Report

Iodine supplementation in pregnant women living in mild-tomoderately iodine deficient areas in India and Thailand: effects on pregnancy outcome and infant development
The MITCH Study

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	Foundation
Independent physician(s)	**To be appointed locally**
Laboratory sites <*if applicable*>	**1. St. John's Medical College and Research Institute, Bangalore, India** **2. Institute of Nutrition, Mahidol University (INMU), Bangkok, Thailand** **3. Dept. of Endocrinology, University Children's Hospital, Switzerland**
Pharmacy <*if applicable*>	***Not applicable***

PROTOCOL SIGNATURE SHEET

Name	Signature	Date
Sponsor or legal representative: ***<please include name and function>*** **For non-commercial research,** **Head of Department:** ***Prof. Dr. Frans Kok***		
Coordinating Investigator/Project leader/Principal Investigator: ***Dr. Alida Melse-Boonstra***		

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LIST OF ABBREVIATIONS AND RELEVANT DEFINITIONS

ABR	**ABR form (General Assessment and Registration form) is the application form that is required for submission to the accredited Ethics Committee (ABR = Algemene Beoordeling en Registratie)**
AE	**Adverse Event**
AR	**Adverse Reaction**
CA	**Competent Authority**
CCMO	**Central Committee on Research Involving Human Subjects**
CV	**Curriculum Vitae**
DSMB	**Data Safety Monitoring Board**
EU	**European Union**
EudraCT	**European drug regulatory affairs Clinical Trials GCP Good Clinical Practice**
IB	**Investigator's Brochure**
IC	**Informed Consent**
IMP	**Investigational Medicinal Product**
IMPD	**Investigational Medicinal Product Dossier**
METC	**Medical research ethics committee (MREC); in Dutch: medisch ethische toetsing commissie (METC)**
(S)AE	**Serious Adverse Event**
SPC	**Summary of Product Characteristics (in Dutch: officiële productinfomatie IB1-tekst)**
Sponsor	**The sponsor is the party that commissions the organisation or performance of the research, for example a pharmaceutical company, academic hospital, scientific organisation or investigator. A party that provides funding for a study but does not commission it is not regarded as the sponsor, but referred to as a subsidising party.**
SUSAR	**Suspected Unexpected Serious Adverse Reaction**
Wbp	**Personal Data Protection Act (in Dutch: Wet Bescherming Persoonsgevens)**
WMO	**Medical Research Involving Human Subjects Act (Wet Medisch-wetenschappelijk Onderzoek met Mensen**

SUMMARY

Rationale: In regions of severe endemic goiter, the adverse effects of in utero iodine deficiency on neuromotor development are well established: randomized controlled trials of iodine supplements given to iodine deficient mothers before pregnancy or during early pregnancy improve motor and cognitive performance of their offspring. However, the potential adverse effects of mild-to-moderate iodine deficiency during pregnancy are unclear. Inadequate thyroid function in the fetus and newborn are the likely cause of brain damage in iodine deficiency.

Objective: To determine whether the daily oral administration of 200 µg iodine to pregnant women in areas of mild-to-moderate iodine deficiency improves maternal and newborn thyroid function, pregnancy outcome, birth weight, infant growth and cognitive performance.

Study design: Double-blind randomized controlled multicentre trial.

Study population: Pregnant women (18-40 years) presenting at the clinic for their first prenatal visit will be recruited at two research sites, namely St. Martha's hospital in Bangalore, India and Ramathibodi Hospital, Mahidol University, Bangkok, Thailand. At each site, 400 women will be recruited.

Intervention: Half of the women will be randomized to iodine treatment (200 µg per day) and the other half to placebo throughout pregnancy.

Main study parameters/endpoints: Differences between group means in indicators of thyroid function, birth outcome, urinary iodine, breast milk iodine, growth, and psychomotor development.

