Providing evidence to support the development of whole grain dietary recommendations in the United Kingdom

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Observational evidence suggests that increased whole grain (WG) intake reduces the risks of many non-communicable diseases, such as CVD, type 2 diabetes, obesity and certain cancers. More recently, studies have shown that WG intake lowers all-cause and cause-specific mortality. Much of the reported evidence on risk reduction is from US and Scandinavian populations, where there are tangible WG dietary recommendations. At present there is no quantity-specific WG dietary recommendation in the UK, instead we are advised to choose WG or higher fibre versions. Despite recognition of WG as an important component of a healthy diet, monitoring of WG intake in the UK has been poor, with the latest intake assessment from data collected in 2000–2001 for adults and in 1997 for children. To update this information we examined WG intake in the National Diet and Nutrition Survey rolling programme 2008–2011 after developing our database of WG food composition, a key resource in determining WG intake accurately. The results showed median WG intakes remain low in both adults and children and below that of countries with quantity-specific guidance. We also found a reduction in C-reactive protein concentrations and leucocyte counts with increased WG intake, although no association with other markers of cardio-metabolic health. The recent recommendations by the UK Scientific Advisory Committee on Nutrition to increase dietary fibre intake will require a greater emphasis on consuming more WG. Specific recommendations on WG intake in the UK are warranted as is the development of public health policy to promote consumption of these important foods.

Whole grains: Dietary guidelines: UK population

What are whole grains and whole-grain foods?

Cereal grains, such as barley, corn, oats, rice, rye and wheat, are a staple food in the human diet and are important sources of dietary carbohydrate and protein. Whole-grain (W-G) foods are foods made from cereals and starch, which contain all three anatomical components of the grain: the outer bran, endosperm and germ. Although there is not one single definition of whole grain (WG), similar definitions from the American Association of Cereal Chemists International and the Healthgrain forum, a European consortium of scientists and industrial partners, are widely used. These definitions state that WG must contain the three component parts of the grain in the same relative proportions found in the intact kernel¹. Additionally, the Healthgrain forum definition allows for small component losses due to processing of the grain². Other definitions found in country-specific reports are very similar to these¹⁻³ giving a general consensus on what constitutes a WG. As such, products that state ‘WG’, ‘wholegrain’ or ‘whole’ followed by the type of cereal or pseudo-cereal (named so because their composition is similar to that of cereal grains) in the ingredients list should comply with...
the consensus WG definitions and can be recognised as W-G foods. Similarly the term wholemeal, a regulated term for breads and flours in the UK^{(6)}, has requirements that align with WG definitions. Some cereal and pseudocereal ingredients, such as oats, oatmeal, brown rice, buckwheat and quinoa, often do not carry the whole/WG prefix. However, in the case of oats the largest majority is consumed as a WG since oats are rarely refined to separate the bran and endosperm. Brown rice is so called because the bran is retained. The small seeds of most pseudo-cereals precludes refining^{(10)}. At present there is no legal definition of a W-G food except for some specific in-country recommendations. For example, in the US foods must contain at least 51 % WG by weight per reference amount customarily consumed to comply with a WG health claim. In Denmark flours, grains and rice must contain 100 % WG, breakfast cereals 60 % and bread 50 % WG and in Germany pasta must contain 100 % WG, whereas wheat and rye breads must contain 90 % WG.

Health benefits of whole grains: observational evidence

CVD, type 2 diabetes, obesity and cancer are the most prevalent non-communicable diseases across the world^{(9)}. CVD (diseases of the heart or blood vessels such as CHD) are the number one cause of death globally (31 % in 2012^{(9)}) and are currently the second biggest killer in the UK, after cancer^{(10)}.

