

GENETIC COUNSELING, PEDIGREE ANALYSIS,
AND THE PREVENTION OF GENETIC DISEASE
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Historically, genetic counseling has ranged from recommendations to not repeat a mating and outbreed, to recommendations to eliminate all relatives of affected animals from the breeding pool. Neither of these two extremes serves the best long-term interest of breeds. Repeated outbreeding to attempt to dilute detrimental recessive genes is not a desirable method of control. Recessive genes cannot be diluted; they are either present or not. Outbreeding can prevent the production of affected animals, but it will propagate and further disperse the detrimental recessive genes. In most purebred animal systems, breeders are working with closed studbooks. Breeders must consider how selection affects genetic diversity in the gene pool. The goal of genetic counseling is to effectively manage the spread of defective genes, while preserving the health and genetic diversity of the population.

Practical genetic disease control recommendations will vary based on several factors including the size of the breeding population, widely dispersed versus recently mutated defective genes, high frequency versus low frequency defective genes, the mode of inheritance, and the availability of tests for carriers.

With widely dispersed or high frequency defective genes, it must be recognized that carriers are spread across the gene pool. Eliminating unique breeding lines because some individuals carry a single defective gene may adversely affect gene pool diversity more than a process that allows a limited number of carriers to reproduce. Conversely, with recently mutated or low frequency defective genes, it is advisable to strictly limit breeding, so as to not spread the defective gene further in the population.

There are no breeding recommendations that will fit every situation. There are, however, guidelines veterinarians can recommend to preserve breeding lines and genetic diversity while reducing the risk of producing carrier or affected individuals. Protocols for genetic counseling and breeding management of genetic disorders can be based on the known mode of inheritance, and the availability of genetic tests.

Without genetic tests, breeders can still reduce the carrier risk in their matings. A quality individual that is found to be a carrier can be retired from breeding and replaced with a quality offspring. The genes of the retired individual can thus be preserved through the selected offspring, but the carrier risk can be reduced by up to half. To further limit the spread of the defective gene, the offspring should be used in only a limited number of carefully planned matings, and should also be replaced with one or two representative offspring. With this vertical mating scheme, you are maintaining the good genes of the line, reducing the carrier risk with each generation, and replacing, not adding to the overall carrier risk in the breeding population.

If gene tests are not available, the storage of frozen semen is important for quality males with high-risk pedigrees. If tests evolve that can differentiate carrier from genetically normal animals, offspring from frozen semen matings can be reintroduced into the gene pool. Both DNA (from blood or cheek swabs) and semen should be stored to utilize this method.

Breeding Practices

As veterinarians, we should understand how matings manipulate genes within our clients' breeding stock. First comes understanding dogs and cats as species, then as genetic individuals. There is little similarity between a Chihuahua and a Saint Bernard, or between a Himalayan and a Sphynx. However, we must understand that while established breeds are separate entities among themselves, they all are genetically the same species. While a mating within a breed may be considered outbred, it still must be viewed as part of the whole genetic picture: a mating within an isolated, closely related, interbred population. Each breed was developed by close breeding and inbreeding among a small group of founding ancestors, either through a long period of genetic selection or by intensely inbreeding a smaller number of generations. This process established the breed's characteristics and made the individuals in it breed true.

Pure-breeds have closed stud books. This means that the diversity of genes in the breed is fixed. Genes cannot be gained through breeding, only lost. In some cat breeds, cats who meet the phenotypic standard of the breed may be introduced into the gene pool. This, of course is an added source of genetic diversity for the breed.

Tens of thousands of genes interact to produce a single individual. All individuals inherit pairs of chromosomes; one from the dam, and one from the sire. On the chromosomes are genes; so all genes come in pairs. If both genes in a gene pair are the same gene (for instance, "aa" or "AA") the gene pair is called homozygous. If the two genes in a gene pair are unlike (for instance, "Aa") the gene pair is called heterozygous. Fortunately, the gene pairs that make a cat a cat and not a dog are always homozygous. Similarly, the gene pairs that make a certain breed always breed true are also homozygous. Therefore, a large proportion of homozygous non-variable pairs - those that give a breed its specific standard - exist within each breed. It is the variable gene pairs, like those that control color, size and angulation that produce variations within a breed.

