

INFLUENCE OF ENZYME AND PHYTOADDITIVE DIETS ON GROWTH PERFORMANCE AND MAINTENANCE STATUS OF SIBERIAN STURGEON (*ACIPENSER BAERII*, BRANDT, 1869)

E. Mocanu^{1*}, F.M. Dima¹, V. Savin¹, M.D. Popa¹

¹Institute of Research and Development for Aquatic Ecology, Fishing and Aquaculture of Galati, Romania

Abstract

The researches were performed on a population of 4271 specimens of Siberian sturgeon (*Acipenser baerii*), with an average mass of 7.64 ± 0.45 g/specimen, obtained in the pilot station of ICDEAPA Galati. The experiment was performed in the pilot recirculating system belonging to the Research and Development Institute for Aquatic Ecology, Fisheries and Aquaculture in Galați, for a period of 12 weeks (May-June 2021). For the experimental group, an experimental feeding diet was conceived, supplemented with 0.05% digestive enzymes and 0.15% phytoadditives consisting of a mixture of aromatics. The aim of this research is to investigate how diets supplemented with enzymes and phytoadditives influence the growth performance and biochemical composition of meat in the 60 days old Siberian sturgeon species (*Acipenser baerii*), reared in a recirculating system. During the 84 experimental days, the biological material from the experimental group fed with feed to which enzymes and phytoadditives were added, recorded an individual growth increase by 33.03 g/specimen higher, compared to the control group, a specific growth rate (SGR) of 2.13% / day and a feed conversion ratio (FCR) of 0.76 kg. The improved diet with enzymes and phytoadditives, used in the nutrition of Siberian sturgeon, favorably influenced the nutritional quality of the biological material involved in the experiment.

Key words: *Acipenser baerii*, recirculating system, growth parameters, maintenance status

INTRODUCTION

Acipenser baerii is one of the most valuable species of sturgeon in aquaculture, with a high commercial value and meat quality for human consumption, and an important caviar source.

It is a species included in the list of endangered wild population, therefore repopulation programs are of great interest [2]. It easily adapts to captivity, with a fast growth rate, a relatively short reproduction cycle (7-8 years), resistant to diseases and environmental conditions.

The aim of this research is to investigate how diets supplemented with enzymes and phytoadditives influence the growth performance and biochemical composition of

meat, in Siberian sturgeon (*Acipenser baerii*), aged 60 days, reared in a recirculating system.

The identification of new and sustainable nutritious ingredients for feed diets in aquaculture is crucial according to Zarantoniello, M. et al., 2021 [14]. In recent years, several studies on nutritional requirements have been conducted [2], [8]. Unfortunately, there are no nutritional requirements for this valuable species [6].

MATERIAL AND METHOD

The biological material used in the experiment was Siberian sturgeon (*Acipenser baerii*), aged 60 days, reared in the pilot station of ICDEAPA Galati.

The experiment was conducted in the pilot recirculating system belonging to the Research and Development Institute for Aquatic Ecology, Fisheries and Aquaculture in Galați, for a period of 12 weeks (May-July 2021).

*Corresponding author:
icpmocelena@yahoo.com

The manuscript was received: 08.10.2021

Accepted for publication: 13.03.2022

The experiment was conducted using 4271 specimens of Siberian sturgeon (*Acipenser baerii*), with an average mass of 7.64 ± 0.45 g/specimen, divided into two rearing basins: Control basin (BM) with Φ 4.4 m and a volume of 13.7 m³ and an experimental basin (Bexp) with Φ 3.6 m and a volume of 9.0 m³.

Physical and chemical analysis

The physico-chemical and biochemical analysis were performed on fish meat at the beginning and end of the experiment, after 84 days of rearing in an intensive recirculating system, fed with experimental diets enriched with enzymes and phytoadditives.

The composition of the fodders and composition of the fish meat

The fish feed and fish meat samples analysis was conducted using procedures indicated by the standard methods of analysis for fish feed and fish meat.

The moisture was determined by Standard Official Methods of the AOAC (1990).

The total ash was determined by Furnace Incineration described by AOAC Official Method 942.05.

The crude protein content of samples was determined using the Kjeldah method of AOAC 17th edition, 2000, Official Method 928.08 Nitrogen in Meat (Alternative II), which involved protein digestion and distillation, where F (conversion factor), is equivalent to 6.25.

