Influence of the technology of processing of meat of snails in anabiosis on the content of toxic elements

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Abstract
An important problem today is the pollution of the territory of Ukraine with heavy metals: lead, cadmium, copper, zinc and others. Relevance of the topic is that nowadays the issues of safety and quality control are not fully resolved in the country, as well as the veterinary and sanitary assessment of the meat of snails of the genus Helix, which are used for food purposes; in particular, there is no data on the content of toxic elements in a meat of different species of snails for different processing modes. The aim of the study was to investigate the meat of snails of the genus Helix for different processing to find heavy metals in it. Meats of snails has been studied after different technological processing: live, chilled, and cooked and frozen (n = 30). The research has been conducted in the winter, when the snails have been in anabiosis. Results of the study show that in accordance with the requirements of DSTU 7821:2016 “Live edible gastropod molluscs. Specifications”, the content of toxic elements in the meat of live snails corresponds to the maximum allowable level. One of the key indicators of safety of meat, in accordance with the requirements of DSTU 7821:2016, is the content of toxic elements in it: lead, cadmium, copper, zinc. It has been found that the meat of snails, in addition to the above mentioned heavy metals, contains also arsenic and iron, which relate to toxic elements as well. Therefore, when developing a complex system of veterinary and sanitary assessment of quality and safety of snails’ meat, we propose to control it on the content of such toxic elements as arsenic and iron. In addition, we recommend to improve the “The Required Minimum List…” (2004) with the studies of the meat of snails of the genus Helix for different technological processing on the content of toxic elements: lead, cadmium, copper, zinc, arsenic and iron.

Key words: toxic elements, Helix pomatia, Helix aspersa maxima, Helix aspersa muller, snails’ meat, heavy metals.

1. Introduction

Increase of human impact on ecosystems has led to the environmental pollution with toxic compounds, including heavy metals, that has caused several important issues for veterinary medicine regarding the prevention of distribution, accumulation and control of the content of heavy metals in soil, water, feed, tissues of livestock and livestock products, in particular, in a meat of snails (Butsiak, 2005; Yakubchak et al., 2018).

The snails’ meat is a delicacy in many countries of the world. It is not only a tasty dish, but also a medication. Broth with snails is used as a healing tool for bleeding treatment, treatment of eyes diseases, digestive system organs, wounds healing. There are special species of snails, which are used for cooking. The most common one is a grape snail (the same for Helix pomatia, Burgundy snail, Roman snail). Snail dishes are considered to be dietary: 10 g of protein, 32 g of fat, 5 g of carbohydrate and vitamin B6, B12; Iron, Calcium, Mangan contain in 100 g of this product. The meat of snails is soft, it is easily digested and cleaved. For example, the digestion of pork or chicken usually lasts 4–5 hours, and the digestion of snails’ meat takes two hours. Therefore, to ensure the useful properties of this dietary product, it must meet the requirements of safety and quality as a food product (Gudyma & Krazhan, 1996).

An important problem today is the pollution of the territory of Ukraine with heavy metals – lead, cadmium, copper, zinc and others. These elements, which are not being destroyed in the soil, migrate in a chain “soil – plant (feed) – animals – products” and have negative impact on the physiological functions of organs and tissues, the clinical condition of animals (Zasiekin, 2002). It is known, that the pollution of the environment with heavy metals over the past few years has increased, which is particularly visible in the countries with a high level of pollution of the territory with heavy metals (Gudyma & Krazhan, 1996).
decade has increased several times and, according to scientists’ forecasts, will increase further. In big cities and industrial regions, the source of the pollution can be different activities of enterprises, vehicles, human waste. The use of some fertilizers and pesticides, containing toxic elements in their composition, also leads to the accumulation of heavy metals by plants (Davydova & Tagasov, 2002).

From the point of view of the problem, today the whole world community is in a worry. Thus, monitoring of the content of heavy metals in raw materials and finished products of the food group is carried out everywhere. As a result of these studies, high concentrations of Cu, Cd, Zn, Pb and Hg have been recorded in some species of sea fish and seafood exported to the markets of Spain (Bordajandi et al., 2004; Falcó et al., 2006), high average values of Hg have been observed in a meat of Norwegian lobster (Visciano et al., 2013), fish, shellfish and red meat products from small species of cetaceans, which are sold in the markets of Japan (Endo et al., 2005). In sunflower honey made in Turkey they have found the maximum content of Cu, Mn, Zn, Fe, Cd, Pb, Cr, Ni (Citak et al., 2012), in medicinal plants of Pakistan – Cd, Cr, Cu, Pb, Fe, Mn, Ni, Zn (Cordell & Colvard, 2012).

