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Modelled gradual changes in protein intake to increase nutrient adequacy lead to greater sustainability when systematically targeting an increase in the share of plant protein

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Abstract

In line with sustainability issues, we are currently seeing a transition towards a lower consumption of animal protein. How ongoing gradual rearrangements in protein patterns impact sustainability and climatic change remains unknown. We used data from a French representative survey and selected for each individual the dual substitution of a serving of a protein food that most improved nutritional adequacy (using the probabilistic PANDiet score), with an increase in the percentage of plant protein required (SP) or not (SN). This was iterated 20 times incrementally and we monitored the evolution of sustainability endpoints, including greenhouse gas emissions (GHGE) and predicted premature deaths avoided. After 20 iterations, the plant protein intake (31.1% total protein) decreased under SN (30.0%) and increased under SP (37.7%). The food groups whose contribution to protein intake increased the most were legumes (+225%), fatty fish (+151%) and lean chicken (+82%) under SN and legumes (+502%), pizzas and quiches (+190%) and fatty fish (+102%) under SP. The PANDiet score rose slightly more under SN (+7.5±0.1) than SP (+6.2±0.1). GHGE levels increased from 5.4±0.05 to 5.7±0.04 kg eq.CO_2/d under SN and decreased to 5.1±0.04 under SP. Diet costs increased from 7.4±0.06 to 8.2±0.05 €/d under SN and 7.6±0.05 under SP. Predicted avoided premature deaths annually in France were 2,200 [1700;2700] under SP and 1,700 [1400;2000] under SN. In those series of small realistic changes in the individual diets, systematically increasing the plant share slightly limits the gain in nutritional adequacy but result in diets that are far more sustainable.

Keywords: Plant protein, diet modeling, nutrient adequacy, simple changes, sustainability parameters, French population.

1. Introduction

The Food and Agriculture Organization of the United Nations (FAO) defined sustainable diets as "*diets with low environmental impacts which contribute to food and nutrition security and to healthy life for present and future generations. Sustainable diets are protective and respectful of biodiversity and ecosystems, culturally acceptable, accessible, economically fair and affordable; nutritionally adequate, safe and healthy; while optimizing natural and human resources*" (Burlingame and Dernini 2012). The consumption of animal-based food, and particularly of meat, has been associated with negative effects on several sustainability parameters when compared to plant sources of protein (Godfray et al. 2018). Indeed, a high consumption of red and processed meats has been associated with a risk of colorectal cancer and cardiovascular diseases (Anses 2016b; Norat et al. 2005; Rohrmann et al. 2013) which are central to the disease burden in western countries (Bauer et al. 2014). Animal protein sources generate more greenhouse gas emissions (GHGE) than plant protein sources (Gerber et al. 2013), in a context where GHGE are a crucial feature of climate change issues (Cleveland and Gee 2017). With regards to the cost of diets, meat and fish are among the principal contributors while cereals and legumes contribute much less (Darmon and Drewnowski 2015). However, animal-based and plant-based food have completely different nutrient profiles and so their substitutability needs to be studied in the context of the complex dietary models that govern their intake. Indeed, because animal products are the main sources of protein, iron, zinc and vitamin B-12 in western diets, a drastic reduction of their consumption could lead to an inadequacy of these nutrients of importance to nutrition security (Phillips et al. 2015). Animal and plant protein sources also differ in terms of the contaminants profiles, with fish containing higher concentrations of dioxin, furans and methyl-mercury and vegetables or cereal-based foods containing more lead, nickel, cadmium, mycotoxins and pesticide residues (Anses 2011b; Millour et al. 2011; Noël et al. 2012; Nougadère et al. 2012; Sirot et al. 2013; Sirot et al. 2012).

Since the early 2000s, the trend towards reducing meat consumption in western countries because of the concerns regarding sustainability issues mentioned above (FranceAgriMer 2015; Godfray et al. 2018) has caused a general and gradual rearrangement in the consumption of protein sources (FAO 2018). More plant-based diets such as semi-vegetarian and vegetarian are becoming increasingly popular and they are expected to favor long term-health and be more sustainable when compared to standard western diets (Appleby and Key 2016; Orlich et al. 2013). Dietary transitions operate mainly via a series of gradual changes to individual dietary choices rather than an abrupt adherence to predefined diets such as those proposed in national guidelines (Anses 2016e; U.S. Department of Health and Human Services and U.S. Department of Agriculture 2015) or consumed by a small part of the population (e.g. vegetarians) (Niva et al. 2017). It nevertheless remains unclear how the general population might make gradual, simple and appropriate changes to their diet in order to improve its quality while reducing its content in animal foods. Recent studies have shown that the dietary patterns of individuals are key to identifying the best modifications that can be made to protein intake in order to increase overall nutrient adequacy (de Gavelle et al. 2019). As we showed in that recent study, the simple and gradual changes to protein patterns in a population would be the first steps in rearranging protein intake and can be studied by modeling unitary modifications to portion sizes with nutritional objectives such as improving nutrient adequacy. Studying the effect of constraints to achieve a higher plant:animal protein ratio could be useful to understand how this affects different sustainability endpoints.

The objective of the present study was therefore to identify pathways of gradual modifications to the consumption of protein components in the diet that could lead to improved nutrient adequacy, with or without the systematic targeting of an increase in plant protein. We thus assessed the effects of these change scenarios on several parameters covered by the FAO definition of sustainable diets: diet costs, GHGE, exposure to food contaminants and the number of premature deaths avoided.

2. Subjects and Methods

2.1. Population, food intake and dietary composition

The study sample was composed of adult participants of the Second Individual and National Study on Food Consumption (INCA2) of the French population conducted in 2006-2007, as previously described (de Gavelle et al. 2017; Dubuisson et al. 2010). In summary, we excluded adults over 65 years of age (because their nutrient requirements differ from those of younger adults) and energy under- and over-reporters identified by comparison of the reported energy intake and the basal metabolic rate, as estimated using Henry equations (Henry 2005), and a cut-off value as defined by Black (Black 2000), resulting in a final sample of 1,678 adults representative of the French population (717 males and 961 females aged 18 to 65 years old) (**Online Resource 1**). Food intakes were calculated from seven-day food records, and individual characteristics (e.g. sex, age, body weight or physical activity) were extracted from self-reported questionnaires and in-person interviews. The nutritional composition of the foods involved has already been described in detail (de Gavelle et al. 2018). In brief, data on nutritional composition were extracted from the 2016 CIQUAL (*Centre d'Information sur la Qualité des Aliments* – Centre for Information on Food Quality) food composition database (Anses 2016c), an amino acid database as previously described (de Gavelle et al. 2017), and databases on phytate (Amirabdollahian and Ash 2010), and heme and non-heme iron in animal foods (Centre d'Information des Viandes 2005; Centre d'Information des Viandes & INRA 2009; Kongkachuichai et al. 2002). The digestibility of protein (de Gavelle et al. 2017) and the bioavailability of iron (Armah et al. 2013; Hallberg and Hulthen 2000) and zinc (Miller et al. 2007) were taken into account. The percentages of animal and plant protein in each food item were obtained by assigning food items to both categories and breaking down the mixed food into ingredients from the recipes, as described in detail elsewhere (Camilleri et al. 2013). The food groups presented in this publication were adapted from the INCA2 food groups, taking account of their fat content. Indeed, meat (excluding poultry) and poultry were both split into two groups depending on whether or not fat contributed to more than 35% of energy; 71% of meat foods were "fatty meat" and 61% of poultry foods were "fatty poultry". Fish was considered as "fatty" if the EPA+DHA content was >1g/100g (Anses 2016e); 34% of fish-based foods were considered to be "fatty".

