Plasmodial pigmentation of placenta and outcome of pregnancy in West African mothers

M WATKINSON, D I RUSHTON

Abstract

In a region where falciparum malaria is endemic and where pregnant women traditionally receive only curative treatment for parasitaemias and no chemoprophylaxis 65 placental biopsy specimens were examined histologically for malaria pigment. Twenty seven placenta had pigment, but parasitaemias had been diagnosed antenatally in only 12 of these women despite their frequent attendance at antenatal and other clinics. The incidence of parasitaemia in pregnant primigravidas was 17.7%, seven times greater than that in lactating primiparous mothers; pregnant primigravidas also had the highest incidence (67%) of pigmented placentas. First born babies with pigmented placentas had a mean (SD) birth weight of 2580 (260) g, significantly less than the 3150 (400) g of unaffected first babies. All babies weighing less than 2500 g at birth had pigmented placentas.

Pigmentation was associated with parasitaemias in the second half of pregnancy, and, although some recovery from early parasitaemias may occur, the fetoplacental unit is inadequately protected by curative treatment alone. Chemoprophylaxis currently remains the procedure of choice.

Introduction

In any given community many factors influence birth weight. In 1978 Whitehead et al reported a significantly lower mean birth weight in rural Gambian babies born in the rainy agri-cultural season than in those born in the dry months of December to June, when there is little agricultural work to be done. The dietary intake and energy expenditure of these mothers correlated significantly with the variation in birth weight, but the influence of other, unanalysed, factors was recognised, in particular the impact of gestational malaria.

The incidence and effects of falciparum malaria during pregnancy have been extensively reported. The placenta is a privileged site where Plasmodium falciparum parasites may become sequestered and develop into older forms. Such infected placentas have aggregates of reticuloendothelial cells in the intervillus spaces, areas of focal necrosis, thickening of the basement membranes, and loss of syncitial microvilli. One sequela to such damage, which is thought to impair the diffusion of oxygen and nutrients to the fetus, is retardation of intraterine growth. Within given communities babies born with placentas containing parasites weigh a mean of 89 g to 311 g less than their uninfected contemporaries.

Falciparum malaria is reported to be hyperendemic in The Gambia, but in 1978-80 it was not government policy to provide malaria prophylaxis during gestation. The introduction of such prophylaxis in a single village (Keneba) was considered inadvisable by the responsible Gambian committee until a national policy had been implemented. We therefore assessed the effectiveness of the policy whereby pregnant women in Keneba received antimalarial treatment only if they had proved parasitaemia. We studied the prevalence of parasitaemia in pregnant and lactating women in Keneba, the incidence of certain placental pathology related to falciparum malaria, and the relation between this pathology and the outcome of pregnancy.

Methods

Once a pregnancy was confirmed either by examination or urine testing (Prepurex, Wellcome) the mother was seen at least every six weeks at regular antenatal clinics, where she was weighed, measured, and examined. Blood was taken for estimation of haemoglobin and examination of a thick film for malaria parasites. Such films were declared negative only after 100 high power fields had been examined. Plasmodium falciparum accounted for more than 99% of the positive results, and although Plasmodium ovale and Plasmodium malariae were occasionally seen they are not considered further in this paper.
In addition to the antenatal clinics there was a daily open door clinic which women could attend with acute illnesses; thick films were examined for any febrile episode. All confirmed parasitaemias were treated with standard doses of chloroquine. Attendance at the antenatal clinics was excellent, with 547 of 557 requested attendances being fulfilled. Information from these clinics has therefore been used for cross sectional analysis.

From February 1979 to August 1980 a biopsy specimen was taken from each freshly delivered placenta whenever practicable. A 1 cm² full thickness section was taken from the maternal surface and immediately placed in formol saline; it was later examined in England. A blood smear from the maternal side of the placenta was also taken and examined for parasites. Birth weight and placental weight were recorded, and the gestational age of the baby estimated within five days of birth by the Dubowitz score,¹⁹ which is accurate in African babies.²⁰²¹ The weight for gestational age was then calculated and expressed as a percentage of the Aberdeen standards²² after correcting for pregnancy number and sex.

Histological examination of the placenta was performed "blind" by an investigator without knowledge of the mother's health in pregnancy or the outcome of the pregnancy. As a simple screening procedure the placentas were divided into two groups; the affected cases, in which aggregates of macrophages containing malarial pigment in the intervillous spaces were identified; and the unaffected, in which these cells were absent. This criterion was used because the presence of such cells appears to reflect the severity and extent of the infection.¹⁷ Single blocks of placenta cut vertically to the chorionic plate were stained with haematoxylin and eosin, Masson's trichrome, periodic acid Schiff, and Giemsa stains.

Student's $t$ test and the $\chi^2$ test (with Yates's correction where appropriate) were used in statistical analysis.

