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Columella

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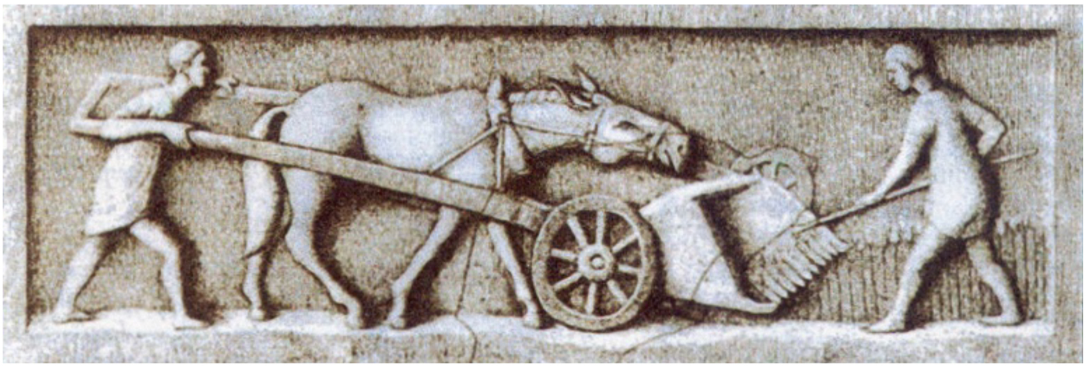

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Columella

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Table of contents

MAGYAR AGRÁR- ÉS ÉLETTUDOMÁNYI EGYETEM

Columella

Volume 11, Number 2, 2024

Columella
Journal of Agricultural and Environmental Sciences

Examination of earthworm abundance; biomass and correlations of soil organic matter in an irrigated (with river and catfish effluent water) and mulched agroforestry system

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Abstract: The aim of our study was to evaluate a complex agroforestry system with the intercropping of aerobic rice and the utilization of reclaimed water for sustainability and climate change adaptation. The foreseeable positive outcomes of the intercropping system could be higher yields for the arable crops, additional woody products and indirectly favourable microclimate, water conservation, increased biodiversity and wind damage reduction. In this study, a special rice-energy willow/poplar agroforestry system was used to analyze the effects of reused water irrigation and mulching on soil salinity, earthworm biomass and abundance, soil organic matter (SOM) content and weed coverage in treerow-dependent habitats. After a three-year irrigation period, we investigated the woody line (WL), the buffer zone (BZ) and the crop line (CL) habitats. In our small-scale (0.3 ha) experiment, aerobic rice production took place between poplar and willow rows. The rice cultivar and woody lines were irrigated with different doses of river water and effluent water from an intensive catfish farm. The effect of irrigation and organic mulching on earthworm abundance, biomass and species composition was also investigated. In conclusion, this study demonstrated the beneficial effects of straw mulching on reducing soil salinity and improving soil health indicators. Based on our results, significantly greater earthworm abundance (274 ind m^2) and earthworm biomass (54.0 g m^2) values were measured in WL than in BZ or CL habitats. There was no significant difference in weed coverage between the CL (0.61%) and BZ (1.91%), but weeds were significantly denser on the WL (12.3%). These findings emphasize the potential advantages of reused water irrigation, mulching, and agroforestry systems in promoting soil health and effective weed control. Further research is warranted to explore the long-term effects and scalability of these practices. Agroforestry systems have the potential to enhance soil biodiversity and microbial activity, which play crucial roles in nutrient cycling and soil health. By studying the effects of agroforestry practices on soil biology, we can provide valuable insights into the mechanisms underlying soil quality enhancement in these systems.
Keywords: irrigation, aerobic rice, organic mulching, earthworm, agroforestry system

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Introduction

The world population is projected to increase to 9 billion by 2050 (Shiferaw et al., 2011). Such growth increases the global need for the production of more food, fiber and fuel. However, future climate extremes are projected to threaten the goal of meeting the ever-increasing population's food, fibre and fuel demands. Therefore, growers must adapt to future environmental changes by practicing, climate-smart agriculture (Zizinga et al., 2022).

Agroforestry practices globally distribute numerous ecological benefits (for instance, climate change mitigation and climate adaptation), and land preservation (e.g., erosion control, enhanced soil fertility). Additionally, methods of combating modern agricultural practices complications, such as loss of moisture quality due to overuse of synthetic nutrient supplements (e.g., N and P) and pesticides. However, the incorporation of trees in agriculture may cause competition for available water with planted crops. In southern Australia, studies revealed a positive correlation between crop yield reduction and moisture competition alley systems (Hall et al., 2002; Unkovich et al., 2003). Kowalchuk and Jong (1995) discovered that competition for moisture is the main factor impacting intercropped grain quantity of spring wheat and shelterbelts in Western Saskatchewan, particularly in dry periods.

Fish effluent water rich in organic matter resulting from excretion and fish feed can be used as a supplement to curb water competition between crops and plants in agroforestry systems (AFS). Additionally, the soil and plants' physical, chemical, and biological properties will benefit from its high nutrient status (Kolozsvári et al., 2021).

Furthermore, organic mulch improves the physical, chemical and biological properties of the soil. The two common materials utilized as ground cover mulch that may decrease soil moisture loss are stubble and plastic. The two further may improve water use efficiency while regulating the soil temperature to aid crop development (Cook et al., 2006; Vincent-Caboud et al., 2019; Yu et al., 2018). Similar to Paulis (2007), an increase in soil organic carbon (SOC) in the top 200 mm was observed owing to the addition of tree stubble residue (Youkhana & Idol, 2009) predominantly in more stable fractions.

Earthworms are biological soil tillers and they are subjected to changes in the physical and chemical properties of the soil (Bakti et al., 2017). Several authors mentioned that the impact of tree plantations on earthworm abundance can be achieved by changing the chemical and/or physical soil properties (moisture regime, pH, SOM levels, biomass additions) (Bakti et al., 2022; Kun et al., 2023; Tian et al., 2000; Gonzalez & Zou, 1999). Earthworm abundance prefers minimal disturbance and more conventional agroforestry management methods (Römbke et al., 2009). Studies by Vaupel et al. (2023) observed higher earthworm density and biomass in agroforestry system tree rows systems compared to monoculture cropland.

AFS effectively improves organic matter addition, consequently improving physical properties, including soil structure, and pores (Kumar et al., 2020). Moreover, AFS as a universal system, can decrease nitrate leaching (Kay et al., 2018), and enhance biodiversity, while sustaining agri-productivity (Pardon et al., 2018; Swieter et al., 2019) and food safety of wheat and barley (Beule et al., 2019). In this context, incorporating the production of food, fibre and fuel in AFS play a significant role in carbon sequestration. For example, Rizvi et al. (2011) observed 50t/ha carbon was sequestered under 7 years of rotation in poplar AFS. Moreover, due to the direct relationship involving soil organic carbon (SOC) and soil organic matter (SOM), an increase in SOC storage by AFS will increase SOM (Gyuricza et al., 2018).

