CHAPTER 1

Introduction and Outline of the Thesis
1. **JUSTIFICATION**

1.1. **Iron and vitamin A deficiencies**

Micronutrient deficiencies are highly prevalent and affect millions of people worldwide, in particular in South and South East Asia (1,2). Anemia is the most widespread deficiency, largely due to iron deficiency, and often aggravated by concurrent vitamin A deficiency (VAD) (3-7). Despite many efforts, little sign of improvement in anemia prevalence has been seen lately (8).

The main direct cause of iron and vitamin A deficiency is inadequate intake of bioavailable iron and vitamin A. The main staple in many South-East Asian countries, as in Indonesia, is rice which has low iron content and is rich in phytate, which reduces iron bioavailability. The consumption of animal products, a good source of iron with relatively high bioavailability, is low. In addition, other factors play a significant role, such as excessive blood loss, infections, and deficiencies of other complementary micronutrients that facilitate the physiologic functions of iron and vitamin A (7,9).

Consequences of anemia include irreversible damage to the physical and mental development of young children (10,11), impaired immune function (12-14) and reduced work performance (15-17), as well as increased peri-natal and maternal morbidity and mortality (18), the exact mechanism of which is still not entirely clear (19). Anemia can induce maternal and fetal stress, which stimulates corticotrophin releasing hormone (CRH) synthesis. Elevated CRH concentrations in turn are a major risk factor for preterm labor, eclampsia, and premature rupture of the membranes (19). In addition, iron deficiency may lead to increased oxidative damage of the fetus and placenta, and the risk of maternal infections (19).

VAD also has detrimental effects on morbidity and mortality (20), not only directly but also partly by contributing to the development of anemia (5,7).

1.1.1. **Anemia and Vitamin A deficiency in Indonesia**

Anemia is widespread in Indonesia. The 1992 nationwide household survey found that 63.5% of pregnant women and 55.5% of underfives suffered from anemia. In 1995, the prevalence among pregnant women, underfives and female workers was 50.9%, 40.5% and 30%, respectively (21). On the whole, as many as 50-70 million people in the country are estimated to suffer from anemia (22). Iron deficiency is indicated as the most common cause of nutritional anemia among pregnant women and preschool children (22). The IDA (iron deficiency anemia) control program consists chiefly of provision of iron/folic acid tablets (each containing 60 mg iron and 0.25 mg folic acid) to pregnant women throughout pregnancy, but coverage is only about 60% (22).

Vitamin A deficiency (serum retinol concentration <0.70 μmol/L) is still prevalent in
underfives (23). It is also found among women of reproductive age, in particular during pregnancy and lactation (24-26), but also among adolescents (27,28). Moreover, there was an increase in the prevalence of nightblindness among young children and women of reproductive age caused by the economic crisis that started in 1997 (29).

Combating these deficiencies is a priority in Indonesia as reflected in the national five year plan for 1994-1999 (Repelita VI) which aimed to eliminate the vitamin A deficiency problem and to reduce the anemia prevalence in pregnant women from 63.5% to 40%, in underfives from 55.5% to 40% and in low income workers from 30% to 20% (30). The target for the end of the second Long Term Development Plan (PJP II, 1993-2018) is a reduction in the prevalence of anemia to 9% in pregnant women and to 10% in both underfives and female workers (21).

1.2. Current approaches

Although improvement of habitual diets through nutrition education and food fortification is the ultimate goal, the success of nutrition education can only be expected in the far future as it is highly dependent on other factors such as availability of food at the household level and intra-family food-distribution (31). Food fortification is an intermediate option with constraints in the field of marketing and acceptability. Supplementation is the best option when prevalence of deficiency is high and nutritional requirements are difficult to fulfill through the habitual diet.

1.2.1. Existing interventions

At the time the studies described in this thesis were designed (1995-1996), micronutrient interventions were focused on iron supplementation for those with the highest needs: underfives and pregnant/lactating women. Iron supplementation, usually combined with folic acid, for all pregnant women was introduced in most countries in the 70s and 80s, including in Indonesia (1974), but without much impact (32).

