

AMERICAN UNIVERSITY OF ARMENIA
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**RISK FACTORS OF IRON-DEFICIENCY ANEMIA
IN INFANTS UNDER 2 YEARS OF AGE IN YEREVAN:
A CASE-CONTROL STUDY.**

(Grant Proposal)

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YEREVAN 99

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Executive summary

Iron-deficiency anemia (IDA) is the most prevalent nutritional disorder among infants throughout the world, especially in developing countries. In low-income countries IDA causes poor performance on mental and motor tests among infants and children.

It is a serious public health problem among children in Armenia as well. According to statistical data from the Ministry of Health that was available only for 1998, the national figure of iron deficient anemia from mild to moderate among infants was 26 %. In urban areas IDA was found in 15%-16% of infants, while in rural areas this figure accounts for 38% of infants.

Since IDA is a disease with significant consequences on child development, the only practical way to approach this problem is prevention of IDA in infancy. Thus, the purpose of this study is the identification of potential risk factors mostly contributing to IDA in infants in Armenia. As a result, data could be used to develop programs for effective prevention. No epidemiological studies have been done before in Armenia concerning risk factors of IDA in infants.

A case control study will be used as a tool for identification of main risk factors contributing to IDA in children under 2 years of age in Yerevan.

The case definition for this study includes all children under 2 years of age from Yerevan, who are diagnosed for the first time with iron-deficiency anemia identified from the Children's Republic Clinical Hospital and the Children's Clinical Hospital # 3 during the time of the survey. The control definition includes all children under 2 years of age from Yerevan, who do not have iron-deficiency anemia identified from the Children's Republic Clinical Hospital and the Children's Clinical Hospital # 3 during the time of the survey and in the medical history.

Cases and controls will be matched on age and gender of children, and hospital of admission. The study will be conducted during the time period of January 1, 2000 - October 1, 2000. 173 cases and 346 controls are necessary for this study.

The influence of following factors will be measured: demographic characteristics of the child (age, gender, birth weight, and maturity); personal medical history of the child (previous diseases, blood losses, and surgical manipulations); diet of the child; anemia of pregnancy;

maternal coffee consumption; educational level of caretakers; and expenditure on food per person.

The data will be collected using the same questionnaire for cases and controls, and from medical records. Following a definition of the variables of interest a questionnaire was developed. It will be used in face-to face interviews or in phone interviews, if a patient had blood test and left the hospital before the interview.

Data will be entered into Stata computer program. Statistical analysis will be done with use of chi-square tests and logistic-regression models, and will be based on estimation of odds ratios and 95% confidence intervals.

INTRODUCTION

Background

Iron deficiency anemia (IDA) is the most prevalent nutritional disorder among infants throughout the world, especially in developing countries. In the US, among poor African-American and Hispanic infants anemia is as high as 20%-24%, while in low-income countries IDA causes poor performance on mental and motor tests among infants and children [1]. These data suggest that IDA is a major public health problem among poor minority children.

Iron-deficiency anemia is a serious public health problem among children in Armenia as well. According to statistical data from the Ministry of Health that was available only for 1998, the national figure of mild to moderate iron deficiency anemia among infants was 26 %. In urban areas IDA was found in 15%-16% of infants, while in rural areas this figure accounts for 38% of infants which is more than two times higher than that in urban areas.

Anemia due to poor dietary iron is seen most often between 9-24 months of age in infants being fed large amounts of milk during periods of growth, and between 3 months and 2 years of age iron deficiency is the leading cause of anemia [2]. Some mothers practice poor habits such as giving tea to their newborn children, which inhibits iron absorption and can produce anemia.

Iron-deficiency anemia is a disease with significant consequences on child development. IDA can have effects on the immune system [3], behavior [4], and mental [5] development. Several studies have indicated a strong correlation between iron deficiency anemia and a delay in the psychomotor development of infants [6,7]. The areas most effected are language and body balance. For these infants iron therapy in most cases is not sufficient to reverse the psychological effects. Thus, if anemia occurs, even timely and adequate iron therapy seems to be ineffective in reversing behavioral and cognitive disadvantages. The only practical way to approach this problem is prevention of iron deficiency anemia in infancy.

Thus, the first step should be identification of potential risk factors mostly contributing to iron-deficiency anemia in infants in Armenia in order to develop effective prevention programs.

No epidemiological studies have been done before in Armenia concerning risk factors of IDA in infants.

Risk Factors for Iron-Deficiency Anemia in Infants

Some epidemiological studies of iron-deficiency anemia done in the world that have included infants have found factors that can predict risk of IDA development.

Figure 1 summarizes the underlying and immediate causes of iron-deficiency anemia.

