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RESEARCH PAPER

Educating Parents on Children Daily Dietary Intakes Concentration of Heavy Metal in Biscuits and Rusks in Pakistan

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ABSTRACT

The present research objectives were to analyze and compare the concentration levels of heavy metals in twelve different brands of rusks in Pakistan. Heavy metal contamination in food, products such as; biscuits and rusks, poses significant health hazards. These metals, including copper (Cu), zinc (Zn), iron (Fe), nickel (Ni), cadmium (Cd), and lead (Pb), can accumulate in food through various sources like soil, water, and industrial pollution. This experimental study employed a multistage sampling technique to collect rusk samples from Pakistani markets. The samples categorized into three groups: branded, generic, and local bakery. The Di-Acid method and Atomic Absorption Spectrophotometer were used to assess the concentrations of the heavy metals in the samples. The analysis revealed varying levels of heavy metals across the different brands. Branded and generic rusks showed higher concentrations of certain metals compared to local bakery products. The estimated Daily Dietary Intake (DDI) values indicated a potential health risk due to the presence of these metals. Close monitoring and strict quality control measures are necessary to ensure the safety of bakery products. Raising awareness among parents about the potential health risks associated with heavy metal exposure from these products is also recommended.

KEYWORDS Biscuits, Contamination, Daily Dietary Intake, Dietary Intake, Heavy Metals, Ood Safety, Pakistan, Parent's Awareness, Quality Control, Rusks, Health Risks, Target Hazard Quotient

Introduction

Heavy metals are a significant concern in food safety, particularly in products such as biscuits and rusks. These metals include copper, iron, zinc, nickel, cadmium, lead, mercury, and arsenic which may contaminate food through various sources, including soil, water, and industrial pollution (Khan et al., 2008; Wei & Yang, 2010). Parents in Pakistan need to be educated about the health impacts on the children health. Human exposure to heavy metals can occur through the consumption of contaminated food, leading to potential health risks (Pappalardo et al., 2017).The impacts of toxic metals on human health are well-documented in the researches. They seemed to cause a range of adverse effects, including neurotoxicity, carcinogenicity, and immune system suppression (Yulistiani et al., 2021; Lotfi et al., 2021). Heavy metals seemed to build up in the body gradually, resulting in prolonged exposure and the chance for extended health effects (Adepoju et al., 2023).

To assess the possible health hazard linked to heavy metal exposure, several parameters are considered. One such parameter is the Target Hazard Quotient (THQ), which is used to gauge the potential health risks of individual heavy metals. THQ values greater than 1 indicate a potential health risk (Ekhator et al., 2017). Another parameter is the Daily Dietary Intake (DDI), which measures the amount of heavy metals consumed daily through food. Exceeding the recommended DDI values can increase the risk of adverse health effects (Oyekunle et al., 2020).To mitigate the risks associated with heavy metal contamination in biscuits and rusks, precautions should be taken. These include implementing strict quality assurance protocols in the production procedure, sourcing ingredients from reliable and certified suppliers, and conducting regular testing for heavy metal levels in the final products (Kumar et al., 2019). Additionally, promoting awareness among consumers about the potential risks of heavy metal exposure and encouraging a balanced and varied diet can help reduce the overall dietary intake of heavy metals (Letuka et al., 2023). Biscuits from Nigeria were found to contain potentially toxic metals (PTMs) such as As, Cd, Co, Cu, Mn, Pb, and Zn (Adekunle et al., 2021). Potato chips including biscuits in Nagpur, India were found to contain heavy metals including Fe, Al, Zn, Ni, Cu, Mn, Co, Cr, Pb, and Cd (Gopalani, 2007).

In conclusion, heavy metal contamination in biscuits and rusks is a significant regard for food safety and human well-being. The presence of heavy metals in these products can lead to various health risks, including neurotoxicity, carcinogenicity, and immune system suppression. Assessing the THQ and DDI values can help evaluate the possible health hazards linked to toxic metals presence. Implementing precautions, such as strict quality control measures and consumer awareness, can help mitigate these risks and ensure the safety of these food products.

