

Breeding Improved Honey Bees

I. Bee Breeding and Animal Breeding

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Preface

Within recent years the technique of artificial insemination has been improved and used successfully in experiments on bee breeding. Thus a major barrier in our efforts to produce better breeds of bees has been successfully hurdled.

Readers of the bee journals now find advertisements telling them that improved hybrid bees are available. Since these are said to be superior to existing races or strains, the wide-awake beekeeper will want to know how they are produced.

This is the first of a series of articles on the use of controlled breeding to improve honey bees. This article discusses the relationship of bee breeding to animal breeding in general. Articles on heredity and variation, sex determination and bee breeding, inbred and hybrid bees, and production of hybrid queens will follow.

I — Bee Breeding and Animal Breeding

Almost everything that we eat or wear is the product of our domesticated plants and animals. Most of these living forms have been cared for and propagated by man since long before the beginning of written history.

Domestication implies several things, no one of which is sufficient to define it completely. It usually means tameness; but individual bears, lions, or even snakes can be tamed, and few of us would call them domesticated animals. Domestication further implies bringing the growth and reproduction of the plant or animal at least partly under man's control, but certain pigeons and geese usually choose their own mates and accept no others. Man uses domesticated animals or their products for his own advantage, and he usually keeps them in or near his dwelling. From the variable wild species man has, by selection and reproduction, developed individuals

that suit his purpose.

Breeding and selection have therefore led to the establishment of types or varieties useful to man. Selection for usefulness and survival under man's care has frequently resulted in varieties incapable of survival without man's care. Man did not create these genetic forms. By controlled breeding and selection he merely succeeded in putting together or fixing these varieties from the genetic variability that existed in their wild ancestors.

The honey bee has a definite place in our modern world. Its products of honey and wax are useful to man, although perhaps not essential to all men. However, the pollination activities of bees affect the lives of most of the people of the world. Although we may never succeed in taming the honey bee or in getting it to change its mating habits, it seems evident that the definition of domestication is sufficiently broad to include this most useful insect. The ways and means of improving the economic value of honey bees by controlling their growth and reproduction will be discussed in this article.

The breeder of animals must decide which animals shall produce the next generation. His choice is determined by his knowledge of the variability that exists in the potential breeding population and the usefulness that he desires in the succeeding generations. The variability in the breeding population is due in part to differences in inherited factors and also in part to differences in environment. A knowledge of the science of heredity should make it easier to choose intelligently among the many animals in the potential breeding population.

Because of the social nature of the honey bee, the colony is the unit upon which selection must be based for most economically important characteristics. The individuals of a colony consist of a queen and her

worker and drone offspring. The queen is mated before she begins egg laying, and her mate is thus the father of the workers in the hive. She may have more than one mate, and therefore the genetic variability of the colony may be due to several individuals. Each of these individuals can be influenced by environmental factors that are not always easy to evaluate. The bee breeders' problems in properly evaluating the genetic and environmental factors affecting each individual colony are much more difficult than those of other animal or plant breeders.

Although honey bees have been propagated by man for 5,000 years or more, there is little evidence that much progress in bee breeding has been made. No superior breeds of bees have been established. The species has been subdivided into races, and all these races show considerable variability. Apparently they are the product of many generations of random matings within populations that have been partially isolated by geographic barriers. Man may have had some influence in fixing these races by selection that altered the frequency of certain inherited factors.

The aim of the bee breeder is to produce better or more profitable bees. This may mean more honey per colony or it may mean more efficient pollinators. The breeder must work with the reproductive individuals that are available. Since colonies of bees differ in many characteristics, the breeder has variability from which to select. This variability may be due to both genetic and environmental factors. The successful bee breeder must observe his colonies closely so that he can make proper allowances for environmental factors affecting the genetic variability in his breeding stocks.

His next problem is to mate the

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breeding individuals so as to obtain genetic improvement. From the variable colonies he attempts to unite the genes (the genetic factors) for good qualities from many stocks into one line or breed while eliminating inferior or less desirable qualities. In order to succeed he must have some knowledge of the science of heredity. Geneticists have accomplished this with many plants and animals, and therefore it is possible with bees.

Since the honey bee is an animal, it would appear that animal breeding methods should be used. The honey bee, however, is quite different from the economically important animals. Its method of reproduction is in a sense more like some plants. We shall attempt to describe these differences and likenesses and show how they affect methods of bee breeding.

The science of heredity is relatively new, having only become recognized as a science in the 20th century. Yet a great amount of real progress through selective breeding was accomplished in most of our economically important plants and animals long before Mendel, the father of genetics, was born. Most every breed of horses, cattle, hogs and chickens used in the United States today was established before the 20th century. Yet even today there are no fixed breeds of bees—we still have only races.

Let us review a few of the methods employed in the establishment of improved breeds of livestock. The fundamental biological principles of inheritance are the same whether we are breeding plants or animals. Therefore, much about bee genetics can be learned by studying the genetics of other animals and plants.

All our domestic animals except the guinea and the turkey originated and were domesticated in prehistoric times in the Old World. Most domestication evidently occurred in central or western Asia, although some is believed to have taken place in Europe, Egypt, India and China. It is still disputed whether most animals descended from a single wild species or from two or more species.

Early naturalists supposed that species were fixed and had no genetic variability. However, modern geneticists have shown that wild populations of one species of animal were not genetically uniform. They have also shown that selected breeding within these wild populations, with their existing genetic variability, can

produce distinctly contrasting races or breeds within a few generations.

It seems probable that all the races of bees, as we know them today, developed from a single wild species of honey bee. In time they became dispersed over a large part of the Old World and were somewhat isolated into many small groups. There was, of course, some interbreeding between groups, but geographical barriers prevented a great many matings between groups. As a result a certain amount of inbreeding took place within each geographic group. By mutations, natural selection, and perhaps some human intervention in selection, the bees within each geographic area became different from those in other areas.

The bees from the various areas are called races. Probably the most geographically isolated race of bees is the Caucasian. A Russian author who studied this race in its native home divided it into six separate varieties. These varieties ranged in color from the banded appearance of the Italian to completely black. Other genetic characteristics within this and other races of bees are equally variable. The bees of one race are fertile when crossed with those of another race.

Wild cattle, horses, and hogs were also variable animals. The recognized breeds of these animals were developed by selected breeding of outstanding individuals from variable populations. The history of the formation of the beef breeds of cattle that originated in the British Isles is typical of successful animal-breeding methods.

History tells us that migrating people and invading armies usually carried with them much livestock from their native lands. Migrations were slow, and the stocks mingled with and interbred with stocks present in the countries through which they passed. It is not known just how many and what kinds of cattle were introduced into the British Isles throughout the ages. However, it is known that the Norsemen and the Dutch carried cattle into the British Isles, and it is suspected that the Romans and the Normans also brought in some cattle. These cattle interbred with the cattle of the islands, and a great amount of genetic variability existed in these stocks before the formation of the breeds that we know today as Hereford, Shorthorn, and Aberdeen-Angus.

About the beginning of the 18th century the common lands of Great

Britain were enclosed so that the intermingling of stocks on a wide scale was stopped. The cattle population was thus broken up into small groups and inbreeding within groups was the result. About this time the cities began to grow and a demand for meat developed. Some types of cattle were more useful for this purpose than others, and breeders attempted to produce these meat-type cattle. A few breeders gathered together some of the best of the desired type into one or a few herds. There followed years of rather intense inbreeding between these selected animals and their descendants. Very little outside breeding stock was permitted to enter these herds. After several generations the cattle within these herds were uniform and were distinct from the other animals in the community. Every recognized breed of cattle has arisen from the concentration of the genes (blood) of one or a few animals of greater than ordinary merit.

Breeds have developed in a similar manner among other types of livestock. The Poland China hog was evolved or originated in Butler and Warren counties in southwestern Ohio. The exact combination of breeds or types that resulted in this hog is not clear. Prior to 1830 there existed in this area a number of types, among which were the Bedfordshire, Byfield, Russian, and Big China. Some Berkshire blood was introduced in 1835 and some Irish Glazier blood in 1839. There was intermixing of these breeds without a definite plan for 20 years or more. In 1870 the Warren County hog was black and white spotted. It was regarded as a breed in 1872 and was given the name "Poland China." The diverse races of swine from which this breed was found could hardly have come together anywhere on earth without the aid of man. From this heterogeneous mixture the breed arose by inbreeding of selected sires.

Why have not bee breeders done the same thing with bees? Many diverse races and strains from all areas of the world have been brought into the United States. There is thus great genetic variability in our bees. When we look around we find outstanding individuals (colonies). We have seen that successful breeders of other animals succeeded in establishing superior breeds of animals by inbreeding and selecting from among outstanding individuals and their descendants.

