

# Concentration of Arsenic in the Muscle, Liver and Kidney of Consumed Calves in the West of Syria

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## Abstract

This study was conducted on 240 samples, 80 muscle, 80 kidney and 80 liver samples, which were collected from 80 calves' carcasses (between 1-3 years old) from Western Syria (Latakia, Tartus, Jableh and Safita). Samples were analyzed for the arsenic contamination using Atomic Absorption Spectrometry (AAS). The results showed that concentrations of arsenic in the kidney, liver and muscle of the calves in Latakia region were 0.7175, 0.602 and 0.4715 mg/kg, respectively. In Jableh region they were 0.43, 0.3425 and 0.2665 mg/kg, respectively, in Tartus region 0.5035, 0.4545 and 0.2918 mg/kg, respectively, and in Safita region 0.3615, 0.342 and 0.2168 mg/kg, respectively. Results also revealed that the highest arsenic concentration was detected in the kidney followed by liver and muscle, and was higher in the samples from Latakia and Tartus regions compared to the samples from Jableh and Safita regions.

## Keywords

arsenic — muscle — liver — kidney — calves

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## Introduction

Metals and chemicals from the food affect the human health. Some are useful having biological functions necessary for the body, such as copper, iron, selenium, and zinc, which can become toxic when highly concentrated (29). Some do not have any known function, for example cadmium, lead, mercury and arsenic. They may be harmful to the public health especially with high exposures (16). Heavy metals including arsenic, are considered the most widespread and dangerous pollutants to the environment.

Arsenic is a general pollutant to the environment since it can be found everywhere. Its components are considered hazardous to the public health (2). Actually, arsenic can enter the human body by inhalation, ingestion or through the skin (29). Almost 80% of the arsenic compounds are used in the industry, and particularly in agricultural medicines such as wood preservatives and pesticides for insects, weeds and fungi. Arsenic is also used in making dyes, textiles, papers, ceramics, paints, cleaning tools and medicines against some internal parasites such as tapeworm (17). Moreover, some arsenic compounds were used to treat diseases such as syphilis, dysentery, trypanosomiasis and psoriasis.

Arsenic-based drugs are still used in treating diseases such as parasitic diseases of the cattle, canine filariasis and blackhead disease in turkey and chicken (8; 17). Arsenic can be found in the environment in different concentrations: less than 1-3 ng/m<sup>3</sup> in the air in places far from pollution; 2-100 ng/m<sup>3</sup> in the cities with industrial factories, less than 0.01 mg/l in water, and 20-140 ng/kg in food. Arsenic concentrations vary according to the places increasing in the industrial areas, garbage dumps, and with other pollutants (29; 21).

Also, one can be professionally exposed to arsenic and

its compounds, for example in the factories of ceramics, glass, cement, melting factories, pharmaceuticals, mineral processing factories, pesticide manufacturing and wood preservatives (7). The toxicity of arsenic depends on the oxidation state, solubility, the dose, duration of exposure, age, sex, the way it gets into the body and genetic predisposition (12).

Triple inorganic arsenic compounds are considered one of the most toxic compounds especially the gallium arsenide compound (GaAs), which is widely used in industries (18). Arsenic causes disorders of the circulatory system, especially the vessels as well as neurological disorders and inflammation of the lymphatic tissues. Also, it disrupts the activity of enzymatic systems dependant on sulphhydryl groups, and can cause anemia, leukemia and diabetes. Arsenic can also cause an increase in white blood cells (specifically eosinophilia), necrosis of the cell walls, liver, kidney and intestine changes, gangrene, reproductive disorders, pigmentation and skin changes, and cancer particularly of the skin, bladder, liver, lung and urethra (5;14;19).

In addition, arsenic urges the programmed cell death of infected cells with leukemia (25). Animals differ in how much arsenic they accumulate according to the type of nutrition and provender (9). High concentration of arsenic in animals causes neurological changes, disruption of the reproductive system and acute inflammation of the digestive system (3).

The aims of this study were to estimate the concentration of arsenic in the meat, liver and kidney of the consumed calves in different areas in western Syria in addition to examining the arsenic concentration in consumed calves in the places nearby industrial facilities, and comparing them with those in the rural places. Also, the results were compared to the international reference values.

