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Assessment of Heavy Metals and Microbial Load in Tomato Ketchup Used in Local Market of Southern Punjab

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ABSTRACT

Tomatoes are globally renowned as one of the most widely cultivated agricultural commodities, indispensable staples in people's everyday diet. Tomato ketchup is susceptible to several types of microbiological contamination that could potentially pose a risk to the product purity. So, the main purpose of this study is to evaluate the presence of heavy metals and microbial containments in locally available ready to eat tomato ketchup in Multan. Results indicated that while levels of heavy metals such as lead and cadmium in some samples were near or exceeded permissible limits, unlikely can cause health hazards. Nonetheless, prolonged exposure could pose significant health hazards. The widespread use of wastewater in agriculture and increasing urbanization contribute to this contamination. All tested samples exhibited bacterial counts exceeding recommended reference levels, highlighting potential health concerns. The study underscores the importance of regular monitoring and provides a basis for effective risk communication and management strategies related to food safety. The study concludes that locally branded ready-to-eat tomato ketchup in Multan poses minimal risk regarding heavy metal contamination. However, non-branded ketchup samples showed higher chances of containing heavy metals and microbial contaminants, making them potentially harmful. While current levels of lead (Pb) and cadmium (Cd) may not cause immediate health effects, prolonged exposure could lead to serious health concerns. Urbanization, metal processing technologies, and wastewater use in farming contribute to increased contamination. All samples exceeded acceptable bacterial counts, highlighting the need for strict monitoring. Ensuring food safety requires regular surveillance of both heavy metal content and microbiological quality in processed foods.

INTRODUCTION

Tomatoes are globally renowned as one of the most widely cultivated agricultural commodities, indispensable staples in people's everyday diet (Collins et al., 2022). Yet, when ingested as part of a diet including tomatoes, tomatoes are recognized as the primary source of the carotenoid lycopene that helps to strengthen the defense against some forms of cancer and cardiovascular disorders (Bhowmik et al., 2012). As fresh tomatoes are harvested in their raw state, they lack durability and, once matured, they deteriorate rapidly (Arah et al., 2015). Our tomatoes undergo conversion into various products such as tomato juice, puree, cocktail, paste, ketchup, sauce, jelly, soups, powder, and tomato chutneys. Driven by market demands and the substantial profits potential in this sector, there has been a swift proliferation of companies specializing in the sale of sauce and ketchup. Typically, the individual engaged in the manufacturing of the final product will fail to detect the existence of heavy metals (Porretta, 2019).

Food safety refers to the procedures implemented to ensure that food is safe for human consumption, by following the necessary standards and regulatory guidelines. It includes measures to prevent the primary factors contributing to instances of foodborne illnesses and contamination, such as proper handling, processing, storage, and preparation of food, as well as thorough cleaning. The primary tenets of food safety are the regulation of temperature, prevention of cross contamination, and maintenance of cleanliness throughout food processing (Beyene et al., 2023). Food contamination refers to the presence of pathogenic microorganisms, poisonous substances, chemicals, viruses, or parasites in food that may pose health hazards (Beyene et al., 2023). Contamination can arise at any distinct stage, ranging from the production of food items to their consumption. There exist various forms of food contamination. Biogenic molecules refer to the existence of microorganisms such as bacteria (e.g., Salmonella, E. coli), viruses (e.g., nor virus), and parasites or parasitic waste. Chemical pollution occurs when hazardous chemical compounds are present in the form of pesticides, food additives, or naturally poisonous substances (Thompson & Darwish, 2019). Physical contamination refers to the introduction of undesirable substances such as glass, metal, or plastic into food products. To obtain comprehensive information about food contamination and its impact on public health, one might refer to the resources provided by the World Health Organization (WHO) and French Academy of Nutrition (FAO). Statistics indicate that Salmonella is widely recognized as the premier foodborne pathogen on a global scale. The finding underscores the presence of small quantities of Salmonella cells that might lead to an infection, therefore underscoring the necessary adherence to rigorous food hygiene protocols (Mkangara, 2023).

Tomato ketchup is susceptible to several types of microbiological contamination that could potentially pose a risk to the product purity (Arah et al., 2015). The microorganisms that may be present include Salmonellae from raw materials, which can cause serious foodborne illness (Yar et al., 2024); Escherichia coli, both pathogenic and those that can contaminate ketchup from raw tomato spoilage or processing; and Wisteria monocytogenes, which thrive in refrigeration and make a challenge especially to sensitive personnel. Moreover, yeasts and molds have the potential to contaminate the ketchup and perhaps generate mycotoxins that may be harmful to the body's systems if consumed. Although less prevalent, viral infections, ideally transmitted by water sources contaminated with the virus, or through the management of food products infected by the virus, result in epidemics of the virus. Hence, the presence of elevated levels of heavy metals in tomato ketchup might be a significant potential issue, as these contaminants may be acquired from sources (Mehrin et al., 2020). Currently, commercially available tomato ketchup in major retailers contains very hazardous heavy metals like lead, cadmium, and arsenic, which often exceed the acceptable levels established by food safety authorities.

In the light of this context, the main aim of this research was to conduct an analysis of possible origins of heavy metals and microbiological contamination throughout the manufacturing and processing stages as well as the evaluation of the possible health hazards linked to the ingestion of contaminated ketchup, particularly for susceptible demographics.

MATERIAL AND METHOD

Study Site

The study was conducted in the Southern Punjab of Pakistan, Multan. Multan is located at 30.18° North latitude, 71.49° East longitude. Multan has a expected population of 1.872 million. Four tomato ketchup brands were selected namely as brand A, brand B, brand C and brand D (brand identities are hidden).

