Body iron is associated with cognitive executive planning function in college women

Cynthia A. Blanton[1]*†, Michael W. Green[2] and Mary J. Kretsch[1]‡

[1]*USDA, ARS, Western Human Nutrition Research Center, University of California, Davis, CA, USA*
[2]*Nutrition and Behaviour Laboratory, School of Life and Health Sciences, Aston University, Birmingham, UK*

Abstract

Evidence of the relationship between altered cognitive function and depleted Fe status is accumulating in women of reproductive age but the degree of Fe deficiency associated with negative neuropsychological outcomes needs to be delineated. Data are limited regarding this relationship in university women in whom optimal cognitive function is critical to academic success. The aim of the present study was to examine the relationship between body Fe, in the absence of Fe-deficiency anaemia, and neuropsychological function in young college women. Healthy, non-anaemic undergraduate women (n 42) provided a blood sample and completed a standardised cognitive test battery consisting of one manual (Tower of London (TOL), a measure of central executive function) and five computerised (Bakan vigilance task, mental rotation, simple reaction time, immediate word recall and two-finger tapping) tasks. Women's body Fe ranged from $-4\cdot2$ to $8\cdot1$ mg/kg. General linear model ANOVA revealed a significant effect of body Fe on TOL planning time ($P=0\cdot002$). Spearman's correlation coefficients showed a significant inverse relationship between body Fe and TOL planning time for move categories 4 ($r-0\cdot39$, $P=0\cdot01$) and 5 ($r-0\cdot47$, $P=0\cdot002$). Performance on the computerised cognitive tasks was not affected by body Fe level. These findings suggest that Fe status in the absence of anaemia is positively associated with central executive function in otherwise healthy college women.

Key words: Iron: Cognition: College women

Fe deficiency affects individuals of all ages and classes but to a larger extent infants, children and women of reproductive age[1,2]. Evidence of the relationship between Fe status and neuropsychological function in young women is beginning to accumulate[3–9]. The WHO estimates that anaemia, the most severe form of Fe deficiency, affects 30 % of women of reproductive age[10], and in the USA, recent reports have indicated that approximately 15 % of females aged 20–49 years are Fe-deficient[11]. These data emphasise the need to give attention to Fe deficiency in developed as well as developing countries. The present study focuses on women of childbearing age, in particular women in college. The prevalence of Fe deficiency in college women has been reported at 30–51 % when using a serum ferritin cut-off of $<20\,\mu g/l$[5,12,13] and 28–50 % when using criteria of serum ferritin of $<15\,\mu g/l$[14–16], or absent or trace bone marrow Fe[17]. Fe-deficiency anaemia has been observed in 16–18 % of female university undergraduate students[18,19]. This population is of particular concern considering the cognitive demands of higher education.

Fe deficiency manifests as alterations in cognitive function, behaviour and mood[20–22]. Despite the high rate of Fe deficiency in women of reproductive age and evidence showing that maternal deficiency affects the fetus and child[23,24], large gaps exist in this area of investigation. Additional studies in women of reproductive age are needed to elucidate the degree of Fe deficiency that is associated with negative neuropsychological outcomes. The impact of mild or moderate Fe deficiency on cognitive function has not been clearly described. Studies to date in women have found significant relationships only when Fe-deficiency anaemia was present. The present study explores the relationship between body Fe status, in the absence of anaemia, and neuropsychological function in young college women.

Experimental methods

Study design

The present study was performed at the United States Department of Agriculture, Western Human Nutrition Research

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Center located at the campus of the University of California, Davis, California. The study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human subjects were approved by the university's Institutional Review Board of the Office of Human Research Protection. Written informed consent was obtained from all participants. The study comprised one laboratory visit, approximately 2 h in duration, preceded by two mornings of at-home saliva collections. On the test morning, women arrived at the laboratory after a 12 h fast and provided samples of blood and urine before height and weight were measured. Blood was tested as described below; urine was tested to confirm the absence of pregnancy. Participants then consumed completely a controlled snack of 18 g whole-grain crispbread crackers (Wasa), 100 g 2 %-fat cottage cheese and 500 ml bottled water. The snack was provided as a standardised countermeasure to negative effects of fasting on cognitive function[25]. Following snack consumption, participants completed written questionnaires. At 60 min after completion of the snack, a battery of standardised cognitive tests was administered, lasting approximately 60 min. Saliva samples were collected immediately preceding cognitive testing and following completion of the sustained attention cognitive task.

