

X-Radiographic Observations of Food Passage through Digestive Tracts of Lemon Sharks

BRADLEY M. WETHERBEE, SAMUEL H. GRUBER, AND ANNA L. RAMSEY

*Rosenstiel School of Marine and Atmospheric Science, University of Miami
4600 Rickenbacker Causeway, Miami, Florida 33149, USA*

Abstract.—The time required for a meal to be completely eliminated from the digestive tract of the lemon shark *Negaprion brevirostris* was determined X-radiographically, with barium sulfate as a contrast medium. Two food markers and X-radiography showed that initial voidance of fecal matter began 16–17 h after feeding. Alimentary tracts emptied completely within 68–82 h after food ingestion. Passage of a meal through the digestive tract took substantially longer than in most teleosts. A slow rate of food passage may contribute to the low consumption of food and slow growth that have been observed for the lemon shark.

Rates of digestion and passage of food through the digestive tracts of fishes have been studied extensively because of their importance and usefulness in estimating consumption and production in fish populations (Windell 1978; Fange and Grove 1979). Consumption, evacuation, and absorption of food are factors of fundamental importance in the development of trophic models that ultimately represent energy flow in marine food webs (Talbot 1985). Food passage studies have also played an important role in the rationale for development of efficient feed supply and dietary formulations in captive and cultured fish (Fange and Grove 1979).

Rate and time for complete gastric evacuation are frequently measured in digestion studies, but the time required for food to be completely eliminated from the gastrointestinal tract of fish (total gut passage time) is not commonly reported (Smith 1980). Regular observation of the entire alimentary tract is technically difficult and most values of total gut passage time reported in the literature were measured at low levels of food consumption (Edwards 1973; Hofer et al. 1982). There have been few estimates of gastric evacuation rate and gastric evacuation time for elasmobranchs (Van Slyke and White 1911; Jones and Geen 1977; Stillwell and Kohler 1982; Medved 1985).

Total gut passage time can be measured either by observing the disappearance of food from the digestive tract (serial slaughter, X-radiography), or by observing production of fecal matter at regular intervals. Serial slaughter, the most frequently used method, prevents examination of variation in food passage within an individual and requires the sacrifice of potentially valuable animals. Molnar and Tolg (1960) developed an X-ray method for monitoring the disappearance

of bony and other hardparts from fish stomachs. Similar X-radiographic observations of the movement of a single meal through the alimentary canal of fish have been made by Edwards (1971, 1973), Jobling et al. (1977), Grove et al. (1978), and Ross and Jauncey (1981), all of whom used barium sulfate (BaSO_4) as a contrast medium.

Our study was conducted to obtain minimum and maximum values for transit time of food within the alimentary tract of the lemon shark *Negaprion brevirostris*. Experimental lemon sharks were fed at varying ration levels to establish criteria for a fecal collection schedule and for comparison of total gut passage times between elasmobranchs and teleosts.

Methods

Lemon sharks caught near Lower Matecumbe Key, Florida, were transported to the laboratory and housed in a 4,600-L indoor aquarium (see Gruber and Keyes 1981). They were maintained at 25°C and 30‰ salinity under a simulated natural photoperiod (12 h light : 12 h darkness) for a minimum of 2 weeks before use. Lemon sharks were fed to satiation three times weekly with blue runner *Caranx crysos*; 11 healthy animals (1.4–1.7 kg) that fed actively and without hesitation were selected, marked, and used in each of the ration level experiments. After a 96-h starvation period, lemon sharks were fed blue runner fillets containing one 2-g BaSO_4 gelatin capsule per 50 g blue runner. In three successive experiments, lemon sharks were fed at ration levels of 0.67%, 1.6%, and 2.7% body weight per day (% BW/d). All were anesthetized with tricaine (MS222, 1:5,000, Sandoz) and X-rayed at 3-h intervals from 65 to 80 h after feeding. No lemon shark was X-rayed more than once in a 12-h period and, in

all but four instances, they were X-rayed only once during an entire ration level trial. X-radiographs were made of both the dorsal and lateral aspect of each fish at 49 kV and 5 mA, and images were recorded on Trimax XDL film (3M Corporation) housed in an enhancement cassette (Kodak, 17 × 7 cm).