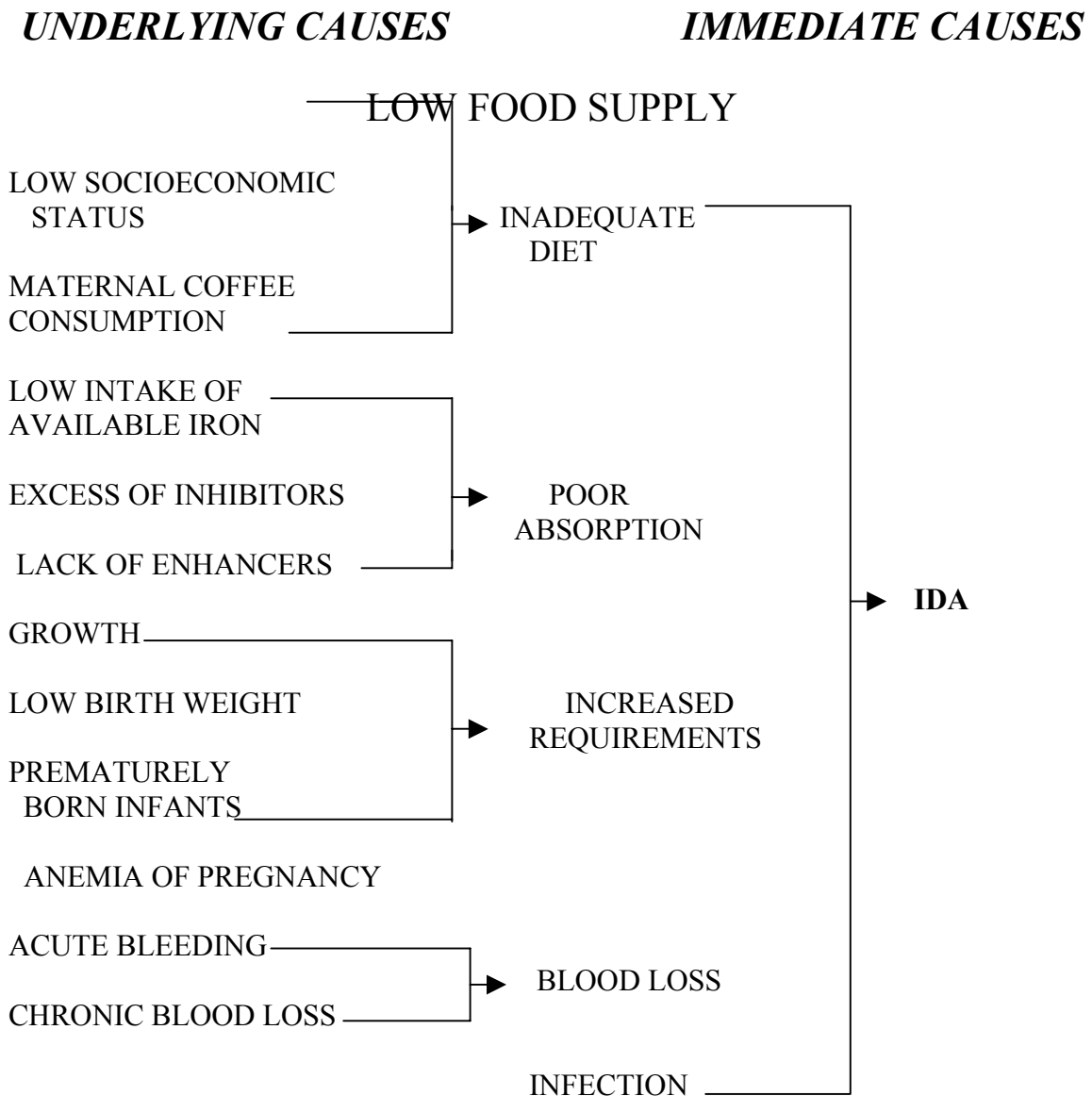


Figure 1. The development of iron-deficiency anemia in infants.

The general risk factors for IDA for infants are of dietary origin due to low food supply and low intake of iron; low socioeconomic status; poor absorption of iron due to low bioavailability of iron, excess of inhibitors, lack of enhancers; increased requirements of iron in infants, especially in those who are low birthweight and premature; anemia of pregnancy; blood loss, and infectious diseases.

Low Socioeconomic Status

Inadequate and poor quality of diet together with infections are the most common immediate causes of IDA [8, 9]. However, these are linked very much to socioeconomic development, so that it is expected that the prevalence of iron-deficiency anemia will not be reduced unless socioeconomic conditions will be improved. Data indicate that low-income countries have a higher prevalence of IDA than those with high socioeconomic status. Even within one country IDA is widespread among poor minorities [8].