Managing high-saline agricultural irrigation water is particularly important during droughts, when plants are already growing in stressful environments. Integrated soil and water management can help minimize adverse effects. In a water conservation

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context, high-saline water can be used in drought-affected areas where freshwater is limited, especially with systems such as drip irrigation, where salt accumulation can be minimized (Hussain et al., 2020). Cavalcante et al. (2022) showed that during drought periods, supplementary irrigation with saline water can reduce plants' water stress levels and increase the physical productivity of water. However, the higher salt content in the soil hinders the growth of plants, and among other things inhibits the uptake of nutrients, and causes an ion imbalance, thereby reducing the crop yield. Some waters with high salt content may contain trace elements such as magnesium, potassium or sodium, which can improve the nutrient supply of the soil in small amounts. The salt content may also improve the stress response capacity of some plants (induced stress mechanisms). In addition, irrigation with saline water for several years can increase the content of exchangeable sodium in the soil, which affects the physicochemical properties of the soil, such as soil bulk density, conductivity, and soil organic carbon content decrease, which also has a negative effect on rice production (Liu et al., 2019; Sun et al., 2019). The benefits only occur when the salinity level is precisely controlled and adapted to the needs of salt-tolerant crops and applied under controlled conditions. The leaching of salts and the maintenance of soil structure are of paramount importance for the long-term sustainability of this type of irrigation. Although high-saline irrigation water is not an ideal solution, under the right conditions and with careful management, it can provide certain benefits for crop production.

Our study aimed to evaluate a complex agroforestry system with the intercropping of aerobic rice and the utilization of reclaimed water for sustainability and climate change adaptation. The examination of the ecological role of the agroforestry system from the point of view of the biological properties of the soil (earthworm numbers and biomass) and the weed coverage of the different ground covers. The diversity of the project, which results from the complexity of the research's goals covering each slice of the biosphere, hydrosphere, lithosphere and atmosphere, requires that the topic has dealt with consortium level (the Hungarian University of Agriculture and Life Sciences and the University of Sopron).

Materials and Methods

2.1. Site Description and Climatic Condition

The experiment was set up at the agroforestry research site (0.3 ha) of the Hungarian University of Agriculture and Life Sciences (MATE), Institute of Environmental Sciences (IES), Research Center for Irrigation and Water Management (ÖVKI) in Szarvas, Hungary (Fig 1).

Hungary has a temperate continental climate; the specific area of the experimental site is described as a warm and dry climate region. Meteorological data from the three-year experiment (2019, 2020, and 2021) were collected at an automatic weather station (Agromet Solar, Boreas Ltd., Érd, Hungary) 1600 m from the experimental site. The precipitation was 516.4 mm, 611.4 mm and 433.9 mm in the consecutive experimental years. There was also a significant difference between the years in the annual average mean temperature values, the warmest year was 2019 with 13.8 °C, the second (12.1 °C) and third year (11.6 °C) were cooler.

At the experimental plantation, two types of trees were planted in 2013, one is the candidate variety willow (*Salix alba* L."82" Naperti clone) of the University of Sopron Forest Research Institute, Department of Plantation Forestry and *Populus* × *euramericana* cv. *Kopecky* poplar. This clone is an artificial hybrid created by Ferenc Kopecky at the Sárvár Experimental Station of the University of Sopron Forestry Science Institute

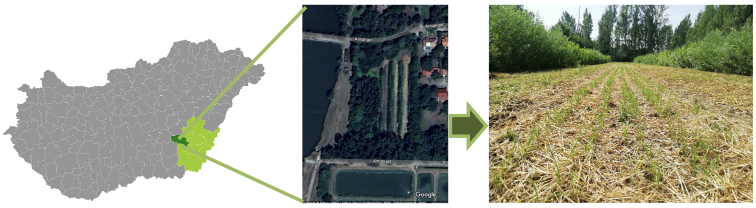

Figure 1: Localization map of the experimental site Szarvas, Hungary.

(B. Tóth, 2006). The geographical origin of the willow clone included in the experiment is Eastern Hungary. The use of the breed cultivation is for energy purposes, the variety was included in the national variety protection list in 2013 under the variety name "Naperti".

The area of the experimental site is 0.3 ha, there are 18 plots of the same size in the area: 136.5 m^2, one plot is 13 m long and 10.5 m wide. There are 8 rows of trees in each plot. The row spacing is 2.5 m and the stem spacing is 0.5 m. All sites are irrigated (a non-irrigated plot was not found in this experiment) from two types of irrigation water sources. In 2018, the plantation was transformed into an agroforestry system. Now, there are six rows of trees in the area (two single rows at the borders, two double rows in the middle) and 3 fields for intercrop cultivation (10-meter width) (Fig 1). The soil type belongs to Vertisols (IUSS Working Group WRB, 2022) with clay texture, 8.3 pH, 5.4% total calcium carbonate, and 2.4% total organic carbon.

2.1. Site Description and Climatic Condition

For irrigation, two types of water were used in the experiment. 1) The "Körös River" (natural surface water, K) originated from the the Bikazug Oxbow Lake of the Körös River, which is suggested for irrigation due to its appropriate quality for irrigation purposes (Kun et al., 2017). 2) The "Effluent water" (E) originating from an intensive African catfish farm. The fish farm used water from deep-groundwater wells to fill their fish farming pools, and then to treat the used water the farm established a constructed wetland (Kolozsvári et al., 2021). The combined constructed wetland system consists of two stabilization ponds and two wetlands (F. Tóth et al., 2016). The water from the first stabilization pond was used to irrigate this experimental site. The effluent is characterized by a high concentration of sodium and bicarbonates due to the geothermal origin of the water and relatively high nutrient content because of the remaining material after fish production. This wastewater contains large amounts of debris such as fish faeces, and organic materials (F. Tóth et al., 2020). According to the irrigation water classification of USDA (Gregory, 1982; Richards, 1954), the wastewater belongs to the C3-S2 group with high salinity and medium sodium hazard.

2.3. Treatments and mulching

The experimental site (80 $\times$ 10 m) was divided into two parts, on one we used mulch (mulched plots) (400 m^2), and on the other (400 m^2) there was no ground cover (unmulched plots). Both sites were irrigated with wastewater using micro-sprinkler irrigation. In each experimental year (2019, 2020, and 2021) the amount of irrigation water was 150 mm $year^{-1}$. Irrigation was done five times each year with 30 mm. The results of the salt content and other values were published earlier (Kun et al., 2023), here we only focus on the effect of mulching on earthworms and soil health.