Protein foods were defined as INCA2 food items that met two criteria: 1) the percentage of energy from protein was >10%, (referring to their intrinsic protein content), and 2) the level of intake at the 90th percentile was >5g protein/portion, referring to their potential contribution to protein intake at a relatively high level of consumption. The percentages of protein foods in each food group are detailed in **Online Resource 2**.

2.2. Nutrient adequacy

Nutrient adequacy was assessed using PANDiet probabilistic scoring (Verger et al. 2012), as previously described (de Gavelle et al. 2019). Briefly, the PANDiet score is calculated as the mean of an Adequacy Sub-score (AS), which is the average of the probabilities of adequacy (PAs) of nutrients for which an Estimated Average Requirement is defined, and a Moderation Sub-score (MS), which is the average of the PAs of nutrients with an existing upper bound reference value. The reference values applied were mainly the most recent values published by the French Agency for Food, Environmental and Occupational Health & Safety (Anses) (Anses 2016a). For each individual, we therefore calculated the PANDiet score (from 0 to 100) where a higher score reflected the higher overall nutrient adequacy of the diet. Details on the PANDiet score are given in **Online Resource 3**.

2.3. Statistical analyses and models

Stepwise dietary models of changes were used to improve the initial PANDiet score calculated for each individual. The models consisted of pairing an increase in the portion size of a protein food with a reduction in the portion size of another protein food in the 7-day diets of the INCA2 individuals. Every possible paired modification was simulated, and the PANDiet score was calculated for each of them. The algorithm then selected the paired modification that most markedly increased the PANDiet score, and implemented the modification in the 7-day diet of the individual. This process was iterated 20 times. We limited the models to 20 steps in order to avoid any drastic change to the diets, as performed in other studies (Verger et al. 2014). Two different scenarios were implemented, as adapted from the scenarios previously described (de Gavelle et al. 2019).

Under the first scenario (SN, standing for scenario with no constraint), individuals could have paired modifications between two protein foods that they were already consuming. Moreover, they could reduce the portion size of a protein food consumed, while introducing a portion of food not consumed in the observed diet but consumed by >10% of individuals with a similar pattern of protein food intake. The scenario SN was the same as the one that we described in detail in a previous publication and referred to as S2 (de Gavelle et al. 2019). Under the second scenario (SP, for constrained by an increase in the plant:animal protein ratio), the same modifications were permitted but the percentage of plant protein (among total protein) in the diet of each individual had to increase at each iteration of the stepwise models, i.e. the modification was constrained by an increase in plant:animal protein. Therefore, the percentage of animal protein among total protein also decreased at each iteration. Details on the paired modifications of portion sizes are given in **Online Resources 4 and 5**.

The weighting schemes proposed in INCA2 (for unequal sampling probabilities and differential non-responses by region, agglomeration size, age, sex, occupation of the household head, size of the household and season) were used to ensure statistical representativeness. An overall level of significance of 5% was used for statistical analyses. When ANOVA identified a significant effect of the type of scenario, we used pairwise post hoc tests with Bonferroni correction to examine differences between the observed and each of the two final simulated diets. All analyses were performed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA).

2.4. Other sustainability parameters

In addition to nutrient adequacy, other sustainability parameters covered by the FAO definition were assessed in the present study: diet costs, GHGE, exposure to food contaminants and the number of premature deaths avoided. The prices of food items were obtained from the 2006 Kantar WorldPanel household consumer panel for France (Kantar WorldPanel 2006), which gives the annual expenditures on and quantities purchased of each food item available on the market by a representative sample of 12,000 French households. A price was assigned to each INCA2 food commodity and a mean price per individual and per day was calculated. Estimates of the GHGE associated to the consumption of food commodities were obtained from the database published by Hartikainen and Pulkkinen based on statistics and existing literature (Life Cycle Assessment (LCA)-based studies). The LCAs accounted for agricultural production and processing steps. A GHGE estimate, expressed in grams of CO_2 equivalents (g CO_2eq), was assigned to each of the food items in the INCA2 nomenclature using the 151 food categories in the database (Hartikaiinen and Pulkkinen 2016).

The dietary exposure of INCA2 individuals was assessed by combining individual consumption data and occurrence data from the French Second Total Diet study (TDS2). This study listed the concentrations of 440 substances (including trace elements, persistent organic pollutants, additives, pesticide residues, mycotoxins, etc.) in 212 core foods sampled in 2007-2009. The food items analyzed during TDS2 were defined from the INCA2 nomenclature and covered about 90% of the entire French diet (Sirot et al. 2009). Sampling took account of seasonality and geographic variability. The management of left-censored data under different hypotheses (lower-bound (LB) and upper-bound (UB)), the hypotheses of speciation drawn for some substances and how the estimated daily intakes were calculated are detailed in **Online Resource 6.**

The Health-Based Guidance Values (HBGVs) considered were those defined in the TDS2 (Anses 2011b) or, when more recent, values determined in Anses opinions and in the HBGV database (Anses 2016d;

Anses 2016e; Anses 2018). The risks associated with dietary exposure to each chemical were evaluated by calculating the percentage of individuals exceeding HBGVs and their 95% confidence intervals ($CI_{95\%}$), or, in the case of a benchmark dose lower limit (BMDL) by calculating the corresponding margin of exposure (MOE) according to international recommendations (EFSA 2005). In the present study, the substances considered were those for which a risk was identified in the population before or after 20 steps of the stepwise models. We chose not to consider certain pesticides which have been banned since TDS2 as the occurrence data are no longer appropriate. Moreover, those pesticides were not detected during more recent studies (Anses 2016d) using lower limits of detection than those applied in TDS2.

The PRIME (Preventable Risk Integrated ModEl) (Scarborough et al. 2014b) is a scenario model that links behavioral risk factors with mortality from noncommunicable diseases (NCD), either directly or mediated by body mass index (BMI), blood cholesterol or blood pressure. This was used to assess the effects of the modifications simulated under scenarios N and P on premature NCD mortality (mainly diet-related cardiovascular diseases, cancers and diabetes). For the analyses reported in this paper, the following dietary risk factors were included: fruit, vegetables, fiber, total fat, saturated fat, polyunsaturated fat, monounsaturated fat, dietary cholesterol and salt. The method parameters, including relative risks estimates and CI, were the same as originally reported (Scarborough et al. 2014b). The 25-65 years old French population in 2006 (63 million inhabitants) was considered in order to fit with the population in the dietary survey. The situation in INCA2 was considered as the baseline situation in France in 2006, and the scenarios SN and SP were considered as the counterfactual scenarios. The input variables were adjusted for energy intake using the residual method (Willett et al. 1997), as the PRIME model is highly sensitive to even small variations in energy intake (Cobiac et al. 2018), whereas dietary surveys involve considerable inaccuracy with respects to energy intake estimates. To assess the difference between SN and SP regarding premature deaths avoided, we calculated, for each individual at each step, the difference between SN and SP for each parameter, which we called δ_{SN-SP}. Then we added the δ_{SN-SP} to the observed value, to assess the effect of the difference between the two scenarios, and used the PRIME model to estimate the number of premature death avoided by the difference between SN and SP. A Monte Carlo simulation was performed to estimate 95% uncertainty intervals around the results. Uncertainty intervals (CI) were estimated, based on the 2.5th and 97.5th percentiles of results generated from 10^5 iterations of the PRIME models, where the estimates of relative risks used to parameterize the model were allowed to vary stochastically according to the distributions reported in the literature.

