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Research Article

Analysis of accessible and total phosphate contents in different foods, to assess their suitability for use by dialysis patients

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Abstract: There is a direct link between hyperphosphatemia and increased mortality in end-stage kidney disease, which can be attributed to the higher consumption of ultra-processed foods. Thus, the study aimed to analyze the total phosphorus content and relative amount of available phosphorus in various manufactured processed foods (MPF) and home processed foods (HPF), specifically for use by dialysis patients. The results of the total phosphorus content were found in the range of (26 to 290 mg P/100g). The highest concentration (290 mg/100g) was found in nan bread (HPF), followed by boiled eggs (255mg/100g), chicken (192mg/100g) and beans (91mg/100g). For MPF samples, the chicken (195mg/100g), whole meal bread (193mg/100g), and wheat and white bread wraps for both Asda and Tesco (99, 124 mg/100 respectively) respectively exhibited high phosphorus content. In this study, relative available P concentrations were in the range of (5-42.5 mg P/100g) for MPF and (6-38 mg P/100g) for HPF. While the highest concentration (42.5 mg/100g) was found in chicken (MPF), followed by whole meal bread, and Coca-Cola. For HPF Nan bread (37.8 mg P/100g) showed high content followed by boiled eggs and Chicken. HPF generally exhibited higher levels of both total phosphorus content and extractable phosphorus compared to manufactured processed foods (MPF). This is an important consideration for patients, particularly those with kidney disease, as they should be mindful of their phosphorus intake when consuming HPF.

1. INTRODUCTION

Phosphorus is one of the most abundant minerals in the crustal rock on the Earth. It is found widely in nature and roughly exists in nearly all foods (Metson *et al.,* 2016). Phosphorus is an essential mineral that is necessary for a well-balanced diet, as it plays a critical role in bone composition and the regulation of numerous physiological processes within the body (Hifizah, 2011). In an individual with normal renal function, the kidney has a crucial role in regulating phosphorus and calcium homeostasis by excreting excess phosphorus. When kidney function deteriorates, the phosphorus levels increase in the blood, which is common in patients with chronic kidney disease (Kestenbaum *et al*., 2005; González-Parra *et al*., 2012). Despite the increase in the prevalence of the condition in many countries, chronic kidney diseases (CKD) remain a problem in the world. For example, CKD is a widespread clinical condition that affects

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over 800 million individuals suffering from various kidney disorders (Thongprayoon *et al*., 2021) and affects more than 20 million American people (Kovesdy *et al*., 2006). According to the Global Burden of Disease research, there were 697.5 million cases of CKD worldwide, representing a 9.1% prevalence rate (Okpechi *et al*., 2022). Phosphorus additives in food have been reported" to be the main cause of increased serum phosphate levels. Processed foods with food additives contain 70% more phosphorus than similar foods without food additives (Cupisti *et al*., 2004). Thus, manufactured meals may contain higher levels of phosphorus compared to naturally available foods, and the body may absorb this phosphorus more readily (Noori *et al*., 2010). However, a crossover experimental design on 8 patients with chronic kidney disease was conducted to compare the effect of meat-based and vegetarian diets on serum phosphate levels, with both diets containing equivalent concentrations of protein and phosphorus. The results revealed that over the course of seven days, participants following the vegetarian diet exhibited significantly lower blood serum phosphate levels compared to those that followed the meat-based diet (Moe *et al*., 2011). This can be attributed to the greater bioavailability and absorption of phosphorus from animal sources compared to that from plant sources (Noori *et al*., 2010). It has been illustrated that after three months, a significant reduction in blood serum phosphate levels (0.6 mg/day) was observed in dialysis patients who were advised to restrict their intake of food additives containing phosphorus, such as certain beverages and processed meats (Sullivan *et al*., 2009). In patients with renal failure, it is a crucial aspect of effective therapeutic management to restrict phosphorus intake, particularly after the initiation of dialysis (Calvo & Uribarri, 2021). To understand the effect of phosphorus on the patient with end-stage CKD, it is essential to gather data regarding the phosphorus content in commonly consumed ready-made meals. Typically, the Western diet provides phosphorus intake ranging from 1000 to 2000 mg per day. However, the recommended daily phosphorus intake for patients with advanced-stage chronic kidney disease (CKD) is considerably lower, ranging from 550 to 1100 mg per day. Therefore, the bioavailability of phosphorus in the diet is generally about 60% (Kooienga, 2007).

Numerous pre-clinical and clinical investigations report that high levels of phosphate can adversely affect health, both in healthy individuals and in patients with various conditions such as cardiovascular disease, bone disorders, or kidney disease. Therefore, the recommended dietary allowance (RDA) for phosphorus is approximately 700 mg per day for adults; however, the majority of individuals consume significantly higher levels of phosphate, nearly double the RDA (Erem & Razzaque, 2018). Thus, the main objective of the present study is to investigate the phosphorus content in both MPF and HPF, as well as to estimate the relative available phosphorus content in these foods and their relationships.

