

NAME OF MILK	DESCRIPTION AND SOURCE
1. BELLMA	Fresh, sterilized milk, German Export Quality. Contains no chemical ingredients for preservation. Butter fat 3.1%. Especially good for children. (Produced, July '80., expirty date July '81), by Rudolf G.W. Kaiser GMBH, Hamburg, W.Germany. Contents: 350ml in tin.
2. LONG LIFE	Processed and packed in Nigeria by MADARA Ltd. P.O.Box 3, Vom. Expiry date: 30th April 1981. Contents: 500ml in packet.

YOGHURTS

NAME	DESCRIPTION AND SOURCE
1. SWEET YOGHURT	Processed and packed in Nigeria by National Livestock Production Company P.O. Box 479, KADUNA. Contents:250ml (packet), Expires 3/3/81.
2. YOGHURT SWEETENED AND SKIMMED	Processed and packed in Nigeria by MADARA Ltd. P.O. Box 3, Vom. Contents : 250ml (packet).
3. YOGHURT DRINK YOGO	Produced by FAN MILK LTD., Nigeria. Contents: 200ml (packet).

4.1.2 HISTORY OF THE FOUR COWS FROM WHICH MILK WAS OBTAINED AT N.A.P.R.I., SHIKA.

All the four cows are crossbreeds Friesian/Friesian crossbreed or White Fulani/Friesian Crossbreed. Their parents were obtained from either Vom, Nigeria or from abroad. They were all T.B. tested which showed negative, and had received following vaccines: TCV, CBPP, BQ and Anthrax. They are all bucket fed on compounded rations of ground nut cakes, maize or guinea corn (18% crude protein) as well as free grazing or grass hay supplied.

Each cow has a tag number (ear no) normally attached to its ear unless it has fallen off. This number is written against each cow as a means of identifying it (in its records).

None of them has had any serious sickness or injury since birth except cow no 61 which had inflamed udder (mastitis) in October 1969 and was treated.

The weight of each cow given under the information about each of them was as at March ending 1981. All the four were still lactating at the time.

NOTE: N.A.P.R.I. = National Production Research Institute

TCV = Tissue Culture Vaccine (for Rinderpest disease)

C.B.P.P. = Contagious Bovine Pleuro pneumonia.

Q.B. = Black Quarter

SHIKA = Village about 12km from ABU main campus where NAPRI is situated.

SOME DATA ABOUT THE FOUR LACTATING COWS AT N.A.P.R.I (A.B.J., ZARIA)

COW NUMBER	61	201	772	813
Date of birth	2 : 1 : 65	17 : 6 : 68	17 : 10 : 72	3 : 2 : 73
Weight at birth	54lb. (49.1kg.)	60lb.(54.5kg.)	70lb.(63.6kg)	65lb.(59.1kg)
Present weight	444 kg.	456 kg.	532 kg.	556 kg.
Date of first calving	11 : 4 : 67	13 : 9 : 71	6 : 4 : 76	27 : 10 : 75
No of lactation periods till date (March,1981)	13	9	5	5
Date of commencement of present lactation period	8 : 11 : 80	29 : 12 : 80	6 : 12 : 80	4 : 2 : 81
Notable lengths of lactation periods	396, 369, 367, 233 days.	187 - 301 days	242, 297, 365 days	127 - 353 days.
Notable weights of milk produced at different lactation periods (with date)	3,260.1kg. in 1967/68 & 1,611.0 kg. in 1979.	1,888.6 kg. in 1972 & 1012.3 kg. in 1980	2,110.3 kg. in 1976 & 2,078.8 kg. in 1979/80	2,809.9 kg. in 1975/76 & 1.024.1 kg. in 1979.

4.2 MATERIALS AND METHODS

MATERIALS

1. Powdered milk in tins - contents: 400g & 450g (all imported).
2. Liquid milk in tins - contents: 170g, 360g and 397g (All imported, except 'peak' and 'Black and White').
3. Fresh milk - 'Bellma' in 350ml tin (imported); 'long life' in 500ml packet (from Vom, Nigeria).
4. Fresh milk from N.A.P.R.I., Shika - 1kg wt. (each cow sample).
5. Yoghurts - 200 and 250ml contents in packets.

METHODS

REAGENTS AND SOLUTIONS

As used in fruits and vegetable analysis.

APPARATUS

1. Furnaces: Gallenkamp FR-645 (0-1500^{o}C) and Gallenkamp FR 555 (0-1000^{o}C).
2. Porcelain dishes: 75ml, 150ml, 280ml capacities.
3. Others as in 'fruits and vegetables' analysis.

PROCEDURE

50g powdered milk was weighed in porcelain dishes, wetted and dried overnight at 100^{o}C in ovens. Liquid milk contents in tin or packet was transferred whole into

crucible and 250ml of fresh milk from Shika was also measured into crucible and evaporated to dryness (more or less) and then transferred to oven for further drying at 100°C overnight. Few drops olive oil were then added to each sample and burnt over bunsen flame until slight ash began to appear. They were removed into furnaces and ashed for 24hr. at 525°C. Ash solutions were prepared as before from 0.15g ash and readings taken of standard solutions of metal ions and the sample solutions on Atomic Absorption Spectrophotometer.