3. Results

3.1. Contribution of different food groups to total protein intake

After 20 steps in the SN, the mean (± SD) plant protein intake fell from 31.1% (±8.0) to 30.0% (±7.6) in the whole population (P<0.0001). It remained constant among men, and fell from 31.5% (±7.7) to 29.4% (±7.2) (P <0.0001) of total protein intake among women. In the SP, the mean plant protein intake rose (P <0.0001) from 31.1% (±8.0) to 37.7% (±8.2) in the whole population, and from 30.8% (±8.4) and 31.5% (±7.7) to 36.7% (±8.5) and 38.7% (± 7.9) in men and women, respectively. The mean contribution to protein intake of high-fat meat, high-fat poultry, deli meat, sandwiches and bread decreased in men and women under both scenarios. The mean contribution to protein intake of fatty fish, pizzas and quiches and legumes increased in both sexes and both scenarios. The mean contribution to protein intake of lean meat (women only), lean poultry, yogurts and prepared dishes (men only) increased in SN only, whereas the contribution of pasta and vegetables (women only) increased in SP only (**Fig. 1**).

The highest rates of increase were for legumes (+225%), fatty fish (+151%) and lean poultry (+82%) in SN and for legumes (+502%), pizzas and quiches (+190%) and fatty fish (+102%) in SP. Likewise, the highest rates of decrease were for deli meat (-52%), sandwiches (-31%) and cheese (-25%) in SN, and for deli meat (-49%), high-fat meat (-38%) and high-fat poultry (-37%) in SP.

3.2. Nutrient adequacy

The two scenarios significantly increased overall nutrient adequacy, as assessed using the PANDiet score. The mean (±SEM) increase in the PANDiet score was smaller (P <0.0001) under SP (+6.2±0.1 points) than SN (+7.5±0.1) in the whole population. The increase was also smaller (P<0.0001) under SP (5.9±0.1 and 6.5±0.1 points) than SN (6.9±0.1 and 8.0±0.1) in men and women, respectively (**Table 1**). The AS increase was higher under SN than SP in both men and women, which was explained by higher increases in PAs for EPA+DHA, iron, iodine, potassium, zinc, riboflavin, vitamin B-6 and B-12 but lower PAs for fiber and folate. The rise in the MS was similar under both SN and SP, because of similar PAs for total fat, SFA and sodium.

3.3. Diet costs

When comparing observed diets with the rearranged diets resulting from SN, the mean (±SEM) diet cost increased (P<0.0001) from €7.4±0.06/d to €8.2±0.05/d in the whole population, and from 8.4±0.07 and €6.5±0.04/d to €9.2±0.09 and €7.4±0.04/d among men and women, respectively. Under SP, the mean cost of the rearranged diets slightly increased (*P*<0.01) in the whole population (€7.6±0.05/d), did not significantly change in men and slightly increased (*P*<0.05) in women (€6.7±0.06/d). Among both men and women, the mean diet cost was lower under SP than SN (*P*<0.0001). Under SN, the increase in the intake of fatty fish (+€0.44 and +€0.35/d for men and women, respectively), prepared dishes (+€0.28 and +€0.20/d for men and women, respectively), and lean poultry (+€0.14 and +€0.30/d for men and women, respectively) contributed the most to the higher cost. Under SP, the increases in the intake of pizzas and quiches (+€0.36 and +€0.14/d for men and women, respectively) and fatty fish (+€0.28 and +€0.20/d for men and women, respectively) contributed the most to the higher diet cost.

3.4. GHGE

Under SN, mean (±SEM) GHGE levels increased (*P*<0.0001) from 5.4±0.05 kg eq. CO_2/d to 5.7±0.04 kg eq. CO_2/d in the whole population, and 6.3±0.08 and 4.5±0.06 kg eq. CO_2/d to 6.5±0.09 and 5.1±0.06 kg eq. CO_2/d, for men and women, respectively. Conversely, under SP, mean GHGE levels decreased (*P*<0.01) to 5.1±0.04 kg eq. CO_2/d in the whole population, and to 5.9±0.07 and 4.3±0.04 kg eq. CO_2 for men and women, respectively (**Fig. 2**). Under SN, the increase in the intake of lean meat (+0.10 and +0.32 kg eq. CO_2/d for men and women, respectively) and prepared dishes (+0.21 and +0.14 kg eq. CO_2/d for men and women, respectively) contributed the most to the higher GHGE. Under SP, the reduction in the intake of high-fat meat (-0.38 and -0.35 kg eq. CO_2/d for men and women, respectively) and deli meat (-0.10 and -0.07 kg eq. CO_2/d for men and women, respectively) contributed the most to the lower GHGE.

3.5. Risks related to exposure to food contaminants

A risk (as a percentage of the population >HBGV significantly different from 0) was identified for 13 substances in the observed diets or at least one of the scenarios and hypotheses for censored data **(Table 2)**. However, under the LB hypothesis, the risks related to exposure to acrylamide, cadmium, chromium VI, methylmercury and sulfites did not differ between under SN and SP and the observed diets. In SP only, the risk related to mycotoxins increased (Deoxynivalenol (DON) in men in LB and UB and T2 + HT-2 in women, in UB only). The risks related to exposure to dioxins, furans and PCB-DL increased in SN only for both sexes and the risk related to PCB-NDL increased for men only in SN. Finally, the risks related to lindane exposure decreased in SP for women only, and the risk related to nickel exposure increased in SN and was higher in SP than in SN and the observed diet in women.

The risks related to acrylamide, chromium VI, inorganic arsenic and lead were characterized using the calculation of MOEs based on BMDLs **(Table 3)**. In the observed diet, the MOEs for acrylamide at the 95th percentile were around 150 for men and women, and slightly decreased in SN and SP. The MOEs for chromium VI at the 95th percentile were between 1,641 and 2,844 for men and women under LB and UB, and were the same in the observed diet, SN and SP. Thus the MOEs were <10,000 and a risk could not be ruled out for acrylamide and chromium VI. The MOEs for inorganic arsenic at the 95th percentile ranged from 0.45 to 16.22 for men and women under LB and UB, which was too low to rule out a health risk. The MOE fell in SN and SP, leading to a higher risk related to inorganic arsenic exposure. The MOEs of lead at the 95th percentile were <10 in the observed diet which indicated a risk related to lead exposure, and decreased in the rearranged diets using both scenarios for men and women.

The differences in risk related to contaminant exposure could be explained by the differences in intake of a few food groups. Indeed, the contribution of fatty fish to exposure to dioxins, furans and PCB-DL increased from 13% in the observed diet to 21% and 31% in the rearranged diets under SP and SN, respectively. The contribution of fatty fish to methylmercury exposure also increased from 10% in the observed diet to 18% and 26% under SP and SN, respectively, while its contribution to PCB-NDL rose from 23% in the observed diet to 35% and 47% under SP and SN, respectively. The contribution of bread to cadmium exposure fell from 21% in the observed diet to 17% and 16% under SN and SP, respectively, with concomitant increases in the contributions of vegetables (from 10% to 14% and 16%), legumes (from 0% to 1% and 3%) and pizza (from 2% to 4% and 7%). The contribution of pizza to DON exposure rose from 6% in the observed diet to 13% and 20% under SN and SP, respectively. The contributions to nickel exposure of legumes (from 1% to 5% and 9%) and pizza (from 1% to 3% and 5%) increased in the observed diet, SN and SP, respectively. The contribution of pizza to inorganic arsenic increased from 2% in the observed diet to 5% and 8% under SN and SP, respectively. Finally, the contribution of legumes to lead exposure increased from 1% in the observed diet to 4% and 6% under SN and SP, respectively.