Reasons for this relative failure include health services-related factors (insufficient availability and accessibility, inadequacy of distribution and availability of tablets, high cost of health services or transport), client-related factors (insufficient knowledge and awareness among the target population of the importance of iron supplementation, low compliance related to gastro-intestinal side effects), biological factors (inadequate daily dosage in the presence of inhibitors of absorption, iron depletion at the time of conception leading to severe anemia in pregnancy), lack of monitoring and evaluation of programs, and other causes for anemia such as vitamin A deficiency and infections (32-35).

Less experience has been accumulated with vitamin A supplementation during pregnancy, partly because of the teratogenic effects of high doses of vitamin A. It is, however,
now considered safe to give low-dose vitamin A supplementation (10,000 IU daily or 20,000 IU once a week) to pregnant women (36,37).

Distribution of high-dose vitamin A capsules to underfives was introduced in Indonesia in 1974 (32), and other countries followed. The program was extended to post-partum women from the early 1990s (38). Studies on iron supplementation for underfives were performed (39,40), but no programs were put in place yet.

1.2.2. Daily vs. weekly iron supplementation

The optimal frequency for iron supplementation is still being debated (41,42). In a recent review, it was concluded that daily and weekly supplementation are both efficacious but neither has been very effective (43). The authors maintained that daily supplementation is indicated in pregnancy, whereas in other target groups (in particular school-aged children) supervised weekly supplementation could be the most suitable approach because of higher compliance and lower cost of supplements and logistics.

1.3. Adolescents as a target group

The prevalences of anemia and vitamin A deficiency are highest among pregnant women and underfives (8). However, the pubertal growth spurt is the period of most rapid growth after infancy that the human being experiences (44), which greatly increases physiological needs for micronutrients, such as iron and vitamin A (45,46). Among girls, menstruation further increases iron requirements. The few studies involving adolescents report high prevalences of anemia and vitamin A deficiency (27,47,48). These deficiencies, specifically of iron, have important negative impact on the health and development of adolescents by hampering their capacity to grow (49), learn and earn (50).

Considering the fact that the main cause of iron deficiency anemia in pregnancy is marginal iron status in women of reproductive age prior to conception, the appropriate alternative approach would be to improve iron status prior to pregnancy, and to continue supplementation during pregnancy (51). Thus, not only would this be beneficial to adolescents, but it would also contribute to healthy pregnancies and thus to a decrease in neonatal and maternal morbidity and mortality (18).

Also in terms of “nutrition economics” a focus on adolescents makes sense. Although it seems logical to target supplementation to population groups with the highest prevalence, it is not likely that this approach can be sustainable if the inflow of anemic primigravidae continue to increase through population growth. This issue can only be addressed if the prevalence of micronutrient deficiencies among the age group just prior to maximum prevalence, in this case adolescent girls, is reduced.

Adolescents also offer a window of opportunity for interventions. Changing pregnant
women's diets is hampered in traditional communities by the many food taboos for pregnant women (52), which generally do not apply to adolescent girls. Changes in habits, both dietary habits and behavior with regard to supplements, may represent the most promising sustainable approach, and introduction of such concepts during adolescence may be particularly effective for making changes in future generations. As adolescence spans several years, weekly supplementation is a promising strategy which has been shown to be efficacious in improving iron and vitamin A status (27,53).

Schools offer an opportunity to reach adolescents and to supply supervised supplementation. In Indonesia, school enrolment is 80% since the implementation of nine years' compulsory education (age range: 6-15 years) (54), while only 50% of pregnant women made the requested four antenatal visits (55). In addition, almost 25% of Indonesian girls marry before the age of 16 (54).

