ORIGINAL ARTICLE

Environmental risk factors for iron deficiency anemia in children 12-24 months old in the area of Thessalia in Greece

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Abstract

Background and aim: Iron deficiency anemia (IDA) is a common problem all over the world, which attacks mainly pregnant women, infants and children. The aim of the study was to estimate the prevalence of IDA in children 12-24 months old in a specific area of Thessalia, located in the central part of Greece, and to identify the environmental risk factors associated with it.

Patients and Methods: In the first part of this cross-sectional and case-control study, the hemoglobin (Hb) levels of 938 children were estimated by a mobile photometer analyzer. In the second part of the study, children with Hb $<$ 11 gr/dl were compared with matched random selected controls in hematological, anthropometric and environmental parameters. The estimated laboratory values were Hb, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, zinc protoporphyrin, serum iron, serum ferritin, transferring saturation, total iron binding capacity and Hb electrophoresis. Finally 75 children (34 boys, 41 girls, mean age 17.51$\pm$3.5 months), who were found with IDA, constituted the case group while 75 healthy children constituted the control group.

Results: The prevalence of IDA in the region was 7.99%. At the same time, a number of children with stigma of b-thalassaemia (2.13%) was discovered, something that had escaped identification. There were no differences due to the method of determination (mobile or laboratory) in the values of Hb between the two groups. Significant differences were recorded ($p<0.001$) in all hematological and anthropometric parameters except for head circumference. Regarding environmental factors, significant differences were found in the following parameters: ratio rooms/number of family members ($p=0.01$), number of family members ($p=0.01$), number of children in the family ($p<0.001$), birth rate ($p<0.001$), education and profession of the parents ($p<0.001$), source of drinking water and sewage system ($p<0.001$), duration of breast feeding ($p<0.001$), milk consumption by the child during the period of the reported research ($p<0.001$), child's health status according to the mother ($p<0.001$), and frequency of seeking pediatric care ($p=0.02$).

Conclusions: Although the prevalence of IDA in this area of Greece is similar to the one observed in the rest of the developed world, it still consists a public health problem. The mobile method for Hb estimation should be introduced in Greece since its reliability to detect IDA has been, once more, confirmed. The application of simple questionnaires for the detection of the environmental IDA risk factors could help in the prognosis and prevention of anemia. Further improvement of the IDA status in Greece could be achieved through the dissemination of information about iron rich foods, the amelioration of environmental conditions and the application of reliable, easy to use and cheap methods for Hb estimation. Hippokratia 2008; 12 (4): 240-250

Key words: children, iron deficiency anemia, environment, questionnaire, Greece

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Iron deficiency anemia (IDA) constitutes the most frequent nutritional problem for both developing and developed countries. It affects mainly infants, children during early childhood and pregnant women[1]. IDA involves a decrease in concentration of red blood cells or hemoglobin (Hb) in the peripheral blood[1-3].

There are several risk factors involved in IDA. Most frequently they involve lack of iron in nutrition, intestinal infestations, virus infections along with factors related to socio-economic conditions. Apart from Hb determination, IDA diagnosis is based on several hematological and biochemical parameters. It also demands an etiologic explanation and an understanding of IDA pathogenesis[4].

IDA sets in by following three distinct biochemical steps: At first there is a pre-latent iron deficiency stage during which iron stores are gradually depleted; during the second stage, that of latent iron deficiency, the level of serum iron starts decreasing in parallel with an offsetting increase in siderophylin synthesis; finally, during the last stage microcytic IDA sets in and IDA becomes a clinically identifiable disease[5-7].

IDA is preventable. Countries, which realized the magnitude of the problem and identified the associated risk factors, were able to intervene directly. At the same time, the application of intervention programs by several countries, for the last twenty years, resulted in a worldwide decrease in the occurrence of anemia. Those programs were based on methods of IDA diagnosis that combined ease of use, swiftness and reliability in diagnosis. Mobile methods that are reliable in assessing Hb levels are considered particularly significant[8-10].

IDA is a serious public health problem in Greece whose estimation is complicated by the high incidence of thalassaemia, in central and northern Greece. The percentage of IDA in Greece is thought to remain at high levels especially in infancy and childhood[11-13].