RESULTS

4.3: MINERAL CONSTITUENTS OF MILK - POWDERED, LIQUID (NON-FRESH AND FRESH) AND YOGHURTS

TABLE 4.1: POWDERED MILK minerals (mg/100g)

NAME OF MILK	ASH%	Na	K	Fe	Ca	Mg	Zn	Mn	Ni
1. COAST	5.68	356	1,250	0	1,440	100	5.4	0	0
2. DANO	5.85	410	1,424	0	1,385	95.5	5.7	0	0
3. FRISIANA	5.88	400	1,431	0	1,380	96.0	6.3	0	0
4. LACTOGEN	4.50	305	1,018	2.0	1,440	73.5	4.3	0	0
5. Mix Me	5.96	517	1,232	0	1,300	81.5	5.4	0	0
6. Nan	2.50	188	531	2.0	590	45.0	1.6	0	0
7. NIDO	5.67	363	1,172	0	1,305	92.6	4.9	0	0

TABLE 4.2 LIQUID MILK (NON-FRESH) Minerals (mg/100g)

NAME OF MILK	ASH%	Na	K	Fe	Ca	Mg	Zn	Mn	Ni
1. BLACK AND WHITE	2.04	186	394	0	460.0	28.0	2.0	0	0
2. DIAMOND BRAND	2.02	166	418	0	405.0	27.6	1.6	0	0
3. FARM CHOCOLATE	0.81	51.0	233	0.8	177.5	21.2	1.0	*	0
4. FARM	0.73	46.4	138	0	170.0	11.4	1.0	0	0
5. FRISIAN FLAG	0.75	42.5	193	0.3	145.0	16.3	0.9	0	0
6. GREENLAND	0.93	184	425	0	421.5	26.0	1.7	0	0
7. LIBBY's	3.50	321	678	0	825.0	53.7	4.4	0	0
8. Little Mary	0.73	51.0	171	0.9	159.0	12.2	0.8	0	0
9. N. N. S. C.	2.01	194	429	0	422.0	27.5	1.5	0	0
10. PEAK BRAND	2.01	165	389	0	420.0	31.6	2.3	0	0
11. UNIFOOD	0.45	31.6	108	0	609.0	7.4	0.6	0	0

NOTE: * = Trace.

MINERAL CONSTITUENTS OF MILK (CONTD.)

TABLE 4.3 minerals
FRESH MILK (mg/100ml)

NAME OF MILK	ASH%	Na	K	Fe	Ca	Mg	Zn	Mn	Ni	Wt/100ml (g)
1. Bellma	0.71	49.5	156	0	159.0	12.3	0.8	0	0	102.9
2. Long Life	0.66	46.0	141	0	165.0	11.4	0.7	0	0	101.6
FRESH MILK FROM N.A.F.R.I., ABU ZARIA										
COW NUMBER	ASH%	Na	K	Fe	Ca	Mg	Zn	Mn	Ni	Wt/100ml (g)
3. 201	0.68	46.0	104	0	95.0	9.3	0.6	0	0	101.9
4. 61	0.76	31.0	132	0	148.0	10.4	0.6	*	0	102.0
5. 813	0.74	55.0	163	0	197.5	11.0	0.7	0	0	101.9
6. 772	0.75	55.5	155	0	195.0	13.0	0.8	0	0	102.1

NOTE: * Trace

TABLE 4.4: YOGHURTS minerals (mg/100ml)

N A M E	ASH%	Na	K	Fe	Ca	Mg	Zn	Mn	Ni	Wt/100ml (g)
1. Sweet Yoghurt	0.79	51.0	153	0	180.0	11.6	0.8	0	0	104,3
2. Yoghurt⊛	0.87	56.0	191	0	200.0	13.9	0.7	0	0	103.6
3. YOGHURT YOGO	0.71	47.0	147	0	152.5	9.0	0.5	*	0	104.4

NOTE: Yoghurt⊛ = Yoghurt Sweetened and Skimmed.

YOGHURT YOGO: Full name is YOGHURT DRINK YOGO.

* = Trace

4.4: DISCUSSION

Table 3 shows the mineral content of milk (cow milk) of different brands. There is abundance of certain mineral elements, while others are present in trace amounts or absent.

4.4.1 ASH CONTENT

The relatively high ash content in milk (Table 4.1 to 4.4) indicates abundance of mineral salts in milk (Appendices 8 and 9 show major and trace element constituents of milk - fresh samples).

Among the powdered milk samples 'Mix Me' has the highest total mineral (ash) content. It also has very high percentage values for many of the individual minerals, notably sodium, potassium and calcium. This milk may be relatively drier than the other samples. One of the milk samples - Nan, a popular milk food rich in nutrients specially formulated for infants is conspicuously low in mineral (ash) content. A value of 2.5% was obtained for this milk compared to 5.67% in Nido (another good milk for growing children). The merit of Nan may lie in its enrichment with iron and other additions such as vitamins. Some of the inclusions in the nutrient content of the milk (Nan) comprise demineralised whey and corn oil.

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Of the liquid milk samples, Libby's milk has the highest ash content (Table 4.2). Infact it has the highest percentage value for each of the individual mineral studied. Manganese and nickel are also absent in this milk. It is observed that the liquid milk samples with the highest mineral content (Libby's inclusive) are all condensed or evaporated milk. The other milk samples (non-evaporated) have similar mineral contents to the fresh milk samples.

The milk products - Yoghurts do not vary much in ash content as well as in individual mineral contents from the fresh milk samples.

