

**1. The following paragraph, which describes some of the vegetation in the Mojave Desert is out of:**

Barbour, M.G., J.H. Burk, and W.D. Pitts. 1980. Terrestrial Plant Ecology. The Benjamin/Cummings Publishing Company, Inc. Menlo Park, CA.

Mojave summer annuals germinate in August or September after heavy rains, generally are  $C_4$  in metabolism, remain small, and mature by the time of autumn frosts. Their life span is measured in terms of weeks rather than months.

Based on this description, if you had to hazard a guess as to the DNA contents of the Mojave's summer annuals, what would it be? What factor(s) is your answer based on?

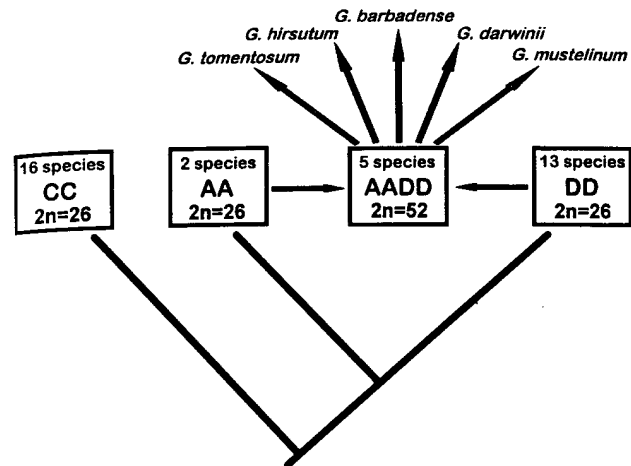
**2. In the following paper:**

Wendel, J.F., A. Schnable, and T. Seelanan. 1995. Proc. Natl. Acad. Sci. USA 92:280-284.

The authors examined the rDNA repeats from the five allopolyploid species of cotton, which are thought to have evolved as illustrated in the diagram at right.

As it turned out, both genomes from *G. mustelinum* have rDNA derived from the AA diploid species, while both genomes of *G. tomentosum*, *G. hirsutum*, *G. barbadense*, and *G. darwinii* the rDNA that came from the DD diploid species.

Name the phenomenon this represents, and diagram and explain how this happens.



Cultivar	Maturity group	pg/2C nucleus
Hardee	VIII	2.86
Jupiter	IX	2.83
Hartwig	V	2.82
Aojia	III	2.79
Resnick	III	2.78
Gnome 85	II	2.77
Maple Ridge	000	2.75
Sioux	000	2.74
Agazsi	0	2.73
Pando	000	2.71
Dawson	0	2.70
McCall	00	2.68
Corsoy 79	II	2.68
Amsoy	II	2.66
Maple Presto	000	2.51

3. The data below were selected from: Graham, M.J., C.D. Nickell, and A.L. Rayburn. 1994. Relationship between genome size and maturity group in soybean. Theor. Appl. Genet. 88:429-432.

The authors used flow cytometry to measure the DNA content of several soybean cultivars. In the United States, soybean cultivars are classified according to maturity group (MG), whereby soybeans of MG 000 are grown in Canada, MG 0 are grown in the northern United States/Southern Canada, MG I are grown in northern United States, etc., with maturity group increasing towards the southern part of the United States. Maturity group IX types would be grown in southern Mississippi. In essence, the larger the maturity group, the longer the growing season.

A) There is approximately a 12% difference in DNA content between cultivars. Is this difference expected within a species? Why or why not?

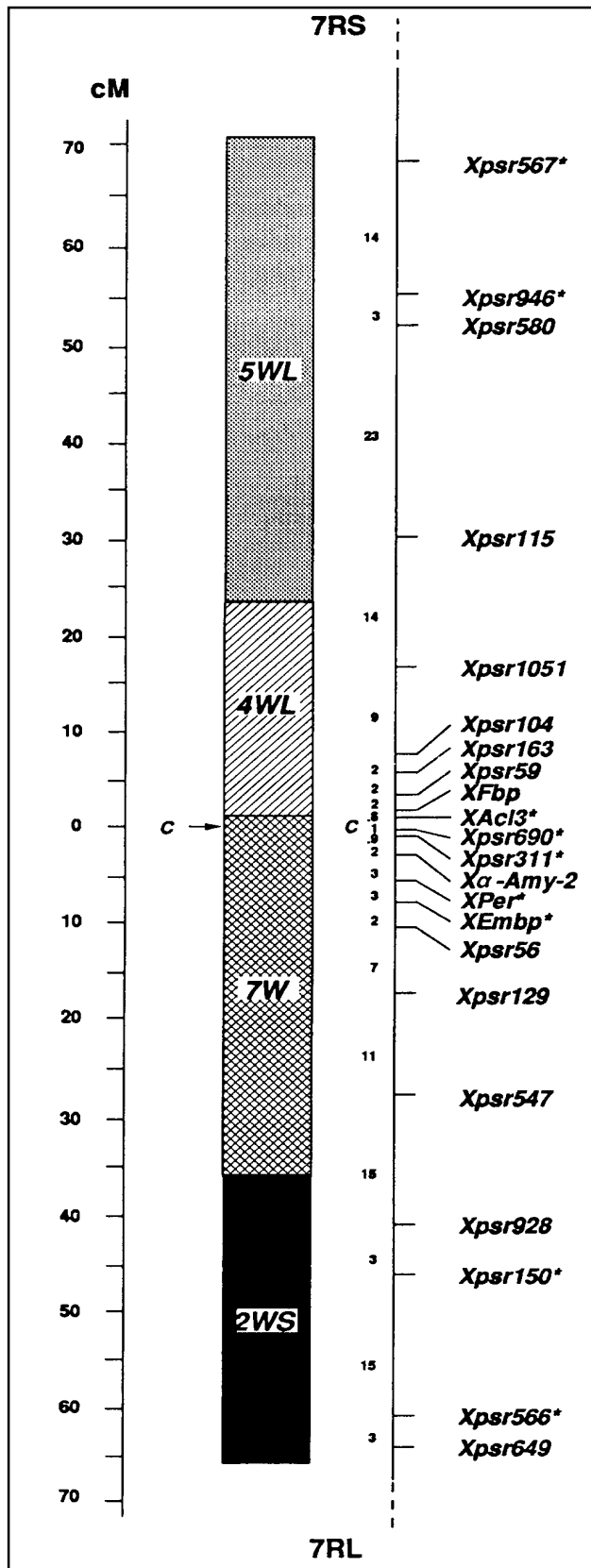
B) In this study, the authors obtained a correlation of  $r = 0.55^{**}$  between DNA content and maturity group. If no information had been provided on maturity group, what conclusions would you have

been able to derive about the region of the country to which a given cultivar was adapted, based on the DNA content? What is the basis for this conclusion?

C) Breeders have started to develop cultivars from inter-maturity group crosses. As this continues, would differences in DNA content between maturity groups be expected to increase, decrease, or stay the same? Why?

D) Describe (as opposed to simply naming) 2 mechanisms that can result in changes in DNA content.

4. The diagram below is from:



Rognli, O.A., K.M. Devos, C.N. Chinoy, R.L. Harcourt, M.D. Atkinson, and M.D. Gale. 1992. RFLP mapping of rye chromosome 7R reveals a highly translocated chromosome relative to wheat. Genome 35:1026-1031.

A) Explain why the authors can use a "W" to represent the wheat genome, as opposed to the conventional A, B, & D.

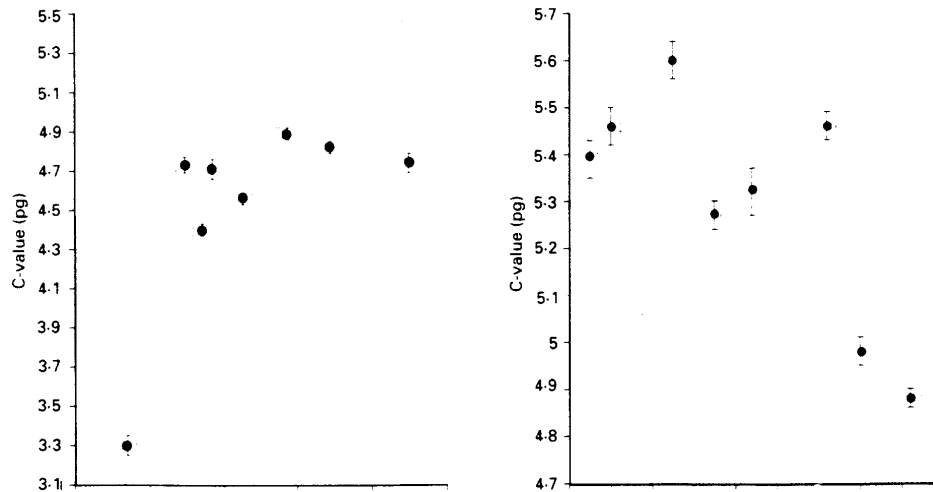
B) Given the composition of the 7R chromosome, what are the implications for its use to derive substitution lines of wheat?