Winter wheat straw (0.25 kg m^{-2}) was applied for mulching (71% soil cover). The mass of the ground cover material was cal-

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culated based on the equation defined in Stefanovits' research (Stefanovits, Filep, & Füleky, 2010).

An energy plantation was installed in the area in 2013, it was transformed and we established the agroforestry system in 2019. Irrigation has been done with agricultural water from fish farming and natural "Körös River" with single and double doses since 2013. In the case of the double-dose treatment, salt accumulation began to appear in the soil due to the negative effect of high-salt irrigation water from fish farming. In order to prevent this and to improve the soil, we used liming and straw mulch covering in the experimental area. The mulch was applied every other year with an amount of 2.5 t/ha, because the straw breaks down slowly.

2.4. Sampling and analyses

Composite soil samples were taken from 0-15 and 15-30 cm depths. Regarding the applied treatments, soil samples were taken from mulched (straw) and unmulched areas in four repetitions. The soil carbon content was determined by CNS analyser (Elementor, Vario MAX Cube), and the soil organic matter was calculated by multiplying the result by 1.74 (Zhang et al., 2022).

Disturbed bulk soil samples were collected before the first irrigation of the three-year experiment (2019 spring) and then each autumn after the irrigations in four repetitions from mulched and un-mulched areas. The sampled soil depths were 0-30 and 30-60 cm, however, we did not differentiate between soil depths during the statistical analysis.

The specific electrical conductivity (EC) of the soil was measured from saturated soil paste (according to Hungarian Standard MSZ-08-0206-2:1978). The available nitrogen content of the soil was characterized by the sum of the nitrite and nitrate contents of the soil ($KCl{-}NO_2^- + NO_3^-{-}N$). Nitrite and nitrate were extracted with potassium chloride and the concentration was measured using FIA spectrophotometer (according to Hungarian Standard MSZ 20135:1999). The sodium (AL-Na) concentration was measured after ammonium-lactate extraction by AAS flame photometry (according to Hungarian Standard MSZ 20135:1999).

Exchangeable cations (K, Na, Ca, and Mg) were extracted with barium-chloride + triethanolamine and their concentrations were measured using atomic adsorption spectrophotometer (AAS) (according to Hungarian Standard MSZ-08-0214-2:1978).

$$ESP(exchangeablesodiumpercantage, \%) = \frac{Na}{(Na+K+Ca+Mg)} \times 100 \quad (1)$$

where, Na^+, K and Mg_2^+, concentrations are expressed in milliequivalents per 100 g of soil (Gregory, 1982; Richards, 1954).

Concerning the earthworm sampling, the sampled habitats were the following: a) *crop line* (CL): in the middle of the interrow section, where soil disturbance, sowing and crop production occurred; b) *buffer zone* (BZ): beside the crop line, which did not receive any soil disturbance or crop production; c) *woody line* (WL): area under the tree line, where no soil disturbance or agricultural cropping occurred. The samples were collected in four repetitions, by hand-sorting method (ISO 23611-1:2018, 2006). Soil blocks (25 × 25 × 25 cm) were excavated onto a plastic sheet, then searched carefully for earthworms. The earthworms were killed in 70% ethanol, transported to the laboratory and fixed in 4% formalin. The number of earthworms (pc m^{-2}), and biomass (g m^{-2}) were determined. The earthworm sampling was carried out in April, 2022.

Weed composition was surveyed by

recording weed cover expressed in the percentage of the total area of 1 m^2 micro-plots on the mulched areas three times in 2022, in spring (April 29), in summer (August 17) and in autumn (October 14). Data collection included all non-crop plants with four replications of all habitats. For each panel, for each distance measured from the edge and for each time, 4 pieces (Zalai et al., 2012), on a 1×1 meter square, were assessed by direct coverage percentage estimation of the coverage of the present weeds by species (Németh & Sárfalvi, 1998). Thus, 4 sample plots were selected per transect, in the tree rows, in the immediate vicinity of the tree rows (buffer zone) and in the cultivated areas. So, the sample plots were selected at the "0" meter (in the undisturbed strip), at the 1 meter directly at the edge of the undisturbed strip at a distance of 0-100 cm from it), at the 2nd meter (at a distance of 100-200 cm from the cultivation edge) and at the 4th meter (at a distance of 300-400 cm from the cultivation edge). The results obtained were averaged per field and per distance from the edge, so the averaged results are presented.

2.5. Statistical analyses

Statistical analyses were done in IBM SPSS statistics 27 software. To model the change of soil parameters affected by mulching (factorial variable; yes or no) and irrigation between 2019-2021 (survey period; factorial variable; 2019 Spring, 2019 Autumn, 2020 Autumn or 2021 Autumn) variables were tested by Multi-Way Analysis of Variance (Multi-Way ANOVA) in the case of soil parameters. Additionally, the sole effect of irrigation between 2019 and 2021 was tested by One-Way Analysis of Variance (ANOVA) separated by mulched and un-mulched conditions, as well. In significant cases, explanatory variables were tested by a two-sample T-test for the mulching variable and a Tukey comparison the for habitat variable.

To model data collection in 2022, both mulching and habitat (factorial variable; WL, BZ or CL) variables were tested by Multi-Way Analysis of Variance (Multi-Way MANOVA) in the case of earthworm abundance, earthworm biomass and soil organic matter. Habitat was tested by Analysis of Variance (ANOVA) in the case of total weed coverage. In significant cases, explanatory variables were tested by a two-sample T-test for mulching and by a Tukey comparison for the survey period variable.

Results

3.1. Earthworm abundance and biomass

Based on MANOVA, (Fig. 2/B; Tab. 1), significantly greater earthworm abundance taken from WL (264 pc m^{-2}) was found as compared to CL (84 pc m^{-2}) under mulched treatments. The unmulched treatments showed a similar tendency, WL (284 pc m^{-2}) was significantly greater than CL (60 pc m^{-2}).

As for earthworm biomass (Fig. 2/A; Tab. 1), significantly greater values were found in WL (55.4 g m^{-2}) and BZ (55.1 g m^{-2}) as compared to CL (24.6 g m^{-2}) habitat in mulched treatments. Whereas, in unmulched treatments, significantly greater biomass values were obtained in WL (52.6 g m^{-2}) as compared to BZ (26.2 g m^{-2}) and CL (14.1 g m^{-2}) habitats.

