1. Creating True Breeding Strains By Vic High

I've been hearing a fair bit of confusion from many on how to create a true breeding strain and so I'm writing this page to try and help shed some light on the subject. There are a few situations where a plant breeder would want to create a true breeding strain (IBL) and a few ways of accomplishing the task. But understanding the subtle differences of the various techniques is not so easy. This paper will attempt to give a basic understanding of what is actually happening with each technique and then apply what is learned to actual projetcs. As a friend worked overtime making sure I didn't forget, breeding is not a black and white subject and as a whole, it would be too complex to put on paper in an easily understood form. Therefore, I will create small fictional examples to reinforce various concepts and then we will take those examples and concepts and apply some reality to them. Try not to get hung up on the erroneous assuptions used here such as flavour being monogenic, the assumption is simply used to make it easier to learn a certain concept.

Just What Is It That We Are Doing?

Before we dive in, maybe we should take the time to understand what we are trying to accomplish when we set out to create a true breeding strain. There are hundreds of possible phenotypic traits that we could observe within a cannabis population. Are we trying to make all of them the same and remove ALL variation? Not likely, the genetic code is just too complex to try. Plus, since phenotype (what we see) is 1/2 genotype + 1/2 environment, everytime the population was grown under new conditions, new heterozygous traits would be observed. Basically, all we are trying to create is an overall uniformity while not worrying about the minor individual varioations. No different than a dog breed. You can look at a germanshepard and recognise it as belonging to a discrete breed. But if you look closer at several germanshepards all at the same time, you will find variations with each and every one of them. Some will be a little taller, some a little wider, some more agressive, some a little fatter, some darker, etc. But they would all fall within an acceptable range for the various traits. Generally speaking, this is what a plant breeder is trying to accomplish when creating a true breeding strain, or IBL.

However this isn't always the case. Sometimes a breeder will just concentrate on a specific trait, like say outdoor harvest date, or mite resistance. You could still have a population where some are 2' bushes and some 10' trees. In this case, you would say that the strain was true breeding for the particular trait, but you wouldn't consider it a true breeding strain per se. In genetics, wording plays a big part in meaning and understanding. As does reference point as my F1 vs F2 comparison page illustrates.

Ok, so we want to make a cannabis population fairly uniform over a few phenotypically important traits, like say flavour for instance. For simplicity sake, we'll just deal with the single trait flavour, it's complex enough. And although flavour is controlled by several gene pairs (polygenic), we'll make the simplistic assumption that it's controlled by a single gene pair (monogenic) for many of the models and examples in this paper. There are many flavours such as chocolate, vanilla, musky, skunky, blueberry, etc, but in this paper we'll just deal with two flavours, pine and pineapple. Either gene in the gene pair can code for either of the flavours. If both genes code for pineapple or both genes code for pine flavour, we say that the gene pair (and individual plant) is homozygous for flavour. If the one gene codes for pine and the other codes for pineapple, we say that the gene pair (and individual plant) is heterozyous with respect to flavour. The heterozygous individual can create gametes (pollen

or ovules) that can code for either pine flavour or pineapple flavour, the homozygous individuals can only create gametes that code for one OR the other. A homozygous individual is considered true breeding and a heterozygous individual is not.

However, as the words imply, when we are creating a true breeding strain, we are looking at a population, not individuals. We are trying to make all the individuals in the population homozygous for a particular trait or group of traits. Lets say we have a population of 50 individual plants, and each plant has has a gene pair coding for flavour. That means that 100 flavour genes make up the flavourgenepool (reality is much more complex). When trying to create a true breeding strain, we are in fact trying to make all 100 of those genes code for the same trait (pineappleflavour in our case). The closer our population comes getting all 100 genes the same, the more homozygous or true breeding it becomes. We use the terminology gene frequency to measure and describe this concept, where gene frequency is simply the ratio or percentage of the population that actually contains a specific gene. The higher the gene frequency, the more true breeding the population is. A fixed trait is where the gene frequency of the trait reaches 100%.

And folks, this is the basic backbone of what breeding is all about, manipulating gene frequencies. It doesn't matter if your making IBL, F1s, F2s, selecting for this or selecting for that, all you are really doing is manipulating gene frequencies. Therefore, to ever really understand what is happening in any breeding project, the breeder must pay attention to gene frequencies and assess how his selective pressures and models are influencing them. They are his measure of success.