Sample Collection

Branded and unbranded (open ketchup) samples were collected from the markets of Multan city of Pakistan. A total of 24 samples including 12 branded (4 brands in triplicate) and 12 unbranded samples (collected from 4 retail shops in triplicate) were collected.

Determination of Heavy Metals by Atomic Absorption Spectrometer (AAS)

Initially, the samples underwent a meticulous washing process to eliminate all traces of mud and grit, followed by a thorough rinsing using distilled water. The tomatoes were peeled. The tomato samples were subsequently chopped into tiny pieces using a knife. Subsequently, the samples were air-dried for a few days and then subjected to microwave heating at temperatures ranging from 45 to 80 °C until a consistent weight was achieved. Once dried, the particles were pulverized using an electric grinder to achieve a minuscule size and promptly placed in new plastic containers for subsequent examination. The wet acid digestion procedure was performed using the standard techniques described in the AOAC (Association of Official Analytical Chemists) report from 1990. 1.0 gram of each dried sample was combined with 10 milliliters of concentrated nitric acid in a 50-milliliter beaker. The mixture was then heated on an electric hot plate for 1 hour to get a partially dried sample. Once again, 10 milliliters of concentrated nitric acid (HNO3) and 4 milliliters of hydrogen peroxide (H_2O_2) were added to the mixture. The solution was then placed on a hot plate and quickly cooked. The addition of HNO_3 and H_2O_2 was maintained until the solution became colorless, and its volume was lowered to 2-3ml. The mixture was subjected to cooling and filtration using a Whatman filter paper. The filtrate was preserved in 10 ml sample bottles or vials. Prior to analysis with the Atomic Absorption Spectrometer, the sample was diluted with de-ionized water to a volume of 25 ml.

The metals that were chosen include arsenic (As), cadmium (Cd), lead (Pb), iron (Fe), zinc (Zn), nickel (Ni), and cobalt (Co). Calibrated standards were created by diluting the commercially available stock solution, which was in the form of a 1000 ppm aqueous solution. Highly purified de-ionized water was utilized to make the working standards. The glass equipment utilized during the analytical procedure were soaked in 8N HNO3 overnight and rinsed with several changes of de-ionized water before being employed.

Microbial Culture Analysis

The samples were diluted by combining 5 grams of food with 50 ml of buffered peptone water and rapidly shaken to remove germs that were attached. The liquid phase is used to create the initial stock sample, which is subsequently diluted to achieve dilutions ranging from 10^1 to 10^{10}. Following the dilution, a volume of 0.1 ml was deposited onto a sterile plate count agar (PCA) (Oxoid Ltd, Basingstoke Hants, England). The sample was then spread evenly on the surface of the agar and then placed in an incubator. The plates were placed in an incubator and kept at a temperature of 37 degrees Celsius overnight. The remaining stock samples were subjected to incubation at a temperature of 37 degrees Celsius for a duration of 4 hours. Following this, they were transferred onto MacConkey Agar plates (manufactured by Oxoid Ltd, located in Basingstoke Hants, England) and incubated overnight at a temperature of 37 degrees Celsius.

Viable Bacterial Count

Following an incubation period of one night, the number of colonies ranging from 30 to 300 on the PCA medium was enumerated. The bacterial counts were quantified by expressing them as the logarithm of colony-forming units per milliliter for liquid food samples or per gram for solid food samples that were tested.

Statistical Analysis

The researchers employed one-way analysis of variance (ANOVA) to ascertain statistically noteworthy disparity, with a 5% probability level, among the examined metals and microorganisms in the different samples. Additionally, Monte-Carlo simulation was utilized to estimation the nutritional consumption of heavy metals by the general population.

RESULTS

Analysis of Heavy Metals in the Tomato Ketchup Samples

The branded samples exhibit arsenic levels ranging from 0.03 to 0.06 mg/kg, but the non-branded samples demonstrate greater amounts, reaching a maximum of 0.13 mg/kg. Significantly, a particular unbranded sample surpasses the recommended limit of 0.1 mg/kg set by the Punjab Food Authority (PFA), suggesting possible health hazards. ANOVA analysis demonstrates a statistically significant disparity in arsenic levels between branded and non-branded samples ($p < 0.05$), indicating that non-branded items are more prone to having elevated quantities of arsenic. The amounts of cadmium in the samples differ, although most branded ketchups typically comply with the PFA limit of 0.05 mg/kg. Nevertheless, two samples without a brand name surpass this threshold, with levels of 0.09 and 0.10 mg/kg. The statistical study reveals notable disparities in cadmium concentrations among the many ketchup varieties, underscoring a potential issue with unbranded items. The levels of lead in both branded and non-branded samples are worrisome. Although branded samples generally stay under the PFA limit of 0.1 mg/kg, a few non-branded samples surpass this barrier, with the highest concentration reaching 0.21 mg/kg. The ANOVA test verifies large disparities in lead levels, indicating an elevated hazard of lead contamination in unbranded ketchups. The iron (Fe) concentration in all samples is significantly lower than the PFA limit of 15 mg/kg. Nevertheless, samples without a brand tend to exhibit somewhat greater levels of iron, but these disparities do not reach statistical significance ($p > 0.05$). This implies that although iron pollution is not a significant worry, it still requires careful observation. The concentrations of zinc (Zn), nickel (Ni), and cobalt (Co) in all samples comply with the PFA standards. There are no notable distinctions in the amounts of these elements in branded and non-branded items. This suggests that these metals do not pose significant issues in the ketchup samples that were examined.

