

Evaluation of the Concentrations of Nitrate, Nitrite and Heavy Metals in Spinach (*Spinacia oleracea*) Irrigated along the Amba Stream Lafia, Nasarawa State

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Abstract

The concentrations of nitrate, nitrite and heavy metals were evaluated in the soil and edible portion of spinach (*Spinacia oleracea*), irrigated along the Amba stream, Lafia, Nasarawa State. Nitrate, nitrite and heavy metals in the soil and spinach samples were determined using spectrophotometric method. The mean nitrate concentrations (\pm SD) ranged from 3042.91 ± 1.62 to 4977.26 ± 9.57 mgkg^{-1} for the spinach while 5899.20 ± 58.98 to 7026.73 ± 12.08 mgkg^{-1} for soil. The mean nitrite concentrations (NO_2^-) (\pm SD) ranged from 0.00029 ± 0.00002 to 0.0102 ± 0.00001 mgkg^{-1} for the spinach while 0.00332 ± 0.00002 to 0.00903 ± 0.00002 mgkg^{-1} for soil. The nitrate and nitrite concentrations of the spinach are above the maximum level of 10 mgkg^{-1} and 2 mgkg^{-1} respectively as specified by WHO. NO_3^- generally recorded higher level of concentration than NO_2^- . The mean concentrations of heavy metals ranged from Cd (1.29 ± 0.01 mg/kg) to Fe (281.60 ± 1.65 mgkg^{-1}) for spinach while Cd (0.93 ± 0.32 mgkg^{-1}) to Fe (1084.1 ± 1.73 mgkg^{-1}) for soil. Cd, Pd and Fe level concentrations in the spinach far exceeded FAO/WHO recommended maximum levels of 0.2, 0.3 and 425.5 mgkg^{-1} , respectively. Transfer factors for the anions between the soil and spinach identify the efficiency of a spinach species to accumulate a given anion. There was a positive correlation at $p = 0.05$ between the anions in the spinach. The transfer factors calculated were lower for all metals except cadmium (1.39).

Keywords: Nitrate, nitrite, heavy metal, spinach, irrigated water

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Introduction

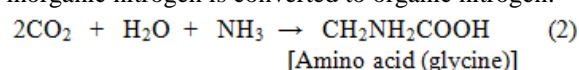
Vegetable and fruit consumption as food offers the most rapid and lowest cost means of providing adequate supplies of vitamins, minerals, and fibres to the people who live in the tropics. In these areas, vegetables are the leafy outgrowth of plants used as food and include those plants and parts of plants used in making soup or served as integral parts of the main sources of our meals [1]. Vegetables are the major sources of the daily intake of nitrate by human beings, supplying about 72 to 94% of the total intake [2]. Nitrate and nitrite are essential nutrients for plant protein synthesis and play a critical role in the nitrogen cycle [3]. Nitrate is a naturally occurring form of nitrogen. Nitrate is formed from fertilizers, decaying plants, manure, and other organic residues. It is found in the air, soil, water, and food (particularly in vegetables) and is produced naturally within the human body. It is also used as a food additive, mainly as a preservative and antimicrobial agent [4]. Due to the increased use of synthetic nitrogen fertilizers and livestock manure in intensive agriculture, vegetables, and drinking water may contain higher concentrations of nitrate now than in the past [5]. Nitrogen is the main limiting factor for most field crops, and nitrate is the major form of nitrogen absorbed by crop plants. Farmers often use nitrogen

fertilizers to increase crop yields. Consequently, many vegetables and forage crops accumulate high nitrate. Leafy vegetables such as spinach, lettuce, and celery contain nitrate at significant levels [6]. The nitrate content of vegetables can be affected by the processing of the food, the use of fertilizers, and growing conditions, especially the soil temperature and (day) light intensity [7-8]. Vegetables such as beetroot, lettuce, radish, and spinach often contain nitrate concentration above 2500 mgkg^{-1} , especially when they are cultivated in greenhouses.

Nitrite content in vegetables is usually very low compared to nitrate [9-10]. Nitrogen fixation makes nitrogen available for use by organisms such as vegetables. The nitrogen in the atmosphere (N_2) is fixed by conversion to inorganic nitrogen [11].