2. MATERIAL and METHODS

2.1. Food Samples

The samples tested in this study included manufactured processed foods (MPF) and home-processed foods (HPF). The MPF samples were acquired from a local market in the United Kingdom, while the HPF samples were prepared using local methods at home. The daily diet of a typical individual consuming manufactured processed foods (MPF) was selected as the study sample, comprising breakfast items such as yogurt, wheat and white bread, and milk; lunch consisting of fried potatoes and Coca-Cola; and dinner featuring basmati rice, chicken breast, baked beans in tomato sauce, and whole meal bread. Additionally, the normal mixed diet for an individual for one day, incorporating both home-processed foods (HPF) and MPF, included breakfast with yogurt, naan bread (from local shops), milk, and eggs; lunch with fried potatoes and Coca-Cola; and dinner of basmati rice, chicken breast, and baked beans in tomato sauce.

A detailed list of the manufactured processed food (MPF) samples used in this study, along with their suppliers, includes items such as whole milk and Mission Deli Wheat and White Wraps (8-pack), both obtained from Tesco. White tortilla bread, Rice, Chicken tikka & basmati rice (weight watcher), and soft drinks (Coca-Cola) were gathered from Asda. Tasty whole meal bread (KINGSNILL), fried potato chips, ready-cooked sliced Chicken breast, yoghurt (Onken) Natural set, beans, baked, in tomato sauce, canned (Beanz) were from Iceland. Naan bread was obtained from local shops.

2.2. Chemicals

All chemicals used in this study include hydrochloric acid, approx. 36% w/w HCl (Fisher chemical), ammonium molybdate tetrahydrate (Santa Cruz), ammonium metavanadate (Santa Cruz), potassium dihydrogen phosphate, KH2PO4 (BDH Laboratory), toluene (Fisher chemical), pepsin from porcine gastric mucosa (SIGMA-ALDRICH), pancreatin (P-1750, from porcine pancreas) (SIGMA-ALDRICH), bile extract porcine (SIGMA-ALDRICH), salivary alpha-amylase from human saliva (SIGMA-ALDRICH), sodium bicarbonate (BDH Laboratory), NaHCO3 (BDH Laboratory).

2.3. Analytical Methods

In this study, MPF samples were sourced from three markets: Asda, Tesco, and Iceland. Home-processed foods (HPF) were prepared using similar methods as the methods used for the MPF samples, but in a home setting. Both the MPF and HPF foods were kept cooled in the laboratory conditions to avoid contamination. All glassware was acid-washed in a laboratory dishwasher, rinsed with distilled water and dried before use. However, deionized iron free water was used during the experiment for the dilution of samples.

2.4. Total Phosphorus Determination

2.4.1. *The preparation of sample solution by dry combustion*

All samples were prepared based on the procedures described by Nielsen (Nielsen, 2017) after a few modifications. Samples of MPF including whole milk, white tortillas (from Asda), wheat and white bread (from Tesco), basmati rice, Coca-Cola, whole meal bread, fried potatoes, chicken breast, yogurt, and beans baked in tomato sauce, along with seven types of home-processed foods (HPF) such as basmati rice, fried potatoes, chicken breast, naan bread, yogurt, beans baked in tomato sauce, and eggs, were collected and transported to the laboratory. One gram of the food sample was weighed and placed into a crucible, which was then positioned in a cool muffle furnace. The temperature was gradually increased to 450 °C and maintained at this level overnight until only a whitish-grey ash remained. After the muffle furnace was turned off, the crucible was rapidly transferred to a desiccator to cool before it was weighed.

Ash content was determined by using this equation: $[(W5 - W4)/W3] *100$

Where W3: the weight of the sample, W4: the weight of the sample and weight of the crucible after dry combustion, W5: the weight of the sample and crucible before dry combustion.

Subsequently, the crucible was placed in a water bath, and 5 mL of approximately 6 M hydrochloric acid was added, followed by the evaporation of the solution to dryness. After that, the residue was moistened with 1 mL of approx. 36% hydrochloric acid, and lightly evaporated to dryness. After approximately 5 mL of water was added, and the solution was evaporated once more. Finally, the residue was collected, transferred to a 25 mL volumetric flask, filtered using Whatman filter paper, and then diluted to a final volume of 25 mL.