An overview of Inbreeding Strategies
What are we trying to create a true breeding strain from?

This a good question. Sometimes a gardener will notice a sport or unique individual in an IBL or F2 population, like say it has pineapple flavour when the rest have pine flavour. For one reason or another he decides he wants to preserve this new trait or combination of traits from that single individual. For the sake of ease of comprehension, we tend to call this special unique individual the P1 mom. He could start by selfing the individual OR breeding that individual with another and create what can be described as F1 offspring. If the F1 route was chosen, then breeders can diverge down two new paths. Some breeders will take the progeny of the F1 crossing and breed it back to the P1 mom, and then repeat for a couple more generations. This is referred to as backcrossing or cubing by cannabis breeders. Another common strategy is to make F2 progeny from the F1 population and then look for individuals that match the P1 mom. They would repeat the process for a few generations. We can call this filial or generational inbreeding since the parents from each cross belong to the same generation.

In another situation, sometimes a farmer will notice a few individuals in his fields that stand out from the crowd in a possitive manner. Like say the are resistant to a problem pest. In this case, he will collect the best of the individuals and his starting population will contain several similar individuals and not a unique single individual as in the previous example. He would skip the hybridizing step (making the F1s) and go straight to the generational inbreeding step.
Cubing the Clone
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A)* In this first situation, we'll deal with the situation where a plant breeder finds a special

individual or clone.
It's a natural thing to be curious and cross a couple of plants that catch your fancy. Grow them out and find a new variation that you like even better. We can preserve the new variation through cloning indefinately, but accidents happen and clones die.* They can get viruses or can suffer clonal deprivation from somatic mutations over time. Plus it's harder to share clones with friends through the mail than seeds. So it's only natural that we would want to create seed backups of this special clone.
But before we start breeding this clone, we should try and figure what exactly it is we want from the seeds we are going to create. Do we want them to simply be able to reproduce individuals like the special clone?* Simple backcrossing (cubing) will accomplish this. Or do we want to to create seeds that will be able to create more seeds like the special clone, a true breeding strain? These are very different in nature. You see, chances are that your special clone will be heterozygous for many of traits she phenotypically expresses. This just means that she will contain genetic information (genes) for two opposing triats, but you can only see one, the dominant one. However, her seeds will only get one or the other of the genes, so her offspring will express all the genetic information she has, including what you can't see within herself. If you want to create a true breeding strain, you need to preserve all the genes you can see, and remove all the genes that you cannot, but may show up in the offspring. Creating homozygosity.* The only way to accomplish this is through selection and generational inbreeding (selecting the homozygous offspring to be parents for the next generation).**
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BackCrossing and Cubing
Backcrossing is where you breed an individual (your special clone) with it's progeny.* Sick in our world, but plants seem to like it*
1) Your first backcross is just a backcross.
2) Your second backcross where you take the progeny from the first backcross and cross back to the SAME parent (grandparent now) is often called SQUARING by plant breeders.
3)* Your third backcross where you take the* progency (squared) from the second* backcross and cross back to the SAME parent (great grandparent now) is often* called CUBING by plant breeders. You can continue the backcrossing but we just call this backcrossing. Cubing is in reference to the number three, as in 3 backcrosses*
Cubing works on the basis of mathamatical probabilities with respect to gene frequencies. The more males you use with each cross, the better the chance that your reality matches the theory. In theory,* with the first backcross, 75% of your genepool will match the genepool of the P1 parent being cubed. Squaring increases this to 87.5% and cubing increases it to 93.75%.* You can arrive at these numbers by taking the average between the two parents making up the cross. For instance, you start by crossing the P1 mom (100%) with and unrelated male (0%)* getting 100% + 0% divided by 2 = 50%. Therefore, the offspring of this first cross are loosly thought of as being 50% like the mom. Take these and do your first backcross and you get 100% (mom) + 50% divided by 2 = 75%. And this is where we get the 75% for the first backcross. Same thing applies as you do more backcrosses. As you will see later, you can apply this same probability math to specific genes or traits, and this can have a dramatic effect on your methodology and selection methods.*
Your selection of the right males for each backcross are the crucial points for success with this technique. In each case, you could select males that contain the genes you want, or you could inadvertedly pick those individuals that carry the unwanted recessive genes. Or more likely, you could just pick individuals that are heterozygous for both genes like the P1 mom being backcrossed. The easiest way to deal with this is to start by only looking at one gene and one trait, like lets assume that flavour is determined by a single gene (in reality it's probably not). And do some punnet squares to show gene frequencies through 3 generations

of* backcrossing. Now lets assume that we found a special pineapple flavoured individual in our pine flavoured population that we wanted to keep. The gene causing the pineapple flavour could be dominant or recessive and the selection abilities and cubing outcome* will be different in both cases.

a) pineappleflavour is dominant.