## Material and Methods

**Sample collection.** This study included a number of 1-3 years old calves from Latakia and Tartus (Western Syria). 240 random samples of flesh, kidneys and livers were taken from 80 slaughtered calves; 40 of which were from Latakia and Tartus (including villages and areas that are close to industrial facilities) in addition to 40 ones from Jableh, Safita and other villages far from any source of pollution. The calves were reared in these areas being fed with the grass and feed. In the summer of 2014 the flesh samples were taken from the calves' thighs, the liver samples from the left lobe, and kidney samples from the kidney margins. In the summer 2014 the flesh samples were taken from the calves' thighs, the liver samples from the left lobe, and the kidney samples from the kidney margins. The samples were then stored in the polyethylene bags and kept in a -20°C refrigerator until being analyzed.

**Chemical Materials and Substances Used.** All the chemical materials used in this study were of high purity according to the Atomic Absorption Standards. The digestive solution consisted of 60% of concentrated nitric acid (HNO<sub>3</sub>) and 40% of perchloric acid (HClO<sub>4</sub>) (24; 30). In each stage, the tubes were washed with distilled water and cleaned with a solution consisting of 520 ml of distilled water, 200 ml of hydrochloric acid and 80 ml of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). The tubes were then washed with 20% nitric acid (HNO<sub>3</sub>). Then, the tools were washed with distilled water and dried by air in an incubator away from any source of dust or pollution (6). The samples were analyzed and the arsenic portion was estimated using Atomic Absorption Spectrometry (Shimadzu AA6800) that uses graphic oven, which works on a (BGC-D2) lamp containing arsenic at a wavelength of 193.7 nm.

### Lab Analysis:

**Samples Digestion.** One gram of the sample (whether a muscle, kidney or liver sample) was accurately weighed and put into a tube with a thermometer set to 0°C. A graduated pipette was used to add 5 ml of digestive solution, and the tube was firmly closed, shaken and held in a vacuum to enable digestion until the next day. Then, the tubes were half-closed and kept for three hours in a 70°C water bath (keeping in mind that the tubes had to be shaken every half an hour), and left to cold to the lab temperature. 5 ml of

distilled water was then added, and a standard sample was prepared but with the deletion of the stage in which a sample was added for determining the arsenic concentration in the materials and solutions (26; 27).

**Filtration.** After digestion, the samples were filtered using Wattman no. 42 filtration paper and prepared to measure the arsenic concentration. Finally, the Atomic Absorption Spectrometry was used according to A.O.A.C. method (20) (standard measuring error = 0.001) to read the results.

**Statistical Analysis:** The American Statistical Analysis (Statistics, version 4.0) and the Analysis of One Way Variance ANOVA were applied for comparing the averages of study results, and Pearson's correlation factor between samples was calculated (23).

## Results

Results of the analysis of samples, rates of arithmetic means, standard deviation, and minimum and maximum levels are shown in Table 1.

The comparison of the arsenic concentration in the samples of the calves from Tartus and Safita demonstrated the highest concentration in the kidney followed by the liver and then the muscle (meat). Besides, the arsenic concentration was higher in the samples from Tartus than Safita (Figure 1). Figure 2 demonstrated higher concentrations in the calves from Latakia in comparison with Jableh, and also higher arsenic concentration in the kidney than in both the liver and meat.

Results showed that arsenic concentration in the kidney and meat was the highest in the samples from the calves of Latakia, and lowest in the samples from the calves of Safita. Moreover, the results showed a significant rise in the arsenic concentration in the liver samples from Latakia compared to a non-significant rise in the samples from Jableh and Safita (Figure 3).

A comparison of the results and the globally permitted limit of pollution of 0.4 mg/kg (22) demonstrated that the lowest pollution rate was detected in the samples of the calves from Jableh and Safita. The highest pollution rate was detected in the kidney samples of the calves from Latakia, which reached 45% (Table 2).

