

## HORMONAL SEX REVERSAL

- Ensure >95% reversal of sex to male.
  - Produced in a commercial scale in Israel, Thailand, Taiwan and the Philippines.
    - 2 Synthetic Androgens used in masculinizing tilapia
  - ethyl testosterone
  - methyl testosterone
    - readily dissolved in 80 to 95% ethyl alcohol
- Synthetic anabolic steroids are expensive, difficult to obtain and maybe completely unavailable in some areas.
- However, the hormone is used for a very short time, and several studies have shown that:
- a). it is cost effective and
  - b). testosterone levels in adult fish that have been hormonally sex-reversed are actually very low

## PRINCIPLE OF SEX REVERSAL

To raise the level of male steroid in the bloodstream of sexually undifferentiated fry i.e. before they have physically become male or female, which occur 17 to 21 days after hatching

## AGE OF FRY TO BE TREATED

- Tilapia fry most suited for sex-reversal are first feeding.
- It should be of know age (less than 17 days).

## PREPARATION OF HORMONE IMPREGNATED DIET

- A). Selection of nutritional ingredients
- Should be of good nutritional quality and highly palatable
  - Crude protein levels of 25 to 45%, at least half of which is of animal origin, and vitamin and mineral supplements are recommended.

Dry ingredients should be sieved, a 0.6 mm mesh screen may be most appropriate

## Alcohol hormone solution

- It is practical to prepare a concentrated 'stock' solution of the hormone that will subsequently be diluted to a greater volume before mixing with the dry ingredients. The alcohol in the stock solution should 80-95% ethyl alcohol.
- The alcohol into which the stock solution is mixed may be 80-95% ethyl or isopropyl alcohol.

## QUANTITY OF TREATED FEED NEEDED

- 250 to 400 g. of treated feed per 1,000 fry.

## POND FOR SEX REVERSAL

- Temperature should not exceed 32°C.
- The salinity of water must be less than 10 ppt.

## FERTILIZATION

- Manuring may be needed to reduce inorganic turbidity.
- Fertilizer induced plankton blooms may improved the dissolved oxygen dynamics.

## SETTING UP OF HAPAS

- Easy access to hapas is important.
- The hapas should be at least 30 cm above the pond bottom.

## SECURING THE EXCLOSURES

- Hapas should extend at least 20 cm above the water.

Stock solution of hormone: Dissolved exactly 4-6 g. of methyl testosterone in exactly 1.0 liter of 80-95% ethyl alcohol. (This quantity is sufficient to treat approximately 300,000 fry)

## LIST OF INGREDIENTS PER KG. OF DIET

Alcohol – hormone stock solution exactly 10 ml. Ethyl or isopropyl alcohol about 500 ml. Dry ingredients 1,000 g.

## PROCEDURE FOR MIXING THE INGREDIENTS

- a). Prepare the ground and sieved dry ingredients
- b). Mix the hormone-alcohol "stock" solution with the alcohol.
- c). Add the above solution slowly and mix with the dry ingredients.
- d). Allow the alcohol to evaporate at room temperature with no direct sunlight by spreading out the mixture to a maximum thickness of 3 to 5 cm. Mix lightly by hand 2 or 3 times.
- e). Treated feeds can be packed once the mixture feels dry to the touch and all the odor of the alcohol has disappeared.
- f). Store at 4 °C

## COVERING THE ENCLOSURES

- Hapas generally need not be covered, but in some regions protection from predatory birds may be appropriate.

## ACCESSORY EQUIPMENT

- Floating feeding rings.
  - A circular ring with a dia. of 50-80cm is adequate for 10,000 to 20,000 fry in a 3-5m<sup>2</sup> hapa.

## STOCKING DENSITY

- 1,000 to 2,000 graded fry per m<sup>2</sup> of net enclosure.
- 12 fry per liter.

## FEEDING RATE

- 15 to 20% of fish weight daily until fry reach an average length of 15 mm (more or less 0.01 g) with gradual reduction down to 10% of fish weight daily until the end of treatment.

## AGE OF FRY TO BE TREATED

The following are the steps involved in the calculation of feeding adjustments:

- Step 1. Determine the initial total weight of the fish stock (in this case, 9 grams for 3,000 fry).
- Step 2. Compute the initial amount of feed to be given. Initial amount of feed = Total weight of fry stock x feeding rate (in this case, 9 g. x 0.20 = 1.8 g/day).
- Step 3. After one week, make some feeding adjustment. This can be done by computing the total weight of surviving fry through sampling (see counting) and multiply it with the feeding rate.

