13

Quantitative Genetic Tools for Development of Superior Brood Stock

Thomas, P. C.

Marine Biotechnology Division,CMFRI, Cochin - 682 018, palahanict@yahoo.com

Success in aquaculture depends on the use of animals with high genetic potential and application of sound management techniques. Though one can bring about improvement in the production performance of a population by environmental manipulations as well as through genetic manipulations, any improvement from the former can not be transmitted to the next generation. The genetic improvement, on the other hand, is inherited by the next generation, and therefore, more important. Any of the different genetic manipulation techniques available can be employed for producing genetically improved brood stock.

In addition to the conventional quantitative genetic techniques like selection and breeding, modern tools like chromosomal engineering (induction of polyploidy, gynogenesis and androgenesis) can also be employed. The most modern technique is genetic engineering where in a desirable gene or set of genes from any source can be transferred into a host animal for producing a transgenic animal with desired characteristics. In farm animals and plants, quantitative genetic techniques like selection and breeding have played an important role in their increased productivity. Although, the plant and animal breeders have conducted scientific breeding programmes on crops and livestock for thousands of years, fish farmers are only beginning to use selection, hybridization and other breeding programmes to improve aquaculture species. Of late, genetics has acquired an important place in aquaculture for producing high yielding strains of fish and shellfish, and for development of disease resistant strains.

Most of the economically important traits in plants and animals are quantitative traits. Quantitative genetics deals with the inheritance of quantitative traits. Quantitative traits are those which are measurable/ quantifiable, and since metric units used for measurement they are also called as Metric traits. They are controlled by many genes which are mostly additive in nature. (In contrast, qualitative traits are controlled by one or a few genes which are non-additive)

In case of additive genes, contribution of each allele (of the same locus or different locus) is additive or cumulative. Contribution of individual gene can vary, and the individual contributions cannot be noticed. Continuous variation of phenotypes is characteristics of quantitative traits. There will be overlapping phenotypes with no sharp distinction between the phenotypes of different genotypes. Genotypes of individuals are indistinguishable from phenotype and hence individuals can not be can be classified into distinct phenotypes as per Mendelian ratios. Instead, many gradations of phenotypes are available leading to continuous variation and bell shaped cure of distribution. In

contrast, qualitative traits show discrete, discontinuous variation, and so individuals can be can be classified into distinct phenotypes as per Mendelian ratios. Study of quantitative traits are made on population basis and quantitative genetic analysis involves description of genetic architecture of the population using statistical procedures. Phenotypic expression of the additive genes of the quantitative traits are very much modified by environment. Genotype sets the maximum limit to which the individual can express the character.

Quantitative genetics is the theoretical basis for all the breeding programmes in animals and plants. It is the logical development of the basic principles of inheritance.

Tools available to the breeder for genetic improvement

Variation

Variation among individuals within the population is the basic and most important pre- requisite for any genetic improvement programme. With out variation no improvement is possible.

In order to study the genetic properties of the population we have to partition the phenotypic variation into component parts attributable to different causes. The phenotypic variation can be divided in to components attributable to the genotype and to the influence of environment. The genotype is the particular assemblage of genes possessed by the individual and the environment is the non-genetic factors affecting it.

Variation is quantified and expressed as variance ($ó^2$). Variance is defined as the average of the squared deviations of individual's value from the population's mean value. The two components of the total variance (phenotypic variance) are the genotypic variance and environmental variance.

$V_P = V_G + V_E + V_{G-E}$

Where,

V_P = Phenotypic variance

V_G = Genotypic variance

V_E = Environmental variance

V_{G-E} = Genotype-environmental interaction variance

The partition of the variance in to its components formulates the question of importance of individual portion in determining its phenotype. The relative importance of the cause of variation means the amount of variation it contributes to the total variation.

Genotype variance: The variance in the population which is due to the difference in the genotype of its members is termed as genotypic variance. The relative importance the genotype as the determinant of phenotype is the ratio of genotypic variance to phenotypic variance (V_G / V_P) is termed as the heritability (h^2).

Environmental variance: It is the variance due to all non-genetic effects starting from feeding to environmental conditions. Environmental variance cannot be eliminated in a population. It plays a major role in determining the phenotypic variance of the population.

