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Does vitamin A supplementation interact with routine vaccinations? An analysis of the Ghana Vitamin A Supplementation Trial[1–3]

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ABSTRACT

Background: The World Health Organization recommends vitamin A supplementation (VAS) at vaccination contacts after 6 mo of age to reduce mortality. However, it is unknown whether the effect of VAS is independent of vaccinations. One of the original VAS trials from Ghana had collected vaccination information.

Objective: We reanalyzed the data to explore the hypothesis that VAS reduces mortality in children who had bacille Calmette-Guérin or measles vaccine as their most recent vaccine but increased mortality when diphtheria-tetanus-pertussis vaccine (DTP) was the most recent vaccine. On the basis of previous studies, we expected the effects to be strongest in girls.

Design: At enrollment, children aged 6–90 mo were randomly assigned to receive VAS or placebo every 4 mo for 2 y. Vaccination status was assessed at enrollment and after 1 and 2 y by reviewing the children's health cards. Lack of a health card was presumed to mean that the child had not been vaccinated.

Results: VAS had a beneficial effect only in children with no record of vaccination at enrollment (n = 5066); the mortality rate ratio (MRR) was 0.64 (95% CI: 0.47, 0.88) compared with 0.95 (95% CI: 0.72, 1.26) in children with one or more vaccinations (n = 6656). Among vaccinated children, the effect of VAS differed between boys (MRR: 0.74; 95% CI: 0.51, 1.08) and girls (MRR: 1.18; 95% CI: 0.84, 1.67) (P = 0.046 for interaction). VAS had a negative effect in measles-vaccinated girls who were missing one or more doses of DTP at enrollment, a group who often received DTP during follow-up (MRR: 2.60; 95% CI: 1.41, 4.80).

Conclusions: The effect of VAS differed by vaccination status. This is potentially problematic because VAS is provided at vaccination contacts. *Am J Clin Nutr* 2009;90:629–39.

INTRODUCTION

Vitamin A supplementation (VAS) to children aged >6 mo in low-income countries has been estimated to reduce overall mortality by 23–30%. Estimates are based on several meta-analyses of VAS trials conducted in the 1980s and early 1990s (1, 2). Currently, the World Health Organization recommends administration of VAS at vaccination contacts after 6 mo of age (3). However, none of the trials that found reduced mortality after VAS examined the link with vaccination status. Hence, it is unknown whether the effect of VAS on mortality is independent of vaccinations.

We proposed the hypothesis that VAS interacts with the nonspecific effects of routine vaccines (4). This hypothesis was based on the observation that the effect of VAS varied by age group. Most VAS trials that addressed the effect of VAS after 6 mo of age (1, 2) and the first 2 trials that examined the effect of VAS at birth (5, 6) found a beneficial effect of VAS on mortality. On the other hand, all trials that have addressed the effect of VAS given between 1 and 5 mo of age have found no effect or even a tendency for a negative effect (7–13). This mortality-age pattern resembled that seen after routine vaccinations. Our group has consistently found that vaccines, apart from disease-specific effects, may also have so-called "nonspecific effects." The live vaccines bacille Calmette-Guérin (BCG), which is recommended at birth, and measles vaccine (MV), which is recommended at 9 mo of age, are associated with decreased overall mortality (14–17), whereas diphtheria-tetanus-pertussis vaccine (DTP), which is recommended at 6, 10, and 14 wk of age, is associated with increased mortality in areas where exposure to pertussis is limited because of high vaccination coverage (18–20). Intriguingly, both beneficial and negative effects of vaccines have been strongest in girls (21–23).

Because vitamin A may act as an adjuvant (24), we proposed that VAS might amplify these nonspecific effects of routine vaccinations on mortality. Hence, VAS would be additionally beneficial when administered in children whose most recent vaccines were BCG and MV, but potentially harmful in children, whose most recent vaccine was DTP (4). This would explain the mortality-age pattern after VAS. Because the nonspecific effects of vaccines are strongest in girls (21–23), the interaction between VAS and vaccines could be expected to be most pronounced in girls.

Since its initial formulation, data consistent with the hypothesis have been reported (25–28). If the hypothesis is correct, it would have major implications for the current vitamin A policy. We sought opportunities to further explore the hypothesis in already existing data sets. One of the original VAS mortality trials from Ghana (29–32) had collected information on vaccinations at

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the initiation of the trial. We analyzed the data to examine whether the effects of VAS on overall mortality differed by vaccine status and sex. The a priori hypothesis was that VAS would be associated with reduced mortality in children who had BCG or MV as their most recent vaccine, but associated with increased mortality when DTP was the most recent vaccine, and these effects would be most pronounced in girls.

SUBJECTS AND METHODS

Setting and study design

The Ghana Vitamin A Supplementation Trial (VAST) took place in Navrongo, a rural area of northern Ghana, from September 1989 to December 1991. The study area and the study design were described in detail elsewhere (29–32). Briefly, the Survival Study enrolled children aged 6–90 mo. At enrollment, children were randomly assigned to receive 200,000 IU VAS (100,000, 6–11 mo of age) or placebo every 4 mo, in a total of 7 rounds during a period of 2 y. The area was divided in 4 geographic zones used for supervision of the fieldworkers: east, west, north, and south. Randomization was done by clusters of 30–77 compounds. Overall, 92 clusters were assigned to VAS and 93 clusters to placebo. A total of 21,906 children were enrolled.

Age determination

At enrollment, the date of birth of the child was determined from the child's health card if available. Otherwise the date of birth was assessed by means of a detailed local events calendar (29). If no exact date could be obtained, the date was set to day 15 of the month of birth. The date of birth may have been determined more precisely among children with a health card because the date of birth was registered on the card. There were far more children who had day 15 in a month as the date of birth among children without a health card (74%) than among those with a card (42%), and the month of birth was grouped close to the date of the enrollment month or 6 mo earlier, which indicated that the children had been said to be, for instance, 1 y, rather than 1 y and some months of age. This would have tended to make the reported age of children with no health card slightly lower than it truly was.

Vaccination information

While the trial was being conducted, the recommended vaccine schedule was as follows: BCG at birth; DTP/oral polio virus (OPV) at 6, 10, and 14 wk of age; and MV at 9 mo of age. Vaccinations were provided by routine government health services.

In Ghana, as in most low-income countries, there are no central vaccination registers. The first time a child comes for a vaccination, the child typically receives the first vaccine(s) and a health card on which the vaccines are noted. The health card is kept by the child's family. Vaccination status is assessed by inspecting the health card. The dates of vaccination for a specific vaccine can only be obtained from children who have a health card available for inspection. Children who have no health card are presumably unvaccinated, because they would have received a health card when first vaccinated. Children who report that they have a health card but the card is not available for inspection may have received one or more vaccines, but the dates and types of vaccination are not available. In the present study, we compared children who had a health card with children without a health card as a crude way of comparing vaccinated and unvaccinated children. Analyses restricted to specific vaccines have been limited to children who had a health card.

