
Project title:

*Dietary and Lifestyle contributors to iron and zinc status in young female
Australian blood donors: a pilot study*

Deakin University in collaboration with Australian Red Cross Blood Bank Service

- **Study Design and Sample:** Pre-menopausal women 173 blood donors (donated blood in the past 2 years) and 49 non-blood donors (not donated past 2 years) with mean age of 28.8 (5.7) (SD) years were assessed for indicators of iron and zinc status (haemoglobin, serum transferrin receptor, serum zinc) and dietary intake of iron and zinc utilising a short iron checklist that provided information on the previous day's intake.

Executive Summary

Key findings: Dietary: Short Iron checklist¹

- The median/mean dietary iron intake for all participants combined (short iron checklist) was 11.6 mg/day (IQR) (9.0-15.3)/12.8 mg/day (SD)(5.9) with 16% of participants below the Estimated Average Requirement (EAR)
- The combined median/mean dietary zinc intake was 8.9mg/day (IQR)(6.7 to 11.7)/9.7 (4.4)mg/day with 23% of participants below the EAR.
- 43% of all participants consumed red meat on the previous day (49% of non-donors, and 41% of blood donors)
- The major single food group contributors to iron intake were vegetables contributing 13% and red meat (beef, lamb, mince meat) contributing 12%.
- The major contributor to zinc intake was beef, lamb, mince meat contributing 20% followed by chicken (17%), then vegetables (12%).
- The same five food groups contributed the greatest percentage of both total dietary iron and zinc (vegetables, beef, lamb, mince meat, wholegrain breads and cereals, other meats, refined breads and cereals)
- Among those who consumed red meat on the previous day the most popular was beef with a mean intake of 64g (66g)/day, median 100g/day.
- Those consuming red meat on the previous day (n=95), consumed a similar amount of vegetables (152g (128)/day to those who did not consume red meat (n=127) 148 (131)g/day.
- The amount of red meat consumed was weakly correlated with the amount of vegetables (excluding salad) $r=0.16$, $p=0.02$.
- The mean dietary iron intake excluding supplements was highest in participants recruited at Deakin University 15.4 ± 1.2 (SE) mg/day (n=39) which was significantly higher than the new donors 9.9 ± 0.7 mg/day (n=21) ($P = 0.039$) and regular donors 12.6 ± 0.4 mg/day (n=182) $P = 0.005$ groups (ANOVA). This provides an indication

¹ Zhou SJ, Schilling MJ, Makrides M Evaluation of an iron specific checklist for the assessment of dietary iron intake in pregnant and postpartum women Nutrition. 2005 21(9)903-13

that education/knowledge of food sources of iron may predict intake as those recruited through Deakin were more likely to have some nutrition knowledge.

➤ **Key findings: Iron and Zinc status**

- There was a positive correlation between serum zinc and serum ferritin ($r=0.23$), a positive correlation between serum zinc and haemoglobin ($r=0.25$) and a positive but low correlation between serum zinc and serum transferrin receptor ($r=0.18$).
- Physical activity was not associated with iron or zinc status
- Blood donation in the past two years was associated with lower serum ferritin and lower haemoglobin.
- Serum zinc tended to be 10% lower in those who had donated blood in the past two years, but this was not statistically significant ($p=0.07$).
- Red meat consumption was not significantly associated with either iron zinc status.
- 36% participants consuming red meat on the previous day had low serum ferritin ($<15 \mu\text{g/L}$), compared to 48% of participants who did not consume red meat on that day ($p=0.09$).

➤ **Key Conclusions:**

- Providing nutrition education to the Blood Service donors on the dietary sources of iron may assist in raising iron intake in this group to a level comparable to 'informed volunteers' from Deakin University.

- As blood donation has a significant impact on iron status and limitations in the dietary assessment tool were identified, we were unable to detect any relationship between dietary iron and zinc intake (assessed by short iron checklist) and iron and zinc status. Similarly we were unable to detect a relationship between red meat intake and iron and zinc status.

- The dietary assessment tool utilised in this study, which although has been validated, has a major limitation in that it only assesses the intake of key foods the day before and does not detect the consumption of iron rich foods that may be consumed on a weekly rather than daily basis, which would have been identified had we obtained the nutrient data from the Cancer Council Food Frequency Questionnaire.

