Rainbow Trout Broodstock Management and Seed Production in Turkey: Present Practices, Constraints and the Future

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Abstract

Trout has been cultured in Turkey since early 1970s, but major developments took place during the 1990s. Currently, with an annual production over 40.000 tons, Turkey became one of the top trout producing countries. Although, production mainly comes from as portion size fish from freshwater farms, seafarming in the Black Sea provides around 2000 tons of large-trout. The paper describes the current rainbow trout seed production practices in Turkey and discusses areas of improvements.

Lack of specialized broodstock or hatchery operators is one of the major characteristics of Turkish trout farming sector. Thus, most of the farms produce their own eggs and fry, and those have not got such facilities buy fingerlings rather than eyed-eggs, but demand for fingerlings vary considerably due to heavy losses as results of water quality fluctuations, disease outbreaks and management issues. There is no genetic breeding or selection programmes and described trout strains. Brood fish are mainly selected on morphological features and no records are kept. Age composition of the broodstocks range from 2 to 7 years, while the sex ratio is often around 1:1. Husbandry and feeding conditions of brood fish are very similar or sometimes worsen than on-growing fish. There is no off-season egg production through genetic or photoperiod manipulations, but natural egg production season ranges from November to April, peaking during January-February. Simply, eggs are stripped, fertilized with milt from at least two males and stocked into incubators of farm-made wooden-wire trays set in concrete or brick troughs, fiberglass troughs with aluminium baskets or vertical-flow stack incubators. Dead eggs are picked during the fist two days or after eyeing. Fluctuating water temperatures and suspended solids are main water quality related problems in hatcheries. Practices such as estimation of egg numbers, disinfections and shocking, and monosex or tripliod stock production are not common applications. Since the water temperature low or fluctuate widely incubation period lasts approximately 45 days and losses may reach 50 percent. Fry are fed commercial starter feeds with containing 45-50% crude protein. Feeding is performed by hand with 3-4 times daily only during the natural day length. Fry can be maintained indoor until size of 0.5-2.0 g and then moved out to small concrete raceways or fingerling ponds with some kind of shading. Growth rates vary widely between the farms and fry may reach mean size of 4-5 g 100 days after hatching. Disorders such as gas bubble, parasites (*Ichthyobodo* or *Costia*, *Ichthyophthirius*, *Saprolegnia*) and diseases (*Flavobacterium branchiophila*, *Flexibacter columnaris*, *Yersinia ruckeri*) may cause serious losses.

Turkish rainbow trout sector has reached stage of maturation, but it has not utilized most of the biotechnological developments widely practices in northern countries. Thus, developments in seed production as specialization in seed production, selective breeding programmes, all year around egg supply, sterile stock production and recirculating systems can be exploited for further development.

Key Words: broodstock, trout culture, seed production, Turkey

Introduction

A journey from McLeod River to Anatolia

The original range of the rainbow trout (*Oncorhynchus mykiss*) was limited to the eastern Pacific Ocean and the freshwater drainage basins mainly west of the Rocky Mountains. As we are now aware, this incredibly versatile or adaptive species is thriving well - particularly as a farmed species - in many countries around the world. It has been introduced to Japan, Europe, most of South and Central America, Africa, Southeast Asia, Australia and New Zealand.

The rainbow trout is probably one of the oldest fish in culture. Eggs have been stripped since 1872. It is also the only fish species distributed around the world through eyed-egg transfer. The first movement of eggs occurred to New York State in 1874 from a native spring spawning stock from Campbells Creek on the McLeod River, California. The pioneering fish culturist Seth Green is attributed with initiating these first transfers (Gall and Crandell, 1992). It is believed that many of the cultured stocks today, were developed from this original stock of McLeod River.

The first successful transfer of rainbow trout eggs out of the North America was made to Tokyo, Japan in 1877. This was followed by the 1885 shipments to England and Scotland. The original stock in Scotland was maintained at historical fish farm Howietown until 1990. According to Laird and Needham (1988) European commercial rainbow trout farming started in Denmark in the 1890s. Eggs from hatchery stocks established in England, Scotland and

Denmark were transferred to other European countries. The first eyed-eggs were introduced in Turkey was in 1970 by Mr. H. Papilla who set up a private farm in Bilecik which is still operating presently. Later researches from Faculty of Agriculture, University of Ankara brought a second party from Italy to pilot trout farm of Ministry of Agriculture in Sarayönü, Konya (Çelikkale *et al.*, 1999).

There have been confusions about the differences between so-called "*normal*", "*steelhead*", "*Shasta*" or "*Kamloops*" rainbow trout. Now these are all identical (i.e. *Oncorhynchus mykiss*). However, there are differences in the life histories and two well recognized forms exist: coastal (steelhead) and inland (redband). The first one generally refers to the large, west coast sea-run fish that utilize both freshwater and saltwater, while the second one always inhabits inland waters. It is believed that today's domesticated or hatchery strains have genes from both forms (Laird and Needham, 1988). That is why they can tolerate almost full-strength seawater after reaching 40-80 g depending on strain and location.

Soon after the initial egg transfers from the McLeod River, many government institutes and private individuals set up hatcheries around the world. The early success of trout culture can be attributed to the following facts:

- Species is highly adaptable to various environmental conditions,
- Exploitable phenotypic and genotypic variations are very high,
- Both sexes easily mature in captivity,
- The gametes could be stripped easily from both sexes,
- Fertilization could be performed externally in almost any holding container,
- The fertilized egg are relatively hardy and development is quite slow,
- The larvae are well developed upon hatching and soon accept artificial feed.

Today the species is widely distributed. On a global basis, eggs are available virtually year-round although it has been classified as a spring spawner (February to June). Geographical locations and thus photoperiod, seasonal variations in water temperature and selective breeding in hatcheries have modified this original spawning season. Since photoperiod is the main determinant of spawning season, fish in southern hemisphere (e.g. Australia and New Zealand) have 6 months off-season spawning compared with those fish in the north. Most of the European stocks have transformed to fall spawning strains. The fish first sexually mature at the age of 9 months to 3 years, and has great range of temperature tolerances.

The main purpose of this presentation is to describe the current general seed production practices in Turkey and discuss areas of improvements. This will include broodstock selection and management, timing of spawning, hatchery management (fertilization, biotechnological manipulations, care of eggs and larvae), and fry and fingerling production.

Global and national trout industry perspective

Farming of trout for domestic consumption has become a very large industry over the last decade. In fact, trout are produced in many parts of world (around 50 countries) virtually wherever there is water cool enough to support them. It is among the first 10 top farmed aquatic species and after the Atlantic salmon it is the second salmonid species widely cultured. Around 500,000 tons of trout, mainly rainbow trout (*Oncorhynchus mykiss*), are produced each year in Europe, North America, Japan and Chile. The top five countries, Chile (77,000 t), Italy (49,000 t), France (48,000 t), Denmark (41,000 t) and Norway (40,000 t) accounted for almost 50% of all production in 1998 (Table 1). Production in marine cages was 119,000 t in 1997, accounting for about one-third of annual global production. International trade in trout is much less than salmon, with exports reaching around 80,000 tonnes in 1997 (Tacon, 1998). Primary consumer countries of trout are Japan, Europe and America, with Japan as the main importer, while Chile as the major exporting country. Trout farms tend to be small and are usually for local consumption and concentrated in trout producing countries, but Norway and Chile have been able to farm particular qualities of large-sized pigmented trout for the Japanese market. Japanese trout imports reached 60,000 tonnes in 1998 (Lem, 1999).

In Europe, large rainbow trout (>1.2 kg individual size) are being increasingly farmed, production rising to 97,000 tons in 1999 with corresponding prices up to 2.76 Euro/kg. It increased to around 110,000 tons in 2000. Production in large-scale trout producing countries remains stable. Turkey also produces around 2,000 tons of trout in this category (Table 1). Countries producing large trout are keen to improve their market knowledge, and discussion on common quality criteria and common marketing efforts has started. With a production of almost 220,000 tons in 1998, portion-size trout production has stabilised in Europe at around 220,000-230,000 tons, divided mainly into "pink" (pigmented) and "white" portion-size fish (Figure 1). Production levelled off in France and Italy, primarily due to market pressures, while slight increases were noted in the UK and Spain. Prices seemed to be more stable for "pink" (Euro 1.96/kg) while average value for "white" was down to Euro 2.1 from Euro 2.2. Most trout associations are seeking to promote this market sector (FAO-HAKI, 1999).

