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FOOD CONSUMPTION AND HEMATOLOGICAL STATUS
OF PREGNANT WOMEN SELECTED FROM TWO PRENATAL CLINICS
IN SHIRAZ, IRAN

By
Atossa Rahmanifar

A DISSERTATION

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ABSTRACT

FOOD CONSUMPTION AND HEMATOLOGICAL STATUS OF
PREGNANT WOMEN SELECTED FROM TWO PRENATAL
CLINICS IN SHIRAZ, IRAN

By
Atossa Rahmanifar

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Four hundred pregnant subjects were selected from Helal and Hafez prenatal clinics in Shiraz, Iran and their dietary intake and hematological status were assessed. Subjects were Shiraz residents, were beyond the 12th week of pregnancy and had not taken iron supplements during the previous year. Socioeconomic characteristics, medical history and household consumption of major food items were obtained by interviews. Blood samples were obtained for assessment of hematological status. Subjects were classified according to household monthly income: very low (VL), low (LO), middle low (ML) and middle and high (M-H).

Individual intake of major dietary iron sources was determined by dividing household consumption by adjusted household size which considered caloric needs of persons within the household. Iron intake was calculated using standard Iranian and U.S. food composition tables.

Average total intake of iron from major food sources was similar between groups; however, the ratio of heme iron to nonheme iron was greater in higher income groups. Bread

Atossa Rahmanifar

was the major source of dietary iron for all groups. Regression analysis showed an inverse relationship between bread consumption and household income. Intake of meat, poultry, greens and herbs and lettuce and heme iron increased as household income increased and size decreased.

Hemoglobin and hematocrit increased as household income increased but the relationship was not linear. Stage of pregnancy had a significant inverse and nonlinear relationship with hemoglobin, hematocrit, serum iron and transferrin saturation. Intake of heme iron increased with increased income but was not related to blood constituents analyzed.

With transferrin saturation less than 15.0% the criterion for anemia, 9.3% of VL, 7.4% of L0, and only 2.0% of ML and M-H income groups were at risk for anemia. Of 17 anemic subjects, 4 were in week 21-30. Thirteen were in week 31-40 of pregnancy and were from VL and L0 groups. Although stage of pregnancy was the most important indicator of hematological status, the higher incidence of anemia in lower income subjects may be due in part to lower availability of dietary iron.

To those Iranian mothers and their progeny
who are in need of adequate medical care.

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INTRODUCTION

Iron deficiency anemia, the most common nutritional anemia worldwide, is a frequent complication of pregnancy (1-3). In many populations, especially in developing countries, the pregnant woman is susceptible to iron deficiency because her demand for iron is increased and usually is not met by her iron stores or by an increase in dietary intake (1).

Geographical differences in iron status may not correlate with the total intake of iron from food (3). Iron status is greatly influenced by the availability of iron in the diet and by the inclusion of dietary factors which enhance or inhibit iron absorption.

The prevalence of iron deficiency anemia and the determination of factors influencing the development of this nutritional anemia have not been reported for pregnant women in central Iran. Thus, it was of interest to assess food consumption, iron intake and hematological status of a population of pregnant women representative of urban areas in central Iran. Shiraz with a population of approximately half a million persons was chosen as the research site.

The objectives of this study were:

- To assess the food and iron intake and hematological status of low, middle and high income pregnant women attending selected prenatal clinics in Shiraz, and
- To determine the effect of household income on dietary factors influencing hematological status.

REVIEW OF LITERATURE

Nutrition plays an important role in the course and outcome of pregnancy because of an increased maternal nutrient requirement. The extent of the increase, however, varies considerably from nutrient to nutrient. Pregnancy imposes a substantial burden on the maternal hematopoietic system because of the need for augmented erythropoiesis for which the two most critical nutrients are iron and folic acid (1).

Nutritional anemia

Nutritional anemia is a world wide problem. Its prevalence is highest in developing countries where it occurs in all age groups, especially in pregnant women and young children (1-3). Although many nutrients are involved in the production of red blood cells and hemoglobin, iron deficiency is by far the most common cause of nutritional anemia throughout the world (2). In some countries, folate deficiency is also an important cause of anemia (2) in certain sections of the population, especially in pregnant women. In some individuals, vitamin B₁₂ deficiency may produce severe anemia, as in classical pernicious anemia; however, this is relatively rare even in countries such as India with a predominantly vegetarian diet. There is little

evidence that vitamin B₁₂ deficiency is a significant cause of anemia worldwide (2).

Iron deficiency

Iron deficiency is most likely to occur when iron requirements are greatest, i.e. during infancy, childhood, the reproductive period in women, and pregnancy (1). In developing countries, apart from increased requirements, the commonest causes of iron deficiency are poor availability of dietary iron and increased iron losses due to parasitic infestation, especially hookworm (1, 3).

Three stages of iron deficiency have been defined (1). The first stage is characterized by decreased iron stores without any other detectable abnormalities. The size of iron stores has been assessed in the past through a variety of invasive techniques, such as histological examination of bone marrow smears for the presence of stainable iron (4). More recently, however, serum ferritin which reflects the amount of storage iron is measured using immunological methods (5).

The second stage of iron deficiency begins after iron stores are exhausted (1). With further iron depletion there is a diminution of hemoglobin synthesis and the percentage saturation of transferrin falls from a normal value of about 30% to less than 15% (6).

The third stage is overt iron deficiency and is reached when the impaired hemoglobin synthesis results in a

measurable decrease in concentration of circulating hemoglobin. Initially, the red blood cells are normocytic and normochromic but ultimately they exhibit the classical morphological changes associated with iron deficiency, i.e. microcytic and hypochromic red blood cells (1).

Assessment of iron status

Nutritional anemia has been defined as "a condition in which the hemoglobin concentration is below the level that is normal, for a given individual, due to deficiency of one or more of the nutrients required for hemopoiesis, and, conversely as a condition in which the hemoglobin concentration can be raised by increasing the amount of nutrient(s) absorbed" (7). However, problems arise when an attempt is made to define a "normal" hemoglobin concentration. A World Health Organization Scientific Group has suggested arbitrary cut-off points for hemoglobin concentration below which anemia is likely to be present in individuals (3). This cut-off point for adult pregnant women is suggested at hemoglobin concentration of 11.0 g/dl of whole blood (3). There will be a small proportion of normal subjects with hemoglobin concentration below this cut-off point who will not show a rise in hemoglobin concentration when supplied with extra hemopoietic nutrients, but the precise percentage in the different groups has not been clearly defined (2). Nevertheless, these arbitrary

values serve as useful guidelines. The greater the percentage of the population with hemoglobin concentration below the cut-off point, the greater is the incidence of anemia in the community (2). As judged by hemoglobin concentrations, the highest prevalence of anemia was found among a population of pregnant women studied in India (3) and in Burma (8) where 80 percent of them had values less than 11 g/100 ml.

A response to iron therapy by an increase in hemoglobin level would be expected in patients with iron deficiency anemia. Correlation of this response with percent transferrin saturation can be a diagnostic test for the iron status of subjects (9). The percent transferrin saturation which is expressed as Serum Iron/Total Iron Binding Capacity (SFe/TIBC) has been shown to accurately assess iron deficiency in non-pregnant patients (6). In iron deficiency, this is depressed, i.e. below 16% with an elevation in the total iron binding capacity (6). An appropriate therapeutic response to iron is marked by an elevation of percent transferrin saturation (SFe/TIBC) to levels of 25 to 35 percent and decline of the TIBC. But, since TIBC also rises in pregnancy (10), it has been questioned whether this test can be employed reliably to assess iron deficiency in pregnancy because the elevated TIBC might affect the interpretation of reduced percent transferrin saturation (SFe/TIBC) during pregnancy. In a group of pregnant anemic patients, the effect of therapeutic doses of oral and parenteral iron on

percent transferrin saturation was studied (9). It was shown that pregnant anemic patients who have an SFe/TIBC under 20% respond to iron therapy, with both an increase in hemoglobin and in SFe/TIBC to above 20%. In contrast, those with SFe/TIBC ratios above 20% failed to increase their hemoglobin after appropriate iron therapy. It was concluded that in pregnancy, SFe/TIBC values below 20% tend to indicate iron deficiency while SFe/TIBC values above 20% tend to indicate iron sufficiency (9). The prevalence of iron deficiency found in collaborative WHO studies in women in the third trimester of pregnancy in different population groups, as judged by the percentage of women having a saturation of transferrin (SFe/TIBC) below 15%, showed a range of 40% to 99% of the population with iron deficiency (7). The prevalence of anemia on the basis of hemoglobin concentration was 22% in Polish women while 40% showed evidence of iron deficiency according to SFe/TIBC values (7). Depressed SFe/TIBC levels can also be seen in other conditions such as chronic inflammatory disease, chronic infection, and malignancy (11). In these cases, however, the TIBC is usually depressed to levels below 300 $\mu\text{g/dL}$ (11). Therefore, one should be cautious in interpreting depressed SFe/TIBC when the TIBC is below 350 $\mu\text{g/dL}$.

Attention to methodology and standardization of measurements are essential for meaningful comparisons between results of surveys in different parts of the world. All

hematological tests should be done on venous blood samples and standard techniques described by Dacie and Lewis (12) have been recommended (7). Determination of hemoglobin concentration by cyanomethemoglobin method and estimation of packed cell volume by microhematocrit method are well standardized methods (13) which have been used in WHO studies (7, 14). Estimation of serum iron by the method of Ramsay (15) or a modification of the method of Bothwell and Merrett (16), and estimation of total iron binding capacity by the method of Ramsay (17) or Herbert et al. (18) have been the methods of choice in WHO and some other studies (7, 14, 19, 20). Serum iron and total iron binding capacity can also be measured by the method of Olson and Hamlin (21) using atomic absorption spectrophotometry. The method is highly specific, accurate and requires less time than most of the colorimetric or atomic absorption methods. Recently, immunoreactive methods have been developed for the measurement of serum ferritin as an estimate of storage iron (5, 22-25).

Iron deficiency anemia during pregnancy

In many populations, the pregnant woman is prone to iron deficiency because her high demand for iron is usually not met by an increase in dietary intake or her own iron stores (26).

Iron balance during pregnancy: In pregnancy, iron loss through menstruation is spared but iron is needed to meet

the basal losses which are similar to those in the nonpregnant state and is about 200 mg for the whole period of pregnancy (27). In addition, the increase in maternal red blood cell mass (about 500 mg), and iron need by fetus (about 290 mg) and placenta (about 25 mg) bring the total requirement to about 1,000 mg for the whole period of pregnancy (27). Except for the daily basal losses, these requirements are concentrated in the second half of pregnancy (27). If iron stores are depleted at the start of pregnancy, the amount of iron which should be absorbed during the second half of pregnancy would have to be about 6.0 mg per day, an absorption which can only be achieved through iron supplements (10, 27). Since most of the women even in developed countries have low or absent iron stores (10, 11), routine administration of iron supplements (30 to 60 mg iron per day) is recommended to all pregnant women (3, 28).

Even in the absence of any deficiency or disease that may decrease erythropoiesis, the increase in plasma volume during pregnancy is relatively greater than the volume of red blood cells added to the circulation, and this led to the concept of "physiological anemia" of pregnancy caused by hemodilution (29, 30). Therefore, a hemoglobin level in the range of 10.5 to 11 g/dl may not necessarily indicate impaired erythropoiesis but may instead be the result of marked increase in blood volume (10). As a result of hemodilution, hemoglobin and hematocrit levels decrease at three

to five months gestation. They then rise slightly at term, and return to normal at six weeks post partum (10).

Anemia and fetal outcome: Severe anemia during pregnancy has been shown to be associated with an increased risk of premature delivery and increased maternal and fetal morbidity and mortality (31-33). Deleterious effects of milder degrees of anemia during pregnancy are less well documented, but premature delivery has been shown to be more common even in mildly anemic mothers (34). A correlation between maternal hemoglobin level and fetal birth weight has also been demonstrated (20) i.e. mothers with lower hemoglobin levels tended to have children of lower birth weight.

Various investigators have not been able to demonstrate a higher prevalence of anemia during the first and second year of life in infants born of iron deficient mothers (10, 19, 35, 36). The main factors affecting the body iron stores of the infant at birth appear to be the variation in birthweight, the cord hemoglobin concentration, and the degree of placental transfer of iron (37).

Iron supplementation: The increased iron requirement during pregnancy associated with inadequate or absent iron stores will inevitably lead to further depletion of maternal stores and without intake of iron supplements, result in iron deficiency anemia as pregnancy progresses (29). Iron supplements are prescribed to restore the circulating hemoglobin mass and to replenish depleted iron storage pools.

Results of a number of longitudinal studies on hemoglobin concentration, hematocrit and erythrocyte count in different gestational stages with and without iron supplements are summarized by Pitkin (38). Hemoglobin concentration, hematocrit, and erythrocyte count declined progressively, particularly in unsupplemented subjects in which mean values at mid-third trimester were in the range of 10.5 to 11.0 g/dl for hemoglobin, 32 to 34 percent for hematocrit, and 3.7 to 4.1 million/mm³ for erythrocyte count. With iron supplementation, the mean values during the mid-third trimester were about 12 g/dl for hemoglobin, 36 percent for hematocrit, and 4 million/mm³ for erythrocyte count. The effect of iron supplementation on hemoglobin concentration has been reviewed by Prichard (29). The mean hemoglobin concentration at or near term in five studies reviewed was 12.3 g/dl with iron supplements and 11.1 g/dl without.

Iron supplements for pregnant women are widely prescribed but different combinations have been used and different doses have been recommended. Ferrous iron salts which are the more available form for absorption are the choice for oral iron supplementation (39). Since iron is absorbed in the proximal small intestine, the use of enteric coated or delayed release preparations is not recommended (39). Ferrous sulfate is the least expensive and is usually administered as 300 mg tablets of the hydrated salt of ferrous sulfate which provides 60 mg of iron. Ferrous

gluconate tablets which are also frequently prescribed are equally well absorbed, but the dose must be doubled since each tablet of 300 mg contains only 36 mg of elemental iron which will increase both the cost and the inconvenience of treatment (39).

The WHO (1972) recommendation is that 30 to 60 mg iron per day should be given to pregnant women in populations with low incidence of iron deficiency and 120 mg to 240 mg per day in populations with high incidence of iron deficiency (14). In two WHO collaborative studies in Dehli and Vellore, India, supplements were given for 10-12 weeks, starting at about 26th week of pregnancy (19). The control group received a placebo and showed a progressive fall in hemoglobin level. The women who received vitamin B₁₂ and folic acid alone (without iron) showed the same response as those receiving the placebo. In most of the women receiving iron in a single daily dose of 30, 60, 120 or 240 mg of elemental iron (as ferrous fumarate) there was a significant increase in the hemoglobin concentration and the best results were in the group receiving 120 and 240 mg of iron. However, even in these groups there was still a high prevalence of anemia and iron deficiency at the end of treatment period. Since time of administration of tablets with respect to food intake was not controlled, it was suggested that iron absorption might have been depressed by some inhibitory factors present in their vegetarian diet (19).

The availability of dietary iron

Although iron deficiency is global in its occurrence, the prevalence varies substantially in different geographical areas of the world (3, 27). Some of these population differences might be due to pathological iron losses caused by hookworm infestation in certain geographical areas. However, the most important etiological factor appears to be an inadequate intake or assimilation of dietary iron. Since the geographical differences in iron status do not correlate with the total intake of iron from food (3, 40, 41), the most important determinant of iron status may be the availability of iron in different diets. Therefore, the amount of iron potentially available from foods depends not only upon the amount of iron supplied but the nature of that iron and the composition of the meal in which it is consumed (42, 43). The total iron content of the diet is thus a relatively poor indicator of the adequacy of the diet with regards to iron.

With respect to its availability, food iron can be divided into two main parts: heme and nonheme iron, each of them forming a different pool of iron (42, 44). Heme iron which comes from hemoglobin and myoglobin i.e. from animal tissue, is absorbed directly as the intact iron porphyrin complex, and its iron is freed in the intestinal mucosal cell (44, 45). Iron from other foods such as vegetables, fruits, cereals, eggs, dairy products as well as nonheme

iron of meats, poultry and fish and from soluble iron supplements in the form of vitamin and mineral tablets all form a common nonheme iron pool (44) which forms the major contribution of iron in most meals.

The efficiency of absorption of heme iron is considerably higher than that of nonheme iron and is affected by the iron status of the individual (44). A person with little or no iron stores may be expected to absorb approximately 35% of heme iron ingested as meat, while a person with adequate iron stores of 500 mg would be expected to absorb approximately 25% (44).

The absorption of nonheme iron depends not only on the iron status of the individual but also on the general composition of the meal. In the iron deficient individual, nonheme iron may be absorbed to as much as 20% when factors enhancing iron absorption are present while in the iron replete individual, nonheme iron absorption may decrease to as much as 2% when enhancing factors are lacking and/or inhibitory factors of iron absorption are present (44).

Until recently, the absorption of iron from composite meals could not be measured. This is now possible by the introduction of the extrinsic tag technique to label food iron (42, 43). The heme iron absorption can be measured using hemoglobin labeled with radio labeled iron as a tracer. The nonheme iron absorption can be measured by mixing the food with an inorganic radiolabeled iron salt.

It is thus possible to study to what extent the availability of iron is influenced by the general composition of a particular meal.

Factors enhancing iron absorption: In several studies, the presence of animal flesh in the diet has been shown to result in an increase in the absorption of nonheme iron from a variety of its sources (43, 45-48). The substitution of 100 g beef for an equivalent amount of protein as egg albumin increased iron absorption to nearly 5-fold in young healthy female volunteers (48). The absorption of nonheme iron from corn or black beans was also shown to increase in human subjects as much as 3-fold when fed with fish or veal (49). Similarly, the absorption of inorganic iron was significantly greater when taken with veal muscle than when ingested with a meal of maize (47). However, all sources of animal proteins are not equivalent in their effect on nonheme iron absorption. While the protein equivalent substitutions of beef, lamb, pork, liver, fish and chicken for the egg albumin resulted in a significant increase in nonheme iron absorption in female subjects, no increase was observed when milk, American cheese or powdered egg were substituted (50). Martinez-Torres and Layrisse (46) found that absorption of iron from black beans in human subjects was doubled when they were fed together with either fish or with an amino acid mixture similar quantitatively and qualitatively with that present in 100 g of fish. These

authors suggested that amino acids derived from dietary protein probably affect iron absorption. Of the amino acids tested, only cysteine proved to enhance iron absorption from black beans. The enhancing effect of certain amino acids such as cysteine and histidine (51-53) and reducing sugars such as fructose (54-56) on nonheme iron absorption has been explained to be due to the formation of chelates with iron which remain soluble during transit through the upper part of the small intestine where iron absorption most rapidly occurs.

Ascorbic acid has also been shown to increase the absorption of nonheme iron from foods (44, 49, 57, 58) as well as from iron supplements in several human studies (59, 60). The increase in iron absorption from a semisynthetic meal (containing ovalbumin as the protein source) was directly proportional to the amount of ascorbic acid added over a range of 25 to 1,000 mg in healthy male volunteers (58). Although ascorbic acid added to the standard meal (containing beef as the protein source) also resulted in a significant increase in iron absorption, the relative enhancement was substantially lower compared to the iron absorption from test meals containing no meat. It was concluded that the effects of ascorbic acid and of meat in facilitating iron absorption were not additive. Similar findings have been reported by Layrisse and co-workers in adult peasants studied in Venezuela (43). The addition of

papaya, containing approximately 70 mg of ascorbic acid to a meal of maize increased absorption from 4.6 to 24.7%. However, when fish which normally enhances iron absorption was added to the meal of maize and papaya, no further increase in iron absorption was observed. It was suggested from these observations that animal tissue and ascorbic acid may enhance iron absorption by the same mechanism. Conrad and co-workers (61) have suggested that ascorbic acid forms a complex with iron at a low pH which remains intact and soluble at the higher pH of the duodenum and donates its iron to the mucosal cells.

Factors inhibiting iron absorption: Egg yolk is a well-known inhibitor of iron absorption (62-65). The iron in egg is strongly complexed to the phosphate of yolk phosphoprotein which accounts for its low iron absorbability (66). Egg albumin, however, does not affect iron absorption (67).

Nonherbal tea is a popular drink in a number of countries where iron deficiency is a problem. Tea has been shown to inhibit nonheme iron absorption from a rice meal (68), a maize-meal porridge (69), and a Western type meal (70). This effect was suggested to be due to the presence of tannates in nonherbal teas forming insoluble complexes with iron which inhibit iron absorption.

Among other dietary factors inhibiting nonheme iron absorption are calcium and phosphate salts (71) and EDTA (72). For example, when EDTA was present at 1:1 molar ratio

with iron, it decreased iron absorption to 72% and at a 2:1 molar ratio, to 50% of that absorbed without EDTA (72). However, several other studies indicated the enhancing effect of EDTA on iron absorption when administered in the form of iron (III) EDTA (73-75) and recommended Fe (III)-EDTA complex as a suitable salt for iron fortification.

The inhibitory effect of bran on iron availability for absorption has been demonstrated in several studies involving human subjects (76-79). Bjorn-Rasmussen has ascribed this inhibitory effect of bran to be due to its high content of phytate (80). It is also postulated that poor iron availability may result in part from the formation of complexes of iron with dietary fiber (81). Various components of fiber in bran, mainly lignin and hemicellulose, were shown to bind with iron and make it unavailable for absorption (82). In an in vitro study by Reinhold et al. (83), the amount of iron bound by neutral detergent fiber prepared from wheat or maize, was shown to depend upon iron concentration, pH, quantity of fiber and the presence and quantity or absence of inhibitors of iron binding. At pH of 6.45, iron binding by fiber was strongly inhibited by ascorbic acid, citric acid, phytic acid, EDTA in low concentrations, and the amino acid cysteine (83). The promotion of iron absorption by these adjuvants may depend in part upon their ability to release iron from its combination with dietary fiber. Kelsay et al. (84) in studies of the effect of high

fiber intakes on mineral metabolism did not find interference with iron availability. However, the high intakes of fruit and vegetables that served as sources of fiber were accompanied by high intakes of ascorbic acid and citric acid and these acids were shown to release iron from its combination with fiber and act as adjuvants to iron absorption (83).

Current methods aimed at promoting adequate iron nutrition in communities that are at risk of iron deficiency have relied on the fortification of dietary staples with iron. However, the value of this approach has been questioned particularly since the demonstration that dietary nonheme iron is absorbed from a common pool (44, 57). If therefore, the diet consists predominantly of cereals such as wheat, maize and rice which have been shown to be extremely poor sources of iron, the added iron is equally poorly absorbed (85). Presumably the dietary factors that restrict the absorption of the intrinsic iron in these foodstuffs have a similar effect on the iron added during fortification.

Dietary fortification with ascorbic acid may be an acceptable alternative to iron fortification in the correction of iron deficiency especially in the nonwestern world where there is a low consumption of meats.

Folate deficiency

Folic acid deficiency is a common cause of megaloblastic anemia in various populations (86). High incidence of folate deficiency during pregnancy has been reported both

in developed and underdeveloped countries (20, 26, 87-91). Among a group of Irish pregnant women at term, 65% had serum folate levels below the lower limit of normal (2 ng/ml) (89). In a Florida study, folacin deficiency was found to be much more prevalent than iron deficiency in low income pregnant women. The serum folacin level was low (3-6 ng/ml) in 48% of all subjects and deficient (<3 ng/ml) in 15% of the subjects (91).

Contrary to these findings, folate deficiency appears to be a relatively uncommon disorder in Central Iran (92). Among a total of 467 subjects studied, only two had serum folate levels below 3 ng/ml and 18 had values in the borderline range (3-5.8 ng/ml). Among the subjects, 170 were pregnant females in different stages of pregnancy and from low socioeconomic class. The well maintained serum folate levels in this population were assumed to be due to the diet which contains large amounts of wholemeal bread or bread prepared from flours of high extraction rate (93). The folate content of these breads was significantly higher than that of white bread from refined flour and no interference with folate absorption by fiber was found in normal subjects (94).

The iron status of pregnant population in Iran

Maternal nutrition, as a result of a particular socioeconomic status, clearly affects the mother's nutritional status (26, 95, 96). In a study by Geissler et al. (95),

dietary intake and nutritional status during the last two weeks of pregnancy and in the third month postpartum were assessed in Iranian women selected from the out-patient populations of two hospitals in Tehran, one of primarily low socioeconomic status (LSE) and the other generally of middle socioeconomic status (MSE). Significant differences were found between the socioeconomic groups in hemoglobin and hematocrit concentration. Mean hemoglobin concentrations were lower in LSE than MSE when measured during pregnancy (10.2 ± 1.1 versus 12.3 ± 0.9 g/dl) and postpartum (10.8 ± 1.4 versus 13.3 ± 1.2 g/dl). In LSE, 33% of the measured hemoglobin levels were below 10 g/dl before parturition and 5% after, whereas in the MSE, there were only 3% below 10 g/dl both before and after delivery. The lower hemoglobin and hematocrit levels in the LSE compared to the MSE during both pregnancy and lactation confirmed the expectation of lower nutritional status, associated with economic condition (26). Assessment of the dietary intake through a 24-hour dietary recall showed a lower consumption of animal products, fruits and vegetables in the LSE compared to the MSE. A similar pattern was found by Froozani et al. (96) among pregnant women attending a public maternity hospital (serving the indigent and low income patients) and those attending a private maternity hospital (for a higher income group) in Tehran. The average daily consumption of foods by the mother was calculated using the total family's consumption

of major foods (obtained from the mother) which assumed an even distribution among family members. The intake of animal proteins, animal fat and citrus fruit was lower and that of legumes and bread was higher in the public group compared to the private group. The analysis of blood samples taken from subjects within 30 minutes of delivery showed a higher hemoglobin concentration in the private group compared to the public group (13.8 ± 1.3 versus 11.1 ± 1.8 g/dl). Mean serum iron, percent saturation of transferrin and mean corpuscular hemoglobin concentration were also significantly higher in the private group than in the public group. However, the public group showed significantly higher hematocrit levels compared to the private group which was not explained. In both studies (95, 96), there was a higher consumption of meat and fruits by the higher income groups compared to the lower income groups. Meat and vitamin C in the diet have been shown to increase nonheme iron absorption (49, 50, 57, 58). The higher hemoglobin values in the higher income groups might possibly be due to a lower parity and higher consumption of meat and vitamin C by these groups.

In a survey of the frequency of anemia in rural populations around Shiraz, Iran, 24% of nonpregnant females and 7% of the males over 16 years old had hemoglobin concentrations of 12 g/dl or less (40). It was suggested that the sex differences in the prevalence of anemia would be due to

iron losses in menses and also due to frequent pregnancies in females. Although iron deficiency was found to be the cause of anemia, diet analysis of 13 year old village and city school boys revealed an average intake of 44.4 mg iron/day (97) which is far in excess of the 18 mg recommended in the dietary allowances (98). However, the question is the availability of ingested iron for absorption. Phytate and fiber have been shown to decrease iron availability for absorption (80-83). Poor iron availability brought about by high intakes of phytate and fiber-rich unleavened wholemeal breads was suggested to be a major factor in etiology of iron-deficiency anemia among villagers (40), despite the high intakes of iron in this region.

METHODS AND PROCEDURES

The methods used in this study included administration of an interview schedule in Farsi, two anthropometric measurements and blood sampling of all subjects selected. Personal visits to the home of some subjects were made for collection of food samples consumed by the subject. All procedures used were performed by the researcher. The specific factors studied were each subject's medical history, socioeconomic characteristics of the family, the family's weekly consumption of specific food items and the subject's hematological status. Food samples were collected for analysis to estimate the range of daily iron intake of the subjects.

Preliminary Work

Prior to the development of the preliminary research proposal, the researcher contacted the chairperson of the Department of Community Medicine at Shiraz University Medical School in Iran to determine whether this research could be conducted through Shiraz Medical School in the city of Shiraz. He agreed to cooperate and provide the required facilities for this research. The preliminary research proposal was written while the researcher was at

Michigan State University. Before the researcher entered the field, informed consent and interview schedules were developed in both English and Farsi (the main language spoken in Iran) and data sheets were developed in Farsi. The entire interview schedule was developed for administration during the initial visit of the patients to the clinic laboratory and it was anticipated that each interview would take 45 minutes to complete. The interview schedules were original and questions were selected after review of the literature and discussion with several professionals. The project was approved by the Michigan State University Committee for Research Involving Human Subjects. The consent forms and the original English interview schedules and data sheets are included in the Appendix A-1 and A-2.

Research proposed in the preliminary proposal was designed to determine the iron status of pregnant women from the low socioeconomic population in the prenatal clinic of the Helal Ahmar Maternity Hospital (Helal clinic) and to conduct an iron supplementation study on anemic subjects. At least 60 subjects with anemia or at high risk of being anemic (hemoglobin level below 11.0 g/dl) were to be selected. If they consented to participate in the study, iron supplements were to be given to them from at least the 24th week of pregnancy. They were to be followed throughout the rest of their pregnancy to determine the effectiveness of the supplementation by the

changes in blood parameters related to iron status.

However, during the winter of 1980, of 73 subjects in the Helal clinic who had Complete Blood Counts (CBC), only six subjects had hemoglobin values below 11.0 g/dl. Four of these subjects had their first referral to the clinic beyond their 30th week of pregnancy, one was in the 28th week and the other in the 23rd week of pregnancy. Therefore, out of 73 possible subjects during a three month period, only one woman was eligible to participate in the proposed supplementation study. As a result, selection of 60 eligible pregnant women with hemoglobin levels below 11.0 g/dl for the iron supplementation study as planned in the preliminary proposal was not feasible within a reasonable period. This problem could not be recognized before entering the field.

Since the iron supplementation study was impractical, the research was redesigned to compare the iron status of pregnant women relative to the family income, food consumption, iron in diet and medical history of the subjects.

Research Site Selection

The location of Shiraz University Medical School in Shiraz was the most important factor in selecting this city because the financial support and laboratory facilities required for conducting this research were available to the researcher. Shiraz University Medical School has been

involved with research in medical and nutritional areas and was supportive of new research projects relevant to community needs. Other factors which were considered in selecting the research site were the community size, geographic location and site physical and social accessibility. Shiraz is a large city where urban pregnant women from low, middle and high socioeconomic populations could be contacted through different maternity clinics.

The specific clinic sites for the proposed research were selected after consultation with several physicians in Shiraz Medical School and after the researcher visited several of the prenatal clinics in Shiraz. The Helal clinic was found to serve the most indigent patients among all prenatal clinics in Shiraz. The prenatal clinic of the Hafez Hospital (Hafez clinic) was suggested as an appropriate location for the selection of subjects who were primarily from middle income population. The prenatal clinic of the Nemazi Hospital was found as a site to which most high income pregnant women were referred. Therefore, these three clinics were selected for the study. The researcher was introduced to the administrators of these clinics by physicians in the Shiraz Medical School.

Pretesting

The pretesting of the interview schedules for item clarity and question sequencing was conducted prior to the

study in the Helal clinic using 40 pregnant women. Several questions on the original interview schedules were omitted subsequent to the pretest due to the non-specific and unquantifiable nature of the responses given. Other changes made in the interview schedule included rewording of certain questions for increased clarity. However, as the study proceeded, it was necessary to streamline the questionnaires to include the most important questions because of the crowdedness of the laboratory room and the desire of the subjects to finish the interview in a shorter period. After interviewing the first 80 subjects, the questions relating to food patterns and beliefs and the 24 hour recall were omitted and some questions relating to socioeconomic characteristics and medical history were shortened. The final revised interview schedules and data sheets concentrated on information related to iron status and nutriture of the subjects and are included in Appendix A-3.

A written consent form for literate subjects and a verbal consent form for illiterate subjects were prepared before the researcher entered the field. Most subjects in the Helal clinic were illiterate and their consent had to be obtained verbally. The verbal consent form was used also for all literate subjects because the researcher and Iranian advisers anticipated that it would be easier for all subjects to comprehend.

Subject Selection

In order to be selected, subjects had to meet all the following criteria:

1. Should be beyond the first 12 weeks of pregnancy at their first visit to the clinic laboratory. Twelve weeks of pregnancy was chosen since it is the earliest period when the change in plasma volume begins (30).
2. Should not have taken any iron supplement during the last 12 months. This was an important criterion because if an iron supplement is or has been taken by the subject, hematological indices will not reflect the subject's dietary iron intake.
3. Should have been a Shiraz resident for at least the last year, because this study was designed to survey an urban population.

If eligible for the study, the women were informed orally of the purpose of the study and were asked to participate. Two hundred ninety four subjects in the Helal and 106 subjects in the Hafez clinics were selected from January 1980 to February 1981. No subjects could be selected from the Nemazi prenatal clinic during a two month period because most had taken iron supplements prior to their first visit with the doctor. Therefore, this clinic was deleted from the study.

In order to have a reference for the iron status of non-pregnant women in the same geographical area and from middle or high socioeconomic population, 21 women, mostly laboratory technicians and students at Shiraz University who volunteered to participate in the study were selected. The criteria for selection of subjects in this group were:

1. Should not have had any pregnancies during the last year,
2. Should not have taken any iron supplements during the last year,
3. Should have been a Shiraz resident for at least the last year,
4. Should be in day 10 to 20 of their menstrual cycle at the time of blood sampling.

Data Collection

All data were collected from January, 1980 to February, 1981. The researcher resided in the City of Shiraz during this period.

Informed Consent

The research project was explained to eligible women and they were asked to participate before the blood samples were taken. Those who agreed to participate verbally consented, indicating that they comprehended the project, participated on their own free will, were free to reject the participation in the study without affecting their treatment and care in the clinic, and knew that information obtained would be treated confidentially.

Biochemical Assessment

All blood samples were obtained between 7:30 and 11:00 a.m. by a laboratory technician. Blood preparation and analysis were completed by the researcher. Approximately

8-9 ml of blood (in addition to what was needed by the clinic laboratory) were obtained from each subject before the interview was administered. Blood was withdrawn by venipuncture using disposable syringes¹. One ml of each blood sample was heparinized immediately using EDTA powder as the anticoagulant. The remainder was transferred to an acid washed test tube, covered with parafilm and allowed to clot. The heparinized samples were used for a complete blood count (CBC) which included a white blood cell (WBC) count, a red blood cell (RBC) count, hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) within 1 to 4 hours after withdrawal, using a Coulter Counter.² The clotted samples were centrifuged³ at 2000 rpm and 22°C for 10 minutes within 2 to 4 hours of withdrawal. The serum was removed and transferred to another acid washed test tube and centrifuged again. The clear serum samples were transferred to small acid washed glass containers, capped and frozen at -18°C for later analysis. One drop of fresh blood was used for preparation of a peripheral blood smear which was later stained with methylene blue for microscopic examination. At the end of

¹JMS Disposable Syringes with Needle. Japan Medical Supply Co., LTD, Hiroshima, Japan.

²Coulter Counter Model S. Coulter Electronics Ltd., Coldharbour Lane, Harpenden, Herts, England.