Nature and extent of the burden and risks associated with participation, benefit and group relatedness: In 2004, the dose, safety and efficacy of iodine supplementation in mild to moderately iodine-deficient pregnant women were reviewed from 6 European trials. In all 6 trials, supplementation resulted in a significant increase in maternal urinary iodine. Iodine doses varied between 50 and 230 µg per day, and the data indicate no clear dose response relationship for urinary iodine or parameters of thyroid function. Iodine supplements during pregnancy appeared to be generally safe; there was no increase in maternal thyroid autoimmunity, or in the prevalence or severity of postpartum thyroid dysfunction (PPTD) in the trials. For the newborn, most data suggest supplementation is safe, although increases in cord blood TSH upon maternal iodine supplementation have been reported, which are probably transient but which have not been followed up until now.

So far, no studies have been conducted in mild-to-moderate iodine deficient areas in which the effects of maternal iodine supplementation on child development have been followed up. Worldwide, millions of children are born each year unprotected against the damaging effects of mild iodine deficiency. This large multicentre study will provide conclusive evidence on the

global need for iodine supplementation during pregnancy in countries where moderate-to-mild iodine deficiency is prevalent.

1. INTRODUCTION AND RATIONALE

Severe iodine deficiency during pregnancy increases the risk of spontaneous abortion, low birth weight and infant mortality (1, 2-6). In regions of severe endemic goiter, the adverse effects of in utero iodine deficiency on neuromotor development are well established: randomized controlled trials of iodine supplements given to iodine deficient mothers before pregnancy or during early pregnancy improve motor and cognitive performance of their offspring (7-9). A meta-analysis of cross sectional studies suggested severe iodine deficiency in a population lowers mean IQ scores by 13.5 points (10). However, the potential adverse effects of mild-to-moderate iodine deficiency during pregnancy are unclear.

Inadequate thyroid function in the fetus and newborn are the likely cause of brain damage in iodine deficiency. Maternal thyroxine (T4) crosses the placenta to support neural development before onset of fetal thyroid function at 10-12 week (11). Maternal T4 still represents up to 20-40% of T4 measured in cord blood at birth (12). Two prospective case-control studies, using different measures of impaired maternal thyroid function, have reported developmental impairment in offspring of affected mothers. In the study by Haddow et al (13), 7-9 yr-old children of mothers with subclinical hypothyroidism during pregnancy (an increased thyroid stimulating hormone (TSH) in the 2nd trimester) were compared to children from mothers with normal thyroid function during pregnancy. None of the children had hypothyroidism as newborns. They were given 15 tests relating to intelligence, attention, language, reading ability, school performance, and visual-motor performance. The children in the first group (n=62) performed slightly less well on all 15 tests. Their IQ scores on the Wechsler Intelligence Scale for Children averaged 4 points lower than those of the 124 matched controls (13).

Pop et al (14) reported a prospective study of pregnant women and their children up to the age of 2 years. Child development was assessed using the Bayley Scales of Infant Development in children of women with hypothyroxinemia (free T4 below the tenth percentile at 12 weeks' gestation) (cases), and in children of women with free T4 between the 50th and 90th percentiles at 12 weeks' gestation (controls). Infants from mothers whose maternal hypothyroxinemia did not improve over the course of pregnancy had delayed mental and motor function compared to controls: 10 index points on the mental scale and eight on the motor scale at the age of 1 year, as well as eight index points on the mental, and 10 on the motor scale at the age of 2 years. A more recent study suggested maternal hypothyroxinemia is a risk factor for neurodevelopmental difficulties that can be identified in neonates as young as 3 weeks of age (15). Despite the potential limitations of their case-control design, these studies suggest even marginal maternal thyroid dysfunction can cause fetal harm. However, in these studies, the maternal thyroid abnormalities were not due to iodine deficiency.