C) What are the implications for chromosome-specific gene introgression (I.e., the use of any technique that relaxes pairing to get crossovers, and hence introgression, between homoeologues).

D) Given the cytogenetic stocks available for wheat and rye, what is the most probable technique used to map the centromere?

5. The following data are from:

Creber, H.M.C., M.S. Davies, D. Francis, and H.D. Walker. 1994. Variation in DNA C value in natural populations of *Dactylis glomerata* L. New Phytol. 128:555-561.



In this paper, the authors collected DNA samples of orchardgrass, at the sites depicted on the map above. In one of the graphs, they plotted DNA content vs the latitude at which the samples were collected. In the other graph, they plotted DNA content vs the altitude at which the samples were collected.

Which graph represents which? Explain the biological consideration you used to formulate your answer. Your answer will be graded on the soundness of the justification you give for your answer. Limit your answer to one short paragraph on the other side of this page.

6.

The following question is based on a March, 1990 article in *Discovery* magazine, titled "Spring fever," by G. Martin. (pp 70-74).

Vernal pools exist in the Sacramento Valley, north of San Francisco in California. These pools fill with water from winter rains, then dry quickly. They have existed for 50,000 years, allowing the evolution of unique plant and animal species. The article describes the plant species inhabiting the vernal pools as follows:

Their highly seasonal habitat has had some obvious effects on the pools' native plants. Most are minute, just an inch or so high, as result of the exigencies of a short growing season. Because they require copious amounts of water to live and reproduce and because water fills the pools completely only during the winter and early spring, the plants must sprout, bloom, and set their seeds in a fraction of the time used by their less-specialized cousins.

A) Make an educated guess as to how the number and size of chromosomes of plants in this habitat would have, as compared to those of their ancestral species. In the space provided, give the rationale for your answer.

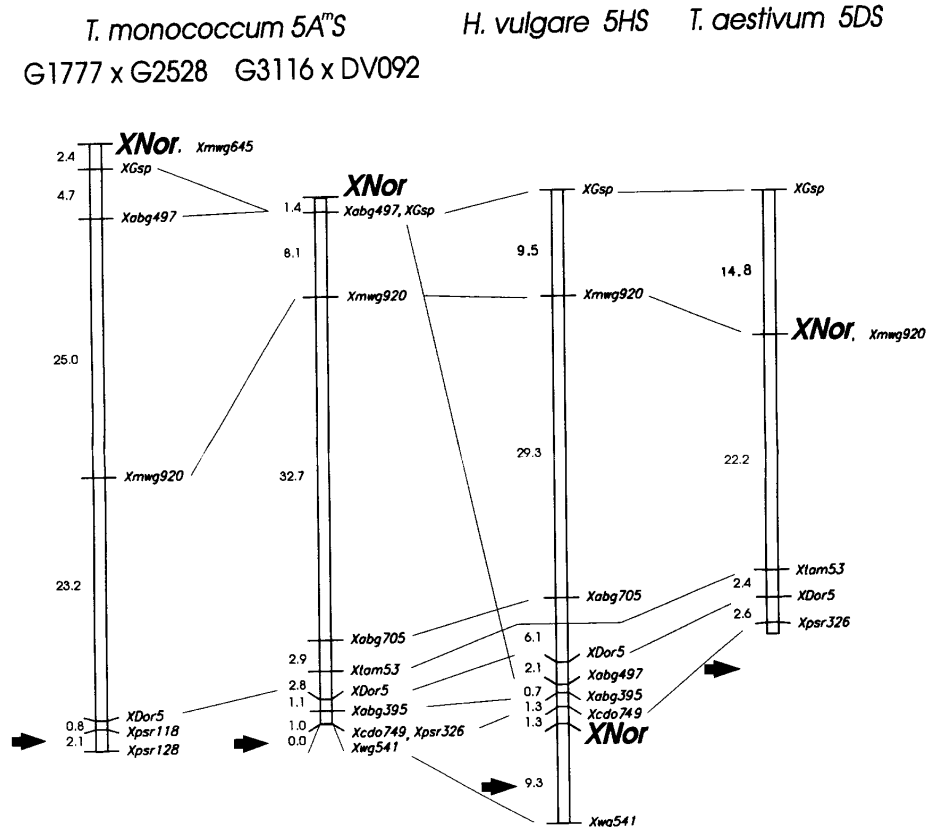
B) How would you check to see if your initial hypothesis is correct? Limit your answer to the space provided.



Greene's Orcutt grass

7. Dubcovsky, J. and J. Dvorák. 1995. Ribosomal RNA multigene loci: nomads of the Triticeae genomes. *Genetics* 140:1367-1377.

In the above paper, the authors looked at the position of the NOR region in different homoeologues of the wheat 5S chromosome, and found that while the sequential order of the genes has remained the same across the homoeologues, the location of the NOR had moved relative to other loci whose location has remained constant relative to each other as follows:



Describe and diagram one possible mechanism which could explain this.

8. The following is from:

Goldblatt, P and M. Takei. 1997. Chromosome cytology of Iridaceae – patterns of variation, determination of ancestral base numbers, and modes of karyotype change. *Ann. Missouri Bot. Gard.* 84:285-304.

In this paper, the authors study chromosomal patterns in various members of the iris family, including *Gladiolus*. *Gladiolus* belongs to subfamily Ixioideae, tribe Ixieae.

The base chromosome number for subfamily Ixioideae, and tribe Ixieae appears to be  $x = 10$ . However, the base chromosome number for *Gladiolus* is  $x = 15$ , although *Gladiolus* is a diploid. **For A-C, limit your answers to the space provided.**

A. Explain why a diploid like *Gladiolus* would have a higher  $x$  number than its ancestors.

Within the genus *Gladiolus*, there are small-flowered African species in which  $n = 14, 13, 12, 11$ . According to the authors, these “species appear to more specialized than those with the presumably ancestral  $x = 15$ .”

B. What is the term used to denote a set of descending chromosome numbers within a genus, such as that found within *Gladiolus*? How does this differ from aneuploidy?

C. What is the advantage to reduced chromosome number as a species specializes to adapt to a new habitat?

D. Diagram the process whereby the base chromosome number might be reduced. Use the back side of this page for your answer.

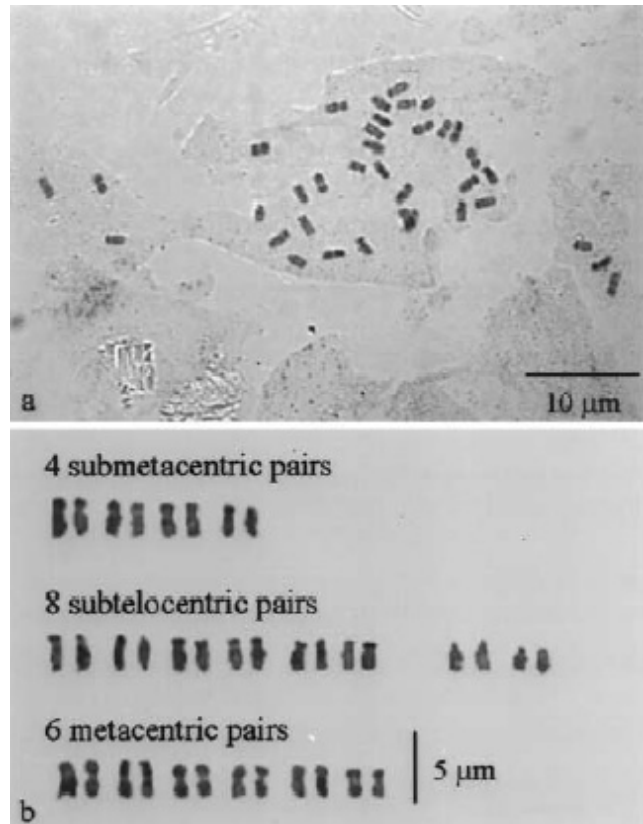


9. The following is from:

Cerbah, M., E. Mortreau, S. Brown, S. Siljak-Yakovlev, H. Bertrand, and C. Lambert. 2001. Genome size variation and species relationship in the genus *Hydrangea*. *Theor. Appl. Genet.* 103:45-51.

*Hydrangea* spp. are commonly used as ornamentals. These are considered diploids, but due to their high chromosome number ( $2n = 36$ ) are thought to be ancient polyploids.

A. Based on the karyotype at right, what inferences, if any, can be made as to whether *Hydrangea* originated as an autotetraploid, or an allotetraploid? Explain your answer.



B. DNA content was found to range from 1.95 pg/2C for *H. quercifolia* to 4.30 pg/2C for *H. macrophylla* subspecies *macrophylla*. Both species are  $2n = 36$ . Describe two mechanisms that would lead to such differences in DNA content.