Although trout culture in Turkey began as early as 1970s, main developments started in the 1980's and accelerated during early 1990s (Figure 2). Since

Table 1. Production of rainbow trout (tons/year) in major countries (derived from various sources).

Countries	Product Forms	1989	1998
Australia (Tasmania)	Portion-size and Salmon-trout [a]	2,200	2,150 [c]
Austria	Portion-size	3,200	3,000
Canada	Portion-size	4,450	5,250 [c]
Chile (1)	Portion-size white and salmon-trout	2,900	77,100 [c]
Colombia	Portion-size	800	7,800 [c]
Denmark (4)	Portion-size white and salmon-trout	32,200	43,000
	Sea trout (*S. trutta*)		250
Finland	Salmon-trout	18,500	17,000
France (3)	Portion-size pink and large pink [b]	30,820	48,000
	Sea trout (*S. trutta*)	1,300	1,300
Germany	Portion-size white and pink	23,100	25,000
Greece	Portion-size white	2,000	2,300
Iran	Portion-size white	1,000	2,600 [c]
Italy (2)	Portion-size white and pink	33,000	49,000
Japan	Portion-size white	15,300	13,400 [c]
Norway (5)	Salmon-trout	3,500	40,000
Peru	Portion-size	1,000	7,500
South Korea	Portion-size white	1,250	3,650
Spain	Portion-size pink and white	16,000	26,000
Sweden	Salmon-trout and portion-size white	2,800	5,300
Turkey	Portion-size white	2,500	34,600
United Kingdom	Portion-size pink and Salmon – trout	15,000	16,000
U.S.A.	Portion-size	26,000	25,700 [c]

[a]: Salmon - trout (2-3 kg) produced in seawater; [b]: Large (>1 kg) pink trout produced in freshwater; [c]: 1997

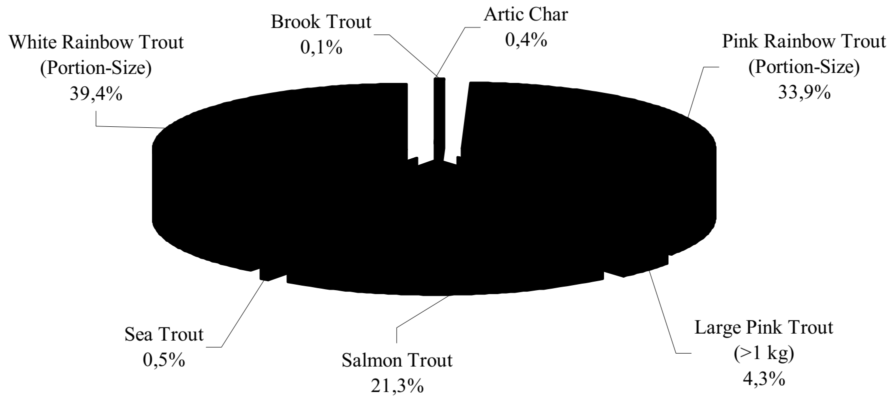

Figure 1. Distribution of cultured trout products in Europe (average of 1996, 1997, 1998) (FAO - HAKI, 1999).

then it has been increasing steadily. The growth was exceptionally intense during the second half of the 1990's and the production grew from around 7,000 t in 1994 to 38,500 t in 1999 (Figure 2). It is almost the only species cultured in freshwater and consists of over 60% of countries total aquaculture production. Nearly half of the production has been produced by farms in the Black Sea (25%) and Aegean (24%) regions, followed by the regions of Marmara (20%), Mediterranean (15%), Central Anatolia (12%), and Eastern and South-eastern Anatolia. One third of the country's trout farms are located in the Black Sea region, but this followed by Mediterranean (22%), Marmara (20%), Aegean (12%) and Central Anatolia (8%).

Only about 2,000 t of the production comes from 24 cage farms located in the Black Sea and the rest is from around 700 freshwater farms. More than 50% of farms are using stream water, while 15% using spring water, 18% both, i.e., spring water for hatchery

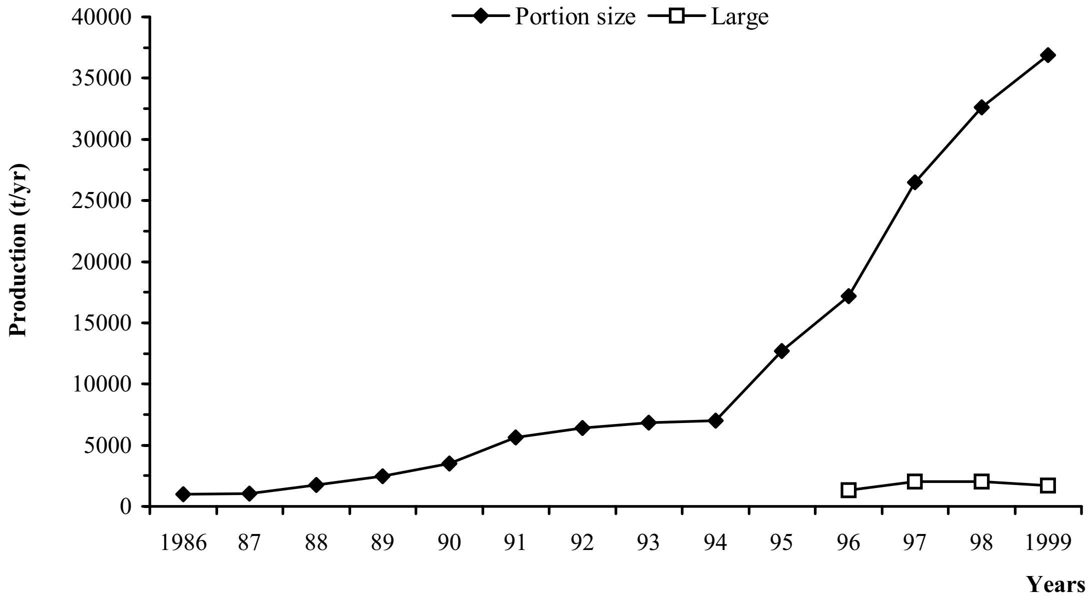

Figure 2. Development of trout production in Turkey (Çelikkale *et al*., 1999; DIE, 1999 and 2000).

operations and stream water for grow-out, and around 5% utilizing lakes and dams. Nearly half of the farms have an annual capacity of 5 t, 45% of them 5 to 30 t and the rest over 30 t. Almost 80% of the farms are family owned and only 16% of them operated by companies. Despite some production disadvantages, small farms have been preferred and seem to be economically more reliable because of local market opportunities and use of family work force. Over 50% of the farms have their own hatcheries.

Trout produced in freshwater farms are marketed as white portion size fish, while those produced in sea cages are sold as large trout (>0.5 kg). This is the only product diversification. Portion size trout is mainly marketed locally, as fresh product either cooked and served in restaurants in or near the farm or as gutted whole fish.

Although at present the total annual egg production in the world is unknown, it can be estimated that around 5 billion eggs are needed per annum to support current world production of some 500,000 tons. Data from Chile's salmonid egg production and imports can be given as an example to illustrate global importance of egg production and trade. Combining domestic production and imported salmonid eggs, total utilization in 1999 was 368.5 million of which 30 percent or 109.8 million with a value of $8.65 million were imported. Approximately 59 percent (65 million) of imports were Atlantic salmon, 40 percent (44 million) rainbow trout and 1.0 percent Coho salmon. Ireland, Norway, Denmark, U.K., U.S.A., Iceland, Sweden, Canada and Finland were the main supplier in order of declining share (Stockard, 2001).

Broodstock Management and Seed Production

Life cycle of fish embraces several distinct phases, such as spawning and egg fertilization, hatching and larval development, and subsequent growth of juvenile fish up to adulthood. The production cycle on a fish farm needs to take account of the different biological and environmental requirements of fish as they grow as shown in Figure 3.