To calculate nitrogen-free extract (NFE), the following formula was used:

$$\text{NFE}\% = 100 - (\text{moisture}\% + \text{crude protein}\% + \text{total fats}\% + \text{crude fibers} + \text{total ash}\%)$$

The total fats were determined using the Soxhlet method, equipped with Gerhardt Brand Multistate Controller, with modified ether extraction methods AOAC 960.39.

Increase protein or retained protein (RP) and increase lipid or retained lipid (RL) were calculated using the following formulas:

$$\text{RP (g protein)} = \text{Final mass / specimen} \times \text{protein} - \text{initial mass / specimen} \times \text{protein}$$

$$\text{RL (g lipid)} = \text{Final mass / specimen} \times \text{lipid} - \text{initial mass / specimen} \times \text{lipid}$$

Assessment of growth performance and feed efficiency

The biometric data collected in this experiment was the total length of the fish (Lt) in cm.

Individual Weight Growth (WG_i, g) and total Weight Growth (WG_t, kg), Food Conversion Ratio (FCR, kg/kg), Specific growth rate (SGR, %/day) were determined as follows:

$$\text{WG}_i = \text{Final weight} - \text{Initial weight (g/fish)}$$

$$\text{WG}_t = \text{Final weight lot} - \text{Initial weight lot (kg/total fish)}$$

$$\text{FCR} = \text{feed fed (kg)} / \text{weight gain (kg)}$$

$$\text{SGR} = 100 \times [(\ln \text{Final fish weight}) - (\ln \text{Initial fish weight})] / \text{experimental days}$$

$$\text{IFC} = \text{kg weight gain} / \text{kg feed}$$

Statistical analysis

All analyses were carried out in triplicate. Statistical analysis was carried out by means of Excel tools. The average values are reported together with standard deviations. The statistical interpretation of the considered data shows a variation within the allowable threshold of $P < 0.05$.

RESULTS AND DISCUSSIONS

Fish feeding experiments

For the control lot, standard feed (FS coded) was used. For the experimental group, an experimental feed diet was conceived, supplemented with 0.05% digestive enzymes (protease, lipase, amylase, gluco-amylase, beta-glucanase, lactase, hemi-cellulase, pectinase, cellulase, alpha-galactosidase maltodextrin) and 0.15% phytoadditives consisting of a mixture of herbs (*Zingiber officinale*, *Mentha piperita*, *Foeniculum vulgare*, *Pimpinella anisum*, in equal proportions).

The feed ration ranged from 0.5% / day of biomass weight to 3% / day of biomass weight, depending on water temperature. The total amount of food calculated for one day was administered every four hours, demonstrating the negative effect of starvation on juvenile sturgeons [1].

The composition of the two types of fodders used in the experiment is presented in Table 1.

Table 1 Chemical composition of the fodders

Fodder sample	Moisture, g%	Proteins, g%	Fats g%	Fiber g%	Ash, g%	NFE** %	Energy value* kcal/100g
FS	6.72	44.95	14.33	3.3	6.9	23.8	415.14
FExp	6.83	44.15	14.0	3.7	7.1	24.22	410.51

The concentrations of nutrients found in the feed administered during the experiment are in accordance with the literature recommendations for Siberian sturgeon 60 days of age [9]. Babaei S., et al., 2017 recommends to improve the quality of sturgeon fillet, high protein diets and lower amount of lipids [2].

Physico-chemical analyses of water

The water's physico-chemical parameters evaluation was done daily, in order to monitor the aquatic environment and the

possibility to intervene in case of deviation of water quality conditions from the allowed limits (table 2).

The recirculating system was designed to ensure all the technological water quality conditions necessary for sturgeon rearing (temperature, dissolved oxygen, pH, organic matter, ammonia, nitrates, nitrite and ammonium), according to Order no. 161/2006 on the classification of surface water quality in order to establish the ecological status of water bodies.

