# Red Grouper Mean Size at Age: An Evaluation of Sampling Strategies Using Simulated Data 

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Abstract. - The influence of sampling protocol on estimation of mean lengths of red grouper (Epinephelus morion) at age was evaluated using computer simulation. Variation in length at age was simulated using platoons of similar length within each year class. Growth of each platoon was assigned using the normal distribution and mean size at age from a previous growth study. Natural mortality (M) was assumed to be 0.2 . Simulated samples were obtained at random or using length stratification from either the population or its fishery. Fishing mortality was assumed to be either a function of fish age or fish length. Estimates of mean length at age were contrasted with the known true mean lengths. The results indicated that reliable estimates of mean size at age requires random sampling with respect to age. Stratification of samples by length biases the estimates of mean length at age. Similarly, samples drawn from size selective gears or fisheries yield biased estimates of mean length at age. Growth models fitted to such data will not generally reflect the mean growth of individuals in the population, and even slight changes in sampling protocol may result in misleading temporal shifts of estimates of size at age.

Determination of mean lengths at age is an important task in many fishery investigations. Such data are often used to characterize growth and can lead to models of growth with age, or studies of densityindependent and density-dependent effects of environmental conditions on population structure and dynamics. The application of age structured methods for the assessment of population status requires estimation of the age composition of the catch. Such estimates may be derived by randomly sampling the catch for age composition, or by similarly sampling the catch for lengths with subsamples stratified by length to obtain age-length keys that can be expanded to the entire catch using the length samples. Both procedures require long term dedication of significant resources. As a consequence, growth models are sometimes used to estimate age composition from length samples of the catch. This convention necessarily assumes that the samples used to construct the model of size at age are representative of size at age for each of the gears encountered in the fishery. Factors such as gear selectivity, bias in the sampling protocol, or variability in actual growth may seriously impinge on the utility of growth models for estimating the age composition of the
catch.
Recent evaluations of the growth of red grouper (Epinephelus morio) of the U.S. Gulf of Mexico documented an important increase in size at age through time (Eklund, 1992; Goodyear and Schirripa 1993; Johnson and Collins ms). This trend led to the development and application of a time-corrected growth model based on the vol Bertalanffy growth equation to estimate the age composition of the catch (Goodyear and Schirripa 1993). The results of application of virtual population analysis methods (Gavaris 1988, Powers and Restrepo 1991) to the resulting catch at age data lead to widely disparate views of the status of the stock (Goodyear and Schirripa 1993). Inadequate estimates of age composition of the catch may have contributed to this unsatisfactory result. Vaughn and Burton (1994) simulated red grouper growth in the presence of size selective mortality to explore methods to reduce the influence of Lee's phenomenon on estimates of the parameters of the Von Bertalanffy growth equation. The present study uses a similar approach to explore the influences of gear selectivity and sampling protocol on the accuracy of estimates of mean size at age using computer simulation.

## Methods

The population simulation model used in this analysis (Goodyear 1989) employed 20 discrete ages with an instantaneous annual natural mortality ( Z ) of 0.2 for all ages. Each year class was further partitioned into 201 platoons with discrete length attributes. The position of a platoon in the distribution of size at age was fixed so that the larger individuals of a year class at age 1 remained larger throughout their lifetime. Mean sizes at age at the beginning of January were assumed to be equal to the values estimated for 1992 from the time corrected growth model of Goodyear and Schirripa (1993), and the coefficient of variation of length at age was estimated from mean backcalculated length at age 5 (0.16). The mean length of individuals of age a in platoon $p, l_{\text {ap }}$, was determined from mean size at age $\left(\mathrm{L}_{\mathrm{a}}\right)$ using the normal distribution and the coefficient of variation of length at age (v) for age 5 , ie.,
$1_{a p}=L_{a}+L_{a} \mathbf{z}_{\mathrm{p}} \mathrm{v}$, where:
$\mathrm{z}_{\mathrm{p}}=$ standard normal deviate for the $\mathrm{p}^{\text {th }}$ percentile of the distribution, and
$\mathrm{v}=\mathrm{L}_{5} / S D_{5}$.
The resulting distribution of size at age in the simulations is given in Figure 1. Although the basic intent of the current exercise is to test the robustness of these estimates, it is less important that they be accurate than it is that they be known with certainty. Systematic sampling-induced biases in the simulated data would also be expected for actual data collected under conditions similar to those evaluated here.

