Short communication

The utility of homoplastic pituitary glands for spawning induction of the African catfish *(Clarias gariepinus)* in commercial aquaculture in Africa

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Abstract

The use of homoplastic pituitary glands for spawning induction in *Clarias gariepinus* is briefly reviewed and compared with alternative agents. A protocol for spawning catfish with homoplastic pituitaries is outlined. For reasons of practicality and low cost, the use of homoplastic pituitary glands is advocated for use in African countries.

Introduction

The African sharptooth catfish, *Clarias gariepinus,* has been shown to be an ideal species for the development of aquaculture in Africa (De Kimpe and Micha, 1974; Richter, 1976; Clay, 1977; Hogendoorn, 1983; Hecht, 1985). Airbreathing *Clarias* catfish are ideally suited to intensive culture (Huisman and Richter, 1987, Areerat, 1987) which is at present being practised on a small, but rapidly expanding scale, in high density recirculating units in the Netherlands and in intensive pond culture in Africa (Huisman and Richter, 1987; Wray, 1987; Hecht *et al.,* 1988). The technology for catfish farming in South Africa has largely been developed locally and differs significantly from the more intensive style of catfish farming practised in Holland (Hecht and Britz, 1988). In both countries, however, broodfish are induced to spawn artificially by means of hormonal treatment. A variety of natural and synthetic hormones have been used (Table 1), and as a consequence of recent studies in the Netherlands the understanding of catfish endocrinology has been greatly advanced (see review of Van Oordt and Goos, 1987). Most literature on methods of hormonal induction of spawning *Clarias* describes the use of synthetic hormones or carp pituitary suspension (CPS) (Table 1).

Recent reviews and research papers discussing the different methods of spawning induction in *C. gariepinus* (e.g. Hecht *et al.,* 1982; Huisman and Richter 1987; Van Oordt and Goos, 1987) have, however, largely overlooked the use of homoplastic pituitary glands; that is pituitaries taken from the species being hypophysised. Apart from being the most widely used technique of artificial spawning induction on cultured fish species in general, the method is of particular significance in Southern Africa because it is universally used to induce spawning on the more than 30 catfish farms in the region (Uys, 1991).

Despite the wide use of this method, and the fact that the first artificial induction of catfish was performed using *Clarias* pituitaries (Ramaswami and Sundararaj, 1957), literature on the topic is anecdotal and no detailed description of the currently practised spawning protocol has been published. It is the objective of this communication to draw attention to the advantages of this convenient and reliable method of spawning induction and present a protocol for hypophysation of *C. gariepinus* using homoplastic pituitary glands.

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Methods

Following a period of experimentation with various inducing agents (Schoonbee *et al.,* 1980; Hecht *et al.,* 1982; Polling *et al.,* 1987), spawning induction of *C. gariepinus* using homoplastic pituitaries has become standard practise on commercial catfish farms in Southern Africa (Hecht *et al.,* 1988). In our hatchery, the following procedure has been developed for the induction of female catfish by means of homoplastic pituitary glands.

Collection of pituitary glands

The pituitary gland is a distinctive white, pea-shaped organ (± 1 mm diameter in a 1 kg fish) which is situated at the base of the brain between its 2 lobes. The main difficulty in removing the pituitary gland is the heavily ossified catfish skull which is difficult to penetrate without damaging the soft brain tissue and pituitary gland. Various methods have been described to remove the bone surrounding the brain which include the use of bone cutters (Viveen *et al.,* 1985), power drill with hole saw attachment (Schoonbee and Swanepoel, 1988) and hammer and chisel (Hecht *et al.,* 1988). Most commonly the pituitary gland is exposed by removing the gills and skin on the roof of the buccal cavity and then cutting through the bridge of fused basi-occipital and paraspenoid bone forming the floor of the brain case. If these bones are cut anteriorly and posteriorly, it is possible to lift this plate of bone, thereby exposing the pituitary gland and brain. Alternatively, the method of Schoonbee and Swanepoel (1988) may be used in which a circular plug is cut through the dorsal surface of the skull by means of a 45 mm diameter hole-saw. The hole is made through the pariental and frontal bones just in front of the posterior fontanel and is then cut down through the pro-otic and exoccipital bones stopping just short the parasphenoid at the base of the brain. After the saw has been removed, the circular plug of bone may be lifted drawing with it the attached brain and pituitary gland which should be clearly visible. To the practised hand, either of these methods is highly effective and allows for rapid removal of the gland.

