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Zinc content in different muesli samples

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Abstract. There is no specialized zinc storage system in the body, therefore there must be a daily intake of zinc to achieve a steady state. Long-term zinc deficiency due to inappropriate nutrition may result in immunological or autoimmune diseases. The aim of this study was to develop muesli with naturally high zinc content. Zinc is found in various plant-based foods as grain flakes and seeds which often are one of the raw materials for muesli. Muesli is one of the grain-based food trends nowadays as people`s life habits are changing due to fast-paced life, still, it is important to obtain all nutrients. According to zinc content in raw materials five different muesli samples were prepared, from all samples, muesli with the highest zinc content with 3.80 mg 100 mg^{-1} was chosen for further analysis. Zinc bioavailability is assessed through the determination of absorbability. The kinetic *in vitro* intestinal digestion suggests how much zinc is released during digestion and could be absorbed in the small intestine. The results show that during digestion approximately 22% of zinc was absorbed in the small intestine. Such characteristics of *in vitro* digestion test shows that by one meal (50 g of muesli) it is possible to replenish our body zinc level by 0.42 mg. To check additional nutrient content in selected muesli samples different mineral and vitamin analyses were done. Mineral and vitamin content in the muesli sample was calculated according to their content in raw materials. Per portion, muesli is source of iron, magnesium, phosphorus, zinc, vitamin B1, vitamin B2, vitamin B3, vitamin B6 and vitamin E.

Key words: absorption, digestion, in vitro, muesli, zinc.

INTRODUCTION

Recent years plant-based product purchasing is growing since vegetarian and vegan diet is getting more popular. Vegetarians and vegans may potentially be at risk in zinc deficiency (Saunders et al., 2013) well planned diets can provide amounts from plant sources (Rose & Strombom, 2019), yet it is important to consume more products with naturally high zinc content. The role of breakfast cereals in balanced diet has been recognized for many years. Breakfast cereals are important source of vitamins and minerals, also, potentially important source of antioxidants and phytoestrogens, and wholegrains (Williams, 2014). Zinc is found in various plant-based foods as grain flakes and seeds which often are one of the raw materials for muesli.

Zinc is second most abundantly distributed trace element after iron. Zinc functions in biology are numerous but can be separated into three main categories: catalytic, regulatory, and structural roles (Bhattacharya et al., 2016). It catalyses enzyme activity, contributes to protein structure, and regulates gene expression (Saper & Rash, 2009; Kambe et al., 2015). Zinc is cofactor for over 1,000 enzymatic reactions and is necessary for over 2000 transcription factors. Zn-fingers protein function for DNA interaction, RNA packaging, activation of transcription, regulation of apoptosis, folding and assembly of protein, and lipid binding. Also, around 10% of human proteins bind to zinc. This is the reason why zinc is associated with a wide variety of organic activities such development, differentiation, and cell growth (Ogawa et al., 2017).

Zinc absorption occurs throughout the small intestine. Approximately 30% of zinc in the diet is absorbed in the small intestine. Of the absorbed zinc, 80% and 20% are bound to blood albumin and alpha-2-macroglobulin. However, this protein-bound zinc comprises only 0.1% of the total body zinc, indicating that only this amount is replenished daily. This serum zinc is delivered and stored in peripheral tissues including skeletal muscle (60%), bones (30%), skin (5%) and liver (5%). Moreover, the skin is the third among tissues with the most abundance of zinc in the body. By strict regulation, zinc is stably maintained in the human body in a weight 2–3 g (Ogawa et al., 2016). It is generally agreed that most of the dietary zinc is absorbed in the proximal small intestine, through a transcellular saturable carrier-mediated mechanism. Even though the jejunum appears to have the highest rate of zinc absorption, the duodenum is first exposed to zinc during the postprandial period and is likely to also contribute to zinc absorption (Gopalsamy et al., 2015).