3.6. Avoidance of premature deaths

Using the PRIME model, we predicted that that the diet modifications after 20 steps of rearrangement would avoid a total of 1,667 [CI 95% 1,354;1,984] premature deaths/year in France under SN (1,325 [1,076;1,576] men and 343 [277;410] women) and 2,173 [1,701;2,637]/year in France under SP (1,666 [1,311;2,016] men and 507 [385;627] women) when compared to the observed diets (**Fig. 3**). These premature deaths were mainly linked to changes in serum cholesterol levels (44% of deaths avoided under SN and SP) and fiber intake (35% under SN and 49% under SP), mainly leading to an avoidance of cardiovascular diseases (92% of deaths avoided under SN and 89% under SP). When assessing the impact of the differences between SN and SP, we predicted that the SP would avoid 511 [281;813] more premature deaths per year than the SN.

4. Discussion

This study identified that targeting the rearrangements that increased nutrient adequacy towards an increase in the percentage of plant protein intake (i.e. in SP vs SN) led to a smaller increase in overall nutrient adequacy but better values regarding all the other sustainability parameters studied in a French representative population.

The constraint of increasing the percentage of plant protein at each step resulted in certain differences regarding the intakes of food groups: under SP, the intake of lean meat, lean poultry and yogurts did not increase, that of fatty fish increased less than under SN and the intake of pizzas, pasta, vegetables and legumes was much higher than under SN. These differences explained most of the differences between the two rearranged diets in terms of the sustainability parameters studied. Indeed, a higher intake of lean meat and poultry under SN explained the better PAs for iron, zinc and vitamin B-12, but a higher cost and higher GHGEs when compared to SP. A higher fatty fish intake under SN led to higher PAs for EPA + DHA, a higher cost of the diet and a higher risk related to dioxins, furans and PCBs when compared to SP. A higher intake of pizzas, bread, legumes and vegetables explained the higher PAs for fiber and folates, the lower cost, lower GHGEs but some trends towards higher risk regarding exposure to mycotoxins and nickel under SP when compared to SN. Under SP, the higher intake of vegetables and fiber and lower intake of dietary cholesterol explained the smaller estimated number of deaths avoided. The evolution of nutrient adequacy is particularly interesting when compared to the results of a study assessing the nutritional status of adults in France (Castetbon et al. 2009). Indeed, in this study, 8.7% of women had a mean serum ferritin < 15.0 µg/L, 6.7% of men and 5.8% of women had a deficiency in plasma folate and 2.9% of men and 3.7% of women had a deficiency of plasma vitamin B-12. Both scenarios helped to identify which dietary modifications could increase the nutrient adequacy of these 3 nutrients without decreasing the adequacy of the other nutrients, depending on whether or not individuals aim at eating more plant protein. These findings could be useful to address nutrient deficiencies in dietary transition.

How the intake of specific food groups changed in both scenarios were in line with French national dietary guidelines, which recommend limiting red meat intake and markedly reducing deli meat intake while increasing that of fatty fish, legumes and vegetables (Anses 2016e). However, as pizzas were consumed by a large part of the population in the observed diets, the pizza intake rose markedly, particularly under SP. However, it appeared that when the selected modifications consisted of an increase of a portion size of pizza (or the introduction of a small portion of pizza), 53% of the foods which had their portion size reduced in parallel were deli meats, which contained more salt and SFA and less calcium, zinc, fiber and vitamin B-9 than pizzas. As a result, our results should not be interpreted as meaning that pizzas are a “healthy food”, but that they are simple substitute when gradually reducing the consumption of deli meat in order to increase overall nutrient adequacy. Conversely, the increases in portions of legumes were mostly limited to clusters that contained legumes in their cluster repertoires. This underlines that account must be taken of both initial dietary patterns and the acceptability of the change, as we discussed previously (de Gavelle et al. 2019). Furthermore, it has been argued elsewhere that the consumption of plant protein from existing sources is more likely to increase in groups that are already consuming them (Niva et al. 2017). The change model used here indeed reflected this reality of a barrier to change by allowing increases among actual consumers or by introducing these foods into the apparent repertoire of likely consumers only. It could seem surprising that fish intake increased in SP, as there was a constraint of increasing the percentage of plant protein at each iteration. However, this was the case when a portion of animal protein food (e.g. meat) was replaced by a portion of fish with a lower protein content, which led to an increase in the share of plant protein among total protein at the end of the iteration.

The SN findings revealed that increasing overall nutrient adequacy, even by means of minor changes, led to a higher diet cost (+11%) and higher GHGE levels (+7%) than in the observed diets, which was in line the fact that the different sustainability parameters are not always compatible with each other (Perignon et al. 2017). For example, observational studies (also conducted as part of the INCA2 study) showed that diets with high nutritional quality were associated with significantly higher diet-related GHGEs than low nutritional quality diets (Vieux et al. 2013), or that the higher quality diets were associated with higher costs and higher GHGEs (Masset et al. 2014). However, Seconda et al. identified, on a large observational French cohort, that the diets with the lowest GHGE were also diets with the lowest cost and high nutritional quality (Seconda et al. 2018). Our finding that the SP diets generated lower GHGE than the SN and observed diets shows that favoring plant protein in change scenarios is a pivotal factor even when only simple and moderate changes to protein patterns are considered. This finding therefore well complements those of previous studies which demonstrated much lower GHGE and higher nutrient adequacy with plant-based diets than with typical western diets, and reductions in GHGE in the context of more radical change models involving plant-based diets (Biesbroek et al. 2018; Macdiarmid et al. 2012; Scarborough et al. 2014a; Springmann et al. 2016; Tilman and Clark 2014). In our study the parallel decrease in GHGE levels and the number of deaths avoided when systematically increasing the percentage of plant proteins (SP vs SN) was in line with previous findings regarding an increase in plant protein intake or a reduction in animal protein intake (Perignon et al. 2017). For example, under a scenario in the UK where the proportions of vegetarians was doubled and the rest of the sample adopted a dietary pattern that was low in red and processed meats, the health risk of colorectal cancer was significantly reduced (Aston et al. 2012). Likewise, in the EPIC-NL cohort, replacing 35 g of meat/day with an equal amount of vegetables, fruits, fish, or cereal/rice/couscous lowered GHGE levels and decreased the all-cause mortality risk (Biesbroek et al. 2014). The contaminants identified as involving a significant risk were almost the same as in the French TDS2 (Anses 2011b; Nougadère et al. 2012), except for nickel, for which no risk was identified in TDS2, and dioxins and dioxin-like PCBs, for which the risk was much lower in TDS2 (which was published before the HBGVs for nickel and dioxins and dioxin-like PCBs were markedly revised downwards) (EFSA CONTAM Panel 2015; EFSA CONTAM Panel 2018).

When comparing SP with SN in our study, there was a lower overall nutrient adequacy but a higher number of deaths were avoided, which could be understood as a discordance between nutrition-related indicators. However, these indicators fundamentally differ. The PRIME model is based on 7 nutrients and account for fruit and vegetable intakes, whereas the PANDiet score takes account of 32 nutrients and no food group, at least directly. The PANDiet also does not include cholesterol, as no reference value has been set by Anses (Anses 2011a), the EFSA (EFSA Panel on Dietetic Products 2010), or the Dietary Guidelines for Americans (U.S. Department of Health and Human Services and U.S. Department of Agriculture 2015). The PANDiet was not developed to predict mortality but rather nutritional status and security and it accounts for many nutrients that have little known association with a risk of chronic disease. The PRIME model was a better indicator than the PANDiet in terms of predicting the number of premature deaths avoided, but did not account for morbidity and other health effects related to the intake of other important nutrients whose intakes were increased by the PANDiet score. Nor did the PRIME model account for potential premature deaths linked to exposure to food contaminants at levels higher than HBGVs.