Results

There were 95 singleton deliveries in the study period. Six mothers, including one who delivered a stillborn infant, were referred for hospital delivery; 15, including one with a stillbirth, had buried the placenta before the midwife arrived, and in nine cases a biopsy specimen was not taken as there had been excessive delay in calling the midwife. Thus biopsy specimens were taken after 65 of the 95 deliveries. Table I shows that these 65 babies did not differ significantly from the 30 babies from whom placental biopsy specimens were not taken, in birth weight, gestational age, or incidence of preterm deliveries. Nor did they differ in the distribution of births by gravidity ($\chi^2=0.47; df=2; p>0.5$) or calendar month ($\chi^2=17.4; df=11; p>0.05$).

Of the 65 biopsy specimens studied, 27 had plasmoidal pigment in them, showing that the mother had had malaria during pregnancy. Table II shows that falciparum parasitaemia had been detected during pregnancy in only 12 of these women, compared with 12 of the 38 women with negative biopsy findings. There was no significant difference in the mean gestations at which pregnancy was diagnosed between the two groups or in the two subgroups of 12 women with parasitaemias. In 9 of the 12 women with parasitaemia but unpigmented placentas, however, malaria parasitaemia had been last detected before 20 weeks' gestation (calculated back from the Dubowitz score), whereas this occurred in only three of the 12 women with parasitaemia and a pigmented placenta ($\chi^2=4.17; p<0.05$). Table II also shows the distribution of the babies by gravidity. Eight of the 12 placenta from primigravidas, nine of the 35 from women of gravidity 2-7, and 10 of the 18 from women of gravidity 8 pregnancy or more had plasmoidal pigment ($\chi^2=8.19; df=2; p<0.02$).

None of the placental smears contained parasites; nor were parasites identified in any of the histological sections.

Table III shows that babies with pigmented placentas had a lower mean weight for gestational age than those without pigmented placentas. When this difference was further analysed by the gravidity of the mother, all groups showed a difference in favour of the non-pigmented group, but the significance persisted only in primigravidas. There were no significant differences between the two groups in the mean gestational ages, the proportion of preterm babies, or the mean placental weights.

Figure 1 shows the mean birth weights in the pigmented and non-

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<th>TABLE I — Details of pregnancies and parasitaemia in mothers with pigmented placentas and those without.</th>
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<td>Mean (SD) gestation at diagnosis of pregnancy (weeks)</td>
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NS = Not significant.

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<th>TABLE II — Details of babies with and without pigmented placentas. Numbers of babies are given in parentheses preceding the value in each column.</th>
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<td>Mean (SD) weight for gestational age (g)</td>
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<td>Mean No of preterm births (before 37 weeks)</td>
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<td>Mean (SD) placental weight (g)</td>
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NS = Not significant.

FIG 1 — Birth weight according to gravidity. Mean birth weight for first-born babies with pigmented placentas was significantly lower than that for babies with non-pigmented placentas ($p<0.01$).
pigmented groups analysed by the gravidity of the mother. The mean birth weight of 3150 (400) g in firstborn babies with non-pigmented placentas was significantly higher than the mean of 2580 (260) g in firstborn babies with pigmented placentas (p < 0.01). These results paralleled those of weight for gestational age, and both analyses showed that babies weighed less with increasing gravidity when the placenta was free of pigment. All nine babies weighing less than 2500 g at birth had pigmented placentas.

Figure 2 shows the incidence of pigmented placentas by calendar month together with the overall prevalence of parasitaemia in all pregnant women attending the regular antenatal clinics. These data are also broken down by gravidity and compared with the prevalence in lactating women attending postnatal clinics up to 18 months after delivery. The data are in two monthly blocks to reduce the variation of small numbers. The prevalence of parasitaemia and the incidence of placental pigmentation peaked in October at the end of the rains (the rains start in May-June and reach a peak in August and September). The pigmentation rate correlated highly with the prevalence of parasitaemia (r = 0.99, p < 0.001), with no time lag.

Table IV summarises the data on parasitaemia in pregnant and lactating women according to gravidity. There were significantly more parasitaemias in primigravidas than in multigravidas during pregnancy (χ² = 23.4; p < 0.001) but not during lactation. There were no significant differences in the prevalence of parasitaemia in the pregnant and lactating groups between women of middle gravidity and those of high gravidity. The third column gives the ratio of the incidence of parasitaemia in pregnant women to that in lactating women. The ratio was highest (7.2:1) and most significant in primigravidas (χ² = 13.2; p < 0.001), less so in women of middle gravidity (ratio = 3.3:1, χ² = 5.95, p < 0.02), and not at all significant in women of the highest gravidity, although malaria was 1-6 times more prevalent in pregnant than in lactating women who had had eight pregnancies or more. Overall, malaria was 3-8 times more prevalent in pregnant than in lactating women (χ² = 20-5, p < 0.001).

**Discussion**

The mothers from whom a placental biopsy specimen was taken did not differ significantly from those who did not undergo biopsy. The results may therefore be considered to represent the general pattern of gestational malaria in Keneba. The village itself, however, had a smaller incidence of malaria than surrounding areas because of the presence of a curative service, which reduced the size of the parasite pool. Thus these results almost certainly underestimate the prevalence and morbidity of malaria in pregnant women in this region as a whole.

