BMJ Open

SWedish IoDine in Pregnancy and Development In Children (SWIDDICH): Protocol of an On-going Randomized Controlled Trial
The Role of Iodine during Pregnancy for Children's Brain Function

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2017-019945
Article Type:	Protocol
Date Submitted by the Author:	17-Oct-2017
Complete List of Authors:	Manousou, Sofia; Department of Medicine; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Johansson, Birgitta; Institute of Neuroscience and Physiology, Sahlgrenska Academy, Chmielewska, Anna; Department of Clinical Sciences, Pediatrics, Umeå University; Department of Pediatrics, Medical University of Warsaw Eriksson, Janna; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Gutefeldt, Kerstin; Department of Endocrinology, University Hospital of Linköping Tornhage, Carl-Johan; Department of Pediatrics, Skaraborg's Hospital; Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg Eggertsen, Robert; Mölnlycke Health Care Center; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Malmgren, Helge; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Hulthen, Lena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; 10Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg Domellöf, Magnus; Umea University, Department of Clinical Sciences Nystrom Filipsson, Helena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Department of Endocrinology, Sahlgrenska University Hospital
Keywords:	IODINE, CHILD DEVELOPMENT, COGNITION, Thyroid disease < DIABETES & ENDOCRINOLOGY, PUBLIC HEALTH

SWedish **I**o**D**ine in Pregnancy and **D**evelopment **I**n **Ch**ildren (SWIDDICH): Protocol of an On-going Randomized Controlled Trial

The Role of Iodine during Pregnancy for Children's Brain Function

Sofia Manousou[1,2], Birgitta Johansson[3], Anna Chmielewska[4,5], Janna Eriksson[1], Kerstin Gutefeldt[6], Carl-Johan Törnhage[7,8], Robert Eggertsen[1,9], Helge Malmgren[1,] Lena Hulthén[1,10], Magnus Domellöf[4] and Helena Filipsson Nyström[1,11]

[1] Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[2]Department of Medicine, Kungälv's Hospital, Kungälv, Sweden
[3]Institute of Neuroscience and Physiology, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[4] Department of Clinical Sciences, Pediatrics, Umeå University, Umeå, Sweden
[5] Department of Pediatrics, Medical University of Warsaw, Warsaw, Poland
[6] Department of Endocrinology, University Hospital of Linköping, Linköping, Sweden
[7] Department of Pediatrics, Skaraborg's Hospital, Skövde, Sweden
[8] Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
[9] Mölnlycke Health Care Center, Mölnlycke, Sweden,
[10]Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[11]Department of Endocrinology, Sahlgrenska University Hospital, Göteborg, Sweden

Corresponding author:
Sofia Manousou
Department of Internal Medicine
Kungälv's Hospital
Lasarettsgatan
SE-442 34 Kungälv
Sweden

Tel: +46 303 98000
Fax: +46 303 15438
Email: sofia.manousou@vgregion.se

Protocol Version: 1.0, 01 September 2017.

Conflicts of Interest: none

Abstract

Introduction

Iodine is essential for normal brain development. Moderate and severe fetal iodine deficiency result in substantial to serious developmental delay in children. Mild iodine deficiency in pregnancy is associated with neurodevelopmental deficits in the offspring, but evidence from randomized trials is lacking. The aim of the SWIDDICH study is to determine the effect of daily supplementation with 150 µg iodine during pregnancy on the offspring's neuropsychological development up to 14 years of age.

Methods and Analysis

Thyroid healthy pregnant women (n=1275: age range 18 to 40 years) at ≤12 weeks gestation) are randomly assigned to receive multivitamin supplements containing 150 µg iodine or non-iodine containing multivitamin daily throughout pregnancy. As a primary outcome, intelligence quotient (IQ) will be measured in the offspring at 7 years (Wechsler Intelligence Scale for Children, WISC-V). As secondary outcomes, IQ will be measured at 3.5 and 14 years, psychomotor development at 18 months and 7 years, and behavior at 3.5, 7 and 14 years. Iodine status (urinary iodine concentration) will be measured during pregnancy and in the offspring at 3.5, 7 and 14 years. Thyroid function (thyroid hormones, thyroglobulin), and deiodinase type 2 polymorphisms will be measured during pregnancy and in the offspring at 7 and 14 years. Structural magnetic resonance imaging (MRI) or other relevant structural or functional brain imaging procedures will be performed in a subgroup of children at 7 and 14 years. Background and socioeconomic information will be collected at all follow-up times.

Ethics and Dissemination

The Bioethics Committee of Gothenburg approved the study protocol (Approval Number: 1089-16 and 431-12). Study results will be submitted to peer-reviewed journals in the fields of endocrinology, pediatrics and nutrition, and presented at relevant conferences.

Trial Registration Number: ClinicalTrials.gov identifier: NCT02378246

Keywords: Iodine; Pregnancy; Child Development; Cognition; Thyroid Hormones

Strengths and Limitations of this study

- **Large interventional controlled trial** on iodine supplementation during pregnancy, powered to detect a difference of 3 IQ points in children.
- **Long observational follow-up** of the children, up to 14 years, with complex assessment of neurocognitive development.
- **Future implementation** of the study is feasible, as the intervention tablet exists on the market.
- **Lack of pure iodine** and pure placebo tablets implies careful interpretation of results.
- **Dropout rate** may be high.

BACKGROUND

Iodine Deficiency as an International Issue

Iodine is essential for the production of thyroid hormones and important for growth and brain development during fetal and early postnatal life [1]; a knowledge obtained after a long history of iodine deficiency (ID) associated disorders. For centuries, goiter with hypothyroidism, mental retardation and cretinism have been an entity. During the 1920s in the United States, Marine and Kimball performed the classic experiment of treating schoolgirls with iodine, leading to a dramatic reduction in the prevalence of goiter. Iodine prophylaxis was established in United States in 1921. After some debate, iodine prophylaxis was introduced in Switzerland in 1922, and then worldwide over the following decades. The combat against severe and moderate ID has been successful in reducing the number of children with ID-caused mental retardation. However, mild ID is widely apparent, especially during pregnancy [2], when dietary iodine demand increases from 150 to 250 µg/day [3].

Iodine Status in Sweden as the Country for this Study

Before iodination of table salt in 1936, ID was common in Sweden [4]. Current iodine intake is sufficient in the general population [5, 6] and was judged adequate during pregnancy during the 1990s [7, 8]; there is no recommendation on iodine supplementation during pregnancy. However, since the 1990s, the situation may have changed because dairy product consumption in adults is lower; milk iodine levels are lower than before [9, 10]; a reduction in salt intake is recommended for reducing the risk of hypertension; new salt forms (flake salt, gourmet salt) without iodine are popular; there is a reluctance to consume "food additives"; awareness of ID among the younger population is generally low; and, the main proportion of total salt intake (≈80%), i.e. from ready-made foods and dishes, does not provide iodine. Unless iodine is added to all salts used, the risk of decreased iodine intake is apparent, and arouses concerns especially for pregnant women. Retrospective, local data on pregnancy highlights this assumption is realistic [11].

Iodine Deficiency during Pregnancy: Effects on Child's Development

Severe and moderate ID leads to lower serum thyroid hormone levels and thereby to lower availability of thyroid hormones in the brain. During fetal life and early years, the growing brain is vulnerable [12, 13] and severe ID results in mental retardation in the newborn, unless thyroid hormone is replaced [14]. In addition, an increased incidence of attention deficit hyperactivity disorders (ADHD) has been associated with mild-to-moderate ID [15].

In mild ID, thyroid hormone levels are maintained, whereas, thyroglobulin (TG) levels are increased as a biomarker of goiter. The brain's use of thyroid hormones depends on the local conversion of inactive hormone thyroxine (T4) to active hormone triiodothyronine (T3), a process mediated by deiodinase type 2 (D2) [16]. D2 is found in the hippocampi and the cerebral cortex and its activity is increased by ID, to maintain sufficient T3 levels [16, 17]. In the presence of normal thyroid hormone in blood, it is unclear how mild ID affects brain development. One theory is that this depends on deiodinases, which can change thyroid hormone signalling locally in different tissues, without affecting serum hormone concentration [16, 18].

Mild ID during pregnancy might have an impact on brain development, despite maintained normal thyroid hormone levels [19-22]. In the United Kingdom, a longitudinal study [19] found 8-year-old children have an increased risk of being in the lowest quartile of verbal IQ if their mothers had urinary iodine concentration (UIC) indicating mild ID in early pregnancy than children of mothers with normal iodine nutrition. In a similar association study from Australia [20], mild ID is linked with lower cognitive performance in 9-year old children. Results from an observational pilot-study from Italy [21] indicate mild-to-moderate ID during fetal life affects cognitive development, especially verbal abilities, even in absence of maternal thyroid insufficiency. In Norway, a large observational study [22] found that maternal iodine intake below the estimated average requirement during pregnancy was associated with reduced fine motor skills and verbal abilities and with more behavior problems at the age of 3 years.

As the randomized controlled trial (RCT) [23] evaluating 150 µg iodine/placebo in pregnant women in an iodine sufficient country was small (n=86) and lacked cognitive assessment in children, there were many expectations about the ongoing MITCH study [24], where 839 women are randomized to 200 µg iodine/placebo. Dr. Zimmermann presented the preliminary data from the MITCH study, which is not yet published, at the the 15th International Thyroid Congress of Iodine Global Network (IGN) at Orlando, Florida on October 18th, 2015. The data revealed that both groups were iodine sufficient according to UIC and that there was no difference in cognitive outcome in 1-2-year-old children. The question if mild ID during pregnancy affects fetal brain development remains unanswered. To prevent subnormal fetal brain development, many international authorities recommend 150 µg extra iodine/day during pregnancy, despite the lack of studies on causality [25, 26].

Knowledge Gaps and Background to the SWIDDICH Study

There is a substantial gap in knowledge about mild ID during pregnancy and its potential negative consequences on neuropsychological development. Therefore, there is a need for a placebo-controlled trial that compares neuropsychological outcome in children exposed to mild ID during fetal life and children with normal iodine nutrition during pregnancy.

In 2012-2015, a pilot randomized placebo-controlled trial involving 200 pregnant women on a daily supplementation with either a multivitamin containing 150 µg iodine/day or a multivitamin without iodine (placebo) was conducted by our group. This study (ClinicalTrials.gov identifier: NCT02378246) aimed to evaluate the effects of iodine supplementation on UIC and thyroid function. As the MITCH study failed to

answer the question if mild ID during pregnancy affects fetal brain development, it is evident to us that our trial needs to be expanded to include a sufficient number of pregnant women to enable a sufficiently powered child follow-up regarding neuropsychological development. This is the **SW**edish **I**o**D**ine in Pregnancy and **D**evelopment **I**n **Ch**ildren (SWIDDICH) study.

Objectives

The primary aim is to assess whether cognition (especially verbal competence) in children whose mothers received 150 µg iodine daily during pregnancy is higher than children whose mothers received placebo (a multivitamin without iodine) and probably remained in mild ID. The purpose is to determine whether all pregnant women who live in a country where the general population is iodine sufficient, but live in conditions that can result in mild ID during pregnancy, should be recommended extra iodine during pregnancy.

METHODS

Design of the SWIDDICH Study

This is a randomized placebo-controlled study in which children are followed-up as an observational cohort, separated into two groups by the fetal iodine exposure.

Setting and Participants

Pregnant women will be recruited from more than ten maternal healthcare centers in Sweden. At the first scheduled pregnancy visit, information about the study will be provided and written informed consent collected. All procedures during pregnancy will be combined with routine pregnancy visits.

Inclusion

The following inclusion criteria will apply: woman aged 18 to 40 years, pregnant at ≤12 weeks, willing to refrain from iodine supplementation and take a multivitamin supplement instead, without current thyroid disease, not in another pregnancy or lactating less than 6 months before inclusion, and non-vegan.

Randomization, Allocation, Concealment and Blinding

Randomization numbers, with an allocation ratio is 1:1, are prepared centrally and sent to each participating center. Consecutive numbers are used and the information regarding the study group allocation of each number stored securely at the premises of the University of Gothenburg, Sweden. Mothers are provided with a random container of pills, by either drawing a lot or blindly drawing a container. All containers are identical, with tasteless pills of the same size for both groups. Recruiting staff, study participants and those involved in laboratory work and developmental assessment are blinded to the group allocation. The code will only be broken by the central study team for data analyses before publications, but will still be blinded to all groups that work in the follow-up. The code has been broken for the 200 women of the pilot study, but all (ie. study participants, psychologists and lab engineers) except the central study team are still blinded.

Intervention

Women in the experimental group receive a daily multivitamin supplement containing 150 µg iodine and those in the control group receive a daily multivitamin supplement containing no iodine (the contents of

the two supplements are presented in Table 1). The intervention lasts throughout pregnancy to the day of delivery, except for the first 200 women who are given tablets until they leave a urine and breast milk sample, within the first 5 days after delivery.

The reason for choosing iodine-containing multivitamins instead of pure iodine tablets as the intervention is to ensure future implementation of the study is feasible. There are currently no pure iodine tablets on the market. In the planning state of the study, discussions about pure iodine tablets with pharmaceutical companies producing multivitamins revealed no interest in launching such a product in the future. Therefore, iodine in multivitamins will be the only available supplement source in an implementation situation.

Other components in the multivitamin products, besides iodine, can intervene with outcomes. It is proposed that vitamin B12 [27, 28] and iron [29] can have positive effects on the brain, and iron and selenium influence thyroid hormone levels [30, 31]. Iron is found in thyroperoxidase (TPO) enzyme that couples iodine to thyroglobulin and selenium is found in deiodinases, such as D2 that converts T4 to T3. Sweden is a selenium deficient country [32], but it is unclear whether selenium deficiency affects cognitive outcome in humans [33]. B12 is higher in iodine-containing multivitamin where iron and selenium are also included. However, B12 content in both placebo and intervention tablets is, at least, equal to the recommended daily intake for B12, thus, B12 deficiency is not anticipated in any of the groups. In addition, the iron contents are low. Many pregnant Swedish women take a separate 100 mg iron supplement, which makes the 12 mg iron in the intervention tablet negligible. Iron and selenium will be measured in a subpopulation to evaluate possible group differences.

Compliance

Participants are asked to bring the container with the remaining pills to the visit in the third trimester. The container is weighted and the percentage of intended doses used is calculated.

Outcomes

Outcomes in Mothers

Outcomes in mothers will be assessed in the first, second and third trimester of pregnancy. UIC and thyroid hormones will be measured in all three trimesters, and TPO-antibodies and TG in the first and third trimester. Breast milk iodine concentration and UIC will be measured in a sub sample of mothers (the first 200 included), once during the first five days after delivery.

Primary Outcome in Children

Cognition measured by intelligence quotient (total IQ) with focus on the verbal compound (verbal IQ) at 7 years is the primary outcome (Wechsler Intelligence Scale for Children, WISC-V [34]).

Secondary Outcomes in Children

Cognition measured by IQ at 3.5 years (Wechsler Preschool and Primary Scale of Intelligence, WPPSI-IV [35]) and at 14 years (Wechsler Intelligence Scale for Children, WISC-V [34] or an equivalent adequate version at the time) are secondary outcomes, together with outcomes related to psychomotor development, behavior and attention deficit hyperactivity disorder (ADHD). Psychomotor assessment will be done by the parents at 18 months (The Ages and Stages Questionnaire, ASQ-3) [36], and by a physiotherapist at 7 years (Movement Assessment Battery for Children, Movement ABC test) [37]. Behavior will be assessed

through parental questionnaires, the Child Behavior Checklist (CBCL); first at 3.5 years (CBCL 1-5) [38], then at 7 and 14 years (CBCL 6-18) [39]. At 7 and 14 years, the Nordic questionnaire 5-15 [40] will be used to assess ADHD-related symptoms.

UIC in a subgroup of newborn (from the first 200 included mothers) will be measured once during the first 5 days of life. In a subgroup of children (n=200), magnetic resonance imaging (MRI) of the brain (with a 3T Philips MR scanner) is planned at 7 and 14 years to evaluate structural brain changes. Automatic segmentation of the whole brain will be done with Freesurfer [41] and Maper, multi-atlas propagation with enhanced registration [42]. Medio-temporal lobe (MTL) structures will be analyzed through manual segmentation using custom software developed in previous projects [43, 44]. Subregional analyses directed at regions of neurogenesis will be included. Intracranial volume measured manually will enable reliable normalization of MTL volumes. Other structural and/or functional brain imaging methods may supplement, or even replace, the described protocol, depending on the state of knowledge at the time of study.

Possible Confounding Variables and Background Information

In children, UIC will be measured from the 3.5-year visit and forward and dry blood spots will be collected for thyroid hormones, TG and deiodinases at 7 and 14 years. Background and confounding variables will be assessed in all three trimesters of pregnancy, at 18 months, 3.5, 7 and 14 years.

Timeframe for the Study Actions

Recruitment to the SWIDDICH study began in 2017 and is planned to be completed in 2019. The follow-up of children is offered to the families who participated in the pilot study (2012-2015), before the decision on study extension was made. The time points for all study actions are presented in Table 2.

CONSIDERATIONS

Power Calculation and Statistical Considerations

The sample size needed, excluding dropouts, is calculated to 788 children (394 in each group) for an effect size of 3 IQ points with SD 15 and power 0.80. Currently, there are no similar randomized studies for power calculation. The smallest significant effect of 3 IQ points is in accordance with an observational study [19], where children of mothers with UIC< 150 µg/L during pregnancy had a 3-point lower IQ at school-age than children of mothers with normal UIC during pregnancy. This expected effect from iodine supplementation in mild iodine deficiency is also suggested by Troendle [45], where statistical considerations are discussed for the possibility that the needed placebo-controlled study is conducted. Assuming a drop out frequency of 22% during pregnancy (which is in accordance with preliminary data from our pilot study with 200 pregnant women) and 20% during the children follow up, 1263 pregnant women need to be recruited to the study. This sample size is in general agreement with Troendle [45], thus, the decision was made to try and recruit 1275 pregnant women. The dropout frequency for the children follow-up could be lower than estimated as there are two occasions for dropout and mothers who remain in the study after the first follow-up can be assumed willing to continue the study. The power calculation assumes the use of an unpaired t-test between groups; however, more advanced analyses could decrease variance, thus requiring a lower sample size.

The sample size will be reassessed by calculating the dropout frequency when 750 women are included and when half of the children from the first 200 included women have been invited to the 3.5-year

neuropsychological evaluation. Sample size reassessment will be conducted without unblinding the study groups.

A 100% compliance to the study medication is assumed, as the results will be based on an intention to treat (ITT) analysis. Compliance is monitored to enable a per protocol analysis (only the compliant participants included) to be added. However, the ITT approach reflects the real life clinical situation, in which a certain number of patients are not compliant with the recommended treatment, and this will be the foundation for future recommendations on iodine supplementation to all pregnant women.

A separate power calculation for the MRI investigation has been done. This assumes the described protocol will be followed, and a previous study of 11-year old children has been used for guidance [46]. To detect a 5% difference with power 0.80, 60 children are needed in each group. As the variation in the hippocampal volumes in 7-year-old children could be slightly larger than in the previous study [46] and since the dropout from the MRI at 14 years needs to be reckoned with, 100 children will be included in each group.