Bee breeders did try methods used by breeders of other animals but they did not succeed. It is true that they could not control matings so successfully as other animal breeders, but this was not the major cause

of their failure. Studies indicate that a major cause for this failure was a lack of knowledge of sex determination in honey bees. Its importance to bee breeding will be explained in a later section.

combinations of genes possible.

The separation of genes going into daughter cells followed by the creation of new combinations by fertilization is called segregation and recombination. This is the mechanism that brings about genetic variation between individuals in a family. Since in the honey bee all the sperms produced by a drone are identical, no variation between sisters (queens or workers) is introduced from the drone.

All the genes at a certain locus may be the same and have exactly the same effect in one individual as in another. Occasionally a gene may change so that it has a different effect. Such a change is called a **mutation**. Both the mutant and the parent gene are then referred to as **alleles**. The new allele multiplies in cell division and may become common in the bee population if it has a favorable action. There can be several alleles at a single locus.

Let us consider a simple case of alleles in the honey bee. It has been shown that the difference between a mutant white-eyed bee and the normal or wild-type black eyed bee is due to the action of alleles at a single locus on one of the chromosomes. It has further been shown that the normal or wild-type allele for the black eye is **dominant** to the mutant allele for white eye, which is then said to be **recessive**. A dominant gene masks the effect of its recessive allele.

In genetics it has become customary to denote genes by means of letters. The allele for black eyes may be designated by a capital **W**, since it is dominant, and the opposing recessive allele by **w**. One of these alleles is located at a particular locus on one of the chromosomes of all honey bees. Let us call this the **W** locus on chromosome number 1.

Since female honey bees have pairs of chromosomes, they can have any two of the alleles at this locus—**WW**, **Ww**, or **ww**. The **WW** and **Ww** genotypes (gene combinations) have black eyes, since **W** is dominant over **w**. The **ww** females have white eyes. Since drones have only one chromosome of each pair, they are either **W** (black eyes) or **w** (white eyes).

Inheritance of eye color in the female bee is thus duplicate and particulate. Each female receives either the **W** or **w** gene from its mother, and it also receives **W** or **w** from its

II. Heredity and Variation

HEREDITY is concerned with the transmission of characters from parent to offspring. Animals of one species resemble each other, and this resemblance is due to heredity. Variations between individuals within any given species may be due to heredity or environment or both.

The body of the honey bee, like that of all the higher animals, is made up of many cells. Each cell contains a sort of cell within a cell, which is called a nucleus. Within the nucleus are the many genes, which are the hereditary determiners of form and function of all the parts and organs of the body.

Let us begin with the unfertilized egg. The egg is one cell having one nucleus and one set of genes. The genes are arranged in the nucleus in definite linear order on a series of string-like structures called **chromosomes**. The position of each gene on a chromosome is called its **locus**. A sperm cell also contains a nucleus with a single set of chromosomes and genes.

In fertilization the sperm enters the egg and its nucleus unites with the nucleus of the egg. Thus the fertilized egg nucleus has two sets of chromosomes and hereditary determiners.

In the development of an individual from the fertilized egg, the egg cell first divides into two cells and then the two daughter cells divide again. This process continues until the adult consists of millions of cells. Each time a cell divides each gene duplicates itself and all the chromosomes split longitudinally into two identical halves. These halves pass to opposite sides of the nucleus then divide into two. The cell then divides and each daughter cell gets one of these genetically identical nuclei. Each cell grows to the original size of its mother cell and the process of division is repeated. As growth proceeds there is an interaction between cells that deter-

mines the form and function of the body parts.

In the honey bee the queens and workers develop from fertilized eggs in this manner. Therefore all the cells in their bodies contain two sets of chromosomes and genes and are said to be **diploid**. The drone, however, develops from an unfertilized egg, has only one set of chromosomes and genes in all the cells of its body and is said to be **haploid**.

During development reproductive organs are formed in which certain cells are set aside for the production of germ cells, sperms or eggs. In the formation of the egg a cell division occurs in which the chromosome behavior is different from that in the multiplication of body cells. As a result of this division the egg has only one member of each pair of chromosomes, or one set of chromosomes and genes as mentioned at the beginning of this discussion. This mechanism assures the maintenance of a constant number of chromosomes in the species.

In most organisms a similar reduction takes place in the formation of the sperm, but as the honey bee drone has only one set of chromosomes no reduction takes place.

In the formation of egg cells it is a matter of chance which member of a given pair of chromosomes reaches a given daughter cell. It receives an assortment of 16 chromosomes (one member of each pair) that might range from all those from the queen's mother to all from her father or any mixture of the two. The genes on one chromosome usually pass as a unit and are said to be linked. Frequently, however, parts of a pair of chromosomes exchange places, so that an egg cell may receive chromosomes that are partly of maternal and partly of paternal origin. This exchange is called **crossing over** and, of course, increases the possibilities for variation tremendously by making many

drone father. Inheritance of eye color in the drone bee is particulate but not duplicate, for he receives either **W** or **w** from his mother since he does not have a father. The particulate nature of inheritance is illustrated by the gene **W** or **w**. In the drone if **W** is present the eye color is black, but it is white when the gene **w** is substituted for **W**.

If a queen bee has a **W** gene on both of her number 1 chromosomes, she is represented by the formula **WW** and is said to be homozygous for the **W** locus. Homozygous means likeness of genes. The drone can never be said to be homozygous, because he has only one chromosome of each pair and thus only one of the alleles, **W** or **w**.

Let us assume that a black-eyed virgin queen homozygous for **W** is

mated to a white-eyed drone. The mating can thus be represented as follows:

$$\begin{aligned} \text{queen} &= \mathbf{WW} \\ \text{drone} &= \mathbf{w} \\ \mathbf{WW} \times \mathbf{w} &= \mathbf{Ww} \end{aligned}$$

Since the queen is homozygous, **WW**, all her eggs will have a **W** gene. All sperms from the drone will have the gene **w**, and the female workers or queen offspring of such a mating will all be **Ww**, or heterozygous. Since **W** is dominant over **w** and all offspring get one **W** and one **w**, the eyes of these females will be black.

Let us suppose that from one of these **Ww** fertilized eggs a virgin queen is produced. She will be **Ww**, or heterozygous for the **W** locus. Now let us mate her to a white-eyed drone that is thus **w**. Since the

queen is **Ww**, she will produce eggs that will have only one of these alleles. On an average 50 percent of the eggs of such a queen will receive a **W** gene and the other 50 percent will receive the **w** gene. The mating and the offspring will be as follows:

$$\begin{aligned} \text{queen} &= \mathbf{Ww} \text{ eggs either } \mathbf{W} \text{ or } \mathbf{w} \\ \text{drone} &= \mathbf{w} \text{ sperm all } \mathbf{w} \\ \mathbf{Ww} \times \mathbf{w} &= \begin{array}{l} 50\% \mathbf{Ww} \text{ heterozygous black-eyed females} \\ 50\% \mathbf{ww} \text{ homozygous white-eyed females} \end{array} \end{aligned}$$

It can be seen that the heterozygous **Ww** black-eyed queen produces 50 percent of heterozygous black-eyed daughters and 50 percent

INHERITANCE OF ABDOMINAL COLORATION IN TWO STRAINS OF HONEY BEES.



Figure 1. An inbred yellow queen artificially mated to a black drone produces worker progeny that are like neither parent but are banded intermediates. The first 12 rows are abdomens of her worker progeny. The last 8 rows are abdomens of drones produced by the yellow queen and are yellow like the queen because they develop from unfertilized eggs.

Figure 2. An F-1 hybrid queen intermediate in color mated to a black drone produces progeny that range in color from intermediate to complete black.