One method to gauge the genetic diversity of a population is to measure the average inbreeding coefficient (or Wright's coefficient) for a breed. The inbreeding coefficient is a measurement of the genetic relatedness of the sire and dam. If an ancestor appears on both the sire and dam's side of the pedigree, it increases the inbreeding coefficient. The inbreeding coefficient gives a measurement of the total percentage of variable gene pairs that are expected to be homozygous due to inheritance from ancestors common to the sire and dam. It also gives the chance that any single gene pair can be homozygous.

The types of matings chosen for breeding animals will manipulate their genes in the offspring, affecting their expression. Linebreeding is breeding individuals more closely related (a higher inbreeding coefficient) than the average of the breed. Outbreeding involves breeding individuals less related than the average of the breed. Linebreeding tends to increase homozygosity. Outbreeding tends to increase heterozygosity. Linebreeding and inbreeding can expose deleterious recessive genes through pairing-up, while outbreeding can hide these recessives, while propagating them in the carrier state.

Most outbreeding tends to produce more variation within a litter. An exception would be if the parents are so dissimilar that they create a uniformity of heterozygosity. This is what usually occurs in a mismating between two breeds, or a hybrid, like a Cockapoo. The resultant litter tends to be uniform, but demonstrates "half-way points" between the dissimilar traits of the parents. Such litters may be phenotypically uniform, but will rarely breed true due to the mix of dissimilar genes.

One reason to outbreed would be to bring in new traits that the breeding stock does not possess. While the parents may be genetically dissimilar, a mate should be chosen that corrects the breeding animal's faults but phenotypically complements its good traits. It is not unusual to produce an excellent quality individual from an outbred litter. The abundance of genetic variability can place all the right pieces in one individual. Many top-winning show animals are outbred. Consequently, however, they may have high heterozygosity and may lack the ability to uniformly pass on their good traits to their offspring. After an outbreeding, breeders may want to breed back to individuals related to their original stock, to attempt to solidify newly acquired traits.

Linebreeding attempts to concentrate the genes of specific ancestors through their appearance multiple times in a pedigree. It is better for linebred ancestors to appear on both the sire's and the dam's sides of the pedigree. That way their genes have a better chance of pairing back up in the resultant offspring. Genes from common ancestors have a greater chance of expression when paired with each other than when paired with genes from other individuals, which may mask or alter their effects.

Linebreeding on an individual may not reproduce an outbred ancestor. If an ancestor is outbred and generally heterozygous (Aa), increasing homozygosity will produce more AA and aa. The way to reproduce an outbred ancestor is to mate two individuals that mimic the appearance and pedigree of the ancestor's parents.

To visualize some of these concepts, the pedigree of a Gordon Setter, Laurel Hill Braxfield Bilye will be used. The paternal grandsire, CH Loch Adair Foxfire, and the maternal grandam, CH Loch Adair Firefly WD, are full siblings, making this a first-cousin mating. The inbreeding coefficient for a first cousin mating is 6.25%, which is considered a mild level of inbreeding.

In Bilye's pedigree, an inbreeding coefficient based on four generations computes to 7.81%. This is not significantly different from the estimate based on the first-cousin mating alone. Inbreeding coefficients based on increasing numbers of generations are as follows: five generations, 13.34%; six generations, 18.19%; seven generations, 22.78%; eight generations, 24.01%; ten generations, 28.63%; and twelve generations, 30.81%. The inbreeding coefficient of 30.81 percent is more than what you would find in a parent-to-offspring mating (25%).

