Effect of *Taraxacum officinale* Weber ex Wiggers extract on growth performance, biochemical blood parameters and meat quality of rainbow trout (*Oncorhynchus mykiss* W.), cultivated in a recirculating system

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Abstract. Plant extracts are natural products, safe for fish and the environment and also are not expensive supplements. The purpose of this study is to determine the effect of the dandelion (*Taraxacum officinale* Weber ex Wiggers) extract on the growth performance, meat quality and biochemical blood parameters (glucose, urea, creatinine, total protein, albumin, aspartate aminotransferase (ASAT), alanine transaminase (ALAT), alkaline phosphatase (ALP), Ca, P, Mg, triglycerides, cholesterol) of rainbow trout (*Oncorhynchus mykiss*). For the experiment a recirculating system in the Aquaculture Base of the Faculty of Agriculture at the Trakia University was used. A control group (no added) and an experimental (with added 272 mg kg^{-1} of dandelion extract) option were set, each of them with two replicates and mean initial weight of fish 13.32±3.07 g and 13.41±3.21 g, respectively. Forty specimens'rainbow trouts with in good health condition were placed in each tank and cultivated for 60 days. At the end of the experiment final weight, specific growth rates, feed conversion ratio were calculated average, and the meat quality and blood biochemical parameters were evaluated. Trouts from the experimental group, fed with supplement had with 9.3% higher average final weight compared to the one of individuals from the control ($p < 0.05$). The blood biochemical parameters urea, creatinine, albumin, ALP, ALAT, cholesterol and triglycerides in control variant were higher compare to values of this parameter of fish from the experimental ($p > 0.05$). Better growth performance and blood parameters were measured in trouts fed with dandelion supplement.
Key Words: biochemical blood parameters, dandelion, growth, meat quality, rainbow trout.

Introduction. Aquaculture is looking for an effective feed to meet the nutritional needs of cultivated species. The antibiotic substances used so far have stimulated the growth of aquatic animals, but their frequent use leads to resistance to pathogenic microorganisms, suppression of the immune system of the cultivated hydrobionts and pollution of the environment (Valkova et al 2015; Valkova et al 2016; Sirakov et al 2019c; Velichkova et al 2019c). It is therefore a need of another solution to this problem. Feed additives are divided into two groups, namely live and non-living. The use of low-cost live feed supplements is highly recommended and encouraged due to their eco-friendly nature (Velichkova et al 2018b). Into the live category come probiotics (Sirakov et al 2018; Kurdomanov et al 2019), some algae (Velichkova & Sirakov 2018) and plants (Kirjakov & Velichkova 2016). The use of parts of herbs or the whole plant is a relatively new strand in aquaculture and a relatively new field of research (Sirakov et al 2019a; Velichkova & Sirakov 2019). Plant extracts are natural products, safe for fish and the environment and also are not expensive supplements (Velichkova et al 2016). Herbs can be applied in many ways, such as dried, fresh, powdered, extracted in difference solvents (water, alcohol, acetone, ether, etc.) (Raman 2017). The use of single herbs or a mixture of them could lead to the overcoming of diseases in fish and may supplement the inadequacy of nutrient and phytochemicals (Velichkova et al 2018a). Phytotherapy with herbs has a great advantage that it does not stress the fish. It is very useful for

aquaculture to trace the effects of herbal extracts that lead to increased appetite, improves immunity and have anti-pathogen properties. Dandelion is a plant of the genus *Taraxacum*, a member of the Asteraceae family and is a rich source of vitamins (especially A and C) and minerals (Modaresi & Resalatpour 2012). *T. officinale* is grown from wild sources or cultivated for medicinal purposes and food. It contains various phytochemicals, including oligosaccharides, flavonoids, phenolic acids, terpenes, polysaccharides, peptides, alkaloids. *T. officinale* are used to enhance the immune systems, as a liver tonic, tract infections, pneumonia, antimicrobial, anti-inflammatory activities (Amin et al 2016). Extract of dandelion is used in the treatment of jaundice, toxicity, purifying the blood, gastrointestinal problems, even cancer (Mahesh et al 2010). The influence of dandelion extract as a dietary supplementation on growth and blood parameters in chickens, pigs, rabbits were tested by different authors (Yan et al 2012; Yang et al 2013; Qureshi et al 2017). Widespread of this plant and its unexplored effect on fish led us to study an extract from *T. officinale* in the cultivation of rainbow trout. The purpose of this study is to determine the effect of the dandelion (*Taraxacum officinale* Weber ex Wiggers) extract on the growth performance, meat quality and biochemical blood parameters (glucose, urea, creatinine, total protein, albumin, aspartate aminotransferase (ASAT), alanine transaminase (ALAT), alkaline phosphatase (ALP), Ca, P, Mg, triglycerides, cholesterol) of rainbow trout (*Oncorhynchus mykiss*).