The simulation model permits specification of seasonal fractions of the annual growth, and for the purpose of this study the growth during the first season was set to zero. This convention causes the simulated lengths during the first season to remain constant so that mean size of the fish in the simulated catch for that season are unaffected by growth or mortality. All sampling from the simulation was restricted to this first season to facilitate comparisons of estimated mean lengths and the true underlying means. Student's $t$ was used to test for significant differences between the true and estimated mean lengths at age at the 0.05 level of probability.

Recruitment in the model was specified by year class, and in most simulations examined it was held constant. Several levels of fishing mortality were evaluated, but fishing mortality rates were constant within each simulation Except where size limit and year-class-strength effects were evaluated, the age distribution of the simulated population was stationary when sampled.

The value of fishing mortality for any individual cell (platoon) in the model is the product of the maximum for any cell and the selectivity for the individual cell. The consequence of sampling for growth from fisheries where gear selectivities are age specific and from fisheries where the gears select fish by length were also examined. For both extremes the selectivity ogives examined included ones which increased asymptotically and ones which decreased asymptotically with increasing size or age (Figures 2 and 3). Each also was evaluated for a U-shaped and dome shaped selectivity curve. Other situations evaluated included the impact of size limits when sampling is restricted to the fishery and the
effects of abnormally strong and weak year classes.
Two strategies for sampling were evaluated. The first was simple random sampling without respect the numbers of samples by length. The second strategy was to stratify the samples by 5 cm length interval. Simulated observations of length and age were obtained from either the population or its fishery. In either case, a cell within the population structure or catch was picked at random. It was evaluated for inclusion as a


Figure 2. Age specific proportions of the maximum exposure to fishing mortality evaluated in this study.


Figure 3. Length specific proportions of the maximum exposure to fishing mortality evaluated in this study.
observation based on the ratio of its abundance $\left(\mathrm{N}_{\text {cell }}\right)$ to the maximum abundance of any other eligible cell $\left(\mathrm{N}_{\text {max }}\right)$. This was accomplished by drawing a uniform random number ( R ) between 0 and 1.0 . If the ratio $\mathrm{N}_{\text {cell }} / \mathrm{N}_{\text {max }}>=\mathrm{R}$ the length and age attributes of the cell were considered eligible to be included as an observation; otherwise, it was discarded. This convention caused the sampled cells to be proportional to their abundance in the simulated population. The process was repeated until 50,000 eligible samples had been drawn.

For the random sampling strategy, all eligible samples were retained as observations. For the length stratified strategy, the first 500 samples within each strata were retained as observations. This resulted in 50,000 observations for the random-sampling strategy and about 7500 observations for the length-stratified sampling strategy for each condition evaluated. No error was added to either the age or length attributes to simulate measurement error.

The performance of each strategy was evaluated by plotting error against age where error $=$ (estimatetrue)/true. Estimates of sample means were made only for ages where sample sizes were 10 or greater. Estimates that are statistically different than the underlying true values are denoted with an asterisk in the plots.

A minimum size of 50.8 cm ( 20 inches) total length for red grouper in federal waters of the Gulf of Mexico was established in 1990. One effect of this regulation was to greatly reduce the availability of small fish available to biologists sampling fishermen catches. Consequently, the promulgation of this minimum size regulation caused a change in selectivity of fish to the fishery that is a function of fish size rather than age. The possible importance of this phenomenon was investigated by sampling simulated catches before and after the size limit was imposed. It was assumed for this test that recruitment was constant, fishing mortality ( F ) was 0.25 , the pre-regulation selectivity ogives were constant with age, and no sublegal fish were available for sampling from the fishery after the minimum size was imposed. Both random and length stratified sampling strategies were evaluated.