Increasingly, sophistication of the retail market particularly supermarket chain for trout, is demanding a continuity of product both in terms of quality and size of fish produced as well as maintenance of regular supplies throughout the year. Realisation of these objectives requires both knowledge and control of all aspects of the trout's biology wherein reproduction is the most important. The life history of the fish is dominated by reproduction and this may pose special problems for farmers. Thus, primary requirement for commercial intensive culture as far as broodstock management and good farming practice are concerned is an ability to control fully the sexual maturation and spawning of the species cultured. Present approaches for achieving this control are showing in Figure 3. Furthermore, deteriorative changes in flesh coloration and texture together with reductions in growth and increased susceptibility to disease, particularly in males, have encouraged the development of methods to delay or prevent maturation. For the trout industry, to meet the 5 billion eggs requirement yearly, there is a need to optimize the reproductive potential of existing

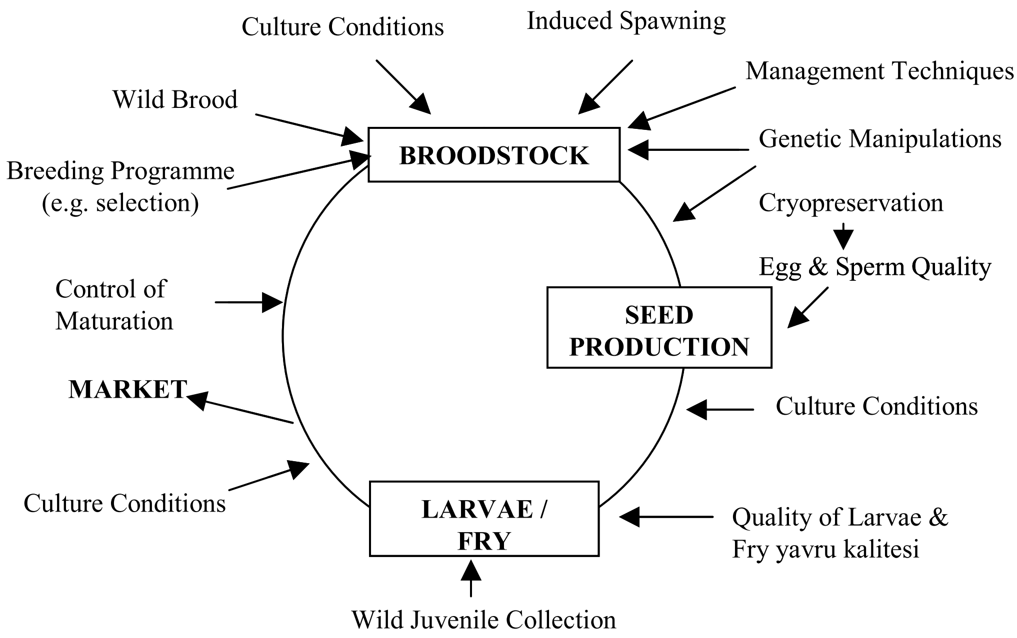

Figure 3. Methods of broodstock management and seed production (larvae and fry) for fish culture (Bromage, 1995).

broodstocks and to make fertilized eggs available during "off-season". Presently complete control of the reproductive cycle of the rainbow trout is possible.

Broodstock Selection

As mentioned earlier, the McCloud River population was widely used for development of hatchery stocks. The species has been introduced to Turkey from European hatchery stocks, which have been evaluated under hatchery conditions for around 90 years. Initially most of the stock material stems from Denmark. After the official introductions during the early 1970s there is no known transfers until 1990s. Thus, it is possible to conclude that today's stocks have originated from a few European stocks. Unfortunately, there are no records or pedigree studies on rainbow trout stocks in Turkey. However, a few regional farms (e.g., public pilot farm in Konya, Papilla in Bilecik and Çağlayan in eastern Black Sea) played an important role as a resource for new stocks. For example, recent chromosome analysis of stocks in Eastern Black Sea Region showed that these fish have similar chromosome karyotypes indicating immediate common past (Ulupınar and Okumuş, 1999).

In general, new farms start production with fingerlings bought from nearest farms and from these fish establish their own broodstocks. Broodstock selection is often based on their morphological conditions. In many cases, unsold fish after marketing season ended as broodstock. Although there seems to be a tendency maintaining fewer males than females, sex ratio is often around 1:1. Age composition of the broodstocks ranges from 2 to 7 years weighing of over 0.5 to 3.0 kg (Table 2). Due to irrational farm management, the number of brood fish might have exceeded the needs of the farms and some of the fish may not be stripped. Generally, females are kept longer than males probably due to physical deterioration of the males. Broodstocks are recruited with candidates from the same stock, but not regularly. Since gene flow from a distance stock is hardly considered, undesirable effects of inbreeding in some stocks have already reached considerable levels.

In spite of annual production of around 40,000 t and around 700 operational farms, so far no genetic breeding programme has been implemented in Turkey and there is not any "described" strain. In fact, rainbow trout is the fish species with most estimates of genetic and phenotypic parameters. Heritabilities

Table 2. Mean values of brood size, fecundity, and egg size of a few rainbow trout broodstocks in north-eastern Turkey (Okumuş *et al.*, 1997; Kurtoğlu *et al.*, 1999).

Variable	Mean	Range
Brood size (g)	1,871	726-3,231
Total fecundity (egg/fish)	3,870	1,509-9,244
Relative fecundity (egg/kg bw)	2,123	1,006-4,281
Egg size (mm)	5.1	4.2-5.8

for economically important traits are of low to moderate size but with large phenotypic variance, particularly for body weight, body shape, colour of fillet and age at maturation (Gjedrem, 1992). Thus, USA and some European countries have conducted successful breeding programmes. Among them the programmes conducted by Norwegian Research Institute Akvaforsk and Aqua Gen AS since 1971 is worth mentioning (Gjerde, 2001). They have been conducting selective breeding programmes in relation to rate of growth, delayed sexual maturity, phenotype for better colour and shape, and better colour of filets. Outlines and gene flow of such a programme are presented in Figure 4. Akvaforsk used to finance the programmes through egg, fingerling and fish sales. Recently, the programmes have been taken over by Aqua Gen AS owned by fish farmers and their organisations (1/3), the National Industrial and Development Bank (1/3) and the others (fish feed manufacturers, fish farming equipment suppliers, banks and insurance). Results of this breeding work were:

- Growth: A gain of 6% per generation, or 2% per year.
- Sexual maturity: A delay of 12% per generation or 4% per year.
- Shape and external colour: Dramatic changes with one generation towards shiny skin and good shape.

It may not be realistic to consider similar well planned and managed breeding programmes in Turkey in immediate future. However, some kind of selection programmes based on at least individual performances of fish should be performed.

Broodstock husbandry and feeding

The primary objective of salmonid broodstock management programme is the production of the maximum number of high quality eggs and fry from available broodstock. It is well known that egg production (fecundity) and quality of eggs is related not only to the genetics of broodstock but also to their rearing conditions. These include husbandry and feeding.

Appropriate rearing temperatures for maturation of broodstock have great importance since undesirable variations can lead to major reductions in egg production and an increase in atresia. Reproduction is very sensitive to stress. This may be especially important for the males. Among the major sources of stress, handling, water quality parameters and stocking density should be watched carefully. Although it depends on the water temperature, dissolved oxygen level and flow conditions, stocking density for broodstock should be lower than for grow-out and Ingram (1988) suggested 8-10 kg/m^3 as maximum for rainbow trout during non-spawning season.