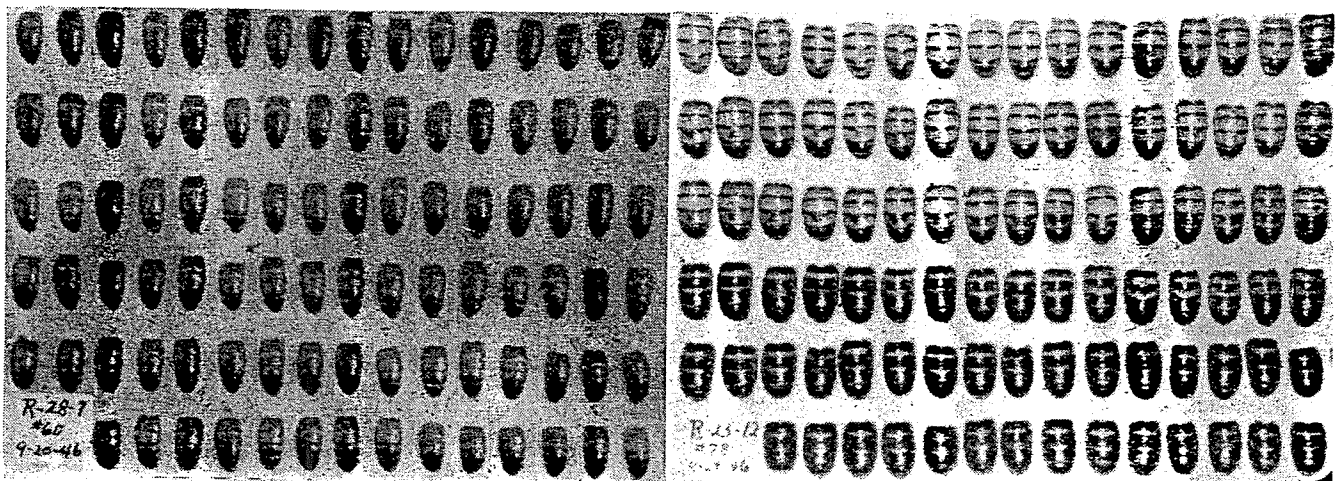


Figure 3. An F-1 hybrid queen intermediate in color mated to a yellow drone produces progeny that range in color from intermediate to parental yellow.

Figure 4. Drones produced by F-1 hybrid queens show gametic segregation for color. These range from yellow to complete black. (Photos courtesy North Central States Bee Culture Laboratory, Madison, Wisconsin.)

of homozygous white-eyed daughters after mating to a drone that is whited-eyed and thus *w*. Now this heterozygous black-eyed queen can produce both black-eyed and white-eyed sons, since her eggs have either a *W* or a *w*. Since these eggs can develop without fertilization, the *W* eggs will develop into black-eyed drones while the *w* eggs will develop into white-eyed drones. This illustrates how it is possible for a drone to exhibit and transmit to his offspring something that was not expressed in his mother, yet all his inheritance is from his mother because he has no father.

In the discussion of the *W* locus we have considered only two alleles, one completely dominant over the other. It is not always true that one allele is completely dominant. The heterozygous condition (such as *Ww*) may have intermediate effects. Furthermore, there can be several alleles of one locus that interact in various ways. When one considers that there may be thousands of loci with interaction of genes at different loci as well as interaction of alleles, it becomes apparent that the possibilities for genetic variation are tremendous.

Bees vary in many factors, not all of which are so easy to evaluate as eye color. It has been shown that there are at least seven different loci for genes that affect the color of the abdomen. Thus many abdominal color patterns are possible, since seven pairs of genes can produce 2187 different genotypes. If the workers of a mixed population are arranged according to color from the darkest to the lightest abdomen, they will fall into a continuous series.* Such a variation is said to be **continuous** in contrast to one such as white eye color, which is

said to be discrete. In most characters of economic importance, such as honey production, egg-laying ability, vigor or longevity, variation is continuous. Although such variation may be due to the interaction of a great many genes, it does not always imply a large number of inherited factors. It is possible that only a few genes are concerned and that much variation is due to environmental influences. These may be many and occur at various times from the beginning of queen rearing all the way through the life of the colony up to the time of harvesting the honey crop.

By a study of variation as such, we are never able to find out which part of the variation is certainly due to environment and which to heredity. The bee breeder must be ever alert and try to reduce the environmental variation to a minimum so that the variation he observes and upon which he bases his selection will be due to genetic factors. Queens in colonies to be compared as to performance should be reared under optimum conditions. (Queen rearing will be treated more fully in a later paper.) It is best to place the colonies in a single yard so that they will all be exposed to the same weather conditions and will have the same forage available. At the beginning of a performance test they should be equalized as to bees, pollen, and honey so that none start the test under a handicap. Colonies should be scattered and isolated as much as possible to prevent drifting and robbing.

The colonial nature of the honey bee makes the selection of breeding individuals particularly difficult. In contrast to the situation in most farm animals, the colony rather

than the individual is the unit of performance upon which the selection of breeding individuals must be based. The colony performance depends upon the performance of a queen of one generation and all her worker daughters of the next generation. We cannot breed this unit. We can in effect breed the queen by using her sons, because, as we have seen they represent her germ cells. For breeding females we can only use virgin queens that are sisters to the workers of the colony.

It has frequently been stated that a beekeeper can improve his stock, even though he may know little about animal breeding, if he is willing to make the effort. Nevertheless, one cannot find conclusive evidence that much genetic improvement has been accomplished.

Among the principles of breed improvement employed by early plant and animal breeders were "like produces like or the likeness of some ancestor," "inbreeding produces nepotency or refinement," and "breed the best to the best." In following these principles why did the bee breeder fail to make the progress made by the early plant and animal breeders? Inadequate control of mating and the difficulty of recognizing genetic superiority of individuals were factors that hindered progress. There is yet another factor that adds complexity to bee breeding, which was not understood until very recently. This is the inheritance of sex determination, and it is a very important factor because it is associated with egg hatchability. The relationship of sex determination to bee breeding is the subject of the next article.

* Inheritance of abdominal coloration in two strains of honey bees is illustrated in figures 1, 2, 3, and 4.

III. Sex Determination and Bee Breeding

EVERY beekeeper knows that the economically productive colony has a large population of worker bees. In order to provide this force the queen must lay a large number of fertile eggs, and they must hatch and be nurtured to develop into adults.

Many years ago Dzierzon discovered that male bees develop from unfertilized eggs. Drones have a mother but no father. This is known as parthenogenesis. It is not peculiar to bees, for many other animals

have this method of reproduction. The female bees, workers and queens, develop from fertilized eggs. They have both father and mother. From each parent they receive 16 chromosomes and so have 16 pairs of chromosomes. The drone, who receives all his inheritance from his mother, has only 16 single chromosomes. Having **paired** chromosomes, the workers and queens are called **diploid** individuals. Since the chromosomes of drones are **not paired**, they are called **haploid** individuals.

Recent work indicates that the fundamental determiner of sex in bees is not the number of chromosomes or whether or not the egg is fertilized. Sex is determined by the action of certain genes at one locus on one pair of chromosomes. In the wasp *Habrobracon*, a relative of the honey bee, the females are diploid and the males are normally haploid, but under certain genetic conditions diploid males have been produced. No diploid males have been discovered in honey bees, but there is experi-

mental evidence that sex determination in honey bees is similar to that in *Habrobracon*.

The position of a gene on a chromosome is called a locus. On one of the chromosomes there is a locus that is called the X, or sex-determining, locus. At this locus in honey bees there is a series of multiple allelic genes—instead of only two alleles, such as capital W and small w, which are alleles to each other, there are many alleles. We will call this the X locus and the alleles X^a, X^b, X^c, etc. To simplify matters we can drop the X and use only the letters a, b, c, etc., remembering always that these are the X alleles.

According to the theory of sex determination in bees, females develop from fertilized eggs that are heterozygous at the X locus. These eggs have two unlike alleles, such as ac, ad, bc, bd, etc. Any fertilized egg that happens to be homozygous at this locus would be a drone if it developed. However, it does not develop but dies instead. Thus all fertilized eggs that are homozygous—that is, having two sex alleles alike, such as aa, bb, cc, etc.—are lethal and do not hatch. Drones develop from unfertilized eggs and are haploid, having only one of these alleles—a or b or c or d, etc.

If a queen with sex alleles ab is mated to a drone having sex allele c, the fertilized eggs from this queen will be ac and bc. Since these eggs are heterozygous, their hatchability is near 100 per cent. An ab queen mated to an a or b drone has fertilized eggs that are ab and aa. Since the aa eggs do not hatch, the viability of fertilized eggs from this queen will be only 50 per cent.

It can thus be seen that egg hatchability and the brood quality of a queen are determined by the sex alleles of the queen and the drone or drones that mate with this queen. To illustrate this point let us set up a breeding example. A beekeeper chooses a breeder queen from which he will raise a number of daughter queens. This queen has high-quality brood because she mated with a drone or drones whose sex alleles were different from hers. Since the queen is diploid and heterozygous for the sex-determining locus, we will assume that she is ab. Having good-quality brood, we know that she mated with a drone or drones that were not a or b. Let us assume that she mated with two drones, one c and one d. Fertilized eggs from this queen will be ac, ad, bc, and bd. Notice that all are heterozygous for the

X locus, an indication of high hatchability and good-quality brood. Let us further assume that these queen daughters ac, ad, bc, and bd are allowed to mate with drones from another unrelated queen, which we will designate as yz, and that each queen mates with two drones—that is, drones y and z, y and y, or z and z. Their fertilized eggs will be either ay, cy, by, and dy or az, cz, bz, and dz. Consequently the brood quality will be good because the queens and drones have different sex alleles.