## Results

Selectivities based on age - Random sampling of the catches derived with the selectivity ogives of Figure 2 with fully recruited fishing mortality of $\mathrm{F}=0.25$ resulted in unbiased estimates of mean lengths at age, but the length stratified samples are strongly biased (Figure 4). Note that the results in Figure 4A1 and 4A2


Figure 4. Error in the estimates of mean length at age for random and length-stratified samples of the catches simulated with the selectivity ogives of Figure 2. Values denoted * are significantly different than the true value ( $\mathrm{p}<0.05$ ).
are for simulations using the selectivity ogives from Figure 2A. Those in 4B correspond to 2B, 4C to 2C and 4D to 2D, respectively. Two of the 48 estimates of mean lengths derived from the random-sampling strategy were statistically different than their true values (Figures $4 \mathrm{~A} 1,4 \mathrm{~B} 1,4 \mathrm{C} 1$ and 4D1). This result is about that expected given the number of sample means and consequently these differences are most likely the result of chance.

In contrast to the results of the random sampling, sample stratification by length resulted in significant differences between the estimates and true values for almost every age for each of the selectivities examined (Figures 4A2, 4B2, 4C2 and 4D2). Inspection of these length stratified sampling results indicates a consistent bias of underestimating the mean size at age of the first or first few ages and overestimating the mean size at age of some or all older ages. Errors of $\pm 10$ percent of the true mean were typical with the maximum error somewhat in excess of about 10 cm about 15 percent of true value at about age 8 to 10 , depending on the selectivity examined.

Length-stratified samples were taken from population simulations at two levels of total mortality ( $\mathrm{Z}=0.25$ and $\mathrm{Z}=0.75$ ) applied to ages 1 through 20 for three recruitment conditions. These were constant recruitment (Figure 5A and 5B), a poor year class equal to 10


Figure 5. Error in mean length at age estimates for length-stratified samples from the population at two levels of total mortality and three recruitment conditions. Values denoted * are significantly different than the true value ( $\mathrm{p}<0.05$ ).
percent of average (Figure 5C and 5D), and a strong year class equal to 10 times normal (Figure 5E and 5F). Constant recruitment was maintained for all but the perturbed year class which was age 4 at the time samples were taken. Overall, the results were biased low for age 1 and high for some or all subsequent ages (Figure 5). Higher total mortality reduced the sample availability for older ages for all recruitment conditions (Figure 5) and reduced or eliminated the bias at older ages. The abnormally weak year class slightly elevated the bias on the corresponding age ( $=4$ ) and reduced it slightly for the subsequent age (Figure 5C and 5D). In contrast, the single stronger than normal year class decreased the magnitude of the bias evident in the estimates of mean lengths of the corresponding age (Figure 5E and 5F).

Selectivities based on length - Random and lengthstratified sampling of the catches derived with the selectivity ogives of Figure 3 with constant recruitment and fully recruited fishing mortality of $F=0.25$ lead to the results of Figure 6. Note that the results in Figure 6A1 and 6A2 are for simulations using the selectivity ogives from Figure 3A, and those in 6B correspond to $3 B, 6 C$ to 3 C and 6 D to 3D, respectively. The ogive of increasing selectivity with size in Figure 3A resulted in


Figure 6. Error in the estimates of mean length at age for random and length-stratified samples of the catches simulated with the selectivity ogives of Figure 3. Values denoted ${ }^{*}$ are significantly different than the true value ( $\mathrm{p}<0.05$ ).
a strong upward bias in sampled mean size in the youngest ages (Figure 6A1 and 6A2). This pattern disappeared with increasing age for both sampling strategies but was replaced with an upward bias in the older ages where length-stratified sampling was employed

Random sampling from the catches produced by the dome shaped ogive of Figure 3B resulted in a pattern of errors that underestimated mean size at age for ages 3 through 6 and an overestimation of mean size at age for ages 8 and above (Figure 6B1). Length-stratified samples from the same catches were relatively unbiased for the younger ages and upwardly biased for the oldest ages (Figure 6B2).