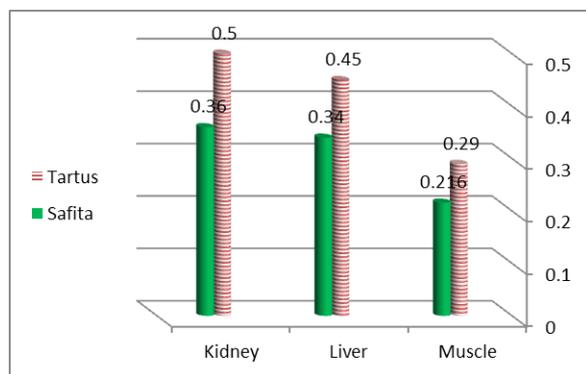
**Table 1.** Statistical data on muscle, kidney and liver samples in study regions (mg/kg) (n=20 of each organ)

Lo 95% CI- UP 95% CI	Max - Min	Mean±SD	Samples	Region
0.6444 – 0.2986	1.23 – 0.05	0.4715 ± 0.3695	Muscle	Latakia
0.8832 – 0.3748	1.84 – 0.07	0.629* ± 0.5431	Liver	
1.0185 – 0.4165	2.13 – 0.06	0.7175* ± 0.6432	Kidney	
0.4636 – 0.1199	1.21 - 0.005	0.2918 ± 0.3672	Muscle	Tartus
0.7097 – 0.1993	1.94 – 0.01	0.4545* ± 0.5453	Liver	
0.7732 – 0.2338	2.21 – 0.02	0.5035* ± 0.5761	Kidney	
0.3727 – 0.1603	0.78 – 0.04	0.2665 ± 0.2269	Muscle	Jablah
0.5020 – 0.1830	103 – 0.07	0.3425* ± 0.3408	Liver	
0.6475 – 0.2125	1.62 – 0.07	0.4300* ± 0.4647	Kidney	
0.3078 – 0.1258	0.8 – 0.016	0.2168 ± 0.1944	Muscle	Safita
0.4859 – 0.1981	1.31 – 0.07	0.3420* ± 0.3075	Liver	
0.5228 – 0.2002	1.42 – 0.05	0.3615* ± 0.3446	Kidney	

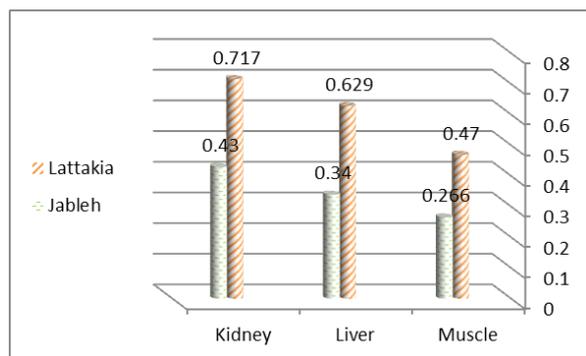
\*p<0.05 : statistically significant

**Table 2.** The percentage of polluted samples exceeding the globally permitted arsenic limit (0.4 mg/kg) in different samples of the studied organs of the calves in all study areas

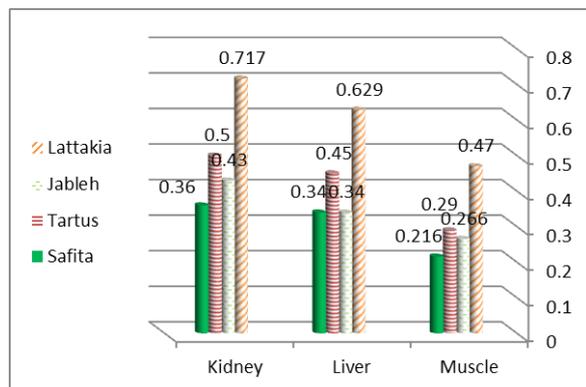
Animal	Meat	Liver	Kidney
Calves of Latakia	30%	40%	45%
Calves of Jableh	15%	20%	25%
Calves of Tartus	15%	30%	35%
Calves of Safita	10%	25%	25%



**Figure 1.** Comparison of the arithmetic means of the kidney, liver and meat of the calves from Tartus and Safita in mg/kg (no significant differences  $p > 0.05$ )



**Figure 2.** Comparison of the arithmetic means of the meat, liver and kidney of the calves from Tartus and Jableh in mg/kg (no significant differences  $p > 0.05$ )



**Figure 3.** Comparison of the arithmetic means of the meat, liver and kidney of the calves from the areas of study in mg/kg.

