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Full Length Research paper

The growth performance and resıdue levels ın Raınbow trout (*Oncorhynchus mykiss walbaum 1792*) after applıcatıon of porcıne growth hormone (pgh) ın different tıme ıntervals

Deniz D. Tosun[1] and Mehmet S. Celikkale[2]

[1]Istanbul University, Faculty of Fisheries, Aquaculture Department, Ordu cad. No:200 Istanbul Universitesi, Su Urunleri Fakultesi,Istanbul/Turkey.
[2]Istanbul Aydın University, Beşyol Mah.Inönü Cad.No: 38 Sefaköy-Küçükçekmece / İSTANBUL.

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In this study, effects of porcine growth hormone on the growth performance of rainbow trout (*Oncorhychus mykiss*) and its possible residue in fish meat and blood were investigated.

The study was defined in two phases. The first phase is the rearing of the fish which were chosen to be in the same weight range as close as possible (90g-110g) till they reach the table size. Through this phase fish were injected with porcine somatotropin in periods differing from one group to another. First group received one injection, second two injections and third group received three injections. At the end of the study, fish in the experimental groups weighed more than the control groups. A1–K1 and A1–K2 groups were significantly different ($p<0.05$) in terms of weight, A2–K1, A2–K2, B1–K1, B1–K2, B2–K1, B2– K2, C1–K1 and C2–K2 groups were significantly different ($p<0.01$) as well. In the second phase, fish carcasses were analyzed for porcine growth hormone residues. There were no significant difference with the control groups in terms of residue levels.

Key words: Growth performance, porcine growth hormone, rainbow trout, residue, aquaculture.

INTRODUCTİON

Trout Farming is one of the oldest forms of commercial fish production. Trout farming dates back over 400 years in Europe, about 150 years in the United States and about 100 years in South Africa (Hinshaw 1990).Trout are farmed for food and for recreational purposes. Rainbow trout (*Oncorhynchus mykiss*), is the most commonly raised species. Brown trout (*Salmo trutta*), and brook trout (*Salvelinus fontinalis*), are also farmed.

Trout farming is an ideal option for sustainable use of water resources. In mountainous regions, it is possible to use both surface and underground waters for trout production purposes. It is possible to help ensure employment and steady incomes by the means of trout farming in areas where employment opportunities are scarce (Woynarovich *et al*., 2011).

Rainbow trout (*Oncorhynchus mykiss*) is of high importance in sport and economic purposes. A normal adult rainbow trout weighs between 2–3 kg, while its maximum length, weight and age are 120 cm total length (TL), 25.4 kg and 11 years, respectively (Froese and Pauly, 2009). Rainbow trout lives in the upper, cold water sections of rivers and seas (Woynarovich *et al*., 2011).

The widely cultured commercial strains of rainbow trouts have been improved from those original rainbow trout populations that possessed advantageous qualities, such as; hardiness, fast growth, resistance to diseases and reliable reproduction under farm conditions. (Woynarovich *et al*., 2011). Many of the production costs in an aquaculture venture are time dependent, abbreviating the time to market size will reduce these expenditures and also lower the

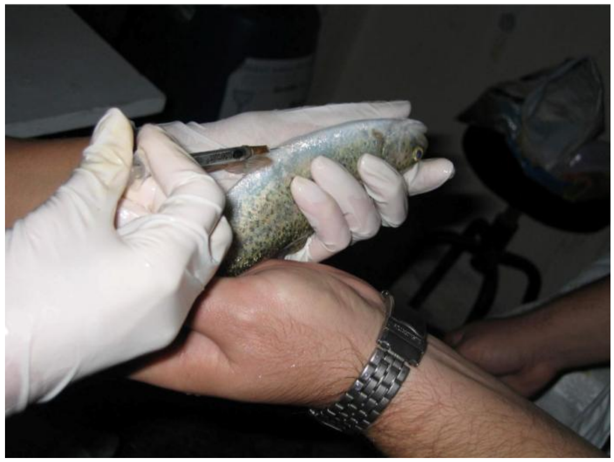
*Corresponding author: deniztosun@gmail.com. Tel: +902124555700/16475

exposure time to a variety of risks like disease vectors, predators, and losses due to climatic damages

(Cook, 2000).