C. *H. macrophylla* has another subspecies, *serrata*, the DNA content of which is 3.85 pg/2C. One of these subspecies grows at sea level, and the other grows in the mountains. Based on DNA content, which species would you predict to be the one that grows at sea level and which one grows up in the mountains? What is the reason for your answer? Use the back side of this page.

D. Whereas almost all species of *Hydrangea* had  $2n = 36$ , *H. aspera* was found to have  $2n = 34$  and *H. involucra* was found to have  $2n = 30$ . This is an example of what? Diagram a process that would lead to reduction in chromosome number. Use the back side of this page.

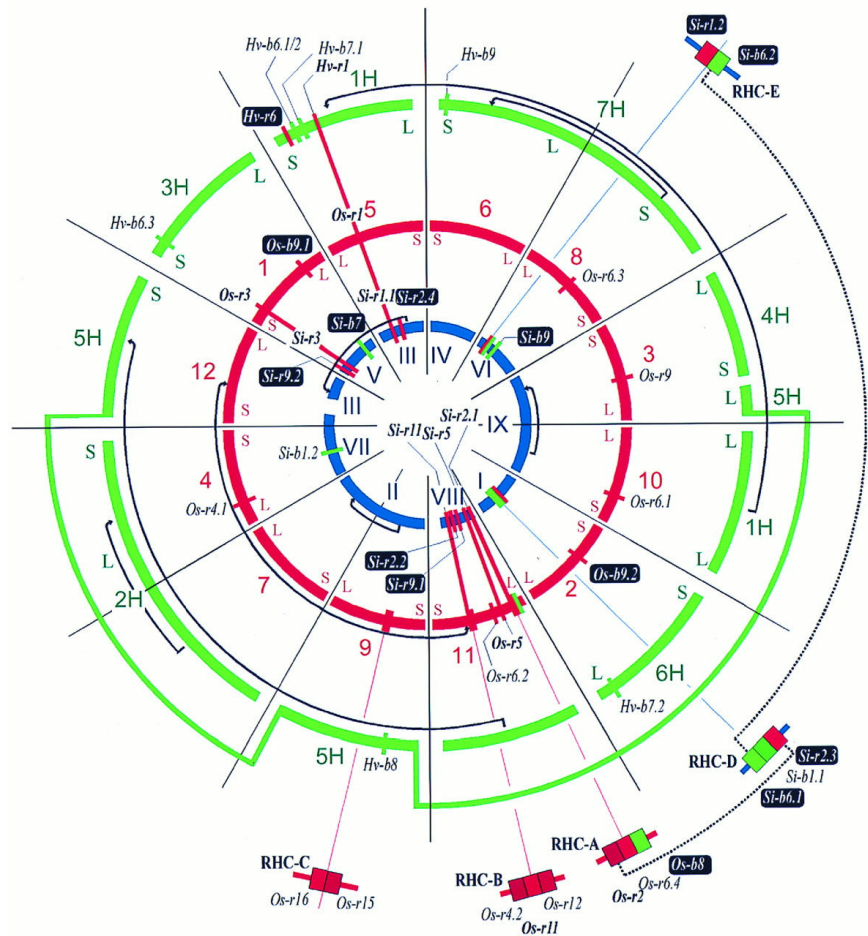


10. The following is from:

Leister D, J Kurth, DA Laurie, M Yano, T Sasaki, K Devos, A Graner, and P Schulze-Lefert. 1998. Rapid reorganization of resistance gene homologues in cereal genomes. *Proc. Natl. Acad. Sci. USA* 95:370-375.

In plants, genes which code for resistance against pathogens (R genes) are found in clusters of almost identical repeating units, called RHCs or R gene homologue clusters. In this paper, the authors examined the presence of RHCs in barley (GREEN), rice (RED), and foxtail millet (BLUE), arranged as circles so that homoeologous segments could be aligned with each other. In the diagram, the RHCs are shown in **black**. According to the authors, "Interspecific analyses of R-like genes frequently revealed nonsyntenic map locations between the cereal species rice, barley, and foxtail millet although tight collinear gene order is a hallmark of monocot genomes. Our data suggest a dramatic rearrangement of R gene loci between related species."

Explain the most likely explanation as to why RHCs are not in syntenic locations across these 3 different grass species. In other words, explain how the RHC's moved from one location to another.



11. The following question is from:

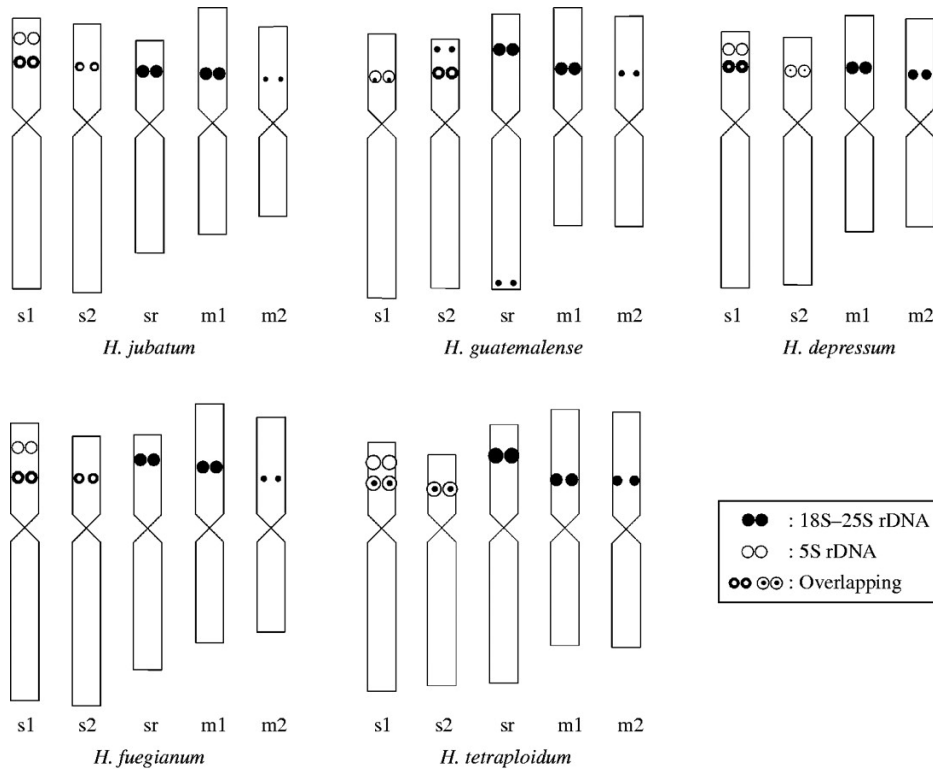
Vershinin, A.V., E.A. Salina, V.V. Solovyov, and L.L. Timofeyeva. 1990. Genomic organization, evolution, and structural peculiarities of highly repetitive DNA of *Hordeum vulgare*. *Genome* 33:441-449.

In this paper, the authors cloned 19 highly repeated sequences out of barley, then looked for these sequences in 6 different wild species closely related to barley. They also looked for them in wheat and rye. Only three of the 19 repeats were present uniquely in *Hordeum*. Another was present in the barley and the rye, but absent in the wheat. Given that barley, wheat, and rye all evolved from a common ancestor, how can you account for the presence of some repeated sequences in some genera, but not in others?

## 12. The following is from:

Taketa, S., H. Ando, K. Takeda, M. Ichii, and R. von Bothmer. 2005. Ancestry of American polyploid *Hordeum* species with I genome inferred from 5S and 18S-25S rDNA. *Ann. Bot.* 96:21-33.

Below are idiograms of the NOR-bearing chromosomes from 5 different allotetraploids of *Hordeum* (wild barleys):



Use the back of this page for your answers. Limit each answer to one sentence, + a diagram if need be.

A. Provide one possible mechanism that explains how the sr chromosome of *H. depressum* lost its NOR after polyploidization (the diagram only shows NOR-containing chromosomes, which is why the sr chromosome is not shown for *H. depressum*).

B. Provide one possible explanation or diagram as to why the NOR regions on chromosome m2 of *H. depressum* and *H. tetraploidum* are larger than the ones on the m2 chromosome of the other 3 species.

C. Explain or diagram the most likely way the s2 and sr chromosomes of *H. guatemalensis* acquired additional NOR sites?

### 13. The following is from:

Shan, F., G. Yan, and J.A. Plummer. 2003. Karyotype evolution in the genus *Boronia* (Rutaceae). Bot. J. Linn. Soc. 142-309-320.

The genus *Boronia* contains several ornamental species, and is from the same family as the genus *Citrus*. Chromosome numbers in *Boronia* include  $n = 7, 8, 9, 10, 11, 16, 18$ , and  $36$ .