The general aim of this study was to estimate the prevalence of IDA in children 12-24 months old in the area of Thessalia, which is located in the central part of Greece, and to identify by means of a simple questionnaire the environmental risk factors associated with it.

Material and Methods

The reported research was conducted for one year in the prefecture of Larissa; in particular, in four rural areas of the prefecture of Larissa (Deskati, Elassona, Tirnavos and Gonni) served by National Health System (NHS) Primary Health Care Centers (PHCCs) and in the city of Larissa. The selection of the area was based on the homogeneity of the population in terms of its origin, the significant improvement in the standard of living that has taken place during the last decade, the general opinion that this area had one of the highest percentages of IDA in Greece and the lack of similar studies.

The total population of children 12-24 months old, living in the study area was 3,012 according to the National Statistical Service of Greece[14]. The distribution of the population per region is shown on the table 1. The study population consisted of children who requested health care services at the recruitment places (Table 1). The screening of the children took place in the PHCCs of the rural areas or in the Immunization Center of the city of Larissa which also belongs to the NHS. The selection of the recruitment places was based on the fact that PHCCs in the rural areas are the principle providers of pediatric care (Table 1) while in urban areas several NHS Services (including hospitals and Immunization Centers) provide pediatric care along with privately practicing pediatricians. The Immunization Center of the city of Larissa was selected because it provides preventive pediatric care to the highest percentage of the Larissa children population compared to privately practicing pediatricians (>60) or the other local NHS services (Table 1).

Nine hundred and thirty eight (938) children 12-24 months old participated in the study (Tables 1 and 2). A blood drop was taken from each child at the beginning of the study with a finger prick. An immediate examination of Hb levels (Hb-He) was performed through a mobile analyzer (b-hemoglobin photometer, HaemoCue, Sheffield, Great Britain). Hb values <11 gr/100ml of blood (as defined by WHO) were used as the diagnostic limit of anemia while a number of other measurements (Table 3) of iron status were used, especially ferritin levels below 10 μg/L and low red-cells indices, in order to identify whether the anemia was due to iron deficiency[2,3].

Children who presented with Hb levels < 11 gr/100 ml (according to the mobile analyzer) were examined by taking venous blood samples for the following: Hb (Hb Lab), hematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular Hb (MCH), mean corpuscular Hb concentration (MCHC), zinc protoporphyrin (ZPP), serum iron (SFe), serum ferritin (SF), transferring saturation (TS), total iron binding capacity (TIBC) and Hb electroforesis[15].

Table 1: Number of children per region taking part in the research.

	Children per region	Children attended	Children studied
Larisa	2,401	530	493
Deskati	84	73	70
Elassona	201	155	151
Tirnavos	253	166	161
Gonni	73	65	63
Total	3,012	989	938

Table 2: Characteristics of the children studied.

Area	Cases of b-thalassaemia	Carriers of b-thalassaemia	Participants remaining	Total
Deskati	5 (M=2, F=3)	3 (M=2, F=1)	62 (M=34, F=28, ma=17.7±3.4)	70 (M=38, F=32)
Elassona	13 (M=5, F=8)	4 (M=2, F=2)	134 (M=66, F=68, ma=18.0±3.3)	151 (M=73, F=78)
Tirnavos	21 (M=11, F=10)	4 (M=1, F=3)	136 (M=61, F=75, ma=17.4±3.1)	161 (M=73, F=88)
Larisa	32 (M=14, F=18)	6 (M=4, F=2)	455 (M=218, F=237, ma=18.0±3.2)	493 (M=236, F=257)
Gonni	4 (M=2, F=2)	3 (M=2, F=1)	56 (M=29, F=27, ma=7.5±3.3)	63 (M=33, F=30)
Total	75 (M=34, F=41, ma=17.5±3.0)	20 (M=11, F=9, ma=18.5±2.7)	843 (M=408, F=435)	938 (M=463, F=475)

M= male, F=female, ma = mean age

The study was planned as a cross sectional and a case control study. Seventy-five children (34 boys, 41 girls, mean age 17.51±3.5 months), who were found with IDA, constituted the case group (Group A).

The control group (Group B) with Hb values ≥11 gr/dl was selected randomly within the next three days of the case recruitment. They underwent the same blood tests as the case group and, finally, 75 children were selected as the control group having being matched, one by one, with the case group by sex, age (± 1 month) and origin.

