

FAMILIAL TENDENCY AND DIETARY ASSOCIATION OF GOITRE IN GAMO-GOFA,
ETHIOPIA. 9

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Science Degree in Applied Human Nutrition,
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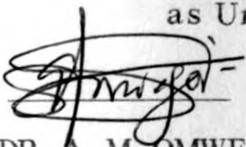
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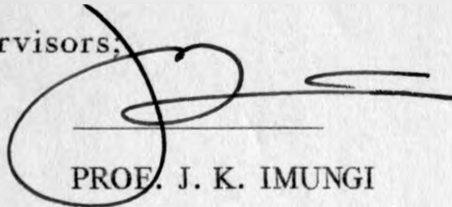


DR. A. M. OMWEGA

Dept. of Food Technology

and Nutrition

Date 12/10/98



PROF. J. K. IMUNGI

Dept. of Food Technology

and Nutrition

Date 12/10/98

Department of Food Technology
and Nutrition, Applied Nutrition
PROGRAMME

DEDICATION: CONTENTS

This work is dedicated to my father Abuye Tirore and my mother Erkale Wanore who always sacrificed so much for my education and 'play' the part of the parents and has seen me grow up and love the Lord.

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DEFINITIONS AND ACRONYMS

Abbreviations:

IDD= Iodine deficiency disorders

TGR= Total goitre rate-is a percentage of all grades of goitre (Grade 1a, 1b, 2 and 3).

UIE= Urinary iodine excretion.

VGR= Visible goitre rates-is a percentage of visible goitre grades (grade 2 and Grade 3).

Definitions:

Woreda= Ethiopian term for a district, sub-division of a province

parents= Both biological parents (mothers and fathers) of the study children included in this study.

Thyroxine (T4)= Thyroid hormone which contains four atoms of iodine; chemically known as tetraiodothyronine.

Triiodothyronine (T3)= Thyroid hormone produced by the thyroid gland and contains three atoms of iodine .

Thyroid Stimulating Hormone (TSH)= secreted from pituitary gland at the base of the brain.

Goitrogens= Chemical substances present in some food stuffs and aggravate iodine deficiency and produce goitre when consumed.

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ABSTRACT

A cross-sectional study which was descriptive and analytical in nature was conducted in 1997 among elementary school children age 6-18 years and both their biological parents in Kucha and Gofa-Zuria woredas, Gamo-Gofa, Ethiopia. The study aimed at assessing the familial tendency and dietary association of goitre. The investigation comprised clinical examination for goitre, collection and laboratory analysis of water and urine samples and interviewing of mothers about foods eaten and about goitre history of grand parents of the children.

A total of 1791 subjects of which 597 were school children and the rest their corresponding biological parents, were selected from seven schools of two woredas by systematic random sampling method.

Prevalence of goitre in the study population was found to be 51.7% of which 21.7% was visible goitre. Goitre ratio between parents (fathers: mothers) was 2:5 for parents age group 20-45 years and 2:9 for those 46 years of age and above.

Familial tendency of goitre between parents and their children was strongly significant. Level of association of goitre was stronger between mothers and their children ($p < 0.0000$ and Odds Ratio = 13.08) than fathers and the children ($p < 0.001$ and Odds Ratio = 4.27).

Halleko (*Moringa stenopetala*), was commonly consumed as leafy vegetable in the study area, and those who consume halleko more than two times

per day were found to have high prevalence of goitre than those who consume less than two times per day ($p < 0.005$).

Laboratory results indicate that the mean urinary iodine excretion level was greater than $9 \mu\text{g}/\text{dL}$ in almost all study groups which showed that the study group had adequate dietary intake of iodine. The results also showed that other factors besides iodine deficiency were playing role in goitre causation.

Laboratory analysis showed that drinking water was contaminated with *E.coli* and coliform micro-organisms to the level which could be considered unfit for human consumption. However, the relationship between microbial load in the drinking water and the goitre prevalence was not significant.

The results therefore indicate that the prevalence of goitre in Gamo-Gofa, Ethiopia was due to hereditary as well as dietary factors.

It is recommended that further investigations be carried out to elucidate the role played by both dietary factors and familial tendency in goitre prevalence. Appropriate action is also required to revitalize and effect the interrupted IDD control programme in the country so as to reduce the prevalence of goitre in the study area and country as a whole.

CHAPTER ONE

1. INTRODUCTION

Iodine deficiency is one of the major nutritional problems in developing countries. An estimated 800 million people in the world are at risk from iodine deficiency (Hetzel, 1987), while 190 million suffer goitre and 3 million suffer overt cretinism and consequently millions some degree of intellectual deficit (Grant, 1991).

Iodine is a constituent of the thyroid hormones thyroxin (T4) and triiodothyronine (T3). It occurs naturally in the human body in amounts between 15–20 mg, while the essential requirement for normal growth is only 100–150 µg per day. Thyroid hormone plays an important role in growth, development, and metabolism in virtually all tissues (Bernal *et al.*, 1977). Lack of iodine causes Iodine Deficiency Disorders (IDD).

A deficiency of iodine manifests itself in a number of ways. These include goitre which was first described in the second century AD and cretinism which was first recognised in the 17th century (Hetzel, 1989). Iodine deficiency also affects intellectual capacity, which has an important mediating role between education and entrepreneurial activity. Therefore, it is believed that those ailments which have greater negative effects on intellectual capacity are likely to cause relatively greater reduction in development.

In a severe iodine deficiency, there is abnormality exhibiting irreversible anomalies of intellectual and physical development. The anomalies are grouped under the general heading of endemic cretinism. Cretinism is associated with goitre and severe iodine deficiency. These comprise mental deficiency, together with a predominant neurological syndrome consisting of defects of hearing and speech, and with characteristic disorders of stance and gait of varying degree, or predominant hypothyroidism and stunted growth (Querido *et al.*, 1974).

Much work has been carried out on the epidemiology, aetiology, pathogenesis and to determine the adequate measures for control of goitre (Delange *et al.*, 1968; Ermans *et al.*, 1983; Kelly, 1960; Thilly *et al.*, 1972).

In severe endemic goitre areas, maternal thyroid status could influence development of the thyroid function of the fetus (Stanbury, 1972). More stillbirths, infant deaths, and endemic cretinism occur among offspring of women with low T₄ and elevated thyroid stimulating hormone (TSH) during pregnancy (Pharoah *et al.*, 1976).

Thyroid carcinoma is also thought to occur more frequently in populations with endemic goitre (Wahner *et al.*, 1956). The distribution of the type of cancer is different from that found in nonendemic populations. Hence, endemic goitre influences an accompanying factor related to incidences of different histological types of thyroid cancer. It has been established that aggressive tumor types prevail in endemic

goitre areas, and these are responsible for the relatively serious progress of thyroid malignancy in these areas. It was also reported that a high proportion of thyroid cancers in endemic goitre areas occurs in recurrent goitres, prophylaxis will also reduce the size of this therapeutic problem group (Riccabona, 1972).

Several studies suggest a possible relationship between increased level of dietary iodine and the occurrence of Hashimoto's thyroiditis and thyrotoxicosis (Vidor, 1973). However, some opinions indicate the benefits of iodine prophylaxis against endemic goitre far outweigh its undesired side effects.

Thyroid response to excess iodine are autoregulatory in nature, since they are mediated within thyroid itself, rather than by change in TSH secretion. Large doses of iodine, administered acutely, produce an absolute or relative inhibition of organic iodinations and iodothyronine synthesis (Wolff-Chaikoff effect), preventing increase in hormone synthesis that would otherwise occur. This effect which is independent to that of TSH, secretion change is distinctly evident in patients with Graves' disease or those with autonomously functioning toxic nodules (Wartofsky *et al.*, 1970).

In Ethiopia, the existence of goitre was recorded at the turn of the century. Results of studies done in various parts of the country, however, indicated that urinary iodine excretion (UIE) did not increase linearly with the decreasing total goitre rate (TGR), which suggests

goitrogenic factors and/or micro-organisms from water sources might be contaminants

(Cherinet *et al.*, 1995).

A minor survey of goitre done in Gamo-Gofa (ENI, 1994) indicated a high prevalence of goitre of 65% in elementary school children. However, the level of UIE of between 3.5 - 5 $\mu\text{g/dL}$, suggested an adequate iodine intake. This gave rise to speculation that this high prevalence of goitre was probably aggravated by other factors which inhibit iodine uptake by the thyroid gland. Also, in some countries, iodine prophylaxis has not been wholly successful (Gaitan *et al.*, 1986). These findings imply that factors other than iodine deficiency play a role in the causation of goitre. In the light of the aforesaid, it is hoped that the findings from this study will serve as baseline information for the Ethiopian National IDD control Programme, which is just about to start.

1.1 Justification of Research Problems

Goitre has been recognised as a problem in Ethiopia since 1903 (ICNND,1959; Hofvander,1970; Kelly, 1960). Since then, studies (Wolde-Gebriel *et al.*, 1993) have indicated the existence of this disease in its varied forms in hilly as well as low land areas of Ethiopia. Wolde-Gebriel *et al.*, also revealed that the proportion of people with goitre was more than expected being in the range of 0.4 - 66.3% among school children. However, unlike this study non of these studies considered the possible role of other factors in contribution to iodine deficiency.

1.2 Objective

This study has the overall objective of determining the familial tendency and dietary association of goitre in Gamo-Gofa, Ethiopia.

1.2.1 Specific-Objectives

1. to determine the prevalence of goitre among (elementary) school children (6–18 years) and their parents.
2. to determine urinary iodine excretion in school children and their biological parents.
3. to determine type and the frequency of food consumption.
4. to determine the association between microbial contamination of drinking water and goitre.

1.3 Study Hypothesis

Familial tendency and diet have no role causing in iodine deficiency problems.

1.4 Expected Benefits of the Study

The causes of goitre in Ethiopia have been determined as iodine deficiency. However, there are areas in which adequate level of iodine intake was recorded but had high prevalence of goitre. This gave rise to speculations that, much of the goitre stems from other factors, which may include familial tendency, consumption of goitrogens, and contamination of drinking water with faecal E. coli and other Coliforms.

The Ethiopian National Iodine Deficiency Disorders Control Programme is in the process of being revitalised. The present study will generate data which will assist the IDD control programme to formulate a viable plan of action. It will also provide baseline information for the evaluation of the programme and give a good insight in causation of IDD, and may also suggest possible means of alleviating the problem.

CHAPTER TWO

2. LITERATURE REVIEW

2.1 IDD Status in Ethiopia

Iodine deficiency disorders (IDD) manifest themselves in a variety of conditions such as goitre, abortions, stillbirths, mental disorders and milder psychomotor defects, and increased perinatal and infant mortality.

Several isolated surveys, reports of foreign travellers and physicians during the Italian invasion of Ethiopia in the second World War suggest a wide occurrence of goitre of different magnitudes in different parts of the country (Kelly *et al.*, 1960). Studies done by ICNND (1959) and Hofvander (1970) also demonstrated that goitre is one of the nutritional problem of public health significance in Ethiopia. Following the results of these studies, W-Gebriel *et al.*, (1993) made a nation-wide goitre assessment in representative samples of the population from 1979–1981. The survey considered school children and households and the final results revealed that a large proportion of school children and households in the country were affected. The prevalence rate was significantly higher in areas of high altitude (>2000m) than low altitude areas (<2000m) (W-Gebriel *et al.*, 1993). The highlands in the north, western and central regions were the ones most affected according to this study. The study also estimated cretinism from an epidemiological model based on existing data to be 59,000 persons (1.17 per 1,000). Based on the recommendations of the aforementioned studies, it was

agreed to institute an IDD programme in the country. However, prior to the initiation of the programme, it was necessary to also make an assessment in the unsurveyed areas. The results of this assessment, combined with baseline data generated in 1987/88 were to be used to plan a viable IDD control programme. The study of unsurveyed areas was conducted in 1987/88 and included biochemical analysis of urinary iodine, T3, T4, and TSH (ENI, 1987/88). Results of the study revealed that the goitre prevalence rates in the urban and rural areas ranged from 1.92 to 58.86 and 0.16 to 73.55% respectively, while the urinary iodine excretion rates varied from 22.38 to 142.44 $\mu\text{g}/\text{day}$ and 10.35 to 97.88 $\mu\text{g}/\text{day}$ in the urban and rural areas respectively.

It was concluded that IDD is a public health problem in Ethiopia. Observation of laboratory results revealed UIE values correlated well with goitre prevalence however, the correlation was not linear which indicated the possible existence of goitrogenic factors.

2.2 Goitre and the Thyroid Gland

Goitre means a thyroid gland bigger than normal. People with iodine deficiency have goitre because they do not manufacture enough thyroid hormones.

A normal thyroid gland should have a minimum size compatible with euthyroidism (a condition of normal thyroid function) under conditions of normal iodine intake (100–150 $\mu\text{g}/\text{day}$) Perez *et al.*, (1960). This gland should be non-palpable. A thyroid gland whose lateral lobes have a

volume greater than the terminal phalanges of the thumbs of the person examined will be considered goitrous.

2.2.1 Estimation of Thyroid Size

The most commonly used method for estimation of thyroid size is palpation and defined by Hetzel (1989).

Grade 0 no goitre

Grade 1a goitre detectable

Grade 1b goitre palpable and visible only when the neck is fully extended. This stage also includes nodular glands, even if not goitrous.

Grade 2 goitre is visible when the neck is in normal position.

Grade 3 very large goitre that can be seen from distance.

The total goitre rate is the occurrence of stages 1a–3; the visible goitre rate is the prevalence of stages 2 and 3.

However, this classification was summarized by WHO–Geneva 1990 with 3 grades.

Grade 0 – no goitre.

Grade 1 – palpable but not visible goitre.

grade 2 – visible goitre of all size.