For instance, in Iran a study was conducted that showed that socioeconomic status affects mothers' nutritional status and consequently that of their infants. This study involved 194 pregnant women randomly selected from a public hospital representing people of low socioeconomic status. Data were compared with those of pregnant women from private maternity hospital representing high socioeconomic status. According to the results, the mean values of hemoglobin, serum iron, and percent transferrin saturation were significantly lower in the low socioeconomic group than in the higher socioeconomic group. The mean birth weight and the mean values of hemoglobin, serum iron, percent transferrin saturation of newborns from the mothers of low socioeconomic status were also significantly lower [10].

Poor Iron Absorption

Iron deficiency is not only caused by a low amount of iron in the diet but also by its poor absorbability. The poor absorption of iron from cereal- and vegetable-based diets is well known.

Milk is a major source of calories during the first year of life. Both breast milk and cow's milk are iron poor foods. The iron content of breast milk falls from about 0.5 mg/liter during the first month after birth to about 0.3 mg/liter between 4 and 6 months of age. The iron content of cow's milk or unfortified cow's milk formulas ranges from 0.5 mg/liter to 1.0 mg/liter [11]. Infant formula is one of the major sources for iron fortification in infancy. Most iron-fortified cow's milk formulas contain about 6.8 mg/liter, 12.8 mg/liter of iron. (See Figure 2).

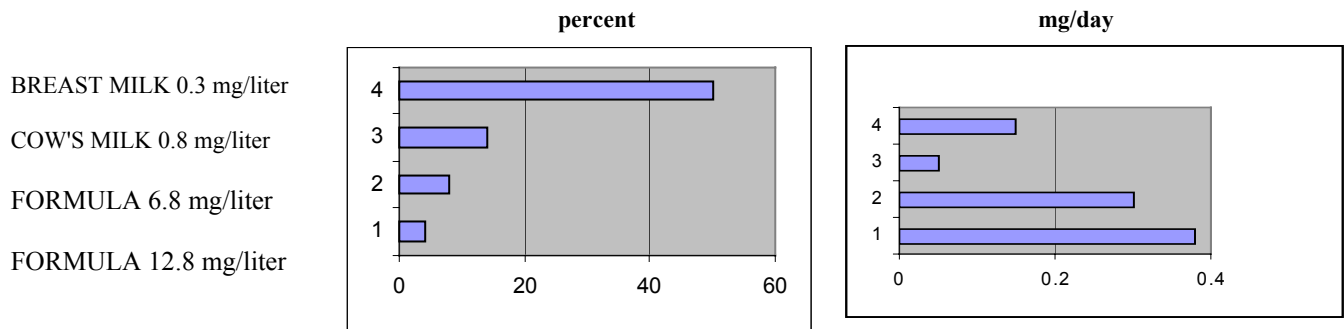


Figure 2. Iron absorption in infants. The calculations are based on a daily intake of 750 ml of milk or formula. The percentage of iron absorbed from each food is shown on the left. The total amount of iron assimilation predicted from the usual daily intake of these foods is shown on the right.

Despite the small amount of iron in breast milk, bioavailability makes an important difference in the amount absorbed.

Literature shows that on average 49 % of the iron is absorbed from breast milk, a high figure compared with about 10 % of iron absorbed from cow's milk or unfortified cow's milk formula [12, 13]. Mean absorption of iron from iron-fortified formulas is about 3 % [13]. Although the percentage absorbed is less about two times than that of unfortified formula, the absolute amount absorbed is more than ten times greater [13].

These data show that infants absorb iron from breast milk more efficiently than from cow's milk and formulas. Breast milk also carries some protection against the development of iron-deficiency anemia while early introduction of cow's milk to infants can cause iron-deficiency.

Iron absorption is influenced by the interaction of various foods consumed in the same meal and the amounts of non-heme and heme iron that they contain [14].

Heme iron, present primarily in foods of animal origin, is well absorbed and is relatively unaffected by diet composition. Non-heme iron, present in vegetables, generally is poorly absorbed and is greatly affected by enhancing or inhibiting substances in the diet [9].

Infants during weaning need highly bioavailable dietary iron to ensure optimal iron status. Meat and ascorbic acid are the main enhancers of non-heme food iron absorption.

There was a study aimed to examine the effect of meat intake on hemoglobin concentration. 41 healthy, term, partially breast fed 8-month-old infants were randomized into two groups: 1. a low-meat group (LMG), where infants received a diet with a mean meat content of 10 g/day; 2. a high-meat group (HMG), where infants received a diet with a mean meat content of 27 g/day. At the beginning of the intervention no significant differences were found in hemoglobin concentrations between the two groups. After the intervention that lasted two months, there was a significant difference in the change in hemoglobin concentration. The intake of iron from meat was significantly higher in the HMG than in the LMG.

The results suggest that an increase in meat intake can prevent a decrease in hemoglobin in late infancy, probably by enhancing iron absorption [15].

Other investigations show that the use of orange juice containing ascorbic acid can more than double the absorption of iron from an entire meal [16].