There is, however, a great paucity of knowledge on the nutritional status of adolescents. In their extensive review of anemia prevalence in the world, DeMaeyer and Adiels-Tegman (47) found that only 39 of 325 studies were done in adolescents, and none comprised of more than 5,000 subjects. Furthermore, the information that is available pertains almost exclusively to prevalence of anemia among post-menarcheal girls and the efficacy of interventions among anemic menstruating girls. The only program for reducing anemia among adolescents that we are aware of is the Egyptian government school-based iron supplementation and nutrition education program reaching 70,000 students, started in 1999. Government and schools were fully involved and the intervention was able to reduce anemia rates by 20% (56).

In 1993, USAID developed, initiated and funded the Opportunities for Micronutrient Interventions (OMNI) project (57) to help countries meet the goals set by the 1990 World Summit for Children, the Ending Hidden Hunger Conference in 1991 and the International Conference on Nutrition in 1992 of virtually eliminating, by the year 2000, both vitamin A and iodine deficiency and their related consequences and reducing by one-third the 1990 levels of iron deficiency anemia in women. The project's objective was to develop comprehensive and integrated micronutrient strategies and programs, and Helen Keller International/Indonesia, as one of the partners in this project, proposed to evaluate the effectiveness of supplementation on anemia among adolescents.

2. OBJECTIVES

These studies in an adolescent population (enrolment age 12-15 years) in East Java, Indonesia, were designed to assess the effect of supplementation with iron-folate, vitamin A,
or both on anemia and vitamin A status. In addition, the underlying causes of poor nutritional status were investigated. More specifically, this project aimed to:

1. Determine the prevalence of anemia and vitamin A deficiencies among adolescents in rural and urban communities
2. Assess underlying factors of anemia and vitamin A deficiency in female and male adolescents in rural and urban communities
3. Test the effectiveness of iron-folate and vitamin A given alone or in combination on reducing anemia and vitamin A deficiency
4. Assess the appropriateness of the school as a channel to address the nutritional problems of adolescents

3. STUDY DESIGN AND METHODS

The preparations for the GIRLS (Gizi: Intervensi Pada Remaja Lewat Sekolah; School-based Nutrition Interventions for Adolescents) project started in January 1996 and field activities commenced in August of the same year. The project ended on 31 July 1999. All intervention and data collection activities were implemented during the school year and schedules were adapted to the schools' schedules.

3.1. Location
3.1.1. Indonesia

The Republic of Indonesia is an island nation with over 18,000 islands, of which approximately 6,000 are inhabited. With a surface area of almost 2 million square kilometers and a population of over 220 million people in 2002, it is the world's fourth most populous country. Life expectancy at birth is 62 years, with infant mortality 71/1000 (58) and the population increases at an annual rate of approximately 2.5%. About 75% of the population lives in the rural areas, with agriculture as the main source of income. The normal diet in most areas is rice-based and low in animal products, and in the case of Madura, also low in dark green leafy vegetables. School enrolment until the age of 16 is 80% owing to the government policy of nine years’ compulsory education. (54)

3.1.2. Study location

The studies were implemented in 24 junior high schools and 10 boarding schools (pondok pesantren) in urban Surabaya, the provincial capital of East Java, and the two westernmost districts of the rural island of Madura: Bangkalan and Sampang. (figure 1)
3.1.3. Schools

The main types of school available in Indonesia for the 12-15 year age group were covered by this project: three-grade junior high schools (both state- and privately run, and both general and Islamic) and Islamic boarding schools or *pondok pesantren*. In this document the latter term is used, because there is no equivalent found outside this particular cultural context. The term *pondok pesantren* is applied to a heterogeneous group of schools, which have in common that the pupils live in the school, and that a strong emphasis is laid on Islamic religious education. Most *pondok pesantren* offer some degree of general education as well, but some focus entirely on religion. They are privately owned and run by religious leaders, and largely independent of government regulations. Some *pondok pesantren* use a class-system like general schools but others apply a more individual approach to teaching in which students have to complete a given number of books and the duration of their education depends largely on their diligence. These characteristics made the *pondok pesantren* less appropriate for the supplement distribution studies which were randomized per class, but more appropriate for a dietary intervention because students all board and kitchens are available at the schools (which was conducted (60) but is not described further in this thesis). Thus, pupils from *pondok pesantren* were included in the anemia survey described in chapter 3, but not in the other studies.