➤ **Lessons learnt and issues identified**

- Due to an inability of the Cancer Council (Vic) to provide nutrient results from our completed questionnaires we have been unable to:
 - Validate the short Iron specific checklist with the Cancer Council revised Food Frequency Questionnaire
 - Assess the relationship between food groups including red meat utilising characterisation of dietary patterns and indicators of iron and zinc status.

- In undertaking this study we now have identified some key issues relating to the validity of the short iron checklist to measure iron intake, as dietary intake is only assessed on one day (previous day) and only 43% of participants reported consuming

meat on the day assessed. This is likely to be an underestimation of the number who consume red meat regularly but not daily.

- **Recommendations:** This study has enabled us to recommend the modification of dietary questionnaires for future studies to ensure a more accurate assessment of dietary iron and zinc intake and potential enhancers and inhibitors of absorption. Recommendations include:
 - That a dietary assessment tool has a low subject burden and has the ability to measure meal distribution and food combinations (including potential iron absorption enhancers or inhibitors) be utilised in future studies. As no tool is readily available we recommend the use of repeat 24hr recalls that have the ability to capture meal distribution. We also recommend the inclusion of some additional questions regarding usual intake of high iron and zinc containing foods e.g. meat, chicken, fish, vegetables and fortified cereals.

Identification of modifications required to the Cancer Council revised Food Frequency questionnaire (FFQ) and iron specific checklist to enable accurate assessment of iron and zinc intake

We would recommend the following additional dietary information should be incorporated in any future studies to accurately assess dietary iron and zinc intake if this FFQ is to be used.

1. Question 27 only asks for frequency of iron use or multivitamin use and there is no way to accurately assess the amount of iron present in iron supplements or multivitamin supplements. Another question is required about specific dose and brand name of vitamin and/or iron supplement and frequency of use needs to be included.
2. Question 8 has combined all types of cow's milk with soymilk and rice milk and some of these products are iron fortified and some are not.
3. Questions 24, 24, and 25 (A) relate to breakfast cereals and allow identification of porridge consumption and two breakfast cereals, but it is not clear how information from these questions are combined to provide an accurate estimate of the amount of each type of cereal consumed. The list of cereals is limited and we would recommend the option of being able to specify specific cereals, and the exact iron content and frequency of use could be used to adjust the final nutrient analysis output for iron by researchers interested in iron intake.

Limitations of short iron checklist¹

1. The questionnaire needed to be updated to include new fortified foods and remove foods no longer available. The updated questionnaire comprised of 76 items, compared to 68 items in the original version by Zhou et al (2005).

2. Items added to the checklist included three items to represent soy foods and meat replacements (e.g. Sanitarium soy products), one item to represent fortified breads, three items to represent a wider range of breakfast cereals, and four items to represent different milk-based drinks and milk additives (e.g. Sustagen Sport, Coles MX11). Items removed from the checklist included 'White Stuff' bread, Uncle Toby's 'Pasta Plus', and Milo in a carton.
3. The iron checklist specifically asked about dietary intake only on the previous day. As consumption of meat products may not be daily for many premenopausal women and the previous day's intake may not be typical we would recommend that the time frame for dietary assessment be changes to "usual" during the last 3 months and that were possible the short iron checklist should be completed on more than one occasion.
4. The fact that data from this checklist relates of only one days intake is likely to deviate considerably from any FFQ, particularly for the Cancer Council FFQ which is measuring usual dietary intake over the last 12 months.

Limitations of both Cancer Council FFQ and short iron checklist

1. Neither dietary assessment method measures meal patterns e.g. the combinations of foods consumed together.
2. Neither dietary assessment method includes the capacity to measure the presence of inhibitors or enhancers consumed at each meal and with snacks.

Recommendations for further studies

1. Consider repeat 24hr recalls which could provide an estimate of "usual dietary iron and zinc". The advantages of 24hr recalls are that they have low subject burden and are easy to conduct. Disadvantages are that one day's intake is not representative, but the inclusion of at least two, 24hr recalls with a calculated average is more likely to capture "usual intake".
2. Use the Meal-based assessment tool of 630 items developed by Heath et al. 2005 to measure dietary iron and zinc intake² may be easier/quicker to administer but this is not clear. Currently no computer program available to allow this tool to be utilised.

² Heath AL, Roe MA, Oyston SL, Fairweather-Tait SJ. Meal-based intake assessment tool: relative validity when determining dietary intake of Fe and Zn and selected absorption modifiers in UK men. Br J Nutr. 2005 Mar;93(3):403-16.