Some studies show that absorption of non-heme food iron can be inhibited by tea, carbonates, phytate, bran and egg yolk. However, these foods have not any appreciable effect on heme iron assimilation. Therefore, iron-deficiency anemia can not develop in non-vegetarian subjects as a consequence of this food consumption only [16,17].

Thus, on the one hand, it is possible to develop iron-deficiency anemia while ingesting large amount of foods rich in iron but in which iron is poorly absorbed. On the other hand, it is possible to escape deficiency ingesting far less than the recommended amount of iron but in forms that are particularly well absorbed.

Increased Iron Requirements

Despite the much smaller body size of infants and children, their iron requirements are almost higher than those of adults [8]. This situation is responsible for the high prevalence of nutritional anemia during infancy and childhood.

Prematurely born and low birth weight infants are at risk of iron-deficient anemia, because of the doubled requirements necessary for rapid growth [18]. In premature infants the absolute amount of total body iron is less at birth than in the full-term infants [9]. Since low birth weight

infants also have more rapid rate of growth, iron stores are exhausted earlier and the demands for iron are much greater than in the term infants [19].

Anemia of Pregnancy

Many case-control studies were done to evaluate the association between anemia during pregnancy and iron-deficiency anemia in infancy. One of them is a prospective case-control study of infants under 1 year of age conducted in Jordan among middle-class urbans [20].

A sample of 107 anemic (Hb < 11 g/dl) and 125 non-anemic mothers were selected at 37 weeks gestation and matched for age and parity, infant data at birth were obtained. The infants were reviewed at 3, 6, 9, and 12 months of age to assess growth, current nutrition, infection rates and iron status.

According to the results the incidence of iron-deficiency anemia was very high in these infants (72%), but significantly higher in the infants born to anemic mothers at all stages of the year with the incidence of 81% (n = 91) compared to 65% in controls (n = 112).

Thus, infants of mothers who had anemia during pregnancy appear to be at increased risk of developing iron-deficiency anemia undetected at birth.

Maternal Coffee Consumption

An interesting study was conducted in Costa-Rica among low-income women to find out whether maternal coffee intake contributes to maternal and infant anemia. Pregnant women of low socio-economic status were contacted through the Prenatal Clinic of the Social Security System in Cartago, Costa-Rica, from September 1985 through October 1986. Based on habitual coffee consumption, these women were divided into coffee drinkers (≥ 10 g ground coffee/d): n = 22; and coffee non-drinkers (0 g/d): n = 26. Women who drank 1-2 cups coffee/d were excluded from the study as well as coffee non-drinkers who consumed tea. Women in this population rarely consumed cola and hot chocolate.

Groups had similar income, education, prenatal care, age, parity, weight, height, pregnancy weight gain, prenatal iron supplementation, energy, protein, Fe, and vitamin C intake and infant sex and gestational age.

According to the results, maternal hemoglobin (Hb) and hematocrit (Hct) at 8 month gestation, cord blood Hb and Hct, infant birth weight and Hb and Hct at 1 month of age, and iron concentration in breast- milk were significantly lower in the coffee drinking group than in the noncoffee drinking group. In addition, the association of coffee with infant Hb and Hct was independent of maternal Fe status and birth weight.

Thus, these results indicate that maternal coffee intake may contribute to maternal and infant anemia [21].

Blood Loss

Some data suggest that occult intestinal blood loss during infancy contribute significantly to the development of iron-deficiency anemia [22]. It may be associated with the consumption of excessive amounts of fresh or pasteurized cow's milk [23].

Childhood Infectious Diseases

Common childhood infectious diseases were found to be risk factors for development of iron-deficiency anemia. A retrospective study was carried out to determine the role of infection on hemoglobin level of a cohort of Alaska Native infants. Hemoglobin and health records were available on 308 children between 6-11 and 12-17 months of age. Additional records were available on 187 children at 18-23 months of age. Episodes of chickenpox, measles, pertussis, and lower respiratory infections were reviewed.

According to the results the mean hemoglobin level of infants 6-11 and 12-17 months of age decreased with the increasing number of total infectious episodes occurring within the three months before hemoglobin measurement. This trend was not apparent for infants 18-23 months of age. At the 6-11 and 12-17 month age interval the number of lower respiratory infections were most associated with a decreased hemoglobin level [24].

These observations emphasize the role of infection in the development and maintenance of iron-deficiency anemia in children under 1 year of age.

Thus, these are general factors that should be accounted to predict risk of development of iron-deficiency anemia in infants.

Specific Aims

This study is a case-control study that aims to identify main risk factors contributing to iron-deficiency anemia in children under two years of age in Yerevan. Since, many risk factors of IDA are known world-wide, this study proposes to investigate the risk factors that most contribute to IDA in infants in Yerevan.