4.4.2: MACRO MINERAL CONSTITUENTS

Calcium and potassium are the most abundant minerals in milk. Milk calcium is in a form readily available to animals for metabolism and hence is the principal source of this element to animals.

Sodium and magnesium which have several important roles in man and other animals and are also found stored in moderate amounts in bones, are also abundant in milk (Tables 4.1 to 4.4). Milk is thus superior to plant sources in supply of calcium and sodium.

One of the fresh milk samples (from cow No. 201) is notably low in calcium and potassium contents. It is observed that cow no. 201 is quite aged (born 1968)

although not as old as cow no. 61 (born 1965). The latter has had much longer lactation periods and far greater quantity of milk produced. The other fresh milk samples from cow No. 813 and 772 (which are younger), have higher calcium and potassium contents than those from the older cows.

4.4.3 MICRO MINERAL CONSTITUENTS

Milk is conspicuously most defecient in IRON compared to many food substances (see also Tables 1, 2, and 3), except in manufactured brands enriched with iron. This deserves special attention since milk is the sole food of infacts in their early (first few months of) life, and the metal is required for the making of red blood cells. The need for iron supplement in infant food arose from this deficiency in milk. Two of the milk samples analysed contain iron supplements. The value obtained in this analysis (2.0mg/100g sample) is much lower than 6.0mg/100g sample as indicated on the containers of these milk samples (Lactogen & Nan). The other values agree with those found by other workers (Appendix 8). The difference mentioned above may be due to different laboratory methods and effectiveness of measures to completely eliminate interference from sulphate ions when the iron content is high.

From the preceding tables it is evident that certain plant materials such as benisseed (Sesamum radiatum), locust bean (Parkia filicoidea), 'Ayoyo' (Cochorus olitorius) and spinach (Spinacea oleracea) are more important than milk as sources of iron. Zinc is fairly abundant in milk. Shroeder et al., (1967) reported 4.18 mg/Kg in whole raw milk.

Manganese and Nickel are shown to be absent more or less in milk (Tables 4.1 to 4.4) These may not necessarily be completely absent. They may be present in limits (few micrograms per kilogram) not detectable by methods employed in this analysis. This applies to all cases where the concentration of a given element is given as zero (0) in Tables in these analyses. Archibald (1951) also reported absence of nickel in milk but found manganese at 0.002 Mg/Kg milk.

In Appendix , it is shown that even when nickel is added to cow feed, it still doesn't show in milk. It is probable hence that nickel is never found in milk.

CONCLUSION:

The proximate analysis as well as the determination of some metals and non-metals in the various samples of bread show that bread is quite a nutritious food in these respects. The carbohydrate, lipid and protein contents are reasonably adequate for energy and body building. Since it is a commodity which is relatively cheap and a ready-to-eat one, it could serve well as an avenue for introducing certain essential nutrients lacking in a population by incorporating these into bread during or after baking (for substances susceptible to destruction by heat), by putting in such medium as butter and applying as a polish on the baked loaves.

The fruits and vegetables provide a number of essential mineral elements (the vegetables being the better sources of these minerals). The essence of mixed diet need be re-emphasised since some plants used as food and which are deficient in some metals are compensated for by those present in others.

SUGGESTIONS FOR FURTHER WORK

1. Study of variations (day to day) in the bread constituents in each of some chosen bakeries for a period of upto 30 - 50 days - (proximate analysis only).

2. Determination of some metals and non-metals in a similar manner to that suggested for proximate analysis in (1) above.

3. Study of factors that enhance keeping quality of bread.

4. Factors affecting availability of metal ions to humans from bread, fruits and vegetables.

5. Analysis for nutrients in non-edible portions of fruits and vegetables.

APPENDIX 1

COMPOSITION OF WHEAT AND PRODUCTS OF MILLING

A. APPROXIMATE COMPOSITION OF WHEAT(%)

MOISTURE :	9 - 18
STARCH :	60 - 68
PROTEIN :	8 - 15
CELLULOSE (FIBRE) :	2 - 2.5
FAT :	1.5 - 2
SUGAR :	2 - 3
MINERAL MATTER (ASH):	1.5 - 2

B. APPROXIMATE COMPOSITION OF FLOUR(%)

	WHITE FLOUR- 72% EXTRACTION	'NATIONAL' FLOUR- 80% EXTRACTION	WHOLE MEAL FLOUR 95% EXTRACTION
MOISTURE:	13 - 15	13 - 15	13 - 13.5
STARCH :	65 - 70	64 - 69	63 - 67
PROTEIN (Nx5.7):	8 - 13	9 - 14	10 - 14
CELLULOSE:	TRACE - 0.2	0.2 - 0.35	1.6 - 2.1
FAT:	0.8 - 1.5	1 - 1.6	1.6 - 2.2
SUGARS	1.5 - 2.0	1.5 - 2.0	2.0 - 3.0
ASH:	0.3 - 0.6	0.6 - 0.8	1.4 - 1.6

From: Modern Cereal Chemistry by Kent-Jones, D.W. and Amos, A.J. 6th ed, p. 9 (1967).