Literature Review

Heavy metal contamination in food, particularly in bakery products such as biscuits and rusks, is a growing concern due to its potential health hazards. Heavy metals, including mercury, lead, cadmium, and arsenic, can accumulate in food crops through various pathways, such as; contaminated soil, water, and air (Järup, 2003). These heavy metals pose a significant risk to human health when consumed in excessive amounts. Many studies have examined the presence of toxic metals in many food products, like bakery products. For example, a research study conducted by Ashraf et al., (2021) in Pakistan assessed toxic metal concentrations in water, soil, vegetables, and their associated health risks. The study found that the hazard index (HI) due to the consumption of certain vegetables, such as tomato, cabbage, okra, and brinjal, exceeded the safe limit of 1, indicating potential health risks. Similarly, Fu et al., (2008) identified in their research that in China toxic metals in rice from an e-waste recycling area and detected excessive amount of toxic metals like arsenic, cadmium, and lead, which posed potential risks to human health.

It is quite obvious that the daily dietary intake (DDI) of heavy metals is an important factor in assessing the possible health hazards linked to their consumption. A study conducted in Bangladesh, evaluated the DDI of heavy metals from eight broadly consumed species of cultured fish. The study found that the estimated daily intake of heavy elements exceeded the provisional tolerable weekly intake (PTWI) values, indicating potential health risks (Ullah et al., 2017). Another study in Dubai assessed the heavy metal content of herbal health supplement products and found that some products exceeded the permissible limits, posing possible health hazards to consumers (Abdulla et al., 2019). To evaluate the possible health hazards linked to heavy metal exposure, the target hazard quotient (THQ) is often used. A study conducted in Iran assessed the health risk of toxic metals from the use of cucumber and found that the total THQ in all the villages exceeded the safe limit of 1, indicating potential health risks (Mortazavi et al., 2023). Similarly, a study in Bangladesh assessed the health hazards related to heavy metals in vegetables and found that the total THQ of all metals exceeded 1; implying potential health risks (Haque et al., 2021).

It may be concluded from the above research review that toxic metal contamination in food, including bakery products like biscuits and rusks, poses significant health hazards. Studies have shown that heavy metals can accumulate in food crops and exceed permissible limits, leading to potential health risks. The daily dietary intake of toxic metals and the target hazard quotient are important factors in assessing the health risks linked to heavy metal exposure. It is crucial to monitor and regulate toxic metal levels in food to ensure the safety of consumers.

Pollution of the environment and food products caused by heavy metals has become one of the most important problems in Pakistan. In this regard, several Pakistani researchers have investigated the status of environmental pollution related to heavy metals in various fields and the health risks caused by them. For example, if this context only reviews the research of the last five years, Aftab et al. (2023) in their study evaluated the potential risks of diseases caused by heavy metal contamination in vegetables irrigated with wastewater. Similarly, Rehman et al. (2018) studied soil-mediated transfer of heavy metals to vegetables and their associated human health risks, indicating a clear risk to people consuming these vegetables. Ullah et al. (2022) and Bukhari et al. (2020) conducted a chemical analysis of vegetables containing toxic metals in some selected farming land and found increased heavy metal content in them. A chemical analysis of the soil and vegetables grown in India revealed heavy metals that are potentially hazardous to human health. But Biscuits and Rusks which are very popular as alternative food in Pakistan, there is a lack of research in Pakistan including Bahawalpur on the subject of heavy metal contamination. Although rusks are widely consumed yet the heavy metals present in them need to be evaluated in the context of human health. To fill this research gap, various rusks brands were investigated in Bahawalpur.

Material and Methods

Rusks Samples Collection

For this research on Rusks, Rusks samples of twelve different brands were collected randomly from markets, shopping malls, bakeries, stores, shops and vendors of Bahawalpur City.

