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A rainbow trout feeding behavior assessment (*Oncorhynchus mykiss*) when introducing a complex feed additive increasing digestibility

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Abstract. The potential to enhance the nutritional appeal of standard formulated feeds for rainbow trout by incorporating an additive with attractant and/or stimulant properties is demonstrated in the study. To assess the feeding behavior of the fish the evaluation algorithm was developed using a specialized setup with two feeding points, along with automatic video recording of the outcomes. The components within the feed additive (89% rapeseed oil, 4% hemp oil, 5% lycopene, and 2% cinnamyl aldehyde) not only aid in improving its palatability but can also have a positive influence on the fatty acid composition of the feeds. It was revealed that the feed additive (at quantities of 0.3% and 0.5%) significantly augmented the attractiveness of the feeds resulting in an increased number of fish congregating in the feeding area and an elevated consumption rate. The computation of the index of relative attractiveness allowed to identify a noteworthy disparity between the experimental and control feeds. The data obtained from the study can recommend the investigated comprehensive feed additive for enhancing the digestibility of standard fish feeds.

Keywords: feeding behavior, attractant, feed additives, rainbow trout, aquaculture

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Научная статья

Оценка кормового поведения радужной форели (*Oncorhynchus mykiss*) при внесении в корма комплексной кормовой добавки с включением компонентов, повышающих его поедаемость

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Аннотация. Показана возможность улучшения пищевой привлекательности кормов стандартной рецептуры для радужной форели путем внесения добавки, обладающей аттрактантными и/или инсайтантными свойствами. Для оценки кормового поведения рыб был разработан алгоритм оценки с использованием специализированного стенда с двумя точками кормления и автоматической фиксацией результатов. Входящие в состав кормовой добавки компоненты (89 % рапсового масла, 4 % конопляного масла, 5 % ликопина и 2 % коричного альдегида) не только способствуют улучшению его поедаемости, но могут положительно влиять на жирнокислотный состав кормов. В результате исследования было установлено, что внесение кормовой добавки (в количестве 0,3 и 0,5 %) достоверно повышает привлекательность кормов, увеличивая количество рыб в кормовой зоне и скорость поедаемости. Расчет индекса относительной привлекательности позволил установить достоверное различие между опытными и контрольными кормами. Полученные данные позволяют рекомендовать разработанную комплексную добавку для улучшения поедаемости кормов, приготовленных по стандартной рецептуре.

Ключевые слова: кормовое поведение, аттрактант, поедаемость, кормовые добавки, форель

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Introduction

Feeding is a critical technological process in aquaculture. The efficiency of fish feeding in industrial aquaculture is determined by numerous factors, each exerting different effects on fish growth processes. When designing industrial feed formulations special attention is given to the nutrient composition, pellet size, and the incorporation of mineral and vitamin premixes [1]. Balanced feed formulations should yield high fish growth rates with efficient feed utilization [2]. However, the feed productivity values reported by manufacturers are not always attainable in industrial aquaculture. The primary reasons for this discrepancy include non-compliance with recommended feed pellet sizes, feed losses due to deviations from feeding technologies [3], and diminished feed attractiveness due to the presence of various anti-nutritional. For instance, the incorporation of microbial protein and plant-based components in feeds can significantly diminish the nutritional appeal of feeds for specific fish species [4].

One of the most rapidly developing areas of feed production for aquatic organisms cultivated in controlled environments is the incorporation of additional components into feeds to enhance their productive characteristics [5, 6]. The most prevalent components include probiotic and trace element additives [7], alongside biologically active components [8]. Substances that enhance the attractiveness and palatability of feeds are considered promising for supplementation [2, 9]. This category of additives encompasses extracts of plant origin [10], diverse salts of bile acids [11], free amino acids [12], saturated and unsaturated fatty acids, as well as other low molecular weight (water-soluble) organic compounds and certain protein components [13]. Protein hydrolysates and amino acids are likely recognized by chemosensory cells as they could serve as stimuli for specific recognition [9]. The impact of these components on feed intake and physiological parameters of fish species such as *Oreochromis niloticus*, *Cyprinus carpio*, *Ctenopharyngodon idella*, and *Ictalurus punctatus* was investigated.

Vegetable oils can serve as safe and effective substitutes for fish oil, a primary source of essential fatty acids in fishmeal. Numerous studies have concentrated on incorporating rapeseed oil and soybean oil into feeds, demonstrating their effectiveness for omnivorous and herbivorous aquaculture species [14], while sometimes hindering feed intake in predatory fish species [15]. It has been observed that introducing various attractants and stimulants to aquaculture feeds significantly enhances their nutritional appeal due to synergistic effects [2].

Various behavioral assessment methods are extensively employed to study the palatability of feed additives. Different automated behavioral techniques are widely utilized both in laboratory settings and for evaluating the feeding behavior of fish in industrial aquaculture. The adoption of these methods facilitates the monitoring of fish condition, alterations in size and weight, feed intake, and shifts in behavior. Among the primary approaches being developed are continuous video monitoring, interactive automatic feeding systems that regulate the quantity of applied feed, the utilization of infrared sensors, and several other systems (see Li et al. [16] for a comprehensive review). Many of these systems have been adapted for fish following their successful application in domestic and farm animals.

The pet food industry has long employed methods to assess feeding behavior in order to ascertain the appeal of different foods. Among the most widely utilized methods is the selective choice from various feed formulations (multiple bowl method) [17]. Similar techniques have been previously employed for laboratory testing of feeds in aquaculture, enabling the examination of alterations in feeding behavior and the assessment of the attractiveness of feeds and feed additives [16].

The objective of this study was to assess the developed feed additive based on plant extracts for its feeding attractiveness and to test the application of the feeding behavior evaluation method on rainbow trout (*Oncorhynchus mykiss*).