Let us assume that the next year the beekeeper raises queen daughters from one of these queens and drones from one of the other queens and allows them to mate together. These are called matings of first cousins. The sex alleles of the daughters of ac x yz will be az, cz, ay, and cy in equal proportions, if the queen is ac and mated to one y and one z drone and received equal amounts of sperm from each.

Assume that the queen selected to produce the drones for mating with these virgins had sex alleles ad and that she was mated to z and y drones. Since drones develop from unfertilized eggs, all drones from an ad queen will be either a or d. If a large number of drones are produced, it is likely that half of them will be a and the other half d. If each virgin—az, cz, ay, or cy—mates with two drones, matings will be to drones a and d, or a and a, or d and d.

The queen az that mated with one a and one d drone will produce the following types of fertilized eggs with equal frequency: aa, ad, za, and zd. Since ad, za, and zd are heterozygous, they hatch, but the homozygous aa fertilized egg dies. Thus one fourth of the fertilized eggs produced by this queen fail to develop. Consequently she has low-quality brood and does not develop a colony with a large population of worker bees. This queen would have produced high-quality brood and a large colony if she had mated with two d drones instead of one a and one d, but only 50 per cent of her eggs would have hatched if she had mated with two a drones.

It is quite evident that the productivity of a colony or of a queen depends to a great extent on the sex alleles represented in the queen and in the drones that mated with the queen. Three sister queens can be identical genetically, and yet if each queen is mated to two drones of a group of brother drones, their egg hatchability can be 50, 75, or 100 per

cent, depending upon the sex alleles in these drones. Other things being equal, if only 50 per cent of the fertilized eggs of a queen hatch, the colony is very unproductive. On the other hand, if the eggs are 100 per cent hatchable, very populous colonies will result. If these queens should mate only once, there would be only 50 per cent and 100 per cent hatchability of fertilized eggs from the several queens, with no 75 per cent hatchability matings.

What then can be expected of a breed-improvement program attempted by a queen breeder? Let us assume that from his own or selected stocks he chooses three mated (tested) queens as the source of all future breeding stock. He will try to improve his stock by breeding the best to the best. He will control matings by isolation so that they will be between progenies of these queens. It is assumed that each of these queens has different sex alleles. If each queen is mated to two drones and all these drones have sex alleles different from any of those in the three selected queens, there will be four different sex alleles in the progeny of each breeder, and a total of 12 alleles for all breeders. These sex alleles are given the following distribution:

Queen No.	Queen	Drones Mated to Queens	Daughter Queens
1	ab	c and d	ac, bc, ad, bd.
2	ef	g and h	eg, eh, fg, fh.
3	jk	l and m	jl, jm, kl, km

Each of these queens with her offspring is a family. From each family the queen breeder may produce drones and queens the first year and allow the matings to occur at random or he may produce only drones from one or two families and queens from the other one or two families. Furthermore, he may segregate the drones and thus divide his population into three mating groups. Now it can be shown that the relationship of matings for the third year is essentially the same by any of these methods. In the first two years the breeder has an opportunity to select between three groups of sister queens and the three groups of brother drones produced by these queens.

If the breeder has a separate yard for drones of each family and allows queens from one of the other families to mate at this isolated location, he can avoid poor viability of brood due to the mating of individuals having sex alleles in common for the

first two years only. This can be illustrated as follows: First-year daughters of queen 1 mate with drones of queen 2; second-year queens (1 x 2) mate with drones from family 3. The third-year queens (1 x 2) x 3 must mate to drones produced by (2 x 3), (1 x 3), or (1 x 2) queens. A queen (1 x 2) x 3 will have one sex allele from the 3 family and the other from either the 1 or 2 family.

Let us assume that a daughter queen has one allele from family 1 and one allele from family 3. She is designated as having sex alleles *ak*. She could have any one of several combinations, but she must have one allele of the four possessed by family 3—that is, *j*, *k*, *l*, or *m*. The other of her sex alleles must come from family 1 or 2 and could be any one of the eight alleles originally present in these lines.

The drones with which this queen mates are sons of (2 x 3) queens. If a large number of (2 x 3) queens produce these drones, then the *e*, *f*, *g*, *h*, *j*, *k*, *l*, and *m* drones will occur with equal frequency. If this *ak* queen mates with only one drone and that drone happens to be *k*, then the eggs of this queen will be only 50 per cent hatchable, since the fertilized eggs will be *ak* and *kk* in equal proportions, and all *kk* eggs will fail to hatch. However, should she mate with a single drone having the sex allele *h*, her eggs would then be 100 per cent hatchable.

If a large number of queens ((1x2) x3) mate with (2x3) drones, half of them with only one drone and the other half with two drones, then 11.3 per cent of the queens will have 50 per cent viable brood, 14.8 per cent of them will have 75 per cent viable brood, and 73.8 per cent will have 100 per cent viable brood.

If the queen producer has maintained three separate groups, each group will be similar to the above, for ((2 x 3) x 1) daughters will be mated to drones produced by (3 x 1) queens and ((3 x 1) x 2) daughters will mate at the yard that produces (1 x 2) drones. The ratio in each yard will average the same for brood viability.

If these stocks are maintained separately or united into one breeding population, the viabilities will be similar in all yards after the second year. Only 12 alleles were present originally in the stocks, and as long as these are present in equal ratios, the brood viabilities of mated queens will approximate the figures given

above (also see Table I). These percentages are based on the assumption that half of all queens mate once and the other half mate twice. If the proportion differs, the percentage of 50 per cent viable matings will vary accordingly, but the percentage that will give 100 per cent viable brood will not change.

It is thus evident that one fourth of all mated queens will produce brood of low viability (50 or 75 per cent egg hatchability) after inbreeding begins, if attempts are made to improve a line by breeding within a closed population started from three selected breeder queens. During the first two seasons there is no inbreeding, for the three lines are being crossed only with each other.

The queen breeder has an opportunity to select for desired type or color, but cannot increase the percentage of matings with high brood viability because he does not know what alleles are carried by each queen.

It can be seen from Table I that the percentage of matings that give high-viability brood increases with the number of sex alleles in the breeding population. However, even with 40 sex alleles in a breeding population, approximately 5 per cent of all single-mated queens and 10 per cent of all double-mated queens will produce 50 or 75 per cent of viable brood. The only sure way to have high-viability brood from all queens is to mate queens of one sex-allele

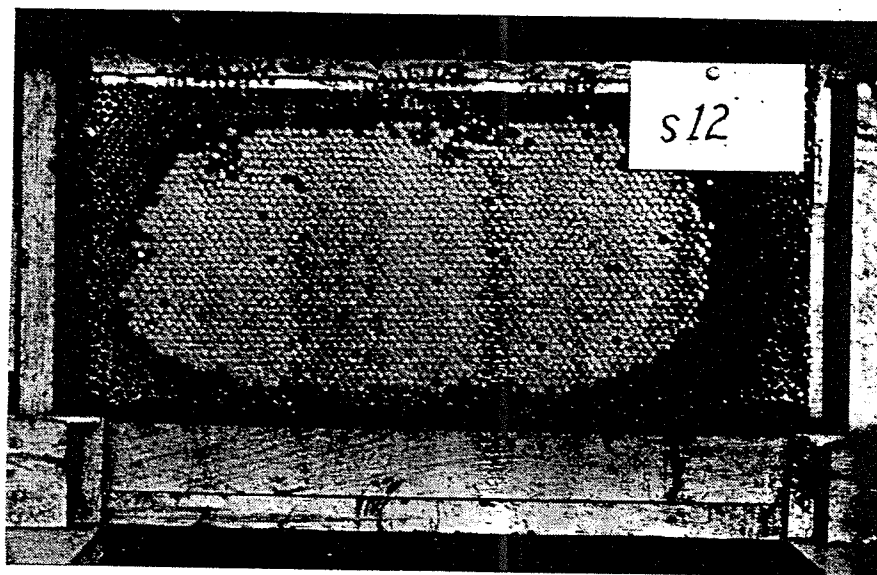


Figure 1. Brood of high viability — 96 per cent. Queen and drones carried unlike alleles, for example, *ab* queen mated to *c* and *d* drones.