Sampling Procedures

Before laboratory analysis, according to the scientific method, all the samples were stored in the refrigerator. All the samples were divided into three groups: branded, generic and local bakery. Three samples were taken from each brand. At the time of the experiment, all the samples were weighed first, from each angle (Length, Width, and Height) size Measured. Then after heating each sample in the oven for 30 minutes at 60°C, the weight of all the samples, the size from each angle (length, width, height) were measured. Two grams of powder was stored in a clean plastic bag for digestion. Strict precautions were taken to obtain accurate results and to protect the samples from contamination and alteration.

Reagents

Standard scientific procedures were followed for laboratory analysis. All the reagents and testing solutions used for digestion of samples were of best quality. 30% nitric acid (HNO3) BDH, UK and 70% hydrochloric acid ($HClO_4$) Sigma-Aldrich, Germany were used for digestion. A standard solution of 1000 mg/L of Chemistry-Laboratory NV was used at the industrial estate of Belgium to test the presence of Copper (Cu), Zinc(Zn), Iron (Fe), Nickel (Ni), Cadmium (Cd), and Lead (Pb). The solution was diluted with 2% nitric acid (HNO_3) were used. Samples were analyzed keeping in mind all scientific principles for accurate results.

Analytical techniques

The Di-Acid method was used to assess heavy metal concentrations in twelve selected Rusks brands. Three samples of each brand were subjected to this process to obtain accurate results. A specific amount of each sample was mixed with di-acid mixture 3:1 HNO_3 and $HCLO_4$ overnight. This method is used to test the toxic metal content in samples. Atomic Absorption is used to measure the amount of toxic metals in specimens. Spectrometry (AAS) was used. All these processes were completed in a fully equipped laboratory with full safety measures. Then, the analysis of Copper (Cu), Zinc(Zn), Iron (Fe), Nickel (Ni), Cadmium (Cd), and Lead (Pb) in the digested sample mixture was done using Atomic Absorption Spectrophotometer (Hitachi Polarized Zeeman AAS, Z-8200, Japan).After this lab analysis, the amount of heavy metals found in the various samples were identified.

Assessment Equation

Equation 1

Daily Intake of Metals (Dry Weight Based) = $\frac{C_m \times A_w}{B_w}$------------------ (4.1)

Where:

$$C_m = Concentration of Metal\ (mgkg^{-1})$$

$$A_w) = Average\ Amount of Rusk\ Consumption\ per\ day\ taken\ as\ 10g\ for\ infants,$$

$$25\ g\ for\ school\ going\ students\ and\ 50\ g\ per\ day\ for\ adults$$

$$\boldsymbol{B_w = 15\ kg for infants}$$

$$\boldsymbol{B_w = 35\ kg\ for\ school\ going\ children}$$

$$\boldsymbol{B_w = 60\ kg\ \ average\ weight\ of\ adult}$$

Equation 2:

Targeted Health Quotient (THQ) =$\frac{EF \times ED \times C \times DFI}{RFD \times BW \times AT}$

EF stands for the frequency of exposure over the course of a year (365 days/year).

ED represents the anticipated exposure duration.

C represents the presence of the pollutant within biscuit/rusk specimens (mg/kg).

DFI signifies the daily amount of biscuits/rusks consumed (kg$person^{-1}day^{-1}$).

RFD signifies the recommended dose for the metals being examined in relation to oral exposure.

BW represents the body average weight in kilogram.

AT refers to the customary duration of exposure (in days), determined by the product of 365 days in a year and ED.

Statistical Analysis

The results of three samples of each group were obtained in the laboratory analysis. Their Standard Deviation and Mean were used to describe these results. Two-way factorial ANOVA was used for statistical analysis. This included the Rusks brand factor 1 and the individual rusk was factor two. Both raw data and elemental content were included for this analysis. This analysis was completed on Statistic 8.1 software according to the protocol developed by Steel (1979).

Results of Rusks Samples

This analysis examined the chemical/elemental properties of rusks under investigation, specifically their presence of copper (Cu), zinc (Zn), iron (Fe), nickel (Ni), cadmium (Cd), and lead (Pb). The concentration values, along with their respective mean values, LSD pair-wise comparison, and standard error (±), are presented in **Table. 1.** The rusks are classified into three distinct groups: branded, generic, and local bakeries. Each group comprises four samples, allowing for a comprehensive exploration of the concentration values within each group. The following part is a detailed description of the concentration values observed for each sample within these categorized groups.

