

# Effects of antenatal multiple micronutrient supplementation on birthweight and gestational duration in Nepal: double-blind, randomised controlled trial



David Osrin, Anjana Vaidya, Yagya Shrestha, Ram Bahadur Baniya, Dharma S Manandhar, Ramesh K Adhikari, Suzanne Filteau, Andrew Tomkins, Anthony M de L Costello

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## Summary

**Background** Neonatal mortality is the biggest contributor to global mortality of children younger than 5 years, and low birthweight is a crucial underlying factor. We tested the hypotheses that antenatal multiple micronutrient supplementation would increase infant birthweight and gestational duration.

**Methods** We did a double-blind, randomised controlled trial in Dhanusha district, Nepal. Women attending for antenatal care with singleton pregnancies at up to 20 weeks' gestation were invited to participate. Participants were randomly allocated either routine iron and folic acid supplements (control; n=600) or a multiple micronutrient supplement providing a recommended daily allowance of 15 vitamins and minerals (intervention; n=600). Supplementation began at a minimum of 12 weeks' gestation and continued until delivery. Primary outcome measures were birthweight and gestational duration. Analysis was by intention to treat. The study is registered as an International Standard Randomised Controlled Trial, number ISRCTN88625934.

**Findings** Birthweight was available for 523/600 infants in the control group and 529/600 in the intervention group. Mean birthweight was 2733 g (SD 422) in the control group and 2810 g (453) in the intervention group, representing a mean difference of 77 g (95% CI 24–130; p=0.004) and a relative fall in the proportion of low birthweight by 25%. No difference was recorded in the duration of gestation (0.2 weeks [−0.1 to 0.4]; p=0.12), infant length (0.3 cm [−0.1 to 0.6]; p=0.16), or head circumference (0.2 cm [−0.1 to 0.4]; p=0.18).

**Interpretation** In a poor community in Nepal, consumption of a daily supplement containing a recommended daily allowance of 15 micronutrients in the second and third trimesters of pregnancy was associated with increased birthweight when compared with a standard iron and folic acid preparation. The effects on perinatal morbidity and mortality need further comparisons between studies.

## Introduction

A third of global deaths happen in children younger than 5 years,<sup>1</sup> most in the neonatal period.<sup>2</sup> Low birthweight (<2500 g, irrespective of gestation) underlies many of these deaths. 25 million low-birthweight infants are born every year, and associations with neonatal mortality have been well described.<sup>3–6</sup> Low birthweight is also associated with diminished childhood growth,<sup>7</sup> morbidity,<sup>8</sup> compromised cognitive and behavioural development,<sup>9,10</sup> and disease in adulthood.<sup>11</sup> It is, however, rather a blunt indicator of fetal history and infant health: low-birthweight infants can be small for gestational age, preterm, or both. Most birthweight data from poor countries do not discriminate between these factors, a dilemma that has led to several approaches to classification and uncertainty about disease burden.<sup>12,13</sup> Although the epidemiology and associations of low birthweight have been examined closely,<sup>14</sup> approaches to alleviate the burden have met with limited success.<sup>15,16</sup>

Maternal nutritional status is linked with fetal weight, particularly small-for-gestational-age birth, as a result of presumptive intrauterine growth restriction. Increases in macronutrient consumption during pregnancy do lead to increased birthweight,<sup>17,18</sup> but control of fetal growth is

complex,<sup>19</sup> and the effectiveness of interventions remains patchy.<sup>20,21</sup> The possibility that improvements in vitamin and mineral status might lead to reductions in low birthweight is attractive in terms of policy planning. A wealth of descriptive epidemiology has drawn links between deficiencies of such micronutrients—particularly several at the same time<sup>22</sup>—and birth outcomes.<sup>23</sup> Many women in wealthy and poor countries already take multiple micronutrient supplements before and during pregnancy, and iron and folic acid supplements are generally recommended. If multiple micronutrient supplement tablets were shown to be helpful, only minor adjustments to policy would be needed.

Repletion might improve birth outcomes,<sup>24</sup> a proposal lent support by findings of a study of infants of women infected with HIV.<sup>25</sup> An expert group has formulated a combination of ten vitamins and five minerals at levels of about one recommended daily allowance for this reason.<sup>26</sup> However, the evidence base for policy change is insufficient.<sup>27,28</sup> We undertook a trial with the aim of establishing whether second and third trimester supplementation with a multiple micronutrient regimen at one recommended daily allowance would increase birthweight and prolong gestational duration.