2.4.2. *Analysis of sample solution*

In the spectrophotometric procedure for total phosphorus determination using a Libra S12-Biochrom (UK) spectrophotometer, various sample volumes (3 mL, 5 mL, and 10 mL) were transferred into a 50 mL volumetric flask, depending on the type of food. Following this, 5 mL of approximately 5 M hydrochloric acid and 5 mL of ammonium molybdate were added. Ammonium metavanadate reagent was ten added and the solution was filled up to the mark. The solution was allowed to stand for 30 minutes to ensure the development of the yellow color, after which the absorbance was measured against a blank at 400 nm

The calibration curve was employed to determine the phosphorus concentration (y) in the working standard solution. This curve was generated at a detection wavelength of 400 nm, using a series of working standard solutions with concentrations ranging from 0 to 50 µg/mL. The calibration curve was then applied to calculate the phosphorus concentration in each sample, following the equation below:

$$C1V1=C2V2 \ \mu g/mL,$$

2.5. In vitro Model

In vitro enzyme digestion procedure was performed according to the procedure of Miller (Miller *et al*., 1981), with a few modifications in the preparation of reagent, enzymes and digestion process to adapt methods with the equipment and glassware that were available in the laboratory.

2.5.1. *Preparation of sample*

Samples of MPF (whole milk, white tortillas bread (Asda), wheat & white bread (Tesco), basmati rice, coca cola, whole meal bread, fried potatoes, chicken breast, yoghurt, beans baked in tomato sauce) and HPF (basmati rice, fried potatoes, chicken breast, Nan bread, yoghurt, beans baked in tomato sauce and eggs) were used in these experiments to estimate relative available phosphorus content. The samples were transported to the laboratory, where 20 g of each was weighed and transferred into 100 mL beakers to initiate the digestion process.

2.5.2. *Preparation of reagent and enzymes*

A pepsin solution was prepared by dissolving 16 g pepsin (from porcine gastric mucosa) in 100 mL of 0.1 M HCl (Miller *et al*., 1981). A Pancreatin-bile extract mixture was prepared by weighing 4 g pancreatin (P-1750, from porcine pancreas) and 25 g of porcine bile extract, then dissolving the mixture in 1 L of 0.1 M $NaHCO_3$, as described by Miller *et al*. (1981) α-amylase: 20mL of 0.85% saline solution (sodium bicarbonate) was added to a bottle of Salivary alpha-amylase from human saliva 1KU to obtain α-amylase 50 KU.

2.5.3. *In vitro digestion methods*

For both MPF and HPF samples (20 g), 15 mL of deionized water was added to 100 mL beakers and the mixture was stirred using a glass rod. For the following food samples: white tortilla bread (Asda), wheat & white bread (Tesco), naan bread, basmati rice, whole meal bread, fried potatoes, and beans baked in tomato sauce, 0.125 mL of salivary amylase was added. The mixture was shaken and then incubated at 37°C for 5 minutes at a shaking speed of 95 rpm. This step aimed to digest the starch in these foods, which are known to have a high starch content. The pH of all samples was first adjusted to 2.0 by adding 6 M HCl. Subsequently, 1.5 mL of pepsin was introduced to each sample, and they were incubated in a shaking water bath at 37°C for 2 hours. After the incubation period, the pH was readjusted to below 7.0 by adding 1 M $NaHCO_3$. A 2.5 mL portion of the pancreatin-bile extract mixture was added to the samples, which were then incubated again in a shaking water bath at 37°C for 2 hours. At the end of the incubation period, the digested tubes were removed and centrifuged at 4000 rpm for 40 minutes, and then 2 mL of supernatant was removed into Eppendorf tubes for more centrifuges. After the centrifuge, digested samples were removed for estimation of the relative available phosphorus content. 0.1mL of samples was transferred into a 50 mL volumetric flask followed by the addition of 5 mL of approximately 5M hydrochloric acid and 5 mL of ammonium molybdate – ammonium metavanadate reagent and filled up to the mark with water. The mixture was allowed to stand for 30 minutes to ensure color development, after which the absorbance was measured against a blank at 400 nm.

2.6. Conversation

The phosphorus values can be converted to phosphate by using the following equation:

Phosphate (mg/100g) = phosphorus (mg/100g) *3.06

Find out this equation by using the molecular weight (MW) of PO4:

Where: P (MW) =31, O (MW) =16, and then PO4 (4*16) +31 = 95

It means that every 95 phosphate contains only 31 phosphorus. However, the µg/mL values can be converted to mg/mL dividing by 1000.

2.7. Percentage of Extractable Phosphorus in Food Samples

The extractable phosphorus value can be converted to a percentage by using this equation:

In vitro extractable P % = (C1/C2) *100.

Where: C1 = concentration of relative available phosphorus; C2 = concentration of total phosphorus.

2.8. Statistical analysis

In this study, each sample was repeated three times and values are given as mean + standard deviation. For both methods, Data was entered into Microsoft Excel, and the IBS SPSS software program 2010 (version 20, USA) was utilized to conduct a normality test using the Shapiro-Wilk test to assess the normality of the data. The data were normally distributed; therefore, One-Way ANOVA and Independent Sample *t*-test at confidence interval for mean 95% were used to determine significant differences between samples.