The premise of this article is that the base chromosome number for this genus, based on the karyotypes determined by the authors, is  $x = 18$ , as opposed to  $x = 9$ , as others have argued.

They justify their conclusion thusly:

“Results in this study support the basic chromosome number of  $x = 18$ . In general, symmetrical karyotypes are regarded as more primitive and asymmetrical karyotypes as more specialized (Stebbins, 1971). The karyotype asymmetry is shown here to increase during the evolution of *Boronia* species where chromosome number descended.”

“Evidence was sought for the occurrence of polyploidization during *Boronia* evolution. If polyploidization had occurred, the species with  $x = 18$  would have twice the total chromosome length of species with  $x = 9$ . In addition, multivalents might be expected in species with  $x = 16$  or  $18$  if autopolyploids were produced. However, species with  $x = 18$  and  $9$  had similar total chromosome length and no multivalents were observed at metaphase I of meiosis in *Boronia* species with  $n = 18$  or  $16$  (Shan, Yan & Plummer, unpublished). Thus no evidence was found for polyploidization from  $x = 9$  to  $x = 18$  in *Boronia*.”



The final conclusion by the authors is that  $x = 18$ , and the  $x = 9$  species have been derived from the  $x = 18$  species, as opposed to  $x = 9$  and the species with  $18$  are really tetraploid.

A) Evaluate the soundness of this paper's conclusions, namely that basal chromosome number for this genus is really  $\times = 18$ , based on the 3 points (symmetry, total chromosome length, and lack of multivalents) raised by the authors.

**Limit your answer to no more than 6 sentences**

B) Based on these criteria and everything you learned in class, did the authors reach the right conclusion? Explain your answer.

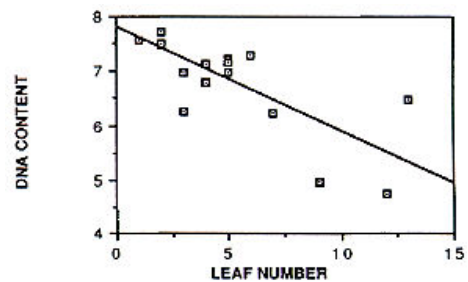
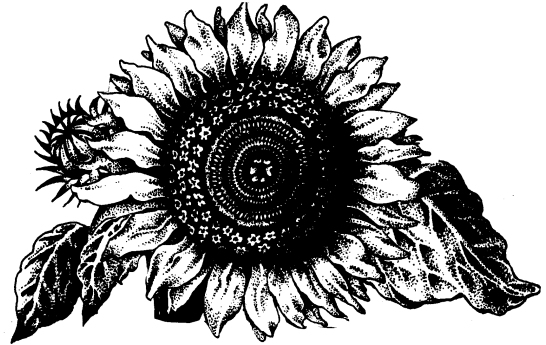
**Limit your answer to a brief paragraph.**

14. The following graph is from:

Michaelson, MJ, HJ Price, JS Johnston, and JR Ellison. 1991. Variation in nuclear DNA content in *Helianthus annuus* (Asteraceae). Amer. J. Bot. 78:1238-1243.

In this paper, the authors found that DNA content of sunflowers varied within and between genotypes. They also found \*intraplant\* variation. The younger leaves had less DNA than the older leaves. Leaf number in the graph refers to the stem node from the bottom. Chromosome counts eliminated aneuploidy as a cause for the discrepancy in DNA content.

Note that this does not necessarily imply loss of DNA in the younger leaves; it just as easily could be gain of DNA in the older leaves. Explain one mechanism that can explain the DNA gain and one that can explain loss as shown in the graph.

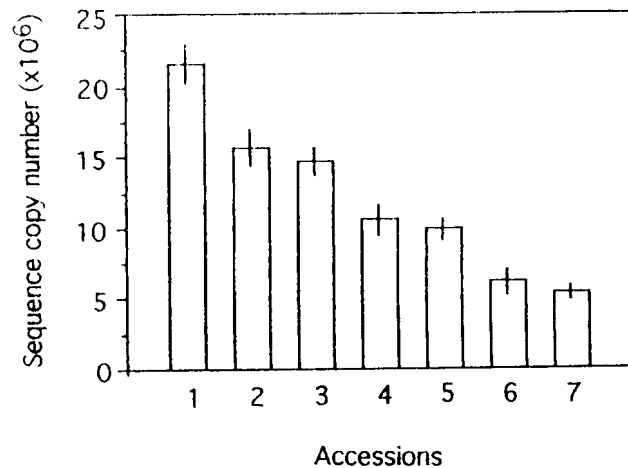


**15. In the following paper:**

Maggini, F., R. D'Ovidio, M.T. Gelati, M. Frediani, R. Cremonini, M. Ceccarelli, S. Minelli, and P.G. Cionini. 1995. *FokI* DNA repeats in the genome of *Vicia faba*: species specificity, structure, redundancy modulation, and nuclear organization. *Genome* 38:1255-1261.

The authors looked at the numbers of a given DNA repeat (*FokI*) in different accessions of faba bean, and obtained the following results:

Accession	Origin
1	Morocco
2	Italy
3	Algeria
4	Greece
5	Egypt
6	Italy
7	Greece



Notice that there was great variability of in the copy number of the repeats per haploid (1C) genome among the accessions. Explain or diagram 2 possible mechanisms which might account for this difference in *FokI* copy number.

## 16. The following is from:

Sorrells, M. E. et al., 2003. *Comparative DNA sequence analysis of wheat and rice genomes. Genome Res. 13:1818-1827.*

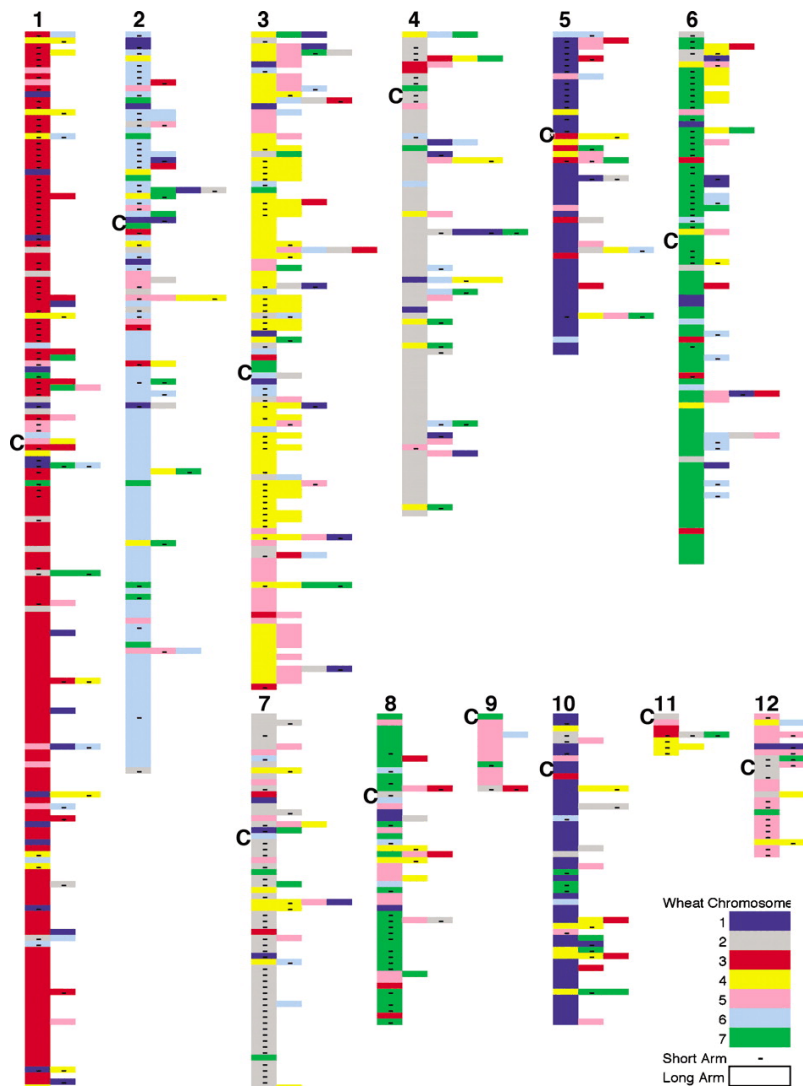
This diagram shows the 12 chromosomes of rice. The main bar under each chromosome has been color-coded to indicate its correspondence with the orthologous wheat chromosomes, numbered 1 through 7.

A) First thing to notice is that  $\times = 7$  for wheat, and  $\times = 12$  for rice. Since most people believe the ancestral number for  $\times$  was 7 or 8 or 9, and since the last whole-genome duplication event took place before the divergence of wheat and rice, it is necessary to explain how rice could have ended up with  $\times = 12$ . **List** the 3 mechanisms covered in class that lead to an increase in base chromosome number (i.e., value of  $\times$ ).

B) Draw a diagram of **one** of these mechanisms to explain how it works.