International Perinatal Care Unit (D Osrin MRCP, Prof A M de L Costello FRCP), and Centre for International Child Health (S Filteau PhD, Prof A Tomkins FRCP), Institute of Child Health, University College London, 30 Guilford Street, London WC1N 1EH, UK; Mother and Infant Research Activities (MIRA), PO Box 921, Kathmandu, Nepal (A Vaidya MD, Y Shrestha RN, R B Baniya MPH, Prof D S Manandhar FRCP); and Institute of Medicine, Tribhuvan University, Kathmandu, Nepal (Prof R K Adhikari MD)

Correspondence to:  
Dr David Osrin  
[d.osrin@ich.ucl.ac.uk](mailto:d.osrin@ich.ucl.ac.uk)

## Participants and methods

### Study location and population

Nepal is a south Asian country challenged by geography, poverty, and a violent insurrection. The most recent estimates of neonatal and perinatal mortality rates are 39 per 1000 livebirths and 47 per 1000 births, respectively.<sup>29</sup> More than half of women cannot read.<sup>30</sup> About a third have low body-mass index ( $<18.5 \text{ kg/m}^2$ )<sup>31</sup> and a quarter report limiting their own food consumption to provide food for their children;<sup>32</sup> half of children have stunted growth.<sup>31</sup> Deficiencies of several micronutrients have been well described in individual studies<sup>33–36</sup> and in a national sample.<sup>37</sup> Hospital-based figures for low birthweight suggest a prevalence of 27%,<sup>38</sup> this number is certainly an underestimate—a level of 40% has been reported in a southern rural population.<sup>39</sup>

The fifth most populous of Nepal's districts, Dhanusha, lies in the southern plains of the central zone. It has a population of about 670 000, 13% of whom are younger than 5 years. The human development index is 0.329, the population per doctor about 19 000, and the population per hospital bed 6700.<sup>40</sup> Two-thirds of households have access to safe drinking water. The urban population forms 11% of the district. Janakpur, the district municipality and former capital of the Mithila kingdom, is a town of great cultural and historic significance.

The sampling frame for potential participants included all women attending a designated antenatal clinic at Janakpur zonal hospital. The clinic was supervised by a senior nurse (YS) and run by auxiliary nurse midwives. It was open six mornings a week on a walk-in basis and provided all the routine services specified in Nepal's national maternity care guidelines and antenatal care protocol.<sup>41,42</sup>

Women were eligible for enrolment at up to 20 completed weeks of gestation. After screening on the basis of history, dates, and examination, we invited potential participants to a room serving as the study centre. We made a firm offer of enrolment if further history, examination, and ultrasound screening confirmed: (1) a gestation of up to 20 completed weeks; (2) a singleton pregnancy; (3) no notable fetal abnormality; (4) no existing maternal illness of a severity that could compromise the outcome of pregnancy; and (5) that the participant lived in an area of Dhanusha or the adjoining district of Mahottari accessible for home visits. If preliminary ultrasound examination suggested a congenital anomaly, we referred participants for repeat ultrasound by a consultant radiologist and management by obstetric specialists. We covered the costs of such unexpected procedures on their behalf.

We explained the nature and process of the trial to potential participants if they met the inclusion criteria. Oral and written information was available in English, Nepali, and Maithili. We deferred consent until women had discussed the trial with their families, and we encouraged them to bring senior family members to the

study centre for the consent process. Literate participants provided signed consent and those unable to write provided witnessed thumbprints.

The trial was approved by the Nepal Health Research Council and the ethics committee of the Institute of Child Health and Great Ormond Street Hospital for Children, London, UK, and was undertaken in collaboration with His Majesty's Government Ministry of Health, Nepal. Benefits to participants included the supply of supplements, free health care, and expedited referral in the event of complications. Information provided by participants remained confidential. Access was restricted to supervisory and research staff at the analytical level. No analyses or outputs included the names of participants.

### Procedures

We did randomisation in advance of recruitment. One of us (DO) randomly allocated 1200 participant identification numbers by computer into two groups in permuted blocks of 50. The allocation code was kept on file in Kathmandu and London. We allocated every identification number a supplement container to last throughout the trial. Containers were filled with either intervention or control tablets in Kathmandu by a team member who was otherwise uninvolved in the trial; these containers were then marked only with identification numbers and transported to the study centre in Janakpur. Intervention and control supplements were manufactured by Danish Pharmaceutical Industries (Ballerup, Denmark) to look, smell, and taste identical.