3. FINDINGS

3.1. Total Phosphorus Content

The results of the total phosphorus content for all food samples showed different variances between samples as illustrated in Table 1, Figure 1a. It can be observed that the phosphorus concentrations in the food samples ranged from 26 to 290 mg P/100 g of fresh samples. The lowest concentration, 26 mg/100 g, was found in Coca-Cola, while the highest concentration of 290 mg/100 g was observed in naan bread, which was followed by boiled eggs (255 mg/100 g), chicken (192 mg/100 g), and beans (91 mg/100 g). In the MPF samples, chicken (195 mg/100 g), whole meal bread (193 mg/100 g), and wheat & white bread wraps from both Asda and Tesco (99 and 124 mg/100 g, respectively) exhibited high phosphorus content. The results indicated a significant difference in the total phosphorus content between the bean and rice samples ($p < 0.05$). Furthermore, the analysis revealed that the effects of manufactured processed foods did not statistically influence the total phosphorus content in chicken, potatoes, and yogurt. Ash content represents the amount of ash material that remained in fresh food samples after putting them in a muffle furnace overnight. All food samples exhibited varying levels of ash content. Generally, phosphorus is well recognized as an essential mineral in the human body. As a result, phosphorus is abundantly found in both plant and animal food sources, including meat, poultry, fish, eggs, and dairy products, as well as in nuts and legumes. It is widely available as a food additive in processed foods such as meats, soft drinks and bakery goods (Willett, 2008). There is a scarcity of research on the determination of total phosphorus and the estimation of in vitro extractable phosphorus across a diverse range of food samples. The difference in food and technological processes account for the observed variation in phosphorus content. When compared to other data, the results obtained are generally consistent with those published by Esperance (2018). For instance, the phosphorus concentrations in whole milk, basmati rice, and Coca-Cola were reported to be 83 mg, 33 mg, and 16 mg per 100 g of fresh sample, respectively. In contrast, our results indicated concentrations of 69 mg, 30–38 mg, and 26 mg per 100 g, respectively. Research indicates that food additives are likely the most significant factor influencing total phosphorus content. This is primarily due to the fact that certain products, such as chicken and meat, are often enhanced with phosphate additives, which can elevate their total phosphorus content (Kalantar-Zadeh *et al*., 2010). Nonetheless, the phosphorus content in boiled eggs was found to be higher than that reported in the data from Esperance (2018), which may be attributed to the type of chicken and their feed.