Zimmermann and Delange (16) reviewed the potential need for iodine supplementation to pregnant women in Europe**;** the dose, safety and efficacy of iodine supplementation in mild to moderately iodine-deficient pregnant women were reviewed from 6 trials. In all 6 trials, supplementation resulted in a significant increase in maternal urinary

iodine. Iodine doses varied between 50 and 230 µg per day, and the data indicate no clear dose response relationship for urinary iodine, TSH, Thyroglobulin, thyroid hormones, or thyroid volume. Iodine supplements during pregnancy appear to be generally safe; there was no increase in maternal thyroid autoimmunity, or in the prevalence or severity of postpartum thyroid dysfunction in the trials. For the newborn, most data suggest supplementation is safe. A tendency to a slight increase in neonatal TSH level after iodine supplementation may be of little importance, as, in these infants, the serum concentration of T4 (which may be the major thyroid hormone influencing brain development) does not show a concomitant reduction.

Both India and Thailand are countries where mild-to-moderate iodine deficiency is still prevalent despite public health measures through production of iodized salt. Southern India is iodine deficient (17), and it is estimated that household coverage with adequately iodized salt is only 40-50% (1, 17). Mild iodine deficiency is common among Indian pregnant women (18-19). A recent study found that 17% of Indian newborns had transient elevations of serum TSH > 5 mU/ml, indicating iodine deficiency in this critical period (20). A recent Indian review emphasized the pressing need for controlled trials evaluating micronutrient interventions in pregnancy (21). In 2005, the Ministry of Public Health in Thailand reported data on iodine deficiency (using median urinary iodine cut-off levels) in pregnant women. The percentage of iodine deficiency was shown to have increased from 2000 to 2004. Over these years, the percentage of mild (median urinary iodine <100 µg/l) and moderate (median urinary iodine <50 µg/l) iodine deficiency increased from 34.4% to 49.4% and from 14.9% to 25.5%, respectively.

Therefore, this study aims to determine the effects of daily oral iodine supplementation in pregnant women with mild-to-moderate iodine deficiency on pregnancy outcome and infant development in India and in Thailand.

2. OBJECTIVES

Primary Objective:

To determine whether the daily oral administration of 200 µg iodine to pregnant women in areas of mild-to-moderate iodine deficiency improves maternal and newborn thyroid function, pregnancy outcome, birth weight, infant growth and cognitive performance.

Secondary Objective(s):

To investigate new indicators for assessing iodine status in pregnancy and infancy: dried blood spot thyroglobulin, neonatal TSH and newborn median urinary iodine concentration.

3. STUDY DESIGN

The study will be a randomized double blind controlled clinical multicentre trial among pregnant women in low-to-middle income neighbourhoods with incomplete household iodized salt coverage in Bangalore, India and in Bangkok, Thailand. Women will be followed up at the 20-24th and 30-34th weeks of gestation and at delivery, with postnatal follow-up visits at 6 weeks, 6, 12 and 24 months. Maternal and infant measures will include: anthropometry, dietary assessment, birth outcomes, thyroid function, iodine nutrition, and infant development (see Table 1)

Table 1: Overview of the study schedule: baseline and follow-up visits

	Prenatal				Post natal				
Time of follow-up	Baseline ≤14 wks	FU 1 24 wks	FU 2 34 wks	FU 3 Delivery	FU 4 72 hrs, pre discharge	FU 5 6 wk	FU 6 6 mo	FU 7 12 mo	FU 8 24 mo
Eligibility assessment and informed consent	X								
Medical history and demographic data	X								
Maternal blood sample by venipuncture	X	X	X			X	X	X	X
Newborn/infant blood sample (cord at delivery, after by heelprick)				X	X	X		X	
Anthropometry	X	X	X			X	X	X	X
Dietary assessment Psychosocial assessment	X	X	X			X	X	X	X
Maternal urine sample (2 x 24-hr collections)	X	X	X		X	X	X	X	X
Infant urine sample (spot)					X	X	X	X	X
Maternal breast milk sample					X	X	X	X	X
Maternal thyroid ultrasound	X	X	X		X	X	X	X	X
Infant thyroid ultrasound					X	X	X	X	X
Infant development tests						X	X	X	X

4. STUDY POPULATION

4.1 Population (base)

In India, subjects will be recruited from the outpatient prenatal clinic at St. Martha's Hospital in Bangalore. On a weekly basis, 40 pregnant women come to the clinic for their first prenatal visit spread over two days. In Thailand, subjects will be recruited from the prenatal clinic at Ramathibodi Hospital of Mahidol University, a large teaching hospital in Bangkok, where approximately 100 pregnant women present weekly for their first prenatal visit spread over three days.