There is no specialized zinc storage system in the body, therefore there must be a daily intake of zinc to achieve a steady state. Long-term zinc deficiency due to inappropriate nutrition may result in immunological or autoimmune diseases. During zinc deficiency different immune functions are decreased. All these impaired functions are completely restored by zinc supplementation (Rink & Gabriel, 2000). Zinc bioavailability should also be considered which is determined in three basic stages - absorbability, mucosal transfer into systemic circulation and utilization within the body. As for human dietary studies, bioavailability is assessed through the determination of absorbability (Bel-Serrat et al., 2014). Phytate have considerable impact on percentage of zinc accessible for absorption. It can bind zinc in the intestinal lumen and form an insoluble complex that cannot be digested or absorbed because humans lack the intestinal phytase enzyme. Inhibitory effect on zinc absorption in adults can be substantial as there is no evidence of an adaptive response to habitual high-phytate intakes (King, 2016). Still some studies show that the total amount of zinc in a meal may have greater effect on zinc absorption than the presence of phytate (Saunders et al., 2013).

The aim of this study was to develop muesli with naturally high zinc content. Zinc is found in various plant-based foods as grain flakes and seeds which often are one of the raw materials for muesli. As previously mentioned, Bel-Serrat et al. (2014) reported that in human dietary studies, zinc bioavailability is assessed through the determination of absorbability. The kinetic *in vitro* intestinal digestion suggests how much zinc is released during digestion and could be absorbed in small intestine. *In vitro* digestion could be helpful after different high nutrition product development to formulate product which would have effect on consumer health. There are not many studies about zinc *in vitro* bioavailability from muesli or other grain products. This study will show the amount of zinc absorbed after one muesli portion. To see additional nutrient content in selected muesli sample, different mineral and vitamin analysis were done.

MATERIALS AND METHODS

Muesli development experiments were carried out at the laboratory of Felici LLC. Three repeated *in vitro* gastrointestinal digestion tests were carried out at the laboratory of Kauna Technical University (KTU). All ingredients used for muesli sample preparation and analysis were supplied by Felici LLC.

Raw material and product analysis

To check zinc content six different whole grain samples and two seed samples were tested. Muesli samples were prepared according to zinc content in selected raw materials, also, according to raw material price to develop market price relevant muesli. The main requirement was 70% flakes, 25% seeds, 5% additional raw materials as freeze-dried fruits. Freeze-dried fruits are needed to achieve a more pleasant taste. According to previously mentioned points, five different muesli samples were prepared. Raw material and muesli sample list and their codes tested for zinc content are listed in Table 1.

Table 1. Sample codes used in research

Raw materials	Sample code
Toasted wheat flakes	F1
Toasted triticale flakes	F2
Toasted oat flakes	F3
Toasted barley flakes	F4
Rolled oat flakes	F5
Extruded buckwheat flakes (without sugar)	F6
Pumpkin seeds	S1
Sunflower seeds	S2
Muesli samples	**Sample code**
Muesli with F1, F6, S1, S2	M1
Muesli with F1, F5, F6, S1, S2	M2
Muesli with F4, F6, S1, S2	M3
Muesli with F4, F5, F6, S1, S2	M4
Muesli with F2, F5, S1, S2	M5

Raw material and muesli sample zinc concentration was determined at food quality testing laboratory by ICP-MS (inductively coupled plasma mass spectrometry) according to PB-223/ICP, ed. II of 12.01.2015. method according to EN 15763:2010 standard.

In vitro analysis

Static *in vitro* simulation operated under adult conditions. Muesli was digested *in vitro* according to INFOGEST 2.0 protocol (Minekus et al., 2014) INFOGEST static in vitro simulation of gastrointestinal food digestion protocol (Brodkorb et al., 2019). Muesli sample was grinded (Braun GmbH, Australia) into homogenous mix, particle size from 0.5 to 2.0 mm. To form a bolus grinded sample was taken in a beaker and same amount of distilled water (1:1) was added. 5 ± 0.01 g of sample was weighted (KERN&SOHN GmbH, Germany) in beaker. To initiate oral phase simulated saliva fluids (SSF) with salivary alpha-amylase from porcine pancreas (75 U mL^{-1} of digest) to a final volume of 10 mL were added. Oral step lasted 2 minutes, pH 7 ± 0.1. Gastric phase was initiated by adding simulated gastric fluids (SGF) containing pepsin (2,000 U mL^{-1} of digest) till 20 mL ± 1 and incubated for 120 min, under pH between 2 ± 0.1 and 3 ± 0.1. After incubation intestinal phase was initiated by adding simulated intestinal fluids (SIF) containing pancreatin (100 U mL^{-1} of digest), lipase (2,000 U mL^{-1} of digest), and bile salts (10 mM of digest) till 40 mL ± 1 and incubated for 120 min, under pH 7 ± 0.1. Temperature controlled water bath at 37 ± 0.1 °C with continuous shaking at 150 rmp (Thermolab, Germany, GFL 1092) was used.