As a result, data could be used to develop programs to prevent IDA in infants that will be directed toward those risk factors identified in this study. The designing of these programs will be hopefully cost-effective and helpful for Armenia.

The main research question:

What are the main and specific risk factors contributing to iron-deficient anemia in children 0-2-year-old in Yerevan?

The objectives of this study:

1. To assess the diet of children with IDA
2. To determine the rate of low birth weight and prematurely born infants amongst study participants.

3. To determine how the health status of women during pregnancy influences the development of IDA in their 0-2-year-old children
4. To determine the living conditions (by food expenditure) in the families of children with and without IDA
5. To determine the educational level of mothers that might influence on health status of children with IDA.

According to the literature and interviews with doctors from the Children's Republic Clinical Hospital, the main variables of the study are the following:

1. Demographic characteristics of the child, which included age, gender, and birth weight, maturity.
2. Personal Medical History of the child concerning previous diseases, such as chickenpox, measles, pertussis, lower respiratory diseases, etc.; blood losses, surgical manipulations.
3. Diet of the child, which includes breast-feeding, introduction of cow's milk before or after 6 months of age, tea intake.
4. Anemia of pregnancy.
5. Maternal coffee consumption.
6. Educational level of mothers.
7. Expenditure on food per person.

Subjects and Methods

Study Design

A case-control study will be used as a tool for identification of main and specific risk factors mostly contributing to iron-deficiency anemia in children under 2 years of age in Yerevan.

This type of analytical design is chosen because of efficiency in time and expense. This method allows the study of multiple, potential causes of a disease among cases and controls. In

addition, one of the strengths of a case-control study is that the prevalence of the condition being studied can be unknown.

It is very difficult to assess the overall situation of iron-deficiency anemia among infants in Armenia because of the lack of national data. It is even more difficult to get estimates of IDA, because parameters other than hemoglobin are not measured. In order to distinguish IDA from other types of anemia there is a need for additional blood tests on serum ferritin level.

Thus, the main outcome measure in the study is iron-deficiency anemia defined as Hb < 10 g/dl, and serum ferritin < 10 mg/l.

Study Population

This study will be conducted among children 0-2-year-old, residing in Yerevan, who will attend the Children's Republic Clinical Hospital and the Children's Clinical Hospital # 3 during the time period of March 1, 2000 - June 1, 2000; excluding children attending at reanimatological, gastroenterological, and pulmonary departments.

Case definition: All children under two years of age from Yerevan, who will be diagnosed as iron-deficiency anemic for the first time in the Children's Republic Clinical Hospital and the Children's Clinical Hospital # 3 during the time of the survey.

Control definition: All children under 2 years of age from Yerevan, who do not have iron-deficiency anemia in the Children's Republic Clinical Hospital and the Children's Clinical Hospital # 3 during the time of the survey and in the medical history.

Cases and controls will be matched by age and gender of children, and hospital. Two controls will be assigned for each case.

Drawing the controls from the same source as the cases provides convenience and some strength, because it can be assumed that study participants are drawn from the same segment of the population and matched on social, economic, and environmental factors. In addition, drawing cases from different hospitals increases generalizability of the results to all patients with the disease.

I exclude children admitted to some departments of the hospitals, and purposely choose patients whose conditions will be independent of IDA. It also allows to generalize the results to the population, which is problem with doing a hospital-based study.

Exclusion criteria for cases and controls: 1) multiple births; 2) age over two-year-old; 3) not residing in Yerevan; 4) diagnosed as having anemia without iron-deficiency; 5) children from reanimatological, gastroenterological and pulmonary departments of the hospitals.

Data collection

The data will be collected using the same questionnaire for cases and controls. Six trained interviewers who are students of the Public Health Department will conduct interviews. Medical records of the study participants will also abstracted by these students. The interview will be face-to-face for main caretakers of children, particularly for mothers, and phone interviews will be conducted for those who had blood tests and left the hospitals before the interview (phone numbers are available from medical records).

The interview will take about 15 minutes. Before beginning consent will be obtained in face-to-face and in phone interviews [See appendix A]. A questionnaire has been developed and pretested at the Children's Republic Clinical Hospital. After pretesting this questionnaire revision were made [See appendix B]. The questionnaire covered the following items: demographic characteristics (age, gender, and birth weight of the child, education of mothers, expenditure on food per person), experiences during pregnancy, nutritional status of the child.

Sample Size

In order to calculate sample size the Stata statistical program was used. Instead of using alpha as 0.05, alpha was split up amongst all the different study variables that should be detected. Since, there are 7 study variables, an alpha of 0.007 (0.05/7) was used for each one.

α -error = 0.007;

β -error = 0.2;

Power = 1 - β -error = 0.8

A two-sample sample size of proportions was computed for each of the variables. As a result, 7 different sample sizes were obtained, and the largest of those sample sizes was chosen.