4.2 Inclusion criteria

1. Age 18-40 years;
2. Gestational age: ≤ 14 weeks (as judged by the date of the last menstrual period);
3. Single pregnancy;
4. Non-lactating;
5. Planned residence in the area for the duration of the study (3 years).

4.3 Exclusion criteria

6. TSH levels outside the range of 0.2- 4.0 mIU/L;
7. History of serious medical conditions such as HIV, hypertension, diabetes, renal, hepatic or cardiovascular disease, thyroid disorders, mental disorders;
8. Use of iodine supplement.

4.4 Sample size calculation

Power calculations based on an anticipated decrease in the prevalence of elevated newborn TSH indicates that a sample size of 250 pregnant women will be needed to have 80% power to detect a decrease of 0.8 (from an anticipated incidence of 10% in the control to 2% with treatment) in the proportion of elevated newborn TSH values with a significance level of 0.05 (two tailed). This is based on the prevalence of transiently elevated newborn TSH of 6-14% in areas of mild-moderate ID (Australia, Thailand, some European countries) and of 2% in iodine sufficient countries (such as Switzerland). Recruitment will be done anticipating over 35% loss to follow-up after delivery, thus up till children have reached the age of 2 years. Thus, a cohort of 400 pregnant women will be enrolled at each research site.

5. TREATMENT OF SUBJECTS

5.1 Investigational product/treatment

Women will be randomly assigned to receive a daily oral supplement of 200 µg iodine (as 260 µg KI; experimental group) or an identical placebo tablet (control group). The regimen will be taken daily from enrolment until delivery.

5.2 Use of co-intervention (if applicable)

Subjects are allowed to take any additional nutritional supplements that do not contain iodine. At both research sites, iron/folic acid supplements will be given to the women as standard procedure in the local hospitals.

5.3 Escape medication (if applicable)

Not applicable.

6. INVESTIGATIONAL MEDICINAL PRODUCT

Not applicable.

7. METHODS

7.1 Study parameters/endpoints

7.1.1 Main study parameter/endpoint

1) Thyroid function as determined by serum TSH, total T4 and T3, free T4 and T3, thyroid-binding globulin (TBG), thyroglobulin (Tg), anti-thyroid peroxidase (TPO) and anti-Tg antibodies; and by ultrasound measurement of maternal and infant thyroid gland volume;
2) Infant development as determined by newborn development assessed at 6 weeks of age using the Neonatal Behavioral Assessment Scale (16); and by the Bayley Scales of Infant Development (17) at 6 months, 1 and 2 years (13).

7.1.2 Secondary study parameters/endpoints (if applicable)

1) Pregnancy outcome, as assessed by:
    - Maternal: details of premature birth/ premature rupture of membranes, eclampsia, mode of delivery, maternal distress (if any)
    - Newborn: birth weight, length, head circumference, APGAR score
2) Urinary iodine concentration (maternal/ infant)
3) Breast milk iodine concentration

7.1.3 Other study parameters (if applicable)

1) Baseline health, socioeconomic and demographic data
    - Details on mother's age, parity, general health, previous pregnancy outcomes and socio-economic status will be obtained.
2) Dietary assessment using validated food frequency questionnaires and 24 hr recall
    - Estimation of maternal iodine intake, salt intake, use of iodized or non-iodized salt
    - Infant feeding practices, including duration and frequency of breastfeeding
3) Anthropometric measurements in mother and infant
    - Weight, height, mid-arm circumference (MAC) and head circumference (infant only)
4) Compliance
    - During each prenatal clinic visit, compliance with supplement use will be assessed in 3 ways: (a) Direct questioning (b) Capsule counting (c) Urinary iodine measurement

7.2 Randomisation, blinding and treatment allocation

Randomisation will be done by blocks per day and will be carried out by an experienced co-worker at each research site who is not involved in the research. These co-workers will lock the key of the code away throughout the study. There is no indication to break the randomisation code before the last follow-up measurements have been performed.