3.2. Soil Organic Matter Content and Total Weed Coverage

Regarding the soil organic matter (SOM) content, the effect of mulching was statistically significant (Fig. 2/C; Tab. 1). Significantly greater SOM content was detected under the mulched treatment in WL (4.7%) as compared to CL (4.5%) habitat. Concerning the unmulched treatments, the following decreasing order was obtained: 4.0 (WL), 3.9 (BZ), and 3.8% (CL), with significant differences only between WL and CL.

As for the total weed coverage (Fig. 2/D; Tab. 1), the mulched plots were examined

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Table 1: Effect of mulching and habitat on earthworm abundance (pc m^{-2}), earthworm biomass (g m^{-2}), soil organic matter (%) and total weed coverage (%) in an agroforestry experiment (Szarvas, Hungary, 2022).

Earthworm abundance						
Variable	df	MANOVA		Tukey comparison		
		F	p-Value	Group	Avg Value (pc m^{-2})	Sign. Class
Mulching	1	0.051	ns	-		
Habitat	2	10.825	0.001	WL	274.00	b
				BZ	186.00	b
				CL	72.00	a
Earthworm biomass						
Variable	df	MANOVA		Tukey comparison		
		F	p-Value	Group	Avg Value (g m^{-2})	Sign. Class
Mulching	1	1.849	ns	-		
Habitat	2	3.573	0.049	WL	54.00	b
				BZ	40.01	ab
				CL	19.39	a
Soil organic matter						
Variable	df	MANOVA		Tukey comparison/T-test		
		F	p-Value	Group	Avg Value (%)	Sign. Class
Mulching	1	155.451	<0.001	mulched	4.597	b
				un-mulched	3.883	a
Habitat	2	3.879	0.050	WL	4.336	b
				BZ	4.244	ab
				CL	4.140	a
Total weed coverage						
Variable	df	MANOVA		Tukey comparison		
		F	p-Value	Group	Avg Value (%)	Sign. Class
Habitat	2	6.184	0.020	WL	12.303	b
				BZ	1.913	a
				CL	0.608	a

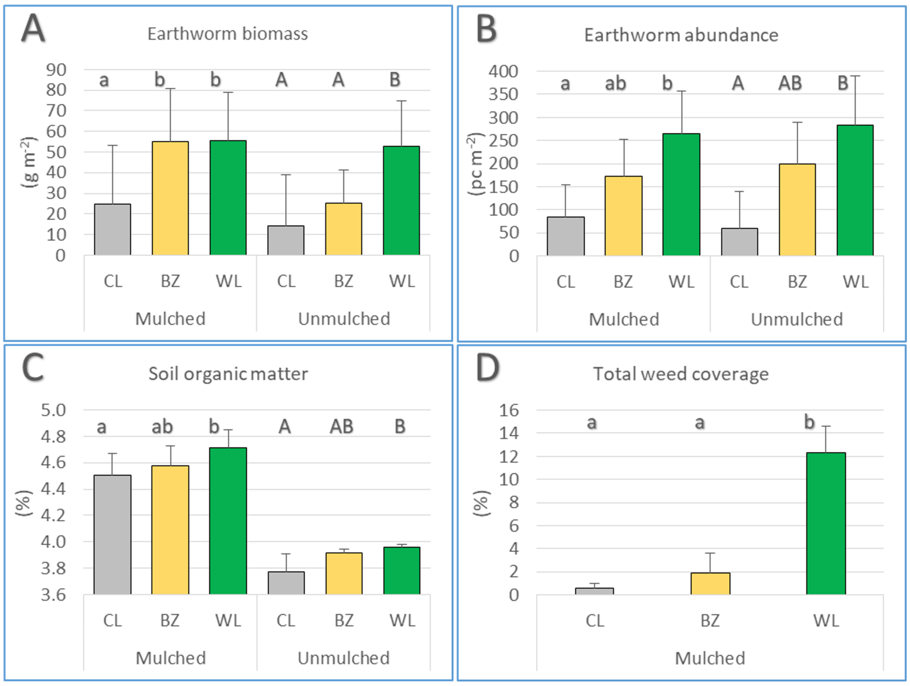

Figure 2: Effect of mulching and habitat on earthworm abundance (pc m^{-2}) (B), earthworm biomass (g m^{-2}) (A), soil organic matter (%) (C) and total weed coverage (%) (D) in an agroforestry experiment with different habitats: crop line (CL), buffer zone (BZ) and woody line (WL). (Szarvas, Hungary, 2022).

only. We found the greatest weed coverage values in the case of the WL (12.3%) as compared to BZ (1.9%), and CL (0.6%) locations. WL was significantly greater than BZ and CL. Based on the results obtained, we found that there was a significant difference in vegetation depending on the different cultivation. The largest part of the cover of the WL was made up of Geophyta species, such as *Rubus caesius, Convolvulus arvensis, Elymus repens* and *Calystegia sepium*. We experienced a smaller degree of weeding in the CL between the WL, and both Geophyta and Therophyta species appeared in these areas. The weeds most typical of the CL were the following: *Convolvulus arvensis, Taraxacum officinale, Elymus repens, Cirsium arvense, Veronica spp., Calystegia sepium, Setaria glauca, Digitaria sanguinalis* and *Ballota nigra*.

Applying mulch is a sustainable, economical and environmentally friendly way to support crop production, while offering numerous benefits in maintaining soil and plant health (El-Beltagi et al., 2022). Mulch reduces evaporation from the soil surface, thus preserving soil moisture for a longer period of time, protecting the soil from the direct effects of large amounts of precipitation, preventing soil compaction and surface crusting. Soil cover protects the soil from erosion caused by wind and water, slows down water runoff, thus reducing soil erosion. The mulch layer covering the soil suppresses the germination and growth of weeds, as it blocks their access to light (Clare et al., 2015). Organic mulches (e.g. straw, compost, tree bark) form humus when they decompose, which improves the structure and fertility of the soil. They promote the activity of beneficial microorganisms, fungi and earthworms living in the soil. These organisms help the nutrient cycle and health of the soil (Akter & Oue, 2018).

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Discussion

Several authors stated that organic mulching materials increase water retention capacity, enhance soil health and fertility, and protect the soils against environmental extremes (e.g. erosion) (Chen et al., 2014; Prosdocimi et al., 2016) and also provide habitat, carbon input and food source for soil organisms (Jodaugienė et al., 2010). However, in our study, mulching treatment did not result in any significant growth of earthworm abundance. Despite other studies (Radics et al., 2022; Simon et al., 2022; Tian et al., 1997), they found greater earthworm abundance due to mulching materials compared to unmulched areas.

