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Abstract

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- Aim: to analyse the dietary habits, alcohol consumption, healthy eating index and student
- 4 performance of a sample of students at a Spanish university to determine if their intake
- 5 of nutrients reach optimal levels for fertility.
- 6 Methos: Descriptive cross-sectional study of female university students in Madrid, data
- 7 were collected in Nutrition classes. Participants were 470 women nursing studying at a
- 8 private university in Madrid, non-random sample was used. An AUDIT test was
- 9 conducted to determine alcohol consumption. Habits and dietary assessment with a three-
- 10 day record using DIAL® program to know main nutrients intake. Quantitative variables
- 11 appeared as mean ± standard deviation, adjusted for all pairwise comparisons using the
- 12 Bonferroni correction Statistical analysis was performed using SPSS 25®.
- 13 Results: The majority of the Spanish university women of reproductive age participating
- 14 in the study did not have the recommended intake of some macro and micronutrients
- 15 carbohydrates, vitamins D and B9, Mg, Fe, and I.
- 16 Conclusions: This study opens further lines of research. It is necessary to carry out
- 17 qualitative research into students' self-perception and their dietary and sleeping habits.
- 18 Addressing ways to improve food access, dietary quality, and healthy lifestyle, should be
- 19 focused on future intervention programs and policies for college students.
- 20 Key words: women, Spanish university students, childbearing age, nutrients, sleep

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1. Introduction

- A healthy lifestyle at a young age is an important factor in enjoying a long and
- 24 healthy life. This lifestyle includes a healthy diet, consuming the appropriate number of
- 25 healthy foods with an optimum protein-carbohydrate ratio, regular physical activity, and
- 26 moderate alcohol consumption (Capurso, 2021). The eating habits of university students
- 27 often depart from the recommendations of medical authorities and nutrition experts and
- 28 their nutritional habits were moving away from the traditional Mediterranean diet towards
- a more western type of diet during the transition from adolescence to adulthood. The

Mediterranean diet is characterised mainly by high intake of vegetables, fruits, nuts, cereals, legumes and fish, and low consumption of meat and poultry (Trichopoulos & Costacou, 2003).

Diet is a modifiable lifestyle factor linked with fertility (Gaskins & Chavarro, 2018). The adequate intake of monounsaturated fatty acids, the choice of plant rather than animal proteins, the use of dietary supplements containing iron and folic acid, vitamin 212, the sufficient intake of antioxidants and a well-balanced diet all seem to play an important role in preventing infertility in women of reproductive age and may help to prevent gynecological disorders (Castelló et al., 2014)(Smits et al., 2018)(Gaskins &

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37 38 Chavarro, 2018). It appears that higher intake of Polyunsaturated Fatty acids (PUFA), 39 specifically omega 3 fatty acids, can enhance female fertility(Gaskins & Chavarro, 2018). 40 It has also been observed that high intake of saturated fats, trans fatty acids and animal 41 42 proteins can be detrimental to fertility, while the intake of complex carbohydrates, fiber, monounsaturated fats and omega-3 fatty acids can be beneficial. Also, it is essential to 43 have an adequate intake of folic acid, vitamins B12, A, D, C, E, calcium, iron, zinc, 44 selenium and iodine to prevent impairments in fertility(González et al., 2018). Adherence 45 to a Mediterranean dietary pattern was associated with better fertility; regarding other 46 gynecological disorders(Onieva et al., 2020)(Fernández et al., 2018), observed that the 47 risk of endometriosis was inversely related to vegetable and fruit consumption in the 48 49 Italian population(Parazzini et al., 2004). For women with pelvic pain has been observed that those who received vitamins C and E reported less pain or dysmenorrhea compared 50 to the placebo group (Santanam et al., 2013) while for vitamin D there were heterogeneity 51

of results(Gaskins & Chavarro, 2018). Specifically in Spanish university students, an

association has been found between lifestyle and menstrual problems; typical Spanish

foods part of a Mediterranean diet, such as extra virgin olive oil and strawberries, may

influence in menstrual cycles and menstrual pain(Onieva et al., 2020)(Fernández et al., 2018), although more study is necessary.