Materials and methods

Animal maintenance. The research was conducted at the infrastructural resource of the Unique Scientific Unit (USU) with registration number 3662433, known as the “Complex of Advanced Aquaculture and Hydroecology Technologies”, situated within the laboratory named “Aquaculture Technologies”. This laboratory falls under the Research Complex of Applied Aquaculture and Fish Health (Aquaculture Center), which operates within the Faculty of Biotechnology and Fisheries at the Moscow State University of Technologies and Management (FCU). The research was conducted in alignment with ongoing research programs under the umbrella of the Research and Education Center titled “Innovative Solutions in the Agroindustrial Complex”. This center is part of the world-class scientific and educational establishment situated in the Belgorod region, dedicated to the study of feeding and behavioral reactions of freshwater aquaculture subjects.

The rainbow trout from the “Adlerskaya” breed group (*Oncorhynchus mykiss*), sourced from the fish farm “Trout Paradise” LLC in the Belgorod region, underwent preliminary acclimatization within the experimental Recirculating Aquaculture System (RAS) at the aforementioned USU facilit

Experimental feed preparation. Experimental feeds were produced using the feed section of the faculty through the cold pelleting method utilizing a two-roll pelletizer named “ZLSP-120”. Concurrently, the pelleted feed mixtures underwent drying in a dehydrator until they reached a moisture content of 13%. These feeds were prepared right before the experiment and subsequently stored in a refrigerated chamber maintained at 4 ºC. The primary feed was formulated following the standard trout feed formulation and featured a pellet size of 4 mm.

The components of the utilized feed additive included rapeseed oil, lycopene extract, hemp oil, and cinnamyl aldehyde. These phytonutrients exhibit the potential to positively impact several physiological parameters of the growing subjects and possess attractant properties. Building upon literature insights and preliminary experiments (unpublished data), the following composition for the feed additive was proposed: 89% rapeseed oil, 4% hemp oil, 5% lycopene, and 2% cinnamyl aldehyde.

Rapeseed oil presents itself as a promising feed component due to its balanced fatty acid composition and cost-effectiveness. Hemp oil, comprising a blend of fatty acids and other biologically active compounds [18], has shown potential for enhancing several physiological parameters while augmenting feed attractiveness [19]. Lycopene, a promising antioxidant with diverse biological activities, finds applications in both the food industry [20] and aquaculture [8].

The incorporation of lycopene at the reported concentrations (15 and 25 mg) is not expected to adversely affect the production and physiological parameters of rainbow trout; instead, it can imbue the feed with a more vibrant hue and enhance its palatability. Cinnamaldehyde, an aromatic aldehyde compound found in cinnamon, showcases antibacterial, antifibrogenic, and antioxidant properties [21]. Its allure for certain farm animals and fish has been noted by various researchers [22].

The experimental feeds were enriched with the feed additive at two concentrations: 0.3 and 0.5 g/kg feed (EF3 and EF5 abbreviation).

Experimental setup for evaluating feeding behavior. To assess the feeding behavior of rainbow trout when with formulated feed compositions, a model experimental setup was constructed. This configuration entailed a 6-meter-long, 0.5-meter-wide, and 0.5-meter-high pool, with a volume of 1.5 tons and a 10% water exchange rate (Fig. 1).

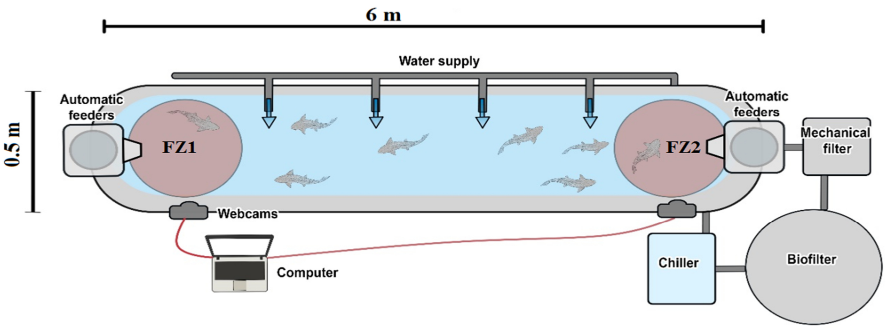

Fig. 1. Schematic diagram of a model experimental setup for evaluating feeding behavior of rainbow trout, including basic elements: FZ1, FZ2 – feeding zones

The water temperature in the fish tanks was maintained at 16.3 ± 0.7 °C, regulated by a Hailea HC-220BH chiller (China, Guangdong Hailea Group Co., Ltd.). The pH level was kept stable at 7.8 ± 0.2. The oxygen concentration remained above 8 mg/L, averaging 8.4 ± 0.4 mg/L, due to the implementation of an Armed 7F-10L oxygen concentrator (Russia, Armed LLC).

The RAS was outfitted with a comprehensive filtration setup incorporating a 500-liter biofilter along with a mechanical filter. Throughout the course of the experiment, monitoring of the hydrochemical parameters of the water was conducted to ensure compliance with the established standards for cultivating rainbow trout. Over the entire experimental period, the levels of ammonium (0.1 ± 0.4 mg/l), nitrite (0.15 ± 0.05 mg/l), and nitrate (12.75 ± 3.5 mg/l) remained well below the specified limits. Water supply to the experimental tank was orchestrated by drawing water from various sources to establish a uniform rheophilic flow across the entire length of the experimental tank. This approach aimed to yield dependable results for behavioral evaluations within the feeding zones. For the experimental setup, a group of 10 rainbow trout specimens, each measuring 22.3 ± 0.1 cm in length and weighing 158.2 ± 11.6 g, was introduced into the pool. Synchronized automatic feeders were strategically positioned at the opposing ends of the pool. In one of these feeders, control food was dispensed, while in the second feeder, experimental food containing the feed additive was provided. The feeding rate amounted to 36.34 g per day per fish biomass (18.16 g for the control and 18.16 g for the experimental group).

The method devised for assessing feed attractiveness is built upon the foundation of prior research by various scholars [23]. In these studies, the authors delineated the key stages of fish feeding behavior and elucidated the underlying principles for behavior regulation. To capture the eating activities in the designated feeding zones (FZ1 and FZ2), webcams linked to a computer were positioned alongside the feeders. This setup facilitated the recording of food consumption. By employing video recording to monitor fish behavior within the experimental environment, the process of data capture is automated and the subsequent data analysis is streamlined [24]. Depictions of the experimental setup are provided in Figure 1.