Random sampling from the catches derived with the U-shaped selectivity ogive of Figure 3C produced overestimates of mean lengths for ages 2 through 5 and underestimates of mean lengths for ages older than 6 (Figure 6C1). The length-stratified samples from the same catches were similarly biased for age 2 but less so for the older ages (Figure 6C2). Further, the bias that was present in the estimates beyond age 4 was toward overestimating the mean size, and it tended to disappear in the oldest ages.


Figure 7. Error in mean length at age estimates from samples before and after the imposition of a minimum size in the fishery for random and length-stratified sampling. Values denoted * are significantly different than the true value ( $p<0.05$ ).

Random sampling from the catches simulated with the ogive of declining selectivity with size of Figure 3D resulted in underestimation of mean lengths at age for all ages above age 3 where the number of observations was at least 10 (Figure 6D1). This trend was also true for the length-stratified samples for ages older than 7, but age 1 was underestimated and ages 3-6 were slightly overestimated (Figure 6D2).

Effects of minimum size - The random sampling from the pre-minimum size condition provided unbiased estimates of mean size at age (Figure 7A). However, the mean lengths at age for the youngest ages available from the post-regulation condition were overestimated using the same sampling strategy (Figure 7B). The lengthstratified samples for the pre-regulation condition (Figure 7C) demonstrated bias similar to that observed for this sampling strategy in previous analyses (Figures 4-6). However, the post-regulation sampled mean lengths at age showed the same strong positive bias for the youngest ages available that was observed for the post-regulation random sampling (Figure 7D).

## Discussion

Of the sampling strategies evaluated here only random sampling from gears that were non-selective with respect to fish length produced unbiased estimates of mean size at age, and then only in the absence of size limits. Length-stratified samples and samples drawn from size selective fisheries all provided biased estimates of mean length at age. Although selectivity patterns that were soley a function of the age of the fish did not result in biased estimates of mean size at age, they often produced samples that were strongly biased with respect to the distribution of lengths of fish in the population. This effect suggests that it may be difficult to decide from the length compostion of samples whether they are from length selective sources or not.

The IBP handbook on methods for assessment of fish production in fresh waters provides the following guidance for the conduct of growth studies: the first step is to "procure a sample of fish representative of all the sizes of the species in the population, as far as possible," (Tesch 1970). This advice favors the adoption of length stratification where simple random sampling would provide overwhelming numbers of individuals of similar sizes and few very large or very small fish. Such sample stratification is relatively common in growth studies (e.g. Guteurer and Childress 1990; Hammers and Miranda 1991; Miranda et al. 1987; Newman and Weisberg 1987). Indeed except
a strong upward bias in sampled mean size in the youngest ages (Figure 6A1 and 6A2). This pattern disappeared with increasing age for both sampling strategies but was replaced with an upward bias in the older ages where length-stratified sampling was employed

Random sampling from the catches produced by the dome shaped ogive of Figure 3B resulted in a pattern of errors that underestimated mean size at age for ages 3 through 6 and an overestimation of mean size at age for ages 8 and above (Figure 6B1). Length-stratified samples from the same catches were relatively unbiased for the younger ages and upwardly biased for the oldest ages (Figure 6B2).

Random sampling from the catches derived with the U-shaped selectivity ogive of Figure 3C produced overestimates of mean lengths for ages 2 through 5 and underestimates of mean lengths for ages older than 6 (Figure 6C1). The length-stratified samples from the same catches were similarly biased for age 2 but less so for the older ages (Figure 6C2). Further, the bias that was present in the estimates beyond age 4 was toward overestimating the mean size, and it tended to disappear in the oldest ages.