Material and Method

Experimental fish and feeding. Eighty specimens from the rainbow trout with an average weight of 13.32+3.07 g (control, C) and 13.41+3.21 g (experimental, T.o.) in good health condition were placed in each tank and cultivated for 60 days. The concrete tanks have an effective water volume of 0.8 m^3, which are part of a recirculation system. Fish from the control group (no added) and the experimental (with added of dandelion extract) option, each with a two repetitions, were set in a recirculating system in the Aquaculture Base of the Faculty of Agriculture at the Trakia University. Fish were fed with 6 mm extruded pellets “Aqua UNI“, produced by “Aqua garant”. To the fish feed of trouts from the experimental group was added 272 mg kg^{-1} extract of dandelion, as well as oiling the granules with 5 mL of sunflower oil for every 100 g of pellets. Rainbow trout from control group were fed with granules only greased with the same amount of sunflower oil. The nutrient content in the feed of the two groups is: 25% crude protein, 3.10% crude lipids, 6% crude fiber, 8% crude ash, 1.04% P, 11.10 MJ kg^{-1} ME, 4800 IU kg^{-1} Vitamin A, 900 IU kg^{-1} Vitamin D. The daily ration that the studied fish received was 1.8% of their live weight and they were fed three times per day (8 am, 12 pm, 16 pm). The tanks were daily cleaned and excreta were siphoned (Sirakov et al 2015). Light was about 12:12 h light: dark cycle throughout the day.

Fish growth performance. The average individual weight (g) of the fish was calculated at the start, middle and end of experiment in order to study the extract of dandelion influence on the weight gain and feed conversion ratio in the rainbow trout, cultivated in recirculation system. At the end of the trial the weight gain (g), survival rate (%) and the feed conversion ratio in fish were determined.

The growth calculations were carried out according to the following formulas:

- Specific growth rate (SGR) (Zhou et al 2006):

$$SGR = \frac{LnWf - LnWi}{n} \times 100$$

where: SGR = specific growth rate, %;
Wi = initial weight, g;
Wf = final weight, g;
n = number of days.

- Feed conversion ratio (FCR):

$$FCR = \frac{\text{Feed given}}{\text{Fish weight gain}}$$

where: FCR = feed conversion ratio;
feed given, g;
fish weight gain, g.

Hydrochemical parameters. The oxygen content (mg L^{-1}), pH, water temperature (°C) and electrical conductivity (µS cm^{-1}) were measured daily with a portable meter (HQ30D), accordingly with LDO, pH (liquid) and conductivity electrodes (Stoyanova et al 2019). Other water quality parameters, ammonium (mg L^{-1}) and phosphates (mg L^{-1}) were monitored on a weekly in Ecolab Agriculture Faculty.

Chemical analyses of meat samples. The musculature samples of rainbow trout were determined on atomic absorption spectrometer (AAS) "A Analyst 800" – PerkinElmer. Crude protein content (%) was calculated by converting the nitrogen content, quantified by Kjeldahl's method, using an automatic Kjeldahl system (Kjeltec 8400, FOSS, Sweden). Lipid content (%) was determined by the method of Soxhlet, using an automatic system (Soxtec 2050, FOSS, Sweden). Ash content (%) was investigated by incineration in a muffle furnace (MLW, Germany) at 550°C for 8 h. Crucibles were brought about the room temperature and weighed (mg).

