

ESTIMATION OF *RAPANA VENOSA* (VALENCIENNES, 1846) QUALITY, A MARINE LIVING RESOURCE FROM THE ROMANIAN BLACK SEA WITH BIOECONOMIC IMPORTANCE

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*The paper presents some aspects regarding the biochemical composition (dry weight, moisture, organic substance, ash, total proteins, carbohydrates and lipids), the level of chemical contaminants (heavy metals - Cu, Cd, Cr, Pb, Ni and organochlorine pesticides - OCPs) and microbiological indicators (total coliforms - TC, fecal coliforms - FC - Escherichia coli, and enterococci - fecal streptococci - FS) of the rapa whelk *Rapana venosa* species, for estimation of its quality with bioeconomic importance. The assessment of those parameters indicates the marine organism quality, the chemical and microbiological contaminant level, nutritive value and the potential for biotechnological utilization. The obtained values for the analyzed parameters confirm the potential for use of this marine resource for bioeconomy and biotechnological purposes.*

Keywords: *Rapana venosa*, Romanian Black Sea coast, quality, biotechnology, bioeconomic importance

1. Introduction

The Asian marine gasteropod species *Rapana venosa* (Valenciennes, 1846) (Fig. 1), common name the veined rapa whelk or Asian rapa whelk, is a mollusk in the family Muricidae, the rock shells and a large sea snail, which has become an invasive species in different regions around the world. It is native to the Sea of Japan and since the 1980's it has become a valuable commercial bioresource [1 - 4]. It has recently also been included in the diet of those native to

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the Black Sea area and represents over 90% of the total marine living resources catches along the Romanian Black Sea coast [5]. The exploitation of the marine living resources started off back in National Institute for Marine Research and Development "Grigore Antipa" (NIMRD) of Constanța, Romania, since 1987, and become in time one of the objectives and directions for biotechnological research with bio economical utilization.

Consumption of seafood is proven to be preventing the life style diseases and giving a long-term health claims. This invertebrate is also used in marine biotechnology, biomedical and pharmaceutical domain [6, 7]. *Rapana venosa* species from Romanian Black Sea coast is an available marine resource and in sufficient quantities to be exploited for human food and marine biotechnology.

The tendency of the last period is to give special importance to the study and selection of bioactive substances of marine origin for use in human and veterinary medicine, the researchers being concerned about the future extracts obtained from the marine environment vegetal or animal species.



Fig. 1. *Rapana venosa* species - aspect of the natural environment (left) and detail (right)

2. Experimental

The biological samples were collected in August 2018, from Vama Veche, Constanța County, an area in the Southern part of the Romanian Black Sea coast, which is considered less affected by pollution and recognized as a marine protected zone. The biochemical composition (total proteins – Lowry method [8], carbohydrates - Dubois method [9] and lipids - Marsh and Weinstein method [10]) was analyzed using an UV-Vis spectrophotometer and gravimetric method (dry weight - DW, moisture, organic substance - OS, ash) according to Romanian Pharmacopoeia, 10th Edition [11].

The heavy metals (Cu, Cd, Pb, Cr, Ni), were analyzed using graphite furnace-atomic absorption spectrometers [12].

The analytical determination of the organochlorine pesticides-OCPs content (hexachlorobenzene - HCB, lindane, heptachlor, aldrin, p,p'dichlorodiphenyldichloroethylene - p,p'DDE, dieldrin, endrin, p,p'dichlorodiphenyldichloroethane - p,p'DDD, p,p'dichlorodiphenyltrichloroethane - p,p'DDT) was made by the gas-chromatographic method.

The microbiological aspects were assessed by "most probable number" (MPN) method which was used to analyze the concentrations of total coliforms - TC, fecal coliforms - FC - *Escherichia coli*, and enterococci (fecal streptococci - FS).

3. Results and discussion

In the Black Sea, mollusks are one of the most important and valuable group of benthic animals as they are widespread in the biotic area of the Pontic basin, dominating the total biomass of the benthos of the continental shelf, and represent an essential trophic element [13].

Rapana venosa is a carnivorous species, performing seasonal migration and preferring habitats occupied by bivalve and crustacean species in a prey-predator relationship [14].

From the biochemical point of view, in the gastropod *Rapana venosa*, the following parameters were investigated: dry weight - DW, moisture, organic substance - OS, ash, total proteins, total carbohydrates, total lipids (Fig. 2).