Relevance of the topic is that now safety and quality issues are not fully resolved in the country, as well as the veterinary and sanitary assessment of the meat of snails of the genus Helix, used for food purposes. The importance of the topic is also confirmed by the fact that in Ukraine, except of DSTU 7821:2016 “Live edible gastropod molluscs. Specifications”, there is no normative base for the meat of snails.

Today, methods of control of safety of meat in Ukraine are introduced, according to a. 3.6. “The Required Minimum List...” (2004), according to which in the non-fish commercial objects (shellfish, crustaceans, invertebrates, seaweed) and products of their processing, the earthworms, reptiles, the mass fraction of lead is 10 μg/kg, cadmium – 2.0 μg/kg, arsenic – 5.0 μg/kg, hydrargyrum – 0.2 μg/kg.

Taking into account the above facts, we conclude that it is impossible to guarantee an objective assessment of the indicators of safety and quality of snails’ meat, as there is no data in this document on the content of toxic elements in the meat of snails of various species for different technological treatment.

The aim of the study is to study the meat of snails of the genus Helix for different technological processing on the content of heavy metals.

2. Materials and methods

Studies have been carried out on the meat of snails of genus Helix, sub-species: Helix pomatia, Helix aspersa maxima and Helix aspersa muller. 30 samples of snails of each species grown in snails farm in the Kyiv region have been selected for the study. Meat of snails has been investigated for different technological treatments: live, chilled, and cooked and frozen (n = 30). The research has been conducted in the winter, when the snails have been in anabiosis. Grape snail (Helix pomatia) has been studied only in a cooked and frozen state with the shelflife up to 6 months under the temperature of -18°C.

In the meat of snails for different technological processing modes, the content of such toxic elements was investigated: mass fraction of lead, cadmium, arsenic, hydrargyrum, copper, zinc and iron.

Mass fraction of lead, arsenic and cadmium has been determined by the method of atomic absorption spectrometry with electrothermal atomization, using the atomic absorption spectrophotometer Varian 240 Z (Australia) to find lead; and a thermo-absorption spectrophotometer Thermo Solaris (the USA) – to find arsenic; atomic absorption spectrophotometer Varian 240 G (Australia) – to find cadmium.

Mass fraction of copper, zinc, iron has been determined by atomic absorption spectrometry method with atomization in a flame by means of an atomic absorption spectrophotometer of type B-55 (Varian, Austria); to determine the indicators of iron they used atomic absorption spectrophotometer of type AA – 55 B (Varian, Austria).

Determination of heavy metals in the meat of snails of the genus Helix for different technological processing has been carried out in accordance with GOST 30178-96, DSTU 7670:2014, MAL 77-12-97.

Mass fraction of hydrargyrum has been determined by the method of atomic absorption spectrometry (without previous sample preparation). The method of quantitative determination of hydrargyrum is based on the selective absorption of free atoms of hydrargyrum with a certain wave length. Absorption measurements has been carried out at a wavelengths of 253.65 nm. Calculation of the hydrargyrum concentration in the meat of snails has been carried out according to the calibrated schedule (depending on the amount of absorption from the concentration of hydrargyrum).

Determination of hydrargyrum in the meat of snails has been carried out in accordance with ISO 11212 – 2: 1997, GOST 26927 – 86, using an atomic absorption spectrophotometer (single-beam spectrometer with two successive measurement cells) “Milestone DMA-80”.

The reliability of the difference between the arithmetic means of the two variation series was determined according to the Student’s criterion, taking into account the validity limit: P ≤ 0.05; P ≤ 0.01, P ≤ 0.001. The obtained research results were processed using the Microsoft Excel computer program.

3. Results and discussion

Results of the study on the meat of snails of different species for different technological processing show that the content of lead in the body of the live H. aspersa muller snails is by 18.2% (P ≤ 0.05) higher, compared to the indicators of H. aspersa maxima snails. However, in the chilled meat of snails of H. aspersa muller the content of lead is by 23.9 % (P ≤ 0.001) higher, compared to the chilled meat of H. aspersa maxima snails (Table 1).
The study found that in the cooked and frozen meat of *H. aspera maxima* snails the content of lead is by 9.4%, and in the meat of *H. aspersa mullerr* – by 3.1% lower, compared with its content in the cooked and frozen meat of *H. pomatia*.