Participants

Healthy, female undergraduate college students enrolled at the University of California, Davis were recruited over an 18-month period using printed and electronic advertisements. A telephone screening questionnaire was administered on initial contact and qualified women proceeded to an in-laboratory health screening that included anthropometric measurements, blood and urine testing, and self-administered questionnaires. Exclusion criteria included the following: age <19 or >30 years; BMI >29·9 kg/m²; current pregnancy or pregnancy within the previous year; current lactation; hormonal contraceptive use; smoking; regular high-intensity exercise level; dieting for weight loss; recent history of eating disorders; inflammatory or endocrine disorders; chronic infections; anaemia (Hb <120 g/l); vitamin B_{12} or folic acid deficiency; haemolytic anaemia; polycythaemia; haemoglobinopathies; race other than Caucasian; excess alcohol consumption or use of recreational drugs, prescription drugs or herbal preparations that could interfere with Fe absorption and/or affect mental performance. Caucasian, non-obese participants were selected to avoid introducing the confounding effects of race and obesity, respectively, on haematology[26,27]. Women were selected because Fe deficiency is less prevalent in men[5] and were tested during the luteal phase of their menstrual cycle in order to control for cycle-related changes in cognitive function[28] and haematology[29]. Women's progression through the recruitment process is depicted in Fig. 1. The final number of research participants was forty-two.

Data collection

For the details on haematological and salivary methods, anthropometric measurements, and questionnaires, see Supplementary Material available online at http://www.journals.cambridge.org/bjn

Assessment of iron

Blood from overnight-fasted participants was analysed for complete blood count, serum ferritin and serum soluble transferrin receptor. Serum Fe was measured to test for associations with cognitive function that we have previously reported[8]. Body Fe was calculated using the following equation: body Fe (mg/kg body weight) = − (log(transferrin receptor/ferritin) − 2·8229)/0·1207[30]. Body Fe was selected as the indicator of Fe status in the present study due to its ability to detect slight differences in Fe levels in non-anaemic individuals that could otherwise be missed by methods of classification utilising multiple defined cut-off values[31]. Furthermore, using body Fe as an assessment measure is consistent with the recent adoption of body Fe as an Fe status indicator for the US National Health and Nutrition Examination Surveys[32,33] and its recommendation for use by the WHO[34].

Assessment of cognitive performance

Each participant was tested individually, in a session lasting approximately 1 h, under uniform lighting conditions inside a soundproof chamber. A research staff member (C. A. B.) trained in administering the standardised cognitive battery and blinded to participants' Fe status conducted all testing. The staff member was present with the participant in the
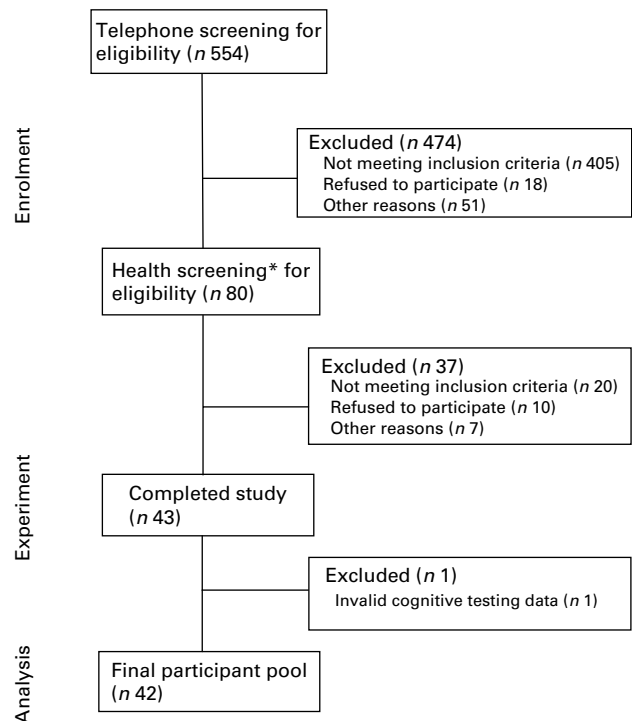