Genotype and environmental interaction variance: Apart from the above two components, variance can arise from the interaction between the genotype and environment also. A single genotype

may perform differently in two different environments. It may show good performance in one environment but may be poor in second. The performance of a population may be reduced or increased in different environments.

Genotypic variance consists of additive genetic variance (breeding value) and non-additive genetic variance (Dominance and interaction deviations).

$V_G = V_A + V_D + V_I$

Where,

V_A = Additive genetic variance

V_D = Dominace variance

V_I = Interactions (Epistatic) variance

Additive genetic variance: It is the sum of the effects of all the additively acting alleles that help to produce a phenotype. It is the breeding value. It is an important component because it is the chief cause of resemblances between the relatives, especially with respect to the quantitative traits. This is the portion of the variance which is definitely and surely being transmitted to the offsprings from the parents.

Non-additive genetic variance: These variations are due to the dominance and epistatic alleles, and therefore, consists of the following.

Dominance variance: This is due to the dominance effects of the alleles at the loci. V_D may not be transmitted to the next generation from the parents since the alleles are disrupted during the meiosis.

Interaction (Epistatic) variance: It occurs due to the interaction between the alleles two or more genes. These interactions may be two factor or three factor or so on. The amount of variation due to interaction is rather small; therefore, the breeder can ignore it.

Heritability (h^2)

The heritability is one of the most important properties of the economically important quantitative traits. It expresses the proportion of the total variance attributable to genetic causes.

Heritability is defined as the ratio of genetic variance to phenotypic variance in a broad sense.

$$\text{Heritability } (H^2) = \frac{\text{Genotypic Variance}}{\text{Phenotypic Variance}}$$

It in narrow sense is defined as the ratio of additive genetic variance to the phenotypic variance.

$$\text{Heritability } (h^2) = \frac{\text{Additive genetic Variance}}{\text{Phenotypic Variance}}$$

Heritability ranges from 0-1 or expressed in percentage. It plays an important role in selection programmes for genetic improvement. An important function of heritability in quantitative genetics / animal or plant breeding is for indicating the reliability of using the phenotypic value as a guide to the breeding value. It can be used to predict the genetic gain from selective breeding. It is the property of the population for the trait under consideration, and not of an individual. It is valid for only the given population in a given environment.

Genetic correlations

Economic traits are usually correlated. Because of this, selection for one trait may lead to simultaneous increase or decrease in the other traits. These are called correlated responses and the traits are called correlated traits. This is expressed in terms of correlation co-efficient (r) which ranges from -1 to +1. Correlated response may be positive or negative. It is due to the pleotrophic effect of the alleles. This plays an important role in trait selection for improvement.

Selective Breeding for fish stock improvement:

Breeding is the practical aspect of the genetics. Clear and thorough understanding of the genetic rationale and genetic principles will avoid the inadvertent mistakes during the breeding. Therefore, the breeder should have clear understanding of its pedigree records, genetic parameters like heritability, correlations and of course reproductive biology and behavior of the species before planning a breeding programme. Therefore, breeding studies for the evaluation of the genetic and phenotypic parameters are the essential pre-mediated step. Planning for the breeding programme could be made only after careful consideration of the above parameters. As for example, when there is relatively larger additive genetic variance, simple selection methods like individual/mass selection should yield good progress. On the other hand if non additive genetic variance is predominant, special selective breeding schemes are to be formulated to exploit them. If heterosis is found to be high, cross breeding programmes could be given priority. When 'over dominance' is important for a trait, reciprocal or recurrent reciprocal selection needs to be employed. Genotype environmental interactions of high magnitude calls for developing different strains to suit each of the environments.

Selective breeding means careful selection of superior individuals from the population as parents for the next generation. This is based on the Robert Backwell theory of "like begets like". The breeder selects (saves) the individuals that possess certain desired phenotypes and culls (removes) those that do not in the hope that the selected individuals will be able to transmit their superiority to their offspring, thereby creating a genetically improved population. Thus two elite/best performing individuals are selected and bred to produce the best performing progeny. In animals and plants this lead to production of pure strains and also improvement in production. Selective breeding of fishes is a very useful approach for genetic improvement of cultured fish populations. There are a number of methods, which can be used for selection of superior genotypes for scientific breeding. Careful consideration of the species and trait to be improved are before taking up the selective breeding programme.