Results of the study of meat of snails for different technological processing also indicate, that in the chilled meat of *H. aspera maxima* snails the content of lead is lower by 38.8% (P ≤ 0.01) compared to the indicators of the live *H. aspersa mullerr* snails. However, in the chilled meat of *H. aspersa mullerr* snails the content of cadmium is by 22.7% (P ≤ 0.001) higher than that of the living snails of the same species, and in the meat of cooked and frozen snails – by 40.9% (P ≤ 0.01), respectively.

According to the requirements of the current DSTU 7821:2016 “Live edible gastropod molluscs. Specifications”, the content of lead in live snails has to be 10 μg/kg. Based on the results of our research, it is evident that the content of lead in live snails does not exceed the permissible level, however natural presence of lead in a soil and a water caused the presence of its residues in the environment, including the body of snails.

Typically, lead gets into food from polluted environment and during the products’ processing with lead itself. The addition of lead compounds to gasoline as an anti-knocker led to a significant release of lead into the surrounding area, especially near the highway roads, so the attention should be paid to the location of the snails farms. Pollution of the environment occurs during the smelting of lead and in case of dumping of water from mines. Long-term use of pesticides, that contain lead, causes a situation when lead gets into the body of snails directly from the contaminated soil; at the same time, the content of lead in feed, which is used for gastropod molluscs feeding, increases. Drinking water, that is used for meat processing of snails, also increases the absorption of lead.

Results of the study show, that the content of cadmium in the body of live *H. aspersa mullerr* snails is by 22.7% (P ≤ 0.001) higher, compared to the indicators of the live *H. aspera maxima* snails. However, in the chilled meat of *H. aspersa mullerr* snails the content of cadmium is by 12.8% (P ≤ 0.001) higher, compared to the chilled meat of *H. aspera maxima* snails (Table 1). In the cooked and frozen meat of *H. aspersa maxima* snails the content of cadmium tends to decrease by 0.76%, in the meat of *H. aspersa mullerr* snails – a similar tendency – lower by 1.5%, compared to the meat of *H. pomatia* snails.

Table 1
The content of toxic elements in the meat of snails of various species and for different processing technology, M ± m, n = 30

| Indicator                  | Technological processing of snails’ meat | Snails’ species |
|----------------------------|----------------------------------------|----------------|
|                            | H. aspera maxima                        | H. aspersa mullerr | H. pomatia |
| Mass fraction of lead, mg/kg | Live                                   | 0.018 ± 0.001 | 0.022 ± 0.001* | – |
|                            | Chilled                                | 0.011 ± 0.001▲ | 0.046 ± 0.001***▲| – |
|                            | Cooked and frozen                      | 0.029 ± 0.001▲ | 0.031 ± 0.001▲ | 0.032 ± 0.001 |
| Mass fraction of cadmium, mg/kg | Live                                   | 0.005 ± 0.001 | 0.022 ± 0.001*** | – |
|                            | Chilled                                | 0.005 ± 0.001 | 0.039 ± 0.001*** | – |
|                            | Cooked and frozen                      | 0.130 ± 0.001 ▲ | 0.129 ± 0.001 ▲ | 0.131 ± 0.001 |
| Mass fraction of arsenic, mg/kg | Live                                   | 0.009 ± 0.001 | 0.010 ± 0.001 | – |
|                            | Chilled                                | 0.008 ± 0.001 | 0.009 ± 0.001 | – |
|                            | Cooked and frozen                      | 0.008 ± 0.001 | 0.009 ± 0.001 | 0.010 ± 0.001 |
| Mass fraction of hydrgyrum, mg/kg | Live                                   | 0.004 ± 0.001 | 0.004 ± 0.001 | – |
|                            | Chilled                                | 0.005 ± 0.001 | 0.004 ± 0.001 | – |
|                            | Cooked and frozen                      | 0.003 ± 0.001 | 0.003 ± 0.001 | 0.005 ± 0.001 |
| Mass fraction of copper, mg/kg | Live                                   | 6.29 ± 0.074 | 10.44 ± 0.101*** | – |
|                            | Chilled                                | 8.49 ± 0.006▲ | 5.66 ± 0.006☆ | – |
|                            | Cooked and frozen                      | 36.24 ± 0.050****▲▲▲ | 35.42 ± 0.063****▲ | 37.30 ± 0.063 |
| Mass fraction of zinc, mg/kg | Live                                   | 7.16 ± 0.008 | 13.82 ± 0.007*** | – |
|                            | Chilled                                | 7.47 ± 0.010▲ | 21.38 ± 0.009*** | – |
|                            | Cooked and frozen                      | 14.18 ± 0.024****▲▲▲ | 13.24 ± 0.008****▲ | 15.10 ± 0.008 |
| Mass fraction of iron, mg/kg | Live                                   | 8.37 ± 0.011 | 20.81 ± 0.006*** | – |
|                            | Chilled                                | 10.12 ± 0.011 ▲ | 23.68 ± 0.014▲ | – |
|                            | Cooked and frozen                      | 19.71 ± 0.009****▲▲▲ | 20.24 ± 0.008****▲ | 21.77 ± 0.011 |