Estimated required sample sizes:

cases n1 = 173

controls n2 = 346

Analysis

Data will be entered on computer using Excel, and the Stata statistical program will be used for analysis of data.

Statistical analysis will be done with use of

- chi-square tests, where odds ratios with 95% confidence intervals will be determined; and
- multivariate logistic-regression models to control for potential confounding, where the dependent variable is positive iron-deficiency anemia status in children, and the independent variables are birth weight, maturity of the infants, previous diseases, blood losses, surgical manipulations, diet of the child, anemia during pregnancy, maternal coffee consumption, expenditure on food, educational level of mothers.

In logistical regression model odds ratios with 95 percent confidence intervals will be used to characterize the association of independent variables with the likelihood of being diagnosed as iron-deficiency anemic (i.e. case vs. control status).

The Study Limitations

- There are limitations in conducting a hospital-based study. In particular, it is difficult to generalize the results to the population as a whole. However, this problem can be solved to some extent: patients from the hospitals can be chosen whose conditions are entirely independent of IDA (excluding patients from some departments of the hospitals). In addition, cases can be selected from different hospitals in order to increase the generalizability of findings to all patients with this disease.
- Another limitation is that the cases are only from Yerevan. This is due to the fact that it is difficult to access patients from other regions. As a result, this may create selection bias.

- ❑ A potential problem in all case-control studies is recall bias. The interviewees may be more likely to remember the information that in their opinion had a connection with the disease.
- ❑ Another type of bias that may influence on the results of the study is instrumental bias. It may be created by the questionnaire.

Human Subjects

Ethical considerations:

During a population survey issues of privacy, confidentiality, and informed consent should be taken into account.

Interviewers will be trained in a way that they will be able to interact properly with caretakers of children. Before interview, a consent form will be provided giving detailed explanations of the aim of the study and procedures necessary for the study. Study participants will be asked for permission to abstract their medical records and to have additional blood test.

Giving a unique identifier to each interview will ensure confidentiality of informants. Only the principal investigator will have access to the names and identification numbers of the participants.

Time Frame for the Project

The time frame for the project was planned in a way to manage all activities. At the beginning of the study it is necessary to hire personnel who will conduct the study. During the period of March 1, 2000 -1, 2000 cases and controls will be interviewed. It was computed as follow:

Key-informants interview stated that on average 10 children under two years of age are admitted to the hospital. Using a conservative estimate it can be assumed that among these 10 children at least one of them will have iron-deficiency anemia and at least 2 two of them will not have anemia and will be eligible for the study. For the two hospitals where the study will be conducted, each day there should be 2 cases and 4 controls

obtained. In order to obtain the 173 cases and 346 controls it is estimated that 3 months for data collection is necessary. Data entry will begin in a month after starting the interviews and finish a month after all interviews will be completed. Then, analysis of the study will be done.[See appendix C].

Budget

The estimated expenditures for implementing the proposed study are given in detail in the budget [See appendix D]. The overall estimated budget is about thirty four thousand US dollars (\$34,000) for the study with nine months duration.

Personnel Responsibilities:

- ❑ The program Co-ordinator is responsible for administration and management of the study.
- ❑ Two Project Assistants are responsible for administration and monitoring of interviewer's activities, their training, and data entry into computer.
- ❑ Six trained interviewers will complete the questionnaire in two hospitals.

Indirect cost is estimated at 10% of direct cost.

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References

1. Pollitt, E. Poverty and child development: relevant of research in developing countries to the United States. *Child Development* 1994 Apr; 65: 283-95.
2. Kaye, R, Oski, FA, Barness, LA. *Core Textbook of Pediatrics*. Third Edition.
3. Dallman, PR: Iron deficiency and the immune response. *Am J Clin Nutr* 1987; 46: 329-334.
4. Pollitt, E, Soemantri, AG, Yunis, F et al: Cognitive effects of iron deficient anemia [C]. *Lancet* 1985: 1:158.