Therefore, we pooled the earthworm abundance values of the mulched and unmulched treatments together. As a result, we obtained significantly greater earthworm abundance in WL (274 pc m^{-2}) and BZ (186 pc m^{-2}) as compared to CL (72 pc m^{-2}) (Figure 4/B). This might be due to low soil disturbance in BZ and WL habitats and high natural input of raw organic matter (plant leaves, roots, etc.) Our result was in line with Norgrove et al. (2011), who stated that greater earthworm abundance was found under undisturbed timber plantation in tropical agrisilvicultural systems compared to cropped plots.

As for earthworm biomass (Fig. 2/A; Tab. 1), almost similar values were gained for BZ (54.9 g m^{-2}) and WL (55.4 g m^{-2}) in mulched treatments. It means that these individuals could gain greater biomass, even with lower abundance (BZ: 172 pc m^{-2}) values, suggesting that mulching provided a great/better weight increase as a food source. Since mulching did not show any significant differences, we pooled these values together, and found that earthworm biomass in WL differed significantly from CL (Tab. 1).

Regarding the soil organic matter (SOM) content, the mulched treatments provided significantly higher values (Tab. 1), suggesting the positive effect of organic mulching on soils. The WL habitat probably provided greater raw organic residue and, thus, resulted in significantly greater SOM content compared to the CL habitat. The intensive disturbance in CL resulted in lower SOM content.

The difference in weed composition was highly collated to the intensity of tillage and coverage of trees. There was no significant difference in weed coverage between the tilled CL (0.61%) and the non-tilled BZ (1.91%) but weeds were significantly denser on WL (12.3%) than both not-covered habitats (CL and BZ).

Conclusion

In conclusion, this study highlights the positive impact of straw mulching in reducing soil salinity and enhancing soil health indicators. It was found that the woody line (WL) and buffer zone (BZ) habitats exhibited higher earthworm abundance compared to crop line (CL) habitats. The use of mulching also contributed to increased earthworm biomass and higher soil organic matter content. Weed coverage, influenced by both tillage intensity and tree cover, showed higher weed density in the woody line habitat. These results underscore the advantages of reused water irrigation, mulching, and agroforestry systems in promoting soil health and controlling weeds. This research provides valuable insights into sustainable soil management practices and supports the integration of agroforestry systems with reused water irrigation. Further investigation into the long-term effects and scalability of these practices is essential. Agroforestry systems have the potential to improve soil biodiversity and microbial activity, which are key drivers of nutrient cycling and soil health. Studying the effects of these practices on soil biology will enhance our understanding of how agroforestry can contribute to soil quality improvement.

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Influence of temperature conditions on the mobile fish hatchery efficency

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Abstract: The demand for angling native fish species and their conservation value are steadily increasing nowadays. Mobile fish hatchery developed by the Department of Aquaculture at MATE AKI allows for immediate on-site propagation after capturing the broodstock. Several experiments have demonstrated the usefulness of the system, but there are certain aspects changing dynamically such as temperature conditions due to the small size of system and environmental exposure, which require further investigation. In this study, applicability of mobile hatchery was compared with a closed recirculation hatchery system. Model species used in the experiments was the chub (*Squalius cephalus* L.). Results show the difference in daily heat input and average temperature between differently positioned Zuger jars in mobile hatchery; however, it does not affect hatching rate, larval mortality rate or body length of freshly hatched larvae. The closed recirculation system had a higher proportion of deformed larvae than the mobile hatchery; in addition, the hatching rate was positive in all Zuger jars. Based on statistical analysis, no statistically significant difference was detected in body length between Zuger. Body length of freshly hatched larvae in the closed recirculation system was significantly smaller than in case of groups incubated in Zuger jars 1 ($P < 0.05$) and 2 ($P < 0.05$). Results show that water temperature of mobile hatchery is affected by the temperature outside, but hatchery units provide optimal temperature for developing eggs even at low air temperatures.

Keywords: mobile hatchery, temperature, embryonic development, incubation

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Introduction

The management of natural waters in Hungary has changed significantly over the past 15 years. While commercial fishing in natural waters used to be common, Law CII on Fisheries, which came into force on 1 September 2013, links the primary use of waters to angling and the development of angling tourism (Ferincz & Staszny, 2020). At the same time, it can be observed that natural fish populations have been reduced in many areas due to overexploitation, loss of spawning grounds and water pollution (Keijzer et al., 2024; Sallai & Juhász, 2020). It has led to the need to rebuild and continuously replenish fish stocks in many areas of our natural waters (Daupagne et al., 2021; Weiperth et al., 2021). However, restocking is a complex and often costly process (Mickiewicz & Wołos, 2012). On the one hand, it has been proven that the population of large fish of a size that can be caught by anglers can be associated with serious risks, for instance, inter- and intraspecific competition (Simonović et al., 2014), reduction of the genetic stock of populations (Hargrove et al., 2022), and loss of natural spawning populations. On the other hand, the positive impact of these programmes on usable yields is also questionable (Simonović et al., 2014). Therefore, research that had been conducted on this topic so far shows that the best means to conserve natural populations is the restoration of natural habitats and spawning grounds (Manfrin et al., 2019). If it is not possible for economic or other reasons, efforts should be made to reduce the potential risks of stocking. It is required that the widest possible range of broodstock from natural habitats be used and the youngest possible age class be released (Araki, 2008). The aim of stocking programmes is to ensure that artificial propagation only helps the species through critical points that cause the greatest losses and which, if eliminated, do not cause significant genetic selection (Klütsch et al., 2019). Such critical points causing high losses may be the lack of spawning habitat (Gao et al., 2016), the inadequate incubation environment of eggs (Crane & Farrell, 2013) or the presence of a starter food (Meira et al., 2022; Skaramuca et al., 1994).

Mobile fish hatchery developed by the Department of Aquaculture at MATE AKI can offer a solution to all these problems. This device allows for immediate on-site propagation of broodstock after capture in natural waters in a way that is as humane as possible for the broodstock. Rapid on-site propagation can maximise the number of broodstock used, and this in turn reduces the risk of genetic narrowing caused by stocking. A mobile hatchery provides opportunity to incubate eggs in situ and to release the fry quickly and efficiently while eliminating mortality due to transport (Csorbai & Urbányi, 2020; Hekli, 2022; Ketut Suwetja et al., 2017).