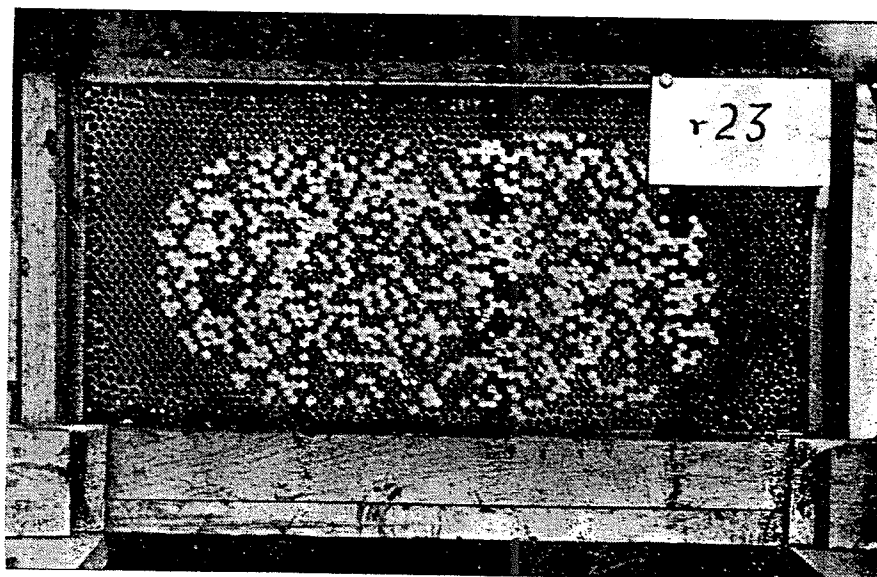


Figure 2. Brood of low viability — 48 per cent. Queen and drones carried same sex alleles, for example, *cd* queen mated to *c* and *d* drones.

combination with drones originating from queens having other sex alleles.

Insofar as other characteristics of economic importance are concerned, little progress can be made after the first few generations by selection within a closed population. Homozygosis, or fixation of genetic characteristics, will increase slightly each generation in the first few generations after inbreeding is begun. The strain of bees developed at the end of 10 to 15 years of breeding by this method will still be variable. However, selections will have reduced some of the variability for

visible traits such as color, and the stock will appear more nearly uniform than at the beginning of the breeding program. Progress may also have been made in selecting for temper and other highly heritable characteristics. However, it is very doubtful whether significant progress can be expected in characteristics such as vigor or honey production for, like most economically important characteristics, they have low heritability. Characteristics that are highly heritable, such as color, are varied only slightly by environmental factors. Low-heritability characteristics, such as egg produc-

tion, are varied considerably by the environment.

It should be pointed out that the small real gain by this method of breeding is made at the expense of brood viability, owing to matings of relatives having sex alleles in common. If the breeder starts with three selected queens and mates their offspring together, one fourth of all matings will result in brood of poor viability. This is a high price to pay for commercial experiments in bee breeding.

The bee breeder is thus unable to establish a superior breed of bees by using the methods that have been successful with cattle and hogs. Breeding of the best to the best and continuing with their descendants results in poor-quality brood in a large number of matings. The animal breeder fixed his breeds by inbreeding within closed populations. The bee breeder using this method fixes some characteristics, but cannot fix the breed for the important characteristic of high egg hatchability. This characteristic is necessary for quality brood and thus populous colonies.

The next article will tell how the bee breeder can insure high-quality brood in all colonies by adopting a breeding plan based on controlled hybrids.

Table 1.

EXPECTED BROOD VIABILITY WITH RANDOM MATINGS IN POPULATIONS HAVING EQUAL FREQUENCIES OF VARIOUS NUMBERS OF SEX ALLELES

Number of Sex Alleles	Single-Drone Matings		Two-Drone Matings			Matings Half Single and Half with Two Drones		
	50%	100%	50%	75%	100%	50%	75%	100%
2	100	0	100	0	0	100	0	0
4	50	50	25	50	25	37.5	25	37.5
6	33.3	66.7	11.1	44.4	44.4	22.2	32.2	55.6
8	25	75	6.3	37.5	56.3	15.6	18.8	65.6
12	16.7	83.3	2.8	27.8	69.4	9.7	13.9	76.4
16	12.5	87.5	1.6	21.9	76.5	7	10.9	82.1
20	10	90	1	18	81	5.5	9	85.5
24	8.3	91.7	.7	15.3	84	4.5	7.6	87.9
28	7.1	92.9	.5	13.3	86.2	3.8	6.6	89.6
32	6.3	93.7	.4	11.7	87.9	3.3	5.9	90.8
36	5.6	94.4	.3	10.5	89.2	2.9	5.2	91.9
40	5	95	.2	9.5	90.3	2.6	4.8	92.6
n	2	n-2	(2) ²	2(2)(n-2)	(n-2) ²			
	<u>n</u>	<u>n</u>	<u>n</u>	<u>n</u>	<u>n</u>			

IV. Inbred and Hybrid Bees

MANY beekeepers have produced excellent queen daughters of imported stock or daughters of some selected queen that they obtained from other beekeepers. The new stock queens usually mate with drones of the beekeeper's own stock. If the sex alleles of the two stocks are different, the eggs from most matings are high in hatchability.

The new stock may also be genetically different from the stock that the beekeeper already has. Thus the queen daughters of the imported queen produce hybrid progeny after mating with the drones of the beekeeper's own stock. These may be racial hybrids or hybrids between strains of bees. Such queens often produce excellent colonies with high-quality brood. The beekeeper then praises the imported stock apparently without realizing that his own drones contribute half to the genetic make-up of the workers in these superior colonies.

The story is often quite different

after a few generations of backcrossing to the original drones or to drones produced by the hybrid daughter queens. Segregation and recombinations occur, and in the third or fourth year the queens and their progenies are frequently more variable than the original stocks. By this time the beekeeper concludes that the original imported stock was not so good after all or had "run out"; so he gets some other new stock for breeding purposes.

By this procedure the colony superiority may again be restored to a high level. It also falls again after a few years. To keep the quality of his bees up, the beekeeper continues to get new or different breeding stock every few years.

These results are best explained as due to hybrid vigor and the crossing of stocks having different sex alleles. The first-generation colonies were superior because they were hybrids—that is, they were headed by cross-mated queens whose worker progeny were hybrids between

the imported and local stocks. By mathematical analysis of the action of genes in future generations of segregation and recombination after outcrossing, it can be shown that by selection alone the beekeeper is powerless to keep this stock at the high level of production and uniformity exhibited by the first-cross progeny.

Most of the complicated physiological characteristics, such as egg production and vigor are dependent for their maximum expression on the harmonious interaction of many genes and their alleles. Since these genes are located on many chromosomes and there is linkage of genes on the same chromosomes, the likelihood that certain factors conducive to vigor will be linked with injurious factors reduces the chance of obtaining maximum vigor by selection within inbred lines. Moreover, in breeding within a line the sex-allele problem again arises and selection is considerably weakened by a high proportion of low viability matings.

In plant and animal breeding it has been found that certain hybrids are superior to the lines from which they are produced. The superiority

of hybrid corn is unquestioned. Animal breeders have also obtained superior hybrids. Because a large proportion of the progeny is required for breeding purposes to maintain the lines, animal breeders have not used hybrids for extensive production. However, in hogs a modification of the controlled-hybrid program is often used. This is the crisscrossing of three lines, using hybrid sows and purebred boars in each generation. The three-breed crossed pigs are more productive than those of the pure breed. Hybridizing inbred lines of chickens has become more popular and profitable within recent years.

Hybrid vigor is generally thought to be a characteristic of dominance. Dominant genes tend to have more favorable effects than their recessive alleles, and in a hybrid the dominant genes find expression at more loci than do the recessive alleles. If one inbred line is dominant (AA) at one locus and recessive at another (bb) and another inbred line has the reverse (aa and BB) characteristics, the hybrid will have one dominant gene at each locus (AaBb). If both loci contribute to vigor, then the hybrid will be superior to either inbred line. It is also thought that certain genes contribute more to vigor in the heterozygous than in the homozygous condition.

An ideal breeding system is the production and maintenance of purebred but unrelated lines and the crossing of these lines for the production of superior hybrids. Not all—in fact, only a very few—of the inbred lines will give outstanding hybrids when crossed. The breeder, however, can perpetuate those lines that nick favorably for the repeated production of superior hybrids.

Plant breeders can take advantage of hybrid vigor because of the large number of seeds produced in each generation and the relatively low cost of producing seed for the production of hybrids. Animal breeders, on the other hand, are not in this favorable position. The expense of producing and maintaining highly inbred lines together with the small number of offspring makes this practice unprofitable, except with chickens and to some extent hogs.