The Student's t-test or the Mann-Whitney test will be used to compare the main outcome between the experimental and control groups. Repeated-measures analysis of variance will be used for IQ scores. If needed, possible confounders, such as socioeconomic factors, background information, thyroid hormones, TG, deiodinase polymorphisms and UIC, will be adjusted for during data analysis. In addition, repeated measurements in a mixed model (where groups are compared repeatedly at 3.5, 7 and 14 years) and within-group analyses are planned. The models will also consider the dropout frequency and recruitment from different maternal health care centers. A multivariate analysis with total grey, total brain volume, intracranial volume, MTL volumes and possibly other regional brain volumes as independent variables will be done. The data analyses will be undertaken by an experienced statistician.

MRI considerations – Where Are Changes from ID Located?

T3 receptors are distributed among all brain areas with high levels in the hippocampi and the cerebellar cortex. Rodent data indicate that T3 receptors are involved in the regulation of hippocampal structure and function [47]. In the human cerebral cortex, thyroid receptors are already present in week 9 and concentrations increase up to 18 weeks of gestation [48]. Considerable amounts of D2 are also found in the cerebral cortex [49]. In the first half of pregnancy, the fetus is dependent on the mother's supply of thyroid hormones. In mild ID, the mother maintains serum thyroid hormone levels through unknown compensatory mechanisms. In the second half of a mild ID pregnancy, when the fetus partly relies on its own thyroid hormone production, the fetus will be hypothyroid as it has not developed compensatory mechanisms and there is a lack of sufficient iodine levels transferred by the mother [49].

The description of neuropathology caused by ID is limited to few observations from adult cretins, ranging from severe cortical atrophy to almost normal appearance. In areas with endemic goiter, fetuses aborted in the second half of the pregnancy have a less differentiated cerebral cortex [49]. In rats, transient periods of thyroid hormone insufficiency during periods of cortical development affect cortical and hippocampal cytoarchitecture [49].

Human data from maternal hypothyroidism support an effect on the brain, specifically on the hippocampus [50]. These data are in line with the recent publication by Korevaar *et al* [51], who conclude the relationship of IQ with FT4 (in peripheral blood) exhibits a u-shaped configuration with lower IQ levels in both ends of the normal range. FT4 in this study is also associated with total grey matter volume.

Considerations on the Neuropsychological Evaluation

Neuropsychological development can be divided into three domains: psychomotor, cognitive (IQ) and socio-emotional development (Figure 1). There are five landmark studies in the iodine field evaluating neuropsychological development in the off-spring that use neuropsychological tests: the ALSPAC (United Kingdom) [19], INMA (Spain) [52], Generation R (Netherlands) [51], MITCH (India and Thailand) [24] and the study by Hynes *et al* (Australia) [20]. Verbal cognitive function appears to be the most susceptible subdomain for ID. In SWIDDICH, verbal cognitive function together with total IQ were chosen (as the latter is the best understood and requested) as primary outcome measurements. As cognitive testing is less valid in younger ages, verbal IQ at 7 years was chosen as the primary evaluation time point and all three domains of neuropsychological development will be evaluated at several follow-up times.

Implications for Society and the Individual

Impaired child development increases economic burdens for society. Lowered IQ is associated with worse economic outcomes and lower lifetime earnings. Small decrements in IQ around the mean are linked to lower incomes [53, 54]. IQ may be the easiest factor to quantify, but may not be the factor with the most serious consequence for a "good life". Environmental factors, including ID, that place the nervous system at risk may affect executive functions, such as planning and initiating ideas and result in attention problems, impulsive behavior, and inability to handle stress and disappointments, thus, impeding success in school and in life and possibly leading to antisocial behavior [55].

If the average IQ of a population drops, the IQ distribution shifts and the number of individuals with low IQ (e.g below 75 or 85, classified as intellectually disabled) increases. In turn, this will also decrease the number of gifted and exceptionally gifted people with high IQs (e.g above 130), who may have major positive impacts on the immediate future for a company or a country.

Based on the dollar value in 1987 in the USA, the cost in terms of reduced income for a one point IQ reduction has been calculated to nearly 20.7 billion USD per year [56]. A 3-point decline in IQ also impacts social costs in the United States [55] and increases the risk of: poverty by 20% during the first three years; low birth weight by 12%; being a recipient of welfare by 18%; and, high school dropout by 28%. Even though a decline of a few IQ points may be small for the individual, the societal effects are considerable. As a small general risk reduction entails a large social benefit, iodine supplementation could be a cost-effective action if the main hypothesis of this study holds true.

Considerations on Possible Adverse Effects of Iodine or Placebo

Iodine supplementation may increase the frequency of post-partum thyroiditis (PPT), as iodine affects autoimmunity [57]. 10-15% of women already have post-partum thyroiditis and this number may increase slightly with iodine supplementation. As PPT is not a dangerous condition and most cases resolve spontaneously, we consider the reduced risk for subnormal brain development in a child motivates accepting the risk for PPT. In Denmark, postpartum thyroiditis was evaluated in a placebo-controlled trial in mild-moderate iodine deficiency, and treatment did not increase or worsen PPT [57].

Excess iodine in the mother may block thyroid function in the fetus leading to hypothyroidism and goiter. However, the iodine multivitamin tablets proposed for this study are already readily available and contain 150 µg iodine, which is lower than the 250 µg, i.e. the dietary iodine demand during pregnancy.

The placebo group is at risk of iodine deficiency during pregnancy. However, as there are no current recommendations in Sweden for iodine supplementation in pregnancy, this group follows normal management.

CONCLUSION

The aim of this paper is to describe the study protocol for the SWIDDICH research project and the considerations that led to its design. The study attempts to further understand the consequences of mild ID during pregnancy and to test whether treatment of the mothers improves outcome in the children. As the study is the largest of its kind, it offers the potential for influencing future recommendations on iodine supplementation to pregnant women living in conditions of mild ID.

ABBREVIATIONS

IQ: intelligence quotient; ADHD: attention deficit hyperactivity disorders; TG: thyroglobulin; T4: thyroxine; T3: triiodothyronine; D2: deiodinase type 2; UIC: urinary iodine concentration; RCT: randomized controlled trial; TPO: thyroperoxidase; MRI: magnetic resonance imaging; MTL: medio-temporal lobe.

Data Sharing

Data from this trial may be shared for individual data analysis in the future. A detailed plan for data sharing will be developed during the later phase of the project.

Authors' Contributions

Sofia Manousou, Birgitta Johansson, Anna Chmielewska, Janna Eriksson, Kerstin Gutefeldt, Carl-Johan Törnhage, Robert Eggertsen, Helge Malmgren, Lena Hulthén, Magnus Domellöf and Helena Filipsson Nyström have contributed to the design of the SWIDDICH study. HFN wrote the first version of the manuscript and SM was responsible for pushing the work forward together with the other coworkers. All co-authors critically reviewed and approved the final version of the manuscript. SM is the guarantor.

Funding Statement

This work was supported by the ALF agreement (grant number ALFGBG-58777), Regional FOU (grant number VGFOUREG-664301**),** Lilla barnets fond (grant number 20160917), Svenska Läkarsälskapet (grant number SLS-688891), and Lars Hiertas minne foundation (grant number FO2016-0016). Multivitamins for the first 200 women were provided by courtesy of Recip medical company, Solna, Sweden.

Acknowledgements: Elisabeth Gramatkovski for her invaluable help as coordinator of the study.

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Table 2. Summary of SWIDDICH study actions.

Figure 1. Neuropsychological domains. Verbal cognitive function, as a subdomain of cognition, appears the most susceptible subdomain for iodine deficiency and is the primary outcome of the SWIDDICH study at the age of 7 years.

References

[1] M.B. Zimmermann, P.L. Jooste, C.S. Pandav, Iodine-deficiency disorders, Lancet 372(9645) (2008) 1251-62.
[2] M.B. Zimmermann, M. Gizak, K. Abbott, M. Andersson, J.H. Lazarus, Iodine deficiency in pregnant women in Europe, The lancet. Diabetes & endocrinology 3(9) (2015) 672-4.
[3] W.H.O. United Nations Children's Fund & International Council for the Control of Iodine Deficiency Disorders, Assessment of iodine deficiency disorders and monitoring their elimination. A guide for programme managers, 3:e edition, Geneva, 2007.
[4] H.F. Nystrom, S. Jansson, G. Berg, Incidence Rate and Clinical features of Hyperthyroidism in a long-term iodine sufficient area of Sweden (Gothenburg) 2003-2005, Clin Endocrinol (Oxf) (2013).
[5] M. Andersson, G. Berg, R. Eggertsen, H. Filipsson, E. Gramatkovski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Adequate iodine nutrition in Sweden: a cross-sectional national study of urinary iodine concentration in school-age children, Eur. J. Clin. Nutr. 63(7) (2009) 828-34.
[6] H. Filipsson Nystrom, M. Andersson, G. Berg, R. Eggertsen, E. Gramatkowski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Thyroid volume in Swedish school children: a national, stratified, population-based survey, Eur. J. Clin. Nutr. 64(11) (2010) 1289-95.
[7] B. Elnagar, A. Eltom, L. Wide, M. Gebre-Medhin, F.A. Karlsson, Iodine status, thyroid function and pregnancy: study of Swedish and Sudanese women, Eur. J. Clin. Nutr. 52(5) (1998) 351-5.
[8] A. Eltom, B. Elnagar, M. Elbagir, M. Gebre-Medhin, Thyroglobulin in serum as an indicator of iodine status during pregnancy, Scand J Clin Lab Invest. 60(1) (2000) 1-7.
[9] H. Lindmark Månsson, the Swedish Milk composition Svensk Mjölk (Swedish milk) 2010.
[10] H. Lindmark Månsson, Den svenska mejerimjölkens sammansättning 2009, Svensk Mjök (Swedish Milk), 2012.
[11] M. Granfors, M. Andersson, S. Stinca, H. Akerud, A. Skalkidou, I. Sundstrom Poromaa, A.K. Wikstrom, H. Filipsson Nystrom, Iodine deficiency in a study population of pregnant women in Sweden, Acta Obstet. Gynecol. Scand. (2015).
[12] M.B. Zimmermann, The adverse effects of mild-to-moderate iodine deficiency during pregnancy and childhood: a review, Thyroid 17(9) (2007) 829-35.
[13] P.N. Taylor, O.E. Okosieme, C.M. Dayan, J.H. Lazarus, Therapy of endocrine disease: Impact of iodine supplementation in mild-to-moderate iodine deficiency: systematic review and meta-analysis, Eur. J. Endocrinol. 170(1) (2014) R1-R15.
[14] J.B. Stanbury, A.M. Ermans, B.S. Hetzel, E.A. Pretell, A. Querido, Endemic goitre and cretinism: public health significance and prevention, WHO Chron. 28(5) (1974) 220-8.
[15] F. Vermiglio, V.P. Lo Presti, M. Moleti, M. Sidoti, G. Tortorella, G. Scaffidi, M.G. Castagna, F. Mattina, M.A. Violi, A. Crisa, A. Artemisia, F. Trimarchi, Attention deficit and hyperactivity disorders in the offspring of mothers exposed to mild-moderate iodine deficiency: a possible novel iodine deficiency disorder in developed countries, J. Clin. Endocrinol. Metab. 89(12) (2004) 6054-60.
[16] R. Peeters, C. Fekete, C. Goncalves, G. Legradi, H.M. Tu, J.W. Harney, A.C. Bianco, R.M. Lechan, P.R. Larsen, Regional physiological adaptation of the central nervous system deiodinases to iodine deficiency, Am. J. Physiol. Endocrinol. Metab. 281(1) (2001) E54-61.

[17] N. Markova, A. Chernopiatko, C.A. Schroeter, D. Malin, A. Kubatiev, S. Bachurin, J. Costa-Nunes, H.M. Steinbusch, T. Strekalova, Hippocampal gene expression of deiodinases 2 and 3 and effects of 3,5-diiodo-L-thyronine T2 in mouse depression paradigms, BioMed research international 2013 (2013) 565218.
[18] F. Courtin, H. Zrouri, A. Lamirand, W.W. Li, G. Mercier, M. Schumacher, C.L. Goascogne, M. Pierre, Thyroid hormone deiodinases in the central and peripheral nervous system, Thyroid 15(8) (2005) 931-42.
[19] S.C. Bath, C.D. Steer, J. Golding, P. Emmett, M.P. Rayman, Effect of inadequate iodine status in UK pregnant women on cognitive outcomes in their children: results from the Avon Longitudinal Study of Parents and Children (ALSPAC), Lancet 382(9889) (2013) 331-7.
[20] K.L. Hynes, P. Otahal, I. Hay, J.R. Burgess, Mild iodine deficiency during pregnancy is associated with reduced educational outcomes in the offspring: 9-year follow-up of the gestational iodine cohort, J. Clin. Endocrinol. Metab. 98(5) (2013) 1954-62.
[21] M. Moleti, F. Trimarchi, G. Tortorella, A. Candia Longo, G. Giorgianni, G. Sturniolo, A. Alibrandi, F. Vermiglio, Effects of Maternal Iodine Nutrition and Thyroid Status on Cognitive Development in Offspring: A Pilot Study, Thyroid 26(2) (2016) 296-305.
[22] M.H. Abel, I.H. Caspersen, H.M. Meltzer, M. Haugen, R.E. Brandlistuen, H. Aase, J. Alexander, L.E. Torheim, A.L. Brantsaeter, Suboptimal Maternal Iodine Intake Is Associated with Impaired Child Neurodevelopment at 3 Years of Age in the Norwegian Mother and Child Cohort Study, J. Nutr. 147(7) (2017) 1314-1324.
[23] F. Brucker-Davis, P. Panaia-Ferrari, J. Gal, P. Fenichel, S. Hieronimus, Iodine Supplementation throughout Pregnancy Does Not Prevent the Drop in FT4 in the Second and Third Trimesters in Women with Normal Initial Thyroid Function, European thyroid journal 2(3) (2013) 187-94.
[24] A. Melse-Boonstra, S. Gowachirapant, N. Jaiswal, P. Winichagoon, K. Srinivasan, M.B. Zimmermann, Iodine supplementation in pregnancy and its effect on child cognition, J. Trace Elem. Med. Biol. 26(2-3) (2012) 134-6.
[25] A. Stagnaro-Green, M. Abalovich, E. Alexander, F. Azizi, J. Mestman, R. Negro, A. Nixon, E.N. Pearce, O.P. Soldin, S. Sullivan, W. Wiersinga, P. American Thyroid Association Taskforce on Thyroid Disease During, Postpartum, Guidelines of the American Thyroid Association for the diagnosis and management of thyroid disease during pregnancy and postpartum, Thyroid 21(10) (2011) 1081-125.
[26] J. Lazarus, R.S. Brown, C. Daumerie, A. Hubalewska-Dydejczyk, R. Negro, B. Vaidya, 2014 European thyroid association guidelines for the management of subclinical hypothyroidism in pregnancy and in children, European thyroid journal 3(2) (2014) 76-94.
[27] V. Bhate, S. Deshpande, D. Bhat, N. Joshi, R. Ladkat, S. Watve, C. Fall, C.A. de Jager, H. Refsum, C. Yajnik, Vitamin B12 status of pregnant Indian women and cognitive function in their 9-year-old children, Food Nutr Bull 29(4) (2008) 249-54.
[28] M.M. Black, Effects of vitamin B12 and folate deficiency on brain development in children, Food Nutr Bull 29(2 Suppl) (2008) S126-31.
[29] E.L. Prado, K.G. Dewey, Nutrition and brain development in early life, Nutr. Rev. 72(4) (2014) 267-84.
[30] M. Ventura, M. Melo, F. Carrilho, Selenium and Thyroid Disease: From Pathophysiology to Treatment, Int. J. Endocrinol. 2017 (2017) 1297658.
[31] S. Hu, M.P. Rayman, Multiple Nutritional Factors and the Risk of Hashimoto's Thyroiditis, Thyroid 27(5) (2017) 597-610.
[32] O. Selinus, Medical geology: Iodine - a classical element (jod ett klassiskt element), Medical geololgy (Medicinsk Geologi), Authors and Studentlitteratur2010, pp. 387-400.
[33] E. Wasantwisut, Nutrition and development: other micronutrients' effect on growth and cognition, Southeast Asian J. Trop. Med. Public Health 28 Suppl 2 (1997) 78-82.
[34] D. Wechsler, Wechsler Intelligence Scale for Children – Fifth Edition (WISC-V), Swedish version, Stockholm, Sweden, 2016.
[35] D. Wechsler, Wechsler Preschool and Primary Scale of Intelligence – Fourth Edition (WPPSI-IV) (Swedish version), Stockholm, Sweden, 2014.
[36] T.E. Squires J, Bricker D, Potter L, ASQ-3 User's Guide. 3rd ed. , Baltimore, MD, US, 2009.

[37] S.E. Henderson, Sugden, D. A., & Barnett, A. L. , Movement assessment battery for children [examiner's manual] (2nd ed.). , London, UK, 2007.
[38] R.L. Achenbach TM, Manual for the ASEBA Preschool Forms & Profiles. , University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2000.
[39] T.M. Achenbach, Rescorla, L.A. , Manual for the ASEBA School-Age Forms & Profiles, University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2001.
[40] B. Kadesjo, L.O. Janols, M. Korkman, K. Mickelsson, G. Strand, A. Trillingsgaard, C. Gillberg, The FTF (Five to Fifteen): the development of a parent questionnaire for the assessment of ADHD and comorbid conditions, Eur. Child Adolesc. Psychiatry 13 Suppl 3 (2004) 3-13.
[41] B. Fischl, D.H. Salat, E. Busa, M. Albert, M. Dieterich, C. Haselgrove, A. van der Kouwe, R. Killiany, D. Kennedy, S. Klaveness, A. Montillo, N. Makris, B. Rosen, A.M. Dale, Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain, Neuron 33(3) (2002) 341-55.
[42] R.A. Heckemann, S. Keihaninejad, P. Aljabar, D. Rueckert, J.V. Hajnal, A. Hammers, Improving intersubject image registration using tissue-class information benefits robustness and accuracy of multi-atlas based anatomical segmentation, Neuroimage 51(1) (2010) 221-7.
[43] C. Eckerstrom, E. Olsson, M. Borga, S. Ekholm, S. Ribbelin, S. Rolstad, G. Starck, A. Edman, A. Wallin, H. Malmgren, Small baseline volume of left hippocampus is associated with subsequent conversion of MCI into dementia: the Goteborg MCI study, J. Neurol. Sci. 272(1-2) (2008) 48-59.
[44] E. Olsson, C. Eckerstrom, G. Berg, M. Borga, S. Ekholm, G. Johannsson, S. Ribbelin, G. Starck, A. Wysocka, E. Lofdahl, H. Malmgren, Hippocampal volumes in patients exposed to low-dose radiation to the basal brain. A case-control study in long-term survivors from cancer in the head and neck region, Radiat. Oncol. 7 (2012) 202.
[45] J.F. Troendle, Statistical design considerations applicable to clinical trials of iodine supplementation in pregnant women who may be mildly iodine deficient, Am. J. Clin. Nutr. 104 Suppl 3 (2016) 924S-7S.
[46] L. Bunketorp Kall, H. Malmgren, E. Olsson, T. Linden, M. Nilsson, Effects of a Curricular Physical Activity Intervention on Children's School Performance, Wellness, and Brain Development, J. Sch. Health 85(10) (2015) 704-13.
[47] A. Guadano-Ferraz, R. Benavides-Piccione, C. Venero, C. Lancha, B. Vennstrom, C. Sandi, J. DeFelipe, J. Bernal, Lack of thyroid hormone receptor alpha1 is associated with selective alterations in behavior and hippocampal circuits, Mol. Psychiatry 8(1) (2003) 30-8.
[48] B. Ferreiro, J. Bernal, C.G. Goodyer, C.L. Branchard, Estimation of nuclear thyroid hormone receptor saturation in human fetal brain and lung during early gestation, J. Clin. Endocrinol. Metab. 67(4) (1988) 853-6.
[49] G.M. de Escobar, M.J. Obregon, F.E. del Rey, Iodine deficiency and brain development in the first half of pregnancy, Public Health Nutr. 10(12A) (2007) 1554-70.
[50] K.A. Willoughby, M.P. McAndrews, J.F. Rovet, Effects of maternal hypothyroidism on offspring hippocampus and memory, Thyroid 24(3) (2014) 576-84.
[51] T.I. Korevaar, R. Muetzel, M. Medici, L. Chaker, V.W. Jaddoe, Y.B. de Rijke, E.A. Steegers, T.J. Visser, T. White, H. Tiemeier, R.P. Peeters, Association of maternal thyroid function during early pregnancy with offspring IQ and brain morphology in childhood: a population-based prospective cohort study, The lancet. Diabetes & endocrinology 4(1) (2016) 35-43.
[52] M. Rebagliato, M. Murcia, M. Alvarez-Pedrerol, M. Espada, A. Fernandez-Somoano, N. Lertxundi, E.M. Navarrete-Munoz, J. Forns, A. Aranbarri, S. Llop, J. Julvez, A. Tardon, F. Ballester, Iodine supplementation during pregnancy and infant neuropsychological development. INMA Mother and Child Cohort Study, Am. J. Epidemiol. 177(9) (2013) 944-53.
[53] J. Schwartz, Societal benefits of reducing lead exposure, Environ. Res. 66(1) (1994) 105-24.
[54] D.S. Salkever, Updated estimates of earnings benefits from reduced exposure of children to environmental lead, Environ. Res. 70(1) (1995) 1-6.
[55] T. Muir, M. Zegarac, Societal costs of exposure to toxic substances: economic and health costs of four case studies that are candidates for environmental causation, Environ. Health Perspect. 109 Suppl 6 (2001) 885-903.