Biochemical blood analyses. Blood was taken from the examined fish directly from the heart with disposable sterile plastic syringes (3 mL) with a needle (Velichkova et al 2019a). As an anticoagulant Heparine sodium (1%) was used. The blood samples were instantly transmitted and analyzed in a hematological laboratory (NCPTC-Trakia University) and reported in Mindray BC-120 hematology analyzer. Follow biochemical blood parameters were investigated: glucose (mmol L^{-1}), urea (mmol L^{-1}), creatinine (µmol L^{-1}), total protein (g L^{-1}), albumin (g L^{-1}), ASAT (U L^{-1}), ALAT (U L^{-1}), Ca (mmol L^{-1}), P (mmol L^{-1}), Mg (mmol L^{-1}), triglycerides (mmol L^{-1}) and cholesterol (mmol L^{-1}).

Statistical analysis. The data received from the trial were statistically analyzed with ANOVA single factor (MSOffice 2010).

Results and Discussion. Water temperature is an important indicator for the optimal development of the cultivated species. In our trial it was 16.8-17.8°C in control and experimental tanks which were within the optimum values for trout farming (Velichkova et al 2019b) (Figure 1).

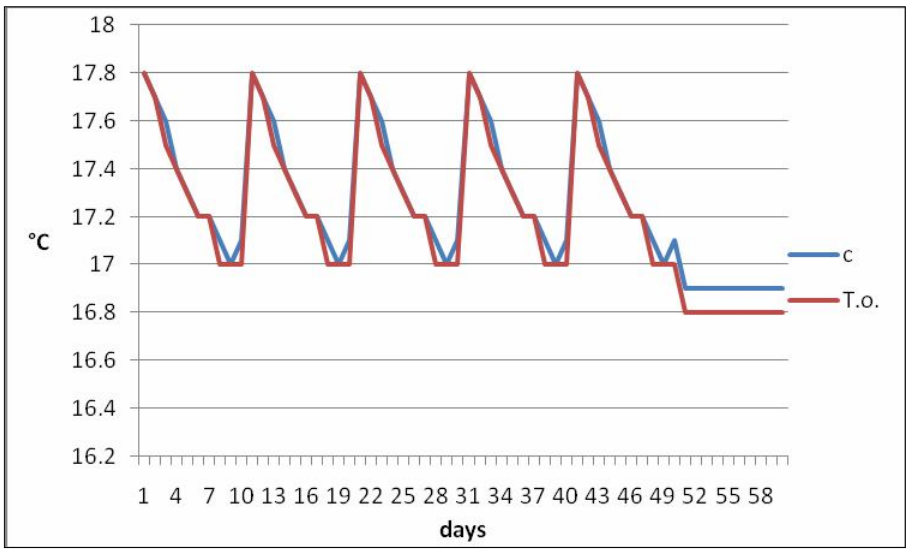

Figure 1. Water temperature in control and experimental tanks.

Water pH values in the recirculation system varied between 7.4 and 8.06 which were slightly alkaline (Figure 2).

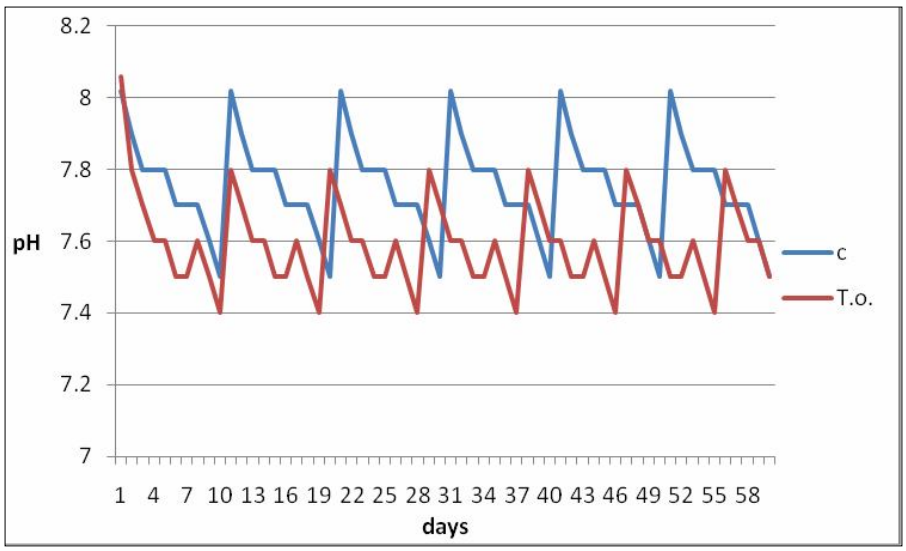

Figure 2. Water pH in control and experimental tanks.