The ANOVA analysis reveals statistically significant variations in the levels of arsenic, cadmium, and lead between the ketchup samples from different brands. The unbranded samples typically exhibit elevated amounts of these heavy metals, with several samples beyond the permitted thresholds established by the Punjab Food Authority. This presents a possible hazard to consumers' health, particularly in relation to lead, which is recognized for its neurotoxic properties. The ANOVA analysis revealed a statistically significant relationship between arsenic, cadmium, and lead. This highlights the need for stricter monitoring and regulation of non-branded food goods. The elevated levels of these toxic metals in generic ketchups indicate potential problems with the acquisition of raw materials, manufacturing procedures, or inadequate quality assurance protocols.

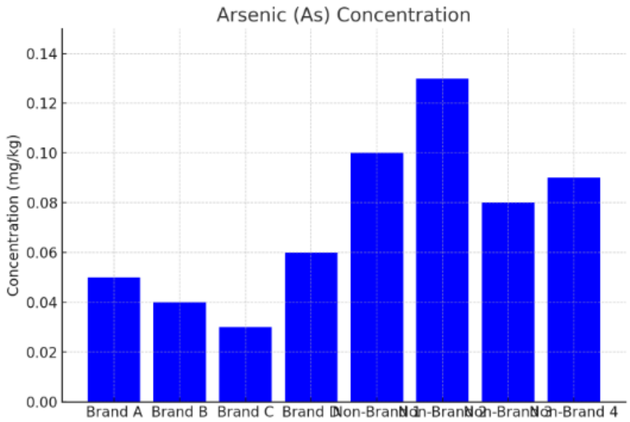
However, the absence of notable disparities in iron, zinc, nickel, and cobalt concentrations across the various varieties of ketchup suggests that these metals are of lesser importance in this particular situation. Nevertheless, it is vital to engage in ongoing surveillance to guarantee that these levels persist under secure thresholds, since any escalation might conceivably provide a health hazard. Ultimately, the research affirms that branded ketchups generally exhibit a higher level of safety in relation to heavy metal contamination. Conversely, non-branded goods may necessitate more stringent supervision and regulation to safeguard public health. These findings emphasize the need of consumers exercising caution when buying unbranded food goods and the necessity for regulatory agencies to implement stricter quality control methods.

Table 1
Tukey HSD All-Pairwise Comparison Test for Heavy Metals

Ketchup Type	As (mg/kg)	Cd (mg/kg)	Pb (mg/kg)	Fe (mg/kg)	Zn (mg/kg)	Ni (mg/kg)	Co (mg/kg)
Brand A	0.0466 ± 0.0088^{bc}	0.0259 ± 0.0111^{b}	0.0684 ± 0.0106^{b}	1.2672 ± 0.0005^{a}	0.7123 ± 0.1567^{a}	0.1511 ± 0.0267^{b}	0.0366 ± 0.0091^{b}
Brand B	0.0556 ± 0.0066^{bc}	0.0352 ± 0.0103^{b}	0.0757 ± 0.0111^{b}	1.2663 ± 0.0014^{a}	0.4273 ± 0.1661^{a}	0.1456 ± 0.0210^{b}	0.0443 ± 0.0375^{b}
Brand C	0.0345 ± 0.0237^{c}	0.0251 ± 0.0102^{b}	0.0572 ± 0.0116^{b}	1.2662 ± 0.0022^{a}	0.6839 ± 0.2813^{a}	0.1163 ± 0.0200^{b}	0.0272 ± 0.0094^{b}
Brand D	0.0678 ± 0.0100^{b}	0.0498 ± 0.0368^{b}	0.1087 ± 0.0312^{b}	1.2656 ± 0.0021^{a}	0.5904 ± 0.3827^{a}	0.1757 ± 0.0279^{b}	0.0572 ± 0.0225^{b}
Non-brand 1	0.1245 ± 0.0105^{a}	0.0886 ± 0.0236^{bc}	0.1678 ± 0.0336^{b}	1.2674 ± 0.0028^{a}	0.5301 ± 0.1986^{a}	0.2781 ± 0.0205^{b}	0.1112 ± 0.0126^{ab}
Non-brand 2	0.1188 ± 0.0237^{a}	0.0766 ± 0.0102^{cd}	0.1467 ± 0.0116^{b}	1.2653 ± 0.0022^{a}	0.6612 ± 0.2813^{a}	0.2476 ± 0.0200^{b}	0.0838 ± 0.0094^{bc}
Non-brand 3	0.1263 ± 0.0100^{a}	0.1128 ± 0.0368^{ab}	0.2105 ± 0.0312^{a}	1.266 ± 0.0021^{a}	0.4084 ± 0.3827^{a}	0.3493 ± 0.0279^{a}	0.1322 ± 0.0225^{a}
Non-brand 4	0.1374 ± 0.0105^{a}	0.1207 ± 0.0236^{a}	0.2402 ± 0.0336^{a}	1.2659 ± 0.0028^{a}	0.5659 ± 0.1986^{a}	0.3778 ± 0.0205^{a}	0.1359 ± 0.0126^{a}

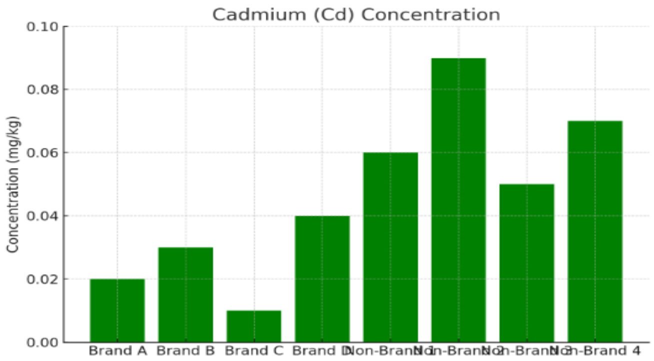
The concentration of arsenic in the analysed ketchups differed among the specific brands and non-branded goods. The branded items in general had reduced amounts of arsenic, suggesting superior management of raw materials and manufacturing procedures as shown in Fig. 1. The Punjab Food Authority (PFA) has established the highest permissible thresholds for arsenic in processed food items. Although non-branded samples were closer to the threshold than the allowed limits, all examined samples were within the acceptable range, underscoring the necessity for rigorous quality control.

