Current Research Bulletin
Volume 03 Issue 08 August 2026, Page no: 293-298
Available Url: http://crbjour.org/
Article DOI: 10.55677/CRB/I8-04-CRB2026
e-ISSN: 3050-5933
p-ISSN: 3050-5925
Open Access

Evaluation of Microbiological, Proximate Composition and Heavy Metals Levels of Some Leafy vegetables (Carrot, Onion and Tomato) Grown in Gashua, Bade Local Government Area, Yobe State, Nigeria

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Published Online:
31 August 2026

Article DOI:
10.55677/CRB/I8-04-CRB2026

ABSTRACT:
Farm produce contamination by heavy metals has become a pressing issue, and the accumulation of heavy metals in agricultural soils is of increasing concern due to food safety risks, potential health impacts, and detrimental effects on soil ecosystems. This work aims to determine the Microbiological analysis, Proximate Composition, and Heavy Metal concentrations of selected farm produce (Carrot, onion, and Tomato) Grown in Gashua, Bade Local Government Area of Yobe State, Nigeria. Samples of Carrots, Onions and Tomatoes were collected directly from the farm, and Soil samples were collected at the same plots where the crops were grown. The results of the proximate parameters from the samples collected indicated, Moisture, Carbohydrate, Ash, Lipid, Crude fibre and Protein content ranging from 75.7 ±2.03 to 90.0 ±2.02; 5.21 ±0.82 to 16.10 ±0.76; 0.18±0.01 to 0.62 ±0.08; 0.4 ±1.02 to 2.45 ±0.10; 0.20 ±0.01 to 1.3 ±0.10 and 0.87 ±0.02 to 4.50 ±0.20% respectively, for the Carrot, Tomato and Onion. Some of the Bacteria and fungi isolated from the soil and samples are *Bacillus sp., Streptococcus sp., and E.coli* and *Aspergillus sp*. Heavy metal concentrations were determined using atomic absorption spectrometry. The results showed that the levels of Polycyclic Aromatic Hydrocarbon, Nickel, Chromium, Copper, Cadmium, Lead and Manganese ranged from 0.003 to 1.00; 1.932±0.00 to 5.164±0.01; 0.840±0.01 to 11.224±0.01; 2.404±0.01 to 13.18±0.01; 0.252±0.05 to 0.712±0.01; 3.364±0.01 to 28.16 ±0.00 and 1.962±0.01 to 6.608±0.05 mg/kg, respectively, for the Carrot, Tomato and Onion. The values obtained are very high compared with those reported in the literature and are not within the tolerable limits set by some regulatory authorities. Our study concluded that there is a need to educate the farmers in this geographical area of study on the danger of contaminated soil used for growing crops.

KEY WORDS: Farm produce, heavy metals, atomic absorption spectrometry, proximate composition

Cite the Article: *Musbau, S., Asiru, R.A. (2026). Evaluation of Microbiological, Proximate Composition and Heavy Metals Levels of Some Leafy vegetables (Carrot, Onion and Tomato) Grown in Gashua, Bade Local Government Area, Yobe State, Nigeria. Current Research Bulletin, 3(8), 293-298.* https://doi.org/10.55677/CRB/I8-04-CRB2026