Usability of the new system has already been tested by the developers in a number of field trials, which have focused on the system's load-bearing capacity. In order to determine the practical usability, it was important to determine how many eggs and larvae the mobile hatchery could incubate. In the first experiment, the system was fed with 350 g of 54% protein fry feed to model the nitrogen load of fry rearing. Nitrogen content of the fish feed was 30 g per day. Total ammonium nitrogen (TAN) was measured at 0.3 ± 0.2 mg/L, while nitrite was measured at 0.1 ± 0.05 mg/L (Csorbai & Urbányi, 2020). These values provide sufficient conditions for the incubation of most fish species (Tilak et al., 2002). Following theoretical investigations, two model fish species (African catfish *Clarias gariepinus* Burchell 1822, and common carp *Cyprinus carpio* L.) and three target fish species (ide *Leucisus idus* L., chub *Squalius cephalus* L., tench *Tinca tinca*) have shown that the hatchery can be used in practice (Csorbai, 2021).

There are, however, certain dynamically changing aspects, such as temperature parameters, which are key to long-term applicability due to the small size of the system and its exposure to the environment, and which merit further investigation. Water temperature is the most important environmental parameter that influences egg development and embryonic development period (Avakul & Jutagate, 2015), hatching success, growth and survival of newly hatched larvae (Kucharczyk et al., 1997; Roessig et al., 2004). Previous experiments have shown that chub larvae are more sensitive to extreme conditions during incubation (Kupren et al., 2008), but also tolerate increasing temperature variation and temperature fluctuations (i.e. incubation at non-constant temperatures) under optimal conditions. Although the incubation time is shorter in both increasing and fluctuating temperature incubations, no difference in larval deformation rate and hatching percentage can be detected (Kupren et al., 2011). Literature also shows that temperature plays a crucial role in the embryonic development of fish and its rate; therefore, the exploration of this parameter is crucial when developing a breeding technique for a fish species.

For the investigation of the influence of temperature conditions on the mobile fish hatch-

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ery efficency the chub as a model species was chosen, as this species is becoming increasingly popular among anglers, but its natural populations have been decreasing in many areas, so that its establishment has become necessary (Weiperth et al., 2021) and the reference data to accurately evaluate the results were available (Krejszeff et al., 2008; Nagy et al., 2023).

Materials and Methods

The broodstock was collected in the Ipolytölgyes section of river Ipoly using an electric fishing machine (Samus 725M). Research licence no. HaGF/154/2021 was at disposal to use the electric fishing machine. The temperature of the river was 14 °C. The 12 female and 31 male chubs were captured and transported to Gödöllő and placed into the mobile fish hatchery which was setted to the MATE AKI Aquaculture Department. Ovopel AUV®was used to induce ovulation and stimulate spermiation. The priming dose (0.1 Ovopel/1 kg of broodstock) was followed by the final dose (0.9 Ovopel/1 kg of broodstock) after 24 hours. After mixing the eggs and milt, system water was used for fertilization. After that, Woynárovich's solution (40 g table salt and 30 g urea dissolved in 10 L system water) was used to prevent eggs from sticking together. The final removal of stickiness was done with tannin solution (5 g tannin, 10 L system water). The batches of eggs were mixed and then divided into four equal parts to obtain 4×620 mL of svollen egg batches. One batch was placed in a 7-L Zuger jar in the recirculation system of the Department of Aquaculture at MATE AKI and the remaining three batches of eggs were placed in three different Zuger jars of the mobile fish hatchery numbered serially. Number 1 was the Zuger jar closest to the entrance, number 2 was the middle one and number 3 was the Zuger jar furthest from the door. The mobile fish hatchery system is based on a basin with a 1.2 m^3 capacity, which is suitable for temporary housing of broodstock and short-term rearing of the fry. Mechanical filtration of water in the system is provided by a sponge filter downstream of the basin. Water from the basin flows through three filter sponges, each of which is 50 mm thick, then reaches the UV filter where four 25 W UV tubes ensure the elimination of unwanted bacteria, viruses and fungi. The pump (Grundfos Alpha 1) is located after the UV filter and transfers system water to a 0.288 m^3 biological filter tank containing 0.144 m^3 of biomedia fill. The K1 biomedia has a useful surface area of 836 m^2/m^3. Water flows by gravity from the biofiltration reactor to the Zuger jars or rearing units. Five 8 L Zuger jars (SDK Sp. z o. o.) can be installed for the hatchery at the same time, which can be easily exchanged during the rearing phase for five larval rearing jars of 100 L (Csorbai & Urbányi, 2020). Temperature stability of the system is ensured by two 250 W aquarium heaters (Eheim Jäger 250) controlled by a thermostatic socket (Sygonix TX3; hysteresis 0.5 °C). Mechanical filtration of the Zuger jar in the control unit was also provided by a sponge filter. It contained a 40-W UV filter and a 150-L biological filter. Temperature and dissolved oxygen content in the system were continuously monitored (probe type WTW FDO-IQ) and the measured data were fed into a computer. When the dissolved oxygen level reached the critical lower limit, the system notified the operators on telephone. Moreover, when the system reached the lower or upper limit of the set desired temperature, the computer reacted by controlling the system accordingly and in case the water temperature reached the lower limit, heating was provided by a 500 W (Eheim Jäger 500) aquarium heater. In cases when the water temperature during the measurement was higher than the desired upper limit, cooling was provided automatically by a 375 W Aqua Medic Titan Chiller 1500.

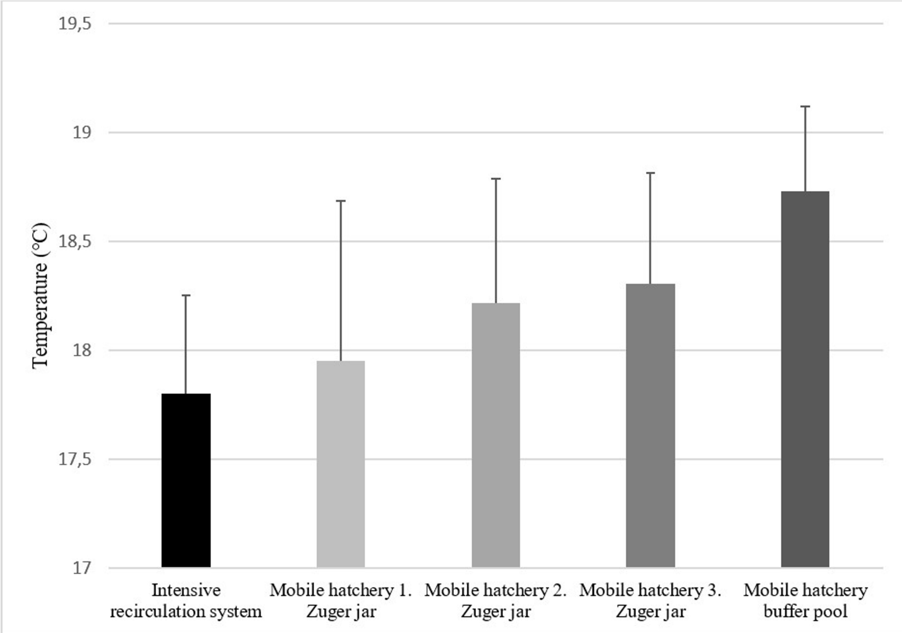

Figure 1: Evolution of average water temperatures during incubation.