Fig. 1. Flow diagram of volunteer progression through recruitment and participation. * Health screening consisted of measurements of height, weight, blood pressure, pulse, temperature, a fasting blood draw, a spot urine collection, and completion of six written questionnaires related to health history, physical activity, psychological state and eating behaviour.

chamber throughout testing. The mostly computerised but standardised assessment battery, described in Green *et al.*[35], measures multiple modalities of cognitive function and is comprised of the following tasks, presented in the same order across the sessions: planning and working memory (Tower of London (TOL)); sustained attention (Bakan vigilance task); spatial reasoning ability (mental rotation); simple reaction time; immediate verbal memory (verbal free recall); motor performance (two-finger tapping task). A manual version of the TOL task[36] was the only non-computerised cognitive test used in the present study.

Tower of London. The TOL task was comprised of three movable discs (one green, one white and one black) that sit on three pegs mounted on a wooden board. Participants were required to move the discs from a standard initial position to a goal position within a set number of moves. There were two goal positions that could be achieved in a minimum of two moves, two that could be achieved in three moves, four that could be achieved in four moves, and four that could be achieved in five moves. Participants were instructed to plan mentally the entire sequence of moves before executing the sequence. A stopwatch was used to measure planning and total time for each trial. Planning time began when the participant was presented with the diagrammed goal position and ended when the participant first touched a disc. Total time encompassed planning through achievement of the goal position. Participants successfully completed three practice trials before beginning the experimental trials.

Bakan task. This sustained attention task was a 6 min visual analogue of the procedure developed by Bakan[37] in which participants were presented with a continuous stream of single numbers (1–9) that appeared one at a time in the centre of the computer screen. Participants were asked to press a response key as soon as they detected a target sequence of three consecutive odd or even numbers.

Mental rotation. This task assessed the visual–spatial component of working memory. Participants were presented with a number of pairs of random geometric forms, with each pair being presented simultaneously. The task was to mentally rotate the form on the right side of the screen to make it identical in orientation to the form on the left side of the screen. Participants then were to decide whether the form on the right side was a duplicate or a mirrored image of the form on the left. They were instructed to press the 1 or 2 key on a keyboard to indicate duplicate or mirrored image, respectively.

Simple reaction time. Participants were presented with 100 reaction time trials with instructions to press the space bar on a keyboard as quickly as they detected a single star appearing in the centre of the computer screen.

Verbal free recall. Participants were presented with two lists of twenty words each, one list being presented at 1 s per word and the other at 2 s per word. Immediately after the presentation of each list, participants were allowed 4 min to recall as many words as possible.

Two-finger tapping. This task of motor speed required participants to alternatively tap the 1 and 2 keys of the keyboard as quickly as possible, using the index and second finger of their dominant hand.

Assessment of covariates

Serum C-reactive protein was measured to test for inflammatory status, which can alter Fe status indicators[38]. Serum plasma Zn was measured to test for associations with cognitive function that we have previously reported[39]. Salivary cortisol was measured to assess the association of corticosteroid levels, an index of stress, with cognitive performance[35]. Saliva samples were obtained using the Salivette device (Sarstedt), consisting of a cotton swab and plastic vial. On each of the 2 d preceding the laboratory appointment, participants were instructed to collect two saliva samples at home: the first upon waking from night-time sleep and the second, 30 min after collecting the first sample. During the laboratory appointment, two additional saliva samples were collected: the first immediately preceding cognitive testing and the second, 30 min after completion of the Bakan task. The mean cortisol level of samples from days 1 and 2 preceding cognitive testing was calculated for each time point for each participant.

Participants completed five questionnaires during the 60 min interim between snack consumption and cognitive testing: the Beck Depression Inventory-II[40]; the Perceived Stress Scale (PSS10)[41]; the State-Trait Anxiety Inventory[42]; the Block Food-Frequency Questionnaire (Block 98 FFQ)[43,44]; a menstrual information questionnaire. Questionnaires were self-administered but checked for completeness upon submission.