³MSE Centrifuge (Cool Spin) - FISONs, England.

the study, all of the serum samples were thawed and prepared to be analyzed for serum iron and total iron binding capacity using an atomic absorption spectrophotometer¹ and percent transferrin saturation was calculated. For serum analysis and preparation of iron standards, the method of Olson and Hamlin (21) was followed. All glassware used for storage and analysis of serum samples was soaked for at least 2 hours in 50% (v/v) nitric acid, rinsed thoroughly in deionized water and air dried.

Administration of the Interview Schedule

All interviews were administered in Farsi by the researcher. The interviews were conducted in the clinic laboratory after the blood samples were taken and in the same room where the laboratory technicians were working and patients referred for all kinds of biochemical tests were waiting. Information obtained through the interview included questions related to the socioeconomic characteristics of the subject and her family, her medical history and the weekly consumption of the major food items by her household. Subjects indicated the amount of food consumed either by weight in kilograms or grams or by showing the researcher the measuring cup which was estimated to be closest to the amount of the food item consumed.

¹Varian Techtron Model AA6 Atomic Absorption Spectrophotometer, Australia.

During all interviews, participants had difficulty answering some questions. When this occurred, the questions were reworded to help the participant understand them.

At the end of the interviews, anthropometric measurements (height and weight) of the subjects were obtained by the researcher. Height was measured to the nearest centimeter with the subject in stocking or bare feet, light clothing, eyes straight ahead, with heels and shoulders touching the wall to which a plastic measuring tape was attached. A light board was placed on the top of the subject's head and perpendicular to the wall. Weight was measured to the nearest kilogram with a medical beam scale with the subject in stocking or bare feet and light clothing. In each clinic, all subjects were weighed using the same scale.

Dietary Assessment

The weights of the major food items consumed by all members of each household during one day, one week, two weeks or one month were calculated from family food consumption data. However, the household consumption of some food items like fruits and vegetables could not be easily estimated by most of the subjects and therefore, the information regarding the household intake of these food items was missing.

To estimate the total intake of iron per day, the amount of iron contained in the foods consumed was calculated. Values for the iron content of local food items that have been analyzed by the Food and Nutrition Institute of Iran (99) were compared with those given for the same item in Bowes and Church (100) as a reliable and generally acceptable reference. Due to the wide discrepancy in the iron content of some food items between these two food composition tables, the food items were assigned the iron content in the food tables of Bowes and Church except for some special items peculiar to Iran. Analyzed values for some green herbs that are used fresh or as a mixture of different fresh herbs and green leaves in Iranian dishes were unavailable in Bowes and Church tables. Therefore, for all greens and herbs consumed, values for iron content as analyzed and reported by the Food and Nutrition Institute of Iran (99) were assigned. The greens include spinach, green onions and radishes with leaves. The herbs include parsley, dill, coriander, fenugreek, leek, tarragon, mint and basil.

Different kinds of Iranian breads also did not have an equivalent in Bowes and Church, so values published by Kowhestani et al. (93) were used. Since the iron content of each is presented per 100 g weight, the approximate weight of different kinds of bread made in Shiraz had to be estimated. Three kinds of breads are made in Shiraz in

small bakeries: Bazari (also called Taftoon), Sangak and Mashhady. Each bakery is specialized to make only one kind of bread. Twelve bakeries were randomly selected from different regions of the city, four bakeries for each kind of bread. Duplicate samples of the whole bread loaves were bought from each bakery. All were weighed on the same day using a laboratory balance.¹ See Appendix B for bread weights. Tanok, the village bread made from whole wheat flour is consumed by certain sections of the urban population. This bread is made at home and the size varies in different households. Since subjects indicated the amount of bread consumed by the household as the number of loaves, the total weight of Tanok bread consumed daily in the household could not be estimated due to the variation in the size of bread made in each household. As a result, the subject's intake of iron from Tanok bread could not be estimated although this bread has been analyzed for iron content. Therefore, for those subjects who consumed Tanok bread, the information related to the daily consumption of bread and iron intake from bread was regarded as missing. The iron content of Mashhady bread has not been determined, but due to its similarity in basic ingredients to Bazari bread, the iron level in Bazari bread was used for Mashhady bread.

¹Mettler P1200 Balance.

Food Collection

In order to have a more precise idea about the average daily iron intake in the diet of the pregnant women from the low socioeconomic population, attempts were made to make home visits and collect the foods consumed by the subject per day. At the end of the interviews in the Helal clinic, subjects were randomly asked if a visit to their home for food collection was possible. The purpose of the home visit and food collection was described to them and those willing to participate were selected and told that it would be a surprise visit. No home visits could be made before summer 1980 due to the lack of transportation. Many of the subjects selected for home visits could not be located or reached at home for several reasons (e.g. incorrect address, out of town, moved). Sixteen subjects were located and were visited in their homes. Each subject was asked to collect exactly the same amount of all foods she ate during the next 24 hours, in plastic bags and bowls provided by the researcher. It was emphasized that there should not be any special change made in their selection and preparation of foods due to this collection. Food samples were picked up at the end of the 24-hour period and new bags and bowls were provided for collection of food samples for another 24 hours. Food samples were picked up again after one day. During the fall season, no home visits could be made because of the lack of transportation.

Therefore, subjects were randomly asked if they were willing to participate in the food collection study and bring the samples of the foods consumed by them during the next 24-hour period to the clinic. Six subjects participated. Instructions for food collection were described to them as for the first group and plastic bags and bowls were given to them to bring their collected foods to the Helal clinic. Subjects were paid 200 Rials (approximately \$2.50) for each 24-hour food collection. However, most of them did not accept the money and participated in the study as a favor to the researcher. All home visits and food collections were made by the researcher.

Food Analysis

Collected foods were weighed in the laboratory¹ and were blended with an equal weight of deionized water in a glass blender.² A sample of the homogenates (approximately 200 grams) was transferred to small glass containers and stored at -20°C for later analysis of iron content. The food samples collected through home visits from the sixteen subjects in the first group were destroyed when the freezer failed during a 3-day weekend and therefore had to be discarded. The homogenates of the food samples collected from the six subjects in the second group were

¹Mettler P1200 Balance.

²Waring Blender.

saved because they were stored in another freezer. A duplicate of each food sample weighing 20 grams was mixed with 30 ml of perchloric and nitric acid (1:1 ratio) and digested on hot plates. Digested clear samples were quantitatively transferred to 50 ml volumetric flasks and diluted with deionized water to 50 ml. A second dilution with deionized water (1:2 ratio) was necessary before analyzing the samples by atomic absorption spectrophotometry for the iron concentration. The standard iron solutions used in analyzing the iron content of foods were the same used for serum analysis. All glassware was acid washed and then used for food analysis.

Data Analysis

All data were analyzed on a CDC 6500 computer, utilizing Statistical Package for the Social Sciences (101) programs.

Subject Classification

Data are presented by comparing the subjects selected from the Helal with those from the Hafez clinic and also by comparing different income groups. All subjects were classified into one of the four income groups on the basis of the approximate monthly household income level they reported (Table 1). Subjects with a household income of 20,000 Rials or below were classified as the very low income group (VL).

Table 1. Classification of subjects by their reported household monthly income.

Income Group	Symbol	Household's monthly income range	
		Rials	U.S. Dollar ¹
Very Low	VL	<20,000	<250.00
Low	LO	20,000-34,999	250-437
Middle Low	ML	35,000-49,999	438-624
Middle and High	M-H ²	>50,000	>625

¹One U.S. Dollar is equivalent to approximately 80 Rials.

²This group included the middle income families (MO) with an income of 50,000 to 64,999 Rials, middle high income families (MH) with an income of 65,000 to 79,999 Rials and high income families (HO) with a monthly income of 80,000 Rials or above.

Those with an income level in the range of 20,000-34,999 Rials were regarded as the low income group (L0) while those with a household income of 35,000-49,999 Rials were classified as the middle low income group (ML). All subjects with a monthly income of 50,000 or above were regarded as the middle and high income group (M-H).

Food Consumption

To estimate the subject's mean daily consumption of different food items, the amount consumed in the household should be divided among its members in a manner that differentiates the uneven distribution of foods among them on the basis of different ages, caloric needs and food consumption. Based on caloric needs of different ages (Appendix C-1) published by FAO/WHO (102) and on personal experience and observation, children and adults were classified into three age groups: 1) 1-4 years of age, 2) 5-11 years of age, and 3) 12 years and above. This classification assumed that the members within the age groups had a similar energy requirement (Appendix C-2) and consumed the same amount of food. Children in the first age group (1-4 years) were assumed to consume 1/2 of an adult's total food intake, those in the second age group (5-11 years) to consume 4/5 of an adult and those beyond 12 years of age were estimated to consume the same amount as an adult (Appendix C-3). Therefore, the household weekly consumption

of food was divided not by the total number of persons living in the household but by an adjusted number which reflects the age distribution within the household. See Appendix C-4 for the derivation of the adjusted total family number in each household.

Iron Intake

The average daily intake of iron from different food items was estimated by determining the amount of iron in each of the food items consumed by the subjects per day. The intakes of iron from the meat and poultry were added to represent the mean daily intake of heme iron. The intake of iron from bread, rice, legumes, eggs, greens and herbs and lettuce were added together to estimate the mean daily intake of iron from the nonheme sources. The sum of mean daily heme and nonheme iron intake was regarded as the mean daily total iron intake although it did not include all the possible minor sources of iron in the diet. Since the majority of the subjects could not easily estimate their household consumption of fruits, vegetables, greens and herbs, the mean daily intakes of iron from bread, rice, meat, poultry, legumes and eggs were added together, representing the partial intake of iron by the subjects.

Hematological Status

The guidelines used to interpret the hematological status of the subjects are those proposed by WHO for

pregnant women (3). According to these guidelines, the subject is considered anemic or at high risk of being anemic if hemoglobin, hematocrit, serum iron or percent transferrin saturation falls below the following:

Hemoglobin = 11.0 g/dl
Hematocrit = 33%
Serum iron = 50 µg/dl
Transferrin saturation = 15%

Transferrin saturation level was used as the criterion for the evaluation of the iron status of the subjects. If one subject showed a transferrin saturation below 15.0 percent, she was regarded as having iron deficiency anemia or at high risk of being anemic even if the hemoglobin, hematocrit or serum iron levels were higher than the levels mentioned above.

Statistical Analysis

Data were analyzed by comparing the subjects in different clinics and in different income groups. Data included the following variables: the subject's age, her stage of pregnancy (weeks), number of pregnancies, number of miscarriages, household size, mean daily consumption of the major food items and dietary iron, height, weight and the hematological indices including RBC count, hemoglobin, hematocrit, MCV, MCH, MCHC, serum iron, TIBC and percent transferrin saturation. Mean, standard deviation and frequency distribution of the above variables were calculated.

The Student's t-test (103) was used to compare the subjects selected from two clinics for the mean daily consumption of the major food items, dietary iron intake, height, weight and blood parameters of the pregnant subjects.

Four different sets of comparisons were made between different income groups for the mean daily consumption of the major food items and dietary iron, height and weight of the pregnant subjects. The comparisons were made between:

1. The M-H and the VL income groups
2. The M-H and the LO income groups
3. The M-H and the ML income group, and
4. The VL and the LO income groups.

Bonferroni t-statistics were used to analyze the means for the significance of difference (103). All income groups were compared to the M-H income group which was regarded as a control group of pregnant women including those from the middle and high income families which were assumed to be economically able to purchase adequate amounts of food.

The household monthly income, household size and season which were hypothesized to affect the subject's mean daily consumption of the major food items and dietary iron were entered into separate regression models for each food item and dietary iron to ascertain the significance of the above three independent variables (the household

income, size and season) as they related to food and iron intake. The linear and quadratic relationships and interactions between the independent variables as they related to the dependent variables (i.e. consumption of each food item and dietary iron) were analyzed and the statistically significant variable(s) was (were) selected for the final regression equation. The variables used in first models are given in Appendix D-1.

Seven variables hypothesized to affect the hematological status of subjects were included in regression models. Regression analysis was used to ascertain the statistical significance of the independent variables which included the approximate household monthly income of the subject, the size of household, her age, weeks of pregnancy, number of pregnancies, and mean daily intake of iron from meat and poultry (expressed as heme iron) and season. The linearities as well as nonlinearities and interactions between the above independent variables as affecting four dependent variables (hemoglobin, hematocrit, serum iron and percent transferrin saturation) were analyzed. Therefore, four different sets of regression models were specified, one for each of the selected indicators of hematological status. After analyzing the first regression models, the statistically significant variables were selected and incorporated into reduced regression models and reanalyzed. The final regression equations included the significant variables.

The empirical variables used in the first models are given in Appendix D-2.

In the regression analysis where the subject's household monthly income was required, the average monthly income of each income group was used (Table 1) instead of each subject's income. Since the majority of the subjects gave an estimated range of their household monthly income instead of an accurate figure due to lack of information or fluctuation in income, use of the average monthly income of each group seemed to be logical for regression analysis.

THE RESEARCH SITE AND POPULATION

The Locale

Information about the geographic, demographic, social and economic characteristics of the locale was obtained from the Iran National Census of Population and Housing, November, 1976 (104).

Geographic Characteristics

The city of Shiraz is located in Fars province in the south-central plateau of Iran (Figure 1). This province with an area of 133,299 square kilometers (8% of Iran's area) is located between 27⁰, 3 and 30⁰, 42' north latitude of equator and between 50⁰, 37 and 55⁰, 38' east longitude on Greenwich Meridian. The major city and capital of the Fars province, Shiraz, is at an altitude of 1,750 meters and approximately 100 kilometers north of the Persian Gulf and 950 kilometers south of the Tehran, the capital of Iran. The climate is quite dry with hot summers, generally mild winters, and much sunshine throughout the year.

Demographic Characteristics

Of 2,035,582 persons living in Fars province in 1976, 42.9% lived in urban areas and 57.1% lived in rural areas.



Figure 1. Map of Iran. Location of Tehran, the capital, and Shiraz, the research site are shown.

The population density of this province was 15.3 persons per square kilometer and the median age was 16.8 years for the male and 17.3 years for the female population. In the urban areas, the average number of members per private household was 4.9. Shiraz had a population of 425,813 persons in November, 1976, of which 227,758 (53.5%) were male and 198,055 (46.5%) were female with a sex ratio (males/females) of 115.0.

Medical Care

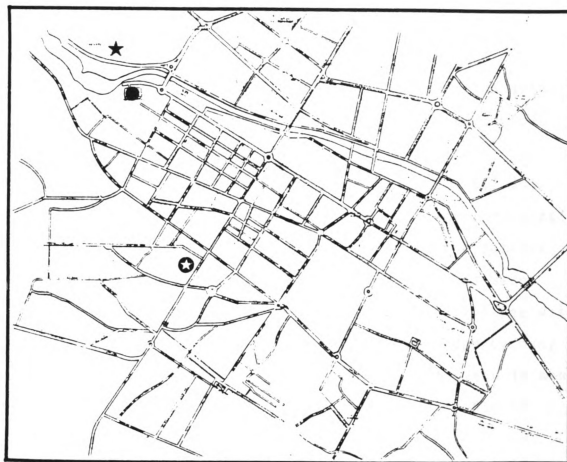
During 1980 there were 25 hospitals and maternity hospitals with 2,233 beds in Shiraz (105). The Shiraz Medical School had four major hospitals in Shiraz, the Sadi, the Nemazi, the Khalili and the Hafez Hospitals, which cared for both free and private patients. The School and its hospitals were serving as the major medical center in the Fars Province. Among the hospitals in Shiraz, there were ten private hospitals and maternity hospitals, five hospitals financed and managed by the Organization of Health (a governmental organization under the jurisdiction of the Ministry of Health), one Army hospital and one maternity hospital, Helal Ahmar, financed by the Treasury General and under the jurisdiction of the Ministry of Health.

The Research Site

Three prenatal clinics in Shiraz were selected for this research project, the prenatal clinic of the Helal Ahmar Maternity Hospital, the prenatal clinic of the Hafez Hospital and the prenatal clinic of the Nemazi Hospital. However, the latter was deleted from the study because attendants did not meet the criteria for participation. Locations of these three hospitals in Shiraz are shown in Figure 2.

The Helal Ahmar Maternity Clinic and Hospital

The Helal Ahmar Maternity clinic and Hospital (Helal) is located in the south-central part of Shiraz. At the time of the study, its participants were primarily from the low socioeconomic population and did not have health insurance because neither they nor their husbands were government employees. Shiraz women as well as women from villages around Shiraz came to this clinic for prenatal care and/or delivery. The clinic and hospital had limited facilities and budget and were financed by the Treasury General. Shiraz University was also involved in serving the patients through the medical students and residents who trained and practiced in the Helal Maternity Hospital. There were 28 beds with an average of 30 to 50 deliveries per 24 hours. Patients were allowed to stay in the hospital from 3 to a maximum of 24 hours after the



★ Helal Ahmar Maternity Clinic and Hospital

★ Hafez Hospital

● Nemazi Hospital

Figure 2. Map of Shiraz. The three prenatal clinics (in the Hospitals) selected for the study are located.

delivery. Due to the lack of facilities, patients recognized to have a complicated delivery (e.g. twins, Cesarean section) were transferred to Hafez Hospital before or during the labor.

Upon their first referral to the clinic, patients had to register and obtain an identification card that would allow prenatal care and hospitalization for birth. The registration fee was 1,000 Rials (approximately \$12.50) which covered the charges for the prenatal care throughout the rest of their term and the delivery. If the patient could not afford to pay that amount, the services were free. During the summer of 1980, the registration fee was raised to 3,000 Rials which could not be afforded by many patients; this resulted in fewer persons coming and in many complaints. The registration fee was lowered again to 1,000 Rials after about one month.

After registration, all patients were sent to the clinic laboratory for several blood tests needed for the completion of their medical records prior to their first visit with the doctor. Blood samples were taken in the morning from 8 to 11 am by laboratory technicians. The average number of patients coming for blood tests was 20 per day which could increase to 40 or decrease to seven or eight during rainy days.

During the Fall of 1980, the clinic's physicians decided not to do a blood test on patients beyond the seventh month of pregnancy because they would not be able to have any prenatal care and their next visit would be for their delivery. This decision was reversed after a few weeks.

After the first visit for registration and a blood test, patients had their first appointment with the doctor one to two months later depending on the clinic load. However a large percentage of the patients attended the first time during their last month of pregnancy and therefore received no prenatal care. They obtained the identification card for hospitalization and had a blood test for their medical records. There were also patients who gave birth at home although they had registered at the clinic.

At their first visit with the doctor, patients with hemoglobin levels below 12.0 g/dl usually received Ferro-sulin pills (200 mg ferrous sulphate) from the clinic pharmacy but if they were able to afford it, a prescription for an iron supplement was given.

The Hafez Hospital

The Hafez Hospital and its clinics are located in the northern part of Shiraz. The participants of the prenatal clinic of the Hafez Hospital (Hafez clinic) were primarily from a middle socioeconomic population living in Shiraz

and most had government health insurance because either they or their husbands were government employees. The prenatal care, delivery and hospitalization expenses ranged from about 15,000 Rials (approximately \$187.50) for normal deliveries to 40,000 Rials (approximately \$500.00) for Cesarean sections. If insured, the patient paid only 10 percent of the expenses.

The Hafez Clinics and Hospital were run by Shiraz University and their budget and administration allowed for much better service and facilities compared to the Helal Maternity Clinic and Hospital. In the maternity department, there were 48 beds; the average number of deliveries ranged from 8 to 10 during 24 hours and the patients were usually hospitalized for 2 to 4 days after delivery.

After registration, patients had their first appointment for blood tests and visiting the gynecologist after about one or two months. Patients were not registered if they presented beyond their fourth or fifth month of pregnancy in which case, they had to go to other public or private clinics. Many of the patients went to private clinics while waiting for their first appointment in the Hafez prenatal clinic and therefore most of them had received iron supplements prior to their first visit to the clinic laboratory. Iron supplements were routinely prescribed for all pregnant women regardless of their blood test results and the patients were responsible for

purchasing them.

The Research Population

Income

Although the Helal clinic served primarily women from the low socioeconomic population and the Hafez prenatal clinic served primarily those of the middle socioeconomic population, there were some overlaps of estimated household monthly income between the two clinic populations. Therefore, subjects selected from both sites were classified on the basis of the approximate household monthly income into one of the four different income groups which are shown in Table 1.

Of 294 subjects selected from the Helal clinic, 66 subjects (22.5%) were in the very low (VL) income group, 158 subjects (53.9%) were in the low (L0) income group, 54 subjects (18.4%) were in the middle low (ML) income group and 15 subjects (5.1%) were in the middle-high (M-H) income group (Table 2). The family income level of one subject could not be estimated. In the Hafez clinic, out of 106 subjects selected, only 1 subject (0.9%) was in the VL income group, 23 subjects (21.7%) were in the L0 income group, 28 subjects (26.4%) were in the ML income group and 54 subjects (50.9%) were in the M-H income group (Table 2).

Table 2. Distribution of subjects selected from Helal and Hafez clinics according to income groups.^a

Clinics	Income Group			
	VL	LO	ML	M-H
	<div style="text-align: center;">% (n)</div>			
Helal (n=293)	22.5 (66)	53.9 (158)	18.4 (54)	5.1 (15)
Hafez (n=106)	0.9 (1)	21.7 (23)	26.4 (28)	50.9 (54)
Total (n=399)	16.8 (67)	45.4 (181)	20.6 (82)	17.3 (69)

^aSee Table 1 for income group classification.

Occupation

Differences in types and remuneration of occupations make comparisons between Iran and the equivalent occupations in industrialized countries difficult.

The Helal clinic attendants or their husbands were not employed by the government while the majority of the husbands of the women attending the Hafez maternity clinic were government employees. This makes a difference between these two clinics in the type of the household head occupations even for the same income reported.

Helal Clinic. Almost all of the attendants were homemakers who had no outside occupation. The occupation of their husbands varied in skill and status. In the very low income group (VL), unemployment was common and the family was spending limited savings or using money borrowed from a relative for living expenses. If the husbands of the attendants in the VL income group had an occupation, it was mainly as an unskilled worker such as a construction laborer, a porter or a peddler and his job was not steady. In the low income group (LO), husbands were in an unskilled to semiskilled job such as a factory worker, plumber, mechanic's helper or construction worker and part-time employment was common. Husbands in the middle-low income groups (ML) had occupations in more skilled and prestigious areas. A husband in the ML income group was a driver, plumber, repair worker, painter or owner of a

small business like a vegetable grocer. Typical examples of occupations of the middle to high income group (M-H) attending the Helal clinic included mechanics, construction builder, or owner of a business such as a fruiter, a grocer or owner of a car repair shop.

Hafez Clinic. Some women attending the Hafez clinic were employed outside of the home, primarily as government employees. Most husbands were employed by the government and the type of occupation ranged from janitors with a low income to university students, teachers and office workers with middle to high income level. Ownership of a private business was the usual source of income for those attendants whose husbands were not government employees and who paid all their clinic and hospital expenses.

Household Size

The mean household size in the Helal and the Hafez clinics was 4.62 and 3.81 persons respectively. The number of persons living in the households ranged from 2 to 16 in the Helal clinic and from 2 to 11 in the Hafez clinic. In the Helal clinic, 17.7% of the households included only 2 persons while 34.9% in the Hafez clinic were composed of 2 persons (Table 3). Households with 7 to 16 persons was 18.7% of the Helal and 5.6% of the Hafez clinic population. In general, the household size increased as family income level decreased (Table 4). While

Table 3. Household size of subjects selected from Helal and Hafez clinics.

Household size	Clinic	
	Helal	Hafez
	% (n)	
2	17.7 (52)	34.9 (39)
3-4	40.1 (118)	38.7 (41)
5-6	23.5 (69)	20.8 (22)
7-8	12.2 (36)	2.8 (3)
9-10	5.1 (15)	1.9 (2)
11-16	1.4 (4)	0.9 (1)
Mean±SD (n)	4.62±2.29 ^a (294)	3.81±2.1 ^b (106)

^aRanged from 2-16.^bRanged from 2-11.

Table 4. Household size of subjects in different income groups.

Household size	Income Groups			
	VL	LO	ML	M-H
	% (n)			
2	14.9 (10)	21.0 (38)	23.2 (19)	33.3 (23)
3-4	40.3 (27)	41.4 (75)	41.4 (34)	33.3 (23)
5-6	26.9 (18)	21.5 (39)	20.7 (17)	23.2 (16)
7-8	10.4 (7)	10.5 (19)	9.8 (8)	7.2 (5)
9-10	7.5 (5)	3.3 (6)	4.9 (4)	2.9 (2)
11-16	0.0 (0)	2.2 (4)	0.0 (0)	1.4 (1)
Mean±SD (n)	4.64±2.1 ^a (67)	4.61±2.43 ^b (181)	4.45±2.14 ^c (82)	4.07±2.2 ^d (69)

^aRanged from 2-10.^bRanged from 2-16.^cRanged from 2-10.^dRanged from 2-11.

14.9% of the subjects in the VL income group had a household size of 2, 33.3% of the M-H income group were a part of 2 member household. In the VL income group, 67.2% of the subjects had a household size of 3 to 6 and 17.9% had a household size of 7 to 16 compared to the M-H income group with 56.5% and 11.5% respectively.

Households which included additional relatives were seen more often in the families of the subjects selected from the Helal clinic compared to those selected from the Hafez clinic. The individuals most likely to be living with the couple and their children were the husband's parents and/or brother(s), sister(s), cousin(s) or a similarly related young person. The wife's relatives seldom lived in her household. This pattern of extended families living together was more common among the families of the subjects from the lower socioeconomic population.

RESULTS

Medical History

The medical history obtained from the subjects included the following information: age of the subject, her stage of pregnancy, number of pregnancies and number of miscarriages. These characteristics are presented by clinics (Table 5, 7, 9 and 11) and by different income groups (Tables 6, 8, 10 and 12).

Age

Women participating in the study were from 13 to 40 years of age. There was a similar age distribution in the two clinics (Table 5). The majority of the subjects in both locations (82% in the Helal and 76.4% in the Hafez clinic) were from 18 to 29 years old; 4.1% of the subjects in the Helal and 5.7% of those in the Hafez clinic were from 13 to 17 years old. The Helal clinic had 13.9% of the subjects in the 30 to 40 years interval while 17.9% of those in the Hafez clinic were in this group. When different income groups are compared, a similar pattern of age distribution is shown between the VL, the L0 and the ML income groups as compared to the M-H income group (Table 6). The percentages of pregnant women in the 18 to

Table 5. Age of subjects selected from Helal and Hafez clinics.

Clinics	Age (years)					Mean±SD
	13-17	18-23	24-29	30-35	36-40	
			% (n)			years
Helal	4.1 (12)	52.8 (152)	29.2 (84)	9.4 (27)	4.5 (13)	23.1 ±6.35
Hafez	5.7 (6)	54.7 (58)	21.7 (23)	15.1 (16)	2.8 (3)	23.7 ±5.43

Table 6. Age of subjects in different income groups.

Income Groups	Age (years)					Mean±SD
	13-17	18-23	24-29	30-35	36-40	
			% (n)			years
VL	6.0 (4)	52.2 (35)	31.3 (21)	6.0 (4)	4.5 (3)	23.7 ±5.57
LO	3.4 (6)	56.8 (100)	27.3 (48)	8.5 (15)	4.0 (7)	22.5 ±6.61
ML	7.3 (6)	52.4 (43)	26.8 (22)	11.0 (9)	2.4 (2)	23.2 ±5.32
M-H	2.9 (2)	46.4 (32)	23.2 (16)	21.7 (15)	5.8 (4)	25.0 ±5.88

23 years and 24 to 29 years group were 46.4% and 23.2% in the M-H income group compared to 52.2% and 31.3% in the VL income group, respectively. In the VL income group, 6.0% of the subjects were between 30 to 35 years of age, compared to 8.5% in the L0, 11.0% in the ML and 21.7% in the M-H income groups in this age interval.

Stage of Pregnancy

Of 294 subjects selected from the Helal clinic, 22.5% were in their 12th to 20th week of pregnancy, 42.5% in the 21st to 30th week and 35.0% in the 31st to 40th week of pregnancy compared to 45.3%, 42.5% and 12.3% of the 106 subjects from the Hafez clinic respectively (Table 7). The percentage of the subjects in the 12th to 20th week of pregnancy in the Hafez clinic was about two-fold that in the Helal clinic, while in the 31st to 40th week of pregnancy, the percentage of the subjects in the Helal clinic was about threefold that in the Hafez clinic. When different income groups are compared, it is shown that with a higher family income, the percentage of the subjects attending in their 12th to 20th week of pregnancy increased and the percentage attending in their 31st to 40th week decreased (Table 8). While 19.4% of the subjects in the VL income group were in the 12th to 20th week of pregnancy and 37.3% in their 31st to 40th week of pregnancy, in the M-H income group, there were 49.3% and 11.6% of the subjects in these

Table 7. Stage of pregnancy of subjects selected from Helal and Hafez clinics.

Clinics	Weeks of pregnancy			Mean±SD
	12-20	21-30	31-40	
		% (n)		weeks
Helal	22.5 (66)	42.5 (125)	35.0 (103)	27.0±7.38
Hafez	45.3 (48)	42.5 (45)	12.2 (13)	20.6±6.08

Table 8. Stage of pregnancy of subjects in different income groups.

Income Groups	Weeks of pregnancy			Mean±SD
	12-20	21-30	31-40	
		% (n)		weeks
VL	19.4 (13)	43.3 (29)	37.3 (25)	26.9±7.40
LO	23.2 (42)	42.0 (76)	34.8 (63)	26.7±7.52
ML	30.5 (25)	45.1 (37)	24.4 (20)	24.4±7.42
M-H	49.3 (34)	39.1 (27)	11.6 (8)	20.9±6.36

two stages of pregnancy respectively.

Number of Pregnancies

As presented in Table 9, 26.2% of the subjects in the Helal clinic and 34.9% of the subjects in the Hafez clinic were primigravida. There were 48.0% of the subjects in the Helal clinic and 51.9% of the subjects in the Hafez clinic who had two to four pregnancies (including the present pregnancy). The percentage of the subjects with five or more pregnancies was 25.5% in the Helal clinic and 13.2% in the Hafez clinic. When different income groups are compared (Table 10), it is shown that with a higher income there was a higher percentage of the subjects who referred with their first pregnancy (26.9% of the subjects in the VL income group compared to 37.7% of the subjects in the M-H income group) and a lower percentage with five or more pregnancies (38.8% of the subject in the VL income group compared to 15.9% of the subjects in the M-H income group).

Number of Miscarriages

Among subjects selected from the Helal clinic, 23.1% had a history of at least one miscarriage compared to 15.1% of the subjects in the Hafez clinic (Table 11). Comparison of different income groups shows that with a higher family income, there was a lower percentage of subjects with a history of miscarriage (Table 12). While 35.8% of the subjects in the VL income group had a history of miscarriage,

Table 9. Number of pregnancies of subjects selected from Helal and Hafez clinics.

Clinics	Number of pregnancies ^a			Mean±SD
	1	2-4	≥5	
		% (n)		pregnancies
Helal	26.2 (77)	48.0 (141)	25.5 (75)	3.2±2.08
Hafez	34.9 (37)	51.9 (55)	13.2 (14)	2.54±1.69

^aIncluding the present pregnancy.

Table 10. Number of pregnancies of subjects in different income groups.

Income Groups	Number of pregnancies ^a			Mean±SD
	1	2-4	≥5	
		% (n)		pregnancies
VL	26.9 (18)	34.3 (23)	38.8 (26)	3.7±2.28
LO	25.4 (46)	53.0 (96)	21.5 (39)	3.1±2.01
ML	29.3 (24)	54.9 (45)	15.9 (13)	2.6±1.64
M-H	37.7 (26)	46.4 (32)	15.9 (11)	2.7±1.96

^aIncluding the present pregnancy.

Table 11. History of miscarriage(s) of subjects selected from Helal and Hafez clinics.

Clinics	No Miscarriages	≥ 1 miscarriage	Mean \pm SD
		% (n)	miscarriages
Helal	76.9 (226)	23.1 (68)	0.36 \pm 0.85
Hafez	84.9 (90)	15.1 (16)	0.25 \pm 0.77

Table 12. History of miscarriage(s) of subjects in different income groups.

Income Groups	No Miscarriages	≥ 1 miscarriage	Mean \pm SD
		% (n)	miscarriages
VL	64.2 (43)	35.8 (24)	0.54 \pm 0.84
LO	79.0 (143)	21.0 (38)	0.37 \pm 1.01
ML	85.4 (70)	14.6 (12)	0.17 \pm 0.47
M-H	87.0 (60)	13.0 (9)	0.19 0.52

21.0% of the subjects in the L0, 14.6% of the subjects in the ML and 13.0% of the subjects in the M-H income groups have had one or more miscarriages.

Food Consumption

The mean daily intake of major food items by the pregnant subjects was obtained from dietary recall data. All subjects were asked about the amount and frequency of consumption of major food items in their household. Some subjects, however, could not estimate their household consumption of all food items requested. The majority of the subjects had problems with estimating the household consumption of fruits and some could only estimate the frequency of fruit consumption by themselves as the number of servings of fruits per week. The household consumption of fresh greens and herbs that are eaten both as cooked and raw was also hard for some subjects to estimate. The vegetable consumption data are not presented because of the wide variation in the types available during different seasons and lack of accurate estimates of consumptions by most of the subjects. Lettuce was the kind of vegetable available most of the time and its consumption is presented. Food items like fish, shrimp and organ meats were used rarely and only by a minority of the subjects. Subjects could not estimate the exact amount consumed and therefore the

mean daily intake of these items was not calculated. The majority of the subjects, however, could give a precise estimate of their household consumption of bread, rice, meat, poultry, eggs and legumes. The amount of food items consumed was indicated by the subjects in the form that was purchased (i.e. raw meat and poultry, uncooked rice, dried legumes).

The mean daily intake of the major food items by the pregnant subjects will be presented both by comparing two clinics and also by comparing different income groups.

Clinics

The mean daily consumption of meat, poultry, eggs and lettuce was significantly lower in the subjects selected from the Helal clinic than those from the Hafez clinic, while the mean daily intake of bread was significantly higher in the former compared to the latter (Table 13). The subjects in the Helal clinic had lower mean daily consumption of greens and herbs than those from the Hafez clinic but the difference between the means was significant only with 90 percent confidence. The mean daily consumption of rice was lower in the subjects from the Helal clinic than those from the Hafez clinic but the difference was not statistically significant. The intake of legumes however, was similar between the subjects of the two clinics (Table 13).

Table 13. Daily consumption of major food items by pregnant subjects selected from Helal and Hafez clinics.

Food Items	Food Intake	
	Helal	Hafez
	g/day	
Bread	405±183.7 ^{a, b} (222)	345±167.5 (51)
Rice	122.5±82.0 (276)	134.8±67.1 (51)
Meat	59.7±48.5 ^c (273)	92.3±42.9 (51)
Poultry	35.1±41.8 ^d (279)	49.4±49.4 (51)
Legumes	21.4±27.0 (273)	20.2±15.5 (50)
Egg	22.9±22.4 ^e (271)	32.3±18.5 (49)
Greens and herbs	83.1±67.0 ^f (213)	106.4±82.0 (39)
Lettuce	77.4±107.5 ^g (239)	172.5±170.6 (44)

^aMean±S.D. The number (n) is shown below the mean.