2.2.2 Endemic Goitre

An area is arbitrarily defined as endemic with respect to goitre if more than 10 per cent of the population or of the children aged 6 to 12 years are found to be goitrous (Hetzel, 1989).

The figure of 10% was chosen

because higher figures usually imply that environmental factors are involved. However, prevalence of several per cent has been found even when all known environmental factors are controlled for.

In recent years, it has been recognized that the consequences of iodine deficiency extend beyond goitre and cretinism to conditions such as

failure of normal human reproduction. In order to cover these various sequelae of iodine deficiency, the term iodine deficiency disorders (IDD) was introduced by Helzel (1983).

2.3 Biochemical Markers of Iodine Intake

2.3.1 Urinary Iodine Excretion (UIE)

The concentration of iodine is currently the most widely used indicator of iodine consumption level in a given community for several reasons. Over 90% of the body's iodine is excreted by urine; thus the iodine level in urine reflects subject's intake. Since the absolute minimum daily iodine requirement is about 50 μg , a urinary iodine level of less than 50 μg per day means iodine deficiency. Urine is also easy to obtain at field condition when compared with serum. Iodine in urine is stable and can withstand collection and transportation under field conditions. Urinary iodine measurements have usually been technically simpler and cheaper than other iodine indicator methods (Dunn, 1993). Thus, it can be used to classify the extent of iodine deficiency in a given area as follows:

1. Mild IDD with goitre prevalence in the range 10–30% and with average urinary iodine excretion (UIE) 3.5–5.0 $\mu\text{g}/\text{dL}$.

2. Moderate IDD with goitre prevalence 20–50%, some hypothyroidism with average UIE level in the range 2.0–3.5 $\mu\text{gI/dL}$.
3. Severe IDD indicated by high prevalence of goitre 30–100%, average urinary iodine below 2.0 $\mu\text{gI/dL}$.

2.3.2 Hormonal Measurements in Serum.

2.3.2.1 Triiodothyronine (T3).

Triiodothyronine (T3) level in human serum is used in the differential diagnosis of thyroid disease. It is transported in serum primarily by thyroid binding globulin (TBG). T3 is known to have more metabolic activity than T4. A substantial part of the endogenous T3 pool is derived from the extra-thyroid deiodination of T4 (Larsen, 1972).

A normal T3 level is generally an indication of the absence of thyrotoxicosis, provided that a reduced TBG level is excluded. However, while T3 level is the most sensitive test for hyperthyroidism the T3 level alone is not sufficient for the diagnosis of hypothyroidism, since a poroportion of hypothyroid patients have normal T3 values. The approximate normal range for T3 concentration suggested was 0.8 to 2.7 nmol/L.

2.3.2.2 Thyroxine (T4).

T4 normally present in plasma in approximatly fifty-fold of circulating T3 (Evered, 1994) and accounts for at least 90% of the circulating protein bound iodine. It is transported bound principally to thyroxine binding globulin (TBG) and secondly to thyroxine binding prealbumin (TBPA) and

albumin (Oppenheimer, 1968). Total thyroxine levels in serum or plasma have long been recognised as an important indicator of thyroid status. In overt hypothyroidism, T4 levels are generally depressed and in overt hyperthyroidism T4 levels are generally raised.

Approximate normal value range for serum T4 is 4.8–12.8 $\mu\text{g}/100\text{ml}$.

2.3.2.3 Thyroid Stimulating Hormone (TSH)

The hypothalamus and the pituitary and thyroid gland are the main participants in a regulatory system that ensures the constant supply of thyroid hormone to tissues. The principal regulatory step involves stimulation of pituitary TSH secretion by the hypothalamic tripeptide, thyroid releasing hormone (TRH) (Wilber, 1973; Jackson, 1982). TSH, in turn, stimulates all aspects of thyroid follicular cell activities, including hormone synthesis and secretion. The pituitary, a small control gland in the brain, detects the low thyroid hormone levels in the blood, and makes more of its control hormone, called thyroid stimulating hormone (TSH), acts upon the thyroid gland to stimulate the production of thyroxine (T4) and triiodothyronine (T3). T3 and T4 levels in turn regulate the secretion of TSH by negative feedback mechanism (Evered, 1974). Approximate normal value range for TSH serum concentration is 1.0–5.5 $\mu\text{IU}/\text{ml}$.

Serum TSH levels are raised in cases of primary hypothyroidism. This increased TSH stimulation is a normal adaptation but it produces a goitre, particularly if stimulation becomes chronic because of continued iodine deficiency. The goitre is a sign that the body is trying to compensate for

lack of iodine. Other causes of goitre exist, but increased TSH stimulation is responsible for goitres in areas of iodine deficiency. The diagnosis of overt hypothyroidism by the low free T4 values is readily confirmed by a raised TSH level.

2.4 Familial Tendency of Goitre

Iodine deficiency has always been present to some extent in goitre, but it is not the only factor which precipitates this disease. Genetic tendency, including defects in enzymes involving iodine and thyroid metabolism have a role in the development of goitre. Familial tendency to develop goitres was studied quantitatively in villages in which goitre prevalence was relatively homogeneous (Ermans *et al.*, 1980). Measurements were made of the proportion of goitrous children in families in which both parents were goitrous, in families in which one parent was goitrous and in families in which neither of the parents was goitrous. The results showed a definite association between parents and their children regarding the prevalence of goitre. However, the difference between goitrous and non goitrous parents were not significant. The authors concluded that differences between the percentage of goitre observed in children of goitrous and non- goitrous parents were not clear-cut and were insufficient to explain the disease merely on the basis of monogenic heredity.

Thyroid hormones which are responsible for the body metabolism are transported in the blood stream by the thyroid binding globulines (TBG). The regulation of TBG synthensis appears to be dependent on a gene

located on the sex chromosome. This condition is based on the observation that inherited TBG deficiency and excess are transmitted as X chromosome linked traits. Affected individuals appear to have an abnormality in the rate of TBG synthesis, since they do not exhibit alteration in TBG degradation (Refetoff *et al.*, 1976).

Another genetic variant of TBG has been recently described: a polymorphic, electrophoretic variant of TBG found with variable frequency in population of Africa and oceanic origin, inherited in an X-linked fashion (Daiger *et al.*, 1981) and a TBG variant present in approximately 40% of Australian Aborigines which, despite normal isoelectric mobility, exhibits a decreased binding affinity for thyroid hormone and great heat lability (Sarne *et al.*, 1983).

2.5 Role of Goitrogens in Goitre

The antithyroid properties of thiocyanate (goitrogen) were first shown by Barker (1936) in patients with hypertension, treated with large doses of thiocyanate. Thiocyanate or thiocyanate-like compounds appear primarily to inhibit the active concentration mechanism of iodide. They also stimulate the release of iodide from the thyroid by inhibiting iodide binding mechanism to form thyroxine (Gaitan, 1980).

Goitrogens are present at varying degrees in many food stuffs, notably rutabagas, turnips, and cabbages. They cause goitre by inhibiting the synthesis and secretion of thyroid hormones.

Several plants, used as food, also contain cyanogenic glucoside – beans, corn, sorghum, sweet potato, lettuce and peas. With the exception of cassava and certain beans (*phaseolus lunatus*), these glucosides are mainly localized in the inedible portions of the plant. The amounts in the edible portions are low enough for the food to be safe for consumption. Cyanogenic glucoside exert goitrogenic effect when converted to thiocyanate in living organisms (Gaitan, 1980).

Goitre and cretinism (a form of mental retardation) are caused by iodine deficiency and are common in areas with low iodine intake. These disorders are aggravated considerably by continuous dietary exposure to insufficiently processed high cyanide cassava. Another nutritional factor that may be involved in the goitrogenic action of cassava or other diets in human is protein calorie intake because the indigenous conversion of thiocyanate (SCN) requires sulfur amino acids. Experiments with pigs have indicated that deficiency protects against antithyroid action of cassava, by reducing the quantity of SCN arising from hydrocyanic (HCN). Other food sources have also been considered because in some countries, iodine prophylaxis has not been wholly successful (Gaitan, 1980) and the fact that despite adequate iodine intake, few cases of goitre still appear (Gaitan *et al.*, 1986). Certain marginal iodine intake areas have been shown to have high and low endemicity such as Andean region of Colombia (Gaitan *et al.*, 1978), Venezuela (Roche, 1959), Western New Guinea (Choufoer *et al.*, 1965).

The presence of polysulfides in the several vegetables of the cruciferae

family has also been described, as antithyroid substances. The major components of onion and garlic have been identified as small aliphatic disulfides, which have marked antithyroid activity in rats (Gaitan, 1980).

Millet or sorghum consumption has also been incriminated in goitre causation in the Sudan (Osman, 1981) in which, a thionamide like substance was incriminated.

In Tasmania seasonal variation in goitre prevalence in schoolchildren was noted despite adequate iodine intake, and in which chlorine, and isothiocyanate, were suspected as the principal goitrogens in the milk (Gibson *et al.*, 1960). In Finland, also goitrin and thioglucoside present in cow's milk from the goitrous region, was considered the causative factor for prevalence of endemic goitre. This information was confirmed by feeding rats with milk for one to two years (Gaitan, 1980).

The nut of the *Araucaria araucana* (pinon) has been suspected to have a role in the pathogenesis in an endemic goitre Indian reserve in Chile (Gaitan, 1980). A pinon diet showed goitrogenic activity in rats that was not due to iodine deficiency.

An excess iodine intake, arbitrarily defined as 2 mg or more per day, inhibits the proteolysis and release of thyroidal hormones and eventually produce "iodide goitre" and hypothyroidism. Sustained ingestion of seaweeds (kelp) rich in iodine cause "endemic coast goitre" (Suzuki, 1980).

2.6 Water Contamination and Goitre

Some bacterially and chemically polluted drinking waters have also been found to cause goitre. In northern Virginia, a study by Vought *et al.*, (1974) indicated that *E.coli* contained an antithyroid compound. In Greece, a non-endemic area obtained drinking water from deep boreholes in contrast to an endemic whose drinking water came from surface sources which contain few salts, iodine, chlorine, carbonate, bicarbonate, sodium, zinc and which were more often polluted with *E.coli* and coliform organisms (Malamos *et al.*, 1971). Organic antithyroid compounds derived from sedimentary rocks (shales, coals, etc) contaminates water supply in areas where goitre persists despite iodine supplementation.

Bacteriological studies carried out in 21 of the 41 localities in western Colombia have indicated some evidence that the overall concentration of bacteria in the pipe line system is related to a high goitre prevalence (Gaitan, 1980). On another study (Gaitan, 1973), noted a decrease in goitre prevalence after introduction of a piped drinking water programme. The author concluded that pollution of water with microorganisms was a contributory factor to goitre endemia.

Some microorganisms like *Klebsiella pneumoniae* (found to degrade goitrogenic substance) in water sources associated with decreased goitre prevalence because of natural biodegradation of the organic contaminants that produce goitre (Maugh 1979; Ermans 1980).

Studies in Trujillo state, Venezuela, showed higher lithium (Li^+) concentration in the water supply of a high-incidence endemic goitre locality than in that of nearby nonendemic locality. Experimental observations indicated that Li^+ at this concentration could be goitrogenic (Cevallos, 1982). More recent studies (Gaitan, 1980; Koutras, 1980) in endemic and nonendemic areas of Greece showed that the concentration of both E.coli antibodies and IgG were higher in the goitrous population than in nongoitrous population of the endemic area, and the IgG concentrations were higher in the endemic area, where drinking water was subject to pollution, than in nonendemic area which had a non-polluted water supply. Evidence by Weiss *et al.*, (1983) suggested immunologic cross-reactivity between human thyroid plasma membrane and antigenic determinants in E.coli and *Y. enterocolitica*, as well as demonstration of thyroid-growth-stimulating immunoglobulins in some goitrous individual (Weiss *et al.*, 1983). All these studies emphasize the fact that factors other than iodine deficiency play a role in causation of goitre.

2.7 Gaps in Knowledge.

There is no work done on association between goitre prevalence and familial tendency of goitre in Ethiopia. There are also some food sources which may be contributing to endemicity of goitre that have not yet been assessed, especially in the present study area.

CHAPTER THREE

3 METHODOLOGY

3.1 Study Site

Gamo-Gofa is found in south western part of Ethiopia and has hot, humid and rainy climate conditions. Gamo-Gofa shares borders with Kenya to the south, Sidamo zone to the west, Shoa to the north and Kaffa zone to the west (Appendix 10).

The study sites were Gofa-Zuria and Kucha Woredas from which the schools were selected, is said to have a high prevalence of goitre in school children which was not attributed fully to the iodine deficiency (ENI, 1994). The study sites are located 600 km south of Addis Ababa the capital city of Ethiopia. These two woredas were selected based on the following criteria:

- *Rural communities – to avoid problems related to the use of foods which are not produced in the region.
- *Limited population movement.

The goitre survey carried out by Ethiopian Nutrition Institute (ENI, 1994) was used as the basis for selecting the woredas with high prevalence of goitre. This was confirmed by conducting a pilot survey in 4 elementary schools, two from each woreda. School children were used because they are more readily available than other community members.

3.2 Study Population

Gamo-Gofa is located in the Southern Nations, Nationalities and peoples' region. The total population of the region was indicated to be more than 10 million. Female to male ratio in the population was about 1:1 ratio (CSA, 1995).

The population in the two woredas is about 268,648 of which about 50% were females. They are not densely populated like other southern regions of the country. More than 98% of the population depend on subsistence farming. The people grow a variety of roots, tubers, leafy vegetables, cereals and pulses. Some of the leafy vegetables and tubers are peculiar to the region.

Christianity is the predominant in the region with more than 97% of the population being followers.

3.3 Type of Study

This was a cross-sectional study involving descriptive and analytical components carried out between 1996 and 1997 among school children and their parents in Kucha and Gofa-Zuria woreda, Gamo-Gofa, Ethiopia. The study was done among elementary school children aged 6 to 18 years and their parents. The study involved goitre assessment and urine samples analysis, interviewing mothers about diet frequency, analysis of water samples from drinking sources.