Table 2 Water chemical parameters in the recirculating system, for Siberian sturgeon rearing

Analysed Parameters/ U.M.	Source of sampling	Min	Max	Average \pm SD*	CV** (%)
Dissolved oxygen (mg/l)	Source	7.55	9.23	8.38 \pm 0.49	0.06
	BM	5.55	7.37	6.47 \pm 0.53	0.08
	BExp	6.13	7.15	6.54 \pm 0.33	0.05
Temperature ($^{\circ}$ C)	Source	18.97	26.30	22.83 \pm 2.92	0.13
	BM	19.14	26.39	22.91 \pm 2.91	0.13
	BExp	19.17	26.30	22.89 \pm 2.90	0.13
Saturation (%)	Source	94.71	101.96	97.88 \pm 2.33	0.02
	BM	68.90	98.70	80.09 \pm 10.68	0.13
	BExp	64.13	98.75	79.65 \pm 9.92	0.12
Ph (upH)	Source	8.00	8.71	8.40 \pm 0.23	0.03
	BM	8.02	8.60	8.38 \pm 0.18	0.02
	BExp	8.01	8.72	8.43 \pm 0.22	0.03
Organic matter (mg KMnO ₄ /l)	Source	19.59	58.08	29.65 \pm 13.23	0.45
	BM	14.28	60.07	30.95 \pm 14.17	0.46
	BExp	23.57	58.08	32.08 \pm 10.92	0.34
Nitrates, (NO ₃ ⁻) (mg/l)	Source	1.54	21.3	8.59 \pm 5.75	0.67
	BM	3.3	21.25	9.40 \pm 5.65	0.6
	BExp	2.41	20.98	9.57 \pm 5.97	0.62
Nitrites, (NO ₂ ⁻) (mg/l)	Source	0.003	0.086	0.033 \pm 0.025	0.75
	BM	0.029	0.085	0.050 \pm 0.015	0.306
	BExp	0.025	0.084	0.054 \pm 0.016	0.308
Ammonia (NH ₃) (mg/l)	Source	0.002	0.008	0.005 \pm 0.002	0.4
	BM	0.002	0.009	0.006 \pm 0.002	0.44
	BExp	0.001	0.0087	0.006 \pm 0.003	0.47
Ammonium (NH ₄ ⁺) (mg/l)	Source	0.01	0.12	0.04 \pm 0.033	0.75
	BM	0.01	2.00	0.19 \pm 0.57	2.94
	BExp	0.01	2.00	0.22 \pm 0.56	2.54

* Standard deviation, ** coefficient of variability

During the study, in the two basins there were no deviations regarding the concentrations of the water's physico-chemical parameters outside the optimal interval. According to FAO 2009, this species

can live in temperatures that vary greatly, from only 1 $^{\circ}$ C to 25-26 $^{\circ}$ C, being quite resistant to a low O₂ content but, no biomass accumulates in such conditions [5].

Analysis of the biologic material involved in the experiment

Fish growth parameters

Fish weighing and measuring was performed at the beginning of the experiment, during the weekly experimental period and at the end of it, in order to

monitor the growth dynamics of Siberian sturgeon in the two groups.

At the end of the experiment, following the analysis of biometric data, the average individual mass and total length of sturgeon specimens in the two enclosures had values that can be found in table 3 and 4.

Table 3 Minimum, maximum, average values, standard deviation and coefficient of variability of the growth parameters recorded in Lot C at the end of the experiment

Growth parameter	U.M	Minimum	Maximum	Average ± SD*	CV** (%)
Individual average mass (W)	g/specimen	28,82	47,99	35,57±6,88	0,19
Total length (LT)	cm	17,50	24,90	21,30±2,37	0,11

* standard deviation,
** coefficient of variability

Table 4 Minimum, maximum, average values, standard deviation and coefficient of variability of growth parameters recorded in the Experimental Group at the end of the experiment

Growth parameter	U.M	Minimum	Maximum	Average ± SD*	CV** (%)
Individual average mass (W)	g/specimen	63,12	80,12	69,24±5,96	0,09
Total length (LT)	cm	23,4	30,00	27,28±2,08	0,08

* standard deviation,
** coefficient of variability

The coefficient of variability is below 15%, therefore the sample is homogeneous and the average is representative. At the end of the experimental period, the weight of the Siberian sturgeon fry registered significant differences ($P<0.05\%$) between the control group and the experimental group.

Following the analysis of biometric data, the values of the individual average mass of the Siberian sturgeon specimens had an ascending evolution in both growth basins (Lot C, Lot Exp), the biological material accumulating linear biomass (fig. 1 and 2) [3].