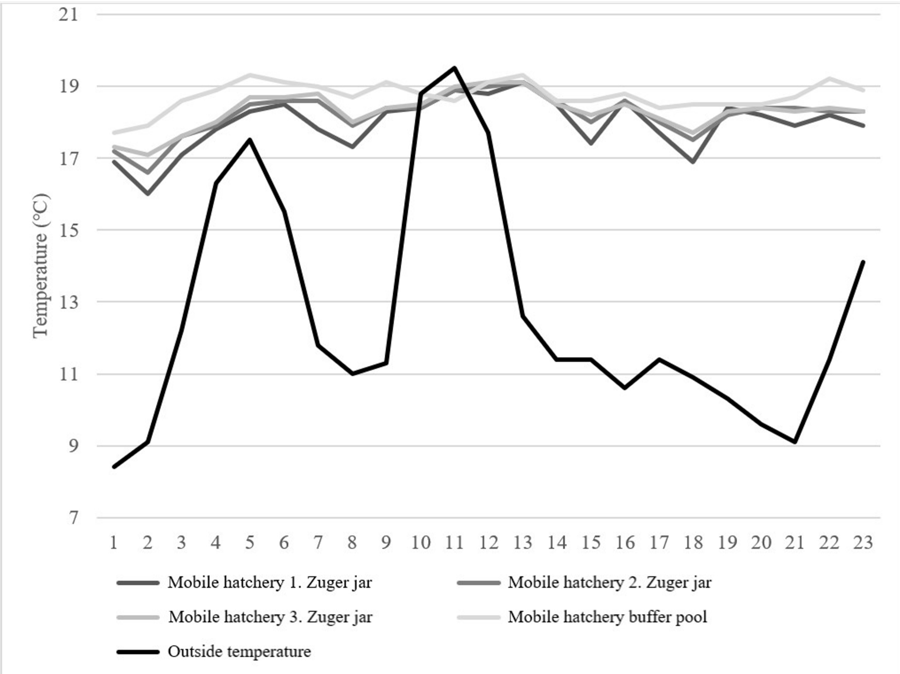

Figure 2: Comparison of the water temperature values measured in the mobile fish hatchery and the outside ambient temperature.

In the indoor recirculation system, the replenishment water volume was constantly 18 L/hour, which means a daily water change rate of 20%. In the mobile fish hatchery, water was changed once a day, also at a rate of 20%. In both cases, the make-up water was mains water at the temperature of 16 °C. Water change was not only to wash

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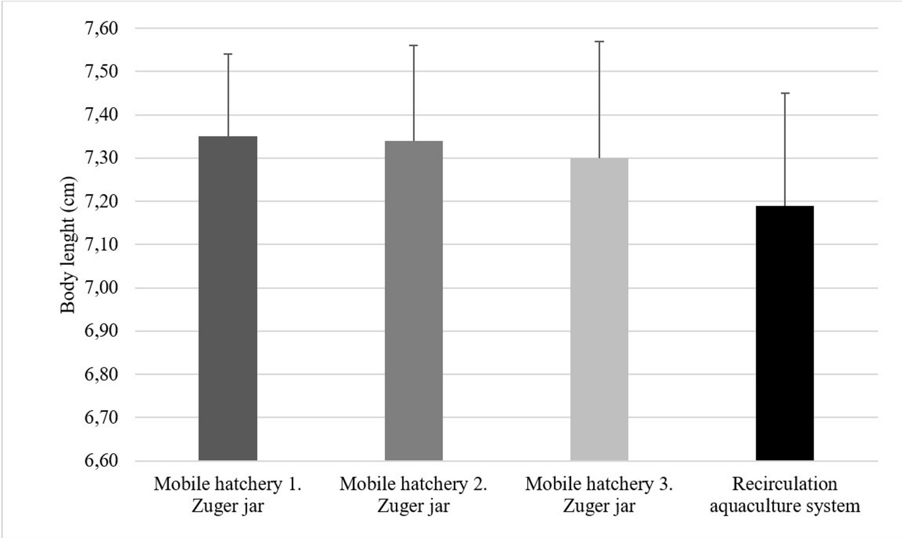

Figure 3: Body length (mean±SD) of freshly hatched larvae for groups incubated in different units. Groups with different letters proved to be significantly differend by ANOVA ($P < 0.05$).

out unwanted nitrogen forms but also to reduce temperature, so it was carried out in the early afternoon. The desired temperature during incubation was 18 °C in both systems. Temperature measurements in the mobile fish incubator and in the indoor recirculation system were carried out every 4 hours at 0:00, 4:00, 8:00, 12:00, 16:00 and 20:00 throughout the incubation period. Measurements were conducted in the four experimental Zuger jars and in the brood fish holding tank of mobile fish hatchery located in the middle of the water body. A Hach HQ 2200 manual measuring device was used for the measurements. For comparison with external air temperature parameters, data of the Hungarian Meteorological Service in Aszód (Station No. 44214), the nearest station to Gödöllő were used.

Hatching rate was determined at the moment of hatching from 150 to 200 eggs using a Leica EZ4E microscope. At the time of hatching, 2×25 eggs were taken from each Zuger jar and after, the properly positioned fish were photographed with a Leica M205 FA microscope and a DFC7000 T camera. The 50 larvae per each Zuger jar in the images were measured using ImageJ (Developed by Wayne Rasband, version 1.52). The fish photographed were reviewed one by one and deformations were noted.

Results of body length measurements were compared using one-point analysis of variance, with 95% significance level for each difference in all cases and GraphPad Prism 4.0 Statistical software was used for statistical analysis.

Results

Average water temperature in the Zuger jar connected to the closed recirculation system was 17.8 ± 0.45 °C during the incubation of eggs. The mean temperature measured and standard deviation of water temperatures measured were the lowest in the Zuger jar mentioned. The average temperature of Zuger jar number 1 in mobile fish hatchery was 18.0 ± 0.73 °C and the same value of number 2 was 18.2 ± 0.57 °C, while in case of number 3, it was 18.3 ± 0.51 °C. In all cases, the standard deviation values of water temperatures measured in the Zuger jars in mobile fish incubator were higher than the standard deviation of incubator parameters measured in the closed recirculation system. It was observed that the average temperature of Zuger jar no. 1 in mobile fish hatchery is the lowest and its standard deviation is the highest; in addition, the average temperature increases while the standard deviation decreases as the distance from the door increases (Fig. 1). The average water temperature of the 1.2 m^3 fish holding pool in mobile fish hatchery was found to be the warmest, namely 18.7 °C during the experiment. Standard deviation of the measured results was the lowest in this case (±0.39 °C). The fact that the temperature of pool is more stable than regarding any of the incubators is probably due to the fact that the large volume of water has a much smaller specific surface area and is, therefore, less exposed to environmental influences than small (8-L) Zuger jars.