The children of both groups were measured for weight (electronic scale), height (rollametre 102.8 for the length) and head circumference (lasso) and their percentiles were defined[16].

Before the children were recruited and blood sampled, the parents were duly informed about the research and consented for their children to participate. They then answered to a specific questionnaire concerning the environmental conditions of their household and the feeding practices of the child. In particular the following information was asked: number of rooms in the house, number of members living in the house, number of children in the family, order of birth for the participating child among its siblings, education and profession of the parents, level of care offered to the child, other income in the family (pensions and/or financial support from Community or Social Services), source of drinking water and sewage system, child's health status according to the mother, frequency of seeking pediatric care, duration of breast feeding, milk consumption by the child during the period of the reported research, iron intake and the history of b-thalassaemia in the family.

The analyzers and methods employed for the blood examinations in the lab were as follows: a) Coulter counter T660, in vitro diagnostic use for Hb, Hct, MCV, MCH, and MCHC, b) ZPP Helena Protofluor reagent, Helena Laboratories, Texas, USA, for ZPP, c) colorimetric method, Elitech Diagnostics, Sees, France for Sfe, d) immunoassay method, IMX, Abbot Diagnostics divisions for SF, e) iron liquicolor CAB method, Human France for TS and TIBC and f) microchromatographic method HbA_2, quick column equipment and supplies, Helena Laboratories, USA for the Hb electrophoresis. The central-core laboratory for blood tests was settled in the Unit of Thalassaemia Prevention in the General Hospital of Larissa.

Children who were found to be carriers of b-thalassaemia (Hb electrophoresis) during the study did not take part in the formation of the two groups.

Two-tailed t test and Wilcoxon non-parametric test for the quantitative variables, Mc Nemar test and test of symmetry for qualitative variables and conditional logistic regression analysis for odds ratio estimation ($p<0.05$)[17] were used for statistical analysis. The SPSS 1.0 version was used for this purpose.

Table 3: Two - tailed t test between cases and controls for the biochemical and anthropometric indices.

Variables	Lower limits for IDA	Average Cases – Controls	Standard deviation	P value
Hb He (gr/dl)	<11	9.9 – 12.7	.784	<0.001
Hb La (gr/dl)	<11	9.8 – 12.6	.803	<0.001
Ht (%)	<33	30.9 – 37.1	2.422	<0.001
MCV (Fl)	<70	63.1 – 76.4	5.021	<0.001
MCH (pg)	<23	19.2 – 25.9	2.157	<0.001
MCHC (g/dl)	<30	27.5 – 33.7	1.743	<0.001
ZPP (μmol/dl)	>75	175.4 – 70.2	7.06	<0.001
Ferritin (μg/l)	<10	11 – 24.2	4.956	<0.001
Sfe (μg/dl)	<20	32.9 – 78.4	17.69	<0.001
TIBC (μg/dl)	>400	351.7 – 275.5	45.58	<0.001
TransSat (%)	<14	9.4 – 28.4	5.962	<0.001
Weight (gr)	-	11,170 – 11,578	767.73	<0.001
Height (cm)	-	81.6 – 82.2	1.804	0.007
Head circumference (cm)	-	47.1 – 47.1	.741	0.828
Weight scale (%)	-	37.1 – 54	30.14	<0.001
Height scale (%)	-	41.9 – 51.4	29.04	0.006

IDA=Iron deficiency anemia, Hb He=hemoglobin Hemoque, Hb La=hemoglobin laboratory, Ht=hematocrit, MCV=mean corpuscular volume, MCH=mean corpuscular hemoglobin, MCHC=mean corpuscular hemoglobin concentration, ZPP=zinc protoporphyrin, SFe=serum iron, TransSat=transferring saturation, TIBC=total iron binding capacity.

Children that were not residents in the study area, children coming from economic immigrant families, children with active infection, and children with a history of blood disease such as thalassaemia or a known history of stigma in the family were excluded from the study.

Results

Seventy-five children (7.99%) out of 938 children participated in the study had IDA and 20 children (2.13%) were carriers of b-thalassaemia (Table 2).

A. The analysis of the continuous variables with t-paired test and Wilcoxon test revealed the following:

The differences between the groups of cases and controls concerning the values of Hb-He and Hb-La were strongly significant ($p<0.001$) indicating that the choice of the photometric mobile method of the Hb count is very reliable (Table 3).