Key Results

PARTICIPANTS

A total of 233 completed the study and included pre-menopausal women recruited through the Australian Red Cross Blood Service (ARCBS) in Sydney (n=192) and Deakin University, Melbourne (n=41). The group recruited through Deakin included some women who had donated blood in the last year.

Eleven subjects were excluded on the basis of high levels of C-reactive protein (CRP; n=9; >10 mg/L), or high serum ferritin levels (n=2; >150 µg/L). The final sample size consisted of 222 women (173 blood donors and 49 non-donors; 185 recruited by ARCBS and 37 recruited by Deakin University).

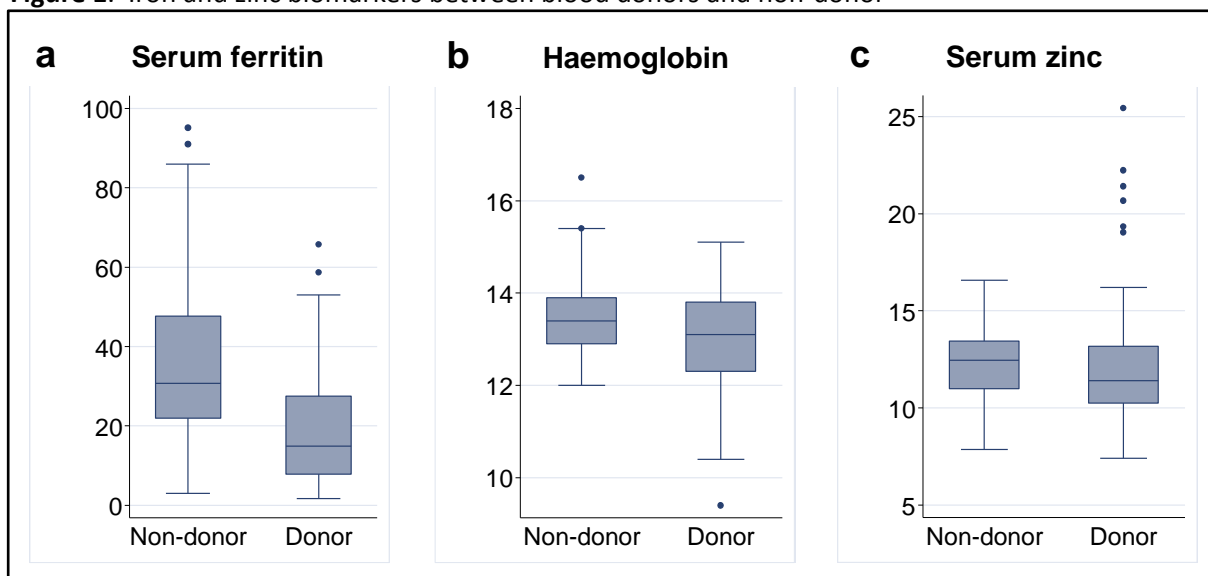
Table 1. Dietary Intake of participants recruited by ARCBS vs participants recruited by Deakin University

	Participants recruited by ARCBS (n=185)		Participants d by Deakin University (n=37)	
	Mean (SD)	Median (IQR)	Mean (SD)	Median (IQR)
Total dietary iron intake (mg/day)	12.3 (5.0)	11.6 (8.7 to 14.9)	15.5 (8.7)	12.5 (10.3 to 19.1)
Beef (grams per day)	27 (47)	0 (0 to 50)	30 (79)	0 (0 to 0)
Lamb (grams per day)	13 (46)	0 (0 to 0)	11 (39)	0 (0 to 0)
Mince meat (grams per day)	13 (33)	0 (0 to 0)	2 (13)	0 (0 to 0)
Liver (grams per day)	0.5 (6.2)	0 (0 to 0)	5 (28)	0 (0 to 0)
Total red meat (grams per day)	53 (71)	0 (0 to 100)	48 (86)	0 (0 to 100)

BLOOD DONATION

Blood donation in the past two years was associated with lower serum ferritin (median (IQR) / mean (SD) in non-donors: 30 (22 to 46) / 36 (23) µg/L; in donors: 15 (8 to 28) / 19 (14) µg/L; t-test with square root transformed serum ferritin: p<0.001) and lower haemoglobin (median (IQR) / mean (SD) in non-donors: 13.4 (12.9 to 13.9) / 13.4 (1.0) g/dL; in donors: 15 (8 to 28) / 13.0 (1.0) g/dL; t-test with untransformed haemoglobin p<0.001).