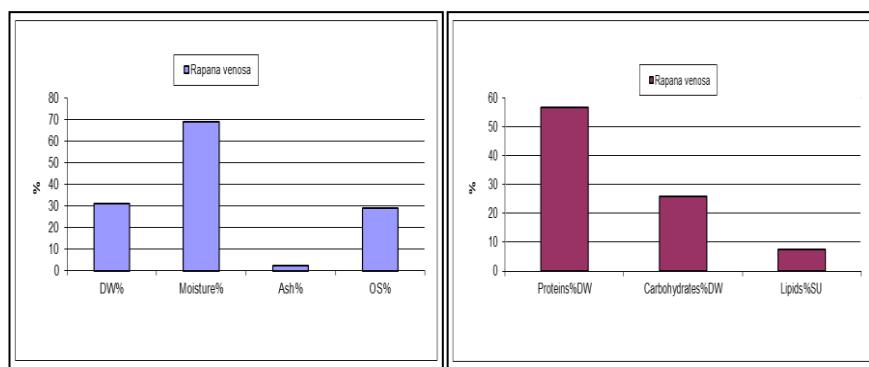


Fig. 2. The *Rapana venosa* biochemical content (dry weight- DW, moisture, organic substance - OS, ash, total proteins, carbohydrates and lipids)

The ash content and dry wet were low in August period (summer) and that is confirmed by other previous studies [6].

According to the sea snail's data sampled month, dry weight and ash content dropped their lowest level in August that clearly showed spawning period; mating period occurred during winter and spring and egg cases were laid in May to August. Similar results were obtained that ash rate was obtained lower in August and September than other months [15].

The analysis of the biochemical composition indicates a high concentration of protein and a low concentration of lipid content in *Rapana venosa* (Fig. 2). The high nutritional value of the marine organism is given by the high concentration of protein, as confirmed by the specialized literature. The rapa whelk has high protein rate and satisfactory amino acid content. Even though the nutritional values may vary depending on the season, feeding habits or habitat of the organism; rapa whelk provides a relatively consistent nutritive value year-round [16].

In terms of lipid content even though rapa whelk has a well-balanced fatty acid composition, it is deficient alone for the diet. *Rapana venosa* appears to be best as diet with relatively high protein and low lipid [17]. The heavy metal concentrations level in *Rapana venosa* from Vama Veche area were generally within the range of normal variation and decreased in the order: Cu > Cr > Cd > Ni > Pb. The concentration of cadmium (9.49 $\mu\text{g/g}$ DW) exceeds the maximum allowable limit (MAC), for human consumption (EC 1881/2006), 5.00 $\mu\text{g/g}$ DW, but it must be taken into account that the total tissue (sole and viscera) has been analyzed. The concentration of lead (0.56 $\mu\text{g/g}$ DW) measured in marine organism is well below the maximum allowable limit (MAC), 7.50 $\mu\text{g/g}$ DW (Fig. 3).

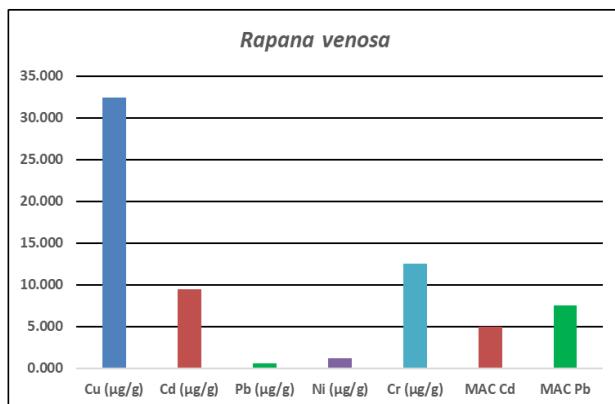


Fig. 3. The level of chemical contaminants, heavy metals (Cu, Cd, Pb, Cr, Ni) in *Rapana venosa* and maximum allowable limits (MAC)

Comparing the heavy metals concentration of *Rapana venosa* with other species from the Black Sea coast, bivalve *Mytilus galloprovincialis* (Lamarck, 1819), a marine mollusk in the family Mytilidae, the results showed the following variation (NIMRD data): Cu (10.19 $\mu\text{g/g}$ DW) > Cd (3.02 $\mu\text{g/g}$ DW) > Ni (2.04 $\mu\text{g/g}$ DW) > Cr (0.67 $\mu\text{g/g}$ DW) > Pb (0.31 $\mu\text{g/g}$ DW).

Generally, *Rapana venosa* presents a higher concentration of the heavy metals than bivalves because it feeds with them. In that case the bioaccumulation of the heavy metals in *Rapana venosa* is linked with the level of pollutants from

its food. Because the metals pollution in aquatic environments can be harmful to human health, it is necessary to understand and periodical control the hazard levels of pollution in seafood. The heavy metals are very toxic because, as ions or in compound forms, they are soluble in water and may be readily absorbed into aquatic organisms [18].