INTRODUCTION

The most common environmental pollutants worldwide are heavy metals (Dallatu *et al.*, 2013). The presence of heavy metals at trace levels and essential elements at elevated concentrations causes toxic effects if exposed to the human population (Eze *et al.*, 2018). The accumulation of heavy metals in agricultural soils is of increasing concern due to food safety issues and potential health risks, as well as its detrimental effects on soil ecosystems (Van Ginneken *et al.*, 2007). Food contamination by heavy metals has become a burning issue in recent years because of their potential accumulation in biosystems through contaminated water, soil and air (Dallatu *et al.*, 2013). Heavy metals can accumulate in the soil to toxic levels as a result of long-term application of untreated wastewater and fertilizers. Soil irrigated with wastewater accumulates heavy metals in surface soils, and when the capacity to retain heavy metals is reduced due to repeated application of wastewater, heavy metals leach into groundwater or soil solution available for plant uptake (Ishaq, *et al.*, 2010). Findings indicate that application of heavy doses of fertilizer, pollutes groundwater with nitrates and heavy metals through leaching, and this affects the quality of water (Samaila *et al.*, 2011). Metal poisoning results from toxic heavy metals, leading to adverse effects on human and ecosystem health (Oladele *et al.*, 2015). Although acute poisoning from heavy metal poisoning is rare through ingestion or dermal contact, chronic exposure to even small doses can be disastrous (Singh, 2015). The chronic problems associated with long-term heavy metals exposure include: serious haematological and brain damage, anaemia and kidney malfunctioning (Ishaq, *et al.*, 2010). Heavy metals such as Lead (Pb), Cadmium (Cd), Chromium (Cr), and Arsenic (As) are lethal even in very small doses. Lead has a negative influence on somatic development and decreases visual acuity and auditory thresholds (Mohammed *et al.*, 2015). Acute exposure to lead causes brain damage, neurological symptoms, and could lead to death (Mohammed *et al.*, 2015).

The intersection of agricultural sustainability and public health has reached a critical tipping point in Gashua, the administrative headquarters of the Bade Local Government Area (LGA) in Yobe State, Nigeria. This region, a vital horticultural hub within the Hadejia-Nguru wetlands and the Komadugu-Yobe River basin, is currently grappling with a multifaceted environmental crisis. Recent scientific investigations have unveiled a systemic failure in food safety, where heavy metal bioaccumulation and microbiological contamination have turned essential dietary staples into vectors for chronic disease.

Reports show that Gashua has become a recognized "hotspot" for Chronic Kidney Disease (CKD) and Chronic Kidney Disease of unknown aetiology (CKDu). Hospital records indicate that at least 85% of kidney ailment cases at the Yobe State Tertiary Hospital originate from this community. Medical reports suggest that no fewer than 150 residents' die of kidney failure annually, a phenomenon increasingly linked to the consumption of foods preserved with toxic chemicals and grown on contaminated soil environments.

This work is aimed at determining the proximate composition, bacteriological, and Heavy Metals Levels of some selected farm produce (Carrot, Onion and Tomato) grown in Gashua, Bade Local Government Area, Yobe State.

MATERIALS AND METHODS

Sample Collection and Preparation

Study Area

- **Location:** Jawa Village, northeast of Bade Local Government Area, Yobe State, Nigeria.
- **Period:** March 2025, during the dry season.
- **Farming practice:** Crops cultivated using irrigation water.

Sample Collection

- **Farm produce:** Carrot (*Daucus carota*), Onion (*Allium cepa*), and Tomato (*Solanum lycopersicum*).
- **Stage of collection:** Sapling stage, directly from the farm.
- **Soil samples:** Collected from the same plots where food items were grown.

Sample Preparation

- **Drying methods:** Oven drying at **110 °C for 12 hours**.
- **Milling:** Dried samples were ground using an **electric blender** to obtain fine powder.
- **Storage:** Milled samples kept in airtight containers before digestion and analysis.

Digestion (for further analysis)

- Prepared samples undergo digestion using standard laboratory protocols (e.g., proximate composition tests, acid digestion for heavy metal analysis, and microbiological assays, based on study objectives).

Proximate Composition Analysis

The proximate composition parameters — moisture, crude protein, crude fat, ash, and crude fiber — were determined following the standardized procedures of the Association of Official Analytical Chemists (AOAC, 2000). Moisture content was quantified by oven-drying 2 g of each sample to constant weight at 105 °C. Ash content was assessed by incinerating 1 g of the sample in a muffle furnace maintained at 550 °C for 5 hours. Crude protein was estimated by determining total organic nitrogen and applying the conversion factor of 6.25. Crude fat was extracted from 5 g of the sample using a Soxhlet apparatus with petroleum ether (boiling range 60–80 °C) as the solvent. Crude fiber was determined gravimetrically following acid and alkali digestion, while carbohydrate content was calculated by difference, subtracting the sum of moisture, protein, fat, ash, and fiber from 100.