The dissolved oxygen during the experiment ranged between 7.38 and 8.30 mg L^{-1} (Figure 3). The values of this parameter during the trial period were higher with 2.1% in the experimental tanks comparison to these of the controlled.

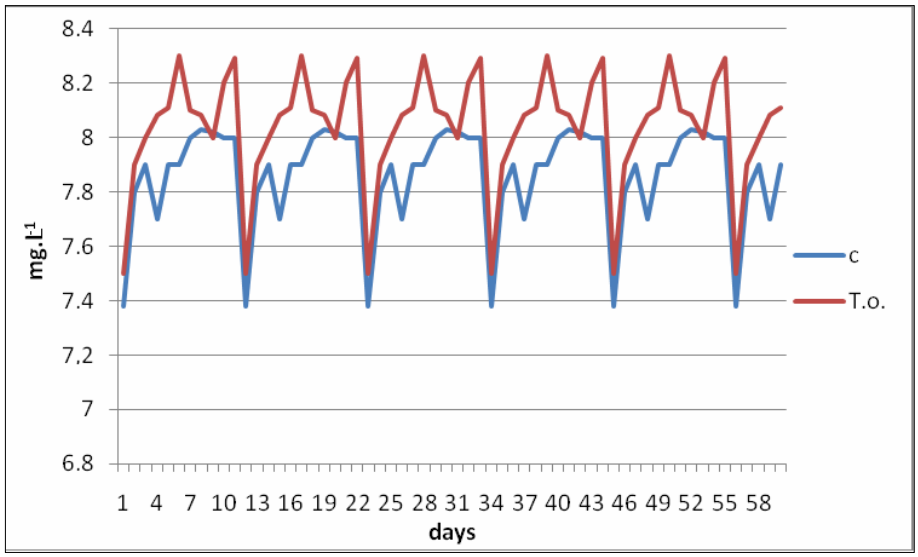

Figure 3. Dissolved oxygen in control and experimental tanks.

Electric conductivity of water varied from 264 to 269 µS cm^{-1} (Figure 4). The conductivity values of experimental variant were with 0.8% higher compared to these one of the control.

The analysis of hydrochemical data showed that during the experiment, they were optimum for the farmed species. Tanks were cleaned three times per day, with addition of fresh water in amount of 10% from the total recirculation system volume. To maintain the optimum water chemical parameters during the experiment, the mechanical filter and the biofilter in particular was of major significance. This led to good results with respect to survival, weight gain and feed conversion ratio in experimental fish. The hydrochemical parameters of water in aquaculture recirculation systems have been found to be better affected by plant cultivation or inclusion as an additive (Velichkova et al 2019b).