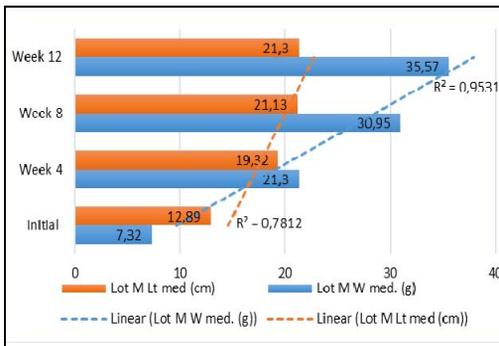


Fig. 1. Dynamics of the average weight (W_{med}) and average length (L_{med}) of the biological material from the control group (Lot C) fed with standard feed

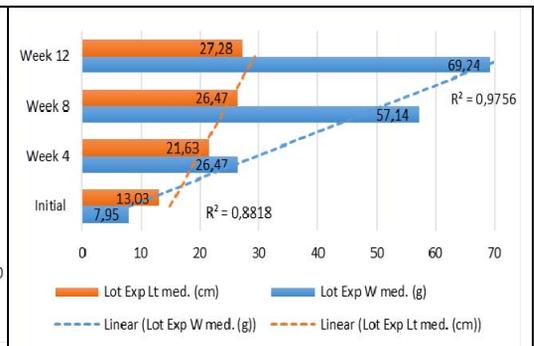


Fig. 2. Dynamics of the average weight (W_{med}) and average length (L_{med}) of the biological material from the experimental group (Lot Exp)

Table 5 exemplifies the bioproductive indicators obtained after experimenting with rearing Siberian sturgeon (*Acipenser baerii*) in a recirculating system and fed diets enriched with enzymes and phytoadditives.

Table 5 Bioproductive indicators obtained when rearing Siberian sturgeon (*Acipenser baerii*) in the recirculating system and fed diets enriched with enzymes and phytoadditives

Growth parameters	UM	Control Lot	Experimental Lot
Initial Parameters			
Number of Specimens	-	2426	1845
Mean individual weight	(g/specimen) mass±SD*	7.32	7.95
Initial Biomass	Kg	17.76	14.67
Density of the initial population	kg/m ³	1.30	1.63
Final Parameters			
Number of Specimens	-	851	1269
Mean individual weight	(g/specimen) mass±SD*	35.58	69.24
Final Biomass	kg	30.28	87.87
Density of the final population	kg/m ³	2.21	9.76
Growth parameters			
Number of days	days	84	84
Weight growth individual (WGi)	g	28.26	61.29
Weight growth total (WGt)	kg	12.52	73.20
Total Shared Food	kg	42.00	55.53
Feed Conversion Rate (FCR)	kg/kg	3.35	0.76
Daily growth rate (DGR)	g/day	0.34	0.73
Specific growth rate (SGR)	%/ day	0.64	2.13
Index of feed conversion (IFC)	Kg/kg	0.30	1.32

The total increase in growth was higher in the Exp lot (73.20 kg), compared to the Control lot (12.52 kg), which indicates the stimulatory effects of enzymes and phytoadditives incorporated in the experimental diet, for the accumulation of biomass.

The same system of subordination of values exists in the calculation of daily growth rate (0.34 g/day in BM, and 0.73 g/day in BExp). This demonstrates that the use of diets with the addition of enzymes and herbs leads to better growth parameters, the fish reaching the market size faster.

The survival of the biological material registered a value of 35.08% in the control basin (PM) where the biological material is fed with standard feed and 68.78% in the experimental basin (BExp) where the biological material is fed with feed with the addition of enzymes and aromatic herbs.

Ryszard K. in 2000 studying the growth rate of *Acipenser baerii* at the age of 30 days, obtained a survival of 47.3%, lower even than the survival of the control group in the present experiment [11].

Fodder with additional enzymes and phytoadditives was more efficiently recovered and assimilated, the sturgeons in

the experimental group accumulating a higher growth rate, with a specific consumption of 0.76 kg feed / kg increase and a feed conversion index of 1.32 kg increase / kg feed consumed. The results are similar to those obtained by Zoltan T., et al., 2011 [13].

The values regarding the specific growth rate (% / day) in the control lot are comparable to those obtained by Ferit R., et al., 2003 [7], who experimented with different rations of food for Siberian sturgeon and lower than the values obtained by Bocioc, 2011 [3].