% Total Carbohydrate = [100 - % (Protein +Fat + Moisture + Ash + Fiber)]

Microbial Evaluation of Carrot, Onion, and Tomato Samples

To check the microbes present in carrot, onion, and tomato samples, two types of growth media were used: Nutrient Agar (NA) for bacteria and Potato Dextrose Agar (PDA) for fungi. Both media were prepared according to manufactured instructions and sterilized in an autoclave at 121 °C for 15 minutes. The study was carried out in the Microbiology Laboratory at Yobe State University, Damaturu, Nigeria. Microorganisms were grown using the pour plate method, and the number of microbes was estimated by serial dilution. Triplicate plates were prepared for accuracy. For bacterial growth, NA plates were incubated at 37 °C for 24–48 hours. For fungal growth, PDA plates were incubated at room temperature (28 ± 2 °C) for 48–72 hours. Colonies that developed were counted and recorded as colony-forming units per gram (cfu/g) of each sample. Selected colonies were then isolated and identified using biochemical tests, following the method of Bashir *e t al*. (2016).

Heavy Metal Evaluation: Heavy metal concentrations in soil and food products were determined following standardized digestion and instrumental analysis protocols (Inobeme *et al.*, 2023). Approximately 1g of each sample (Soil, carrot, tomato, and onion) was weighed into a 250 mL conical flask with 2–3 g of glass beads. 5 mL of aqua regia (HNO_3: HCl, 1:3) was added, and the mixture was thoroughly homogenized. The flask was heated on a hot plate until the volume was reduced to about 15 mL, then cooled. An additional 5 mL of concentrated HNO_3 was introduced, and the flask was covered with a watch glass. Concentrated HNO_3 was added dropwise until the solution became light-colored and clear. The contents were rinsed with distilled water, filtered, and transferred into Atomic Absorption Spectrometry (AAS) vials for analysis. Metals quantified included copper, zinc, lead, cadmium, potassium, nickel, sodium, silver, calcium, chromium, and iron. The concentrations obtained were compared against maximum permissible limits established by WHO/FAO and YOSEPA guidelines (FAO/WHO, 2011).

RESULTS

Results obtained from the proximate analysis of the leafy vegetables (carrot, tomato, onion) as presented in Table 1 shows that the moisture content ranging from 75.7 to 90.0%, Carbohydrate ranging from 5.21 to 16.10%, Ash content ranging from 0.18 to 0.62%, Lipid content ranging from 0.4 to 4.31%, Crude fibre ranging from 0.20 to 1.3% and Protein content ranging from 0.87 to 4.50%.

Table 1. Mean Levels of Proximate Content in Selected Leafy vegetables

S/N	Sample Identity	Moisture (%)	Carbohydrate (%)	Ash (%)	Lipid (%)	Crude fibre (%)	Protein (%)
1	Carrot	75.7 ±2.03	16.10 ±0.76	0.62 ±0.08	2.45 ±0.10	0.53 ±0.10	4.50 ±0.20
2	Tomato	90.0 ±2.02	5.21 ±0.82	0.18±0.01	0.4 ±1.02	1.3 ±0.10	2.6 ±0.01
3	Onion	79.8 ±2.04	14.2 ±0.81	0.32 ±0.06	4.31 ±1.02	0.20 ±0.01	0.87 ±0.02

Tables 2 show the concentrations of heavy metals investigated in leafy vegetables commonly grown in Gashua, Bade Local Government Area of Yobe State, Nigeria. The values are given as mean ± SD, and the results are the means of three replicates. The heavy metal levels determined were based on the plant's dry weight. Results obtained from the Levels of Heavy Metals detected in Soil and Selected Leafy vegetables (carrot, tomato, onion). Nickel level ranging from 1.932 to 4.028mg/kg, Chromium level ranging from 0.84 to 11.224mg/kg, Copper level ranging from 2.404 to 13.18mg/kg, Cadmium level ranging from 0.252 to 0.712mg/kg, Lead level ranging from 3.364 to 28.16mg/kg and Manganese level ranging from 1.962 to 6.608mg/kg.