^bSignificantly higher intakes than subjects from Hafez clinics (P<0.05).

^cSignificantly lower intakes than subjects from Hafez clinic (P<0.01).

^dSignificantly lower intakes than subjects from Hafez clinic (P<0.05).

^eSignificantly lower intakes than subjects from Hafez clinic (P<0.01).

^fSignificantly lower intakes than subjects from Hafez clinic at P<0.10.

^gSignificantly lower intakes than subjects from Hafez clinic (P<0.01).

Information relating to the family food consumption was obtained from all but one of the subjects selected from the Helal clinic and from the last 54 of 106 subjects selected from the Hafez clinic.

Income Groups

Bread. The frequency of consumption of city breads, Sangak, Bazari and Mashhady and the village bread, Tanok, among different households are shown in Table 14. Although all households consumed more than one kind of bread, Table 14 shows the kind of bread preferably consumed by the households. Tanok bread was consumed by 31.8% of the subjects in the VL and 23.5% of the subjects in the L0 income groups compared to 6.2 and 1.9% of the subjects in the ML and the M-H income groups respectively (Table 14). In general, with the increase in family income level, the frequency of consumption of Tanok bread decreased and that of Bazari bread increased. The frequency of consumption of Sangak bread was higher in the VL, the L0 and the ML income groups than in the M-H income group. Mashhady bread was the city bread that was least consumed by all income groups (Table 14).

The mean daily intake of bread by the subjects in the M-H income group was lower than the other three income groups, but only the difference between the VL (457 g/day) and the M-H (356 g/day) income groups was statistically

Table 14. Types of breads consumed by pregnant subjects in different income groups.

Bread Type	Income Group			
	VL	LO	ML	M-H
	% (n)			
Sangak	27.3 (18)	29.0 (47)	30.8 (20)	15.4 (8)
Bazari	31.8 (21)	41.4 (67)	53.8 (35)	75.0 (39)
Mashhady	9.1 (6)	6.2 (10)	9.2 (6)	7.7 (4)
Tanok	31.8 (21)	23.5 (38)	6.2 (4)	1.9 (1)

significant (Table 15). In the VL income group 37.2% of the subjects consumed more than 500 g of bread per day compared to 21.6% of the subjects in the M-H income group (Table 16). Consumption of bread was shown to have a significant linear and inverse relationship to family monthly income (Figure 3), but the income explained only 2.9% of the variation in the subjects' bread consumption (Table 17).

Rice. The mean daily consumption of rice in the M-H income group was higher than the VL and the L0 and lower than the ML income groups and only the difference between the VL and the M-H income groups was significant at $P < 0.10$. In the M-H income group, 13.7% of the subjects consumed below 60 g of rice per day, 23.5% consumed between 60 and 99.9 g and 62.8% consumed more than 100 g of rice compared to 30.6%, 29.1% and 40.3% in the VL income group respectively (Table 16). Consumption of rice was linearly related to the family income (Figure 3) indicating a higher consumption of rice by the higher income families. The household size, however, had an inverse and linear relationship with rice intake (Figure 4). These variables explained 5.7% of the variation in rice consumption (Table 17).

Meat. The mean daily consumption of meat by the subjects in the M-H income group was 91.6 g which was 61.3 g higher than the VL and 32.4 g higher than the L0 income groups. Both differences were statistically significant with 99%

Table 15. Daily consumption of major food items by pregnant subjects in different income groups.

Food Items	Food Intake			
	VL	L0	ML	M-H
	g/day			
Bread	457±211.3 ^{a,b} (43)	395±172.7 (122)	377±171.2 (57)	356±179.6 (51)
Rice	102.1±65.1 ^c (62)	118.2±80.7 (154)	151.6±90.2 (60)	138.2±71.0 (51)
Meat	30.3±32.0 ^{d,e} (61)	59.2±42.9 ^f (154)	92.7±53.9 (59)	91.6±46.8 (50)
Poultry	17.6±29.6 ^{g,h} (61)	32.1±35.3 ⁱ (158)	54.0±46.9 (60)	57.5±58.8 (51)
Legumes	16.0±14.1 (58)	22.2±26.3 (157)	22.4±32.9 (59)	22.9±23.6 (49)
Egg	20.2±20.3 (56)	23.1±23.3 (155)	26.9±22.8 (60)	29.7±17.9 (49)
Greens and herbs	48.7±50.6 ^{j,k} (45)	88.3±67.3 (123)	103.4±76.6 (46)	106.5±74.0 (38)
Lettuce	49.9±77.3 ^l (54)	83.9±105.4 ^m (135)	101.2±145.5 ⁿ (51)	160.5±166.7 (43)

^aMean±SD. The number (n) is shown below the mean.^bVL intake significantly greater than M-H at p<0.02.

Table 15. (cont'd.).

c _{VL}	intake significantly lower than M-H at $p < 0.10$.
d _{VL}	intake significantly lower than M-H at $p < 0.01$.
e _{VL}	intake significantly lower than L0 at $p < 0.01$.
f _{L0}	intake significantly lower than M-H at $p < 0.01$.
g _{VL}	intake significantly lower than M-H at $p < 0.01$.
h _{VL}	intake significantly lower than L0 at $p < 0.10$.
i _{L0}	intake significantly lower than M-H at $p < 0.01$.
j _{VL}	intake significantly lower than M-H at $p < 0.01$.
k _{VL}	intake significantly lower than L0 at $p < 0.01$.
l _{VL}	intake significantly lower than M-H at $p < 0.01$.
m _{L0}	intake significantly lower than M-H at $p < 0.10$.
n _{ML}	intake significantly lower than M-H at $p < 0.10$.

Table 16. Distribution of consumption of major food items among pregnant subjects in different income groups.

Food	Range of intake	Percent of subjects			
		VL	LO	ML	M-H
	g/day	%			
Bread	100-249	16.3	16.4	21.1	29.4
	250-500	46.5	57.4	56.1	49.0
	>500	37.2	26.2	22.8	21.6
		<u>100.0</u> (n=43)	<u>100.0</u> (n=122)	<u>100.0</u> (n=57)	<u>100.0</u> (n=51)
Rice	0-59.9	30.6	23.4	3.3	13.7
	60-99.9	29.1	30.5	31.7	23.5
	100-200	29.0	29.9	40.0	49.1
	>200	11.3	16.2	25.0	13.7
		<u>100.0</u> (n=62)	<u>100.0</u> (n=154)	<u>100.0</u> (n=60)	<u>100.0</u> (n=51)
Meat	0-9.9	32.8	5.2	1.7	2.0
	10-24.9	26.2	9.7	5.1	6.0
	25-49.9	19.7	36.4	13.5	10.0
	50-100	14.7	35.1	39.9	32.0
	>100	6.6	13.6	40.7	40.0
		<u>100.0</u> (n=61)	<u>100.0</u> (n=154)	<u>100.0</u> (n=59)	<u>100.0</u> (n=50)
Poultry	<25	72.1	52.5	28.3	31.4
	25-49.9	14.8	21.5	26.7	19.6
	50-100	9.8	18.3	26.7	33.3
	>100	3.3	7.6	18.3	15.7
		<u>100.0</u> (n=61)	<u>100.0</u> (n=158)	<u>100.0</u> (n=60)	<u>100.0</u> (n=51)
Legumes	<10	31.0	24.8	33.9	32.7
	10-24.9	48.3	43.3	30.5	30.6
	25-40	17.2	21.0	22.0	18.4
	>40	3.4	10.8	13.6	18.3
		<u>100.0</u> (n=58)	<u>100.0</u> (n=157)	<u>100.0</u> (n=59)	<u>100.0</u> (n=49)
Eggs	<10	35.7	32.9	28.3	10.2
	10-29.9	39.3	36.8	26.7	46.9
	30-50	16.1	21.3	35.0	24.5
	>50	8.9	9.0	10.0	18.4
		<u>100.0</u> (n=56)	<u>100.0</u> (n=155)	<u>100.0</u> (n=60)	<u>100.0</u> (n=49)

Table 16. (cont'd.).

Greens and herbs	<25	40.0	14.6	17.4	15.8
	25-49.9	20.0	22.8	6.5	7.9
	50-100	26.7	25.2	32.6	28.9
	>100	13.3	37.4	43.5	47.4
		<u>100.0</u> (n=45)	<u>100.0</u> (n=123)	<u>100.0</u> (n=46)	<u>100.0</u> (n=38)
Lettuce	<40	61.1	40.7	51.0	27.9
	40-99.9	22.2	26.7	15.7	20.9
	100-250	13.0	25.9	13.7	25.6
	>250	3.7	6.7	19.6	25.6
		<u>100.0</u> (n=54)	<u>100.0</u> (n=135)	<u>100.0</u> (n=51)	<u>100.0</u> (n=43)

Figure 3. Effect of household income on bread and rice consumption of pregnant subjects.

Regression Equations:

$$Y_{\text{bread}} = 459.1 - 0.019 X_I \quad R^2 = 2.9\%$$

$$Y_{\text{rice}} = 96.6 + 0.00848 X_I \quad R^2 = 2.9\%$$

where X_I = Household Income

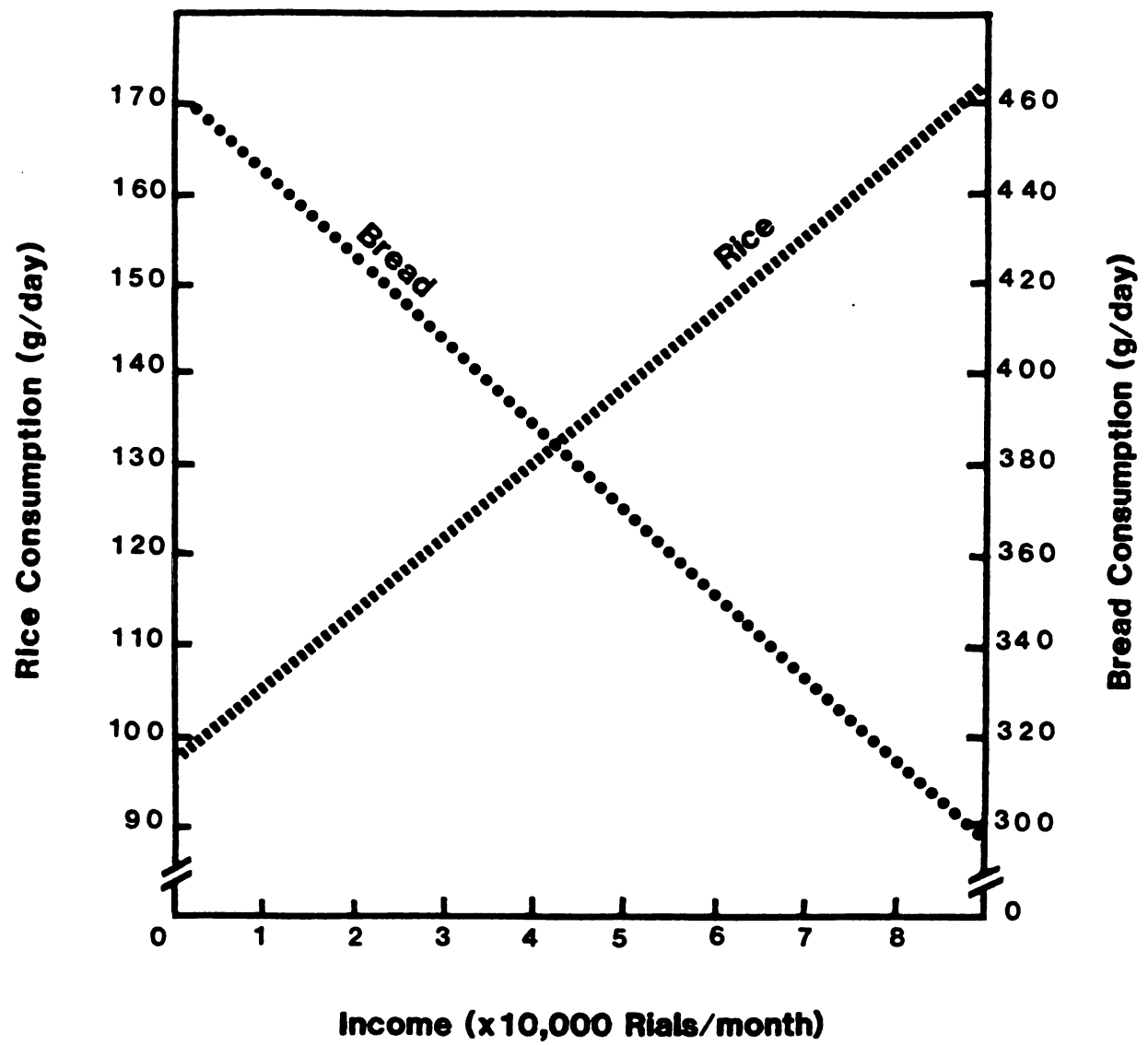


Table 17. Regression equations and regression coefficients for the significant independent variables related to the consumption of the major food items and dietary iron by pregnant subjects.

Dependent Variable (Y)	Regression equation	Independent ^a Variable	Regression Coefficient ±SE	Significance	Percentage Variance Explained
Bread (Y _B)	$Y_B = 459.1 - 0.019X_I$	C: 459.1 ± 25.3 X _I : -0.019 ± 0.0066		0.000 0.005	2.9
Rice (Y _R)	$Y_R = 126.5 + 0.008 X_I - 6.01 X_{HS}$	C: 126.5 ± 13.71 X _I : 0.008 ± 0.0027 X _{HS} : -6.01 ± 1.925		0.000 0.005 0.002	5.7
Meat (Y _M)	$Y_M = 53.7 + 0.024 X_I - 1.37 \times 10^{-6} X_S^2 - 3.51 X_{HS} - 28.7 X_S + 5.01 X_S^2$	C: 53.7 ± 16.96 X _I : 0.024 ± 0.0056 X _I ² : -1.37 × 10 ⁻⁶ ± 0.656 × 10 ⁻⁶ X _{HS} : -3.51 ± 1.074 X _S : -28.7 ± 12.34 X _S ² : 5.01 ± 2.418		0.002 0.000 0.037 0.001 0.021 0.039	24.4
Poultry (Y _P)	$Y_P = 14.2 + 0.0177 X_I - 1.23 \times 10^{-6} X_I^2 - 4.17 X_{HS}$	C: 14.2 ± 10.46 X _I : 0.0177 ± 0.00512 X _I ² : -1.23 × 10 ⁻⁶ ± 0.6057 × 10 ⁻⁶ X _{HS} : -4.17 ± 0.0986		0.176 0.001 0.044 0.000	15.1

Table 17. (cont'd.).

Legumes	$Y_{Lg} = 28.2 - 1.58 X_{HS}$	$C: 28.2 \pm 3.20$ $X_{HS}: -1.58 \pm 0.651$	0.000 0.016	1.8
Egg	$Y_E = 38.1 - 14.2 X_S + 2.89 X_S^2$	$C: 38.1 \pm 6.90$ $X_S: -14.2 \pm 6.28$ $X_S^2: 2.89 \pm 1.227$	0.000 0.024 0.019	1.7
Greens and herbs	$Y_{GH} = 51.7 + 0.035 X_I - 3.11 \times 10^{-6} X_I^2 - 8.60 X_{HS}$	$C: 51.7 \pm 20.66$ $X_I: 0.035 \pm 0.010$ $X_I^2: -3.11 \times 10^{-6} \pm 1.159 \times 10^{-6}$ $X_{HS}: -8.60 \pm 1.977$	0.013 0.001 0.008 0.000	15.0
Lettuce	$Y_{Lt} = 138.2 + 0.043 X_I - 0.006 X_I X_{HS} + 5.52 X_{HS} - 114.9 X_S + 21.0 X_S^2$	$C: 138.2 \pm 56.30$ $X_I: 0.043 \pm 0.009$ $X_I X_{HS}: -0.006 \pm 0.002$ $X_{HS}: 5.52 \pm 7.591$ $X_S: 114.9 \pm 35.0$ $X_S^2: 21.0 \pm 6.75$	0.015 0.000 0.005 0.468 0.001 0.002	21.0
Heme iron (Y_{HFe})	$Y_{HFe} = 1.22 + 1.36 \times 10^{-3} X_I - 7.47 \times 10^{-8} X_I^2 - 0.221 X_{HS}$	$C: 1.22 \pm 0.589$ $X_I: 1.36 \times 10^{-3} \pm 0.288 \times 10^{-3}$ $X_I^2: 7.47 \times 10^{-8} \pm 3.41 \times 10^{-8}$ $X_{HS}: -0.221 \pm 0.0563$	0.039 0.000 0.029 0.000	27.5

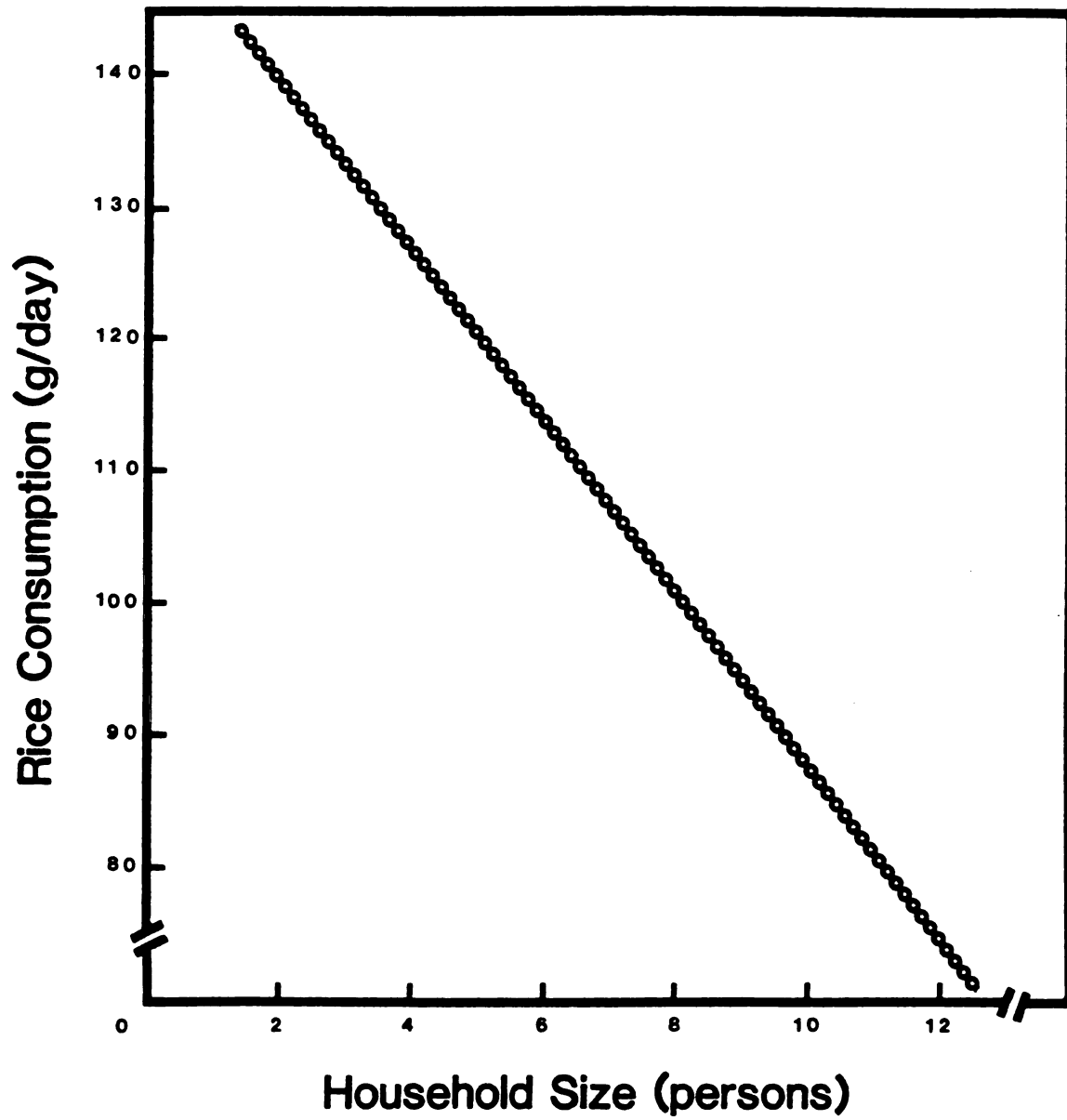
^a C=Constant; X_I =Household Income; X_S =Season; X_{HS} =Household Size;

Figure 4. Effect of household size on rice consumption of pregnant subjects

Regression Equation:

$$Y_{\text{rice}} = 154.0 - 6.61 X_{\text{HS}} \quad R^2 = 3.5\%$$

where X_{HS} = Household Size



confidence (Table 15). Although subjects in the L0 income group had a lower consumption of meat than the ML and the M-H income groups, the former (L0) consumed a higher amount of meat per day than the VL income group and the difference of 28.9 g was highly significant ($P < 0.01$). The mean daily consumption of meat in the ML income group was very close to that of the M-H income group which confirmed the results of regression analysis (Table 17) that the relationship between the meat consumption and the family income was highly significant but not linear (Figure 5). Meat consumption was significantly related to household income and size and season which explained 24.4% of the variation in consumption. The subject's meat intake decreased linearly as household size increased (Figure 6). Meat consumption was lower during spring and summer compared to fall and winter (Figure 7). Household size and season explained 3.8 and 1.8% of the variations in the subjects' meat consumption respectively while the income explained 20.3% of the variation. In the VL income group, 32.8% of the subjects consumed below 10 g of meat per day, while in the L0, the ML and the M-H income groups, only 5.2%, 1.7% and 2.0% consumed less than this amount of meat respectively (Table 16). While 78.7% of the subjects in the VL income group consumed less than 50 g of meat per day, 51.3% of the subjects in the L0, 20.3% in the ML and 18.0% in the M-H income groups consumed less than 50 g of meat daily (Table 16).

Poultry. In general, the poultry consumption by different income groups followed a similar pattern to the meat

Figure 5. Effect of household income on meat and poultry consumption of pregnant subjects.

Regression Equations:

$$Y_{\text{meat}} = 6.64 + 0.022 X_I - 1.04 \times 10^{-6} X_I^2, \quad R^2 = 20.3\%$$

$$Y_{\text{poultry}} = -3.90 + 0.0167 X_I - 1.016 \times 10^{-6} X_I^2 \quad R^2 = 10.5\%$$

where X_I = Household Income

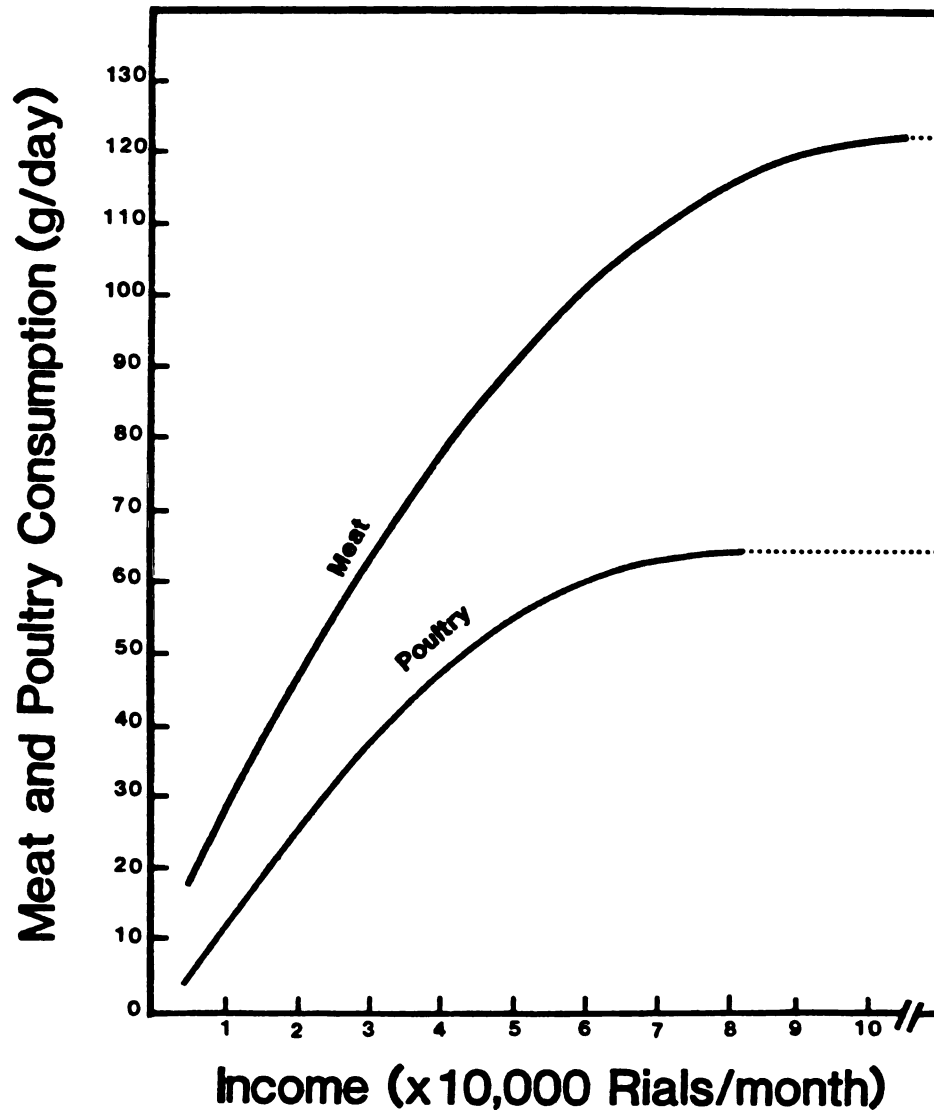


Figure 6. Effect of household size on meat and poultry consumption of pregnant subjects.

Regression Equations:

$$Y_{\text{meat}} = 83.9 - 4.28 X_{\text{HS}} \quad R^2 = 3.8\%$$

$$Y_{\text{poultry}} = 58.1 - 4.61 X_{\text{HS}} \quad R^2 = 5.8\%$$

where X_{HS} = Household Size

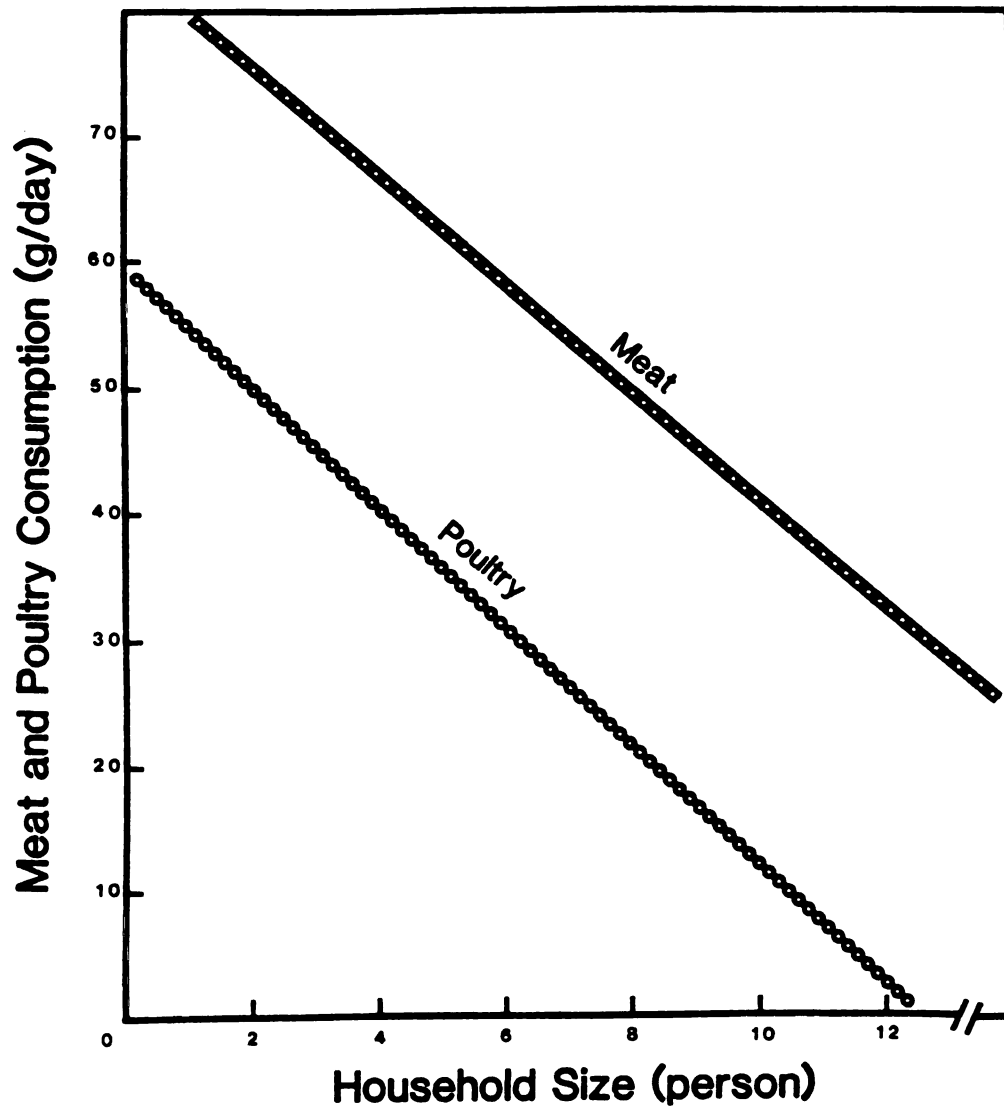
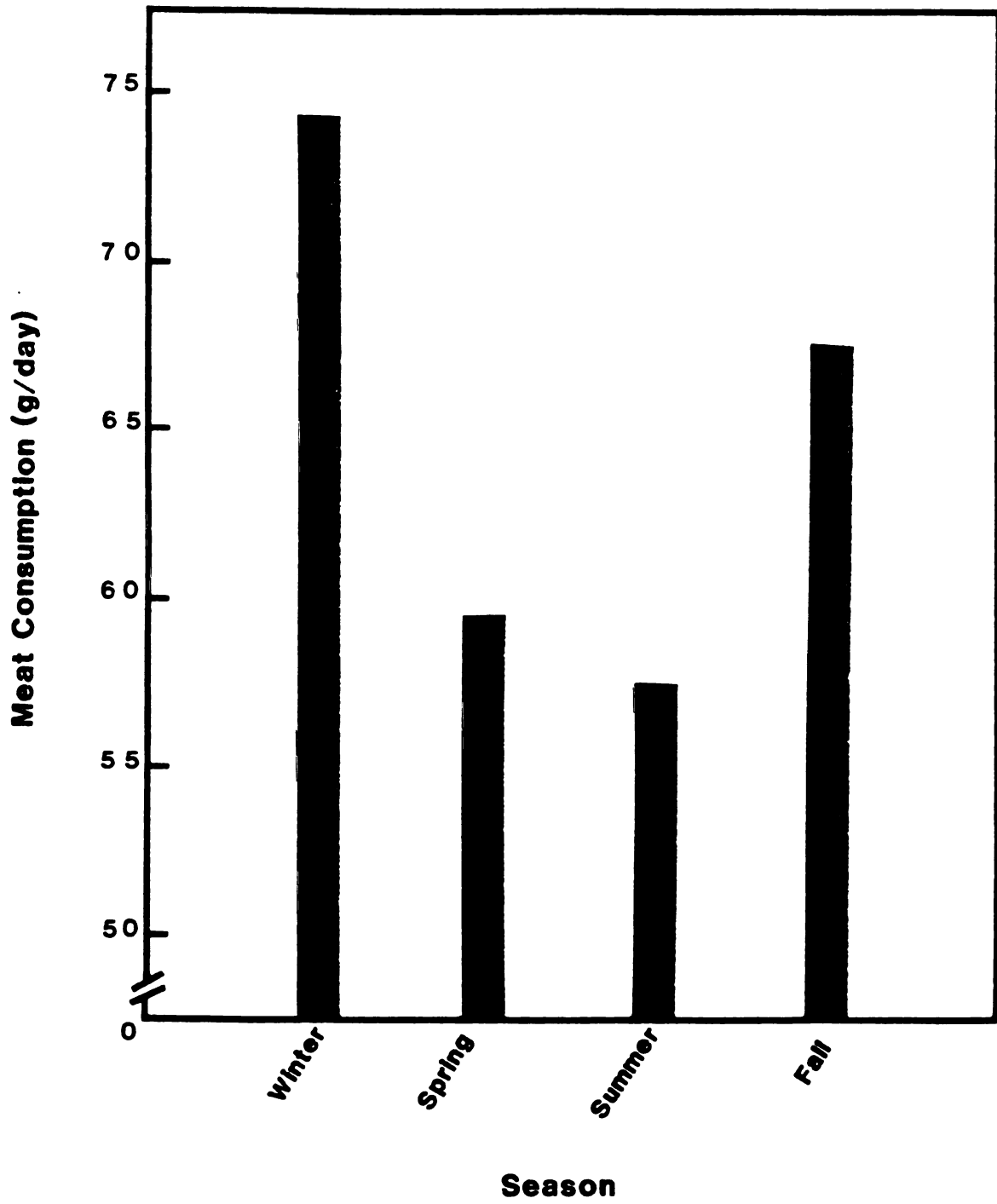


Figure 7. Effect of season on meat consumption of pregnant subjects.

Regression Equation:

$$Y_{\text{meat}} = 101 - 33.1 X_S + 6.18 X_S^2 \quad R^2 = 1.8\%$$

where X_S = Season



consumption. The mean daily intake of poultry in the M-H income group was 57.5 g which was 39.9 g higher than the VL income group and 25.4 g higher than the L0 income group. Both differences were highly significant (Table 15). The difference of 14.5 g between the mean daily consumption of poultry in the VL and the L0 income groups was significant but only with 90% confidence which was due to the wide individual variation. Consumption of poultry in the ML income group was not significantly different from the M-H income group and the difference was only 3.5 g of poultry per day. Poultry consumption increased as household income increased, but the relationship was not linear (Figure 5). The household size, however, had a significant inverse and linear relationship with consumption of poultry (Figure 6). Household income and size explained 15.7% of the variation in poultry consumption (Table 17). In the VL income group, 72.1% of the subjects consumed less than 25 g of poultry per day compared to 52.5% in the L0, 28.3% in the ML and 31.4% in the M-H income group (Table 16).

Legumes. The mean daily consumption of beans, peas and lentils in the L0 and the ML income groups was very close to that of the M-H income group. The VL income group had lower consumption of legumes than all the other income groups but the differences were not significant (Table 15). While 3.4% of the subjects in the VL income group consumed more than 40 g of legumes per day, in the L0, the ML and the M-H income groups, there were 10.8%, 13.6% and 18.3% of the

subjects respectively who consumed more than 40 g daily (Table 16). Regression analysis showed no significant relationship between the family income level and the consumption of legumes, while the household size had a significant inverse relationship to the legume consumption (Figure 8) which explained only 1.8% of the variation in legume intake (Table 17).