APPENDIX 2

PERCENTAGE NUTRIENTS RETAINED DURING MILLING FOR PRODUCTION OF AMERICAN 70% EXTRACTION FLOUR USED FOR MAKING WHITE BREAD

VITAMINS		MINERALS	
Pantothenic acid	50	Selenium	84
Folic acid	33	Chromium	60
Vitamin B_6	28	Molybdenum	52
Thiamin	23	Calcium	40
Riboflavin	20	Copper	32
Nicotinic acid	19	Phosphorus	29
Vitamin E	14	Iron	24
		Potassium	23
		Zinc	22
		Sodium	22
		Magnesium	15
		Manganese	14
		Cobalt	12

Adapted from Millfeed Manual, Millers National Federation, Chicago, 1967.

APPENDIX 3

COMPOSITION OF FOODS, 100 GRAM EDIBLE PORTION - BREAD

	BREAD TYPE	WATER %	PRO-TEIN%	FAT %	ASH %	Ca mg	P mg	Fe mg	Na mg	K mg
1.	Cracked Wheat	34.9	8.7	2.2	2.1	88	128	1.1	529	134
2.	French or Vienna (enriched)	30.6	9.1	3.0	1.9	43	85	2.2	580	90
3.	French or Vienna(unenriched)	30.6	9.1	3.0	1.9	43	85	0.7	580	90
4.	Italian (enriched)	31.8	9.1	0.8	1.9	17	77	2.2	580	74
5.	Italian (unenriched)	31.8	9.1	0.8	1.9	17	77	0.7	585	74
6.	Raisin	35.3	6.6	2.8	1.7	71	87	1.3	365	233
7.	Rye, American	35.5	9.1	1.1	2.2	75	147	1.6	557	145
8.	White, unenriched made with 1-2% NFDM.	35.8	8.7	3.2	1.9	70	87	0.7	507	85
9.	White, unenriched made with 5-6% NFDM	35.0	9.0	3.8	2.0	96	102	0.7	495	121
10.	Whole wheat + 2% NFDM	36.4	10.5	3.0	2.4	99	228	2.3	527	273

Note: NFDM = non-fat dry milk.

From: COMPOSITION OF FOODS - RAW, PROCESSED, PREPARED by Bernice K. Watt and Annabel L. Merrill, in Agriculture Handbook No. 8, United States Department of Agriculture, 1975.

APPENDIX 4

TABLE SHOWING COMPARATIVE VALUE OF BREAD(percent) IN PRESENT DAY DIETS

	ENERGY VALUE	PROTEIN	CAL-CIUM	IRON	VITA-MIN B_1	NICOTI-NIC ACID
Milk, Cream & Cheese	12.7	23.1	60.8	3.8	13.1	3.7
Vegetables	7.2	8.8	5.9	17.0	21.3	17.9
Bread	+++ 16.6	+ 19.4	++ 15.7	++ 20.4	++ 22.1	++ 18.7
Meat	15.3	26.1	2.1	28.1	24.5	39.3
Fruit	2.0	1.0	1.6	4.0	3.3	2.8
Cereals other than Bread	15.0	10.1	8.1	13.4	10.6	9.8
Sugar and preserves	12.4	-	0.3	0.9	-	0.1
Fats	14.4	0.2	0.3	0.6	-	0.4
Eggs	1.9	5.1	2.0	6.7	3.3	0.2
Fish	0.9	4.7	1.7	2.0	0.9	3.7

+++ Best source ++ Second best source + Third best source. From a cost point of view present day enriched bread in U.K. is the cheapest source of calories, protein, iron, vitamin B_1 and nicotinic acid in the diet and as cheap a source of calcium as dairy products.

Source: - Modern Cereal Chemistry by Kent-Jones,D.W. and Amos, A.J. 6th ed. p. 440 (1967).

APPENDIX 5

TABLE OF CONVERSION OF MILLILITRE SODIUM THIOSULPHATE (TITRE) IN THE DETERMINATION OF SUGAR FOR 25ml LUFF-SCHOORL REAGENT

SODIUM THIOSULPHATE ($Na_2S_2O_3$)0.IM	GLUCOSE, FRUCTOSE, INVERT SUGARS ($C_6H_{12}O_6$)	SODIUM THIOSULPHATE ($Na_2S_2O_3$)).IM	GLUCOSE, FRUCTOSE INVERT SUGARS ($C_6H_{12}O_6$)
ml	mg	ml	mg
1.0	2.4	13.0	33.0
2.0	4.8	14.0	35.7
3.0	7.2	15.0	38.5
4.0	9.7	16.0	41.3
5.0	12.2	17.0	44.2
6.0	14.7	18.0	47.1
7.0	17.2	19.0	50.0
8.0	19.8	20.0	53.0
9.0	22.4	21.0	56.0
10.0	25.0	22.0	59.1
11.0	27.6	23.0	62.2
12.0	30.3		

From: Official Journal of European Communities, p.41, 1974.

APPENDIX 6

A. THE MINERAL COMPOSITION OF SOME COMMON LEAFY VEGETABLES (mg/100g) DRY SAMPLE

NAME	Ca	P	Mg	Mn	K	Na	Fe	Zn	Cu	Cl
Celosia argentea	5,100	525	7,250	7.50	7,500	30.00	80	12.75	1.50	140
Corchorus olitorius	3,300	420	8,150	6.75	3,150	25.00	74	7.88	1.00	515
Vernonia amygdalina	2,950	332	405	14.72	4,000	86.25	48	9.50	2.75	910
Amaranthus spp.	5,750	465	1,550	5.25	4,100	58.75	48	7.50	1.00	500
Talinum triangulare	3,000	320	1,400	13.0	8,850	100.00	102	11.63	1.25	890
Solanum nigrum	3,200	367.5	400	4.75	4,325	53.75	36	8.88	2.50	940