Figure 1
Concentration of Arsenic in Different Ketchup Samples



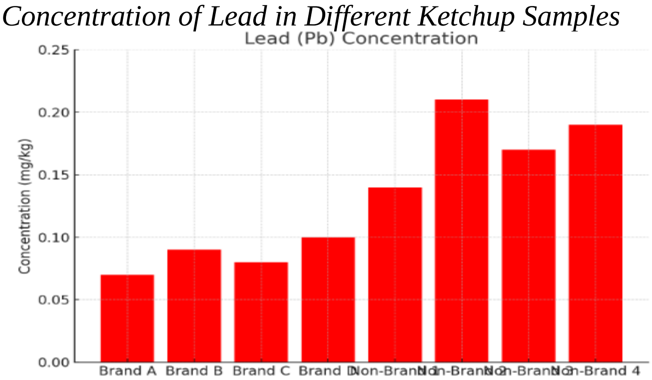
Cadmium levels were seen to be consistently low in all samples, with branded ketchups exhibiting notably lower quantities compared to non-branded ones. This implies the use of improved sourcing and processing criteria in branded products as shown in Fig. 2. The PFA imposes stringent restrictions on the cadmium levels in food goods. The findings suggest that all ketchup samples fell within the acceptable limit, however more vigilant supervision is necessary, particularly for unbranded items.

Figure 2
Concentration of Cadmium in Different Ketchup Samples



Significant variations in lead content were observed across branded and non-branded ketchup samples. Less rigorous quality standards may have contributed to the increased lead levels observed in non-branded items as shown in Fig. 3**.** As to the PFA standards, the lead concentration in all examined ketchup samples was below the highest permissible thresholds. But the elevated amounts detected in unbranded ketchups justify worry and demand more thorough testing.

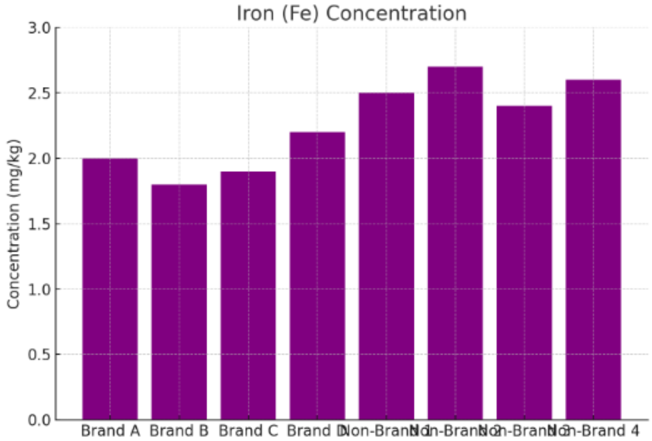
Figure 3
Concentration of Lead in Different Ketchup Samples



The iron concentrations in the ketchup samples fell within the permissible threshold. Branded ketchups often had more uniform iron levels, whereas non-branded varieties displayed greater fluctuation as shown in Fig. 4. The PFA generally does not establish stringent upper thresholds for iron unless it is a potential hazard. Analysis of the ketchup samples indicates that iron levels are well controlled in all cases.

Figure 4

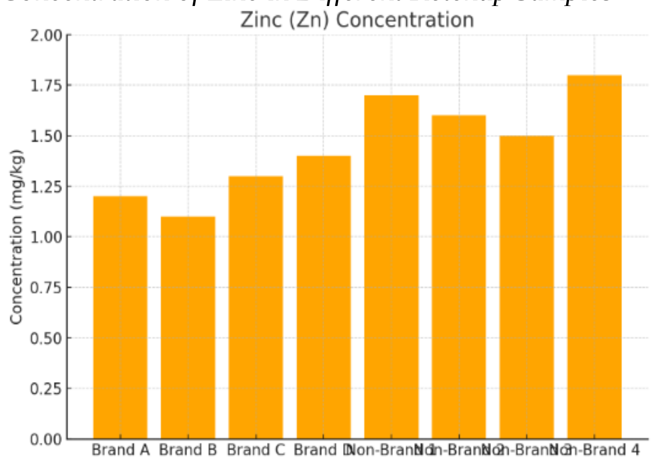
Concentration of Iron in Different Ketchup Samples



The zinc levels in the ketchup samples were within acceptable specifications, with branded goods exhibiting somewhat lower amounts compared to non-branded ones as shown in Fig. **5.** These findings indicate that all products are safe in terms of zinc concentration. Zinc is traditionally regulated in food items, and the concentrations detected in these samples fell below the permissible limit according to PFA standards.