[56] J. Schwartz, Low-level lead exposure and children's IQ: a meta-analysis and search for a threshold, Environ. Res. 65(1) (1994) 42-55.
[57] S.B. Nohr, A. Jorgensen, K.M. Pedersen, P. Laurberg, Postpartum thyroid dysfunction in pregnant thyroid peroxidase antibody-positive women living in an area with mild to moderate iodine deficiency: is iodine supplementation safe?, J. Clin. Endocrinol. Metab. 85(9) (2000) 3191-8.

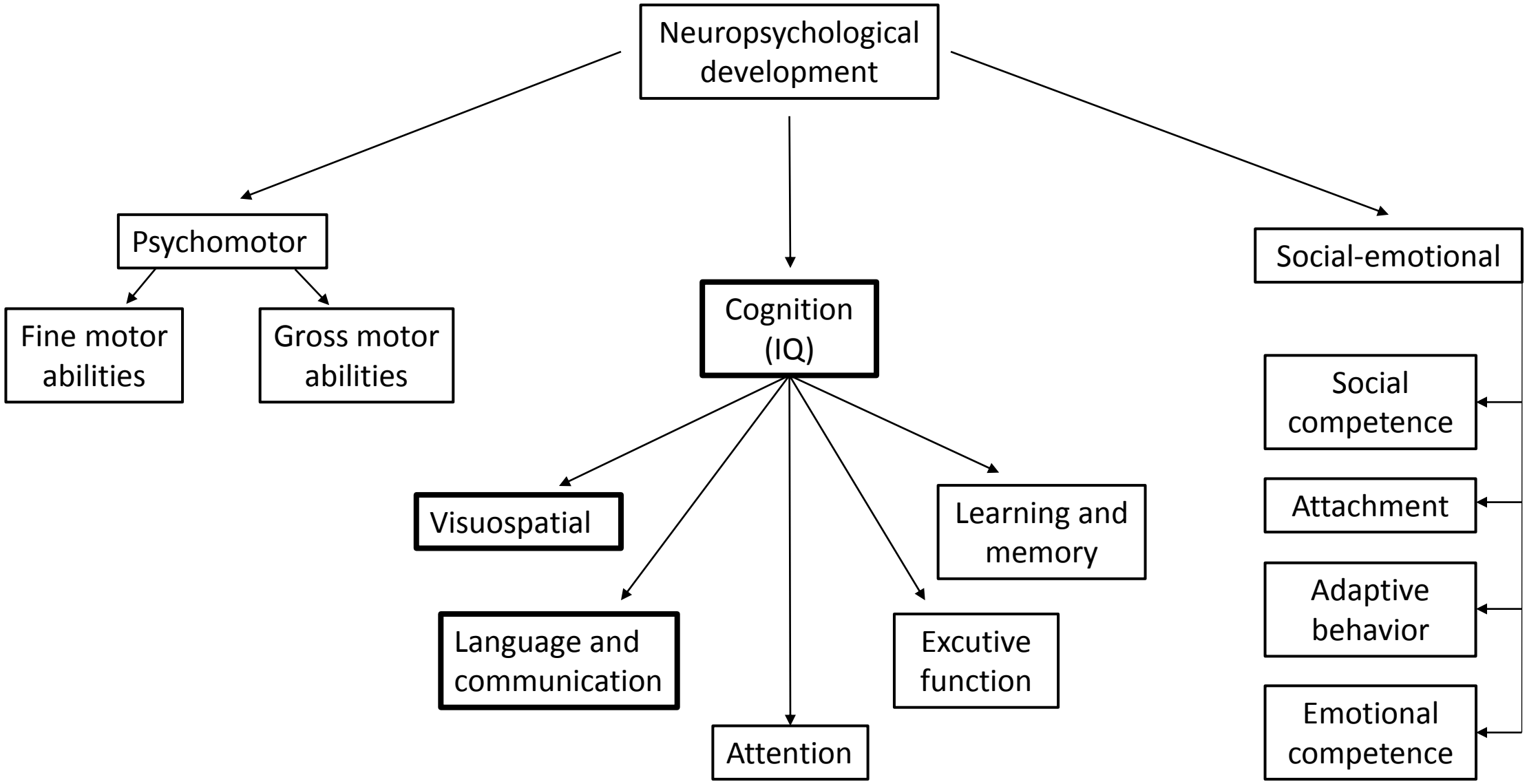

Neuropsychological development
Psychomotor
Fine motor abilities
Gross motor abilities
Cognition (IQ)
Visuospatial
Language and communication
Attention
Excutive function
Learning and memory
Social-emotional
Social competence
Attachment
Adaptive behavior
Emotional competence

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Intervention (iodine 150 microgram) **Commercial name: MITT VAL VEGETARIAN**	**Placebo (no iodine)** **Commercial name: ENOMDAN**
B2 1,4 mg (100%)* B12 15 microgram (600%) Iron 12 mg (86%) Zink 12 mg (120%) Iodine 150 microgram (100%) Selenium 50 microgram (91%) Calcium 50 mg (31%)	Vitamin A 400 microgram (50%) Vitamin B1 1,4 mg (127%) Vitamin B2 1,7 mg (121%) Vitamin B6 1,8 mg (129%) Vitamin B12 3 microgram (120%) Vitamin C 60 mg (75%) Vitamin D 5 mikrogram (100%) Vitamin E 10 mg (83%) Niacin 19 mg (119%) Folic acid 200 microgram (100%)

* (%RDI) = % of Recommended Daily Intake

Table 2. Summary of SWIDDICH study actions.

	Pregnancy					Child follow-up			
TIMEPOINT	**First pregnancy visit (<12 weeks)**	**Week 8 – 12**	**Week 25-28**	**Week 34-38**	**Post Partum Day 1-5** (subgroup)	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
ENROLMENT:									
Information given	X								
Eligibility screen	X								
Informed consent	X								
Allocation		X							
INTERVENTION ***Iodine 150 µg or Placebo in multivitamins***		←	—	→					
ASSESSMENTS:	**First pregnancy visit (<12 weeks)**	**Week 8 – 12**	**Week 25-28**	**Week 34-38**	**Post Partum Day 1-5** (subgroup)	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
Urinary iodine concentration		X	X	X	X (Child and mother)		X	X	X
Thyroid function*		X	X	X				X	X
Milk iodine concentration					X				
COGNITION: IQ							X WPPSI	X WISC	X WISC
Behavior							X CBCL	X CBCL Nordic 5-15	X CBCL Nordic 5-15
Psychomotor development						X ASQ-3		X Mov ABC	
Brain MRI *(subgroup)*								X	X
BACKGROUND INFORMATION:									
EUthyroid SES questionnaire *adults*						X	X	X	X
EUthyroid SES questionnaire *children*									X

Own questionnaire		X	X	X		X	X	X	X

mo: months, ys: years, WPPSI: Wechsler Preschool and Primary Scale of Intelligence, WISC: Wechsler Intelligence Scale for Children, ASQ-3: The Ages and Stages Questionnaire, Mov ABC: Movement Assessment Battery for Children, EUthyroid SES questionnaire: Socioeconomic Status questionnaire validated by EUthyroid foundation
* FT4 TSH, thyreoglobuline: serum sampling during pregnancy and dry blood spot sampling during children follow-up

BMJ Open

The Role of Iodine Containing Multivitamins during Pregnancy for Children's Brain Function: Protocol of an On-going Randomized Controlled Trial- the SWIDDICH study

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2017-019945.R1
Article Type:	Protocol
Date Submitted by the Author:	21-Dec-2017
Complete List of Authors:	Manousou, Sofia; Department of Medicine; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Johansson, Birgitta; Institute of Neuroscience and Physiology, Sahlgrenska Academy, Chmielewska, Anna; Department of Clinical Sciences, Pediatrics, Umeå University; Department of Pediatrics, Medical University of Warsaw Eriksson, Janna; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Gutefeldt, Kerstin; Department of Endocrinology, University Hospital of Linköping Tornhage, Carl-Johan; Department of Pediatrics, Skaraborg's Hospital; Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg Eggertsen, Robert; Mölnlycke Health Care Center; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Malmgren, Helge; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Hulthen, Lena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; 10Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg Domellöf, Magnus; Umea University, Department of Clinical Sciences Nystrom Filipsson, Helena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Department of Endocrinology, Sahlgrenska University Hospital
Primary Subject Heading:	Obstetrics and gynaecology
Secondary Subject Heading:	Paediatrics, Diabetes and endocrinology, Nutrition and metabolism
Keywords:	IODINE, CHILD DEVELOPMENT, COGNITION, Thyroid disease < DIABETES & ENDOCRINOLOGY, PUBLIC HEALTH

The Role of Iodine Containing Multivitamins during Pregnancy for Children's Brain Function:

Protocol of an

On-going Randomized Controlled Trial- the SWIDDICH study

Sofia Manousou[1,2], Birgitta Johansson[3], Anna Chmielewska[4,5], Janna Eriksson[1], Kerstin Gutefeldt[6], Carl-Johan Törnhage[7,8], Robert Eggertsen[1,9], Helge Malmgren[1,] Lena Hulthén[1,10], Magnus Domellöf[4] and Helena Filipsson Nyström[1,11]

[1] Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[2]Department of Medicine, Kungälv's Hospital, Kungälv, Sweden
[3]Institute of Neuroscience and Physiology, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[4] Department of Clinical Sciences, Pediatrics, Umeå University, Umeå, Sweden
[5] Department of Pediatrics, Medical University of Warsaw, Warsaw, Poland
[6] Department of Endocrinology, University Hospital of Linköping, Linköping, Sweden
[7] Department of Pediatrics, Skaraborg's Hospital, Skövde, Sweden
[8] Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
[9] Mölnlycke Health Care Center, Mölnlycke, Sweden,
[10]Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[11]Department of Endocrinology, Sahlgrenska University Hospital, Göteborg, Sweden

Corresponding author:
Sofia Manousou
Department of Internal Medicine
Kungälv's Hospital
Lasarettsgatan
SE-442 34 Kungälv
Sweden

Tel: +46 303 98000
Fax: +46 303 15438
Email: sofia.manousou@vgregion.se

Protocol Version: 1.0, 01 September 2017.

Conflicts of Interest: none.

Abstract

Introduction

Iodine is essential for normal brain development. Moderate and severe fetal iodine deficiency result in substantial to serious developmental delay in children. Mild iodine deficiency in pregnancy is associated with neurodevelopmental deficits in the offspring, but evidence from randomized trials is lacking. The aim of the SWIDDICH study is to determine the effect of daily supplementation with 150 µg iodine during pregnancy on the offspring's neuropsychological development up to 14 years of age.

Methods and Analysis

Thyroid healthy pregnant women (n=1275: age range 18 to 40 years) at ≤12 weeks gestation) are randomly assigned to receive multivitamin supplements containing 150 µg iodine or non-iodine containing multivitamin daily throughout pregnancy. As a primary outcome, intelligence quotient (IQ) will be measured in the offspring at 7 years (Wechsler Intelligence Scale for Children, WISC-V). As secondary outcomes, IQ will be measured at 3.5 and 14 years, psychomotor development at 18 months and 7 years, and behavior at 3.5, 7 and 14 years. Iodine status (urinary iodine concentration) will be measured during pregnancy and in the offspring at 3.5, 7 and 14 years. Thyroid function (thyroid hormones, thyroglobulin), and deiodinase type 2 polymorphisms will be measured during pregnancy and in the offspring at 7 and 14 years. Structural magnetic resonance imaging (MRI) or other relevant structural or functional brain imaging procedures will be performed in a subgroup of children at 7 and 14 years. Background and socioeconomic information will be collected at all follow-up times.

Ethics and Dissemination

This study is approved by the Ethics Committée in Göteborg, Sweden [Diary numbers: 431-12 approved 18th June 2012 (pregnancy part) and 1089-16 approved 8th February 2017 (children follow-up)]. According to Swedish regulations, dietary supplements are governed by the National Food Agency and not by the Medical Product Agency. Therefore, there is no requirement for a monitoring committée. The National Food Agency does not perform any audits of trial conduct. The trial is conducted according to the Declaration of Helsinki. The participating sites will be contacted regarding important protocol changes both orally and in writing, and the trial registry database will be updated accordingly. Study results will be presented at relevant conferences, and submitted to peer-reviewed journals with open access in the fields of endocrinology, pediatrics and nutrition. After the appropriate embargo period, the results will be communicated to participants, healthcare professionals at the maternal health care centers, the public, and other relevant groups, such as the national guideline group for thyroid and pregnancy and the National Food Agency.

Trial Registration Number: ClinicalTrials.gov identifier: NCT02378246, first registered 4 March 2015

Keywords: Iodine; Pregnancy; Child Development; Cognition; Thyroid Hormones

Strengths and Limitations of this study

- **Large interventional controlled trial** on iodine supplementation during pregnancy, powered to detect a difference of 3 IQ points in children.
- **Long observational follow-up** of the children, up to 14 years, with complex assessment of neurocognitive development.
- **Future implementation** of the study is feasible, as the intervention tablet exists on the market.
- **Lack of pure iodine** and pure placebo tablets implies careful interpretation of results.
- **Dropout rate** may be high.

BACKGROUND

Iodine Deficiency as an International Issue

Iodine is essential for the production of thyroid hormones and important for growth and brain development during fetal and early postnatal life [1]; a knowledge obtained after a long history of iodine deficiency (ID) associated disorders. For centuries, goiter with hypothyroidism, mental retardation and cretinism have been an entity. During the 1920s in the United States, Marine and Kimball performed the classic experiment of treating schoolgirls with iodine, leading to a dramatic reduction in the prevalence of goiter. Iodine prophylaxis was established in United States in 1921. After some debate, iodine prophylaxis was introduced in Switzerland in 1922, and then worldwide over the following decades. The combat against severe and moderate ID has been successful in reducing the number of children with ID-caused mental retardation. However, mild ID is widely apparent, especially during pregnancy [2], when dietary iodine demand increases from 150 to 250 µg/day [3].

Iodine Status in Sweden as the Country for this Study

Before iodination of table salt in 1936, ID was common in Sweden [4]. Current iodine intake is sufficient in the general population [5, 6] and was judged adequate during pregnancy during the 1990s [7, 8]; there is no recommendation on iodine supplementation during pregnancy. However, since the 1990s, the situation may have changed because dairy product consumption in adults is lower; milk iodine levels are lower than before [9, 10]; a reduction in salt intake is recommended for reducing the risk of hypertension; new salt forms (flake salt, gourmet salt) without iodine are popular; there is a reluctance to consume "food additives"; awareness of ID among the younger population is generally low; and, the main proportion of total salt intake (≈80%), i.e. from ready-made foods and dishes, does not provide iodine. Unless iodine is added to all salts used, the risk of decreased iodine intake is apparent, and arouses concerns especially for pregnant women. Retrospective, local data on pregnancy highlights this assumption is realistic [11].

Iodine Deficiency during Pregnancy: Effects on Child's Development

Severe and moderate ID leads to lower serum thyroid hormone levels and thereby to lower availability of thyroid hormones in the brain. During fetal life and early years, the growing brain is vulnerable [12, 13] and severe ID results in mental retardation in the newborn, unless thyroid hormone is replaced [14]. In

addition, an increased incidence of attention deficit hyperactivity disorders (ADHD) has been associated with mild-to-moderate ID [15].

In mild ID, thyroid hormone levels are maintained, whereas, thyroglobulin (TG) levels are increased as a biomarker of goiter. The brain's use of thyroid hormones depends on the local conversion of inactive hormone thyroxine (T4) to active hormone triiodothyronine (T3), a process mediated by deiodinase type 2 (D2) [16]. D2 is found in the hippocampi and the cerebral cortex and its activity is increased by ID, to maintain sufficient T3 levels [16, 17]. In the presence of normal thyroid hormone in blood, it is unclear how mild ID affects brain development. One theory is that this depends on deiodinases, which can change thyroid hormone signalling locally in different tissues, without affecting serum hormone concentration [16, 18].

Mild ID during pregnancy might have an impact on brain development, despite maintained normal thyroid hormone levels [19-22]. In the United Kingdom, a longitudinal study [19] found 8-year-old children have an increased risk of being in the lowest quartile of verbal IQ if their mothers had urinary iodine concentration (UIC) indicating mild ID in early pregnancy than children of mothers with normal iodine nutrition. In a similar association study from Australia [20], mild ID is linked with lower cognitive performance in 9-year old children. Results from an observational pilot-study from Italy [21] indicate mild-to-moderate ID during fetal life affects cognitive development, especially verbal abilities, even in absence of maternal thyroid insufficiency. In Norway, a large observational study [22] found that maternal iodine intake below the estimated average requirement during pregnancy was associated with reduced fine motor skills and verbal abilities and with more behavior problems at the age of 3 years.

As the randomized controlled trial (RCT) [23] evaluating 150 µg iodine/placebo in pregnant women in an iodine sufficient country was small (n=86) and lacked cognitive assessment in children, there were many expectations about the MITCH study [24]. In that trial, 832 women were randomized to 200 µg iodine/placebo. The data revealed that both groups were iodine sufficient in the second and third trimester, and that there was no difference in cognitive outcome in 5-6 year-old children. Hence, the MITCH study failed to answer this question, due to a lower than expected prevalence of ID in the studied population.To prevent subnormal fetal brain development, many international authorities recommend 150 µg extra iodine/day during pregnancy, despite the lack of studies on causality [25, 26].

Knowledge Gaps and Background to the SWIDDICH Study

There is a substantial gap in knowledge about mild ID during pregnancy and its potential negative consequences on neuropsychological development. Therefore, there is a need for a placebo-controlled trial that compares neuropsychological outcome in children exposed to mild ID during fetal life and children with normal iodine nutrition during pregnancy.