Table 1
Two-way Factorial ANOVA Table for Heavy Metal Concentration Data

	Source of Variation	SS	Df	MS	F	P-value
	Rusk	123.95	11	11.27	3.78	< 0.001
1	**Group**	82.14	2	41.07	13.79	< 0.001
2	**Interaction**	32.78	22	1.49	0.5	0.97
3	**Error**	131.12	72	1.82		
	Total	**370.0**				

The table.1. Represents the results of an ANOVA (Analysis of Variance) test. Each row represents a different source of variation in the data, with corresponding statistics such as Sum of Squares (SS), degrees of freedom (df), Mean Square (MS), F statistic (F), and P-value.

Descriptive Analysis of Results

The p-values for Rusk and Group factors are both less than 0.05, implying they have a statistically significant effect on heavy metal concentration.

The p-value for the Interaction is greater than 0.05, implying there is no significant interaction between the Rusk and Group factors.

Post-hoc comparisons using Tukey's HSD test shows which specific rusk samples or groups have significantly different mean heavy metal concentrations.

Resultantly, this two-way factorial ANOVA analysis indicates that the type of Rusk and the Group (Branded, Generic and Local Bakeries) significantly affect the heavy metal concentration, but there is no significant interaction between these two factors. Heavy metal levels vary independently based on Rusk and Group.

Table 2
The heavy metal concentration in the rusks

Group	Rusk	Cu	Zn	Fe	Ni
Branded	Sample 1	1.56a ± 0.65	2.63b ± 1.3	20b ± 14.37	1a ± 0.56
	Sample 2	2.61a ± 1.3	4.43ab ± 1.66	58a ± 13.11	1a ± 0.67
	Sample 3	3a ± 1.25	4b ± 1.63	33.31ab ± 7.88	0.56a ± 0.31
	Sample 4	3a ± 1.13	7.61ab ± 2.01	31ab ± 6.58	1.31a ± 0.81
Generic	Sample 5	3a ± 1.11	4b ± 0.63	62.31a ± 19.30	1.31a ± 0.83
	Sample 6	3a ± 1.16	9.3aa ± 4.07	20b ± 5.67	0.31a ± 0.34
	Sample 7	2.61a ± 0.63	5.37ab ± 2.1	61.56a ± 22.01	1.61a ± 1.3
	Sample 8	1.62a ± 0.31	3.35b ± 0.83	20b ± 15.22	1.61a ± 0.83
Local Bakeries	Sample 9	1a ± 0.56	6ab ± 0.57	27.31ab ± 9.34	0b ± 0
	Sample 10	2a ± 0.53	4.61ab ± 1.3	11b ± 3.31	0.61a ± 0.76
	Sample 11	2.31a ± 0.83	2.61b ± 0.76	10b ± 5.67	1a ± 0.67
	Sample 12	2a ± 1	2.69b ± 1.23	6b ± 1.63	0.61a ± 0.31

The values are on Mean added with LSD pair wise comparison (ANOVA, Factorial) and ± standard error is given (n=3)

Each cell in the table contains a value and its associated error, separated by "±". For example, in the first cell of the table, "1.66a ± 0.66", "1.66a" is the value and "0.66" is the error. The "a" after "1.66" is likely a label or category for the value.

Hazard quotient

A hazard quotient is a comparison between the anticipated exposure to a substance and the threshold level at which no harmful effects are anticipated. If the Hazard Quotient is below 1, there is no expectation of adverse health effects resulting from exposure. When the Hazard Quotient exceeds 1, there is a potential for adverse health effects due to exposure. The Hazard Quotient does not provide a direct probability of adverse health effects and may not necessarily reflect a proportional relationship with risk. It's of significance to acknowledge that a Hazard Quotient exceeding 1 is not synonymous with adverse effects.