B. THE MINERAL COMPOSITION OF THE MORE COMMON FRUITS (mg/100g) EDIBLE PORTION

	Ca	P	Mg	Fe	Cu	Na	K	Cl	S
Banana	7.9	29.4	41.9	0.52	0.24	1.5	348	79.8	13.5
Grape	4.2	16.1	5.2	0.43	0.09	2.1	344	1.8	7.6
Guava	230.0	46.2	13.8	0.94	0.32	4.2	295	52.4	8.7
Lime	122.0	21.4	12.3	0.37	0.22	7.2	184	4.9	13.2
Mango	11.4	13.8	11.3	0.43	0.31	6.4	192	24.2	-
Orange	42.6	24.8	11.8	0.42	0.08	3.2	201	3.1	9.3
Pine Apple	12.2	10.2	17.3	0.54	0.09	2.1	263	32.4	3.2
Pawpaw	178.4	11.5	-	2.24	0.22	-	-	-	-

By Oyenuga, V.A. and Fetuga, B.L.
Extracted from: Topic 9: National Seminar on Fruits and Vegetables, Ibadan, October, 1975.

APPENDIX 7

COMPOSITION OF FOODS 100g EDIBLE PORTION

	% WATER	% ASH	Ca(mg)	P(mg)	Fe(mg)	Na(mg)	K(mg)
Banana: Raw, common	75.7	0.8	8	26	0,7	1	370
Eggplant: Raw	92.4	0.6	12	26	0.7	2	214
Grape fruit: Raw, all varieties	88.4	0.4	16	16	0.4	1	135
Plantain (baking banana)	66.4	0.9	7	30	0.7	5	385
Grapefruit juice: all varieties	90.0	0.2	9	15	0.2	1	162
Guavas: Raw, common	83.0	0.6	23	42	0.9	4	289
Orange juice:Raw varieties(all)	88.3	0.4	11	17	0.2	1	200
Orange: Raw, all varieties	86.0	0.6	41	20	0.4	1	200
Pawpaw: Raw, common, North American type	76.6	0.5	-	-	-	-	-
Papayas: Raw	88.7	0.6	20	16	0.3	3	234
Pineapple: Raw	85.3	0.4	17	8	0.5	1	146
Pine apple juice, unsweetened	85.6	0.4	15	9	0.3	1	149
Tomatoes, green, raw	93.0	0.5	13	27	0.5	3	244
Tomatoes, ripe, raw	93.5	0.5	13	27	0.5	3	244
Tomato juice, canned	93.6	1.1	7	18	0.9	200	227
Spinach: raw	90.7	1.5	93	51	3.1	71	470
Newzealand Spinach: raw	92.6	1.8	58	46	2.6	159	795

Source: Composition of foods - raw, processed, prepared by Bernice K.Watt and Annabel L. Merril. Agriculture Handbook No 8, United States Dept. of Agric., Washington, D.C., (1963 and 1975).

APPENDIX 8

AVERAGE VALUES FOR MILK SALT CONSTITUENTS

CONSTITUENT	CONTENT IN WHOLE MILK(mg/100ml)	NUMBER OF SAMPLES
Calcium	123	824
Magnesium	12	759
Phosphorus	95	829
Sodium	58	491
Potassium	141	472
Chlorine	119	1,579
Sulphur	30	80
Citric acid	160	307

FROM: FUNDAMENTALS OF DAIRY CHEMISTRY By
B. H. Webb and Johnson, A. H. p.17 (1965).

APPENDIX 9

SOME TRACE ELEMENTS IN COW's MILK ($\mu g l^{-1}$)

ELEMENT	COW RECEIVING NORMAL RATION	COW WITH SUPPLEMENT OF ELEMENT IN RATION
Aluminium	460	810
Arsenic	50	450
Barium	QUALITATIVE	---
Boron	270	660
Chromium	15	---
Cobalt	0.6	2.4
Copper	130	NO INCREASE
Fluorine	150	---
Iodine	43	UP TO 2700
Iron	450	NO INCREASE
Lead	40	---
Lithium	QUALITATIVE	---
Manganese	22	64
Molybdenum	73	371
Nickel	0	0
Rubidium	QUALITATIVE	---
Silicon	"	---
Silver	"	---
Strontium	"	---
Tin	"	---
Vanadium	"	---
Zinc	3,900	5,100

Source: FUNDAMENTALS OF DAIRY CHEMISTRY By B.H. Webb and Johnson, A. H. p.19 (1965).

APPENDIX 10

SOME TRACE ELEMENTS IN THE HUMAN BODY

ELEMENT	ppm	APPROXIMATE AMOUNT IN A 70-Kg MAN(mg)	FUNCTION
Strontium	2.0	140	UNKNOWN
Copper	1.4	100	ESSENTIAL
Aluminium	1.4	100	CONTAMINANT
Lead	1.1	80	"
Tin	0.43	30	"
Iodine	0.43	30	ESSENTIAL
Cadmium	0.43	30	CONTAMINANT
Manganese	0.3	20	ESSENTIAL
Vanadium	$\pm$0.3	$\pm$20	ESSENTIAL?
Barium	0.23	16	UNKNOWN
Arsenic	$<$1.4	100	UNKNOWN
Antimony	$<$1.3	90	CONTAMINANT
Lanthanum	$<$0.7	50	"
Niobium	$<$0.7	50	"
Nickel	$<$0.14	10	UNKNOWN
Boron	$<$0.14	10	ESSENTIAL?
Chromium	$<$0.09	6	ESSENTIAL
Ruthenium	$<$0.09	6	CONTAMINANT
Molybdenum	$\pm$0.07	$\pm$5	ESSENTIAL
Cobalt	$\pm$0.04	$\pm$3	ESSENTIAL
Gold	$<$0.01	1	CONTAMINANT
Silver	$<$0.01	1	"

Source: HAWK's PHYSIOLOGICAL CHEMISTRY By B. L. Oser, p.561 (1965).