Experimental design. During the feeding sessions (conducted twice daily), the subsequent parameters were meticulously recorded in accordance with the methodology outlined by Geurden et al. [15]: fish presence near the feeders and within the field of view of the webcams. These aspects were noted separately for each of the two designated Feeding Zones (FZ1 and FZ2). Specifically, the data collected encompassed the count of fish and the duration required to completely consume the feed portion, termed as the Complete Eating Duration (CED). Two distinct sets of experiments were undertaken, each involving varying quantities of the feed additive, denoted as EF3 and EF5. Furthermore, the Relative Feed Eating Rate (RFR), determined by calculating the ratio of the fish count within the feeding area to the time taken for feed consumption, was also computed. For the purpose of differentiation, Control 1 (Con1) pertained to the assessment involving the feed formulation EF3, while Control 2 (Con2) was linked to the feed formulation EF5.

The Relative Preference Coefficient (RP) was used as an integral indicator of food attractiveness. This indicator reflects the number of fish in a feeding zone relative to the average number of fish in the same zone during the adaptation period. This coefficient was calculated separately for the testing period and the validation period.

The experiment extended over a span of 17 days for each of the experimental concentrations. The experimental design adhered to the subsequent schedule: *Days 1 to 3 (adaptation period):* A period allocated for adapting to the feeding regimen. As established by the research of other scholars, fish typically require up to 7 days to adjust to the prevailing conditions and feeding schedule [15]. Feeding transpired twice daily, specifically at 11 : 00 AM and 6 : 00 PM. Preliminary assessments of feeding behavior revealed that the maximum duration for consuming a feed dose was 200 seconds. Guided by this observation, the primary experiment involved a 5-minute video recording interval to monitor feeding behavior. *4-10 days (experimental period).* This phase encompassed the testing period, constituting the core of the experiment. Throughout each day, the fish were randomly distributed within the tank, displaying minor movements linked to foraging activities and the selection of an optimal rheophilic regime. Upon the activation of the automated feeders, the following parameters were meticulously recorded: the fish count in the distinct feeding zones (FZ1, FZ2), the rate of complete food consumption, and the behavioral responses of rainbow trout concerning the feed. These recordings were performed over a 5-minute timeframe. 11-17 days – validation period. This period was designated as the validation period. During this phase of experimental feeding, the positions of the experimental and control feed groups were interchanged. This strategic switch aimed to prevent the formation of enduring conditioned reflexes among the fish. The primary objective of this testing interval was to affirm the consistent presence of stable attractant properties within the evaluated feed additive.

Statistical analysis. Statistical analysis was performed using R software (v3.5.2)/RStudio and GraphPad Prism version 8.0 software (GraphPad, San Diego, CA, USA). Data in the tables are presented as mean ± SD, p value < 0.05 was taken as statistically significant. Data were tested for normality and homogeneity of variance using the Shapiro-Wilk test and Bartlett's test, respectively. If the data conformed to normal distribution, the unpaired t-test with Welch's correction was used when comparing two samples, and the Mann–Whitney test was used for non-normal samples.

Results

Feeding behavior in adaptation period. During the adaptation period, when provided with the control food, the fish displayed random positioning within the feeding zones during feeding times, without exhibiting any distinct preference. At other instances, the trout engaged in exploratory behavior, manifesting as movement throughout the tank area with minor pauses occurring within the feeding zones.

Over the span of 1-3 days, a total of 6 measurements were conducted, corresponding to the number of feeding instances. These measurements yielded numerical data. The count of fish within the feeding areas during feeding moments exhibited no notable variation between the recorded results and throughout the entirety of the adaptation period, revealing no substantial differences. The numerical findings from the measurements of trout behavior have been detailed in Table 1.

Feeding behavior when using EF3 feed additive. Starting from the 4th day, one of the feeders saw the introduction of the experimental formulation. In the initial series of experiments (EF3 – 0.3%), a discernible shift in feeding behavior was observed. A noteworthy rise in the number of fish within the feeding area fed with the experimental feed was evidenced when compared to the control group (Con1) ($p < 0.05$; see Fig. 2, *a*).

Table 1

Results of evaluation of feeding behavior of rainbow trout using control feed formulation (adaptation period, 1-3 days)

Measured parameter	Results on 1-3 days of experiment (adaptation period)	
	Control 1	Control 2
Number of fish in Z1	4.83 ± 0.75	4.66 ± 1.03
Number of fish in Z2	5.16 ± 0.75	5.33 ± 1.03
CED FZ1, s	144.50 ± 18.27	131.50 ± 10.25
CED FZ2, s	125.75 ± 4.23	123.66 ± 15.48
RFR FZ1, s	30.65 ± 6.77	29.24 ± 6.19
RFR FZ2, s	25.75 ± 4.23	23.84 ± 4.94

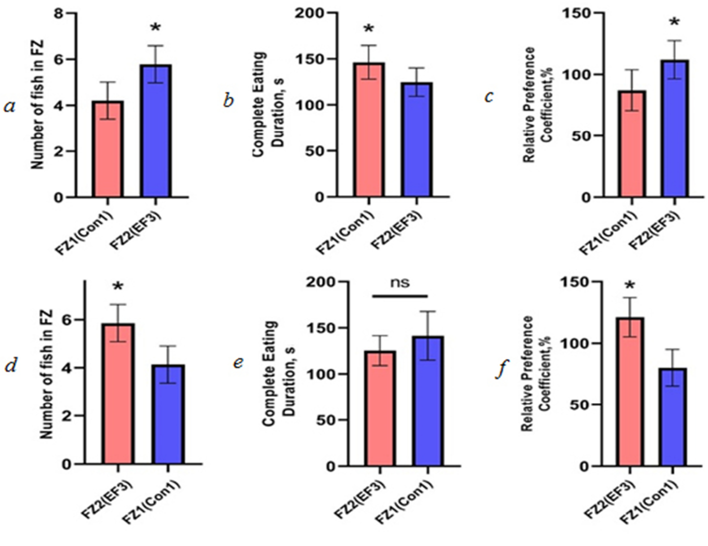

Fig. 2. Measured parameters of feeding behavior of a rainbow trout during the testing and validation period for the first series of experiments (EF3): *a*, *d* – number of fish in the feeding area; *b*, *e* – time of food eating, CED; *c*, *f* – relative attractiveness of the food, RP; *ns* – no significant difference; * – significant difference by t-test with Welch's/Mann–Whitney correction

Over the period spanning from the 4th to the 10th day, the mean fish count within the FZ2 zone amounted to 5.7 ± 0.8. Within the first experimental series, the control group exhibited the lengthiest feeding behavior duration (146.2 ± 18.2 s), exhibiting significant disparity from the experimental group's 121.9 ± 1.8 s ($p < 0.05$; refer to Fig. 2, *b*; Table 2).