It is recommended that pituitary glands only be collected during the summer months when levels of pituitary gonadotrophic hor-

TABLE 1
SUBSTANCES USED FOR HORMONALLY INDUCED SPAWNING OF CLARIID CATFISH

Substance	Species	Author
Desoxycorticosterone acetate (DOCA)	*C. gariepinus*	De Kimpe and Micha, 1974 Hogendoorn and Wieme, 1975 Pham, 1975, Micha, 1975 Richter and Van den Hurk, 1982
Carp pituitary suspension (cPS)	*C. gariepinus*	Hogendoorn, 1977 Hogendoorn and Vismans, 1980 Hecht *et al.*, 1982
Human chorionic gonadotropin (hCG)	*C. gariepinus*	Eding *et al.*, 1982
Chorionic gonadotropin + *Clarias* PS	*C. gariepinus*	Hecht *et al.*, 1982 Polling *et al.*, 1987
Carp PS + hCG	*C. gariepinus*	Schoonbee *et al.*, 1980 Hecht *et al.*, 1982
Clarias pituitary suspension	*C. batrachus* *C. macrocephalus* *C. gariepinus*	Ramaswami and Sundararaj, 1957 Carreon *et al.*, 1973 Van der Waal, 1974; 1978 Schoonbee *et al.*, 1980 Britz and Hecht, 1987 Prinsloo *et al.*, 1989
Progestagen (17-alpha-progesterone)	*C. gariepinus*	Richter, 1985
Pimozide + LHRHa	*C. gariepinus*	De Leeuw *et al.*, 1985 Richter *et al.*, 1987

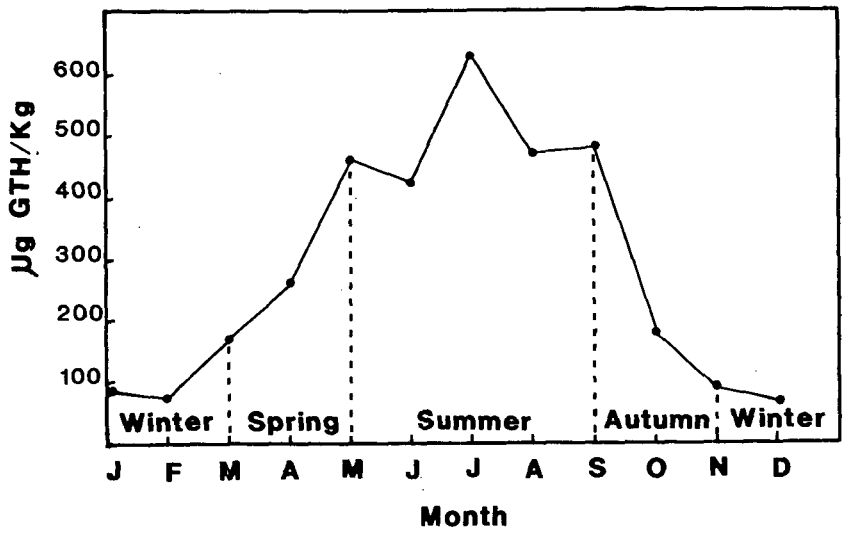

Figure 1
Annual changes in pituitary gonadotrophic hormone (GTH) content of feral female African catfish Clarias gariepinus *in their natural habitat in the Hula Nature Reserve, Israel. Note that the northern hemisphere seasonal cycle applies in this study. Pituitary GTH levels are expressed per kilogram body weight. After Peute* et al., *1986.*

mone (GTH) are high. As illustrated in Fig. 1, the summer pituitary GTH levels are 4 to 6 times that of the winter levels.