From 29 November 2012 until 1 June 2015, a pilot randomized placebo-controlled trial involving 200 pregnant women on a daily supplementation with either a multivitamin containing 150 µg iodine/day or a multivitamin without iodine (placebo) was conducted by our group. This study (ClinicalTrials.gov identifier: NCT02378246) aimed to evaluate the effects of iodine supplementation on UIC and thyroid function. As the MITCH study failed to answer the question if mild ID during pregnancy affects fetal brain development, it is evident to us that our trial needs to be expanded to include a sufficient number of pregnant women to enable a sufficiently powered child follow-up regarding neuropsychological development. In contrast to the MITCH study, we have reasons to believe, from our own data, that mild ID is prevalent in the third trimester in Sweden, with UIC 98 µg/L [11]. Therefore we conduct the **SW**edish **I**o**D**ine in Pregnancy and **D**evelopment **I**n **Ch**ildren (SWIDDICH) study. We hypothesize that

the use of an iodine containing multivitamin during pregnancy results in a better cognitive development of the child, compared with a multivitamin without minerals (superiority trial).

Objectives

The primary aim is to assess whether cognition (especially verbal competence) in children whose mothers received 150 µg iodine daily in a multivitamin during pregnancy is higher than children whose mothers received placebo (a multivitamin without iodine) and probably remained in mild ID. The purpose is to determine whether all pregnant women who live in a country where the general population is iodine sufficient, but live in conditions that can result in mild ID during pregnancy, should be recommended extra iodine during pregnancy.

METHODS

Design of the SWIDDICH Study

This is a randomized placebo-controlled study in which children are followed-up as an observational cohort, separated into two groups by the fetal iodine exposure.

Setting and Participants

Pregnant women will be recruited from more than ten maternal healthcare centers in Sweden with the aim to form several clusters, to facilitate children follow-up. The main study site will be in Gothenburg with secondary sites in Umeå, Linköping and other areas, where materal health care centers are recruited. At the first scheduled pregnancy visit, information about the study will be provided and written informed consent collected by the mid-wife. All procedures during pregnancy will be combined with routine pregnancy visits.

All informed consents, and blood and urine for future analyses will be sent to the main study site in Gothenburg. To promote participant retention and a complete follow-up, a contact from the study coordinator will be taken after childbirth. Also, information will be shared with participants on the homepage gu.se/swiddich.

Inclusion

The following inclusion criteria will apply: woman aged 18 to 40 years, pregnant at 7-12 weeks, willing to refrain from iodine supplementation and take a multivitamin supplement instead, without current thyroid disease, not in another pregnancy or lactating less than 6 months before inclusion, and non-vegan.

Randomization, Allocation, Concealment and Blinding

Randomization numbers, with an allocation ratio is 1:1, are prepared centrally and sent to each participating center. Consecutive numbers are used and the information regarding the study group allocation of each number stored securely at the premises of the University of Gothenburg, Sweden. Mothers are provided with a random container of pills, by either drawing a lot or blindly drawing a container. All containers are identical, with tasteless pills of the same size for both groups. Recruiting staff, study participants and those involved in laboratory work and developmental assessment are blinded to the group allocation. The code will only be broken by the central study team for data analyses before publications, but will still be blinded to all groups that work in the follow-up. The code has been broken for the 200 women of the pilot study, but all (ie. study participants, psychologists and lab engineers) except the central study team are still blinded. No other interimsanalyses are planned.

Intervention

Women in the experimental group receive a daily multivitamin supplement containing 150 µg iodine and those in the control group receive a daily multivitamin supplement containing no iodine (the contents of the two supplements are presented in Table 1). The intervention lasts throughout pregnancy to the day of delivery. Women in both groups are recommended, as all pregnant women in Sweden, to take extra folic acid during the first trimester and even extra iron, when the hemoglobin status indicates it. These administrations do not interfere with the study tablet. The women are not, however, allowed to take any other multivitamins, besides the study supplement.

Compliance - discontinuation

Participants are asked to bring the container with the remaining pills to the visit in the third trimester. The container is weighted and the percentage of intended doses used is calculated. Mothers who do not longer want to participate in the study during pergnancy will be regarded as drop-outs and no further data collection will be done. If there is a disconitinuation in the children follow up, children can come to the next visit. If the discontinuation is permanent, the registry search will still be done.

Outcomes

Outcomes in Mothers

Outcomes in mothers will be assessed in the first, second and third trimester of pregnancy. UIC and thyroid hormones will be measured in all three trimesters, and TPO-antibodies and TG in the first and third trimester.

Primary Outcome in Children

Cognition measured by intelligence quotient (total IQ) with focus on the verbal compound (verbal IQ) at 7 years is the primary outcome (Wechsler Intelligence Scale for Children, WISC-V [27]).

Secondary Outcomes in Children

Cognition measured by IQ at 3.5 years (Wechsler Preschool and Primary Scale of Intelligence, WPPSI-IV [28]) and at 14 years (Wechsler Intelligence Scale for Children, WISC-V [27] or an equivalent adequate version at the time) are secondary outcomes, together with outcomes related to psychomotor development, behavior and attention deficit hyperactivity disorder (ADHD). Psychomotor assessment will be done by the parents at 18 months (The Ages and Stages Questionnaire, ASQ-3) [29], and by a physiotherapist at 7 years (Movement Assessment Battery for Children, Movement ABC test) [30]. Behavior will be assessed through parental questionnaires, the Child Behavior Checklist (CBCL); first at 3.5 years (CBCL 1-5) [31], then at 7 and 14 years (CBCL 6-18) [32]. At 7 and 14 years, the Nordic questionnaire 5-15 [33] will be used to assess ADHD-related symptoms.

Parents also admit to a registry search at 3.5, 7 and 14 years. This search concerns the In and Out-patients registries to collect information on medical diagnoses, the Drug registry, the Medical Brith registry, quality registrries, maternal- child- and school-health care for medical and growth data, and Educational registries.

In a subgroup of children (n=200), magnetic resonance imaging (MRI) of the brain (with a 3T Philips MR scanner) is planned at 7 and 14 years to evaluate structural brain changes. Automatic segmentation of the whole brain will be done with Freesurfer [34] and Maper, multi-atlas propagation with enhanced registration [35]. Medio-temporal lobe (MTL) structures will be analyzed through manual segmentation using custom software developed in previous projects [36, 37]. Subregional analyses directed at regions of neurogenesis will be included. Intracranial volume measured manually will enable reliable normalization

of MTL volumes. Other structural and/or functional brain imaging methods may supplement, or even replace, the described protocol, depending on the state of knowledge at the time of study.

Possible Confounding Variables and Background Information

In children, UIC will be measured from the 3.5-year visit and forward and dry blood spots will be collected for thyroid hormones, TG and deiodinases at 7 and 14 years. Background and confounding variables will be assessed at 18 months, 3.5, 7 and 14 years.

Timeframe for the Study Actions

Recruitment to the SWIDDICH study began in March 2017 and is planned to be completed in 2019. Currently 75 of 1075 pregnant women have been included. Several strategies are used to reach target sample size: a study coordinator is emplyoyed with only purpose to have contact with maternal health care centers, a step-wise reinburshment model is applied to the maternal health care centers with higher rates in case of high recruitment rates, and the National Food Agency promotes study participation in their communication with maternal health care centers. Also, local pediatricians are involved to faciliate the children follow-up. The follow-up of children is also offered to the families who participated in the pilot study (2012-2015), before the decision on study extension was made. The time points for all study actions are presented in Table 2.

CONSIDERATIONS

Considerations on the content of the intervention and the "placebo" tablets

The reason for choosing iodine-containing multivitamins instead of pure iodine tablets as intervention is to ensure that future implementation of the study is feasible. There are currently no pure iodine tablets on the market. In the planning state of the study, discussions were made with pharmaceutical companies to provide pure iodine tablets and placebo, but the interest was low. Iodine in multivitamins will be the only available supplement source in most countries in the future. Therefore, a multivitamin containing 150 µg iodine was chosen for the intervention and a multivitamine without minerals as the comparator.

Other components in the multivitamin products, besides iodine, may intervene with outcomes. It is proposed that vitamin B12 [38, 39] and iron [40] can have positive effects on the brain, and iron and selenium influence thyroid hormone levels [41, 42]. Iron is found in thyroperoxidase (TPO) enzyme that couples iodine to thyroglobulin and selenium is found in deiodinases, such as D2 that converts T4 to T3. Sweden is a selenium deficient country [43], but it is unclear whether selenium deficiency affects cognitive outcome in humans [44]. B12 is higher in iodine-containing multivitamin where iron and selenium also are included. However, B12 content in both placebo and intervention tablets is, at least, equal to the recommended daily intake for B12, thus, B12 deficiency is not anticipated in any of the groups. In addition, the iron contents are low. Many pregnant Swedish women take a separate 100 mg iron supplement, which makes the 12 mg iron in the intervention tablet negligible. Iron, B12 and selenium will be measured in a subpopulation to evaluate possible group differences and contributions to thyroid metabolism.

Considerations in choosing realistic starting point of intervention.

Fetal brain development during the first 12 weeks is dependent on maternal T4 levels. By initiating the intervention at pregnancy week 7-12, a substantial part of the first trimester is missed. Iodine supplements may ideally be initiated before conception. Practically, a recruitment of women who plan a pregnancy is difficult, as these women are not known by health care before pregnancy. The only way would be an advertisement in the newspaper to recruit women that are planning pregnancy. This would be ineffective

and would create a selection bias, as only 50% of those that get pregnant have planned it and not every woman responds to an advertisement. We include women at the earliest possible stage and this is far earlier that in the recent publication by Casey et al [45] with negative results, that included pregnant women in mean gestational week 16.6-18.0. The inclusion in our study is similar to the one in the MITCH study, where women were included in gestational week 10-11 [24].

Power Calculation, Data Management, Statistical Considerations, and Authorship

The sample size needed, excluding dropouts, is calculated to 788 children (394 in each group) for an effect size of 3 IQ points with SD 15 and power 0.80. Currently, there are no similar randomized studies for power calculation. The smallest significant effect of 3 IQ points is in accordance with an observational study [19], where children of mothers with UIC< 150 µg/L during pregnancy had a 3-point lower IQ at school-age than children of mothers with normal UIC during pregnancy. This expected effect from iodine supplementation in mild iodine deficiency is also suggested by Troendle [45], where statistical considerations are discussed for the possibility that the needed placebo-controlled study is conducted. Assuming a drop out frequency of 22% during pregnancy (which is in accordance with preliminary data from our pilot study with 200 pregnant women) and 20% during the children follow up, 1263 pregnant women need to be recruited to the study. This sample size is in general agreement with Troendle [45], thus, the decision was made to try and recruit 1275 pregnant women. The dropout frequency for the children follow-up could be lower than estimated as there are two occasions for dropout and mothers who remain in the study after the first follow-up can be assumed willing to continue the study. The power calculation assumes the use of an unpaired t-test between groups; however, more advanced analyses could decrease variance, thus requiring a lower sample size.

The sample size will be reassessed by calculating the dropout frequency when 750 women are included and when half of the children from the first 200 included women have been invited to the 3.5-year neuropsychological evaluation. Sample size reassessment will be conducted without unblinding the study groups.

A 100% compliance to the study medication is assumed, as the results will be based on an intention to treat (ITT) analysis. Compliance is monitored to enable a per protocol analysis (only the compliant participants included) to be added. However, the ITT approach reflects the real life clinical situation, in which a certain number of patients are not compliant with the recommended treatment, and this will be the foundation for future recommendations on iodine supplementation to all pregnant women.

A separate power calculation for the MRI investigation has been done. This assumes the described protocol will be followed, and a previous study of 11-year old children has been used for guidance [46]. To detect a 5% difference with power 0.80, 60 children are needed in each group. As the variation in the hippocampal volumes in 7-year-old children could be slightly larger than in the previous study [46] and since the dropout from the MRI at 14 years needs to be reckoned with, 100 children will be included in each group.

Coded collected data will be entered into a database, with appropriate back-up from the university servers. Key lists will be kept safe. The transfer of data to the databases will be validated by random cross-checks with the original data set. UIC analyses will be run in duplicate to promote validity. For further deatails see ethical applications and clinical.trials.gov. All authors will have access to all data. The statisticians will have access to the data needed.

The choice of methods for comparing the main outcome between the experimental and control groups will be guided by the data distributions. In case of deviation from normality assumptions, transformations of data may be done. Non-parametric tests will be used for non-normal and ordinal data. Possible confounders, such as socioeconomic factors, other background information, gestational age, thyroid hormones, TG, deiodinase polymorphisms and UIC, will be considered in the data analysis. Repeated measurements in a mixed model (where groups are compared repeatedly at 3.5, 7 and 14 years) and within-group analyses are planned. The models will also consider the dropout frequency and recruitment from different maternal health care centers, which will be used as a factor in the analysis. For all drop-outs, relevant background variables will be studied. Adjustments for bias may be performed. For non-informative drop-outs, methods for multiple imputations will be considered. A multivariate analysis with total grey, total brain volume, intracranial volume, MTL volumes and possibly other measures of brain structure and function as independent variables will be done. The data analyses will be undertaken by an experienced statistician. Authorships will be decided according to the Declaration of Vancouver.

MRI considerations – Where Are Changes from ID Located?

T3 receptors are distributed among all brain areas with high levels in the hippocampi and the cerebellar cortex. Rodent data indicate that T3 receptors are involved in the regulation of hippocampal structure and function [47]. In the human cerebral cortex, thyroid receptors are already present in week 9 and concentrations increase up to 18 weeks of gestation [48]. Considerable amounts of D2 are also found in the cerebral cortex [49]. In the first half of pregnancy, the fetus is dependent on the mother's supply of thyroid hormones. In mild ID, the mother maintains serum thyroid hormone levels through unknown compensatory mechanisms. In the second half of a mild ID pregnancy, when the fetus partly relies on its own thyroid hormone production, the fetus will be hypothyroid as it has not developed compensatory mechanisms and there is a lack of sufficient iodine levels transferred by the mother [49].

The description of neuropathology caused by ID is limited to few observations from adult cretins, ranging from severe cortical atrophy to almost normal appearance. In areas with endemic goiter, fetuses aborted in the second half of the pregnancy have a less differentiated cerebral cortex [49]. In rats, transient periods of thyroid hormone insufficiency during periods of cortical development affect cortical and hippocampal cytoarchitecture [49].

Human data from maternal hypothyroidism support an effect on the brain, specifically on the hippocampus [50]. These data are in line with the recent publication by Korevaar *et al* [51], who conclude the relationship of IQ with FT4 (in peripheral blood) exhibits a u-shaped configuration with lower IQ levels in both ends of the normal range. FT4 in this study is also associated with total grey matter volume.

Considerations on the Neuropsychological Evaluation

Neuropsychological development can be divided into three domains: psychomotor, cognitive (IQ) and socio-emotional development (Figure 1). There are five landmark studies in the iodine field evaluating neuropsychological development in the off-spring that use neuropsychological tests: the ALSPAC (United Kingdom) [19], INMA (Spain) [52], Generation R (Netherlands) [51], MITCH (India and Thailand) [53] and the study by Hynes *et al* (Australia) [20]. Verbal cognitive function appears to be the most susceptible subdomain for ID. In SWIDDICH, verbal cognitive function together with total IQ were chosen (as the latter is the best understood and requested) as primary outcome measurements. As cognitive testing is less valid in younger ages, verbal IQ at 7 years was chosen as the primary evaluation time point and all three domains of neuropsychological development will be evaluated at several follow-up times.

Implications for Society and the Individual

Impaired child development increases economic burdens for society. Lowered IQ is associated with worse economic outcomes and lower lifetime earnings. Small decrements in IQ around the mean are linked to lower incomes [54, 55]. IQ may be the easiest factor to quantify, but may not be the factor with the most serious consequence for a "good life". Environmental factors, including ID, that place the nervous system at risk may affect executive functions, such as planning and initiating ideas and result in attention problems, impulsive behavior, and inability to handle stress and disappointments, thus, impeding success in school and in life and possibly leading to antisocial behavior [56].

If the average IQ of a population drops, the IQ distribution shifts and the number of individuals with low IQ (e.g below 75 or 85, classified as intellectually disabled) increases. In turn, this will also decrease the number of gifted and exceptionally gifted people with high IQs (e.g above 130), who may have major positive impacts on the immediate future for a company or a country. A cost-benefit analysis of iodine supplementation in mild-to moderate ID is recently proved positive [57].

Based on the dollar value in 1987 in the USA, the cost in terms of reduced income for a one point IQ reduction has been calculated to nearly 20.7 billion USD per year [58]. A 3-point decline in IQ also impacts social costs in the United States [56] and increases the risk of: poverty by 20% during the first three years; low birth weight by 12%; being a recipient of welfare by 18%; and, high school dropout by 28%. Even though a decline of a few IQ points may be small for the individual, the societal effects are considerable. As a small general risk reduction entails a large social benefit, iodine supplementation could be a cost-effective action if the main hypothesis of this study holds true.

Considerations on Possible Adverse Effects of Iodine or Placebo

Iodine supplementation may increase the frequency of post-partum thyroiditis (PPT), as iodine affects autoimmunity [59]. 10-15% of women already have post-partum thyroiditis and this number may increase slightly with iodine supplementation. As PPT is not a dangerous condition and most cases resolve spontaneously, we consider the reduced risk for subnormal brain development in a child motivates accepting the risk for PPT. In Denmark, postpartum thyroiditis was evaluated in a placebo-controlled trial in mild-moderate iodine deficiency, and treatment did not increase or worsen PPT [59].

Excess iodine intake in the mother may block thyroid function in the fetus, leading to hypothyroidism and goiter, and has been associated with poorer mental and pshychomotor development or behavior problems in children [22, 52, 60]. However, the risk of iodine excess seems larger, if the normal population is iodine deficient, which is in contrast to the Swedish normal population.

The placebo group is at risk of iodine deficiency during pregnancy. However, as there are no current recommendations in Sweden for iodine supplementation in pregnancy, this group follows normal management.

The intervention and the comparator are diet supplements, and the total intake of nutrients depends on the diet. Information on adverse reactions is not collected.

CONCLUSION

The aim of this paper is to describe the study protocol for the SWIDDICH research project and the considerations that led to its design. The study attempts to further understand the consequences of mild ID during pregnancy and to test whether treatment of the mothers with an iodine-containing-multivitamin improves outcome in the children. As the study is the largest of its kind, it offers the potential for

influencing future recommendations on iodine supplementation with multivitamins to pregnant women living in conditions of mild ID.

ABBREVIATIONS

IQ: intelligence quotient; ADHD: attention deficit hyperactivity disorders; TG: thyroglobulin; T4: thyroxine; T3: triiodothyronine; D2: deiodinase type 2; UIC: urinary iodine concentration; RCT: randomized controlled trial; TPO: thyroperoxidase; MRI: magnetic resonance imaging; MTL: medio-temporal lobe.

Data Sharing

Data from this trial may be shared for individual data analysis in the future. A detailed plan for data sharing will be developed during the later phase of the project.

Authors' Contributions

Sofia Manousou, Birgitta Johansson, Anna Chmielewska, Janna Eriksson, Kerstin Gutefeldt, Carl-Johan Törnhage, Robert Eggertsen, Helge Malmgren, Lena Hulthén, Magnus Domellöf and Helena Filipsson Nyström have contributed to the design of the SWIDDICH study. HFN wrote the first version of the manuscript and SM was responsible for pushing the work forward together with the other coworkers. All co-authors critically reviewed and approved the final version of the manuscript. SM is the guarantor.