7.3 Study procedures

Socioeconomic status and demographics

Data on socioeconomic status, demographics and general data (e.g. mother's age, parity, previous pregnancy outcomes) will be collected by questionnaire.

Anthropometric measurements

Maternal weight, height and mid-upper arm circumference (MUAC):

- Weight will be measured after voiding using a beam balance or spring balance scale. The balance will be placed on a hard flat surface, and checked for zero-balance before each measurement. The subject will wear light clothing and will be asked to stand relaxed in the center of the platform and to look straight ahead. Body weight will be recorded to the nearest 0.1 kg. The balance will be calibrated with a set of weights regularly throughout the study, and whenever it is moved to another location.
- Height will be measured using a stadiometer with a sliding headpiece. Subjects will be asked to remove shoes and socks and to stand straight with the head positioned such that the Frankfurt plane is horizontal, feet together, knees straight, and heels, buttocks, and shoulder blades in contact with the vertical surface of the wall. Height will be recorded to the nearest millimeter. If the reading falls between two values, the lower reading will always be recorded. Successive measurements should agree within five millimeter.
- Mid-upper-arm circumference (MUAC) will be measured by using a flexible, non-stretch tape. The subject should stand erect and sideways to the measurer, with the head in the Frankfurt plane, arms relaxed and legs apart. If the subject is wearing a sleeved garment, it should be removed or the sleeves rolled up. The measurement is taken at the midpoint of the upper left arm, between the acromion process and the tip of the olecranon. After locating the midpoint, the left arm is extended so that it is hanging loosely by the side, with the palm facing inwards. The tape is wrapped gently but firmly around the arm at the midpoint without squeezing the arm. Measurements are taken to the nearest millimeter.

Infant weight, length, head circumference:

- A pediatric scale with a pan will be used for weighing infants and children less than two years of age. They will be weighed naked or with minimal clothing. Once the infant is lying quietly, weight is recorded to the nearest 10 g. If there is no alternative, the mother and infant will be weighed together, and then the mother alone, using a beam balance. The infant's weight will then be calculated by subtraction.
- Length of infants and children less than two years of age will be measured with a measuring board. The infant will lie down; face upward, with the head towards the fixed end and the body parallel to the long axis of the board. The shoulder blades should rest against the surface of the board and with the head so that the Frankfurt plane is vertical. The reading will be taken to the nearest millimeter. If the subject is restless, only the left leg will be positioned for the measurement.
- Head circumference: for the measurement of head circumference, a flexible non-stretch tape will be used. The mother will hold the infant with the left side facing the measurer, looking straight ahead so that the line of vision is perpendicular to the body

and the Frankfurt plane of the head is in a horizontal position. The tape is placed just above the supra-orbital ridges covering the most prominent part of the frontal bulge, and over the part of the occiput, which gives the maximum circumference. Measurements will be made to the nearest millimeter.

Dietary assessment:

Maternal dietary iodine intake will be assessed using 24-h recalls. An additional questionnaire will be used to record data on use of iodized salt, foods containing iodine such as seafood, fortified foods and supplements; and infant feeding practices including duration and frequency of breastfeeding.

Pregnancy outcome:

- Details of premature birth / premature rupture of membranes, eclampsia, duration of labor, cephalo-pelvic disproportion, mode of delivery, maternal distress and postpartum hemorrhage will be taken from the hospital record;
- Birth weight, length and head circumference of the infant will be measured;
- APGAR score at one, five and ten minutes will be recorded.

Thyroid volume:

Maternal and infant thyroid gland volume will be measured using a portable echo camera (Aloka, Mure, Japan), and thyroid volume will be calculated as follows:

Volume of each lobe (mL) = AP diameter (cm) x ML diameter (cm) x CC diameter (cm) x 0.479, and the lobe volumes will be summed.