Figure 5

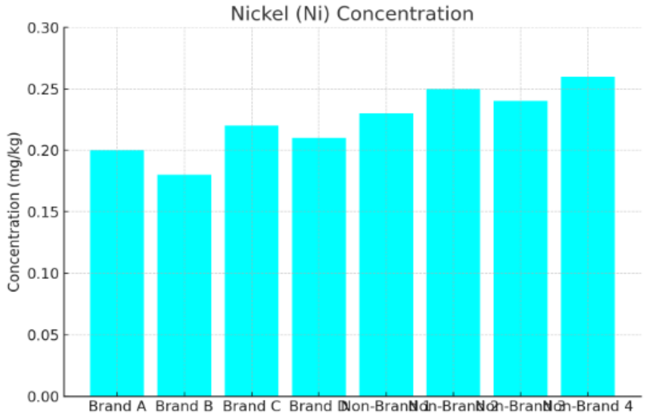
Concentration of Zinc in Different Ketchup Samples



Differences in nickel amounts were observed across the ketchup samples, with non-branded items exhibiting greater levels than branded ones as shown in Fig. 6. This phenomenon may be attributed to variations in production procedures. While all samples examined fell under the nickel content limitations defined by the PFA, the elevated levels in non-branded ketchups may be worrisome for those with nickel allergies.

Figure 6

Concentration of Nickel in Different Ketchup Samples



Concentrations of cobalt were found to differ among the ketchup samples, with non-branded items exhibiting greater amounts. This indicates manufacturing settings with less rigorous control as shown in Fig. 7. The PFA has established criteria for cobalt concentrations in food, and the results showed levels that were within these specified limits. Nevertheless, it is advisable to closely monitor, particularly for non-branded items.

Figure 7

Concentration of Cobalt in Different Ketchup Samples

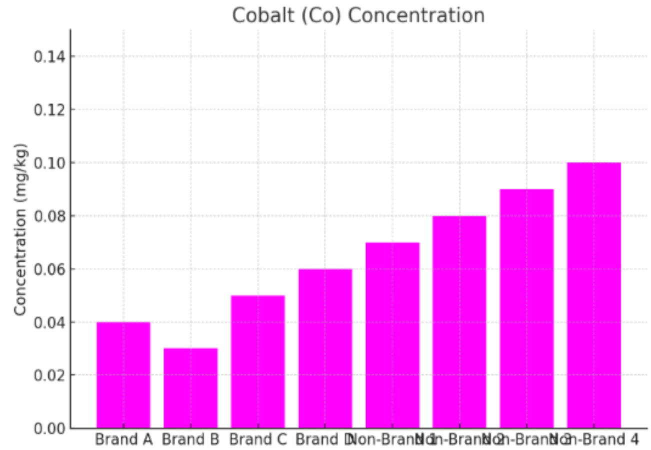


Figure 8

Concentration of All Heavy Metals in Different Ketchup Samples

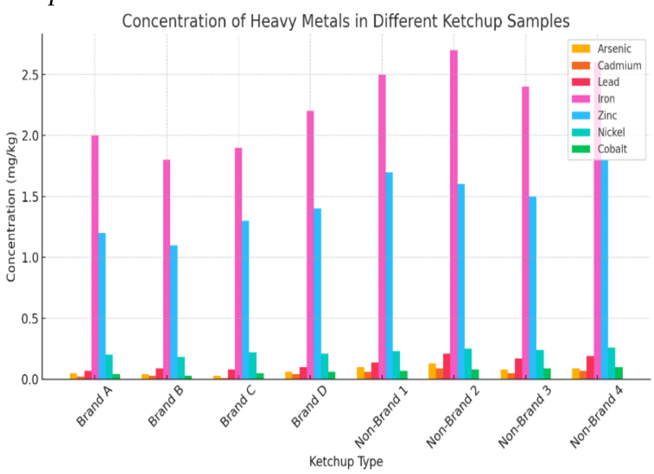


Table 2

Analysis of Variance Table for Heavy Metals

Completely Randomized AOV for As

Source	DF	SS	MS	F	P
Ketchup	4	0.03509	0.00877	55.6	0.0000
Error	19	0.00300	0.00016		
Total	23	0.03809			

Summary Statistics Table

Parameter	Value
Grand Mean	0.0884
Coefficient of Variation (CV)	14.20
Bartlett's Test of Equal Variances (χ^2)	2.56
Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.6340
Cochran's Q	0.3735
Largest Variance / Smallest Variance	8.4510
Component of Variance (Between Groups)	0.00209
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.0467	7.25E-03
Brand B	3	0.0556	7.25E-03
Brand C	3	0.0345	7.25E-03
Brand D	3	0.0678	7.25E-03
Non-brand	12	0.1257	3.62E-03

Completely Randomized AOV for Cd

Source	DF	SS	MS	F	P
Ketchup	4	0.02705	0.00676	14.6	0.0000
Error	19	0.00878	0.00046		
Total	23	0.03583			

Summary Statistics Table

Parameter	Value
Grand Mean	0.0669
Coefficient of Variation (CV)	32.14
Bartlett's Test of Equal Variances (χ^2)	7.44
Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.1143
Cochran's Q	0.6774
Largest Variance / Smallest Variance	15.892
Component of Variance (Between Groups)	0.00153
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.0260	0.0124
Brand B	3	0.0352	0.0124
Brand C	3	0.0252	0.0124
Brand D	3	0.0498	0.0124
Non-brand	12	0.0997	0.0062

Completely Randomized AOV for Co

Source	DF	SS	MS	F	P
Ketchup	4	0.03466	0.00866	15.7	0.0000
Error	19	0.01047	0.00055		
Total	23	0.04512			

Summary Statistics Table

Parameter	Value
Grand Mean	0.0786
Coefficient of Variation (CV)	29.87
Bartlett's Test of Equal Variances (χ^2)	6.23
Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.1827
Cochran's Q	0.6363
Largest Variance / Smallest Variance	10.348
Component of Variance (Between Groups)	0.00197
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.0366	0.0136
Brand B	3	0.0444	0.0136
Brand C	3	0.0272	0.0136
Brand D	3	0.0573	0.0136
Non-brand	12	0.1158	0.0068