Salmonids are cultured under a variety of environment-

Figure 1. Injection procedure.



al conditions which affects the growth performance of the fish during intensive production. Fish growth is under the influence of a wide range of biotic and abiotic factors and as in all vertebrates, can be regulated by growth hormone (GH) / insulin like growth factor-1 (IGF-1) interaction (Weatherley and Gill, 1987). Growth hormone is the pituitary hormone that regulates somatic growth. Exogenous GH applications enhances growth in fish, triggers IGF-1 production and affects the growth metabolism (Devlin *et al.,* 2004, Gabillard *et al*., 2003). Many scientists worked on the effects of bovine GH, genetically engineered rainbow trout GH and recombinant salmon GH on the growth rates and feed conversions of salmonid fish extensively in the last 30 years. (Higgs *et al*., 1975; Kayes, 1977; Markert *et al*., 1977; McLean *et al*., 1997; Higgs *et al*., 1979; Yu *et al*., 1979; Danzmann *et al*., 1990; Moriyama *et al*., 1993; Petersson *et al*., 2004; Inoue *et al*., 2003; Riley *et al*., 2003). The results showed that immersion, injection or usage of slow-release implants containing GH stimulated significant increases in growth and feed conversion with various efficiencies depending on the method used to deliver GH (Cook, 2000).

Growth hormone, which is secreted by the posterior pituitary gland, is the most important organic compound that affects growth. GH enhances aminoacids entrance to the muscels and bone cells and thus increases protein synthesis. Sulphate accumulation in the cartilage is increased and tissues bind glucose better. GH promotes burning of fats, in animals, weight gain is %5-8 more with enhanced feed conversion rates and food intake. Many studies shows that with the increase of GH in blood plasma, growth is increased (Mclean *et al.,* 1997; Etherton, 1999; Arıman, 2000; Wille *et al.,* 2002).

Although hormone usage enhances growth, there are concerns about its accumulation and transfer to humans via the consumption of the animals that are fed or injected with the hormones. Some scientists believe that exogenous hormones may have the potential to cause cancers in humans. In this study, our objective was to observe and identify the effects of porcine growth hormone on the growth performance of rainbow trout which was administered by intraperitonal injections and detect possible pGH residues in meat samples.

MATERİALS AND METHODS

Experimental Fish

Rainbow trout (*Oncorhynchus mykiss*) used in this experiment (120 fish, 100±10 g) were supplied from the I.U. Sapanca Inland Fisheries Research and Application Center, Sapanca, Sakarya, Turkey.

Hormone and Residue Analyzes

Porcine hormone bought from Sigma® (Growth Hormone from Porcine Pituitaries (9002-72-6) EC No 232-666-5) was used for the intraperitonal hormone injections of 0.1µg/gr. Fish of GH was injected in 1 ml saline solution for every injection period (15 days starting from day 0). GH IRMA IMMUNOTECH kit (cat#1397) from Epsilon Electronic Co. Ltd. (Ankara/Turkey) which is capable of calculating 0.1 mIU/L GH concentration was used in GH residue analyzes. Analysis were conducted at Biochemistry Department of Ankara University, Veterinary Faculty. Meat samples were prepared according to Ishıkawa *et al*. (1987). Each group had 7 randomly chosen muscle meat samples of fish which was limited with kit capacity for hormone residue analysis.

Experimental Design

At I.U.Sapanca Inland Fisheries Research and Application Center Hatchery, acclimated rainbow trout (100±10 g) were randomly distributed into eight 1m^3 circular fiberglass tanks (15 fish per tank) as four experimental groups (A, B, C, and K). Duplicate groups of fish were injected with porcine growth hormone and the control group was injected with isotonic sodium chloride solution (Figure 1). First Group (A) received a single intraperitonal injection at day 0, second group (B) received two injections at day 0 and 15, and the third group (C) received injections at days 0, 15 and 30. Every group which did not receive hormone injections, just like the control group (K), were injected with isotonic sodium chloride to even the handling stress between the groups. The injection procedure and time table is given in Table 1. Before every injection fish were anesthetized with tranquil,weighed (±0.01g presician scales) and measured for length. After the injections, fish were fed till table size (200-250 gr) and sampled for analysis.

Table 1. Injection timetable.