The strengths of this study were that nutrient adequacy was assessed using the most precise estimates of the probabilities of adequacy for each nutrient, using the most recently published nutrient reference values and taking account of the bioavailability of iron and zinc and the digestibility of protein. The rearrangements were realistic because changes to portion sizes have been acknowledged as being highly acceptable (Bianchi et al. 2018; Poquet et al. 2017; Vanhonacker et al. 2013), they remained within the food repertoires of clusters of individuals sharing similar dietary protein patterns (de Gavelle et al. 2019), and they were limited to 20 changes of portion sizes (each food having around 5 portions sizes), which limited the deviation from observed diets. Very few studies have implemented such acceptability constraints. Horgan et al. have designed constraints aiming at modeling, firstly, changes of portions sizes, then adding new foods and finally removing foods, which was in line with the hypotheses in our study (Horgan et al. 2016). However, our findings have some limitations and uncertainties. We chose scenarios which considered that the introduction of a food not already consumed by individuals but consumed by a cluster of people with a similar dietary pattern was acceptable, which had not been tested. However, this could be viewed as a probabilistic approach to identify foods that were indeed consumed by individuals but were not found in their records covering a 7-day period. The difference of death avoided between SP and SN could not be tested statistically using the confidence intervals around of avoided death generated using the PRIME model because these intervals are not related to inter-individual variability but uncertainty derived from propagation of model parameter uncertainty. However, the estimate of the premature death avoided by the difference between SN and SP had a uncertainty interval that did not include 0, and thus we concluded that SP did avoid more premature deaths than SN. The food and contamination data concerned the period 2006-2009 and did not provide precise information on the foods considered (for example if the food was organic or produced locally). This lead to uncertainties about the extrapolation of the results to the current situation, as food intake profiles have evolved since 2006 (e.g. the mean intake of organic foods has increased between 2006-2007 and 2014-2015 (Anses 2017)). Another limitation affecting this study is that we considered that the food prices were not affected by the rearrangements, which would not be true in the case of changes involving the entire population. Indeed, under our modeling of an increase in legume consumption in the population (5-fold increase under SP), the price of legumes would have increased in line with demand. Yet, changes in food intakes remain limited. The data used to assess GHGE estimates was extracted from a review study based on European data, and not only French data. There might be differences between GHGE estimates in France and the mean estimate in the European Union, as the agricultural and livestock production systems or the electricity production system are not the same in each country. However there is no complete database about GHGE of food commodities in France, and the database used in the present study was the most recent and complete. Using aggregated data from European system also allow more generalization to the GHGE impact of the changes that were identified here on a nutritional criterion. Finally, the sustainability parameters considered during this study were not all-encompassing as many environmental parameters (eutrophication, water footprint, land use or biodiversity indicators, etc.) were not included. Although reductions in environmental footprints indicators such as land and water use were associated with those of GHGE with more sustainable dietary patterns involving restrictions on animal-based foods (Aleksandrowicz et al. 2016; Pimentel and Pimentel 2003), we did not measure these indicators during the study, nor any other non-environmental indicators of sustainability.

In summary, we were able to identify that modifications to portion sizes of protein foods in the diets of French adults that markedly improved the overall nutrient adequacy. The risk related to exposure to contaminants did not evolve to any great extent except for dioxins, furans and nickel, and most changes were dependent on the intake of specific food groups such as fatty fish, bread, pizzas, vegetables and legumes. Introducing constraints to increase the intake of plant protein slightly limited the improvement in nutrient adequacy but gave rise to more marked reductions in the number of premature deaths avoided and improvements in other sustainability parameters (diet-related GHGEs and diet cost).

Contributors

The authors' contributions were as follows - E.G., J-F.H. and F.M. designed research, with the contribution of C.O. and L-G.S. E.G., M.P., P.L., J-F.H. and F.M. conducted the research; P.L., V.S. and L-G.S. provided essential materials; E.G., M.P. and P.L. analyzed data and performed statistical analysis; E.G., J-F.H., V.S., C.O., H.F., L-G.S. and F.M. wrote the paper; E.G. and F.M. had primary responsibility for final content.

Conflict of Interest

The authors declare that they have no conflict of interest.

Fig. 1 Mean contributions (%) of different food groups to protein intake in the observed diets and rearranged diets after 20 steps of the stepwise model in men (a) and women (b) in the INCA2 population (2006-2007) (n=1,678). Under scenario N, dual changes to portion size (i.e. a reduction in one protein food and an increase in another) were permitted between foods already consumed by the individual and "new" protein foods could be also introduced to balance reductions in the protein foods consumed, inasmuch as these foods formed part of the food repertoire of the cluster to which the individual belonged. Scenario P was the same except that the overall percentage of plant protein in individual diets had to increase at each iteration. "+" means that the mean contribution was higher than in the observed diets and "-" means it was lower, as tested by pairwise post hoc comparisons with Bonferroni correction (P<0.05). For example, after 20 steps under scenario N in women, lean meat contributed to 7.4% of total protein intake in men, which was significantly higher than the 5.9% recorded in the observed diets. The food groups shown contributed to >2% of total protein intake in at least one gender in the observed diets, SN or SP.

Fig. 2 Mean GHGE levels associated with observed diets and rearranged diets after 20 steps of the stepwise model under scenarios N and P among men (a) and women (nb) in the INCA2 population (n=1,678). Under scenario N, dual changes in portion size (i.e. a reduction in one protein food and an increase in another) were permitted between foods already consumed by the individuals and "new" protein foods could be introduced to balance reductions in the protein foods consumed, inasmuch as these new foods formed part of the food repertoire of the cluster to which the individual belonged. Scenario P was the same except that it was necessary for the overall percentage of plant protein in individual diets to increase at each iteration. "+" means that the mean contribution was higher than in the observed diets and "-" means it was lower, as tested by pairwise post hoc comparisons with Bonferroni correction (P<0.05). Labeled means without a common letter differ, as tested by pairwise post hoc comparisons with Bonferroni correction (P<0.05). Men and women were stratified so that the labels of men and women were independent. The food groups shown contributed to >2% of total protein intake in at least one gender in the observed or rearranged diets, and gained or lost >1% in the rearranged diets.