The digestion was stopped during gastric (G) phase after 0, 60, 120 minutes and duodenal (D) phase at 5, 60, 120 minutes. To neutralize sample to a pH of 7.0 ± 0.1 and stop digestion process the samples were cooled to 0–4 ± 0.1 °C in ice water. Sample pH was controlled by MW102-FOOD (Milwaukee Instruments, USA). After digestion samples were centrifuged at 4,000 rmp, at +4 °C temperature (MPW-260RH, MPW Med. Instruments, Warsaw, Poland) and filtered. The soluble fraction was collected and freeze-dried before analysis. Digestion procedure was performed thrice.

Zinc concentration was determined by atomic absorption spectroscopy method, which is based on selective absorption of electromagnetic waves by non-exited atoms of analyte. Samples were analysed with Perkin Elmer AAnalyst 400 (Perkin Elmer, USA) spectrometer utilizing flame as atomizer and zinc hollow cathode lamp as the source of electromagnetic radiation. Atomizer was produced by burning acetylene with air with oxidizing lean blue flame and absorption. Specific to zinc, it was measured at 213.9 nm, this wavelength provides linear range for measurements up to 1.0 mg L^{-1} with characteristic concentration of 0.018 mg L^{-1}. Using method of calibration curve using series of zinc standards with concentration ranging up to 1.0 mg L^{-1} quantification was performed. After each digestion step zinc content was determined, digestion procedure was performed twice.

Data processing

To analyse obtained data MS Excel 2016 was used. ANOVA analysis were performed to determine the difference between the samples. Factors were defined as significant if *p*-value was below 0.05.

RESULTS AND DISCUSSION

Zinc content in different raw materials and muesli samples

As base for muesli production Felici LLC usually is using two types of wholegrain flakes, toasted, and rolled. Both used for their different structure, but the main reason is their nutrition. To check zinc content different grain flakes as toasted triticale, barely, oat, wheat flakes, rolled oat flakes and extruded buckwheat flakes were chosen and tested, as for seeds pumpkin and sunflower seeds were tested. Table 2 shows zinc content in different grain flakes and seeds. Zinc amount between grain flakes ranged from 1.21 to 2.83 mg 100 g^{-1}. Lowest zinc content was in extruded buckwheat flakes (without sugar), highest 2.23 and 2.82 mg 100 g^{-1} were in toasted triticale flakes and rolled oat flakes. As for seeds it was 5.00 mg 100 g^{-1} for sunflower seeds and 7.50 mg 100 g^{-1} for pumpkin seeds. To decide on the most appropriate raw materials, a review for zinc content in different grain flakes and seeds before analysis was done. Different studies emphasize zinc content in grains or rolled flakes, there are not many studies about zinc content in toasted and extruded flakes. Frolich et al. (2013) reported that 100 grams barley has 1.4 mg, rye 2.4 mg, wheat 2.6 mg and oats 2.9 mg of zinc, highest content was in whole grain oats. Biel et al. (2020) reported results showed that per 100 g oat grains has the lowest zinc content of 2.1 mg, as for other flakes, barley 2.3 mg, triticale 2.5 mg, and wheat have the highest content around 3.2 mg of zinc per 100 grams. In other study, Kruma et al. (2018) compared zinc content in triticale grains and flakes.