Biological samples:

- Five-mL blood samples will be collected from mothers by venipuncture, from which serum will be derived and aliquoted into 1x1 ml and 2x500 µl. Blood spots will be collected by heel stick from infants and blotted onto high quality filter paper. The blood spot will be air dried and then stored in low gas permeable bags that contain desiccant to reduce humidity. Serum and blood spot samples will be frozen at -20°C. One aliquot of each will be packed on dry ice and shipped to the Laboratory for Human Nutrition, Zürich, Switzerland, and be analyzed at the Swiss Children's Hospital, University of Zürich, under supervision of Prof. Zimmermann. Serum TSH, total T4 and T3, free T4 and T3, and thyroid-binding globulin (TBG) will be analyzed by immunoassays. Anti-thyroid peroxidase (TPO) and anti-Tg antibodies will be measured by radioimmunoassay (RIA TgAb, RSR, Cardiff, United Kingdom). Thyroglobulin (Tg) will be measured on dried whole blood spots using an immunoassay (PerkinElmer Life Sciences, Wallac, Turku, Finland) adapted for dried blood spots (Zimmermann, 2003, 2006).
- Maternal spot urine samples will be collected in plastic cups. Infant urine samples will be collected with a special collection pad used as an inlay in the diaper. Urine samples will then be aliquoted into microcentrifuge tubes and frozen at -20°C until analyzed. Urinary iodine concentration will be measured using the Pino modification of the Sandel-Kolthoff reaction (Pino, 1996). Urinary creatinine will be measured using a modification of the

Jaffe method (Clarke, 1961). Urinary measurements will be performed locally in the laboratories at both research centers.

- Breast milk samples will be collected by hand excretion (or pump) straight into an eppendorf with cap and then frozen at -20°C until analyzed. Iodine concentration in breast milk will be measured using the Pino modification of the Sandell-Kolthoff reaction (Pino, 1996) (same procedure as urine sample analysis).

Infant growth and development testing:

- Newborn development will be assessed at 6 weeks of age using the Neonatal Behavioral Assessment Scale (Kooistra, 2006).
- Bayley Scales of Infant Development will be done at 6 months, 1 and 2 years (Pop, 1999). Local adaptations will be made for these tests.

7.4 Withdrawal of individual subjects

Subjects can leave the study at any time for any reason if they wish to do so without any consequences. The investigator can decide to withdraw a subject from the study for urgent medical reasons.

7.4.1 Specific criteria for withdrawal (if applicable)

Not applicable

7.5 Replacement of individual subjects after withdrawal

Subjects that withdraw during the first two weeks of enrolment will be replaced.

7.6 Follow-up of subjects withdrawn from treatment

Subjects that withdraw from the treatment will be followed up as scheduled.

7.7 Premature termination of the study

There are no criteria for premature study termination.

8. SAFETY REPORTING

8.1 Section 10 WMO event

In accordance to section 10, subsection 1, of the WMO, the investigator will inform the subjects and the reviewing accredited METC if anything occurs, on the basis of which it appears that the disadvantages of participation may be significantly greater than was foreseen in the research proposal. The study will be suspended pending further review by the accredited METC, except insofar as suspension would jeopardise the subjects' health. The investigator will take care that all subjects are kept informed.

8.2 Adverse and serious adverse events

Adverse events are defined as any undesirable experience occurring to a subject during a clinical trial, whether or not considered related to the investigational drug. All adverse events reported spontaneously by the subject or observed by the investigator or his staff will be recorded.

A serious adverse event is any untoward medical occurrence or effect that at any dose results in death;

- is life threatening (at the time of the event);
- requires hospitalisation or prolongation of existing inpatients' hospitalisation;
- results in persistent or significant disability or incapacity;
- is a congenital anomaly or birth defect;
- is a new event of the trial likely to affect the safety of the subjects, such as an unexpected outcome of an adverse reaction, lack of efficacy of an IMP used for the treatment of a life threatening disease, major safety finding from a newly completed animal study, etc.