5. Lozoff, B, Brittenham, GM, Wolf, AW et al: Iron deficiency anemia and iron therapy effects on infant developmental test performance. *Pediatrics* 1987; 79: 981-995.
6. Wallter, T. Effect of iron-deficiency anemia on cognitive skills in infancy and childhood. *Bailliers Clin Haematol* 1994 Dec; 7(4): 815-27.
7. Major, P. Iron deficiency anemia and psychomotor development in infants. *Tidsskr Nor Laegeforen* 1994 Jun 30; 114(17): 1995-6.
8. Florentino, RF, Guirriec, RM. Prevalence of nutritional anemia in infancy and childhood with emphasis on developing countries. New York, Raven Press, 1984: 61-74. Nestle Nutrition Workshop Series. Vol.4.
9. Dallman, PR. Iron nutrition in infancy. In: Bond JT, Filer LJ JR, Leveille GA, Thomson AM, Weil WB JR, ed. *Infant and child feeding*. New York, Academic Press, 1981; 363-373.
10. Froozani, MO, Vadhani, F, Montazani, K, et al. Maternal and newborn iron status in a public and private maternity hospital at delivery in Tehran. *Trop Pediatr Environ Child Health* 1978; 24:182.
11. Siimes, MA, Vuori, E., and Kuitunen, P. Breast milk iron: A declining concentration during the course of lactation., *Acta Paediatr. Scand* 68, 1979, 33-37.
12. Saarinen, UM, and Siimes, MA. Iron absorption from infant formula and the optimal level of iron supplementation. *Acta Paediatr. Scand* 66, 1977, 719-722.
13. Rios, E, Hunger, RE, Cook, JD, Smith, NJ, and Finch, CA. The absorption of iron as a supplements in infant cereal and infant formulas. *Pediatrics* 55, 1975, 686-693.
14. Monsen, ER, Hallberg, L, Layrisse, M, Hegsted, DM, Cook, JD, Mertz, W, and Finch, CA. Estimation of available dietary iron. *Am J Clin Nutr.* 1978; 31: 134-141.
15. Engelman, MD, Sandstrom, B, Michaelsen, KF. Meat intake and iron status in late infancy: an intervention study. *J Pediatr Gastroenterol Nutr* 1998 Jan; 26(1): 26-33.
16. Callender, ST, Marney, SR, and Warner, GT. Eggs and Iron absorption. *Br J haematol* 1970; 19: 657-665.
17. Stekel, A, Amaz, M, Calvo, E, Chadud, P, Hertrampt, E, Laguno, S, Olivares, M, Pizarro, F. Nutritional significance of interactions between iron and food components. *Arch Latinoam Nutr.*1983 Mar; 33(1): 33-41.
18. Kilbride, J, Baker, TG, Parapia, LA, Khoury, SA, Shuqaidef, SW, Jerwood, D. Anemia during pregnancy as a risk factor for iron-deficiency anemia in infancy: a case-control study in Jordan. *Int J Epidemiol* 1999 Jun; 28(3): 461-8.

19. Lafuente Mesanza, P, Ojembarrena Martinez, E, Sasieta Altuna, M, Pinan Frances, MA, Urreta Dolora, MJ, Lombardera Jimenez, JL. Anemia and depletion of iron reserves in healthy 12-month-old infants. *An Esp Pediatr* 1992 Jul; 24-8.
20. Lundstrom, U. Iron release from the stores: a mechanism in maintenance of concentration of hemoglobin in low-birth-weight infants. *Acta Paediatr Scand* 1980 Mar; 69(2): 249-52.
21. Munoz, LM, Lonnerdal, B, Keen, CL, and Dewey, KG. Coffee consumption as a factor in iron deficiency anemia among pregnant women and their infants in Costa Rica. *Am J Clin Nutr* 1988; 48: 645-51.
22. Hoag, MS, Wallerstein, RO, and Pollycove, M. Occult blood loss in iron deficiency anemia of infancy. *Pediatrics* 1961; 27: 199-203.
23. Wilson, JF, Lahey, ME, and Heiner, DC. Studies on iron metabolism. V. Further observations on cow's milk-induced gastrointestinal bleeding in infants with iron-deficient anemia. *J Pediatr* 1974; 84: 335-344.
24. Cruz, A, Parkinson, AJ, Hall, D, Bulkow, L, Heyward, W. Association of early childhood infections and reduced hemoglobin levels in a historic cohort of Alaska Native infants. *Arctic Med Res* 1990 Oct; 49(4): 175-9.

APPENDICES

Appendix A

Informed Consent for Mothers

I _____ give my consent to participate in a study about factors that contribute to iron-deficiency anemia in children under two years of age. I understand that as a part of this study I will be answering questions about my pregnancy history, my infant's health and diet. I also understand that as a part of this process my infant will have additional blood tests to make clear whether my child has iron-deficiency anemia or not, and my infant's medical records may be reviewed to get further information about health status of my infants.

I understand that my participation in this study is completely voluntary, and my name will not be attached to any of the records that will be reviewed for the study. The information that I provide will be used only for this program and will not be shared with anybody outside of the research team. I understand that I am free to end the interview at any time and withdraw from this study.

I realize that I have a right to refuse to answer any questions I am not comfortable answering. I do understand that my participation in this project will provide valuable information in improving the health of pregnant women and infants. I understand that I will be provided a copy of this consent form, at my request, and have access to the Principal Investigator of this study if I have any questions about my participation.