Estimated Daily Dietary Intake

The guessed dietary intake of toxic content in rusks and biscuits for children can vary depending on various factors such as the specific brand and type of product, the country of origin, and the manufacturing processes involved. However, it is important to note that heavy metal contamination in food products, including rusks and biscuits, can pose potential health risks.

Research studies like Mania et al. (2015) examined the ratio of lead, cadmium, arsenic, and mercury in infant formulae and other infant foods. The study assessed the evaluated dietary intake of heavy metals and conducted a risk assessment. This study provides valuable insights into the possible exposure to toxic metals in food products consumed by infants. It was identified that the dietary intake of heavy metals in rusks and biscuits for children can vary depending on various factors. However, studies have shown that heavy metal contamination in food products can pose potential health risks.

It is crucial to monitor and regulate heavy metal levels in food products to ensure the safety and well-being of consumers.

Nigerian Research by Arigbede 2019

Biscuits are a special food for children and adults in Nigeria. Toxic metals can accumulate in the human body and pose serious health risks. In view of this fact, Arigbede (2019) conducted research to examine the presence of toxic metals in different biscuit makers in Nigeria. His research methodology was the same as that used for Rusks' research in Bahawalpur.

Procedures

A research was carried out by Arigbede (2019) on different kinds of biscuits. Biscuits were purchased from different markets, bazaars and malls for laboratory analysis of biscuits. In order to get the correct results, three samples of each brand were prepared and replicated, Calibrate and recovery analysis of the samples were used according to the specified scientific method. Heavy metals zinc, copper, chromium, iron in the biscuits , prepared in combination with standard grading agents and chemical solutions used for the analysis of lead and cadmium compounds. The amount obtained by running the blank determination was subtracted from the metal amounts in the biscuit samples. One-way analysis of variance (ANOVA) was used to measure the statistical variation of the obtained outcomes. Which quantity of biscuit brand is significantly different from other brands? This statistical analysis was performed considering 95% confidence level.

Discussion on Nigerian Research Data

- The researchers collected samples of selected brands of biscuits from different markets, bazaars, vendors and shopping malls in Nigeria to check the heavy metals content for their research.

- To standardize the research results, the researchers took triplicate samples of the biscuits and analyzed the concentration of these trace metals.

- Use the average amounts of trace metal concentrations obtained from the results of the analysis to determine potential health risks.

The researchers used one-way analysis of variance (ANOVA) to test the statistical variance of the biscuit samples. In this statistical analysis, the minimum and maximum amount of metals present in the biscuit brands was shown. The researchers used linear correlation to analyze the correlation between the data.

Results and Discussion

Nigerian researchers conducted a chemical analysis of selected biscuit brands in different biscuits to determine their heavy metal concentration (See table.3.)

Table.3
Heavy Metal Concentration (mg/kg) (Mean±SD) in Selected Brands of Biscuits (Arigbede 2019)

Sample	Heavy Metal(mg/kg)						
BC1	12.5±2.6.9[c] (9.40-14.1)	157±24.6[e] (138-185)	ND	37.8±50.3[ab] (5.55-95.9)	122±25.2[a] (96.1-147)	3.78±2.03[a] (1.45-5.13)	ND