Completely Randomized AOV for Fe

Source	DF	SS	MS	F	P
Ketchup	4	4.026E-06	1.006E-06	0.24	0.9092
Error	19	7.807E-05	4.109E-06		
Total	23	8.210E-05			

Summary Statistics Table

Parameter	Value
Grand Mean	1.2663
Coefficient of Variation (CV)	0.16
Bartlett's Test of Equal Variances (χ^2)	3.53
Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.4734
Cochran's Q	0.4191
Largest Variance / Smallest Variance	26.978
Component of Variance (Between Groups)	–7.521E-07
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	1.2672	1.17E-03
Brand B	3	1.2664	1.17E-03
Brand C	3	1.2663	1.17E-03
Brand D	3	1.2656	1.17E-03
Non-brand	12	1.2662	5.85E-04

Completely Randomized AOV for Ni

Source	DF	SS	MS	F	P
Ketchup	4	0.17001	0.04250	19.9	0.0000
Error	19	0.04065	0.00214		
Total	23	0.21067			

Summary Statistics Table

Parameter	Value
Grand Mean	0.2300
Coefficient of Variation (CV)	20.11
Bartlett's Test of Equal Variances (χ^2)	6.69
Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.1530
Cochran's Q	0.6499
Largest Variance / Smallest Variance	10.700
Component of Variance (Between Groups)	0.00978
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.1511	0.0267
Brand B	3	0.1456	0.0267
Brand C	3	0.1163	0.0267
Brand D	3	0.1757	0.0267
Non-brand	12	0.3129	0.0134

Completely Randomized AOV for Pb

Source	DF	SS	MS	F	P
Ketchup	4	0.08209	0.02052	16.7	0.0000
Error	19	0.02328	0.00123		
Total	23	0.10537			

Summary Statistics Table

Parameter	Value
Grand Mean	0.1344
Coefficient of Variation (CV)	26.04
Bartlett's Test of Equal Variances (χ^2)	9.56

Degrees of Freedom (DF)	4
P-value (Bartlett's Test)	0.0485
Cochran's Q	0.5862
Largest Variance / Smallest Variance	26.865
Component of Variance (Between Groups)	0.00468
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.0684	0.0202
Brand B	3	0.0758	0.0202
Brand C	3	0.0573	0.0202
Brand D	3	0.1088	0.0202
Non-brand	12	0.1913	0.0101

Completely Randomized AOV for Zn

Source	DF	SS	MS	F	P
Ketchup	4	0.17167	0.04292	0.65	0.6347
Error	19	1.25696	0.06616		
Total	23	1.42862			

Statistic	Value
Grand Mean	0.5724
CV	44.93
Bartlett's Test of Equal Variances	$\chi^2 = 2.82$, DF = 4, P = 0.5892
Cochran's Q	0.5449
Largest Var / Smallest Var	8.4422
Component of Variance (between groups)	-0.00563
Effective Cell Size	4.1

Ketchup	N	Mean	SE
Brand A	3	0.7123	0.1485
Brand B	3	0.4274	0.1485
Brand C	3	0.6839	0.1485
Brand D	3	0.5904	0.1485
Non-brand	12	0.5414	0.0742

Microbial Analysis Total Plate Count (TPC)

The TPC results show significant variation, especially across branded and non-branded samples. Typically, branded ketchups have lower Total Plate Count (TPC) readings, which suggests a higher level of microbiological quality. Conversely, samples without a brand name have TPC (Total Plate Count) values reaching as high as 5.6 million cfu/g, which greatly beyond the acceptable thresholds for safety. The statistical study demonstrates a substantial disparity in TPC (Total Plate Count) between branded and non-branded ketchups ($p < 0.01$), indicating that non-branded goods may have a greater likelihood of microbiological contamination.

The detection of E. coli in unbranded samples is concerning, since it suggests the existence of fecal contamination and poses a substantial threat to public health. Not a single sample from the branded products showed any signs of E. coli, highlighting the significance of brand reputation and rigorous quality control measures. The chi-square test demonstrates a statistically significant correlation between the type of ketchup and the presence of E. coli ($p < 0.05$), hence highlighting the safety problems linked to non-branded items.

Figure 9

Total Plate Count (tpc) and e. Coli Presence in Ketchup Samples

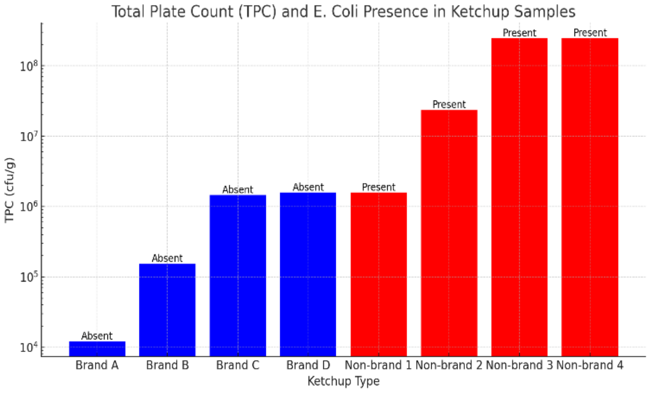


Table 3

Total Plate Count (TPC) in Ketchup Samples

Ketchup Type	TPC (cfu/g)
Brand A	12160±70d
Brand B	153171±59d
Brand C	1456602±101c
Brand D	1578805±103c
Non-brand 1	1578822±109c
Non-brand 2	23567817±67b
Non-brand 3	245685752±6383a
Non-brand 4	248678288±1056a