Honey bees more nearly approach the condition existent in plant breeding. Less than 1 percent of the daughter queens and drones of a queen is needed to perpetuate the line. Whereas only 4 to 10 queens are needed in each generation to in-

sure the continuance of an inbred line, 3 or 4 outcrossed queens can be used to produce 10,000 hybrid queens.

The low cost of producing and maintaining inbred lines of bees, together with the large number of offspring that can be produced from a single breeding individual, suggests that a bee-breeding program based on hybrid vigor is practical. The effect of the sex alleles on egg hatchability further indicates that other breeding plans, such as line breeding, are slow and expensive because too many poor-viability matings occur.

To produce hybrid bees the bee breeder may cross different races, strains, or inbred lines of bees. Unless these bees are homozygous for the desired characters, the resulting hybrids will be variable. Furthermore, other crosses of the same races or strains will differ from each other. A sure method of having uniform hybrids is to have strains homozygous for the desired characters.

The fastest method of increasing homozygosity in a line of stock is to inbreed. Inbreeding is the mating of closely related individuals, such as parent-offspring, brother-sister, or cousins. Related individuals are likely to have many of the same inherited qualities, and mating of these individuals tends to fix these qualities in a homozygous, or pure, condition. Inbreeding is used chiefly for the one purpose of producing homozygosity, or genetic likeness of offspring.

Inbreeding within a population tends to separate the population into many distinct families. Each family becomes uniform within itself but distinctly different from other families. Selection between families of such inbred lines can then be made with more accuracy than selection between individuals. This is especially true for characteristics of low heritability, such as egg production and vigor.

The inbred individuals become lower in average merit than non-inbreds. They are not nearly so vigorous or productive. Inbreeding is the severest test of the heredity worth of the individual that can be made, for it causes fixation of both good and bad traits.

Because of the mating habits of bees, it is necessary that all inbreeding matings be made by artificial insemination. After highly inbred lines are obtained it is necessary to control matings to insure keeping

the stocks pure. Artificial mating is the only safe method known at present with bees.

If the breeder makes very close matings, such as mother-son, brother-sister, or backcrossing to a queen, he can accomplish very little by individual selection. It is true that he can accomplish a great deal when selecting for color or other visible characteristics that are highly heritable. However, most of these characters have little economic importance. Characteristics of greater importance, such as brood production, vigor, or resistance to disease, are not visibly detectable in individual queens or drones. Selection for these traits is more effective after the lines are inbred.

To determine traits in inbred lines, crosses are made to a line or lines known to possess certain inherited qualities. Thus the test of inbred lines for characteristics of low heritability is to cross all inbred lines to certain "tester" lines and compare the unknown inbred lines with each other. Selection then is between inbred lines.

The mathematical consequences of the various systems of inbreeding have been worked out by several authors. Kalmus and Smith (1948), Crow and Roberts (1950), and Polhemus, Lush, and Rothenbuhler (1950) have reported on the various systems of inbreeding possible with honey bees. The formulas of Crow and Roberts can be used to determine the inbreeding and relationship coefficients of stocks of bees.

Before beginning to produce inbred lines the bee breeder chooses the stocks that he will use. He may desire to cross two or more of the selected noninbred stocks before beginning to inbreed.

He should obtain a few queens each from a large number of sources and test all queens for one season under uniform conditions in a single yard if possible. In this way he can get first-hand knowledge of the potential breeding stocks. He may select some colonies for immediate inbreeding and perhaps allow the others to cross freely for use as future breeding stocks.

It has been shown in both plant and animal breeding that the best hybrids usually come from crosses of unrelated stocks of different origin. The breeder should therefore select a number of different types of bees. He should select some black, some yellow, and some queens intermediate in color rather than all of one color type. He should also select

some long, slender and some short, stubby queens. He should obtain some gentle bees and perhaps even one or two lines with a lot of evil temper!

The superior hybrids are genetic heterozygotes. They are the result of crosses between stocks that are unlike genetically. Differences in type, color, or temper denote genetic differences for these characteristics and probably also for other characteristics that the breeder cannot readily see or measure.

There is some evidence that gentleness in bees is dominant over viciousness. In one of our experiments the hybrids of a cross between a vicious and a gentle line were gentle rather than vicious. The vicious line may bring into the hybrid the few genes that mean the difference between an average and a superior hybrid bee. By no means are we suggesting that we need viciousness in bees. This is merely an illustration of how genetic diversity might be obtained.

Once the stocks are chosen, close inbreeding is begun to fix the lines. The breeder should know which matings to make to obtain the desired inbreeding in the shortest time with the least expense. Figure 1 shows the percentage of inbreeding obtained in successive generations by the various systems of inbreeding possible in honey bees. The percentage of inbreeding is the percentage of heterozygous loci of the original selected individuals that become homozygous by inbreeding. Inbreeding has no effect on genes already homozygous in the lines; so we are concerned only with those genes that are not homozygous. Since the bee breeder cannot tell which genes were originally heterozygous and what effect each gene has, he can only measure the increase in purity of the stocks by the percentage of inbreeding.

With present techniques the two systems of mating that increase inbreeding fastest, backcrosses to a male and a mother-son, are not advisable economically. Loss of breeding individuals, and consequently of inbred lines, is high when these systems are followed.

The next most rapid method of increasing inbreeding is by brother-sister matings. This system is recommended, because at present it is the most practical of all systems shown. For the first two generations the systems of brother-sister and aunt-nephew matings and backcrossing to a female are identical in

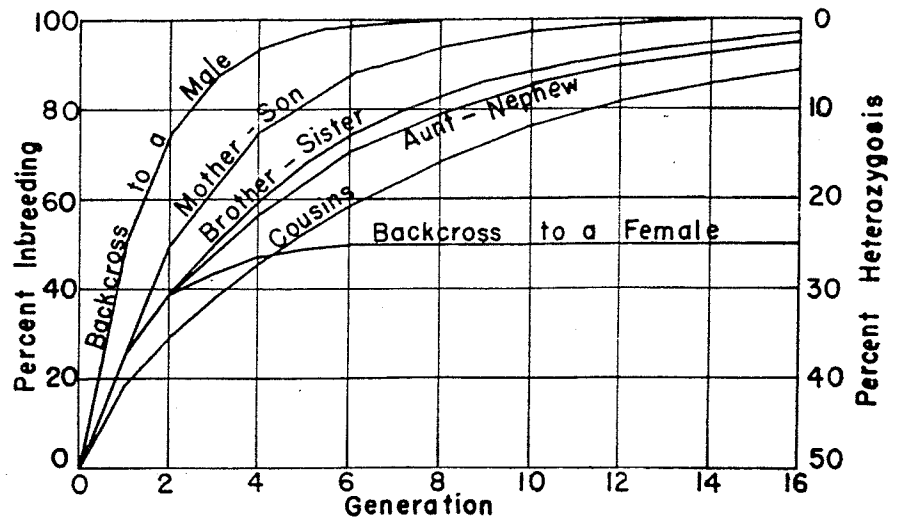


Figure 1. Percentages of inbreeding and of heterozygosity (assuming the initial value to be 50 per cent) in successive generations of various systems of inbreeding in honey bees. (From Crow and Roberts, 1950.)

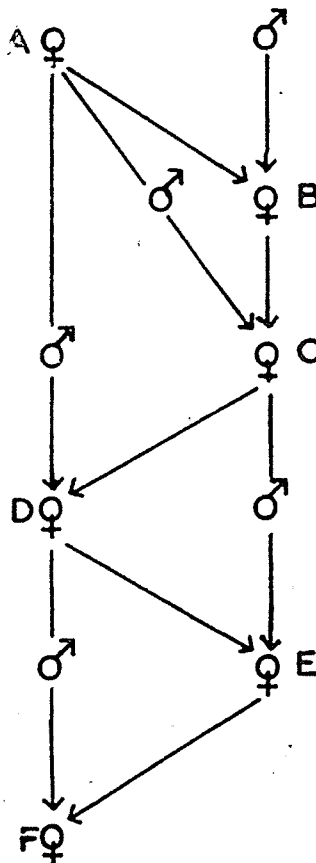


Figure 2. Arrow diagram of a recommended system of mating for inbreeding in bees. After two generations of backcrossing to a selected queen, the line is continued by brother-sister matings.

percentage of inbreeding. Because the aunt-nephew system depends upon the use of only one drone for each mating, this system is not so dependable for insuring the survival of an inbred line. Using only one drone to inseminate a queen often results in a poorly inseminated queen, who may turn into a drone layer before the next generation is produced.