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P = pineapple flavour and p = pine flavour

Therefore since each individual will have two flavour genes paired up, the possible genotypes are PP, Pp, and pp. Since P is dominant, PP and Pp will express pineapple flavour while pp will exhibit pine flavour, these are their phenotypes. Now since the pineapple is a new flavour, chances are that the special individual will be heterozygous, or more specifically, Pp. Therefore, the only possible parent combination is Pp X pp with the Pp being the parent to be cubed.

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Figure 1. The F1 cross

Now most will find it tough to pick males with the gene for pineapple flavour since males don't produce female flowers. Therefore, they will select males randomly and blindly with respect to this trait. The ratio of* P to p genes of the male F1 generation to be used in the first backcross will be 2:6. Another way to look at it is to say that the P gene fequency is 25%.* This means that one out of four pollen grains will contain the gene for pineapple flavour. Here is how this plays out in the first backcross.

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Figure 2. The B1 cross

Now it's this first backcross that first creates an individual that is homozygous (PP) for the pineapple flavour. However, again because of our limited selection abilities, we choose males randomly. From the random males we should expect* three out of eight pollen grains to to contain the gene for pineapple flavour. The P1 female will still contribute one P gene for every p gene. I'll spare your computor's memory and* and not post the table, feel free to do it yorself though on paper to be sure you understand what happening*

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The* second backcross (Squaring) will produce the following:

3 PP 8 Pp** 5 pp**

Therefore,* 68.75% will have pineapple flavour and 31.25% will have pine flavour. The frequency of the P gene has risen to 7/16 or* 43.75%.

And finally, the third backcross (Cubing) will net the following genotypic ratios:

7PP 16Pp*** 9pp**

Therefore,* 71.875%* will have pineapple flavour after cubing has been completed. Roughly 22% (7/32*100) of the cubed progeny will be true breeding for the pineapple flavour. The frequency of the P gene has risen to roughly 47% (30/64).

In conclusion, if the backcrossing continued indefinately with random selection of males and with large enough of a population size,* the frequency of the P gene would max out at 50%. This means that the best that can be expected from cubing is 25% true breeding for pineapple flavour and 75% that will display the pineapple flavour. You would never be rid of the 25% that would maintain the pine flavour. This model would hold true when trying to cube any heterozygous trait.

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b) Pineapple flavour is recessive

In this case, P is for the pine flavour and p is for pineapple flavour. Convention is that the capital letter signifies dominance. For the breeder to have noticed the interesting trait, the mom to be cubed would have to be homozygous for the pineapple flavour (pp). Depending where the male came from and whether it was related, it could be Pp or PP, with PP being

more likely. It won't make much difference which in the outcome.
F1 cross* is pretty basic, we'll skip the diagram. We simply cross the female (pp) with the male (PP) and get offspring that are all Pp. Since the pine flavour is recessive, none of the F1 offspring will have pineapple flavour (hint*). However, the frequency of the gene p will be 50%.
pp X PP = Pp + Pp + Pp + Pp
Since the F1 generation are all the same (Pp), the pollen it donates to the first backcross will contain a p gene for every P gene. The first backcross will be:
B1 = pp X Pp = Pp + Pp + pp + pp
As you can see, 50% of the offspring will be pineapple flavoured and the frequency of the p gene is 6/8 or 75%. This B1 generation will generate pollen containing 6 p genes for every 2 P genes.
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Figure 3.The second backcross.