The collection of vaccination information is illustrated in **Figure 1**. Vaccination information was collected at the initiation of the trial. Of the 21,906 children in the trial, 13,462 were enrolled in the first round. One year later, in the fourth round, and 2 y later, in the seventh and last rounds, vaccination information was collected again. We used the information from the fourth and the seventh rounds (obtained from children who had a health card or who had no health card at the respective rounds) to assess the chance of getting vaccinations in the year (s) after enrollment (**Appendix A**).

Outcomes

Survival status was assessed at each of the 7 rounds. If a child had died, the date and the likely cause of the death were noted. The fieldworkers were trained to screen for night blindness and to identify Bitot's spot and corneal xerosis or ulcer or scarring. If they thought that the child might have any of these, the child was referred to a weekly research clinic, where the child was screened by a clinician who had received additional training in recognizing xerophthalmia. The clinician's decision was taken as the definitive diagnosis.

Statistical analyses

Vaccination information was only available at enrollment and during the fourth and seventh rounds, and no vaccination information was collected from dead children. Hence, it was not possible to directly assess the effect of VAS within the time window in which a vaccine was the most recent vaccine, because information on vaccines received after enrollment would only be available for children who survived and had their vaccination status assessed the next year(s). We analyzed the effect of VAS according to vaccination status at enrollment. We analyzed 3 phases of vaccination delivery: *1*) children who had received only BCG, *2*) children who had received DTP but not MV, and *3*) children who had received MV. Most children had registered an OPV at the same time as a DTP vaccine, but we focused on DTP because previous studies have indicated that DTP rather than OPV has negative nonspecific effects (20). We assessed the effect of VAS on survival in both the first 4-mo follow-up period (first to second rounds) of the trial and during the full 2-y study period (first to seventh round). The interpretation of this analysis was weakened by the fact that it did not take into account vaccines given during follow-up. When we analyzed the data, it became clear that the vaccination incidence was high in the months after enrollment. Hence, we conducted further analyses in which we focused on vaccination status at enrollment as a predictor of vaccines likely to be received during follow-up, eg, children with a health card who had not received MV during follow-up often received MV, and, likewise, children who had not received 3 doses of DTP often received an additional dose of DTP (Appendix A). We used vaccination information in the fourth round and mortality in the subsequent second year of the study to assess whether the changes in vaccination status after enrollment were in fact associated with survival.

Comparisons of background factors for children with and without a health card were carried out by using Poisson and linear

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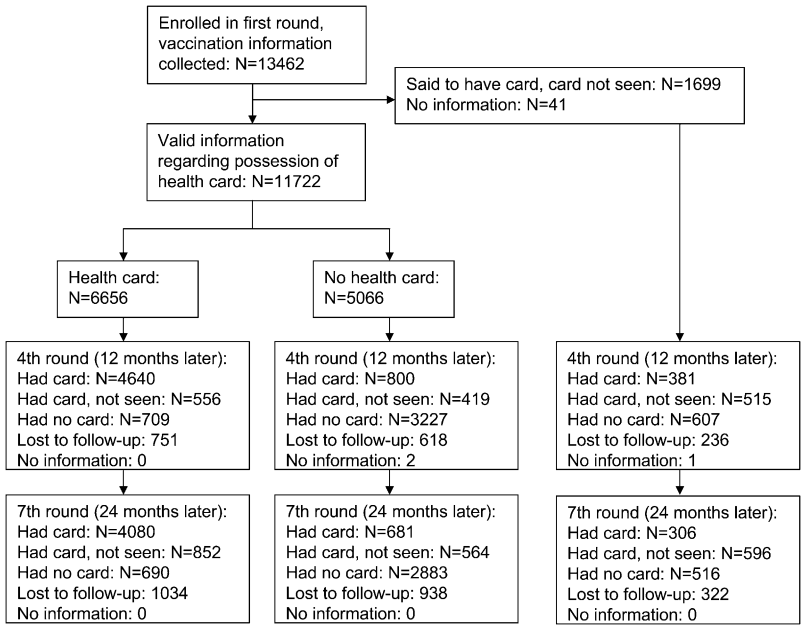

FIGURE 1. Overview of the collection of information on vaccination status. Note that children were lost to follow-up if they moved or were withdrawn from the trial.

regression models with control for age groups (<12, 12–17, 18–23, 24–29, 30–35, 36–47, and ≥48 mo), sex, and zone. Comparisons of compound assets were furthermore adjusted for the number of participating children in the compound to take into account the fact that compound size varied considerably. Robust variance estimation allowing correlations within clusters was applied (33).

The survival data were analyzed in Cox proportional hazards models with age as the underlying time, ie, with analysis time starting at the date of birth and delayed entry at the time of the first round. Hence, age was inherently controlled for in the models. We adjusted all analyses for zone. We tested the effect of all background factors presented in **Table 1** on the estimate of the effect of VAS on mortality after 2 y. None of the factors changed the estimate by ≥5% and were therefore not adjusted for. The comparisons of the effect of VAS among children with and without a health card were conducted with and without adjustment for socioeconomic factors (possession of zinc roof and radio) and markers of a pro-vitamin A–rich diet (mango trees, pawpaw trees, and red palm oil in compound controlled for number of participating children in the compound) to allow for the probability that children without a health card may have been more vitamin A–deficient. Robust variance estimation allowing correlations within clusters was applied (33). The effect of VAS was studied in all children (adjusted for sex as well as the abovementioned factors) and in a model allowing differential effects in boys and girls. Within all models, tests for proportionality of hazard rates were computed by using Schoenfeldt's residuals (34). Data were analyzed by using Stata 10.0 (StataCorp, College Station, TX).

RESULTS

Of the 13,462 children enrolled in the first round, 49% presented a health card, 13% were said to have had a card but it was not seen, 38% were reported not to have had a card, and 0.1% had no information on health card status. At the fourth round 1 y later, 35% (595/1699) of those who had been reported in the first round to have had a health card but this was not available to be seen were reported not to have a card, and the information for this group is therefore less valid. Hence, the analysis has focused on the 11,722 children whose card was seen (n = 6656) or who were reported not to have had a card (n = 5066) during the first round (Figure 1). The comparison of background factors indicated that those who were excluded from the present analysis (n = 1740) generally resembled children who had a health card more than those who were reported not to have a health card, although they were older (Table 1).

Comparison of children with and without a health card

There were major differences in the proportion of children having a health card between the 4 zones (Table 1). Children with a health card were younger than those having no card. They more often came from compounds with a zinc roof, radio, and access to pro-vitamin A–rich foods. They were more likely to have a BCG scar and not to have had measles illness before enrollment. The advantage in measles protection was substantially reduced when the children who had received MV were excluded from the comparison. Children with a health card had significantly lower midupper-arm-circumference-for-age z score and weight-for-age z score. They were also more likely to have been hospitalized.