After screening, consent, and enrolment, one of us (YS) allocated participants sequential identification numbers and the corresponding supplement containers. Supplements were provided in a take-home bottle at monthly visits to the study centre. Bottles were labelled with the participants' names and identification numbers only. The allocation code was broken on two occasions: (1) for the first 500 participants to inform the interim data monitoring committee (by DO: the committee were not told which group represented the intervention); and (2) for this analysis.

In accordance with recommendations for trial design,<sup>43</sup> controls received current nationally advised tablets containing iron 60 mg and folic acid 400  $\mu\text{g}$ .<sup>41</sup> The intervention group received tablets containing vitamin A 800  $\mu\text{g}$ , vitamin E 10 mg, vitamin D 5  $\mu\text{g}$ , vitamin B1 1.4 mg, vitamin B2 1.4 mg, niacin 18 mg, vitamin B6 1.9 mg, vitamin B12 2.6  $\mu\text{g}$ , folic acid 400  $\mu\text{g}$ , vitamin C 70 mg, iron 30 mg, zinc 15 mg, copper 2 mg, selenium 65  $\mu\text{g}$ , and iodine 150  $\mu\text{g}$ . These amounts adhere to the suggested composition of multiple micronutrient supplements for antenatal use recommended by UNICEF, WHO, and the United Nations University.<sup>26</sup> We sent a sample of tablets from participants' allocations for composition analysis midway through the trial. Vitamin E concentrations were 25% higher than expected, a surplus added by the manufacturer in view of probable

degradation. Retinol, iron, and vitamin C concentrations were about 10% lower than expected.

Participants received supplements from enrolment (at no earlier than 12 weeks' gestation) to delivery. We advised women to take one tablet daily, preferably after food and at the same time, and to avoid other supplements and drugs unless recommended by a study obstetrician. The follow-up plan included a contact visit every 2 weeks—a combination of monthly clinic visits and monthly home visits. At every contact visit, we asked the participant about morbidity, questions about the trial, her plans for delivery, and her consumption of supplements. We encouraged all women to make plans for delivery and advised them to consider hospital birth.

We offered the following blood tests to all participants at enrolment: haemoglobin concentration; blood group; rhesus status; and rapid plasma reagin test for syphilis. We prepared blood samples and stored them for micronutrient assays with the woman's permission. We undertook urine dipstick testing for protein and sugar at every antenatal care visit, and we offered repeat testing for blood haemoglobin concentration and amounts of selected micronutrients at about 32 weeks' gestation.

We assayed blood haemoglobin spectrophotometrically with a HemoCue system (Dronfield, UK), with daily calibration checks. Blood samples were spun in a centrifuge and plasma was stored at  $-20^{\circ}\text{C}$  until transport to the UK in liquid nitrogen, after which samples were stored at  $-80^{\circ}\text{C}$ . We measured vitamins A and E in a randomly selected 10% subsample of specimens taken at enrolment and 32 weeks' gestation. Plasma retinol and  $\alpha$  tocopherol concentrations were assayed simultaneously by high performance liquid chromatography.<sup>44</sup> We standardised the method with control serum purchased from the National Institute of Standards and Technology (Gaithersburg, MD, USA) and quality control samples were run within every batch. The interassay coefficient of variation for the quality control plasma was 6% for both retinol and  $\alpha$  tocopherol.  $\alpha$  tocopherol was expressed as a ratio to plasma triglycerides, measured on a COBAS Fara autoanalyser with a commercial kit (Roche Diagnostics, Basel, Switzerland). The interassay coefficient of variation for triglycerides was 4.6%.

In the event of significant illness, we arranged for the participant to be seen by a consultant obstetrician or doctor. We prespecified two deviations from protocol: (1) if a participant's enrolment blood haemoglobin concentration was less than 70 g/L, she was given an extra 60 mg of iron daily, anthelmintic treatment, and her blood haemoglobin was rechecked after 1 month; and (2) if a participant described night blindness at any time, she was given 2000  $\mu\text{g}$  of vitamin A daily and referred for medical follow-up.

For hospital births, study team midwives identified participants presenting at the maternity unit and obtained birth details and infant anthropometric measurements. For home births, we encouraged participants' families to

contact the study team, after which a field supervisor visited as soon as possible to collect birth details and undertake infant anthropometry. Babies who became unwell were referred for management at the hospital paediatric unit. Women exited the study when their babies reached 1 month of age, at which time mother and child attended for postnatal checks. We met the cost of any medical care needed during pregnancy, delivery, or postpartum and transport for deliveries or emergencies.