Eggs. The subjects in the M-H income group had a higher mean daily consumption of eggs than the subjects in the VL, the LO and the ML income groups but none of the differences were statistically significant (Table 15). In the VL income group, 35.7% of the subjects consumed less than 10 g of egg per day compared to 10.2% of the subjects in the M-H income group consuming below this amount (Table 16). There was no significant relationship between the family income level and the egg consumption in this population. Season, however, was shown to be significantly related with the egg intake (Table 17). Eggs were consumed less during the spring and summer compared to the fall and winter, but season explained only 1.7% of the variation in egg consumption (Figure 9).

Greens and herbs. The mean daily consumption of greens and herbs increased as the family income level increased. The consumption in the VL income group was significantly lower than the LO and the M-H income groups while the mean daily

Figure 8. Effect of household size on legumes and greens and herbs consumption of pregnant subjects.

Regression Equations:

$$Y_{\text{legume}} = 28.2 - 1.58 X_{\text{HS}} \quad R^2 = 1.8\%$$

$$Y_{\text{greens and herbs}} = 13.01 - 9.60 X_{\text{HS}} \quad R^2 = 9.3\%$$

where X_{HS} = Household Size

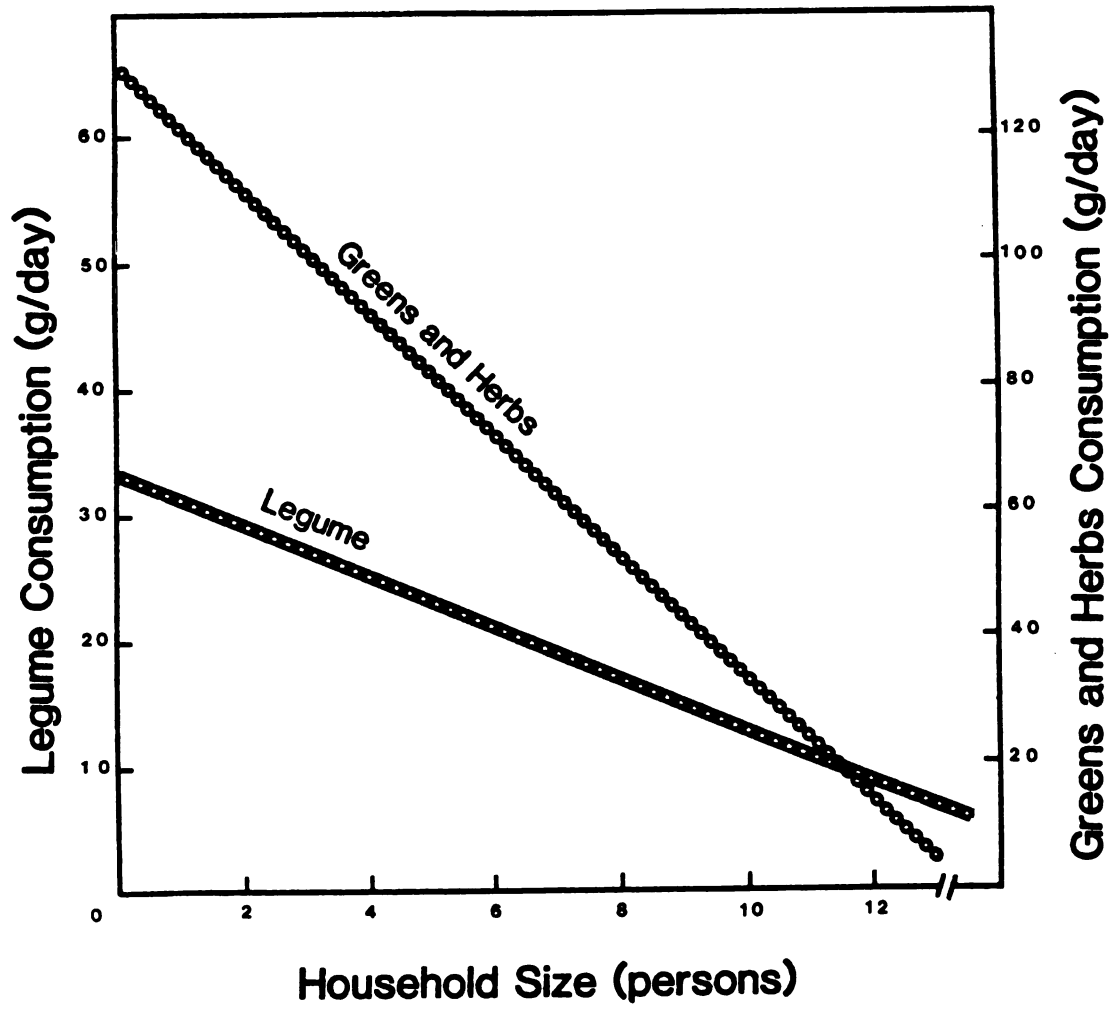
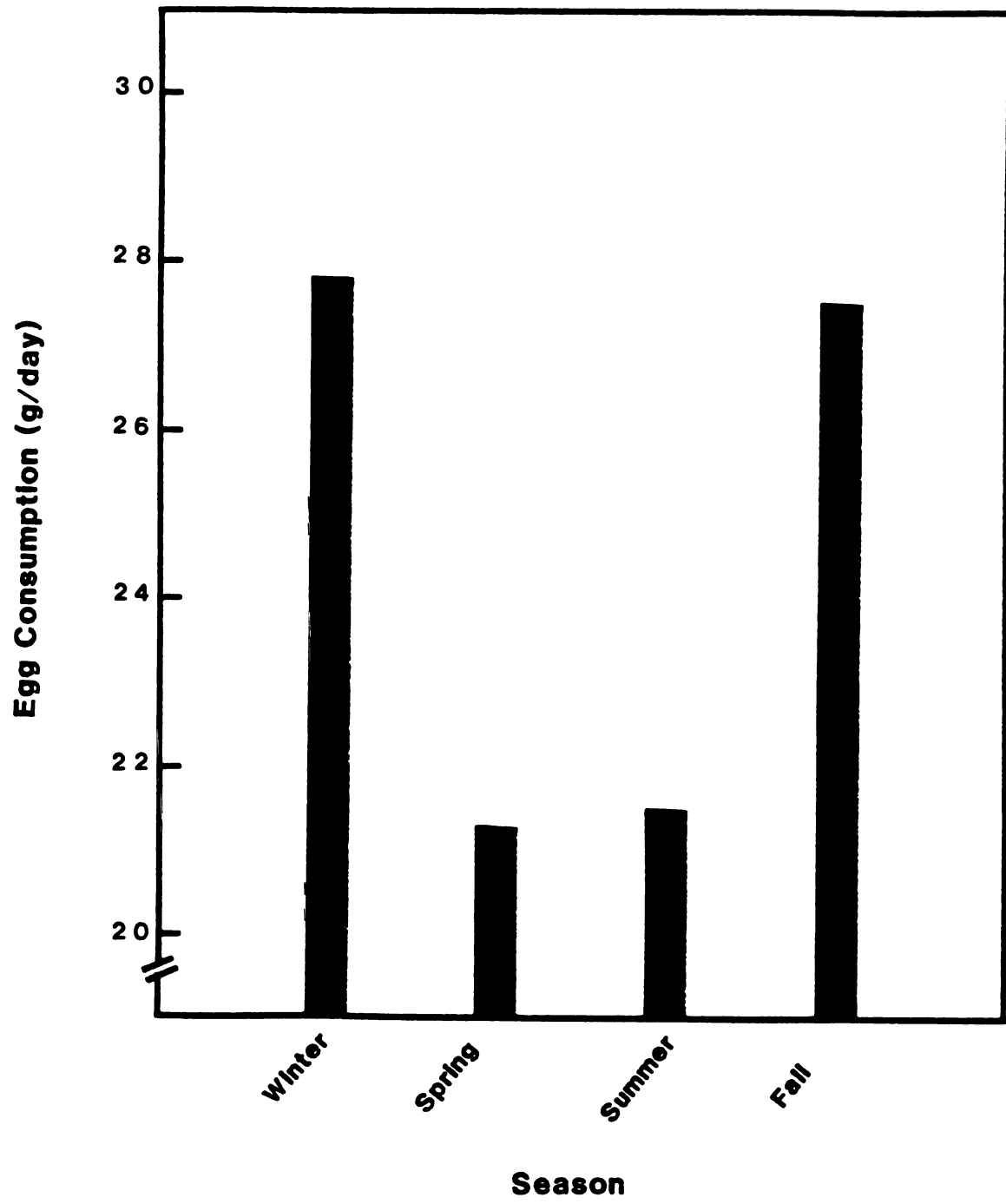


Figure 9. Effect of season on egg consumption of pregnant subjects:

Regression Equation:

$$Y_{\text{Egg}} = 38.2 - 14.2 X_S + 2.89 X_S^2 \quad R^2 = 1.7\%$$

where X_S = Season



consumption in the ML income group was close to that of the M-H income group (Table 15). The mean daily consumption of greens and herbs in the L0 income group was 18.2 g lower than the M-H income group but the difference was not statistically significant. In the VL income group, 60.0% of the subjects consumed less than 50 g of greens and herbs per day compared to 37.4% in the L0 and 23.7% in the M-H income group (Table 16). A significant relationship was found between greens and herbs consumption and the household income and size which explained 15.0% of the variation in their consumption (Table 17). The income and consumption of greens and herbs had a significant and non-linear relationship indicating no further increase in their intake beyond a certain income level (Figure 10). Household size, however, had a negative and linear relationship with the greens and herbs consumption which explained 9.3% of the variation in consumption (Figure 8).

Lettuce. The mean daily consumption of lettuce in the M-H income group was higher than the VL, the L0 and the ML income groups and all differences were statistically significant (Table 15). The household income, the interaction between the household income and size and season were significantly related to the subjects' consumption of lettuce and explained 21.1% of the variation in consumption (Table 17). Income had a linear relationship with the lettuce intake

Figure 10. Effect of household income on lettuce and greens and herbs consumption of pregnant subjects.

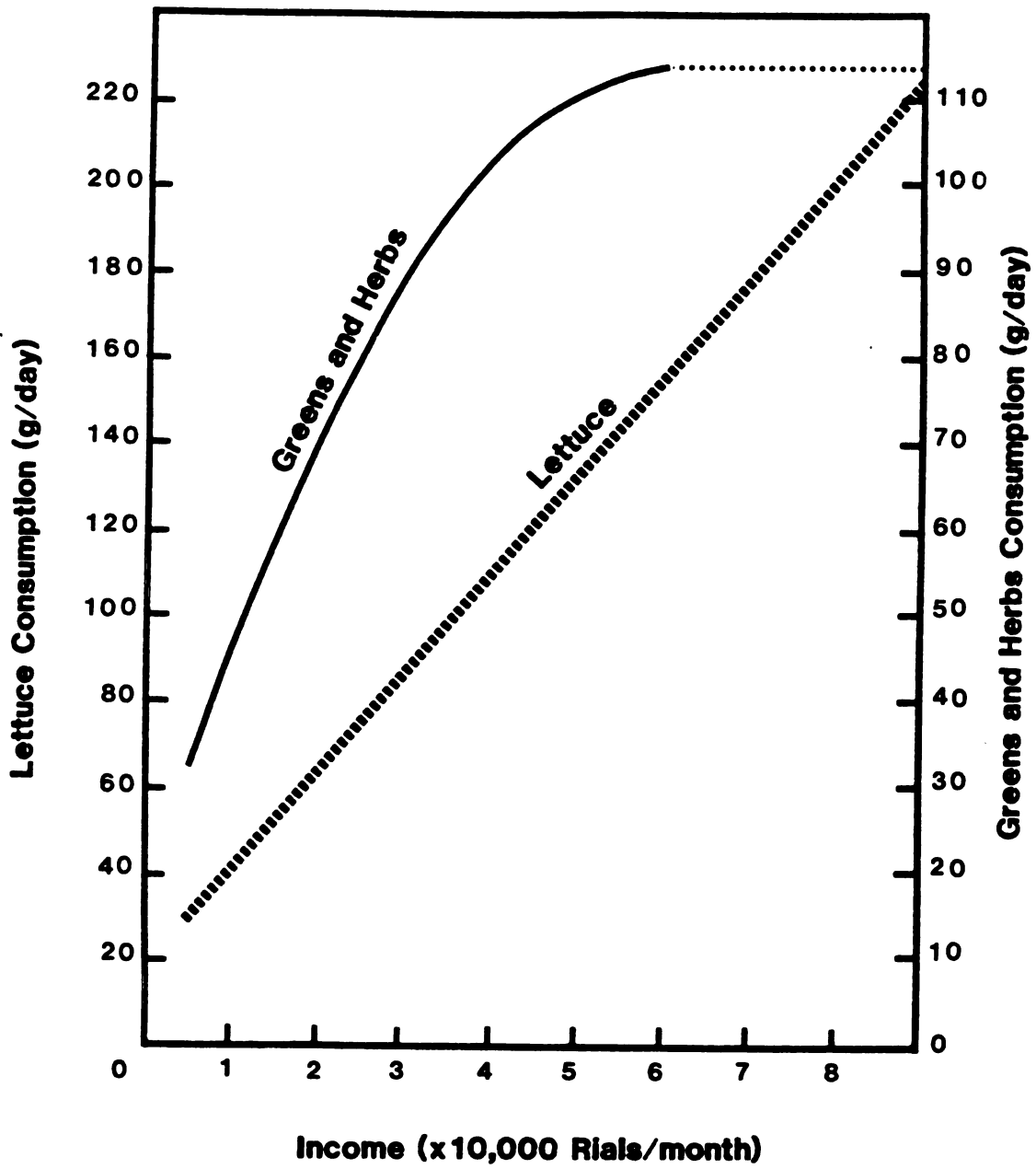
Regression Equations:

$$Y_{\text{greens and herbs}} = 18.4 + 0.0311 X_I - 2.54 \times 10^{-6} X_I^2$$

$$R^2 = 7.5\%$$

$$Y_{\text{lettuce}} = 17.6 + 0.0227 X_I \quad R^2 = 8.9\%$$

where X_I = Household Income



and explained 8.9% of the variation in lettuce consumption (Figure 10). Intake of lettuce was higher during the fall and winter compared to the spring and summer (Figure 11). Season explained 5.4% of the variation in lettuce consumption. While 61.1% of the subjects in the VL income group consumed less than 40 g of lettuce per day, there were 40.7, 51.0 and 27.9% of the subjects in the L0, the ML and the M-H income groups who consumed less than 40 g (Table 16).

Fruits. Less than a quarter of the subjects in the VL income group had at least one serving of fruit every day, while about half of the L0 income group, the majority of the ML income group and nearly all of the M-H income group consumed fruit every day of the week (Table 18). In the VL income group, 23.3% of the subjects either did not consume fruits at all or did so rarely compared to 8.7% of the subjects in the L0 income group. In the ML and the M-H income groups, however, all subjects consumed fruit at least once per week.

In general, the M-H and the ML income groups followed a similar pattern in the consumption of the above food items as compared to the VL and the L0 income groups. The VL and the L0 income groups had a higher consumption of bread and a lower consumption of the other major food items compared to the ML and the M-H income groups.

Figure 11. Effect of season on lettuce consumption of pregnant subjects.

Regression Equation:

$$Y_{\text{lettuce}} = 255.9 - 149.7 X_S + 283 X_S^2 \quad R^2 = 5.4\%$$

where X_S = Season

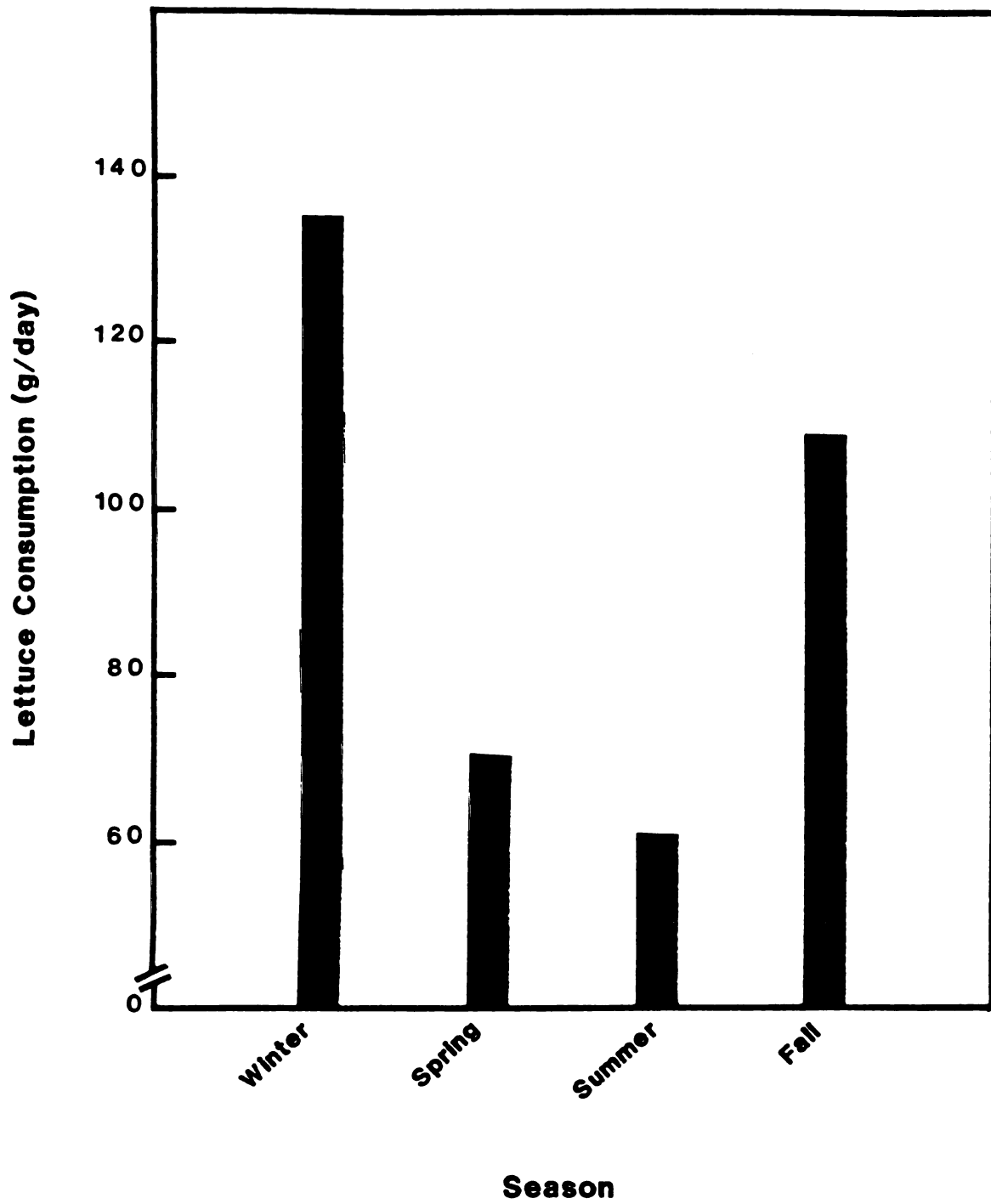


Table 18. Frequency of fruit consumption by pregnant subjects in different income groups.

Fruit Consumption	Percent of Subjects			
	VL	LO	ML	M-H
Day/week ¹	% (n)			
7	23.3 (14)	55.7 (89)	88.7 (55)	96.0 (49)
3-4	28.4 (17)	25.0 (40)	4.8 (3)	2.0 (1)
1	25.0 (15)	10.6 (17)	6.5 (4)	2.0 (1)
0	23.3 (14)	8.7 (14)	0.0 (0)	0.0 (0)
	<u>100.0</u> (60)	<u>100.0</u> (160)	<u>100.0</u> (62)	<u>100.0</u> (51)

¹At least one serving per day.

Iron Intake

The mean daily intake of iron from its major food sources by the pregnant women who were interviewed is presented according to clinic and income.

Clinics

The mean daily intake of iron from the bread was higher in the subjects selected from the Helal clinic than those selected from the Hafez clinic, while the intake of iron from legumes was similar between two clinics and that of rice, meat, poultry, eggs, greens and herbs and lettuce was higher in the latter than in the former (Table 19). All differences between the means were statistically significant except those of rice and legumes. When the major sources of iron in the diet were classified according to heme or nonheme iron, the mean daily intake of heme iron (from the meat and poultry) was shown to be significantly lower in the subjects from the Helal clinic (Table 20). The mean daily intake of nonheme iron from the major food sources (bread, rice, legumes, eggs, greens and herbs, and lettuce) however, was higher in the subjects from the Helal clinic but the difference was not statistically significant. When the heme and nonheme iron intakes were added together, the results indicated a similar total intake of iron between the subjects of the two clinics (Table 20) which were above the recommended allowances of 28 mg per day, set

Table 19. Daily intake of iron from major food sources by pregnant subjects selected from Helal and Hafez clinics.

Source of Iron	Iron Intake	
	Helal	Hafez
	mg/day	
Bread	29.12±15.12 ^{a, b} (222)	23.49±12.77 (51)
Rice	0.962±0.644 (276)	1.06±0.527 (51)
Meat	2.91±2.36 ^c (273)	4.49±2.09 (51)
Poultry	0.53±0.63 ^d (279)	0.74±0.74 (51)
Legumes	1.48±1.86 (273)	1.39±1.07 (50)
Egg	0.508±0.497 ^e (271)	0.719±0.410 (49)
Greens and herbs	3.76±3.05 ^f (213)	4.80±3.69 (39)
Lettuce	1.08±1.51 ^g (239)	2.41±2.39 (44)

^aMean±SD. The number (n) is shown below the mean.

^bSignificantly higher intakes than subjects from Hafez clinic (p<0.05).

^cSignificantly lower intakes than subjects from Hafez clinic (p<0.01).

^dSignificantly lower intakes than subjects from Hafez clinic (p<0.05).

^eSignificantly lower intakes than subjects from Hafez clinic (p<0.01).

^fSignificantly lower intakes than subjects from Hafez clinic at p<0.10.

^gSignificantly lower intakes than subjects from Hafez clinic (p<0.01).

Table 20. Daily intake of heme^a, nonheme^b, partial^c and total^d iron by pregnant subjects selected from Helal and Hafez clinics.

	Iron Intake	
	Helal	Hafez
	mg/day	
Heme iron	3.41±2.59 ^{e, f} (272)	5.23±2.30 (51)
Nonheme iron	37.37±16.38 (144)	34.53±16.42 (38)
Partial iron	36.15±15.62 (196)	32.58±12.90 (47)
Total iron	40.97±16.60 (144)	39.50±16.04 (38)

^aIron from meat and poultry.

^bIron from bread, rice, eggs, legumes, greens and herbs and lettuce.

^cIron from bread, rice, meat, poultry, eggs and legumes.

^dSum of heme and nonheme iron intake.

^eMean±SD. The number (n) is shown below the mean.

^fSignificantly lower intakes than subjects from Hafez clinic (p<0.01).

by the FAO/WHO (27). The average daily intake of iron from the major food sources except vegetables, greens and fruits were added and presented as the partial iron (Table 20). Mean daily intake of partial iron was higher in the Helal subjects than in the Hafez subjects which was mainly due to a higher intakes of iron from bread by the former.

Income Groups

The mean daily intakes of iron from the major food items by the subjects in different income groups are presented in Table 21. Bread was the major source of iron in the diet of all income groups and its intake decreased as the family income level increased. In the VL income group, the average daily intake of iron from bread was 33.30 mg which was significantly higher than that in the L0 and the M-H income groups with a mean of 28.27 mg and 24.50 mg per day, respectively. In the VL income group, 82.3% of the total daily iron intake was derived from bread and this ratio decreased to 68.2% in the L0, 61.7% in the ML and 59.0% in the M-H income group (Table 22). The VL income group had a mean daily intake of 0.80 mg of iron from rice which was 0.28 mg lower than the intake in the M-H income group, a statistically significant difference. The average daily intake of iron from the meat and poultry in the M-H income group was close to that of the ML income group and significantly higher than that of the VL and the L0 income groups.

Table 21. Daily intake of iron from major food sources by pregnant subjects in different income groups.

Source of Iron	Iron Intake			
	VL	L0	ML	M-H
	mg/day			
Bread	33.30±17.86 ^{a,b} (43)	28.27±13.92 (122)	26.90±14.11 (57)	24.50±14.20 (51)
Rice	0.80±0.51 ^c (62)	0.93±0.63 (154)	1.19±0.71 (60)	1.09±0.56 (51)
Meat	1.47±1.56 ^{d,e} (61)	2.88±2.09 ^f (154)	4.51±2.62 (59)	4.46±2.28 (50)
Poultry	0.26±0.44 ^{g,h} (61)	0.48±0.53 ⁱ (158)	0.81±0.70 (60)	0.86±0.88 (51)
Legumes	1.10±0.97 (58)	1.53±1.81 (157)	1.55±2.27 (59)	1.58±1.63 (49)
Egg	0.45±0.45 (45)	0.51±0.52 (155)	0.60±0.51 (60)	0.66±0.40 (49)
Greens and herbs	2.20±2.32 ^{j,k} (45)	3.99±3.08 (123)	4.57±3.52 (46)	4.80±3.26 (38)
Lettuce	0.70±1.08 ^l (54)	1.17±1.48 ^m (135)	1.42 ⁿ ±2.04 (51)	2.25±2.33 (43)

^aMean±SD. The number (n) is shown below the mean.^bVL intake significantly greater than M-H at p<0.02.

Table 21. (cont'd.).

c _{VL}	intake significantly lower than M-H at $p < 0.10$.
d _{VL}	intake significantly lower than M-H at $p < 0.01$.
e _{VL}	intake significantly lower than L0 at $p < 0.01$.
f _{L0}	intake significantly lower than M-H at $p < 0.01$.
g _{VL}	intake significantly lower than M-H at $p < 0.01$.
h _{VL}	intake significantly lower than L0 at $p < 0.10$.
i _{L0}	intake significantly lower than M-H at $p < 0.01$.
j _{VL}	intake significantly lower than M-H at $p < 0.01$.
k _{VL}	intake significantly lower than L0 at $p < 0.01$.
l _{VL}	intake significantly lower than M-H at $p < 0.01$.
m _{L0}	intake significantly lower than M-H at $p < 0.10$.
n _{ML}	intake significantly lower than M-H at $p < 0.10$.

Table 22. Ratio of bread, heme^a and nonheme^b iron in diet to total iron intake^c of pregnant subjects in different income groups.

Source of Iron	Percent of the total iron intake per day			
	VL	LO	ML	M-H
	%			
Bread iron	82.3	68.2	61.7	59.0
Heme iron	3.7	9.3	14.0	15.2
Nonheme iron	96.3	90.7	86.0	84.8

^aIron from meat and poultry.

^bIron from bread, rice, eggs, legumes, greens and herbs and lettuce.

^cSum of heme and nonheme iron intake.

Although the L0 income group had a significantly lower intake of iron from meat and poultry than the M-H income group, it had significantly higher iron intakes from these sources than the VL income group (Table 21). The same pattern is shown when the mean daily intakes of iron from meat and poultry are added together representing the heme iron intakes of different income groups (Table 23). Although the L0 income group had significantly higher intakes of heme iron than the VL income group, both had lower heme iron intakes than the M-H income group and the differences were statistically significant ($P < 0.01$). The mean daily heme iron intake below 3.0 mg was seen in 78.7% of the subjects in the VL, 51.6% in the L0, 25.4% in the ML and 16.0% in the M-H income group (Table 24). The mean daily heme iron intake of above 6.0 mg was seen in 3.3% of the subjects in the VL income group compared to 42.0% of those in the M-H income group (Table 24). Heme iron formed 3.7% of the average total iron intake per day of the subjects in the VL income group while in the L0, the ML and the M-H income groups, 9.3%, 14.0% and 15.2% of the total iron intake were from the meat and poultry respectively (Table 22). Results of regression analysis showed a significant relationship between the household income and size and daily intake of heme iron which explained 27.5% of the variation in intake (Table 17). Heme iron intake increased with the increase in household income (Figure 12) and decrease in household size (Figure 13). The

Table 23. Daily intake of heme^a, nonheme^b, partial^c and total^d iron by pregnant subjects in different income groups.

	Iron Intake			
	VL	L0	ML	M-H
	mg/day			
Heme iron	1.74±1.76 ^{e,f,g} (61)	3.33±2.21 ^h (153)	5.31±2.77 (59)	5.33±2.55 (50)
Nonheme iron	38.53±20.40 (27)	36.44±14.39 (79)	37.80±15.74 (40)	35.05±18.28 (36)
Partial iron	38.16±18.2 (37)	35.17±14.77 (108)	36.20±14.61 (52)	33.14±14.14 (46)
Total iron	39.58±20.12 (27)	39.88±14.62 (79)	43.32±16.18 (40)	40.23±17.88 (36)

^aIron from meat and poultry.

^bIron from bread, rice, eggs, legumes, greens and herbs and lettuce.

^cIron from bread, rice, meat, poultry, eggs and legumes.

^dSum of heme and nonheme iron intake.

^eMean±SD. The number (n) is shown below the mean.

^fVL intake significantly lower than M-H at p<0.01.

^gVL intake significantly lower than L0 intake at p<0.01.

^hL0 intake significantly lower than M-H intake at p<0.01.

Table 24. Distribution of heme^a, nonheme^b and total^c iron intake among pregnant subjects in different income groups.

	Range of Intake mg/day	Percent of Subjects			
		VL	LO	ML	M-H
		%			
Heme iron	<3.0	78.7	51.6	25.4	16.0
	3.0-5.9	18.0	37.2	39.0	42.0
	>6.0	3.3	11.2	35.6	42.0
		100.0	100.0	100.0	100.0
		(n=61)	(n=153)	(n=59)	(n=50)
Nonheme iron	8-23.9	22.2	16.5	20.0	30.6
	24-39.9	44.4	54.4	37.5	41.7
	>40	33.4	29.1	42.5	27.7
		100.0	100.0	100.0	100.0
		(n=27)	(n=79)	(n=40)	(n=36)
Total iron	12-23.9	22.2	10.1	10.0	11.1
	24-39.9	40.7	44.3	32.5	50.0
	>40	37.1	45.6	57.5	38.9
		100.0	100.0	100.0	100.0
		(n=27)	(n=79)	(n=40)	(n=36)

^aIron from meat and poultry.

^bIron from bread, rice, eggs, legumes, greens and herbs and lettuce.

^cSum of heme and nonheme iron intake.

Figure 12. Effect of household income on heme iron intake of pregnant subjects.

Regression Equation:

$$Y_{\text{heme iron}} = 0.267 + 0.00131 X_I - 0.064 \times 10^{-6} X_I^2 \quad R^2 = 24.0\%$$

where X_I = Household Income

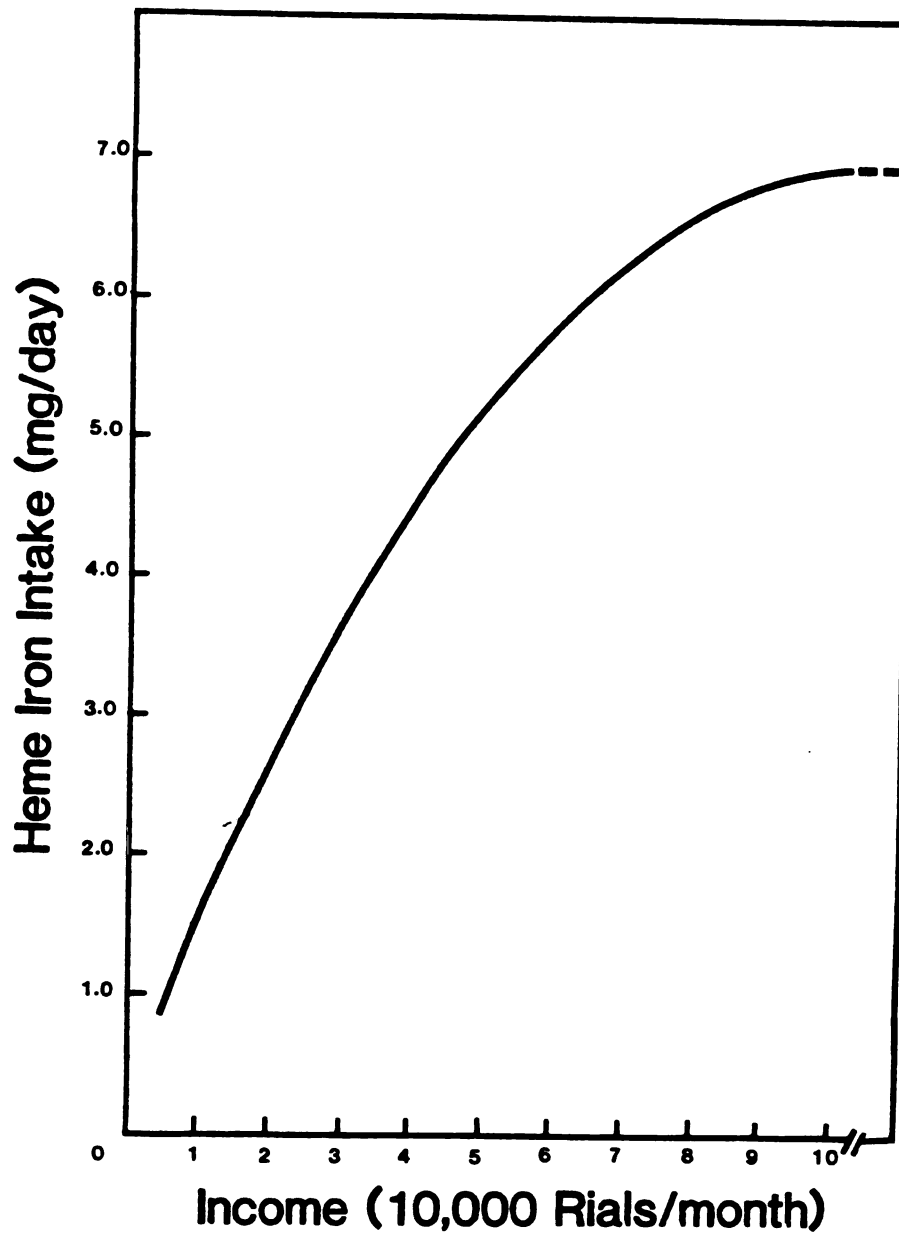
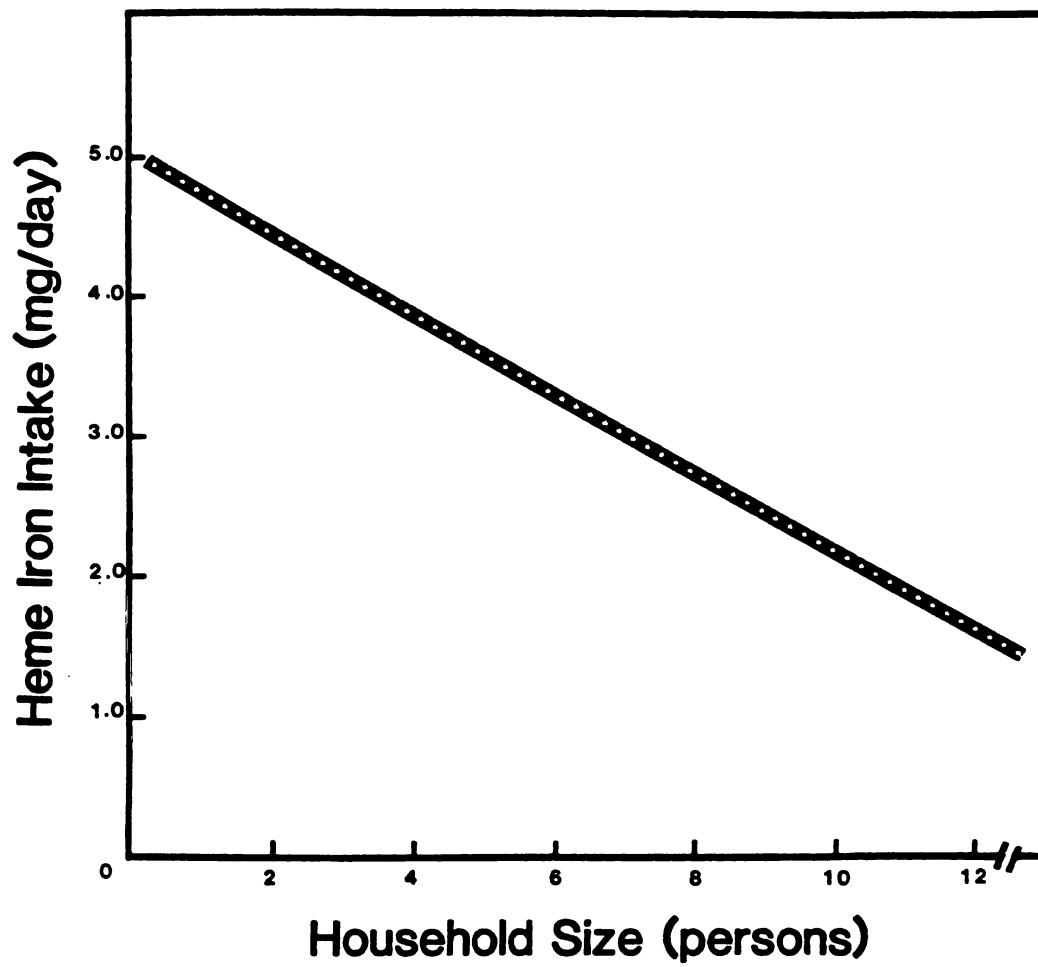


Figure 13. Effect of household size on heme iron intake of pregnant subjects.