C). Suppose you are a wheat breeder, in general, how could you use the information available for a particular disease resistance gene in rice to identify the same disease resistance gene in wheat?

D) What do you think is the purpose of repetitive DNA? (I am just curious to know what you think, so there is no right or wrong answer): ☺



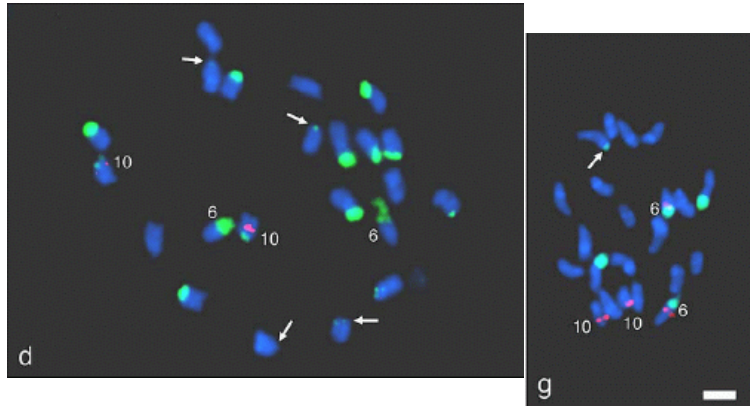


**17. The following is from:**

Pedrosa-Harand A, CC Sonza de Almeida, M Mosiolek, MW Blair, D Schweizer, and M Guerra. 2006. Extensive ribosomal amplification during Andean common bean (*Phaseolus vulgaris* L.) evolution. Theor Appl Genet 112:924-933.

In this case, the authors noted that the number of 45S ribosomal DNA sites varied between 3 and 8 on chromosomes 6 and 10, depending on genotype.

The photos illustrate 2 examples.



Chromosomes of cv 'Nuña Mani'. Green indicates hybridization to the 45S rDNA probe. Arrows indicate signals too faint to see well in the photo.

Describe or diagram one phenomenon that could possibly explain the variability in 45S rDNA loci, ie, how would the 45S rDNA move around the genome and change in location number?

**18. The following is from:**

Garnatje T, S Garcia, R Vilatersana and J Vallès. 2006. Genome size variation in the genus *Carthamus* (Asteraceae, Carduaceae): systematic implications and additive changes during allopolyploidization. *Ann Bot* 97: 461-467.

*Carthamus* is a genus native to the Mediterranean, and is divided into two sections, *Carthamus*, which are all diploid, and includes the commercially important safflower. Section *Atractylis* contains diploids and their allopolyploids. Here, the authors measured C values in the various species in *Atractylis*, and came up with the following averages for:

- $2x \text{ species} = 1C = 1.42$ ;  $4x \text{ species} = 1C = 1.19$ ;  $6x \text{ species} = 1C = 1.20$

Though the 4 and 6x species have more total DNA due to their higher C number (ie, 4 and 6C), 1C values (in pg) for the diploid progenitors were higher than the 1C values for their allopolyploid derivatives.

Briefly explain why there might be lower C values in the derived allopolyploids compared to their ancestral genomes.

**19. The following is from:**



Naganowska B, B Wolko, E Śliwińska, Z Kaczmarek, and MT Schifino-Wittman. 2006. 2C DNA variation and relationships among New World species of the genus *Lupinus* (Fabaceae). *Pl Syst Evol* 256:147-157.

DNA contents for North American species are given in the table below. Why does there appear to be a correlation between DNA content and the growth habit of these species?

*L. polyphyllus*.

[http://tr.wikipedia.org/wiki/Resim:Lupinus\\_polyphyllus3.JPG](http://tr.wikipedia.org/wiki/Resim:Lupinus_polyphyllus3.JPG)

Species	Life cycle	2C DNA (pg)	Species	Life cycle	2C DNA (pg)
<i>albicaulis</i>	Perennial	2.68	<i>sericeus</i>	P	1.44
<i>texensis</i>	Annual	2.36	<i>leucophyllus</i>	P	1.42
<i>mexicanus</i>	P	2.00	<i>garfieldensis</i>	?	1.41
<i>rivularis</i>	P	1.76	<i>argenteus</i>	P	1.39
<i>latifolius</i>	P	1.74	<i>arbustus</i>	P	1.39
<i>arboreus</i>	P	1.74	<i>arizonicus</i>	A	1.27
<i>polyphyllus</i>	P	1.70	<i>luteolus</i>	?	1.17
<i>lepidus</i>	P	1.69	<i>microcarpus</i>	A	1.14
<i>verricolor</i>	P	1.67	<i>concinus</i>	A	1.13
<i>albifrons</i>	P	1.48	<i>nanus</i>	A	1.09

**20. The following is from:**

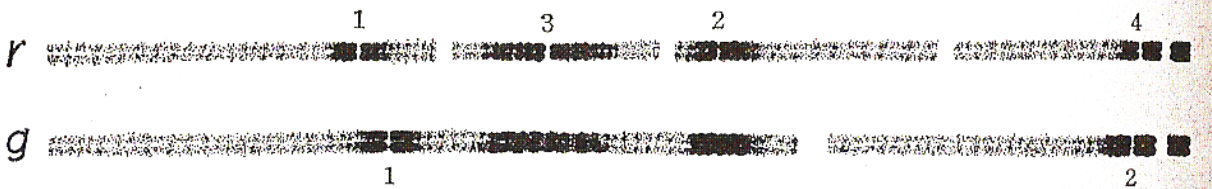
Tanaka, R. 1967. A comparative karyotype analysis in *Haploppapus gracilis* ( $2n = 4$ ) and *H. ravenii* ( $2n = 8$ ). *Cytologia* 32:542.

This is a composite flower native to the western US. A photo is at right. This species is  $x = 2$ , and evolved from *H. ravenii* ( $x = 4$ ). In this paper, karyotypes from both species are aligned below. Each chromosome is numbered above its centromere:



Photo credit:

<http://wc.pima.edu/~bfiero/tucsonecol>



A) It is evident that chromosome 2 of *gracilis* is made up from parts of chromosomes 2 and 4 of *ravenii*. What type of chromosomal reconfiguration does this represent?

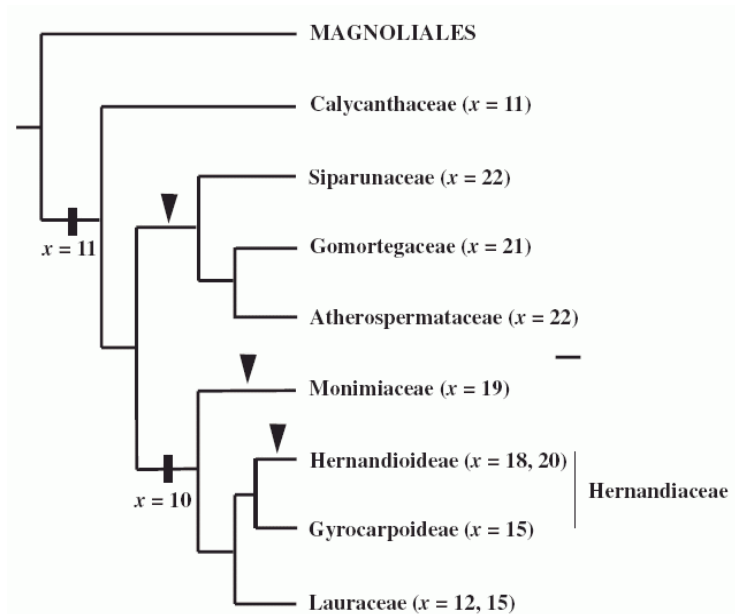
B) It is likewise evident that chromosome 1 of *gracilis* contains chromosomes 1 and 3, and part of chromosome 2 of *ravenii*. To what extent, if any, can this be explained by Darlington's progressive reduction model? Explain your answer.

C) What is the advantage or disadvantage to having fewer chromosomes?

21. *The following is from:*

Oginuma K and H Tobe. 2006.  
Chromosome evolution in the Laurales  
based on analyses of original and  
published data. J. Plant Res 119:309-  
320.

The Laurales are an order within the  
magnolias, an ancient dicot group that  
predates the eudicot-monocot split.  
This order has 7 families and over 90  
genera in it. Among the better-known  
products are things like laurel,  
avocado and cinnamon, along with  
camphor and sassafras. The  
phylogenetic tree at right illustrates the  
chromosome relationships within the  
order. Arrowheads denote  
polyploidization events.



A) Name the phenomenon that led from a base number of  $x=11$  to  $x=10$ , and diagram how it is thought to happen.

B) Which other families show this phenomenon?

C) How does one get from  $x = 10$  to  $x = 12$ ?

**22. The following was inspired by:**

Picault et al. 2009. Identification of an active LTR retrotransposon in rice. *Plant J.* 58:: 754-765.