Primary outcomes were birthweight and gestational duration. We measured birthweight on Seca 835 electronic scales (Hamburg, Germany) accurate to 10 g, tared before every measurement. We attempted to establish birthweight as soon after birth as possible, but defined late birthweight as a measurement recorded after 72 h. We estimated gestational age on the basis of transabdominal ultrasound fetal biometry with an Aloka SSD 900 with obstetric probe (Tokyo, Japan). In pregnancies less than 13 weeks and 6 days, we used crown-rump length and the chart of Robinson.<sup>45</sup> Between 14 and 20 weeks, we used biparietal diameter and head circumference and the charts of Chitty.<sup>46</sup> We added in measurements of femur length if necessary. One of us (AV) did the scans, with the exception of 30 women scanned by DO. Ultrasound training and quality control were provided by the superintendent ultrasonographer of University College London Hospitals. Scan stills were printed and stored in the participant's file, and scan videotapes were sent to the UK for regular quality control examination.

Secondary outcomes included infant length and head circumference. We measured infant length on a Kiddimetre board accurate to 1 mm (Raven Equipment, Castlemead, UK) for hospital and home births where vehicular access was possible. When severe monsoon conditions made transport of the large, heavy Kiddimetre to homes impractical, we measured some infant lengths on a Rollametre (Raven Equipment). We assessed occipitofrontal head circumference with a plastic length tape accurate to 1 mm, taking the middle value of three consecutive measurements.

We defined loss to follow-up as failure to attend the antenatal clinic for 3 months and failure to meet the participant after three home visits. We identified miscarriage as cessation of confirmed pregnancy before 23 weeks' gestation; stillbirth as delivery of an infant showing no signs of life—movement, breathing, or heartbeat—after 23 weeks' gestation; early neonatal death as death of a liveborn infant in the first 7 days after birth; and late neonatal death as death of a liveborn infant after 7 but within 28 days.

We assessed adherence by the discrepancy estimate method.<sup>47</sup> Supplements were provided every month in a varying (but known) quantity in excess of requirements. Team members were aware of the numbers of tablets remaining and added to take-home bottles, but did not imply to participants that they were important. We told

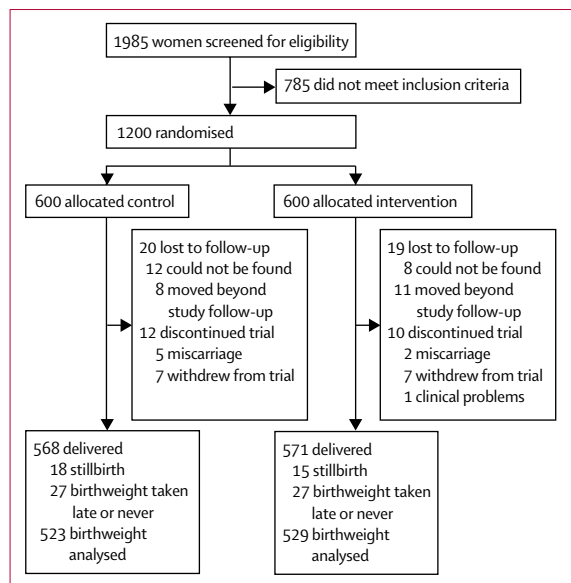


Figure: Trial profile

participants that the bottles contained more than enough tablets to last a month, but that they were formulated for pregnancy and should not be given to other family members.

### Statistical analysis

We calculated the sample size at a power of 90% and a two-sided significance level of 5%. We allowed for the possibility of 30% loss to follow-up, a figure that seemed pessimistic but reasonable in view of the logistic difficulties of such trials. At 600 participants per arm, the study would attribute significance to a change in mean birthweight of 100 g (assuming a control mean of 2800 g [SD 450]) and a change in mean gestational duration of 3 days (assuming a control mean of 275 days [12.4]). Analysis was by intention to treat.

An independent data monitoring committee met in November, 2003, to consider interim data from 500 participants. The committee reported that there had been high adherence to the study protocol, that randomisation had resulted in comparable groups, that no evidence of harm could be attributed to either supplement, that there was no evidence of substantial mortality differences, and that the sample size should remain as projected.

We obtained information about participants, their progress, and outcomes in individual files that we checked manually for completeness. Data were entered into a relational database management system (FileMaker Pro 5.5), which incorporated validation constraints. We subjected the data to range checks and case-by-case examination for completeness and accuracy after export to SPSS version 11. Birthweight and gestational duration were normally distributed and were assessed with independent sample *t* tests assuming equal variances. We

assessed categorical outcomes (low birthweight and preterm birth) with logistic regression. The distribution of plasma retinol conformed to normality. Data for  $\alpha$  tocopherol/triglycerides needed natural logarithmic transformation, and geometric means were calculated.