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There are several barriers to following a healthy lifestyle for nursing students, especially the lack of time and motivation due to their stressful academic studies, even though they recognize the importance of a healthy diet (Fernández et al., 2020). Some studies found a relationship between short sleep duration and poor diet: fewer servings of fruits and vegetables, whole grains and beans and overall poorer dietary quality (Kim et al., 2011). It has also been observed that compared to normal sleepers (7-8 h), short sleepers (5-6 h) reported higher intakes of absolute protein, carbohydrate, and total fat but a lower intake of dietary fibre, whereas very short sleepers (<5 h) reported lower intakes of protein, carbohydrates, dietary fibre, and total fats(Grandner et al., 2013). In a study of adolescents across several European countries, those who slept <8 h were more sedentary and demonstrated a decrease of healthy food intake (Bel et al., 2013). In Spanish study 25% of women skipped breakfast, mainly young women, with no differences between weekdays and weekend days(Ruiz et al., 2018). It has been observed that shorter sleep durations are associated with unhealthier dietary habits or skipping breakfast (Dashti et al., 2015)(Otsuka et al., 2019). Skipping breakfast has been associated with menstrual problems(Onieva et al., 2020). The number of studies investigating the role of individual dietary factors and overall dietary patterns in cases of sleep disorders provide an interesting body of evidence. The intake of specific nutrients such as n-3 polyunsaturated fatty acids, B vitamins, zinc, and magnesium have also been linked to brain functions. B-group vitamins may modulate cognitive performance and improve cerebral and cognitive functions and calcium, potassium, and magnesium can

modulate sleep(Lassale et al., 2019) (Smith & Refsum, 2016).

Nursing students tend to be predominantly women of childbearing age and during their training they acquire knowledge about nutrition and a healthy lifestyle, although they may depart from the recommendations received in their academic training(Rizo, 2014)(Sánchez et al., 2015)(Montero & Úbeda, 2006).

The objective was to to determine if nutrient intake reach an optimal value in terms of fertility in nursing female students.

2. Methods

2.1 Study, Participants and Procedure

This descriptive cross-sectional study was conducted in Nursing students at a private university in Madrid, a non-random sample was used. The students were recruited at classes and the Nutrition professor provided information about the purpose of the study. Those who wished to voluntarily take part in the study, provided a written consent before the starting. The data was collected during class time and analyzed anonymously. The study was conducted in March-April 2021-The data was uploaded onto a database for analysis. The academic performance was calculated as the average score of all subjects studied.

Inclusion criteria was to be a female Nursing student. Exclusion criteria were based on age (> 30 years), missing data, as well as unusual energy intake (either too low or too high). The total initial sample consisted of 581 students and after discarding students and uncompleted tests, a final simple of 470 women were included in the study.

Questions relating to daily hours of sleep were also included in this section and are divided into: > 7, 6-7 and < 6 hours sleep(Jahrami et al., 2019).

Participants completed self-reported sociodemographic information on their age, course, nationality, height, weight. The participants took themselves their height, weight,

waist and hip measurements and entered the results into the questionnaires. These variables were used for the body mass index (BMI) analysis (Table 1). The mean age of the students was $21.2 \pm 3.0 (18-30 \text{ years})$.

With respect the academic results, in Spain marks correspond to: 1 good grades (7-10); 2 average grades (5-6.9) and 3 poor grades (≤4.9)(López-Moreno et al., 2021).

2.2. Dietary assessment and healthy eating index

 Food intake was assessed using three-day food records (two weekdays and one weekend day/holiday), after receiving instructions during class time. The dietary intake of some nutrients was compared with age and sex-specific requirements of these, according to the dietary recommended index (DRI) for Spanish population(Moreiras et al., 2016). The number of nutrients with inadequate intake was determined for each participant. Nutrient intakes and healthy eating index were then determined using Spanish DIAL® software (Ortega et al., 2007). The HEI score is: 50 inadequate, 50–70 acceptable-good and 70 very good-excellent(Ortega et al., 2007).