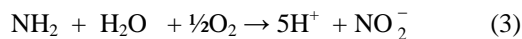


Animals then get their nitrogen from plants from amino acids, through a process called assimilation where the inorganic nitrogen is converted to organic nitrogen:

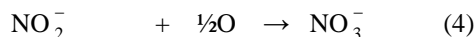




The above process also operates when microorganisms decompose the remains of dead plants and animals in both terrestrial, e.g. soil and marine environments. Ammonia can be oxidized to *nitrate and nitrite* [11]: *Nitrosomonas* bacteria oxidize the fixed nitrogen to nitrite through the denitrification process:

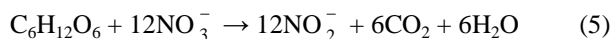


The *Nitrobacter* bacteria oxidize nitrite to nitrate:

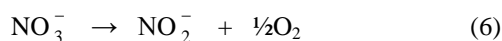


The small sub-cycle between assimilation, death, and mineralization represents the recycling of nitrogen within the soil system in most animals and plant life including all agricultural participants. It is critically dependent on fixation as the major independent natural source of available inorganic nitrogen. However, industrial fixation represents man's attempt to supplement the available inorganic nitrogen in the soil system by synthesizing ammonia from nitrogen, hydrogen, and methane in the Haber process and thus forming salts for fertilizers [11]. The nitrate content in crops is one of the important indicators of farm production quality. Nitrate content in food is strictly regulated because of its toxicity, especially to young children. The actual toxin is not the nitrate ion itself but rather the nitrite ion, which is formed when nitrate is reduced by intestinal bacteria [12].

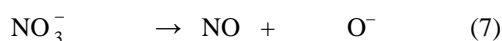
Nitrate which is naturally present in green leafy vegetables and also in the water, supply is rapidly converted to nitrite by the naturally occurring bacteria residing on our tongues, as well as in the intestines, and then absorbed into the bloodstream:



The amount of nitrate that is supplied by leafy vegetables and in drinking water is generally about 100-170 mgkg⁻¹ [13]. High concentrations of nitrates in vegetables can reflect the over-application of nitrate-containing fertilizer to the soils on which these vegetables are grown [14]. Investigations have shown that a high nitrate accumulation in plants results in nitrite production:



This is converted into nitric oxide (NO):



This together with O₂⁻ could be rapidly catalyzed by nitrate reductase into peroxy-nitrite (ONOO⁻) which has been reported to be highly toxic to plants [15].



The factors responsible for nitrate accumulation in plants are mainly nutritional, environmental, and physiological. Nitrogen fertilization and light intensity have been identified as the major factors that influence the nitrate content in vegetables [16]. Changes in light

intensity lead to a diurnal pattern of nitrate accumulation in plants. Nutrients like chloride, calcium, potassium, sulphate and phosphorus are involved in the nitrate accumulation process in plants. The nitrate content varies in various parts of plants [17-18] and with the physiological age of the plant [6-18]. A reduction in nitrate content can add value to vegetable products already popular for their nutritional and therapeutic properties [5]. Vegetables, an important component of the human diet and a major source of nitrate and nitrite constitute nearly 72 to 94% of the average daily human dietary intake [5]. Unfortunately, leafy vegetables grown under different agro-ecological conditions accumulate nitrate to potentially harmful concentrations. Generally, nitrate-accumulating vegetables belong to the families *Brassicaceae* (Rocket, radish and mustard), *Chenopodiaceae* beetroot, Swiss chard and spinach), *Amaranthaceae* (*Amaranthus*), *Asteraceae* (lettuce and parsley) [5].

It has been reported that vegetables that are consumed with their roots, stems and leaves have a high nitrate accumulation, whereas melons and those vegetables with only fruit as consumable parts have a relatively low nitrate accumulation [19–20]. Stopes *et al.* [21] compared nitrate content in organic and conventional produced vegetables sampled during winter in Great Britain. They found that the peak nitrate content was lower in inorganically produced vegetables, although there was considerable variation. A study at Salinas Valley, California, [22] reported that a relationship was found between the nitrate content of lettuce and the amount of nitrogen fertilizer applied. They found out that the nitrate content of the plant was largely dependent on the application of nitrogen fertilizer and on the growth rate of the plant.