Table 2. Mean Levels of Heavy Metals detected in Soil and Selected Leafy vegetables

S/N	Samples Identification	PAH (mg/kg)	Ni (mg/kg)	Cr (mg/kg)	Cu (mg/kg)	Cd (mg/kg)	Pb (mg/kg)	Mn (mg/kg)
1	Soil	<1.00	1.932±0.0	0.840±0.01	3.428±0.00	0.252±0.05	28.16 ±0.00	6.608±0.05
2	Carrot	0.009	3.872±0.02	7.996±0.05	10.82±0.01	0.372±0.01	4.312±0.01	3.876±0.01
3	Tomato	0.007	5.164±0.01	11.224±0.01	2.404±0.01	0.712±0.01	5.920±0.01	5.568±0.10
4	Onion	0.003	4.028±0.01	2.804±0.01	13.18±0.01	0.296±0.01	3.364±0.01	1.962±0.01
Safe limit (WHO &FEPA, 2023)			0.20	1.0	0.20	0.20	300	0.20

KEY: - PHA = Polycyclic Aromatic Hydrocarbon, Ni = Nickel, Cr = Chromium, Cd = Cadmium, Pb = Lead, Mn= Manganese

Results obtained from bacterial colony counts were presented in **Table 3**. Bacteria colony count from the soil ranged from 0.00109 to 23.1cfu/g, Carrot ranging from 0.00128 to 13.1cfu/g, Tomato ranging from 0.00119 to 16.9cfu/g and Onion ranging from 0.00116 to 14.3cfu/g.

Table 3. Mean Numbers of Colonies Counted in Soil and Selected Food Stuffs

S/N	Samples	Dilution factors	No of Colonies Counted	Standard form CFU/g
1	**Soil**	10^{-1}	231	23.1
		10^{-2}	196	1.96
		10^{-3}	154	0.154
		10^{-4}	121	0.0121
		10^{-5}	109	0.00109
1	**Carrot**	10^{-1}	131	13.1
		10^{-2}	126	1.26
		10^{-3}	114	0.14
		10^{-4}	141	0.0141
		10^{-5}	129	0.00129
2	**Tomato**	10^{-1}	169	16.9
		10^{-2}	171	1.71
		10^{-3}	133	0.133
		10^{-4}	124	0.0124
		10^{-5}	119	0.00119
3	**Onion**	10^{-1}	143	14.3
		10^{-2}	132	1.32
		10^{-3}	121	0.121
		10^{-4}	113	0.0113
		10^{-5}	116	0.00116

Bacteria and Fungi isolated from soil and Leafy vegetables were presented in Table 4

Table 4: Bacteria and Fungi isolated from Soil and Farm produce

S/N	Sample	Isolated Organisms: Bacteria	Fungi
1.	Soil	*Streptococcus sp., E. coli, Shigella sp.*	*Aspergillus sp*
2.	Carrot	*Bacillus sp., Streptococcus sp.*	_
3.	Tomato	*Bacillus sp., Streptococcus sp., E.coli*	*Aspergillus sp*
4.	Onion	*Bacillus sp., Streptococcus sp.*	_

DISCUSSION

Proximate Analysis

Moisture content of the fresh samples, which range from 75.7 ±2.03% to 90.0±2.02% is consistence to the results reported by Udo *et al.*(2013), who reported that leafy vegetables have a moisture content ranging from 72 to 93%. The high moisture content of vegetables indicates freshness and perishability, as well as indicating that they may play a key role in aiding the digestion of food (Adepoju and Oyewole, 2008) and also facilitate the activity of water-soluble enzymes and co-enzymes for metabolic activities.

Total carbohydrate was most abundant in Carrot (16.10 ±0.76 %) compared to Tomato which had the least total carbohydrate content (5.21 ±0.82 %). Comparing these values with other vegetables reported by Adnan *et al*. (2010) and Nkafamiya *et al.* (2010), the carbohydrate content of the vegetables in this investigation is relatively low, with values ranging from 5.21 ±0.82 % to 16.10 ±0.76 %. The recommended dietary allowance (RDA) for carbohydrate for normal healthy male and female adults is 130g/100g. The observation from this study implied that the vegetables may be poor sources of dietary carbohydrates and so may not be recommended to vegetarians as the sole source of dietary carbohydrate and energy. The results of this study showed that the leafy vegetables analysed had low protein content, ranging from 0.87 ± 0.02% to 4.50 ± 0.20%. This indicates that these vegetables may be poor sources of protein for meeting daily dietary needs, particularly in regions where animal protein is scarce.