## DAILY FEED RATIOS ACCORDING TO FISH TOTAL LENGTH

Average Length (mm)	Daily Ration per 1,000 fry (g)	Average Length (mm)	Daily Ration per 1,000 fry (g)
8	2	17	13
9	3	18	15
10	4	19	16
11	5	20	17
12	6	21	19
13	7	22	21
14	8	23	24
15	10	24	27
16	11	>24	30

**COUNTING**

Before treatment, fry are too small and delicate. To get the average weight, (use two to three samples of fish) use the following formula:

Average weight =  $Tw/Nf$

Where Tw = Total weight of fish

Nf = Total number of fish sampled

Weight of fish to be treated

=  $\frac{[Ave. wt. (g)] [No. of fry to be treated]}{1,000 g.}$

- a). Select light colored container with vertical sides and flat bottoms.
- b). Add about 5g of clear water in the container.
- c). Count 500 pcs. fry into each container.

**Example:**

1<sup>st</sup> sample: 500 pcs. = 6.6 g. - 5g (water) = 0.0032g

2<sup>nd</sup> sample: 500 pcs. = 6.4 g. - 5g (water) = 0.0028g

3<sup>rd</sup> sample: 500 pcs. = 6.7 g. - 5g (water) = 0.0034g

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Meant wt. = 0.0030g

One thousand fry to be treated in 1 m<sup>2</sup> hapa

=  $\frac{1,000 \times 0.0030g}{1,000 g}$

Total weight of fry to be treated = 0.003 kg or 3 grams

- d). Add in 3g. of fry to 5g of water then this will be good for one hapa.

**ESTIMATING FINAL NUMBERS**

- It is more convenient and accurate to estimate the final number of surviving fish by dividing the total final weight of fish in the hapa by the average weight of the subsample (about 100 to 150g)

**Example of Estimating Final Numbers**

Bucket with water	=	430 g.
Bucket with water plus fry	=	550 g.
Counted number of fry in bucket	=	480
Weight of all treated fry in bucket	=	4,250 g.
Estimated total number of fry	=	<u>4,250</u>

$(550-430) / 480 = 0.25 = 17,000 \text{ pcs.}$

**TREATMENT DURATION**

- Fry can be effectively sex reversed in 20 days, but occasionally only 95% of the fry develop as phenotypic males. Sex reversed success is more consistent when the treatment duration is 25 to 28 days.

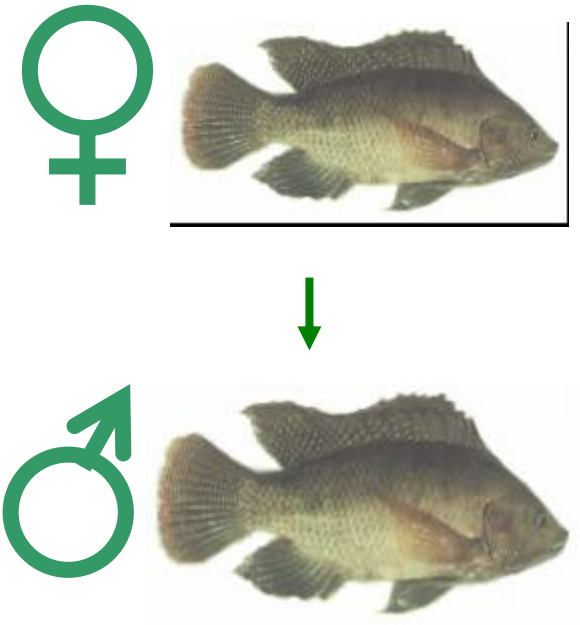
**MINIMUM ACCEPTABLE SIZE**

- After a 25 – 28 days treatment, few fry are less than 14 mm. However, if more than 5% are 13mm or less, those individuals should be culled because 25% of them may be females.

**FACTORS WHICH ARE IMPORTANT TO ENSURE GOOD AND EFFICIENT SEX REVERSAL INCLUDES:**

- a). Age of fry (<17-24 days)
- b). Correct hormone dose (40-60 mg)
- c). High palatability of feed (25-45% protein)
- d). High frequency of feeding (3-6x per day)
- e). Lack of disease
- f). Optimum temperature (<32 °C)
- g). Even size to prevent cannibalism
- h). Control level of natural food
- i). Protect storage of the hormone and hormone treated feed (4 °C)
- j). Optimum fry density (1,000/m<sup>2</sup> or 12 fry/liter)

# SEX REVERSAL OF TILAPIA



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