A large population consumes this spinach and the fact that such a population is exposed to nitrate and nitrite suggests the need for the evaluation of the concentration of nitrate and nitrite in selected vegetables particularly spinach cultivated in spinach farm along Amba Stream in Lafia, Nasarawa State.

Sources of heavy metals contamination include atmospheric depositions, previous site used, and use of metal garden containers, paints, and burn fires. Through precipitation or ion exchange, heavy metals and their compounds enter soil, water, and plant ecosystems and thus, result in the accumulation or spread of heavy metals pollutants. Other important sources of heavy metals are liquid waste from metal galvanization, the tanning industry and other industries, sludge from sewage treatment plants, as well as municipal wastes. Unlike organic pollutants, they do not decay and thus pose a different kind of challenge for remediation and therefore call for thorough research on them.

The absorption of heavy metals by soil is regulated by several factors among which are pH and the organic matter content of the soil. When water from polluted sources is used for irrigation farming, it will eventually lead to increased levels of heavy metal uptake by the crops and consequently endangering human health [23].

Plants grown on land polluted with municipal, domestic, or industrial wastes can absorb heavy metals in the form of mobile ions present in the soil solution through their roots or foliar absorption. These absorbed metals get bio-accumulated in the roots, stems, fruits, grains, and leaves of plants [24]. When any of these plant parts are consumed by humans, it becomes hazardous particularly when the levels in the plant parts are well above tolerable limits as recommended by regulatory bodies such as WHO/FAO, NAFDAC, and SON. The health risks depend on the chemical composition of the waste materials, its physical characteristics, the vegetables cultivated and the consumption rate [25]. This study aims to assess the accumulation of heavy metals in spinach (*Spinacia oleracea*) irrigated with water from Amba stream in Lafia, Nasarawa State, to ascertain whether or not the consumption of these vegetables is safe for human consumption.

Materials and Methods

Study area

The study area is Lafia town, the headquarters of Lafia Local Government Area, and the capital of Nasarawa State, Nigeria. It is located between latitude 8,4833°E and longitude 8,5167°N and is bordered in the north by the Lafia North Development Area, and in the east by the Lafia East Development Area (Fig. 1). The water from the Amba stream has been regularly used for irrigation of vegetables grown and consumed in Lafia town and its environs. Waste from homes (domestic wastes), mechanic workshops, car wash stations, the cottage industries, etc empty into the stream. To enhance the yield of the vegetables, fertilizers, and manures are occasionally added to the soil. There are possibilities for over-application of these fertilizers and manures. Hence the uptake and storage of nitrate and nitrite from these waters, fertilizers and manures by these vegetables are very likely since these salts are soluble and mobile in groundwater.