Observational epidemiology studies suggest there is an inverse association between increased habitual WG intake and reduced risk of non-communicable disease. Results of systematic reviews and meta-analyses suggest that there is a 20–30 % risk reduction of CVD and type 2 diabetes, comparing low or non-consumers with WG intakes of 48–80 g/d^{(11,12)}. These reported associations may also be dose-responsive with meta-analyses suggesting 22 % CVD risk reduction by increasing W-G food intake to 90 g/d (i.e. three daily servings of foods containing WG) and an absolute risk reduction of 0.3 % in the type 2 diabetes rate for each 10 g/d of WG consumed^{(13,14)}. Increased WG intakes have also been associated with reduced risks of cancer, particularly colorectal cancers where a 10 % relative risk reduction was estimated for an increment of three daily W-G food servings^{(15)}. Furthermore, high v. low WG intakes have been associated with reduced risks of digestive tract cancers as well as breast, prostate and pancreatic cancers^{(16–18)}. One of the major risk factors for CVD, type 2 diabetes and cancer is obesity, which has become a global health concern as the rate has more than doubled since the 1980s^{(19)}. Intake of WG may have a beneficial role in weight management or maintenance^{(20)}. A meta-analysis of three prospective cohort studies with a total of 119 054 participants, concluded that three to five daily servings of WG was associated with consistently less weight gained during 8–13 years of follow-up, compared with never/rare consumers^{(21)}. These three cohort studies were conducted in US populations and the data are supported by a recent examination of trends in national data over 12 years, which confirmed that W-G foods may contribute to weight management^{(22)}. Most recently, meta-analyses have focused on investigating WG consumption in relation to mortality from non-communicable diseases, with similar results to the non-communicable disease risks^{(22)}. All-cause mortality and disease-specific (CVD or events, diabetes, cancers and respiratory disease) mortality risks are shown to be reduced by 5–30 % for a doubling of W-G food intake as well as per 16 g/d and three daily servings of WG^{(23–26)}.

Many of the observational studies included in the systematic reviews and meta-analyses adjust their risk estimations for confounding factors, which may also explain the suggested associations. Age, sex, smoking, physical activity, body mass, history of disease, health markers and other dietary intakes such as energy intake, have all been considered and included in multivariable analyses. Varying effects of confounding are seen, for example, an age- and sex-adjusted hazard ratio of 0.61 (95 % CI 0.59, 0.62) was attenuated to 0.77 (95 % CI 0.75, 0.79) after adjustment for smoking and further attenuated to 0.83 (95 % CI 0.81, 0.86) after additional adjustments for race/ethnicity, alcohol intake, education, marital status, health status, obesity, physical activity, red meat, fruit, vegetables and total energy intakes and hormone use^{(22)}. These results show that there is evidence of confounding, but the reported significant 17 % reduction in all-cause mortality hazard ratio, for the highest WG consumers compared with the lowest, appears be independent of the confounders tested. Although this study, and the majority of the observational studies, consistently report independent inverse associations even after adjustment for relevant confounding factors, it is important to note that the potential for residual confounding may still remain. In addition, the majority of the studies included in the meta-analyses come from US and Scandinavian populations with a small number of studies from European cohorts and a very few from middle or far-Eastern populations. Therefore, the applicability of the findings to populations with differing dietary patterns and cultural habits should be further investigated.

Heath benefits of whole grains: intervention evidence

The results of intervention studies do not consistently corroborate the findings from observational studies. Some interventions show beneficial effects of consuming WG on health markers, whereas others fail to find significant results. For example, obese participants with metabolic syndrome who were given a 12-week dietary advice intervention to obtain all grain servings from W-G foods, showed a reduction in plasma C-reactive protein concentrations and percentage body fat in the abdominal region compared with a WG avoidance group (refined grain group). In another randomised controlled trial, markers of inflammation were reduced in overweight and obese but otherwise healthy participants, with suboptimal diets, following a W-G wheat intervention v. a refined grain control group for 8 weeks. However, no
significant variations in body composition, plasma lipids or glycaemia were found in these participants or between intervention and the refined grain control (27). Similarly, in a 16-week WG intervention, no changes in CVD markers between intervention groups and controls were seen (28). A meta-analysis of randomised controlled WG intervention studies on body weight and body composition concluded that the trials did not support the role of WG in body weight management. However, beneficial effects of WG on body weight may be more apparent for body fat percentage or abdominal adiposity, which may be mediated through decreased inflammatory responses (29).

Explanation for the differing findings of intervention studies to those of observation studies are thought to be due mainly to the differences in study design. Intervention studies are time-restricted with no reported trial lasting longer than 4 months, which may not be long enough for sustained health benefits to be seen. Sample sizes are often small, although usually authors claim that they are powered to be able to detect any significant meaningful changes in disease markers resulting from the intervention. The type, variety and quantity of WG used in intervention studies differ and this may be another reason for inconsistent results. Some WG, such as oats, rye and barley, contain higher fibre, particularly soluble fibre, than wheat and rice. Therefore, the physiological effects on the body may differ between grain types and if one grain type or a mix of grains are included in the diet.