APPENDIX 11(a)

LIMITS OF DETECTABILITY BY ABSORPTION FLAME PHOTOMETRY:

ELEMENT	RESONANCE LINE (nm)	LIMIT OF DETECTABILITY($\mu g\ ml^{-1}$)+
Antimony	231.1	1.5
Barium	553.6	8.0
Bismuth	306.8	2
Cadmium	228.8,326.1	0.03,2
Calcium	422.7	0.08
Cesium	852.1	0.15
Chromium	357.9,425.4	0.15,50
Copper	324.7	0.1
Gold	242.8,267.6	0.3,2
Iron	248.3	0.1
Lead	283.3	0.5
Lithium	670.8	0.03
Magnesium	285.2	0.01
Manganese	279.5,403.4	0.05,1.0
Mercury	253.7	10
Nickel	232.0,341.5	0.2,5
Potassium	766.5	0.03
Rubidium	785.0	0.1
Silver	328.1,338.3	0.05,2
Sodium	589.0	0.03
Tin	286.3	5
Zinc	213.9	0.03

Source: See below + concentration giving 1% absorption.

APPENDIX 11(b)

EXCITATION POTENTIALS FOR SOME ELEMENTS

ELEMENT	EXCITATION POTENTIAL(eV)
Potassium	1.61
Lithium	1.85
Calcium	2.93
Aluminium	3.14
Iron	3.30
Copper	3.81
Magnesium	4.34
Lead	4.37

SOURCE: (a) & (b) From Instrumental Methods of Analysis by H. H. Willard, L. L. Merritt(Jr.) and J. A. Dean, 4th ed. (1965).

REFERENCES

1. Adeneye, J.A., Oyenuga, V.A., and Olaloke, F.A., Preliminary findings on the composition of milk of white fulani cattle. Nig. J. Sc. 4, (2), 181-186 (1970).

2. Akinrele, I.A. and Shambe, T., Bread and confectionaries baking tests with composite flour. Nig J. Sc. Vol. 8, nos 1 & 2 (1974).

3. Alexeyev, V., Quantitative Analysis (Translated from Russian) by E. Uvarov. MIR Publishers, Moscow, 1969.

4. Allen, S.E., Grimshaw, H.M., Parkinson, J.A. and Quarmby, C., Analysis of Ecological Materials, 1st ed. Blackwell Scientific Publications, Oxford, London (1974).

5. Allison, J.B., The Nutritive Value of Dietary Proteins in Mammalian Protein Metabolism. Vol. 2. Academic Press, New York and London (1968).

6. Ankrah, E.K., The Chemical Composition of Fresh and Evaporated milk of cow marketed in Ghana. Ghana J. Agric. Sc., 26 (2), 109-111.

7. Archibald, J.G., J. Dairy Sci., 34, 1026 (1951).

8. Balls, A.K. and Hale, W.S., J. Assoc. Off. Agric. Chem., XVIII, 135 (1935).

9. Balls, A.K. and Hale, W.S., Cereal Chem., XIII, 54 (1936).

10. Basir, O. and Melnoo, A.M., Methods for determination of calcium in biological materials. W. Afr. J. Biol. Appl. Chem., 13(1), 20-28 (1970).

11. Beeson, K.C., The mineral composition of crops with particular reference to the soils in which they were grown. U.S.D.A. Misc. pub. 369, Washington D.C. (1941).

12. Benton, A.G., J. Dairy Sci., 12, 481(1929).

13. Bergman, E.L., Overall aspects of plant analysis in vegetable production. Symposium: Nutrition and fertilization of vegetables. Warsaw, Poland (Sept. 1971).

14. Bressani, R., Wilson, D.L., Behar, M., and Scrimshaw, N.S., Supplementation of cereal proteins for adult human subjects with amino acids. J. Nutr., 70, 176 (1960).

15. Bricker, M. Mitchell, H.H. and Kinsman, G.M. The protein requirements of adult human subjects in terms of proteins contained in individual foods and food combinations. J. Nutr., 30, 269 (1945).

16. Chakraborty, R., Studies on Hydrocyanic, oxalic and phytic acid content of foodstuffs. M.Sc thesis (ABU)., Zaria

17. Chopra, J.G., Enrichment and fortification of foods in Latin America. Amer. J. Public Health, 64, 1944.

18. Cobble, J.W., and Herman, H.A., Mo. Agric. Expt. Sta. Research Bull. 485 (1951).

19. Dean, J.A., Flame Photometry. McGraw-Hill, New York (1960).

20. Dean, J.A. and Carnes, W.W., Analyst, 87, 743 (1962).

21. Devadas, R.P., Devadoss, M.V. and Rowlands, R., The supplementary value of wild greens ....a mixture of leafy vegetables to a poor indian rice diet. J. Nutr. Dietet. 1:34-39 (1964).