As you can see, the second backcross or squaring produces pineapple flavour in 75% of the offspring. And the p gene frequency within those offspring is roughly 88%. (Remember C88*). Of the pollen grains from this squaring, 14 out of 16 will carry the p gene for pineapple flavouring. When they are backcrossed to the P1 mom for the third time, they net the following cubed progeny:
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Figure 4. The third backcross

Aftercubing of a homozygous gene pair, we end up with roughly 88% of them displaying the desired trait (pineapple flavour in this case) and also being true breeding for that same trait. The frequency of this desired gene will be roughly 94%. If the backcrossing was to continue indefinately, the gene frequency would continue to approach 100% but never entirely get there.
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It should be noted that the above examples assume no selective pressure and large enough population sizes to ensure random matings. As the number of males used in each generation decreases, the greater the selective pressure whether intended or not. The significance of a breeding population size and selective pressure is much greater when the traits to be cubed are heterozygous. And most importantly, the above examples only take into account for a single gene pair.
In reality, most of the traits we select for like potency are influenced by several traits. Then the math gets more complicated if you want to figure out the success rate of a cubing project. Generally speaking, you multiply the probabilities of achieving each trait against each other. For example, if your pineapple trait was influenced by 2 seperate recessive genes, then you would multiply 87.5% * 87.5%* (.875 * .875 *100) and get 76.6%. This means that 76.6% of the offspring would be pineapple flavoured. Now lets say the pineapple trait is influenced by 2 recessive traits and and a heterozygous dominant one. We would multiply 87.5% by 87.5% by 71.9% (.875*.875*.719*100) and get 55%. Just by increasing to three genes, we have decreased the number of cubed offspring having pineapple flavouring down to 55%. Therefore, cubing is a good technique where you want to increase the frequency of a few genes (this is an important point to remember*), but as the project increases, the chance of success decreases .... at least without some level of selective pressure.
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Applying the pressure
The best way to significantly increase your chances of success is to apply intended selective

pressure and eliminate unintentional selective pressure. Try to find clearcut and efficient ways to isolate and select for and against certain traits. Find ways to be sure your males are passing along the intended traits and remove all males that do not. This includes ALL traits that may be selected for. Some traits you will be able to observe directly in the males. Other traits like flowering duration you may not. If you are selecting for a trait you can't directly observe, you want to do some*progeny tests and determine which males pass on the most desireable genes. I'll explain more on progeny tests later.
It's important that when chosing your best males to ignore the superficial traits having nothing to do with the real traits your looking for. You see, cannabis has several thousand genes residing on just 10 chromosome pairs or* 20 individual chromosomes. Therefore each chomosome contains hundred of genes. Each gene residing on the same chromosome is said to be linked to each other. Generally speaking, they travel as a group* . If you select for one of them, you are actually selecting for all of the traits on the chromosome. There is an exception to this rule refferred to as breaking linked genes via crossing over, but for simplicity sake, we will ignore that for now. Getting back to selection, you could decide to select for a trait such as you like the spikey look of the leaves while really being interested in fixing the grapefruit flavour. But as it may happen, both traits may be on the same chromosome pair but opposite chromosomes. If so, as long as you select the plants with spikey leaves, you will never get the grapefruit flavour you really want. It's good to keep in mind that each time you select for a triat, you are selecting against several hundred genes* This is why most serious breeders learn to take small methodical steps and work on one or two traits at a time. Especially with inbreeding projects such as selfing and backcrossing.
Now lets see what kind of improvements we can make in the first example of trying to cube a heterozygous dominant trait using some selective pressure. Lets say that with each generation, we are able to remove the individuals recessive for the pine flavour (pp), but can't remove the heterozygous ones (Pp). If you recall, our P1 mom had the genotype (Pp) in that model and the F1 cross yielded (Pp + Pp + pp + pp) as possible offspring combinations. We remove the two (pp) individuals leaving us with only Pp. Therefore our first backcross will be:
Pp * Pp = PP + Pp + Pp + pp
Again we remove the pp individual leaving us with PP + 2Pp. Going into the second backcross we have increased our P gene frequency from 37.5% up to 66.7%. This means that going into the second backcross 4 of every six pollen grains will carry the P gene. The outcome is as follows
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As you can see, after selecting against the homozygous recessives for 2 backcrosses, we have increased our P gene frequency to 58% from 44% in our squared population. If we again remove the homozygous recessives, our gene frequency increases to 70% (14/20) going into the third backcross, meaning that 7 out of 10 pollen grains will carry the P gene. Again, I'll spare your PC's memory and just give your the results of the third backcross.
B3 cross = 7 PP + 10 Pp + 3 pp
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This translates to mean that 95% of the progeny will taste like pineapple after cubing a heterozygous dominant strain if* the homozygous pine tasting ones are removed prior to to each backcross. This is an improvent from 72% when no selection occurred.* The frequency of individuals true breeding for the pineapple flavour rose to 35%.* But more importantly, the P gene frequency improves to 60%. This will be an important consideration when we discuss progeny testing* .
But for now lets recap the percentage of individuals true breeding for the pineapple taste in each of the models. In the case where the pineapple flavour trait is heterozygous dominant