3.4 Ethical Considerations

- * Obtain consent from respondents.
- * Do not exploit informants
- * Research permit
- * Safe guard informants rights, interests and sensitivities.

3.5 Study Instruments

1. Questionnaire – A structured questionnaire (Appendix 1) consisting of questions on demography, clinical examination of goitre, drinking water sources, pedigree history of goitre of grand parents and food consumption frequency was administered to parents.
2. Iodine free vacuum test tubes used for collection of urine samples at the field.
3. Spectrophotometer is an instrument used for measuring urinary iodine concentration.
3. Colony counter – was used to quantify the microbial count in water from drinking water sources.

3.6 Selection and Training of Interviewers

3.6.1 Selection Procedure

There were two steps in the selection procedure:

Firstly, a recruitment notice was posted seeking a registration of candidates. The following criteria was used in selection of interviewers:

- * Sex; female. This is because it is culturally difficult for male interviewers to interact with women particularly in rural areas.
- * Good command of the local language of the communities, living in the

selected area.

- * Physically fit and healthy to move around in the villages.
- * Agree to the terms of payment
- * Priority was given to those with experience in household surveys and those who grew up in the rural areas.

Second stage involved administration of a questionnaire for purposes of assessing their understanding, accuracy and legibility of their hand writing. Finally four were selected and another four were registered as reserve.

3.6.2 Training

To obtain reliable information it depends greatly on the quality, good training and supervision of the interviewers. Thus, training of the field staff was given utmost attention.

The training was given by the principal investigator covering the purpose, objectives and method of the study and how to address and approach subjects in culturally acceptable manner.

3.7 Pilot Survey

Pilot survey started with preparatory stages; which included writing letters to responsible organizations to get research permit and other supports, making preliminary visit to study sites assess laboratory conditions to arrange for laboratory analytical components of the study, translating questionnaire to Amharic and getting it ready for pilot survey. After getting to the field assistants were recruited and trained

for five days. The pilot study was done in four schools two from each woreda. The questionnaire and interview techniques were tested around the study areas. Intra-interviewer and inter-interviewer variation was assessed and appropriate measures were taken to reduce variations. Some of the measures taken were, interviewers were asked to do interview several times until inter-interviewer variation was reduced and not significant. In the mean-time intra-interviewer variation was also assessed and found to be not significant. During this time the ability of the assistants was evaluated practically. Although, clarity and relevance of the questionnaire was checked by administering to parents of the children. Clinical examination of goitre was also done both in parents and their children. After obtaining the results of pilot study relevant adjustments were made on the study questionnaires. Actual survey followed almost immediately after reviewing and modification of the methodology and questionnaire.

3.8 Sampling Frame and Sampling Procedure

Sampling frame of the study was an elementary schools children age 6–18 years in the two woredas of Gamo-Gofa from which samples of study were identified.

Prior to the actual study pilot survey was conducted in four schools of the region. During the pilot study it was identified that sample size of fathers having goitre was found to be very rare, which could make a problem during statistical analysis. As the result of this the sample size was maximized than intended.

For actual study, multi-stage sampling was applied. Based on the previous studies and pilot study, Gofa-Zuria and Kucha woreda were selected purposely from which seven elementary schools accessible in rainy season were taken for this study. This is because apart from the main road from Addis Ababa to Sawla town, all others were earth roads making some areas inaccessible in rainy season. Identification of schools was done in collaboration with regional office for ministry of education. Schools were found in a distance of 40 to 150 km from each other. Before starting actual work letter of cooperation from regional office was distributed to identified schools and health institutions.

After introduction of team members to teachers, explanation about the objectives of the study was given to them. Clinical examination for goitre was done in all selected schools, in all children. Five hundred ninety seven children age 6 to 18 years were identified by systematic random sampling from seven elementary schools. Identification was effected after undertaking clinical examination of goitre in each school. When two children from one household were encountered, one of them was taken by random selection method. If one of the parents of the child was missing for any reason during the day of the clinical examination then that child and his parents were substituted by reserve. Of these 597 school children sub-samples were again selected by systematic random sampling for urine sample collection. It was established and found to be most practical to collect 40-50 urine samples from an area to assess dietary iodine status of a given community. To get a true reflects of iodine intake the size of urine samples was doubled in this study. After identification of the children, call cards were prepared and given to all selected

children by school teachers to come to school with their biological parents on the day of appointment for clinical examination and collection of other information. Call cards bearing the names of children (from school register) included; name of the head of the household, message, date, time, place of appointment and signature of school director. (Summary of sampling procedure indicated in figure 1).

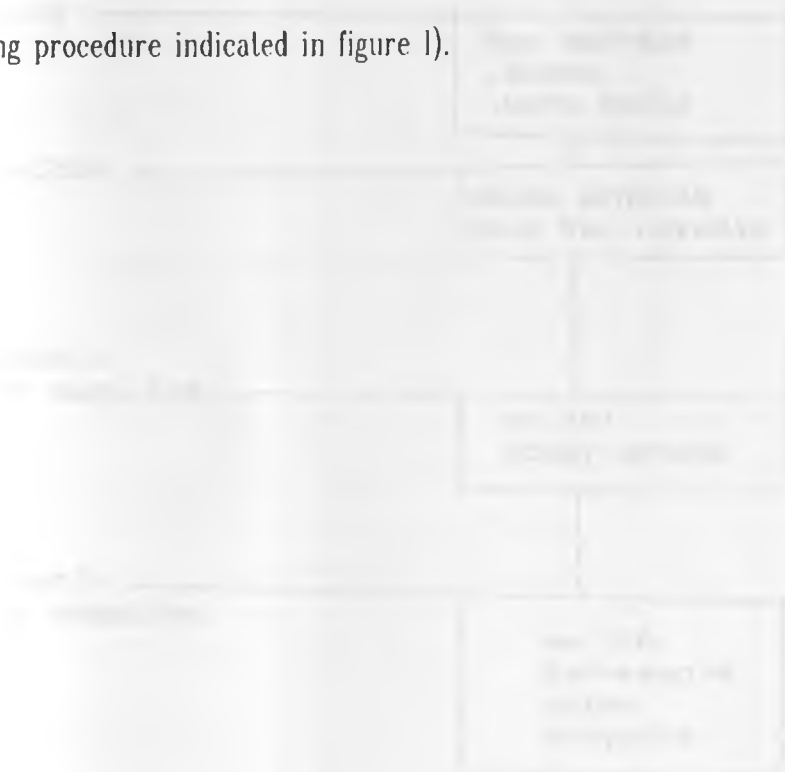
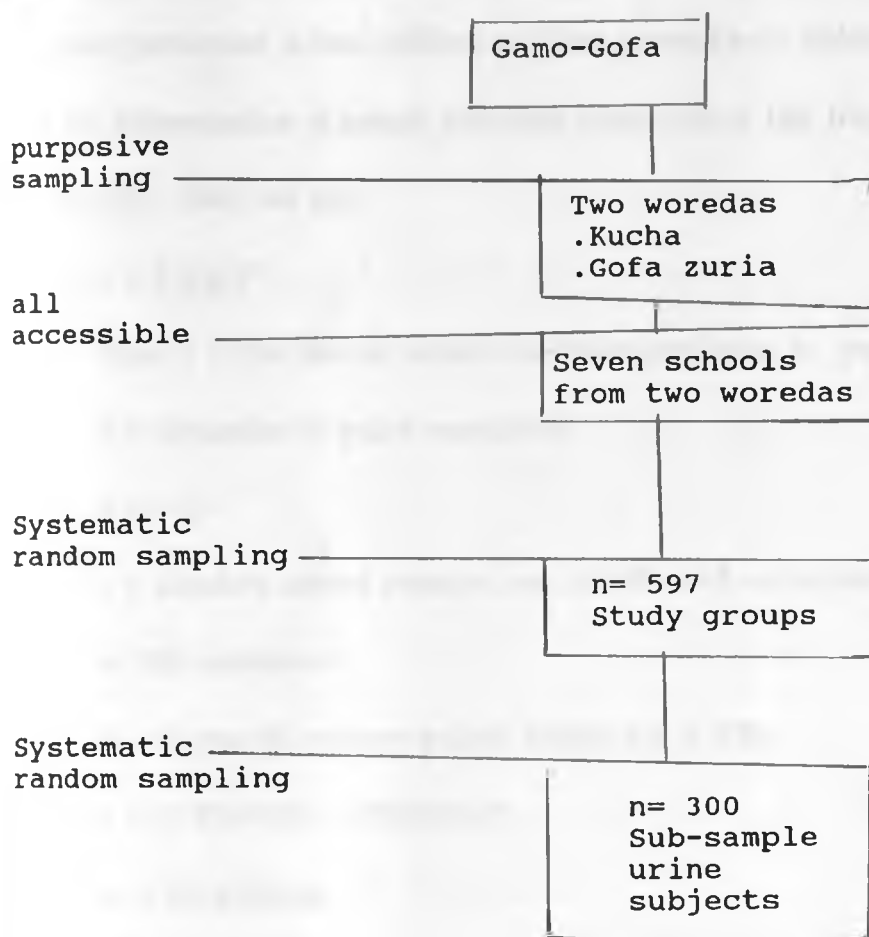


Figure I.FLOW CHART - MULTISTAGE SAMPLING PROCEDURE

3.8.1 Sample Size Determination

In order to test null hypothesis that goitre has no familial tendency and diet has no effect on goitre prevalence, school children and their parents were included.

For the determination of sample size total goitre rate of 65% from previous goitre survey results (ENI, 1994) was used.

$$n = Z^2 pq/d^2$$

Where n = the desired sample size (when population is greater than 10,000)

p = proportion of goitre prevalence.

$$q = 1-p$$

z = standard normal deviation, set at 1.96 which correspond to 95% confidence.

d = degree of accuracy desired. Usually set at 0.05.

$$n = (1.96)^2(0.65 \times 0.35)/(0.05)^2$$

$$n = 350 \text{ children}$$

10% substitutes

$n = 350 + 35 = 385$ children, when both parents were included this figure would be

1155.

However, in view of avoiding shortage of samples in statistical analysis the sample size was maximized to 1791 children and their parents.

3.9 Activities

3.9.1 Determination of Prevalence of Goitre

In order to identify subjects eligible for inclusion in this study all children, in seven schools were registered by the trained interviewers. The data recorded on a prepared form.

All registered children were assembled on examination post with their biological parents where they were examined for goitre and interviewed.

Goitre examination was carried out according to PAHO/WHO recommended method (Delange *et al.*, 1986)

3.9.1.1 Clinical Examination of Goitre.

Clinical examination for goitre was done by the principal investigator and a health personnel (nurse). Before starting actual work, the two standardized estimation of thyroid size between them selves to minimize variation between them.

The examiner stood or sat (depending on the height of the examinee) facing the subject, placed two thumbs on either side of the subjects windpipe several centimeters below the notch of the thyroid cartilage and rolled the thumbs gently over the thyroid, which lies next to the windpipe. If each lobe of the thyroid was smaller than the parts of the subject's thumb (the "terminal phalanx"), the examiner classified the thyroid as grade 0 (no goitre). If the lobes were bigger, and when the subjects head was tilted backwards and the goitre could not be seen but was palpable, the goitre was classified as grade 1A. If the goitre could be

seen as well as palpated, the subject's head is returned to a position, looking straight ahead. If the goitre could be seen only with the head tilted back, it was called grade 1B. If it could be seen with the subject looking straight ahead it was called grade 2. If it was quite large and could easily be seen from a distance of about 10 meters, it was classified as grade 3. The two biggest goitre grades, grade 2 and grade 3 were also classified as visible goitre rates (VGR). All goitre grades together excluding grade 0 were termed as total goitre rate, (TGR).

3.9.2 Determination of type and frequency of Commonly Consumed Foods

The questionnaire for collecting diet frequency and other information was translated into national language/Amharic (Appendix II) then administered to parents of the children.

In view of assessing magnitude of goitre prevalence in relation to the food being consumed, type of food and frequency of consumption in the village was assessed by interviewing mothers. Mothers were asked to list types of foods consumed commonly by household members. Foods were listed in a given form starting from more common to least. Then for each type of food, frequency of consumption was assessed.

The interview part also included retrospective history of goitre of the grand parents. Mother and father of the child were asked whether their parents had goitre or not. This question only determined visible goitre because, respondents might not know the presence of nonvisible goitre in their parents.

3.9.3 Analysis of Urinary Iodine

3.9.3.1 Principles

Urinary iodine determination was done according to the method of Sandell-Kolthoff (Donn *et al.*, 1993).

This method is based on the principle of urinary iodine determination include wet digestion in strong acid (perchloric acid) at high temperature (110° C). Iodine was measured by its catalytic action on the reduction of the ceric ion to the cerous ion coupled to the oxidation of arsenite to arsenic. The analysis was done at the Ethiopia National Health and Research Institute (ENIIRI), Addis Ababa.

3.9.3.2 Materials and Reagents

Digestion block, spectrophotometer (Beckman model 25), thermometer, pyrex test tubes (13 mm O.D. x 100 mm L), reagents, flasks, bottles, micro-pipettes adjustable volume (5-50 and 100-1000 μ l), laboratory balance (Mettler H35), distilled and deionized water, gloves, french curve and graph paper.

Potassium chlorate (KClO₃, dry powder) (AR)

Perchloric acid 70% (HClO₄, liquid solution) (AR)

Arsenic trioxide (As₂O₃, dry powder) (AR)

Sodium chloride (NaCl, dry powder) (AR)

Sulphuric acid 36 N (H₂SO₄, liquid) (AR)

Ceric ammonium sulphate (Ce(NH₄)₄(SO₄)₄.H₂O, dry powder) (AR)

Potassium iodate (KIO₃, dry powder) (AR) and Distilled and deionized water.