Regression Equation:

$$Y_{\text{heme iron}} = 4.91 - 0.269 X_{\text{HS}} \quad R^2 = 5.3\%$$

where X_{HS} = Household Size



VL income group had lower intakes of iron from legumes, eggs, greens and herbs and lettuce than the M-H income group but only the differences in greens and herbs and lettuce were significant (Table 21). The mean daily intake of nonheme iron from bread, rice, legumes, eggs, greens and herbs, and lettuce in the M-H income group was lower than all the other income groups but the differences were not statistically significant (Table 23). The mean daily nonheme iron intake below 24 mg was seen in 22.2% of the subjects in the VL income group compared to 30.6% of those in M-H income group (Table 24). Nonheme iron formed 96.3% of the total daily iron intake of the subjects in the VL income group (Table 22). This ratio decreased to 90.7% in the L0, 86.0% in the ML and 84.8% in the M-H income groups. No significant relationship was found between the nonheme iron intake and the household income, household size or season. When the total intakes of heme and nonheme iron were obtained, the M-H income group had slightly higher intakes than the VL and the L0 and slightly lower intakes than the ML income groups but none of the differences were statistically significant (Table 23). The partial iron intake was lower in the M-H income group and higher in the VL income group compared to the other income groups but none of the differences were statistically significant (Table 23).

Food Analysis

The iron content of foods consumed by six subjects of the Helal clinic during a 24 hour period ranged from 18.41 to 52.39 mg with an average of 32.37 mg (Table 25). The lowest iron intake from bread was 12.8 mg, the highest was 38.88 mg and the ratio of iron from bread to the total iron of the diet ranged from 67.8 to 85.0% indicating that bread was the major source of iron in the diet of these subjects.

Height and Weight

Height

The height of the subjects selected from the Helal and the Hafez clinics is presented in Table 26. The mean and distribution of height between the subjects of two clinics were similar. Among different income groups, the M-L income group had the lowest and the M-H income had the highest mean height (Table 27). In the VL income group, 12.2% of the subjects were less than 150 cm tall compared to 11.7% in the LO income group, 15.4% in the ML income group and 6.1% in the M-H income group below 150 cm high. The percentages of the subjects between a height of 160 and 170 cm ranged from 19.9% in the ML to 38.8% in the M-H income group (Table 27).

Table 25. Iron content of foods collected by pregnant subjects of Helal clinic during a 24-hour period.

Food Sample	Iron Content			
	Bread ^a	Cooked portion ^b	Fruits and Greens ^c	Total
	mg/24 hour			
1	18.16	4.15	4.25	26.56
2	31.20	7.42	0	38.62
3	33.47	3.93	1.96	39.36
4	38.88	10.24	3.27	52.39
5	13.84	3.99	0.58	18.41
6	12.80	5.03	1.04	18.87
Average	24.72	5.79	1.85	32.37

^aIron content was calculated using the analyzed values reported by Kouhestani et al. (93).

^bIron content was determined as analyzed by the researcher.

^cIron content was calculated using the food tables of Bowes and Church (100) for fruits and food tables of Food and Nutrition Institute of Iran (99) for greens and herbs.

Table 26. Height of pregnant subjects selected from Helal and Hafez clinics.

Height	Percent of subjects	
	Helal	Hafez
cm		% (n)
<150	10.7 (27)	14.3 (9)
150-159	63.1 (159)	54.0 (34)
160-170	26.2 (66)	31.7 (20)
Mean height ±SD (n)	156.1±5.26 (252)	156±5.38 (63)

Table 27. Height of pregnant subjects in different income groups.

Height	Percent of subjects			
	VL	LO	ML	M-H
cm	% (n)			
<150	11.5 (6)	11.7 (17)	14.9 (10)	6.1 (3)
150-159	59.6 (31)	61.4 (89)	71.7 (48)	55.1 (27)
160-170	28.9 (15)	26.9 (39)	19.4 (13)	38.8 (19)
Mean Height ±SD (n)	156.1±4.73 (52)	156.2±5.59 (145)	155.3±5.01 (67)	156.9±5.21 (49)

Weight

The average body weights of the pregnant subjects at three different stages of pregnancy are presented in Tables 28 and 29. During the 12th to 20th week of pregnancy, the average body weight was 55.6 kg in the VL and 55.2 kg in the L0 income group compared to 60.7 kg in the ML and 59.2 kg in the M-H income group. The same pattern in the mean body weight was seen between the VL and the L0 and between the ML and the M-H income groups during the 21st to 30th week of pregnancy. However, during the last nine weeks of pregnancy, the average body weight of subjects from different income groups was similar. The mean body weight of the subjects in the M-H income group was not significantly different from the other income groups during any of the stages of pregnancy (Table 29).

Hematological Status

The following indices of maternal hematological status are presented: complete blood count (RBC, hemoglobin, hematocrit, MCV, MCH and MCHC), serum iron, total iron binding capacity (TIBC) and percent transferrin saturation. Data are presented by different clinics and different income groups.

Table 28. Body weight of pregnant subjects selected from Helal and Hafez clinics at different weeks of pregnancy.

Weeks of Pregnancy	Body Weight	
	Helal	Hafez
	kg	
12-20	57.5±9.14 ^a (63)	57.7±8.92 (35)
21-30	57.7±8.54 (117)	59.9±11.02 (24)
31-40	58.1±7.93 (97)	62.6±6.53 (7)

^aMean ± SD. The number (n) is shown below the mean.

Table 29. Body weight of pregnant subjects in different income groups and at different weeks of pregnancy.

Weeks of Pregnancy	Body Weight			
	VL	LO	ML	M-H
	kg			
12-20	55.6±5.55 ^a (12)	55.2±8.12 (39)	60.7±12.72 (21)	59.2±7.13 (26)
21-30	57.2±7.25 (26)	57.2±9.09 (55)	59.1±10.30 (33)	60.8±8.38 (17)
31-40	58.8±10.23 (24)	58.2±7.46 (56)	58.0±6.84 (17)	59.3±5.77 (7)

^aMean±SD. The number (n) is shown below the mean.

Clinic

Mean values for the constituents analyzed in blood of the pregnant subjects selected from the Helal and the Hafez clinics are shown in Table 30. Helal clinic subjects had significantly lower hemoglobin concentration, serum iron level and percent transferrin saturation than the subjects from the Hafez clinic while the other blood constituents were either similar for these two groups of subjects or the differences were not statistically significant. When the means of values for the constituents in blood of the subjects in each clinic were compared based on different stages of pregnancy, it was found that in both clinics all of the constituents analyzed were lower during the 21st to 30th and 31st to 40th week of pregnancy than during the 12th to 20th week of pregnancy (Table 31). The percentage and number of pregnant subjects in each clinic who had deficient values for hemoglobin, hematocrit, serum iron and transferrin saturation are tabulated in Table 32. The guidelines used to classify the deficient levels are those proposed by WHO for pregnant women (3). According to these guidelines, 8.2% of the subjects selected from the Helal clinic had deficient hemoglobin and 6.2% had deficient hematocrit levels, whereas in the Hafez clinic, the corresponding percentage for both deficient hemoglobin and hematocrit levels was 2.8% (Table 32). In the Helal clinic, 6.8% of the subjects had transferrin saturation levels

Table 30. Mean of values for constituents analyzed in blood of pregnant subjects selected from Helal and Hafez clinics.

Constituent	Blood Level	
	Helal	Hafez
RBC ($\times 10^{12}/L$)	4.59 \pm 0.44 ^a (290)	4.63 \pm 0.45 (104)
Hemoglobin (g/dl)	12.9 \pm 1.42 ^b (294)	13.2 \pm 1.15 (106)
Hematocrit (%)	38.4 \pm 3.85 (290)	39.1 \pm 2.92 (106)
MCV (fl)	82.9 \pm 7.3 (286)	83.1 \pm 6.46 (104)
MCH (pg)	28.0 \pm 2.96 (290)	28.0 \pm 2.73 (104)
MCHC (g/dl)	33.8 \pm 1.46 (290)	33.9 \pm 1.31 (106)
Serum iron (μ g/dl)	118.3 \pm 38.9 ^c (262)	133.6 \pm 37.7 (55)
TIBC (μ g/dl)	460.6 \pm 72.7 (257)	461.2 \pm 59.4 (52)
Transferrin Saturation (%)	25.3 \pm 6.8 ^d (237)	28.2 \pm 6.4 (52)

^aMean \pm SD. The number (n) is shown below the mean.

^bSignificantly lower than subjects from Hafez clinic (p<0.10).

^cSignificantly lower than subjects from Hafez clinic (p<0.01).

^dSignificantly lower than subjects from Hafez clinic (p<0.01).

Table 31. Mean of values for constituents analyzed in blood of pregnant subjects selected from Helal and Hafez clinics at different weeks of pregnancy.

Parameter	Clinic	(n)	Weeks of Pregnancy		
			12-20	21-30	31-40
RBC ($\times 10^{12}/L$)	Helal	(290)	4.74 \pm 0.45 ^a (66)	4.48 \pm 0.42 (123)	4.63 \pm 0.43 (101)
	Hafez	(104)	4.75 \pm 0.53 (46)	4.52 \pm 0.38 (45)	4.64 \pm 0.31 (13)
Hemoglobin (g/dl)	Helal	(294)	13.7 \pm 1.30 (66)	12.7 \pm 1.24 (125)	12.7 \pm 1.51 (103)
	Hafez	(106)	13.6 \pm 1.04 (48)	12.9 \pm 1.21 (45)	13.0 \pm 1.05 (13)
Hematocrit (%)	Helal	(290)	40.4 \pm 3.61 (66)	37.8 \pm 3.52 (125)	38.0 \pm 4.0 (99)
	Hafez	(106)	39.7 \pm 2.75 (48)	38.4 \pm 2.92 (45)	39.1 \pm 3.19 (13)
MCV (fl)	Helal	(286)	84.4 \pm 5.99 (66)	83.3 \pm 7.22 (123)	81.5 \pm 8.0 (97)
	Hafez	(104)	83.2 \pm 6.74 (46)	82.8 \pm 5.97 (45)	84.0 \pm 7.49 (13)
MCH (pg)	Helal	(290)	28.8 \pm 2.25 (66)	28.1 \pm 2.73 (123)	27.3 \pm 3.45 (101)
	Hafez	(104)	28.5 \pm 2.71 (46)	27.7 \pm 2.76 (45)	27.5 \pm 2.70 (13)
MCHC (g/dl)	Helal	(290)	34.2 \pm 1.41 (66)	33.8 \pm 1.40 (125)	33.5 \pm 1.53 (99)
	Hafez	(106)	34.3 \pm 1.43 (48)	33.7 \pm 1.19 (45)	33.3 \pm 0.64 (13)

Table 31. (cont'd.).

SFe ($\mu\text{g/dl}$)	Helal (262)	142.2 \pm 32.2 (61)	118.1 \pm 38.3 (103)	103.6 \pm 36.2 (98)
	Hafez (55)	142.4 \pm 37.5 (33)	120.6 \pm 34.2 (18)	119.0 \pm 43.2 (4)
TIBC ($\mu\text{g/dl}$)	Helal (257)	476.4 \pm 76.7 (54)	466.2 \pm 80.9 (103)	446.2 \pm 58.2 (100)
	Hafez (52)	471.7 \pm 51.7 (30)	448.3 \pm 65.6 (18)	439.5 \pm 84.3 (4)
Transferrin Saturation (%)	Helal (237)	30.0 \pm 6.04 (51)	25.1 \pm 6.39 (89)	22.9 \pm 6.28 (97)
	Hafez (52)	29.1 \pm 6.21 (30)	26.8 \pm 6.16 (18)	27.1 \pm 9.54 (4)

^aMean \pm SD. The number (n) is shown below the mean.

Table 32. Pregnant subjects in Helal and Hafez clinics with deficient values for hemoglobin, hematocrit, serum iron and transferrin saturation.

Constituent	Evaluation ^a criteria	Clinic	
		Helal	Hafez
		% (n)	
Hemoglobin (g/dl)	<11.0	8.2 (24)	2.8 (3)
Hematocrit (%)	<33.0	6.2 (18)	2.8 (3)
Serum iron (µg/dl)	<50.0	1.5 (4)	0.0 (0)
Transferrin saturation (%)	<15.0	6.8 (16)	1.9 (1)

^a Criteria used were those proposed by WHO for pregnant women (3).

below 15.0% whereas in the Hafez clinic 1.9% of the subjects were found to have low transferrin saturations. When WHO guidelines for deficient serum iron levels were used, lower percentages of iron deficient subjects were found in both clinics. Among the above four criteria, percent transferrin saturation was chosen for the evaluation of the subjects' iron status. According to this criteria, 6.8% of the subjects in the Helal clinic and 1.9% of those in the Hafez clinic had iron deficiency anemia or were at high risk of being anemic.

The results of microscopic examination of the peripheral blood smears of all pregnant subjects who had a hemoglobin concentration below 12 g/dl are shown in Table 33. Of 48 subjects with hemoglobin level between 11.0 and 11.9 g/dl, 77.1% had normal and 22.9% had slightly microcytic red blood cells with or without slight hypochromia. Among 18 subjects who had a hemoglobin concentration between 10.0 and 10.9 g/dl, 22.2% had normal erythrocytes while 22.2% had slightly microcytic, 38.9% had slightly microcytic and slightly hypochromic, and 16.7% had a characteristic microcytic red blood cells with slight hypochromic. Of the nine subjects with hemoglobin concentrations below 10.0 g/dl, none had normal red blood cells while four subjects (44.4%) had slightly microcytic red blood cells with or without slight hypochromia, three subjects had microcytic erythrocytes with slight hypochromia and two

Table 33. Results of microscopic examination of peripheral blood smears of pregnant subjects with hemoglobin concentrations below 12.0 g/dl.

Red Blood Cell morphology	Hemoglobin Concentration (g/dl)		
	11.0-11.9	10.0-10.9	<10
		% (n)	
Normal	77.1 (37)	22.2 (4)	0.0 (0)
Slightly Microcytic	14.6 (7)	22.2 (4)	22.2 (2)
Slightly microcytic and slightly hypochromic	8.3 (4)	38.9 (7)	22.2 (2)
Microcytic and slightly hypochromic	0.0 (0)	16.7 (3)	33.4 (3)
Microcytic and hypochromic	0.0 (0)	0.0 (0)	22.2 (2)
Megaloblastic or sickle cell	0.0 (0)	0.0 (0)	0.0 (0)
	<u>100.0</u> (48)	<u>100.0</u> (18)	<u>100.0</u> (9)

subjects with the lowest hemoglobin concentrations among all, showed characteristic microcytic and hypochromic red blood cells. No cases of megaloblastic or sickle cell anemia were seen.

Income Group

The mean values for the constituents analyzed in blood of the pregnant subjects in different income groups are given in Table 34. The subjects in the VL and the L0 income groups had lower mean values for all blood constituents than those in the ML and the M-H income groups. The subjects in the VL income group also had lower mean values for the majority of blood constituents than those in the L0 income group. When the mean of values for the constituents in blood of the subjects at different stages of pregnancy were compared, it was found that for most constituents, there were lower mean values during the last 20 weeks of pregnancy than during the 12th to 20th week of pregnancy regardless of the subjects' income group (Table 35). The percentage and number of subjects in different income groups with deficient values for hemoglobin, hematocrit, serum iron and transferrin saturation are presented in Table 36. For these blood constituents, there were a smaller percentage of the subjects in the ML and the M-H income groups with deficient levels compared to the VL and the L0 income groups. While 1.4% of the M-H and 3.7% of

Table 34. Mean of values for constituents analyzed in blood of pregnant subjects in different income groups.

Constituent	Blood Level		
	VL	L0	M-H
RBC ($\times 10^{12}/L$)	4.58 \pm 0.47 ^a (66)	4.59 \pm 0.42 (180)	4.62 \pm 0.48 (82)
Hemoglobin (g/dl)	12.6 \pm 1.21 (67)	12.9 \pm 1.50 (181)	13.4 \pm 1.24 (82)
Hematocrit (%)	37.6 \pm 3.40 (64)	38.4 \pm 4.04 (180)	39.5 \pm 3.27 (82)
MCV (fl)	81.7 \pm 7.50 (63)	82.7 \pm 7.30 (179)	84.5 \pm 6.49 (82)
MCH (pg)	27.3 \pm 3.10 (66)	27.8 \pm 2.97 (180)	28.7 \pm 2.58 (82)
MCHC (g/dl)	33.6 \pm 1.52 (64)	33.7 \pm 1.41 (180)	34.0 \pm 1.34 (82)
Serum iron (μ g/dl)	114.2 \pm 40.4 (58)	116.1 \pm 38.1 (148)	131.6 \pm 35.8 (57)
TIBC (μ g/dl)	446.8 \pm 68.8 (57)	457.4 \pm 71.9 (144)	482.7 \pm 74.2 (53)
Transferrin saturation (%)	25.1 \pm 7.5 (54)	24.8 \pm 6.93 (134)	27.3 \pm 5.73 (51)

^aMean \pm SD. The number (n) is shown below the mean.

Table 35. Mean of values for constituents analyzed in blood of pregnant subjects in different income groups and at different weeks of pregnancy.

Parameter	Income Level	(n)	Weeks of Pregnancy		
			12-20	21-30	31-40
RBC ($\times 10^{12}/L$)	VL	(66)	4.78 \pm 0.39 (13)	4.37 \pm 0.46 (29)	4.73 \pm 0.43 (24)
	LO	(181)	4.70 \pm 0.44 (42)	4.52 \pm 0.39 (77)	4.60 \pm 0.44 (62)
	ML	(82)	4.75 \pm 0.51 (25)	4.53 \pm 0.48 (37)	4.64 \pm 0.41 (20)
	M-H	(65)	4.76 \pm 0.56 (32)	4.47 \pm 0.29 (25)	4.57 \pm 0.22 (8)
Hgb (g/dl)	VL	(67)	13.0 \pm 1.10 (13)	12.4 \pm 1.13 (29)	12.5 \pm 1.34 (25)
	LO	(182)	13.8 \pm 1.11 (42)	12.7 \pm 1.36 (77)	12.6 \pm 1.68 (63)
	ML	(82)	14.1 \pm 1.40 (25)	13.0 \pm 1.17 (37)	13.2 \pm 0.76 (20)
	M-H	(69)	13.5 \pm 1.08 (34)	12.9 \pm 0.97 (27)	12.8 \pm 1.22 (8)
Hct (%)	VL	(64)	39.0 \pm 2.63 (13)	36.9 \pm 3.53 (29)	37.7 \pm 3.52 (22)
	LO	(181)	40.5 \pm 3.39 (42)	37.9 \pm 3.70 (77)	37.6 \pm 4.41 (62)
	ML	(82)	40.8 \pm 3.60 (25)	38.4 \pm 3.13 (37)	40.0 \pm 2.35 (20)
	M-H	(69)	39.5 \pm 3.03 (34)	38.3 \pm 2.32 (27)	38.2 \pm 2.93 (8)

Table 35. (cont'd.)

MCV (fl)	VL	(63)	80.8±7.46 (13)	83.7±8.13 (29)	79.6±6.25 (21)
	LO	(180)	85.0±5.40 (42)	82.5±6.48 (77)	81.2±8.84 (61)
	ML	(82)	84.8±5.12 (25)	83.8±7.34 (37)	85.5±6.49 (20)
	M-H	(65)	83.0±7.39 (32)	83.6±6.13 (25)	82.7±5.55 (8)
MCH (pg)	VL	(66)	26.9±2.54 (13)	28.2±3.06 (29)	26.5±3.18 (24)
	LO	(181)	28.9±2.18 (42)	27.7±2.52 (77)	27.2±3.69 (62)
	ML	(82)	29.3±1.97 (25)	28.5±2.98 (37)	28.4±2.56 (20)
	M-H	(65)	28.5±2.78 (32)	28.2±2.69 (25)	27.8±2.93 (8)
MCHC (g/dl)	VL	(64)	33.4±1.38 (13)	33.8±1.57 (29)	33.6±1.58 (22)
	LO	(181)	34.2±1.27 (42)	33.7±1.36 (77)	33.4±1.48 (62)
	ML	(82)	34.6±1.44 (25)	33.9±1.24 (37)	33.4±1.13 (20)
	M-H	(69)	34.3±1.49 (34)	33.8±1.18 (27)	33.5±1.86 (8)
SFE (μg/dl)	VL	(58)	146.0±36.3 (11)	120.2±41.7 (23)	94.0±29.4 (24)
	LO	(149)	142.7±30.5 (34)	110.7±33.5 (57)	103.1±38.5 (58)
	ML	(57)	133.6±34.6 (17)	130.0±41.3 (27)	132.1±26.0 (13)
	M-H	(53)	145.1±37.0 (32)	125.1±35.2 (14)	96.9±37.6 (7)

Table 35. (cont'd.)

TIBC ($\mu\text{g/dl}$)	VL	(57)	445.8 \pm 69.1 (11)	469.9 \pm 79.5 (22)	426.0 \pm 52.0 (24)
	LO	(145)	484.4 \pm 71.3 (27)	446.3 \pm 80.2 (60)	455.4 \pm 59.7 (58)
	ML	(58)	467.5 \pm 80.1 (18)	504.3 \pm 77.4 (25)	464.9 \pm 53.7 (15)
	M-H	(49)	481.4 \pm 56.7 (28)	454.9 \pm 47.1 (14)	395.0 \pm 45.6 (7)
TS (%)	VL	(54)	33.0 \pm 7.11 (11)	24.5 \pm 6.72 (19)	21.9 \pm 5.51 (24)
	LO	(135)	30.1 \pm 6.18 (26)	24.8 \pm 6.04 (52)	22.3 \pm 6.65 (57)
	ML	(51)	28.0 \pm 5.33 (16)	26.3 \pm 6.56 (22)	28.2 \pm 4.74 (13)
	M-H	(49)	28.0 \pm 5.74 (28)	27.3 \pm 6.84 (14)	24.0 \pm 6.58 (7)

Table 36. Pregnant subjects in different income groups with deficient values for hemoglobin, hematocrit, serum iron and transferrin saturation.

Constituent	Evaluation ^a criteria	Income Group			
		VL	LO	ML	M-H
%					
(n)					
Hemoglobin (g/dl)	<11.0	9.0 (6)	9.4 (17)	3.7 (3)	1.4 (1)
Hematocrit (%)	<33.0	7.8 (5)	7.2 (13)	2.4 (2)	1.4 (1)
Serum iron (μg/dl)	<50.0	1.7 (1)	2.0 (3)	0.0 (0)	0.0 (0)
Transferrin saturation (%)	<15.0	9.3 (5)	7.4 (10)	2.0 (1)	2.0 (1)

^aCriteria used were those proposed by WHO for pregnant women (3).

the ML income group had deficient hemoglobin levels, in the VL and the L0 income groups, there were 9.0% and 9.4% of the subjects respectively with hemoglobin levels below 11.0 g/dl. A similar pattern was found for low hematocrit levels showing a lower percentage of the subjects in the ML and the M-H income group with the hematocrit below 33% than those in the VL and the L0 income groups. However, when the serum iron level below 50 $\mu\text{g/dl}$ was used as a criterion for evaluation of the iron status of the subjects, none of the subjects in the M-H and the ML income groups and only 1.7% of the VL and 2.0% of the L0 income groups showed low serum iron levels. Transferrin saturation levels below 15.0% were seen in 2.0% of the subjects both in the M-H and the ML income groups, 7.4% of those in the L0 and 9.3% of those in the VL income group which are regarded as having iron deficiency anemia or at high risk of being anemic (Table 36). Frequency distribution of hemoglobin, hematocrit, serum iron and transferrin saturation among the subjects in different income groups are given in Table 37. Hematocrit levels below 34.0% were found in 12.5% of the VL, 10.6% of the L0, 2.4% of the ML and 4.3% of the M-H income groups. The percentages of the subjects with serum iron levels below 100 $\mu\text{g/dl}$ were 44.8 in the VL and 41.2 in the L0 income groups whereas in the ML and the M-H income groups, there were 22.8% and 20.8% of the subjects respectively with serum iron levels below

Table 37. Frequency distribution of hemoglobin, hematocrit, serum iron and transferrin saturation among pregnant subjects in different income groups.

Constituent		Income Group			
		VL	LO	ML	M-H
		% (n)			
Hemoglobin (g/dl)	<10	4.5 (3)	2.8 (5)	1.2 (1)	0.0 (1)
	10-10.99	4.5 (3)	6.6 (12)	2.5 (2)	1.4 (1)
	11-12.99	52.5 (35)	38.1 (69)	28.0 (23)	40.6 (28)
	≥13	38.8 (26)	52.5 (95)	68.3 (56)	58.0 (40)
Hematocrit (%)	<34	12.5 (8)	10.6 (19)	2.4 (2)	4.3 (3)
	34-37.9	46.9 (30)	33.3 (60)	24.4 (20)	34.8 (24)
	≥38	40.6 (26)	56.1 (101)	73.2 (60)	60.9 (42)
Serum iron (µg/dl)	<60	5.1 (3)	5.4 (8)	0.0 (0)	5.7 (3)
	60-99.9	39.7 (23)	35.8 (53)	22.8 (13)	15.1 (8)
	100-139.9	29.3 (17)	33.1 (49)	40.3 (23)	37.7 (20)
	≥140	25.9 (15)	25.7 (38)	36.8 (21)	41.5 (22)

Table 37. (cont'd.)

Transferrin saturation (%)	<16	13.0 (7)	9.7 (13)	2.0 (1)	4.0 (2)
	16-19.9	11.2 (6)	17.2 (23)	7.8 (4)	6.1 (3)
	20-27.9	48.1 (26)	39.6 (53)	43.1 (22)	36.7 (18)
	≥28	27.8 (15)	33.6 (45)	47.1 (24)	53.1 (26)

this level. Transferrin saturation levels below 20.0% were seen in 24.2% of the subjects in the VL and 26.9% of those in the LO income group which were more than twice of the corresponding percentages seen in the ML and the M-H income groups which were 9.8% and 10.1% respectively. In general, the frequency distribution of the above four constituents in blood showed that the percentages of the subjects with low blood levels of these constituents were higher in the VL and the LO income groups compared to the ML and the M-H income groups.

Regression analysis was used to find the relationship between the independent variables (Appendix D-2) and each subject's hemoglobin, hematocrit, serum iron and transferrin saturation (the dependent variables). Selected variables assumed to affect these constituents in blood were entered into four different regression models and the following variables were found to have a significant relationship with the level of the above blood constituents:

Hemoglobin. The household monthly income and its square and the weeks of pregnancy and its square were the significant dependent variables related to the subject's hemoglobin level. These variables, however, explained 9.7% of the variation in hemoglobin (Table 38). Income had a significant nonlinear relationship with the hemoglobin level which explained 3.4% of its variation. With the increase

Table 38. Regression equations and regression coefficients for the significant independent variables related to the hemoglobin, hematocrit, serum iron and transferrin saturation levels (dependent variables) of the pregnant subjects.

Dependent Variable	Regression Equation	Independent ^a Variable	Regression Coefficient \pm SE	Significance	Percentage variance explained
Hemoglobin	$Y_{Hgb} = 14.7 + 4.37 \times 10^{-4} X_I - 4.46 \times 10^{-8} X_I^2 - 0.169 X_{wp} + 2.48 \times 10^{-3} X_{wp}^2$	C	14.7 \pm 0.72	0.0	
		X_I	4.37 $\times 10^{-4} \pm 1.553 \times 10^{-4}$	0.005	
		X_I^2	-4.46 $\times 10^{-8} \pm 1.854 \times 10^{-8}$	0.017	9.7
		X_{wp}	-0.169 \pm 0.054	0.002	
		X_{wp}^2	2.48 $\times 10^{-3} \pm 1.042 \times 10^{-3}$	0.018	
Hematocrit	$Y_{Hct} = 43.5 + 1.20 \times 10^{-3} X_I - 1.32 \times 10^{-7} X_I^2 - 0.49 X_{wp} + 7.69 \times 10^{-3} X_{wp}^2$	C	43.5 \pm 1.96	0.00	
		X_I	1.20 $\times 10^{-3} \pm 0.423 \times 10^{-3}$	0.005	
		X_I^2	-1.32 $\times 10^{-7} \pm 0.504 \times 10^{-7}$	0.009	8.1
		X_{wp}	-0.493 \pm 0.146	0.001	
		X_{wp}^2	7.69 $\times 10^{-3} \pm 2.812 \times 10^{-3}$	0.007	
Serum Iron	$Y_{Sfe} = 234.1 - 7.10 X_{wp} + 0.097 X_{wp}^2$	C	234.1 \pm 24.66	0.000	
		X_{wp}	-7.10 \pm 1.989	0.000	17.4
		X_{wp}^2	0.097 \pm 0.0376	0.010	
Transferrin Saturation	$Y_{TS} = 44.9 - 1.19 X_{wp} + 0.016 X_{wp}^2$	C	44.9 \pm 4.60	0.000	
		X_{wp}	-1.19 \pm 0.369	0.001	15.7
		X_{wp}^2	0.016 \pm 0.007	0.020	

^a C=Constant, X_I =Household income, X_{wp} =Weeks of pregnancy.

in family income from very low to higher levels, the hemoglobin level also increased and its maximum level was predicted at the household monthly income of 60,360 Rials (Figure 14). Weeks of pregnancy had a significant inverse relationship to the subjects' hemoglobin level, explaining 7.6% of the variation in its level. The relationship was not linear and the lowest hemoglobin level was predicted at 34.8 weeks of pregnancy (Figure 15).

Hematocrit. The same variables which related significantly to the subjects' hemoglobin level were shown to be related to the hematocrit and explained 8.1% of the variation in hematocrit (Table 38). Income had a significant nonlinear relationship to the subjects' hematocrit level and the highest level was predicted at the household income level of 53,090 Rials per month (Figure 14). Weeks of pregnancy was significantly related to the hematocrit but the relationship was inverse and nonlinear, predicting the minimum hematocrit level at 32.3 weeks of pregnancy (Figure 15).

Serum Iron. The only significant variable shown to be related to the subjects' serum iron level was the weeks of pregnancy and its square which explained 17.4% of the variation in serum iron level (Table 38). Weeks of pregnancy had a nonlinear and inverse relationship to the serum iron level and the lowest level was predicted at 36.6 weeks (Figure 16).

Figure 14. Effect of household income on hemoglobin concentration and hematocrit level of pregnant subjects.

Regression Equations:

$$Y_{\text{Hgb}} = 12.1 + 4.17 \times 10^{-4} X_I - 3.46 \times 10^{-8} X_I^2 \quad R^2 = 3.4\%$$

$$Y_{\text{Hct}} = 36.3 + 1.12 \times 10^{-3} X_I - 0.106 \times 10^{-6} X_I^2 \quad R^2 = 2.4\%$$

where X_I = Household Income

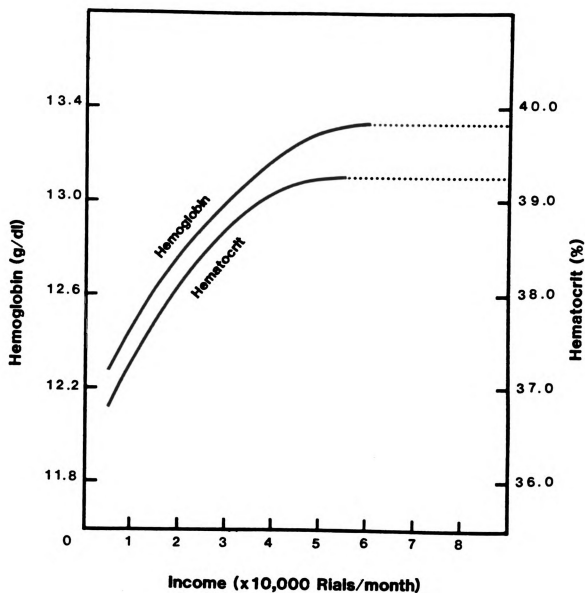


Figure 15. Effect of stage of pregnancy on hemoglobin concentration and hematocrit level of pregnant subjects.