Zinc content per 100 g triticale grains was 3.12 mg, rolled flakes 2.84 mg, but content in toasted flakes prepared by different technological parameters varied from 2.49 till 2.68 mg. Study reported that loses of minerals were mainly influenced by rolling thickness. The major losses could occur due to the solubilisation of minerals during the steaming process. This explains lower zinc content in toasted grain flakes. In whole obtained results showed that zinc content in grains and flakes in each study is different. Studies does not mention grain variety or flakes thickness what could affect zinc content in crops.

Table 2. Zinc content in analysed raw materials and muesli samples

Raw material sample code	Zinc content, mg 100 g^{-1}
F1	1.44 ± 0.01
F2	2.23 ± 0.01
F3	1.72 ± 0.01
F4	1.35 ± 0.01
F5	2.82 ± 0.01
F6	1.21 ± 0.01
S1	7.50 ± 0.01
S2	5.00 ± 0.01
Muesli sample code	**Zinc content, mg 100 g^{-1}**
M1	1.89 ± 0.01
M2	2.44 ± 0.01
M3	2.09 ± 0.01
M4	2.54 ± 0.01
M5	3.80 ± 0.01

As for seeds, Saunders et al. (2013) reports that pumpkin seeds contain 7.5 mg, sunflower seeds 5.8 mg, sesame seeds 5.5 mg, and flaxseed 4.3 mg of zinc per 100 grams. Other studies report that zinc content in pumpkin seeds is 7.8 mg 100 g^{-1} (Syed et al., 2019), sunflower seeds is 5.0 mg 100 g^{-1} (Anjum et al., 2012), sesame seeds is 3.6 mg 100 g^{-1} (Alyemeni et al., 2010), flaxseeds is 4.0 mg 100 g^{-1} (Bernacchia et al., 2014). All studies reported that pumpkin seeds and sunflower seeds has the highest zinc content. Obtained results showed similar zinc content in pumpkin and sunflower seeds as in previously mentioned studies.

After results five different muesli samples were prepared and analysed, results shown in Table 4. Zinc content in muesli samples varied from 1.89 till 3.80 mg 100 g^{-1}. Looking at the results, it was decided to use M5 muesli sample with 3.80 mg 100 g^{-1} zinc content for *in vitro* digestion test and for additional mineral and vitamin analysis. M5 sample contained following raw materials - toasted triticale flakes, rolled oat flakes, pumpkin seeds, and sunflower seeds, in total 96%. To improve muesli, taste freeze-dried pomegranate seeds, freeze-dried mango, banana, passiflora smoothie pieces and safflower petals were used.

According to regulation (EU) No 1169/2011 of the European Parliament and the council of 25 October 2011 and to regulation (EC) No 1924/2006 of the European Parliament and the council of 20 December 2006 daily reference intake for zinc is 10 mg so 3.80 mg 100 g^{-1} in developed muesli sample is high zinc content. By one portion, 50 g, it is possible to consume 1.90 mg of zinc, what will be 19% of daily reference intake, what would be source of zinc.

After European Commission request European Food Safety Authority (EFSA) has written scientific opinion on dietary reference values for zinc (EFSA panel on dietic products, nutrition, allergies (NDA)) which includes zinc average requirements and population reference intake for four different amounts of dietary phytate. This information is shown on Table 3.

As for EFSA (2014) scientific opinion on dietary reference values for zinc, for women varies from 7.5 mg day^{-1} till 12.7 mg day^{-1} for but for men from 9.4 mg day^{-1} till 16.3 mg day^{-1}. Reference intake is based on level of phytate intake, from 300 till 1,200 mg day^{-1}. Estimations show that starting from phytate intake of 600 mg day^{-1} zinc reference intake is higher than 10 mg day^{-1}. Further analysis for phytate content in muesli should be done.