The outcomes of intervention studies are reported as the outcome on risk markers for disease, not the occurrence of a disease as this would not be possible within the time frame of an intervention study. Therefore, comparison with observational studies where a particular disease or event has occurred may not be appropriate. Interestingly observational studies that report on markers for disease risk also have varying results (30). For example, in our recent analysis of UK WG intakes, no significant differences or trends in blood pressure, blood lipids or anthropometric measures between non-consumers and increasing tertile of WG intake were observed. However, a reduction in leucocyte counts by increasing tertile of WG intake, and a difference in adults with a small intake of WG compared with non-consumers was seen in C-reactive protein concentrations (31). This suggests that intervention studies and observational studies, which report biomarkers of disease risk, are more aligned in the inconsistency of their results compared with those that only report disease outcomes. Finally, intervention studies that report no changes in disease markers are most often carried out in healthy or overweight but otherwise ‘healthy’ volunteers. This raises the question, whether improvement in disease risk markers should be expected, if the participants are otherwise healthy. Some of the largest effects are seen in intervention studies with ‘at risk’ participants with dyslipidaemia or obesity. Furthermore, it is known that as we age our health and health markers in general decline. We should reconsider the pharmacological paradigm, which suggests that short-term dietary intervention with WG should improve or reduce disease risk in favour of a longer-term model, which suggests that increased WG intake in the longer-term reduces age-related declines in health.

Despite the inconsistent results from WG interventions, to our knowledge, no study has shown or reported negative effects or outcomes of increasing WG intake on health markers. Therefore, advice to consume more WG could be a low-risk public health strategy. Of course it is important to note that for a small proportion of the population with gluten intolerance, caution must be made when consuming WG containing gluten. However, gluten-free WG alternatives such as amaranth, brown rice, buckwheat and quinoa are available and their consumption by those with gluten intolerance can be encouraged. WG oats do not contain gluten, but are sometimes cross-contaminated with wheat during harvesting or factory processing. Thus, consumers should always check product labels for gluten-free oat ingredients for clarification.

Health benefits of whole grains: mechanisms of action

There is no one clear mechanism identified for which WG benefit the body, instead there are a combination of several processes suggested, which may also interact with one another. Essentially the accepted pathways in which WG have an effect on chronic diseases can be split into two: dietary fibre and bioactive components (Fig. 1).

WG contain cereal fibre and have increased amounts compared with refined grain counterparts. For example, the Association of Official Analytical Chemists dietary fibre content of wholemeal bread is more than twice as high at 7 g/100 g, whereas for white bread it is 2.9 g/100 g (32). Insoluble and soluble dietary fibres improve digestive health through a range of effects such as stool bulking, altered intestinal transit time as well as increased colonic fermentation, which induces the production of SCFA (33). Beta-glucan, a soluble fibre found in higher concentrations in oats and barley, has been shown to lower blood cholesterol concentrations and improve postprandial insulin and glucose responses (34,35). In addition, these physiological effects of both soluble and insoluble fibres may also have satiating effects on appetite, which may have a role in weight management (36). Cereal fibre, in particular, has been highlighted as one fibre source that may reduce the CHD risk (37), and the need for trials investigating the effects of cereal fibre on type 2 diabetes risk has been emphasised (38). It is also important to consider that the associated benefits of WG are above and beyond those of just the cereal fibre. WG also contain a large amount of bioactive components such as phenolic acids, lignans, plant sterols, tocolts, benzoazinoids and alkylresorcinols as well as a variety of vitamins and minerals (39,40). Many of these have antioxidant and anti-inflammatory properties as well as providing essential nutrients into the diet of WG consumers, which could lead to protection from later disease (41–43).

New and emerging research into the gut microbiome suggests that WG may influence the type of bacteria that
make up the gut microbiota, which has beneficial effects on the host gut health\cite{44-46}. In a human trial, it was shown that a mixture of WG types, a combination of WG barley and brown rice, increased gut microbial diversity, which induced some beneficial changes on the profile of bacterial populations in the host; evidence that in the short term, increased intake from a mixture of WG alters the gut environment and results in improvements in systematic inflammation\cite{47}.