Table 2

Estimation of rainbow trout feeding behavior using control and experimental feed formulation in the first experimental series (EF3)

Measured parameter	Results on 4-17 days of experiment	
	4-10 days	11-17 days
Number of fish in FZ1	4.21 ± 0.80	5.85 ± 0.77
Number of fish in FZ2	5.78 ± 0.80	4.14 ± 0.77
CED FZ1, s	146.28 ± 18.23	125.42 ± 16.15
CED FZ2, s	124.78 ± 15.35	141.42 ± 26.32
RFR FZ1, s	35.83 ± 3.97	21.76 ± 5.56
RFR FZ2, s	21.99 ± 1.81	35.20 ± 12.00
RP FZ1, %	87.19 ± 16.02	121.18 ± 10.34
RP FZ2, %	111.98 ± 15.51	80.18 ± 14.9

Utilizing the numerical data, the Coefficient of Relative Feed Preference was computed, yielding a value of 111.9 ± 15.5%. This value significantly exceeded the values recorded in the control group ($p < 0.05$; see Fig. 2, *c*). The feed containing 0.3% feed additive not only significantly decreased the feeding duration but also notably increased the fish count within the feeding area.

The validation period, initiated on the 11th day of the experiment with the interchange of the positions of feeders containing experimental and control feed, saw its commencement. Over the initial three days, the fish exhibited no discernible preference for either feeding zone, mirroring the behavior observed during the adaptation phase. Subsequently, an escalation in the number of fish within the feeding zone featuring the experimental food (FZ1) was noted. The average fish count within FZ1 significantly surpassed that of the control group ($p < 0.05$; Fig. 2, *d*). Nevertheless, no noteworthy distinctions in the duration of feed consumption were recorded (Fig. 2, *e*).

The relative speed of feed consumption within the experimental group averaged at 21.76 ± 5.5 seconds, representing a considerable 61.7% reduction compared to the pace of control group feed intake ($p < 0.05$; Table 3). The computation of the index of relative feed attractiveness revealed that the experimental feed significantly ($p < 0.05$) exceeded the control parameters, with an attractiveness index of 121.1 ± 10.3% (Fig. 2, *f*).

Feeding behavior when using EF5 feed additive. In the second series of experiments, where the feed additive dosage was elevated to 0.5% (EF5), akin outcomes were achieved. Hence, throughout the testing period, the mean fish count within feeding zone FZ2 when administered the experimental feed averaged 6.07 ± 0.92. This count surpassed the value observed within the control feeding zone (Con2) ($p < 0.05$; Table 3).

Table 3

Estimation of rainbow trout feeding behavior using control and experimental feed formulation in the first experimental series (EF5)

Measured parameter	Results on 4-17 days of experiment	
	4-10 days	11-17 days
Number of fish in FZ1	3.92 ± 0.91	5.71 ± 0.99
Number of fish in FZ2	6.07 ± 0.91	4.28 ± 0.99
CED FZ1, s	137.07 ± 15.51	127.64 ± 17.75
CED FZ2, s	118.07 ± 13.65	143.92 ± 15.19
RFR FZ1, s	37.36 ± 17.17	22.85 ± 4.61
RFR FZ2, s	19.83 ± 2.78	35.48 ± 7.80
RP FZ1, %	84.18 ± 20.22	122.44 ± 18.55
RP FZ2, %	113.83 ± 17.19	80.35 ± 18.64

A markedly longer duration and relative pace of feed consumption were evident in the control feed compared to the experimental counterpart. The relative feed attractiveness within the feeding area containing the experimental feed was significantly higher than the controls ($p < 0.05$; Fig. 3, *c*).

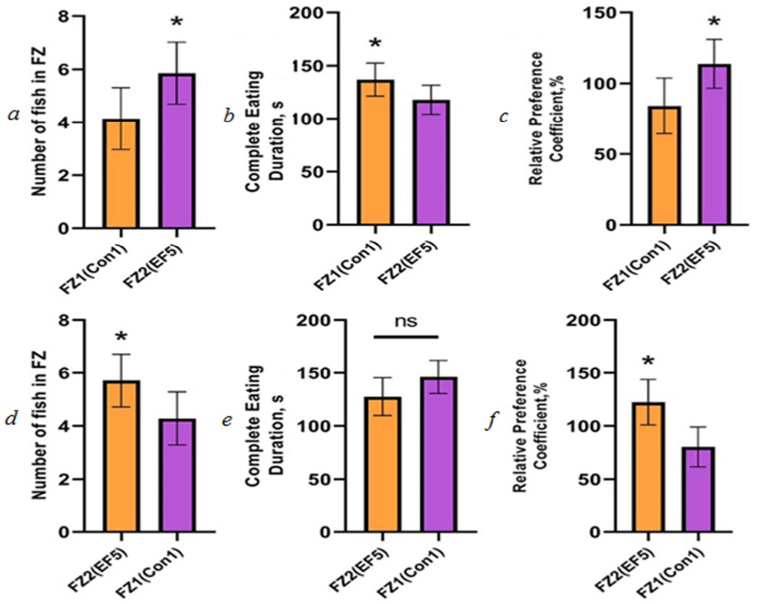

Fig. 3. Measured parameters of feeding behavior of rainbow trout during the testing and validation period for the first series of experiments (EF5): *a*-*f* – see fig. 2

The overarching trends in feeding behavior did not exhibit noteworthy variations from those documented in the initial series of experiments. Enhanced behavioral engagement was noted among fish within the feeding zone featuring the experimental feed.