In a pilot study, 17 lemon sharks (0.9–1.7 kg) were each fed a 50-g meal of blue runner fillet, which included 2 g BaSO₄. These fish were X-rayed at 14, 16, 18, 30, 45, 48, 65, 74, 77, 80, 82, and 87 h after feeding to obtain a preliminary estimate of the range of total gut passage times. Preliminary studies were also conducted to establish the minimum BaSO₄ concentration and X-ray dosage that would yield radiographs of sufficient quality. All lemon sharks voluntarily consumed the desired ration and resumed normal swimming activity when returned to the aquarium at the end of each experiment. Experimental animals recovered from short-term experimental stress and voluntarily consumed food 96 h after having been X-rayed.

To test for possible alterations of total gut passage time caused by BaSO₄, four lemon sharks were fed a 1.6%-BW/d meal of blue runner dyed red with erythrosin. In these cases, no BaSO₄ was included in the meal. Two lemon sharks were killed 60 h and two 80 h after feeding, and food location in the alimentary tract was recorded. This allowed a comparison between the location of BaSO₄ and location of dyed food in fish that had been fed meals of equal size.

To determine minimum gut transit time, blue runner fillets (50 g) were either marinated in erythrosin dye or marked with activated charcoal and fed to lemon sharks kept under constant temperature and salinity, as in the radiography trials. These markers radically alter the color and appearance of fecal matter, which is normally pale yellow-green, almost neutrally buoyant, and diaphanous. Feces resulting from an erythrosin-dyed meal were dull red and greater in density, whereas activated charcoal caused the feces to become black and more solidified. Neither marker appeared to be absorbed or to affect the health or appetite of the experimental subjects. Lemon sharks and aquarium water were observed at 30 min intervals for 21 h after feeding, and presence or absence of dyed excreta in the tank water was recorded. Dyed excreta were removed from the aquarium after each observation. Minimum gut transit time was taken as an average of time intervals during which dyed excreta were first ob-

served for each of 11 experimental animals. Lemon sharks ranging in weight from 0.9 to 1.7 kg were used in each dye experiment.

Results

Passage of food marked with BaSO₄ through the digestive tracts of lemon sharks could be observed clearly (Figure 1). In preliminary studies, when a constant 50-g ration was fed to individuals, BaSO₄ was last observed 77 h after ingestion. This suggested a time span during which radiographic examination should be concentrated.

Pooled data from all experimental ration levels represented 59 observations. From 14 to 68 h after feeding, BaSO₄ was present in all 22 observations of alimentary tracts. In 9 of 10 lemon sharks observed 80 to 87 h after feeding, BaSO₄ was completely eliminated from digestive tracts. Although sample size limits interpretation of the extent to which meal size influenced total gut passage time, passage time appeared to increase linearly with ration within the period 68–82 h. If total gut passage time for each ration is taken as the first interval during which at least 66% of alimentary tracts examined showed no BaSO₄, a 0.67%-BW/d meal was completely emptied from the alimentary tract 71 h after feeding, whereas 1.6%-BW/d and 2.7%-BW/d rations were not emptied until after 74 h and 80 h, respectively.

In both marking procedures, dyed feces passed through the digestive tract quickly and feces appeared normal at the conclusion of the observation periods. Onset of excretion of fecal material began 16.3 h after feeding in lemon sharks given blue runner fillets marked with activated charcoal and 16.8 h after feeding in those fed erythrosin-dyed fillets. Minimum passage times did not differ significantly between the two marking methods ($t = -1.6609$; 10 df; $P < 0.05$). These values of minimum gut passage time are supported by the radiographic study, which showed that BaSO₄ required at least 16 h to become disseminated throughout the digestive tract to the end of the spiral valve.