Table 4: Two - tailed t test between cases and controls for the ratio rooms/members leaving in the house.

	Average	Standard deviation	P value
Ratio rooms/members			
• **Cases**	1.34	$\pm$ 1.03	0.012
• **Controls**	1.66		

The differences for each variable separately between cases and controls for the values of Hct, MCV, MCH, MCHC, ZPP, Sfe, SF, TIBC and TS are also statistically very significant (p <0.001) which confirms the diagnosis of IDA (Table 3).

Concerning the anthropometric indices, the highest statistical difference between the two groups was observed with respect to the weight of the children with p value <0.001. The height also differentiates the two groups in a statistically significant way (p < 0.003). By contrast the head circumference does not seem to differentiate (p = 0.82) the two groups of children (Table 3).

Regarding the ratio of the rooms of the house to the household members, the ratio was significantly higher (p=0.012) in the control group than in the case group (Table 4).

B. The analysis of the qualitative variables of the questionnaire with Mc Nemar test and the test of symmetry reveals the following for each variable:

Number of members leaving in the house: the families of the cases are more crowded than the families of the controls (p=0.016) (Table 5a).

Number of the children leaving in the house: the families of the cases have 3 or more children leaving in the house than the controls (p<0.001) (Table 5a).

Order of birth for the participating child among its siblings: the cases have a reverse order of birth among their siblings than the controls (p<0.001) (Table 5a).

Education of the parents: in the case group parents have had either primary or secondary education. On the contrary most of the parents in the control group have had technical or university education (fathers p<0.001, mothers p<0.001) (Table 5a).

Occupation of the parents: most of the parents in the case group are farmers or manual laborers unlike the parents of the control group who are either state employees or self employed (fathers p=0.006, mothers p<0.001) (Table 5a).

Source of drinking water: almost all the cases drink either continuous or interrupted tap water. On the contrary most of the controls drink bottled water (p<0.001) (Table 5b).

Sewage system: all the controls leave in areas with sewers. A number of the cases leave in areas with cesspools in the houses (p<0.001) (Table 5b).

Other income in the family: There was not any significant difference between the two groups (p=0.86) (Table 5b).

Child's health status according to the mother: Mothers of cases estimate their children's health status as of a lower level that mothers of controls do (p<0.001) (Table 5b).

Frequency of seeking pediatric care: cases were visiting the pediatrician mostly when they were sick while the controls visited the pediatrician on a regular basis (p=0.02) (Table 5b).

The duration of breast-feeding was logger in the controls than in the cases (p<0.001) (Table 5b).

Milk consumption by the child during the period of the reported research: the consumption of fresh cow's milk vis-a-vis of fortified, was greater in the cases than in the controls (p<0.001) (Table 5b).

Iron intake: there was not statistical difference between cases and controls (p=0.25).

C. Analysis of the qualitative variables of the questionnaire with conditional logistic regression analysis reveals the following:

Number of members leaving in the house: the chances for IDA increase when the members of the family leaving in the house are 5 or more (p=0.002) (Table 6).

Number of children leaving in the house: as the number of children in the family increases the chances for IDA also increase (p<0.001) (Table 6).

Order of birth for the participating child among its siblings: Children who were born later are more likely to present with IDA (p<0.001) (Table 6).

Care taking of the child (mother, grand parents, another person): the chances for IDA increase when the caretaker is not part of the immediate family (p=0.003) (Table 6).

There was no history of b-thalassaemia in the families of both groups.

Discussion

IDA is a common problem for both the developed and the developing world. According to WHO, the prevalence of anemia among children of 0-4 years old in 1980 is as follows: Africa 56%, North America 8%, Latin America

Table 5a: McNemar and test of symmetry analysis of the environmental factors between cases and controls.