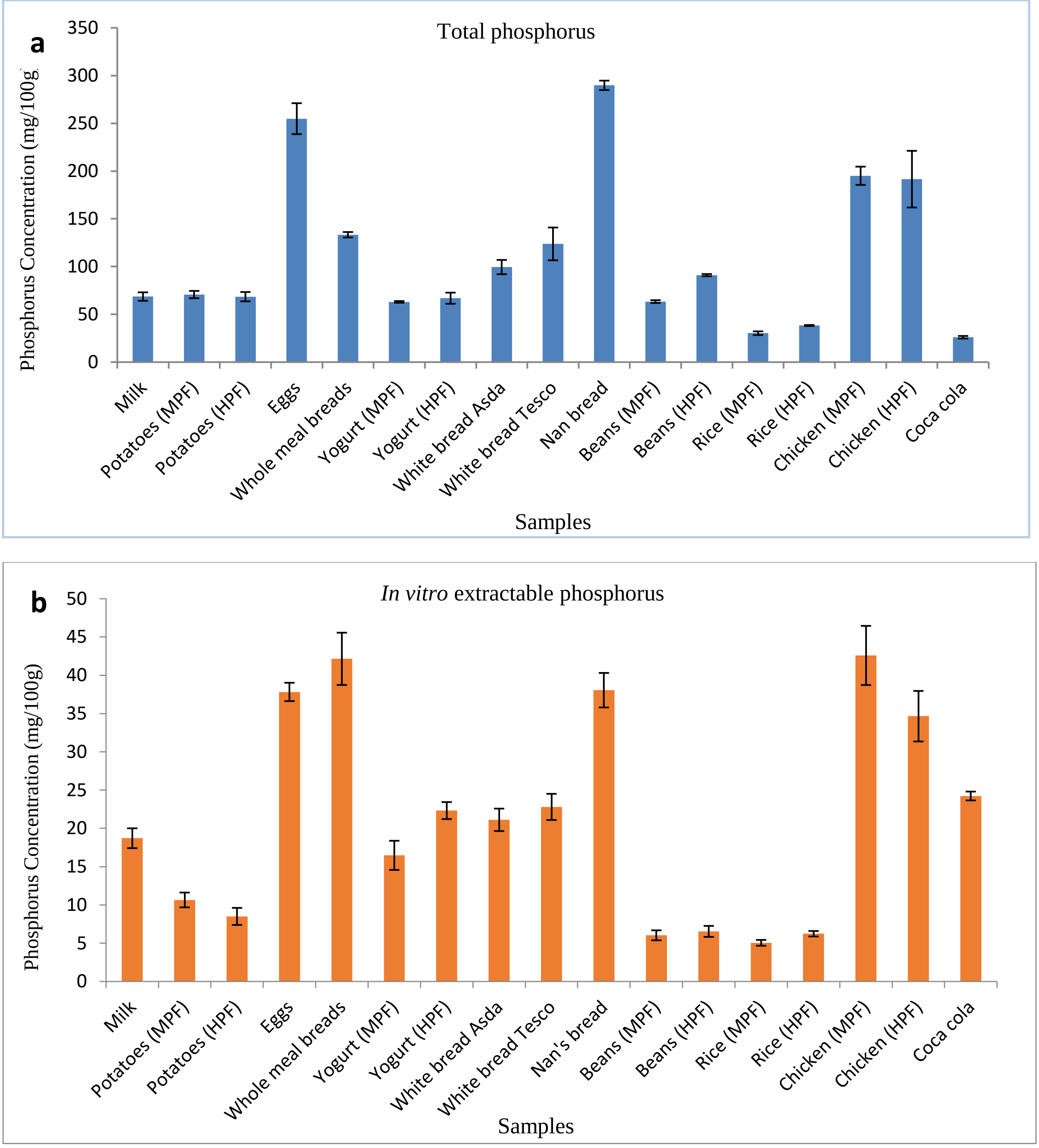

Figure 1. a: Shows total phosphorus concentration in different food samples (MPF and HPF) by using the vanadomolybdate methods; **b:** Shows the extractable amount of phosphorus content of different food samples.

A notable observation was that the phosphorus content was significantly higher in naan bread, which was prepared in a local bakery using a specific recipe unique to that establishment. These findings suggest that the phosphorus content of Nan bread may help increase phosphorus intake more effectively than other sources. Additionally, it was also observed that there was no significant difference in total phosphorus content between the white tortilla bread from Asda and the wheat and white bread from Tesco, as both types of bread contain similar ingredients. Interestingly, in this study, the manufacturing process did not affect the phosphorus content of the beans and rice samples. However, the phosphorus content in high-processed foods (HPF) was found significantly higher than that in minimally processed foods (MPF). This discrepancy may be attributed to the factors as the types of seeds, harvest season, tomato sauce, and the quantity of beans used during production.

Table 1. Shows the total phosphorus and phosphate concentration in 11 food samples with vanadomolybdate methods (measured as mg P/100 g fresh sample). Where, MPF: manufacture processed foods, HPF: home processed foods.

No	Food Samples	Process type	Total phosphorus (mg P/100g fresh sample)	Total phosphate (mg PO_4/100g fresh sample)	Ash content (g)
1	Whole milk	MPF	69 ± 0.3^d	211 ± 0.5^f	0.018 ± 0.1
	Fried Potatoes	MPF	71 ± 0.9^d	217 ± 0.1^f	0.031 ± 0.3
2		HPF	69 ± 0.12^d	211 ± 1^f	0.042 ± 0.6
3	Whole chicken eggs, boiled	HPF	255 ± 0.77^a	780 ± 1.2^a	0.015 ± 0.9
4	Whole meal breads	MPF	133 ± 1^c	407 ± 1.6^c	0.021 ± 0.6
	Yogurt	MPF	63 ± 0.83^d	193 ± 0.8^g	0.005 ± 0.2
5		HPF	67 ± 0.9^d	205 ± 0.7^f	0.009 ± 0.7
6	Wheat & white breads wraps	MPF Asda	99 ± 0.7^c	303 ± 0.6^e	0.016 ± 0.1
		MPF Tesco	124 ± 1^c	379 ± 0.4^d	0.002 ± 0.0
7	Nan bread	HPF Local	290 ± 1.9^a	887 ± 1.9^a	0.028 ± 0.1
8	beans baked in tomato sauce	MPF	63 ± 1.4^d	193 ± 1^g	0.011 ± 0.3
		HPF	91 ± 1.1^c	278 ± 1.2^e	0.015 ± 0.1
9	Basmati rice	MPF	30 ± 1.9^d	92 ± 0.8^g	0.009 ± 0.4
		HPF	38 ± 0.5^a	116 ± 0.8^g	0.012 ± 0.0
10	Chicken	MPF	195 ± 1.5^b	597 ± 0.9^b	0.014 ± 0.2
		HPF	192 ± 2.6^b	588 ± 1.8^b	0.011 ± 06
11	Soft drink (Coca cola)	MPF	26 ± 0.09^e	80 ± 0.9^h	0.007 ± 0.1

$^{a\text{-}h}$Data are given as mean ± *SD* (n=3) and various letters are significantly different at $p \leq 0.05$.