Location of BaSO₄ in lemon sharks fed a 1.6%-BW/d meal of blue runner and X-rayed 60 h after feeding was identical to food position in fish fed a 1.6%-BW/d meal of blue runner marked with erythrosin and killed at 60 h. Dyed food and BaSO₄ had been completely eliminated from both control and test animals 80 h after ingestion. Differences in fish size could not be correlated with differences in total gut passage time observed.

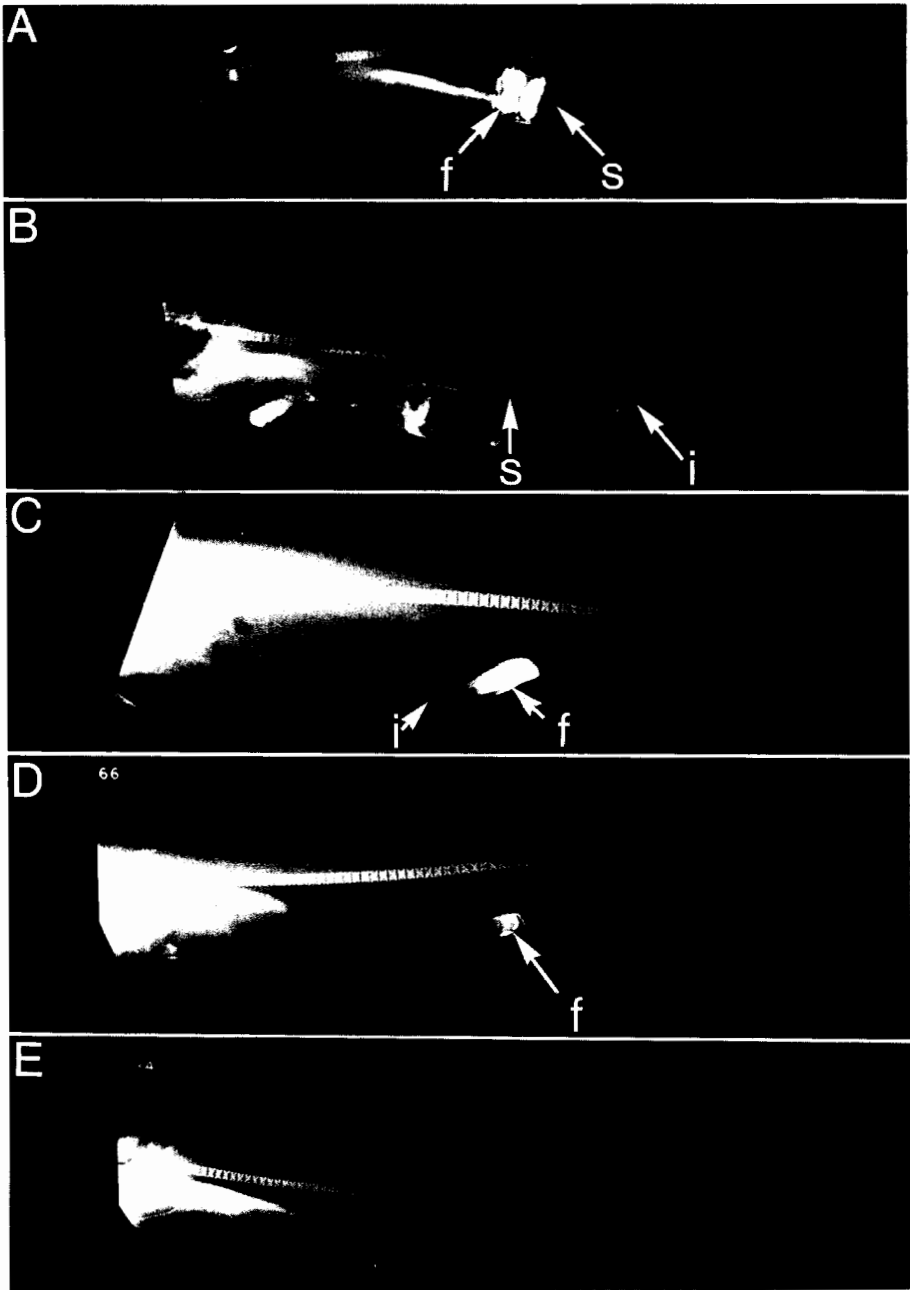


FIGURE 1.—Passage of radiopaque barium through the gastrointestinal tract of lemon sharks shown in X-radiographs taken after ingestion of a meal containing BaSO_4 . (A) Dorsal view at 1 h. (B–E) Side views at 22, 43, 66, and 84 h. Stomach (s), intestine (i), and food (f) are labeled.