Preservation and storage of pituitaries

Whole pituitary glands are usually preserved in 95% alcolhol, then stored in a refrigerator at 2 to 5°C. Alternatively pituitaries may be acetone or freeze dried and stored either whole or in a powder form (Pickford and Atz, 1957; Schoonbee and Swanepoel, 1988). We find the first method the most convenient; however, all three are effective and pituitaries may be stored for 2 to 3 years without a noticeable loss in gonadotrophic activity.

Selection of spawners

Hypophysation is over 90% successful in inducing female catfish to spawn (Uys, 1991) provided the pituitary glands contain sufficient GTH and the ova of the females are ripe. It is therefore important to know how to recognise a gravid female. Catfish display a seasonal gonadal cycle and gravid animals may be found from late spring (October) throughout summer until water temperatures drop in autumn (March/April). Ripe females are easily identified by their distended bellies and the genital papilla is usually red and slightly swollen. The ripeness of the ova may be confirmed by inserting a 2 mm glass canula into the genital papilla and gently sucking a few ova into the tube. Ripe eggs have a firm, translucent appearance and a diameter ≥ 1mm. Their colour is usually green; however, shades of brown or red are commonly observed. If the ova are yellow and opaque with a "runny" texture, reabsorption has begun and it is too late to attempt spawning induction. This usually occurs in late summer or autumn following a temperature drop greater than 5°C lasting more than a week.

It is not possible to judge externally whether male catfish have developed testes; however, if gravid females are present in a body of water, viable sperm should be present in the males. Males are not hypophysised since viable sperm is obtained from their excised testes without any hormonal treatment.

Pituitary dosage

In the practical fish farming context, the dosage to be administered is most conveniently measured in terms of whole pituitary glands. The number of pituitaries used in each dose is dependent on the relative weights of the donor and recipient animals, and the time of year the pituitaries were collected as the gonadotropic activity of feral catfish pituitaries varies seasonally (Fig. 1). For donor and recipient fish of equivalent weight a single homogenised pituitary gland collected during the summer months is sufficient to induce spawning of a gravid female. However, if the pituitaries were collected in spring or early summer the dosage is increased to 1,5 glands per spawner due to the lower gonadotropic activity of the pituitaries (Britz and Hecht, 1988).

Preparation and administration of pituitary dosage

In a commercial hatchery, an average of 4 to 8 females of 1 kg each are hypophysised in a batch. The appropriate number of pituitary glands are removed from storage in alcohol and placed on a paper towel for a minute to allow the alcohol to evaporate. The pituitary glands are homogenised together in a tissue grinder with a small volume of sterile water (±0,5 mℓ), and the homogenate then further diluted with sterile water so that each fish will receive 1 mℓ of solution. The pituitary solution is drawn into a hypodermic syringe (5 or 10 mℓ volume) and injected intramuscularly next to the dorsal fin. A single injection of *Clarias* pituitary suspension is administered to induce spawning and no primer or follow-up doses are required.

TABLE 2
LATENCY TIME (TIME BETWEEN HYPOPHYSATION AND STRIPPING OF FEMALE BROODFISH) IN RELATION TO TEMPERATURE. AFTER VIVEEN *et al.*, 1985

Water temperature (°C)	Latency time (h)
20	21
21	18
22	15,5
23	13,5
24	12
25	11
26	10
27	9
28	7,5
29	7

The latency time between hypophysation and spawning is temperature dependent (Table 2).

Hypophysation stimulates vigorous activity and aggressive behaviour and the injected females are thus placed individually in covered holding tanks to prevent them injuring one another or jumping out of the containers. When the estimated latency time has elapsed, the female is examined to check the condition of the ova. If ova are spontaneously extruded from the genital papilla, the fish is ready for stripping. If not, it is returned to the holding tank and the process repeated hourly until the ova flow freely.