Whole grain intake recommendations

There are currently some recommendations to consume WG across the globe. These vary by country with some offering generic advice and others, which give merit to the observational evidence, providing quantity-specific daily target intakes\cite{48}. For example, in the USA and Canada advice to 'make one-half of your grains whole grains' is followed by a quantity recommendation of a minimum 3–5 ounce-equivalents (servings)/d (48–80 g/d)\cite{49}. Similarly quantity-specific dietary intake targets are advised in Singapore where advice for adults is to consume 75 g/d WG per 10 MJ diet (four daily portions). Semi-quantity-specific intake targets are advised in the UK, where advice is given for adults to consume at least one serving of rice and alternatives from W-G foods\cite{5}. In the majority of other countries with food-based dietary public guidance (including Australia, China, France, Germany and Ireland) generic advice to choose or include WG and consume more WG is given\cite{11,48}. The variety of WG recommendations across many countries could be confusing, particularly where quantity-specifics do not match. Target intake are largely based on dietary fibre and endorsing WG as a source of fibre. Clearly targets need to reflect country-specific cultural and traditional diets; however, some consistency between countries based on scientific evidence would be useful.

Current whole grain intake

As with WG recommendations, WG intake varies across countries. Assessing intake of WG is challenging. Along with the universal issues of dietary intake reporting from either FFQ or diet records, further difficulties arise for WG since the identification of these partly rely on participant knowledge, manufacturer information and standardised databases on the content of W-G in foods. Such databases are publicly available in the USA through the United States Department of Agriculture Patterns Equivalents Database\cite{50} and recently updated data have been published for W-G foods consumed in Australia\cite{51}. We have recently published our database of W-G foods consumed in the UK covering the period 1986–2016\cite{52}. Despite the difficulties in assessing and measuring WG intake, the available data show that consumption and intake of WG in the majority of countries is low (Table 1). Average WG intakes for adults range from as little as 4 g/d in Italy, measured in 2005–2006, and 5 g/d in France measured in 2009–2010, to as high as 58 g/d (63 g/d/10 MJ) in Denmark measured in 2011–2013\cite{53-55}. The higher reported WG intakes in the Danish population are attributed to a combination of traditional diets that include WG foods, such as rye bread, and the recent success of the Danish WG campaign\cite{56}. The campaign, a public and private company partnership, aiming to increase accessibility and awareness of WG and the associated health benefits, has seen an increase in average Danish intakes of 75%.
from 2004 to 2013. In the UK, WG intake from foods with at least 10 % WG content was reported to be 7 g/d for children and 14 g/d for adults from the 2000–2001 and 1997 national dietary surveys, respectively (57,58). We (the authors) have worked to update these data using the UK National Diet and Nutrition Survey data from 2008 to 2011, covering the first 3 years that the survey has been run on a rolling programme (59). Although our assessment included previous surveys (59). We reported average WG intakes of 20 and 13 g/d in adults and children, a slight increase on the previous survey (59). Although our assessment included children. However, white bread and 'pasta, rice, pizza and other miscellaneous cereals' were the two most commonly consumed cereals and cereal products, eaten by more than 70 % of the total population (60). This gives opportunity for replacement of refined grain breads and pastas with WG varieties and the potential to increase population WG intakes. In particular, increased consumption of WG provides opportunity to increase dietary fibre intakes since our analysis showed that dietary fibre intakes were 5
grams (21).