The total inbreeding coefficient is the sum of the inbreeding from the close relatives (first cousin mating), and the background inbreeding from common ancestors deep in the pedigree. Such founding ancestors established the pedigree base for the breed. **The background inbreeding has far more influence on the total inbreeding coefficient than the first-cousin mating, which only appears to be its strongest influence.**

Knowledge of the degree of inbreeding in a pedigree does not necessarily help you unless you know whose genes are being concentrated. The relationship coefficient, which can also be approximated by what is called the *percent blood* coefficient, represents the probable genetic likeness between the individual whose pedigree is being studied, and a particular ancestor. It is a measurement of the average percentage of genes the individual and the ancestor should have in common.

We know that a parent passes on an average of 50% of its genes, while a grandparent passes on 25%, a great-grandparent 12.5%, and so on. For every time the ancestor appears in the pedigree, its percentage of passed-on genes can be added up and its "percentage of blood" estimated. **In many breeds, an influential individual may not appear until later generations, but then will appear so many times that it necessarily contributes a large proportion of genes to the pedigree.** This can occur in breeds, due either to prolific ancestors (usually males), or a small population of animals originating the breed. Based on a twenty-five generation pedigree of Bilye, there are only 852 unique ancestors who appear a total of over twenty-million times.

In Bilye's pedigree, CH Afternod Drambuie has the highest genetic contribution of all of the linebred ancestors. He appears 33 times between the sixth and eighth generations. One appearance in the sixth generation contributes 1.56% of the genes to the pedigree. His total contribution is 33.2% of Bilye's genes, second only to the parents. **Therefore, in this pedigree, the most influential ancestor doesn't even appear in a five-generation pedigree.**

Foundation dogs that formed the Gordon Setter breed also play a great role in the genetic makeup of today's dogs. Heather Grouse appears over one million times between the sixteenth and twenty-fifth generations, and almost doubles those appearances beyond the twenty-fifth generation. He contributes over ten percent of the genes to Bilye's pedigree. Any detrimental recessive genes carried by Heather Grouse or other founding dogs, would be expected to be widespread in the breed.

The average inbreeding coefficient of a breed is a measurement of the breed's genetic diversity. When computing inbreeding coefficients, you have to look at a deep pedigree to get accurate numbers. An inbreeding coefficient based on 10-generation pedigrees is standardly used, but requires a computerized pedigree database to compute.

The average inbreeding coefficient for a breed will be based on the age and genetic background of the breed. A mating with an inbreeding coefficient of 14 percent based on a ten generation pedigree, would be considered moderate inbreeding for a Labrador Retriever (a popular breed with a low average inbreeding coefficient), but would be considered outbred for an Irish Water Spaniel (a rare breed with a higher average inbreeding coefficient).

Looking at the historical pedigrees of Bull Terrier breeding dogs (males and females that have five or more registered offspring), we find that for dogs born in the decade 1970-1979, the average ten generation inbreeding coefficient was 23.11% +/- 6.04%. For Bull Terriers born 1980-1989, this number is 21.54% +/- 5.69%. For 1990-1999, the average inbreeding coefficient is 19.01% +/- 6.23. It is obvious that the 10 generation inbreeding coefficient of the Bull Terrier

breed is going down with each decade. This shows that the breeders are utilizing the diversity of the gene pool, and not breeding themselves into a corner with popular sires.

Of course, the actual diversity of genes and inbreeding in the breed is not going down. It is just that the earlier ancestors producing background inbreeding are falling beyond the 10th generation, and are no longer included in the computation. As long as the health and vitality of the breed is being maintained, and there is no epidemic of breed-related disease from detrimental recessives, this pure-bred population should be able to be maintained.

Most breeds start from a small founding population, and consequently have a high average inbreeding coefficient. If the breed is healthy and prolific, the breadth of the gene pool increases, and the average inbreeding coefficient can go down over time. Some dog breeds were established on a working phenotype, and not on appearance. These breeds usually start with low inbreeding coefficients due to the dissimilar backgrounds of the founders. As certain individuals are linebred on to create a uniform physical phenotype, the average inbreeding coefficient can increase.