### Discussion and conclusion

All samples were collected randomly from the calves without any particular conditions, causing high standard devia-

tions and high ranges of the obtained results. It is clear that high concentrations of arsenic were recorded in the internal organs (kidney and liver), and then in the meat (muscles). Arsenic concentration in the areas of Latakia and Tartus was remarkably higher than that in the samples from Jableh and Safita, which may be due to industrial activities such as oil refinery, textile industry, tanneries, plastics industries, ceramic plants, glass and smelting plants, pharmaceuticals, mineral processing, wood preservatives, cement plants, port and free zone, and sewage etc. The lowest concentrations were found in Safita and Jableh due to the remarkable decline in arsenic emissions and the distance from the industrial activities. Arsenic concentration also varies according to age, breed and amount of food intake among calves. Since arsenic effect is cumulative in the body, it increases with age (11).

High arsenic concentration in the studied samples could be attributed to arsenic transfers to animals from the plants growing on the polluted soil in addition to adding fertilizers, chemical and organic tonics, and spreading pesticides (1). It was shown that arsenic concentration was remarkably highest in the kidneys due to their function and vital role in filtering the blood and removing toxins (13). Arsenic levels were also high in the liver samples due to its physiological activity in storing minerals, fat, iron, zinc and potassium, metabolizing toxins and biologically active substances, and detoxifying them into the harmless substances in the body. Despite the liver is the cemetery of the red blood cells in the body, it cannot eliminate arsenic. By comparing our results with other published studies and considering the differences of the places and aspects of the global studies, this study demonstrated significantly lower arsenic concentrations in the meat, liver and kidney in comparison with the results from Pakistan concerning the concentration of some heavy minerals in the meat and organs of cows, sheep and poultry. In the latter studies, arsenic concentration in the kidney, liver and meat of calves was 46.99, 52.44 and 46.46 mg/kg, respectively (13). Moreover, the arsenic concentrations obtained from our study were also lower than those from another study, which demonstrated the arsenic concentration in the small intestines, liver, kidney and meat of calves of 0.82, 1.4, 1.23, and 2.02 mg/kg, respectively (4). However, one study on the effect of pollution with toxic elements on 9-12 months old calves in Spain showed that arsenic concentration in kidney, liver and meat was 0.0131, 0.0124, 0.0387 mg/kg, respectively (15), which proved to be remarkably lower than arsenic levels detected in our study. Besides, the results obtained by our study were also higher than those of existence of heavy minerals in calves in Egypt where arsenic concentration in the kidney, liver and meat of calves was 0.01492, 0.00464, 0.005 mg/kg, respectively (10).

All in all, there were no significant differences in the arsenic concentrations in the studied areas ( $p > 0.05$ ). However, there was a significant difference in the arsenic concentrations between different types of collected samples (muscle samples, kidney samples and liver samples) in all areas ( $p < 0.05$ ). While the arsenic concentrations were high in the cities, they were lower in the rural areas. Additionally,

the higher arsenic concentrations in the kidneys compared to liver and muscles could be due to the nature of these tissues as they contain fat, and thus are more likely to accumulate arsenic. The difference of arsenic concentrations in calves' samples and their variations in different organs command finding suitable consumption systems to limit the harmful effects of arsenic on the consumers. Although significant differences in arsenic concentrations are present in the regions with contaminating industries, there is a need for overall scanning of the areas surrounding pollution-causing areas in order to identify the intensity of arsenic spread, and also scanning of the rest of Syria. Therefore, there is a need to relate the environmental map to the arsenic concentrations.

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## Koncentracija arsena u mišićima, jetri i bubrezima teladi konzumirane u zapadnoj Siriji

### Sažetak

#### Uvod i ciljevi

Teški metali, uključujući i arsen, se smatraju jednim od najrasprostranjenijih i najopasnijih zagađivača okoliša. Arsen je opći zagađivač okoliša s obzirom na svoju sveprisutnost. Arsen uzrokuje poremećaje u cirkulatornom sistemu, posebno na krvnim žilama, neurološke poremećaje i upale limfatičnih tkiva. Arsen, također, ometa rad enzimatskih sistema ovisnih o sulfhidrilnim grupama, i može uzrokovati anemiju, leukemiju i dijabetes uz još neke štetne efekte.