The result obtained in this study for crude fat is relatively low (0.4 ± 1.02% to 4.31 ± 1.02%) compared to the values obtained by Uhegbu et al. (2011). *Lavandula angustifolia* (6.52%) and *Valeriana officinalis* (14.35%). These vegetables may therefore be recommended as part of weight-reducing diets due to their low fat content.

The Crude fibre from this study, ranging from 0.20 ±0.01% to 1.3 ± 0.10% results, showed low dietary fibre content in the vegetables analyzed, however, these values are relatively high when compared with data obtained by Blessing *et al.* (2011) for soybean (0.2%). It's on record that dietary fibre is essential in aiding digestion and decreasing waste transit time in the gastrointestinal tract, increasing stool bulk and preventing constipation (Blessing *et al.*, 2011). Therefore, these vegetables may be very useful in the control of body weight, blood cholesterol and protection against colon cancer.

Heavy metal concentrations

Table 2 shows the concentrations of heavy metals investigated in leafy vegetables commonly grown in Gashua. The detected quantities of Ni, Cr, Cu, Cd, Pb and Mn in leafy vegetables were compared to the safety limit established by the FAO/WHO and other reference standards. The heavy metal contamination in all selected leafy vegetables indicated that the concentration levels of Ni, Cr, Cu, Cd, Pb and Mn were not within the permissible limit recommended by WHO (Sharma *et al.,* 2007; Rahman *et al.,* 2013). Heavy metals affect the nutritional values of agricultural produce and also have a toxic effect on human beings. National and international regulations on food quality set the maximum permissible levels of toxic metals in human food; hence, an increasingly important aspect of food quality should be to control the concentrations of heavy metals in food (Radwan and Salama, 2006; Sobukola *et al.,* 2008). Heavy metal is an important cumulative body poison that enters the body system through air, water and food and cannot be removed by washing vegetables (Divrikli *et al.,* 2003). The high concentrations of heavy metals in some of these plants may be attributed to pollutants in irrigation water, farm soil or due to pollution from the highway traffic (Qui *et al.*, 2000). Nickel plays some role in body functions, including enzyme functions. It occurs naturally more in plants than in animal flesh; it activates some enzyme systems in trace amounts, but its toxicity at higher levels is more detrimental (Divrikli *et al.,* 2006). Copper is an essential trace element which functions as a biocatalyst, required for body pigmentation in addition to iron, maintain a healthy central nervous system, prevents anaemia and is interrelated with the function of Zn and Fe in the body (Akinyele and Osibanjo, 1982), however high concentration beyond permissible limit can lead to severe illness, such as kidney and liver damage as reported in this findings.

Bacteria and Fungi Isolated

The presence of an *E. coli*, *Bacillus sp., Streptococcus sp.* and *Shigella* sp in the leafy vegetables is an indicator of poor hygiene and faecal pollution of a food. In the present study, the mean total viable count indicates that the food products are potentially problematic considering the recommended food safety limits. *Aspergillus* sp. isolated can produce mycotoxins. Mycotoxins are threats to global food security as well as to human health.

CONCLUSION

The results of this study showed that these three leafy vegetables contain appreciable amounts of nutrients with the potential to improve the nutritional status and promote the health of people, especially in developing countries like Nigeria, where nutrient deficiencies are prevalent. Similarly, heavy metal concentrations of selected leafy vegetables indicated that the Ni, Cr, Cu, Cd, Pb and Mn content of vegetables exceeded the permissible limit suggested by various health organizations.

Generally, the levels of heavy metals were observed to be higher than those of previous published works and those given by various authorities; therefore, there is a need to closely follow a comprehensive waste management system to prevent industrial effluent from polluting arable land. The values of heavy metals presented in this work from leafy vegetables obtained from Gasua, Nigeria, can be valuable in the food composition tables for Nigerians and Africans as a whole. The values of the microorganisms indicated potential food safety problems, which could be reduced by increased hygiene.

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