Discussion

Despite the paucity of aquaculture literature relating to the hypophysation of catfish it has proved to be the most effective means of spawning induction for the commercial catfish farmer in Africa. Although a trend exists to develop alternative synthetic substances for the induction of spawning in catfish and other cultured fish (see reviews of Jhingran and Pullin, 1985; Van Oordt and Goos, 1987), hypophysation remains the most common technique used to induce spawning in a number of cultured species. Examples include the common carp *(Cyprinus carpio)*, the Chinese carps *(Ctenopharyngodon idella, Hypothalmichthys molitrix, Aristichthys nobilis)*, Indian major carps *(Catla catla, Labeo rohita, Cirrhinus mrigala)*, mullet *(Mugil cephalus)*, tench *(Tinca tinca)*, pike *(Esox lucius)*, and wels *(Silurus glanis)* (Singha *et al.*, 1974; Liao 1976; Horvath and Lukowicz, 1982; Jhingran and Pullin, 1985). The first recorded use of homoplastic pituitary glands for the spawning induction of *Clarias* species was by Ramaswami and Sundararaj (1957) on *C. batrachus* and subsequently *C. macrocephalus* and *C. gariepinus* were first induced to spawn using the same method (Carreon, 1973; Van der Waal, 1972; 1978). *Clarias* pituitaries have also been shown to be effective in the spawning of other species such as the common carp *(Cyprinus carpio)*, grass carp *(Ctenopharyngodon idella)* and butter catfish *(Eutropius depressirostris)* (Schoonbee and Prinsloo, 1986; Prinsloo and Schoonbee, 1988).

Despite the utility of using homoplastic pituitaries to hypophysise catfish, endocrinological research efforts have largely been directed towards the development of synthetic hormone

analogs for the induction of spawning (Table 1). The reason for this is probably that hypophysation is regarded by the scientific community as a relatively crude technique, because pituitaries are usually not bioassayed and may vary in gonadotropic activity resulting in possible uncertainties with respect to dosage. In practise, however, variability in pituitary gonadotropic activity is not a serious problem since broodfish do not require a precise dosage of pituitary gonadotropin, but simply sufficient to trigger the ovulation process. The recommended hypophysation protocol thus prescribes a dosage which, in effect, is slightly higher than the minimum required to induce spawning, to make allowance for any variability in the levels of pituitary gonadotropin. The administration of this slight overdose has no detrimental effect on the fish or its spawning success. If greater accuracy with respect to pituitary dosage is nevertheless desired, a simple bioassay technique is available to determine the potency of pituitary extracts (Yaron *et al.*, 1982).

The use of *Clarias* pituitary suspension for spawning induction holds several advantages over other methods for the commercial catfish farmer. Firstly, when hypophysation is performed on a regular basis by experienced personnel using gravid females the method is highly reliable and broodfish seldom fail to spawn. Secondly, the single dose hypophysation protocol is simpler than other methods described which require multiple doses of synthetic hormones to induce spawning (e.g. Schoonbee *et al.*, 1980; Hecht *et al.*, 1982; Polling *et al.*, 1987). Furthermore, the method is cheap and convenient since the culturist is in a position to be self-sufficient in the provision of pituitaries, which are obtained from fish slaughtered for market or from males sacrificed during spawning. Independence in the provision of hormones is particularly important to fish farmers in African countries, for whom commercial hormone preparations are expensive and often unobtainable due to foreign exchange controls. For the foreseeable future therefore, hypophysation of broodfish using homoplastic pituitaries would appear to be the most viable option for the commercial catfish culturist in Africa.

Acknowledgements

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Errata

Vol **17**(2) 147-154.
At-site flood frequency analysis for Thailand by HN Phien and N Laungwattanapong
Page 148, column 1, line 7:
....distribution for $\mathbf{b = 0}$....should read...$\mathbf{b \neq 0}$.

Vol. **16**(4) 227-236.
Methods to convert American class A-pan and Symon's tank evaporation to that of a representative environment by HH Bosman
Page 228, Table 2, column 2:
5,4586 should read **-5,4586**

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Grabow, WOK, Coubrough, P, Nupen, EM and Bateman, BW (1984) Evaluations of coliphages as indicators of the virological quality of sewage-polluted water. *Water SA* **10**(1) 7-14.

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