Generalno se životinje međusobno razlikuju po tome koliko arsena mogu akumulirati ovisno o vrsti prehrane i stočne hrane. Ciljevi ove studije jesu odrediti koncentraciju arsena u mesu, jetri i bubrezima konzumirane teladi u različitim područjima zapadne Sirije te ispitati koncentraciju arsena u konzumiranoj teladi u mjestima blizu industrijskih objekata, i usporediti je sa onom u ruralnim područjima. Rezultati su uspoređeni sa međunarodnim standardima.

#### Materijal i metode

Otopina za digestiju se sastojala od 60% koncentrirane nitratne kiseline ( $\text{HNO}_3$ ) i 40% perhloratne kiseline ( $\text{HClO}_4$ ). Korištene su i druge hemijske supstance visoke čistoće. Uzorci su analizirani, a udio arsena je određen atomskom apsorpcijskom spektrometrijom (Shimadzu AA6800) uz upotrebu grafičke peći koja koristi lampu (BGC-D2) sa arsenom na valnoj dužini od 193.7 nm.

Ova studija je uključila telad staru 1-3 godine porijeklom iz Latakije i Tartusa (Zapadna Sirija). Kupno 240 slučajnih uzoraka mesa, bubrega i jetre uzeto je od 80 teladi od kojih je 40 bilo iz Latakije i Tartusa (uključujući sela i područja u blizini industrijskih objekata), a 40 iz Jableha, Safite i sela koja su udaljena od bilo kakvog izvora zagađenja. Telad je uzgojena u ovim područjima gdje je prehranjivana travom i stočnom hranom. U ljeto 2014. go-

dine uzorci mesa sa uzeti sa butina, uzorci jetre iz lijevog režnja, a uzorci bubrega sa rubova. Nakon toga su uzorci spakirani u polietilenske vrećice, i držani u frižideru na  $-20^\circ\text{C}$  do analize.

#### Rezultati i interpretacija

Jasno je da su najviše koncentracije arsena zabilježene u unutarnjim organima (bubrezi i jetra), a zatim u mesu (mišići). Koncentracija arsena u područjima Latakije i Tartusa je bila znatno veća od one u uzorcima iz Jableha i Safite što se može obrazložiti postojanjem industrijskih aktivnosti kao što su rafinerije nafte, tekstilna industrija, kožare, industrija plastičnih materijala, tvornice keramike, staklare i topionice, farmaceutska industrija, obrada ruda, zaštita drveta, cementare, luke i slobodne zone, odlagališta otpada, itd. Najniže koncentracije su pronađene u Safiti i Jablehu zahvaljujući znatnom padu emisije arsena i udaljenosti od industrijskih aktivnosti. Pokazalo se da su koncentracije arsena daleko najveće u bubrezima zahvaljujući njihovoj funkciji i vitalnoj ulozi u filtriranju i uklanjanju toksina. Razine arsena su također bile visoke u uzorcima jetre zahvaljujući njenoj fiziološkoj ulozi u akumulaciji minerala, masti, željeza, cinka i kalija, metabolizmu toksina i biološki aktivnih supstanci koje detoksicira u supstance koje nisu štetne za organe. Nadalje, jetra je "groblje" crvenih krvnih zrnaca, ali se ipak ne može osloboditi arsena.

#### Zaključak

Nivoi arsena su bile veći u gradovima u odnosu na ruralna područja. Osim toga, veća koncentracija arsena u bubrezima u odnosu na jetru i mišiće se može pripisati prirodi ovih tkiva koja sadrže mast, i sklonija su akumulaciji ovog elementa. Razlika u koncentraciji arsena u uzorcima mesa teladi i njena varijacija u različitim organima nalaže pronalazak odgovarajućeg sistema konzumacije kako bi se ograničili štetni učinci arsena na potrošače.