3.2. In vitro Digestion Procedure

The purpose of this experiment was to estimate to what extent phosphorus from those food samples was available for human consumption and to show different relative availability levels between samples. Several MPF and HPF samples were analyzed through in vitro digestion procedures to calculate the relative available phosphorus content, and the obtained results of the relative available phosphorus content for all food samples are shown in (Table 2, Figure 1b). It can be observed that there were different variances between samples. In this study, the results for relative available phosphorus varied, ranging from 10% to 93% for minimally processed foods (MPF) and 7% to 37% for highly processed foods (HPF). The mean concentrations of phosphorus were also variable, ranging from 5 to 43 mg P/100 g for MPF and 6 to 38 mg P/100 g for HPF. The highest concentration of phosphorus (43 mg/100 g) was observed in chicken (MPF), while the lowest concentration (5 mg/100 g) was found in basmati rice (MPF). Figure 1b illustrates that highly processed foods (HPF) exhibited the highest relative available phosphorus content in yogurt, beans, and rice, while minimally processed foods (MPF) showed elevated levels in chicken and potatoes. The results showed that the amount of relative available P was significantly different in yoghurt and rice at (p<0.05). Table 2 indicates that the effects of home-processed foods significantly influenced the extractable phosphorus content in yogurt and rice. Three HPF (yoghurt, beans and rice) contained slightly higher relative available P than MPF. However, the relative available P content of coca cola was rather high, which may be due to food additive content. Usually, this type of beverage includes little to no protein or other organic compounds, so the phosphorus is completely from a food additive. Therefore, Sherman (2007) states that a wide range of food additives contain phosphorus and phosphorus-containing additives are currently being added to a wide variety of processed foods such as soft drinks (Sherman, 2007). The results from Coca-Cola suggest a potential impact of food additives on patients with chronic kidney disease. In addition, the difference in relative available phosphorus content can be can be attributed to the diversity of food samples and the different processing methods employed. Moreover, the results show that the amount of relative available P was only significantly different in yoghurt and rice samples at (p<0.05).

Table 2. Shows concentration of relatively available phosphorus and phosphate in 11 food groups *in vitro* digestion methods (measured as mg P/100 g fresh sample). Where MPF: manufacture processed foods, HPF: home processed foods.

No	Food Samples	Process type	Extractable phosphorus (mg P/100g fresh sample)	Extractable phosphate (mg PO_4/100g fresh sample)	*In vitro* extractable phosphorus (%)
1	Whole milk	MPF	18.7 ± 1.2[d]	58 ± 0.12[d]	27
		MPF	10.6 ± 0.9[e]	34 ± 0.33[e]	14
2	Fried Potatoes	HPF	8.5 ± 1.1[f]	28 ± 0.4[f]	11
3	Whole chicken eggs, boiled	HPF	37.8 ± 1.1[b]	116 ± 0.7[b]	15
4	Whole meal bread	MPF	42.1 ± 1.4[a]	129 ± 0.01[a]	12
		MPF	16.4 ± 1.9[d]	49 ± 1.1[d]	23
5	Yoghurt	HPF	23 ± 1.10[c]	70 ± 0.77[c]	37
6	Wheat & white bread wraps	MPF Asda	22.3 ± 1.4[c]	64 ± 2.1[c]	16
		MPF Tesco	22.8 ± 1.7[c]	70 ± 1.9[c]	22
7	Nan bread	HPF Local	38 ± 2.2[b]	116 ± 0.11[b]	18
8	beans baked in tomato sauce	MPF	6 ± 0.6[g]	18 ± 1.3[g]	10
		HPF	6.5 ± 0.7[g]	21 ± 0.66[g]	7
9	Basmati rice	MPF	5 ± 0.3[g]	15 ± 0.99[g]	17
		HPF	6.2 ± 0.3[g]	18 ± 1.4[g]	17
10	Chicken	MPF	42.5 ± 1.8[a]	132 ± 0.1[a]	20
		HPF	34.6 ± 1.2[b]	107 ± 0.12[b]	20
11	Soft drink (Coca cola)	MPF	24.2 ± 0.5[c]	73 ± 1[c]	93

Interestingly, the relative available phosphorus percentage in white tortillas from Asda was higher than that in wheat and white bread from Tesco, although both types contained lower levels compared to naan bread, with values of 23%, 21%, and 38%, respectively. It was observed that the lowest amount of relative available P was found in home-processed beans 7%. This is because plant-based phosphorus (beans and legumes) is poorly digested and absorbed by humans' gastrointestinal tract, ranging from 40% to 50%, the reason is that phosphorus from plants is in the form of inorganic phytic acid (Fukagawa *et al*., 2011; Noori *et al*., 2010). In contrast, Weremko *et al*. (1997) reported that the availability of phosphorus in plants varies from 10% to 60%, which can be attributed to the presence of phytate.