3.9.3.3 Sample Collection Procedure

Urine samples were collected both from children and their parents in iodine free test tubes and were kept in deep freezer in a near by health institute until completion of the field work. Water samples from drinking sources were collected in sterilized brown bottles, and transported for analysis within 24 hour. Both urine and water samples were transported from the field to the laboratory by using ice-box in order to avoid any chemical reactions.

3.9.3.4 Procedure for Determination of Iodine in Urine.

1. 250 μ l of evenly mixed urine samples were pipetted into a 13 x 100 mm pyrex test tubes.
2. 250 ml of water was pipetted into another 13 x 100 mm tube, for blank.
3. Into 13 x 100 mm tubes, 250 μ l of each iodine standard (2, 5, 10, 15, μ g/dL) was pipetted, then deionized water was added to make a final volume of 250 μ l for each tube.
4. Then 750 μ l of Chloric acid solution was added to each tube (samples, blank, and standard), mixed gently.
5. All tubes were heated for 50-60 minutes in a heating block at 110° C.
6. After cooling the tubes to the room temperature 3.5ml arsenious acid solution was added to each tube, then was mixed, let it stand for 15 minutes.
7. 350 μ l of ceric ammonium sulphate solution was added to each tube and quickly mixed. Constant time interval was kept between additions to successive tubes.

8. Exactly 20 minutes after addition of ceric ammonium sulphate to first tube, absorbance was be read at 405 nm in spectrophotometer, and successive tubes were be read at the same interval.

9. A standard curve was constructed by plotting iodine concentration of each standard against absorbance. For each sample, absorption was read off the standard curve and then the corresponding iodine concentration was located on the abscissa. The unit of urinary iodine concentration was termed as, microgram of iodine per one hundred litre of urine ($\mu\text{g}/\text{dL}$). A standard curve was prepared for every batch of analysis. Average standard curve for all individual curves is given in Appendix IV.

3.10 Determination of Micro-organisms in Water Samples

Water samples were collected in sterilized brown bottles from drinking sources. Determination of microorganism was carried out in ENHRI laboratory as follows (Harrigan *et al.*, 1976):

1. Mix sample of water.
2. prepare the dilutions.
 - a. holding a sterile 1 ml blow-out pipette vertically, introduce the pipette tip not more than 3 cm below the surface of the sample and suck up and down 10 times to the 1 ml mark. Withdraw 1 ml sample, touching the tip of the pipette against the neck of the bottle to remove the excess of liquid adhering to the outside of the pipette. Transfer to the first tube of the dilution series. The pipette must not contact the diluting fluid. Blow out the content of the pipette. Discard this pipette and label the first dilution tube 1/10.
 - b. Using a fresh sterile pipette, mix the contents of the first dilution

tube by sucking up and down to the 1 ml mark 10 times. Then withdraw 1 ml of first dilution and transfer to a second tube of sterile diluent expelling the content of the pipette as described above. Discard this pipette and label the second dilution tube 1/100.

c. Further dilutions of 1/1000, 1/10000 can be made similarly as required, depending on the probable bacterial count of the sample.

3. preparing the plates.

a. Using fresh sterile pipette, mix the content of the final dilution tube, eg. 1/100, by sucking up and down ten times. Withdraw 1 ml of the dilution and transfer the contents to a sterile petri-dish.

b. In the same way, the same pipette may be used to transfer into petri-dish 1 ml of the 1/10 dilution and then do the same for the original sample.

4. Pouring the plates.

a. To each plate, add 10 ml of molten agar medium at 45°C and immediately mix the medium and inoculum by a combination of to-and-fro shaking and circular movements lasting 5-10 sec. Then the plates are incubated and then the number of the colonies is counted. Counts should be made on plates which contain fewer than 300 colonies.

This method (Most Probable Number count (MPN)) provides an estimate of the number of living organisms in a sample which are capable of multiplying in a given liquid medium. The results are given as number of cells count in milli-liter of water sample.

3.11 Monitoring Validity and Reliability of the Data

Data on the questionnaire were edited every day after work.

Laboratory instruments were been calibrated before and in between use.

The assistants were regularly supervised. The data on the questionnaire was checked every day for gross errors at field and for completeness of the information, consistency of answers and for the proper filling of the forms by enumerators.

3.12 Data Processing

Suitable software packages (dBASE, EPIINFO version 5 and SPSS) were employed for data entering, cleaning and analysis. Both descriptive and analytical methods were used to present the results of this study. The following statistical methods were used to assess level of significance between and among variables used in the analyses.

Odds ratio and chi-square with cross-tabulation were used to assess level of significance between proportions (to assess familial tendency of goitre between children, and their biological parents, and dietary association of goitre).

Further more to assess strength of association between dependent and predictor variables logistic regression was applied. For this purpose 300 subjects whose data was complete with respect to hereditary factors, diet and urinary iodine excretion were considered.

t-test and correlation were used between group means (UIE and TGR) to assess level of significance. One-way analysis of variance was used among group means (mean UIE's) to assess level of difference. In general it was intended:

* to determine the relationship between prevalence of goitre and iodine status (UIE)

* to assess the trend of goitre between groups with and without goitre.

* to examine the relationship of goitre with diet (food consumed) and type of drinking water.

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CHAPTER FOUR

4. RESULTS:

4.1 Demographic Nature of Study Population

Mean frequencies and percentages are used to present information in this chapter.

Of the 1791 subjects included in this study, 597 were school children, while the remaining (1194) were their parents (both mother and father). The male to female ratio of the study children was 2:1. The mean ages of male and female children were 11.4 ± 2.2 years and 11.3 ± 2.3 years respectively. Statistically there was no significant age difference ($p > 0.05$) between the male and the female children. Overall mean age of the study children was found to be 11.4 ± 2.2 years ranging from 6–18 years. Majority of the children (71.2%) were within the age range 6–12 years. The mean ages of the mothers and fathers were 35.7 ± 7.4 years and 45.5 ± 10.5 years respectively. The mean age of the fathers was significantly higher ($P < 0.001$) than the mean age of the mothers.

Majority of the population (67%) were Gofa tribe, while Gamo accounted for 13% and the Wolayta and Amahara tribes constituted the rest. For religion, protestants were the majority at about 66%, while orthodox was 30% and the Moslems only 1.2%.

4.2 Goitre Prevalence in School Children and Their Parents

Table 1 gives the magnitude of total goitre rates among the children and

their biological parents. Children, both male and female, were divided into two age groups 6–12 years (category for children) and 13–18 years (category for adults), while the parents, both male and female, were divided into two age groups 20–45 years (average child bearing age) and 46 years and above, for examination of both the presence of total goitre and visible goitre. It was observed that the prevalence of goitre was strongly dependent upon both age and sex. The higher age range children had lower goitre rates except in female children where the total goitre rate remained almost unchanged between the two age groups, while visible goitre rate increased from 35.4% to 42.0% from age range 6–12 years to the 13–18 years. This was noted to be due to increase in grade III goitre.

For both parents, the goitre rates of the age range 20–45 years were higher than the goitre rates of the age range 46 years and above. In children as well as their parents and for all age categories, goitre rates (gross and visible) were higher in females than in males, and the trend was more pronounced in the parents than in the children. Total goitre rates (sub-total for children and parents, all sex) were 69.4%, 74.8%, 19.6% and 64.2% for male children, female children, and fathers and mothers respectively, while visible goitre rates were found to be 13.9%, 36.9%, 4.9% and 35.5% for male children, female children, and mothers and fathers respectively.

From these results it could be noted that total goitre rates (%TGR) were considerably higher than visible goitre rates (%VGR) for all groups. Both total goitre rates and visible goitre rates were significantly lower ($p < 0.01$) in fathers than in all the other groups. Overall total goitre rate

(%TGR) in the study population was 51.5% while visible goitre rate was 21.7%.

On moving from low grades of goitre (1a and 1b) to high grades of goitre (2 and 3), it was noticed that the subjects had problems with breathing, swallowing and voice commonly associated with goitre increase. These problems were more pronounced in higher age groups than in the low age groups.

Table 1. Goitre rates by age and sex of children and parents

Sex	Age (Yrs)	Total Exam.	Total Goitre (%)	Visible Goitre (%)
Children				
Male	6 to 12	277	71.5	22.7
	13 to 18	122	64.4	11.5
Total		399	69.4	13.9
Female	6 to 12	148	75.0	35.1
	13 to 18	50	74.0	35.1
Total		198	74.8	36.9
Parents				
Fathers	20 to 45	349	25.8	6.0
	46+	248	10.9	3.2
Total		597	19.6*	4.9*
Mothers	20 to 45	549	65.4	36.4
	46+	48	50.0	25.0
Total		597	64.2**	35.5**
Grand Total		1791	51.7	21.7

* Significantly ($p < 0.001$) lower than **

4.3 Goitre Prevalence Against Physiological Conditions of Mothers

Table 2 summarizes the desegregated values of both total and visible goitre rates against physiological conditions (pregnant, lactating, and nonpregnant nonlactating) of the mothers. There were no significant differences ($p>0.05$) among the groups of mothers in both total goitre and visible goitre. Visible goitre rates were 34.8%, 33.6% and 37.1% for pregnant, lactating and nonpregnant nonlactating respectively. For all groups, the total goitre rates were approximately double that of the visible goitre rates. The total goitre rate was however, slightly higher in pregnant women than in both lactating and nonpregnant nonlactating women but, there were no significant ($p>0.05$) among the rates.

Table 2. Goitre rate by physiological conditions of the mothers

	Total exam.	Total goitre (%)	Visible goitre (%)
Pregnant	46	73.9	34.8
Lactating	241	63.9	33.6
Nonpregnant nonlactating	310	62.9	37.1
Total	597	64.2	35.5

4.4 Urinary Iodine Excretion Categories with Goitre

Table 3 shows urinary iodine excretion (UIE) in relation to total goitre rate and sex. Number of individuals were placed in the categories to which their UIE values belong. For all categories of urinary iodine excretion the number of individuals having goitre was greater than those without goitre except fathers. In the case of fathers, those with goitre were less than those without goitre for all UIE categories. Rate of increase in individuals without goitre with the increasing UIE categories was higher in fathers than other groups. Of the 300 urine samples analyzed only 31 had UIE values less than 2 $\mu\text{gI/dL}$, while 145 had UIE values greater than 10 $\mu\text{gI/dL}$.

Table 3. Goitre rate by desegregated categories of urinary iodine excretion and by sex.

		urinary iodine excretion categories ($\mu\text{gI/dL}$)					
		=====					
Group	goitre	<2 n	2.1-5.0 n	5.1-10 n	10 ⁺ n	Total n	
<hr/>							
children							
	no	-	(33) 2	(38) 3	(16) 3	(22) 8	
female	yes	(100) 4	(67) 4	(62) 5	(84) 16	(78) 29	
<hr/>							
	no	(20) 1	(25) 2	(41) 11	(26) 6	(32) 20	
male	yes	(80) 4	(75) 6	(59) 16	(74) 17	(68) 43	
<hr/>							
parents							
	no	(40) 4	(38) 6	(54) 14	(40) 19	(43) 43	
mothers	yes	(60) 6	(62) 10	(46) 12	(60) 29	(57) 57	
<hr/>							
	no	(66) 8	(90) 10	(59) 13	(55) 30	(61) 61	
fathers	yes	(34) 4	(10) 1	(41) 9	(45) 25	(39) 39	
<hr/>							

(n) denotes number of individuals in that category
Number in parenthesis is percentage

4.5 Goitre Prevalence in Relation to Level of Urinary Iodine

Excretion.

Goitre prevalence in relation to urinary iodine excretion is shown in Figures 2 and 3. Figure 2 indicates total goitre rates by level of urinary iodine excretion while, Figure 3 shows visible goitre rates by level of urinary iodine excretion. The mean levels of urinary iodine in all the study groups were greater than $9.0 \mu\text{g}/\text{dL}$. As indicated there were weak positive correlations among urinary iodine and goitre rates in all groups except female children (figure 2) having negative correlation but still weak. The correlations were, however, not significant in all the groups.