All SAEs will be reported to the accredited METC that approved the protocol, according to the requirements of that METC.

8.2.1 Suspected unexpected serious adverse reactions (SUSAR)

Not applicable.

8.2.2 Annual safety report

Not applicable.

8.3 Follow-up of adverse events

All adverse events will be followed until they have abated, or until a stable situation has been reached. Depending on the event, follow up may require additional tests or medical procedures as indicated, and/or referral to the general physician or a medical specialist.

8.4 Data Safety Monitoring Board (DSMB)

A DSMB will be established consisting of the following persons:

- Prof. Dr. Richard Hurrell (Chair, Laboratory of Human Nutrition, ETH Zürich, Switzerland);
- Ms. Kaethe Santagate (Administration, Laboratory of Human Nutrition, ETH Zürich, Switzerland);
- Local member 1 (Institute of Nutrition, Mahidol University, Bangkok, Thailand)
- Local member 2 (St. John's Medical College, Bangalore, India)

Both local members will not be involved in the underlying study. In all three countries, the key to the treatment code will be kept in a sealed envelope, so that the code can be broken in case the DSMB sees reason for that. Any adverse events will be reported to the DSMB by the researchers on a monthly basis, whereas serious adverse events (SAE) will be reported and handled immediately.

9. STATISTICAL ANALYSIS

9.1 Baseline characteristics

Baseline characteristics of both treatment groups will be given as percentages, means with standard deviations, or medians with percentiles.

9.2 Maternal iodine status, thyroid function and pregnancy outcome

Differences in changes between treatment groups at 24 and 34 months of gestation and at delivery will be tested for statistical significance by t-test (two-sided, $\alpha = 0.05$) for the following maternal and newborn variables:

- Maternal urinary iodine;
- Maternal thyroid volume;
- Maternal serum thyroid hormone concentrations (Thyroid Stimulating Hormone (TSH), total T3 and T4, free T3 and T4), thyroid binding globulin (TBG), thyroglobulin (TG), anti-thyroid peroxidise (TPO) and anti-TG antibodies;
- Dietary iodine intake;
- Gestational age at delivery;
- Obstetric complications;
- Birth weight, length, head circumference and APGAR score of newborn;
- Cord blood thyroid hormone concentrations (TSH, total T3 and T4, free T3 and T4), TBG, TG;
- Infant urinary iodine;
- Breast milk iodine.

Statistical tests will be corrected for repeated measurements and multiple comparisons. Per protocol data analysis will be conducted, since no selection-bias in drop-out due to the type of treatment is to be expected.

9.3 Post-delivery follow-up data

Follow-up data collected from mothers and children at 6 weeks and 6, 12 and 24 months after delivery will be used to test for statistical significant differences between treatment groups by t-test for the following variables:

- Maternal and infant urinary iodine;
- Maternal and infant thyroid volume;
- Maternal and infant thyroid hormone concentrations (TSH, total T3 and T4, free T3 and T4), TBG, TG Maternal breast milk iodine;
- Maternal and infant dietary iodine intake;
- Differences in scores on the Neonatal Behavioral Assessment Scale and the Bailey Scales of Infant Development (psychomotor development index (PDI) and motor development index (MDI))

Univariate and multivariate analysis will be used to determine associations between maternal urinary iodine concentrations at 34 weeks of gestation and parameters of pregnancy outcome, maternal and infant thyroid function and child development at follow-up. Models will be adjusted for confounders and effect modifiers such as socioeconomic status, gestational age at delivery, initial iodine status, initial thyroid function, age, and parity. Logistic regression will be used to determine the impact of iodine supplementation on estimators of risk for impaired psychomotor development of the infant.

9.4 Interim analysis (if applicable)

Baseline data will be used for cross-sectional analysis without breaking the treatment code. Baseline urinary iodine concentrations will be associated with indicators of maternal thyroid function at the start of the study using multivariate regression models. Treatment codes will only be broken after all data have been collected, or in case the DSMB sees reason to break the codes.