Figure 1. Iron and zinc biomarkers between blood donors and non-donor



PHYSICAL ACTIVITY

No aspects of physical activity (total time, time in moderate physical activity, time in vigorous physical activity, or metabolic equivalents) were associated with iron or zinc status. Although 60% of participants performed at least one hour of vigorous physical activity per week, and 80% of the sample met national guidelines for physical activity, it is likely the duration and/or intensity of exercise performed by participants was insufficient to induce substantial iron losses.

DIET

The median/mean dietary iron intake (short iron checklist) was 11.6 mg/day (IQR) (9.0-15.3)/12.8 mg/day (SD)(5.9) with 16% of participants below the Estimated Average Requirement (EAR). The median/mean dietary zinc intake was 8.9mg/day (IQR)(6.7 to 11.7)/9.7 (4.4)mg/day with 23% of participants below the EAR.

43% of all participants consumed red meat on the previous day (49% of non-donors, and 41% of blood donors).

Top contributors to dietary iron and zinc intake

Based on responses to the short iron checklist, food groups (e.g. beef, lamb and mince meat; vegetables; sweets) were ranked on their average contribution to total dietary iron and total dietary zinc intakes. The five food groups contributing the greatest percentage of total dietary iron intake and total dietary zinc intake are presented in Table 1 and Table 2. The same five food groups contributed the most to both iron and zinc dietary intakes. Beef, lamb and mince meat were collectively the second greatest contributor to dietary iron intake (11.6%), and were the greatest contributor to dietary zinc intake (20.0%). Data from the iron checklist should be cautiously used to estimate dietary zinc intake as the iron checklist does not include dairy products and thus may not provide a true indication of total dietary zinc intake – e.g. 53% of participants usually consume at least one cup of milk per day, with one cup containing approximately 1 mg of zinc which would represent approximately 11%.

Table 2 Top 5 food groups contributing to total dietary iron intake

Food group	Average contribution to total iron intake (%)
Vegetables	13.4
Beef, lamb, mince meat	11.6
Wholegrain breads and cereals	9.8
Chicken, pork, fish, seafood	9.2
Refined breads and cereals	9.1

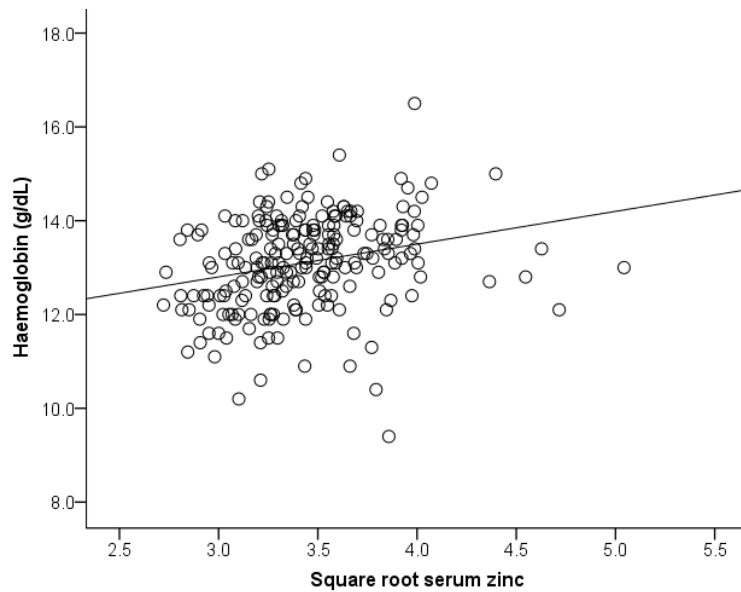
Table 3 Top 5 food groups contributing to total dietary zinc intake

Food group	Average contribution to total zinc intake (%)
Beef, lamb, mince meat	20.0
Chicken, pork, fish, seafood	17.0
Vegetables	12.3
Refined breads and cereals	7.6
Wholegrain breads and cereals	7.2

Association between serum zinc and markers of iron status

Serum zinc and serum haemoglobin

Figure 2. Serum zinc vs Haemoglobin (n=217)
(zinc values square root transformed); $r=0.25$, $p<0.001$



Serum zinc vs serum ferritin

Figure 3. serum zinc v serum ferritin (n=217)
(values square root transformed); $r=0.23$, $p=0.001$