In this study, the authors used micro array technology to detect gene expression from retrotransposons, and thus identified a new type called *Lullaby*. This LTR retrotransposon is transposing in ‘Nipponbare’<sup>1</sup>, a traditional Japanese variety and parent to many modern rice cultivars.



‘Nipponbare’ rice

A) Transposable elements are classified into two classes. What class is this type of transposable elements did they study in this case?

B) What is the defining characteristic of this class of transposable elements?

C) In most cases these LTRs are not active due to silencing, or because they no longer have functional copies of the genes needed for transposition. However, when they are active, they can facilitate genetic changes. Give one example of how they may do so:

D) “Gene expression is subject to a regulatory network of a complexity that it only just being realised. ... The often forcible insertion of DNA into a tightly controlled genetic regulatory network is likely to produce unintended effects.”

<http://www.greenpeace.org/raw/content/international/press/reports/50-years-since-the-double-heli.pdf>

<http://www.greenpeace.org/raw/content/international/press/reports/illustrating-the-problems-of-g.pdf>

The above sentences are used by Greenpeace to highlight the reasons why GE crops are dangerous. Use the information from Picault et al to support or refute the Greenpeace argument.

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<sup>1</sup>[http://www.shigen.nig.ac.jp/rice/rgn/vol22/image/22\\_13\\_1.jpg](http://www.shigen.nig.ac.jp/rice/rgn/vol22/image/22_13_1.jpg)

**23. The following is from:**

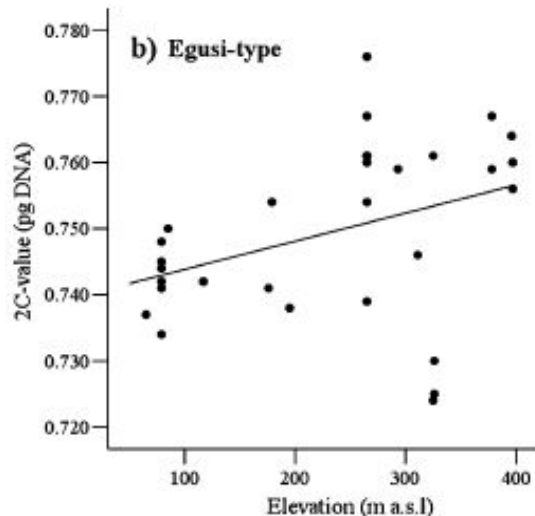
Achigan-Dako EG, J Fuchs, A Ahanchede and FR Blattner. 2008. Flow cytometric analysis in *Lagenaria siceraria* (Cucurbitaceae) indicates correlation of genome size with usage types and growing elevation. Plant Syst Evol. 276-9-19.

This is an annual species also known as bottle gourd or calabash. It is native to the tropical parts of Africa, and is grown for its edible seed.

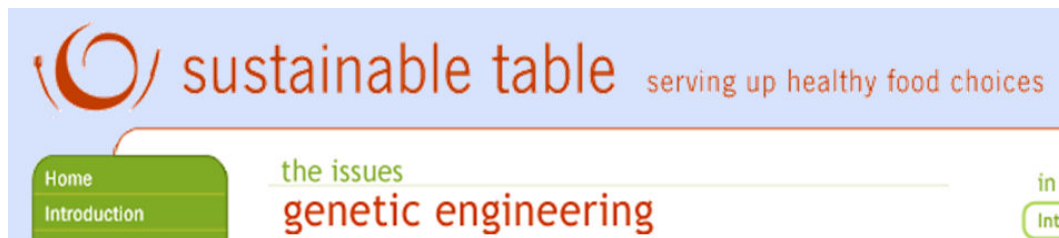
The authors studied genome size in 366 individuals from 117 accessions of bottle gourds, and found large differences, of up to 12%, in 2c values. DNA varied by ecotype and by elevation planted, as seen in the graph below.

**Extra credit.** The examples from class tended to have lower DNA amounts at higher altitudes. The results here are the opposite.

What advantage would there be to having high 2C amounts at higher elevations? Are there other possible explanations? Note that the elevation gradient ran south-north in this case. Discuss your answer.



24. The following question is from <http://www.sustainabletable.org/issues/ge>



In their arguments as to why we need to desist from genetic engineering, this web site says, “Scientists do not know if the forced insertion of one gene into another gene could destabilize the entire organism, and encourage mutations and abnormalities.”

Using your newly acquired knowledge and understanding of the plant genome, either support or refute the above argument. Limit your self to no more than 2 well constructed, well thought out points.



Correlation between stem growth rate and DNA content. These parameters were not significantly correlated in native (non invasive) genotypes, but very negatively correlated with invasive genotypes.

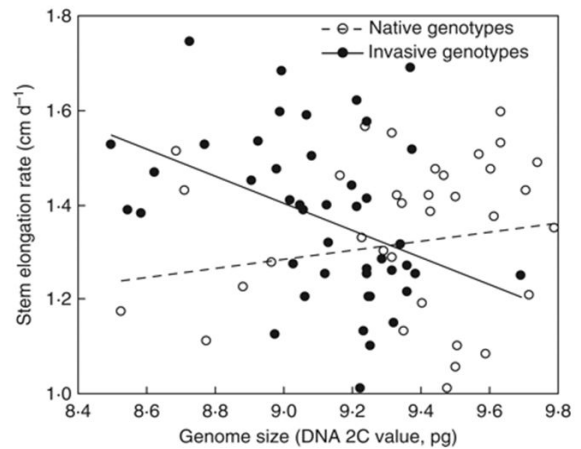
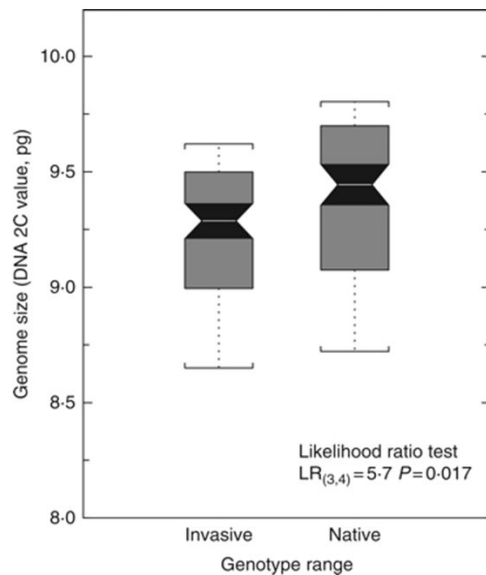
## PBGG 8990, Problem set VII

## Evolution of the Karyotype, Page 25

### 25. The following is from:

Lavergne S, NJ Muenke and J Molfasky. 2010. Genome size reduction can trigger rapid phenotypic evolution in invasive plants. *Annals of Botany* 105: 109-116.

In this paper, the authors were working with reed canarygrass (*Phalaris arundinacea*). Genotypes vary in invasiveness, with the more invasive genotypes having on average less DNA and a faster growth rate.



Are these results believable? Explain why or why not.

Average DNA content of invasive and non invasive genotypes.

## 26. The following is from:

Liu, Jin, Rouse, Friebe, Gill and Pumphrey.  
2011. Development and characterization of wheat- *Ae. searsii* Robertsonian translocation and <....> conferring resistance to stem rust. Theoretical and Applied Genetics 122:1537-1545.

## A 'time bomb' for world wheat crop

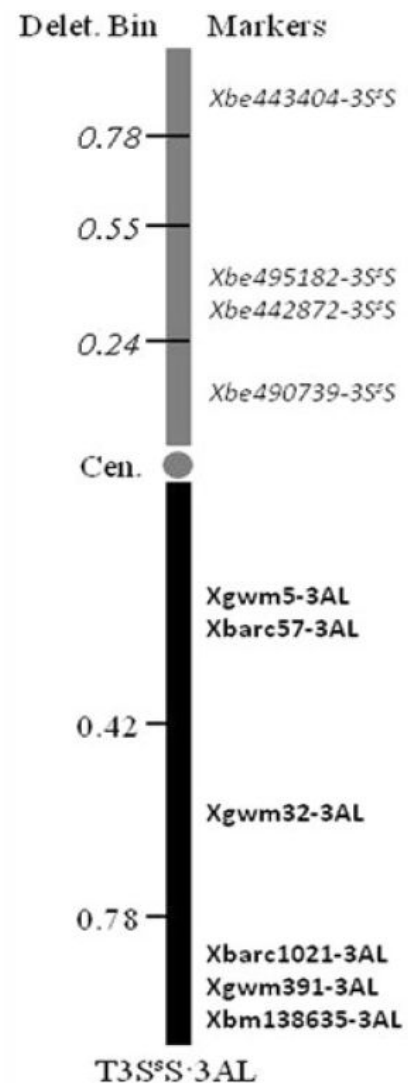
*The Ug99 fungus, called stem rust, could wipe out more than 80% of the world's wheat crops as it spreads from Africa, scientists fear. The race is on to breed resistant plants before it reaches the U.S.*

June 14, 2009 | Karen Kaplan

In this paper, the authors screened alien addition lines of wheat, and found those containing the 3S chromosome from *Aegilops searsii* ( $2n = 2x = 14$ ) were resistant to the Ug99 strain.