VAS effects according to health card status

Because health cards are typically handed out in connection with a vaccination, we assumed that children having a health card had been vaccinated with one or more vaccines, and according to the information collected in the first round, 6244 (93.8%) had one or more vaccines documented on their health card. Of the remaining 412 children, who had a health card but no

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TABLE 1
Characteristics of children according to health card status at enrollment

	Health card (n = 6656)	No health card (n = 5066)	Adjusted relative risk (95% CI) or P values for health card versus no health card[1] (n = 11,722)	Health card not seen (n = 1699)	No information (n = 41)
Male sex [*n*/total *n* (%)]	3371/6656 (51)	2558/5066 (50)	1.01 (0.98, 1.05)	852 (50)	21 (51)
Median age (mo)[2,3]	30.2 (17.8, 47.3)	39.9 (23.7, 53.4)	$P < 0.0001$	48.0 (35.0, 58.9)	37.1 (17.2, 55.6)
Zone [*n* (%)][2]					
East	1709 (26)	1616 (32)	1 (ref)	342 (20)	21 (51)
North	2085 (31)	1124 (22)	1.33 (1.12, 1.59)	709 (42)	8 (20)
South	1666 (25)	1985 (39)	0.88 (0.75, 1.04)	366 (22)	10 (24)
West	1196 (18)	341 (7)	2.39 (1.73, 3.30)	282 (17)	2 (5)
No. of participating children in compound [*n* (%)][2]					
1–2	2550 (38)	2313 (46)	1 (ref)	711 (42)	12 (29)
3–4	2145 (32)	1609 (32)	1.10 (1.03, 1.17)	510 (30)	18 (44)
5–6	1074 (16)	689 (14)	1.23 (1.07, 1.41)	275 (16)	2 (5)
≥7	887 (13)	455 (9)	1.42 (1.23, 1.64)	203 (12)	9 (22)
Zinc roof [*n*/total *n* (%)][2]	1901/6643 (29)	984/5051 (19)	1.48 (1.33, 1.65)[4]	475/1693 (28)	12/41 (29)
Radio in compound [*n*/total *n* (%)][2]	2192/6643 (33)	928/5050 (18)	1.61 (1.45, 1.79)[4]	556/1693 (33)	12/41 (29)
Mango tree in compound [*n*/total *n* (%)][2]	2580/6643 (39)	1566/5050 (31)	1.09 (1.00, 1.19)[4]	687/1693 (41)	14/41 (34)
Pawpaw tree in compound [*n*/total *n* (%)][2]	861/6643 (13)	460/5048 (9)	1.19 (1.04, 1.38)[4]	219/1693 (13)	2/41 (5)
Red palm oil in compound [*n*/total *n* (%)][2]	266/6643 (4)	94/5050 (2)	1.68 (1.11, 2.54)[4]	53/1693 (3)	2/41 (5)
Ever breastfeed [*n*/total *n* (%)][2]	6648/6654 (100)	5059/5065 (100)	1.00 (1.00, 1.00)	1697/1699 (100)	Only 7 observations
Still breastfeeding [*n*/total *n* (%)][2]	3328/6655 (50)	1836/5064 (36)	1.03 (0.99, 1.07)	279/1699 (16)	Only 8 observations
Median mid-upper-arm circumference *z* score[2,3]	−1.31 (−2.01, −0.65)	−1.28 (−1.98, −0.64)	$P = 0.04$	−1.17 (−1.86, −0.56)	Only 3 observations
Median weight-for-age *z* score[2,3]	−1.72 (−2.58, −0.97)	−1.60 (−2.48, −0.79)	$P < 0.0001$	−1.42 (−2.19, −0.71)	Only 3 observations
Bacille Calmette-Guérin scar [*n*/total *n* (%)][2]	5330/6653 (80)	1167/5062 (23)	3.44 (2.99, 3.97)	1285/1698 (76)	Only 8 observations
Ever had measles [*n*/total *n* (%)][2]	174/6646 (3)	249/5046 (5)	0.67 (0.53, 0.85)	111/1690 (7)	Only 5 observations
Ever had measles [*n*/total *n* (%)][2,5]	94/2354 (4)	249/5046 (5)	1.20 (0.92, 1.56)	111/1689 (7)	Only 5 observations
Admitted to hospital [*n*/total *n* (%)][2]	263/6643 (4)	118/5057 (2)	1.87 (1.43, 2.44)	59/1694 (3)	Only 6 observations
Received vitamin A in study [*n*/total *n* (%)]	3259/6656 (49)	2489/5066 (49)	0.97 (0.86, 1.11)	817/1699 (48)	15/41 (37)
Incidence of xerophthalmia during study [*n*/total *n* (%)]					
All	45/6656 (1)	36/5066 (1)	1.03 (0.66, 1.61)	12/1699 (1)	0/41 (0)
Placebo group	29/3397 (1)	27/2577 (1)	1.02 (0.59, 1.76)	6/882 (1)	0/26 (0)

[1] Derived from Poisson and linear regression models adjusted for age, sex, and zone. Analyses of age, sex, and zone, respectively, are adjusted only for the other 2 variables. ref, reference.
[2] Information at enrollment.
[3] Interquartile range in parentheses.
[4] Also adjusted for the number of participating children in the compound.
[5] Children with recorded measles vaccination were excluded.

vaccinations noted, 10% (39 of the 374 children who were seen again during the fourth round) had vaccination dates that preceded the first round. Hence, it appears that some field assistants had noted that the child had a health card, but forgotten to note the vaccination dates. Other children may have received vaccines along with the vaccination card, but the health worker who provided the vaccine(s) had forgotten to write it on the health card. Hence, we compared children with and without a health card as a proxy for children with and without vaccinations. However, we also tested whether the main conclusions remained the same if we compared only children with a documented vaccination at enrollment with children with no health card.

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As shown in **Table 2**, within the 2-y trial period, VAS only had a beneficial effect for children without vaccinations; the mortality rate ratio (MRR) was 0.64 (95% CI: 0.47, 0.88) in these children compared with 0.95 (95% CI: 0.72, 1.26) in children with vaccinations (test of interaction between VAS and vaccination status, $P = 0.057$). This result did not change substantially after control for socioeconomic factors and provitamin A–rich food in the compound (Table 2). The beneficial effect of VAS on mortality in children without vaccinations was similar for boys (MRR: 0.68; 95% CI: 0.47, 0.99) and girls (MRR: 0.60; 95% CI: 0.40, 0.92). There was no overall beneficial effect among children with vaccinations (MRR: 0.95; 95% CI: 0.72, 1.26). However, the effect was not the same in boys and girls (test of interaction between VAS and sex, $P = 0.046$). There was a strongly differential effect of VAS among girls with and without vaccinations (test of interaction, $P = 0.009$) whereas there was no differential effect for boys (test of interaction, $P = 0.777$). The 3-factor interaction between VAS, sex, and vaccinations reached a P value of 0.050. The pattern was the same if the analysis was restricted to the first 4 mo after the first round, in that the effect of VAS tended to be different for girls with and without vaccinations (Table 2), although this difference was not statistically significant (test of interaction, $P = 0.152$), possibly because of the small number of mortality events.