	A	B	C	K
First injection Day 0	GH 0.1µg/gr.fish	GH 0.1µg/gr.fish	GH 0.1µg/gr.fish	ISC*
Second injection Day 15	ISC*	GH 0.1µg/gr.fish	GH 0.1µg/gr.fish	ISC*
Third injection Day 30	ISC*	ISC*	GH 0.1µg/gr.fish	ISC*

*ISC: Isotonic sodium Chloride

Table 2. Chemical Composition of the diet.

Feed composition	%
Fish meal	48.00
Bone meal	12.70
Soybean meal	20.00
Wheat meal	13.00
Blood meal	3.00
Fish oil	2.00
Antioxydant(Ethoxyguin)	0.03
[1]Vitamin mixture	0.90
[2]Mineral mixture	0.10

[1]Guaranteed levels per kg of product.
[2]Guaranteed levels per kg of product.

Diet

A commercial trout feed with 45% crude protein (Bioaqua Extruded Trout Feed) was used throughout the experiment. Pellets number 3 and 4 (Table 2) were offered as 3% of body weight/day, three times a day. Table.2 Feed composition.

Weight Gain

Fish were weighed every 15 days during injections and at the time of sampling. Below formulas were used to calculate growth parameters (Celikkale, 2002):

Gmb = (A2 – A1) / n

Gy = {(A2 – A1)/ A1} x 100

Feed Conversion Ratio (FCR) = weight of feed given (g) / fish weight gain (g)

K = (P x 100) / L^3

Gmb = weight gain of an individual (g), Gy= Relative Growth Rate (%)

K: Condition factor, P: live weight, L: lenght (cm), A1 = initial weight (g), A2 = final weight (g)

STATİSTİCAL ANALYSİS

Mean values were analyzed with Student's T-test. Microsoft® Office Excel 2003 and SPSS 12.0 for Windows were used for statistical analyzes.

RESULTS & DİSCUSSİON

Weight Measurements

All fish in every group was weighed individually at the beginning of the trials (initial weight) and at the end of every 15 day periods. Results were analyzed using single factor T-test and mean values with standard deviations are given in Table 3. At the end of the last period, control groups had the lowest weight measurements which was 218.37g. Group A was following the control group with average of 261.29g. The highest measurements belonged to the groups B and C which were very similar, 282.92g and 277.54g, respectively. Students T-test showed that weight measurement results for A and K groups groups had significant difference at the significance level of $p<0.05$ and B-K and C-K groups had significant differences at the significance level of $p<0.01$. These results show that intraperitonal porcine hormone injection resulted in more weight gain in rainbow trout. However, trial groups were not significantly different from each other and this can be interpreted as increased intervals do not conclude in more weight gain.

Table 3. Weight measurement results.

Groups	Starting weight	1. Period	2. Period	3. Period	4.Period
A	111.84±2.25	134.68±3.40	167.42±5.28	210.35±7.35	261.29±11.73
B	111.94±1.62	135.95±1.71	173.39±3.53	213.56±6.65	282.93±4.91
C	113.26±2.20	136.13±2.70	173.70±4.09	225.97±6.36	277.54±4.53
K	105.76±1.82	121.65±2.50	158.01±4.57	194.23±6.61	218.37±7.82

Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

Table 4. Calculated Relative Growth Rates.

Groups	Day 0 – Day 15	Day 15 – Day 30	Day 30 – Day 45	Day 45 – Day 60	X±Sx(%)
A	22.99	32.58	42.53	51.33	37.35±6.13
B	24.34	37.44	40.17	69.36	42.83±9.47
C	22.86	37.57	52.27	51.57	41.07±6.99
K	15.89	36.36	36.22	24.14	28.15±5.01

Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

Table 5. Feed Conversion Ratios.

Groups	Day 0 – Day 15	Day 15 – Day 30	Day 30 – Day 45	Day 45 – Day 60	X±Sx(%)
A	1.90	1.33	1.02	0.84	1.20±0.22
B	1.79	1.17	1.09	0.62	1.17±0.24
C	1.93	1.17	0.83	0.84	1.19±0.28
K	2.86	1.20	1.20	1.80	1.76±0.39

Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

Relative Growth Rate

Relative growth rates for the groups were calculated by using the following formula;

$Gy = \{(A2 - A1)/ A1\} \times 100$

Calculated results for the periods and mean values with deviations are given in Table 4,Table 4 Calculated Relative Growth Rates.