Table 3. Estimations of Average Requirement (AR) and Population Reference Intake (PRI) for zinc according to phytate intake and body weight (adults) (EFSA, 2014)

Level of phytate intake, mg day^{-1}	Body weight, kg	AR	PRI [c]
300	58.5 [a]	6.2	7.5
	68.1 [b]	7.5	9.4
600	58.5 [a]	7.6	9.3
	68.1 [b]	9.3	11.7
900	58.5 [a]	8.9	11.0
	68.1 [b]	11.0	14.0
1,200	58.5 [a]	10.2	12.7
	68.1 [b]	12.7	16.3

(a) Median body weight of 18 to 79 year old women based on measured body weight of 19,969 women in 13 EU Member States assuming a BMI of 22 kg m^{-2} (see Appendix 11 in EFSA NDA Panel (2013)). At this body weight the psychological zinc requirements are 2.9 mg day^{-1} (EFSA, 2014);

(b) Median body weight of 18 to 79 year old men based on measured body weight of 16,500 men in 13 EU Member States assuming a BMI of 22 kg m^{-2} (see Appendix 11 in EFSA NDA Panel (2013)). At this body weight the psychological zinc requirements are 3.2 mg day^{-1} (EFSA, 2014);

(c) Dietary zinc intake of subjects with a body weight at the 97.5 percentile of the reference body weights (i.e., 79.4 kg for men, 68.1 kg for women) (EFSA, 2014).

Mineral and vitamin content in selected muesli sample

Nutritional quality of breakfast cereals is very important, as they can contribute significantly to daily intake of energy, carbohydrate, protein, dietary fibre, vitamin and mineral (Jones & Poutanen, 2020). To check additional nutrient content in selected muesli samples different mineral and vitamin analysis were done. Mineral and vitamin content in muesli sample were calculated according to their content in raw materials. As for minerals - iron, calcium, magnesium, phosphorus, potassium, and zinc and for vitamins - vitamin B1, vitamin B2, vitamin B3, vitamin B6 and vitamin E. Content and percent of daily reference intake is shown in Table 4.

Table 4. Mineral and vitamin content in selected muesli sample

Minerals and vitamins	Content, mg 100 g^{-1}	Daily reference intake*, %
Iron	5.98 ± 0.01	42.71 ± 0.01
Calcium	49.80 ± 0.01	6.23 ± 0.01
Magnesium	220.73 ± 0.01	58.86 ± 0.01
Phosphorus	519.62 ± 0.01	74.23 ± 0.01
Potassium	460.85 ± 0.01	23.04 ± 0.01
Zinc	3.80 ± 0.01	38.00 ± 0.01
Vitamin B1	1.07 ± 0.01	97.27 ± 0.01
Vitamin B2	0.89 ± 0.01	63.57 ± 0.01
Vitamin B3	11.33 ± 0.01	70.81 ± 0.01
Vitamin B6	1.17 ± 0.01	83.57 ± 0.01
Vitamin E	8.04 ± 0.01	67.00 ± 0.01

* According to regulation (EU) No 1169/2011 of the European Parliament and the council of 25 October 2011.s