Selection of species

Selection of the species is very important aspect of selective breeding programme. While selecting a species the breeder should consider two things, the first is that it should have well established breeding and seed production technology and the second is its economic importance.

Selection of trait

The trait or traits under selection should be economically important, highly heritable, positively correlated and it should be easily measurable. Some economically important traits in fishes are growth rate, body size & shape, meat quality and disease resistance.

Methods of selection :

Selection of the parents for next generation is based on its additive genetic makeup or breeding value. The genetic potential of an individual is judged by the phenotypic performance of the individual in several ways.

Individual selection/ Mass selection

In this method the individuals are selected based on their own performance, viz. on the basis of their phenotypic value. This selection method is very effective when the h^2 of the trait is high. It is usually simple to operate and it yields the rapid response.

Pedigree selection

In this method individuals are selected based on the performance of their parents or grand parents. Pedigree records of the ancestors are considered for selection of parents.

Family selection

Families are selected or rejected as a unit according to the mean phenotypic value of the family. Within the family, variations are not considered. The accuracy of the family selection depends on the heritability of the trait, family size, family type and the variation due to the environment. This is more useful when selection is practiced for less heritable traits like reproductive traits, carcass quality and disease resistance.

With in family selection

In this method, families are considered as a sub population and individuals are selected based on their performance in relation to the mean performance of their family. Individuals with better performance from good families are selected and the others are culled. This method is more efficient than individual selection and family selection. Selection within families would eliminate the non-genetic variation.

Progeny testing

Parents are selected based on the performance of the progeny. This method is reliable, and practicable if cryopreservation of the gametes is possible.

Selection programme for improvement of more than one trait:

Tandem selection

In this method selection is aimed at improving one trait at a time for generations till the goal is reached. Then other trait is selected and it continued for all the traits. It is simple but time consuming, it is rarely practiced because it takes long time and there is a chance of negative correlation between traits which decreases the overall performance.

Independent culling levels

Independent culling level is a selection programme employed when two are more traits are considered for simultaneous improvement. In this, the breeder has to fix the minimum performance level for each trait, and the individual must perform above the minimum level to get selected. Disadvantage of this method is that an individual should be outstanding in all the traits to get selected.

An individual with best performance in one trait and average performance in the other is liable for rejection. The breeder may end up with only a few individuals which satisfy the set performance levels of all the traits leading to inbreeding depression.

Selection index

Index selection is the best and most efficient method of selection progrmme. In this method all the traits considered for selection are given scores, in every individual, according to their performance. An index is made for each animal based on the scores of the different traits considered for selection, giving weightage to each trait according to their relative economic importance using linear regression equation.

Breeding plans :

Selective breeding programme intends to exploit the genetic variations existing in the population, so there should be wide genetic base in the foundation stock. For this, different stocks may be inducted from different sources, making sure the presence wide genetic base.

Superior individuals selected from the population are *inter se* mated to produce the next generation. Continuous selection eliminates the undesirable alleles present in the population. It makes the population homozygous. This is very much useful in evolution of different strains and lines. Mating of related individuals can lead to inbreeding depression which a breeder should be concerned about. To overcome this, crossing between the strains or inbred lines is employed. Out crossing is very useful method in selective breeding wherein unrelated individuals of same species are bred to minimize the effect of inbreeding depression. Crossing of two inbredlines increases the performance because of heterosis. Heterosis is the observed superiority of the hybrid progeny over the parental mean. Hybridization is the rapid method of bringing about genetic modification. Species hybridization is the inter specific crossing.

The salient results of a selective breeding programme carried out in Artemia at CMFRI, Cochin to study the quantitative genetic parameters as well as the response to selection are presented briefly below.

Methodology:

Artemia franciscana (Kellogs1906) from Great Salt Lake, Utah was used for the study. Method of selection followed was Mass selection (individual selection) and the trait under selection was the naupilar size (length in ìm). Bi-directional mass selection was practiced in two sub-populations derived from the base generation viz., SNS line & BNS line with the aim of developing two divergent stocks. While selection for reducing the naupliar size was practiced in SNS line, BNS was selected for bigger naupliar size. Six selected generations were raised. Intensity of selection (i) common for male and female together was estimated as the mean of two sexes i.e. i = ½ (im + if) (Falconer, 1981). Intensity of selection for male and female separately calculated as ratio of effective selection differential to the phenotypic standard deviation. The heritability values of the selected trait (naupliar size) were estimated from full sib data and from regression of offspring on parent as per the procedure given by Becker (1975).The predicted genetic response per generation was calculated for each line separately within sex as per the procedure described by Falconer (1960).