Since there is no breeding programme in hatcheries in Turkey, broodstock is not marked or tagged at all. Broodstock are maintained under similar conditions with on-growing juveniles, but often worse conditions due to lack of space and inadequate water supply. Generally, sexes are stocked together and after spawning season they are not handled until the next breeding season. Although stocking densities may vary from farm to farm and in the same farm with time, generally around 10 kg/m^3 or less is

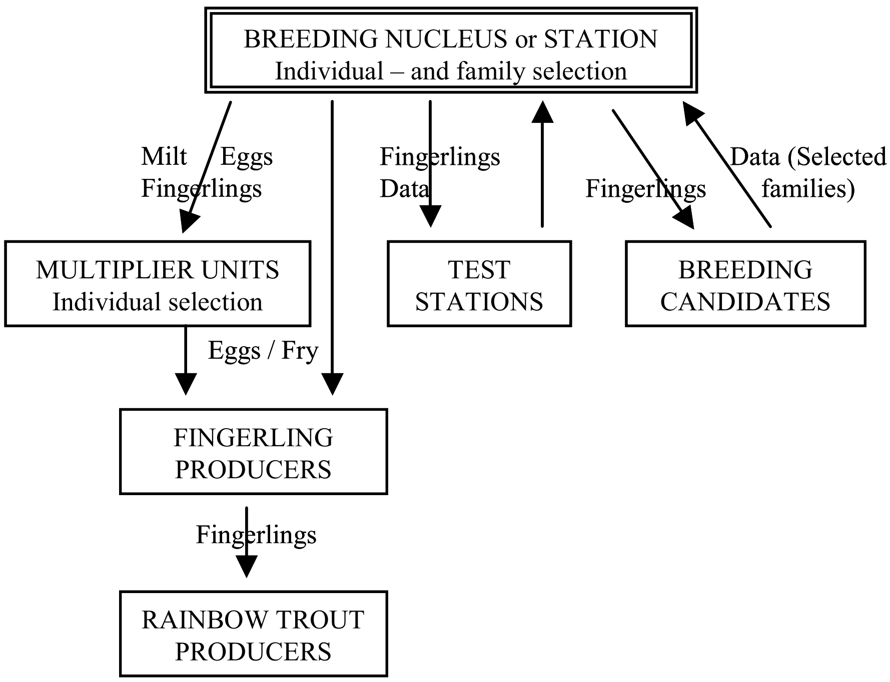

Figure 4. Design and gene flow in a typical selective breeding programme (Gjedrem, 1992 and Gjerde, 2001).

practised. Health of the fish may worsen during the summer months due to high water temperatures, low water quality, and for those fish, which are not, stripped, particularly the males, they die. Saprolegniaceous infections particularly in males are often seen. Infected fish may be treated by a salt bath.

Feeding and nutritional quality of broodstock diets have both direct and indirect effects on egg production, egg size and probably egg quality (Bromage *et al.*, 1992). Because fecundity becomes established early in ovarian development and as yolk proteins constitute >80% of the egg dry weight, food restriction at the beginning of development of the ovaries (vitellogenesis) may reduce egg production (Pepper and Crim, 1996). In addition, egg quality is also correlated to the quality of the diet. Unfortunately, since no growth is expected from brood fish, most farmers sometimes ignore regular daily feeding. Studies showed that feeding broodstock trout at half or three-quarters of their recommended daily ration (0.5% to 1% of body weight per day) throughout the year results in up to 25% reduction in egg production. In addition, significant numbers of the fish fed insufficiently do not spawn in that particular year (Bromage, 1995). Brood fish may show different reaction to seasonal variations in daily feeding rates. For example, Bromage *et al.* (1992) divided the annual reproductive cycle of the rainbow trout in the three 4-month periods and fed the fish at either high (1.0% of body weight) or low (0.4%) rates for different periods (Figure 5). The result clearly showed that those fish fed on high rations for the first 4 months had higher egg production and included a higher percentage of spawned individuals. In contrast feeding high rations during the latter stages of the cycle did not affect egg production, but increased the weight of the broodfish (Table 3).

Broodstocks in commercial farms are fed with ordinary grower pellets of 4-6 mm. As it been mentioned, these fish are not fed regularly in many farms. Some farmers seem to be keener on over-feeding and fattening of the fish. However, it is unlikely that daily feeding rate of around 1% will cause such an undesirable effect.

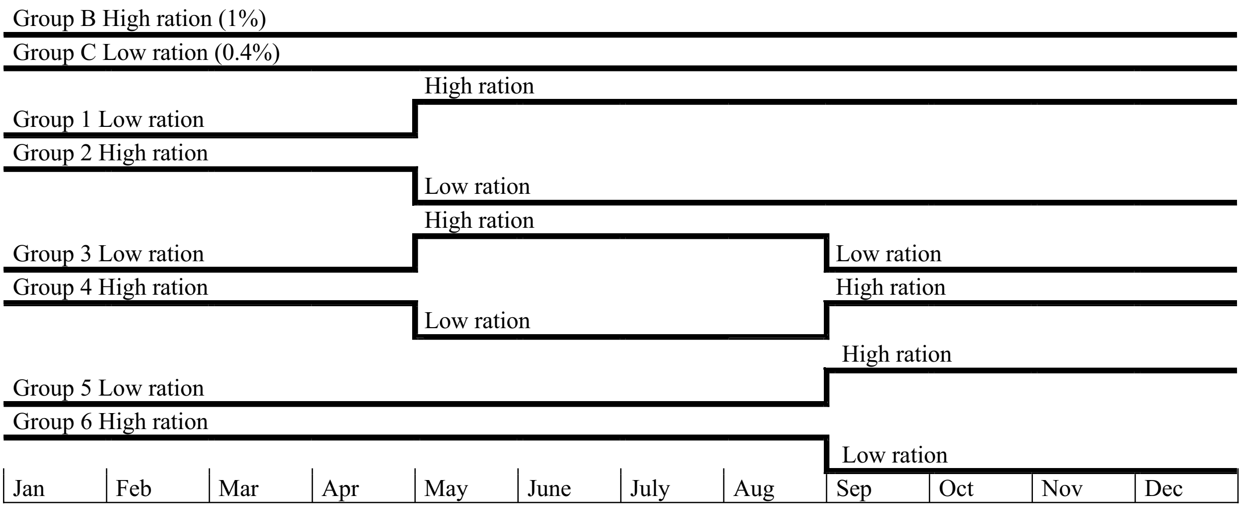

Figure 5. Experimental protocol for feeding 8 groups of rainbow trout with high and low daily rations for different periods of the year. Groups B and C received high and low rations throughout the year respectively, whereas ration levels varied for other groups, for example, Group 3 received low ration for the first period, followed by high and low ration again (Bromage *et al.*, 1992).

Table 3. Effects of high and low daily feeding rates for different periods of the year on spawning rate, egg production and weight of fish (See Figure 3). The last two columns show the number and weight of fish required to produce 1 million eggs (Bromage *et al.*, 1992).

Group	% Spawning	Mean fecundity (egg/kg)	Mean weight (kg)	For 1 million eggs	
				Number of fish	Total biomass (kg)
B	68.2	3,036	1.571	483	759
C	34.5	1,864	0.775	1,555	1,205
1	48.2	2,562	1.321	810	1,070
2	63.6	2,355	0.903	668	603
3	47.3	2,693	1.047	785	822
4	68.2	2,268	1.305	646	843
5	40.9	1,865	1.081	1,311	1,417
6	70.0	3,060	1.163	467	543

Final maturation and spawning time

The rainbow trout originally spawns in spring or early summer, but today's hatchery stocks spawn during autumn-winter period (Table 4). It seemed that autumn spawning has been developed either by selective breeding or as a response to seasonal variations in photoperiod and water temperature. However, there seems to be large variations between stocks and individuals within the same stock. For example, in Turkey, even in the same geographical region, spawning starts in late November and lasts up to April, but the peak occurs during January-February. There is no off-season egg production through selective breeding or photoperiod manipulations in Turkey. Water temperature during the winter months is too low in many of the trout farms and the incubation period is extended and the farm losses Thus, advancing or delaying the time of egg stripping might be one of the major developments for Turkish trout industry. For this purpose, first of all, existing natural variations between individuals and/or stocks should be evaluated. If this would not answer the needs of the industry, then various photoperiod regimes can be developed according to demands.

Temperature would appear to exert a modifying influence on the phasing of reproduction. For example, part of the same stock maintained from September onwards on river water at 1-2°C spawned in March-April, whereas their siblings kept in 10°C spring water temperature spawned in December-February (Morrison and Smith, 1986). Similarly, broodfish transferred to sea cages in October spawned at same time at beginning of January, while their cousins stayed in freshwater spawned one to three weeks later (Okumuş *et al.*, 1997). Low freshwater temperature was the reason for this variation.

In spite of important effect of temperature, it is well known that maturation, ovulation and spawning in rainbow trout are primarily determined by the seasonal waxing and waning of day length, i.e., photoperiod. According to this mechanisms, exposing the broodfish long days of artificial lighting, early ovarian development and in the year, that is, before the longest day of the summer solstice (21^{st} June) would advance spawning time, whereas long days later than summer solstice would produce a delay of spawning. For example, if the fish exposed to long days during January - March and short days after that, 3-4 months advancement in spawning can be achieved (Figure 6). In contrast when exposure of the fish to longest days occur in September, spawning will be delayed up to 3 months. Although various regimes can be developed, at present constant long days (each day) around February – March followed by constant short days (6 hours light and 18 hours dark) during the rest of the year is recommended. This regime may produce 3 to 4 month advances in spawning time, is obviously less complex, easier to manage and practised in commercial farms. The photoperiod manipulations require broodstock tanks that are totally dark, with light provided by white light fluorescent and adjusted weekly or monthly if necessary with 24 hours cycle electronic timers. It is important that light or dark periods should not be interrupted. Once the spawning time of a stock has been modified in this way, the fish must be maintained permanently under photoperiod regime. Usually, after the spawning of a stock has been advanced or delayed 3 to 4 months, then the fish are maintained under same photperiod cycle, which will produce spawning at the same time of the year.