By signature form I acknowledge that I am willing to participate in this study

Participant's Signature

Date

Witness Signature

Date

Appendix B

Questionnaire for Mothers

May I speak to mother of

_____?

Name of the child

If mother is not available, use proxy (father, grandmother)

1. Who is the person being interviewed?

Mother _____

Father _____

Other family member _____ (please, specify)

Now I will ask questions concerning your pregnancy

2. Did you have anemia during your pregnancy with this child?

Yes _____

No _____ (if No, go to Q.3)

2.1. During what period of pregnancy did you have anemia? (check all responses)

1-st trimester _____

2-nd trimester _____

3-rd trimester _____

3. What diseases did you have during your pregnancy?

None _____

DK _____

4. Did you drink coffee during this pregnancy?

Yes _____

No _____ (if No, go to Q.5)

4.1. On average, how many cups of coffee did you drink per day?

DK _____

Now, I will ask questions concerning child birth

5. What kind of complications did you have during delivery? (Cesarian section, etc.)

None _____
Other _____ (please, specify)
DK _____

6. Was your child born on time or not?

Term _____
Preterm _____
DK _____

7. What was the birth weight of your child?

< 2,500 g _____
2,500-3,000 g _____
> 3,000 g _____

8. What is the gender of your child?

Male _____
Female _____

9. What is the age of your child?

_____ (months)

10. What diseases did your child have?

DK _____

11. Did your child have any surgical manipulations?

Yes _____
No _____
DK _____

12. Did your child have any blood loss?

Yes _____
No _____
DK _____

Now, I am going to ask questions concerning your child's diet

13. Did you breast-feed your infant?

Yes _____
No _____ (if No, go to Q.14)

13.1. How long did you breast feed your infant?

_____ (months)

13.2. Did you supplement breast feeding with something?

Yes _____
No _____ (if No, go to Q.14)

13.2.1. With what? (Specify type of food) At what age ?

_____ -- _____ (months)

14. Did you give your child cow's milk?

Yes _____
No _____ (if No, go to Q.15)

14.1. At what age did you introduce cow's milk to your child?

_____ (months)
DK _____

14.2. How long did you give it to your child?

_____ (months)
DK _____

15. Did you give your child tea?

Yes _____
No _____ (if No, go to Q.16)

15.1. At what age?

_____ (months)
DK _____

15.2. How long?

_____ (months)
DK _____

15.3 How much? _____ ml

16. What is your educational level?

_____ School (8)
_____ High school (10)
_____ College (2)
_____ University (5-6)
_____ Post graduate

Total years _____

17. How much do you spend on food each month?

_____ (drams)
DK _____

18. How many people live in your household?

Thank you.

Medical Record Abstraction

Child's:

Age _____ (months)

Gender: Male _____
Female _____

Birth Weight _____ (kg)

Maturity _____ (weeks)

Previous Diseases:

Any Surgical Manipulations:

Any Blood Loss:

Appendix D

Budget

	\$	Duration	Total
<u>Personnel</u>			
Program Coordinator /mo	\$400	9 months	\$3,600
Project Assistants /mo	\$300	2 persons@9months	\$5,400
Subtotal			\$9,000
<u>Training of 6 Interviewers</u>			
Food person/day	\$3	6 persons @ 2days	\$36
Training Materials	\$100		\$100
Trainees /day	\$5	6 persons @ 2days	\$60
Subtotal training			\$196
<u>Testing</u>			
Testing on Hemoglobin	\$1	520 children	\$520
Testing on Serum Ferritin	\$1	520 children	\$520
Subtotal testing			\$1,040
<u>Capital Assets</u>			
Computer	\$900	2	\$1,800
Printer	\$400	1	\$400
Copier	\$600	1	\$600
Subtotal			\$2,800
<u>Operating Costs</u>			
Office Rent /mo	\$50	9 months	\$450
Office Supplies /mo	\$150	9 months	\$1,350
Communication	\$300		\$300
Transportation: car rental & maintenance /mo	\$200	2 cars @ 9months	\$3,600
fuel /mo	\$40	9 months	\$360

Subtotal operating costs			\$6,060
<u>Interview</u>			
Interviews & filling Questionnaires and Medical Records Abstraction /q-re	\$1	520	\$520
Checking & cleaning the Data /q-re	\$0.5	520	\$260
Data entry into Electronic Files /q-re	\$1	520	\$520
Analysis	\$1000		\$1,000
Subtotal			\$2,300
Direct Cost			\$21,396
Indirect Cost (10% of direct cost)			\$2,140
Overall Total			\$23,536

Appendix C

Time Frame for the Project

Activities Planning For 2000										
Activities/month	Jan	Feb	March	April	May	June	July	Aug.	Sept	Oct.
Staff Hiring										
Office rent, supplies, equipment, preparation of materials, etc.										
Training of the interviewers										
Conducting interviews										
Coding and data entry into computer										
Analysis										
Report preparation										