Variables	**Cases**		**Controls**		**P value**
Members in the house					
• **3-5**	45 (60%)		59 (78.7%)		
• **5+**	30 (40%)		16 (21.3%)		0.01
Number of children					
• **1**	10 (13.3%)		34 (45.3%)		
• **2**	28 (37.3%)		30 (40%)		
• **3+**	37 (49.3%)		11 (14.7%)		<0.001
Child's birthrate					
• **1st**	11 (16.7%)		32 (42.7%)		
• **2nd**	34 (45.3%)		33 (44%)		
• **3rd+**	30 (40%)		10 (13.3%)		<0.001
Parents' education	Mother	Father	Mother	Father	
• **primary**	16 (21.3%)	19 (25.3%)	1 (1.3%)	8 (10.7)	
• **secondary**	38 (50.7%)	38 (50.7%)	16 (21.3%)	18 (24%)	
• **higher**	18 (24%)	14 (18.7%)	29 (38.7%)	22 (29.3%)	<0.001 (Mother)
• **university**	3 (4%)	4 (5.3%)	29 (38.7%)	27 (36%)	<0.001 (Father)
Profession of the father	Mother	Father	Mother	Father	
• **farmer**	25 33,3%)	36 (48%)	8 (10.7%)	13 (17.3%)	
• **handworker**	20 (26.7%)	20 (26.7%)	22 (29.3%)	10 (13.3%)	
• **state employee**	6 (8%)	8 (10.7)	29 (38.7%)	28 (37.3%)	
• **business**	19 (25.3%)	11 (14.7%)	9 (12%)	24 (32%)	<0.001 (Mother)
• **housework**	5 (6.7%)		7 (9.3%)		<0.001 (Father)

26%, Eastern Asia 20%, North Asia 56%, Europe 14% and Oceania 18%. Around 43% of the world's children (12% in developed countries and 51% in developing countries) suffer from anemia[18-23].

Turkey possesses the greatest percentage of IDA among the Mediterranean countries: 44% of the children between 6 and 24 months of age has Hb values <11gr/dl[24]. In Morocco 35.4% of children 6 months to 5 years of age were found with Hb values below the normal limit[25].

It is reported that the prevalence of IDA in the developed countries has fallen under 20% in comparison to 70% of the developing countries [26]. The 9.4% of iron deficiency and 4.3% of IDA that was reported in Spain has been attributed to insufficient iron diet[27]. In Britain the prevalence of IDA in childhood varies widely depending on the socio-economic status, culture and nationality of the population, i.e. for Pakistanis it is 29% while for Indian 20%[28]. In Ireland, the prevalence of IDA is reported as 2.6% in children aged 12 months and 9.2% in children aged 24 months[29].

Regarding Greece, the prevalence of IDA is reported in the following cross-sectional studies: For the prefecture of Rodope in North-Eastern Greece, IDA was found to be 15.2% and iron deficiency 32.8% in children 2-24 months of age[11]; for the prefecture of Achaia in Southern Greece, in children of 6 months to 9 years of age, it is reported that 18% suffer from IDA and 57% from iron deficiency[12]; for the prefecture of of Thessaloniki in Central Macedonia, Greece, in children 5 months to 6 years of age it is reported that 20% suffer from IDA and 39% from iron deficiency[13].

In the present study almost one-third of the total children population of the specific area was studied (Tables 1 and 2). The percentage of IDA (7.99%) was lower in

Table 5b: McNemar and test of symmetry analysis of the environmental factors between cases and controls.

Variables	Cases	Controls	P value
Source of drinking water			
• **tap water**	49 (65.3%)	41 (54.7%)	
• **interrupted**	25 (33.3%)	18 (24%)	
• **bottle water**	1 (1.3%)	16 (21.3%)	0.003
Sewage system			
• **sewer**	65 (86.7%)	75 (100%)	
• **cesspool**	10 (13.3%)	0	<0.001
Child's health status			
• **Healthy**	33 (44%)	57 (76%)	
• **Invalid**	18 (24%)	14 (18.7%)	
• **Weak**	24 (32%)	4 (5.3%)	
• **Total**	75	75	<0.001
Pediatric care			
• **Regularly**	27 (36%)	41 (54.7%)	
• **In sickness**	48 (64%)	34 (45.3%)	0.02
Breast-feeding duration			
• **Not at all**	35 (46.7%)	5 (6.7%)	
• **3 months**	26 (34.7%)	18 (24%)	
• **6 months**	11 (14.7%)	39 (52%)	
• **6+ months**	3 (4%)	13 (17.3%)	
• **Total**	75	75	<0.001
Milk during the study time			
• **Fresh cow's milk**	71 (94.7%)	7 (9.3%)	
• **Fortified**	4 (5.3%)	68 (90.7%)	
• **Total**	75	75	<0.001

comparison to the previous studies (Table 2). This significant deviation in the percentage of IDA may be due to the design and implementation of the present study or to the more widespread consumption of iron fortified foods in combination with the intake of medical iron preparations that has become much more widespread during the last 15 years.