Note: *P ≤ 0.05 – compared to the live *H. aspera maxima* snails

**P ≤ 0.001 – compared to the chilled meat of *H. aspera maxima* snails

****P ≤ 0.001 – compared to the live *H. pomatia* snails

▲▲▲P ≤ 0.001 – compared to the cooked and frozen meat of *H. aspersa maxima* snails

▲▲P ≤ 0.01 – compared to the live *H. aspersa mullerr* snails

▲▲P ≤ 0.01 – compared to the live *H. aspersa maxima* snails

▲▲▲P ≤ 0.001 – compared to the live *H. aspersa maxima* snails

▲▲ P ≤ 0.001 – compared to the live *H. aspersa mullerr* snails

▲▲▲ P ≤ 0.001 – compared to the cooked and frozen meat of *H. aspersa maxima* snails

▲▲▲▲ P ≤ 0.001 – compared to the live *H. aspersa maxima* snails
cies, and in the cooked and frozen meat – by 17.05% (P ≤ 0.001), respectively.

According to the requirements of DSTU 7821:2016, the content of cadmium up to 2 μg/kg in a meat of live snails is allowable. The results of our study show that the content of cadmium in a meat of live snails does not exceed the maximum allowable level (MAL), but its residual amounts have been found in a meat.

Recently, it has been found that the increased use of sewage drains with a high content of cadmium can cause an increase of this toxic element in the environment. Biological accumulation of cadmium in edible plants and bioavailability in soils treated with sewage is influenced by several factors, including plants species, pH of the soil, presence of other microelements and the intensity of treatment with sewage (Fowler, 1979; Bingham, 1979). There are plants that accumulate cadmium and it is possible that they can be used for molluscs feeding.

Kneip and Hazen (Kneip & Hazen, 1973) have studied the marsh ecosystem, contaminated with cadmium, and have found that this toxic element is absorbed by marsh and aquatic plants, as well as by animals. It is important to note that toxicity of cadmium depends on the content of zinc in products. Symptoms of zinc deficiency are amplified with large doses of cadmium, but the toxicity of cadmium is reduced after increased zinc dozes receiving.

Results of our study show that in the meat of live H. aspersa mullerr snails the content of arsenic tends to increase by 10%, compared to the meat of live H. aspersa maxima snails. However, in the chilled meat of H. aspersa mullerr snails its content tends to increase by 12.5%, as compared to the chilled meat of H. aspersa maxima snails (Table 1).

In the cooked and frozen meat of H. aspersa maxima snails the content of arsenic tends to decrease by 20%, and in the meat of H. aspersa mullerr snails – by 10%, respectively, compared with the meat of H. pomatia snails.

Results of the study on the content of hydrargyrum in the meat of live and the chilled meat of H. aspersa maxima and H. aspersa mullerr snails show, that the hydrargyrum content is in equal quantities in these samples (Table 1). However, in the cooked and frozen meat of H. aspersa maxima, H. aspersa mullerr snails the content of hydrargyrum tends to decrease by 40%, compared to the meat of H. pomatia snails.

According to the requirements of DSTU 7821:2016, the content of hydrargyrum up to 2 μg/kg in a meat of live snails is allowable. This means that the results of our study meet the requirments and do not exceed the maximum allowable level.

From the sources of literature, it is known that all living organisms can accumulate hydrargyrum. The release of a significant amount of hydrargyrum in the environment took place before the introduction of rules prohibiting the discharge of hydrargyrum into the environment, which led to the pollution and accumulation of its small amounts in environmental objects, in particular – in the meat of snails.

Analysis of the results of the study show, that the content of copper in the meat of live H. aspersa mullerr snails is by 2.8% (P ≤ 0.001) lower, and in the meat of H. aspersa mullerr snails – by 5.04% (P ≤ 0.001) lower, compared to the meat of H. pomatia snails.