Primary sponsor is Helena Filipsson Nyström (principal investigator), Sahlgrenska Academy and University of Gothenburg and Sahlgrenska University Hospital, Göteborg, Sweden. The main study site is in Gothenburg, but additional sites are Umeå and Linköping.

Funding Statement

This work was supported by the ALF agreement (grant number ALFGBG-58777, ALFGBG-717311), Regional FOU (grant number VGFOUREG-664301**),** Lilla barnets fond (grant number 20160917), Svenska Läkarsällskapet (grant number SLS-688891), and Lars Hiertas minne foundation (grant number FO2016-0016), Formas grant (grant number 2017-0095), Grant from General Maternity Hospital Foundation 2017. Multivitamins for the first 200 women were provided by courtesy of Recip medical company, Solna, Sweden, but they are not involved in the study design and they do not contribute in any other way. The National Food Agency is a stakeholder of this trial. The authors of this manuscript are the only ones contributing to design, management, future analyses with the support of unbound statisticians, interpretation of data, writing the manuscript and decision on where to submit. The maternal health care centers are reimbursed for the collection of patients by the principal investigator (HFN).

Acknowledgements: Elisabeth Gramatkovski for her invaluable help as coordinator of the study.

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Table 2. Summary of SWIDDICH study actions.

Figure 1. Neuropsychological domains. Verbal cognitive function, as a subdomain of cognition, appears the most susceptible subdomain for iodine deficiency and is the primary outcome of the SWIDDICH study at the age of 7 years.

References

[1] M.B. Zimmermann, P.L. Jooste, C.S. Pandav, Iodine-deficiency disorders, Lancet 372(9645) (2008) 1251-62.
[2] M.B. Zimmermann, M. Gizak, K. Abbott, M. Andersson, J.H. Lazarus, Iodine deficiency in pregnant women in Europe, The lancet. Diabetes & endocrinology 3(9) (2015) 672-4.
[3] W.H.O. United Nations Children's Fund & International Council for the Control of Iodine Deficiency Disorders, Assessment of iodine deficiency disorders and monitoring their elimination. A guide for programme managers, 3:e edition, Geneva, 2007.
[4] H.F. Nystrom, S. Jansson, G. Berg, Incidence Rate and Clinical features of Hyperthyroidism in a long-term iodine sufficient area of Sweden (Gothenburg) 2003-2005, Clin Endocrinol (Oxf) (2013).
[5] M. Andersson, G. Berg, R. Eggertsen, H. Filipsson, E. Gramatkovski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Adequate iodine nutrition in Sweden: a cross-sectional national study of urinary iodine concentration in school-age children, Eur. J. Clin. Nutr. 63(7) (2009) 828-34.
[6] H. Filipsson Nystrom, M. Andersson, G. Berg, R. Eggertsen, E. Gramatkowski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Thyroid volume in Swedish school children: a national, stratified, population-based survey, Eur. J. Clin. Nutr. 64(11) (2010) 1289-95.
[7] B. Elnagar, A. Eltom, L. Wide, M. Gebre-Medhin, F.A. Karlsson, Iodine status, thyroid function and pregnancy: study of Swedish and Sudanese women, Eur. J. Clin. Nutr. 52(5) (1998) 351-5.
[8] A. Eltom, B. Elnagar, M. Elbagir, M. Gebre-Medhin, Thyroglobulin in serum as an indicator of iodine status during pregnancy, Scand J Clin Lab Invest. 60(1) (2000) 1-7.
[9] H. Lindmark Månsson, the Swedish Milk composition Svensk Mjölk (Swedish milk) 2010.
[10] H. Lindmark Månsson, Den svenska mejerimjölkens sammansättning 2009, Svensk Mjök (Swedish Milk), 2012.
[11] M. Granfors, M. Andersson, S. Stinca, H. Akerud, A. Skalkidou, I. Sundstrom Poromaa, A.K. Wikstrom, H. Filipsson Nystrom, Iodine deficiency in a study population of pregnant women in Sweden, Acta Obstet. Gynecol. Scand. (2015).
[12] M.B. Zimmermann, The adverse effects of mild-to-moderate iodine deficiency during pregnancy and childhood: a review, Thyroid 17(9) (2007) 829-35.
[13] P.N. Taylor, O.E. Okosieme, C.M. Dayan, J.H. Lazarus, Therapy of endocrine disease: Impact of iodine supplementation in mild-to-moderate iodine deficiency: systematic review and meta-analysis, Eur. J. Endocrinol. 170(1) (2014) R1-R15.
[14] J.B. Stanbury, A.M. Ermans, B.S. Hetzel, E.A. Pretell, A. Querido, Endemic goitre and cretinism: public health significance and prevention, WHO Chron. 28(5) (1974) 220-8.
[15] F. Vermiglio, V.P. Lo Presti, M. Moleti, M. Sidoti, G. Tortorella, G. Scaffidi, M.G. Castagna, F. Mattina, M.A. Violi, A. Crisa, A. Artemisia, F. Trimarchi, Attention deficit and hyperactivity disorders in the offspring of mothers exposed to mild-moderate iodine deficiency: a possible novel iodine deficiency disorder in developed countries, J. Clin. Endocrinol. Metab. 89(12) (2004) 6054-60.
[16] R. Peeters, C. Fekete, C. Goncalves, G. Legradi, H.M. Tu, J.W. Harney, A.C. Bianco, R.M. Lechan, P.R. Larsen, Regional physiological adaptation of the central nervous system deiodinases to iodine deficiency, Am. J. Physiol. Endocrinol. Metab. 281(1) (2001) E54-61.
[17] N. Markova, A. Chernopiatko, C.A. Schroeter, D. Malin, A. Kubatiev, S. Bachurin, J. Costa-Nunes, H.M. Steinbusch, T. Strekalova, Hippocampal gene expression of deiodinases 2 and 3 and effects of 3,5-diiodo-L-thyronine T2 in mouse depression paradigms, BioMed research international 2013 (2013) 565218.
[18] F. Courtin, H. Zrouri, A. Lamirand, W.W. Li, G. Mercier, M. Schumacher, C.L. Goascogne, M. Pierre, Thyroid hormone deiodinases in the central and peripheral nervous system, Thyroid 15(8) (2005) 931-42.
[19] S.C. Bath, C.D. Steer, J. Golding, P. Emmett, M.P. Rayman, Effect of inadequate iodine status in UK pregnant women on cognitive outcomes in their children: results from the Avon Longitudinal Study of Parents and Children (ALSPAC), Lancet 382(9889) (2013) 331-7.
[20] K.L. Hynes, P. Otahal, I. Hay, J.R. Burgess, Mild iodine deficiency during pregnancy is associated with reduced educational outcomes in the offspring: 9-year follow-up of the gestational iodine cohort, J. Clin. Endocrinol. Metab. 98(5) (2013) 1954-62.

[21] M. Moleti, F. Trimarchi, G. Tortorella, A. Candia Longo, G. Giorgianni, G. Sturniolo, A. Alibrandi, F. Vermiglio, Effects of Maternal Iodine Nutrition and Thyroid Status on Cognitive Development in Offspring: A Pilot Study, Thyroid 26(2) (2016) 296-305.
[22] M.H. Abel, I.H. Caspersen, H.M. Meltzer, M. Haugen, R.E. Brandlistuen, H. Aase, J. Alexander, L.E. Torheim, A.L. Brantsaeter, Suboptimal Maternal Iodine Intake Is Associated with Impaired Child Neurodevelopment at 3 Years of Age in the Norwegian Mother and Child Cohort Study, J. Nutr. 147(7) (2017) 1314-1324.
[23] F. Brucker-Davis, P. Panaia-Ferrari, J. Gal, P. Fenichel, S. Hieronimus, Iodine Supplementation throughout Pregnancy Does Not Prevent the Drop in FT4 in the Second and Third Trimesters in Women with Normal Initial Thyroid Function, European thyroid journal 2(3) (2013) 187-94.
[24] S. Gowachirapant, N. Jaiswal, A. Melse-Boonstra, V. Galetti, S. Stinca, I. Mackenzie, S. Thomas, T. Thomas, P. Winichagoon, K. Srinivasan, M.B. Zimmermann, Effect of iodine supplementation in pregnant women on child neurodevelopment: a randomised, double-blind, placebo-controlled trial, The lancet. Diabetes & endocrinology 5(11) (2017) 853-863.
[25] A. Stagnaro-Green, M. Abalovich, E. Alexander, F. Azizi, J. Mestman, R. Negro, A. Nixon, E.N. Pearce, O.P. Soldin, S. Sullivan, W. Wiersinga, P. American Thyroid Association Taskforce on Thyroid Disease During, Postpartum, Guidelines of the American Thyroid Association for the diagnosis and management of thyroid disease during pregnancy and postpartum, Thyroid 21(10) (2011) 1081-125.
[26] J. Lazarus, R.S. Brown, C. Daumerie, A. Hubalewska-Dydejczyk, R. Negro, B. Vaidya, 2014 European thyroid association guidelines for the management of subclinical hypothyroidism in pregnancy and in children, European thyroid journal 3(2) (2014) 76-94.
[27] D. Wechsler, Wechsler Intelligence Scale for Children – Fifth Edition (WISC-V), Swedish version, Stockholm, Sweden, 2016.
[28] D. Wechsler, Wechsler Preschool and Primary Scale of Intelligence – Fourth Edition (WPPSI-IV) (Swedish version), Stockholm, Sweden, 2014.
[29] T.E. Squires J, Bricker D, Potter L, ASQ-3 User's Guide. 3rd ed. , Baltimore, MD, US, 2009.
[30] S.E. Henderson, Sugden, D. A., & Barnett, A. L. , Movement assessment battery for children [examiner's manual] (2nd ed.). , London, UK, 2007.
[31] R.L. Achenbach TM, Manual for the ASEBA Preschool Forms & Profiles. , University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2000.
[32] T.M. Achenbach, Rescorla, L.A. , Manual for the ASEBA School-Age Forms & Profiles, University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2001.
[33] B. Kadesjo, L.O. Janols, M. Korkman, K. Mickelsson, G. Strand, A. Trillingsgaard, C. Gillberg, The FTF (Five to Fifteen): the development of a parent questionnaire for the assessment of ADHD and comorbid conditions, Eur. Child Adolesc. Psychiatry 13 Suppl 3 (2004) 3-13.
[34] B. Fischl, D.H. Salat, E. Busa, M. Albert, M. Dieterich, C. Haselgrove, A. van der Kouwe, R. Killiany, D. Kennedy, S. Klaveness, A. Montillo, N. Makris, B. Rosen, A.M. Dale, Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain, Neuron 33(3) (2002) 341-55.
[35] R.A. Heckemann, S. Keihaninejad, P. Aljabar, D. Rueckert, J.V. Hajnal, A. Hammers, Improving intersubject image registration using tissue-class information benefits robustness and accuracy of multi-atlas based anatomical segmentation, Neuroimage 51(1) (2010) 221-7.
[36] C. Eckerstrom, E. Olsson, M. Borga, S. Ekholm, S. Ribbelin, S. Rolstad, G. Starck, A. Edman, A. Wallin, H. Malmgren, Small baseline volume of left hippocampus is associated with subsequent conversion of MCI into dementia: the Goteborg MCI study, J. Neurol. Sci. 272(1-2) (2008) 48-59.
[37] E. Olsson, C. Eckerstrom, G. Berg, M. Borga, S. Ekholm, G. Johannsson, S. Ribbelin, G. Starck, A. Wysocka, E. Lofdahl, H. Malmgren, Hippocampal volumes in patients exposed to low-dose radiation to the basal brain. A case-control study in long-term survivors from cancer in the head and neck region, Radiat. Oncol. 7 (2012) 202.
[38] V. Bhate, S. Deshpande, D. Bhat, N. Joshi, R. Ladkat, S. Watve, C. Fall, C.A. de Jager, H. Refsum, C. Yajnik, Vitamin B12 status of pregnant Indian women and cognitive function in their 9-year-old children, Food Nutr Bull 29(4) (2008) 249-54.

[39] M.M. Black, Effects of vitamin B12 and folate deficiency on brain development in children, Food Nutr Bull 29(2 Suppl) (2008) S126-31.
[40] E.L. Prado, K.G. Dewey, Nutrition and brain development in early life, Nutr. Rev. 72(4) (2014) 267-84.
[41] M. Ventura, M. Melo, F. Carrilho, Selenium and Thyroid Disease: From Pathophysiology to Treatment, Int. J. Endocrinol. 2017 (2017) 1297658.
[42] S. Hu, M.P. Rayman, Multiple Nutritional Factors and the Risk of Hashimoto's Thyroiditis, Thyroid 27(5) (2017) 597-610.
[43] O. Selinus, Medical geology: Iodine - a classical element (jod ett klassiskt element), Medical geololgy (Medicinsk Geologi), Authors and Studentlitteratur2010, pp. 387-400.
[44] E. Wasantwisut, Nutrition and development: other micronutrients' effect on growth and cognition, Southeast Asian J. Trop. Med. Public Health 28 Suppl 2 (1997) 78-82.
[45] J.F. Troendle, Statistical design considerations applicable to clinical trials of iodine supplementation in pregnant women who may be mildly iodine deficient, Am. J. Clin. Nutr. 104 Suppl 3 (2016) 924S-7S.
[46] L. Bunketorp Kall, H. Malmgren, E. Olsson, T. Linden, M. Nilsson, Effects of a Curricular Physical Activity Intervention on Children's School Performance, Wellness, and Brain Development, J. Sch. Health 85(10) (2015) 704-13.
[47] A. Guadano-Ferraz, R. Benavides-Piccione, C. Venero, C. Lancha, B. Vennstrom, C. Sandi, J. DeFelipe, J. Bernal, Lack of thyroid hormone receptor alpha1 is associated with selective alterations in behavior and hippocampal circuits, Mol. Psychiatry 8(1) (2003) 30-8.
[48] B. Ferreiro, J. Bernal, C.G. Goodyer, C.L. Branchard, Estimation of nuclear thyroid hormone receptor saturation in human fetal brain and lung during early gestation, J. Clin. Endocrinol. Metab. 67(4) (1988) 853-6.
[49] G.M. de Escobar, M.J. Obregon, F.E. del Rey, Iodine deficiency and brain development in the first half of pregnancy, Public Health Nutr. 10(12A) (2007) 1554-70.
[50] K.A. Willoughby, M.P. McAndrews, J.F. Rovet, Effects of maternal hypothyroidism on offspring hippocampus and memory, Thyroid 24(3) (2014) 576-84.
[51] T.I. Korevaar, R. Muetzel, M. Medici, L. Chaker, V.W. Jaddoe, Y.B. de Rijke, E.A. Steegers, T.J. Visser, T. White, H. Tiemeier, R.P. Peeters, Association of maternal thyroid function during early pregnancy with offspring IQ and brain morphology in childhood: a population-based prospective cohort study, The lancet. Diabetes & endocrinology 4(1) (2016) 35-43.
[52] M. Rebagliato, M. Murcia, M. Alvarez-Pedrerol, M. Espada, A. Fernandez-Somoano, N. Lertxundi, E.M. Navarrete-Munoz, J. Forns, A. Aranbarri, S. Llop, J. Julvez, A. Tardon, F. Ballester, Iodine supplementation during pregnancy and infant neuropsychological development. INMA Mother and Child Cohort Study, Am. J. Epidemiol. 177(9) (2013) 944-53.
[53] A. Melse-Boonstra, S. Gowachirapant, N. Jaiswal, P. Winichagoon, K. Srinivasan, M.B. Zimmermann, Iodine supplementation in pregnancy and its effect on child cognition, J. Trace Elem. Med. Biol. 26(2-3) (2012) 134-6.
[54] J. Schwartz, Societal benefits of reducing lead exposure, Environ. Res. 66(1) (1994) 105-24.
[55] D.S. Salkever, Updated estimates of earnings benefits from reduced exposure of children to environmental lead, Environ. Res. 70(1) (1995) 1-6.
[56] T. Muir, M. Zegarac, Societal costs of exposure to toxic substances: economic and health costs of four case studies that are candidates for environmental causation, Environ. Health Perspect. 109 Suppl 6 (2001) 885-903.
[57] M. Monahan, K. Boelaert, K. Jolly, S. Chan, P. Barton, T.E. Roberts, Costs and benefits of iodine supplementation for pregnant women in a mildly to moderately iodine-deficient population: a modelling analysis, The lancet. Diabetes & endocrinology 3(9) (2015) 715-22.
[58] J. Schwartz, Low-level lead exposure and children's IQ: a meta-analysis and search for a threshold, Environ. Res. 65(1) (1994) 42-55.
[59] S.B. Nohr, A. Jorgensen, K.M. Pedersen, P. Laurberg, Postpartum thyroid dysfunction in pregnant thyroid peroxidase antibody-positive women living in an area with mild to moderate iodine deficiency: is iodine supplementation safe?, J. Clin. Endocrinol. Metab. 85(9) (2000) 3191-8.

[60] M. Murcia, M. Rebagliato, C. Iniguez, M.J. Lopez-Espinosa, M. Estarlich, B. Plaza, C. Barona-Vilar, M. Espada, J. Vioque, F. Ballester, Effect of iodine supplementation during pregnancy on infant neurodevelopment at 1 year of age, Am. J. Epidemiol. 173(7) (2011) 804-12.

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Intervention (iodine 150 microgram) **Commercial name: MITT VAL VEGETARIAN**	**Placebo (no iodine)** **Commercial name: ENOMDAN**
B2 1,4 mg (100%)* B12 15 microgram (600%) Iron 12 mg (86%) Zink 12 mg (120%) Iodine 150 microgram (100%) Selenium 50 microgram (91%) Calcium 50 mg (31%)	Vitamin A 400 microgram (50%) Vitamin B1 1,4 mg (127%) Vitamin B2 1,7 mg (121%) Vitamin B6 1,8 mg (129%) Vitamin B12 3 microgram (120%) Vitamin C 60 mg (75%) Vitamin D 5 mikrogram (100%) Vitamin E 10 mg (83%) Niacin 19 mg (119%) Folic acid 200 microgram (100%)

* (%RDI) = % of Recommended Daily Intake

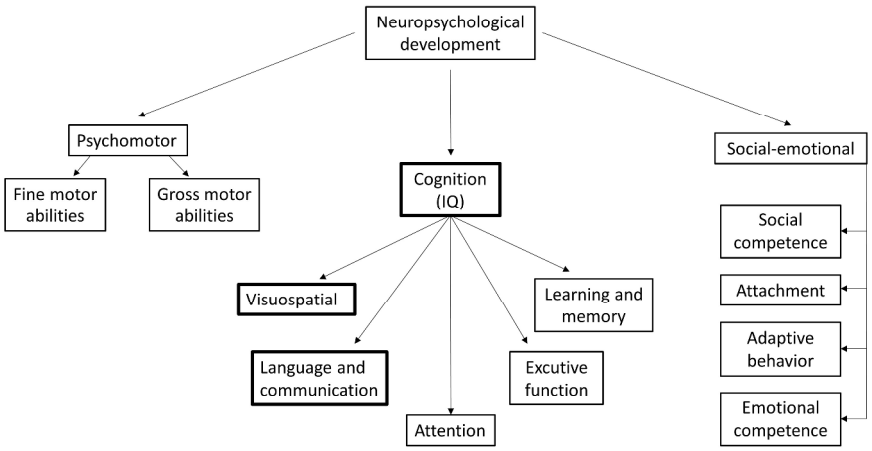
Table 2. Summary of SWIDDICH study actions.