Regression Equations:

$$Y_{\text{Hgb}} = 15.6 - 0.169 X_{\text{wp}} + 0.00243 X_{\text{wp}}^2 \quad R^2 = 7.6\%$$

$$Y_{\text{Hct}} = 45.6 - 0.481 X_{\text{wp}} + 0.00744 X_{\text{wp}}^2 \quad R^2 = 6.2\%$$

where X_{wp} = weeks of pregnancy

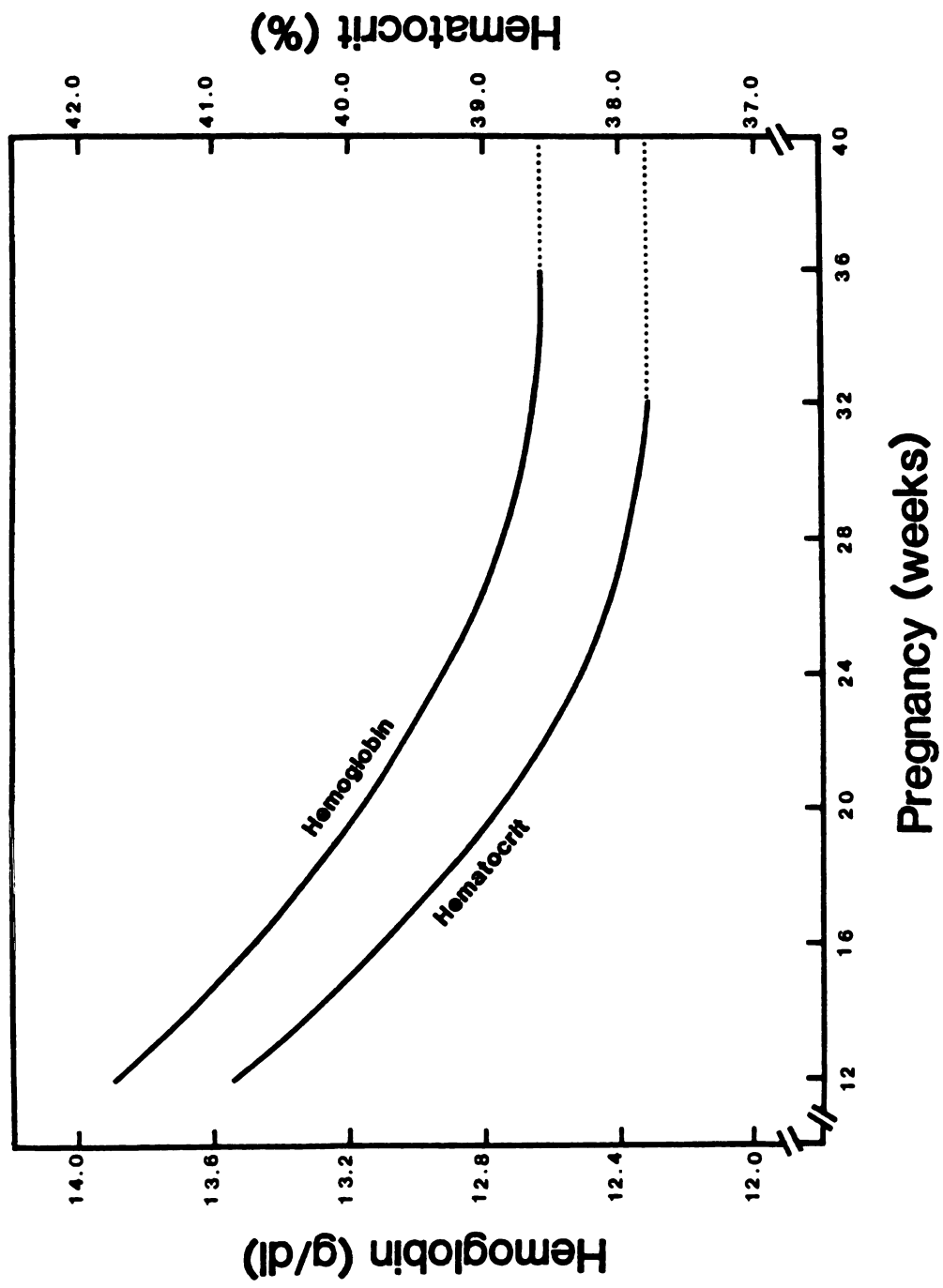


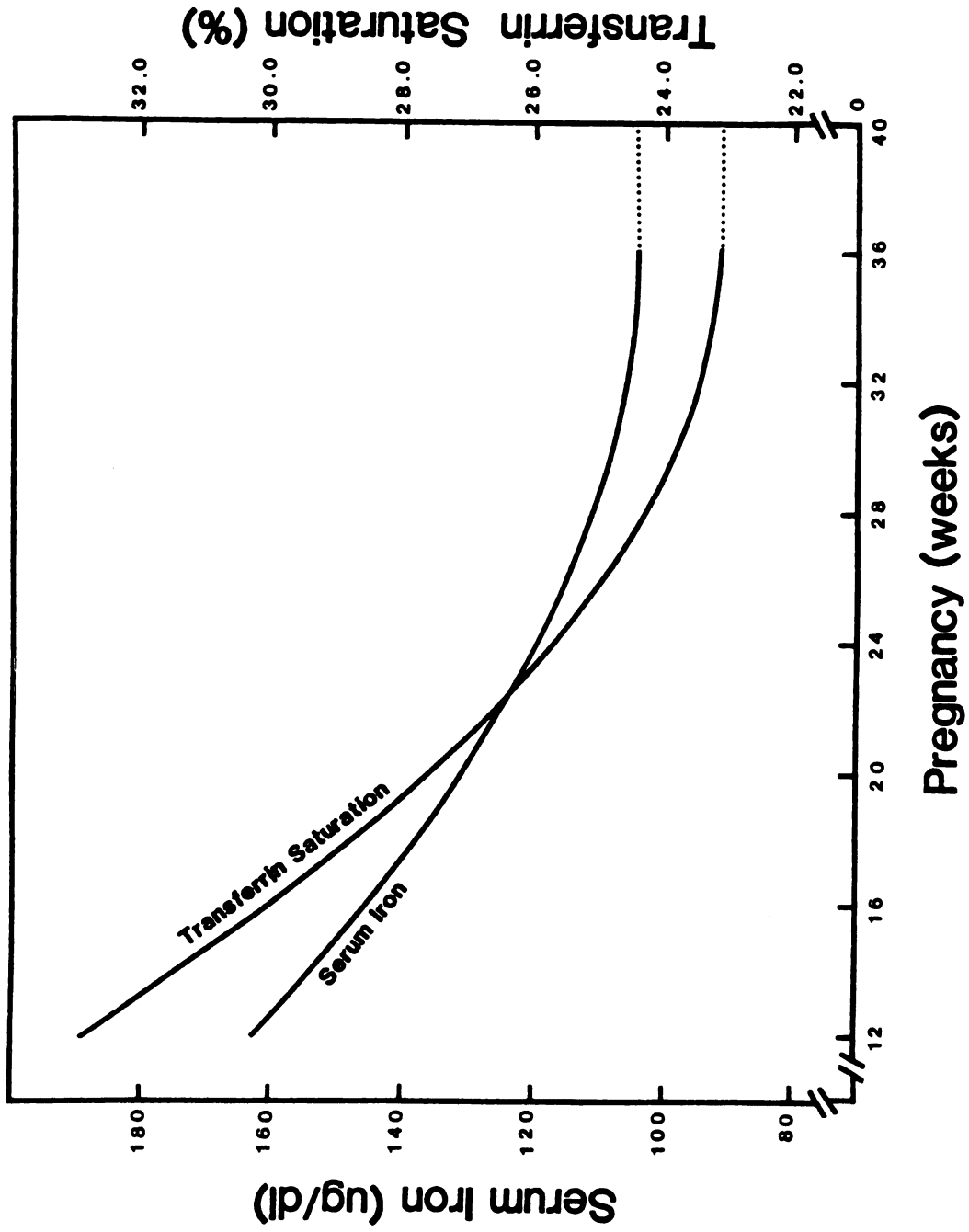
Figure 16. Effect of stage of pregnancy on serum iron concentration and percent transferrin saturation of pregnant subjects.

Regression Equations:

$$Y_{SFe} = 234.1 - 7.10 X_{wp} + 0.0969 X_{wp}^2 \quad R^2 = 17.4\%$$

$$Y_{TS} = 44.9 - 1.19 X_{wp} + 0.016 X_{wp}^2 \quad R^2 = 15.7\%$$

where X_{wp} = Weeks of pregnancy



Transferrin saturation. Similar to serum iron, the only independent variable with significant relationship to the subject's percent transferrin saturation was her weeks of pregnancy and its square which explained 15.7% of the variation in percent transferrin saturation (Table 38). Weeks of pregnancy had an inverse and nonlinear relationship with the subject's transferrin saturation and its minimum level was predicted at 36.4 weeks of pregnancy (Figure 16).

Hematological Status of the Non-pregnant Population

Mean, standard deviation and range of values for the constituents analyzed in blood of 21 non-pregnant subjects with middle-high to high income levels are shown in Table 39. The mean RBC, hemoglobin, hematocrit, MCV, MCH and transferrin saturation in this population were higher than the means in the pregnant population of all income groups (Table 34). The mean MCHC of the non-pregnant subjects was higher than that of the subjects in the VL and the L0 income groups and equal to that of the ML and M-H income groups. The mean serum iron level in the non-pregnant population was higher than that in the VL and the L0 and lower than that in the ML and the M-H income groups.

Table 39. Mean of values for constituents analyzed in blood of the non-pregnant subjects.^a

Constituent	Mean±SD (n=21)	Range
RBC ($\times 10^{12}/L$)	4.97±0.43	(4.39-6.17)
Hemoglobin (g/dl)	14.6±0.73	(13.4-16.4)
Hematocrit (%)	43.1±2.09	(39.5-47.0)
MCV (fl)	87.3±6.67	(69.0-96.0)
MCH (Pg)	29.6±2.47	(22.6-33.2)
MCHC (g/dl)	34.0±0.90	(32.4-35.9)
Serum iron ($\mu g/dl$)	124.3±32.2	(73.0-181.0)
TIBC ($\mu g/dl$)	431.6±51.0	(368-543)
Transferrin saturation (%)	28.8±6.76	(18.4-43.5)

^aMean age was 27.6±4.7 years.

However, the mean TIBC in the pregnant subjects of all income groups was higher than the TIBC of the nonpregnant subjects.

DISCUSSION

The course and outcome of pregnancy are influenced by a woman's preconceptional nutritional status as well as her diet during pregnancy. Increased nutritional needs during pregnancy have led researchers to assess the nutritional status of pregnant women as a section of the population at high risk of developing nutritional deficiencies. Augmented erythropoiesis during pregnancy increases the woman's demand for two critical nutrients, iron and folic acid (1). In many populations, pregnant women are prone to develop iron and folate deficiencies (20, 26, 87, 91, 96) mainly because their high demand for these two nutrients is not met by an increase in dietary intake.

In Central Iran, folate deficiency was shown to be an uncommon disorder even among pregnant women in both urban (Shiraz) and rural areas (92). Iron deficiency anemia, however, was frequently reported in a study of nonpregnant rural women in Central Iran (40). Pregnant women were excluded in that study. Considering the increase in iron requirement during pregnancy and low availability of iron for absorption due to high fiber and phytate intake in the rural diet (40), iron deficiency is likely to be a nutritional problem among the rural pregnant women in central

Iran. Since 57% of the population in Fars province lived in rural areas (103), the need for and importance of assessing the iron status of pregnant women in rural Iran was realized by the researcher. However, due to problems with physical accessibility to and social climate of the rural areas and also lack of required facilities and funds, it was impossible to use the rural pregnant population.

A high incidence of anemia among the low income pregnant women of Tehran (95, 96) indicated problems in the urban population and led the researcher to study the iron nutriture and hematological status of low income urban pregnant women in Central Iran and compare them with middle and high income pregnant women of the same region. Shiraz was chosen as a feasible locale because of its social and physical accessibility and because the required facilities and funds to conduct the study were available.

Geissler et al. (95) compared the nutritional and hematological status of pregnant women in two hospitals in Iran. They classified subjects as low or middle socioeconomic status only on the basis of hospital. It is likely that the populations were more heterogeneous than was assumed (95).

Of the two prenatal clinics selected in Shiraz, the Helal clinic was assumed to serve the low income population and the Hafez clinic to serve the middle income population. However, due to the overlap of the household income levels

of subjects in these two clinics, all results were analyzed according to household income as well.

The majority of the subjects in the Helal clinic did not know the exact monthly income of their husband, and gave an estimated average or range of his daily, weekly or monthly income. Lack of accurate information about the husband's income seemed to be mainly because husbands did not have a steady job or the remuneration varied from day to day. Some subjects reported that their husband did not wish to share income information with them. In contrast, most subjects in the Hafez clinic knew their husband's income because the majority of them were government employees with steady jobs and salaries and also because the husbands usually shared income information with them.

Although household income is an important determinant of the food consumption of a family, it is not the income per se, but the food purchasing power of the family and number of people living in the household which affect the food intake of family members. Determination of per capita income by dividing household income by size i.e. assuming even distribution of income between household members is open to question since the living expenses of children and adults might be considerably different. Therefore, in this study, the mean daily consumption of different food items and dietary iron by the subjects are presented by classifying the subjects into different income groups based on their

estimated household monthly income. For the statistical analysis, however, multiple regression was used so as to analyze the effect of household income as well as household size and other independent variables on food and iron intake and hematological status (dependent variables) of the pregnant subjects.

Food Pattern and Consumption

Variables assumed to affect the food and iron intake of pregnant subjects were the household income and size and season (Figure 17). The linearities as well as nonlinearities and interactions among these variables related to the food and iron intake were included in regression models and analyzed to determine the statistically significant variables affecting the subjects' consumption of major food items and iron (Appendix D-1).

All subjects generally followed the traditional Iranian food pattern although differences in food preferences and consumption were evident. The main meals of the day were the midday and evening meals while breakfast was generally light. Breakfast consisted of tea with bread alone or accompanied by butter and jam, white cheese or eggs and milk occasionally. The dishes served for lunch were usually heavier than those for dinner but they could be interchanged. In general if one meal included rice, the other did not. If the meal did not include rice, more or less

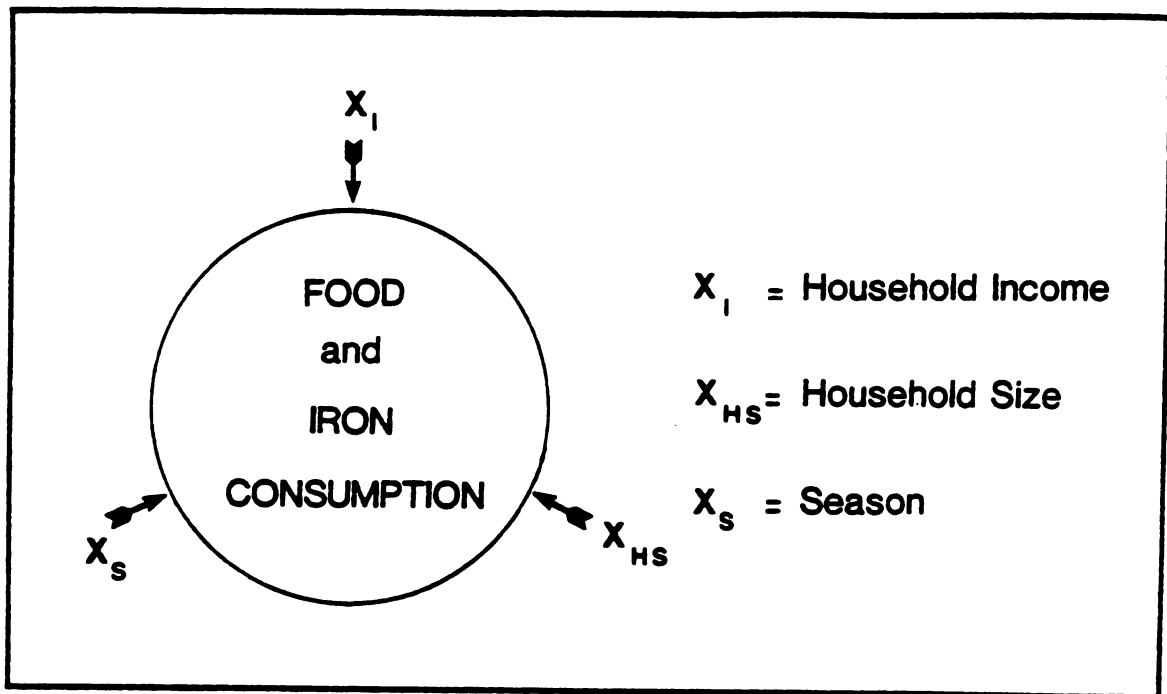


Figure 17. Causal model developed to assess relationships between independent variables (household income, household size and season) affecting food and iron intake (dependent variables) of pregnant subjects.

bread was consumed depending on the affluence of the family. Snacks were usually fruits and sometimes vegetables like lettuce which might be consumed every day in the higher income families and rarely or not at all in the lower income families.

The results of food consumption and dietary iron intake will be discussed with the emphasis on intake relative to income.

Bread

Bread is an important dietary item in the entire Iranian population. It is the staple food of low income families and supplies 50 to 70% of the energy intake of the villagers in central Iran (97). Tanok, the unleavened paperish bread made from whole wheat flour of nearly 100 percent extraction, is used by villagers around Shiraz and by certain sections of low income families in the city. In the pregnant population studied, 31.8% of the VL income group consumed the village bread, Tanok, while only 1.9% of the M-H income group ate this bread (Table 14). More frequent consumption of Tanok bread by the VL and the LO income groups may be because the majority of the subjects in these two income groups have migrated from the villages and still follow their traditional food pattern. It may be that the homemade village bread costs less than the city breads and is, therefore, more economical for the low income

families. The village bread is made at home while the three kinds of city breads are prepared in small commercial bakeries in Shiraz. The majority of the subjects consumed at least two different kinds of breads. In all income groups, Bazari and Sangak breads were more frequently consumed than the Mashhady bread (Table 14). Bazari bread was more often consumed as family monthly income increased; for example, in the M-H income group, 75.0% of the subjects consumed this bread compared to 31.8% of the subjects in the VL income group. A factor that may influence the family's choice of bread besides the taste preferences is the distance of the bakeries from the residence. Bakeries are located more in the central and south part of the city and the higher income families residing in the northern part of the city may prefer to buy bread that is sold in the nearest bakery or a kind of bread which does not become stale as quickly, such as Bazari bread. Fewer bakeries made Mashhady bread than the other two city breads. This is probably one of the major reasons why this bread was used by less than 10% of the households in all income groups. Mashhady bread is a saltier bread so taste preferences may also have been a factor affecting the bread choice.

The mean daily intake of bread increased as the family income level increased (Table 15) and the difference of 101 grams between the M-H and the VL income groups was statistically significant. Consumption of other food items

generally followed an opposite trend, i.e. higher consumption in higher income families. These results explain that foods are generally accompanied with more bread in the lower income families than in the higher income families. Bread is the most economic source of calories which may partly explain the reason for its higher consumption in the low income families with limited food budgets. The household income level showed a significant inverse and linear relationship to the bread consumption of pregnant subjects (Figure 3). Although household income was shown to be a significant variable affecting the subject's consumption of bread, it was not a strong predictor of bread consumption since it explained only 2.9% of the variation in bread intake. Bread is a popular food which might be consumed to considerable amounts by the middle or high income families which are economically able to purchase the more expensive foods like rice and meat. Therefore, different food preferences among the families of similar income level might be considered as one of the factors explaining the variation in the consumption of bread as well as other food items by the pregnant subjects.

The mean daily bread consumption by the subjects selected from the Hafez clinic was 345 g and it was 405 g for those in the Helal clinic. These amounts were similar to the reported bread consumption by low income pregnant subjects selected from a public maternity clinic in Tehran

(96). However, Geissler et al. (95) using subjects from two prenatal clinics in Tehran, reported lower bread consumption, a mean daily bread intake of 300 g by the pregnant women of low socioeconomic status and an intake of 230 g by those of middle socioeconomic status. Bread was usually bought daily and in some families, it was purchased two or three times a day. Fresh warm bread is preferred to day-old stale bread which may be discarded. Since no adjustments could be made for leftover bread that was discarded, the mean daily consumption of bread by some subjects might have been slightly overestimated. However, the researcher's experience suggests that little bread is usually wasted.

Rice

Rice is a popular and widely consumed food item in Iran. It is usually cooked plain and eaten with one kind of stew or is mixed with different food items like legumes, green herbs, green beans, tomatoes or cabbage. This mixture is usually eaten with pieces of meat or chicken.

The intake of rice decreased linearly as the household income decreased and household size increased (Figures 3 and 4). The household income and size, however, explained only 5.7% of the variation in rice consumption indicating that other factors affected rice intake. In the M-H income group, some subjects consumed rice every day while some

others consumed it less frequently although it could be afforded daily. This suggests that there are different food preferences among the families within the same income range. Mean daily intake of rice by pregnant women of low and middle socioeconomic status in Tehran reported in 1978 (95) were higher than that of pregnant women of Shiraz in this study. This difference may be due to different food patterns between the residents of these two cities, the higher price of rice in 1980 or to the different method of dietary assessment used in the Tehran and Shiraz studies, i.e., 24 hour recall versus weekly household food consumption.

Meat

The major kinds of meats consumed in Iran are lamb, veal and beef. Lamb is the most expensive and is preferred to veal and beef. Pork, if available, might be consumed by the non-Moslem minorities. Meat is prepared and consumed in different ways. It is the central item of stews, which are frequently consumed. The amount of meat consumed in stews may vary from a generous serving to a few pieces or none depending on the affluence of the family. Stew meat is usually accompanied by a vegetable like eggplant, zucchini squash, tomatoes, celery or okra or by green leaves and herbs mixed with beans or by other vegetables. Many different stews were consumed by families of low to high

income levels but the proportion of meat to the vegetable or legume as reported by the subjects was lower in the lower income families. Meat is also the main ingredient of the most widely consumed soup "abgusht" (water-meat), a national dish of Iran. Its ingredients are meat, white beans, chick peas, potatoes, tomatoes, spices and dried limes. "Abgusht" was reported as a popular food among all income groups but it was more frequently consumed by the lower income families. The proportion of meat to other ingredients was lower in the low income families compared to the higher income families according to subjects' reports. Besides stews and "abgusht", meat might be consumed roasted in pieces called "kabob" which are eaten with plain rice. Meat was eaten more often by the higher income families due to its high cost. Ground meat might also be consumed as roasted patties and be eaten with rice or bread or it might be mixed with potatoes or chick pea flour, egg and onion and fried in small patties.

Regression analysis showed a significant relationship between the meat consumption and the household income, size and season which explained 24.4% of the variation on consumption (Table 17). As household income increased from very low to higher levels, meat consumption also increased but the relationship was not linear. The effect of income on increasing the meat consumption diminished at the higher income levels. When the income level increased from 10,000

to 40,000 Rials per month, the mean daily meat consumption was predicted to increase by 50.3 g. When the household income increased from 40,000 to 70,000 Rials, however, the predicted increase in meat consumption was only 31.6 g daily. Beyond a certain level of income (105,523 Rials per month), the increase in income had no further effect on increasing the meat intake (Figure 5). Although the consumption of meat had a significant inverse and linear relationship with the household size (Figure 6), income was a more important predictor of the meat intake as it explained 20.3% of the variation in its intake compared to household size which explained 3.8% of the variation. Results of regression analysis were in agreement with the differences in the mean daily meat consumption in different income groups. Meat consumption by the subjects of the M-H income group was similar to that of the ML income group while the LO and the VL income groups had significantly lower intakes of meat (Table 15). In the high income families, the leaner and more expensive cuts of meats might be consumed rather than a higher amount of meat compared to the middle income families. In the low income families, meats with a higher amount of fat which cost less and which provide more energy relative to cost are consumed. Therefore, the mean daily consumption of meat as reported by the subjects in the VL and the LO income groups might have been slightly overestimated since they reported the weight of

meat as purchased which may have included a higher amount of fat than the meats purchased by the higher income families.

Meat consumption was shown to be related to the season. Lower consumption of meat during the spring and summer might be due to a higher consumption of vegetables which are plentiful and inexpensive during the warm seasons. Season, however, was not a strong predictor of the meat consumption since it explained only 1.8% of the variation in consumption (Figure 7).

Poultry

Chicken is usually served with rice. It may also be accompanied by different vegetables like potatoes or carrots and be served with bread. Generally, in Shiraz, meat was preferred to chicken. Chicken was less frequently consumed than meat by the subjects of all income groups. Although the cost of chicken was less than that of meat, low income families could not afford frequent consumption of chicken. The mean daily consumption of chicken by the subjects of the VL income group was less than one third of that of the subjects of the M-H income group. Poultry consumption increased as household income increased but the relationship was not linear and the maximum consumption of chicken was predicted at the family monthly income level of 82,110 Rials (Figure 5). Poultry consumption, however, was less

influenced by household income level than the meat consumption. Income explained 10.5% of the variation in poultry consumption and 20.3% of the variation in meat consumption. Therefore, income was a better predictor of the meat consumption than the poultry consumption. This may partly be due to lower cost of poultry than that of meat. Household size was shown to have a significant inverse and linear relationship with poultry consumption indicating lower intakes by the subjects living in bigger households (Figure 6). Income, however, accounted for higher percentage of variation in poultry consumption than household size.

Legumes

Legumes are one of the relatively cheap sources of calories and proteins that are widely used in Iran by different income groups in soups and stews as well as in rice dishes. Different kinds of thick soups called "ash" are made of beans, peas or lentils with greens and herbs. The mean daily consumption of legumes by subjects of the VL income group was lower than the other income group but was not statistically different. In the Geissler et al. study in Tehran (95), pregnant women of middle socioeconomic status consumed more beans than those of low socioeconomic status. There was no significant relationship between legume consumption and household income while household size showed an inverse linear relationship with the

consumption of legumes (Figure 8). Although most pregnant subjects reported a higher consumption of legumes during winter than summer, no relationship was found between their consumption and season.

Eggs

Eggs are consumed as an entree at breakfast or occasionally as an evening meal or mixed with vegetables like potatoes or greens and herbs. Although the mean daily consumption of eggs by the subjects of different income groups showed a higher intake by those of a higher income group, the regression analysis did not indicate any significant relationship between egg consumption and household income or size. Eggs are not an expensive item in Iran and, therefore, consumption is not often limited to household income as is the case for the meat and poultry consumption. However, in the very low income families, eggs might be consumed in a limited amount. Fewer eggs were consumed during the warm seasons. Abundance of vegetables during spring and summer might have been the cause for the decrease in egg intake. Eggs are generally regarded as a "hot" food by some Iranians. This may partly explain their lower consumption during the warm and hot seasons. Lower availability of eggs in the market during spring and summer of 1980 may also be regarded as another factor contributing to lower consumption of eggs during

part of the research period. Season, however, was a weak predictor of egg consumption as it explained only 1.7% of the variation in intake. The greatest difference in egg intake between the cold and hot weather was predicted to be only 7.8 g per day or a difference of approximately one egg per week (Figure 9).

Greens and Herbs

In Iran, greens and herbs are consumed fresh, either raw or cooked. They are rarely consumed in the dried state or used for garnishing dishes as in U.S.A. Green onions, radishes, tarragon, basil and mint are served raw along with the main dish. They might be consumed every day or less frequently depending on the income and food preferences of the family. Fresh greens and herbs including parsely, leek, dill, fenugreek, mint, spinach and coriander are cooked in stews, or "ash" or are mixed with rice. All kinds of greens and herbs are available and are plentiful during all seasons of the year. Thus it was not surprising that their consumption was not related to the time of the year (Table 17). Intake of greens and herbs was significantly related to household monthly income which explained 7.5% of the variation in their consumption. The relationship was not linear and the maximum consumption was predicted at the monthly income of 61,100 Rials (Figure 10). Household size, however, was linearly related to greens and herbs

consumption showing a lower intake by the subjects living in households with a higher number of people (Figure 8). Although greens and herbs are not an expensive food item, their consumption by the low income families could be limited because it is preferable to spend the limited food budget primarily on items which provide more energy and are more filling than the items that are served as a sidedish.

Vegetables

A variety of vegetables is used in Iranian dishes. In Shiraz, most of the vegetables are available only during one or two seasons or are more abundant in one season relative to others. Vegetables like tomatoes, eggplants, zucchini squash or green beans are grown mainly during the spring and summer and are more plentiful and cheaper during these seasons. Some vegetables like beets, parsnips and lettuce are on the market mainly during the cold seasons. Vegetables like potatoes and carrots are available all through the year. The kind of vegetables used by the pregnant subjects varied widely depending on the time of the year. Subjects could give more accurate estimates of their household consumption of the vegetables which were available in the season when they were interviewed. Lettuce is an example of a vegetable that was available during all seasons but more plentiful during the fall and winter. Most of the subjects could give an estimate of their household

consumption of this vegetable. Therefore, lettuce was chosen as an example of a vegetable eaten by this population frequently. Lettuce is used in salads or dipped in vinegar and eaten as a snack which might be accompanied by bread. Its consumption was significantly related to season which explained 5.6% of the variation in its intake (Figure 11). Lettuce consumption was also significantly related to the household income and the interaction between the household income and size (Table 17). The subjects' consumption of lettuce increased as the household income increased (Figure 10) and income explained 8.9% of the variation in lettuce consumption. Although household size was not significantly related to the lettuce consumption, it had to be included in the final regression equation due to its significant interaction with household income which was shown to be an important factor influencing the lettuce intake (Table 17).

A similar pattern to lettuce consumption might be postulated for most other vegetables consumed by the subjects, predicting a lower consumption by the subjects living in the households with lower income and larger size and higher consumption during the seasons when the vegetable is abundant and inexpensive. Although most of the vegetables cost much less than food items like meats and poultry, their consumption is expected to be lower by the subjects in low income families with a limited food budget than by those in higher income families.

Fruits

Fruits are consumed after meals or as a snack between meals and the kind of fruit used varies widely depending on the time of the year. Pomegranates and quince are inexpensive in fall and citrus fruits are available mostly during the fall and winter. Persian melons and watermelons are grown during the summer and cost less than other summer fruits like grapes, cherries, nectarines and apricots which are consumed mainly by the middle and high income families. Cucumber is regarded as a fruit in Iran and was widely consumed in summer because it is popular and inexpensive. Limes, a good source of vitamin C, are plentiful and inexpensive during the summer and lime juice is widely used as a seasoning or is consumed as limeade, both very popular in Shiraz. Since lime is not considered as a fruit in Iran, subjects did not report consumption of limes when asked for their fruit consumption.

All of the fruits in season were more frequently consumed by the subjects of the M-H and the ML income groups than those in the L0 and the VL income groups. There were 23.3% of the subjects in the VL and 8.7% of those in the L0 income groups who did not consume fruits at all probably because they could not afford fruits.

Iron Intake

The subjects' mean daily intake of iron from the major food sources was determined by calculating the amount of iron in the food items consumed by the subjects (obtained from dietary recall) using information in standard food consumption tables.

The average total daily iron intakes of pregnant subjects in different income groups were similar and averaged 39.58 mg in the VL, 39.88 mg in the LO, 43.32 mg in the ML and 40.23 mg in the M-H income groups. Therefore, the average daily iron intake of all income groups was in excess of the 28 mg of iron per day recommended by FAO/WHO during pregnancy in populations where less than 10% of caloric intake is from animal foods (27). In the VL income group 78.8% of the subjects had a daily iron intake above 24 mg compared to 89.9% of the LO, 90.0% of the ML and 88.9% of the M-H income group. However, the total iron content of the diet is a relatively poor indicator of the adequacy of dietary iron. For example, although iron intakes of 18 village boys 13 to 14 years old in central Iran averaged 44.4 mg per day which exceeded the estimated requirements recommended by FAO/WHO (27), iron deficiency anemia was prevalent, i.e. 30% of the children had a hemoglobin concentration of 12 g/dl or less. The important determinant of the adequacy of dietary iron is its availability for

absorption. Iron absorption from foods depends not only upon the amount of iron supplied, but by the nature of that iron and the composition of the meal in which it is consumed (42, 43). Heme iron which comes from animal tissues is absorbed to a considerably higher degree than nonheme iron which comes primarily from cereals, legumes, eggs, vegetables and fruits as well as iron supplements (44). The absorption of nonheme iron, however, might be increased in the presence of enhancing factors like animal flesh (43, 45-48) and vitamin C (44, 49, 57, 58) or decreased when the factors inhibiting iron absorption like fiber and phytate (40, 80-82) are present. The prevalence of anemia among the villagers around Shiraz was explained to be due to high intakes of phytate from the unleavened whole wheat village bread which might have interfered with nonheme iron absorption although bread was the main source of iron in the village diet (40). Low iron availability from the village diet might have also been due to high contents of fiber in bread which has been shown to inhibit the iron absorption (81).

Bread was the major source of iron in the diet of pregnant subjects in all income groups. In the VL income group, iron from bread made up 82.3% of the average total daily iron intake of the subjects while in the L0, the ML and the M-H income groups, 68.2, 61.7 and 59.0% of the mean daily iron intake was derived from bread (Table 23). Thus, bread

provided a lower proportion of the total iron intake as household income increased. In the VL and the L0 income groups, the mean daily intake of iron provided only by bread (Table 21) exceeded the 28 mg per day recommended during pregnancy by the FAO/WHO (27).

Tanok, the unleavened village bread made from whole wheat flour of 100% extraction was consumed more frequently by subjects in the lower income families than by those in the higher income families (Table 14). This village bread has a higher fiber and phytate content than the city breads since the latter are made from wheat flours of lower extraction rate (97) and the leavening used in city breads partially destroys the phytate (106). Higher intakes of fiber and phytate through a higher consumption of the village bread by the VL and the L0 income groups might be responsible for lower availability of nonheme iron for absorption in the diet of the subjects from lower income families.

Besides fiber and phytate, tea has been shown to interfere with the absorption of nonheme iron (68-70). Nonherbal tea was widely consumed by the subjects at breakfast, after meals, or between meals. Thus, tea consumption may have also contributed to decreased iron absorption.

Rice, legumes, eggs, greens and herbs and lettuce were the other sources of nonheme iron in the diet besides breads for which the average daily consumption could be estimated.

Among these nonheme sources of dietary iron, greens and herbs provided higher amounts of iron in the daily diet of all income groups than rice, legumes, eggs or lettuce (Table 21). In the M-H income group, lettuce was also an important source of dietary iron as it provided an average of 2.25 mg of iron per day. The mean daily intake of iron provided by greens and herbs and lettuce was 7.05 mg in the M-H income group which was more than twice of the amount (2.90 mg) provided by these sources in the diet of the VL income group. The iron intakes from other vegetables and fruits could not be estimated due to the lack of accurate information about their average daily consumption by the subjects. Nonheme iron provided 96.3% of the average total iron intake of the subjects in the VL income group compared to 90.7% in the L0, 86.0% in the ML and 84.3% in the diet of the M-H income groups. Therefore, with increased family income, nonheme iron formed a lower proportion and heme iron formed a higher proportion of the total dietary iron intake.

Significant relationships were found between heme iron intake and household income and size which explained 27.5% of the variation in its intake (Table 17). With increase in household income from low to higher levels heme iron intake also increased, but beyond the monthly income of 102,270 Rials there was no further increase in the heme iron intake (Figure 12). Intake of heme iron decreased in a linear manner as the household size increased (Figure 13).

Income, however, was a more important indicator of heme iron intake than the household size as the former explained 24.0% of the variation and the latter explained 5.0% of the variation in its intake. In the M-H income group, heme iron formed 15.2% of the average total daily iron intake. This ratio decreased in lower income groups and in the VL income group only 3.7% of the total iron was derived from heme sources. While 84.0% of the subjects in the M-H income group consumed more than 3.0 mg of heme iron per day, there were only 21.3% of the subjects in the VL income group who had a daily heme iron intake above 3 mg.