If we limited the analysis to a comparison of children with a documented vaccine at enrollment ($n = 6244$) and to children with no health card ($n = 5066$), the conclusions remained the same. The MRR was 0.96 (95% CI: 0.72, 1.26) in children with a documented vaccination (test of interaction between VAS and vaccination status, $P = 0.057$). Among children with a documented vaccination, the effect of VAS tended to be beneficial for boys (MRR: 0.72; 95% CI: 0.49, 1.05; $P = 0.090$) but not for girls (MRR: 1.22; 95% CI: 0.87, 1.72; $P = 0.246$). Hence, the difference in the effect of VAS in boys and girls became slightly stronger (test of interaction between VAS and sex, $P = 0.025$).

Overall, VAS reduced the risk of xerophthalmia by 54% (relative risk: 0.46; 95% CI: 0.28, 0.75). This effect was seen in children with and without a health card and in both boys and girls (Table 2). Among children with a health card, the risk of xerophthalmia was the same in children who had only received DTP and in children who had received MV at enrollment (relative risk: 0.77; 95% CI: 0.29, 2.09).

Vaccination coverage

The overall vaccination coverage was low; at enrollment, only 46% of the children who were aged 6–90 mo had at least one documented DTP vaccination and only 37% had MV (**Table 3**). For those with a health card, the coverage continued to increase until ≈24 mo of age. Most children had received their routine vaccinations out-of-sequence; 78% of those who had received both BCG and DTP had received BCG at the same time as one of their DTP vaccinations. A total of 61% had received DTP at the same time as MV, and 49% had received DTP after MV. Some had received DTP both with and after MV; hence, a total of 80% (3423/4297) had received at least one of their doses of DTP simultaneously with or after MV (Table 3).

Among children who had their vaccination status assessed during the first round and 12 mo later during the fourth round, the vaccination intensity in the 12 mo after enrollment was high for children with a health card at enrollment (Appendix A). Almost 60% received additional doses of DTP if they had not received 3 doses of DTP at enrollment (71% among children with no MV at enrollment, and 51% among children with MV at enrollment). The incidence of DTP vaccination was highly significantly different between those who had received the third or fourth DTP (DTP3-4) and those who had not yet received 3 doses of DTP (DTP0-2). Approximately 65% of those who had not received MV at enrollment subsequently received this vaccine (56%, 66%, and 61% among children with no DTP, DTP1-2, and DTP3-4 at enrollment, respectively). Among children who did not have a health card at enrollment, but who were seen during the fourth round, only ≈11% received DTP (11%) or MV (12%) during the first year of follow-up. The vaccination intensity was comparable in boys and girls (data not shown).

VAS and BCG vaccine

Only 119 children had received BCG and no DTP or MV at enrollment (data not shown). Only one of these children died within the next 2 y; therefore, it was not possible to analyze any differential effect of VAS.

VAS and DTP

Among children who had received DTP but not MV at enrollment, VAS tended to be associated with lower mortality (**Table 4**). The effect appeared to be independent of the number of DTP vaccines received before enrollment, although the study lacked the power to examine this (data not shown).

Among the children who had their vaccination status assessed 1 y later, we assessed whether the effect of VAS could be related to the children receiving MV during follow-up. The MRR for VAS versus placebo during the second year of the study was 0.45 (95% CI: 0.15, 1.37) for those who had received MV during the first year ($n = 747$, 73%), whereas the MRR was 1.19 (95% CI: 0.28, 5.02) for children who had not received MV during the first year ($n = 282$; 27%) (test of interaction, $P = 0.25$).

VAS and MV

Among MV recipients, VAS was associated with significantly higher mortality for girls in both the first 4 mo and the full 2 y of the trial (**Table 5**). Female VAS recipients had roughly 2-fold higher mortality than female placebo recipients and than male VAS recipients. VAS was not associated with mortality among boys. The effect of VAS over 2 y of follow-up was significantly different for boys and girls (test of interaction, $P = 0.009$)

Because MV children who had <3 doses of DTP at enrollment were likely to receive additional doses of DTP during follow-up, we analyzed mortality among MV-vaccinated children according the number of doses of DTP received before enrollment (Table 5). There was little difference in mortality between VAS and placebo recipients who had received DTP3-4 at enrollment. However, among children who had received 0–2 doses of DTP, mortality was 2.6-fold higher for girls who received VAS than for the placebo group ($P = 0.002$), but only half among boys who received VAS compared with placebo ($P = 0.056$). Hence, the effect of VAS differed significantly between boys and girls among the children who were likely to have received additional doses of DTP during follow-up (test

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TABLE 2
Mortality and xerophthalmia for vitamin A supplementation (VAS) and placebo recipients according to sex and health card status at enrollment[1]