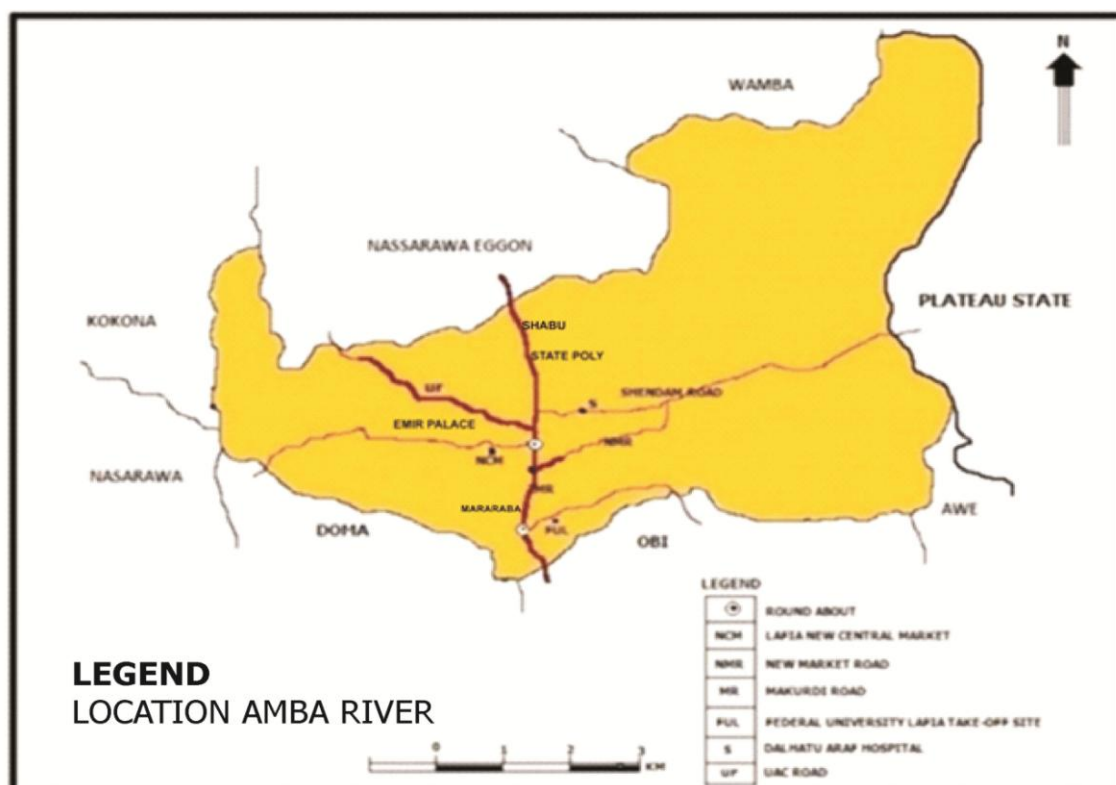


Figure 1: Map showing the study area

Sample and sampling

For this study, a total of three different spinach (*Spinacia oleracea*) and three soil samples were each collected simultaneously from the three distinct farmlands irrigated with the Amba stream water, cultivated with the application of fertilizers, manures, herbicides, and pesticides. During each collection, samples were randomly collected from different plots. Sample collections were carried out according to the methods described by [12, 26] into cleaned new polyethylene bags and transported to the laboratory [12, 26].

Spinach sample preparations for nitrate and nitrite analysis

Spinach (*Spinacia oleracea*) samples were cleaned to remove visible soil and then washed with tap water, thereafter with distilled water several times, and then sliced into nearly uniform sizes to facilitate drying at the same rate. The sliced samples were then dried in an oven at 105°C for 24 h until they were brittle and crisp. At this stage, no micro-organisms can grow, and care is taken to avoid any source of contamination.



The dried samples were mechanically ground into fine particles using clean mortar and pestle and sieved to obtain < 2 mm fractions. A portion (1 g) of each of the sieved samples was taken separately in 100 mL polyethylene or glass bottles, and 40 mL of distilled water was added, capped, and shaken for 30 min. The solutions were filtered, and the filtrates were made up to the marks in 100 mL volumetric flasks [12].

Digestion procedures for selected heavy metal

A 0.5 g of the powdered spinach sample was placed into the digestive tube followed by 5 mL of concentrated HNO₃ and 3 mL of concentrated H₂O₂. The mixture was heated at 160°C for 1 h in a fuming hood and then cooled to room temperature. After the addition of 20 mL of distilled water, the mixture was filtered, transferred to a 50 mL volumetric flask, diluted to the mark, and allowed to settle for 15 h. The digestion was done in triplicate.

Determination of nitrate [NO₃⁻] and nitrite [NO₂⁻] concentration in the spinach samples

The determination of nitrate in each of the spinach (*Spinacia oleracea*) sample solutions was performed by using a spectrophotometer (model 2000) at a wavelength of 543 nm. The equipment was scrolled to select the stored program number for nitrate (64 Nitrate-N). The result, which was obtained as nitrate-nitrogen (NO₃⁻ N) was converted to mg/kg nitrate (NO₃⁻) by multiplying by 4.4 (conversion factor) [27]. The concentration levels of nitrate (mg/kg) in the samples were calculated from:

$$\text{NO}_3^- \text{ (mg/kg)} = C \times V/M \quad (1)$$

Where, C is the concentration of NO₃⁻ in the sample (mg/kg), V is the total volume of the sample solution (100 mL) and M is the weight of the sample (1 g) [12].

Nitrite levels in the sample solutions were similarly determined except that in this case, different reagents were used. The program number for nitrite was 67 nitrite-N ((NO₂⁻ - N). and the reaction period was 5 min as against 10 min in the case of nitrate.

Determination of nitrate, nitrite and heavy metals concentrations in soil

Concentrations of nitrate and nitrite in soil samples obtained from the irrigated farm where spinach leaves were obtained were determined by spectrophotometric method like those previously described for spinach leaves [27]. The heavy metals Cu, Zn, Fe, Cr, Cd, Pb and Mn were determined by flame atomic absorption spectrophotometer (FAAS) that uses air/acetylene as an oxidant.