The mobile Hb estimation by finger prick is not a common practice in Greek pediatric care. However it applies widely in public health systems in many countries since it has been proved reliable, quick and easy to use[8-10]. The present study represents the first time where it was applied for estimating anaemia in a sample of this size and for the particular age of the population although it was first used by Columbia University researchers in a field study in Crete in 1962. The repetition of the Hb estimation in the laboratory for both groups, with intravenous blood, provided the cross validation of the values reported by the mobile Hb estimation (Table 3).

The Hb concentration and the Hct are the main criteria widely used for estimating the sufficiency of iron nutrition in infants and children[15,30-32]. Although the quick estimation of Hb and Hct have been accepted as a very efficient methods for the prognosis of IDA[33], they lack sen-

Table 6: Conditional logistic regression analysis of environmental factors between cases and controls.

Group	Odds ratio	Standard error	P value	95% confidence interval
Members 5+	0.45	0.16	0.02	0.19-0.87
Number of children 3+	2.84	0.73	0.001	1.71-4.7
Birthrate 3rd +	2.39	0.58	0.001	1.43-3.84
Care taking				
• **grandmother**	0.51	0.23	0.14	0.21-1.23
• **another person**	0.33	0.17	0.03	0.11-0.89

sitivity and specificity for the IDA diagnosis and they fail to identify patients with iron deficiency but no IDA[34].

The indices MCV, MCH, and MCHC are useful for the diagnosis of ID[11,29,31]. Determination of concentration of Sfe and TIBC constitutes the classical and established methods used for the diagnosis of ID[15,30,31,35,36]. The use of the transferin saturation percentage (serum iron divided by TIBC), which characterizes the transferin percentage occupied by iron atoms, represents the relation between iron sufficiency and bone marrow requirements**.** Approximately 20% of the iron in the body is stored in the tissues mainly as ferritin and haemosiderin. The concentration of serum ferritin in normal conditions is directly proportional to the iron stores available in the organism[11,37]. The last step in the bio-composition of heme is the incorporation of iron into the ring of protoporphyrin-9. In the case of complete or relative iron deficiency, the space usually occupied by it, remains available and so, within the ring of protoporphyrin, zinc is incorporated with the formation of zinc protoporphyrin (Table 3)[31,35,38,39].

In the case of the present study, all of the above blood tests indicated that the anemia was due to iron deficiency.

In Greece the most common condition, which should be differentially diagnosed from IDA, is the simple form of b-thalassaemia. The carriers of this disorder constitute about 8% of the Greek population, with a fluctuation from region to region between 2 and 15%[40]. The differential diagnosis for the carriers of b-thalassaemia is based upon the normal levels of protoporphyrin and ferritin unlike those of MCV, which are always low, and especially in electrophoresis of Hb[31,36,41,42]. In the population sample that we studied, the percentage of the carries was 2.13% (Table 2).

The significant ($p<0.001$) statistical difference found for each of the blood tests used between the two groups, confirms their effectiveness to differentiate between cases and controls, while it confirms the reliability of the mobile counter for the diagnosis of IDA (Table 3).

The anthropometric indices of weight and height imply the existence of significant statistical differences ($p<0.001$ and $p=0.007$ respectively) that extends to the percentiles ($p<0.001$ and $p=0.06$ respectively) between the two groups, thus confirming (Table 3) the significance and the consequences of IDA[16]. It is noted that a number of studies support a strong relation between nutritional iron intake and weight and height development in childhood[43-46].

In the present study the size of the household and of the family apparently increases the difference between the children with IDA and them with normal blood levels. A reasonable explanation is the apparently lower iron intake in the crowded families along with a greater exposure to infections and parasitic infestations[47] (Table 5a).

The chances for IDA increase as the number of the children in the family or the order of birth for the participating child among its siblings increase. The appearance of the problem is possibly related to the variance of the daily-recommended iron intake and the generally poor nutrition[48,49] (Table 5a).