22. Dicke, W.K. Weigers, H.A. and Kamer, J.H. Van de., Coeliac disease. The presence in wheat of a factor having deleterious effect in cases of coeliac disease. Acta Paed. Uppsala, 42, 34 (1953).

23. Diggins, R.V., Bundy, C.E. and Christensen V.W., Dairy Production. 4th ed. © 1979.

24. Doby, G., Plant Biochemistry. Revised ed. Inter-science publishers, London, New York and Sydney (1965).

25. Entenan, C.J., American oil chem. soc., 38, 534(1961).

26. Feinstein, L. and Hart, J.R., Cereal chem., 36, 191 (1959).

27. Freilich, J. and Frey, C.N., Cereal chem., XX, 30 (1943).

28. Fosnot, R.H., and Haman, R.W., Cereal chem, XXII, 41 (1945).

29. Goodall, D.W. and Gregory, F.G ., Chemical composition of plants as an index of their nutritional status. Imp. Bureau of Hort. and Plantation crops. Eás Malling, Kent, England. Tech. Comm., 17, 167 (1947)

30. Graham, G.G., Placko, R.P., Acevedo, G., Morales, E and Cordano A., Lysine enrichment of wheat flour: evaluation in infants; Amer. J. Clin. Nutr., 22, 1459 (1969).

31. Harper, A.E., The nutritive value of cereal proteins with special reference to the availability of amino acids, Proc. 5th intern. congr. Biochem., 8, 82 (1965).

32. Hart, F.L. and Fisher, H.J., Modern Food Analysis. Springer-Verlag, New York (C) (1971).

33. Hashmi, M.H., Ali, E. and Umar, M. Anal. Chem., 34, 988.

34. Herd, C.W. and Amos, J., Cereal Chem., VII,251 (1930).

35. Horwitz, W. (Ed.), Official Methods of Analysis of the Association of Official Analytical Chemists. 11th ed., 1970 and 12th ed., 1975. AOAC, Washington D.C.

36. Hurtwitz, C. and Beeson, K.C., Cobalt content of some edible food plants. Fed. Res. 9, 348-359

37. Jacobs, Morris B., Chemical Analysis of Foods and Food Products. D. Van-Nostrand Co., New York (1938).

38. JAOAC 6, 508 (1923).

39. JAOAC 9, 42, (1936).

40. JAOAC 11, 38 (1938).

41. JAOAC 27, 86,395(1944).

42. JAOAC 28, 77 (1945).

43. Kapu, M.M., Zinc and Copper in Sympton-Free Nigerians, Those with Sickle Cell Disease and Pathological Conditions. Ph.D. Thesis, ABU., Zaria (1979).

44. Kefford, J.F., Chemical constituents of citrus fruits. Adv. Food Res. 9 225.

45. Kent, N.L., Technology of Cereals with special Reference to Wheat. 2nd ed. Pergamon Press. Oxford and New York (1975).

46. Kent-Jones, D.W. and Amos, A.J., Modern Cereal Chemistry. 6th ed., 1967. Food Press Ltd., London.

47. Ketiku, A.O., Chemical composition of unripe (green) and ripe plantain (Musa paradisiaca). J. Sc. Food and Agric. 24 (6), 703-707 (1973) OR Nutr. Abstr. Rev. 44 (1974).

48. Kitson, R.E. and Mellon N.G., Ind. Engng. Chem. Analyst. Edu. 16, 379 (1944).

49. Klemm, G.C., General methods for ashing. J.Assoc. Offic. Agr. Chemists, 47, 40 (1964).

50. Krebs, H. and Mellanby, K., Lancet, 319 (1942).

51. Kyaruzi, V.K., Nutrition in East Africa. 1st ed. Longmans, Arusha and Nairobi (1966).

52. Lane, J.H. and Eynon, L., J. Soc. Chem. Ind., 42, 32T (1923).

53. Legates, J.E., J. Dairy Sci., 43, 1527 (1960).

54. Litchfield, C., Analysis of Triglycerides. Academic Press, New York and London (1972).

55. Luff-Schoorl Method: Official J. European Comm; P.38 (1974).

56. Macy, I.G., Kelly, H.J. and Sloan, R.E., National Research Council Publication 254(1953)

57. Mc Cance, R.A., Widdowson, E.M., Moran, T., Pringle, W.J.S. and Macrae, T.F., The Chemical composition of wheat and rye and of flours derived therefrom. Biochem. J., 39, 213 (1945).

58. McCance, R.A. and Widdowson, E.M., Chemical Composition of foods, Med. Res. Council, H.M.S.O., London (1946).

59. Mc Cance, R.A. and Widdowson, E.M., J. Hyg., 59, (1947).

60. Mc Dowell, R.E., State of the dairy industry in warm climates of less developed countries. Cornell international Agric. mimeograph 57 (1977).

61. Moran, T., Nutritional significance of recent work on wheat, flour and bread. Nutr. Abs. Revs., 29, (1959).

62. Moran T. and Pace, J., Nature, CL, 224 (1942).

63. Moore: Iron metabolism and nutrition. Harvey lectures, 55, 67 (1959-1960).

64. Moss, P., J. Sc. Food and Agric., 12, 30.

65. Narayana Rao, M., Satry, L.V.L. and Parpia, H.A.D., What nutrition fruits contain. Indian Horticulture (July-Sept), 87-95 (1967).