There is no specific level or percentage of inbreeding that causes impaired health or vigor. If there is no diversity (non-variable gene pairs for a breed) but the homozygote is not detrimental, there is no effect on breed health. The characteristics that make a breed reproduce true to its standard are based on non-variable gene pairs. There are pure-bred populations where smaller litter sizes, shorter life expectancies, increased immune-mediated disease, and breed-related genetic disease are plaguing the population. In these instances, prolific ancestors have passed on detrimental recessive genes that have increased in frequency and homozygosity. With this type of documented inbreeding depression, it is possible that an outbreeding scheme could stabilize the population. However, it is also probable that the breed will not thrive without an influx of new genes; either from a distantly related (imported) population, a natural landrace population, or crossbreeding.

Diversity Issues

Fortunately, most breeds do not find themselves in the position of this amount of limited diversity and inbreeding depression. However, the perceived problem of a limited gene pool has caused some breeders to advocate outbreeding of all individuals. Studies in genetic conservation and rare breeds have shown that this practice actually contributes to the loss of genetic diversity. By uniformly crossing all “lines” in a breed, you eliminate the differences between them, and therefore the diversity between individuals. Eventually, there will not be any “unrelated line” to be found. Everyone will have a mixture of everyone else’s genes. This practice in livestock breeding has significantly reduced diversity, and caused the loss of unique rare breeds.

A fallacy of using outbreeding to maintain genetic diversity is the belief that the diversity of a breed must be maintained in every single animal. Breeders must concentrate on the specific goals of breeding (selecting for the health and quality of the breed), versus the tools used to get there (outbreeding, linebreeding, etc.) Selecting breeding stock simply to produce the lowest possible inbreeding coefficient is not a goal that will guarantee a quality animal. Animals who are poor examples of the breed should not be used simply to maintain diversity. Related individuals with desirable qualities will maintain diversity, and improve the breed.

The process of maintaining healthy “lines” or families of animals, with many breeders crossing between lines (outbreeding) and breeding back (linebreeding) as they see fit maintains diversity in the gene pool. **It is the varied opinion of breeders as to what constitutes the ideal representative of the breed, and their selection of breeding stock that maintains breed diversity.**

A basic tenet of population genetics is that gene frequencies do not change from the parental generation to the offspring. This will occur regardless of the homozygosity or heterozygosity of the parents, or whether the mating is an outbreeding, linebreeding, or inbreeding. This is the nature of genetic recombination. Selection, and not the types of matings used affect gene frequencies and breed genetic diversity.

If two parents are both heterozygous (both Aa) for a gene pair, on the average, they would produce 25% AA, 50% Aa, and 25% aa. (These are averages when many litters are combined. In reality, any variety of pairing up can occur in a single litter.) If a prolific male comes out of this litter, and he is homozygous aa, then the frequency of the “a” gene will increase in the population, and the frequency of the “A” gene will decrease. This is known as the popular sire syndrome. Of course, each individual has thousands of genes that vary in the breed, and everyone carries some deleterious recessive genes. The overuse of individual breeding animals contributes the most to decreased diversity (population bottlenecks), and the increased spread of deleterious recessive genes (the founders effect). Again, it is selection (use of this stud to the exception of others), and not the types of matings he is involved in that alters gene frequencies. Breeders should select the best individuals from all lines, so as to not create new genetic bottlenecks.

Decisions to linebreed, inbreed or outbreed should be made based on the knowledge of an individual's traits and those of its ancestors. Inbreeding will quickly identify the good and bad recessive genes the parents share, based on their expression in the offspring. However, unless there is prior knowledge of what the offspring of milder linebreedings on the common ancestors were like, the litters (and buyers), may be exposed to extraordinary risk of genetic defects. In matings, the inbreeding coefficient should only increase because of specific linebreeding (increasing the percentage of blood) to selected ancestors.