The analysis of technological indicators highlighted a favourable influence of digestive enzymes and phytoadditives on growth parameters, a similar conclusion stated by Stroe et al., 2013, studying the effect of vitamin C [4].

The biochemical composition of the material involved in the experiment

The effect of interaction of diets with enzymes and phytoadditives on the biochemical components of the Siberian sturgeon species can be appreciated from the study of the data revealed in table 6.

Table 6 Biochemical composition of Siberian sturgeon reared (12 weeks) in recirculating system and fed with diets enriched with enzymes and phytoadditives

	Fish sample	Moisture, g%	Proteins, g%	Fats, g%	Ash, g%	RP, g proteins	RL, g lipids	M/P**
		Media±SD*						
Initial	Lot C	92.51	6.21	0.5	0.78	-	-	14.90
	Lot Exp	92.74	5.98	0.62	0.66	-	-	15.51
Final	Lot C	76.13±1.65	14.48±1.15	8.13±0.45	1.26±0.55	469.6	285.5	5.26
	Lot Exp	72.75±2.01	14.68±1.12	11.28±0.52	1.29±0.45	968.9	776.1	4.96

* standard deviation

** M/P= Moisture, (g%)/ Proteins, (g%)

The protein content does not differ significantly between the two batches, which suggests an almost similar evolution of the biological material in retaining protein. However, a variation of the protein content over time for the two batches is observed, from an average of 6.10 g%, in the beginning, to a maximum of 14.68 ± 1.12 g% (experimental lot) and 14, 48 ± 1.15 g% (Lot C) characteristic at the end of experiment. The values of protein in the two groups at the end of experiment are similar to values identified by Soleiman H., et al., 2019, with the mention that the specimens analysed by him had a double individual mass compared to the specimens in the present experiment [12].

Analysing the data referring to the lipid content, significant differences are observed both at the beginning of experiment and the end of experiment, but also between the two groups at the end of experiment, the differences being significant ($p > 0.05$).

Increase protein or retained protein (RP) and increase lipid or retained lipid (RL) are indicators, in which biochemical constituents are involved and through which the quality of the biological material involved in the experiment is assessed.

The highest amount of protein and lipids accumulated was quantified for the specimens from the Experimental Group, fed diets in which enzymes and phytoadditives were incorporated. The values are comparable to those identified by Pelic M., et al., 2019, analysing the fatty acid composition of different sturgeon species [10].

The ratio between the percentage of water and protein in the muscle tissue of Siberian sturgeon (U / P) characterizes the biological material in terms of nutritional value and maintenance status.

At the beginning of experiment, the biological material was characterized by an average U / P ratio equal to 14.9 (lot C) and 15.51 (lot Exp). During the experiment, this ratio decreased in the two groups, reaching 5.26 in lot C and 4.96 in the experimental lot.

CONCLUSIONS

Fodder enriched with enzymes and phytoadditives used in the feeding diets of the Siberian sturgeon species (*Acipenser baerii*) do not change the quality of the aquatic environment, parameters being appropriate to the growth and development of the species under study;

The body mass of the Siberian sturgeon specimens registered, during the 12 weeks, an increase of 386% in the control lot and 771% in the experimental lot, fed with fodder with additional enzymes and phytoadditives.

The daily growth rate is higher in the experimental lot compared to the control lot, which shows that the fodder enriched with enzymes and phytoadditives causes faster body mass increases.

The survival of the biological material was at 35% in the control lot and 69% in the experimental lot fed with feed with the addition of enzymes and phytoadditives, all values being within the normal survival limits for Siberian sturgeon.

Fodder with additional enzymes and phytoadditives was more efficiently recovered and assimilated, the sturgeons in the experimental lot achieving a higher growth rate, with a specific consumption of 0.76 kg feed / kg increase and a feed conversion index of 1.32 kg increase / kg feed consumed.

Enhanced diets with enzymes and phytoadditives, used for the nutrition of the

Siberian sturgeon species have favourably influenced the nutritional quality of the biological material involved in the experiment.

The fish in the two lots (Lot C, Lot Exp) at the end of experiment presented a better state of maintenance, highlighted by the U / P ratio, compared to the start of the experiment.