Because drones mature more slowly than queens, backcrossing to a female produces inbreeding to 37.5 percent faster than brother-sister mating in time consumed per generation for the first two generations. Thus backcrossing to the original queen for the first two generations followed by brother-sister matings in all future generations is recommended for the production of inbred lines in the shortest time with the greatest chance of success in maintaining the inbred lines. Multiple drone matings may be made in all generations to insure good inseminations.

The bee breeder should know what inbreeding will do to his stocks. If he starts an inbred line by backcrossing and then makes brother-sister matings as illustrated in Figure 2, he may expect that each line will become more uniform as inbreeding progresses. Most noticeable, however, for the first few generations will be the quality of the brood.

If queen B, a daughter of A, is mated to several drones that are sons of A, the egg hatchability of queen B will average 75 percent. A daughter of queen A, mated to sons of queen A will have egg hatchability that will average either 75 or 50 percent. If her egg hatchability is 50 percent, the line has been reduced to two sex alleles, and queens D, E, and F will also have eggs of 50-percent hatchability if mated as shown in the figure. If queen C has egg hatchability of 75 percent, then D may also be 75 percent but somewhere not far from E or F egg hatchability will probably drop to 50 percent and all future generations will remain at that level.

However, by selection it is possible to keep egg hatchability at 75 percent, but the breeder would be reducing the effectiveness of inbreeding slightly by selection against certain alleles. It is advisable to select the 50-percent egg-hatchability matings in the C or D generation and thus quickly reduce all inbred lines to two sex alleles, so that they will also have 50-percent egg hatchability. If this is done, an analysis of the sex alleles is more readily accomplished. By test crossing to identify the sex alleles in each line the breeder can then predict which crosses will give high egg hatchability in hybrids and which crosses will give intermediate or low egg hatchability.

Other than improving egg hatchability, the bee breeder can accomplish very little by selection while inbreeding. He can select queens and drones in each generation for color and general appearance, but selection for most other characteristics will be very inefficient.

In one season of inbreeding it is possible to produce a number of sister queens of the D generation and mate them to their brothers (drones produced by their mother queen C). These queens will be wintered, and the following year the breeder can make test crosses while continuing to inbreed the lines by brother-sister matings. It is advisable to test-cross the inbred lines at the E generation of queens (50-percent inbred). Brother-sister mating should be made each year after the first to insure continuing the inbred

lines until they are selected in hybrid combinations.

Since egg hatchability in inbred lines with two sex alleles is only 50 percent, the inbreds do not develop colonies with sufficient populations to permit accurate evaluation of such economically important characteristics as honey production, swarming tendencies, and wintering. However, such qualities as tongue length, wing length, and temper may be measured in inbred workers. It is also possible to select for such characters in the queen as size and number of ovarian tubules. This selection in inbreds as inbreds is supplemental to rather than a substitute for selection between inbred lines when in crosses with other inbred lines.

All crosses should be made artificially and the colonies tested under environmental conditions as nearly uniform as possible. The various hybrid crosses will differ, but the individuals of a given hybrid will be uniform as a group.

After it has been found that the colonies having (line 3 x line 4) hybrid queens mated to drones produced by (line 1 x line 2) hybrid queens are superior in production, then the breeding stock to produce this four-way combination can be distributed for commercial production. A commercial queen producer then needs only two artificially mated breeder queens from the breeding source to produce several thousand hybrid queens.

One of these breeder queens is used to produce daughter queens that will head the 50 or more drone-producing colonies necessary to supply ample drones. An inbred queen of line 1, artificially mated to drones from inbred line 2, is the mother of the hybrid daughters needed to furnish drones. Since drones are produced parthenogenetically, those drone-producing queens can be naturally mated to the drones of the breeder's own stock.

An inbred queen of line 3 artificially mated to drones from inbred line 4 is used as the breeder queen. Her daughters will be hybrids (3x4). These virgins are naturally mated to drones produced by the (1x2) hybrid queens. The honey producer receives (3x4) hybrid queens mated to (1x2) drones, and the worker offspring in his colonies will be four-way hybrids—(1x2) x (3x4).

The test queens produced at Kelleys Island and distributed by the Honey Bee Improvement Cooperative Association are produced in this manner. In the last article of this series we will describe the methods and practices used in large-scale production of four-way hybrid bees at Kelleys Island.

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V. Production of Hybrid Queens

EVERY commercial queen producer has his own system of management. Just what system will produce the best quality queens at the most economical cost is not definitely known. Having been engaged in the breeding and testing of bees for a number of years, we have naturally tried many methods. Here-with we will describe the method that we use for producing large numbers of hybrid test queens at the Kelleys Island, Ohio, mating station. We use this method because we have found that it produces the largest proportion of good queens at the most economical cost of any method that we have used.

In testing stocks we, of course, must have environmental factors of queen rearing as nearly constant as

possible and must also attempt to produce queens of the highest quality. In order to measure genetic differences we must create an environment that will permit the genetic potentialities to be expressed to their fullest extent.

We begin to prepare for queen rearing in the fall. Six months before we intend to make the first graft of queen cells we requeen every colony on the island with queens from which we desire drones the following year. Let us call these queens hybrids (1x2), produced by crossing inbred lines 1 and 2. Into every colony we introduce a (1x2) hybrid queen. Since the island is isolated and every colony has a (1x2) hybrid queen, we will have control of the drone population the

following year. We clip all queens and mark them so that they can be identified and found easily.

All colonies must have ample populations of worker bees to winter properly and build up rapidly in the spring. They must also have enough honey properly organized for wintering plus plenty of reserve for build-up in the spring.

Our colonies are in square hive bodies. Each hive body holds 12 frames of the Dadant shallow size. We winter each colony in four hive bodies. When ready for winter, the colony has a (1x2) hybrid queen, a good population of young bees, 70 to 80 pounds of honey, and several frames of pollen. The bottom entrance block is placed in the entrance with a $\frac{3}{4}$ -inch opening for a

lower entrance. A 1-inch auger hole in the second hive body from the top gives an upper entrance near the location of the winter cluster of bees. Colonies prepared in this manner in the fall need little, if any, attention during the winter.

About March 15 we check the colonies for queens, brood, honey, and pollen. At that time most of the cluster of bees and the brood nest should be in the top hive body. If not, we organize it so that supplemental feeding of pollen cakes can proceed in an orderly fashion. We give the colonies 1- to 1½-pound pollen cakes by placing them on the top bars between the frames and the reversed inner cover. We give additional pollen cakes as needed—one every week or 10 days—until the supply of natural pollen is adequate. We usually have to feed pollen cakes until about May 10, or at the beginning of apple blossoming.

When we give the third pollen cakes to the colonies, we add a drone comb to the brood nest. The colonies get an additional drone comb with the next pollen cakes. All swarm-box and finishing colonies get two to four good drone combs, and they remain part of the brood nest throughout the season.

From the 4 to 6 frames with brood on March 15 the colonies should expand their brood nests to about 20 frames with brood on May 10. As soon as the colonies have brood in two hive bodies, we interchange the upper two bodies so that the brood nest can expand rapidly. When the brood nest expands to the three hive bodies, we interchange the top and bottom bodies around the middle hive body with brood. At about that time we are ready to begin queen rearing.

In our operations we plan to make our first graft of queen cells about May 10. To insure prompt matings it is necessary to have adult drones in the hives when the first graft of queen cells is made. To have adult drones on May 10 we must have drone eggs on April 15. Thus we give our colonies drone combs about April 10 with the third round of pollen cakes.

Drone combs are very important in our operations. We inspect them every time we work the colonies. We never allow them to be shoved outside the brood nest or to be filled with honey. We have a standing rule that when we cannot find plenty of drone eggs, larvae, pupae, and adults, we stop all work and put these good dark drone combs in

the middle of the brood nests so that the queens will fill them with drone eggs. One has to run low on drones only once to find out how valuable they are in queen production.

For starting queen cells we use a modification of the swarm box. To prepare a swarm box we find the queen in the colony and set the frame she is on aside. We set the top hive body on a screen division board on top of the reversed hive cover on the ground. In this hive body we have 10 frames. At least three of the frames in the center must be one-half to two-thirds filled with open honey and pollen. The other six frames should have a little honey but should not be heavy with sealed honey. We shake the bees from 8 to 10 frames of brood into this hive body to get about four pounds of bees into this body. We place the inner cover immediately on this hive body to prevent too many of the bees from flying off or crawling over the sides of the hive body. The rest of the colony with the queen we put together and place this modified swarm box on top of the hive, followed by the outer cover. We thus have a swarm box made up on top of a colony with the bees and equipment from that colony.