In this study, the available P content (percentage) was generally lower than those reported in the literature with the exception of Coca-Cola. It is evident that the in vitro methods include gastrointestinal digestion using pepsin at pH 2 for 2 hours, followed by digestion with pancreatin-bile extract during the intestinal stage at pH 7 for an additional 2 hours. The results indicate that incubation time and pH may have little effect on the digestion process. The variation in pH conditions and the unstable incubation times within the context of this study are the primary factors resulting in the lower available phosphorus content (percentage). However, it was found that the digestibility increased with increasing incubation time (Miller *et al*., 1981). On the other hand, mineral absorption occurs in the earlier part of the small intestine, therefore the reproduction of the situation existing in the small intestine is the most important step for in vitro digestion procedure (Shiowatana *et al*., 2006). It should be noted that the aforementioned situation may not be appropriately applicable to in vitro methods; therefore, pH and incubation time may affect digestive enzymes as well. This may be attributed to the digestive enzymes not being sufficiently active to properly digest the samples; however, the effects could differ if the samples were consumed by humans.

3.3. Phosphorus with Their Impact on Advanced-Stage CKD Patients

The comparison between the total phosphorus and in vitro extractable phosphorus showed that the amount of total and relative available P was significantly different in all food samples at ($p<0.05$). The strong association between diet and health underscores the critical role of dietary guidance in managing and potentially preventing phosphorus imbalances in advanced-stage chronic kidney disease (CKD). It is a leading cause of morbidity and mortality throughout the world. Returning to the two primary findings, it can be concluded that these food samples are suitable for consumption by patients with end-stage chronic kidney disease (CKD), specifically those undergoing dialysis. Patients with advanced-stage chronic disease require specific dietary restrictions to limit the build-up of extra phosphorus in the body. Winger *et al*., (2012) show that control of serum phosphate is achieved by reducing dietary P intake. Moreover, Uribarri (2001) indicated that dialysis alone is insufficient to prevent elevated serum phosphorus levels in patients without dietary restriction. Generally, patients with advanced-stage CKD can face varying lifestyle challenges such as dietary modification as well. Recommendations for these patients are often complex and depend on the stage of the disease and the presence of coexisting medical conditions, such as diabetes (Winger *et al*., 2012). Dietary restrictions are recommended for dialysis patients, particularly regarding protein intake. However, this may lead to malnutrition over time, as high-protein foods are a major source of dietary phosphorus. (Shinaberger *et al*., 2008). Consequently, dietary phosphorus restriction must be balanced with adequate protein intake to prevent additional health complications for the patient. This study demonstrates that home-processed foods (HPF) generally have a higher total phosphorus content but exhibit lower or comparable available phosphorus percentages compared to manufactured processed foods (MPF). For example, home-processed potatoes (HPF) contain 69 mg P/100 g, with only an estimated 11% available, while manufactured processed potatoes (MPF) contain 71 mg P/100 g, with 14% available (Table 3 and 4). It is also obvious that relative available P significantly affects serum PO4 levels in patients with end-stage CKD. Additionally, manufactured processed foods (MPF) such as chicken and soft drinks may contain phosphorus additives that are significantly higher than those found in home-processed foods (HPF). Inorganic phosphorus present in additive foods is absorbed at a rate exceeding 90% (González-Parra *et al*., 2012; Rosenberg, 2000). This study demonstrated that 93% of the phosphorus content in coca cola was available. For this reason, patients with stage 3-5 CKD are advised to avoid using MPF such as coca cola, because clinically it is a leading cause of significant improvement in serum phosphate.

3.4. Measuring Dietary Intake in The Context of Food Analysis

Dietary intake has an important role in increasing and decreasing phosphate content in the blood. The analyzed food samples were categorized into manufactured processed foods (MPF) and home processed foods (HPF) for the purpose of this study. In addition, typical daily diet for an individual was calculated based on both manufactured processed foods (MPF) and home-processed foods (HPF) over two different days. Hence, the recommended daily phosphorus intake for a patient with advanced-stage CKD ranges between (550 – 1100 mg/day) (Kooienga, 2007). The average diet in Europe contains approximately 1,000 to 1,500 mg of phosphorus per day (EFSA, 2019). Table 3 shows each serving of (MPF) samples contains (1237 mg) of phosphorus per day, which is slightly higher than the clinically accepted range of 550 to1100 mg/day for patients with end-stage CKD while indicating lower relative available P content (329 mg). In contrast, Table 4 demonstrates that each serving of home-processed food (HPF) samples contains 1,717 mg of phosphorus per day, which exceeds the clinically accepted range of 550 to 1,100 mg/day for patients with end-stage chronic kidney disease (CKD). However, it indicated higher available P content than the MPF. The difference between manufactured processed foods and home-processed foods may be due to differences in ingredients. The home process ingredients were found to be more effective in increasing daily P intakes compared to MPF, particularly in Nan bread, which contained the highest amount of phosphorus among all other samples. Based on the results obtained in this study, it is proposed that dietary restrictions for both manufactured processed foods (MPF) and home-processed foods (HPF) are influential and essential for balancing phosphate levels in the body. There are considerably fewer data and research published on the total P and relative available P content of (MPF) and (HPF).