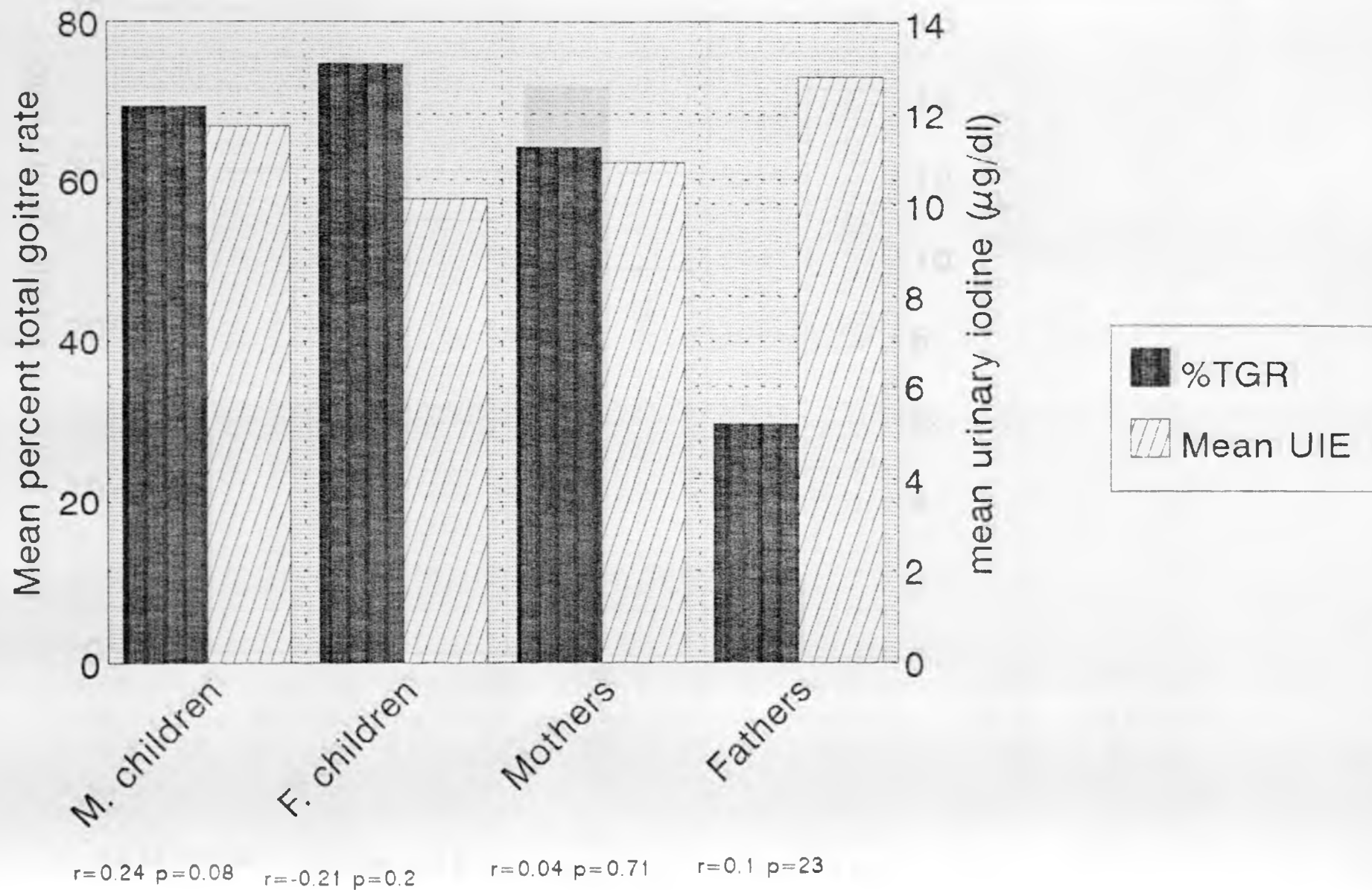


Figure 2 Total goitre rate by sex and level of UIE

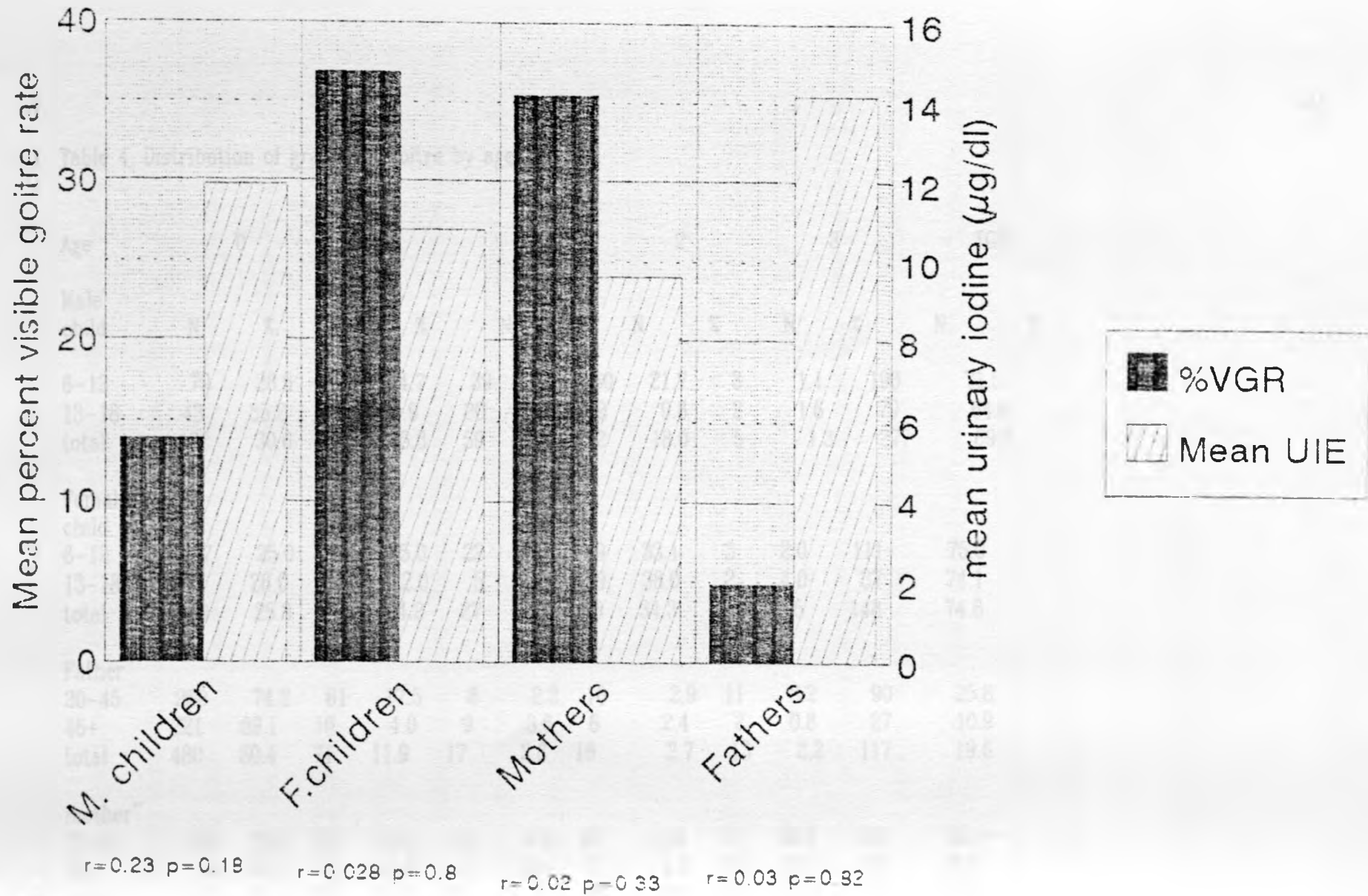


Figure 3 visible goitre by sex and level of UIE

Table 4. Distribution of grades of goitre by age and sex.

Age	0		1A		1B		2		
	N	%	N	%	N	%	N	%	
Male' child									
6-12	79	28.5	96	34.7	39	14.1	60	21.7	3
13-18	43	35.2	45	36.9	20	16.4	12	9.8	2
total	122	30.6	141	35.3	59	14.8	72	18.0	5
Female' child									
6-12	37	25.0	37	25.0	22	14.9	49	33.1	3
13-18	13	26.0	11	22.0	5	10.0	19	38.0	2
total	50	25.3	48	24.2	27	13.6	68	34.3	5
Father''									
20-45	259	74.2	61	17.5	8	2.3	10	2.9	11
46+	221	89.1	10	4.0	9	3.6	6	2.4	2
total	480	80.4	71	11.9	17	2.8	16	2.7	13
mother''									
20-45	190	34.6	121	22.0	38	6.9	87	15.8	113
46+	24	50.0	7	14.6	5	10.4	2	4.2	10
total	214	35.9	128	26.6	43	7.2	89	14.9	123

Significance chi-square test * p < 0.001 ** p < 0.0001

3

TGR

N	%	N	%
1.1	198	71.5	
1.6	79	64.8	
1.3	277	69.4	
2.0	111	75.0	
4.0	37	74.1	
2.5	148	74.8	
3.2	90	25.8	
0.8	27	10.9	
2.2	117	19.6	
20.6	359	65.4	
20.8	24	50.0	
20.6	383	64.2	

Table 4 indicated individual grades of goitre by age and sex. As grades of goitre increase from 1A to 3, prevalence decreased for all age groups, except for female children and mothers. For female children there was an increase of goitre prevalence from 1B to 2, and for mothers from grade 2 to grade 3. Among grades of goitre, the higher prevalence was found in grade 1A than others. Between the sexes, the goitre rate among female children was significantly higher ($p < 0.001$) than in male children. For parents, all grades of goitre were significantly ($p < 0.0001$) higher in mothers than in fathers. This was because all individual grades of goitre were higher in mothers than fathers. Nonvisible grades of goitre (1A and 1B) were higher in male children than female children. However, the rate decreased dramatically in male children from nonvisible to visible and visible goitre rate became higher in female children than male.

4.6 Familial Tendency of Goitre

Cross-tabulation between 597 children and their biological mothers was done to determine the familial tendency of total goitre between them. These results are shown in Table 5. It was found that 313 mothers with goitre had 313 goitrous children, while 102 mothers without goitre had 102 children without goitre. Matched pair analysis indicated that familial tendency of total goitre was strongly associated between mothers and their children. The odds ratio was 4.07 with a 95% confidence limit of 2.76 - 6.02. The Pearson chi-square test, for the correlated proportions, showed that the total goitre rate between children and their biological mothers is associated significantly ($p < 0.0000$).

Table 5. Familial tendency of total goitre between mothers and their children.

	Children (n= 597)		
	without goitre	with goitre	total
without goitre	102	112	214
Mother (n=597)			
with goitre	70	313	383

Statistics

$\chi^2 = 57.71$ $df = 1$ $p < 0.0000$ $CI = 95\%$
 $OR = 4.07$ $2.76 < OR < 6.02$

Visible goitre was also cross-tabulated between mothers and their children. The results are shown in Table 6.

Pearson chi-square test indicated that the familial link of visible goitre between mothers and their children was strongly significant ($P < 0.0000$).

Familial tendency of visible goitre from mothers to their children was stronger ($OR = 13.08$) than total goitre ($OR = 4.07$) for the same groups.

Table 6. Familial tendency of visible goitre between mothers and their children.

	children (n=245)		
	without goitre	with goitre	Total
without goitre	102	26	128
mothers (n=245)			
with goitre	27	90	117

Statistics

$\chi^2 = 78.26$ $df = 1$ $p < 0.0000$ $CI = 95\%$
 $OR = 13.08$ $6.83 < OR < 25.27$

Statistical analysis was also done on the association between fathers and their children (Table 7) in terms of total goitre rates. It was found that there was a strong ($p < 0.0001$) total goitre genetical association between children and their biological fathers. The odds ratio was 3.61 with a 95% confidence limits of 1.49 – 6.82.

Table 7. Familial tendency of total goitre between fathers and their children.

		children (n=597)		
		Without goitre	with goitre	Total
fathers (n=597)	without goitre	158	322	480
	with goitre	14	103	117

Statistics

$\chi^2 = 20.13$ $df = 1$ $p < 0.0001$ $CI = 95\%$
 $OR = 3.61$ $1.94 < OR < 6.82$

The association of visible goitre between children and their fathers is shown in Table 8.

The odds ratio in favor of children having fathers with visible goitre in developing visible goitre was 4.27 times more than those having fathers without visible goitre grade. The 95% confidence limit for the odds ratio was between 1.24 and 16.15. Also pearson chi-square test showed that the association was significant ($p < 0.005$). The odds ratio computed to assess familial tendency of visible goitre between fathers and their children indicated stronger association than odds ratio computed for total goitre rate for the same group of children and their fathers.

Table 8. Familial tendency of visible goitre between fathers and their children.

		Children (n=285)		
		without goitre	with goitre	Total
fathers (n=285)	without goitre	158	111	269
	with goitre	4	12	16

Statistics

$\chi^2 = 6.98$ $df = 1$ $p < 0.005$ $CI = 95\%$
 $OR = 4.27$ $1.24 < OR < 16.15$

Odds ratio was calculated for total goitre association between both parents (in combination) and their children (Table 9). The odds ratio between parents and their children was 4.71 with a 95% confidence limits of 3.14 – 7.05. Pearsons chi-square test also indicated existence of strong ($p < 0.0001$) association of total goitre between parents and their children.

Table 9. Familial tendency of total goitre between both parents and their children.

		children (n=566)		Total
		without goitre	with goitre	
parents (n=566)	without goitre	99	93	192
	with goitre	69	305	374

Statistics

$\chi^2 = 66.65$ $df = 1$ $p < 0.0001$ $CI = 95\%$
 $OR = 4.71$ $3.14 < OR < 7.05$

In order to assess genetic association of visible goitre between children and their biological parents, odds ratio and chi-square test were done (Table 10). The odds ratio was 12.91 with a 95% confidence limits of 6.44 to 26.14. Pearson chi-square test showed that familial tendency of visible goitre between children and their parents had very significant ($p < 0.0001$) association.

Table 10. Familial tendency of visible goitre between parents and their children.

	children (n=220)		
	without goitre	with goitre	Total
without goitre	99	20	119
with goitre	28	73	101

Statistics

$\chi^2 = 68.89$ $df = 1$ $p < 0.0001$ $CI = 95\%$
 $OR = 12.91$ $6.44 < OR < 26.14$

Pedigree study was conducted to investigate familial tendency of visible goitre rate of grandmothers with total goitre rate of mothers of the children (Table 11).

This part of investigation was only done for visible goitres because grand mothers were not clinically examined but the information was obtained by interviewing their daughters who were the mothers of the children. These could only remember what was visible. Statistical analysis (chi-square test) of reported visible goitres indicated strong genetic association ($p < 0.0001$) between grandmothers and mothers of the children. The odds ratio was 2.87 with a 95% confidence interval of 1.52 – 5.50.

Table 11. Familial tendency of reported visible goitre of grandmothers to total goitre rate of the mothers of the children.

		maternal grandmother's have goitre (n=597)	
		No	yes
mothers (n=597)	no	200	14
	yes	319	64

Statistics

$\chi^2 = 12.497$ $df = 1$ $p < 0.0001$ $CI = 95\%$

$OR = 2.87$ $1.52 < OR < 5.50$

Table 12 gives the results of the genetical association between the visible goitres of the biological mothers and the biological maternal grandmothers of the children. Using Pearson chi-square test, strong association ($p < 0.0001$) of familial tendency of visible goitre was noted between mothers and grand mothers of the children. From this, it could be deduced that the association of visible goitre rate was stronger than the association of total goitre between the same groups. When the visible goitre of mothers were matched with the visible goitre rates of the maternal grandmothers of the children, the odds ratio increased to 4.64 with a 95% confidence limits of 2.39 to 9.19.