2.3. Habits and food consumption

We used the following questions: Are you following a diet?; Do you have breakfast every day?; Do you reduce consciously your food intake?; Are you on diet now or during last months?; Do you take something to reduce appetite?. Participants give a response with answers where Yes (1), No (2) and Sometimes (3). Other questions were:

How do you consider your weight? and participants give a response with dichotomic answers where: overweight (1), slightly overweight (2), normal (3), slim (4), very slim (5). How do you consider your lifestyle?: (0) unhealthy, (1) healthy and (2) very healthy. The frequency of consuming foods was assessed on an ordinal scale: never (0), 1-3 times

a month (1), once a week (2), few times a week (3); once a day (4) and few times a day 128 (5) of different food groups(Robledo et al., 2014). 129 130 131 2.4. Alcohol consumption To assess alcohol consumption and behavior, the 10-question AUDIT was used 132 (Bador et al., 2001). The AUDIT test was used to determine the alcohol consumption 133 habits of the participants. The test consists of 10 items, 8 of which are on a Likert scale 134 of 5 categories from 0 (never/1 or 2 units) to 4 (daily/10 or more units). The two remaining 135 items also use a Likert scale but with 3 categories ordered from 0 to 2. The response from 136 participants allows them to be classified into three levels: without risk of dependence, at 137 risk consumption and probable alcohol dependence syndrome (ADS). 138 139 140 2.5. Ethical consideration The study was approved by the Ethical Committee of University (16/2021), and it 141 fully complied with the Helsinki Declaration. 142 143 2.6. Statistical Analysis 144 The qualitative variables were presented as frequencies and percentages while 145 quantitative variables appeared as mean±standard deviation, adjusted for all pairwise 146 147 comparisons using the Bonferroni correction All analyses were performed using the IBM SPSS statistical software package, version 25®. 148 149 150 Figure 1. Flow chart of the study 151 152

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154	3. Results
155	A total of 470 female university students participated in this study with an average
156	age of 21.2±3.0. The average BMI was 21.8 ± 2.5 (Table 1), 7.7 % of participants were
157	classified as underweight, 10.6% as overweight-obese and 81.7% as normalweight.
158	Waist/hip ratio (WHR) was 0.79 \pm 0.9; 60.6% students had healthy ratios while 39.3% had
159	unhealthy ratios. With respect to a cademic performance, 48.3% obtained average grades,
160	28.7% had excellent grades and $23%$ had failing grades. (Table 1).
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162	Table 1
163	Anthropometric characteristics, sleep hours, academic performance, and physical activity
164	of nursing students.
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166	Regarding the intake of breakfast (Table 2) we found that 47.6% of nursing
167	students do not have breakfast every morning. HEI was acceptable, representing 60.2%
168	of the total students (Table 2).
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170	Table 2
171	Descriptive data of breakfast habits, reduce intake of food or take substances to help
172	weight control and personal considering the weight.
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175	Table 3 shows the energy, macronutrient and micronutrient intake of female nursing
176	students. Details of the findings are shown in Table 3.

177 Table 3 178 179 Mean daily intakes of macro and micronutrients from the 3-day food diary record. 180 181 182 Table 4 shows the intake of nutrients with respect to the BMI of nursing students. We note that those who were underweight have less intake of: total energy, % fat, vitamin 183 B9, Fe, Mg and I. When the students were overweight, we observed higher intakes than 184 normal of total energy, proteins and fat %E , trans fatty acids, Na and K and less intake 185 186 of vitamin A, Fe and Mg. 187 188 189 Table 4 Macro and micronutrient intake with respect Body Mass Index (BMI) in nursing female 190 191 students. 192 193 Table 5 shows the results of AUDIT test. Most students need alcohol education 194 195 (68.7%) and only 10% were non-drinkers. We also observed that 58.7% were binge 196 drinkers. According to the question 3 of AUDIT test on hazardous drinking, we observed 197 that monthly 27.2% of the students had ≥ 6 alcoholic drinks. We found a correlation (p<0.05) between students with worse academic marks who drank more alcoholic 198 beverages (data not shown). See more details in Table 5. 199 200 201 Table 5