REFERENCES

- [1] Ashouri, G., Hoseinifar S.H., M. T. Mozanzadeh, A. Mani, A. Khosravi, O. Carnevali, Compensatory growth, plasma hormones and metabolites in juvenile Siberian sturgeon (*Acipenser baerii*, Brandt 1869) subjected to fasting and re-feeding, 2019, *Aquac. Nutr.* 26, p 400–409.
- [2] Babaei S., A. A.-Kenari, M. Hedayati & M. Ali Y.-Sadati, Growth response, body composition, plasmametabolites, digestive and antioxidant enzymes activities of Siberian sturgeon (*Acipenser baerii*, Brandt, 1869) fed different dietary protein and carbohydrate: lipid ratio, *Aquaculture Research*, 2017, 48, p 2642–2654.
- [3] Bocioc Elena, 2011, Research on the use of probiotics in industrial aquaculture in recirculating systems (Doctoral Thesis), „Dunarea de Jos” University of Galați Romania.
- [4] Dicu (Stroe) M. D., Cristea V., Mirea (Ciortan) C., Plăcintă S., Petrea M. Ș., Coadă M. T., Effects of Different Levels of Dietary Vitamins C on Growth Performance of Stellate Sturgeon (*Acipenser stellatus*, Pallas, 1771), *Scientific Papers: Animal Science and Biotechnologies*, 2013, 46(2)
- [5] FAO. 2009, *Acipenser baerii*., Cultured aquatic species fact sheets, http://www.fao.org/fishery/docs/CDrom/aquaculture/I1129m/file/en/en_acipenser.htm
- [6] Falahatkar, Bahram, Nutritional Requirements of the Siberian Sturgeon: An Updated Synthesis, 2018, 10.1007/978-3-319-61664-3 11.
- [7] Ferit R., Gülten K., L, Mevlüt K., Growth Performance and Food Conversion Ratio of Siberian Sturgeon (*Acipenser baerii* Brandt) at Different Daily Feeding Rates, *Turk J Vet Anim Sci* 27, 2003, p 1085-1090.
- [8] Liu, H. Wu X., Zhao W., Xue M., Guo L., Zheng Y., Yu Y. Nutrients apparent digestibility coefficients of selected protein sources for juvenile Siberian sturgeon (*Acipenser baerii* Brandt), compared by two chromic oxide analyses methods, 2009, *Aquac. Nutr.* 15, 650–656.
- [9] Médale F., G. Corraze, S.J. Kaushik, Nutrition of farmed Siberian sturgeon. A review of our current knowledge, Proceedings of international symposium on sturgeon, 6–11 September 1993, Publishing 1995, Moscow-Kostroma-Moscow, Russia, VNIRO.
- [10] Pelic M., Vidakovic S. K., M. Z. Balos, N Popov, N Novakov, M Cirkovic, D Ljubojevic P., Fatty acid composition of *Acipenseridae* – sturgeon fish, *Earth and Environmental Science* 333, 2019, 012092.
- [11] Ryszard K., Mirosław S., Bożena S., Postembryonic Development, Survival and Growth Rate of Siberian Sturgeon (*Acipenser baerii* Brandt) Larvae, *Arch. Pol. Fish.* vol. 8 fasc. 2000, p. 193-204.
- [12] Soleiman H., Najmeh S., Hadi J., Mehdi N.F., Karim M., Growth performance, antioxidant and immune status of Siberian sturgeon (*Acipenser baerii*) fed diets containing green tea extract and oxidized fish oil, *J Appl Ichthyol.* 2019; p 1–10.
- [13] Zoltan T. Szelei, Marian Bura, and Szidonia C. Szücs, Research regarding bioproductive indicators achieved by the Siberian sturgeon (*Acipenser baerii*) juvenile in recirculated system, *AACL Bioflux*, 2011, Volume 4, Issue 4, p 530-535.
- [14] Zarantonello, M., Randazzo B., Nozzi, V., Truzzi, C., E. Giorgini, G. Cardinaletti, L. Freddi, S. Ratti, F. Girolametti, A. Osimani, V. Notarstefano, V. Milanović, P. Riolo, N. Isidoro, F. Tulli, G. Gioacchini & I. Olivotto, Physiological responses of Siberian sturgeon (*Acipenser baerii*) juveniles fed on full-fat insect-based diet in an aquaponic system, *Scientific Reports*, 2021, Articol no. 1057.