Transfer factors for concentration of nitrate and nitrite from soils to spinach

Transfer factor (TF) is the ratio of the concentration of nitrate or nitrite in a plant to the concentration of nitrate or nitrite in soil. TF for nitrate and nitrite were computed based on the method described by [28], according to the following formula:

$$TF = P_s \text{ (mgkg}^{-1} \text{ dry wt)}/S_t \text{ (mgkg}^{-1} \text{ dry wt)} \quad (2)$$

Where: P_s is the plant nitrate or nitrite content originating from the soil and S_t is the total nitrate of nitrite contents in the soil.

Data analysis

Data collected were subjected to statistical tests of significance using the student t-test and Analysis of Variance (ANOVA) at p<0.05 to assess pairs results in the spinach (*Spinacia oleracea*) and soil samples. That is, to assess significant variation in the levels of nitrate, nitrite, and heavy metals in vegetables as well as in soils. Probabilities less than 0.05 (p <0.05) were considered statistically significant. The correlation coefficient was used to determine the association between the nitrate and nitrite in spinach (*Spinacia oleracea*) at p = 0.05. All statistical analyses were done by SPSS software for Windows.

Results and Discussion

Nitrate (NO₃⁻) and nitrite (NO₂⁻) concentrations in spinach

Table 1 presents the concentrations of nitrate and nitrite in the spinach (*Spinacia oleracea*) samples obtained from the three irrigated farmland [Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)]. The concentration ranged from 3042.91 – 4977.26 mgkg⁻¹, with farm Sp2 recording the highest nitrate concentration of 4977.26 mgkg⁻¹. When compared to other farms, farm Sp1 had a nitrate concentration of 3042.91 mgkg⁻¹ which was the least. Statistical analysis showed that significant variation (p<0.05) exists between the farms' nitrate concentrations. The nitrate concentration in the spinach leaves obtained from all the irrigated farms was higher than [28] permissible limit (2500 mgkg⁻¹) and [29] concentration range of 2000 – 3000 mgkg⁻¹. The trend mean concentration of nitrate of spinach in the three different farmlands in descending order were farm Sp1 (3042.91 mgkg⁻¹) >farm Sp2 (4830.13 mgkg⁻¹)>farm Sp3 (4977.26 mgkg⁻¹). Previous work reported by [29] for spinach and other vegetables sold in Lafia showed that spinach nitrate concentration ranged from 792.22 – 5823.86 mgkg⁻¹. Environmental condition had been reported to be a serious factor that contributes to nitrate concentration accumulation [30]. High sunlight intensity increases nitrate levels in plant tissues by increasing the activity of the nitrate reductase. [29] showed that the type of crop is a primary factor that affects the nitrate level such that certain foods tend to accumulate large amounts of nitrate.

Nitrite concentration in the spinach leaves showed values that ranged from 0.00092 – 0.00102 mgkg⁻¹. These concentrations were significantly lower than [28] maximum level (100 mgkg⁻¹). Closed mean concentration values were also observed in irrigated farms Sp1 and Sp2, however, farm Sp3 significantly varied. The trend in the nitrite concentration was in the order: farm Sp3 > farm Sp1 > farm Sp2. The mean nitrite concentration recorded in this study was lower than [29, 31]. This has been opined [32] that continuous intake of low nitrite-containing food substances can cause cancer and methemoglobinemia after bioaccumulation in the body system.

Table 1: Nitrate and Nitrite Concentrations in Spinach (mgkg⁻¹)

Location	Nitrate (NO ₃ ⁻)	Nitrite (NO ₂ ⁻)
Sp1	3042.91±1.62 ^c	0.00094±0.00002
Sp2	4977.26±9.57 ^a	0.00092±0.00002
Sp3	4830.13±4.50 ^b	0.00102±0.00001

Data were expressed as means ± standard deviation of three replicates. Means with different superscript are statistically different (p<0.05). Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)

Table 2: Nitrate and nitrite concentration in soil (mgkg⁻¹)