202	AUDIT test, AUDIT-C test and AUDIT-3 in female nursing students.
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205	The pattern of food intake was characterized by high daily intakes of fruits and
206	cereals, 1-3 times/week of vegetables, meat, fish, legumes and soft drinks and sometimes-
207	never for nuts. See Table 6 for more details.
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209	Table 6
210	Frequency of food intake among female nursing students
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213	In Table 7 we observed that BMI is significative correlated (p<0.05) with HEI
214	and we observed significative correlations between macronutrients. Table 8 shows sleep
215	time and nutrients, details of the findings of each dimension are shown in Table 8.
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218	Table 7
219	Bivariate correlations between BMI, %E of macronutrients, AUDIT and Healthy eating
220	Index.
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224	Sleep time in university students, macro, and micronutrients intake.
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4. Discussion

The aim of the study was to evaluate the nutritional habits of female university students of childbearing age. In our study, participants were generally of normal weight; 7% of the students were underweight, 10.6% overweight/obese and 81.7% were of normal weight, these figures were similar to previous studies carried out in Madrid in both private and public universities in terms of weight perception, some 64% of the students perceived they had a normal weight, like results generally found among university students in Madrid. (Robledo et al., 2014). Regarding nutritional habits, half of the students had breakfasted every day, nearly a quarter were on diet, two-thirds thought they had a healthy lifestyle and one third had an acceptable HEL. In the present study AUDIT test 58.7% of women were binge-drinkers and 68.7% need alcohol education. Mean time of sleep of the students were ≤6 hours/day₂, less than a quarter were very active and had a healthy eating index.

With respect to their lifestyle, 14.3% answered that it was inadequate and 47.6% did not breakfast every day. According to their food intake, over a quarter eat fruit every day, less than a quarter eat vegetables and around the half of the students eat cereals daily. Some 85.7% eat meat and fish and less than a quarter eat legumes 1-3 times/week. Nearly half of the students drink soft drinks with 45% reporting they have soft drinks sometimesnever. Nutrients with intakes \leq 30% RDI: vitamin D and omega 3. Intakes \leq 80% RDI: omega 3, vitamins D, E and B9, minerals: Mg, Fe, K, Zn and I. Our findings show that half of the students breakfasted daily, Fujiwara et al. reported that skipping food for aesthetic reasons during adolescence could have future negative effects on their fertility (Fujiwara et al., 2009). More than 50% considered that they had a healthy lifestyle,

with an acceptable HEI and normal weight. It is important to note that one third of students considered themselves overweight. Regarding alcohol consumption, the AUDIT score was like a study conducted in Madrid with health sciences students (Marchena et al., 2020). Both found female students drink large quantities of alcohol, which may impact their fertility. A high percentage of university students sleep less than recommended for this populations, and this may also affect their fertility (Stocker et al., 2021).

For body weight, no association was observed in this study. Similar results were obtained in a recent study of nursing students(López-Moreno et al., 2021a) and among Moroccan students(Benaich et al., 2020).

Alcohol consumption has been increasing among women, The AUDIT score was like that of a study conducted in Madrid (Marchena et al., 2020). The results show that 68.7% are not at risk of problematic alcohol consumption and 3.6% have a high risk of having alcohol dependence. Among this group it is necessary to suggest the need to reduce their alcohol intake. Women have greater vulnerability to the effects of alcohol and the association found between alcohol consumption and difficulties in getting pregnant deserves further attention (Míguez & Permuy, 2017). Due to the pandemic situation, university students spent less time at the university which may have affected their alcohol consumption, leading to a higher risk of drinking.

Binge drinking refers to drinking of ≥4 alcoholic beverages in a single session.

Our students showed a rate of binge drinking of 10.2% compared with data from Madrid, which showed a mean score for binge drinking of 15.6% (Observatorio Español de Drogas y Adicciones, 2021). Among our sample we found an inverse relationship between alcohol intake and academic performance (López-Moreno et al., 2021).