	All children			Boys			Girls		
	All	Had health card	No health card or card not available	All	Had health card	No health card or card not available	All	Had health card	No health card or card not available
Deaths									
First to second round									
VAS [*n*/total *n* (%)]	82/5748 (1.4)	53/3259 (1.6)	29/2489 (1.2)	41/2901 (1.4)	21/1666 (1.3)	20/1235 (1.6)	41/2847 (1.4)	32/1593 (2.0)	9/1254 (0.7)
Placebo [*n*/total *n* (%)]	91/5974 (1.5)	51/3397 (1.5)	40/2577 (1.6)	44/3028 (1.5)	22/1705 (1.3)	22/1323 (1.7)	47/2946 (1.6)	29/1692 (1.7)	18/1254 (1.4)
MRR (95% CI)[2]	0.94 (0.66, 1.34)	1.05 (0.70, 1.59)	0.79 (0.45, 1.38)	1.00 (0.64, 1.56)	0.96 (0.55, 1.66)	1.08 (0.58, 2.02)	0.89 (0.57, 1.39)	1.13 (0.68, 1.87)	0.51 (0.19, 1.38)
MRR (95% CI)[3]	0.93 (0.66, 1.31)	1.05 (0.70, 1.56)	0.77 (0.44, 1.34)	1.00 (0.65, 1.55)	0.96 (0.56, 1.67)	1.06 (0.57, 1.95)	0.86 (0.55, 1.36)	1.09 (0.66, 1.82)	0.50 (0.18, 1.34)
First to seventh round									
VAS [*n*/total *n* (%)]	210/5748 (3.7)	120/3259 (3.7)	90/2489 (3.6)	95/2901 (3.3)	48/1666 (2.9)	47/1235 (3.8)	115/2847 (4.0)	72/1593 (4.5)	43/1254 (3.4)
Placebo [*n*/total *n* (%)]	274/5974 (4.6)	129/3397 (3.8)	145/2577 (5.6)	142/3028 (4.7)	66/1705 (3.9)	76/1323 (5.7)	132/2946 (4.5)	63/1692 (3.7)	69/1254 (5.5)
MRR (95% CI)[2]	0.79 (0.63, 0.98)	0.95 (0.72, 1.26)	0.64 (0.47, 0.88)	0.71 (0.54, 0.92)	0.74 (0.51, 1.08)[a]	0.68 (0.47, 0.99)	0.88 (0.67, 1.16)	1.18 (0.84, 1.67)[a,b]	0.60 (0.40, 0.92)[b]
MRR (95% CI)[3]	0.79 (0.64, 0.98)	0.97 (0.73, 1.27)[c]	0.63 (0.46, 0.87)[c]	0.71 (0.54, 0.92)	0.75 (0.51, 1.09)[d]	0.67 (0.47, 0.97)	0.88 (0.67, 1.15)	1.19 (0.85, 1.68)[d,e]	0.60 (0.40, 0.90)[e]
Xerophthalmia									
First to seventh round									
VAS [*n*/total *n* (%)]	25/5748 (0.4)	16/3259 (0.5)	9/2489 (0.4)	11/2901 (0.4)	6/1666 (0.4)	5/1235 (0.4)	14/2847 (0.5)	10/1593 (0.6)	4/1254 (0.3)
Placebo [*n*/total *n* (%)]	56/5974 (0.9)	29/3397 (0.9)	27/2577 (1.1)	25/3028 (0.8)	14/1705 (0.8)	11/1323 (0.8)	31/2946 (1.1)	15/1692 (0.9)	16/1254 (1.3)
RR (95% CI)[2]	0.46 (0.28, 0.75)	0.56 (0.32, 0.98)	0.34 (0.16, 0.75)	0.43 (0.21, 0.88)	0.42 (0.17, 1.03)	0.43 (0.16, 1.18)	0.48 (0.25, 0.90)	0.69 (0.33, 1.44)	0.27 (0.08, 0.84)
RR (95% CI)[3]	0.48 (0.29, 0.78)	0.58 (0.33, 1.01)	0.36 (0.16, 0.80)	0.45 (0.22, 0.92)	0.44 (0.18, 1.08)	0.45 (0.16, 1.25)	0.50 (0.26, 0.95)	0.70 (0.33, 1.47)	0.29 (0.09, 0.95)

[1] MRR, mortality rate ratio, derived from Cox proportional hazards models; RR, relative risk, derived from Poisson models. Estimates with the same superscript letters were significantly different, $P < 0.05$.
[2] Adjusted for age, sex, and zone.
[3] Adjusted for age, sex, zone, possession of zinc roof, radio, mango tree, pawpaw tree, red palm oil, and number of participating children in the compound.

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TABLE 3
Vaccination coverage at enrollment in the vitamin A trial[1]

Vaccine	Coverage of vaccines		Coverage by age among children with health card						
	All children (n = 11,722)	Children with health card (n = 6656)	6–11 mo (n = 733)	12–17 mo (n = 961)	18–23 mo (n = 789)	24–29 mo (n = 815)	30–35 mo (n = 616)	36–47 mo (n = 1141)	≥48 mo (n = 1601)
	%	%	%	%	%	%	%	%	%
BCG	44 (5140)	77	83	86	86	80	77	72	67
Any DTP	46 (5425)	82	85	90	89	87	83	77	70
DTP3-4	25 (2896)	44	22	38	49	50	54	48	44
Any OPV	46 (5401)	81	86	89	89	86	83	77	70
OPV3-5	24 (2801)	42	21	35	47	50	52	48	42
MV	37 (4297)	65	26	54	76	77	74	72	67
BCG at same time as a DTP vaccination	—	78[2]	84	84	82	78	77	71	62
MV at same time as a DTP vaccination	—	61[3]	75	69	68	67	62	60	49
At least one DTP vaccination after MV	—	49[3]	13	29	43	51	56	54	62

[1] n in parentheses. BCG, bacille Calmette-Guérin; MV, measles vaccine; DTP, diphtheria-tetanus-pertussis vaccine; OPV, oral polio virus; DTP3-4, 3–4 doses of DTP; OPV3-5, 3–5 doses of OPV.
[2] Proportion of children with both a BCG and a DTP vaccine.
[3] Proportion of children with a measles vaccine. Note that a child can belong to both groups.

of interaction, P = 0.0004), but not among the children who were not likely to receive further doses of DTP (P = 0.704).

To explore whether the MRR between the high VAS and placebo recipients in girls who had received 0–2 doses of DTP at enrollment was in fact related to having received additional doses of DTP after enrollment, we assessed the effect of VAS during the second year of follow-up in children who had and in children who had not received DTP during the first year (**Table 6**). Among the girls who had a health card both at enrollment and 12 mo later at the fourth round and who had between 0 and 2 doses of DTP at enrollment, VAS was associated with higher mortality among those who had received DTP after MV, but lower mortality among those who had not received DTP after MV) (test of interaction between VAS and DTP after MV, P = 0.03) (Table 6).

DISCUSSION

Principal findings

The original study found a beneficial effect of VAS on overall mortality; the MRR between VAS and placebo recipients was 0.81 (95% CI: 0.68, 0.98) (22). In the present reanalysis, which focused on the children enrolled at the first round who had or did not have a child-held health card, the overall effect of VAS on mortality was similar: 0.79 (95% CI: 0.63, 0.98).

As hypothesized, the reanalysis suggests important interactions between VAS, sex, and vaccines. VAS was associated with a strong beneficial effect in children with no record of vaccination, whereas there was almost no effect for those who had been vaccinated. This differential effect was due to a difference in girls, in whom VAS was associated with a decrease in mortality in the unvaccinated but in whom VAS was associated with a nonsignificant increase in mortality in the vaccinated (Table 2). This was due to a differential effect of VAS according to vaccination type. Among girls who had already received MV at enrollment, VAS was associated with significantly higher mortality. This was only seen in girls who were missing doses of DTP at enrollment and were therefore likely to receive them during follow-up (Table 5). Consistent with this interpretation, VAS was associated with significantly higher mortality during the second year of the trial in girls who had received DTP after MV during the first year of the trial (Table 6).