Table 4. Reputed spawning times of farmed stocks of rainbow trout (Bromage and Cumaranatunga, 1988; Okumuş *et al.*, 1997; Kurtoğlu *et al*., 1999).

Location	Latitute	Spawning month(s)
Faroes	62ºN 15ºW	February- June
Central Russia	60ºN 60ºE	March-June
Sweden	60ºN 15ºE	April-June
Denmark	55ºN 10ºE	January-May
South Russia	55ºN 20ºE	December
Northern Ireland	55ºN 07ºW	November-January
United Kingdom*	50.6-56.4ºN 5-1º W	Aug-Sept; Sept.-Oct; Oct-Nov; Oct-Jan; Dec-Feb; Jan-Feb; April-May
Poland	55ºN 20ºE	March-April
British Columbia	50ºN 120ºW	March-June
Northern France	47ºN 02ºE	November-January
Northern Italy	45ºN 10ºE	October- January
Northern Turkey	40º57'N 40ºE	December-February
Washington State	47ºN 122ºW	October-February
Japan	35ºN 135ºE	November- January
Mexico	25ºN 105ºW	January-February
Kenya	02ºN 35ºE	June-August
South Africa	28ºS 20ºE	May-July
Tasmania	42ºS 147ºE	June-August

* : Different strains / stocks

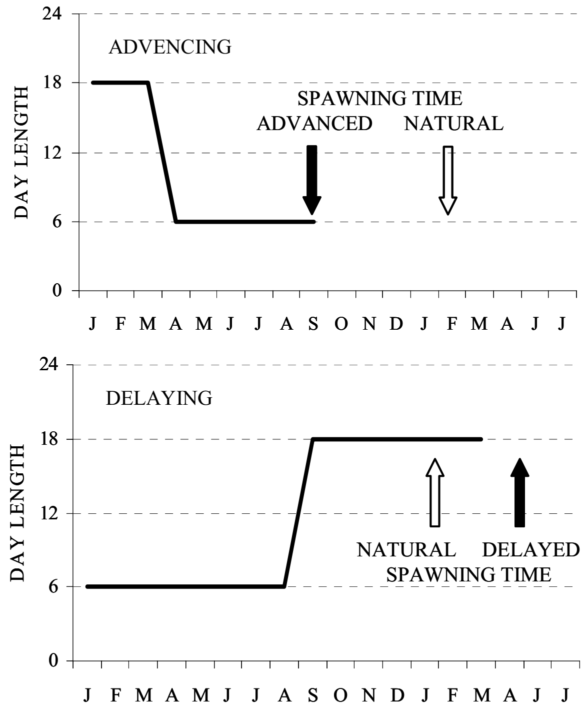

Figure 6. Advancing and delaying the spawning time of rainbow trout broodstock through photoperiod control.

Seed Production

The specialised hatcheries are well developed in many of the trout-producing countries, but this not the case in trout seed production in Turkey. Most of the farms produce their own eggs and fry and those farms without broodstock mainly buy fingerlings rather than eyed-eggs. Only farms with hatchery may buy eggs in case of heavy losses. Eggs and fingerlings are mostly purchased from regional or national suppliers. Only during the early 1990s summer, eggs were imported from Northern Europe, Canada, Kenya and New Zealand. Apart from limited unofficial exports to former Soviet Republics there is no eyed-egg export from Turkey.

Hatchery buildings are simple constructions often designed and built by the farmers. Generally, hatchery-rearing facilities may include broodstock units, incubators and fry rearing troughs or tanks. There are also areas for feed storage, room(s) for workers and an office. Most of the hatcheries take advantage of gravity and choose a location that is downstream from the water source.

In spite of a long history of farming, there are many new techniques and procedures in the rainbow trout hatcheries particularly with regard to off-season egg production, sex control and disease prevention. Figure 7 shows the range of practices and treatment currently available in the modern trout hatchery and since the most of the recent developments have not been employed in Turkish trout hatcheries, it also shows the potential developments in the new future.

Water quality requirements during incubation

It is well known that water quality requirements for salmonid fishes are quite high, and their eggs and larvae require waters of highest quality (Table 5). Although some hatcheries may have good quality surface water supplies, ground water sources are to be preferred because of their year around constancy of temperature and lack of disease or pollutant hazards and silt. Approximately one-third of trout farms in

Table 5. Suggested water quality parameters for rainbow trout hatchery water supplies (Piper *et al*., 1982).

Parameters	Values (mg/*l*)	Parameters	Values (mg/*l*)
Temperature	8-12 ºC	Zinc	<0.05
Dissolved oxygen	> 7	Suspended solids	<3 (eggs) < 15 (rearing)
Carbon dioxide	< 10	Ammonia (NH_3)	< 0.012
Total gas pressure (%)	104-115	Nitrate	0-3.0
Total alkalinity ($CaCO_3$)	10-400	Nitrite	0.015
Total hardness ($CaCO_3$)	10-400	Hydrogen sulfide (µg/*l*)	< 1.0
PH	6.5-8.0	Copper (µg/*l*)	2.0 (SW) 3-4 (HW)
Calcium	4-160	Cadmium (µg/*l*)	0.2-0.8 (SW) 1.3-1.8 (HW)
Chlorine	< 0.15	Lead (µg/*l*)	1-2 (SW) 4-7 (HW)
Manganese	< 0.01	Nickel (µg/*l*)	25-64 (SW) 110-150 (HW)
Iron (total)	< 0.15	Mercury (µg/*l*)	0.1
Phosphorous	0.01-3.0	Water requirement (for each litre of eggs)	1-5 l/min
Aluminium	0.005 (pH<6.5) 0.1 (pH>6.5)		