It has been found that in the chilled meat of H. aspersa maxima snails the copper content is by 34.9% (P ≤ 0.001) higher, and in the cooked and frozen meat – by 17.3% (P ≤ 0.001) higher, compared to the body of live snails of the same species. While in the chilled meat of H. aspersa mullerr snails the content of copper is by 54.2% (P ≤ 0.001) lower, and in the cooked and frozen meat of snails – by 29.4% (P ≤ 0.001) higher, compared to the body of live snails of the same species.

According to the requirements of DSTU 7821:2016, the content of copper up to 30 μg/kg in a meat of live snails is allowable. Our research shows that in the body of live snails in the winter period the content of this toxic element does not exceed the maximum allowable level.

In the meat of live H. aspersa mullerr snails the content of zinc is by 51.8% (P ≤ 0.001) higher than in the body of live H. aspersa maxima snails; but in the chilled meat of H. aspersa mullerr snails the content of zinc is by 34.9% (P ≤ 0.001) higher, compared to the indicators of the chilled meat of H. aspersa maxima snails (Table 1).

In the cooked and frozen meat of H. aspersa maxima snails the content of zinc is by 6.09% (P ≤ 0.001) lower, and in the meat of H. aspersa mullerr snails – by 12.3% (P ≤ 0.001) lower, compared to the meat of H. pomatia snails.

The results of the study show that in the chilled meat of H. aspersa maxima snails the content of zinc is by 4.3% (P ≤ 0.001) higher, and in the cooked and frozen meat of snails – by 98% (P ≤ 0.001) higher, respectively, than the zinc content in the body of live snails. In the chilled meat of H. aspersa mullerr snails the content of zinc is by 54.7% (P ≤ 0.001) higher, whereas in the cooked and frozen meat of the same species of snails it is by 4.1% (P ≤ 0.001) lower, compared to the zinc content in live H. aspersa mullerr snails.

Results of the study show that the content of zinc in the body of live snails does not exceed the maximum allowable level, as according to the DSTU 7821:2016 the content of zinc in a body of live snails has to be below 200 μg/kg.

The content of iron in the body of live H. aspersa mullerr snails is by 40.2% (P ≤ 0.001) higher than in the body of the live H. aspersa maxima snails; but in the chilled meat of H. aspersa mullerr snails the content of iron is by 42.7% (P ≤ 0.001) higher than in the chilled meat of H. aspersa maxima snails (Table 1). In the cooked and frozen meat of H. aspersa maxima snails the content of iron is by 9.46% (P ≤ 0.001) lower, and in the meat of H. aspersa mullerr – by 7.02% (P ≤ 0.001) lower, compared to the meat of H. pomatia snails.

Results of the study show, that in the chilled meat of H. aspersa maxima snails the content of iron is by 20.9% (P ≤ 0.001) higher, while in the cooked and frozen meat of snails – by 42.4% (P ≤ 0.001) higher, respectively, as compared to the indicators of the iron content in a body of live snails. In the chilled meat of H. aspersa mullerr snails the content of iron is by 13.7% (P ≤ 0.001) higher, and in the cooked and frozen meat of the same species – by 2.8% (P ≤ 0.001) lower, in comparison with iron content in live H. aspersa mullerr snails.
4. Conclusions

1. According to the requirements of the current DSTU 7821:2016 “Live edible gastropod molluscs. Specifications”, the content of toxic elements in the body of live snails, which have been under study in the winter period in the snail farm of the Kyiv region, corresponds to the maximum allowable level.

2. It has been found that the content of toxic elements in a chilled and cooked and frozen meat of snails is higher, than in bodies of live snails of various species. Apparently this is due to the technological processing of meat, as the insufficient quality equipment may affect the process of the final product contamination with high-toxic elements.

3. One of the basic indicators of the safety of snails’ meat, according to DSTU 7821:2016, is the content of toxic elements in it: lead, cadmium, hydrargyrum, copper and zinc. In the meat of *H. aspersa maxima*, *H. aspersa mullerr*, *H. pomatia* snails, in addition to the above mentioned heavy metals, arsenic and iron, which are considered to be toxic elements as well, have been found. Therefore, the complex system of veterinary and sanitary assessment of the safety of meat of snails should include an analysis on toxic elements, such as arsenic and iron.

4. We recommend to improve the “The Required Minimum List…” (2004) with indicators of the content of such toxic elements: lead, cadmium, hydrargyrum, copper, zinc, arsenic and iron for different technological processing.

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