They next made double monosomic lines (3A/3S, 3B/3S, and 3D/3S). The F<sub>2</sub> were screened for stem rust resistance. Resistant lines were screened with molecular markers for the short and long arms of 3S, and plants positive for the 3SL marker were discarded. A result is shown at left.

A) Diagram or explain how a monosomic line is made



B) Diagram the stages whereby they would go from a double monosomic to a Robertsonian fusion

Extra Credit) Describe a procedure you could use to reduce linkage drag from the 3SS arm.

**27. The following is from:**

Shen, Y-G, Z-L Wang and K-Y Guan. 2007. Karyotypical studies on thirteen *Iris* plants from China. Acta Phytotaxonomica Sinica 45:601-618.

Karyotypes for:

*I. dolichosiphon* ( $2n = 11$ )

*I. collettii* ( $2n = 14$ )

*I. laevigata* ( $2n = 16$ )

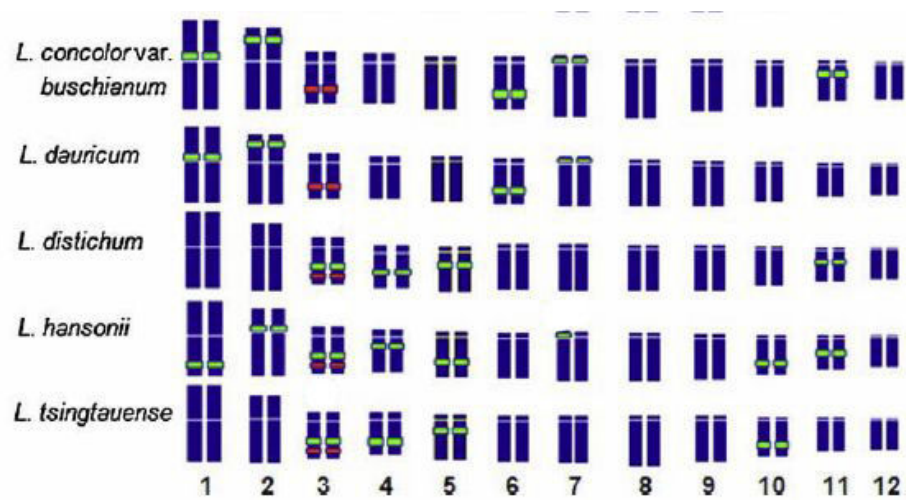
*I. wattii* ( $2n = 20$ )



A) Is different chromosome number expected within a genus? Why or why not?

## 28. The following is from:

Sultana et al, 2010.  
Mapping of rRNA gene  
loci and inter-specific  
relationships in wild  
*Lilium* distributed in  
Korea. J. Plant Biol.  
53:433-443.



In the graphic, red is the NOR, and green is the 5S RNA rDNA, which is also made of tandem repeats. It is evident that the 5S rDNA has been deleted from some chromosomes and moved to others.

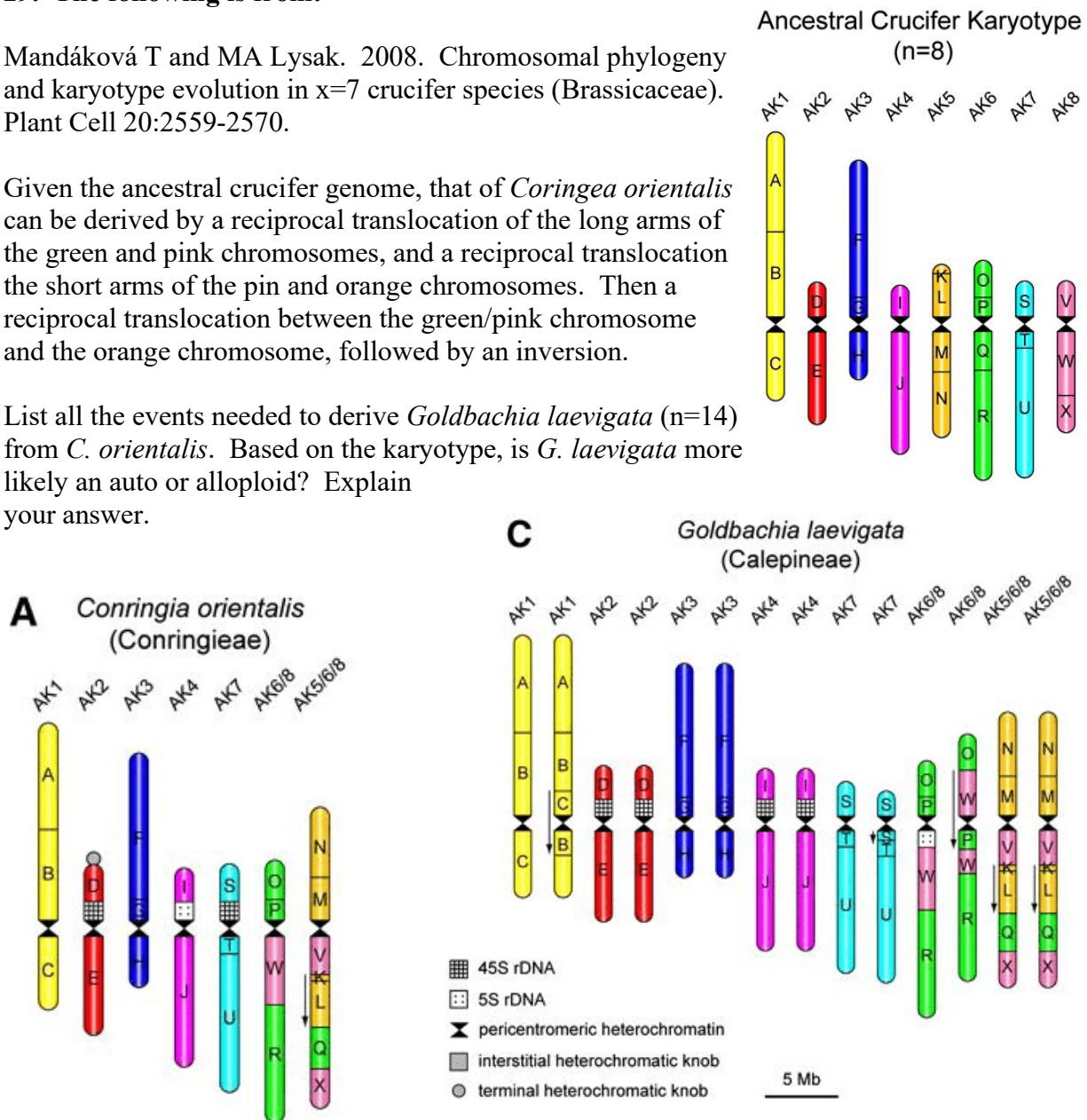
Three phenomena were discussed in class that can move loci from their original positions to nonsyntenic locations. List these three, and then specify which is the more likely to have moved the 5S rDNA to new locations. Don't forget to explain your answer.

**29. The following is from:**

Mandáková T and MA Lysak. 2008. Chromosomal phylogeny and karyotype evolution in  $x=7$  crucifer species (Brassicaceae). Plant Cell 20:2559-2570.

Given the ancestral crucifer genome, that of *Coringea orientalis* can be derived by a reciprocal translocation of the long arms of the green and pink chromosomes, and a reciprocal translocation the short arms of the pin and orange chromosomes. Then a reciprocal translocation between the green/pink chromosome and the orange chromosome, followed by an inversion.

List all the events needed to derive *Goldbachia laevigata* ( $n=14$ ) from *C. orientalis*. Based on the karyotype, is *G. laevigata* more likely an auto or allopolyploid? Explain your answer.



**30. It has been said that:**

*"To create a little flower is the labour of ages..."* - William Blake (1757-1827).

**Others have since elaborated:**

*"Unless there is polyploidization"* - Hans Winkler (1877 - 1945)

*"From interspecific hybridization via  $2n$  gametes"* Cyril Dean Darlington (1903-1981)

*"Combined with genetic control of chromosome pairing"* Ernie Sears (1910–1991)

Based on the latest findings from cytogenetics, in your learned opinion, what should the latest cytogenetic development in our understanding of plant genome evolution be?

**31. In the following paper,**

Liu R, C Vitte, J Ma, AA Mahama, T Dhliwayo, M Lee and JL Bennetzen. 2007. A GeneTrek analysis of the maize genome. PNAS 104:11844-11849,

The following statement is made: "Two major bursts of LTR retrotransposon activity in the last 2 million years are responsible for the large size of the maize genome, but only the more recent of these is well represented in gene-containing BACs, suggesting that LTR retrotransposons are more efficiently removed in these domains."