Overall, our data presents acceptable values and, in many cases, lower values than those observed in the survey carried out among young people in Madrid (Robledo et al., 2014);

in this study, the average intake of fruit and vegetables was 2.4 times/week, with a daily consumption of 31.8%, compared to 5.2 in the regional survey and with 57.7% consuming them daily. Regarding vegetables, women ate they 2.4 times/week and 18.4% ingested them daily, data that contrasts with those observed in university students of Madrid, which was 4.4 times/week and 33.3% of them took them daily. Cereal consumption was 1.8 times/week, with 53.1% reporting daily consumption; this intake is lower than that obtained in the survey of the Community of Madrid showing consumption by university students of 5.4 times/week and 60.4% daily consumption (Robledo et al., 2014). For meat consumption, our findings were like the Community survey of 2.8 times/week. Legume consumption was slightly higher in our study at 2.9 times/week compared to 2 times/week in the Community survey. This was also the case with the consumption of fish, slightly higher in our study at 3 times/week compared to 1.6 according to the Community survey. For nuts, our findings were higher at 3.2 times/week compared to 1.1 times/week for university students from Madrid (Robledo et al., 2014). The consumption of soft drinks was similar, with an average intake of 3.1 times/week. However, unlike the previous survey, 9.6% of our students consumed soft drinks daily compared to 22.7% of our students.

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Mean sleep time in our study was ≤6 hours, similar results were obtained in other studies (Özdişli et al., 2021)(Peltzer & Pengpid, 2019)(de la Portilla et al., 2019). Our study also observed a lack of association of BMI with short and adequate sleepers, similar results were observed by others (Mondin et al., 2019). That non-significant relationship between BMI-sleep time, may be because short sleepers were more actives students (data not shown). Sleep is very important to physical and emotional health. We did not detect differences between sleep duration and the intake of nutrients. Nevertheless, individuals who slept less than 6 hours had significantly higher intakes of vitamin A, folic acid, iron,

selenium and iodine, than those who slept more hours. Some aspects of health and student functionality are related with sleep disorders as explained above, but there has been little study into the relation between poor sleep and fertility(Kloss et al., 2015). Sleep and sleep disturbances are among the most significant health and wellbeing factors related to menstruation, pregnancy and menopause. If sleep directly affects reproductive hormones and/or related physiological process, a direct relation may be expected between sleeplessness and fertility, perhaps because sleep quality is important to oocyte health (Stocker et al., 2021)(Yanık & Tokat, 2021).

Sleep has been studied in young adults in relation to nutrition and physical activity, and adult cardiometabolic risk (RCM)(Ames et al., 2018). Follow-up was conducted before and after university graduation, finding that the duration of sleep, nutrition and physical activity could predict adult RCM. The levels of physical activity and nutrition predicted the RCM for women and men, and in women the duration of sleep was also a predictor. Short, poor-quality sleep is associated with high blood pressure.

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High consumption of fruit, vegetable, and salt during adolescence is associated with lower systolic blood pressure in young adulthood and the duration of sleep with lower RCM risk(Ames et al., 2018).

Diet is known as modifiable lifestyle factor related with fertility (Gaskins & 2 Chavarro, 2018)((Panth et al., 2018); it has been observed that higher protein intake is associated with an increased risk of ovulatory infertility; higher intake of unsaturated fat instead of carbohydrates, is associated with an increased risk of ovulatory infertility. In this study, the students had high intake of protein (115%) and lipids (118%), above recommendations. The intake of carbohydrates, depending on quantity and quality, can have an impact on ovulatory infertility. Carbohydrate intake with a high dietary glycemic load is associated an increased risk of infertility due to anovulation in healthy

women(Chavarro et al., 2009). However, there is no data on the intake of simple carbohydrates which are deleterious to fertility. Omega 3 fatty acids are also important in improving fertility in women (Panth et al., 2018), others noted that high intake of PUFA (omega 3 fatty acids) and low intake of trans fatty acids may enhance female fertility (Gaskins & Chavarro, 2018).

According to micronutrients, the intake of vitamin B9, was insufficient in this study and it has been reported that lower intake of this vitamin is related with sporadic anovulation among young healthy women (Gaskins et al., 2012). With respect folic acid, we found negative correlation with soft drinks (p<0.05) and positive with iron and magnesium (p<0.001). According to vitamin D, 39.3% only have the recommended intake and its deficiency may be detrimental to <u>fertility</u>.