TABLE 4
Mortality of vitamin A supplementation (VAS) and placebo recipients among children vaccinated with diphtheria-tetanus-pertussis vaccine (DTP) who had not yet received measles vaccine at enrollment and who had received at least one DTP, by length of follow-up and sex[1]

	Deaths			P for interaction between VAS and sex
	All children	Boys	Girls	
First to second round				
VAS [n/total n (%)]	15/697 (2.2)	6/348 (1.7)	9/349 (2.6)	—
Placebo [n/total n (%)]	27/738 (3.7)	13/377 (3.4)	14/361 (3.9)	—
MRR (95% CI)	0.60 (0.32, 1.14)	0.51 (0.20, 1.29)	0.69 (0.32, 1.47)	0.589
First to seventh round				
VAS [n/total n (%)]	34/697 (4.9)	16/348 (4.6)	18/349 (5.2)	—
Placebo [n/total n (%)]	54/738 (7.3)	28/377 (7.4)	26/361 (7.2)	—
MRR (95% CI)	0.66 (0.41, 1.06)	0.61 (0.34, 1.11)	0.71 (0.39, 1.31)	0.692

[1] MRR, mortality rate ratio, derived from Cox proportional hazards models, adjusted for age, sex, and zone.

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TABLE 5
Mortality of vitamin A supplementation (VAS) and placebo recipients among measles vaccine–vaccinated children according to length of follow-up, sex, and number of doses of diphtheria-tetanus-pertussis vaccine (DTP) at enrollment[1]

	Deaths			*P* for interaction between VAS and sex
	All children	Boys	Girls	
First to second round				
VAS [*n*/total *n* (%)]	30/2089 (1.4)	10/1085 (0.9)	20/1004 (2.0)	—
Placebo [*n*/total *n* (%)]	19/2208 (0.9)	8/1107 (0.7)	11/1101 (1.0)	—
MRR (95% CI)	1.65 (0.94, 2.91)	1.21 (0.51, 2.90)	1.99 (1.05, 3.77)	0.324
First to seventh round				
VAS [*n*/total *n* (%)]	70/2089 (3.4)	23/1085 (2.1)	47/1004 (4.7)	—
Placebo [*n*/total *n* (%)]	62/2208 (2.8)	33/1107 (3.0)	29/1101 (2.6)	—
MRR (95% CI)	1.17 (0.84, 1.64)	0.69 (0.41, 1.16)	1.75 (1.11, 2.76)	0.009
First to seventh round: children who had DTP0-2 at enrollment				
VAS [*n*/total *n* (%)]	—	10/446 (2.2)	30/385 (7.8)	—
Placebo [*n*/total *n* (%)]	—	19/452 (4.2)	12/409 (2.9)	—
MRR (95% CI)	—	0.50 (0.25, 1.02)	2.60 (1.41, 4.80)	0.0004
First to seventh round: children who had DTP3-4 at enrollment				
VAS [*n*/total *n* (%)]	—	13/639 (2.0)	17/619 (2.8)	—
Placebo [*n*/total *n* (%)]	—	14/655 (2.1)	17/692 (2.5)	—
MRR (95% CI)	—	0.90 (0.45, 1.81)	1.09 (0.54, 2.20)	0.704

[1] MRR, mortality rate ratio, derived from Cox proportional hazards models, adjusted for age, sex, and zone; DTP0-2, had not yet received 3 doses of DTP; DTP3-4, 3–4 doses of DTP.

Strengths and weaknesses

The strength of the present study was its large sample size and the fact that the data were collected by a research team who was unaware of the hypothesis. However, vaccination data were only collected once a year, and we were not able to change the vaccination status for children when they received other vaccines, except annually. Many children received additional vaccines already within the first months of follow-up (Appendix A), possibly because the inspection of heath cards at enrollment made mothers aware that their children were missing vaccines. Furthermore, the vaccination practice varied; few children received the vaccinations according to schedule. This meant that the planned approach of analyzing the effect of VAS using the most recent vaccination at enrollment as a proxy for most recent vaccination became less meaningful. Hence, we conducted a further analyses of survival in which we stratified the children according to the vaccinations they were likely to receive during follow-up. This is obviously less ideal than if the vaccination status had been updated on a more frequent basis.

Bias and confounding

The present study involved randomization by cluster, was placebo-controlled, and was double-blind. Misclassification of exposure (vitamin A or placebo) did not seem likely, and no allocation errors were found after this was checked in a sample of bottles returned from the field. Differential misclassification of outcomes (deaths and xerophthalmia) also seemed unlikely. However, the key analyses in the current study were subgroup analyses, so the important strength of maximizing the chance of equalizing known and unknown confounders that is conferred by randomization may have been lost. We therefore examined the

TABLE 6
Mortality of vitamin A supplementation (VAS) and placebo recipients among measles vaccine (MV)–vaccinated children during the second year of the study according to whether diphtheria-tetanus-pertussis vaccine (DTP) had been received after MV[1]

	Deaths					
	Received DTP after MV during first year		Did not receive DTP after MV during first year		*P* for interaction between VAS and receiving DTP during first year	
	Boys	Girls	Boys	Girls	Boys	Girls
VAS [*n*/total *n* (%)]	4/202 (2.0)	7/162 (4.3)	0/109 (0.0)	1/84 (1.2)	—	—
Placebo [*n*/total *n* (%)]	4/220 (1.8)	1/201 (0.5)	3/96 (3.1)	2/73 (2.7)	—	—
MRR (95% CI)	1.12 (0.28, 4.52)	9.99 (1.26, 79)	0	0.31 (0.03, 3.23)	NA	0.03

[1] Table includes only the children from Table 5 who had a health card inspected in the fourth round and had received <3 doses of DTP in the first round. MRR, mortality rate ratio, derived from Cox proportional hazards models, adjusted for age, sex, and zone.

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effects of vaccination with and without adjustment for known potential confounders.

We analyzed the effect of VAS by health card status. Some of those without a health card may have actually received vaccines, and indeed 23% (1167/5062) of the children without a health card were reported to have a BCG scar by fieldworkers. Furthermore, 11% (709/6656) of those who had their card seen during the first round did not have a card during the fourth round, which indicated that cards did get lost. However, the proportion with a BCG scar among children without a card (1167/5062) was significantly lower than among those with a card (5330/6653), and the proportion reporting measles infection among children with a card (174/6646) was significantly lower than that among children without a card (249/5046). Hence, it seems likely that a large majority of those without a health card had never received any vaccination. The vast majority of children with a health card had one or more documented vaccines (6244/6656). If anything, the differences in VAS effects in children with and without a health card became stronger if we excluded children with a health card but no documented vaccines. Hence, it seemed justified to use possession of a health card as a proxy for having or not having received vaccines. Nondifferential misclassification of exposure information would lead to conservative effect estimates.