In the aquatic environment there are many factors that influence heavy metal accumulation in living organisms, these factors acting differently, depending on the species and type of metal, for example: pH, various organic compounds, the humic substrate, complex particles, the presence or absence of other minerals, anions, various ionic bonds, temperature, salinity, light intensity, the redox potential and dissolved oxygen concentrations [19]. The organochlorine pesticides (OCPs) concentration in *Rapana venosa* varied between detection limits (LOD) and 0,1922 µg/g DW. The highest concentrations were measured for p,p'DDT and its metabolites (Tab. 1).

The organochlorine pesticides (dieldrin, p,p'DDD and p,p'DDT) concentrations determined in *Rapana venosa* were higher than those reported in other species, such as bivalve *Mytilus galloprovincialis* from the Romanian Black Sea coast and shellfish from the Egyptian Mediterranean area (Tab. 1).

Table 1
Comparative OCPs in shellfish samples from the Romanian Black Sea and Egyptian Mediterranean Sea

µg/g DW	LOD	<i>Rapana venosa</i> from the Romanian Black Sea coast	<i>Mytilus galloprovincialis</i> (NIMRD data) from the Romanian Black Sea coast	Shellfish from the Egyptian Mediterranean coast [20]
HCB	0.0005	0.0005	0.0164	-
Lindan	0.0004	0.0227	0.0004	-
Heptaclor	0.0003	0.0003	0.0411	0.0006
Aldrin	0.0003	0.0003	0.0129	0.0018
p,p'DDE	0.0003	0.0002	0.0002	0.0045
Dieldrin	0.0004	0.0316	0.0003	0.0020
Endrin	0.0002	0.0004	0.4441	0.0357
p,p'DDD	0.0002	0.1581	0.0686	0.0086
p,p'DDT	0.0002	0.1922	1.9111	0.0086

The determined organochlorine pesticide values have generally been within the range of values admissible for human consumption (except: dieldrin, p,p'DDE, p,p'DDD and p,p'DDT) and can significantly depend on the age and/or condition of the organisms.

Organochlorinated compounds are part of a class of compounds generically referred to as "persistent organic pollutants" (POCs), which have been

associated with a significant environmental impact on a wide variety of species and virtually on all trophic levels. Many persistent organic pollutants have been implicated in a wide variety of adverse effects on the environment and human health, including impaired reproduction, endocrine disorders, reduced immunity and cancer [20].

The microbiological quality of the marine snail *Rapana venosa* was assessed according to the national and European norms of water quality for mollusks (Council Directive no. 79/923/EEC transposed by the GD no. 201/2002). The level of microbiological indicators concentrations recorded in the meat of these mollusks of commercial interest from the Romanian Black Sea coast in August 2018, was below the accepted limits by regulations (< 300 CF/100 g), ranging from 0 to 20 MPN/100 g (Tab. 2).

Different coastal marine waters and groundwaters host and carry many pathogenic microorganisms that cause disease in both humans and animals. Part of the pathogenic microorganisms are naturally found in marine and ocean environments, being their natural constituents, while others, considered anthropogenic contaminants (e.g. enteric or enterobacterial bacteria, protozoa, viruses, etc.), reach the coastal environments, mostly by pollution with contaminated wastewater [21 - 23].

Table 2

The microbiological indicators level in *Rapana venosa* species

Microbiological indicators	TC MPN/100g	FC - <i>E. coli</i> MPN/100g	FS MPN/100g
<i>Rapana venosa</i>	20	0	0

In the present systems of monitoring, the quality of the marine components (water, sediments, vegetal and animal organisms) [21] and the estimation of the presence of the species of pathogenic or potentially pathogenic bacteria that regularly belong to the animal and human intestinal microflora is determined indirectly with the help of fecal pollution indicators, such as, total coliforms - TC, fecal coliforms – FC- *Escherichia coli* and fecal streptococci - FS, whose presence in certain concentrations denotes the existence of a source of impurification of the respective component with residues from humans or animals (fecal contamination) and hence, increased chances of pathogens.

4. Conclusions

The estimation of the *Rapana venosa* quality parameters indicates a high content of biochemical components (proteins) that confirm its high nutritional value and capacity for biologically active substances production with subsequent practical applications (marine biotechnology). The level of contaminants (heavy metals and OCPs) has generally been within the range of normal variation (EC

1881/2006 for heavy metals) with some exceptions, as Cd, dieldrin, p,p'DDE, p,p'DDD and p,p'DDT content.

The microbiological indicators analyzed emphasized an optimal microbiological quality according to the specific in force norms for marine organisms. The obtained results for the analyzed parameters confirm the increased potential of the *Rapana venosa* species for use in bioeconomy and biotechnological purposes.

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