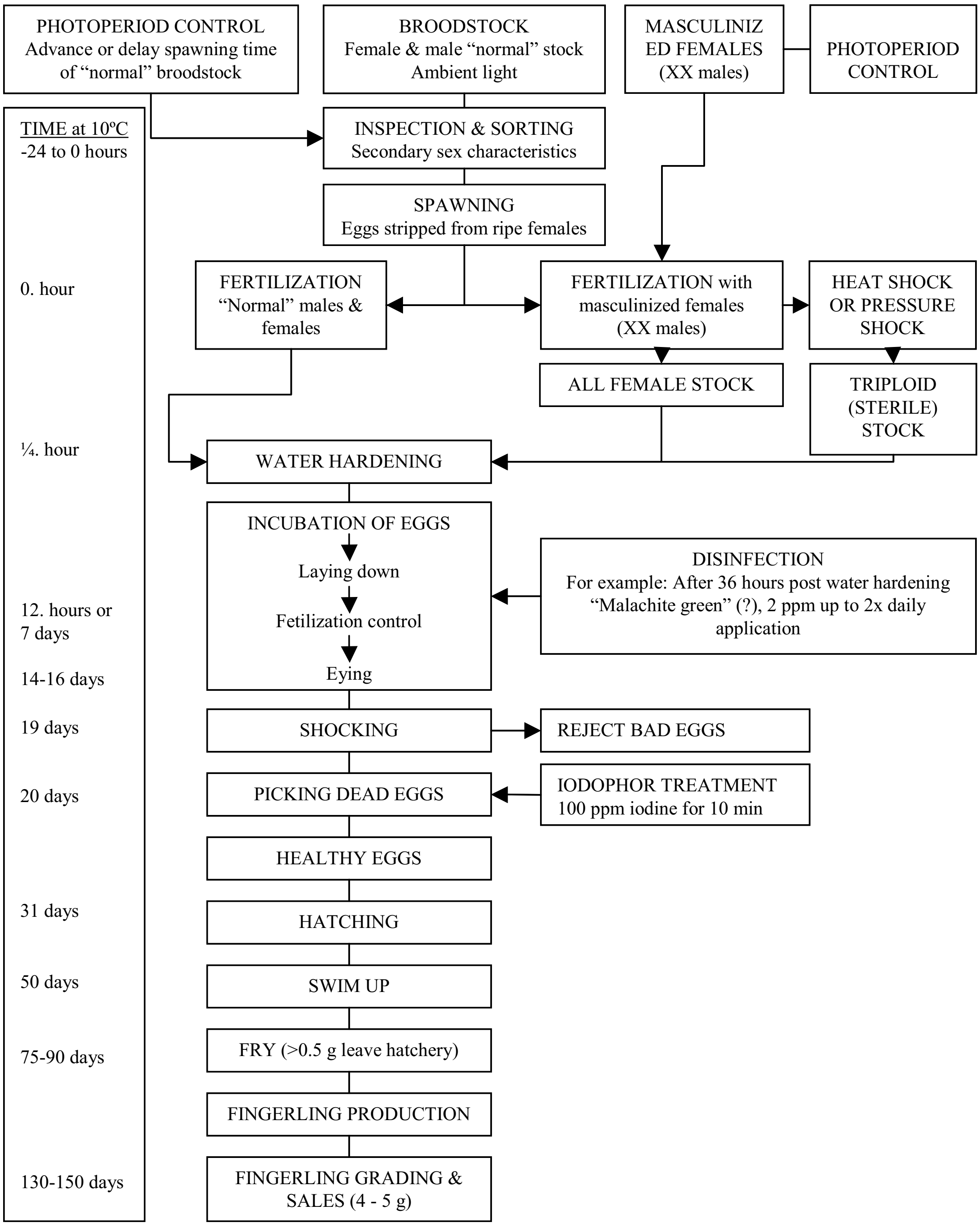

Figure 7. Flow diagram illustrating procedures used in a modern rainbow trout hatchery and timing of development stages at optimum temperature (10ºC). Modified from Bromage (1995).

Turkey are spring water in their hatcheries. In spite of this, losses during incubation and larval period can be quite high due to poor water quality or accidental interruptions. In general there is no regular monitoring, and any water quality improvement activities such as filtration and heating. Thus, low and particularly varying water temperatures extend incubation period, and accumulation of suspended solids increases the losses. Few farms are using spring water, in many cases bringing from long distances.

Although it is not yet used in trout farming in Turkey, as a result of intensifying scrutiny and legislated restrictions in European and North American countries the industry is being forced to consider recirculation technology as an option to traditional fish rearing techniques. In re-circulating systems, advanced water-purification processes are used, allowing a portion or all of the hatchery water to be reused. By implementing recirculation technology, water consumption and effluent volumes may be dramatically reduced. Also, because the heated water is treated and reused, optimized temperatures can be economically achieved despite the outside climate, allowing ideal fish growing conditions to be maintained year-round. Turkish trout producers should consider introduction of recirculation system in the near future.

Stripping, fertilization and incubation

Brood fish are checked regularly 7 to 10 days intervals. However, due to shortages of holding units, ripe fish are not separated previously, but often checked on day of stripping and returned back to the same unit after stripping. Thus some fish might be handled several times. Stripping is performed immediately when they determined to be ripe, but ideally it should proceed about two days after 20% of the brood fish are ready. Timing of striping is very important in relation to ovulation and in fact it is the only factor that has been shown clearly to affect egg quality in rainbow trout. Ideally, eggs should be stripped in four to ten days following ovulation or egg released from the ovary (Needham, 1988).

Artificial spawning involves manually stripping and combining eggs from the female with milt from the male. Stripping is performed in hatchery building or outdoor next to broodstock ponds. In some hatcheries, anaesthetics such as MS-222 (50-100 mg/*l*) or Benzocaine (50 mg/*l*) are used to reduce stress to the fish and simplify the spawning procedures. Those who are not using anaesthetics, generally uses two people stripping. Stripping and fertilization procedures are somewhat different from what is recommended and practised in modern hatcheries (Figure 8). The eggs from 1 to 3 females are collected in a dry plastic pan and at least 2 males are stripped directly over the egg mass. Thus, depending on the sex ratio, males may be stripped more than once not only during the entire spawning period but also during one of the spawning batches. After thorough mixing of the eggs and milt, the mixture is left a few minutes and water is added to activate the sperm. Diluent or fertilization liquid is not used in spite of proven positive effects on fertilization rates and common use in other countries (Figure 9).

Water-hardening occurs within 30 minutes and transferred into incubators. As we know water-hardened eggs can be transported from 1 to 48 hours after fertilization, but it is not a common practice. The total volume or weight of each batch of eggs and their sizes should be measured during this period, but it is mostly ignored. These variables and thus total egg production and fecundities can be determined easily using von Bayer egg counting chart and/or egg enumeration based on displacement method (Table 6 and 7). Eggs are mostly incubated in farm-made wooden - wire trays placed in hatchery troughs built in reinforced concrete or brick. Size of the troughs and baskets may vary but generally the former is around 2-3x0.4-0.5x0.3-0.5 m and the latter 40x50 cm. Recently, fiberglass troughs with aluminium baskets and vertical-flow stack incubators consisting of 20 trays in two stacks are being used. Generally, eggs are placed no more than 2 layers. Water flow in incubators with 5-6 baskets or 20 trays may range from 15 to 25 *l*/min.

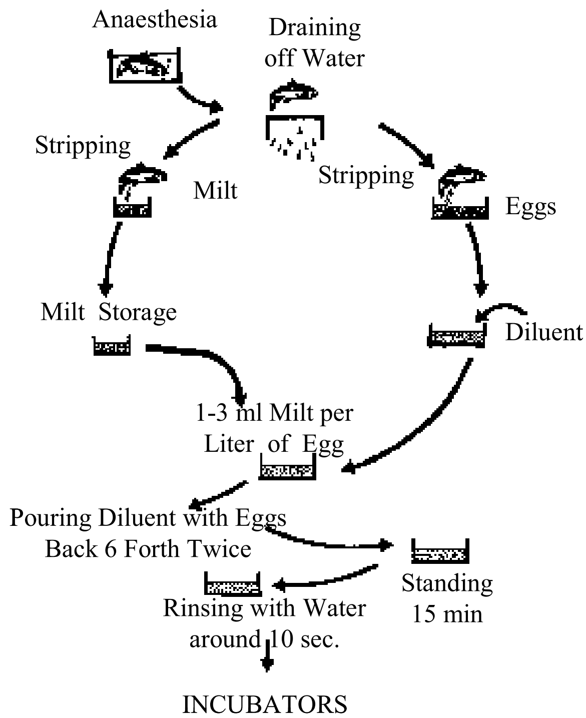

Figure 8. Schematic representation of the artificial fertilization procedure for salmonids.

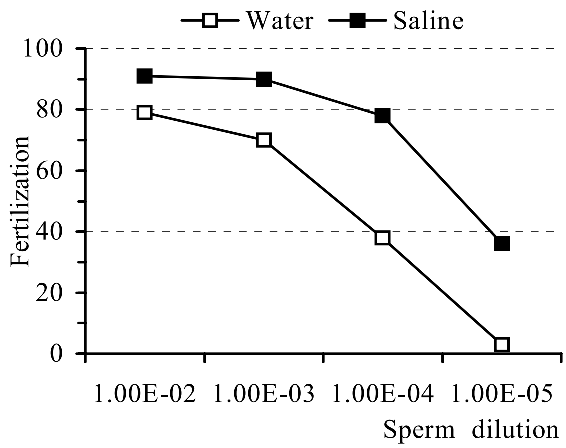

Figure 9. Fertilization (%) of rainbow trout eggs after insemination in saline and water at increasing dilution rates of spermatozoa (10^{-2} to 10^{-5}) (Billard and Jensen, 1996).

Table 6. von Bayer Egg Counting Chart.