Location	Nitrate (NO ₃ ⁻)	Nitrite (NO ₂ ⁻)
Sp1	5899.20±58.98 ^a	0.00332±0.00002
Sp2	7026.73±12.08 ^c	0.00473±0.00002
Sp3	6324.96±24.65 ^b	0.00903±0.00002

Data were expressed as means ± standard deviation of three replicates. Means with different superscript are statistically different (p<0.05). Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)

Nitrate (NO₃⁻) and nitrite (NO₂⁻) concentrations in soil

Table 2 presents the concentration of nitrate and nitrite in the soil samples. The result showed that nitrate concentration ranged from 5899.20 – 7026.73 mgkg⁻¹. Statistical different tests for variation (ANOVA) showed that significant differences exist in the irrigated farm soils. Farm Sp2 had the highest nitrate mean concentration (7026.73 mgkg⁻¹). As compared to previously reported work on farmland soil around irrigated sites by [33], values as low as 198.52 – 398.65 mgkg⁻¹ were obtained. Lokeshwari and Chandrappa [34], also reported nitrate concentration of 8012.05 – 8921.13 mgkg⁻¹ in irrigated soil which was higher than the concentration obtained in this report.

The results of the soil nitrite concentration were reported in Table 2. Nitrite concentration ranged from 0.00332 – 0.00903 mgkg⁻¹. Highest and lowest mean concentration was obtained in farm Sp1 and Sp3, respectively. Nitrite concentration obtained by European Food Safety Authority [35] were higher

(0.00988 mgkg⁻¹) than the result obtained in this study. Uwah *et al.* [36] also reported higher nitrite concentration (0.0100 mgkg⁻¹). The results from this study showed that nitrite accumulates in low concentration.

Heavy metal concentrations in the spinach and soil

Table 3 shows the metal content levels in the spinach (*Spinacia oleracea*) analyzed from all the samples obtained from Amba stream, Lafia. The values recorded for the mean concentrations of heavy metals in spinach ranged from Cd (1.29±0.01 mgkg⁻¹) to Fe (281.60±1.65 mgkg⁻¹). However, the concentrations of heavy metals in spinach are in the following order: Fe (281.60±1.65 mgkg⁻¹), Zn (59.80±3.01 mgkg⁻¹), Cr (54.20±1.00 mgkg⁻¹), Cu (30.60±1.10 mgkg⁻¹), Mn (9.10±1.00 mgkg⁻¹), Cd (1.29±0.01 mgkg⁻¹) and Pb (1.29±0.025 mgkg⁻¹), respectively.

Table 3: Results of heavy metals concentration in spinach (mgkg⁻¹)

Parameter	Sp1	Sp2	Sp3
Cu	33.50	36.80	34.90
Cd	0.58	1.20	1.00
Pb	7.60	7.82	7.80
Cr	65.20	64.10	66.30
Fe	1085.10	1085.09	1082.11
Zn	70.40	72.40	71.40
Mn	27.60	29.82	28.80

Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)

Table 4: Results of heavy metals concentration in soil (mgkg⁻¹)

Parameter	Sp1	Sp2	Sp3
Cu	30.60	29.50	31.70
Cd	1.30	1.28	1.29
Pb	1.49	1.46	1.51
Cr	54.00	53.00	55.00
Fe	280.60	283.50	280.70
Zn	59.78	56.80	62.82
Mn	9.10	8.10	10.10

Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)

Table 5: Average concentration of heavy metals in soil and spinach (mgkg⁻¹)

Metal	Spinach	Soil
Cu	30.60±1.10	35.07±1.6
Cd	1.29±0.01	0.93±0.32
Pb	1.29±0.02	7.74±0.12
Cr	54.20±1.00	68.20±1.10
Fe	281.60±1.65	1084.1±1.73
Zn	59.80±3.01	71.4±1.00
Mn	9.10±1.00	28.74±1.11



The metal content levels in the soil were analyzed from the samples obtained from the Amba stream, Lafia were shown in Table 4. The average concentrations of heavy metals recorded in soil ranged from Cd (0.93 ± 0.32 mgkg⁻¹) to Fe (1084.1 ± 1.73 mgkg⁻¹). The concentrations of the metals in soil are as follows: Fe (1084.1 ± 1.73 mgkg⁻¹), Zn (71.4 ± 1.00 mgkg⁻¹), Cr (68.20 ± 1.10 mgkg⁻¹), Cu (35.07 ± 1.6 mgkg⁻¹), Mn (28.74 ± 1.11 mgkg⁻¹), Pb (7.74 ± 0.12 mg/kg) and Cd (0.93 ± 0.32 mgkg⁻¹), respectively.

