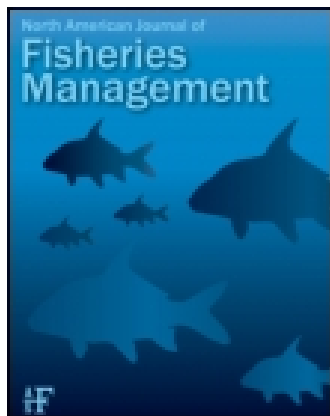


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Efficacy of Identifying Stocked Crappies in a Tennessee Reservoir through Oxytetracycline Marking

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Abstract.—Oxytetracycline (OTC) immersion was used to identify black-nosed crappies, a morphological variation of black crappie *Pomoxis nigromaculatus*, stocked into Normandy Reservoir, Tennessee, during fall 1997. The technique effectively marked 97% of the treated fish. Analysis of one otolith per fish by one reader successfully identified 98% of marked and unmarked fish in a blind test. Marks were formed before annulus formation and were not obscured by annulus-related autofluorescence, suggesting that OTC can be effectively used late in the year (October and November) in Tennessee.

Tetracycline antibiotics have been used to mark fish in age validation research and stocking evaluations for several species (Scidmore and Olson 1969; Lorson and Mudrak 1987; Babaluk and Craig 1990; Heidinger and Brooks 1998). The Tennessee Wildlife Resources Agency (TWRA) currently stocks about 1.5 million crappies *Pomoxis* spp. annually into Tennessee waters (T. Churchill, TWRA, personal communication). This stocking program has not been evaluated, and marking with tetracycline chemicals offered a powerful evaluation tool.

Conover and Sheehan (1994) successfully marked 88% of juvenile black crappies *P. nigromaculatus* with the oxytetracycline (OTC) immersion technique outlined by Brooks et al. (1994). We evaluated this technique as a means for identifying black-nosed crappies (BNC), a morphological variation of the black crappie characterized by a predorsal stripe (Etnier and Starnes 1993), that were stocked during October–December 1997 into Normandy Reservoir, a 1,307-ha tributary impoundment on the upper Duck River in south-central Tennessee.

Methods

Black-nosed crappies ($N = 40,000$) were marked by means of the OTC immersion technique outlined by Brooks et al. (1994). The fish were held for 6 h aboard hauling tanks in a solution

consisting of 500 mg OTC/L and 300 mg sodium phosphate dibasic buffer/L. The pH in the holding tanks was stabilized at 6.8–7.0. After the immersion period, BNC were stocked into Normandy Reservoir at several locations. Stocking dates, numbers, mean total length of fish, hauling tank densities, and water temperatures are reported in Table 1. Although BNC were marked on two separate dates, we considered both groups as one observation in assessing marking efficacy.

Although BNC occur naturally in some Tennessee impoundments and can pass the trait to future generations (Etnier and Starnes 1993), no crappies possessing the predorsal stripe had been observed in Normandy Reservoir between 1990 and 1996 (Sammons et al. 1997). Black-nosed crappies had been stocked into Normandy Lake during 1996; however, individuals of this cohort were not sexually mature during spring 1997. Consequently, all age-1 BNC collected during 1998 were assumed to be stocked fish and the predorsal stripe was used to identify crappies that were immersed in OTC.

Age-1 black crappies and white crappies *P. annularis* were collected in cove-rotenone samples in August 1998, and sagittal otoliths were removed from all fish ($N = 97$). Otoliths were cleaned to remove connective tissue that could cause autofluorescence during viewing and then mounted dorsal side down on microscope slides with cyanoacrylic cement (Secor et al. 1991). Age-1 white crappie otoliths in the cove samples and age-1 black and white crappie otoliths from previous years were used as controls in the viewing process.

Otoliths from all BNC and 33 control fish (one sagittae/fish, $N = 100$) were arranged as a blind test and viewed by one reader using a Nikon Optiphot-2 compound microscope equipped with a 100-W ultraviolet light source and a Nikon B3-A filter cube (450–490 nm excitation filter, a 515-nm barrier filter, and a 510-nm dichroic mirror). Otoliths were wet ground with 600-grit sandpaper to aid in mark detection.

Results and Discussion

Conover and Sheehan (1994) reported unreadable sagittae in 3% of their study fish. Unreadable

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TABLE 1.—Stocking dates, number of fish stocked, mean length (mm), hauling tank densities (g/L), hauling tank, and reservoir water temperatures (°C) for oxytetracycline-marked black-nosed crappies stocked into Normandy Reservoir during late fall 1997.

Date	Number	Mean length (mm)	Tank density (g/L)	Tank temperature (°C)
30 Oct	10,606	53	24	17.2
2 Dec	29,990	76	24	14.0

otoliths resembled sagittae composed of the vaterite polymorph of calcium carbonate (David et al. 1994). All sagittae used in our analysis were readable and appeared normal. Analysis of one otolith by one reader correctly identified 98% of the otoliths as marked or unmarked. Mark clarity in all marked otoliths was excellent. Only 3% (2 out of 70) of the BNC otoliths (i.e., a mark was expected) were incorrectly scored as unmarked; in each case, the second sagittae was subsequently evaluated by the same reader and no mark could be detected. Due to the clarity of the observed OTC marks, these sagittae were scored as unmarked, which resulted in a marking efficacy of 97% (68 out of 70). None of the 30 control otoliths were incorrectly scored as marked.

Application of OTC during periods of slow fish growth can result in mark formation within the annulus and reduce mark clarity (Conover and Sheehan 1994). The marks in our evaluation were formed adjacent to the annulus on the proximal side and were not obscured by annulus-related autofluorescence. Water temperatures at the time of marking (October–December 1997) were higher in this study (14–17.2°C) than marking temperatures (14–14.5°C) reported by Conover and Sheehan (1994) in southern Illinois during October 1993, suggesting that in regions with longer growing seasons, oxytetracycline can yield quality marks when applied late in the year.

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