Fig. 3 Annual numbers of premature deaths (and uncertainty intervals) avoided in the 2006 French population by implementation of the dietary modifications under scenarios N (SN) and P (SP), as estimated by the PRIME model, for men (a) and women (b)

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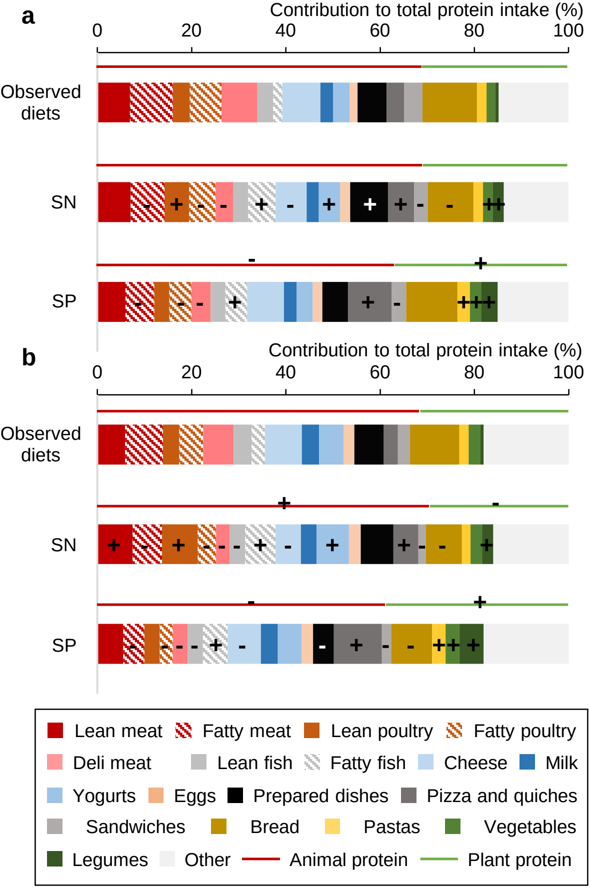
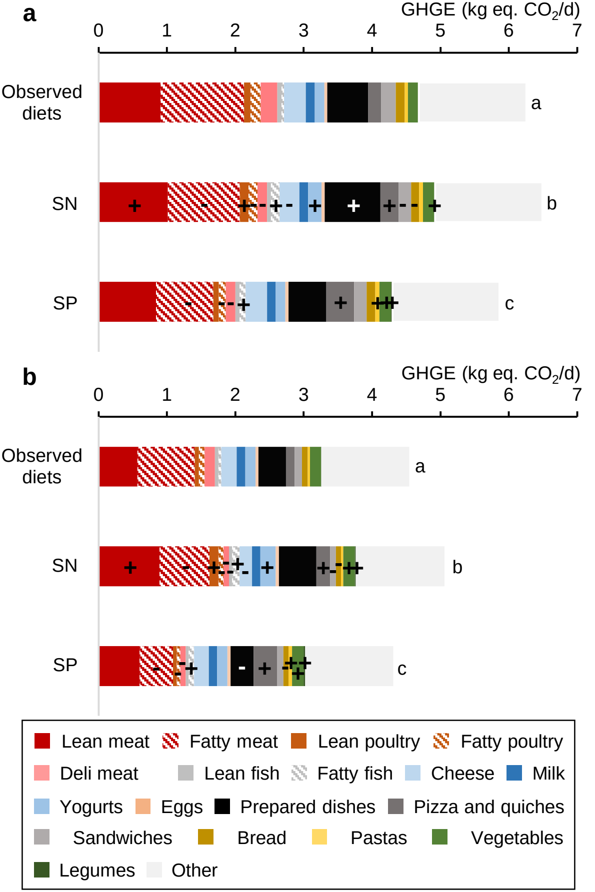

a
Contribution to total protein intake (%)
0
20
40
60
80
100
Observed diets
SN
SP
b
Contribution to total protein intake (%)
0
20
40
60
80
100
Observed diets
SN
SP
Lean meat
Fatty meat
Lean poultry
Fatty poultry
Deli meat
Lean fish
Fatty fish
Cheese
Milk
Yogurts
Eggs
Prepared dishes
Pizza and quiches
Sandwiches
Bread
Pastas
Vegetables
Legumes
Other
Animal protein
Plant protein


a
GHGE (kg eq. CO_2/d)
0 1 2 3 4 5 6 7
Observed diets
SN
SP
a
b
c
b
GHGE (kg eq. CO_2/d)
0 1 2 3 4 5 6 7
Observed diets
SN
SP
a
b
c
Lean meat
Fatty meat
Lean poultry
Fatty poultry
Deli meat
Lean fish
Fatty fish
Cheese
Milk
Yogurts
Eggs
Prepared dishes
Pizza and quiches
Sandwiches
Bread
Pastas
Vegetables
Legumes
Other

Supplementary Material.

Modeling gradual changes in protein intake to increase nutrient adequacy leads to greater sustainability when systematically targeting plant protein

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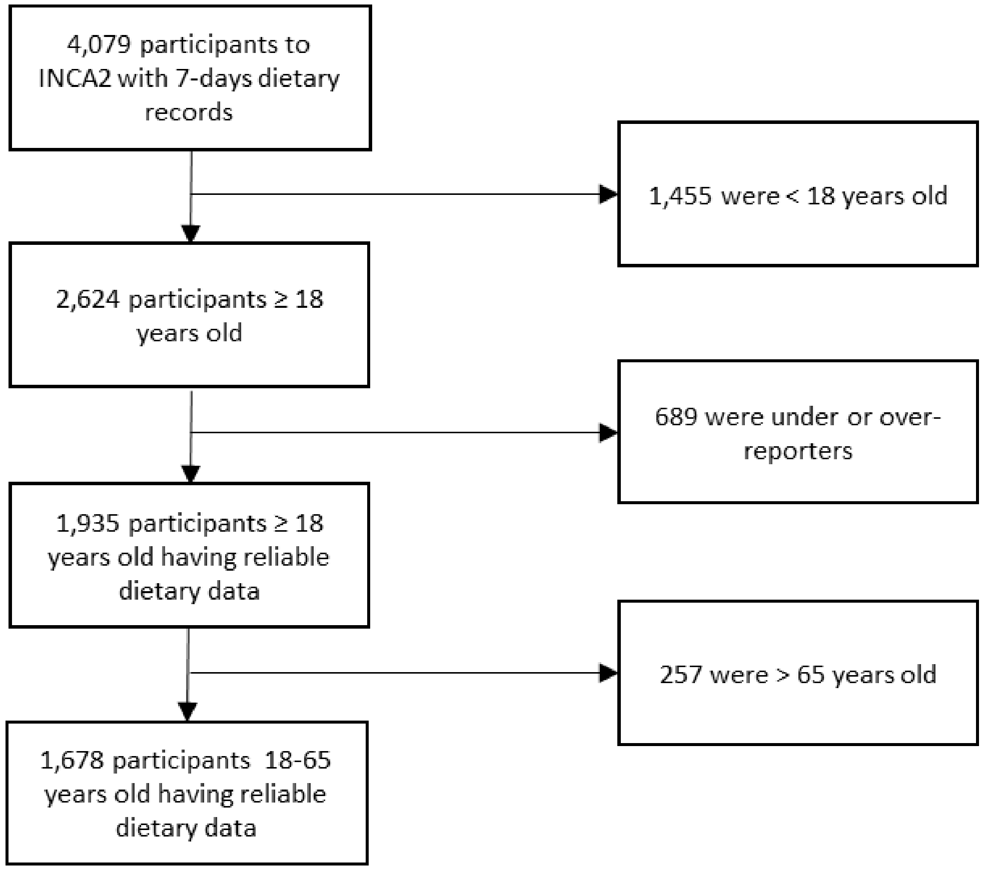


Online Resource 1. Flow chart explaining the sampling of French subjects from the INCA2 study

Supplementary Material.

Online Resource 2. Percentage of food items considered as "protein foods" by food group of the INCA2 nomenclature.