### Role of the funding source

The sponsor had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

### Results

The first participant joined the trial on Aug 11, 2002, and the last on Oct 22, 2003; all women had exited the trial by July, 2004. The figure shows the trial profile. Most exclusions from enrolment were for gestations of more than 20 weeks. Maternal illnesses that led to exclusion were: recently treated recurrent cysticercosis (1); need for chlorpromazine (1) or anticoagulant (1) drugs with changing doses; and symptomatic mitral stenosis (1) or multivalvular heart disease (1). Fetal exclusions were: twin pregnancies (6); anencephaly (1); occipital meningocele (1); encephalocele (1); duodenal atresia (1); and a grossly dilated pelvicalyceal system (1). All participants received their allocated supplements, irrespective of whether they took any. 20 participants enrolled in the trial but were never seen again, even after a thorough search in the areas they had given as their addresses. 19 moved out of the areas in which they could be tracked and we do not know their birth outcomes. Seven participants had spontaneous abortion. 14 women withdrew from the trial because they felt it would not benefit them. One withdrew after developing generalised itching. In deviations from protocol, four participants received treatment for severe anaemia and three for night blindness.

Data for 1139 deliveries were available for the analysis of gestational duration. Because most stillborn infants were not weighed, we included only liveborn infants in the analysis of birthweight. The birthweight outcome was available for 523 (87%) infants in the control group and 529 (88%) in the intervention group.

Table 1 shows baseline characteristics. Both arms were comparable. Most participants belonged to the Maithil group, traditional residents of the area: 518/599 (86%) in the control group and 499/599 (83%) in the intervention group. The remaining women were mainly from Muslim and hill Indo-Aryan groups. Half the participants came from Janakpur municipality and most came from families who owned some land and were supported by non-agricultural income from shops and small-scale manufacture. About half of participants had been to school, at least at primary level. 540 (45%) were in their first pregnancies. Enrolment weight was measured at mean 16.0 weeks (SD 3.1) in the control group and 15.7 weeks (3.7) in the intervention group. Although

	Control (n=600)	Intervention (n=600)
<b>Residence</b>		
Urban	316 (53%)	314 (52%)
Rural	284 (47%)	286 (48%)
<b>Land owned</b>		
None	39 (7%)	29 (5%)
≤10 kattha*	312 (52%)	337 (56%)
>10 kattha*	247 (41%)	227 (38%)
<b>Husband's occupation</b>		
No work	61 (10%)	69 (12%)
Farming	92 (15%)	89 (15%)
Salaried	252 (42%)	261 (44%)
Small business	114 (19%)	109 (18%)
Waged labour	66 (11%)	53 (9%)
Student	8 (1%)	9 (2%)
Out of country	7 (1%)	10 (2%)
<b>Age</b>		
<20 years	171 (29%)	190 (32%)
20–29 years	398 (66%)	387 (65%)
≥30 years	31 (5%)	23 (4%)
<b>Schooling</b>		
None	271 (45%)	273 (46%)
Primary	67 (11%)	56 (9%)
Lower secondary or higher	262 (44%)	271 (45%)
<b>Parity</b>		
0	266 (44%)	274 (46%)
1–2	261 (44%)	276 (46%)
≥3	73 (12%)	50 (8%)
Enrolment weight (kg)	45.1 (6.0)	45.1 (6.2)
Height (cm)†	151.0 (5.7)	150.5 (5.4)
Enrolment body-mass index (kg/m <sup>2</sup> )	19.8 (2.4)	19.9 (2.4)
<18.5 kg/m <sup>2</sup>	170 (28%)	172 (29%)

Data are number of participants (%) or mean (SD). \*10 kattha is about 0.3 hectares.  
†Control, n=598; intervention n=599.

**Table 1: Baseline household and participant characteristics**

mean height was not strikingly short, mean weight at enrolment was low at 45 kg, and 342 (29%) women had a low body-mass index. Participants who were lost to follow-up or withdrew from the trial were less likely to be Maithil and were wealthier than those who remained.

The mean period of potential supplementation was 158 days (SD 30) in the control group and 161 days (29) in the intervention group. Discrepancy estimates of adherence included all enrolled participants: if they withdrew from the trial or were lost to follow-up, we calculated their days of involvement and assumed that they had taken no supplements. We assessed adherence in terms of the number of tablets used over the period of participation, which relies on the assumption that they had been consumed by the participant. The distribution of adherence was J-shaped with clustering towards 100%. Consumption accounted for a median 98% of days of participation in the control group (IQR 91–100) and 97% in the intervention group (91–100).