10. ETHICAL CONSIDERATIONS

10.1 Regulation statement

The study will be conducted according to the principles of the Declaration of Helsinki (latest amendment: Tokyo 2004) and in accordance with the Medical Research Involving Human Subjects Act (WMO). We will follow the International Ethical Guidelines for Biomedical Research Involving Human Subjects issued by the Council for International Organizations of Medical Sciences (CIOMS; http://www.cioms.ch/guidelines_nov_2002_blurb.htm). These guidelines contain standard operating procedures according to Good Clinical Practice (GCP).

10.2 Recruitment and consent

After intake following the routine procedures for first-time visits of pregnant women in each hospital, women who are interested in taking part in the study will be referred to the research team. The study procedures will be explained orally. In India this will be done in English, or one of the local languages (Kannada, Tamil or Telugu), and in Thailand the explanation will be given in Thai. Women who decide to take part in the study will be asked to sign the consent form.

10.3 Objection by minors or incapacitated subjects (if applicable)

Not applicable.

10.4 Benefits and risks assessment, group relatedness

In 2004, the dose, safety and efficacy of iodine supplementation in mild to moderately iodine-deficient pregnant women were reviewed from 6 European trials. In all 6 trials, supplementation resulted in a significant increase in maternal urinary iodine. Iodine doses varied between 50 and 230 µg per day, and the data indicate no clear dose response relationship for urinary iodine or parameters of thyroid function. Iodine supplements during pregnancy appeared to be generally safe; there was no increase in maternal thyroid autoimmunity, or in the prevalence or severity of postpartum thyroid dysfunction (PPTD) in the trials. For the newborn, most data suggest supplementation is safe, although increases in cord blood TSH upon maternal iodine supplementation have been reported, which are probably transient but which have not been followed up until now.

So far, no studies have been conducted in mild-to-moderate iodine deficient areas in which the effects of maternal iodine supplementation on child development have been followed up. Worldwide, millions of children are born each year unprotected against the

damaging effects of mild iodine deficiency. This large multicentre study will provide conclusive evidence on the global need for iodine supplementation during pregnancy in countries where moderate-to-mild iodine deficiency is prevalent.

10.5 Compensation for injury

The investigators have a local institutional liability insurance. Also, insurance for participants in the trial is arranged according to the local guidelines.

10.6 Incentives (if applicable)

Travel costs and other costs due to participation in the study, such as costs for accommodation, will be reimbursed. Also certain medical costs can be reimbursed if health issues arise during physical examination or otherwise during the course of the trial and if the participant is not able to bear those costs through health insurance.

11. ADMINISTRATIVE ASPECTS AND PUBLICATION

11.1 Handling and storage of data and documents

Subjects will be given a unique identity number, the key to which will be kept in a data file protected with a password to which only the local project leaders have access. During data-analysis, data will be connected to ID numbers and not to personal information. Data will not be forwarded to third parties. Biological material will be encoded upon storage. Data and biological material will be stored for a maximum of five years after completion of the study. Documents that contain personal information will be safe locked, and a shredder or burner will be used for final destruction. In any publications resulting from the MITCH study, data will be presented in such a way that it cannot be related to a specific person.

11.2 Amendments

Amendments are changes made to the research after a favourable opinion by the accredited METC has been given. All amendments will be notified to the METC that gave a favourable opinion.

11.3 Annual progress report

The sponsor/investigator will submit a summary of the progress of the trial to the accredited METC once a year. Information will be provided on the date of inclusion of the first subject, numbers of subjects included and numbers of subjects that have completed the trial, serious adverse events/ serious adverse reactions, other problems, and amendments.

11.4 End of study report

The investigator will notify the accredited METC of the end of the study within a period of 8 weeks. The end of the study is defined as the last patient's last visit.
In case the study is ended prematurely, the investigator will notify the accredited METC, including the reasons for the premature termination.
Within one year after the end of the study, the investigator/sponsor will submit a final study report with the results of the study, including any publications/abstracts of the study, to the accredited METC.

11.5 Public disclosure and publication policy

No arrangements concerning public disclosure and publication have been made.

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