Results :

Heritability of naupilar length:

The heritability estimates of naupilar length, from the regression of progeny on parents, pooled over generations, were 0.2123 ± 0.0766 and 0.3885 ± 0.1108 for males and females respectively in SNS line. The corresponding estimates in BNS were 0.5777 ± 0.1154 and 0.3364 ± 0.1176 respectively. The heritability estimates from full sib data, pooled over generations, were 1.3256 ± 0.0474 and 1.1004 ± 0.0522 for males and females respectively in SNS line, whereas, the corresponding estimates in BNS were 1.2580 ± 0.0583 and 1.4221 ± 0.0479 respectively. While the moderate values of heritability estimated from b_{op} indicated existence of fairly good amount of additive genetic variance which can be exploited through simple selective breeding techniques, the very high estimates of heritability from full sib analysis indicated existence of non-additive genetic variances also.

Selection differential:

Selection differentials, averaged over generations, were slightly higher in females of both SNS and BNS lines. Their mean values were -16.6780 ìm and -16. 3966 ìm in SNS males, -19.9266 ìm and -22.3101 ìm in SNS females, 16.2308 ì m and 15.8700 ì m in BNS males and 17.1180 ìm and 17.0019 ì min BNS females.

Phenotypic responses :

Phenotypic responses for naupliar length from selection was quite substantial. The naupliar size in SNS line, from six generations of selection for smaller size, could be reduced from 486.99 ìm and 490.58 ìm in males and females respectively to 441.67 ìm and 453.05 ìm. The cumulative gain for males and females were -44.32 ìm and -37.52 ìm respectively with average gain per generation being -5.76 ìm and -4.96 ìm. In the BNS line, the naupliar size could be increased to 495.58 ìm and 529.37 ìm in males and females from 486.99 ìm and 490.58 ìm in the base generation, through five generations of selection for bigger naupliar size. The total gain worked out in males and females were 8.59 ìm and 38.80 ìm with mean gain of 0.39 ìm and 5.52 ìm respectively. The mean phenotypic responses were statistically significant except for BNS males.

Realised genetic gain:

The observed phenotypic response is the combined effect of both genetic and environmental factors. Since the environment rarely remains the same over the period of selection, separating out these effects becomes rather difficult. One of the most commonly used methods for removing environmental effect from the phenotypic gains and for determining genetic gain is the use of an unselected control population. Such a control line was used in the present study. Most of the phenotypic responses realized from selection were due to genetic gains. In the SNS line, total genetic gain realized from six generations of individual selection for reduction of the naupliar length was -41.7244 ìm in males and -38.7585 ìm in females. Whereas in BNS line, the total genetic gain from five generations of selection were 12.6427 ìm and 39.4836 ìm in males and females respectively.

The realized mean genetic gain per generation, estimated from regression of control corrected generation means on generation numbers was -5.2585 ìm in males and -5.2289 ìm in females of

SNS, and 0.9338 ìm in males and 5.3493 ìm in females of BNS line. The mean genetic gains were fairly high and statistically significant except in BNS males.

Expected genetic gains :

Expected responses were calculated using heritability estimated from regression of offspring on parent (bop) and also full sib heritability. While, estimates as per former were close to realized genetic gains, those from latter were on the higher side. This result indicates that heritability estimates from full sibs are indeed inflated by non-additive genetic variance , unlike the bop which includes only additive genetic variance.

Generation wise phenotypic response in naupliar length realized from bi-directional selection for reducing naupliar length in SNS line and for increasing naupliar length in BNS line showed that the response in both the lines were in the desired direction. It can be seen that while in SNS line, both the sexes readily responded to selection for decrease in naupliar size, there was a differential response to selection for the higher size in BNS line. In BNS line, the females showed 14.5 times higher response than males, while in the SNS line both sexes showed comparable response. It is rather difficult to explain whether this low response in males was due to attainment of the genetically pre-set maximum size for that sex or due to any other reasons.