and no selective pressure is used, cubing produced 22% true breeding individuals. By selecting against the homozygous pine recessive, we were able to increase this too 35%. And finally, when cubing a homozygous recessive gene, we are able to achieve a cubed population that is 87.5% true breeding for the pineapple flavour. And as I pointed out earlier, these numbers only apply to single gene traits. Lets say the pineapple flavour is coded by two seperate genes, one dominant and one recessive, and you are able to select against the homozygous recessive pine flavour while selecting for the dominant pineapple flavour gene. Your cubed population would then contain 87.5% * 35% (.875 * .35 * 100) =* 30% true breeding individuals. As you can see, as long as the cubed source is heterozygous, it doesn't matter how many backcrosses you do, you will never achieve a true breeding strain.

We can often get get hung up on terminology and lose sight of what is really trying to be said. For this paper, when I discuss inbreeding, I'm talking about crossing individuals from the same generation and not backcrossing. I've haphazardly referred to this as generational inbreeding, although I'm not certain such terminology is considered accurate, haha. Also, in my F1 vs F2 discussion paper, I try to finely define terms such IBL, F1, and F2 from the perspective of a seed vendour and seed buyer. Those definitions won't apply here and I'll rely upon the most generic definitions of those terms for this discussion.

Your starting point of an inbreeding project can involve two parents that are related or two parents that are not. You could even start with a single parent and self it. In each case, we will arbitrarily assign the parents making up the starting point the P1 parents. In a typical inbreeding project, the progeny of the P1 parents will be called the F1 cross. When you cross individuals from an F1 generation together, you get an F2 generation. Cross the F2 generation and you get an F3 generation. The F3 generation gives rise to the F4 generation, and likewise, the F4 gives rise to the F5 generation. A similar inbreeding strategy can also be applied as a followup to a selfing or backcrossing project. We will first take a close look at how we can manipulate gene frequencies by solely working with generational inbreeding. Lets say we want to stabilize the pineapple flavour of a special individual within a pine flavoured population. The genes controlling the pine flavour could be dominant or recessive. This fact can greatly influence the success of the project.

If the pineapple flavour is controlled by a dominant gene, there is a good chance the indivual will be heterozygous (Pp) where P symbolizes pineapple flavour and p symbolizes pine flavour. It can also safely be assumed that other individuals in the population are homozygous (pp) for the pine gene. Therefore our F1 cross will be:

F1 cross = Pp x pp = Pp + Pp + pp + pp

50% of the F1 generation will be pineapple flavoured and the frequency of the pineapple (P) gene will be 2/8 or 25%. Natually when selecting parents for the F2 generation, we would choose ones that were pineapple flavoured and therefore they would all be heterozygous (Pp). By being able to select both sets of parents, we call this a full sib cross. Again due to it's common simplicity I'll spare you the punnet square, we can determine the genetic combinations of our F2 population in our heads.

F2 (a) cross = Pp x Pp = PP + Pp + Pp + pp - typical mendelian phenotypic 3:1 and genotypic1:2:1 ratios.

75% of the F2 population will have pineapple flavour and our frequency of the P gene is now 4/8 or 50%. Now moving onto the F3 (a) generation gets a little harder to do in our heads. Again spotting the pine flavoured (pp) individuals should be easy and therefore removed from the breeding population. This leaves us with the PP + Pp + Pp individuals to make up the breeding population. We shorten it to PPPPpp to indicate the breeding population's genotype and frequency of the P gene. Since it can evenly be divided by 2, PPp just as accurately symbolizes the same genotype. Therefore the two parents become PPp x PPp. Each individual letter can represent the frequency of a single gamete (pollen or ovule) in the breeding population.