Effect of vitamin A deficiency

Several significant differences in baseline characteristics were observed between those with and without a health card, which could perhaps have explained the differential effect of VAS. In particular, the effects could be explained by a higher prevalence of vitamin A deficiency among children without a health card. Indeed, vitamin A–rich foods were less common in households of children without a health card, and these children clearly also came from compounds that were poorer. However, in other respects, the children with a health card could seem to represent a more fragile group of children; they apparently had worse nutritional status and were more often hospitalized. The worse nutritional status may indicate that the age of the children without a health card on average had been assessed to be lower than it truly was (*see* Subjects and Methods), and more frequent hospitalization could simply reflect that communities with high vaccination coverage had closer contact with health services. Importantly, the proportion who developed xerophthalmia during the follow-up period was similar among those with and without a card, and, particularly, there was no significant difference in the placebo group (Table 1). Hence, the differential effect of VAS in children with and without a health card may thus have been due to a true differential effect of VAS in vaccinated and unvaccinated children.

VAS and vaccine interactions

Before analyzing the present data, we hypothesized that the effect of VAS would depend on the most recent vaccine at enrollment, the effect of VAS being beneficial when BCG and MV were the most recent vaccines, but negative when DTP was the most recent vaccine, especially in girls. The observed interaction between VAS and health card supports that VAS and vaccines interact. The fact that this interaction was only seen for girls supports the hypothesis that this interaction was due to interactions between VAS and vaccines, because the negative nonspecific effects of vaccines have been observed in girls (21–23).

In contrast with our prior expectations, VAS tended to be beneficial when DTP was the most recent vaccination (Table 4), whereas VAS was associated with increased mortality in girls who had MV as their most recent vaccination before enrollment (Table 5). Initially, this appeared to contradict the hypothesis. However, that hypothesis was based on the expectation that children received the vaccines in the recommended sequence, ie, first BCG, then all doses of DTP, and then MV. In the present study, most children had received vaccines out-of-sequence, and vaccination intensity was high during the months after enrollment (Table 3 and Appendix A). Having DTP as the most recent vaccination before enrollment was associated with a high probability of receiving MV after enrollment, and having MV and 0–2 doses of DTP before enrollment was associated with a high probability of receiving one or more DTP vaccines during enrollment. Hence, the most recent vaccination may in fact have been the vaccine received after enrollment rather than the vaccine received before enrollment. From this perspective, the beneficial effect of VAS in children with DTP as their most recent vaccination before enrollment might indicate that many children were likely to receive MV during follow-up; although data were limited for the last year of the study, they did suggest that the beneficial effect was among children who had received MV during the first year of the study.

The negative effect of VAS in girls with MV before enrollment might reflect that many children received DTP during follow-up. This was clearly indicated by the fact that the strong negative effect of VAS (Table 5) was only found among MV girls who had received <3 doses of DTP before enrollment and hence were likely to receive additional doses of DTP during follow-up. In support of this interpretation was the observation that the excess mortality during the second year of the study among girls who had received VAS was only found among girls who had received DTP after MV during the first year of the study (Table 6). Hence, this reanalysis supports the hypothesis that receiving VAS in the same time window as DTP may have negative consequences for girls.

Conclusions

VAS clearly reduced the risk of developing xerophthalmia in boys and girls with and without a health card. Hence, vitamin A deficiency was present and prevented or treated by means of VAS. Despite this, VAS had no effect on mortality in children who were vaccinated, and it was associated with increased mortality among measles-vaccinated girls who were likely to have received DTP during follow-up.

Note that the first large randomized VAS studies, which led to the current VAS policy, took place when the global vaccination program was in its initial phase. The present study adds to an accumulating number of studies suggesting that the effect of VAS is not independent of routine vaccines and may be harmful in some situations (12, 13, 26–28). This is potentially problematic because the official policy is to give VAS at vaccination contacts. Because the current VAS policy is not based on specific studies, there is an urgent need to reassess interactions between VAS and

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vaccines in data from previously conducted studies if data on vaccination status were collected and to test the effect of VAS and different vaccination practices in future randomized trials.

The authors' responsibilities were as follows—DAR and FNB: responsible for planning and conducting the original trial with colleagues from the London School of Hygiene and Tropical Medicine, Ministry of Health of Ghana, and University of Service and Technology, Kumasi, Ghana; CSB and PA: proposed the present analysis; JN: provided assistance with the statistical analysis; CSB and PA: wrote the first draft of the manuscript; and DAR and FNB: reviewed the first draft of the manuscript. All authors contributed with comments and approved the final version of the manuscript. None of the authors had any conflicts of interest.