Blood haemoglobin samples were available for 1054 women at enrolment and for 1050 in the third trimester (table 2). Enrolment blood samples were taken at a mean gestation of 16.3 weeks (SD 3.0) in the control group and 16.1 weeks (2.9) in the intervention group; third trimester samples were taken at 31.4 weeks (2.0) and 31.6 weeks (2.2), respectively. Blood haemoglobin

concentrations did not differ between arms at enrolment. 401 (38%) women were anaemic at enrolment (<110 g/L) but severe anaemia was rare (one participant in the control group and three in the intervention group had haemoglobin concentrations <70 g/L). Allocation to either supplement was associated with a fall in the prevalence of anaemia. At enrolment, plasma retinol concentration was assayed in 127 participants and vitamin E in 108; respective numbers for the third trimester were 101 and 91. Micronutrient supplementation resulted in significantly higher retinol ( $p=0.01$ ) and vitamin E ( $p=0.03$ ) concentrations at around 32 weeks' gestation.

Of 568 deliveries in the control group, 300 (53%) took place in hospital and 43 (8%) were caesarean sections. In the intervention group, 346 (61%) of 571 deliveries were hospital births and 46 (8%) were caesarean sections. Table 3 presents the analysis of primary and secondary outcomes. 1052 birthweights were available for analysis; 832 (79%) were taken on the first day, 184 (17%) on the second, and 36 (3%) on the third. Birthweight was greater in the intervention group than in the control group; this difference barely changed when restricted to infants born at term. The intervention was associated with a 25% relative reduction in the prevalence of low birthweight. Although gestational duration was 1.5 days longer in the intervention group than in the control group, the difference was not significant. Birth length and infant head circumference did not differ between arms.

Mean birthweight was 91 g higher in male than in female infants (2817 g vs 2726 g; [95% CI for the difference 38–144]), 150 g greater in infants of multiparous than primiparous women (2839 g vs 2689 g; [97–202]), and 116 g higher in infants of participants whose body-mass index was 18.5 kg/m<sup>2</sup> or more compared with less than 18.5 kg/m<sup>2</sup> (2804 g vs 2688 g; [57–175]). Table 3 also summarises the birthweight outcome after stratification for these factors.

The study was not powered to detect differences in mortality, and none of the differences shown in table 4 is

	Control group	Intervention group
<b>Haemoglobin</b>		
Enrolment sample (n)	517	537
Mean (SD) concentration (g/L)	115 (16)	115 (16)
<110 g/L (n [%])	200 (39%)	201 (37%)
Third trimester sample (n)	517	533
Mean (SD) concentration (g/L)	118 (14)	118 (12)
<110 g/L (n [%])	148 (29%)	133 (25%)
<b>Retinol</b>		
Enrolment sample (n)	67	60
Mean (SD) concentration (μmol/L)	1.11 (0.32)	1.17 (0.39)
Third trimester sample (n)	56	45
Mean (SD) concentration (μmol/L)	1.20 (0.39)	1.39 (0.33)
<b>α-tocopherol/triglycerides</b>		
Enrolment sample (n)	56	52
Geometric mean (95% CI) ratio (μmol/mmol)	12.9 (11.7–14.2)	13.3 (11.8–14.8)
Third trimester sample (n)	52	39
Geometric mean (95% CI) ratio (μmol/mmol)	10.7 (9.8–11.6)	12.6 (11.1–14.4)

**Table 2: Maternal nutritional biochemistry**

	Control	Intervention	Difference (95% CI)	p
<b>Primary and secondary outcomes</b>				
Birthweight (g)	2733 (422) [n=523]	2810 (453) [n=529]	77 (24 to 130)	0.004
Gestation at birth (weeks)	39.2 (2.0) [n=568]	39.4 (1.9) [n=571]	0.2 (-0.1 to 0.4)	0.12
Birth length (cm)	48.6 (3.2) [n=517]	48.9 (2.9) [n=526]	0.3 (-0.1 to 0.6)	0.16
Head circumference (cm)	33.6 (2.2) [n=519]	33.8 (2.2) [n=526]	0.2 (-0.1 to 0.4)	0.18
<b>Stratified birthweight outcomes</b>				
Infant sex				
Female	2672 (399) [n=260]	2780 (429) [n=256]	108 (36 to 179)	0.003
Male	2794 (437) [n=261]	2838 (474) [n=273]	44 (-33 to 122)	0.261
Mother's parity				
Primigravid	2664 (404) [n=231]	2714 (418) [n=242]	50 (-24 to 124)	0.189
Multigravid	2787 (428) [n=292]	2891 (467) [n=287]	104 (31 to 177)	0.005
Mother's body-mass index				
<18.5 kg/m <sup>2</sup>	2661 (443) [n=148]	2715 (402) [n=145]	54 (-43 to 152)	0.274
≥18.5 kg/m <sup>2</sup>	2762 (410) [n=374]	2845 (467) [n=383]	83 (20 to 146)	0.010
<b>Categorical outcomes</b>				
Low birthweight	133/523 (25%)	101/529 (19%)	0.69 (0.52 to 0.93)*	0.014
Preterm	54/568 (10%)	47/571 (8%)	0.85 (0.57 to 1.29)*	0.45