Discussion

Based on comparison of sacrificed lemon sharks and X-radiographic observations of the location of BaSO_4 in the alimentary tract of experimental

fish, BaSO_4 and blue runner passed through the digestive tract at the same rate and the marker did not separate from the food during passage. The similar rates of passage of food with and without BaSO_4 support the assumption that BaSO_4 does

not alter digestion rate. Edwards (1971), Jobling et al. (1977), and Ross and Jauncey (1981) observed no effect of BaSO₄ in concentrations up to 25%, on the digestive processes of fish. Therefore, BaSO₄ appears to be an easily measured, non-invidious contrast medium suitable for studies of food transit in both teleosts and elasmobranchs.

Elasmobranchs are susceptible to stress resulting from handling, and the possibility of physiological changes due to stress must be recognized (Cliff and Thurman 1984). Swenson and Smith (1973) have shown that the rate at which food passes through the gastrointestinal tract decreases when fish are subjected to stressful treatment such as handling and forced feeding. Because such stress may result in alterations of digestion kinetics, an attempt was made to minimize handling and time required for anesthetization, transportation, and X-radiography of experimental animals. Lemon sharks were not force-fed, and the period of digestion proceeded undisturbed until capture for X-radiography. Upon return to aquarium tanks, animals maintained continuous swimming movements and fed voluntarily, indicating that they were not heavily stressed while being X-rayed. To obtain an accurate measure of total gut passage time, it would have been advantageous to follow the digestive process to completion in individual lemon sharks, rather than estimating total gut passage time from periodic observations of several fish and pooling results. However, continuous observation of the passage of food through an individual lemon shark would require frequent observations, and results would represent values for stressed animals.

Previous investigations involving gastric evacuation of elasmobranchs have been conducted at single ration levels only, and the effect of increasing ration on gastric emptying for sharks is unknown. Our data suggest that there may be a positive correlation between ration level and total gut passage time for the lemon shark. If such a correlation exists, however, the increase in passage time observed in these experiments was not directly proportional to the increase in ration (i.e., the proportionality constant was less than 1.0), which would imply that the rate of voidance increased as ration was increased. Many authors have reported an increase in the amount of food processed by fish per unit time as ration is increased (Tyler 1970; Beamish 1972; Elliott 1972; Swenson and Smith 1973; El-Shamy 1976; Jobling et al. 1977; Flowerdew and Grove 1979; Gwyther and Grove 1981), thus supporting the present findings.

A total gut passage time of 68–82 h for the lemon shark is slow in comparison to most teleosts, particularly tropical species. Lane and Jackson (1969) reported that voidance was complete within an average of 48.6 h for 20 teleost species examined at 19–25°C. In a summary by Fänge and Grove (1979), total gut passage time averaged approximately 50 h for 48 species of teleosts and 29 h for those tested at 20–30°C. Cortes (1987) found that gastric evacuation in the lemon shark is prolonged (37–41 h) in comparison to values reported for teleosts. Van Slyke and White (1911), Jones and Geen (1977), Stillwell and Kohler (1982), and Medved (1985) have also proposed that complete gastric evacuation takes substantially longer for elasmobranchs than for teleosts.

The metabolic rate of the lemon shark is similar to that of a moderately active teleost predator (Bushnell 1982; Gruber 1984), despite a slow rate of food passage and correspondingly reduced food consumption (Windell 1978). Such a combination of reduced food consumption and a moderate rate of metabolism may mean that the amount of energy available for storage as growth may be relatively low in the lemon shark, resulting in a slow growth rate compared to teleosts. Indeed, low levels of production and growth have been observed for the lemon shark by Gruber (1982, 1984).

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