Fig 1: F3 (a) Cross

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If we continued into the F5 generation using the same selective pressure, we would end up with 144PP + 72Pp + 9pp which translates into 96% of the population tasting like pineapple. The frequency of the pineapple gene would have risen to 80%.
b) But lets say that in all reality, that we can't determine flavour in the males, so we can only remove the pine (pp) flavoured individuals from the female parents. We call this a half-sib cross when we can't select our pollen source. So we would be doing two crosses Pp x Pp and Pp x pp. A shortcut is to combine the various genotypes into one and just write Pp x Pppp. I'll skip the punnet square on this one, but please feel free to do one yourself to be sure you understand what's happening. If you don't, it should become obvious when I go through the F3 cross in detail.
F2 (b) cross = Pp x Pppp = PP + Pp + Pp + Pp + Pp + pp + pp + pp
62.5% (5/8 of the halfsib F2 population would be pineapple flavoured and the frequency of the P gene would be 6/16 or 37.5%. This is quite a decrease by simply not being able to remove the pp males from the breeding population. I'll carry this one more generation (F3 cross) in detail to show you the developing patterns. After removing the pine flavoured (pp) females, our female genepool would be PP+Pp+Pp+Pp+Pp = PPPPPPpppp = PPPpp. Without any selective pressure, our male genepool would remain PP+Pp+Pp+Pp+Pp+pp+pp+pp = PPPPPPpppppppppp = PPPppppp. Here's how the cross plays out.
As you can see, the F3 cross yields pineapple flavour in 75% (30/40) of the offspring. The frequency of the P gene has risen to 48.8% (39/80).
Lets look at the mathematical patterns developing. To recap, this F3 cross was PPPpp x PPPppppp. Lets rewrite it in a simpler fashion that expresses the ratio of each gene (or gamete). We would get 3P2p x 3P5p. If you note, when we add up the numerical value of each side of the cross and then multipy them (3+2)*(3+5) we get the number 40, which turns out is the same as the number of offspring created by the punnett square. Notice that when we multiply the two 3Ps, we get 9P, the same number of PP individuals from the punnett square? The same pattern holds for each combination, so what we have here is a simple way of calculating a punnet square outcome without actually drawing the punnet square. This can save alot of time when we get into complex combinations. So lets use the mathematical method of determining the results from the above F3 cross.
3P2p x 3P5p = (3*3)P + (3*5)Pp + (2*3)Pp + (2*5)pp = 9PP + 15Pp + 6Pp + 10pp = 9PP + 21Pp + 10pp
As you can see we came up with the same number as the punnet square without drawing all the lines. Now lets use the same formula to calculate the F4 and F5 generations. We will remove the 10pp from the female genepool and be left with 9PP + 21Pp. If we add up all the P's and p's, this works out to [2(9) + 21]P and 21p which translates to 39P21p. The male gene pool will work out to be [2(9)+21]P and [21+2(10)]p = 39P41p. Remember, each number infront of each gene simply represents the frequency of that particular gene.
F4 Cross:
39P21p x 39P41p = 39*39PP + 39*41Pp + 21*39Pp + 21*41pp = 1521PP + 1599Pp + 819Pp + 861pp
= 1521PP + 2418Pp + 861pp and these add up to 4800
Therefore (1521+2418/4800 or 82% will have pineapple flavouring and the frequency of the P gene will be 2(1521)+2418/2(4800) begin_of_the_skype_highlighting +2418/2(4800) end_of_the_skype_highlighting = 5460/9600 or 56.8%.
Can you imagine doing that with a punnet square? Even so, as you can see, the literal numbers are getting a little crazy and are becoming hard to follow. It may be easier to start working with gene frequencies in terms of decimals or simply percentages. Percentages are the easiest to follow but there is a trick or two to remember, so I'll stick with simple decimals. So lets move onto the F5 generation using decimals to indicate frequencies. First we have to calculate the gene frequencies of each parental genepool. If you recall, the F4