Table 12. Familial tendency of visible goitres of grandmothers (based on verbal reports) to visible goitres of the mothers of the children.

		maternal Grandmothers have goitre	
		No	Yes
mothers	No	200	14
	yes	160	52

Statistics

$$X^2 = 26.3144 \quad df = 1 \quad p < 0.0001$$

$$OR = 4.64 \quad 2.39 < OR < 9.13$$

4.7 Contribution of Locally Consumed Foods to Goitre Prevalence

More than 98% of the study population depended on subsistence farming. The population cultivated mainly yams, sweet potato, potato, maize, sorghum, teff and halleko (*Moringa stenopetala*). Coffee was occasionally grown as a cash crop.

Cross tabulation between school children's goitre and the most commonly consumed foods (maize, yam, potato, teff, halleko and sorghum) was done (Table 13). It was noted that only halleko (*Moringa stenopetala*) which was consumed in the form of kurkurfa and/or boiled had significant ($p < 0.05$) association with prevalence of goitre. The cut-off point for

frequency of consumption was taken as, consumption frequency, less than or equal to once per day and the other greater than once per day. However, in the cases of halleko consumption cells with expected frequency less than 5 were encountered in statistical analysis. For condition like this, Fisher exact values are applied as recommended. Both one and two tailed Fisher exact values were significant for $p < 0.05$. The Pearson chi-square test for the correlated proportions indicated that the association of goitre prevalence with halleko consumption more than twice per day was statistically significant ($p < 0.05$).

Table 13. Frequency of food consumption against goitre in elementary school children age 6 -18 years.

Type of food	Freq. of consumption	Goitre		OR	χ^2	df	p-value
		no (n)	yes (n)				
Teff	≤ once per day	94	3	2.25 (0.59,10.02)	1.66	1	0.449
	> once per day	209	15				
Potato	≤ once per day	72	64	0.77 (0.05,1.77)	1.70	1	0.193
	> once per day	194	132				
Maize	≤ once per day	14	157	0.64 (0.33,1.22)	2.08	1	0.149
	> once per day	52	371				
Yam	≤ once per day	41	4	2.70 (0.82,9.86)	3.22	1	0.073
	> once per day	91	24				
Sorghum	≤ once per day	24	8	1.32 (0.51,3.54)	0.38	1	0.538
	> once per day	84	37				
Halleko	≤ once per day	10	7	4.57 (1.56,13.67)	10.67	1	0.0011
	> once per day	107	342				

Fisher exact: for halleko consumption 1 tailed p-value= 0.0026862

2 tailed p-value= 0.0026862

n= number of individuals with goitre.

Number in parenthesis is 95% confidence limit for odds ratio.

4.8 Microbial Contamination of Water and its Contribution to Goitre

Drinking water from five sources within the study areas were collected and analyzed for bacterial contamination. Of the five samples, three were found to be contaminated with E.coli which could make them non-potable. Coliform levels of different magnitudes were noted in all the sources of drinking water (Table 14). The highest total goitre rate of 89.7% was however, found in Boarda which had no E.coli in the drinking water, while the least total goitre rate (57.5%) was registered in Morka whose drinking waters had E.coli and highest concentration of coliform count. There was inconsistent dependency between goitre rate and presence of E.coli and/or coliform in water samples from the drinking sources. Water samples from Ganda and Laymatsela were not collected due to logistic difficulties.

Table 14. Bacterial counts of drinking water and goitre rates by study sites.

Study site	water source	Coliform count	E.coli present	Total goitre rate (%TGR)
Mella-Noha	spring ³	17±6	no	82.9
Mishakare	river ³	161±75	yes	78.0
Boarda	spring ²	54±15	no	89.7
Morka	river ²	161±34	yes	57.5
Karza-Dombe	spring ³	161±63	yes	78.0
Ganda ^c	44.1
Lymatsela ^c	86.7

The results are given by number of cells coun per ml of sample.

^c water sample not analyzed.

^{2,3} number of samples collected for analysis.

4.9 Implementation of logistic regression to assess strength of association between dependent and predictor variables.

Logistic regression analyses were used to estimate the likelihood of goitre rate in children which is said to be affected by familial tendency and dietary factors. It is to examine the association in more detailed multi-variate analysis. This analysis provides the opportunity to evaluate the strength of association (seen in bi-variate analysis in the previous section of the results) between particular variables while simultaneously controlling for the confounding effects of others. It is therefore, possible to investigate both genetic and dietary variables described in bi-variate analyses may be functioning as determinants of goitre in children at Gamo-Gofa, Ethiopia.

These variables used as determinants of child goitre rate were chosen because of their influence on child goitre as presented in the earlier cross tabulation results.

Like in the bi-variate analysis the model for goitre in children shows that biological mothers and fathers goitre, and halleko consumption more than once per day significantly affect the odds of goitre of a child. The odds of child having goitre verses child having no goitre increase with more consumption of halleko (more than once per day) and when biological parents have goitre.

Table 15. Logistic regression models to predict the goitre of children coefficient and wald statistics (chi-square) for background variables (n=300).

Beta

Out come	Variables	Beta coefficient	Chi-square	Significance (wald stat.)
children goitre	mother with goitre	1.2264	26.0536	0.0000
	father with goitre	0.8519	6.6863	0.0097
	halleko consumption	0.6552	3.8370	0.0500
	children UIE	1.4083	3.0369	0.0675
	yam consumption	0.3269	0.1760	0.6748

CHAPTER FIVE

5. DISCUSSION

5.1 Prevalence of Goitre

Goitre has been an old age significant public health problem in Ethiopia. Subsequent studies done in Ethiopia have indicated that prevalence of goitre rates have been increasing over time (ICNND, 1959; Hofvander, 1970; Abera et al., 1987/88; Woldie-Gebriel, 1993 and Cherinet, 1995). The study sites chosen were among the highest goitre endemic areas in the country. Almost all the sites had goitre rate greater 50%, except Ganda with 44.1%. This clearly indicated that the study sites were severely affected by iodine deficiency problems. Studies have indicated that goitre rates at this levels were accompanied by other irreversible mental and physical health impairments (Buttfield and Hetzel, 1967). As goitre rates increase above 30%, cretinism may account for 1-10 per cent of the population. This suggests that the number of cretin cases estimated by Wolde-Gabriel in 1993 would be more if the current goitre situation was reassessed in the country.

The prevalence of goitre was found to be a function of place, age and sex. Though our study populations in the two woredas were situated under similar environmental conditions, the prevalence of goitre varied considerably from one school to another. The degree of endemicity ranged from 44.1% in Ganda to 89.7% in Boarda school (Table 14). Goitre prevalence was higher in female children (75.0%) than in male children (71.5%) for age groups of 6-12 years. This high prevalence of goitre in children is classified as severe according to Hetzel (1987). In parents,

male to female ratio for goitre prevalence was 2:5 for the age group 20–45 years and 2:9 in those 46 years of age and above. The higher rate of goitre in female than in male parents could be explained by the higher physiological requirement of iodine by females than males (Braveman *et al.*, 1967). Moving from low age category to the high age category the rate of goitre decreased more dramatically in male than in female parents, while in female children there was an increase in visible goitre rate as we move to higher age groups. This is in-line with the finding of the previous studies (Kelly, 1960; Wolde-Gabriel 1993).

It has also been reported that the major cause for goitre is iodine deficiency (Delange, 1986). Studies from other parts of the world have indicated that goitre endemicity is also associated with goitrogens in food sources (Ermans, 1983 and Osman, 1981), water contamination by different microorganisms (Gaitan, 1973 and Malamas, 1971) and nutrition as protein-calorie deficiency (Gaitan, 1986).

Results of laboratory analysis of urinary iodine revealed that almost all study groups had UIE levels greater than 9 µg/dL (figures 2 and 3) indicating that iodine deficiency was not severe enough to produce the high rates of goitre. Urinary iodine level is an indirect way of evaluating dietary intake of iodine in a given community because, iodine intake is more or less equal to excretion (Hetzel, 1989). A level of 5 µg/dL and above of iodine in urine is considered as indicator for safe range of intake. This level is far below the UIE levels found in this study. It is important to note that in the presence of adequate iodine intake, a

positive though not significant correlation was found between goitre prevalence and urinary iodine excretion (Figures 2 and 3). This strongly suggests that other factors contribute to the high prevalence of goitre in the study area.

5.2 Familial Tendency of Goitre

Familial tendency on endemicity of goitre was assessed by clinical examination of children and their biological parents. The results of familial tendency of goitre between parents and their children indicate that the association was significant between the two groups. Statistical analysis was done between the parents (separately and in combination) and their children. Combination of parents in statistical analysis could be a confounding factor. Therefore, separate and combined analysis was applied. Familial association of goitre between children and their biological parents was therefore found to be strongly significant. In a separate analysis the level of association of visible goitre rate was stronger between mothers and their children ($P=0.0000$ and $OR= 13.08$) than fathers and their children ($p<0.005$ and $OR= 4.27$). Logistic regression analysis was also effected to evaluate the strength association between these variables while controlling for the confounding effect of others. The results indicated that genetical association of goitre between children and their biological parents is strongly significant. Using the logistic analysis (Table 15) it is also confirmed that the association of goitre between children and their mothers is stronger than children and their fathers.

Combination of goitre grades (total goitres) could also be a confounding

factor. Therefore, separate analysis of goitre rates (total goitres and visible goitres) was effected. Odds ratios in all cases for separate analysis were greater than the combined analysis suggesting that the familial tendency of visible goitres were greater than the familial tendency of total goitres. However, in both cases, the association or familial tendency of goitre between parents and their children was found to be statistically significant (Table 5 to Table 10).

Similar statistical analysis was done between grandparents and parents of the children. Goitre association between maternal grandfather, paternal grandfather, and paternal grandmother and parents of the children was not statistically significant. It was only maternal grandmothers and mothers of the children who had familial association of goitre (Tables 11 and 12). Visible goitre rates of maternal grandmothers was cross-tabulated with total goitres and visible goitres of the mothers separately. The results (Table 11 and 12) indicated that there was strong association of goitre between the two groups. The results of the pedigree study mainly depended on the memory of the parents of the children to recall whether grandparents of the children had goitre or not. Hence, this might be the possible explanation for lacking familial association of goitre between aforesaid groups. Studies done in the other part of the world showed a definite association of goitre between parents and their children (Ermans *et al.*, 1980). The authors also found that the difference between goitrous and nongoitrous parents was slight and therefore concluded that it was not possible to explain the disease merely on the bases of monogenic heredity. Other

investigators reported that heredity is one of the contributing factor for the enlargement of the thyroid gland (Benmiloud *et al.*, 1986). This agrees with the findings of this study. Therefore, the null hypothesis "goitre has no familial tendency" is rejected.

5.3 Frequency of Food Consumption and its Relation to Goitre.

The presence of goitrogenic substance in food stuffs has been suspected for many years. In 1928, it was firmly established when development of goitre in rabbits fed on cabbage was demonstrated. Since then, other vegetables of the genus *Brassica* (*cruciferrae* family) have been found to contain goitrogenic properties (Langer and Greer, 1977).

In the present study the influence of naturally occurring dietary factors on the thyroid volume was computed by analyzing the association between dietary consumption (frequency) and goitre prevalence.

All the foods in the study area had no statistically significant influence on the prevalence of goitre except halleko (*Moringa stenopetala*) which was consumed as staple food in the study area. It is cultivated abundantly in the southern regions of Ethiopia. In addition to nutritional value *M. stenopetala* has medicinal and agricultural importance for the communities. Boiled halleko leaves, in the form of soup, locally used for treatment of hypertension, diabetics and diarrhoea. Agriculturally, halleko does not need much labour input for cultivation. It is also used for fencing. The study area has hot and humid climatic conditions and is frequently affected by drought. Due to this reason leafy vegetables were very rare except halleko which is drought resistant and is always

available. Halleko is a tall tree with elongated branches and small green leaves. Stability of greenness of the plant in any kind of drought situation gave an importance to small scale farmers in semi-arid areas of Gamo-Gofa. Accessibility by the hungry during drought and the little labor requirement during growth made the plant important food to the region. It is understood that Ethiopia is frequently affected by drought. Ministry of agriculture is introducing halleko to the northern drought affect regions of Ethiopia. Halleko is consumed by all age and ethnic groups. Kurkurfa is a type of food prepared locally by mixing halleko leaves with maize-barley flour. Some times it is boiled separately and consumed with bread. Only the leaf, flower and young fruit part is consumed.

There are several species of Moringa. *Moringa pterygosperma* grows in East African territories and in Angola and yields a gum known as Moringa gum (CSA, 1994). Investigation shows the gum contains galactose and glycuronic acid. Its oil known as ben oil is applied externally for treatment of goitre and acute rheumatism in the Philippines (Valenzuela P. *et al.*, 1953). The leafs are rich in calcium, iron and phosphorus. Isothiocyanate and hydrocyanic acid were also isolated from this species (Quisumbing E., 1951). Isothiocyanate is the known goitrogenic chemical substance while hydrocyanic acid is toxic substance to human body. Hydrocyanic acid (HCN) from cassava when converted to thiocyanate (SCN) in human body also acts as a goitrogenic substance (Ermans *et al.*, 1980, Ermans *et al.*, 1983, Hetzel, 1987). The other species are *Moringa oliefera* and *Moringa stenopetala* whose seeds were observed to act as

primary coagulants and recommended for domestic water treatment or purification in rural areas of Africa (CSA, 1994).