Data are mean (SD) [n] unless otherwise indicated. \*Odds ratio (95% CI).

Table 3: Primary, secondary, and stratified outcomes

significant. More stillbirths were reported in the control group and more early neonatal deaths in the intervention group. Overall perinatal mortality rates were, therefore, similar. A combination of verbal autopsy and clinical assessment suggested that the commonest causes of death were infection, preterm birth, and birth asphyxia. No obvious imbalance was noted between the groups with respect to these factors.

Morbidity was both a potential outcome and a potential adverse effect. Typical antenatal problems were gastrointestinal symptoms (nausea, dyspepsia, abdominal pain) and backache. We recorded no differences between the groups in morbidity or reported problems during pregnancy or in incidence of complications such as failure to progress, retained placenta, and postpartum haemorrhage. Seven participants in the control group and two in the intervention group were treated for clinical eclampsia. No differences were seen between the groups in postpartum morbidity reports. Likewise, in infants we recorded no differences in modified Apgar scores, cough, breathing difficulties, diarrhoea, feeding problems, or fever. Two infants in the control group had identifiable congenital anomalies (talipes equinovarus; cleft lip and

	Control group	Intervention group
Births	568	571
Stillbirths	18	15
Livebirths	550	556
Neonatal deaths	11	17
Early neonatal deaths	5	13
Late neonatal deaths	6	4
Perinatal deaths	23	28
Neonatal mortality rate (per 1000 livebirths)	20.0	30.6
Early neonatal mortality rate (per 1000 livebirths)	9.1	23.4
Late neonatal mortality rate (per 1000 livebirths)	10.9	7.2
Stillbirth rate (per 1000 births)	31.7	26.3
Perinatal mortality rate (per 1000 births)	40.5	49.0

Table 4: Mortality outcomes

palate), as did two in the intervention group (acyanotic congenital heart disease; tracheo-oesophageal fistula, imperforate anus, and preterm birth). Both infants in the intervention group died in the neonatal period.

## Discussion

We have shown that antenatal supplementation with a multiple micronutrient preparation was associated with increased birthweight when compared with a standard iron and folic acid preparation. Gestational duration was not affected by supplementation. We achieved high retention rates, and imprecision was restricted to tolerances implied by use of electronic scales for weighing, a 72-h window for measurement of birthweight, and ultrasound biometry for gestational assessment.

A trial of this type was undertaken in Tanzania and included 1067 women infected with HIV-1.<sup>25</sup> Supplementation with multivitamins at 2–20 times the recommended daily allowance in the third trimester was associated with a 44% fall in low birthweight and reductions in preterm deliveries (39%), small-for-gestational age infants, and fetal death. In a double-blind randomised controlled trial in Mexico,<sup>48</sup> a supplement containing 1–1.5 times the recommended daily allowance of ten vitamins and three minerals was compared with iron alone. No effects were reported on birthweight, gestational age, or proportion of low birthweight infants. Although the design maximised compliance, loss to follow-up was appreciable (229/874) and—more importantly—a third of participants were overweight and the rate of low birthweight was less than 9%.

In a double-blind cluster randomised controlled trial from a district in southern Nepal, 426 villages received one of five supplement regimens.<sup>39</sup> The researchers noted that folic acid and iron supplementation were associated with a non-significant 60 g increase in birthweight over control or folic acid alone. This effect was not seen when zinc was added to folic acid and iron. Multiple micronutrient supplements (in a generally similar formulation to our study that also contained 100 mg of magnesium, 30 mg of zinc, 60 mg of iron, but no selenium) were associated with a significant 64 g rise in birthweight over control. The interpretation of this trial<sup>39</sup> has been that multiple micronutrients confer no added benefit over that of iron and folic acid supplements alone.