Statistical analyses

Statistical analyses were performed using IBM SPSS version 19.0 (2010). The logarithm (base 10) of planning time for the four levels (two, three, four and five moves) of the TOL task was compared using a general linear model. Log-transformed data were used since the raw data were significantly right-skewed. The continuous between-subjects factor was body Fe. The within-subjects factors were the four TOL levels and the interaction of TOL level and body Fe. This type of analysis is a repeated-measures ANOVA with a covariate allowing for different slopes for the different levels of the within-subjects factor. For tests of correlation, variables for Fe status and the computerised tasks were examined for normality using the Kolmogorov–Smirnov test with the Lilliefors correction. Pearson's product–moment correlation coefficient is reported when both variables fit a normal distribution. Spearman's rank correlation coefficient is reported when either one or both of the variables do not fit a normal distribution. Correlations between salivary cortisol levels and each of the following were also examined: body Fe; cognitive test scores; behavioural questionnaire scores. The significance level was set at $P < 0.05$ and quoted levels are two-sided.

Results

Demographic and haematological measurements

Body Fe ranged from −4·2 to 8·1 mg/kg (Table 1), with five women (12 % of the participants) having values <0 mg/kg. Of the study participants, seven women, including the five with negative body Fe, had abnormal serum ferritin

Table 1. Sample characteristics (*n* 42)

(Mean values, standard deviations, medians, quartiles and ranges)

	Mean	SD	Range	Median	Q1	Q3
Age (years)	21·4	1·6	18·6−28·2	20·9	20·3	22·2
BMI (kg/m^2)	22·4	2·5	19·0−29·2	22·0	20·5	24·0
Body Fe (mg/kg)	3·2	2·9	−4·2−8·1	3·8	1·2	5·5
Ferritin (ng/ml)	27·1	17·0	6·0−92·1	24·0	14·1	33·0
Transferrin receptor (mg/l)	6·6	2·6	3·4−17·6	5·6	4·9	7·6
Hb (g/l)	132	7	120−151	133	126	137
Serum Fe (μg/l)	898	396	293−2045	863	569	1015
Plasma Zn (μg/l)	8	1	7−10	8	7	9
C-reactive protein (mg/l)	0·2	0·4	0·0−2·8	0·1	0·0	0·1
BDI-II score[40,63]	3·1	3·1	0−12	2·0	0	5
STAI state pre-test*	26·9	6·3	20−48	25·0	22	30
STAI state post-Bakan	30·1	8·0	20−50	29·5	23	35
STAI trait	32·3	7·4	22−49	31·5	26	38
Perceived Stress Scale[41]	10·6	4·7	3−19	10·5	7	14

Q1, quartile 1; Q3, quartile 3; BDI-II, Beck Depression Inventory-II; STAI, State-Trait Anxiety Inventory.

* The STAI[42] consists of separate self-report scales that measure the intensity and frequency of state (at this moment) and trait (general) anxiety. Both scales were administered before cognitive testing and the state scale was repeated 30 min after completion of the Bakan task.

$(<12 \, \mu g/l)$[45] and eight women had abnormal transferrin receptor $(>8·3 \, mg/l)$ levels. Serum Fe was below normal $(<500 \, \mu g/l)$[45] in four women, two of whom had negative body Fe. The results are shown in Table 1.

Cognitive performance

Tower of London. The analysis revealed significant main effects of body Fe ($F_{(1,40)} = 11·1$, $P=0·002$) and TOL move category ($F_{(2·5,99)} = 60·1$, $P=0·0001$) on planning time. The interaction of body Fe and TOL move category on planning time approached but did not reach significance ($P=0·057$). The estimated slope of body Fe against planning time became steeper as task difficulty increased; that is, as the target move category advanced from 2 to 5, body Fe had a more pronounced effect on planning speed ($B = -0·02$, $-0·02$, $-0·04$, $-0·05$; $P=0·07$, 0·07, 0·01, 0·002 for categories 2, 3, 4 and 5, respectively). Spearman's correlations showed significant inverse relationships between body Fe and planning time for move categories 4 and 5 (Table 2; Fig. 2). Correlations for move categories (2 and 3) approached but did not reach significance ($P=0·07$ for both). Serum ferritin and serum Fe were also significantly negatively correlated with planning time for target move categories 4 and 5. Serum transferrin receptor level, which rises in Fe deficiency, was significantly positively correlated with planning time for move categories 2 and 4. In contrast to planning time associations, Fe status as measured by body Fe, transferrin receptor or ferritin did not affect the participants' total task execution time or their ability to complete TOL tasks within the allowed number of moves.