Results show that muesli sample contains is 5.98 mg 100 g^{-1} of iron, per portion it would be 21.36% of daily reference intake. Iron, as zinc, is most abundant trace mineral in human body, with 3–4 g of iron present in average adult. Iron and zinc are frequently assessed together as these minerals share common dietary sources, the absorption of both nutrients from food is believed to be enhanced and inhibited by similar compounds, and consequently, deficiency of both nutrients occur simultaneously (Lim et al., 2013). Results show that muesli sample contains 49.80 mg 100 g^{-1} of calcium, per portion it would be 3.11% of daily reference intake. Winiarska-Mieczan et al. (2016) reported similar average results in muesli and crunchy, around 39.22 mg 100 g^{-1}. Calcium is abundant element in human body, it contains around 1 kg of calcium with more than 99% deposit in the bone in the form of calcium phosphate. Through interacting with numerous proteins distributed in different cellular compartments, calcium is involved in large amounts of aspects of life, such as muscle contraction, enzyme activation, cell differentiation, immune response, programmed cell death and neuronal activity (Pu et al., 2016). Results show 220.73 mg 100 g^{-1} of magnesium, per portion it would be 29.43% of daily reference intake. Winiarska-Mieczan et al. (2016) reported average result of 84.33 mg 100 g^{-1} what is 2.6 times less than in tested muesli sample. Magnesium is the fourth most common mineral in the human body after calcium, sodium and potassium and is the second most common intracellular cation after potassium. Within the frame of 70 kg individual, there is an average of 25 g of magnesium in reserve with 53% in bone, 27% in muscle, 19% in soft tissues and less than 1% in the serum (Schwalfenberg & Genius, 2017). It plays an important role in molecular, biochemical, physiological, and pharmacological functions in the body (Faheemuddin & Abdul, 2019). After analysis it is possible to see that muesli sample contains 519.62 mg 100 g^{-1} of phosphorus, per portion it would be 37.12% of daily reference intake. Phosphorus is one of the most abundant minerals in the body, majority stored in bone and teeth. Maintenance of extracellular and intracellular phosphate levels within a narrow range is important for many biological processes, including energy metabolism, cell signalling, regulation of protein synthesis, skeletal development, and bone integrity. The recommended dietary allowance, 700 mg day^{-1}, for healthy adults, is meant to maintain serum phosphorus concentrations within the physiologic range of 2.5 to 4.5 mg dL^{-1} (Kiela et al., 2017). Results show that muesli sample contains 460.85 mg 100 g^{-1} of potassium, per portion it would be 11.52% of daily reference intake. Winiarska-Mieczan et al. (2016) reported average result of 182.9 mg 100 g^{-1} what is 2.5 times less than in tested muesli sample. Potassium is the main intracellular cation in the body and is principally involved in membrane potential and electrical excitation of both nerve and muscle cells and acid-base regulation. On average, the potassium content of an adult human is estimated to be around 1.6 to 2.0 g kg^{-1} body weight (Lanham-New et al., 2012). Results show 1.07, 0.89, 11.33 and 1.17 mg 100 g^{-1} of vitamin B1, B2, B3 and B6 in the product. After eating one portion it is possible to admit 48.63% of daily reference intake for vitamin B1, 31.78% of daily reference intake for vitamin B2, 35.40% of daily reference intake for vitamin B3 and 41.79% of daily reference intake for vitamin B6. Vitamins B comprises a class of water-soluble complexes. It has very important molecular function in the human body. Vitamin B1 (thiamine) strengthen the immune system, helps neuronal communication, maintains processes in cells and tissues. Vitamin B2 (riboflavin) metabolize fats, protect from ischemia reperfusion. Vitamin B3 (niacin) works in DNA proliferation, produces energy (Karunaratne et al., 2017). Vitamin B6 participates in more than one hundred transamination, decarboxylation, and other types of reactions, including the initial step of porphyrin synthesis, glycogen mobilization, amino acid transsulfuration, and neurotransmitter synthesis (Kohlmeier, 2003). Results show that product contain 8.04 mg 100 g^{-1} of vitamin E, for one portion it is 33.5% of daily reference intake. Vitamin E mostly recognized for its antioxidant function that terminates the self-perpetuating cycle of lipid peroxidation (Bruno & Mah, 2014).

Daily reference intake was calculated according to regulation (EU) No 1169/2011 of the European Parliament and the council of 25 October 2011. Per portion muesli is source of iron, magnesium, phosphorus, zinc, vitamin B1, vitamin B2, vitamin B3, vitamin B6 and vitamin E.

***In vitro* analysis**

Zinc digestion results are presented in Fig. 1 as average release. Average results show that in small intestine in total 22% of zinc was absorbed, it means that by one muesli portion it is possible to replenish our body zinc level approximately by 0.42 mg, maximum 0.63 mg of zinc. In the end of gastric (G) phase 67% of zinc was released. At the beginning of duodenal (D) phase 77% of zinc released, after 60 minutes 84% of zinc were released, but in the end of duodenal (D) phase, after 120 minutes, 89% of zinc were released from loaded muesli sample. In one of three measurements in the end of duodenal phase 100% of zinc was absorbed, what shows that maximum 33% of zinc could be absorbed in small intestine. Values indicates significant difference between *in vitro* digestion stages ($p < 0.05$).