Throughout the validation period, a significant increase was observed in the number of fish within the feeding zone with the experimental feed (FZ1), in contrast to the control (Fig. 3, *d*; $p < 0.05$). Concurrently, a decrease in the fish count within the control feeding zone (FZ2) correlated with protracted feed consumption durations and a diminished rate of ingestion (Fig. 3, *e*), presenting noteworthy distinctions compared to the feeding zone featuring FZ1 feed.

The computation of the relative preference coefficient facilitated the identification of a substantial index surplus within the experimental group when juxtaposed against the control formulation. During this stage of the experiment, the fish predominantly positioned themselves within the feeding zones throughout the entire duration, likely as a consequence of developing a conditioned reflex, which served to delimit the experiment's timeframe.

Discussion

The data acquired during the course of this study unequivocally demonstrate that the chosen formulation of the feed additive instigates a discernible alteration in feeding behavior. This change is characterized by a consistent preference exhibited by the experimental fish for the provided sustenance, contrasting the behaviors of the control groups. This preferential tendency persisted over the validation period, thereby affirming the presence of attractive attributes within the scrutinized feed additive. It is conceivable that the components integrated into the developed additive exerted a notable influence on the food's allure, largely attributable to their interaction with the olfactory and gustatory receptors of rainbow trout. Additionally, the increase in number of fish within the feeding zone featuring the experimental feed might be ascribed to the potential release of water-soluble constituents into the aquatic environment.

Plant essential oils consist of aromatic volatile compounds that possess the potential to influence the feeding behavior of fish [2, 22]. Cinnamaldehyde, categorized as an essential oil, has been reported by several researchers as a noteworthy food attractant across diverse animal species [25]. Multiple studies have illustrated that the incorporation of cinnamon aldehyde into feed yields growth enhancement [26]. This enhancement is attributed not solely to its impact on nutrient digestion and absorption, antioxidant capabilities, and immune status, but also to its favorable influence on appetite.

Various systems play integral roles in the regulation of feeding behavior: (*i*) receptors tasked with deciphering signaling molecules emanating from both the external and internal environments, and (*ii*) the hypothalamic-pituitary system, which amalgamates incoming signals to facilitate neurohumoral regulation. Notably, the hypothalamus generates humoral factors that either stimulate or curb food intake. A pivotal characteristic of receptors lies in their exclusive transmission of information pertaining to biologically consequential chemical stimuli, a feature that stems from the evolutionary progression of chemospecificity [9]. The apprehension of minute quantities of molecules, such as individual fatty acids constituting a fractional portion of food components (0.3 and 0.5%), is aptly executed by olfactory receptors.

The devised experimental protocol for appraising the attractiveness of artificial feeds has effectively unveiled substantive contrasts in feeding behavior contingent on varying feed compositions. The fixation of multiple parameters coupled with the precise experimental phase has facilitated the direct quantification of these parameters, thereby yielding intricate indices of feeding preference. Noteworthy about this evolved assessment framework is its emphasis on commercial aquaculture feeds at the juncture of their conception [27]. Prevailing methodologies have primarily been employed to investigate physiological and ethological dimensions of feeding behavior [3].

The more streamlined and concise experimental design presented within this paper, featuring an expanded array of data collection points, yields result that align well with other, more intricate methodologies. Capitalizing on the developed experimental setup, beyond the scrutinized parameters, more intricate investigations into feeding behavior become plausible. These include studies concerning individual/group taste preferences, assessments of attractant impacts on diverse stages of feeding behavior, and the dynamics of group interactions during feeding instances. Such endeavors will require improvements to the video recording system, adjustments to the adaptation and testing durations, as well as simultaneous testing across multiple setups. It might also be imperative to fine-tune the experiment's overall duration and individual stages (adaptation, testing, validation) to preempt the development of feeding reflexes in the fish.

The evident enhancement in feed consumption observed among the experimental groups of fish encourages further exploration of the feed additive's components, permitting their introduction in larger proportions. This progression offers the potential for refining the fatty acid composition of composite feed, as well as substituting fish oil and synthetic phospholipids with ecologically sourced constituents. An inherent constraint regarding the integration of vegetable oils in feed lies in their limited content of long-chain polyunsaturated fatty acids (ω-3), which are essential for fish. Their deficiency can precipitate compromised receptor cell sensitivity and the degradation of muscle tissue quality. Studies have demonstrated that substituting a portion of fish oil with vegetable oils (below 30-50%) in salmonid fish diets does not compromise growth performance [14]. Certain plant-derived fatty acids can undergo desaturation and elongation processes in specific freshwater fish species, converting into the requisite bodily fatty acids [28-30].

The feed additive employed in this study did not exert any influence on the fatty acid composition of the feeds due to its low content (300 and 500 g/kg). Nonetheless, hemp and rapeseed oil contained n-3 fatty acids up to 22 and 13%, respectively, surpassing soybean and olive oil (8 and 1%), yet trailing behind linseed oil (60%). Consequently, the utilization of elevated concentrations of the explored components (up to 30% replacement of the fat component of the feed) could potentially contribute not only to heightened digestibility but also to an improved growth rate via an augmented energy value of the feed.

The observed feed attractiveness is susceptible to alteration as the fish mature and through the process of “habituation”, which might necessitate the refinement or substitution of active components to achieve the intended outcome. Physiologically, receptor cells can reach a threshold beyond which further increases in stimulus concentration result in diminishing responses. Prolonged exposure to feeds of a specific formulation can lead to habituation in certain instances. Consequently, fish might exhibit a preference for “more familiar” feeds over new feeds, even if the latter possess a more comprehensive composition. One could assert that the maximum impact on fish rearing from the utilization of the developed feed additive will likely be realized in cage-based fish farming, where losses stemming from unconsumed feed are minimal.