Relative growth rates of the trial groups were significantly different than control groups but they did not have any significant difference between each other ($P<0.05$).

Feed Conversion Ratio

Calculated feed conversion ratios are given in Table 5. Group B had the lowest mean FCR which was calculated as 1.17 and highest was group K with 1.76. Mean FCR values were significantly different between control groups and the trial groups ($p<0.05$). These clearly suggest that porcine growth hormone increases the utilization of consumed food in rainbow trout. However, mean FCR values of the trial groups did not have any significant difference between each other.

Body Length

Standard lengths of the trouts were measured and mean results with deviations are given in Table 6 T-test was

Table 6. Mean values of measured standard lengths.

Groups	Day 0	Day 15	Day 30	Day 45	Day 60
A	21.57±0.20	22.26±0.22	23.03±0.30	25.15±0.34	27.06±0.49
B	21.56±0.17	22.50±0.15	23.98±0.23	25.56±0.29	27.79±0.35
C	21.73±0.14	22.60±0.14	23.69±0.19	25.79±0.29	27.69±0.37
K	21.41±0.19	22.04±0.17	22.81±0.22	24.50±0.28	25.87±0.37

Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

Table 7.Residue levels in the blood sample analyzes of the fish.

Samples	A	B	C	K
Sample 1	0.40	0.40	0.50	0.50
Sample 2	0.40	0.40	0.55	0.50
Sample 3	0.40	0.40	0.50	0.45
Sample 4	0.45	0.50	0.55	0.45
Sample 5	0.45	0.70	0.55	0.40
X±Sx	0.44±0.02	0.40±0.26	0.52±0.02	045±015

* mili.international unit / litre
** Sample number was limited with the capacity of the kit
Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

Table 8. Residue levels in muscle meat samples (mIU/L*).

Samples	A	B	C	K
Sample 1	0.55±0.02	0.40±0.01	0.40±0,01	0.55±0.05
Sample 2	0.40±0.02	0.40±0.01	0.45±0.01	0.55±0.05
Sample 3	0.55±0.02	0.40±0.01	0.40±0.01	0.40±0.05
Sample 4	0.40±0.02	0.40±0.01	0.50±0.01	0.70±0.05
Sample 5	0.55±0.02	0.40±0.01	0.45±0.01	0.60±0.05
Sample 6	0.40±0.02	0.45±0.01	0.40±0.01	0.75±0.05
Sample 7	0.40±0.02	0.45±0.01	0.40±0.01	0.45±0.05

* mili.international unit / litre
** Sample number was limited with the capacity of the kit
Values are presented as mean ± SD. Values with the different letters within the same column/row are significantly different at P<.05/P<0.01 by Student's T-test.

conducted on the last length measurements of the trouts and according to the results, The results of groups A-B, A-C, B-K and C-K were different at significance level $p<0.01$ while A and K groups had differences at $p<0.05$ significance level.

Blood Analysis

Trout were sampled for blood analysis randomly (n = 2). Amount of samples used were limited with the capacity of the test kit. Results of the blood analyzes is shown in Table 7. These results were compared using single factor ANOVA test at 0.05 significance level and no difference was found between the groups that were injected hormone and/or the control group.

Meat Analysis

Meat samples of the trout were analyzed to observe possible GH hormone residues. Samples were chosen randomly. Control groups were compared with hormone injected groups and results were subjected to single factorial ANOVA at 0.05 significance level (Table 8). It was concluded that GH did not leave residues with intraperitonal injections.

Many studies showed that GH, which is active in growth mechanism in animals, obtained from various organisms are functional in other organisms when delivered via injections, feeding or other means. After the delivery of exogenous GH, receiving animals show enhanced growths. Although in different scales, the affect of growth hormone is mostly positive. Even in low amounts, growth hormone is effective. This positive effect of GH can be a commercially seductive option as seen in milk production from cows in the USA although there are concerns in terms of human health due to possible residues in animal products. There had been studies on the residue levels of GH in dairy products but studies on residue levels of GH in trout are scarce (Cavari *et al.,* 1993, Moriyama *et al* 1993, Deaver *et al.,* 1999, Petersson *et al.,* 2004, Ronsholdt *et al.,* 2004, Rasmussen *et al.,* 2001, Bigaa *et al.,* 2005).