3.2. Study population

Although the main interest of the project was in girls, the school setting provided the additional advantage of being able to collect data on adolescent boys, a group that has received even less attention than their female peers (47). Thus, the population consisted of girls and boys between the ages of 12 and 15 at enrollment, whose parents gave written
Figure 2. Diagram of the three-year study design
consent for participation in the activities.

3.3. Interventions

In 24 junior high schools, randomized weekly supplementation trials with iron/folate and/or vitamin A were performed as described in this thesis. The folate was included in the tablets because the supplementation was designed as a reproductive health effort. Studies were performed during three consecutive school years, which at the time lasted from August through June with a one-month holiday in July. The studies in the second and third years were follow-up studies based on lessons learned in the field and initial data analysis of the preceding year(s).

The original design consisted of a comparison between nutrition education, dietary intervention and supplementation with iron and/or vitamin A. The dietary approach was tested in five boarding schools only as these were more suited for this approach. The results of this intervention will be reported elsewhere (60). Unfortunately, the formative research necessary for the design of the education component was not finalized in time to include this component. Initially, this led to a blank control group of the first intervention study (detailed description in chapter 4), but when it became clear there was no prospect of developing and implementing this component, the blank control group was changed into a placebo group (second and third studies, described in chapters 5 and 6).

The supplementation studies were implemented during three consecutive school years (1996/1997 – 1998/1999) (Figure 2). Baseline data were collected at the start of the project (Oct-Dec 1996). Because of the school setting, each year the new students in grade 1 were enrolled in the studies, and baseline data were collected on these subjects at the start of their first year at school (Aug-Sep 1997 and Aug-Sep 1998). Post intervention data for the three studies were collected between Mar-May 1997, Mar-May 1998 and Mar-May 1999, respectively (Figure 3). These data were also used as baseline data for ‘old’ students in the subsequent studies. Obviously, students who graduated were not enrolled in the subsequent year’s study.

In the second study, despite having moved up a grade, subjects received the same type of supplementation as during the first study, although subjects and field workers were not aware of this; while the new pupils in grade 1 received the same supplementation as the former third grade pupils of the same school.

The study in the last year on the effect of enhanced nutrition education was different, and the third-grade pupils in the urban schools were re-randomized for the iron/folate trial. This approach was based on the observation that the risk of being anemic after the second intervention was higher in urban subjects, and on the assumption that compliance for iron was poor, especially in the urban schools, among the pupils who had participated in the first
rounds of supplementation. (This later turned out to be an erroneous conclusion, as in fact the urban subjects had complied relatively well compared to the rural subjects). Therefore, it was decided to implement a placebo-controlled iron supplementation trial in the third grades of the urban schools, with intensified supervision and interactive classroom activities aimed at raising awareness of the need for iron supplementation and creating better rapport with the pupils (third study; detailed description in chapter 6).

3.4. Data Collection Methods and Moments

Trained field workers (the majority of whom were nutritionists and nurses) collected data on socio-economic background and health status of all new enrollees (data collection moments 1 and 3; figure 3), using a standard questionnaire. In addition, anthropometric measurements were taken and hemoglobin concentration was assessed in peripheral blood by HemoCue™. A subsample was randomly drawn from all grades in all schools for collection of venous blood for HPLC analysis of serum retinol concentration. After each intervention, all data were collected again, except those on socio-economic background. In addition, self-reported compliance for the supplementation was recorded for a subsample of pupils. During the third study, observed compliance was recorded weekly by the field workers.