Breeders should not set too many goals in each generation, or the selective pressure for each goal will necessarily become weaker. Genetically complex or dominant traits should be addressed early in a long-range breeding plan, as they may take several generations to fix. Traits with major dominant genes become fixed more slowly, as the heterozygous (Aa) individuals in a breed will not be readily differentiated from the homozygous-dominant (AA) individuals. Desirable recessive traits can be fixed in one generation because individuals that show such characteristics are homozygous for the recessive genes. Individuals that pass on desirable traits for numerous matings and generations should be preferentially selected for breeding stock. This prepotency is due to homozygosity of dominant (AA) and recessive (aa) genes. However, these individuals should not be overused, to avoid the popular sire syndrome.

Breeders should plan their matings based on selecting toward a breed standard, based on the ideal temperament, performance, and conformation, and should select against the significant breed related health issues. Using progeny and sib-based information to select for desirable traits, and against detrimental traits will allow greater control.

Breeding Recommendations to Manage Genetic Disease

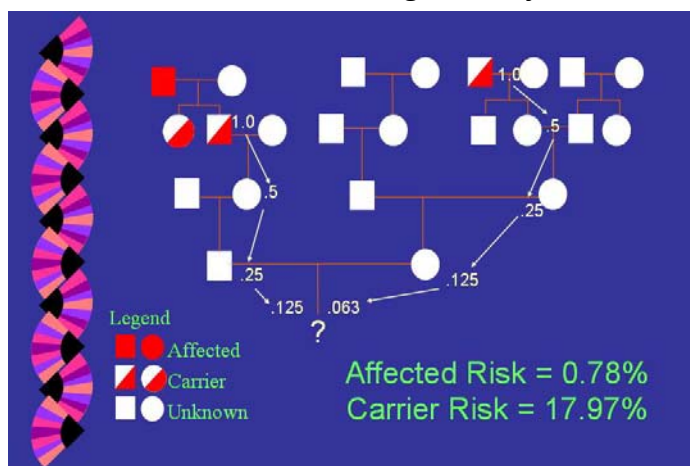
Based on the mode of inheritance of a disorder, and the availability of genotypic or phenotypic genetic tests, breeding management recommendations can be used to prevent or reduce the frequency of carrier or affected offspring.

Autosomal Recessive Disorders

In the case of a simple autosomal recessive disorder for which a test for carriers is available, the recommendation is to test breeding-quality stock, and breed carriers to normal-testing individuals. This prevents affected offspring from being produced. The aim is to replace the carrier breeding-animal with a normal-testing offspring that equals or exceeds it in quality. Breeders may not produce this offspring in a single mating, and may still have to breed another carrier in the next generation, as long as it is again bred to a normal testing animal. Breeders don't want to diminish breed diversity by eliminating quality animals from the gene pool because they are carriers. Additional carrier testing offspring should not be placed in breeding homes; as the goal is to reduce the frequency of the defective gene in the population. As each breeder tests and replaces carrier animals with normal-testing animals, the problem for the breed as a whole diminishes.

The problem with simple autosomal recessive disorder for which no carrier test exists is the propagation and dissemination of inapparent carriers in the gene pool. Breeders must assess whether each individual animal in their breeding program is at high risk of being a carrier. This requires knowledge of the carrier or affected status of close relatives in the pedigree. An open health registry that is supported by the parent club makes it easier for breeders to objectively assess these matters. By determining the average carrier-risk for the breeding population, breeders can select matings that have a projected risk that is lower than the breed average.

If a breeding animal is at high risk of being a carrier, the best advice is to breed to an individual that has a low risk. This will significantly diminish the likelihood that affected animals will be



produced, and can reduce by up to half the risk that there will be carriers among the offspring. Using relative-risk assessment as a tool, breeders should replace higher-risk breeding animals with lower-risk offspring that are equal to or better than their parents in quality. A negative aspect of pedigree analysis is that it selects against families, regardless of an individual's normal or carrier status. On the other hand, it allows for the objective risk assessment and continuation of lines that might

otherwise be abandoned due to high carrier-risk.