Food groups	**Number of protein food items[1] (as % of total food items in the food group)**
Meat	35 (100%)
Poultry and game	23 (100%)
Sandwiches, snacks	29 (100%)
Deli meat	53 (96%)
Fish	71 (93%)
Legumes	10 (91%)
Offal	15 (88%)
Prepared dishes	69 (88%)
Cheese	91 (87%)
Eggs and derivatives	11 (85%)
Pizzas, quiches and salted pastries	17 (81%)
Bread and bread products	18 (72%)
Milk	15 (71%)
Pastas	3 (60%)
Dairy products	36 (59%)
Rice or wheat	2 (50%)
Desserts, puddings and milk jelly	13 (41%)
Foods intended for particular nutritional uses	6 (37%)
Dried fruits and oilseeds	8 (32%)
Soups and broths	6 (32%)
Other hot drinks	3 (27%)
Other cereals	1 (25%)
Breakfast cereals	5 (21%)
Other fat	1 (17%)
Potatoes and related	2 (17%)
Coffee	1 (14%)
Vegetables (excl. potatoes)	10 (10%)
Cakes	4 (9%)
Pastries	1 (8%)
Sweet or savory cookies and bars	1 (3%)
Nonalcoholic soft drinks	1 (2%)
Butter	0 (0%)
Oil	0 (0%)
Margarine	0 (0%)
Fruit	0 (0%)
Ice cream and frozen desserts	0 (0%)
Chocolate	0 (0%)
Sugars and derivatives	0 (0%)
Water	0 (0%)
Alcoholic beverages	0 (0%)
Stewed fruit and compote	0 (0%)
Condiments and sauces	0 (0%)

Supplementary Material.

[1]Some foods were excluded as they were ingredients (e.g. gelatin) (n = 9), or were considered to be too expensive in France (> €50/kg, e.g. lobster) (n = 6). Finally, 564 protein foods were accounted for in the models.

Supplementary Material.

Online Resource 3. Implementation of the PANDiet score to the present study. The PANDiet score, expressed as the average of an adequacy subscore (accounting for 27 nutrients), and a moderation subscore (accounting for six nutrients, plus 12 potential penalty values). DHA and EPA + DHA are weighted by 1/2 as DHA is counted twice. Niacin equivalents were calculated as the sum of dietary niacin and 1/60 dietary tryptophan. The upper reference value for sugars excludes lactose. The tolerable upper intake limit for vitamin A concerns retinol only. ALA, Alpha Linolenic Acid. DHA, Docosahexaenoic Acid. EIEA, Energy Intake Excluding Alcohol. EPA, Eicosapentaenoic acid. LA, Linoleic Acid. NE, Niacin Equivalent. SFA, Saturated Fatty Acid.

PANDiet score

Average of Adequacy and Moderation subscores

Adequacy subscore

Nutrient	Reference value (/day)	Variability	Source
Protein	0.66 g/kg bw	12.5%	(FAO Expert Consultation 2011)
LA	3.08% EIEA	15%	(Anses 2011)
ALA	0.769% EIEA	15%	(Anses 2011)
DHA	0.192 g	15%	(Anses 2011)
EPA + DHA	0.385 g	15%	(Anses 2011)
Fiber	23 g	15%	(Anses 2016)
Vitamin A	570 (men) or 490 (women) µg	15%	(Anses 2016)
Thiamin	0.3 mg/1000 kcal	20%	(EFSA Panel on Dietetic Products 2016b)
Riboflavin	1.3 mg	15%	(EFSA Panel on Dietetic Products 2017)
Niacin	5.44 mg NE/1000kcal	10%	(Anses 2016)
Panthotenic acid	3.62 (men) or 2.94 (women) mg	30%	(Anses 2016)
Vitamin B-6	1.5 (men) or 1.3 (women) mg	10%	(EFSA Panel on Dietetic Products 2016c)
Folate	250 µg	15%	(Anses 2016)
Vitamin B-12	3.33 µg	10%	(Anses 2016)
Vitamin C	90 mg	10%	(Anses 2016)
Vitamin D	10 µg	25%	(Anses 2016)
Vitamin E	5.8 (men) or 5.5 (women) mg	40%	(Anses 2016)
Calcium	860 (<= 24 y.o) or 750 (>24 y.o.)	15% or 13%	(Anses 2016)
Copper	1.0 (men) or 0.8 (women) mg	15%	(Anses 2016)
Iodine	107 µg	20%	(Anses 2016)
Bioavailable iron	0.95 mg for men and non-menstruating women. Lognormal distribution for menstruating women (See formula in de Gavelle et al. (de Gavelle et al. 2018))		(Anses 2016)
Magnesium	5 mg/kg bw	15%	(Anses 2016)

Moderation subscore

Nutrient	Reference value (/day)	Variability	Source
Protein	2.2 g/kg bw	12.5%	(Anses 2016)
Total fat	44% EIEA	5%	(Anses 2016)
SFA	12% EIEA	15%	(Anses 2011)
Carbohydrates	60.5% EIEA	5%	(Anses 2016)
Sugars	100 g	15%	(Anses 2016)
Sodium	3312 (men) or 2483 (women) mg	30%	(Anses 2016)

Tolerable Upper Intake Limits		**Source**
Vitamin A	3000 µg	(Anses 2016)
Niacin	900 mg	(Anses 2016)
Vitamin B6	25 mg	(Anses 2016)
Folate	1170 µg	(Anses 2016)
Vitamin D	100 µg	(Anses 2016)
Vitamin E	300 mg	(Anses 2016)
Calcium	2500 mg	(Anses 2016)
Copper	10 mg	(Anses 2016)
Iodine	600 µg	(Anses 2016)
Dissociable magnesium	250 mg	(Anses 2016)
Selenium	300 µg	(Anses 2016)
Zinc	25 mg	(Anses 2016)

Supplementary Material.

Manganese	1.56 (men) or 1.39 (women) mg	40%	(Anses 2016)
Phosphorus	Calcium (mol) / 1.65 c.f. phosphorus section	7.5% + CV Calcium (mg)	(EFSA Panel on Dietetic Products 2015)
Potassium	2692 mg	15%	(EFSA Panel on Dietetic Products 2016a)
Selenium	54 µg	15%	(Anses 2016)
Bioavailable zinc	0.642 + 0.038 b.w.	10%	(Anses 2016)

Online Resource 4. Details about the dual modifications of portions sizes of protein foods.

As reported by Bianchi et al. (Bianchi et al. 2018), the foods were grouped into “serving size sub-groups” (n = 132), which corresponded to the sets of foods items consumed at the same time in similar amounts. As the models allowed an increase or decrease in the quantity consumed of the protein foods, possible variations in quantity were defined for each sub-group as “portion size steps”. The portion size step for some sub-groups, defined for foods sold in units or packs (e.g. yogurts) (n = 18), was the quantity in one unit or pack. For the other 114 sub-groups, the portion size step was defined as follows:

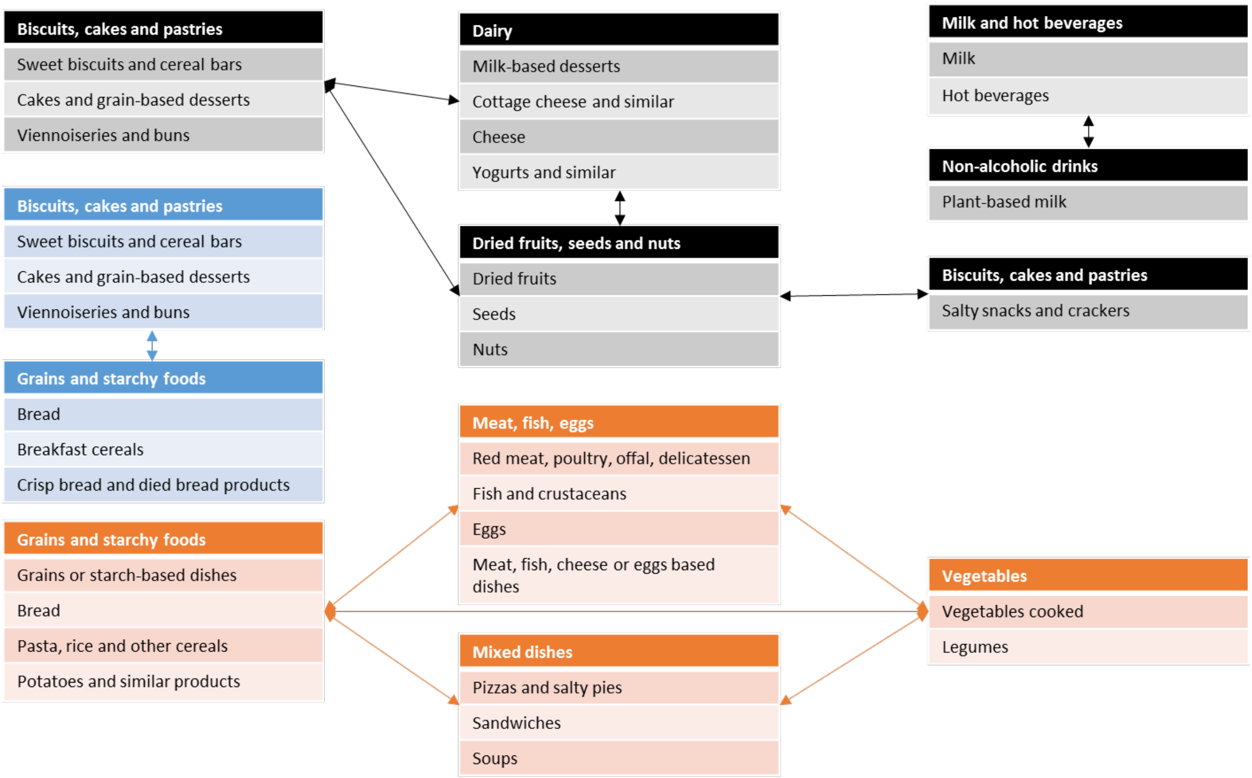
$$STEPi = \left(\frac{(P25_i - P10_i) + (P50_i - P25_i) + (P75_i - P50_i) + \ (P90_i - P75_i)}{4}\right)$$

Each step in the models consisted in decreasing (or increasing) the declared serving size by one portion size step lower (or higher). The quantity of food introduced from outside the food repertoire was one portion size step. The lowest portion permitted when reducing the portion size was 0 g and the highest portion permitted was defined as the 90th percentile of intake of the serving sub-group. The paired modifications of portions were constrained between food groups that could be substituted according to the French cultural meal scheme (e.g. modifications to portions of meat could not be paired with yogurts). The rules of compatibility between food groups for paired modifications, adapted from Bianchi et al. (2018), are described in Online Resource 5.

Finally, the steps of the model in each individual diet could not lead to an increase or decrease in the initial energy intake of more than 10%, and to protein and indispensable amino acids intakes lower than the Estimated Average Requirement.

Supplementary Material.

Online Resource 5. Diagram presenting the possible paired modifications of portion sizes (one protein food gets a lower portion and one protein food gets a higher portion), depending on the meal, between food subgroups belonging to different food groups. The name of the food group is presented in bold in the first row of each box. The names of the food subgroups belonging to the group are presented from the second row to the last row of the box. The paired modifications are allowed between protein foods within the food groups and with other food groups when an arrow connects two food groups only. The paired modifications of portions of protein foods belonging to food subgroups whose name are written in orange are allowed in lunch and dinner only, those whose name are written in blue are allowed in breakfast only and those whose name is written in black are allowed in every occasion.



Online Resource 6

Censored data are defined as values below detection or quantification limits. The censored data were processed according to EFSA, FAO and WHO recommendations (EFSA et al. 2011):

Two concentration assumptions were made: the lower-bound (LB) assumption and the upper-bound (UB) assumption. The low assumption is a scenario where the undetected values are estimated to be 0 and the detected but unquantified values are estimated to be equal to the LOD. The high assumption is a scenario where the undetected values are estimated to be equal to the LOD and the detected but unquantified values are estimated to be equal to the LOQ. The LB scenario is therefore minimalist, the UB scenario maximalist.

As only total arsenic and mercury forms were analyzed, the speciation hypothesis for inorganic arsenic and methylmercury are described as follows:

- for seafood food items, the percentage of inorganic arsenic was taken from the results of French studies on dietary exposure to arsenic (Leufroy et al. 2011; Sirot et al. 2009), as detailed in the iTDS report (Anses 2016a),
- for water, 100% of the arsenic was supposed to be inorganic,
- for the other food items, 70% of the arsenic was supposed to be inorganic, as recommended by the EFSA (European Food Safety Authority (EFSA) 2014).
- for fish items, it was considered that 100% of mercury was methylmercury and 20% inorganic mercury and for mollusks and crustaceans that 80% of mercury was methylmercury and 50% inorganic mercury. For other foods, it was considered that 100% of mercury was inorganic (Anses 2016b).

As in previous publications (de Gavelle et al. 2016; Nougadère et al. 2011), the estimated daily intake (EDI) of each substance “j” for each individual “i” was calculated with a semi probabilistic approach as follows:

$$EDI_{i,j} = \frac{\sum_{k=1}^{n} C_{i,k} \times L_{k,j}}{BW_i} \quad \textit{in µg/kg of body weight/day}$$

n: number of foods consumed by individual i for which concentration data are available
$C_{i,k}$: consumption of food item k by individual i (g/day), calculated as an average consumption on the recorded days
$L_{k,j}$: mean level of substance j in food k (if possible regional and seasonal mean) (µg/g)
BW_i: body weight of individual i (kg)

Supplementary Material.

Online Resource 7. Mean GHGE estimate of the INCA2 food groups after assignment of INCA2 items to GHGE estimates from Hartikainen and Pulkkinen 2016[1]

INCA2 food groups	Mean GHGE estimate (kg CO_2eq/kg food)
Meat	33.5
Poultry and game	6.2
Sandwiches, snacks	7.2
Deli meat	5.6
Fish	3.7
Legumes	0.5
Offal	22.2
Prepared dishes	6.6
Cheese	8.3
Eggs and derivatives	2.9
Pizzas, quiches and salted pastries	5.4
Bread and bread products	1.0
Milk	1.6
Pastas	1.0
Dairy products	1.7
Rice or wheat	1.3
Desserts, puddings and milk jelly	5.3
Foods intended for particular nutritional uses	1.0
Dried fruits and oilseeds	2.0
Soups and broths	0.5
Other hot drinks	0.3
Other cereals	1.0
Breakfast cereals	1.1
Other fat	8.6
Potatoes and related	0.8
Coffee	0.6
Vegetables (excl. potatoes)	1.3
Cakes	2.1
Pastries	2.1
Sweet or savory cookies and bars	2.1
Nonalcoholic soft drinks	0.6
Butter	9.5
Oil	3.4
Margarine	1.7
Fruit	0.7
Ice cream and frozen desserts	6
Chocolate	3.1
Sugars and derivatives	1.0
Water	0.1
Alcoholic beverages	1.5
Stewed fruit and compote	1.5
Condiments and sauces	1.1

[1] The mean GHGE estimate was weighted by the total intake of the food item in the INCA2 study

Supplementary Material.

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