Adherence to healthy diet is related to better fertility in women(Gaskins & Chavarro, 35

2018). A diet rich in fruit and vegetables may help to prevent gynecological disorders, compared 24
to a diet high in animal or dietary fats, red meat, and alcohol (Harris et al., 2018). There is evidence that oxidative stress plays an important role in female fertility and could be an inexpensive treatment to improve fertility outcomes(Smits et al., 2018). Vitamins play an essential role in maintaining health and preventing disease. The intake of vitamins in this study was vitamin C>vitamin A> vitamin E. Our students reach 78.9% RDI and thus may have problems with their future fertility if their iron intake remains insufficient.

The interrelation between different nutritional habits and lifestyles and their potential in improving health and fertility has been demonstrated, and several studies with university students have been conducted with novel approaches discussed below (Semsarian et al., 2021).

The study has certain limitations. It was only conducted with women and some data were self-reported. Another potential limitation was that the results cannot be generalized, and it is necessary to extent research to students of different degree programs.

One strength of our study is that includes dietary intake and lifestyle characteristics collected prior to attempting pregnancy. As future lines of research, it would be interesting to explore the self-perception of participants of their own habits and to analyze the habits of young men, also involved in the issue of fertility. It is possible to improve the health and wellbeing of female students. Physical activity and better nutritional habits can lead to improved fertility although more research is necessary. Further recommendations are the implementation of awareness campaigns on the importance of diet through multi-level nutritional educational intervention promoting healthy behaviour.

5. Conclusions

Most Spanish university women of reproductive age participating in the study did not have recommended intake of carbohydrates, vitamins D and B9, Mg, Fe and I. This is despite that fact that as nursing students they have knowledge about nutrition and should have higher scores than other groups of similar age.

Given the importance of diet to fertility and gestation it may be necessary to develop strategies to raise awareness of these issues both among healthcare professionals and the general population. Messages should emphasize the importance of a healthy lifestyle, diet and sleep patterns to fertility and motherhood. Further studies are warranted to confirm and extend these findings.

This study opens further lines of research. It is necessary to carry out qualitative research into students' self-perception and their dietary and sleeping habits. It is also important to include male students in these studies given that fertility is an issue for both sexes. Similarly, further research into sleep and fertility is recommended to better understand the complex relationship between these two issues.

377 378 **Declaration of Competing Interest** 379 The authors declare that they have no known competing financial interests or personal 380 relationships that could have appeared to influence the work reported in this paper. Commented [MIL(1]: We just added competing interest as Reviewer 1 suggested 381 382 Acknowledgements 383 384 The authors appreciate the support from Universidad Francisco de Vitoria, for the research (UFV2021-39). 385 386 References 387 Ames, M. E., Leadbeater, B. J., & MacDonald, S. W. S. (2018). Health behavior 388 changes in adolescence and young adulthood: Implications for cardiometabolic risk. Health Psychology, 37(2), 103-113. https://doi.org/10.1037/HEA0000560 389 390 Bador, T.F., Higgins-Biddle, J.C., Saunders, J.B., Monteiro, M.G.& Organización Mundial de la Salud (2001). AUDIT. Cuestionario de Identificación de los 391 392 trastornos debidos al Consumo de Alcohol. Pautas para su utilización en Atención 393 Primaria. Organización Mundial de la Salud , WHO/MSD/MSB/01.6a, 40 pp. https://apps.who.int/iris/handle/10665/331321 Field Code Changed 394 Capurso, C. (2021). Whole-Grain Intake in the Mediterranean Diet and a Low Protein to 395 Carbohydrates Ratio Can Help to Reduce Mortality from Cardiovascular Disease, 396 397 Slow down the Progression of Aging, and to Improve Lifespan: A Review. Nutrients 2021, Vol. 13, Page 2540, 13(8), 2540. https://doi.org/10.3390/NU13082540 398 399 Castelló, A., Pollá, M., Buijsse, B., Ruiz, A., Casas, A. M., Baena-Cañ Ada, J. M., Lope, 400 V., Antolı'n, A., Ramos, M., Muñoz, M., Lluch, A., De Juan-Ferré, A., Jara, C., Jimeno, M. A., Rosado, P., Diáz, E., Guillem, V., Carrasco, E., Pé Rez-Gó Mez, B., 401 ... Martı'n, M. M. (2014). Spanish Mediterranean diet and other dietary patterns and 402

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