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## THE FROG CULTURE INDUSTRY, PAST AND PRESENT

By JOHN M. PRIDDY,  
*President, Southern Frog Company*  
*Dumas, Arkansas*

and

DUDLEY D. CULLEY, JR.  
*School of Forestry and Wildlife Management*  
*Louisiana State University*  
*Baton Rouge, Louisiana*

### ABSTRACT

A brief review on the history of bullfrog (*Rana catesbeiana*) culture is covered. Present research programs in frog culture are reviewed.

The need for developing culture techniques are paramount due to the demand for bullfrogs in biological and medical research, education and dwindling of native stocks.

Bullfrogs are reared in 20-foot diameter concrete tanks, housed indoors with controlled temperature. Up to 5,000 young frogs placed in each tank are thinned to about 1,000 as they grow. Crickets, worms, fish, and tadpoles are used for food.

Research needs include the development of a commercial feed, genetically defined strains, simple and effective breeding techniques, methods for disease control, and modification of rearing tanks for more efficient cleaning and temperature control.

### INTRODUCTION

It is not known just when culture, or attempted culture, of the bullfrog (*Rana catesbeiana*) originated. The earliest literature found is an article published by F. M. Chamberlain in 1897, in which he briefly touches on methods of propagation. Additional information on early accounts of frog culture is covered by Cobb, (1903); Meehan, (1913); Herriman, (1933); Anonymous (1938); and Stearns, (1939). These growers collected metamorphosing tadpoles or young frogs from the wilds, placed them in protected ponds and provided them with care and food. In most cases success was marginal due to the inability of growers to prevent disease, control predators, provide sufficient food, and prevent cannibalism. The approach taken today by the authors considers outdoor rearing obsolete and believes that environmental control is essential in every phase of culture, at least until proven differently.

Brief accounts of frog culture in Japan are available. Breeding stock of bullfrogs were shipped to Japan from the United States during the early 1900's. Reasonably successful culture techniques were developed, and the bullfrog has been a favored food in Japan since its introduction. The basic rearing techniques involved confinement and concentrated feeding with fly larvae, silkworm pupae, crayfish, table scraps, etc. Increased use of the bullfrog as food, habitat destruction, high exportation, and DDT contamination has severely reduced the collection and sale of bullfrogs in Japan today. Although the United States still received large imports of froglegs from Japan, most of these legs come from India via Japan. Over-exploitation and DDT contamination in the frogs have seriously threatened the frog population in India today.

With the exception of one known case, all attempts to culture bullfrogs in the United States have been confined to ponds. J. E. Stearns (1939) published the only known account of indoor culture. Although his housing was out-of-doors, he constructed circular concrete tanks, maintained reasonable environmental controls, and was able to successfully raise frogs. Much of the basic research on frog culture conducted at Louisiana State University during 1968-69 has confirmed Mr. Stearns' thinking, i.e., that bullfrogs can be reared successfully, quickly and in large numbers.

In addition to the amphibian research program at Louisiana State University (Culley and Gravois, 1970) two well established amphibian research centers exist. Dr. George Nace (1968), Director of the Amphibian Facility at the University of Michigan maintains colonies of a variety of frogs and other amphibians and has developed culture techniques applicable to bullfrog culture. Dr. T. Kawamura, past president of Hiroshima University has also conducted extensive research with amphibians and has developed many successful frog culture techniques. Although these two facilities have concentrated their work on species other than *Rana catesbeiana*, many practices they now employ are applicable to bullfrog culture. As the amphibian research at Louisiana State University expands, additional species of amphibians will be the object of research.

Much of the biomolecular and medical research in this country utilizes bullfrogs. However, wild-caught frogs can no longer be utilized for the sophisticated level of research now being undertaken. Unless well-defined strains of frogs are developed, much of this research will be terminated. An estimated 50% of all frogs used in education in the United States are now imported due to the increase in demand and dwindling of native stocks. Only by mass production of frogs can we hope to fill the demand and reduce exploitation of our native stocks. The increased pressure on biological supply houses for more frogs has only accelerated the depletion of native populations and suppliers now state that they will be unable to obtain sufficient frogs within the next 10 to 15 years to justify collection and marketing.