Heme iron present in animal flesh is absorbed considerably better than the nonheme iron (44). Heme iron has been shown to enhance the absorption of nonheme iron from different sources (43, 45-48). Meat rather than poultry formed the major source of heme iron in pregnant subjects' diets. Although fish, shrimp and organ meats (especially liver) may contribute heme iron in the diet, they were not included in estimating the daily heme iron intake of the subjects because of the very low consumption of these foods in the diets of most subjects. These foods provided a negligible amount of dietary iron per day.

Ascorbic acid has also been shown to increase absorption of nonheme iron from foods (44, 49, 57, 58). Fruits are the major dietary source of vitamin C. A higher consumption of fruits by the subjects of M-H and the ML income groups than

those of L0 and the VL income groups provided more vitamin C and probably enhanced the availability of nonheme iron for absorption in these subjects to a further extent. Greens and herbs are also good sources of vitamin C and their higher consumption in the higher income families may contribute to a higher iron availability for absorption.

In summary, the average total daily intake of iron was similar between income groups and ranged from 39.6 to 43.3 mg. However, the higher consumption of meat, poultry and fruit by subjects of M-H and ML income group provided higher intakes of heme iron and vitamin C. These are expected to increase the availability of dietary nonheme iron for absorption (43-49, 57, 58). Therefore, the availability of iron for absorption was likely to be higher in the diet of subjects from higher income families than those from lower income families.

To obtain food consumption and dietary iron intake of pregnant subjects, household weekly consumption of major food items was divided by household size which was adjusted for age assuming uneven distribution of foods between children and adults (Appendix C-3). However, no adjustments were made for sex and males and females were assumed to consume the same amount. This may have resulted in an overestimation of food and iron intake by pregnant subjects since the husband or other adult males living in the household are expected to consume more foods to meet their energy

requirements than the pregnant subject. In the VL and LO income families where the husband's occupation is mainly unskilled jobs with heavy activity like construction work, the higher energy requirements of the husband could increase the discrepancy in the food consumption of the husband and wife even more. Besides the higher energy needs and food intakes of the adult males, cultural factors may have influenced the proportion of food eaten by the husband and wife. It is customary in Iran at meal times that foods go first to the husband and then to the wife. This pattern is seen in the low socioeconomic population especially. In low income families where the amount of food is limited, the wife may not eat enough to satisfy her appetite since there is not sufficient food for family members and food for her husband takes top priority. In the middle and high income families, however, food is in sufficient quantity so all may eat as much as they desire. Therefore, the subjects' consumption of major food items and dietary iron may have been overestimated. Any overestimation seems to be more probable in the lower income groups.

Although adjustments were made to differentiate the energy requirements and food consumption of children and adults, all household members above 19 years old were assumed to consume the same amount of food. Thus, no adjustments were made for low food consumption by the elderly. This may have caused an underestimation of food

consumption by the pregnant subjects in households which include elderly persons.

Although using the household weekly food consumption had the disadvantage of possible overestimation (or in some cases underestimation) of the subject's food intake, it was considered to be the best and most feasible method of dietary recall for this population. A majority of the subjects could give precise estimates of the amount of major food items (especially bread, rice, meat, poultry, legumes and eggs) consumed by the household since they were purchased daily or measured to follow recipes. Most Iranian dishes are mixtures of several foods and it is impossible to estimate the amount of each food consumed by an individual. For the first 80 subjects, both 24-hour recall and the household weekly food consumption were used to get dietary intake information. The 24-hour recall was discontinued after these 80 subjects due to its lack of feasibility and reliability in eliciting information about the subject's food intake.

Iron intake of six subjects (2 in the VL and 4 in the L0 income groups from the Helal clinic) as analyzed and calculated from collected food samples showed that the total iron intake of the subjects during a 24-hour period averaged 32.37 mg. Bread was the major source (76.4%) of total iron in the diet of the six subjects. Although six food samples were not enough to give a precise estimate of the daily

iron intake of this pregnant population, the agreement with the iron intakes of subjects as calculated from the household food consumption data supports the appropriateness of this method of dietary recall in estimating the food consumption and iron intake of the subjects.

Hematological Status

To assess the hematological status of pregnant women, all independent variables hypothesized to affect hemoglobin, hematocrit, serum iron and transferrin saturation were included in regression models (Figure 18). All linearities, nonlinearities and interactions between the independent variables were included in the first models (Appendix D-2) and the final regression models included the statistically significant variables related to hematological status of the subjects.

Results of regression analysis showed that among all variables, household income and weeks of pregnancy had statistically significant relationships with hemoglobin and hematocrit level. With the increase in household income from very low to higher levels, hemoglobin and hematocrit levels were also found to increase, but the relationships were not linear indicating no further increase in their level beyond a certain household income (Figure 14). Income, however, was not a strong predictor of the hemoglobin

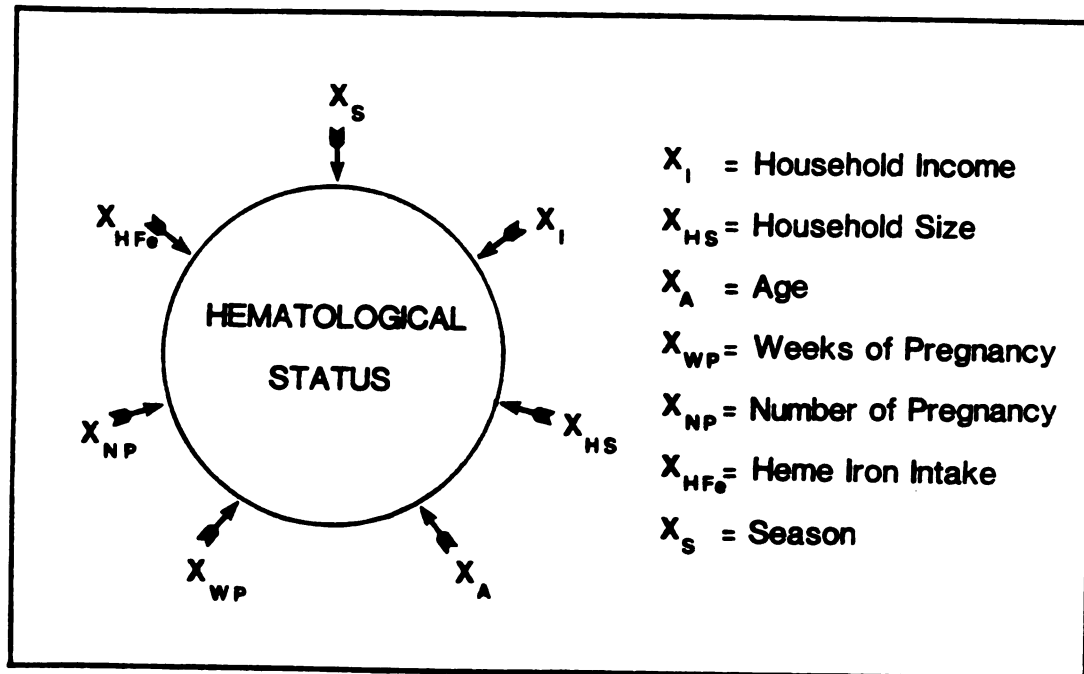


Figure 18. Causal model developed to assess relationships between independent variables (household income, household size, subject's age, weeks of pregnancy, number of pregnancies, heme iron intake and season) affecting the hematological status (dependent variables: hemoglobin, hematocrit, serum iron and transferrin saturation levels).

and hematocrit levels as it explained only 3.4% of the variation in hemoglobin and 2.4% of the variation in hematocrit levels.

Iron status during pregnancy has been shown to be related to the economic background of the pregnant women which will affect the maternal nutrition (26, 95, 96). Foroozani et al. (96) reported lower mean hemoglobin, serum iron and percent transferrin saturation in the subjects selected from a public maternity clinic than those from a private maternity clinic in Tehran (96).

In a study by Geissler et al. (95) in Tehran, mean hemoglobin concentration of women selected from a low socioeconomic status population (LSE) was 10.2 g/dl during the last two weeks of pregnancy and 10.8 g/dl postpartum. Both were significantly lower than the mean hemoglobin values (12.3 g/dl prepartum and 13.3 g/dl postpartum) in subjects from a middle socioeconomic population (MSE). When results of Geissler et al. are compared with the hemoglobin concentration of Shiraz pregnant women in this study, it is seen that the mean hemoglobin levels in the pregnant subjects of the VL and the LO income groups in Shiraz were higher than the postpartum values reported for LSE and also from the prepartum values reported for the MSE group. Mean hemoglobin concentration in the subjects of the ML and the M-H income groups were comparable to the postpartum values observed in the MSE women in Tehran. Hemoglobin levels

below 10 g/dl were seen in 33% of the LSE subjects in Tehran before parturition and in 5% of them postpartum. In the MSE group, the corresponding percentages were 3% both before and after parturition. In Shiraz study, however, only 4.5% of the subjects in the VL income group had hemoglobin levels below 10 g/dl and all of them were beyond their 20th week of pregnancy. This percentage is comparable to the postpartum percentage reported in the LSE group of Tehran. Hemoglobin levels below 10 g/dl were seen in 2.8% of the LO, 1.2% of the ML and none of the subjects in the M-H income group. Therefore, the hematological status of the pregnant subjects with very low (VL) and low (LO) income levels in Shiraz were similar to that of the middle socioeconomic population and much better than that of the low socioeconomic population of pregnant women in Tehran (95). This difference might be due to lower income levels or lower food purchasing power in the LSE group living in Tehran compared to the VL and the LO income groups in Shiraz or due to higher intakes of iron by the Shiraz women. The mean daily intake of iron in the Tehran study was reported to be 18 mg in the MSE and 15 mg in the LSE group. Both were less than half of the total mean daily iron intake found among Shiraz women in this study.

When the mean values of the constituents analyzed in blood of the pregnant subjects (Table 34) are compared to those of the nonpregnant subjects (Table 39), it is shown

that the latter had higher levels of most blood constituents than the pregnant subjects of all income groups. These differences were more evident between the nonpregnant subjects and those in the VL and the L0 income groups. The nonpregnant group was selected as a reference to represent the hematological status of women from an economically able population. Income levels of this group were similar to those of the M-H income group, yet the latter showed lower mean values for the constituents analyzed in blood than the former. This difference is primarily due to the physiological changes and high demands for iron during pregnancy.

In addition to income, weeks of pregnancy had significant inverse relationship with the hemoglobin and hematocrit levels (Figure 15). The weeks of pregnancy explained 7.6 and 6.1% of the variation in hemoglobin and hematocrit respectively. It appears that stage of pregnancy was a better predictor of hemoglobin and hematocrit than income since the former accounted for a higher percentage of variance than the latter. Decline in hemoglobin and hematocrit as pregnancy proceeds was to be expected since one of the fundamental physiological adjustments during pregnancy is an increase in maternal blood volume which exceeds an increase in erythropoiesis (107). The increase in plasma volume begins in the first trimester, continues through the second and early third trimester, and then levels off during the last six to eight weeks of pregnancy (108). Therefore,

increase in plasma volume follows a gently sigmoid pattern and at maximum it reaches 1200 ml or approximately 50% above the nonpregnant state (107). The total erythrocyte volume, however, increases in a more linear pattern and at term reaches 20 to 30% above that of the nonpregnant state (109). By comparing these two changes, it becomes evident that the increase in erythrocyte mass is not sufficient to compensate for the expanded plasma volume and as a result, the red blood cell count, hemoglobin concentration and hematocrit decline, reaching the lowest level at 32 to 34 weeks of pregnancy (108). The state of decreased hemoglobin and hematocrit levels caused by the expanded plasma volume and hemodilution has been termed "physiological anemia" (30). A non-pregnant subject with hemoglobin levels of 13 or 14 g/dl may show a drop to as low as 10 or 11 g/dl during pregnancy, a level which would indicate anemia when non-pregnant but not in pregnancy. Erythrocytes remain normochromic and normocytic in the physiological anemia accompanying pregnancy (30). Although the general pattern of these changes is not particularly influenced by maternal iron status, the degree of change depends substantially on it (109).

The lowest hemoglobin concentration of pregnant subjects was predicted at 34.8 weeks and the lowest hematocrit level at 32.3 weeks of pregnancy (Figure 15). Declines in hemoglobin and hematocrit levels during the course of pregnancy

have been reported in several studies of women who were followed throughout their pregnancies (10, 110, 111). Although the present study was cross-sectional, lower mean hemoglobin and hematocrit levels could be shown in subjects who were in the middle (21-30 weeks) and late (31-40 weeks) stages of pregnancy than in those who were in the early (12-20 weeks) stage of pregnancy (Table 35).

Serum iron concentration and percent saturation of transferrin (serum iron/total iron binding capacity) have also been reported to be inversely related with the length of gestation in women not given supplemental iron (10, 111, 112); however, when iron supplements were given to pregnant women, serum iron concentration did not change significantly (10, 111, 112). Total iron binding capacity (TIBC) increased as pregnancy proceeded in both supplemented and nonsupplemented women, but the increase was greater in those women who did not receive iron supplements. Gerritsen and Walker (113) found increased TIBC in the blood of Bantu pregnant women who showed no reduction of either hemoglobin or serum iron. Thus, it appears that there is a definite effect on the levels of transferrin as a consequence of pregnancy and if iron stores are depleted, the TIBC is further increased.

In the present study, serum iron concentration and percent transferrin saturation had significant inverse and nonlinear relationships with the weeks of pregnancy

(Figure 16). Stage of pregnancy was shown to be a significant independent variable and an important predictor of the serum iron and percent transferrin saturation as it explained 17.4% of the variation in the former and 15.7% in the latter. Therefore, pregnant women who were in their later stages of pregnancy were at a higher risk of developing iron deficiency than those who were in the earlier stages of pregnancy. In the population studied, the proportion of subjects in different stages of pregnancy was not similar between income groups. While 49.3% of the subjects in the M-H and 30.5% of those in the ML income groups were in their early stage of pregnancy (12-20 weeks), there were only 19.4% of the VL and 23.2% of the L0 income groups who were at this stage of pregnancy. In contrast, in the VL and the L0 income groups, 37.3 and 34.8% of the subjects were in their late stages (31-40 weeks) of pregnancy respectively compared to 24.4% of the ML and only 11.6% of the M-H income groups (Table 8). Therefore, the lower mean hemoglobin, hematocrit, serum iron and percent transferrin saturation and the higher incidence of iron deficiency anemia (transferrin saturation below 15%) in the Helal subjects and in the VL and the L0 income groups were due in part to a higher percentage of the subjects from the lower income families (referring mostly to Helal clinic) who were in their later stages of pregnancy. Among 17 anemic subjects with transferrin

saturation below 15.0%, four subjects (23.4%) were in their 21-30th week and 13 (76.5%) were in 31-40th week of pregnancy. Stage of pregnancy, however, did not totally explain the higher frequency of anemia in the lower income subjects. For example, of 25 subjects in the VL group who were between the 31st to 40th week of pregnancy, four (16.0%) were anemic while in the ML income group none of the 20 subjects in the same stage of pregnancy were anemic.

Percent transferrin saturation level was selected as the most sensitive criteria for evaluation of the iron status (9) and pregnant women with levels below 15% were regarded as anemic or at high risk of being anemic according to FAO/WHO standards. According to this criteria, 6.8% of the subjects in the Helal clinic and 1.9% of those in the Hafez clinic had iron deficiency anemia (Table 32). When the pregnant subjects were compared on the basis of different household income ranges, 9.3% of the VL, 7.4% of the LO and 2.0% of the ML and the M-H income groups had percent transferrin saturation below 15. The incidence of iron deficiency anemia previously reported in pregnant women of Tehran was considerably higher than the incidence in this study in Shiraz. In the Tehran study, a transferrin saturation level below 15.0% was seen in 57.1% of the women in a public maternity clinic and in 26.7% of those in a private maternity clinic just before parturition (96). Lower food purchasing power due to higher costs of living

in the capital (Tehran) or a different pattern of food consumption by the pregnant population in Tehran may have resulted in lower iron intakes and/or lower availability of iron for absorption. Carr (9) reported that women whose transferrin saturation was less than 20% demonstrated a response to iron treatment with a significant increase in hemoglobin and a restitution of transferrin saturation greater than 20 percent. Therefore, transferrin saturation level below 20% during pregnancy was regarded by Carr (9) as an indication of iron deficiency. According to this criterion, 24.2% of the subjects in the VL, 26.9% in the L0, 9.8% in the ML and 10.1% in the M-H income group were iron deficient or at high risk of being deficient. Among these iron deficient subjects, 54.2% were in their 31-40th week, 37.3% were in 21-30th week and only 8.5% were in their 12-20th week of pregnancy. Of 88 subjects in the L0 and the VL income groups who were in their last two months of pregnancy, 30 (34.1%) were iron deficient while in 28 subjects of the ML and the M-H income groups, only two (7.1%) of the subjects in the same stage of pregnancy were iron deficient.

Although percent transferrin saturation is a sensitive indicator of iron deficiency, interpretation of low levels should be made with caution since depressed percent transferrin saturation and TIBC levels below 300 $\mu\text{g/dl}$ might be seen in conditions such as inflammatory disease

and chronic infection (11). In the Shiraz study, among the pregnant women with transferrin saturation below 20%, only five had TIBC less than 350 g/dl and these were between 327 and 335 g/dl. Therefore, inflammatory diseases or chronic infections do not seem to be the cause of depressed transferrin saturation in these subjects. Hookworm infestation has been reported as an important cause of anemia in different regions of the world (1, 3). Parasitic infestations are ubiquitous in rural Iran (114), but they are not of a type that would cause blood loss (40). Therefore, it may be assumed that blood loss due to parasitic infestations is not a cause of iron deficiency in the urban population studied.

A high incidence of folate deficiency has been reported in pregnant women both in developed and underdeveloped countries (20, 26, 81-91). In central Iran, however, folate deficiency appears to be a relatively uncommon disorder (92). Even among pregnant subjects in different stages of pregnancy selected from lower socioeconomic populations in urban (Shiraz) and rural (Marvdasht) areas, serum folate levels were normal. A probable explanation for the well maintained serum folate levels during pregnancy may be due to the diet which included large amounts of whole wheat bread or bread prepared from flours of high extraction rate which contain significantly higher folate contents than the white bread made from refined flour (93,

94). Folate deficiency, therefore, does not appear to be a nutritional problem among pregnant women studied in Shiraz.

In conclusion, iron deficiency anemia was not a serious nutritional problem during the first half of pregnancy in the subjects from low as well as high income families. However, as pregnancy proceeded, subjects were at higher risk of developing iron deficiency when iron supplements were not given. Although stage of pregnancy was the most important determinant of the hematological status of the pregnant subjects and there were a higher percentage of the low income subjects in their later stages of pregnancy, stage of pregnancy can only partly explain the higher incidence of anemia in the lower income subjects. Even within the same stage of pregnancy, the percentage of iron deficient subjects was higher among the lower income subjects. Higher consumption of heme iron and vitamin C by the more affluent subjects may in part be responsible for a higher availability of iron for absorption and better iron status compared to the low income subjects.

Since none of the subjects who were in their first half of pregnancy were anemic, it appears that women from all income levels wisely choose a variety of foods within the limitations of their food budget.

SUMMARY

Iron deficiency anemia is a world wide problem with higher prevalence in developing countries where it occurs in all age groups and especially during pregnancy when iron requirements are greatest (1-3). The objective of this study was to assess the food and iron consumption and hematological status of pregnant women from low income families and compare them with those of middle and high income families in the same urban area of central Iran. Shiraz was selected as the locale because pregnant populations and required facilities were available.

The methodological tools used were an interview schedule including information about socioeconomic and medical characteristics and food consumption of the subject and her family, measurement of height and weight and blood analysis. Foods consumed by some subjects during a 24-hour period were also collected to determine the daily iron intake.

To estimate the average amount of food consumed by the subjects, household consumption of the major food items obtained from dietary recall was divided by an adjusted household size developed to differentiate the energy requirement and food consumption of children and adults. The mean daily iron intake of subjects was calculated using standard

food composition tables used in Iran and U.S.A.

The independent variables hypothesized to influence the subjects' consumption of foods and dietary iron were the household income and size and season. Factors assumed to be related to the hematological status were the subject's household income and size, her age, stage of pregnancy, number of pregnancies, average daily intake of heme iron and season. Regression analysis (103) was used to identify the variable(s) which has (have) a statistically significant relationship to the food and iron consumption and hematological status of the subject. Linearities as well as nonlinearities and interactions between variables were entered into regression models and significant variables were determined. To identify the significant differences between mean daily food consumption and mean values for the analyzed blood constituents of the subjects in two clinics and in four different income groups, the Student's t-test and Bonferroni t-test were used respectively (103).

Four hundred subjects were selected from two prenatal clinics in Shiraz, Iran. All subjects were Shiraz residents, were beyond their 12th week of pregnancy and had not taken iron supplements during the previous year. The majority of the subjects selected from the Helal clinic were from low income families and those selected from the Hafez clinic were primarily from middle income families. However, due to the overlap of the household monthly income between the subjects

of the two clinics, all were classified into one of four groups on the basis of household monthly income: very low (VL), low (LO), middle low (ML) and middle and high (M-H). Mean daily consumption of major food items and dietary iron by the subjects and mean values for the constituents analyzed in blood are presented both by clinics and income groups.

In order to have a reference for iron status of non-pregnant women in the same area and from a population with adequate financial resources (middle or high income level), 21 young female volunteers were selected and blood analyzed as for the pregnant subjects. All non pregnant subjects were in the same age range as were the pregnant subjects, had not taken iron supplements within the last year, and had not been pregnant during the previous year.

Subjects in the VL income group had significantly higher intakes of bread and lower intakes of all major food items than those in the M-H income group; however, only the difference in rice, meat, poultry, greens and herbs and lettuce were statistically significant. In general, ML and M-H income groups had similar food consumption. Although the subjects in the LO income group had significantly lower intakes of meat and poultry than the M-H income group, their intakes were significantly higher than those of the VL income group.

Regression analysis showed a significant inverse and linear relationship between household income and bread consumption while consumption of rice and lettuce increased in a linear manner as household income increased. The income, however, was not a strong predictor of bread and rice consumption. Although the consumption of meat, poultry, greens and herbs and heme iron was shown to increase with increased household income, the relationships were not linear which indicates no further increase in their consumption beyond a certain income level. Consumption of legumes and eggs were not affected by the subjects' household income. Intake of heme iron and all those food items (rice, meat, poultry, greens and herbs and lettuce) which were limited by the household income were also affected by the household size; a lower intake was reported by subjects living in larger households. Income, however, was a stronger indicator of meat and poultry consumption and heme iron intake than the household size, while intake of rice and greens and herbs was influenced more by household size than income. Consumption of meat, eggs and lettuce was lower during the spring and summer than during fall and winter, but season was not a strong predictor of their consumption.

The average total daily intake of iron was similar between different income groups and was above the intake of 28 mg per day recommended by FAO/WHO (1970) for pregnant women. However, heme iron as a percentage of the average

daily total intake of iron was higher in higher income subjects. Bread formed the major source of iron in the diet of subjects in all income groups. While heme iron intake increased with higher household income and smaller household size, nonheme iron intake was not influenced by household income or size.

The average iron intake of six subjects (32.37 mg) as analyzed and calculated from the collected food samples (during a 24-hour period) was above the FAO / WHO recommendation for iron intake during pregnancy. Bread formed the major source of iron in the diet of all six subjects. Although the number of food samples analyzed were not sufficient to give a precise estimate of the average daily iron intake of the study population, the results were in agreement with the values for iron intake of the pregnant women as calculated from the household food consumption. Therefore, this method of dietary recall appears to be appropriate for this population.

Mean hemoglobin, hematocrit, serum iron and percent transferrin saturation were lower in VL and LO income groups than in ML and M-H income groups. Regression analysis showed a significant nonlinear relationship between the hemoglobin and hematocrit and income level. Stage of pregnancy had a significant inverse relationship with hemoglobin, hematocrit, serum iron and percent transferrin saturation which was not linear. Stage of pregnancy was a more important predictor

of hemoglobin and hematocrit than household income.

Subjects with transferrin saturation below 15.0% were regarded as anemic or at high risk of being anemic. According to this criteria, 9.3% of the VL, 7.4% of the L0 and 2.0% of the ML and the M-H income groups were anemic. All of these anemic subjects were in their second half of pregnancy with the majority in the last two months. Since in the VL and the L0 income groups the percentage of subjects in their later stages of pregnancy was higher than in the ML and the M-H income groups, the higher incidence of anemia and lower mean hemoglobin, hematocrit, serum iron and transferrin saturation levels among the subjects in the VL and the L0 income groups seems, in part, to be caused by their later stage of pregnancy (a statistically significant variable). Income, however, might have played a role in lower iron status of lower income subjects. Of 17 anemic subjects, 13 were in VL and L0 income groups. Even within the same stage of pregnancy, higher incidence of iron deficiency was seen among the lower income women.

It can be concluded that the iron status of pregnant women from all income groups was generally satisfactory during the first half of pregnancy. In the second half of pregnancy, incidence of iron deficiency anemia increased and more subjects were at risk of developing iron deficiency. This is probably due to the increased need for the hemopoietic substances mainly during the second half of pregnancy.

It needs to be mentioned that consumption of meat and poultry (the major dietary heme iron sources) and vitamin C sources (vegetables, greens and herbs and fruit), which are enhancers of iron absorption, was lower in diets of VL and L0 income groups. In contrast, intake of bread, a major source of fiber and phytate, both inhibitors of iron absorption, was higher in the diet of the subjects from the VL and L0 groups which may have contributed to lower iron availability in the diet and may have caused the pregnant women from lower income families to have a higher risk of developing iron deficiency especially during the second half of pregnancy.

A lower incidence of anemia in pregnant women of Shiraz compared to those previously studied in Tehran (95, 96) may be due in part to a higher food purchasing power or wiser selection of foods by the women of Shiraz within the limitations of their income.

In general, the iron status of most pregnant women in this study was satisfactory during the first half of pregnancy. As pregnancy proceeded, more cases of iron deficiency were seen. Iron supplementation of pregnant women who are considered at risk of developing iron deficiency on the basis of blood analysis is recommended.

STRENGTHS AND LIMITATIONS

Strengths

Although the use of household food consumption had some limitations in estimating the amount of food consumed by individual subjects, it was the most feasible method of dietary assessment. Most Iranian dishes are a mixture of several food items and, for a majority of foods, it is impossible for an individual to give an estimate of consumption. Therefore, the widely used method of 24-hour recall was not a reasonable method for assessing food intakes of this population. A food record was not a practical method of dietary assessment in the population studied since the majority of pregnant women from the Helal clinic were illiterate and it was desirable to use the same method of dietary recall for all subjects. The food record also had the same limitation of a 24-hour recall i.e. for most foods it is not possible to estimate the individual's consumption because of mixed dishes. Food inventory was inappropriate because of the high degree of illiteracy and family food purchasing pattern.

The subject's mean daily intake of major food items was determined by dividing the household consumption by an adjusted household size which considered caloric needs of

different age groups and uneven distribution of foods among the household members. Assuming an even distribution of foods underestimates the adult's food consumption in a household which includes children.

Multiple regression analysis was the most feasible method of statistical analysis since it identified statistically significant independent variable(s) related to consumption of food and dietary iron and to hematological status of the subjects (dependent variables). It identified the linear as well as nonlinear responses and interaction among different independent variables and could predict the response in the dependent variables for given values of the independent variables. Thus, in multiple regression all independent variables affecting the dependent variables are analyzed together. In contrast, in simple correlations the relationship between two variables is tested separately assuming other variables to be constant. This procedure may result in false or incomplete conclusions. Results of blood analysis showed significant differences between the percent transferrin saturation of the VL and the M-H income groups and one could conclude that the only significant variable explaining the lower percent transferrin saturation in the VL income group is the income, while results of regression analysis determined that the stage of pregnancy was a more significant variable than income.

All interviews were administered by the researcher which accounted for a better control and less variation. The researcher, a native of Iran, was familiar with the culture and language.

Limitations

One of the limitations of this study was an inability to determine the exact household income of subjects. A majority of subjects in the Helal clinic could not give a precise estimate of their monthly household income because the household head did not have a steady job, his remuneration fluctuated from day to day, or husbands did not share the income information with wives. Although the income ranges reported by these subjects do not represent the exact income of the households, they do give some information about the overall average monthly income. Per capita income is usually preferred to household income, but the even distribution of household income among members of different ages and with different living expenses could not be assumed in this study. However, by using regression analysis, both household income and size (in addition to other variables) were assumed to affect the subjects' food and iron consumption and hematological status and were included in the preliminary regression models to determine the significance of each variable and their interactions. Although household

income level was shown to be significantly related to the subjects' consumption of most food items, it is not the income per se but the food expenditure of the household which directly influences food intake of household members. Household food expenditure could not be estimated.

Although household weekly food consumption was the most feasible method of dietary recall, adjustments made to differentiate the uneven distribution of foods among the household members did not consider all possible factors. Lack of an adjustment for sexes and elderly persons living in the households might have underestimated or overestimated some subjects' food consumption.

No information on leftover or discarded foods was obtained so, in a few cases, food consumption may have been slightly overestimated. However, little food is usually wasted by the population studied.

A majority of the subjects could not give an accurate estimate of their household consumption of vegetables and fruits especially if they were asked to report those items which were not in season. Therefore, in calculating the average total daily intake of the subjects, iron from vegetables and fruits was not included. As a result, the average daily iron intakes of the subjects might be slightly underestimated. However, fruits and vegetables in the forms consumed in Iran generally contribute small amounts of iron.

Food samples consumed during a 24-hour period by some volunteer subjects were collected for determination of iron content. However sixteen food samples were discarded when the freezer in which they were stored failed during a 3-day weekend. Analysis could be performed on only six food samples which were stored in a different freezer. Although the result of analysis of the six composites gave some information about iron intake of the subjects, the number of samples analyzed was not sufficient to give a reliable estimate of iron intake of this population.

Changes in the country's economy prior to and during the study may have affected employment rate, inflation, food availability and prices of food, all of which could be expected to influence the food consumption of certain sections of the population.

RECOMMENDATIONS AND SUGGESTIONS FOR FURTHER RESEARCH

The incidence of anemia among the lower income subjects, most of whom had not received prenatal care (including iron supplementation) at all or until late in pregnancy, suggests that public health policy in Iran should be evaluated and adjusted to provide earlier and more complete prenatal care and follow-up of the low income pregnant population.

When iron supplements were given to pregnant women who were anemic or at high risk of becoming anemic, there was no follow-up to determine the effectiveness of the supplementation. Subsequent blood tests and determination of compliance are needed for this population.

An iron supplementation program is recommended to be accompanied by some information about the importance of iron supplements to the mother and baby.

As an informal part of interviews at Helal clinic and if the situation (environment and time) allowed, the researcher briefly and informally discussed nutrition with subjects and asked about their food purchasing power. Subjects seemed to be receptive, eager to learn and willing to discuss their problems related to limited food budgets.

At the end of the interviews, the researcher gave iron tablets (Ferrosulin, ferrous sulfate 200 mg) provided by physicians in the clinic, to the pregnant subjects who registered at the clinic during their last two months of pregnancy and received no other prenatal care. The high demand for iron during pregnancy and the importance of iron supplements to meet these needs were explained in simple words and subjects were asked to repeat the information given. Subjects asked questions, listened carefully and showed their interest in this new information. Some women reported that they had side effects including nausea, constipation, indigestion and epigastric pain when they had used iron supplements.

There is a need to design and provide nutrition education which can meet the needs of the population studied. Research is needed to determine appropriate delivery systems and sites for nutrition education. On the basis of this research, prenatal clinics appear to be one of the appropriate sites to reach the pregnant population. Brief sessions of nutrition and health education in classes held in prenatal clinics may be appropriate. At the time of this research no prenatal classes were provided in the clinics studied.

The feasibility of iron fortification of selected food products in Iran needs to be studied.

More information is needed concerning the role of vitamin C, an enhancer of iron absorption, in preventing iron deficiency anemia.

Use of appropriate food models and varieties of measuring cups and bowls (similar to the ones commonly used to measure foods in the household) may help the respondent to give a more accurate estimate of her individual and household food consumption in future studies. Use of 24-hour recall along with the household food consumption may provide more detailed information about the individual's food consumption.

Collection and analysis of food samples obtained from a randomly selected pregnant population will provide useful information about the nutrient intakes of the population and is recommended in future studies.

APPENDICES

APPENDIX A
CONSENT FORM
INTERVIEW SCHEDULES
DATA SHEETS

APPENDIX A-1
VERBAL CONSENT FORM
(asked before a witness)

Since most pregnant women are at high risk of becoming anemic, I would like to ask you if you agree to give a little more (approximately 2 teaspoons) blood (in addition to what is required by the clinic) to detect if you are anemic or not. However, you are free to decide about having this blood test and your treatment and care in this clinic will not be affected if you don't agree to have this blood test.

You will also be asked some questions about your family, number of children you have and their ages, number of pregnancies and miscarriages (if any) you had, you and your husband's occupation and income, your health status and the kind and amount of food consumed by you and your family.

Results of the interview and your blood test will be kept in strict confidence and you will remain anonymous in any report.

Do you agree to give a blood sample and answer the questions?
This subject (name: _____) has
agreed to participate in the study.

Date _____

Researcher _____

Witness _____

APPENDIX A-2

IRANIAN PREGNANT WOMEN

INTERVIEW SCHEDULE

(Original Schedule)

What is your name?

Family and Social History

1. How many children do you have?
How old is each?
2. Can you read and write?
If yes, how many years of schooling have you had?
3. Can your husband read and write?
If yes, how many years of schooling has he had?
4. What is the occupation of your husband?
5. Do you work outside the home?
If yes, what type of work?
6. What is your family income?
What are the sources of income?
7. Do any of your children work outside the home?
If yes, how many are working? What is their occupation and what is their income?
8. Are there other members of your family besides your children who are living with you?
If yes, what are their ages?
do they have any income?
do you receive any financial help from them?
9. Have you always been living in Shiraz?
If no, where did you live before living in Shiraz?
When did you move to Shiraz?

Medical History

10. How old are you?
11. How old were you at your first menstruation?
12. Is your menstrual cycle regular or irregular?
13. Is your blood loss during menstruation heavy, moderate, or scant?
How many days does your menstruation last?
14. How many pregnancies have you had?
15. Tell me about the outcome of each of your pregnancies.
How many miscarriages have you had?
Have any of your babies been stillborn?
Have any of your children died after birth? If yes,
how many and what was the cause?

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16. When do you expect to deliver your baby?
At what month or week of pregnancy are you now?
17. Tell me if you have any important or hereditary diseases like diabetes?
18. Have you had any surgery?
If yes, what?
19. Are you taking any medication(s) now?
If yes, what?
Who prescribed it (them) for you?

Diet History

20. How is your appetite during pregnancy?
21. Do you have nausea or vomiting (or both) during pregnancy?
If yes, how severe is it and how many times a day do you experience it? How long have you had that problem?
22. Do you have any desire or craving to eat some foods or non-food items?
If yes, what are those foods?
23. What kind of foods do you eat more of, or you don't eat (or eat less) because of your pregnancy? Tell me your reasons.
24. What foods do you think are good and what foods are bad to eat during pregnancy?
From whom did you receive this information?
25. How do you feed your babies?
Did you breast feed your babies?
If not, what was the reason?