The technique we used to divide affected from non-affected placentas was extremely simple and our data suggest that it is an effective discriminator. It is open to certain criticisms in that the samples of placental tissue were small, though they were randomly selected and included the full thickness of the organ. The distribution of cells containing pigment was patchy even in heavily affected cases and some of those classed as negative may have been categorised as affected if more tissue had been available for examination.

Our basic finding—namely, that primigravidas suffer the most pronounced increase in the prevalence of malaria, and that first born babies bear the brunt of placental morbidity with the greatest reduction in birth weight and weight for gestational age—confirms the findings of earlier reports. Unlike previous studies, however, our indicator of placental disease was plasmodial pigment in the placenta and not parasites in placental smears taken immediately after delivery. The complete absence of parasites in our smears was in sharp contrast to the incidences of 15% to 33%, reported in these earlier studies. Parasites were also absent in the histological sections of our placentas. Any recent parasitaemias had presumably been treated and no parasites remained in the placentas, though we cannot state this categorically, because other data in our study show that parasitaemias were not always detected.

As fewer than half the women with pigmented placentas had had parasitaemia detected and treated in pregnancy, many of the malarial episodes must have caused either trivial symptoms or no symptoms whatsoever. Neither the easily accessible daily clinic nor regular antenatal clinics ensured diagnosis and adequate protection against the morbidity of plasmodial infection. The ineffectiveness of such a curative service becomes more apparent when it is further recognised that more than a third of the women with non-pigmented placentas also had malaria. Galbraith et al commented that previous infection may leave a heritage of pigment deposition and placental damage, but the corollary that an absence of pigment means that the placenta is intact from plasmodial damage has yet to be shown.
The presence of plasmodial pigment was associated with parasitaemias in the last half of pregnancy, and nine of the 12 women whose last detected parasitaemia was before 20 weeks had not digested placenta. It may be inferred from this that pigment is "cleared" from the placenta in some cases, although our data do not show that this occurs invariably, and we do not know either the factors responsible for the removal of the pigment or the duration of this process. The fact that the peak monthly incidence of placental pigmentation was synchronous with, rather than lagging behind, the peak incidence of parasitaemia offers further evidence that pigmentation is associated with a relatively recent infection.

As pigmented placentas were related to low birth weights and weights for gestational age whereas non-pigmented placentas, even in women with early parasitaemias, were not it may be only the later parasitaemias and later placentation that cause major morbidity for the fetoplacental unit. Either the immature placenta is less susceptible or recovery from earlier attacks may occur in the last trimester of pregnancy. MacGregor and Avery, reporting on the interruption of malaria transmission in the Solomon Islands, noted, "The adverse effects of malaria... on fetal growth were apparently reversible if transmission of infection... was interrupted as late as the third trimester of pregnancy." Our results, gathered in a different manner, offer some support for this finding.

Although highly gravid women in their eighth pregnancy or beyond had only a slight increase in incidence of parasitaemia, they had a higher percentage of pigmented placentas than women of middle gravidity. This again is contrary to the earlier reports based on placental smears, and the reason why the placental pigmentation appears to produce conflicting results is not clear. Although the presence of malaria pigment was associated with an insignificant reduction in the mean weight for gestational age of 6g, the combination of this and the fall off in weight for gestational age with increasing gravidity (Table 1) resulted in a high proportion of babies weighing less than 2500g. All babies who weighed less than 2500 g or less at birth had plasmodial pigment in their placentae.

Other workers have commented on the incidence of prematurity associated with placental malaria.

The definition of prematurity at the time of those reports was an infant with a birth weight of 2500 g or less. With greater understanding of the low birth weight baby, the definition of a preterm infant is now one born before the 37th completed week of gestation. By this criterion there was no excess of preterm infants in the group with pigmented placentas, and neither this nor the almost identical mean gestational ages of babies in the two groups suggest placental parasitisation as a cause of preterm delivery. This does not necessarily contradict the findings of Gilles et al that a high fever associated with a parasitaemia may precipitate labour.

In a community where a curative service has failed to offer pregnant women and their fetuses adequate protection against malaria and placental parasitisation the introduction of prophylaxis should be considered. Our findings suggest that it should be offered to women of all gravidities, even though parasitaemia rates are low in highly gravid women. Both this study and that by MacGregor and Avery suggest that the crucial period for the protection of the fetoplacental unit is the last trimester of pregnancy. For primigravids at least prophylaxis should be started earlier to prevent the mid-trimester anaemia reported by Gilles et al. Indeed, this study in a community which had an otherwise full antenatal service cannot refute the conclusion of those same authors when they state that the administration of antimalarial drugs prophylactically throughout pregnancy is the single most important medical procedure in pregnancy in a country where malaria is endemic.

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References

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