Most current attempts at frog culture in the United States still employ outdoor facilities, mainly ponds. Success has been minimal, and the only economical market has been through the sale of these pond-reared frogs for pond stocking purposes or breed stock. The demand for frogs for educational and research purposes cannot be met by these operations, nor can the frogs be produced cheaply enough to realize a profit by selling to biological supply houses.

In 1970, the senior author utilized the techniques of Stearns (1939) to construct the first known indoor bullfrog culture facility. This facility is dedicated to producing well-defined laboratory strains of bullfrogs, with the idea in mind of providing our university and medical research centers throughout the country with high quality research colonies. From this operation much information is being gathered which will open the door to mass culture of bullfrogs, eventually to include marketing bullfrogs for general educational purposes as well as a source of food.

Although the culture program is still in its infancy, there is reason for optimism, as many well-established attitudes about frog culture have turned out to be more myth than truth. Contrary to present attitudes, bullfrogs can be crowded in large numbers, grow rapidly, and seldom exhibit cannibalism.

Techniques associated with developing commercial frog operations are opening the door to a variety of problems that need researched. The availability of a commercial operation for observation has been a great value in directing research activities at the University level. University research in frog culture must remain sensitive and objective to both the needs of the commercial operations and the biological and medical research centers if the frog culture industry is to succeed. The remaining portion of this paper will describe the bullfrog facility at

Dumas, Arkansas, and discuss some of the problems and research needs that have developed since its inception.

## COMMERCIAL CULTURE TECHNIQUES

### HOUSING

In July 1970, the senior author began construction of facilities for the Southern Frog Company. An all-steel building, 30' x 160', was constructed on a 6-inch concrete slab. The walls and ceiling were covered with 2-inch fibreglas insulation. Six 20-foot diameter tanks were constructed using concrete blocks with a 30-inch clearance between each tank. Five of the tanks were constructed with the walls 30 inches high and one tank wall was 36 inches high which permitted the holding of large frogs gathered from the wild.

Single drains, constructed with 6-inch P.V.C., were installed in the tanks 6 inches from the inside wall and flush with the tank floor. The drains connect to a concrete ditch constructed outside and parallel the building.

A metal collar 4" x 6" with a ¼-inch mesh wire brazed across the top was formed to fit inside the drain. The collar slips down into the drain until flush with the tank floor, thus the frogs are retained but the waste in the tank is flushed. In order to control the water level in the tank, any length of 6-inch diameter P.V.C. is placed in the drain and extends upward into the tank. The wire mesh collar is dropped into the extended P.V.C. and the tank filled to the desired depth.

The interior of the tanks were coated with two coats of Davis Weld No. 303 epoxy paint to waterproof the tanks and prevent the frogs from coming into contact with lime which will cause skin erosion.

Water provided by a well located near the building and piped through 2-inch P.V.C., is introduced into the tanks through 1-inch rubber hoses. The tanks are thoroughly washed out twice a day.

Lighting is kept minimal. The building is void of windows and skylights. Lack of sunlight has produced no detrimental effects on the frogs as far as we can tell. Six fluorescent light fixtures are spaced inside the building to provide lighting only when work is done in the building. It has been noted that when the lights are turned on or when there is any activity in the building, the frogs tend to crowd toward the walls of the tank and will pile on top of each other. To alleviate this nervousness, cover has been provided by placing small wooden shelters in the tanks.

The building is heated during the winter months with two 75,000 B.T.U. kerosene forced air heaters. The air temperature is maintained not lower than 70 F. During the summer months air is permitted to ventilate through the doors at each end of the building.

### FOOD

The frogs are fed inside the tanks where they are housed. In order to have sufficient food, a food chain must be set up and maintained. Since the temperature is never below 70 F., the frogs do not hibernate and must be fed during the winter. Crickets and earth worms are raised in the building. Golden shiners and fathead minnows are provided also, but are purchased because of their abundance and low cost.

Up to 20,000 crickets are raised in boxes 2' x 6' x 30". Worms (5,000) are raised in boxes 2' x 12' x 12". A total of 15 boxes of crickets and 5 worm beds are sufficient to handle 5 to 6 thousand frogs when fed in conjunction with fish.