Conclusion

Following this study, the feed additive designed for rainbow trout, comprising rapeseed and hemp oils, lycopene, and cinnamon aldehyde, exhibited evident attractant and possibly insitant properties. The formulated experimental procedure and apparatus enabled the assessment of disparities in the nutritive allure of industrial pelleted feeds for rainbow trout by monitoring factors such as the duration of fish presence in the feeding region, the number of fish, and the pace of feed consumption (both during the test period and validation period). The integration of the developed feed additive into the standard feed composition, at concentrations of 0.3 and 0.5% of the feed weight, significantly influenced the dietary preference of trout within the simulated environment. Notably, no notable distinction emerged between the two examined concentrations (0.3 and 0.5%) across the scrutinized parameters. However, it's noteworthy that the higher attractant concentration did incite a modification in fish behavior, manifesting as the congregation of fish within the feeding zone between feedings. Based on the acquired findings, we are inclined to endorse the adoption of the developed additive for enhancing the palatability of trout feed created according to the conventional formulation.

References

1. Miroshnikova E. P., Ponomarev S. V. *Akvakul'tura* [Aquaculture]. Orenburg, Izd-vo Orenburg. gos. un-ta, 2013. 184 p.

2. Jannathulla R., Sravanthi O., Khan H. I., Moomeen H. S., Gomathi A., Dayal J. S. Chemoattractants: Their essentiality and efficacy in shrimp aquaculture. *Indian Journal of Fisheries*, 2021, vol. 68 (1), pp. 151-159.

3. Liu H., Xu L., Li D. Detection and recognition of uneaten fish food pellets in aquaculture using image pro-

cessing. *In Sixth International Conference on Graphic and Image Processing (ICGIP 2014)*, 2015, vol. 9443, pp. 86-92.

4. Heikkinen J., Vielma J., Kemiläinen O., Tiirola M., Eskelinen P., Kiuru T., von Wright A. Effects of soybean meal based diet on growth performance, gut histopathology and intestinal microbiota of juvenile rainbow trout (*Oncorhynchus mykiss*). *Aquaculture*, 2006, vol. 261 (1), pp. 259-268.

5. Okrestina N. F., Isaeva O. M. Razrabotka tekhnologii ispol'zovaniia kombikormov s privlekatel'nymi vkusovymi dobavkami iz otkhodov syr'ia gidrobiontov v usloviiakh lososevykh rybovodnykh zavodov na Kamchatke [Development of technology of using mixed fodder with attractive flavor additives from hydrobiont raw material wastes in conditions of salmon fish farms in Kamchatka]. *Sostoianie i puti razvitiia akvakul'tury v Rossiiskoi Federatsii*, 2022, pp. 130-135.

6. Grozesku Iu. N., Bakhareva A. A., Sergeeva Y. V., Zhandalgarova A. D., Vladimirov V. S. Karotinoidnye preparaty v sostave kombikormov dlia osetrovykh ryb [Carotenoid preparations in compound feeds for sturgeon fish]. *Perspektivy razvitiia pishchevoi i khimicheskoi promyshlennosti v sovremennykh usloviiakh*, 2019, pp. 176-181.

7. Tekebaeva Zh. B., Shahabayeva G. S., Sarmurzina Z. S., Bisenova G. N., Urazova M. S., Dosova A. D., Abzhalelov A. B. Probiotiki i ikh primenenie v akvakul'ture [Probiotics and their application in aquaculture]. *Novosti nauki Kazakhstana*, 2020, no. 4, pp. 170-185.

8. Nikiforov-Nikishin A., Smorodinskaya S., Kochetkov N., Nikiforov-Nikishin D., Danilenko V., Bugaev O., Vatlin A., Abrosimova N., Antipov S., Kudryavtsev A., Klimov V. Effects of three feed additives on the culturable microbiota composition and histology of the anterior and posterior intestines of Zebrafish (*Danio rerio*). *Animals*, 2022, vol. 12 (18), p. 2424.

9. Atema J. Chemical senses, chemical signals and feeding behavior in fishes. *Fish Behaviour and its Use in the Capture and Culture of Fishes*, 1980, pp. 57-101.

10. Okrestina N. F., Isaeva O. M., Bonk A. A. Sravnitel'nyi analiz primeneniia standartnogo kombikorma i kombikormov s ispol'zovaniem natural'nykh pishchevykh attraktantov v ratsione kizhucha (*Oncorhynchus kisutch*) v akvakul'ture [Comparative analysis of the use of standard mixed fodder and mixed fodder with natural food attractants in the diet of coho salmon (*Oncorhynchus kisutch*) in aquaculture]. *Sostoianie i puti razvitiia akvakul'tury v Rossiiskoi Federatsii*, 2020, pp. 185-191.

11. Hara T. J., Macdonald S., Evans R. E., Marui T., Arai S. Morpholine, bile acids and skin mucus as possible chemical cues in salmonid homing: electrophysiological revaluation. *Mechanisms of migration in fishes*, 1984, pp. 363-378.

12. Fay R. R., Tavolga W. N. *Sensory biology of aquatic animals*. Springer Science & Business Media, 2012. 936 p.

13. Ponomarev S. V., Sergeev A. V., Sergeeva Iu. V. Primenenie novykh komponentov dlia povysheniia effektivnosti kormleniia tsennykh ob"ektov akvakul'tury [Application of new components to improve the feed efficiency of valuable aquaculture objects]. *Vestnik Orenburgskogo gosudarstvennogo universiteta*, 2006, no. 12-2, pp. 199-201.

14. Nasopoulou C., Zabetakis I. Benefits of fish oil replacement by plant originated oils in compounded fish feeds. A review. *LWT*, 2012, vol. 47, no. 2, pp. 217-224.

15. Geurden I., Cuvier A., Gondouin E., Olsen R. E., Ruohonen K., Kaushik S., Boujard T. Rainbow trout can discriminate between feeds with different oil sources. *Physiology & behavior*, 2005, vol. 85, no. 2, pp. 107-114.

16. Li D., Wang Z., Wu S., Miao Z., Du L., Duan Y. Automatic recognition methods of fish feeding behavior in aquaculture: a review. *Aquaculture*, 2020, vol. 528, p. 735508.

17. Tobie C., Péron F., Larose C. Assessing food preferences in dogs and cats: a review of the current methods. *Animals*, 2015, vol. 5, no. 1, pp. 126-137.

18. Saoud I. P., Babikian J., Nasser N., Monzer S. Effect of cannabis oil on growth performance, haematology and metabolism of Nile Tilapia *Oreochromis niloticus*. *Aquaculture Research*, 2018, vol. 49, no. 2, pp. 809-815.