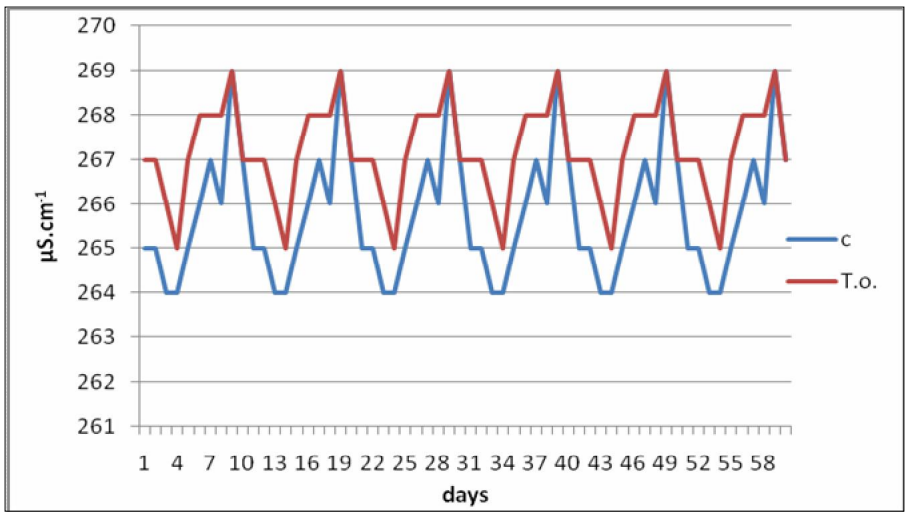

Figure 4. Electric conductivity in control and experimental tanks.

Ammonium concentration was with 32.4% lower in the water of the experimental variant compare to control for 60 days period. Phosphorus was with 18.7% lower in variant with dandelion extract compared to control for the same period (Table 1).

Table 1

Ammonium and phosphorus concentration in control and experimental tanks

Group	*n*	*Parameters (mg L^{-1})*	
		NH_4^+	$P\text{-}PO_4^-$
C	8	0.68±0.17*	0.91±0.35
T.o.	8	0.46±0.12*	0.74±0.37

*P < 0.05.

Growth performance and feed utilization efficiency. The results of the growth performance of cultivated trout in both variants are presented in a Table 2.

Table 2

Growth performance of rainbow trout in control and experimental tanks

Parameter	*n*	*C*	*T.o.*
		$\bar{x} \pm SD$	$\bar{x} \pm SD$
Initial body weight, g	40	13.32±3.07	13.41±3.21
Final body weight, g	40	38.10±7.92*	47.20±12.03*
Survival rate, %		100	100
SGR, % per day		1.86±0.16	2.08±0.01
Average individual weight gain, g	40	24.78±1.02*	33.79±1.06*
FCR		1.83	1.48

*P < 0.05.

The average initial live weight of rainbow trout from control and experimental variants was respectively 13.32±3.07 g and 13.41±3.21 g and the differences were not statistically significant (p > 0.05) (Table 2). By the end of the experiment was received average live weight of fish, fed with dandelion extract supplemented 47.20±12.03g which was with 19.3% higher compared to the value of the same parameter of control trouts (p < 0.05) (Table 2). Survival rate during the experiment showed 100% in fish in experimental and control group (Table 2). Some studies on the use of plant-based feed additives indicate a higher survival of fish fed with feed additives (Sirakov & Velichkova 2018).

The average individual weight gain of rainbow trout from the group fed with dandelion extract supplemented was 33.79±1.06g which was with 26.6% higher

compared to this one of control fish and the differences were statistically significant ($p < 0.05$) (Table 2).

At the end of the trial, the analysis of consumed extruded pellets showed that feed conversion ratio of trout in the experimental group was 1.48 and it was 19.12% lower than this one in control fish (Table 2). The growth parameters of rainbow trout were higher in the group fed with dandelion extract supplemented. According to Sirakov et al (2019b) study of carp feeding with a dandelion supplement, they received also higher growth rates for fish fed a dandelion extract compared to the control.

Biochemical blood parameters. According to Coz-Rakovac et al (2005) the blood glucose level may vary by season and water temperature. In the present study, the glucose level was 4.52% lower in rainbow trout from control group, but the differences was not statistically proven ($p > 0.05$) (Table 3). The urea level was 50.68% higher for control fish compare to these ones from the experimental variant, but with not statistically significant differences ($p > 0.05$). The creatinine level was 39.47% higher in the experiment fed with dandelion extract supplement compare to control group ($p > 0.05$). The level of the total protein was higher by 3.07% in the rainbow trout fed with the supplement compared to control, although this difference did not reach statistical significance ($p > 0.05$). Albumin transports hormones, vitamins, and substances like calcium throughout the body. Also keeps fluid from leaking out of blood vessels and nourishes tissues (Coz-Rakovac et al 2005). The amount of albumin in the blood of experimental fish is lower with 36.7% compare to control group. ALAT and ALP values are higher with 28% and 35.8% respectively in blood of trouts from the control group compare to experimental variant. All these results show that the dandelion extract has a positive effect on liver function. Cholesterol and triglyceride levels of fish may be affected by pollution agents (Yang & Chen 2003). The triglyceride concentrations in serum of rainbow trout fed with supplement were lower with 37.85% than in control fish. The measured cholesterol are higher with 36.53% in control variant compare to the dandelion extract supplement experimental group, but with not statistically significant differences ($p > 0.05$). The change in blood electrolytes may bring disturbances in the normal vital physiological functions of the fish and its growth rate (Prasad et al 2011). Calcium is contained in the bones in combination with phosphorus under the form of calcium phosphate. In this study the blood calcium, phosphorus and magnesium of rainbow trout in control variant is higher than the experimental variant, but it is not statistically proven ($p > 0.05$).