The frogs are fed crickets and worms in the morning. The tanks are kept semi-dry, that is, dry spots are in the tank so the crickets and worms will not drown. At night, after the tanks have been cleaned, about one-half inch of water is put into the tanks, which is sufficient for the fish to survive, but not deep enough for them to swim freely. The fish

are then an easy prey for the frogs. Sufficient fish are placed in the tank so that a few will remain the next morning. Concentrations of 5,000 young frogs to a tank will require two to five pounds of fish daily (225 shiners to a pound) depending upon the temperature.

#### SANITATION

One of the most important aspects of the frog culture is sanitation. Until simple effective disease control methods are worked out, the practice of preventive medicine is a must. Experience justifies the need for extremely clean tanks. After feeding each morning, the tanks and frogs are sprayed down with water. The tanks are then swept with a heavy stock broom and rinsed. Separate brooms are provided for each tank and extreme care is used to keep from cross-contaminating tanks. At night, the tanks are sprayed again, swept out, and water added for the night feeding.

Once a week the tanks are disinfected with Economics Laboratory, Inc.'s Mikrokylene, diluted at the ratio of one-fourth oz. per gallon of water. This is mixed in a 2-gallon garden sprinkler can and sprayed over the frogs as well as the inside of the tanks. Due to the hardness of the water (36 ppm as calcium carbonate) care must be exercised to prevent a build-up of the disinfectant. Prior to disinfecting with Mikrokylene, Clorox was used as a disinfectant and frogs died due to a build-up of clorox in the tanks. Mortality began after using clorox about 2 months, and we lost 20% of our young frogs in some tanks. Since changing to Mikrokylene very little mortality has occurred.

The frogs are inspected two or three times a week for signs of infection or injury. If any frogs are found in poor condition, Mikrokylene is added daily at the rate of  $\frac{1}{8}$  oz. per gallon to control spreading of disease. If necessary the unhealthy frogs are removed.

#### FROGS

At the present time, two types of bullfrogs are being cultured, the laboratory-conditioned frog and the laboratory-reared frog. The laboratory conditioned frog is caught in the wild and brought into the facility. Before the frogs are placed into the tanks, they are dipped in a Mikrokylene solution. The frogs are kept in the tanks until such time as they will adapt to confinement and eat regularly, which takes about 8 weeks. During this period the mortality is about 20%. Most mortality appears to be due to nervous stress. During the first week of confinement, the tanks are disinfected three times. Ample cover is provided to alleviate the nervous stress and activity around the tank is held to a minimum. Food is available at all times.

Laboratory-reared frogs are raised from eggs which are gathered from the wild and brought into the facility for hatching and rearing of the tadpoles through metamorphosis. Detailed records are maintained on each lot of eggs as to the origin of eggs, date hatched, length of time in tadpole stage, date of metamorphosis, diets and medications.

After the eggs hatch, the tadpoles are kept in screen baskets 3' x 1' x 1' which are mounted on 2-inch legs placed in the 20-foot tanks. Each basket contains 200 tadpoles and is placed in sufficient water to have at least 6-inches of water in the basket. The tadpoles are fed a commercial minnow feed (33% protein) and wilted lettuce. In order to eliminate the growth inhibitor (Rose and Rose, 1965) excreted by the tadpoles, the tanks are continuously flushed with water. The flow rate is slow enough to keep from creating a current which the tadpoles would have to swim against. Approximately two water exchanges occur in 24 hours.

After about 4 months, the tadpoles are ready for metamorphosis which occurs naturally. After metamorphosis, the frogs are then placed into the tanks at the rate of 5,000 per tank. The records which were maintained on the eggs are continued on the frogs. As the frogs grow, they are thinned to 1,000 per tank by 6 months of age.

The growth rate of frogs will vary. As the age increases, the size variation will increase. Size variations experienced in the tanks are listed below:

Age	Weight Range	Snout to Vent Range
2 months	10 - 45 g	2.5 - 7.5 cm
4 months	20 - 80 g	5.0 - 8.8 cm
6 months	25 - 110 g	5.0 - 9.5 cm
8 months	40 - 300 g	6.5 - 14.0 cm
10 months	70 - 430 g	7.5 - 16.5 cm

The mortality of tadpoles during metamorphosis has been less than 1% for the senior author. However, the junior author has experienced high mortality (up to 99%) with some populations of wild tadpoles brought into the laboratory. The cause is unknown at the present time, but gives rise for concern in frog culture.