Plasma Zn level was not correlated with performance on the TOL task or any computerised cognitive test.

Bakan vigilance task. Fe status was not significantly correlated with the number of correct or incorrect hits per min. There was a significant effect of block on the number of correct ($P=0·0001$) and incorrect ($P=0·01$) responses for all women; that is, across progressive blocks 1−6, women made fewer correct hits and more incorrect hits.

Mental rotation. Fe status did not have a significant effect on the number of correct decisions made across rotation angle blocks.

Simple reaction time. Reaction time to a stimulus was not correlated with Fe status. There was a significant ($P=0·0001$) effect of block on reaction time: across blocks 1−5, reaction time slowed for all women.

Verbal free recall. Recall of words presented for 1 and 2 min was not affected by Fe status. There was a significant ($P=0·03$) effect of presentation time on recall: women recalled more words presented for 2 *v.* 1 s.

Tapping. Body Fe, transferrin receptor and ferritin were not significantly correlated with two-finger tapping speed. A significant correlation was found between serum Fe and tapping time. That is, higher serum Fe was associated with slower tapping speed (r 0·35, $P=0·04$). There was a significant effect of trial block ($P=0·0001$), with progressive slowing in speed over the course of the task. This rate of slowing was not affected by Fe status.

Covariates. Serum C-reactive protein concentrations were within normal limits for all participants, supporting the absence of a confounding effect of inflammatory or infectious processes on the interpretation of Fe status measures. Plasma Zn was below normal $(<8·5 \, \mu g/l)$[45] in twenty-seven women (64 % of the participants).

Table 2. Relationship between iron status measures and Tower of London (TOL) planning time (*n* 42 women)†

TOL target move	Body Fe	TfR	Ferritin	Serum Fe
2	−0·28	0·30*	−0·26	−0·18
3	−0·28	0·03	−0·36*	−0·17
4	−0·39**	0·37*	−0·33*	−0·44**
5	−0·47**	0·22	−0·51**	−0·35*

TfR, serum soluble transferrin receptor.

* $P<0·05$; ** $P<0·01$.

† Spearman's correlation coefficients for associations between Fe status measures and TOL planning times for target move categories 2−5.
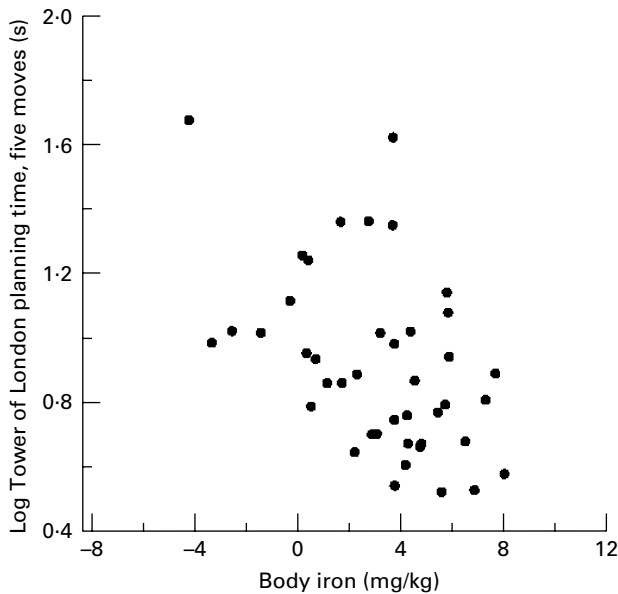
Fig. 2. Scatter plot and regression equation of the relationship between log Tower of London planning time for five-move level and body iron (*n* 42). Slope: −0·047; SE slope: 0·01, *P*=0·001; adjusted R^2 = 0·205.

Salivary cortisol levels measured on the 2 d before cognitive testing and during the test session were not affected by Fe status. Salivary cortisol levels increased significantly (*P*=0·04) in all women, regardless of Fe status, between waking and 30 min after waking during at-home measurements (data not shown). A significant (*P*=0·03) decrease in salivary cortisol between baseline and 30 min post-Bakan task on the cognitive testing day was seen in all women without regard to Fe status.