cross created the following genepool. 1521PP + 2418Pp + 861pp with a total of 4800. When we translate each ratio into a decimal we get 1521/4800PP + 2418/4800Pp + 861/4800pp = .32PP + .50Pp + .18pp after rounding to two decimal places. [Hint: Note that if we add up all our decimals, we get a total of 1. If they don't, a mistake was made.]
So now lets use the ratios we got from the F4 generation to calculate the gene frequencies of the parental genepools of the F5 cross. All we do is add together the frequency of each gene (gamete) and divide by the total of the ratio used in that genepool. Since the Pp is only half P, we divide this one in half. Therefore, the male parental genepool will be (.32+.25)/1 which equals .57P. Once we know P, we automatically know p since it is simply 1-P or .43. Therefore, the male parental genepool is .57P.43p. Now to determine the female gene frequencies, we need to subract the .18pp from the total numbers since they will be removed from the genepool. This is one way to do it. (.32+.25)/1-.18 = .57/.82 = .695P. Again 1-P=p so we end up with the female gene frequencies of .695P.305p. The F5 cross is as follows:
.695P.305p x .57P.43p = (.695*.57)PP + (.695*.43)Pp + (.303*.57)Pp + (.305*.43)pp = .40PP + .47Pp + .13pp
Finally in conclusion, after 4 generations of inbreeding where we only make our selections from the female population, we end up with an F5 population where 87% taste like pineapple. Plus the frequency of the pineapple gene in the F5 population is 63.5%. This is significantly less than if we were able to apply the selective pressure to both parental genepools. If you recall, in that situation we achieved 96% of the population tasting like pineapple. The frequency of the pineapple gene would have risen to 80%. This is just the case where the flavour gene is dominant, the situation when selecting for recessive traits is much nicer.
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When the trait we want is recessive.
In this case, we will assign the symbol p to indicate pineapple flavour and P will indicate Pine flavour. If we find a single pineapple flavoured individual in a population of pine flavoured individuals, and the trait is recessive, then the individual must be homozygous (pp) for the trait. When choosing a mate to cross it with, there is a chance you could select a heterozygous individual, but it's more likely to use a homozygous dominant pine flavoured one so that is what we'll base this next model on. Therefore, our F1 cross will be:
F1 cross = pp x PP = Pp + Pp + Pp + Pp or just simply Pp since all the F1s are the same. Just to maintain consistency, I will point out that none (0%) of the F1 cross will have pineapple flavour but the frequency of the p gene will be 50%. Now when we move onto the F2 population, our parents will both be Pp. Here is the F2 cross:
F2 cross = Pp x Pp = PP + Pp + Pp + pp
Since there was no selection in choosing the parents, the p gene frequency remained at 50%. However, 25% of the offspring will be pineapple flavoured. As has been shown previously, it is this reassortment within the F2 population that is key. Now we can spot the females that are homozygous (pp) for the pineapple flavour. If we can identify the pineapple flavoured males, then we will be finished with an F3 cross as follows:
F3(a) cross = pp x pp = pp + pp + pp + pp - all true breeding for the pineapple flavour, mission accomplished 🙂
But not so fast, many are unable to determine the flavour of a male plant and so therefore wouldn't be able to perform any selections on the male portion of the genepool. Again we are back in a half-sib breeding model. The male population's population be PP + Pp + Pp + pp which equals 4P4p which in turn can be simplified to Pp. The frequency of the p gene in the female genepool will remain 100% from now on in this model. In this case the F3 cross would be:
F3 (b) cross = pp x Pp = Pp + pp + Pp + pp
50% of the F3(b) generation would be pineapple flavoured and the frequency of the pineapple (p) gene has increased to 6/8 or 75%. We would select out the PP female but use

both Pp and pp males in the F4 cross. From what we learned in the previous section we could designate our gene frequencies as the female breeding pool = .5p.5p and the male breeding pool as .25P.75p. You see where those came from? Remember that 6/8 or 75% were p from the F3(b) cross? Well the 75% simply becomes .75 when we convert to decimal form. And 1-p=P to arrive at the .25p. Hence our F4 cross is:
F4 (b) cross = .5p.5p x .25P.75p = (.5*.25)Pp + (.5*.75)pp + (.5*.25)Pp + (.5*.75)pp = .25Pp + .75pp or more simply Pp + pp + pp + pp
We'll skip to the F5 generation, see if you can figure where I get my gene frequencies.
F5 (b) cross = .5p.5p x .125P.875p = .125Pp + .875pp
So after doing half sib inbreeding for 4 generations, we achieve an F5 generation where 87.5% of the offspring will be pineapple flavoured and the frequency of the p gene will be 93.75%. Not bad at all, just as good as cubing, but the 100% we achieved with the previous full sib example was better and with two fewer generations (HINT!!).
Please keep in mind that these models assume that flavour is a trait controlled by a single gene or linked group of genes. Reality isn't as simple, but the principles mentioned here apply to more complex models as well. The main point to take from this is that the degree of selection we use can very much influence our success rate. And that selecting for dominant and recessive traits have some subtle differences.