3.5. Sample size determination for biochemical subsample

Although the subsample was used to determine retinol status, no data were available on prevalence of vitamin A deficiency in Indonesian adolescents. Therefore, the power calculation was based on an assumed anemia prevalence of 30% (Schultink 1995, personal communication). To allow detection of a 50% reduction in this prevalence, and accounting for 25% drop-out from the study, it was calculated that 168 subjects in each cohort were needed to obtain significance at the p<0.05 level, and 80% power. This was rounded to 15 subjects.
per grade in the supplementation schools (15 schools with 3 grades each; n=225 per group) and 15 per school in the control schools (9 schools with 3 grades each; n=405) for the first study (total 630). Of the 764 samples collected at baseline (Control (C) 164, Vitamin A (VA) 204, Iron (Fe) 192, Vitamin A and Iron (VAFe) 204), 625 were included in the analyses; C 128, VA 175, Fe 154, VAFe 168 (DO 139=18.2%; mostly due to absence/refusal of second sample). For the second study, based on the results of the previous study, the anemia prevalence was presumed to be 25%. Allowing detection of a 50% reduction, while accounting for 20% drop out, it was calculated that 184 samples per cohort (total 736) were needed to obtain significance at the p<0.05 level and with 80% power. Thus, at least 10 subjects per grade in each of the 24 schools were included in this subsample. In this study 792 samples were collected, of which 726 were used in the analyses (Placebo (P) 147, VA 195, Fe 176, VAFe 208) (DO 66=8.3%; mostly due to absence/refusal of second sample).

3.6. Indicators for puberty and age

Determining pubertal status is crucial in studying an adolescent population. The Gold Standard for determination of pubertal stage is the method developed by Tanner, which involves physical examination and exact determination of stages of breast development, testicular size and pubic hair (61,62). However, using this method is not possible in field studies in many non-western societies. Therefore, subjects were asked whether they had had their first menstruation or nocturnal ejaculation, respectively. The validity of these indicators is assessed in chapters 2 and 3.

Determination of exact age is another challenge in many settings. In Indonesia, a birth certificate is required to enter formal education and schools have records of official dates of birth of all pupils. However, in some cases these official dates differ from the actual date of birth, for various reasons. In the studies described here, at each data collection moment, all pupils were asked their date of birth and their answers were checked against the school records. If there were discrepancies between their answers at different moments and the school’s records, the most frequently given answer was used. If all answers were different from one another, the individual was excluded from data analysis. If there was no date of birth in the school’s records and the date was not known (but the month and year was), it was recorded as 15. If both date and month was not known (but the year was), it was recorded as 15 June. The inaccuracy introduced by this procedure is acceptable in this age group.
CHAPTER 1

4. OUTLINE OF THIS THESIS

Justification and objectives of the studies have been discussed in this chapter.

To validate the interview method of maturity rating as used in the studies, in chapter 2 an assessment was made of this method against anthropometric indicators of growth and maturity.

In chapter 3 the determining factors of anemia in adolescent girls and boys aged 12-15 years are identified.

Chapter 4 presents the results of the first intervention study, consisting of weekly supplementation with 10,000 IU vitamin A, 60 mg elemental iron, or both, in relation with hemoglobin and serum retinol concentrations.

The second intervention study is reported on in chapter 5, which presents the effects on anemia and serum retinol concentration of placebo-controlled weekly supplementation with 20,000 IU vitamin A and sugar-coated tablets containing 60 mg elemental iron, or both.

Chapter 6 reports the results of the last study, which assessed compliance of urban boys and girls to iron supplementation when they are given more supervision and motivation, and the intervention’s impact on hemoglobin concentration.

In chapter 7, the literature on interventions for anemia among adolescents in developing countries is reviewed, including the studies described in chapters 4-6.

Finally, in chapter 8, the results of the studies are interpreted from a scientific and programmatic perspective.

REFERENCES


60. Sari M, de Pee S, Yip R, Martini E, Sugiatmi, Soekarjo D, Bloem MW, Muhilal. Foods naturally rich in iron increase hemoglobin concentration among anemic adolescent subjects