Gaitan (1980) also indicated that most of the leafy vegetables of the genus *Brassica* including cabbage and turnip, and plants of family *Cruciferae* have naturally occurring goitrogenic substances in them. Goitrogenic substance (cyanogenic glucoside) have also been found in several staple foods such as cassava, maize, bamboo shoots, sweet potatoes, and lima beans around the world. After ingestion the glucosides readily convert to thiocyanate. The goitrogenic action of glucoside is due to the release of thiocyanate from glucoside by the action of enzymes in human body (Ermans *et al.*, 1980).

During statistical analysis by computing chi-square, cells with expected frequency less than 5 were encountered. When the expected frequencies in the table is less than 5, the observed significance level based on the chi-square distribution may not be correct. In condition like this use of fisher exact values is recommended. However, both chi-square and fisher exact values were found to be significant. Consumption of halleko in the form of kurkurfa or boiled more than twice per day was strongly ($p < 0.001$) associated to goitre compared to consumption of less once per day. Odds ratio also indicated that those who consume halleko more than twice per day were 4.57 times likely to have goitre than the other groups. The association of halleko consumption with goitre noted in bi-variate analysis was evaluated using strongest statistical analysis (logistic regression) (Table 15) and found to be significant. Hence, the null

hypothesis "some food sources have no role in goitre causation" is also rejected.

5.4 Drinking Water in Relation to Goitre Causation.

Microorganisms contaminating water supplies have been implicated as causative factors in endemic goitre (Gaitan 1980; Malamos 1971; Vought 1974; Koutras 1980). Bacteriological studies of the drinking waters in villages with high prevalence of goitre showed that the water was more polluted with E.coli and coli-like organisms than waters from nongoitrous localities (Malamos 1971; Koutras 1980). Similarly, Vought (1974) reported observing existence of goitre despite adequate iodine supplementation.

In the present study, three out of five water samples collected from Kucha and Gofa-Zuria woreda, were found to be non-potable and unfit for human consumption due to high bacterial contamination with E.coli and coliform. Among the water sample groups, all have high prevalence of goitre; the least 57.5% in Morka. However, inconsistency between goitre rate and bacterial load was noted in this study.

E.coli contamination was found in water sample from Morka, in-addition to the highest concentration of coliform count. However, the rate of goitre was least when compared to Boarda and Mella-Noha having highest prevalence of goitre with no contaminations of E.coli and minimum contamination of coliform in their drinking waters. Results indicate that bacterial contamination of water had no relationship with goitre size. Neither was a relationship seen with the source of drinking water from

spring or river. This is contrary to epidemiological studies done in other parts of the world, which point to a relationship between drinking water and goitre prevalence (Gaitan 1973,1980; Malamos 1971), that pollution of drinking water with microorganisms like E.coli and coliform are contributory factors to goitre endemia.

Even though, consistency was lacking between goitre rate and bacterial contamination of drinking water, the magnitude of goitre was still high in those sites having contaminated waters and also the presence of E.coli in drinking waters is not accepted at any concentration level.

Comparative statistical analysis was done to assess goitre relation between groups treating drinking water before consumption and those who do not treat. Similarly, the rate of goitre prevalence was computed between those having toilet and those having no toilets. The assumption was that presence of toilet minimizes water contamination by microorganisms, which are suspected to aggravate goitre prevalence and although treating (boiling, chlorination) may annul the existence of bacteria in drinking water and make it fit for consumption. But, neither the presence or absence of toilet nor treating drinking water before consumption had relation with goitre prevalence in this study.

CHAPTER SIX

6. CONCLUSION AND RECOMMENDATION

6.1 Conclusion

This study shows that Gamo-Gofa was one of the highest goitre endemic areas in the country. The results indicate that goitre prevalence is at levels that may be accompanied by other IDD problems like cretinism.

The causation of goitre is characterized as being multiple in nature.

Different factors could contribute to the variation in goitre prevalence among close communities with similar iodine environment.

Familial tendency was found to be one of the factor contributing to the development of goitre. Genetic association of goitre was noted between children and their biological parents. Strongest association was found between mothers and their children than fathers and the children though, the association was statistically significant in both cases.

Diet also contributes to the endemicity of goitre. Consumption of halleko (*Moringa stenopetala*) a leafy vegetable plant, had an association with the prevalence of goitre. It grows in arid and semi-arid areas of Gamo-Gofa. Those who consume halleko more than once per day had higher prevalence of goitre than those who consume less frequently or not at all. Hence, it is understood that diet is one of the etiological factors for goitre prevalence.

In this study both type of source of water (river, spring and well) and microbial load have no association with goitre prevalence.

The endemicity of goitre is highly dependent on sex. In children as well as parents goitre rates are higher in females than in males. Goitre prevalence was also found to be a function of age. For both parents and male children, the prevalence of goitre is higher in low age categories than high.

6.2 Recommendation

The existing evidences and the results of this study suggest that goitre is a public health problem in these study areas. Based on the results of this study it is recommended the following action to be taken.

1. Appropriate steps should be taken to effect the IDD control programme (sea salt or rock salt iodation) without lapse to alleviate this terrifying nature of IDD problem in the country. It is also important to consider the distribution of oral iodized oil which is used as an immediate and fast means alleviation of severe IDD problems. The collaboration of different organizations (NGO's, UN and government) is important to materialize the programme.

2. Further studies are needed on halleko plant to investigate which chemical constituent is responsible in goitre causation, how and means of alleviation.

3. There is need for more systematic and extensive study on familial tendency of goitre and the reason why it is more strongly associated between mother and child than father and child.

4. Further studies should be carried out to determine the real contribution of water sources with faecal material to goitre. Most of the drinking water sources had faecal contamination which is unfit for consumption hence, it is also advisable to take an appropriate action.

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Appendix 1

HOUSEHOLD QUESTIONNAIRE

SOUTH ETHIOPIA REGION - GAMO-GOFA

Zone.....Village/Kebele

Name of school.....Name of student.....

Name of interviewer.....

DEMOGRAPHY

- | | | | |
|--|--|--|--|
| 1. Child identification number _____. | | | |
| 2. Name of the head of the household..... | | | |
| 3. Occupation of the head of the household..... | | | |
| 4. Ethnic group..... | | | |
| 5. Length of stay in the area in years..... | | | |
| 6. Religion (protestant = 1; Orthodox = 2; Catholic = 3;
moslem = 4; Other = 5) | | | |

7. DRINKING WATER SOURCE

- 1= river 2= Unprotected spring
 3= protected spring 4= Pipe water treated
 5= pipe water not treated 6= Well treated
 7= Well not treated

8= Do you treat your drinking water? 1= yes 2= no

9= If yes how? 1= Boiling 2= chlorination

3= Filtration 4= Other (specify)

10= Do you have latrine? 1= yes 2= no

11= What type of latrine 1= private 2= common

12= How far is the latrine from drinking water source?.....km

13: CLINICAL EXAMINATION DATA

a b c d e f g

NAME	SEX	AGE	RHH	PHY CON.	GG	NODUL	HEALTH PROBLEM ASS. WITH GOITRE

a= sex: Male= 1 Female= 2

b= age in years

c= Relation to household (RHH)

1= Father 3= son

2= Mother 4= Daughter

d= physiological condition (phy.con)

1= pregnant

2= lactating

3= non pregnant

non lactating

e= Goitre grade: 0= 0;

1a=1

1b=4; 2= 2 3= 3

f= Nodularity: 1=1, 3=3

2=2, 4=4

g= Health problems

1= swallowing 2= breathing

3= sound/voice 4= other

14. Paternal /Father's line /pedigree studies

a= Grandfather had thyroid swelling 1= yes 2= no

b= Grandmother had thyroid swelling 1= yes 2= no

15. Maternal /Mother's line /pedigree studies

a= Grandfather had thyroid swelling 1= yes 2= no

b= Grandmother had thyroid swelling 1= yes 2= no

16: LABORATORY ANALYSIS

a= Date of analysis.....

b= Iodine level 1= father..... µg/dl

2= mother.....

3= son.....

4= daughter.....

15. U = የአናቱ አባት አገቅርት 1 = አለባቸው 2 = የአባቸውም
 A = የአናቱ አናት አገቅርት 1 = አለባቸው 2 = የአባቸውም

16. የሲቦሪቲ ምርመራ

የአዎኒን መጠን በሸገት

1 = አባት _____ 2 = አናት _____

3 = ወገድ ለጅ _____ 4 = ሴት ለጅ _____

17. የምዝገባ ዓይነትና የአድጋገብ ደገፍቶሽ

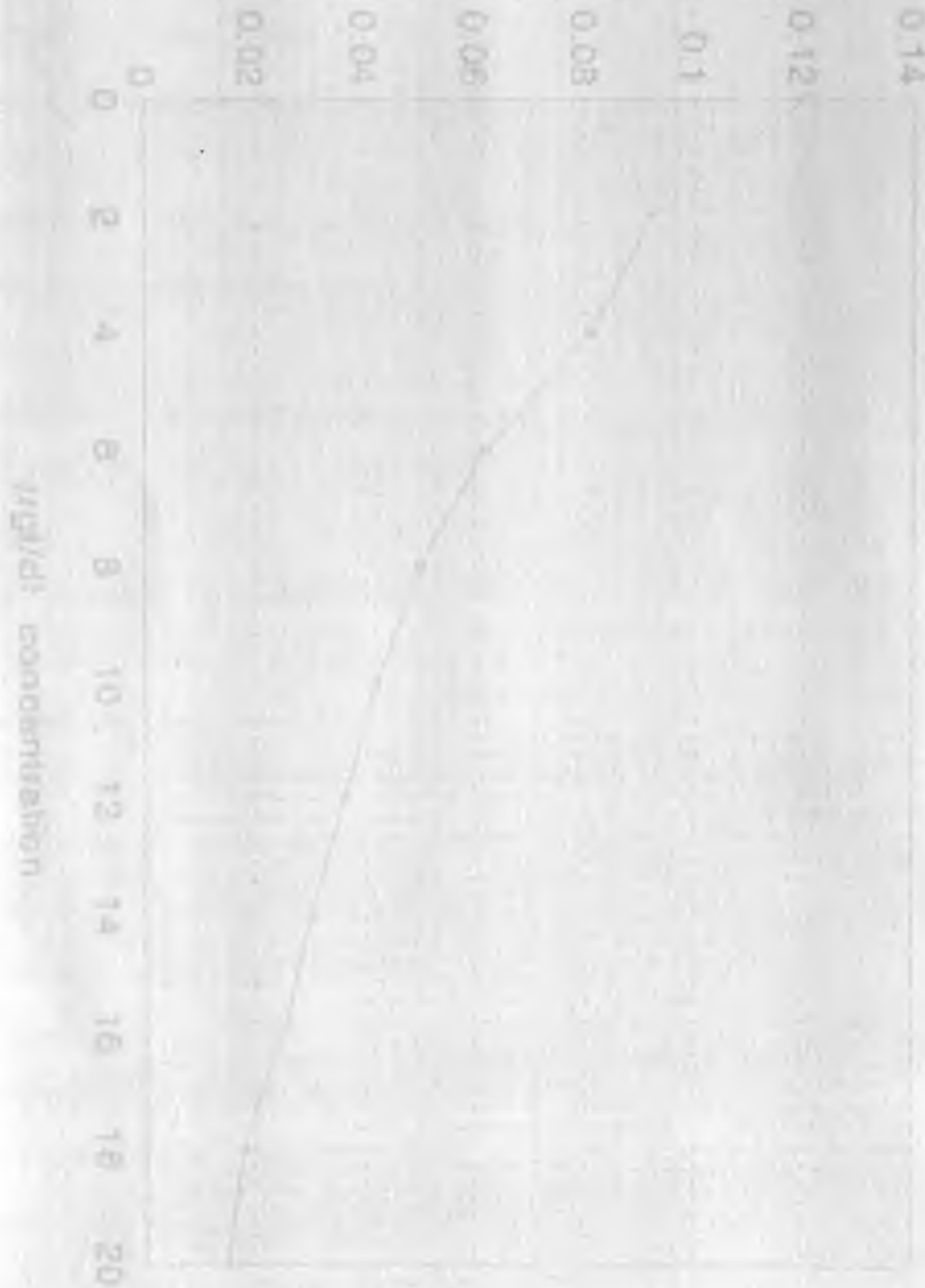
የምዝገባ ዓይነት	በቀን				በሰዎች						1 ጊዜ			2 ጊዜ				
	1	2	3	4	1	2	3	4	5	6	2ባ	3ባ	3ባ	10	20	30	40	