No of Eggs in 30 cm	Egg size mm	No of Eggs per		No of Eggs in 30 cm	Egg size mm	No of Eggs per	
		Litre	30 g			Litre	30 g
33	9.0	1,349	51	57	5.3	6,693	252
34	8.7	1,471	55	58	5.2	7,096	268
35	8.5	1,608	60	59	5.1	7,420	280
36	8.2	1,746	66	60	5.0	7,764	294
37	8.0	1,882	71	61	4.9	8,130	307
38	7.8	2,032	76	62	4.9	8,519	322
39	7.6	2,198	83	63	4.8	8,933	337
40	7.4	2,360	89	64	4.7	9,375	354
41	7.3	2,538	95	65	4.6	9,846	372
42	7.1	2,733	103	66	4.5	10,350	391
43	6.9	2,918	110	67	4.5	10,704	404
44	6.8	3,118	118	68	4.4	11,268	425
45	6.6	3,339	126	69	4.3	11,871	448
46	6.5	3,580	136	70	4.3	12,298	464
47	6.4	3,799	143	71	4.2	12,745	481
48	6.2	4,038	153	72	4.2	13,457	508
49	6.1	4,294	162	73	4.1	13,961	527
50	6.0	4,574	173	74	4.1	14,492	547
51	5.9	4,816	182	75	4.0	15,050	568
52	5.7	5,142	194	76	4.0	15,636	590
53	5.6	5,425	205	77	3.9	16,253	614
54	5.5	5,729	216	78	3.9	16,904	638
55	5.4	6,057	229	79	3.8	17,588	665
56	5.4	6,319	239				

Using a 15 or 30 cm V-shaped trough, align the eggs side-by-side throughout the length of the trough, count the number of eggs in the trough, repeat this process 3-5 times to ensure an accurate count, refer to the above chart for the number of eggs per litre.

Table 7. Trout egg enumeration using displacement method.

ml displaced	No. of eggs in 30 g	*ml* displaced	No. of eggs in 30 g	*ml* displaced	No. of eggs In 30 g	*ml* displaced	No. of eggs in 30 g
3.0	523	6.0	262	9.0	174	12.0	131
3.2	490	6.2	253	9.2	170	12.2	128
3.4	461	6.4	245	9.4	167	12.4	126
3.6	436	6.6	237	9.6	163	12.6	124
3.8	412	6.8	231	9.8	160	12.8	122
4.0	392	7.0	224	10.0	157	13.0	121
4.2	373	7.2	218	10.2	154	13.2	119
4.4	356	7.4	212	10.4	151	13.4	117
4.6	341	7.6	206	10.6	148	13.6	115
4.8	327	7.8	201	10.8	145	13.8	114
5.0	314	8.0	196	11.0	142	14.0	112
5.2	301	8.2	191	11.2	140	14.2	110
5.4	290	8.4	187	11.4	137	14.4	109
5.6	280	8.6	182	11.6	135	14.6	107
5.8	270	8.8	178	11.8	133	14.8	106
						15.0	105

Add 25 *ml* of water to a 50 *ml* graduated cylinder, place 50 eggs in the cylinder and take a final reading of the water displaced, the above

Eggs are left undisturbed until eyeing stage and two major problems may occur during this sensitive period. The first one is fungal (*Saprolegnia parasitica*) infection of eggs and the second one is accumulation of suspended silt on and around the eggs. Some hatcheries treat *Saprolegina*-infected eggs with 2-5 mg/l solution of malachite green that should be applied every other day. Fungal treatment of eggs may also be conducted in formalin (a 37% solution of formaldehyde) at a concentration of approximately 1 part formalin to 600 parts water for about 15 min every 1-3 day. Unfortunately, treatment for fungus is not regularly applied in all hatcheries. Thus, due to both fungal infection and silt sedimentation, large numbers of eggs are lost.

Eggs are siphoned from the incubators and allowed to drop about 50 cm into a bucket of water. Weak or undeveloped eggs rupture, turn white and can easily be removed. This application is not practised in Turkey. Instead dead eggs are picked by hand or siphoning after eyes appear. Because of the large numbers of dead eggs, producers sometimes find impossible to pick dead ones. Idophor treatment for eyed eggs after picking is not also a common practice.

After all of the eggs have hatched in conventional incubators, the baskets are removed and fry are left in the troughs. In case of vertical-flow stack incubators, the yolk-sac fry are transferred into fry troughs or tanks before free-swimming stage. Water depth in these units is kept around 15-20 cm.

Use of biotechnology

Although it has no practical applications in rainbow trout hatcheries it Turkey, biotechnology plays an increasingly important role in both increasing the efficiency of trout production systems and reducing or preventing reproductive developments. The following are the major biotechnological applications used in modern trout rainbow trout hatcheries.

Controlled sex determination (i.e. all female stock production): In many cultured fish species, one of the sexes may have better production characteristics than the other (e.g., rapid growth, delayed sexual maturation) or is more valuable in the market. In rainbow trout, males mature earlier than females and during maturation undergo significant changes in appearance, growth and flesh quality (Figure 10). Thus all-female stocks are preferred for large or salmon-trout production. There are two ways of monosex female trout production.

Direct feminization: In this process, fish are treated during early developemnt in the embryo, alevin, or early feeding stage with estrogens, e.g., oestradiol-17β or ethynyl-oestradiol). The recommended procedure for rainbow trout is feeding the fry with feed containing 20 mg of oestradiol-17β for 60-70 days during the first feeding at around 10ºC (Bromage, 1988). This treated stock is not suitable as a broodstock. In addition, there are various objections and some regulatory limitations to the hormonal treatment of fish that are subsequently to be used as a human food. For these reasons, indirect (i.e., utilizing monosex female sperm) is preferred.

Indirect feminization: Due to limitations of direct feminization this procedure is preferred. It is a genetic modification and requires two generations to accomplish (Figure 11). The first step is to produce broodstock males that are genetically female and called "functional", "phenotypic" male or "masculinized" female. For this purpose, mixed sex first feeding fry are masculinized with methyl testosterone, grown to sexual maturity and developed testes and male secondary sexual characteristics. In the second step, these phenotypic males producing only female sperm (XX) are used to fertilize normal eggs and all female offspring are obtained (Figure 11).

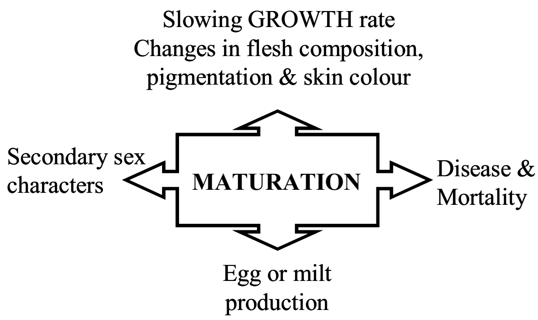

Figure 10. Some morphological and physiological changes accompanying sexual maturation (modified from Bromage, 1988).

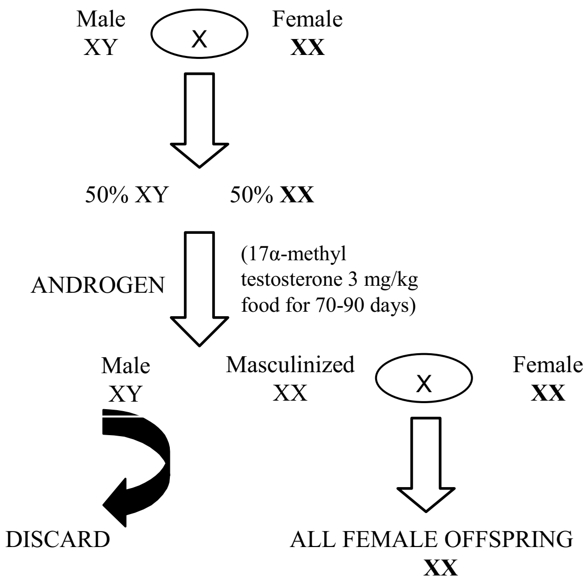

Figure 11. Two stage indirect feminization protocol for rainbow trout. Stage 1: the masculinization of genetically female fish. Stage 2: producing all female stock by crossing milt from masculized females (functional males) and eggs from normal females.

Sterilisation (triploidy): In some cases it may be valuable to produce sterile fish and thus prevent sexual maturation, eg., in salmonids where the development of secondary sexual characteristics results in decreased market value or where it is important to prevent reproductive interaction with wild fish. Although sterilization can also be achieved through hormonal treatment (e.g., 30 mg/kg food 17α-methyl testosterone for 110 days), the most commonly employed method is triploidy which is achieved by subjecting fertilized eggs to shock (e.g., heat or pressure). Thus, the extrusion of the second polar body is prevented and nucleus contains 3n that cannot divide or reproduce successfully. However, the effects of triploidy on reproduction vary greatly with the sex of the fish. Females are most strongly affected. Therefore, triploidy cannot be successfully used in commercial scale unless it is performed on all-female stocks. Temperature is the most widely used for triploid rainbow stock production under farm conditions. Immersing the fertilized eggs into 28ºC for 10 min, beginning 30 min after fertilization induce triploidy rate of over 90%.