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The principal bias in the estimates of mean size of the fish at age from the length stratified samples was the direct result of the sample stratification. The first few strata were smaller than the mean size of age 1 and consequently small age 1 fish were favored in the samples; thus leading to a downward bias. As the mean size increased with age this phenomenon subsided and was replaced by a similar phenomenon only in reverse. Where the distributions of size at age overlap, the length strata can contain fish of many different ages. If a limit is imposed on the number of fish sampled per strata then there is a tendency for the more abundant year classes in a strata to dominate among all samples for that strata. Because the process of mortality reduces a year class with time, fast growing younger fish tend to dominate over the slower growing older fish in each strata. This leads to a positive bias in the estimates of mean size by age because the overall effect is to sample relatively more large fish than small fish of a particular age. This same phenomenon is the cause of the effect that the strong year classes had on the estimates of mean sizes seen in Figure 5.

Means of samples drawn from the catches of size selective fisheries were particularly strongly biased. The pattern depended on the selectivity pattern of the fishery and, to a lesser extent, on whether the estimates were from random or length-stratified samples of the catch. Also, the extent of the bias depended on both the selectivity ogive and the sampling strategy. For some selectivity ogives length stratification reduced the extent of the bias in the estimates of mean size at age for some ages compared to the random samples.

Vaughn and Burton (1994) simulated red grouper growth in the presence of size selective mortality to explore methods to reduce the influence of this phenomenon on estimates of the parameters of the Von Bertalanffy equation. Their results suggested that the best parameter values are obtained by including only lengths backcalculated to the most recent annuli when estimating growth parameters for the population. Although, Lee's phenomenon is commonly interpreted as the result of size selective mortality on the mean size at age of the survivors, Ricker $(1969,1975)$ noted that gears which induce size selective sampling also produce this phenomenon. This effect is the result of gradients
in the bias introduced into the estimates of mean size at age by the size selectivity. The results of the present study imply that if samples are length-stratified or obtained from length selective fisheries or sampling gears, it may not be possible to obtain growth model parameter estimates that are representative of the population.

It might be argued that if the intent of the collection of age-length data is to develop a growth model to be used to age fish taken in the fishery, then the appropriate objective is a model of the age-length relation of the catch rather than actual growth. A model that predicted true mean age of the fish in the population at a particular length would be less useful than one which predicted the mean age at length in the catch. If so, then the random sampling strategy would be appropriate, and a different model may be required for each fishery. The bias introduced by length stratification make this strategy generally ineffective.

The major disadvantage of the random sampling strategy is the large numbers of samples that are required to obtain estimates of size at age for the older fish in the catch because of the cumulative effect of mortality. Although I took 50,000 observations in the simulations, only small sample sizes were obtained for the older ages in the catch. Also if the selectivities are not constant with time then annual growth models will also be required. It is probably noteworthy also that if sufficient randomized age estimates of the catch are available to estimate mean size at age for the majority of ages in the catch, then that data would probably reflect the age composition of the catch at least as well as ages assigned from length frequency samples using a growth model fitted to the size at age data.

Application of age-structured methods for stock assessment requires annual collections of age-frequency information for the harvest of each component of the fishery. Given the large sample sizes needed to support models with which to estimate ages from lengths, it seems more appropriate to abandon the use of age-length models and to rely on other methods wherever possible. Probably the most widely used approach is to estimate age composition from random length-frequency samples of the catch and length-stratified samples of the age structure using age-length keys (Ketchen 1950; Hoenig 1987).

In the particular case of U.S. Gulf of Mexico red grouper, the mean lengths of red grouper caught in various components of the overall fishery are clearly variable (Goodyear and Schirripa 1993). Recreational
anglers harvest fish that average smaller than those harvested by their commercial counterparts. The selectivity patterns for the three commercial gear types (traps, bottom longlines, and handlines) are also clearly different.

It is possible that these differences in mean size are the result of different age-specific selectivity patterns. However, it is also possible that the underlying cause is more a function of the size of the fish than its age. Grouper are notorious in the recreational literature for escaping capture by retreating to cover from which they cannot be extracted, and bigger fish are more likely be successful than the smaller ones. Presumably, this would be more of a problem for the lighter gear used by the typical angler than for the commercial fishermen using electric or hydraulically-operated equipment. Such an effect would lead to differences in size selectivity patterns between these two components of the fishery.
 discussion of the source of the samples that lead to the "observed" change: in growth.

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