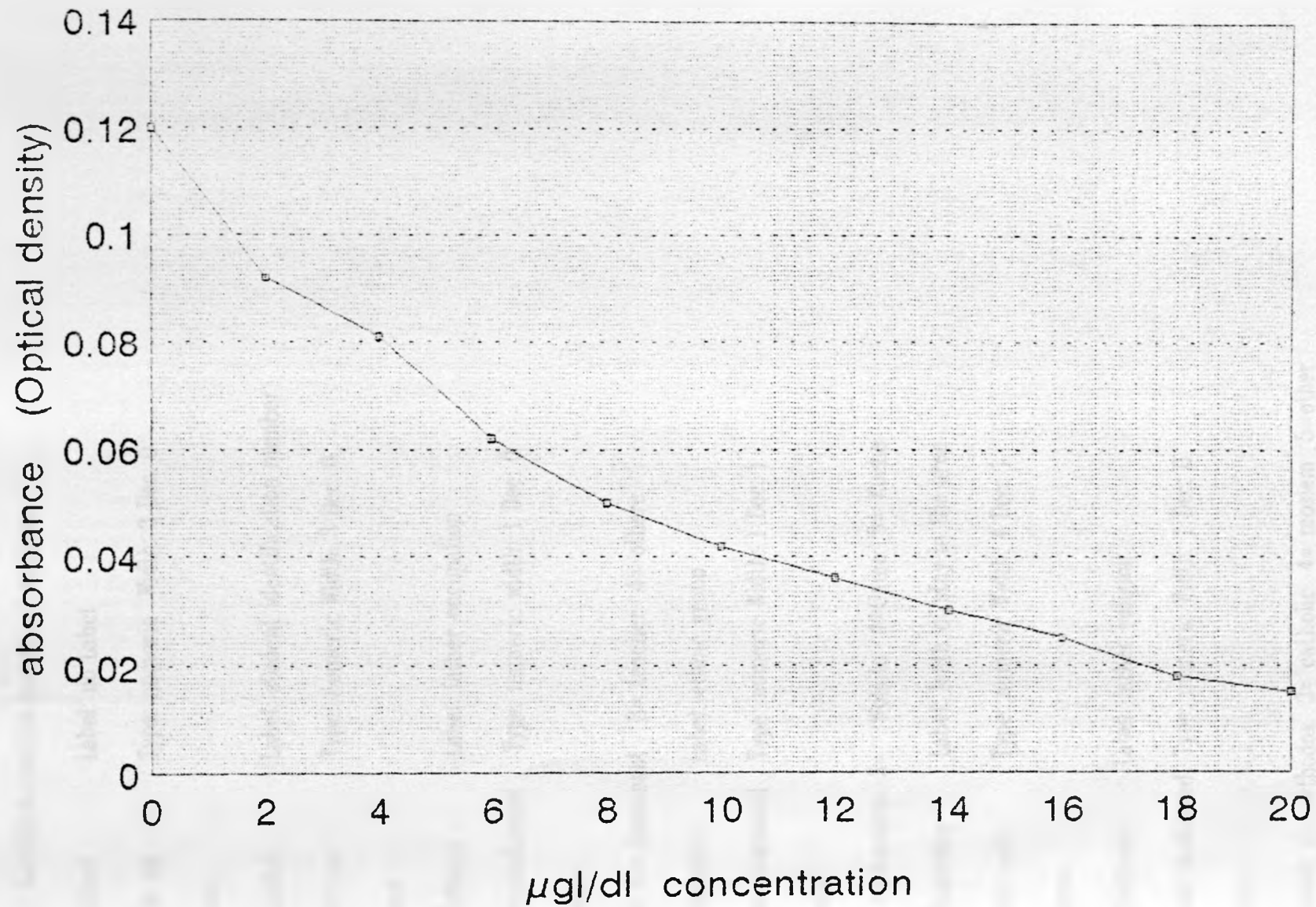
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Appendix 3 Preparation of Solutions.

1. Chloric acid solution: In 2000 ml Erlenmeyer flask containing 910 ml of deionized water, 500 g of KClO_3 was dissolved by heating until it goes into solution. Then 375 ml HClO_4 (perchloric acid, 70%) was added with constant stirring. Stored in a freezer over night and next day filtered with filter paper (Whatman #1).
2. 5N H_2SO_4 : To about 700 ml of deionized water 139 ml of concentrated H_2SO_4 was added and when cool, the volume was adjusted to 1 litre with deionized water.
3. Arsenious acid solution: In a 2000 ml Erlenmeyer flask 20 g As_2O_3 and 50 g NaCl were added, then 400 ml of 5N H_2SO_4 was added slowly. The volume was made to 1 litre and heated to dissolve. Finally after cooling to the room temperature it was diluted to 2 litre and stored in a dark bottle in a room temperature away from light.
4. Ceric ammonium sulphate solution: In a 1000 ml of volumetric flask 48 g of ceric ammonium sulphate was dissolved with 3.5 N H_2SO_4 (3.5 N H_2SO_4 was made by slowly adding 97 ml concentrated H_2SO_4 to about 800 ml deionized water, when cool, it was adjusted to a final volume of 1 litre with deionized water). The bottle was stored in a dark away from light at room temperature.
5. Standard iodine solution, 1 μg iodine per ml: In 100 ml of volumetric flask 0.168 mg of KIO_3 was dissolved. Then kept in a dark bottle. For

Standard curve preparation appropriate dilutions of $1 \mu\text{g}$ per ml solution of KIO_3 was made.





Appendix IV Standard curve for determination of urinary iodine

Appendix 5 Variables used in statistical analysis.

Variable: school Label: no Label

Value labels: no Type: numeric Width: 3 Dec: 0

Missing: none

Variable: studid Label: student identification number

Value labels: no Type: numeric Width: 3 Dec: 0

Missing: none

Variable: fathocc Label: father occupation

Value labels indicated Type: numeric width: 1 Dec: 0

Missing: none

1= Farmer 2= merchant 3= teacher 4= others

Variable: fathethn Label: ethnic group

Value labels indicated Type: numeric Width: 1 Dec:)

Missing: none

1= Gofa 2= Amhara 3= Wolaita 4=Gamo 5= Kucha

Variable: housstay Label: lenth of stay in the area

Value Labels: no Type: numeric Width: 4 Dec: 1

missing: none

Variable: fathrelg Label: father religion

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= protestant 2= orthodox 3= catholic 4= moslem 5=other

Variable: watsorc Label: drinking water source

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= river 2= unprotected spring 3= protected spring 4= pipe water treated 5= pipe water not treated 6= well treated
7= well not treated.

Variable: waterprep Label: water preparation for drinking

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= yes 2= no

Variable: howprep Label: how do you prepare

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= boiling 2= chlorination 3= filtration 4= others

Variable: havetoil Label: have toilet

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= yes 2= no

Variable: howuse Label: what type of toilet

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= private 2= common

Variable: disttoil Label: distance toilet from water

Value Labels: no Type: numeric Width: 3 Dec: 1

Missing: none

Variable: sexstudent Label: no

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= male 2= female

Variable: agestudent Label: no

Value Labels: no Type: numeric Width: 4 Dec: 1

Missing: none

Variable: physstud Label: physiological condition student

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= pregnant 2= lactating 3= non pregnant non lactating

Variable: ggstud Label: goitre grade student

Value Labels: indicated Type: numeric Width: 1 Dec:0

missing: none

0= 0 1a= 1 2= 2 3= 3 1b= 4

Variable: healthstud Label: health problems of student

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= swallowing 2= breathing 3= sound/voice 4= heart problem

Variable: sexmother Label: no

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

2= female

Variable: agemoth Label: no

Value Labels: no Type: numeric Width: 4 Dec: 1

Missing: none

Variable: physmoth Label: physiological condition mother

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= pergnant 2= lactating 3= non pergnant non lactating

Variable: ggmoth Label: goitre grade mother

Value Labels: indicated Type: numeric Width: 1 Dec:0

missing: none

0= 0 1a= 1 2= 2 3= 3 1b= 4

Variable: healthmoth Label: health problems of mother

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= swallowing 2= breathing 3= sound/voice 4= heart problem

Variable: sexfather Label: no

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= female

Variable: agefather Label: no

Value Labels: no Type: numeric Width: 4 Dec: 1

Missing: none

Variable: ggfath Label: goitre grade father

Value Labels: indicated Type: numeric Width: 1 Dec:0

missing: none

0= 0 1a= 1 2= 2 3= 3 1b= 4

Variable: healthfath Label: health problems of father

Value Labels: indicated Type: numeric Width: 1 Dec: 0

Missing: none

1= swallowing 2= breathing 3= sound/voice 4= heart problem

Variable: gpgf Label: goitre paternal grand father

Value Labels: indicated Type: numeric Width: 1 Dec: 0

missing: none

1= yes 2= no

Variable: gpgm Label: goitre paternal grand mother

Value Labels: indicated Type: numeric Width: 1 Dec: 0

missing: none

1= yes 2= no

Variable: gmgf Label: goitre maternal grand father

Value Labels: indicated Type: numeric Width: 1 Dec: 0

missing: none

1= yes 2= no

Variable: gmgm Label: goitre maternal grand mother

Value Labels: indicated Type: numeric Width: 1 Dec: 0

missing: none

1= yes 2= no

Variable: UIES Label: student urinary iodine

Value Labels: no Type: numeric Width 4 Dec: 1

Missing: none Type: numeric Width 4 Dec: 1

Variable: UIEM Label: mother urinary iodine

Value Labels: no Type: numeric Width 4 Dec: 1

Missing: none Type: numeric Width 4 Dec: 1

Variable: UIEF Label: father urinary iodine

Value Labels: no Type: numeric Width 4 Dec: 1

Missing: none Type: numeric Width 4 Dec: 1

Variable: halcons Label: halleko consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: maizecons Label: maize consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: potatcons Label: potato consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: millcons Label: millet consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: pumpcons Label: pumpkin consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: sorgcons Label: sorghum consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: milkcons Label: milk consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Variable: cabcons Label: cabbage consumption per day

Value Labels: no Type: numeric Width: 5 Dec: 2

missing: none

Appendix 6 Distribution of goitre rate by religion.

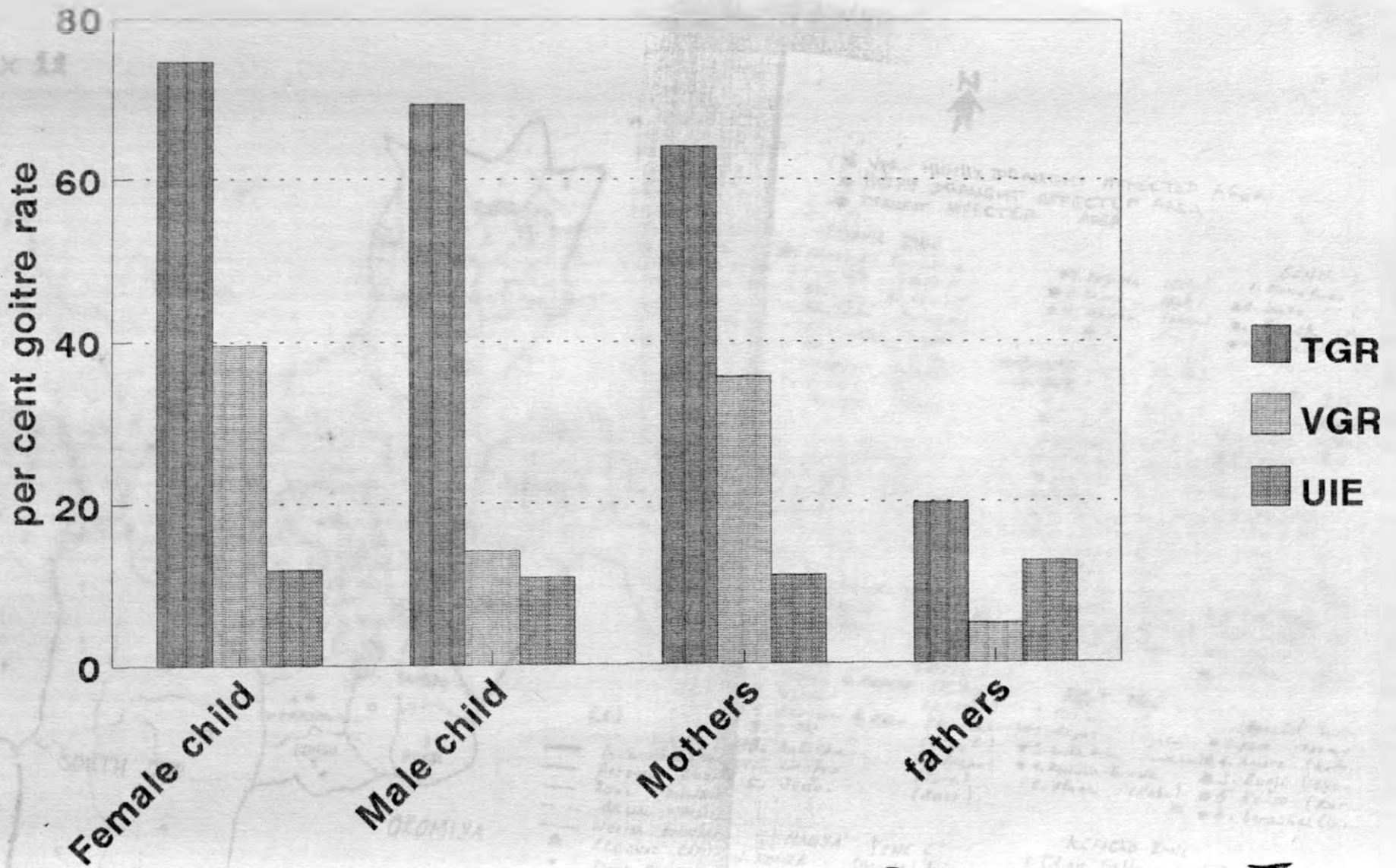
Religion	Children		Mothers		Fathers	
	N	%	N	%	N	%
Protestant	276	64.9	248	64.8	80	68.4
Orthodox	136	32.0	122	31.9	34	29.1
Catholic	2	0.47	2	0.5	-	-
Moslem	6	1.4	5	1.3	1	0.9
No religion	5	1.2	6	1.7	2	1.7

Appendix 7 Distribution of goitre rate by ethnicity.

Ethnicity	Children		Mothers		Fathers	
	N	%	N	%	N	%
Gofa	324	76.2	312	81.5	101	86.3
Amhara	11	2.6	8	2.1	5	4.3
Wolyta	43	10.1	26	6.8	5	4.3
Gamo	43	10.1	32	8.4	6	5.1
Kucha	4	0.9	5	1.3	-	-

Appendix 8 Distribution of goitre rate by occupation.

	Children		Mothers		Fathers	
	N	%	N	%	N	%
Farmers	412	96.9	373	97.4	111	94.9
Merchants	1	0.2	2	0.5	1	0.9
Teachers	10	2.4	7	1.8	4	3.4
Others	2	0.5	1	0.3	1	0.9



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Appendix 9 Total and visible goitre rate in relation with UIE