Early rearing

This phase of the hatchery operation or seed production start with ponding (i.e. transfer of fry from incubation trays or baskets into troughs or tanks) or first feeding. Shallow troughs and fiberglass tanks of 0.5-1 m^3 are most widely used for fry rearing, and intermediate depth tanks and small concrete outdoor raceways for fingerlings. Although at swim-up stage, it is important that water depth in rearing units should be no more than 15 cm to allow the fry to reach the surface and inflate the swimming bladder, in practice, depths of the units are kept around 50 cm. Water inflow vary considerably within and between the farms, but it should be around 1 l/min for 1 kg fry biomass at the beginning and for 2 kg biomass of 5 g (Bromage *et al.*, 1988).

When fish begin to swim-up to the surface, thin yolk sac has been absorbed. In order to manage the hatchery efficiently, proper feeding practices, growth projections, and keeping and up to date inventory are essential. There seems to be considerable variations on initiation of the first feeding. Some producers start as soon as few fry have shown signs of swimming up, while others begin when 50% of fry try to swim up. The best time is when the fry are willing to feed. Most of the fry feeds are commercial microparticulate compound feeds with particle size range of 0.3-0.6 mm containing 45-50% crude protein and 12-14% lipid. Traditional starter feed mash made from mixture of liver and spleen is not utilized anymore. Feeding is performed by hand with 2-3 hours intervals only during the natural day length, but it can be recommended that they should be fed for 20 hours each day by increasing the day length and using automatic feeders. At this stage of development, fry can consume over 10% of their body weight per day and almost double their weight every week. As the fry grow they are fed with proportionately less ration (around 5-7% of bw) and larger particle sizes, mainly granule or crumbles of 0.8-1.0 mm.

Fry can be maintained indoor until size of 2 g and then moved out to small concrete raceways or fingerling ponds with some kind of shading and/or net covers against birds. Growth rates vary widely between and within the farms, and may reach mean size of 5 g around 130-150 days after fertilization or 100 days after hatching (Table 8). At this stage, fingerlings are graded by hand, or hand operated bar graders.

Table 8. Development of rainbow trout fry and fingerlings (Kurtoğlu *et al.*, 1999).

Age (days)	Mean weights (±SD) (mg)	Daily growth	
		mg	%
40	305±56		
55	644±139	22.6	7.4
70	1,072±254	28.5	4.4
85	1,644±496	38.1	3.6
100	5,140±167	233.1	14.2
112	9,000±289	321.7	6.3
118	10,500±343	250.0	2.8
133	19,200±541	580.0	5.5

Survival during incubation and early rearing stages

The primary objective of hatcheries is to produce the maximum number of the highest quality eggs from existing broodstock. Good quality eggs are usually distinguished by their high survival rates during incubation and as hatched fry, but in addition they must also yield healthy fry with good growth characteristics (Springate and Bromage, 1983).

Egg quality is usually defined in terms of survival by hatchery managers and scientists. Fetilization rate can be determined 10-12 hours after fertilization by immersing a sample of 20 eggs in a clearing solution (water: methanol: glacial acetic acid, 1:1:1) and counting the number of eggs with cleavage furrows (Bromage and Cumranatunga, 1988). Alternatively, it can be determined after 7 days by immersion of eggs in clearing solution and the number with a neural (white) streak.

Unfortunately, hatcheries are not keeping accurate production, development and survival records. In addition, there are not any nation-wide surveys. Thus, our knowledge on survival rates is quite limited (Table 9). Incubation and early rearing survival rates are highly variable. Springate and Bromage (1983) recorded fertilization and hatching rates of 90% and 70%, respectively but subsequent survival up to four months of age of only 35-40%. As it has been seen in Figure 12, Bromage and Cumranatunga (1988) recorded highly variable results ranging from 0% fertlization to 85% survival to the age of four month. These observations indicate that there is multitude of intrinsic (genetic, physiological) and environmental (diet, water quality) factors that should be optimized. The principal causes of the losses can be summarized as:

- Timing of stripping or over-ripening of the eggs,
- Suboptimal water quality (low and high water temperatures, silt accumulation),
- Feeding and nutritional states of both broodstocks and fry (particularly in sufficient feeding),
- Other husbandry practices,
- Gas bubble problems
- Parasites (*Ichthyobodo* or *Costia, Ichthyophthirius*=Ich or white spot, *Saprolegnia*) and diseases (IHN, *Flavobacterium branchiophila* = Bacterial gill disase, *Flexibacter columnaris, Yersinia ruckeri*)

Table 9. Mean values of brood size, fecundity, egg size, duration of embryonic development stages and, fertilization and survival rates from a few rainbow trout hatcheries in northeastern Turkey (Kurtoglu *et al.*, 1999; Okumuş *et al.*, 1997).

Stages	T°	Day	% Survival
Fertilization	9.0	0	73.7
Eying	8.4	20	65.5
Hatching	9.5	38	63.2
Swim-up	8.1	60	61.7
Incubation period	January – March		
112. Day	17.2	112	60.6

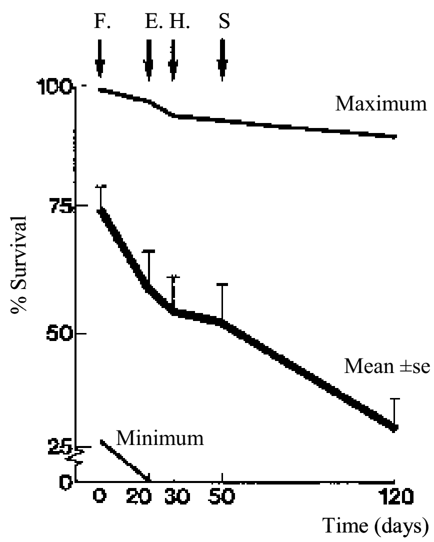

Figure 12. Mean (±sem; n=15) fertilization (F) and survival rates at eyeing (E), hatch (H) and swim up (S) of batches of rainbow trout eggs with the highest and lowest individual values (Bromage and Cumranatunga, 1988).

Future Outlook

World-wide, farmed rainbow trout production is increasing, but progressively with a lower annual rate (Table 1). It likely maintains its position in the top 15 cultured species in the world, and its position in the top 10 species with respect to total value. Particularly in European countries, limitations include the lack of farm sites, increasing concern about the environment and the market prospects. New regulations related to water abstraction and effluent discharge, and control over feed conversion will pose severe constraints to the development of the industry. Thus, in the future little growth is expected in many trout producing countries.

The rainbow trout farming in Turkey, unlike the European and North American countries, has enjoyed very rapid development during the recent years. Lack of strict environmental and quality regulations, declines in capture fisheries, government supports in various ways, suitable sites and cheap labour force are the major factors encouraging this development. Today, in terms of annual production values, Turkish rainbow trout sector has reached a status as those major trout producers of Europe, namely, France, Denmark and Italy without fully utilizing most of the recent biotechnological developments (specialization in hatchery production, selective breeding, recirculating systems, all year around egg production, sterile stock production, product diversity etc). Thus, there seems to be an opportunity that this sector can maintain further developments. However, there are a number of constraints to overcome if it is to realise the full potential and maintain the sustainable development. Environmental regulations, market saturation, economical difficulties and changing consumer tendencies are the foreseeable limitations waiting for the sector. In order to continue the recent success, the industry has to develop well-planned production and good marketing strategies.

Currently, in terms of production-oriented constraints, regular supply of high quality of eggs and fry, collectively known as seed, is amongst the main limitations to further developments. Thus, there seems to be needs for:

- Improvements in the culture conditions and management procedures for the broodstock,
- Initiating long term selective breeding programmes,
- Specialization in broodstock management and seed production,
- Off season egg and fry production,
- Improving egg and fry quality, and reducing losses,
- Monitoring fish health and disease outbreaks, and regulating for egg and fry movements,
- Watching variations in water quality,
- Feeding protocols to meet optimum quantitative and qualitative feed requirements

of the fry and fingerlings,
- Encouraging employment of high technology production techniques such as recirculation systems,
- Establishing close cooperation amongst the producers, government and research organizations.

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