### Table 1. Whole grain (WG) intakes of adults in national surveys

<table>
<thead>
<tr>
<th>Country; Study</th>
<th>Age range (n)</th>
<th>Median (5th – 95th percentile)</th>
<th>% non-consumers (%)</th>
<th>% meet target intake (target)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Italy; INRAN-SCAI (Italian food consumption database) 2005–2006 (53)</td>
<td>18–65 years (TP: 2313, M: 1068, F: 1245)</td>
<td>TP: 4 (12) M: 3 (12)</td>
<td>TP: 0 (3⁰) M: 0 (30°)</td>
<td>TP: 76 M: 84</td>
</tr>
<tr>
<td>France; Comportements et Consommations Alimentaires en France 2010 Survey (54)</td>
<td>18+ years (TP: 1389, M: 588, F: 801)</td>
<td>TP: 5 (0–3) M: 4 (0–5)</td>
<td>TP: 0 (26¹) M: 0 (25¹)</td>
<td>TP: 68 M: 72</td>
</tr>
<tr>
<td>UK; National Diet and Nutrition Survey 2000–2001 (58)</td>
<td>19–64 years (TP: 1692, M: 758, F: 934)</td>
<td>TP: 23 (28) F: 5 (0–5)</td>
<td>TP: 14 (0–98) F: 2 (0–79)</td>
<td>TP: 29 F: 63</td>
</tr>
<tr>
<td>USA; National Health and Nutrition Examination Survey 2011–2012 (51)</td>
<td>19+ years (TP: 4878)</td>
<td>TP: 0.97 (0.05 oz-eq/d)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Australia; Australian Grain and Legumes Consumption and Attitudinal Study 2014 (74)</td>
<td>2–70 years (TP: 3031, M: 1194, F: 1837)</td>
<td>TP: 29 (37) M: 33 (45)</td>
<td>TP: 20 (0–67) M: 19 (0–67)</td>
<td>TP: 20 M: 21</td>
</tr>
<tr>
<td>Norway; Norwegian Women and Cancer Cohort 1992–1998 (76)</td>
<td>30–60 years (F: 1797)</td>
<td>TP: 51 (36) F: 44 (0–120)</td>
<td>TP: 44 (0–120) F: 19 (0–120)</td>
<td>TP: 5 F: 5</td>
</tr>
<tr>
<td>Sweden; Northern Sweden Health and Disease Study Cohort 1992–1998 (78)</td>
<td>30–60 years (TP: 2989, M: 1372, F: 1617)</td>
<td>M: 58 (50) F: 41 (32)</td>
<td>M: 49 (0–149) F: 35 (0–102)</td>
<td>TP: 5 M: 29% F: 27% (75 g/d/10 MJ)</td>
</tr>
<tr>
<td>Denmark; Danish National Survey of Diet and Physical Activity 2011–2013 (55)</td>
<td>15–75 years (TP: 3189, M: 1546, F: 1643)</td>
<td>TP: 58 (–) M: 65 (–)</td>
<td>TP: 0 F: 51 (–)</td>
<td>TP: 0 F: 51 (–)</td>
</tr>
</tbody>
</table>

5% standard error; SD, standard deviation; TP, total population; M, male; F, female.

* Median and 97.5th percentile.

† Median and 95th percentile.

*5 oz-eq can be either 16 or 28.35 g depending on the food source hence is not converted in grams (21).
and 3 g/d significantly higher in adult and child WG consumers compared with non-consumers.

Barriers to new whole grain intake recommendations

Although the new Eatwell Guide and advice from Public Health England has raised the profile of W-G foods\(^{61}\), we believe that a more explicit recommendation is required. We also suggest that a global recommendation would be useful to improve clarity and encourage industry to develop more W-G products. However, before any new recommendation can be made consideration is needed of potential difficulties that may arise. Consumer desirability of WG, particularly in the taste, texture, price and availability will be key factors in aiding an increase in WG intakes. The availability of W-G food products has increased as has the popularity of foods perceived as being more healthy\(^{62}\). However, as a consequence in some cases these foods can be more expensive\(^{63}\). Food manufacturers should seek opportunities to develop new W-G foods, which are appealing and affordable for the consumer. Manufacturers must be allowed to label their foods effectively and in a way which is regulated for the consumer. This is linked to a second potential barrier to a quantity-specific WG recommendation, which is to have a clear definition of a W-G food. As part of the present work on updating national intake data in the UK we have developed a database of the WG contents of foods consumed from several UK surveys\(^{52}\). Following the guidelines by Ross \textit{et al.}\(^{64}\) we report on a dry weight basis from which intakes can be calculated in g/d given the portion size consumed. The database also contains W-G foods as single raw ingredients as well as on an as consumed basis so that intake can be calculated either from recipe ingredients with weights or as the food as eaten. For example, the WG content of dry wholemeal pasta is estimated to be 89.5 % DM, whereas wholemeal pasta boiled is estimated to be 30.9 % WG DM as eaten. This database, as with other databases, highlights the differing amounts of WG contained in food products. In the USA, the health claim ‘Diets rich in wholegrain foods and other plant foods, and low in saturated fat and cholesterol may help reduce the risk of heart disease’ is allowed for use only on foods that contain at least 51 % WG\(^{65,66}\). Previously in 2002 the UK also had a health claim that was allowed for the use on foods containing at least 51 % WG: ‘People with a healthy heart tend to eat more whole-grain foods as part of a healthy lifestyle’\(^{67}\). However, this claim is no longer permitted for use, since in 2010, the European Foods Standards Agency rejected the use of all WG health claims in Europe on the basis that WG was ‘insufficiently characterised’\(^{68}\). More recently the US American Association of Cereal Chemists International and a multidisciplinary expert roundtable have proposed a characterisation that W-G foods must deliver at least 8 g WG per 30 g serving (about 27 % WG content)\(^{1,69}\). The Healthgrain forum have recently proposed that a W-G food should contain at least 30 % WG content on a DM basis with more W-G ingredients than refined grain ingredients in the final product. This is in addition to compliance with country-specific fat, salt and sugar limitations\(^{70}\). A scientific consensus and subsequent studies using one definition of a W-G food would add to the evidence concerning health benefits of WG and aid public bodies in recommending food-based WG guidance.