Table 4

Analysis of Variance Table for TPC

Completely Randomized AOV for TPC

Source	DF	SS	MS	F	P
Ketchup	4	9.997E+16	2.499E+16	2.86	0.0518
Error	19	1.659E+17	8.730E+15		
Total	23	2.658E+17			

Grand Mean: 6.53E+07 CV: 143.00

Chi-Sq	DF	P
Bartlett's Test of Equal Variances	186	4
Cochran's Q	1.0000	
Largest Var / Smallest Var	4.390E+12	

Component of Variance for Between Groups: 3.943E+15
Effective Cell Size: 4.1

Ketchup Summary (N, Mean, SE)

Ketchup	N	Mean	SE
Brand A	3	12160	5.39E+07
Brand B	3	153171	5.39E+07
Brand C	3	1.46E+06	5.39E+07
Brand D	3	1.58E+06	5.39E+07
Non-brand	12	1.29E+08	2.70E+07

LSD All-Pairwise Comparisons Test of TPC by Ketchup

Ketchup	Mean	Homogeneous Groups
Non-brand	1.29E+08	A
Brand D	1.58E+06	B
Brand C	1.46E+06	B
Brand B	153171	B
Brand A	12160	B

Alpha: 0.05 Critical T Value: 2.093

There are 2 groups (A and B) in which the means are not significantly different from one another.

DISCUSSION

The increased concentrations of Pb might arise from several origins, including food, water sources, or the immediate surroundings. In contrast, amounts of Mercury and Arsenic in the individual samples were determined to be beneath all recognised standards. The evaluation of food safety and its effects on consumers necessitates meticulous examination of the quantity of heavy metals ingested via dietary intake. Insufficient literature existed about the mean consumption of the particular meals under investigation in Pakistan. The direct toxicity of arsenic compounds to the body's systems arises from their capacity to accumulate in several organs, including the liver, spleen, kidney, lungs, and gastrointestinal tract, following absorption into the body (Ismail, 2009). The findings show that the amount of Arsenic (0.0003 mg/kg) in the Tomato ketchup was within the permissible thresholds at all designated sampling locations. Repeated consumption of Arsenic at a dosage of 0.03 mg/kg via food might result in transient symptoms including nausea, vomiting, diarrhoea, debility, reduced appetite, cough, and headache. Nevertheless, it can also lead to grave health repercussions. Based on the increased incidence of cancer in those exposed to arsenic in their employment, environment, or diet, the International Agency for Research into Cancer (IARC) has classified it as a human carcinogen. Despite its historical use as a rodenticide, arsenic is by much more toxic than other metallic substances. Prolonged exposure to minimal amounts of arsenic is linked with skin, circulatory system, and brain system adverse effects (Blanco et al., 2008). The average nutritional consumption of Arsenic was 0.04 micrograms per kilogramme of body weight per week. The Joint FAO/WHO Expert Committee on Food Additives (JEFCA) recognised in 2009 that the permissible weekly ingestion of Arsenic without having detrimental effects is 15µg per kilogramme of body weight. Based on the higher concentration levels, it may be presumed that cadmium carriages a noteworthy health threat to consumers. The study conducted by (Sharma et al., 2009) revealed a sophisticated cadmium content of 1.96 mg/kg in tomatoes compared to the value reported in this aforementioned work. Sharma et al. (2009) suggests that the detection of elevated levels of heavy metals, notably cadmium, in plants like tomatoes might be attributed to the use of contaminated water for irrigation purposes.

Furthermore, (Jimoh & Mohammed, 2012) described higher amounts of cadmium that surpass the set upper limits. An investigation in Egypt by (Radwan & Salama, 2006) on Egyptian fruits and vegetables revealed that the cadmium (Cd) content in tomatoes was below the established regulatory limit, with an average measurement of 0.03 ± 0.01 mg/kg. The mean dietary intake of Cadmium is described as 0.2 µg/kg bw/person/week. The objective is to evaluate the amounts of intake in respect to the PTWI guiding principle of 2.5 µg/kg body weight per week as defined by JEFCA.

The major route of lead exposure to the general people is through the ingestion of food that contains this hazardous element. Deviation from the maximum permissible threshold (0.2 mg/kg) of lead concentration in food can result in detrimental consequences, both in the immediate and extended periods. Adverse effects of transient exposure to high levels of lead include neurological dysfunction, paralysis (referred to as lead palsy), anaemia, and gastrointestinal problems. Hence, the amounts of the lead in tomatoes surpass the permissible boundary in maximum of the selected locations for sample collection. Nevertheless, according to the results of this study, those who consume these vegetables are prone to encountering toxicity. Extended duration of exposure can lead to damage to the kidneys, reproductive and immune systems, and can affect the brain system. Even little levels of lead exposure greatly affect the cognitive growth of young infants. Like mercury, lead has the ability to traverse the eutherian blockade and accumulate within the developing foetus. Neonates and young offspring are more susceptible than grown person to the detrimental properties of lead, and they also have a higher capacity to absorb lead. Research by the Food Safety Authority of Ireland in 2009 indicates that even short and insignificant exposures of newborn infants to lead might affect their neurobehavioral development. The study done by Radwan and Salama (2006) examined Egyptian fruits and vegetables and revealed that tomatoes had a content of heavy metals, notably Pb, at 0.26 ± 0.09 mg/kg. In 2009, Aryan Dermisbas conducted a study with the objective of quantifying the concentrations of oil, micronutrients, and heavy metals in tomatoes. The analysis revealed a lead (Pb) concentration of 0.43 ± 0.08 mg/kg. An independent study in the work area of Huludao City, China verified the existence of lead (Pb) at a concentration of 6.62 milligrams per kilogramme.