	Pregnancy				Child follow-up			
TIMEPOINT	**First pregnancy visit (<12 weeks)**	**Week 7 – 12**	**Week 25-28**	**Week 34-38**	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
ENROLMENT:								
Information given	X							
Eligibility screen	X							
Informed consent	X							
Allocation		X						
INTERVENTION ***Iodine 150 µg or Placebo in multivitamins***		◆—	—	—◆				
ASSESSMENTS:	**First pregnancy visit (<12 weeks)**	**Week 7 – 12**	**Week 25-28**	**Week 34-38**	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
Urinary iodine concentration		X	X	X		X	X	X
Thyroid function*		X	X	X			X	X
Milk iodine concentration								
COGNITION: IQ						X WPPSI	X WISC	X WISC
Behavior						X CBCL	X CBCL Nordic 5-15	X CBCL Nordic 5-15
Psychomotor development					X ASQ-3		X Mov ABC	
Brain MRI *(subgroup)*							X	X
BACKGROUND INFORMATION:								
EUthyroid SES questionnaire *adults*					X	X	X	X
EUthyroid SES questionnaire *children*								X

Own questionnaire		X	X	X	X	X	X	X

mo: months, ys: years, WPPSI: Wechsler Preschool and Primary Scale of Intelligence, WISC: Wechsler Intelligence Scale for Children, ASQ-3: The Ages and Stages Questionnaire, Mov ABC: Movement Assessment Battery for Children, EUthyroid SES questionnaire**:** Socioeconomic Status questionnaire, validated by EUthyroid foundation

* FT4 TSH, thyreoglobuline: serum sampling during pregnancy and dry blood spot sampling during children follow-up



Neuropsychological domains. Verbal cognitive function, as a subdomain of cognition, appears the most susceptible subdomain for iodine deficiency and is the primary outcome of the SWIDDICH study at the age of 7 years.

338x190mm (300 x 300 DPI)

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Page
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	2
	2b	All items from the World Health Organization Trial Registration Data Set	1, 2, 4-7, 11
Protocol version	3	Date and version identifier	1
Funding	4	Sources and types of financial, material, and other support	11
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1, 11
	5b	Name and contact information for the trial sponsor	1
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	11
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	5
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	3-4
	6b	Explanation for choice of comparators	7
Objectives	7	Specific objectives or hypotheses	5

Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	5
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	5
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	5
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	5-6 Table 1
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	Non applicable
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	6
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	6
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	6
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	Table 2
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	7-8
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	7
Methods: Assignment of interventions (for controlled trials)			
Allocation:			5

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	5
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	5
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	5
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	5
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	5
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	6
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	5, 6
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	8-9
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	8-9
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	8-9
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	9
Methods: Monitoring			

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	2
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	5
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	11
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	2
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	2
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	2
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	5
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	Non applicable
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	5
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	1
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	9
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	Non applicable
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	2

	31b	Authorship eligibility guidelines and any intended use of professional writers	9, 11
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	Not applicable
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	See appendix
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	5

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

BMJ Open

The Role of Iodine-containing-multivitamins during Pregnancy for Children's Brain Function: Protocol of an On-going Randomized Controlled Trial- the SWIDDICH study

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2017-019945.R2
Article Type:	Protocol
Date Submitted by the Author:	20-Feb-2018

Complete List of Authors:	Manousou, Sofia; Department of Medicine; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Johansson, Birgitta; Institute of Neuroscience and Physiology, Sahlgrenska Academy, Chmielewska, Anna; Department of Clinical Sciences, Pediatrics, Umeå University; Department of Pediatrics, Medical University of Warsaw Eriksson, Janna; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Gutefeldt, Kerstin; Department of Endocrinology, University Hospital of Linköping Tornhage, Carl-Johan; Department of Pediatrics, Skaraborg's Hospital; Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg Eggertsen, Robert; Mölnlycke Health Care Center; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Malmgren, Helge; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg Hulthen, Lena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; 10Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg Domellöf, Magnus; Umea University, Department of Clinical Sciences Nystrom Filipsson, Helena; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg; Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Department of Endocrinology, Sahlgrenska University Hospital
Primary Subject Heading:	Obstetrics and gynaecology
Secondary Subject Heading:	Paediatrics, Diabetes and endocrinology, Nutrition and metabolism
Keywords:	IODINE, CHILD DEVELOPMENT, COGNITION, Thyroid disease < DIABETES & ENDOCRINOLOGY, PUBLIC HEALTH

The Role of Iodine-containing-multivitamins during Pregnancy for Children's Brain Function:
Protocol of an On-going Randomized Controlled Trial- the SWIDDICH study

Sofia Manousou[1,2], Birgitta Johansson[3], Anna Chmielewska[4,5], Janna Eriksson[1], Kerstin Gutefeldt[6], Carl-Johan Törnhage[7,8], Robert Eggertsen[1,9], Helge Malmgren[1,] Lena Hulthén[1,10], Magnus Domellöf[4] and Helena Filipsson Nyström[1,11]

[1] Institute of Medicine, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[2]Department of Medicine, Kungälv's Hospital, Kungälv, Sweden
[3]Institute of Neuroscience and Physiology, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[4] Department of Clinical Sciences, Pediatrics, Umeå University, Umeå, Sweden
[5] Department of Pediatrics, Medical University of Warsaw, Warsaw, Poland
[6] Department of Endocrinology, University Hospital of Linköping, Linköping, Sweden
[7] Department of Pediatrics, Skaraborg's Hospital, Skövde, Sweden
[8] Department of Pediatrics, Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
[9] Mölnlycke Health Care Center, Mölnlycke, Sweden,
[10]Department of Internal Medicine and Clinical Nutrition, Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden
[11]Department of Endocrinology, Sahlgrenska University Hospital, Göteborg, Sweden

Corresponding author:
Sofia Manousou
Department of Internal Medicine
Kungälv's Hospital
Lasarettsgatan
SE-442 34 Kungälv
Sweden

Tel: +46 303 98000
Fax: +46 303 15438
Email: sofia.manousou@vgregion.se

Protocol Version: 1.0, 01 September 2017.

Conflicts of Interest: none.

Abstract

Introduction

Iodine is essential for normal brain development. Moderate and severe fetal iodine deficiency result in substantial to serious developmental delay in children. Mild iodine deficiency in pregnancy is associated with neurodevelopmental deficits in the offspring, but evidence from randomized trials is lacking. The aim of the SWIDDICH study is to determine the effect of daily supplementation with 150 µg iodine during pregnancy on the offspring's neuropsychological development up to 14 years of age.

Methods and Analysis

Thyroid healthy pregnant women (n=1275: age range 18 to 40 years) at ≤12 weeks gestation, will be randomly assigned to receive multivitamin supplements containing 150 µg iodine or non-iodine containing multivitamin daily throughout pregnancy. As a primary outcome, intelligence quotient (IQ) will be measured in the offspring at 7 years (Wechsler Intelligence Scale for Children, WISC-V). As secondary outcomes, IQ will be measured at 3.5 and 14 years, psychomotor development at 18 months and 7 years, and behavior at 3.5, 7 and 14 years. Iodine status (urinary iodine concentration) will be measured during pregnancy and in the offspring at 3.5, 7 and 14 years. Thyroid function (thyroid hormones, thyroglobulin), and deiodinase type 2 polymorphisms will be measured during pregnancy and in the offspring at 7 and 14 years. Structural magnetic resonance imaging (MRI) or other relevant structural or functional brain imaging procedures will be performed in a subgroup of children at 7 and 14 years. Background and socioeconomic information will be collected at all follow-up times.

Ethics and Dissemination

This study is approved by the Ethics Committee in Göteborg, Sweden [Diary numbers: 431-12 approved 18th June 2012 (pregnancy part) and 1089-16 approved 8th February 2017 (children follow-up)]. According to Swedish regulations, dietary supplements are governed by the National Food Agency and not by the Medical Product Agency. Therefore, there is no requirement for a monitoring Committee and the National Food Agency does not perform any audits of trial conduct. The trial will be conducted in accordance with the Declaration of Helsinki. The participating sites will be contacted regarding important protocol changes, both orally and in writing, and the trial registry database will be updated accordingly. Study results will be presented at relevant conferences, and submitted to peer-reviewed journals with open access in the fields of endocrinology, pediatrics and nutrition. After the appropriate embargo period, the results will be communicated to participants, healthcare professionals at the maternal health care centers, the public, and other relevant groups, such as the national guideline group for thyroid and pregnancy and the National Food Agency.

Trial Registration Number: ClinicalTrials.gov identifier: NCT02378246, first registered 4 March 2015

Keywords: Iodine; Pregnancy; Child Development; Cognition; Thyroid Hormones

Strengths and Limitations of this study

- **Large interventional controlled trial** on iodine supplementation during pregnancy, powered to detect a difference of three IQ points in children.
- **Long observational follow-up** of the children, up to 14 years, with complex assessment of neurocognitive development.
- **Future implementation** of the study is feasible, as the intervention tablet exists on the market.
- **Lack of pure iodine** and pure placebo tablets implies careful interpretation of results.
- **Dropout rate** may be high.

BACKGROUND

Iodine Deficiency as an International Issue

Iodine is essential for the production of thyroid hormones and important for growth and brain development during fetal and early postnatal life [1]; a knowledge obtained after a long history of iodine deficiency (ID) associated disorders. For centuries, goiter with hypothyroidism, mental retardation and cretinism have been an entity. During the 1920s in the United States, Marine and Kimball performed the classic experiment of treating schoolgirls with iodine, leading to a dramatic reduction in the prevalence of goiter. Iodine prophylaxis was established in United States in 1921. After some debate, iodine prophylaxis was introduced in Switzerland in 1922, and then worldwide over the subsequent decades. The combat against severe and moderate ID has been successful in reducing the number of children with ID-caused mental retardation. However, mild ID is widely apparent, especially during pregnancy [2], when dietary iodine demand increases from 150 to 250 µg/day [3].

Iodine Status in Sweden as the Country for this Study

Before iodination of table salt in 1936, ID was common in Sweden [4]. Current iodine intake is sufficient in the general population [5, 6] and was considered adequate during pregnancy during the 1990s [7, 8]; there is no recommendation on iodine supplementation during pregnancy. However, since the 1990s, the situation may have changed due to dairy product consumption in adults being lower; milk iodine levels are lower than before [9, 10]; a reduction in salt intake is recommended for reducing the risk of hypertension; new salt forms (flake salt, gourmet salt) without iodine are popular; there is a reluctance to consume "food additives"; awareness of ID among the younger population is generally low; and, the main proportion of total salt intake (≈80%), i.e. from ready-made foods and dishes, does not provide iodine. Unless iodine is added to all salts used, the risk of decreased iodine intake is apparent, and arouses concerns, especially for pregnant women. Retrospective, local data on pregnancy highlights this assumption is realistic [11].

Iodine Deficiency during Pregnancy: Effects on the Child's Development

Severe and moderate ID leads to lower serum thyroid hormone levels and thereby to lower availability of thyroid hormones in the brain. During fetal life and early years, the growing brain is vulnerable [12, 13] and severe ID results in mental retardation in the newborn, unless the thyroid hormone is replaced [14]. In addition, an increased incidence of attention deficit hyperactivity disorders (ADHD) has been associated with mild-to-moderate ID [15].

In mild ID, thyroid hormone levels are maintained, whereas, thyroglobulin (TG) levels are increased as a biomarker of goiter. The brain's use of thyroid hormones depends on the local conversion of inactive hormone thyroxine (T4) to active hormone triiodothyronine (T3), a process mediated by deiodinase type 2 (D2) [16]. D2 is found in the hippocampi and the cerebral cortex and its activity is increased by ID, to maintain sufficient T3 levels [16, 17]. In the presence of normal thyroid hormone in blood, it is unclear how mild ID affects brain development. One theory is that this depends on deiodinases, which can change thyroid hormone signalling locally in different tissues, without affecting serum hormone concentration [16, 18].

Mild ID during pregnancy might have an impact on brain development, despite maintained normal thyroid hormone levels [19-22]. In the United Kingdom, a longitudinal study [19] found 8-year-old children have an increased risk of being in the lowest quartile of verbal IQ, if their mothers had mild ID in early pregnancy, than children of mothers with normal iodine nutrition. In a similar association study from Australia [20], mild ID was linked with lower cognitive performance in 9-year old children. Results from an observational pilot-study from Italy [21] indicate mild-to-moderate ID during fetal life affects cognitive development, especially verbal abilities, even in absence of maternal thyroid insufficiency. In Norway, a large observational study [22] found maternal iodine intake below the estimated average requirement during pregnancy was associated with reduced fine motor skills and verbal abilities and with more behavior problems at the age of 3 years.

As the randomized controlled trial (RCT) [23] evaluating 150 µg iodine/placebo in pregnant women in an iodine sufficient country was small (n=86) and lacked cognitive assessment in children, there were many expectations about the MITCH study [24]. In this trial, 832 women from Thailand and India were randomized to 200 µg iodine/placebo, and there was no difference in cognitive outcome in 5-6 year-old children. However, these results were ambiguous, for several reasons. First, the women had entered MITCH study with urinary iodine concentration (UIC) as in mild ID, but they did have a normal TG, which indicated the iodine stores in prepregnancy may have been sufficiently filled, thus, minimizing any mental effects on the children. Second, some women were already iodine sufficient at baseline [25]. Third, both intervention and placebo groups were iodine sufficient in the second and third trimesters. To prevent subnormal fetal brain development, many international authorities recommend 150 µg extra iodine/day during pregnancy, despite the lack of studies proving causality [26, 27].

Knowledge Gaps and Background to the SWIDDICH Study

There is a substantial gap in knowledge about mild ID during pregnancy and its potential negative consequences on neuropsychological development. Therefore, there is a need for a placebo-controlled trial that compares neuropsychological outcome in children exposed to mild ID during fetal life and children with normal iodine nutrition during pregnancy.

From 29 November 2012 until 1 June 2015, a pilot randomized placebo-controlled trial involving 200 pregnant women receiving a daily supplementation with either a multivitamin containing 150 µg iodine/day or a multivitamin without iodine (placebo) was conducted by our group. This study (ClinicalTrials.gov identifier: NCT02378246) aimed to evaluate the effects of iodine supplementation

on UIC and thyroid function. As the MITCH study had ambiguous results, the question if mild ID during pregnancy affects fetal brain development remains unanswered and it was evident to us that our trial needed to be expanded, to include a sufficient number of pregnant women, to enable a satisfactorily powered child follow-up regarding neuropsychological development.

There are indications [28] that UIC level during pregnancy in Sweden is lower than detected in the MITCH study, and an elevated TG is detected in early pregnancy, implying a lower iodine status at start of study. Moreover, iodine status in the third trimester is clearly lower in a local Swedish study [11] than in the placebo group in the MITCH study, indicating a different iodine situation in Sweden than in Thailand and India. Therefore, the **SW**edish **I**o**D**ine in Pregnancy and **D**evelopment **I**n **Ch**ildren (SWIDDICH) study is conducted. The hypothesis is that the use of an iodine-containing-multivitamin during pregnancy results in better cognitive development in the child than with a multivitamin without minerals (superiority trial) and this effect is stronger on verbal competence, which is in agreement with previous findings [19, 21, 22].

Objectives

The primary aim is to assess whether cognition (especially verbal competence) in children whose mothers received 150 µg iodine daily in a multivitamin during pregnancy is higher, than children whose mothers received placebo (a multivitamin without iodine) and probably remained in mild ID. The purpose is to determine whether all pregnant women who live in a country where the general population is iodine sufficient, but live in conditions that can result in mild ID during pregnancy, should be recommended extra iodine during pregnancy.

METHODS

Design of the SWIDDICH Study

This is a randomized placebo-controlled study in which children are followed-up as an observational cohort, separated into two groups by fetal iodine exposure.

Setting and Participants

Pregnant women will be recruited from more than ten maternal healthcare centers in Sweden with the aim of forming several clusters to facilitate child follow-up. The main study site will be in Gothenburg, with secondary sites in Umeå and Linköping, and other areas where maternal health care centers are recruited. At the first scheduled pregnancy visit, information about the study will be provided and written informed consent collected by the mid-wife. All procedures during pregnancy will be combined with routine pregnancy visits.

All informed consents, and blood and urine for future analyses will be sent to the main study site in Gothenburg. To promote participant retention and a complete follow-up, a contact from the study coordinator will be taken after childbirth. In addition, information will be shared with participants on the homepage https://www.gu.se/swiddich.

Inclusion

The following inclusion criteria will apply: woman aged 18 to 40 years, pregnant at 7-12 weeks, willing to refrain from iodine supplementation and take a multivitamin supplement instead, without current thyroid disease, not in another pregnancy or lactating less than 6 months before inclusion, and non-vegan.

Randomization, Allocation, Concealment and Blinding

Randomization numbers, with an allocation ratio 1:1 are prepared centrally and sent to each participating center. Consecutive numbers are used and the information regarding the study group allocation of each number stored securely at the premises of the University of Gothenburg, Sweden. Mothers are provided with a random container of pills, by either drawing a lot or blindly drawing a container. All containers are identical, with tasteless pills of the same size for both groups. Recruiting staff, study participants and those involved in laboratory work and developmental assessment are blinded to the group allocation. The code will only be broken by the central study team for data analyses before publications, but will still be blinded to all groups working with the follow-up. The code has been broken for the 200 women of the pilot study, but all (ie. study participants, psychologists and lab engineers) except the central study team are still blinded. No other interim analyses are planned.

Intervention

Women in the experimental group receive a daily multivitamin supplement containing 150 µg iodine and those in the control group receive a daily multivitamin supplement containing no iodine (the contents of the two supplements are presented in Table 1). The intervention lasts throughout pregnancy to the day of delivery. Women in both groups are recommended, as are all pregnant women in Sweden, to take extra folic acid 400 µg/day during the first trimester [29], and even extra iron, when the hemoglobin status indicates it. Therefore, women in the placebo group will be on maximum 600 µg daily folic acid supplementation, which is safely below the tolerable upper level of 1000 µg/day [30]. The folic acid and iron administrations do not interfere with the study tablet. However, the women are not permitted to take any other multivitamins besides the study supplement.

Compliance - Discontinuation

Participants are asked to bring the container with the remaining pills to the visit in the third trimester. The container is weighted and the percentage of intended doses used is calculated. Mothers who no longer want to participate in the study during pergnancy will be regarded as drop-outs and no further data will be collected. If there is disconitinuation in the children follow-up, children can come to the next visit. If the discontinuation is permanent, a registry search will still be done.

Outcomes

Outcomes in Mothers

Outcomes in mothers will be assessed in the first, second and third trimester of pregnancy. UIC and thyroid hormones will be measured in all three trimesters, and TPO-antibodies and TG in the first and third trimester.

Primary Outcome in Children

Cognition measured by intelligence quotient (total IQ) with focus on the verbal compound (verbal IQ) at 7 years is the primary outcome (Wechsler Intelligence Scale for Children, WISC-V [31]).