19. Gonzalez S. C., Matsudo V. K. R., Carlini E. A. Effects of marihuana compounds on the fighting behavior of Siamese fighting fish (*Betta splendens*). *Pharmacology*, 1971, vol. 6, no. 3, pp. 186-190.

20. Smorodinskaia S. V., Gribkova V. A., Alekseev A. E., Glebova I. A. Primenenie ekstrakta likopina kak komponenta funktsional'nogo pitaniia v khlebobulochnykh izdeliiakh iz drozhzhevogo testa [Application of lycopene extract as a component of functional nutrition in yeast dough bakery products]. *Vestnik Voronezhskogo gosudarstvennogo universiteta inzhenernykh tekhnologii*, 2022, vol. 84, no. 2 (92), pp. 93-100. DOI: 10.20914/2310-1202-2022-2-93-100.

21. Caipang C. M. A., Mabuhay-Omar J., Gonzales-Plasus M. M. Plant and fruit waste products as phytogenic feed additives in aquaculture. *Aquaculture, Aquarium, Conservation & Legislation*, 2019, vol. 12, no. 1, pp. 261-268.

22. Gu Y., Han J., Wang W., Zhan Y., Wang H., Hua W., Wang W. Dietary Cinnamaldehyde Enhances Growth Performance, Digestion, Immunity, and Lipid Metabolism in Juvenile Fat Greenling (*Hexagrammos otakii*). *Aquaculture Nutrition*, 2022, vol. 2022. DOI: 10.1155/2022/2132754.

23. Kasumyan A., Isaeva O., Oanh L. T. K. Taste Preferences and Orosensory Feed Testing Behavior in Barramundi *Lates calcarifer* (Latidae, Perciformes). *Journal of Marine Science and Engineering*, 2022, vol. 10, no. 9, p. 1213.

24. Kolarevic J., Aas-Hansen Ø., Espmark Å., Baeverfjord G., Terjesen B. F., Damsgård B. The use of acoustic acceleration transmitter tags for monitoring of Atlantic salmon swimming activity in recirculating aquaculture systems (RAS). *Aquacultural engineering*, 2016, vol. 72, pp. 30-39.

25. Wall E. H., Doane P. H., Donkin S. S., Bravo D. The effects of supplementation with a blend of cinnamaldehyde and eugenol on feed intake and milk production of dairy cows. *Journal of Dairy science*, 2014, vol. 97, no. 9, pp. 5709-5717.

26. Wang Y., Wang Q., Xing K., Jiang P., Wang J. Dietary cinnamaldehyde and *Bacillus subtilis* improve growth performance, digestive enzyme activity, and antioxidant capability and shape intestinal microbiota in tongue sole, *Cynoglossus semilaevis*. *Aquaculture*, 2021, vol. 531, p. 735798.

27. Grozesku Iu. N., Bakhareva A. A. Ispol'zovanie attraktivnykh veshchestv v kormlenii osetrovykh ryb [Use of attractants in feeding sturgeon fish]. *Innovacionnoe razvitie rybnoj otrasli v kontekste obespecheniya prodovol'stvennoj bezopasnosti Rossijskoj Federacii: materialy I Natsional'noi zaochnoi nauchno-tekhnicheskoi konferentsii.* Vladivostok, Dalrybvtuz, 2017, pp 16-22.

28. Sargent J. R., Tocher D. R., Bell J. G. The lipids. *Fish nutrition*, 2003, pp. 181-257.

29. Vasil'ev A. A., Rudneva O. N. i dr. *Planirovanie tekhnologicheskikh protsessov v akvakul'ture* [Process plan-

ning in aquaculture]. Saratov, Saratovskii istochnik Publ., 2022. 135 p.

30. Nikiforov-Nikishin A., Nikiforov-Nikishin D., Kochetkov N., Smorodinskaya S., Klimov V. The influence of probiotics of different microbiological composition on histology of the gastrointestinal tract of juvenile *Oncorhynchus mykiss*. *Microscopy Research and Technique*, 2022, vol. 85, no. 2, pp. 538-547.

Список источников

1. Мирошникова Е. П., Пономарев С. В. Аквакультура. Оренбург: Изд-во Оренбург. гос. ун-та, 2013. 184 с.

2. Jannathulla R., Sravanthi O., Khan H. I., Moomeen H. S., Gomathi A., Dayal J. S. Chemoattractants: Their essentiality and efficacy in shrimp aquaculture // Indian Journal of Fisheries. 2021. V. 68 (1). P. 151–159.

3. Liu H., Xu L., Li D. Detection and recognition of uneaten fish food pellets in aquaculture using image processing // Sixth International Conference on Graphic and Image Processing (ICGIP 2014). 2015. V. 9443. P. 86–92.

4. Heikkinen J., Vielma J., Kemiläinen O., Tiirola M., Eskelinen P., Kiuru T., von Wright A. Effects of soybean meal based diet on growth performance, gut histopathology and intestinal microbiota of juvenile rainbow trout (*Oncorhynchus mykiss*) // Aquaculture. 2006. V. 261 (1). P. 259–268.

5. Окрестина Н. Ф., Исаева О. М. Разработка технологии использования комбикормов с привлекательными вкусовыми добавками из отходов сырья гидробионтов в условиях лососевых рыбоводных заводов на Камчатке // Состояние и пути развития аквакультуры в Российской Федерации. 2022. С. 130–135.

6. Грозеску Ю. Н., Бахарева А. А., Сергеева Ю. В., Жандалгарова А. Д., Владимиров В. С. Каротиноидные препараты в составе комбикормов для осетровых рыб // Перспективы развития пищевой и химической промышленности в современных условиях. 2019. С. 176–181.

7. Текебаева Ж. Б., Шахабаева Г. С., Сармурзина З. С., Бисенова Г. Н., Уразова М. С., Досова А. Д., Абжалелов А. Б. Пробиотики и их применение в аквакультуре // Новости науки Казахстана. 2020. № 4. С. 170–185.