Studies conducted by Radwan & Salama (2006) and (Khan et al., 2022) have similarly concluded that there is no adverse effect linked to the consumption of commonly grown food in areas where wastewater is utilised for irrigation. The present investigation primarily attributes the induction of human health dangers to Cadmium (Cd). The study conducted by (Zheng et al., 2007) revealed that the daily consumption of Cd in the area of Huludao Zinc Plant above the permissible daily consumption limit. This presents a hazard to the inhabitants residing in that area. Based on their investigation of fruits and vegetables in Ghanaian markets, (Bempah et al., 2011) determined that the presence of heavy metals in these food items does not now present an imminent threat to human health.

Nevertheless, prolonged ingestion of fruits and vegetables containing even modest amounts of pollution might result in the buildup of these chemical elements in the body, potentially leading to lethal outcomes for the human population. The risk assessment mainly concentrated on the fluctuation in direct exposure resulting from the temporal activity patterns of the human population. Stochastic Monte Carlo Simulation was used to examine sources of variability.
The microbiological safety of all items was evaluated by microbial analysis. The study sought to identify the precise categories of viable bacteria that may potentially contaminate the samples. The standard plate count (SPC), sometimes referred to as the viable count, is a commonly employed technique for evaluating the microbiological integrity of food. The mean total bacterial levels for Brands A, B, C, and D were 12160 ± 70, 153171 ± 59, 1456602 ± 101, and 1578805 ± 103 log10 colony-forming units per gramme, respectively (Table 3). Maximum of the samples examined in this research did not encounter the acceptable microbiological threshold, namely over 10^6 cfu/g, which is the minimum boundary for bacterial count in commercially prepared meals. Mensah et al. (2002) found that salads prepared and sold on the streets of Accra contained a bacterial count of 6.3 ± 0.78. Furthermore, (Christison et al., 2008) also recorded a notable presence of microorganisms in baguettes and salads that were filled. The study conducted by (Bukar et al., 2010) in South Africa revealed that the mean bacterial counts in ready-to-eat meals above the permissible threshold. The occurrence of Escherichia coli in ready-made meals is objectionable as it specifies inadequate sanitary situations that have directed to contamination or insufficient heat treatment. The analysis of 12 food samples revealed that all non-branded items were found to be contaminated with Escherichia coli. More precisely, there were 3 occurrences associated with Brand D, 2 instances associated with Brands B and C, and 1 incidence associated with Brand A. The occurrence of staphylococci in ready-to-eat tomato ketchup is largely attributed to human contact, wherein contamination occurs. Their inclusion in the trial suggests time temperature ratio manipulation is probable to have happened due to indecorous treatment. Contamination upto 10^4 cfu or above are well-thought-out potentially dangerous, as consuming foods contaminated at this level has the capacity to induce foodborne illness. The implementation of appropriate food handling procedures can effectively mitigate contamination, while the maintenance of suitable temperature controls can impede the growth of the organism. The insufficiency of B. cereus in cooked meals is usually attributed to inadequate temperature management. If the concentration of B. cereus exceeds 103 colony-forming units per gramme, it is imperative to carry out an investigation of the food handling procedures used by the food outlet. Concentrations of 104 colony forming units (cfu) per gramme or above are considered to be potentially hazardous. The consumption of foods contaminated at this level has the potential to result in foodborne illness. Furthermore, other Bacillus kind, including B. licheniformis and B. subtilis, have been related with foodborne infection and may require testing. A comprehensive analysis of the geographical distribution, frequency, and levels of Bacillus sp. in particular consumer products is provided in this paper. Overall, 98% of the examined samples were found to have included counts. The Food Standards Australia New Zealand recommendations from 2001 state that the presence of this pathogen at a level of ≥0.6 log cfu/g marks the Bacillus sp. in ready-made foods as undesirable. A minimum of 105 colony-forming units per gramme (cfu/g) of pathogenic Bacillus spp. (B. subtilis, B. pumilis, B. licheniformis) rendered the ketchup samples unacceptable. The study's results indicate the presence of contamination and inadequate hygienic controls throughout the production and processing of ready-to-eat tomato ketchup. Pre-prepared tomato ketchup that may be consumed without additional cooking, but is contaminated with pathogenic bacteria such as Escherichia coli, or has intolerable amounts of S. aureus or B. spp., is ranked as hazardous. These substances are considered harmful to human health and/or inappropriate for consumption by humans. The results emphasise the requirement of maintaining appropriate cleanliness while one handles these specific types of ready-to-eat products.

CONCLUSION

As do not have any harmful impact on people. It is necessary to monitor these heavy metals to ensure that they remain at safe levels. The levels of the other metals, lead (Pb) and cadmium (Cd), are either extremely close to or beyond the permissible limits. The current quantities may not have immediate detrimental or adverse effects. However, continued use of these metals, from any source, might potentially have hazardous health consequences. Consuming ready-to-eat tomato ketchup does not provide an unacceptable danger to the public, despite the fact that the primary way people are exposed to it is by consuming veggies. The rapid urbanization and widespread use of metal technology for size reduction, together with the utilization of wastewater in vegetable farming, have led to elevated levels of heavy metals and microbiological pollutants in our food. All the samples obtained had a bacterial count that above the reference levels, which presents a substantial hazard to users. The findings of this study, however, show that the danger of consuming locally branded ready-to-eat tomatoes in Multan is minimal in terms of the examined heavy metals. But the consumption of non-branded tomato ketchup is harmful and most chances of heavy metals and microbial attacks.

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