Secondary Outcomes in Children

Cognition measured by IQ at 3.5 years (Wechsler Preschool and Primary Scale of Intelligence, WPPSI-IV [32]) and at 14 years (Wechsler Intelligence Scale for Children, WISC-V [31] or an equivalent adequate version at the time) are secondary outcomes, together with outcomes related to psychomotor development, behavior and attention deficit hyperactivity disorder (ADHD). Psychomotor assessment will be done by the parents at 18 months (The Ages and Stages Questionnaire, ASQ-3) [33], and by a physiotherapist at 7 years (Movement Assessment Battery for Children, Movement ABC test) [34]. Behavior will be assessed through parental questionnaires, the

Child Behavior Checklist (CBCL); first at 3.5 years (CBCL 1-5) [35], then at 7 and 14 years (CBCL 6-18) [36]. At 7 and 14 years, the Nordic questionnaire 5-15 [37] will be used to assess ADHD-related symptoms.

Parents also give their consent to a registry search at 3.5, 7 and 14 years regarding the In and Out-patients registries for collecting information on medical diagnoses, the Drug registry, the Medical Brith registry, quality registries, maternal- child- and school-health care for medical and growth data, and Educational registries.

In a subgroup of children (n=200), structural brain changes will be evaluated by magnetic resonance imaging (MRI) of the brain (with a 3T Philips MR scanner) at 7 and 14 years. Automatic segmentation of the whole brain will be with Freesurfer [38] and Maper, multi-atlas propagation with enhanced registration [39]. Medio-temporal lobe (MTL) structures will be analyzed through manual segmentation with custom software developed in previous projects [40, 41]. Subregional analyses directed at regions of neurogenesis will be included. Intracranial volume measured manually will enable reliable normalization of MTL volumes. Other structural and/or functional brain imaging methods may supplement, or even replace, the described protocol, depending on the state of knowledge at the time of study.

Possible Confounding Variables and Background Information

In children, UIC will be measured from the 3.5-year visit and forward and dry blood spots will be collected for thyroid hormones, TG and deiodinases at 7 and 14 years. Background and confounding variables will be assessed at 18 months, 3.5, 7 and 14 years.

Timeframe for the Study Actions

Recruitment to the SWIDDICH study began in March 2017 and is planned to be completed in 2019. Currently 75 of 1075 pregnant women have been included. Several strategies are used to reach target sample size: a study coordinator is employed to contact maternal health care centers, and a step-wise reinbursment model is applied to the maternal health care centers in case of high recruitment rates, the National Food Agency promotes study participation in their communication with maternal health care centers, and local pediatricians are involved to faciliate the children follow-up. The follow-up of children was also offered to the families participating in the pilot study (2012-2015), before the study extension was decided. The time points for all study actions are presented in Table 2.

Patient and Public Involvement statement

Pregnant women were not involved in the planning of the study.

CONSIDERATIONS

Considerations on the Content of the Intervention and the "Placebo" Tablets

The reason for choosing iodine-containing-multivitamins instead of pure iodine tablets as the intervention is to ensure future implementation of the study is feasible. There are currently no pure iodine tablets available on the market. In the planning state of the study, discussions were initiated with pharmaceutical companies to provide pure iodine tablets and placebo, but interest was low. In the future, iodine in multivitamins will be the only available supplement source in most countries. Therefore, a multivitamin containing 150 µg iodine was chosen for the intervention and a multivitamin without minerals as the comparator.

Other components in the multivitamin products, besides iodine, may intervene with outcomes. It is proposed that vitamin B12 [42, 43] and iron [44] can have positive effects on the brain, and iron and selenium influence thyroid hormone levels [45, 46]. Iron is found in thyroperoxidase (TPO) enzyme that couples iodine to thyroglobulin. Selenium is found in deiodinases, such as D2, which converts T4 to T3, and is also an antioxidant of the thyroid gland. Sweden is a selenium deficient country [47], but it is unclear whether selenium deficiency affects cognitive outcome in humans [48]. B12 is higher in iodine-containing multivitamins where iron and selenium also are included. However, B12 content in both placebo and intervention tablets is, at least, equal to the recommended daily intake for B12; thus, B12 deficiency is not anticipated in any of the groups. In addition, the iron content is low and many pregnant Swedish women take a separate 100 mg iron supplement, which makes the 12 mg iron in the intervention tablet negligible. Iron, B12 and selenium will be measured in a subpopulation to evaluate possible group differences and contributions to thyroid metabolism.

Considerations in Choosing a Realistic Starting Point for Intervention.

Fetal brain development during the first 12 weeks is dependent on maternal T4 levels. By initiating the intervention at pregnancy week 7-12, a substantial part of the first trimester is missed. Ideally, iodine supplements may be initiated before conception. Practically, the recruitment of women who plan a pregnancy is difficult, as these women are not known by health care providers before pregnancy. One way would be through advertising in the newspaper to recruit women that are planning pregnancy. However, this would be ineffective and create selection bias, as only 50% of those who fall pregnant have planned the pregnancy, and not every woman responds to an advertisement. Women are included at the earliest possible stage and this is still far earlier that in a recent publication by Casey *et al.* [45] that included pregnant women in mean gestational week 16.6-18.0 and found negative results. The inclusion in the proposed study is similar to in the MITCH study, where women were included in gestational week 10-11 [24].

Power Calculation, Data Management, Statistical Considerations, and Authorship

The sample size needed, excluding dropouts, is calculated to 788 children (394 in each group) for an effect size of 3 IQ points with SD 15 and power 0.80. Currently, there are no similar randomized studies for power calculation. The smallest significant effect of 3 IQ points is in accordance with an observational study [19], where children of mothers with UIC< 150 µg/L during pregnancy had a 3-point lower IQ at school-age than children of mothers with normal UIC during pregnancy. This expected effect from iodine supplementation in mild iodine deficiency is also suggested by Troendle [49], where statistical considerations are discussed for the possibility that the needed placebo-controlled study is conducted. Assuming a drop out frequency of 22% during pregnancy (which is in accordance with preliminary data from the pilot study with 200 pregnant women [28]) and 20% during the children follow-up, 1263 pregnant women need to be recruited to the study. This sample size is in general agreement with Troendle [49], thus, the decision was made to try and recruit 1275 pregnant women. The dropout frequency for the children follow-up could be lower than estimated, as there are two occasions for dropout and mothers who remain in the study after the first follow-up can be assumed willing to continue the study. The power calculation assumes the use of an unpaired t-test between groups; however, more advanced analyses could decrease variance, thus, requiring a lower sample size.

The sample size will be reassessed by calculating the dropout frequency when 750 women are included and when half of the children from the first 200 included women have been invited to the

3.5-year neuropsychological evaluation. Sample size reassessment will be conducted without unblinding the study groups.

A 100% compliance to the study medication is assumed, as the results will be based on an intention to treat (ITT) analysis. Compliance is monitored to enable a per protocol analysis (only the compliant participants included) to be added. However, the ITT approach reflects the real life clinical situation, in which a certain number of patients are not compliant with the recommended treatment, and this will be the foundation for future recommendations on iodine supplementation to all pregnant women.

A separate power calculation for the MRI investigation has been done. This assumes the described protocol will be followed, and a previous study of 11-year old children has been used for guidance [50]. To detect a 5% difference with power 0.80, each group requires 60 children. As the variation in the hippocampal volumes in 7-year-old children could be slightly larger than in the previous study [50], and as dropout from the MRI at 14 years needs to be considered, 100 children will be included in each group.

Coded collected data will be entered into a database, with appropriate back-up from the university servers. Key lists will be kept safe and transfer of data to the databases will be validated by random cross-checks with the original data set. UIC analyses will be duplicated to promote validity. For further details, see ethical applications [Diary numbers: 431-12 approved 18th June 2012 (pregnancy part) and 1089-16 approved 8th February 2017 (children follow-up)] and https://clinicaltrials.gov/. All authors will have access to all data and the statisticians will have access to the data needed.

The choice of methods for comparing the main outcome between the experimental and control groups will be guided by the data distributions. In case of deviation from normality assumptions, transformations of data may be done. Non-parametric tests will be used for non-normal and ordinal data. Possible confounders, such as socioeconomic factors, other background information, gestational age, thyroid hormones, TG, deiodinase polymorphisms and UIC, will be considered in the data analyses. Repeated measurements in a mixed model (where groups are compared repeatedly at 3.5, 7 and 14 years) and within-group analyses are planned. The models will also consider dropout frequency and recruitment from different maternal health care centers, which will be used as a factor in the analysis. For all drop-outs, relevant background variables will be studied. Adjustments for bias may be performed. For non-informative drop-outs, methods for multiple imputations will be considered. A multivariate analysis with total grey, total brain volume, intracranial volume, MTL volumes and possibly other measures of brain structure and function as independent variables will be conducted. The data analyses will be undertaken by an experienced statistician. Authorship will be decided according to the Declaration of Vancouver.

MRI considerations – Where Are Changes from ID Located?

T3 receptors are distributed among all brain areas with high levels in the hippocampi and the cerebellar cortex. Rodent data indicate T3 receptors are involved in the regulation of hippocampal structure and function [51]. In the human cerebral cortex, thyroid receptors are already present in week 9 and concentrations increase up to 18 weeks of gestation [52]. Considerable amounts of D2 are also found in the cerebral cortex [53]. In the first half of pregnancy, the fetus is dependent on the mother's supply of thyroid hormones. In mild ID, the mother maintains serum thyroid hormone levels through unknown compensatory mechanisms. In the second half of a mild ID pregnancy, when the fetus partly relies on its own thyroid hormone production, the fetus will be hypothyroid, as it has not developed compensatory mechanisms and there is a lack of sufficient iodine levels transferred by the mother [53].

The description of neuropathology caused by ID is limited to few observations from adult cretins, ranging from severe cortical atrophy to almost normal appearance. In areas with endemic goiter, fetuses aborted in the second half of the pregnancy have a less differentiated cerebral cortex [53]. In rats, transient periods of thyroid hormone insufficiency during periods of cortical development affect cortical and hippocampal cytoarchitecture [53].

Human data from maternal hypothyroidism support an effect on the brain, specifically on the hippocampus [54]. These data are in line with the recent publication by Korevaar *et al* [55], who conclude the relationship of IQ with FT4 (in peripheral blood) exhibits a U-shaped configuration with lower IQ levels in both ends of the normal range. FT4 in this study [55] is also associated with total grey matter volume.

Considerations on the Neuropsychological Evaluation

Neuropsychological development can be divided into three domains: psychomotor, cognitive (IQ) and socio-emotional development (Figure 1). There are five landmark studies in the iodine field evaluating neuropsychological development in the off-spring that use neuropsychological tests: the ALSPAC (United Kingdom) [19], INMA (Spain) [56], Generation R (Netherlands) [55], MITCH (India and Thailand) [57] and Hynes *et al* (Australia) [20]. Verbal cognitive function appears to be the most susceptible subdomain for ID. In SWIDDICH, verbal cognitive function together with total IQ were chosen (as the latter is the best understood and requested) as primary outcome measurements. As cognitive testing is less valid at younger ages, verbal IQ at 7 years was chosen as the primary evaluation time point, and all three domains of neuropsychological development will be evaluated at several follow-up times.

Implications for Society and the Individual

Impaired child development increases economic burdens for society. Lowered IQ is associated with worse economic outcomes and lower lifetime earnings. Small decrements in IQ around the mean are linked to lower incomes [58, 59]. IQ may be the easiest factor to quantify, but may not be the factor with the most serious consequence for a "good life". Environmental factors, including ID, that place the nervous system at risk may affect executive functions, such as planning and initiating ideas and result in attention problems, impulsive behavior, and inability to handle stress and disappointment, and can impede success in school and in life and possibly lead to antisocial behavior [60].

If the average IQ of a population drops, the IQ distribution shifts and the number of individuals with low IQ (e.g below 75 or 85, classified as intellectually disabled) increases. In turn, this will also decrease the number of gifted and exceptionally gifted people with high IQ (e.g above 130), who may have major positive impacts on the immediate future for a company or a country. A cost-benefit analysis of iodine supplementation in mild-to moderate ID has recently proved positive [61].

Based on the dollar value in 1987 in the USA, the cost in terms of reduced income for a one point IQ reduction has been calculated to nearly 20.7 billion USD per year [62]. A 3-point decline in IQ also impacts social costs in the United States [60] and increases the risk of: poverty by 20% during the first three years; low birth weight by 12%; being a recipient of welfare by 18%; and, high school dropout by 28%. Even though a decline of a few IQ points may be small for the individual, the societal effects are considerable. As a small general risk reduction entails a large social benefit, iodine supplementation could be a cost-effective action if the main hypothesis of this study holds true.

Considerations on Possible Adverse Effects of Iodine or Placebo

Iodine supplementation may increase the frequency of post-partum thyroiditis (PPT), as iodine affects autoimmunity [63]: 10-15% of women already have post-partum thyroiditis and this number may increase slightly with iodine supplementation. As PPT is not a dangerous condition and most cases resolve spontaneously, we consider the reduced risk for subnormal brain development in a child motivates accepting the risk for PPT. In Denmark, postpartum thyroiditis was evaluated in a placebo-controlled trial in mild-moderate iodine deficiency, and treatment did not increase or worsen PPT [63].

Excess iodine intake in the mother may block thyroid function in the fetus, leading to hypothyroidism and goiter, and is associated with poorer mental and psychomotor development or behavior problems in children [22, 56, 64]. However, the risk for adverse effects of iodine supplementation is higher in cases of preconception ID, due to sudden increase of iodine intake, and should therefore not be the case in Sweden where the normal population is iodine sufficient [65].

The placebo group is at risk of iodine deficiency during pregnancy. However, as there are no current recommendations for iodine supplementation during pregnancy in Sweden, this group follows normal management.

The intervention and the comparator are dietary supplements, and the total intake of nutrients depends on the diet. Information on adverse reactions is not collected.

CONCLUSION

The aim of this paper is to describe the study protocol for the SWIDDICH research project and the considerations that led to its design. The study attempts to further understand the consequences of mild ID during pregnancy and to test whether treatment of the mothers with an iodine-containing-multivitamins improves outcome in the children. As the study is the largest of its kind, it offers the potential for influencing future recommendations on iodine supplementation with multivitamins to pregnant women living in conditions of mild ID.

ABBREVIATIONS

IQ: intelligence quotient; ADHD: attention deficit hyperactivity disorders; TG: thyroglobulin; T4: thyroxine; T3: triiodothyronine; D2: deiodinase type 2; UIC: urinary iodine concentration; RCT: randomized controlled trial; TPO: thyroperoxidase; MRI: magnetic resonance imaging; MTL: medio-temporal lobe.

Data Sharing

Data from this trial may be shared for individual data analysis in the future. A detailed plan for data sharing will be developed during the later phase of the project.

Authors' Contributions

Sofia Manousou, Birgitta Johansson, Anna Chmielewska, Janna Eriksson, Kerstin Gutefeldt, Carl-Johan Törnhage, Robert Eggertsen, Helge Malmgren, Lena Hulthén, Magnus Domellöf and Helena Filipsson Nyström contributed to the design of the SWIDDICH study. HFN wrote the first version of the manuscript and SM was responsible for pushing the work forward together with the other coworkers. All co-authors critically reviewed and approved the final version of the manuscript. SM is the guarantor.

The primary sponsor is Helena Filipsson Nyström (principal investigator), Sahlgrenska Academy and University of Göteborg and Sahlgrenska University Hospital, Göteborg, Sweden. The main study site is in Göteborg with additional sites in Umeå and Linköping.

Funding Statement
This work was supported by: the ALF agreement (grant number ALFGBG-58777, ALFGBG-717311); Regional FOU (grant number VGFOUREG-664301**);** Lilla barnets fond (grant number 20160917); Svenska Läkarsällskapet (grant number SLS-688891); Lars Hiertas Minne Foundation (grant number FO2016-0016); Formas grant (grant number 2017-0095); and, a grant from General Maternity Hospital Foundation 2017. Multivitamins for the first 200 women were provided by courtesy of Recip medical company, Solna, Sweden, but they are not involved in the study design and they do not contribute in any other way. The National Food Agency is a stakeholder in this trial. The authors of this manuscript solely contributed to the design, management, future analyses with the support of unbound statisticians, interpretation of data, writing the manuscript and decisions on where to submit. The maternal health care centers are reimbursed for the collection of patients by the principal investigator (HFN).

Acknowledgements: Elisabeth Gramatkovski, Michael Hoppe and Therese Karlsson for their invaluable help as coordinators of the study.

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Table 2. Summary of SWIDDICH study actions.

Figure 1. Neuropsychological domains. Verbal cognitive function, as a subdomain of cognition, appears the most susceptible subdomain for iodine deficiency and is the primary outcome of the SWIDDICH study at the age of 7 years.

References

[1] M.B. Zimmermann, P.L. Jooste, C.S. Pandav, Iodine-deficiency disorders, Lancet 372(9645) (2008) 1251-62.
[2] M.B. Zimmermann, M. Gizak, K. Abbott, M. Andersson, J.H. Lazarus, Iodine deficiency in pregnant women in Europe, The lancet. Diabetes & endocrinology 3(9) (2015) 672-4.
[3] W.H.O. United Nations Children's Fund & International Council for the Control of Iodine Deficiency Disorders, Assessment of iodine deficiency disorders and monitoring their elimination. A guide for programme managers, 3:e edition, Geneva, 2007.
[4] H.F. Nystrom, S. Jansson, G. Berg, Incidence Rate and Clinical features of Hyperthyroidism in a long-term iodine sufficient area of Sweden (Gothenburg) 2003-2005, Clin Endocrinol (Oxf) (2013).
[5] M. Andersson, G. Berg, R. Eggertsen, H. Filipsson, E. Gramatkovski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Adequate iodine nutrition in Sweden: a cross-sectional national study of urinary iodine concentration in school-age children, Eur. J. Clin. Nutr. 63(7) (2009) 828-34.
[6] H. Filipsson Nystrom, M. Andersson, G. Berg, R. Eggertsen, E. Gramatkowski, M. Hansson, L. Hulthen, M. Milakovic, E. Nystrom, Thyroid volume in Swedish school children: a national, stratified, population-based survey, Eur. J. Clin. Nutr. 64(11) (2010) 1289-95.
[7] B. Elnagar, A. Eltom, L. Wide, M. Gebre-Medhin, F.A. Karlsson, Iodine status, thyroid function and pregnancy: study of Swedish and Sudanese women, Eur. J. Clin. Nutr. 52(5) (1998) 351-5.
[8] A. Eltom, B. Elnagar, M. Elbagir, M. Gebre-Medhin, Thyroglobulin in serum as an indicator of iodine status during pregnancy, Scand J Clin Lab Invest. 60(1) (2000) 1-7.
[9] H. Lindmark Månsson, the Swedish Milk composition Svensk Mjölk (Swedish milk) 2010.
[10] H. Lindmark Månsson, Den svenska mejerimjölkens sammansättning 2009, Svensk Mjök (Swedish Milk), 2012.
[11] M. Granfors, M. Andersson, S. Stinca, H. Akerud, A. Skalkidou, I. Sundstrom Poromaa, A.K. Wikstrom, H. Filipsson Nystrom, Iodine deficiency in a study population of pregnant women in Sweden, Acta Obstet. Gynecol. Scand. (2015).
[12] M.B. Zimmermann, The adverse effects of mild-to-moderate iodine deficiency during pregnancy and childhood: a review, Thyroid 17(9) (2007) 829-35.
[13] P.N. Taylor, O.E. Okosieme, C.M. Dayan, J.H. Lazarus, Therapy of endocrine disease: Impact of iodine supplementation in mild-to-moderate iodine deficiency: systematic review and meta-analysis, Eur. J. Endocrinol. 170(1) (2014) R1-R15.
[14] J.B. Stanbury, A.M. Ermans, B.S. Hetzel, E.A. Pretell, A. Querido, Endemic goitre and cretinism: public health significance and prevention, WHO Chron. 28(5) (1974) 220-8.
[15] F. Vermiglio, V.P. Lo Presti, M. Moleti, M. Sidoti, G. Tortorella, G. Scaffidi, M.G. Castagna, F. Mattina, M.A. Violi, A. Crisa, A. Artemisia, F. Trimarchi, Attention deficit and hyperactivity disorders in the offspring of mothers exposed to mild-moderate iodine deficiency: a possible novel iodine deficiency disorder in developed countries, J. Clin. Endocrinol. Metab. 89(12) (2004) 6054-60.
[16] R. Peeters, C. Fekete, C. Goncalves, G. Legradi, H.M. Tu, J.W. Harney, A.C. Bianco, R.M. Lechan, P.R. Larsen, Regional physiological adaptation of the central nervous system deiodinases to iodine deficiency, Am. J. Physiol. Endocrinol. Metab. 281(1) (2001) E54-61.
[17] N. Markova, A. Chernopiatko, C.A. Schroeter, D. Malin, A. Kubatiev, S. Bachurin, J. Costa-Nunes, H.M. Steinbusch, T. Strekalova, Hippocampal gene expression of deiodinases 2 and 3 and effects of 3,5-diiodo-L-thyronine T2 in mouse depression paradigms, BioMed research international 2013 (2013) 565218.
[18] F. Courtin, H. Zrouri, A. Lamirand, W.W. Li, G. Mercier, M. Schumacher, C.L. Goascogne, M. Pierre, Thyroid hormone deiodinases in the central and peripheral nervous system, Thyroid 15(8) (2005) 931-42.
[19] S.C. Bath, C.D. Steer, J. Golding, P. Emmett, M.P. Rayman, Effect of inadequate iodine status in UK pregnant women on cognitive outcomes in their children: results from the Avon Longitudinal Study of Parents and Children (ALSPAC), Lancet 382(9889) (2013) 331-7.
[20] K.L. Hynes, P. Otahal, I. Hay, J.R. Burgess, Mild iodine deficiency during pregnancy is associated with reduced educational outcomes in the offspring: 9-year follow-up of the gestational iodine cohort, J. Clin. Endocrinol. Metab. 98(5) (2013) 1954-62.