A double-blind placebo-controlled randomised trial has been reported from Harare, Zimbabwe.<sup>49</sup> Participants were enrolled at 22–35 weeks' gestation and outcomes were available for only 66%, but the trial is valuable for having included many women with HIV infection. A 50 g increase in birthweight in the supplemented group (in a slightly different tablet composition from our study) was not significant, but birthweights in this population were higher than in south Asia, and the rate of low birthweight was only 10.5%. The concentration of low birthweight in south Asia, with higher birthweights in general in sub-Saharan

Africa, does have implications for roll-out of potential programmes.

How far can our findings be generalised? Micronutrient supplementation has its adherents and its critics. Supplementation began at about 4 months' gestation and lasted until delivery. This strategy seems feasible in settings where antenatal care uptake is high, and continuation into the postpartum period is also a possibility. In settings like rural Nepal where uptake is low, the effectiveness of supplementation programmes has been limited. The discrepancy estimate method probably overestimated adherence, but the changes in plasma vitamin concentrations, the outcome of the study, and our own impression during implementation, all suggest that participants did take substantial numbers of tablets. Incidentally, it is noteworthy that even with regular support and encouragement and an assurance of exemption from fees, only 50–60% of participants availed themselves of hospital delivery.

The external validity of the findings would also be limited if the participants were an unusual group. Census figures suggest that the level of female literacy in Dhanusha district is about 36%; 37% of children attend primary and 18% lower secondary school. In a previous Nepalese trial,<sup>39</sup> 20% of participants were literate and 90% delivered at home. Comparison of these figures with those of our study confirms our impression that the sample was a mix of urban and rural women and favoured the moderately poor population rather than the extreme rural poor or the wealthy: participants had somewhat more schooling and were more literate, were usually from families involved in small-scale urban and periurban businesses, and were less likely to be anaemic than average survey figures suggest.<sup>37</sup> Likewise, birth-weights and low birthweight rates accord with estimates that probably under-represent the rural poor.<sup>38</sup>

Research is justified if the populations in which it is done are likely to benefit from the results.<sup>43</sup> Does an improvement in birthweight of this size translate into reduced mortality or morbidity? Is the improvement associated with better developmental indices? Does it have long-term health benefits? These questions remain open. In common with some other investigators,<sup>50</sup> we suggest that changes in birthweight at term might not have resounding effects. Modest changes in rates of preterm birth or increases in gestational duration could have stronger effects on mortality,<sup>13</sup> but large studies would be necessary to describe them. We did not record significant effects on morbidity or mortality and agree with other authors that multiple micronutrient supplementation needs more evaluation before we consider large-scale programmes.<sup>27,51</sup> We draw attention to the high early neonatal mortality in the intervention group, which—although not significant—is similar to a finding reported by Christian and colleagues.<sup>52</sup> Our study did not address the particular issue of multiple micronutrient supplements for women living with HIV,

in whom impressive effects of high doses of selected micronutrients have been described in Tanzania.<sup>25</sup>

Our tentative finding of a differential effect of the supplement is interesting. The effect of multiple micronutrients on fetal weight seems to have been enhanced in female infants, in births of higher order, and in babies of women with a greater body-mass index. The growth potential for fetuses of larger, parous women is high, but female fetuses are usually smaller than males, and the finding of a greater increment in females is somewhat counterintuitive. We look forward to further work and opinions on our findings.

The public-health implications of our findings await confirmation by the results of other studies currently underway in Bangladesh, Burkina Faso, China, Guinea Bissau, Indonesia, and Pakistan.

#### Contributors

The investigators are members of an international study group, which met in 2002 (<http://www.ich.ucl.ac.uk/ich/html/academicunits/cich/pdfs/micronutrientmi.pdf> [accessed Feb 15, 2005]) and 2004. All authors contributed to implementation of the study and criticised drafts of the paper. D Osrin was responsible for study protocols, methods, and electronic database, advised on implementation, did the analysis, and wrote the first draft of the paper. A Vaidya was the clinical coordinator of the study, undertook ultrasound and medical assessment, entered data, and did the analysis. Y Shrestha supervised all nursing and maternity aspects of the study and undertook data collection and allocation. R B Baniya supervised field activities and was the programme manager in Dhanusha. D S Manandhar and R K Adhikari were principal investigators and took overall responsibility for the study in Nepal. S Filteau, A Tomkins, and A M de L Costello were principal investigators in the UK and contributed to study design and writing of the report.

#### Conflict of interest statement

In the planning phase of the study, DO, SF, and AT attended an international principal investigators' meeting funded by the Micronutrient Initiative. After study completion, but before this paper was written, AV, DSM, and AT attended a second meeting funded by UNICEF. The other authors declare that they have no conflict of interest.

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