The mechanism of growth hormone is not completely understood due to the complexity of the growth process of living organisms but there are studies explaining principals. The relation between GH and insulin like growth factor (IGF-1) is defined. It is well known that growth hormone acts on growth by promoting IGF-1 production. Increase in protein synthesis, food conversion efficiency, appetite increase and nitrogen accumulation is attributed to growth hormone. It is reported that growth hormone increases feed intake and metabolic needs in fish (Deaver *et al.,* 1999, Ronsholdt *et al.,* 2004, Farmanfarmaian *et a.,l* 1999).

In this trial, unlike other GH trials with bovine GH, human GH or poultry GH, porcine growth hormone was used. Although the source of the GH differed, results showed many similarities in terms of growth performance in fish. As resulted in many other trials, GH showed enhanced growth in trout when compared with control groups. (Lin *et al.,* 1995; Kitlen *et al*., 1997; Silverstein *et al* ., 2000; Li *et al* ., 2003). In 1997, Kitlen, Hejol and Zinck reported that bovine GH application to rainbow trout with varying doses resulted in enhanced growth and with increasing GH amount growth was increased as seen in our results. As GH injection increased growth compared to control group, groups receiving more GH injections had more weight gain at the end of the trials. (Kitlen *et al.,* 1997). In another experiment conducted by Silverstein *et al.,* (2000), it was concluded that *Ictalurus punctatus* showed enhanced growth when bovine GH (bGH) is used. In this same experiment, FCR was better compared to the control groups. These two results are in agreement with our results. Silverstein *et al.,* (2000) reported that some species that does not favor cold water conditions may benefit the use of bGH to enhance growth in low temperature waters. In our study, we concluded that less than 9°C, growth hormone did not affect growth rates as much as it did during optimum temperatures. This can be observed between periods 1 and 2 where water temperatures were lower than 9 degrees. Peterson *et al.,* (2004) reported a similar result where bGH use resulted in enhanced growth of *Ictalurus punctatus* which is in agreement with our results where porcine GH (pGH) resulted in enhanced growth for rainbow trout. Farmanfarmaian and Sun (1999) conducted an experiment on striped bass (*Morone saxatilis)* where they injected bovine growth hormone intraperitonally. They concluded that growth enhancement of the hormone was related with amino acid absorption increase, nitrogen retention increase, muscle tissue increase and feed conversion rate decrease. In our study we did find that feed conversion rate was better in pGH administered trout (1,28±0,31) compared to the control groups (1,61±0,25) and this can easily be linked to the enhanced growth of the fish. Ronsholdt and McLean (2004) and Rasmussen *et al.,* (2001) reported enhanced growths for bGH administered (injection) trout. These two experiments are both in agreement with our results. Same result was reported by Willie *et al.* (2001) on blue tilapia (*Oreochromis aureus*) with bovine growth hormone injection. McLean *et al*., (1997); Li *et al.,* (2003); Lin *et al.,* (1995); Leedom *et al.,* (2002); Higgs *et al.,* (1976) and Arıman, (2000) all reported growth enhancement in variety of fish administored with bGH.

Arıman (2000), reported that growth enhanced trout by recombinant human somatotropine added feeds did not yield any residues in fish meat after the trials. The residue analyzes was conducted after 20 days following the feeding period to let the GH be accumulated by the fish. This study differs from our trials both in terms of the administration method, type of hormone used and the experimental design. In our experiment, groups had different amounts of GH administered in different time intervals giving us the opportunity to differentiate the effects of time dependencies on the residue levels if any were to be found. There are hardly any studies to compare our results on the residue analyzes of types of GH in fish meat other than this experiment.

CONCLUSİON

Fish that were injected with pGH showed better growth performance than the control group which only received isotonic sodium chloride injection. This property of growth hormones might lead the producers to use these additives. Meat and blood samples returned no evidence of residual growth hormones for this species. This type of residual hormone monitoring should be widened on other cultured species for the protection of public health bearing in mind that hormone usage has the potential to cause cancer in human beings. Literature on this subject is scarse and further studies are needed on this subject.

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REFERENCES

Arıman H (2000). Effect of growth agent and increasing temperature during daytime on rainbow trout fingerlings *(Oncorhynchus mykiss)*, Dissertation thesis(PhD), Atatürk University Institute of Science. Kindly provide the volume,issue and page number.