Air temperature data of the Hungarian Meteorological Service were also compared with the data of mobile fish hatchery. The diagram shows that although water temperature change follows the trend of air temperature change, the daily heat input remains adequate even in colder temperatures and water temperature did not drop below the critical 16 °C regarding any of the Zuger jars during the incubation period (Fig. 2).

The calculated hatching rate in the mobile fish hatchery was very similar in all three Zuger jars. The Zuger jar located closest to the door had a hatching rate of 67.1%, the Zuger jar in the middle had 67.5%, and the hatchery jar furthest from the door had 66.2%. A lower hatching rate of 63.3% was counted in the closed recirculation system. The average total body length of hatching larvae incubated in Zuger jar 1 of mobile hatchery was 7.35 ± 0.19 mm. The same value for Zuger jar 2 was 7.34 ± 0.22 mm and for Zuger jar 3 was 7.30 ± 0.27 mm. Nevertheless, the average body length of the closed recirculation hatched larvae was 7.19 ± 0.26 mm. Based on statistical evaluation, no statistically verifiable difference in hatching larval body length was detected between Zuger jars 1, 2 and 3. However, the body length of freshly hatched larvae in closed recirculation system was significantly smaller than in case of groups incubated in Zuger jars 1 ($P < 0.05$) and 2 ($P < 0.05$) (Fig. 3).

Differences were also observed in larval deformation: the proportion of malformed and weakly hatched larvae was 12% in Zuger jar 1, 10% in Zuger jar 2 and 14% in Zuger jar 3 of mobile hatchery. For larvae hatched in the closed recirculation system, this rate was 22%.

Discussion

SIn the experiment, temperature conditions in the mobile hatchery and their effect could be determined during the egg incubation period. Results show that water temperature in mobile hatchery is affected by the outside temperature, yet hatchery units provide adequate temperatures for developing eggs even at low air temperatures (Kupren et al., 2008). It was observed that there was a difference in daily heat input and average temperature between the differently located Zuger jars, but it did not affect hatching rate t or body length of newly hatched larvae. In the experiment, mobile hatchery was compared with a closed recirculation system having a lower daily heat input. There was no significant difference in hatching rate, but the percentage of deformed larvae was lower in mobile hatchery. There was a statistically verifiable ($P < 0.05$) difference in mean body length of larvae hatched in the closed recirculation system and the mobile hatchery. The difference was significantly higher only for Zuger jars 1 and 2 with higher heat input and the Zuger jar attached to the closed system. Besides all, results were found to be simi-

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lar to those of Kupren et al. (2011). It was also found that the chub is well tolerant to changes in temperature conditions during the incubation period. While similarly to the Polish research group, no difference was found in hatching rate; however, the body length of freshly hatched larvae was statistically verifiably longer in Zuger jars with higher temperature fluctuations. Larger body size at hatching also affects initial feeding and growth, which may have a positive effect on individuals throughout their development. Based on the results of this research and those cited in literature review, it can be concluded that mobile hatcheries can offer a sustainable option for enhancing natural fish stocks and their temperature conditions are as expected.

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MAGYAR AGRÁR- ÉS ÉLETTUDOMÁNYI EGYETEM

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Editorial note

This is the final issue and volume number under my editorial leadership. As of December 31, 2024, I have resigned as editor-in-chief of this journal and the rector of the Hungarian University of Agriculture and Life Sciences has accepted my resignation.

Several factors prompted this decision. The first 2024 issue of Columella published only three articles, the second issue published only two, and an increase in the number of articles is not anticipated in the current publishing environment. I believe the journal is not appealing to potential authors, anymore. There are numerous competing scientific journals available, the peer review process has become expedited, the scientific prestige (quartile ranking) of competing journals is higher, and researchers are incentivized to publish their results in highly ranked journals.

However, it would be unjust to solely attribute the journal's challenges to external factors. Three years ago, during a personal meeting with the rector, we agreed that Columella should enhance its international profile, be made searchable in Scopus, and increase the publication frequency to four issues per year. These objectives have not been realized in the past three years, and I bear full responsibility for this shortcoming. The protracted editorial and review process exacerbated the already challenging circumstances.

The past three and a half years have been an extraordinary journey for our journal. In 2022 and 2023, Columella became the official journal of the annual International Scientific Conference on Water held each year in Szarvas, Hungary. Thanks to this decision, the second issue of 2022 published 17 peer-reviewed articles, a number that seems incredible from today's perspective.

I would like to express my sincere gratitude to everyone who assisted the editorial work of Columella. This includes the two co-editors of the journal, Mária Kovács-Weber and Miklós Heltai, guest editors Mihály Jancsó, Zoltán Futó, Károly Bodnár, Ágnes Kun, Noémi Valkovszki and the countless reviewers who have given their time and energy to improve the quality of manuscripts submitted to the journal. I would also like to express my sincere gratitude to the auhtors who have honored the journal by sending their manuscripts to be published in Columella. Finally, I would like to thank all readers who found the science in our articles worthy of their attention.

Sincerely yours,

Ákos Horváth
editor-in-chief

Source of the graphics

Front cover:
Gallo-Roman harvesting machine, called Vallus. Source: U. Troitzsch - W. Weber (1987): Die Technik : Von den Anfangen bis zur Gegenwart

Rear cover:
Portrait of Columella, in Jean de Tournes, Insignium aliquot virorum icones. Lugduni: Apud Ioan. Tornaesium 1559. Centre d'Études Supérieures de la Renaissance - Tours

HORVÁTH Ákos, editor-in-chief

DSc (agricultural sciences), Chair of the Department of Aquaculture at the Institute of Aquaculture and Environment Protection of the Hungarian University of Agriculture and Life Sciences, member of the Committee on Animal Sciences of the Hungarian Academy of Sciences. Professional fields: aquaculture, fish reproduction, biology of fish gametes, cryopreservation, transplantation, population genetics of fish as well as aquatic toxicology.

Lucius Junius Moderatus Columella

(AD 4 – 70) is the most important writer on agriculture of the Roman empire. His De Re Rustica in twelve volumes has been completely preserved and forms an important source on agriculture. This book was translated to many languages and used as a basic work in agricultural education until the end of the 19th Century.