#### MARKET POTENTIAL

Due to the initial installation costs involved in developing indoor frog culture techniques and the sustaining cost to maintain the food chain and operations, it appears that the market will have to be research institutions rather than for food. It does not appear to be economical to compete against the cost of frogs which grow as a by-product with the fish culturist or caught in the wilds. Until such time as some of the problems outlined below are overcome, it will be impossible to culture the bullfrog for the food market.

#### PROBLEMS AND NEW DEVELOPMENTS

One of the greatest needs is for a commercial feed. At the present time, the senior writer is experimenting with methods of placing worms in a container with meat and commercial catfish feed. The worm movement gives the meat and catfish food motion and does attract the frogs. This technique is patterned after Dr. T. Kawamura, director of the amphibian facility at Hiroshima University. The results are incomplete at this time. If a commercial feed could be utilized, the costly food chain could be eliminated and medication could be administered for disease control.

Laboratory breeding techniques need to be improved to insure production of genetically defined strains of frogs. At the present time only limited success is enjoyed by induced breeding. Both writers are researching this problem. Selection for such characteristics as rapid growth, identifying markings, blood characteristics, disease resistance, etc. are necessary if the bullfrog is to become a standardized animal for the research market.

Modifications are needed to improve the rearing tanks in order to decrease the labor required in cleaning and to maintain better temperature control, especially during the winter. During the winter the air temperature is maintained no lower than 70 F, but when the temperature outside the building drops below freezing, the outside cold penetrates the concrete floor and a marked decrease of food intake by the frogs is noted. Some type of heating system needs to be installed to keep the floor of the tanks above 70 F for consistent food intake.

Finally, too little is known about bullfrog diseases, both when in the tadpole and frog stage. Little information is available on methods of disease control or how to administer medications. A sick frog will not actively pursue living food and it is impossible to force-feed medicated food to several thousand frogs, and injections are not practical. Until medications are developed and techniques for administering them are found, extreme sanitation is the only method of disease control.

In spite of the many unknowns, the first year of commercial operation was quite successful. Approximately 3,500 frogs were maintained and growth was considered good. Approximately 10% mortality occurred and most of this was due to disinfectant buildups rather than disease. The

second year of operation will be expanded to house approximately 6,000 frogs. The plant has space for approximately 12,000 bullfrogs under maximum operation. Although the market potential looks promising, we must develop an active advertising program to create the demand before we can operate at full capacity.

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## A RESPIROMETER FOR DETERMINING THE EFFECT OF PESTICIDES AND RELATED POLLUTANTS ON OXYGEN CONSUMPTION OF ESTUARINE FISHES<sup>1</sup>

By PATRICK R. PARRISH  
*Environmental Protection Agency  
Gulf Breeze Laboratory  
Sabine Island  
Gulf Breeze, Florida*

#### ABSTRACT

A continuous-flow respirometer was constructed to measure the effect of pesticides and related pollutants on oxygen consumption of estuarine fishes. The parts of the respirometer in contact with pollutants were constructed of glass and teflon for efficiency in cleaning. Filtered, irradiated sea water of constant temperature and salinity was gravity-fed through ten experimental and ten control respiration chambers in which individual fish were held. Flow rates through the chambers were controlled by stopcocks and measured by flowmeters; dissolved oxygen was determined by the Winkler method before and after water passed through each chamber. Pollutants were metered into the experimental chambers by syringe pump.

#### INTRODUCTION

This research was initiated because (1) little is known about the effects of pesticides and related pollutants on the oxygen consumption of fishes, and (2) the criterion for effect in almost all acute fish/pesticide bioassays in previous years has been death. Recently, investigators at this laboratory have begun to use behavioral and physiological responses of fishes to measure sublethal effects of pollutants (Hansen, 1969; Cop-

<sup>1</sup> Gulf Breeze Contribution No. 135.