Bigaa PR, Petersonb B , Schellinga GT, Hardyc RW, Caind KD, Overturf K, Ott TL (2005). Bovine growth hormone treatment increased IGF-I in circulation and induced the production of a specific immune response in rainbow trout (*Oncorhynchus mykiss*), Aquac. 246: 437–445.

Cavari B, Funkenstein B, Chen TT, Gonzalez-Villasenor LI, Schartl M (1993). Effect of growth hormone on the growth rate of the gilthead seabream (*Sparus aurata*), and use of different constructs for the production of transgenic fish. Aquac.111,(1–4):189–197.

Celikkale MS (2002).*Inland fish and aquaculture (Vol 1), Blacksea Technical University Press*, ISBN 975-6983-25-6.Kindly clarify the figure in red or provide the page number.

Cook JT, McNiven MA, Richardson GF, Sutterlin AM (2000). Growth rate, body composition and feed digestibility, conversion of growth-enhanced transgenic Atlantic salmon, *Salmo salar*.Aquac. 188:15–32.

Danzmann RG, Van Der Kraak GJ, Chen TT, Powers DA (1990). Metabolic effects of bovine growth hormone and genetically engineered rainbow trout growth hormone in rainbow trout (*Oncorhynchus mykiss*) reared at a high temperature. Can. J. Fish.Aquat. Sci. 47:1292–1301.

Deaver DR, Bryan KA (1999). Effects of Exogenous Somatotropin (ST) on Gonadal Function in Ruminants and Swine. Domest. Anim. Endocrinol. 17: 287-297.

Devlin RH, Biagi CA, Yesaki TY (2004). Growth, Viability and Genetic Characteristics of GH Transgenic Coho Salmon Strains, Aquacult. 236: 607-632.

Etherton TD (1999). Emerging Strategies for Enhancing Growth: Is There a Biotechnology Better Than Somatotropin?, *Domest. Anim.Endocrinol.* 17: 171-179.

Farmanfarmaian A, Sun LZ (1999). Growth hormone effects on essential amino acid absorbtion, muscle amino acid profile, N-retention and nutritional requirements of striped bass hybrids, Genetic Analysis: Biomolecular Engineering, 15: 107-113.

Froese R, Pauly D (Eds) (2009). FishBase. World Wide Web electronic publication.www.fishbase.org, version (01/2009).

Gabillard JC, Weil C, Rescan PY, Navarro I, Guiterrez J, Le Bail PY (2003). Effects of Environmental Temperature on IGF1, IGF2, and IGF Type I receptor expression in rainbow trout (*Onchorhynchus mykiss*), General and Comparative Endocrinology,133: 233-242.

Hinshaw JM (1990). Trout Farming-A guide to production and inventory management. Southern Regional Aquaculture Center Publication 222, Stoneville, MS. Kindly provide the volume,issue and page number.

Higgs DA, Donaldson EM, Dye HM, McBride JR (1975). A preliminary investigation of the effect of bovine growth hormone on growth and muscle composition of coho salmon (*Oncorhynchus kisutch*). General and Comperative Endocrinology. 27, 240–253.

Higgs DA., Donaldson EM, Dye HM, Mcbride JR (1976). Influence of Bovine Growth Hormone and L-thyroxine on Growth, Muscle Composition, and Histological Structure of the Gonads, Thyroid, Pancreas, and pituitary of Coho Salmon (*Oncorhynchus kisutch*), J.Fish.Res.Board.Canada. 33,(7): 1585-1603.

Higgs DA., Fagerlund UHM, McBride JR, Eales JG (1979). Influences of orally administered L-thyroxine or 3,5,3X-triiodo-L-thyronine on growth, food consumption, and food conversion of underyear-ling coho salmon (*Oncorhynchus kisutch.*) Canadian. J.Zool. 57, 1974–1979.

Inoue K, Iwatani H, Takei Y (2003). Growth hormone and insulin-like growth factor I of a euryhaline fish Cottus kazika : cDNA cloning and expression after seawater acclimation, General and Comparative Endocrinology, 131, 77-84.

Ishıkawa Junko, Fuse Yusuke, Wakabayashi Katsumi (1987). Choice of extraction procedure for estimation of anterior pituitary hormone content, *Endocrinol. Japon*. 31, 755-767.