REFERENCES

1. Beaton GH, Martorell R, McCabe G, L'abbé KA, Edmonston B, Ross AC. Effectiveness of vitamin A supplementation in the control of young child morbidity and mortality in developing countries. Toronto, CA: University of Toronto, 1993.
2. Fawzi WW, Chalmers TC, Herrera MG, Mosteller F. Vitamin A supplementation and child mortality. A meta-analysis. JAMA 1993;269: 898–903.
3. WHO/UNICEF/IVACG Task Force. Vitamin A supplements. A guide to their use in the treatment and prevention of vitamin A deficiency and xerophthalmia, 2nd ed. Geneva, Switzerland: World Health Organization, 1997.
4. Benn CS, Balé C, Sommerfelt H, Friis H, Aaby P. Vitamin A supplementation and childhood mortality: amplification of the non-specific effects of vaccines? Int J Epidemiol 2003;32:822–8.
5. Humphrey JH, Agoestina T, Wu L, et al. Impact of neonatal vitamin A supplementation on infant morbidity and mortality. J Pediatr 1996;128: 489–96.
6. Rahmathullah L, Tielsch JM, Thulasiraj RD, et al. Impact of supplementing newborn infants with vitamin A on early infant mortality: community based randomized trial in southern India. BMJ 2003;327: 254.
7. Daulaire NM, Starbuck ES, Houston RM, Church MS, Stukel TA, Pandey MR. Childhood mortality after a high dose of vitamin A in a high risk population. BMJ 1992;304:207–10.
8. West KP Jr, Katz J, Shrestha SR, et al. Mortality of infants $<$ 6 mo of age supplemented with vitamin A: a randomized, double-masked trial in Nepal. Am J Clin Nutr 1995;62:143–8.
9. Rahman MM, Mahalanabis D, Wahed MA, Islam MA, Habte D. Administration of 25,000 IU vitamin A doses at routine immunisation in young infants. Eur J Clin Nutr 1995;49:439–45.
10. Randomised trial to assess benefits and safety of vitamin A supplementation linked to immunisation in early infancy. WHO/CHD Immunisation-Linked Vitamin A Supplementation Study Group. Lancet 1998;352:1257–63.
11. Newton S, Cousens S, Owusu-Agyei S, et al. Vitamin a supplementation does not affect infants' immune responses to polio and tetanus vaccines. J Nutr 2005;135:2669–73.
12. Mahalanabis D, Rahman MM, Wahed MA, Islam MA, Habte D. Vitamin A megadose during early infancy on serum retinol concentrations and acute side effects and residual effects on six months follow-up. Nutr Res 1997;17:649–59.
13. Newton S, Owusu-Agyei S, Filteau S, Gyan T, Kirkwood BR. Vitamin A supplements are well tolerated with the pentavalent vaccine. Vaccine 2008;26:6608–13.
14. Garly ML, Martins CL, Balé C, et al. BCG scar and positive tuberculin reaction and reduced infant and childhood mortality: a non-specific beneficial effect of BCG? Vaccine 2003;21:2782–90.
15. Roth A, Sodemann M, Jensen H, et al. Tuberculin reaction, BCG scar and lower female mortality. Epidemiology 2006;17:562–8.
16. Aaby P, Samb B, Simondon F, Coll Seck AM, Knudsen K, Whittle H. Non-specific beneficial effect of measles immunisation: analysis of mortality studies from developing countries. BMJ 1995;311:481–5.
17. Aaby P, Bhuyia A, Nahar L, Knudsen K, Francisco A, Strong M. The survival benefit of measles immunisation may not be explained entirely by the prevention of measles disease. Int J Epidemiol 2003;32:106–15.
18. Kristensen I, Aaby P, Jensen H. Routine vaccinations and child survival: follow up study in Guinea-Bissau, West Africa. BMJ 2000;321:1435–8.
19. Aaby P, Jensen H, Gomes J, Fernandes M, Lisse IM. The introduction of diphtheria-tetanus-pertussis vaccine and child mortality in rural Guinea-Bissau: an observational study. Int J Epidemiol 2004;33:374–80.
20. Aaby P, Rodrigues A, Biai S, et al. Oral polio vaccination and low case fatality at the paediatric ward in Bissau, Guinea-Bissau. Vaccine 2004; 22:3014–7.
21. Aaby P, Garly ML, Balé C, Martins C, Lisse I, Jensen H. Routine vaccinations and child survival in war situation with high mortality: effect of gender. Vaccine 2002;21:15–20.
22. Veirum JE, Sodemann M, Biai S, et al. Diphtheria-tetanus-pertussis and measles vaccinations associated with divergent effects on female and male mortality at the paediatric ward in Bissau, Guinea-Bissau. Vaccine 2005;23:1197–203.
23. Aaby P, Jensen H, Rodrigues A, et al. Divergent female-male mortality ratios associated with different routine vaccinations among female-male twin pairs. Int J Epidemiol 2004;33:367–73.
24. Hoag KA, Nashold FE, Goverman J, Hayes CE. Retinoic acid enhances the T helper 2 cell development that is essential for robust antibody responses through its action on the antigen presenting cells. J Nutr 2002; 132:3736–9.
25. Benn CS, Diness BR, Roth A, et al. Randomized trial of the effect on mortality of 50,000 IU vitamin A given with BCG vaccine to infants in Guinea-Bissau, West-Africa. BMJ 2008;336:1416–20.
26. Benn CS, Rodrigues A, Yazdanbakhsh M, et al. The effect of high-dose vitamin A supplementation administered with BCG vaccine at birth may be modified by subsequent DTP vaccination. Vaccine 2009;27:2891–8.
27. Benn CS, Martins C, Rodrigues A, Fisker AB, Christoffersen D, Aaby P. The effect of vitamin A supplementation administered with missing vaccines during national immunisation days in Guinea-Bissau. Int J Epidemiol 2009;38:304–11.
28. Fisker AB, Lisse IM, Aaby P, et al. Impact of neonatal vitamin A supplementation with BCG vaccine on vitamin A status at 6 weeks and 4 months of age. Am J Clin Nutr 2007;86:1032–9.
29. VAST Study Team. Vitamin A supplementation in northern Ghana: effects on clinic attendances, hospital admissions, and child mortality. Ghana Lancet 1993;342:7–12.
30. Binka FN, Ross DA, Morris SS, et al. Vitamin A supplementation and childhood malaria in northern Ghana. Am J Clin Nutr 1995;61:853–9.
31. Ross DA, Kirkwood BR, Binka FN, et al. Child morbidity and mortality following vitamin A supplementation in Ghana: time since dosing, number of doses and time of the year. Am J Public Health 1995;85: 1246–51.
32. Kirkwood BR, Ross DA, Arthur P, et al. Effect of vitamin A supplementation on the growth of young children in northern Ghana. Am J Clin Nutr 1996;63:773–81.
33. StataCorp. Survival analysis and epidemiological tables. College Station, TX: Stata Press, 2007.
34. Armitage P, Colton T, eds. Encyclopedia of biostatistics. 2nd ed. New York, NY: Wiley, 2005.

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APPENDIX A

Proportion of children receiving vaccination between enrollment and 4 mo, between enrollment and 12 mo, and between enrollment and 24 mo among children with a health card and children without a health card at enrollment[1]

Health card status and vaccination status at enrollment	Vaccinated between enrollment and 4 mo later[2]		Vaccinated between enrollment and 12 mo later[2]		Vaccinated between enrollment and 24 mo later[3,4]
	Received ≥1 DTP during follow-up	Received MV during follow-up	Received ≥1 DTP during follow-up	Received MV during follow-up	Received ≥1 DTP during follow-up
	*% (*n/*total* n*)*	*% (*n/*total* n*)*	*% (*n/*total* n*)*	*% (*n/*total* n*)*	*% (*n/*total* n*)*
No health card at enrollment					
Presumably unvaccinated	7 (297/4027)	7 (274/4027)	11 (434/4027)	12 (473/4027)	11 (383/3564)
Had health card at enrollment					
No MV at enrollment					
BCG (no DTP or MV)	38 (30/80)	29 (23/80)	55 (44/80)	56 (45/80)	56 (39/70)
DTP1-2 (no MV)	49 (392/799)[a]	39 (308/799)	71 (566/799)[b]	66 (529/799)	74 (529/717)[c]
DTP3-4 (no MV)	6 (14/230)[a]	49 (113/230)	9 (20/230)[b]	61 (141/230)	9 (17/200)[c]
Had health card at enrollment					
MV at enrollment					
DTP0-2	37 (419/1147)[d]		51 (585/1147)[e]		51 (515/1010)[f]
DTP3-4	4 (91/2036)[d]		6 (124/2036)[e]		6 (103/1774)[f]

[1] BCG, bacille Calmette-Guérin; MV, measles vaccine; DTP, diphtheria-tetanus-pertussis vaccine; DTP1-2, 1–2 doses of DTP; DTP3-4, 3–4 doses of DTP; DTP0-2, had not yet received 3 doses of DTP. Values with the same superscript letters had a significantly different incidence of DTP vaccination, $P < 0.00001$.

[2] All children whose vaccination status was assessed in the first and the fourth round.

[3] All children whose vaccination status was assessed in the first and the seventh round.

[4] Data on measles vaccination from the seventh round are missing. Hence, it was not possible to measure the incidence of measles vaccination in the last year of the study.

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