Both the educational level and the profession of either father and/or mother affect to the same degree the presence of IDA, possibly by influencing the general socioeconomic conditions of the family and consequently the nutrition and development of the child. Several studies have shown that the education level, especially of the mother, has a direct relationship to the iron intake and iron deficiency[50,51]. Similarly occupations that are associated with lower incomes affect the child's nutrition[52,53] (Table 5a).

As far as the care of the child, (by the parents, the grandparents, or other persons, especially when the mother works), it is noted that although there are no differences between the cases and the controls in developing IDA, the chances for IDA increase when the care taker of the child is another person, besides the mother or the grandparents (Table 6). This is probably due to the poor nutrition of the child, resulting mainly from negligence on the part of the caretakers their ignorance or lack of concern about the iron needs of children or their requirements for a healthy nutrition[54].

The source of drinking water is a variable, which differentiates the two groups. Most of the cases used to drink tap water, sometimes provided with interruptions during the day, unlike to most of the controls that drank bottle water. It has been reported that the relationship between the quality of drinking water and the intestinal infections affects the iron levels in the body[55] (Table 5b).

The sewage system of the households that participated in the present study appears to affect the IDA incidence. Children of a few families that are part of the case group were using cesspools (Table 5b). Although there is a lack of reports on cesspool use in Greece, the effect on IDA incidence could be attributed either to intestinal infestations or to a general poor socio-economic condition of the family that cesspool use in Greece usually indicates.

The child's health status is a significant index of IDA[56,57]. In the present study, the cases are considered by their mothers as more prone to disease than the control group (Table 5b).

The frequency of asking pediatric care is an index of developing IDA[34]. The present study reveals that children visiting the pediatrician regularly are less likely to develop IDA than those that request medical care only when sick (Table 5b). It should be noted that all of the families that participated in the study had easy access to the health care.

In the current study, the children that were breast-fed from 3 to 6 months or more had higher iron levels than those that were not breast-fed (Table 5b). Breast milk as well as the complete cow's milk is usually regarded as poor source of iron, containing 0.3 and 0.8 mg/l respectively. Nevertheless the low content of iron in breast milk is offset by its very high absorption rate by the child's organism. It is estimated that around 50% of the iron in the human milk is absorbed versus only 10% of the iron in the cow milk[29,58].

The study results reveal that all the children of the case group were drinking fresh cow's milk. In contrast, most of the children in the control group were drinking fortified milk (Table 5b). It seems that the type of milk constitutes a serious forewarning index of IDA (Table 5b). Cow's milk interferes with the absorption of the food iron and may aggravate the iron deficiency due to the minor bleeding of the intestinal mucus that it causes. Iron absorption out of fortified milk is about six times greater[59,60] than out of cow milk or of non-fortified milk. Fortification with 15 mg of iron and 100 mg of folic acid per liter of milk has proven very effective in preventing iron deficiency[29,59].

The present study proves the participation of all of the environmental factors that were discussed in the development of IDA. In Greece there is a lack of similar studies that investigate the relationship between IDA and the environment, while popular beliefs about the role of environmental factors in the development of IDA complicate the problem. It is reported that countries who have significantly improved their socio-economic status, did not manage to wipe out or proportionally decrease the incidence of IDA[61,62]. This contradiction may be due to the coexistence of two different conditions: On the one hand the existence of an adequate iron supply to the children and on the other the existence of poor absorption of the available iron supply by the children's organism. Meeting these conditions and determining the factors that influence them are important because failure in any one of them could lead to anemia.

Conclusions and Recommendations

Although the prevalence of IDA in this area of Greece is similar to that in most of the developed world, it still constitutes a public health problem. The mobile method for Hb estimation should be introduced and widely used in Greece, especially since its reliability to detect IDA has been adequately established and reconfirmed by the present study. The application of simple questionnaires for the detection of the environmental IDA risk factors could improve prognosis and prevent anemia. Different approaches have been used to overcome the problem of anemia. The major approaches mentioned are: provision with medical treatment, fortification of foods with iron, dissemination of information on the importance of a healthy and balanceed diet and control of virus epidemics and parasitic infestations[63,64].

Further improvement of the IDA status in Greece might be achieved through continuous information dissemination about iron rich foods and iron absorption by the human body, the improvement of environmental conditions and the application of reliable, easy to use and cheap methods for Hb estimation.

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