66. Nickerson, T.A., J. Dairy Sci., 43, 598 (1960).

67. Nuru, S., Self-sufficiency in livestock production. NAPRI Bul., No.2, April/June, 1980.

68. Oke, O.L., Chemical studies on the more commonly used leaf vegetables in Nigeria. J. West Afr. Sc. Assn. 11, (182), 42-48 (1966)

69. Okoh, P.N., Biochemical Studies on Traditionally prepared Food of Northern Nigeria (furanono). MSc. thesis (ABU)., Zaria (1973).

70. Ologunde, O.O., Influence of Potassium and Magnesium on Growth and Composition of Sorghum. PhD. thesis. University of Nebraska (1980).

71. Oomen, H.A.P.C. and Grubben, G.J.H., Tropical Leaf Vegetables in human nutrition communication 69, Dept. of Agric. Res., Royal Trop. Inst. Amsterdam.

72. Oser, B.L. (Ed.), Hawk's Physiological Chemistry. 14th ed. Mc.Graw-Hill, New York, Toronto, Sydney, London (1965).

73. Oyenuga, V.A., Nigeria's foods and feedstuffs: Their chemistry and nutritive value. Ibadan University Press, Ibadan (1968).

74. Oyenuga, V.A. and Fetuga, B.L., Dietary importance of fruits and vegetables. Topic 9: National seminar on fruits and vegetables, Ibadan. October, 1975.

75. Pearson, D., The Chemical Analysis of Foods, 7th ed. Churchill Livingstone, London and New York (1976).

76. Peterson, W.H. and Elvehjem, C.A., The iron content of plants and animal foods. J. Biol. Chem., 78, 215-223 (1928).

77. Riddet, W. Campbell, I.L., Mc Dowall, F.H. and Cox, G.A., New Zealand J. Sc. Tech., 23A, 80, 99 (1942).

78. Sai, F.A., Nutritional value of West African fruits and vegetables in 'Fruits and Vegetables in West Africa', (H.D. Tindall) F.A.O., Rome (1965).

79. Sair, L. and Fetzer, W.R., Cereal Chem., XIX 633 (1942).

80. Sandstead, H.H., Present Knowledge of the minerals in 'Present Knowledge in Nutrition. 3rd ed. Nutrition Foundation, New York (1967).

81. Schmidt, D.R., Yield and Chemical Components of Tropical Leafy Vegetables Species as influenced by Environmental Variables and Plant Nutrition. PhD. thesis Cornell University (1971).

82. Schmidt, R.R., Comparative yields of eight Tropical leafy vegetables grown at two soil fertility levels. Agron. J., 63, 546,550 (1975).

83. Schmidt, G.H., J. Dairy Sci., 43, 213 (1960).

84. Shen, T. and Greddes, W.F., Cereal Chem., XIX, 609 (1942)

85. Shroeder, H.A., Mason, A.P., Tipton, I.A. and Balsa, J.J. Essential trace metals in man: Zinc, Relation to environmental Cadmium. J. Environ. Bio., 20, 179-210 (1967).

86. Spicer, A. (Ed.), Bread-Social, Nutritional and Agricultural Aspects of Wheaten Bread. Applied Science Publishers Ltd., London (1975).

87. Taylor, W.H., Analyst, 82, 488 (1957).

88. Udy, D.C., Cereal Chem., 31, 389 (1954).

89. Udy, D.C., Cereal Chem., 33, 190 (1956a).

90. Vines, A.E. and Rees, H., PLant and Animal Biology Vol.2. 4th ed. Pitman Publishing Co., London, New York, Toronto, Nairobi, Johannesburg, Victoria 3053-Australia (1972).

91. Vogel, A.I., A Textbook of Qualitative Inorganic Analysis including Elementary Instrumental Analysis. 3rd ed. Longmans, London (1961)

92. Vogel, K.P. Johnson, V.A. and Mattern, P.J., Results of systematic analysis for proteins and lysine composition of common wheat (Triticum aestiuum L.) in the U.S.D.A. World Collection, Nebr. Res. Bul. 258, 20p (1973).

93. Watt, B.K. and Merrill, A.L., Composition of Foods. U.S.D.A. Agric Handbook no.8. U.S. Govt. Print Office, Washington D.C. (1963).

94. Webb, B.H. and Johnson, A.H., Fundamentals of Dairy Chemistry. The Avi Publishing Co., Inc., Westport, Connecticut (C) (1965).

95. White, A., Handler, P. and Smith, E.L., Principles of Biochemistry. 4th ed. (International student edition). McGraw-Hill Book Co., New-York, London (1968).

96. Whittle, E.G., Analyst, 68, 247 (1943).

97. Wichmann, H.J., six papers on factors influencing the ash determination. J. Assoc. Offic. Agr. Chemists, 23, 630 (1940).

98. Widdowson, E.M. and Mc Cance, R.A., Studies on the nutritive value of bread and on the effect of variations in the extraction rate of flour on the growth of undernourished children. Med. Res. Council Spec. Rept. Ser. No 287, H.M.S.O; London (1954).

99. Willard, H.H. and Dean, J.A., Anal. Chem., 22, 1264 (1960).

100. Willard, H.H., Merritt, L.L. (Jr.) and Dean, J.A., Instrumental Methods of Analysis, 4th ed. Affiliated East-West Press Pvt. Ltd. New Delhi (1965).