8. Nikiforov-Nikishin A., Smorodinskaya S., Kochetkov N., Nikiforov-Nikishin D., Danilenko V., Bugaev O., Vatlin A., Abrosimova N., Antipov S., Kudryavtsev A., Klimov V. Effects of three feed additives on the culturable microbiota composition and histology of the anterior and posterior intestines of Zebrafish (*Danio rerio*) // Animals. 2022. V. 12 (18). P. 2424.

9. Atema J. Chemical senses, chemical signals and feeding behavior in fishes // Fish Behaviour and its Use in the Capture and Culture of Fishes. 1980. P. 57–101.

10. Окрестина Н. Ф., Исаева О. М., Бонк А. А. Сравнительный анализ применения стандартного комбикорма и комбикормов с использованием натуральных пищевых аттрактантов в рационе кижуча (*Oncorhynchus kisutch*) в аквакультуре // Состояние и пути развития аквакультуры в Российской Федерации. 2020. С. 185–191.

11. Hara T. J., Macdonald S., Evans R. E., Marui T., Arai S. Morpholine, bile acids and skin mucus as possible chemical cues in salmonid homing: electrophysiological re-evaluation // Mechanisms of migration in fishes. 1984. P. 363–378.

12. Fay R. R., Tavolga W. N. Sensory biology of aquatic animals. Springer Science & Business Media, 2012. 936 p.

13. Пономарев С. В., Сергеев А. В., Сергеева Ю. В. Применение новых компонентов для повышения эффективности кормления ценных объектов аквакультуры // Вестн. Оренбург. гос. ун-та. 2006. № 12-2. С. 199–201.

14. Nasopoulou C., Zabetakis I. Benefits of fish oil replacement by plant originated oils in compounded fish feeds. A review // LWT. 2012. V. 47. N. 2. P. 217–224.

15. Geurden I., Cuvier A., Gondouin E., Olsen R. E., Ruohonen K., Kaushik S., Boujard T. Rainbow trout can discriminate between feeds with different oil sources // Physiology & behavior. 2005. V. 85. N. 2. P. 107–114.

16. Li D., Wang Z., Wu S., Miao Z., Du L., Duan, Y. Automatic recognition methods of fish feeding behavior in aquaculture: a review // Aquaculture. 2020. V. 528. P. 735508.

17. Tobie C., Péron F., Larose C. Assessing food preferences in dogs and cats: a review of the current methods // Animals. 2015. V. 5. N. 1. P. 126–137.

18. Saoud I. P., Babikian J., Nasser N., Monzer S. Effect of cannabis oil on growth performance, haematology and metabolism of Nile Tilapia *Oreochromis niloticus* // Aquaculture Research. 2018. V. 49. N. 2. C. 809–815.

19. Gonzalez S. C., Matsudo V. K. R., Carlini E. A. Effects of marihuana compounds on the fighting behavior of Siamese fighting fish (*Betta splendens*) // Pharmacology. 1971. V. 6. N. 3. P. 186–190.

20. Смородинская С. В., Грибкова В. А., Алексеев А. Е., Глебова И. А. Применение экстракта ликопина как компонента функционального питания в хлебобулочных изделиях из дрожжевого теста // Вестн. Воронеж. гос. ун-та инженер. технологий. 2022. Т. 84. № 2 (92). С. 93–100. DOI: 10.20914/2310-1202-2022-2-93-100.

21. Caipang C. M. A., Mabuhay-Omar J., Gonzales-Plasus M. M. Plant and fruit waste products as phytogenic feed additives in aquaculture // Aquaculture, Aquarium, Conservation & Legislation. 2019. V. 12. N. 1. P. 261–268.

22. Gu Y., Han J., Wang W., Zhan Y., Wang H., Hua W., Wang W. Dietary Cinnamaldehyde Enhances Growth Performance, Digestion, Immunity, and Lipid Metabolism in Juvenile Fat Greenling (*Hexagrammos otakii*) // Aquaculture Nutrition. 2022. V. 2022. DOI: 1 0.1155/2022/2132754.

23. Kasumyan A., Isaeva O., Oanh L. T. K. Taste Preferences and Orosensory Feed Testing Behavior in Barramundi *Lates calcarifer* (Latidae, Perciformes) // Journal of Marine Science and Engineering. 2022. V. 10. N. 9. P. 1213.

24. Kolarevic J., Aas-Hansen Ø., Espmark Å., Baeverfjord G., Terjesen B. F., Damsgård B. The use of acoustic acceleration transmitter tags for monitoring of Atlantic salmon swimming activity in recirculating aquaculture systems (RAS) // Aquacultural engineering. 2016. V. 72. P. 30–39.

25. Wall E. H., Doane P. H., Donkin S. S., Bravo D. The effects of supplementation with a blend of cinnamaldehyde and eugenol on feed intake and milk production of dairy cows // Journal of Dairy science. 2014. V. 97. N. 9. P. 5709–5717.

26. Wang Y., Wang Q., Xing K., Jiang P., Wang J. Dietary cinnamaldehyde and *Bacillus subtilis* improve growth performance, digestive enzyme activity, and antioxidant capability and shape intestinal microbiota in tongue sole, *Cynoglossus semilaevis* // Aquaculture. 2021. V. 531. P. 735798.

27. Грозеску Ю. Н., Бахарева А. А. Использование аттрактивных веществ в кормлении осетровых рыб // Инновационное развитие рыбной отрасли в контексте обеспе-

чения продовольственной безопасности Российской Федерации: материалы I Нац. заоч. науч.-техн. конф. Владивосток: Дальрыбвтуз, 2017. С. 16–22.

28. Sargent J. R., Tocher D. R., Bell J. G. The lipids // Fish nutrition. 2003. P. 181–257.

29. Васильев А. А., Руднева О. Н. и др. Планирование технологических процессов в аквакультуре. Саратов: Саратов. источник, 2022. 135 с.

30. Nikiforov-Nikishin A., Nikiforov-Nikishin D., Kochetkov N., Smorodinskaya S., Klimov V. The influence of probiotics of different microbiological composition on histology of the gastrointestinal tract of juvenile *Oncorhynchus mykiss* // Microscopy Research and Technique. 2022. V. 85. N. 2. P. 538–547.

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