Breeding an individual only once and replacing it with an offspring allows breeders to improve their chances of moving away from defective genes and also limits the dissemination of defective genes. When dealing with disorders for which carriers cannot be identified, the number of offspring placed in breeding homes should be kept to a minimum.

Autosomal Dominant Disorders

Autosomal dominant genetic disorders are usually easy to manage. Each affected animal has at least one affected parent, but it can be expected that half of the offspring of an affected animal will be free of the defective gene. With disorders that cause death or discomfort, the recommendation is to not breed affected animals. To produce the next generation of a line, a normal full sibling of an affected animal can be used, or the parent that is normal can be used.

A problem with some autosomal dominant disorders is incomplete penetrance. In other words, some individuals with the defective gene may not show the disorder. Roughly half their offspring, however, may be affected. If a genetic test is available, this is not a problem. Otherwise, relative-risk assessment can identify which individuals are at risk of carrying incompletely penetrant dominant genes.

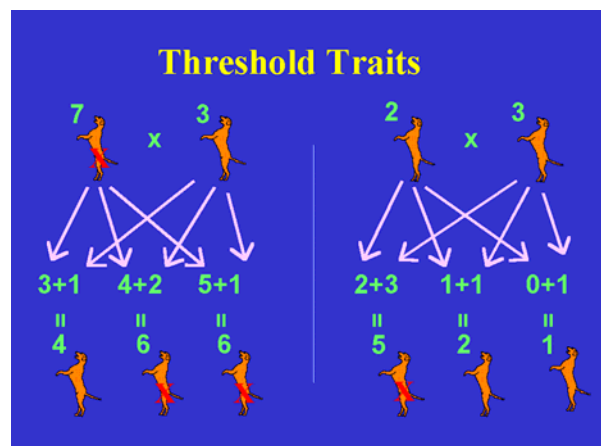
Sex-Linked Disorders

For sex-linked (also known as x-linked) recessive defective genes for which carrier tests exist, breeders should follow the same “breed and replace” recommendations as are outlined above in the discussion of autosomal recessive disorders. If there is no test, the defective gene can be traced through the pedigree. If a male is affected, he would have received the defective gene from his carrier mother. All of his daughters will be carriers, but none of his sons. By using relative-risk assessment to breed him to a female that is at low risk of being a carrier, you can prevent affected offspring, and select a quality son for replacement.

There are rare instances in which a female is affected with a sex-linked disorder. In such cases, she would have received the defective gene from both parents; specifically, an affected father and a mother who is either a carrier or is affected herself. If an affected female is bred, all the sons will be affected, and all the daughters would be carriers, so affected females clearly should not be bred. A normal male that is a littermate to an affected female, however, would be able to carry on the line without propagating the defective gene. Rare sex-linked dominant disorders are managed the same way as autosomal dominant disorders. The difference is that affected males will *always* produce all affected daughters.

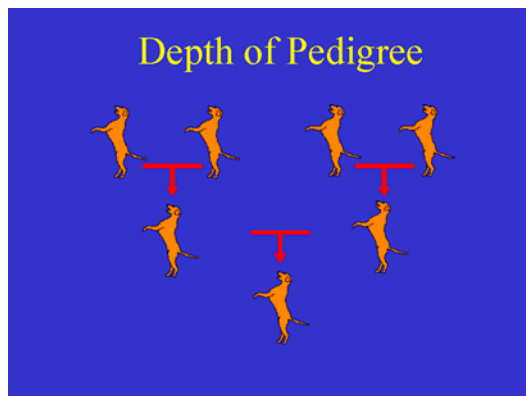
Polygenic disorders/Complex Inheritance

To manage polygenically controlled disorders genetically, they must be considered as threshold traits. A number of genes must combine to cross



a threshold producing an affected individual. As the presence of individual genes may not be all that is required to express the defect, these genes are considered liability or susceptibility genes. If phenotypically normal parents produce affected offspring, both should be considered to carry a subclinical genetic load of liability genes that combined to cause the disorder. In the theoretical mating above), consider that five additive hip dysplasia liability genes must combine to produce an affected dog.