[21] M. Moleti, F. Trimarchi, G. Tortorella, A. Candia Longo, G. Giorgianni, G. Sturniolo, A. Alibrandi, F. Vermiglio, Effects of Maternal Iodine Nutrition and Thyroid Status on Cognitive Development in Offspring: A Pilot Study, Thyroid 26(2) (2016) 296-305.
[22] M.H. Abel, I.H. Caspersen, H.M. Meltzer, M. Haugen, R.E. Brandlistuen, H. Aase, J. Alexander, L.E. Torheim, A.L. Brantsaeter, Suboptimal Maternal Iodine Intake Is Associated with Impaired Child Neurodevelopment at 3 Years of Age in the Norwegian Mother and Child Cohort Study, J. Nutr. 147(7) (2017) 1314-1324.
[23] F. Brucker-Davis, P. Panaia-Ferrari, J. Gal, P. Fenichel, S. Hieronimus, Iodine Supplementation throughout Pregnancy Does Not Prevent the Drop in FT4 in the Second and Third Trimesters in Women with Normal Initial Thyroid Function, European thyroid journal 2(3) (2013) 187-94.
[24] S. Gowachirapant, N. Jaiswal, A. Melse-Boonstra, V. Galetti, S. Stinca, I. Mackenzie, S. Thomas, T. Thomas, P. Winichagoon, K. Srinivasan, M.B. Zimmermann, Effect of iodine supplementation in pregnant women on child neurodevelopment: a randomised, double-blind, placebo-controlled trial, The lancet. Diabetes & endocrinology 5(11) (2017) 853-863.
[25] S.C. Bath, Iodine supplementation in pregnancy in mildly deficient regions, The lancet. Diabetes & endocrinology 5(11) (2017) 840-841.
[26] E.K. Alexander, E.N. Pearce, G.A. Brent, R.S. Brown, H. Chen, C. Dosiou, W.A. Grobman, P. Laurberg, J.H. Lazarus, S.J. Mandel, R.P. Peeters, S. Sullivan, 2017 Guidelines of the American Thyroid Association for the Diagnosis and Management of Thyroid Disease During Pregnancy and the Postpartum, Thyroid 27(3) (2017) 315-389.
[27] L. De Groot, M. Abalovich, E.K. Alexander, N. Amino, L. Barbour, R.H. Cobin, C.J. Eastman, J.H. Lazarus, D. Luton, S.J. Mandel, J. Mestman, J. Rovet, S. Sullivan, Management of thyroid dysfunction during pregnancy and postpartum: an Endocrine Society clinical practice guideline, J. Clin. Endocrinol. Metab. 97(8) (2012) 2543-65.
[28] F. Pacini, B. Nygaard, Abstracts, European thyroid journal 5(Suppl 1) (2016) 57-176.
[29] Nordic Council of Ministers, Nordic Nutrition Recommendations 2012 : integrating nutrition and physical activity, Nordic Council of Minsters, Copenhagen, 2014.
[30] European Food Safety Authority, Folic Acid: an update on scientific developments, 2009. https://www.livsmedelsverket.se/globalassets/matvanor-halsa-miljo/kostrad-matvanor/gravida/folic-acid---an-update-on-scientific-developments.-rapport.-efsa-european-food-safety-authority.-2009..pdf?amp;epslanguage=sv.
[31] D. Wechsler, Wechsler Intelligence Scale for Children – Fifth Edition (WISC-V), Swedish version, Stockholm, Sweden, 2016.
[32] D. Wechsler, Wechsler Preschool and Primary Scale of Intelligence – Fourth Edition (WPPSI-IV) (Swedish version), Stockholm, Sweden, 2014.
[33] T.E. Squires J, Bricker D, Potter L, ASQ-3 User's Guide. 3rd ed. , Baltimore, MD, US, 2009.
[34] S.E. Henderson, Sugden, D. A., & Barnett, A. L. , Movement assessment battery for children [examiner's manual] (2nd ed.). , London, UK, 2007.
[35] R.L. Achenbach TM, Manual for the ASEBA Preschool Forms & Profiles. , University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2000.
[36] T.M. Achenbach, Rescorla, L.A. , Manual for the ASEBA School-Age Forms & Profiles, University of Vermont, Research Center for Children, Youth, and Families, Burlington, Vermont, US, 2001.
[37] B. Kadesjo, L.O. Janols, M. Korkman, K. Mickelsson, G. Strand, A. Trillingsgaard, C. Gillberg, The FTF (Five to Fifteen): the development of a parent questionnaire for the assessment of ADHD and comorbid conditions, Eur. Child Adolesc. Psychiatry 13 Suppl 3 (2004) 3-13.
[38] B. Fischl, D.H. Salat, E. Busa, M. Albert, M. Dieterich, C. Haselgrove, A. van der Kouwe, R. Killiany, D. Kennedy, S. Klaveness, A. Montillo, N. Makris, B. Rosen, A.M. Dale, Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain, Neuron 33(3) (2002) 341-55.
[39] R.A. Heckemann, S. Keihaninejad, P. Aljabar, D. Rueckert, J.V. Hajnal, A. Hammers, Improving intersubject image registration using tissue-class information benefits robustness and accuracy of multi-atlas based anatomical segmentation, Neuroimage 51(1) (2010) 221-7.

[40] C. Eckerstrom, E. Olsson, M. Borga, S. Ekholm, S. Ribbelin, S. Rolstad, G. Starck, A. Edman, A. Wallin, H. Malmgren, Small baseline volume of left hippocampus is associated with subsequent conversion of MCI into dementia: the Goteborg MCI study, J. Neurol. Sci. 272(1-2) (2008) 48-59.
[41] E. Olsson, C. Eckerstrom, G. Berg, M. Borga, S. Ekholm, G. Johannsson, S. Ribbelin, G. Starck, A. Wysocka, E. Lofdahl, H. Malmgren, Hippocampal volumes in patients exposed to low-dose radiation to the basal brain. A case-control study in long-term survivors from cancer in the head and neck region, Radiat. Oncol. 7 (2012) 202.
[42] V. Bhate, S. Deshpande, D. Bhat, N. Joshi, R. Ladkat, S. Watve, C. Fall, C.A. de Jager, H. Refsum, C. Yajnik, Vitamin B12 status of pregnant Indian women and cognitive function in their 9-year-old children, Food Nutr Bull 29(4) (2008) 249-54.
[43] M.M. Black, Effects of vitamin B12 and folate deficiency on brain development in children, Food Nutr Bull 29(2 Suppl) (2008) S126-31.
[44] E.L. Prado, K.G. Dewey, Nutrition and brain development in early life, Nutr. Rev. 72(4) (2014) 267-84.
[45] M. Ventura, M. Melo, F. Carrilho, Selenium and Thyroid Disease: From Pathophysiology to Treatment, Int. J. Endocrinol. 2017 (2017) 1297658.
[46] S. Hu, M.P. Rayman, Multiple Nutritional Factors and the Risk of Hashimoto's Thyroiditis, Thyroid 27(5) (2017) 597-610.
[47] O. Selinus, Medical geology: Iodine - a classical element (jod ett klassiskt element), Medical geololgy (Medicinsk Geologi), Authors and Studentlitteratur2010, pp. 387-400.
[48] H.M. Skroder, J.D. Hamadani, F. Tofail, L.A. Persson, M.E. Vahter, M.J. Kippler, Selenium status in pregnancy influences children's cognitive function at 1.5 years of age, Clin. Nutr. 34(5) (2015) 923-30.
[49] J.F. Troendle, Statistical design considerations applicable to clinical trials of iodine supplementation in pregnant women who may be mildly iodine deficient, Am. J. Clin. Nutr. 104 Suppl 3 (2016) 924S-7S.
[50] L. Bunketorp Kall, H. Malmgren, E. Olsson, T. Linden, M. Nilsson, Effects of a Curricular Physical Activity Intervention on Children's School Performance, Wellness, and Brain Development, J. Sch. Health 85(10) (2015) 704-13.
[51] A. Guadano-Ferraz, R. Benavides-Piccione, C. Venero, C. Lancha, B. Vennstrom, C. Sandi, J. DeFelipe, J. Bernal, Lack of thyroid hormone receptor alpha1 is associated with selective alterations in behavior and hippocampal circuits, Mol. Psychiatry 8(1) (2003) 30-8.
[52] B. Ferreiro, J. Bernal, C.G. Goodyer, C.L. Branchard, Estimation of nuclear thyroid hormone receptor saturation in human fetal brain and lung during early gestation, J. Clin. Endocrinol. Metab. 67(4) (1988) 853-6.
[53] G.M. de Escobar, M.J. Obregon, F.E. del Rey, Iodine deficiency and brain development in the first half of pregnancy, Public Health Nutr. 10(12A) (2007) 1554-70.
[54] K.A. Willoughby, M.P. McAndrews, J.F. Rovet, Effects of maternal hypothyroidism on offspring hippocampus and memory, Thyroid 24(3) (2014) 576-84.
[55] T.I. Korevaar, R. Muetzel, M. Medici, L. Chaker, V.W. Jaddoe, Y.B. de Rijke, E.A. Steegers, T.J. Visser, T. White, H. Tiemeier, R.P. Peeters, Association of maternal thyroid function during early pregnancy with offspring IQ and brain morphology in childhood: a population-based prospective cohort study, The lancet. Diabetes & endocrinology 4(1) (2016) 35-43.
[56] M. Rebagliato, M. Murcia, M. Alvarez-Pedrerol, M. Espada, A. Fernandez-Somoano, N. Lertxundi, E.M. Navarrete-Munoz, J. Forns, A. Aranbarri, S. Llop, J. Julvez, A. Tardon, F. Ballester, Iodine supplementation during pregnancy and infant neuropsychological development. INMA Mother and Child Cohort Study, Am. J. Epidemiol. 177(9) (2013) 944-53.
[57] A. Melse-Boonstra, S. Gowachirapant, N. Jaiswal, P. Winichagoon, K. Srinivasan, M.B. Zimmermann, Iodine supplementation in pregnancy and its effect on child cognition, J. Trace Elem. Med. Biol. 26(2-3) (2012) 134-6.
[58] J. Schwartz, Societal benefits of reducing lead exposure, Environ. Res. 66(1) (1994) 105-24.
[59] D.S. Salkever, Updated estimates of earnings benefits from reduced exposure of children to environmental lead, Environ. Res. 70(1) (1995) 1-6.

[60] T. Muir, M. Zegarac, Societal costs of exposure to toxic substances: economic and health costs of four case studies that are candidates for environmental causation, Environ. Health Perspect. 109 Suppl 6 (2001) 885-903.
[61] M. Monahan, K. Boelaert, K. Jolly, S. Chan, P. Barton, T.E. Roberts, Costs and benefits of iodine supplementation for pregnant women in a mildly to moderately iodine-deficient population: a modelling analysis, The lancet. Diabetes & endocrinology 3(9) (2015) 715-22.
[62] J. Schwartz, Low-level lead exposure and children's IQ: a meta-analysis and search for a threshold, Environ. Res. 65(1) (1994) 42-55.
[63] S.B. Nohr, A. Jorgensen, K.M. Pedersen, P. Laurberg, Postpartum thyroid dysfunction in pregnant thyroid peroxidase antibody-positive women living in an area with mild to moderate iodine deficiency: is iodine supplementation safe?, J. Clin. Endocrinol. Metab. 85(9) (2000) 3191-8.
[64] M. Murcia, M. Rebagliato, C. Iniguez, M.J. Lopez-Espinosa, M. Estarlich, B. Plaza, C. Barona-Vilar, M. Espada, J. Vioque, F. Ballester, Effect of iodine supplementation during pregnancy on infant neurodevelopment at 1 year of age, Am. J. Epidemiol. 173(7) (2011) 804-12.
[65] M. Moleti, B. Di Bella, G. Giorgianni, A. Mancuso, A. De Vivo, A. Alibrandi, F. Trimarchi, F. Vermiglio, Maternal thyroid function in different conditions of iodine nutrition in pregnant women exposed to mild-moderate iodine deficiency: an observational study, Clin. Endocrinol. (Oxf.) 74(6) (2011) 762-8.

Table 1. Multivitamin with iodine (intervention) and multivitamin without iodine ("placebo"): table of contents.

Intervention (iodine 150 microgram) **Commercial name: MITT VAL VEGETARIAN**	**Placebo (no iodine)** **Commercial name: ENOMDAN**
B2 1.4 mg (87%)* B12 15 microgram (750%) Iron 12 mg (30%) Zink 12 mg (133%) Iodine 150 microgram (85%) Selenium 50 microgram (71%) Calcium 250mg mg (28%)	Vitamin A 400 microgram (50%) Vitamin B1 1.4 mg (93%) Vitamin B2 1.7 mg (106%) Vitamin B6 1.8 mg (128%) Vitamin B12 3 microgram (150%) Vitamin C 60 mg (70%) Vitamin D 5 microgram (50%) Vitamin E 10 mg (100%) Niacin 19 mg (111%) Folic acid 200 microgram (50%)

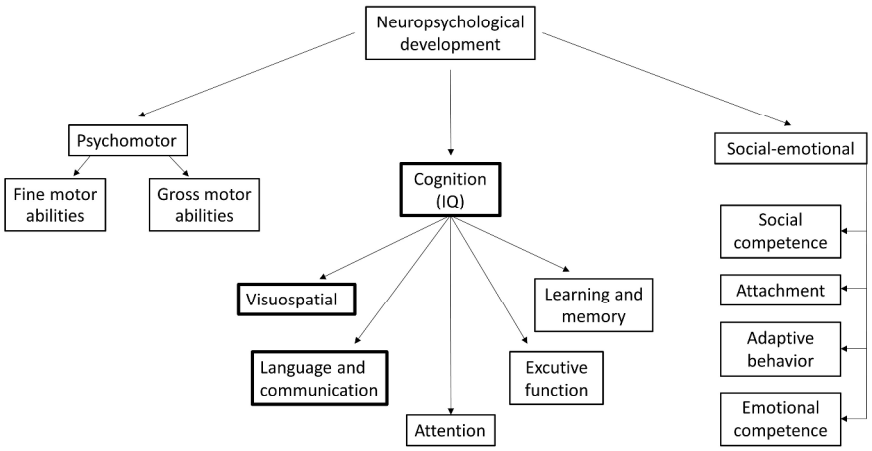
* (%RDI) = % of Recommended Daily Intake during pregnancy[29]

Table 2. Summary of SWIDDICH study actions.

	Pregnancy				Child follow-up			
TIMEPOINT	**First pregnancy visit (<12 weeks)**	**Week 7 – 12**	**Week 25-28**	**Week 34-38**	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
ENROLMENT:								
Information given	X							
Eligibility screen	X							
Informed consent	X							
Allocation		X						
INTERVENTION ***Iodine 150 µg or Placebo in multivitamins***		◆———	———	———◆				
ASSESSMENTS:	**First pregnancy visit (<12 weeks)**	**Week 7 – 12**	**Week 25-28**	**Week 34-38**	**18 mo**	**3.5 ys**	**7 ys**	**14 ys**
Urinary iodine concentration		X	X	X		X	X	X
Thyroid function*		X	X	X			X	X
Milk iodine concentration								
COGNITION: IQ						X WPPSI	X WISC	X WISC
Behavior						X CBCL	X CBCL Nordic 5-15	X CBCL Nordic 5-15
Psychomotor development					X ASQ-3		X Mov ABC	
Brain MRI *(subgroup)*							X	X
BACKGROUND INFORMATION:								
EUthyroid SES questionnaire *adults*					X	X	X	X
EUthyroid SES questionnaire *children*								X
Own questionnaire		X	X	X	X	X	X	X

mo: months; ys: years; WPPSI: Wechsler Preschool and Primary Scale of Intelligence; WISC: Wechsler Intelligence Scale for Children; ASQ-3: The Ages and Stages Questionnaire; Mov ABC: Movement Assessment Battery for Children; EUthyroid SES questionnaire: Socioeconomic Status questionnaire, validated by EUthyroid foundation

* FT4 TSH, thyroglobulin: serum sampling during pregnancy and dry blood spot sampling during children follow-up



Neuropsychological domains. Verbal cognitive function, as a subdomain of cognition, appears the most susceptible subdomain for iodine deficiency and is the primary outcome of the SWIDDICH study at the age of 7 years.

338x190mm (300 x 300 DPI)

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Page
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	2
	2b	All items from the World Health Organization Trial Registration Data Set	1, 2, 4-7, 11
Protocol version	3	Date and version identifier	1
Funding	4	Sources and types of financial, material, and other support	11
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1, 11
	5b	Name and contact information for the trial sponsor	1
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	11
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	5
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	3-4
	6b	Explanation for choice of comparators	7
Objectives	7	Specific objectives or hypotheses	5

Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	5
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	5
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	5
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	5-6 Table 1
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	Non applicable
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	6
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	6
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	6
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	Table 2
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	7-8
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	7
Methods: Assignment of interventions (for controlled trials)			
Allocation:			5

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	5
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	5
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	5
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	5
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	5
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	6
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	5, 6
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	8-9
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	8-9
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	8-9
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	9
Methods: Monitoring			

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	2
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	5
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	11
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	2
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	2
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	2
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	5
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	Non applicable
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	5
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	1
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	9
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	Non applicable
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	2

	31b	Authorship eligibility guidelines and any intended use of professional writers	9, 11
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	Not applicable
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	See appendix
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	5

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.