Menstrual cycle information provided by women (usual cycle length and flow rate) showed no relationship with Fe status. Women self-reported similar (within normal range) levels of perceived stress and trait anxiety; however, state (i.e. at this moment) anxiety measured by the State-Trait Anxiety Inventory immediately before the cognitive test session was significantly positively correlated with body Fe (*r* 0·30, *P*=0·03). No other psychological questionnaire score showed a significant association with body Fe. The scores are listed in Table 1.

During the study, seven FFQ measured energy intakes outside the range and were excluded from the analyses. There were no significant correlations between body Fe and any dietary component analysed on an absolute or energy-adjusted (per 4186 kJ (1000 kcal)) basis (data not shown). Nor did the number of daily servings of any food group, including that containing meat, beans/legumes and eggs (Fe-rich foods), or use of dietary supplements differ according to Fe status.

Discussion

The present study contributes new information on the degree of Fe deficiency associated with negative neuropsychological effects in women of reproductive age. We report significant slowing in planning speed during a test of central executive

function in college women with reduced body Fe but not anaemia. College women experience Fe depletion at a higher rate than the general population of pre-menopausal women[5,14,46]; therefore, it is important to investigate the impact of low Fe status on cognitive function in this subgroup in whom optimal cognition is critical to academic success. The present findings are consistent with the results from previous studies[3–9] performed in women of reproductive age and extend those findings to young college women without anaemia. Kretsch *et al.*[8] observed an association between performance on a test of sustained attention and a decline in Hb and transferrin saturation in obese women consuming a 50 % energy-restricted diet supplemented to provide a complete nutritional profile. Interestingly, the present study did not observe an impact of body Fe on the performance of this same task of sustained attention. Reasons for the dissimilar findings might be related to differences between participant populations, particularly in the level of tissue Fe stores. Neither group of women had Hb values <120 g/l, but body Fe was not measured in the previous study, making comparisons of Fe status between the groups difficult.

Murray-Kolb & Beard[9] demonstrated statistically significant differences in scores on the computerised cognitive abilities tests[47] between the Fe-sufficient and anaemic groups, but not between the Fe-sufficient and Fe-deficient non-anaemic groups. However, a significant trend for coincident declines in cognitive test performance and Fe status category was observed with the Fe-deficient non-anaemic group scores intermediate to the Fe-sufficient and Fe-deficient anaemic groups. The fact that cognitive performance on the computerised test batteries was not significantly affected by Fe status in both the present study and that conducted by Murray-Kolb & Beard suggests that many components of cognition remain intact in preclinical Fe deficiency. It is perhaps important that the present study revealed lower body Fe-related impairment only in planning time for the TOL task, a measure of central executive function. This might indicate a particular sensitivity of the TOL task to detect subtle changes in working memory planning speed, although the finding warrants replication.

Sensitivity of the TOL task to capture alterations in planning ability in relation to exposure to Fe deficiency is supported by a study by Lukowski *et al.*[48]. Male and female participants (*n* 114) were followed from age 12 to 23 months, with measurements of Fe status repeated at ages 5, 11–14 and 19 years, and cognitive function tested at age 19 years. In the follow-up, 95 % of the 19-year-old participants were Fe-sufficient. Participants who as infants had Fe-deficiency anaemia or Fe deficiency that did not correct after 3 months of Fe therapy showed impaired performance on tests of planning, recognition memory, inhibitory control and set-shifting at age 19 years. Planning ability was assessed using Stockings of Cambridge, a computerised form of the TOL. Similar to the present results, group differences in planning ability were found for the highest difficulty (five-move) problems. Together, these findings support both a long-term developmental effect and an acute effect of Fe deficiency on TOL/Stockings of Cambridge performance.