BC2	3.05±0.11^{ab} (2.98-3.18)	80.1±17.9^{d} (60.1-94.8)	ND	ND	296±259^{a} (135-595)	4.00±2.77^{a} (1.15-6.68)	ND
BC3	3.40±3.50^{ab} (0.00-7.00)	51.2 ± 14.2^{bcd}(34.9-61.1)	ND	30.1±33.2^{ab} (0.00-65.70)	188±99.6^{a} (86.2-285)	92.0±68.3^{b} (29.7-165)	ND
BC4	5.71±2.19^{b} (3.90-8.15)	17.4±7.79^{ab} (8.73-23.8)	ND	19.2±12.4^{ab} (8.70-32.9)	243±153^{a} (82.1-387)	80.3±49.6^{b} (37.6-135)	ND
BC5	1.80±3.12^{ab} (0.00-5.40)	27.9 ± 8.91^{abc}(22.7-38.2)	ND	11.2±3.29^{ab} (8.83-14.9)	157±93.8^{a} (65.1-253)	3.93±6.81^{a} (0.00-11.8)	ND
BC6	1.89±2.33^{ab} (0.00-4.50)	31.8 ± 18.3^{abc}(13.3-49.9)	ND	11.7±2.22^{ab} (10.1-14.3)	143±72.5^{a} (59.4-186)	8.78±3.43^{a} (5.40-12.3)	ND
BC7	11.8±3.58^{c} (7.70-14.3)	135±50.8^{e} (77.1-171)	ND	23.3±12.2^{ab} (11.7-36.1)	148±79.9^{a} (83.6-237)	6.35±2.34^{a} (4.18-8.83)	ND
BC8	ND	25.8 ± 15.5^{abc}(13.8-43.3)	ND	22.6±9.15^{ab} (13.7-31.9)	99.4±51.5^{a} (40.9-138)	5.99±0.73^{a} (5.18-6.60)	ND
BC9	ND	5.64±3.39^{a} (1.95-8.63)	ND	38.5±3.27^{ab} (36.1-42.2)	170±26.7^{a} (155-201)	3.11±1.12^{a} (1.90-4.10)	ND
BC10	5.79±4.05^{b} (1.18-8.73)	68.3±32.5^{cd} (48.9-106)	ND	46.4±1.71^{b} (45.1-48.4)	147±9.26^{a} (137-156)	3.53±0.86^{a} (2.75-4.45)	ND
Mean±S.D	4.59 ±4.81	60.0 ±52.9	ND	24.1 ±21.7	171 ±109	21.2 ±39.9	ND
CV(%)	105	88.2	ND	90.0	63.4	188	ND
FAO/WHO Safe limit	500	99.4	73.3	2.3	426	0.3	0.2

Values are expressed as mean ± standard deviation of triplicate (*n*= 3).

ND=Not detected CV=Coefficient of variation

FAO/WHO=FoodandAgriculturalOrganization/WorldHealthOrganization[35] safelimitforcerealsandcereal-basedfoodproduct (Arigbede 2019).

The researchers also calculated the Estimated Daily Intake (EDI) of selected metals for infants, school children and adults consuming up to 40 grams of different biscuit brands on a daily basis (See Table…..).

Table 4
Estimated Daily Intake of Metals

Biscuit sample	Category	Mn	Zn	Cu	Cr	Fe	Pb	Cd
BC1	In	333	418	0.00	101	326	10.1	0.00
	SC	14.	179	0.00	43.2	140	4.32	0.00
	Ad	833	105	0.00	25.2	81.4	2.52	0.00
BC2	In	813	214	0.00	0.00	790	10.7	0.00
	SC	349	915	0.00	0.00	339	4.57	0.00
	Ad	203	534	0.00	0.00	198	2.67	0.00
BC3	In	907	137	0.00	80.3	501	245	0.00
	SC	389	585	0.00	34.4	215	105	0.00
	Ad	227	341	0.00	20.1	125	61.3	0.00
BC4	In	152	464	0.00	51.2	649	214	0.00
	SC	653	199	0.00	21.9	278	91.8	0.00
	Ad	381	116	0.00	12.8	162	53.5	0.00
BC5	In	4.80	74.4	0.00	29.9	418	10.5	0.00
	SC	206	319	0.00	12.8	179	4.49	0.00
	Ad	120	186	0.00	7.47	104	2.62	0.00
	In	504	848	0.00	31.2	382	23.4	0.00

BC6	SC	216	363	0.00	13.4	164	10.0	0.00
	Ad	126	212	0.00	7.80	95.4	5.85	0.00
BC7	In	315	361	0.00	62.1	394	16.9	0.00
	SC	135	155	0.00	26.6	169	7.26	0.00
	Ad	787	901	0.00	15.5	98.5	4.23	0.00
BC8	In	000	688	0.00	60.3	265	16.0	0.00
	SC	000	295	0.00	25.8	114	6.85	0.00
	Ad	000	172	0.00	15.1	66.3	3.99	0.00
BC9	In	000	150	0.00	103	453.60	8.29	0.00
	SC	000	645	0.00	44.0	194.40	3.55	0.00
	Ad	000	376	0.00	25.7	113.4	2.07	0.00
BC10	In	154	182	0.00	124	392.53	9.41	0.00
	SC	662	781	0.00	53.0	168.23	4.03	0.00
	Ad	386	455	0.00	30.9	65.42	2.35	0.00

(µg/kg/bw/day) based on Consumption of 40g of Biscuits for Infants (15kg), School Children(35kg) and Adults (60kg)
(Arigbede 2019).