The bees confined in the swarm box are the nurse bees from the brood nest. They have no brood until they receive grafted queen cells. They are supplied with frames containing honey and pollen and are separated from the hive by a screen division board. The temperature of the bees in the swarm box is partly controlled by the colony below through the screen division board. Unless the colony is in the sun during excessive hot weather, we do not fear overheating the bees in the swarm box. However, these swarm boxes, as well as the other colonies used in queen rearing, should be in the shade.

In our operations we make up swarm boxes on four colonies just before noon. Immediately after lunch we give each swarm box four bars of grafted cells. For the first graft we use large larvae (about 36 to 48 hours old) and graft into dry cells. Any colony, usually a swarm-box colony, is the source of the larvae for this first graft. Each bar of cells has 20 to 23 cells. Two bars of cells to a frame and two frames of grafted cells to each swarm box give approximately 80 to 90 grafted cells to each swarm box.

We place the two frames of grafted cells in the swarm box with one frame containing honey and pollen between the frames of queen cells and the other frames of honey and pollen next to the frames of queen cells. When removing the outer cover and the inner cover to insert the frames of queen cells into the swarm box, we use very little smoke and do not worry about the few bees that fly out. These are field bees anyway and are of little value in the swarm box.

Twenty-four hours later (the next afternoon) we remove the cover from the swarm-box colony, lift the swarm box off, remove the screen division board, replace the swarm box on the top of the colony (now united by the removal of the screen), remove the two frames of queen cells, push the remaining frames together, add two empty combs to fill up the space, put the inner cover on the hive, place the screen division board on it, and finally put the outer cover over all. When the colony is closed, all we have left on the outside are two frames of queen cells that are well supplied with royal jelly and covered with bees.

After we have emptied all four swarm boxes, we shake the bees off the started queen cells and take them to the grafting house. Here we consolidate the accepted cells of the 16 bars of cells into 12 bars. Each bar has 20 to 23 queen cells. We now remove the queen larvae from these queen cells and are ready to regraft into queen cells that are well supplied with royal jelly.

We regraft with 24-hour larvae from our breeder queens and give one bar of regrafted queen cells to each finishing colony.

The finishing colony is most important in the rearing of good queens. It is a very populous, queen-right colony in which brood rearing is never restricted. To provide ample space for such a colony, at least four shallow hive bodies are necessary. We use the upper hive body for finishing queen cells and confine the queen to the lower three hive bodies by an excluder. We place the frame for queen cells in the middle of the upper hive body between frames of unsealed brood.

On the day before giving a finishing colony a bar of queen cells, we raise three to six frames of unsealed brood and move the older sealed brood to the brood nest below the excluder. The main brood nest of the queen in the finishing colony is in the hive body just below the

excluder. She can expand into the lower hive bodies.

When working a finishing colony to raise young brood, we put the heavy combs of honey to the outside in the lower hive body and move up partly filled combs to the upper hive body so that the brood nest does not become crowded. The bees will keep moving the honey up if it is not sealed. It is not necessary to feed a good colony of bees to stimulate or keep brood rearing or queen rearing progressing at a high rate, provided the colony has ample food.

When a heavy honeyflow is on, we add a fifth shallow hive body, placing it on top. This helps to keep the brood nest open by giving more space outside the brood nest for nectar storage. However, it is not advisable to have frames of empty combs just above the queen cells. They should be near the outside of the super.

Seven days later we give another bar of cells to the finishing colony after young brood has been raised above the excluder.

Ten days after the queen cells are regrafted we put the cells into the nuclei at the mating yard. Our nuclei hold four shallow frames. We use a triple "nuc" box formed by dividing a super into three compartments. Each "nuc" has two auger holes one below the other, and the three nuclei in each box have similar holes on different sides of the hives.

No colonies are placed in the mating yard. Strong colonies may incite robbing with the result that nuclei may be robbed out. Our drone-producing colonies are our cell-finishing, swarm-box and reserve colonies located no closer than 250 yards from the "nuc" yard.

The breeder queens used on the island are of one inbred line artificially mated to drones of another. They are not so vigorous or such good layers as noninbred or hybrid queens, and therefore require different care than the commercial queen producer is accustomed to providing.

These breeder colonies must get help from other colonies to insure adequate numbers of young bees to keep them populous. If a breeder colony is allowed to become weak, the egg production of the queen will decline and the larvae used in grafting to produce hybrid queens will not be well supplied with royal jelly previous to grafting. Should this happen, both the quality and the quantity of the hybrid queens will

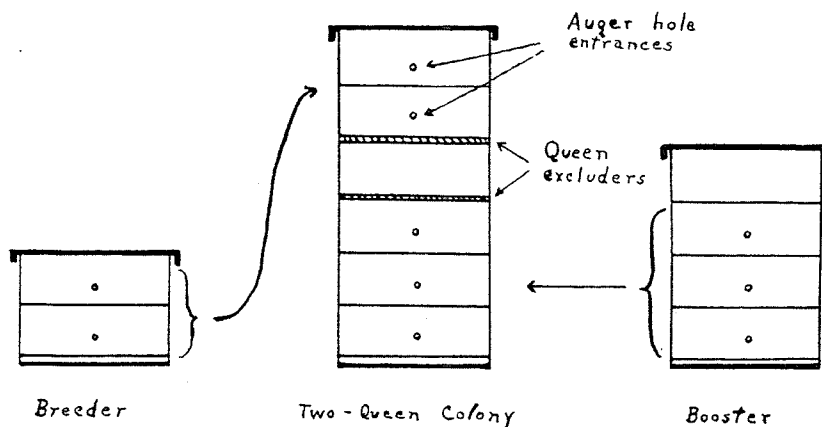


Figure 1. Diagram showing establishment of a breeder colony above a booster colony.

be lowered.

To keep an ample population in a breeder colony, we must add young bees at frequent intervals. In our operations at Kelleys Island we use the two-queen colony system. Figure 1 shows how such a colony is made up and organized. It is not difficult to operate, and its use insures a steady addition of young bees to the breeder colony without great danger of losing the breeder queen.

We introduce the breeder queen into a newly made-up colony. When she is well established and laying satisfactorily, this colony becomes the top unit of a two-queen colony. The queen in the lower unit, or booster colony, is a good vigorous hybrid queen and furnishes young bees for strengthening the upper unit.

The procedure for establishing a two-queen colony is as follows: We move a first-class colony to a location convenient to the grafting house. This colony is in a shallow four-story hive with the brood nest in the lower three hive bodies. After the colony becomes established, in two or three days, we consolidate the queen and all her brood in the lower three hive bodies, place an excluder above and below the fourth hive body, and the two-story breeder colony on top of this booster colony. All hive bodies with brood have 1-inch auger-hole openings at the front of the hive. The entrances to the two units of this now two-queen colony are on the same side of the hive. The bees of both units mingle, but the two brood nests are separated by two queen excluders and a super for storage between the excluders. Bees from the lower brood nest go to the upper brood nest through the excluders and help keep this upper unit populous. Returning field bees, using the auger-hole en-

trances, tend to drift to the upper breeder unit, keeping the population ample in this unit.

During a honeyflow the bees tend to crowd honey into the brood nest of the breeder unit. Since the queen producer is grafting from this colony every few days, he can easily keep an open brood nest in the breeder nest by moving frames of honey to the lower unit as space for brood rearing or honey storage is needed. It is advisable to check the lower unit of this colony once a week to prevent overcrowding or swarming.

Since the two brood nests are separated by two excluders and the intervening super, there is no danger of the two queens coming in contact with each other. Because there is always danger of a virgin queen entering the breeder colony and killing a valuable queen, a small square of zinc excluder is placed over the auger-hole entrances of the breeder unit. Bees passing through zinc excluders lose part of their pollen loads. Thus it is advisable to watch the supply of pollen in the breeder unit and add a frame of pollen when necessary to insure proper feeding of the larvae.

The queens produced at Kelleys Island are two-way hybrids. An inbred queen of line 3 artificially mated to drones from inbred line 4 is the source of the hybrid queens (3 x 4). The beekeeper who purchases these queens has hybrid queens (3 x 4) heading his colonies. Since all drones on the island are sons of (1 x 2) hybrid queens, the worker daughters of these hybrids queens will be (1 x 2) x (3 x 4) double hybrid workers in a colony headed by a single hybrid queen. If, in addition, the opposing sex alleles in the final

cross are different, the hatchability will approach 100 per cent and the brood quality will be excellent.

To provide the bee industry with the foundation stock for the production of such scientifically compounded hybrids, adapted to various purposes and various localities, is the objective of the Kelleys Island breeding project that is being conducted in cooperation with the Honey Bee Improvement Coopera-

tive Association.

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