Food Frequency

Tell me how much of the following food items are consumed in your household. How often? Indicate the amount consumed by the weights as you buy them or by showing me which one of these cups (placed in front of person) is closest to the amount you consumed.

26. How many loaves of bread do you buy each day?
How much bread do you eat yourself per day?
What kind of bread do you usually buy?
27. How often and how much of the following food items are consumed in your household?
 - Rice
 - Meat
 - Poultry
 - Fish and shrimp
 - Bologna and hot dog
 - Liver, heart and kidney
 - Eggs (you can tell the number of eggs you use)
 - Legumes including red beans, white beans, lima beans, pinto beans, chick peas, split peas and lentils
 - Milk
 - Yogurt

- Cheese
- Butter
- Potatoes
- Vegetables of the season
- Fresh greens and herbs used for stew, in rice or in ash*
- Fresh greens and herbs for eating raw
- Fruits of the season
- Soft drinks
- Tea

Food Pattern

28. Name the major food items or kinds of foods that are eaten for breakfast, lunch and dinner in your household in a week and indicate the frequency of their consumption.
29. How many cups of tea do you drink every day? Do you drink it before, with, after or between meals? Is it usually dark, medium or light in color?

24-Hour Recall

30. Now I want you to tell me everything you ate or drank yesterday. Start from breakfast, then lunch and then dinner and also mention anything you had in between the meals. You should also tell the amount of each food items consumed by yourself.

*Thick soups that are mostly made with vegetables, pulses and rice.

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IRANIAN PREGNANT WOMEN
INTERVIEW DATA SHEET
(Original Questionnaire)

Date _____

ID# _____

Name _____

Family and Social History

1. Number of children _____
Each child's age _____
2. Subject's level of education
____no____
____yes. years of schooling _____ Degree _____
3. Husband's level of education
____no____
____yes. years of schooling _____ Degree _____
4. Husband's occupation _____
5. Subject's occupation _____
6. Family income _____
Source of income _____
7. Children's occupation ____no, ____yes
How many working _____ Type of occupation _____
Income _____
8. Number of people living in household besides children ____
Their ages _____
Their income _____
9. Shiraz resident from childhood
____yes____
____no, place of residence _____,
when moved to Shiraz _____

Medical History

10. Age _____
11. Age at first menarch _____
12. Menarch cycle ____regular____
____irregular____
13. Blood flow during menstruation ____heavy____
____moderate____
____scant____
Duration ____days____
14. Gravida _____

15. Number of: miscarriages____, stillbirth____, died
after birth____, age at death____, cause____
16. Expected date of delivery____
Weeks of pregnancy____
17. History of important or hereditary diseases____

18. History of surgery____
19. Medications taken now____
prescribed by____

Diet History

20. Appetite____
21. Nausea____ Vomiting____
Severity____ times per day____
How long you have had the problem____
22. Extreme desire for foods or non-foods
____ no
____ yes, name those____
23. Foods eaten more frequently____
Foods eaten less frequently____
Reason for this change during pregnancy____

24. Foods good for pregnant women____
Foods bad for pregnant women____
Source of information____
25. How did you nurse your baby?____
Breastfeed ____ yes
____ no, why?

Anthropometric Measurement

Height____ cm
Weight____ kg

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Food Frequency

Household Size _____ Name _____

ID# _____

Date _____

How often and how much of the following food items are consumed in your household?

Food item	How often consumed	How much consumed
-----------	--------------------	-------------------

Bread (Type _____)

Rice

Meat

Poultry

Fish and shrimp

Liver, heart and kidney

Eggs

 Legumes: Beans
 Peas
 Lentils

Milk

Yogurt

Cheese

Butter

Potatoes

Vegetables in season

Greens and herbs (cooked)

Greens and herbs (raw)

Fruits in season

Soft drinks

Tea

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Food Pattern

Name _____

ID# _____

Date _____

Meals	Kinds of foods	Frequency of consumption (per week)
-------	----------------	--

Breakfast

Lunch

Dinner

Tea: cups per day _____

Before _____ With _____ After _____ or Between _____ Meals

Dark _____ Medium _____ Light _____

24-Hour Recall

Foods	Amount
-------	--------

Breakfast

Lunch

Dinner

APPENDIX A-3
 IRANIAN PREGNANT WOMEN
 INTERVIEW SCHEDULE
 (Final Revised Schedule)

General

1. What is your name?
How old are you?
2. How many children do you have?
How old is each of them? Start with the oldest.

Socioeconomic Information

3. Do you know how to read and write?
(If yes, how many years of schooling have you had?)
4. Does your husband know how to read and write?
(If yes, how many years of schooling has he had?)
- *5. Do you work outside the home?
(If yes, please describe this work.)
Do you receive a salary for this work?
How much money or other type of payment do you receive
per day or week or month?
- *6. What is your husband's occupation?
What is his income (per day or per week or per month?)
**Is this a fixed income?
If not, how often does he earn this much money?
- *7. How many people in your household receive salaries?
How much do they receive?
8. How many people live in your household?
How old are they?
9. What is your home address?

Medical History

10. How many times have you been pregnant?
How many miscarriages have you had?
 11. Have any of your babies been stillborn?
(If yes, how many?)
 12. Have any of your children died after birth?
(If yes, how many?)
For each child that died, would you tell me his (her)
age at death and the cause of death?
- * Household monthly income was calculated on the basis of
total income earned by the household members as reported
by the subjects.
- **This question was asked only of those subjects whose
husbands did not have a steady job.

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13. When do you expect to deliver your baby?
In what month or week of pregnancy are you now?

Household Food Consumption

Now I would like to ask you some questions about the amount of some food items consumed by your household and by yourself (if you can indicate). Please tell me how much of each item is consumed per day or per week or per month of if not consumed at all. To indicate the amount of consumption, you can tell me by the weights as you buy them or if you don't know the weights, tell me the amount by showing which of these cups that are placed in front of you is closest to the amount you used.

14. How many loaves of bread do you buy each day?
How much bread do you or your family eat each day?
What kind of bread do you usually buy?
15. How often and how much of the following food items are consumed in your household?
- Rice
 - Meat (do not include the weight of bones)
 - Poultry
 - Fish and shrimp
 - Liver, heart and kidney
 - Eggs (you can tell the number of eggs you use)
 - Legumes, like:
 - red beans
 - white beans
 - lima beans
 - pinto beans
 - chick peas
 - split peas
 - lentils
 - Vegetables of the season
 - Fresh greens and herbs for use in stews, in rice and in ash
 - Fresh greens and herbs for eating raw
 - Fruits of the season

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IRANIAN PREGNANT WOMEN
INTERVIEW DATA SHEET
(Final Revised Questionnaire)

Name _____

Date _____

ID# _____

General

1. Subject's name _____
Age _____
2. Number of children _____
Each child's age _____

Socioeconomic Information

3. Subject's education
_____ no
_____ yes...how long _____ Degree _____
4. Husband's education
_____ no
_____ yes...how long _____ Degree _____
5. Subject's occupation _____
income _____
6. Husband's occupation _____
income _____
7. Other sources of income in the household and its
amount _____
8. Number of people living in the household _____
their ages _____
9. Home address _____

Medical History

10. Gravida _____
Number of miscarriages _____
11. Number of stillbirths _____
12. Number of children died after birth _____
Their age at death _____
Cause of death _____
13. Expected date of delivery _____
Weeks of pregnancy _____

Anthropometric Measurement

Height _____ cm
Weight _____ kg

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Household Food Consumption

Name _____

ID# _____

Date _____

How often and how much of the following food items are consumed in the household:

Food item	How often consumed	How much consumed
-----------	--------------------	-------------------

Bread (Type _____)

Rice

Meat

Poultry

Fish and shrimp

Liver, heart and kidneys

Eggs

Legumes: Beans
Peas
Lentils

Vegetables in season

Greens and herbs for
cooking

Greens and herbs for
eating raw

Fruits in season

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APPENDIX B
WEIGHTS OF IRANIAN BREAD

APPENDIX B
WEIGHTS OF IRANIAN BREADS
AS PURCHASED IN SHIRAZ LOCAL BAKERIES^a

Type of Bread		
Sangak	Bazari	Masshady
	g	
560	249	315
564	218	307
551	256	291
582	267	300
656	273	314
668	255	304
687	238	311
725	248	297
624.4 \approx 625 ^b	250.5 \approx 250	304.9 \approx 305

^aBreads were bought from 12 bakeries in different regions in Shiraz, 4 bakeries for each kind of bread. Duplicate samples of the whole bread loaves were bought from each bakery and were weighed within 1 to 3 hours after purchase.

^bAverage

APPENDIX C

ENERGY REQUIREMENT OF DIFFERENT AGE GROUPS

APPENDIX C-1
ENERGY REQUIREMENT OF CHILDREN AND ADOLESCENTS
BY AGE (FAO/WHO, 1972)

Energy per person per day (kcal)		
Age (years)	Males	Females
<1	820	820
1	1180	1180
2	1360	1350
3	1560	1520
4	1720	1670
5	1870	1790
6	2010	1900
7	2140	2101
8	2260	2110
9	2380	2210
10	2500	2300
11	2600	2350
12	2700	2400
13	2800	2450
14	2900	2500
15	3000	2500
16	3050	2420
17	3100	2340
18	3100	2270
19	3020	2200
>19	3000	2200
		(+340 for later half of pregnancy)

APPENDIX C-2
ENERGY REQUIREMENT OF CHILDREN, ADOLESCENTS
AND ADULTS BY AGE GROUPS^a

Energy per person per day (kcal)			
Age (years)	Males	Females	Average for both sexes
1-4	1455	1430	1442.5
5-11	2251	2096	2174
12-19	2960	2385	2672
>19 (Reference adult - moderately active)	3000	2200 +350 (for later half of pregnancy)	2775

^a Average energy requirement of each age group was calculated using the average energy requirement for all the ages within the group.

APPENDIX C-3

THE RATIO OF ENERGY REQUIREMENT OF CHILDREN
AND ADOLESCENTS TO ADULTS
(AVERAGE FOR BOTH SEXES)

Age Groups (years)	Energy Req. (kcal)	Ratio of energy requirements
1-4	1440	$\frac{1440}{2775} = 0.52 \approx 1/2$
5-11	2174	$\frac{2174}{2775} = 0.78 \approx 4/5$
12-19	2672	$\frac{2672}{2775} = 0.96 \approx 1$
>19	2775	$\frac{2775}{2775} = 1 \approx 1$

APPENDIX D
SELECTION OF VARIABLES
FOR REGRESSION ANALYSIS

APPENDIX C-4
DEVIATION OF THE ADJUSTED
HOUSEHOLD SIZE

Adjusted Household Size (Adj. HS) was derived as the following:

$$\begin{aligned} \text{Adj. HS} = & \text{No. of persons in age group (1)} \times 1/2 \\ & + \text{No. of persons in age group (2)} \times 4/5 \\ & + \text{No. of persons in age group (3)} \times 1 \end{aligned}$$

The estimated weekly consumption of the food items by the subject was obtained by dividing the household's weekly consumption of those items by the Adj. HS.

Example: Subject's consumption of Food item A =

$$\frac{\text{Amount of food item A consumed in the household}}{\text{Adj. HS}}$$

APPENDIX D-1
REGRESSION ANALYSIS

Variables Assumed to be Related to Food and Iron Consumption
(To be included in the first regression models)

I. Dependent Variables

A. Food Consumption (g/day):

Y_B =Bread	Y_{Lg} =Legumes
Y_R =Rice	Y_E =Eggs
Y_M =Meat	Y_{GH} =Greens and herbs
Y_P =Poultry	Y_{Lt} =Lettuce

B. Iron Intake (mg/day): Y_{BFe} =Bread iron
 Y_{HFe} =Heme iron^a
 Y_{NHFe} =Nonheme iron^b
 Y_{TFe} =Total iron^c

II. Independent Variables

A. Linear: X_I =Household income (Rials per month)

X_{HS} =Household size (persons)

X_S =Season

B. Quadratic: X_I^2 , X_{HS}^2 , X_S^2

C. Two-way interaction: $X_I X_{HS}$, $X_I X_S$, $X_{HS} X_S$

D. Three-way interaction: $X_I X_{HS} X_S$

^aIron from meat and poultry.

^bIron from bread, rice, legumes, eggs, greens and herbs and lettuce.

^cSum of heme and nonheme iron.

APPENDIX D-2
REGRESSION ANALYSIS

Variables Assumed to be Related to Hematological Studies
(To be included in the first regression models)

I. Dependent Variables: Y_{Hgb} =Hemoglobin (g/dl)

Y_{Hct} =Hematocrit (%)

Y_{SFe} =Serum iron (μ g/dl)

Y_{TS} =Transferrin saturation (%)

II. Independent Variables:

A. Linear: X_I =Household income (Rials per month)

X_A =Age (years)

X_{wp} =Weeks of pregnancy

X_{NP} =Number of pregnancies

X_{HS} =Household size (persons)

X_{HFe} =Heme iron intake (mg/day)

X_S =Season

B. Quadratic: X_I^2 , X_A^2 , X_{wp}^2 , X_{NP}^2 , X_{HS}^2 , X_{HFe}^2 , X_S^2

C. Two-way interaction: $X_I X_A$, $X_I X_{wp}$, $X_I X_{NP}$, $X_I X_{HS}$,

$X_I X_{HFe}$, $X_I X_S$, $X_A X_{wp}$, $X_A X_{NP}$, $X_A X_{HS}$, $X_{wp} X_{NP}$,

$X_{wp} X_{HS}$, $X_{NP} X_{HS}$, $X_{HS} X_{HFe}$, $X_{HS} X_S$

D. Three-way interaction: $X_I X_A X_{wp}$, $X_I X_A X_{NP}$, $X_I X_A X_{HS}$,

$X_I X_{wp} X_{NP}$, $X_I X_{wp} X_{HS}$, $X_I X_{NP} X_{HS}$, $X_A X_{wp} X_{NP}$,

$X_A X_{wp} X_{HS}$, $X_A X_{NP} X_{HS}$, $X_{wp} X_{NP} X_{HS}$, $X_I X_{HS} X_{HFe}$, $X_I X_{HS} X_S$,

$X_I X_{HFe} X_S$

REFERENCES

REFERENCES

1. WHO Technical Report Series, No. 580. Control of nutritional anaemia with special reference to iron deficiency. Geneva, 1975.
2. Baker, S.J. Nutritional anaemia - a major controllable public health problem. Bull. WHO 1978; 56:659-74.
3. WHO Technical Report Series, No. 405. Nutritional Anaemias. Geneva, 1968.
4. Rath, C.E. and Finch, C.A. Sternal marrow hemosiderin: a method for the determination of available iron stores in man. J. Lab. Clin. Med. 1948; 33:81-6.
5. Addison, G.M., Beamish, M.R., Hales, C.N., Hodgkins, M., Jacobs, A., and Llewellyn, P. An immunoradiometric assay for ferritin in the serum of normal subjects and patients with iron deficiency and iron overload. J. Clin. Pathol. 1972; 25:326-9.
6. Bainton, D.F. and Finch, C.A. The diagnosis of iron deficiency anaemia. Am. J. Med. 1964; 37:62-70.
7. Baker, S.J. and DeMaeyer, E.M. Nutritional anemia: its understanding and control with special reference to the work of the World Health Organization. Am. J. Clin. Nutr. 1979; 32:268-417.
8. Aung-Than Batu, Thane-Toe, U-Hla-Pe and Khin-Kyi-Nyunt. A prophylactic trial of iron and folic acid supplements in pregnant Burmese women. Israel J. Med. Sci. 1976; 12:1410-7.
9. Carr, M.L. The diagnosis of iron deficiency in pregnancy. Obstet. Gynecol. 1974;43:15-21.
10. DeLeeuw, N.K.M., Lowenstein, L., and Hsiah, Y.S. Iron deficiency and hydraemia in normal pregnancy. Medicine 1966; 45:291-315.
11. Scott, D.E. and Pritchard, J.A. Iron deficiency in healthy college women. J. Am. Med. Assoc. 1967; 199: 897-900.

12. Dacie, J.V. and Lewis, S.M. Practical Haematology. London:Churchill-Livingstone, 1964.
13. International Committee for Standardization in Haematology. Brit. J. Haemat. 1967; 13 (suppl):71-5.
14. WHO Technical Report Series, No. 503. Nutritional anemias. Geneva, 1972.
15. Ramsay, W.N.M. The determination of iron in blood plasma or serum. Clin. Chim. Acta. 1957; 2:214-20.
16. Bothwell, T.H. and Mallett. B. The determination of iron in plasma or serum. Biochem. J. 1955; 59:599-602.
17. Ramsay, W.N.M. The determination of the total iron binding capacity of serum. Clin. Chim. Acta. 1957; 2:221-6.
18. Herbert, V., Gottlieb, C.W., Lau, K.S., Fischer, M., Gevirtz, N.R. and Wasserman, L.R. Coated charcoal assay of unsaturated iron binding capacity. J. Lab. Clin. Med. 1966; 67:855-62.
19. Sood, S.K., Ramachandran, K., Mathur, M., Gupta, K., Ramalingaswami, V., Swarnabai, C., Ponniah, J., Mathan, V.I., and Baker, S.J. WHO sponsored collaborative studies on nutritional anemia in India. I. The effects of supplemental oral iron administration to pregnant women. Quart. J. Med. 1975; 44:241-58.
20. Yusufyi, D., Mathan, V.I., and Baker, S.J. Iron, folate and vitamin B₁₂ nutrition in pregnancy: a study of 1000 women from southern India. Bull. WHO 1973; 48:15-22.
21. Olson, A.D. and Hamlin, W.B. A new method for serum iron and total iron-binding capacity by atomic absorption spectrophotometry. Clin. Chem. 1969; 15:438-44.
22. Miles, L.E.M., Lipschitz, D.A., Bieber, C.P. and Cook, J.D. Measurement of serum ferritin by a 2 site immunoradiometric assay. Ann. Biochem. 1974; 61: 209-24.
23. Jacobs, A., Miller, F., Worwood, M., Beamish, M.R. and Wardtop, C.A. Ferritin in the serum of normal subjects and patients with iron deficiency and iron overload. Brit. Med. J. 1972; 4:206-8.

24. Lipschitz, D.A., Cook, J.D., and Finch, C.A. A clinical evaluation of serum ferritin as an index of iron stores. *New Engl. J. Med.* 1974; 290:1213-6.
25. Goldie, D.M. and Thomas, M.J. Measurement of serum ferritin by radioimmunoassay. *Ann. Clin. Biochem.* 1978; 15:102-8.
26. Molina, R.A., Diez-Ewald, M., Fernandez, G. and Valazques, N. Nutritional anaemia during pregnancy. A comparative study of two socio-economic classes. *J. Obstet. Gynaecol. Brit. CommonWlth.* 1974; 81:454-8.
27. WHO Technical Report Series, No. 452. Requirements of Ascorbic Acid, Vitamin D, Vitamin B₁₂, Folate and Iron. Geneva, 1970.
28. Recommendations by the Committee on Maternal Nutrition. In: *Maternal Nutrition and the Course of Pregnancy.* Committee on Maternal Nutritional. Food and Nutrition Board, National Research Council. National Academy of Sciences, Washington, D.C., 1970; p. 188.
29. Pritchard, J.A. Anemias Complicating Pregnancy and the Puerperium. In: *Maternal Nutrition and the Course of Pregnancy.* Committee on Maternal Nutrition. Food and Nutrition Board, National Research Council. National Academy of Sciences, Washington, D.C. 1970; p. 74-109.
30. Worthington, B.S., Vermeersch, J., and Williams, S.R. Changes in blood volume and composition. In: *Nutrition in pregnancy and lactation.* Saint Louis: C.V. Mosby Company, 1977; p. 38.
31. Gatenby, P.B.B., and Lillie, E.W. Clinical diagnosis of 100 cases of severe megaloblastic anemia of pregnancy. *Brit. Med. J.* 1960; 2:1111-4.
32. MacGregor, M.W. Maternal anemia as a factor in prematurity and prenatal mortality. *Scott. Med. J.* 1963; 8:134-40.
33. Llewellyn, J.D. Severe anemia in pregnancy. *Aust. N.Z. J. Obstet. Gynecol.* 1965; 5:191-7.
34. Ratten, G.J. and Beischer, N.A. The significance of anemia in an obstetric population in Australia. *J. Obstet. Gynecol. Brit. CommonWlth.* 1972; 79:228-37.

35. Woodruff, C.W. and Brideforth, E.B. Relationship between the hemogram of infant and that of the mother during pregnancy. *Pediatrics* 1953; 12:681-5.
36. Lanzkowsky, P. The influence of maternal iron deficiency anaemia on the hemoglobin of the infant. *Arch. Dis. Childh.* 1961; 36:205-9.
37. Burman, D. Iron requirement in infancy. *Brit. J. Haematol.* 1971; 20:243-7.
38. Pitkin, R.M. Hematologic Indices. In: *Laboratory Indices of Nutritional Status in Pregnancy*. Committee on Nutrition of the Mother and Preschool Child. Food and Nutrition Board, National Research Council. National Academy of Sciences, Washington, D.C. 1978.
39. Savin, M.A. A practical approach to the treatment of iron deficiency. *Ration Drug Therapy* 1977; 11:1-6.
40. Haghshenass, M., Mahloudji, M., Reinhold, J.G. and Mohammadi, N. Iron deficiency anemia in an Iranian population associated with high intakes of iron. *Am. J. Clin. Nutr.* 1972; 25:1143-6.
41. Venkatachalam, P.S. Iron metabolism and iron deficiency in India. *Am. J. Clin. Nutr.* 1978; 21:1156-61.
42. Hallberg, L., and Bjorn-Rasmussen, E. Determination of iron absorption from whole diet. *Scand. J. Haematol.* 1972; 9:193-7.
43. Layrisse, M., Martinez-Torres, C., and Gonzalez, M. Measurement of the total daily dietary iron absorption by the extrinsic tag model. *Am. J. Clin. Nutr.* 1974; 27:152-62.
44. Cook, J.D., Layrisse, M., Martinez-Torres, C., Walker, R., Monsen, E., and Finch, C.A. Food iron absorption measured by an extrinsic tag. *J. Clin. Invest.* 1972; 51:805-15.
45. Layrisse, M., and Martinez-Torres, C. Food iron absorption: iron supplementation of food. *Prog. Haematol.* 1971; 7:137-60.
46. Martinez-Torres, C., and Layrisse, M. Effect of amino acids on iron absorption from a staple vegetable food. *Blood* 1970; 35:669-82.
47. Layrisse, M., Martinez-Torres, C., Cook, J.D., Walker, R., and Finch, C.A. Iron fortification of food: its measurement by the extrinsic tag method. *Blood* 1973; 41:333-52.

48. Cook, J.D., and Monsen, E.R. Food iron absorption. I. Use of a semisynthetic diet to study absorption of nonheme iron. *Am. J. Clin. Nutr.* 1975; 28:1289-95.
49. Layrisse, M., Martinez-Torres, C., and Roch, M. The effect of interaction of various foods on iron absorption. *Am. J. Clin. Nutr.* 1968; 21:1175-83.
50. Cook, J.D. and Monsen, E.R. Food iron absorption in human subjects. III. Comparison of the effect of animal protein on nonheme iron absorption. *Am. J. Clin. Nutr.* 1976; 29:859-67.
51. Kroe, D.T., Kinney, T.D., Kaufman, N., and Klavins, J.V. The influence of amino acids on iron absorption. *Blood* 1963; 21:548-52.
52. VanCampen, D., and Grosse, E. Effect of histidine and certain other amino acids on the absorption of iron-59 by rats. *J. Nutr.* 1969; 99:68-74.
53. VanCampen, D. Enhancement of iron absorption from ligated segments of rat intestine by histidine, cysteine and lysine: Effects of removing ionizing groups and of stereoisomerism. *J. Nutr.* 1973; 103: 139-42.
54. Davis, P.S. and Deller, D.J. Prediction and demonstration of iron chelating ability of sugars. *Nature* 1966; 212:404-5.
55. Davis, P.S. and Deller, D.J. Effect of orally administered chelating agents EDTA, DTPA and fructose on radio iron absorption in man. *Aust. Ann. Med.* 1967; 16:70-5.
56. Amine, E.K. and Hegsted, D.M. Effect of dietary carbohydrates and fats on inorganic iron absorption. *J. Agric. Food Chem.* 1975; 23:204-8.
57. Sayers, M.H., Lynch, S.R., Jacob, P., Charlton, R.W., Bothwell, T.H., Walker, R.B. and Mayet, F. The effects of ascorbic acid supplementation on the absorption of iron in maize, wheat, and soya. *Brit. J. Haematol.* 1973; 24:209-18.
58. Cook, J.D., and Monsen, E.R. Vitamin C, the common cold, and iron absorption in man. *Am. J. Clin. Nutr.* 1977; 30:235-41.
59. Sayers, M.H., Lynch, S.R., Charlton, R.W. and Bothwell, T.H. Iron absorption from rice meals cooked with fortified salt containing ferrous sulphate and ascorbic acid. *Brit. J. Nutr.* 1974; 31:367-75.

60. Bjorn-Rasmussen, E. and Hallberg, L. Iron absorption from maize. *Nutr. Metab.* 1974; 16:94-100.
61. Conrad, M.E., and Schade, S.G. Ascorbic acid chelates in iron absorption: a role for hydrochloric acid and bile. *Gastroenterology* 1968; 55:35-45.
62. Moore, C.V., and Dubach, R. Observations on the absorption of iron from foods tagged with radioiron. *Trans. Assoc. Am. Phys.* 1951; 64:245-56.
63. Elwood, P.C., Newton, D., Eakins, J.D. and Brown, D.A. Absorption of iron from bread. *Am. J. Clin. Nutr.* 1968; 21:1162-9.
64. Callender, S.T., Marney, S.R., and Warner, G.T. Egg and iron absorption. *Brit. J. Haematol.* 1970; 19: 657-65.
65. Monsen, E.R. and Cook, J.D. Food iron absorption in human subjects. V. Effects of the major dietary constituents of a semisynthetic meal. *Am. J. Clin. Nutr.* 1979; 32:804-8.
66. Moore, C.V. Iron nutrition. Iron metabolism: an International symposium, Springer, Berlin, 1964; p. 224.
67. Bjorn-Rasmussen, E. and Hallberg, L. Effect of animal proteins on the absorption of food in man. *Nutr. Metabol.* 1979; 23:192-202.
68. Disler, P.B., Lynch, S.R., Charlton, R.W., Torrance, J.D., Bothwell, T.H., Walker, R.B., and Mayet, F. The effect of tea on iron absorption. *Gut.* 1975; 16:193-200.
69. Derman, D., Sayers, M., Lynch, S.R., Charlton, R.W. and Bothwell, T.H. Iron absorption from a cereal-based meal containing cane sugar fortified with ascorbic acid. *Brit. J. Nutr.* 1977; 38:261-9.
70. Rossander, L., Hallberg, L. and Bjorn-Rasmussen, E. Absorption of iron from breakfast meals. *Am. J. Clin. Nutr.* 1979; 32:2484-9.
71. Monsen, E.R. and Cook, J.D. Food iron absorption in human subjects. IV. The effects of calcium and phosphate salts on the absorption of nonheme iron. *Am. J. Clin. Nutr.* 1976; 29:1142-8.

72. Cook, J.D. and Monsen, E.R. Food iron absorption in human subjects. II. The effect of EDTA on absorption of dietary nonheme iron. *Am. J. Clin. Nutr.* 1976; 29:614-20.
73. Layrisse, M. and Martinez-Torres, C. Fe (III)-EDTA as iron fortification compound. *Am. J. Clin. Nutr.* 1977; 30:1166-74.
74. Viteri, F.E., Garcia-Ibanez, K. and Torem, B. Sodium iron EDTA as an iron fortification compound in Central America. Absorption studies. *Am. J. Clin. Nutr.* 1978; 31:961-71.
75. Martinez-Torres, C., Romano, E.L., Renzi, M. and Layrisse, M. Fe(III)-EDTA as iron fortification. Further studies. *Am. J. Clin. Nutr.* 1979; 32:809-16.
76. Widdowson, E.M. and McCance, R.A. Iron exchanges in adults on white and brown bread diets. *Lancet* 1942; 1:558-90.
77. Husain, R., Walker, R.B., Layrisse, M., Clark, P. and Finch, C.A. Nutritive value of food iron. *Am. J. Clin. Nutr.* 1965; 16:464-71.
78. Callender, S.T. and Warner, G.T. Iron absorption from bread. *Am. J. Clin. Nutr.* 1968; 21:1170-4.
79. Elwood, P.C., Benjamin, I.T., Fry, F.A., Eakins, J.D., Brown, D.A., DeKoch, P.C. and Shah, J.U. Absorption of iron from Chapatti made from wheat flour. *Am. J. Clin. Nutr.* 1970; 23:1267-71.
80. Bjorn-Rasmussen, E. Iron absorption from wheat bread - influence of various amounts of bran. *Nutr. Metabol.* 1974; 16:101-10.
81. Reinhold, J.G., Ismail-Beigi, F. and Faraji, B. Fiber vs. phytate as determinant of the availability of calcium, zinc, and iron in breadstuffs. *Nutr. Rept. Int.* 1975; 12:75-85.
82. Ismail-Beigi, F., Faraji, B. and Reinhold, J.G. Binding of zinc and iron to wheat bread, wheat bran, and their components. *Am. J. Clin. Nutr.* 1977; 30: 1721-5.
83. Reinhold, J.G., Garcia, J.S. and Garson, P. Binding of iron by fiber of wheat and maize. *Am. J. Clin. Nutr.* 1981; 34:1384-91.

84. Kelsay, J.L., Behall, K.M. and Prather, E.S. Effect of fiber from fruits and vegetables on metabolic responses of human subjects. II. Calcium, magnesium, iron and silicon balances. *Am. J. Clin. Nutr.* 1979; 32:1876-80.
85. Martinez-Torres, C. and Layrisse, M. Nutritional factors in iron deficiency: Food iron absorption. *Clin. Haematol.* 1973; 2:339-52.
86. Herbert, V. Introduction to symposium on folic acid deficiency. *Am. J. Clin. Nutr.* 1970; 23:841-2.
87. Giles, C. and Shuttleworth, E.M. Megaloblastic anemia of pregnancy and the puerperium. *Lancet* 1958; 2:134-7.
88. Strieff, R.R. and Little, A.B. Folic acid deficiency in pregnancy. *New Engl. J. Med.* 1967; 276:776-9.
89. Temperley, I.J., Meehan, M.J. and Gatenby, P.B. Serum folic acid levels in pregnancy and their relationship to megaloblastic marrow change. *Brit. J. Haematol.* 1968; 14:13-9.
90. Herbert, V., Coleman, N., Spivack, M., Ocasio, E., Ghanta, V., Kimmel, K., Brenner, L., Freundlich, J. and Scott, J. Folic acid deficiency in the United States: folate assays in a prenatal clinic. *Am. J. Obstet. Gynecol.* 1975; 123:175-9.
91. Bailey, L.B., Mahan, C.S. and Dimperio, D. Folacin and iron status in low-income pregnant adolescents and mature women. *Am. J. Clin. Nutr.* 1980; 33:1997-2001.
92. Russel, R.M., Ismail-Beigi, F., Afrasiabi, K., Rahimifar, M., Pourkamal, D. and Ronaghy, H. Folate levels among various populations in central Iran. *Am. J. Clin. Nutr.* 1976; 29:794-8.
93. Kouhestani, A.J., Ghavifeker, H., Rahmanian, M., Mayurian, M. and Sarkissian, N. Composition and preparation of Iranian breads. *J. Am. Diet. Assoc.* 1969; 55:262-6.
94. Russel, R.M., Ismail-Beigi, F. and Reinhold, J.G. Folate content of Iranian breads and the effect of their fiber content on the intestinal absorption of folic acid. *Am. J. Clin. Nutr.* 1978; 29:799-802.
95. Geissler, C., Calloway, D.H. and Margen, S. Lactation and pregnancy in Iran. Diet and nutritional status. *Am. J. Clin. Nutr.* 1978; 31:341-54.

96. Froozani, M.D., Vahidani, F., Montazami, K. and Malekafzali, H. Maternal and newborn iron status in a public and a private maternity hospital at delivery in Tehran. *Trop. Pediatr. Envir. Child. Health.* 1978; 182-6.
97. Maleki, M. Food consumption and nutritional status of 13-year old village and city schoolboys in Fars province in Iran. *Ecol. Food Nutr.* 1973; 2:39-42.
98. Recommended Daily Dietary Allowances, Food and Nutrition Board, National Academy of Sciences, National Research Council, 1972.
99. Food and Nutrition Institute (Iran). Composition of Iranian Foods, Food and Nutrition Institute Publication No. 112. Tehran (Iran).
100. Church, C.F. and Church, H.N. Food Values of Portions Commonly Used. J.B. Lippincott Co. 12th Edition, 1975.
101. Statistical Package for the Social Sciences - SPSS. Nie, N.H., Hadlai Hull, C., Jenkins, J.G., Steinbrenner, K. and Bent, D.H. McGraw Hill Inc., Second Edition, 1975.
102. Energy and protein requirement. Report of a joint FAO/WHO Expert Group, FAO, Rome 1972.
103. Gill, J.L. Design and Analysis of Experiments in the Animal and Medical Sciences. 1978. The Iowa State University Press, Ames, Iowa, U.S.A.
104. National Census of Population and Housing. Fars Province, Iran. November 1976. Plan and Budget Organization, Statistical Center of Iran, Serial No. 170, August 1980.
105. Fars Province Statistics. No. 817. August 1980.
106. Reinhold, J.G. Phytate destruction by yeast fermentation in whole wheat meals. *J. Am. Diet. Assoc.* 1975; 66:38-41.
107. Hytten, F.E., and Paintin, D.B. Increase in plasma volume during normal pregnancy. *J. Obstet. Gynaecol. Br. Commonw.* 1963; 70:402-7.

108. Pitkin, R.M. Hematologic indices. In: Laboratory Indices of Nutritional Status in Pregnancy. The National Research Council. National Academy of Sciences. Washington, D.C. 1978; pp. 9-17.
109. Hytten, F.E. and Leitch, I. The Physiology of Human Pregnancy, Blackwell Scientific Publications, Oxford, 2nd edition, 1971.
110. Paintin, D.B., Thomson, A.M. and Hytten, F.E. Iron and the haemoglobin level in pregnancy. J. Obstet. Gynaecol. Br. Commonw. 1966; 73:181-90.
111. Svanberg, B., Arvidson, B., Norrby, A., Rybo, G. and Solvell, L. Absorption of supplemental iron during pregnancy - a longitudinal study with repeated bone marrow and absorption measurements. Acta Obstet. Gynecol. Scand. Suppl. 1975; 48:87-108.
112. Hancock, K.W., Walker, P.A. and Harper, T.A. Mobilisation of iron in pregnancy. Lancet 1968; 2:1055-8.
113. Gerritsen, T. and Wilker, A.R.P. The effect of habitually high iron intake on certain blood values in pregnant Bantu women. J. Clin. Invest. 1954;33:23-6.
114. Kohout, E., Dutz, W. and Petrossian, A. Laboratory findings in Iranian villages and tribal communities, 1964. Am. J. Clin. Nutr. 1967; 20:410-4.