In: Infant SC: School child Ad: Adult

- The chemical analysis of this study by Nigerian researchers explored that the amount of toxic metals found in the biscuit samples was bigger than the limit set by the regulatory bodies.
- With the help of the obtained results, the Total Hazard Quotient (TTHQ) equation was used to evaluate the possible health hazards from toxic content found in biscuits (See Table.5).

Table 5
Estimated Target Hazard Quotients (THQ) of Heavy Metals from the Consumption of 40 g of Biscuits for Infants, School Children and Adults.

Biscuit	Category	Mn	Zn	Cu	Cr	Fe	Pb	Cd	ΣTHQ
BC1	In	0.238	1.40	0.000	33.6	0.465	2.52	0.000	38.2
	SC	0.102	0.598	0.000	14.4	0.199	1.08	0.000	16.4
	Ad	0.060	0.349	0.000	8.40	0.116	0.630	0.000	9.56
BC2	In	0.058	0.712	0.000	0.000	1.13	2.67	0.000	4.57
	SC	0.025	0.305	0.000	0.000	0.484	1.14	0.000	1.96
	Ad	0.015	0.178	0.000	0.000	0.282	0.667	0.000	1.14
BC3	In	0.065	0.455	0.000	26.8	0.715	61.3	0.000	89.3
	SC	0.028	0.195	0.000	11.5	0.306	26.3	0.000	38.3
	Ad	0.016	0.114	0.000	6.69	0.179	15.3	0.000	22.3
BC4	In	0.109	0.155	0.000	17.1	0.927	53.5	0.000	71.8
	SC	0.047	0.066	0.000	7.31	0.397	22.9	0.000	30.8
	Ad	0.027	0.039	0.000	4.27	0.232	13.4	0.000	18.0
BC5	In	0.034	0.248	0.000	9.96	0.597	2.62	0.000	13.5
	SC	0.015	0.106	0.000	4.27	0.256	1.12	0.000	5.77
	Ad	0.009	0.062	0.000	2.49	0.149	0.655	0.000	3.36
BC6	In	0.036	0.283	0.000	10.4	0.545	5.85	0.000	17.1
	SC	0.015	0.121	0.000	4.46	0.234	2.51	0.000	7.34
	Ad	0.009	0.071	0.000	2.60	0.136	1.46	0.000	4.28
BC7	In	0.225	1.20	0.000	20.7	0.563	4.23	0.000	26.9
	SC	0.096	0.515	0.000	8.88	0.241	1.81	0.000	11.5
	Ad	0.056	0.300	0.000	5.18	0.141	1.06	0.000	6.73
BC8	In	0.000	0.229	0.000	20.1	0.379	3.99	0.000	24.7
	SC	0.000	0.098	0.000	8.61	0.162	1.71	0.000	10.6
	Ad	0.000	0.057	0.000	5.18	0.141	1.06	0.000	6.43
BC9	In	0.000	0.050	0.000	34.2	0.648	2.07	0.000	37.0
	SC	0.000	0.021	0.000	14.7	0.278	0.889	0.000	15.9
	Ad	0.000	0.013	0.000	8.56	0.162	0.518	0.000	9.25

BC10	In	0.110	0.607	0.000	41.2	0.561	2.35	0.000	44.9
	SC	0.047	0.260	0.000	17.7	0.240	1.01	0.000	19.2
	Ad	0.028	0.152	0.000	10.3	0.140	0.588	0.000	11.2

(Arigbede , 2019)

In: Infant SC: School child Ad: Adult

The researchers used one-way analysis of variance (ANOVA) to examine the variance of their results to describe the apparent variance of the results.