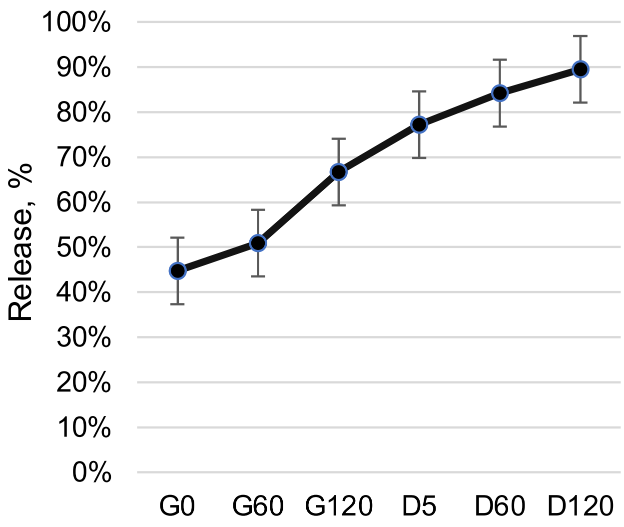

Figure 1. Release of zinc during gastric (G) and intestinal (D) *in vitro* digestion: G0 – gastric phase at 0 min digestion; G60 – gastric phase at 60 min digestion; G120 – gastric phase at 120 min digestion; D5 – duodenal phase at 5 min digestion; D60 – duodenal phase at 60 min digestion; D120 – duodenal phase at 120 min digestion.

As reported by Ogawa et al. (2016) approximately 30% of zinc in the diet is absorbed in the small intestine, obtained results are similar. Looking at other zinc digestion tests, Silva et al. (2020) in their study, where they compared organic and inorganic forms of zinc in salmonid diet, reported that solubility was similar in both diets tested. The amount of soluble zinc was low in the acidic hydrolysis (3–8%) and lower in the alkaline hydrolysis (0.4–2%). Results are significantly lower than in this study, yet they reported that solubility is impacted, also, by the pH of the gastrointestinal environment. In other study, Martinez et al. (1998) reported that the solubility of zinc of green beans evaluated by an *in vitro* method was from 14.7 till 55.1%. Observed results showed wide range. Results can be different in salmonoid diet as it contains both animal and plant ingredients, and it is lipid rich sample (Silva et al., 2020). It is possible to see that zinc solubility during digestion is impacted by different factors as mineral chemical form, pH of the gastrointestinal environment, phytate intake, fibres such as cellulose, etc. (Maares & Haase, 2020).

CONCLUSIONS

This study shows that zinc content in grain flakes varies according to grain variety and used flake type. As mentioned in other studies the major losses could occur due to the solubilisation of minerals during the steaming process. This shows why zinc content in toasted flakes will be lower than in rolled flakes. Also, as different grain varieties during production are used it is needed to follow grain variety and it zinc content each season. *In vitro* digestion test average results showed that by one meal (50 g of muesli) it is possible to replenish our body zinc level by 0.42 mg. Overall results show that this muesli can be used for consumers with different diets to replenish their body zinc level each day. Of course, zinc solubility during digestion is impacted by different factors as mineral chemical form, pH of the gastrointestinal environment, phytate intake, fibres such as cellulose, etc. Different raw material processing methods as thermal processing, malting (followed by germination), fermentation, soaking in water, hydrothermal treatment can reduce dietary phytate content (Gibson et al., 2018). Developed muesli contains raw materials which have had thermal processing and hydrothermal treatment what potentially leads to reduced dietary phytate content in muesli. Further investigation on dietary phytate content, it impacts on digestion should be done. In this study, other mineral and vitamin content in muesli sample were calculated according to their content in raw materials. Per portion, muesli is source of iron, magnesium, phosphorus, zinc, vitamin B1, vitamin B2, vitamin B3, vitamin B6 and vitamin E. Further laboratory tests on total mineral and vitamin content in developed muesli sample for more precise results should be applied. Results show that by one muesli portion it is possible to receive different naturally present minerals and vitamins.

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