Table 5 shows the comparison between concentrations of heavy metals in soils and spinach. All soil samples have higher Pb concentrations compared to the spinach samples. Cr has the lowest concentration in soil samples, where samples; Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima) had 54.00, 53.00 and 55.00 mgkg⁻¹, respectively. Only a fair difference exists between the concentrations of Cd in all the soil and spinach samples analyzed. In general, Mn also recorded very high concentration values in all the soil samples from the irrigated farm Sp1, Sp2 and Sp3. Zn levels in the soil were far higher in all the spinach samples analyzed compared to their corresponding soil samples. All soil samples of Fe have a higher level of concentration than the corresponding spinach samples. The Cu content level is higher in spinach than in soil. The comparison of heavy metals in soils and spinach vegetables would show that all the metals determined were either more concentrated in the soils or compared favorably with the ones in the spinach vegetables on a pairwise basis. This meant that the soil would serve as a reservoir source of all the metals in the environment.

Nitrate and nitrite transfer factor from soil to spinach

Transfer factors of nitrate and nitrite from soil to spinach were presented in Table 6. Bio-availability computation of the transfer factor revealed that nitrate was well accumulated in farm Sp3 (0.76) and nitrites in farm Sp1 (0.283). The transfer factors for the anions between the soil and vegetables identify the efficiency of the spinach leaves to accumulate the nitrate and nitrite anions [37]. The computed transfer factor quantified the relative differences in bioavailability of the anions to the spinach leaves. These factors were based on the root uptake of the anions.

Table 6: Transfer factor from soil to spinach

Location	Nitrate (NO ₃ ⁻)	Nitrite (NO ₂ ⁻)
Sp1	0.520	0.283
Sp2	0.710	0.195
Sp3	0.760	0.113

Sp1 (Rafin Ganiya), Sp2 (Rafin Ganye) and Sp3 (Rafin Angwan Galadima)

Table 7: Transfer Factor (TF) from Soil to Spinach

Metal	TF
Cu	0.10
Cd	1.39
Pb	0.92
Cr	0.83
Fe	0.56
Zn	0.83
Mn	0.32

Heavy metals transfer factor from soil to spinach

The transfer factors from the soil to the spinach leafy vegetable and daily intake of heavy metal (mg/kg) are summarized in Table 7. To estimate heavy metals, the heavy metals transferred to plants are a function of both soil and plant properties due to the representative bioavailability of heavy metals to plants. The values of TF decreased with the increasing respective metal concentration of metal edible vegetables. The heavy metal transfer factors in spinach vegetable from the study area grown at the Amba stream of three sites were 0.10, 1.39, 0.92, 0.83, 0.56, 0.83, 0.32 for Cu, Cd, Pb, Cr, Fe, Zn, Mn, respectively. The trend of metal TF from soil to spinach vegetable grown at Amba Stream was in the order of Cd > Pb > Cr > Zn > Fe > Mn > Cu.

Conclusion

This study has been able to determine the concentration levels of nitrate, nitrite, and heavy metals in the spinach (*Spinacia oleracea*) and soil samples obtained in Amba Stream, Lafia, Nigeria, and suggested possible pollution of the study area due to excessive usage of fertilizer, manures, pesticides, herbicides, and other agrochemicals as well as the use of wastewater in irrigating the soils, and the environmental factors pertinent in the study area. The results were higher than the published maximum permissible contents of nitrate and nitrite in some spinach. Therefore, consumption of this spinach as food may pose a possible health hazard to humans and animals at the time of the study. Growers of spinach in these areas should be educated on the need to grow crops with safe levels of nitrate and nitrite fertilizers. From the result, it was noticed that the toxicity levels of heavy metals were found to be higher in all the samples analyzed. The concentration of the anions and as well as heavy metals in the spinach and soils obtained in this study would go a long way in providing baseline data for the assessment of the levels of nitrate and nitrite in the spinach and soils obtained in Amba Stream, Lafia, Nigeria.

Conflict of interest: Authors have declared that there is no conflict of interest reported in this work.

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