On average patients with end-stage renal disease are facing many challenges in their daily life with dietary restrictions, and the lack of information about dietary content. For example, naan bread is a popular type of bread in many countries worldwide, typically prepared in local bakeries without any labeling or nutritional information. Many people consider Nan bread to be a natural source of nutrients. This leads to the conclusion that it is not accurate to assert that all home-processed foods have high phosphorus content. In addition, eggs are one of those food sources that is prepared at home when was contains a high amount of phosphorus. These results suggest that is important to explore alternative approaches to prevent complications in patients with end-stage chronic kidney disease (CKD). Instead of solely advising patients to avoid manufactured processed foods, there should be a focus on producing specialized foods through manufacturers, while encouraging greater reliance on home-processed foods.

Table 3. shows mean and percentage of the total and relative available phosphorus in manufacture processed foods (MPF) for one-day meals of a normal individual. Where 1 cup milk: 250g, 1oz potatoes: 28.42g, 1 slice whole bread: 22g, 1 slice wheat bread: 23g, 1cup beans: 253g, 1cup rice: 164g, 1oz yoghurt: 227g, chicken breast:140g, Coca-Cola: 370g.

Food samples	Total P (mg/100g)			*In vitro* extractable P (mg/100g)		*In vitro* extractable P %
	mg P/100g	Rate	Daily/mg	mg P/100g	Daily/mg	%
Whole milk	69	1 cup	173	19	48	27
Potatoes	71	2 oz	40	11	6	14
Whole meal bread	133	4 Slice	122	42	39	12
Yoghurt	63	1/8-oz container	143	16	36	23
Wheat &white bread (Asda)	99	2 Slice	40	21	9	16
Beans in tomato sauce	63	1 cup	159	6	15	18
Basmati rice	30	4 cup	197	5	33	17
Chicken breast	195	140 g	273	43	60	20
Coca cola	26	370 mL	90	24	83	93
Total intake			1237		329	27%
Daily intake for a normal individual in one day	1237 (mg/day)					27%

Table 4. presents the mean and percentage of total and relative available phosphorus in home-processed foods (HPF) for a typical day's meals for an individual. Where 1 cup meal: 250g, 1oz potatoes: 28.42g, 1 Nan: 100g, 1cup beans: 253g, 1cup rice: 164g, 1oz yoghurt: 227g, 1 large egg; 50g, chicken breast: 140g, coca-cola: 370g.

Food samples	Total phosphorus (mg/100g)			*In vitro* extractable P (mg/100g)		*In vitro* extractable P %
	mg P/100 g	Rate	Daily	mg P/100 g	Daily/mg	%
Whole milk	69	1 cup	173	19	48	27
Potatoes	69	2 oz	39	9	5	11
Yoghurt	67	8-oz container	152	23	52	37
Nan bread	290	1 Nan	290	38	38	18
Beans in tomato sauce	91	1 cup	230	7	18	7
Basmati rice	38	4 cup	249	6	39	17
Chicken breast	192	140g	269	35	49	20
Coca-cola	26	370 mL	90	24	83	93
Eggs	255	2 large	225	38	38	15
Total intake			1717		370	27
Daily intake for a normal individual in one day		1717 (mg/day)			27%	

4. CONCLUSION

This study aimed to analyze the phosphorus content in two types of processed foods to evaluate their effects on dialysis patients. Most food samples in this study contained a reliable amount of total phosphorus with the exception of Nan bread and eggs. The highest concentration of total phosphorus was found in naan bread, whereas the lowest concentration was found in Coca-Cola. The availability of phosphorus differed considerably depending on the type of food. The lowest values were observed in basmati rice (MPF), while the highest values were found in chicken breast (MPF); both were below the expected range of 50-60%. The high phosphorus intake could be the main reason for the increased PO4 level in patients with end-stage chronic diseases. Therefore, the findings of this study provide valuable information for individuals with advanced-stage chronic kidney disease (CKD), helping them make informed dietary choices to protect their health and avoid foods that may exacerbate their condition. Further in vivo research is needed to evaluate the effects of home-processed foods and manufactured processed foods on patients with end-stage chronic kidney disease with a particular focus on foods such as Nan bread and egg that include high total P content. Patients with end-stage chronic kidney disease are recommended a varied diet consisting of both animal and plant products high in phytate with enough protein content as well. The consumption of processed foods should be restricted due to their high content of food additives. Additionally, certain home-prepared foods, such as naan bread and eggs, should also be consumed with caution. Patients are also recommended to increase their knowledge and awareness of the phosphorus content in commonly consumed foods. their knowledge about dietary phosphorus content in common foods.

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Declaration of Conflicting Interests and Ethics

The authors declare no conflict of interest. This research study complies with research and publishing ethics. The scientific and legal responsibility for manuscripts published in IJSM belongs to the authors.

Authorship Contribution Statement

All authors contributed equally to the writing of the article. In addition, all authors have read and agreed to the published version of the article.

Data Availability

Data supporting the findings of this study are available from the corresponding author upon request.

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