The realized response calculated by subtracting the mean control values of each generations from the corresponding selected generation mean is free of environmental effects and therefore, gives the true genetic gain from selection. Comparison of the genetic and phenotypic gains realized in this study point towards the fact that though the environment had played a role in deviating the phenotypic response from the genetic response, its effect was comparatively low and that the genetic gain was quite substantial. Most of the documented selection studies in the aquatic species have reported the response to selection with out considering the environmental effects and therefore, represents only the phenotypic response and not the genetic response. The substantial genetic gains realized in this study indicate the usefulness of selective breeding for developing genetically altered lines.

Genetic improvement need not be restricted to maximizing growth and feed conversion, but can be in survivability and disease resistance. Like any other biological traits there is naturally occurring genetic diversity in disease resistance also. Selective breeding for disease resistance, supported by marker assisted selection is need of the hour which calls for investigations in the field of genetic markers of enhanced disease resistance.

The decision to conduct a selective breeding programme is a decision that must be made on a case-by-case basis. Because selective breeding programmes require dedication, a certain level of sophistication, record keeping and the investment of extra labour. Additionally, selective breeding programmes are not free; they also require the investment of money. Finally, these programmes usually do not produce immediate improvements. Improvements are usually not seen for at least one growing season, so a breeder must be able to incorporate long-term planning into his programme, and he must be patient. A final requirement that must be met for conducting a selective breeding programme is the existence of proper facilities.

Suggested reading

Bondrai, K., R.A. Dunham, R.O. Smitherman, J.A. Joyce and S. Castillo. 1983. Response to bidirectional selection for body weightin blue tilapia. In: *International symposium on Tilapia in aquaculture*. (Eds.Fishelson, L and Z.Yanon.). Tel Aviv.

Douglas, T. 1986. Genetics for fish hatchery managers. pp 1-298.

Ehlinger, N.F. 1964. Selective breeding of trout for resistance to furunculosis. *N.Y.Fish Game J.* 11: 78-90.

Falconer, D.S. 1961. Introduction to quantitative genetics. pp 1-364.

Padhi, B.K and R.K.Mandal. 2000. Applied fish genetics pp 1-190.

Hershberger, W.K., J.M. Myers, R.N. Iwamoto, W.C. Maculey and A.M. Saxton. 1990. Genetic changes in the growth of coho salmon (*Onchorhynchus kisutch*) in marine net- pens, produced by ten years of selection. *Aquacultutre*, 85:187-197.

Kirpichinikov,V.S., I. Hyasov Ju., L.A. Short, A.A. Vikhman,M.V. Ganchenko, L.A. Ostashevsky, V.M. Shivanov, G.F. Tikhnov and V.V. Jurin. 1993. Selection of Krasnodor common carp (C*yprinus carpio L*) for resistance to dropsy: Principal results and prospects. *Aquaculture*, 111:7-20.

Mallet, A.L. and L.E. Haley. 1983. Effects of inbreeding on larval and spat performance in the American oyster. *Aquaculture*, 33:229-235.

Millenbach, C. 1950. Rainbow brood stock selection and observations on its applicationto fishery management. *Prog. Fish.Cult.* 12:151-152.

Moav,R. and G. Wohlfarth.1976. Two way selection for growth rate in common carp(*Cyprinus carpio L*). *Genetics*, 82: 83-101.

Nell, J. A., I. R. Smith and A. K. Sheridan. 1999. Third generation evaluation of Sydney rock oyster *Saccostrea commercialis* breeding lines. *Aquaculture* 170: 195-203.

Nell. J. A. and R. E. Hand (in press). Evaluation of the progeny of the Second generation Sydney rock oyster *Saccostrea glomerata* breeding lines for resistance to QX disease *Martelia sydneyi. Aquaculture*.

Newkirk, G.F. and L.E Haley. 1983. Selection for growth rate in the European oyster, *Ostrea edulis*: Response to second generation groups. *Aquaculture*, 33:149-155.

Smitherman, R.O. and R.A. Dunham. 1985. Genetics and breeding. In:*Channel catfish culture(*ed.Tucker,C.S*.).* 283-316.