Fe status was assessed in the present study using a calculation of body Fe based on the ratio of serum transferrin receptor: ferritin[30]. Whereas anaemia screening in populations is commonly performed using multiple haematological measurements and defined cut-off values, this more recently developed method expresses body Fe per unit body weight, thus improving the precision of evaluating Fe status in individuals. Body Fe was recently adopted as an Fe status indicator for the US National Health and Nutrition Examination Surveys[32,33] and has been recommended for use by the WHO[34]. Body Fe measurement has shown sensitivity in identifying slight differences in Fe status that are not detected by other blood tests. In an Fe supplementation intervention trial in pregnant Jamaican women, body Fe was significantly higher in women provided with 100 $v.$ 50 mg iron sulphate daily for 3 months; however, no group differences were seen when assessed by Hb or transferrin receptor measures[30]. Body Fe calculation has also shown sensitivity in detecting gradual improvements in tissue Fe resulting from 6 months of 10 mg/d Fe fortification[30]. In the present study, the occurrence of Fe deficiency, defined as body Fe < 0 mg/kg, was 12%. This is similar to the prevalence of 9·2 (SD 1·6)% of non-pregnant women aged 20–49 years with < 0 mg/kg body Fe observed in the US National Health and Nutrition Examination Survey 2003–2006[11]. The present finding contrasts substantially, however, with reported Fe deficiency rates of 30–50% in college women[5,12–17] when serum ferritin serves as the index of Fe status. The lower estimation of Fe deficiency by body Fe $v.$ the ferritin model (where Fe deficiency is defined as two or all three abnormal concentrations of ferritin, transferrin saturation or erythrocyte protoporphyrin) has been reported previously[11,33] and is thought to result from greater sensitivity of the body Fe measure[32].

Salivary cortisol was measured in the present study in order to test whether Fe status-related changes in cognitive function were correlated with stress, but no significant effect of Fe status on cortisol levels was observed. Neither was there a consistent pattern across behavioural questionnaire scores showing a relationship between body Fe and stress or depression. Nonetheless, this is an area of study worthy of pursuit since Fe treatment shows efficacy in alleviating self-reported fatigue in Fe-depleted pre-menopausal women[49]. Fatigue manifests in part as cognitive impairment[50].

Food-frequency measurements were collected to determine whether dietary intake was related to body Fe. A lower intake of Fe-rich foods by women with lower $v.$ higher body Fe might be expected, but no differences in any nutrient measure were found. Nor was Fe supplementation different across the participant groups since this was an exclusionary study criterion. It is possible that dietary behaviours that influence Fe absorption, such as co-ingestion of enhancers and inhibitors of Fe, were not detected by the questionnaire. Peneau et al.[51] found a positive association between serum ferritin and intakes of fibre-poor fruits, vegetables and juices in premenopausal women. Notably, these investigators measured dietary intake using multiple 24 h recalls over a 2-year period, which yielded considerably greater detail on dietary habits than the FFQ used in the present study. Yokoi et al.[52]

also reported that dietary beef intake was a positive predictor and bran breakfast cereal consumption was a negative predictor of serum ferritin.

The strengths of the present study include the use of body Fe to measure Fe status across a continuum and selection of a participant population similar in factors known to affect cognitive task performance including education level, age, sex, drug and alcohol use, and menstrual phase[28,53–57]. The study's design also minimised confounding by testing all participants at the same time in the morning and under standard conditions of feeding status (overnight fast followed by a standard snack before testing) and test administrator (C. A. B.) using a standardised cognitive test battery. However, these defined participant characteristics and experimental design features limit the findings' scope of inference. This was an observational study intended to build existing knowledge, and its results neither imply causation nor claim to be definitive. Further, the sample size was modest and included only a small percentage of women with a tissue Fe deficit. Another important consideration is that only non-obese women were included in the present study. While this inclusion criteria eliminated the confounding effect of obesity on Fe status assessment[58], it does not allow application of the study's findings to an estimated 6% of college women classified as obese[59]. Lastly, the ecological validity of standardised cognitive tests, such as the TOL, remains unconfirmed[60]. This question is being actively investigated with the use of virtual environments[61,62]. Virtual reality employed as an assessment tool would seem to lend itself to elucidating the real-life functional implications of reduced Fe status.

In conclusion, the present study showed a significant association between cognitive planning ability and body Fe while performing the TOL task of central executive function in non-anaemic college women. This finding extends the relatively recent recognition that Fe deficiency with anaemia impairs cognition in women of reproductive age[3,9] to the possibility that reduced Fe status without anaemia can also underlie this functional impairment. Confirmation that preclinical Fe deficiency is indeed causative is contingent upon reversal of cognitive impairment with body Fe repletion. The present study together with our earlier findings[8] and those of other investigators[3–7,9] challenge the paradigm that deleterious functional consequences of Fe deficiency occur primarily in developing children and adolescents and not in adults.

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