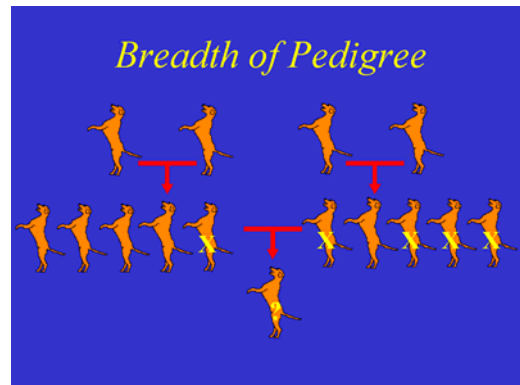
In addition to quantitative or additive genes, many polygenic disorders can have a major recessive or dominant qualitative gene that must be present to produce an affected individual. Molecular genetic research to identify these genes can allow better management of polygenic disorders such as hip dysplasia, epilepsy, and congenital heart anomalies. If one exists, the “trigger” gene in one breed or family may be different from the gene in others. Consequently, the development of a genetic test in one breed may not prove useful in all breeds.



In polygenic disorders, all individuals are not affected due to the same gene combinations. Breeders must break down affected phenotypes into traits that more directly represent the genes that control them. As environmental aspects also affect the expression of many polygenic disorders, they too need to be identified and controlled.

The primary reason for diminished

progress against polygenic disorders is that breeders have been selecting for depth of pedigree; generations of phenotypically normal parents and grandparents. In polygenic disorders, the phenotype of the individual does not directly represent its genotype. The phenotype of the full brothers and sisters more directly represent the range of genes present in the breeding individual. In other words, with polygenic disorders the breadth of the pedigree (that is, consideration of all siblings of individuals in the pedigree) is as important, if not more important than the depth of the pedigree (consideration only of parent-offspring relationships). This can be evaluated through open health registries, such as CHIC (www.caninehealthinfo.org).



Phenotypically normal individuals from litters with a high incidence of disease are expected to have a higher compliment of liability genes that is closer to the disease threshold than the average for the breed. This is why it is important to screen both pet and breeding animals from litters for polygenic disorders. By counseling owners to select for breadth of phenotypically normal littermates of breeding animals, and of parents of breeding animals, all breeds should realize a decrease in polygenically controlled disorders. In addition, the offspring of breeding individuals should be monitored to see which are passing the disorder with higher frequency. By

evaluating all of these aspects of polygenic disorders, we are helping breeders produce healthier animals.

Unknown Inheritance

For disorders without a known mode of inheritance or carrier test, breeders should be counseled to use the same control methods as with polygenic disorders. Animals with a low genetic load for the disorder should be selected for breeding, through the results of examinations of first-degree relatives (littermates, parents, and offspring). If there are multiple generations of normalcy in the breadth of the pedigree, then you can have some confidence that there is less risk that liability genes are being carried. Quality individuals with higher risk should be bred to individuals with lower risk, and replaced with a quality offspring. This recommendation should be repeated in the next generation, to further lower the risk of producing affected or carrier animals.

It is distressing to breeders when we confirm a genetic disorder. As veterinary professionals, we can offer positive and practical genetic counseling recommendations to maintain breed lines and genetic diversity, and improve the overall health of breeds. The total elimination of defective genes will probably be impossible for most breeds. The use of these guidelines can assist breeders in making objective breeding decisions for genetic-disease management, while continuing their breeding lines. The individual breeder can use genetic tests to; 1) identify carriers, 2) work to breed away from the defective gene(s), and 3) ensure (through testing) that the defective gene(s) is not reintroduced in future matings. Each breeder will have their own rate of progress, depending on the frequency of the defective gene(s) in their own breeding animals, and which desirable individuals are carriers.