Conclusion

The observational evidence on the long-term health benefits of higher WG consumption is clear and consistent. As a result some public health groups advise quantity-specific daily WG intake recommendations. Since grains are important dietary sources of energy and other nutrients including dietary fibre, and WG varieties contain higher amounts of fibre compared with refined grain varieties, recommendation to consume them should be emphasised. The current UK advice from the Eatwell Guide, now includes images of W-G foods and the emphasis on choosing ‘wholegrain and higher fibre versions with small amounts of salt fat and sugar’\(^{61}\). The inclusion of W-G food images within the ‘carbohydrates’ section of the plate is a step in the right direction. The recent Scientific Advisory Committee on Nutrition Report on Carbohydrates and Health, has advised that dietary fibre intakes should be raised to a minimum of 30 g/d for adults and 15–25 g/d for children with no more than 5 % of dietary energy coming from free sugars\(^{71}\). We believe that a quantity-specific recommendation for WG intake would be more helpful to the general public than the general statement in the Eatwell Guide, since 30 g/d fibre will be impossible to achieve without the inclusion of WG. For example, the British Nutrition Foundation have developed a 7-d meal plan, which is designed to indicate the amount of different foods needed to achieve the fibre and free sugars targets\(^{72}\). Within this meal plan more than half of the carbohydrate-rich foods are W-G foods. To achieve 30 g/d of dietary fibre an adult would need to consume almost six daily servings of WG, in addition to over eight daily portions of fruits and vegetables (Table 2). This gives a very clear indication of the need to consume substantial quantities of WG, in addition to fruit, vegetables together with high-fibre beans and pulses, which are also included in the meal plan.

Increasing WG intakes may be difficult. Studies in the USA have shown that despite having a quantity-specific recommendation of 3 oz eq/d, this target has not been achieved. Assessing trends across 12 years of the National Health and Nutrition Examination Survey showed that, although recommended intakes of total grains are being met, only small increases in WG intake were observed and <10 % of Americans currently meet the recommendation for WG intakes. This suggests that despite the increasing consumer interest and availability of W-G foods, little progress in replacing intake of refined grains with WG has occurred in the past 12 years\(^{73}\). In contrast, as previously mentioned, the Danish population has shown considerable success in
improving WG intakes, demonstrating that with public and private partnership campaigns population dietary habits can change.

It is important that any new dietary recommendations focus on replacement of refined grain foods with W-G foods, so that overall energy intake does not increase. Finally, there may be potential for co-ordinating a WG recommendation with the current UK fruits and vegetables guidance. For example, the current '5-a-d' campaign for portions of fruits and vegetables could be mirrored by a '3-a-d' campaign for WG. This would require clarity in definitions of WG, W-G foods and mechanisms to enable consumers to identify portions of W-G foods.

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Conflict of Interest

None.

Authorship

K. D. M. collated the evidence, analysed UK data, gave the presentation at the Nutrition Society’s Summer Meeting and drafted the article. C. J. S. supervised the research, contributed to writing and critical review of the manuscript. M. S. P. supervised the research and critically revised the manuscript for intellectual content.

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