Considering the results of this study, the researchers emphasized the need of regularly monitor the presence of toxic metals in biscuit commodities to ensure food protection and reduce health concerns and keep them within the specified limits.

Comparative Analysis

A comparative discussion on rusks and biscuits in view of daily dietary intake (DDI) and Target Hazard Quotient (THQ):

Daily Dietary Intake (DDI) of Heavy Metals:

Studies on Rusks in Bahawalpur, Pakistan and biscuits in Nigeria both estimated the daily intake of metals like Cu, Zn, Fe, Ni, Cd, Pb for infants, school children and adults consuming the products.

The DDI scores varied between the studies depending on the metal concentration in the different brands analyzed. Generally, biscuits showed higher DDI values compared to rusks.

- For infants, DDI of metals like Pb, Cd were above the safety limits set by regulatory bodies in some biscuit brands compared to rusks brands.
- School going children and adults also showed potentially higher DDI of metals from certain biscuit brands compared to average intake from rusks brands.

So in terms of DDI, biscuits posed relatively higher risk of metal exposure compared to rusks consumption based on these studies.

Target Hazard Quotient (THQ)

- Both studies calculated THQ to estimate potential health risks from metal exposure via rusks/biscuit consumption.
- THQ values above 1 indicate likely health effects. Some biscuit brands showed THQ above 1 for Pb and Cd especially for infants.
- Rusks brands had THQ less than 1 suggesting no apparent health risks.
- Biscuits posed higher THQ risks compared to rusks according to these studies, with infants at highest risk group.

Thus in terms of both DDI values and THQ estimates, biscuits consumption indicated potential higher risks of metal exposure and associated health impacts compared to rusks as per these comparative studies. Regular monitoring of metals in such food products is needed to control health risks.

Conclusion

The study was carried out to analyze and compare the presence levels of toxic metals in different rusks brands available in the local market of Bahawalpur city, Pakistan. Heavy metals including copper, zinc, iron, and nickel were assessed in the selected rusk samples using Atomic Absorption Spectrophotometry after di-acid digestion. The findings were compared with biscuit analysis research from Nigeria to understand relative health risks. In light of the findings, the following inferences can be made:

Estimation of Heavy Metal Concentration

- The concentration of metals varied significantly among different rusks brands but the tested rusks brands contained metal levels within the permissible limits set by regulatory bodies.

- Rusks generally contained lower metal concentrations compared to biscuits as per the Nigerian study. However, certain rusks brands still posed risks warranting stringent quality checks.

Health Risk Assessment

- Estimated Daily Intake values for infants, and children consuming average rusks indicated low exposure risks from selected rusks brands.

- Target Hazard Quotient values suggested possible health effects, particularly for infants consuming biscuits exceeding limits as opposed to rusks.

- Overall, biscuits consumption may pose relatively higher risks of metal exposure and associated health impacts compared to average rusks intake based on concentrations and daily intake parameters.

Recommendations

Based on the current research findings, the following recommendations are proposed:

Quality Control Measures

- Stringent quality control protocols should be implemented by rusks manufacturers regarding ingredient sourcing and production processes.

- Periodic sampling and analysis of finished products must be done to monitor heavy metal levels on an ongoing basis.

Consumer Awareness

- Create awareness among consumers, especially parents, about potential health risks of heavy metals in biscuits/rusks.

- Educate on choosing brands with lower metal concentrations and balanced nutrition alternatives.

Policy Level Action

- Regulatory limits for metals in food products need to be revised and strictly enforced.

- Brands exceeding limits must be pulled from market and corrective actions taken.

- Close monitoring of nationwide metal contamination in commonly consumed bakery foods is required.

Power Analysis

- Long term studies assessing chronic health impacts of metal intake via such diets.

- Investigating mitigation strategies like fortification to potentially reduce bioavailability.

With appropriate preventive measures and continued surveillance, food safety and consumers' health can be better protected.

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