Kayes T (1977). Effects of hypophysectomy, beef growth hormone replacement therapy, pituitary autotrans-plantation, and environmental salinity on growth in the black bullhead (*Ictalurus melas*). General and Comparative Endocrinolgy 33, 371–381.

Kitlen JW, Hejbol EK, Zinck T (1997). Growth performance and Respiratory Burst Activity in Rainbow Trout Treated with Growth Hormone and Vaccine, Fish & Shellfish Immunology.7: 297-304.

Leedom TA, Uchida K, Ya Da T, Richmann III H, Byatt JC, Collier RJ, Hirano T, Grau G (2002). Recombinant bovine growth hormone treatment of tilapia: growth

response, metabolic clearance, receptor binding and immunoglobin production, Aquaculture. 207: 359-380.
Li Y, Bai J, Jian Q, Ye X, Lao H, Li X, Luo J, Liang X (2003). Expression of Common Carp Growth Hormone in the Yeast *Pichia pastoris* and growth stimulation of juvenile tilapia (*Oreochromis niloticus*), Aquac. 216: 329-341.
Lin HR, Zhang Q, Peter RE (1995). Effects of Recombinant Tuna Growth Hormone (GH) and Analogs

of Gonadotropin-Releasing Hormone (GnRH) on Growth of Grass Carp (*Ctenopharyngodon idellus*), Aquac. 129, 341-343.
Markert JR, Higgs DA, Dye HM, MacQuarrie DW (1977). Influence of bovine growth hormone on growth rate, appetite, and food conversion of yearling coho salmon (*Oncorhynchus kisutch*) fed two diets of different composition. Canadian. J.Zool. 55, 74–83.
Mclean E, Devlin RH, Byatt JC, Clarke CW, Donaldson EM (1997). Impact of a Controlled Release Formulation of Recombinant Bovine Growth Hormone Upon Growth and Sea Water Adaptation in Coho (*Oncorhynchus kisutch*) and Chinook (*Oncorhynchus tshawytscha*) Salmon, Aquac. 156: 113-128.
Moriyama S, Yamamoto H, Sugimoto S, Abe T, Hiranoc T, Kawauchia H (1993). Oral administration of recombinant salmon growth hormone to rainbow trout, *Oncorhynchus mykiss,* Aquac. 112, (1): 99–106.
Petersson BC, Small BC, Bosworth BG (2004). Effects of bovine growth hormone (Posilac®) on growth performance , body composition and IGFBPs in two strains of channel catfish, Aquac.232: 651-663.
Rasmussen RS, Rønsholdt B, Ostenfeld TH, McLean E, Byatt JC (2001). Growth, feed utilization, carcass composition and sensory characteristics of rainbow trout treated with recombinant bovine placental lactogen and growth hormone, Aquac. 195, (3-4): 367–384.
Riley LG, Hirano T, Grau EG (2003). Effects of transfer from seawater to fresh water on the growth hormone/insulin-like growth factor-I axis and prolactin in the Tilapia, *Oreochromis mossambicus*, Comparative Biochemistry and Physiology, Part B: Biochemistry and Molecular Biology, 136: 647-655.
Ronsholdt B, Mclean E (2004). Effects of Growth Hormone and Salbutamol on Growth Performance, Fillet Proximate Composition and Pigmentation of Rainbow Trout (*O. mykiss*), Aquac. 229: 225-238.
Silverstein JT, Wolters WR, Shimizu M, Dickhoff WW (2000). Bovine growth hormone treatment of channel catfish: strain and temperature effects on growth, plasma IGF-I levels, feed intake and efficiency and body composition, Aquac.190: 77-88.
Weatherley AH, Gill HS(1987). The Biology of Fish Growth. Academic Press, London, 443.
Wille K, Mclean E, Goddard JS, Byatt JC (2002). Dietary lipid level and growth hormone alter growth and body conformation of blue tilapia, *Oreochromis aureus,* Aquac. 209: 219-232.
Woynarovich A, Hoitsy G, Moth-Poulsen T (2011). Small-scale rainbow trout farming. FAO Fisheries and Aquaculture Technical Paper No. 561. Rome, FAO. 81
Yu TC, Sinnhuber RO, Hendricks JD(1979). Effect of steroid hormones on the growth of coho salmon (*Oncorhynchus kisutch*). Aquac.16: 351–359.