Table 3

Biochemical blood parameters of rainbow trout in control and experimental groups

Blood parameters	*n*	*C* ($\bar{x} \pm SD$)	*T. o.* ($\bar{x} \pm SD$)
Glu, mmol L^{-1}	6	5.06±0.67	5.30±1.10
Urea, mmol L^{-1}	6	0.73±0.05	0.36±0.30
Crea, µmol L^{-1}	6	7.60±1.50	4.60±4.04
TP, g L^{-1}	6	34.70±5.60	35.80±4.40
Alb, g L^{-1}	6	16.60±2.08	10.50±9.10
ASAT, U L^{-1}	6	65.00±100.50	94.60±163.90
ALAT, U L^{-1}	6	5.00±7.80	3.60±4.70
ALP, U L^{-1}	6	341.00±178.40	218.60±217.00
Ca, mmol L^{-1}	6	2.40±0.33	1.90±0.34
P, mmol L^{-1}	6	5.03±1.95	4.60±1.92
Mg, mmol L^{-1}	6	0.78±0.07	0.69±0.10
TG, mmol L^{-1}	6	1.77±0.10	1.10±0.90
CHOL, mmol L^{-1}	6	5.83±2.25	3.70±3.20

Note: Glu = glucose, Crea = creatinine, TP = total protein, Alb = albumin, ASAT = aspartate aminotransferase, ALAT = alanine transaminase, ALP = alkaline phosphatase, Ca = calcium, P = phosphorus, Mg = manganesium, TG = triglycerides, CHOL = cholesterol.

Chemical analyses of meat samples. The analysis of the data, concerning the chemical composition of the meat of the rainbow trout, cultivated in recirculation system showed that the dietary dandelion supplementation in amount of 0.73% increased significantly the moisture and decreased the dry matter content of the fillets ($p > 0.05$). It was received with 2.39% higher protein in the fillets from experimental fish fed with dandelion extract compare to control variant and the differences was statistically significant ($p < 0.05$). The higher value of this parameter in trouts from the experimental groups could be results of increased appetite and better assimilation of nutrients by digestive system in fish due to the added supplement. This result is controversial to data received from Satpathy et al (2003) who stated that higher protein level in meat of experimental fish might due to the higher energy level of the died. The fat concentrations in fillets of rainbow trout fed with supplement were lower with 18.7% compared to control fish ($p < 0.05$).

Table 4

Chemical composition of the fillets of the rainbow trout (*O. mykiss*) in control and experimental groups (%)

Indicator (%)	*n*	*Groups*	
		C	*T. o.*
		$\bar{x} \pm SD$	$\bar{x} \pm SD$
Moisture	6	74.46±0.66	75.01±0.26
Dry matter	6	25.54±0.70	24.99±0.27
Crude protein	6	18.71±0.22*	19.17±0.09*
Fat	6	5.29±0.2*	4.30±0.15*
Ash	6	1.61±0.03*	1.52±0.02*

*P < 0.05.

Conclusions. A higher average individual weight gain and feed conversion ratio were measured in rainbow trout fed with *T. officinale* supplement respectively with 26.6% and 19.12% compared to control fish. The better blood parameters were measured in rainbow trout fed with *T. officinale* supplement. A higher crude protein and a lower fat were measured in rainbow trout fed with *T. officinale* supplement respectively with 2.39% and 18.7% compared to control fish, which show a better quality of the meat in trout fed with dandelion supplement extract.

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