A) Explain the main way whereby LTR retrotransposons can increase the size of a genome. Limit answer to 2 sentences

B) Diagram a model that explains how the region between the LTRs of retro transposon might be removed from a genome.

C) List 1 other mechanism whereby a genome can shrink. What is the possible role of LTRs in this?

32. The following is from:

Alves et al., 2011. Chromosome characterization and variability in some Iridaceae from Northeastern Brazil. *Genetics and Molecular Biology* 34: 259-267.

The karyotypes are from 5 species (labeled a through e) from the tribe Tigridene.

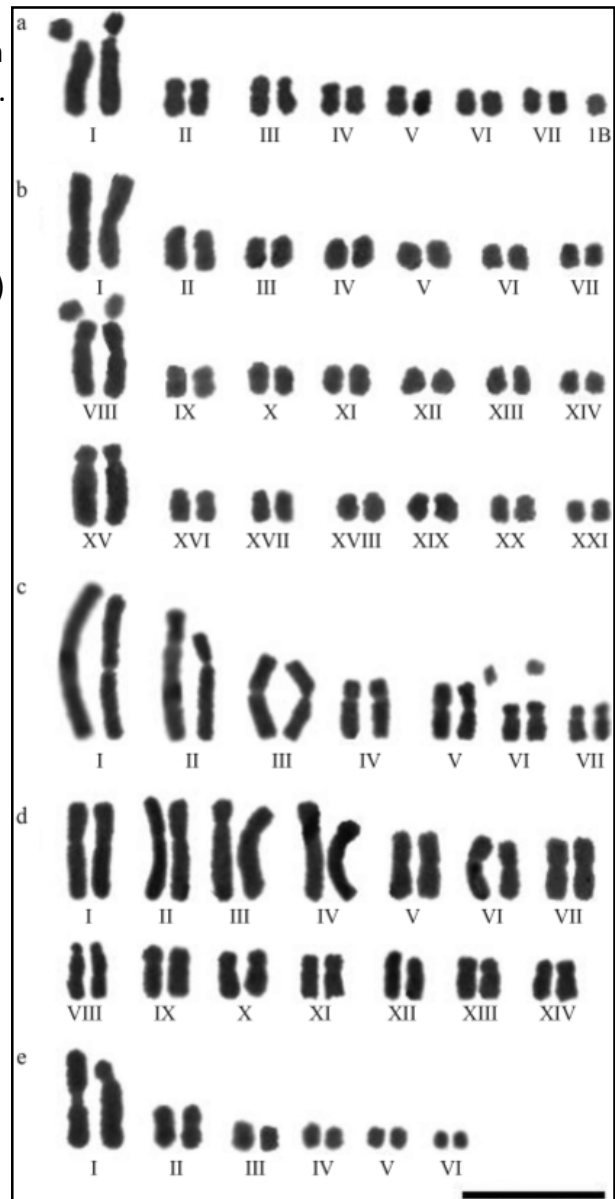
Note they find that species a) (*Alophia drummondii*) has a B chromosome, and thus is  $2n = 2x + 1 = 15$ .

A) How do you know that species a) is really not a triploid,  $2n = 3x = 15$ ?

B) How do you know that species a) is not a trisomic?

C) How do you know that it is not a monosomic? (ie, the normal number is  $2n = 2x = 16$ )

D) Specify **two** additional lines of evidence you would want to see before deciding if the species a) has or does not have a B chromosome.



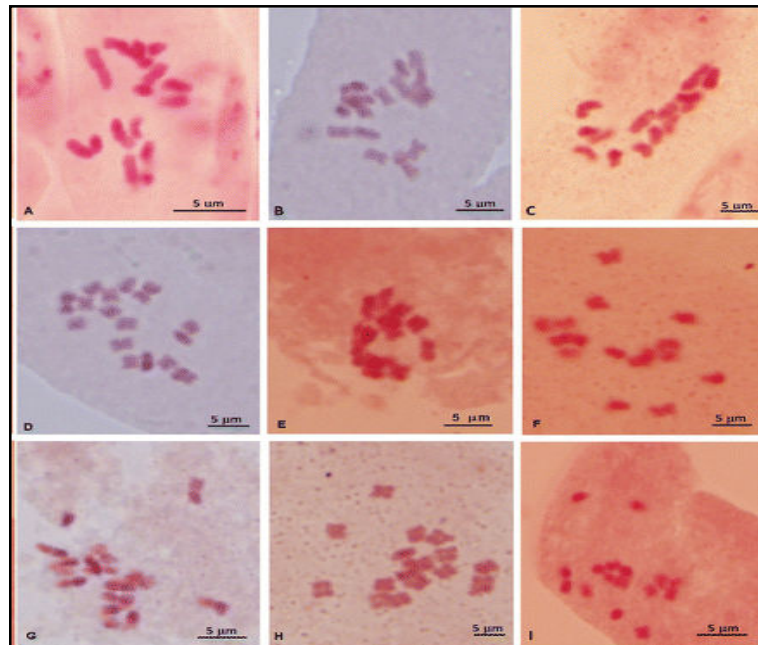


**33. The following is from:**

Uslu. 2012. Karyology of nine *Trifolium* L. taxa from Turkey. Caryologia 65:304-310.

Notice that chromosome numbers for these clover species range from  $2n = 12$  to  $2n = 14$  to  $2n = 16$ .

A) Name the phenomenon illustrated by the different  $n$  numbers.



B) Diagram 1 method by which a  $2n = 14$  species could give rise to a  $2n = 12$  species and another method by which it could give rise to a  $2n = 16$  species.

34. The following is from:

**ALLIANCE FOR BIO-INTEGRITY**

*Preserving the Safety of Our Food, the Health of Our Environment,  
and the Harmony of Our Relationship with Nature*

<http://www.biointegrity.org/Overview.htm>, accessed 20 April 2017

According to them, genetic engineering has "unprecedented risks" because:

"Traditional breeding is based on sexual reproduction between like organisms. The transferred genes are similar to genes in the cell they join. They are conveyed in complete groups and in a fixed sequence that harmonizes with the sequence of genes in the partner cell. In contrast, bioengineers isolate a gene from one type of organism and splice it haphazardly into the DNA of a dissimilar species, disrupting its natural sequence."

Now that you have taken plant cytogenetics, list 3 properties of plant genomes that either support or refute the Alliance statement about unprecedented risks. For each property listed, explain why or why not it supports the statement.

**35. The following is from:** Cao, Vu, Wang, Appenroth, Messing, and Schubert. 2015. The map-based genome sequence of *Spirodela polyrhiza* aligned with its chromosomes, a reference for karyotype evolution. *New Phytologist*. 209:354-363.

According to the authors, "Duckweeds are aquatic monocotyledonous plants of potential economic interest with fast vegetative propagation, comprising 37 species with variable genome sizes (0.158-1.88 Gbp)"

Using synteny analysis researchers determined that duckweed had  $x = 7$  ancestral chromosomes (present day has  $2n = 2x = 20$ ). Analyses have determined that historically, duckweed has experience two rounds of polyploidization.

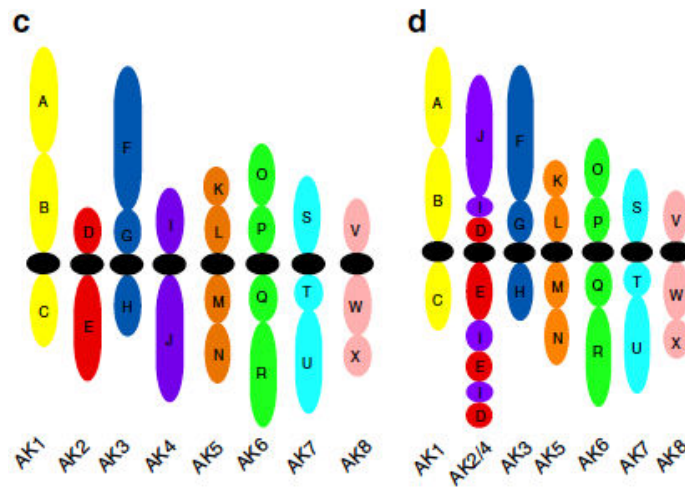


- How many present day chromosomes would we expect given that the ancestral karyotype experienced 'two successive rounds of WGD'?  $2n = \underline{\hspace{2cm}}$
- Name the phenomenon that explains how duckweed has  $2n = 40$  chromosomes as opposed to the  $2n = \underline{\hspace{2cm}}$  answer identified in part a.
- Extra Credit: Describe and/or diagram two mechanisms that contribute to the difference in haploid chromosome number between the ancestral and present day karyotypes.

1 [http://www.illinoiswildflowers.info/wetland/plants/gr\\_duckweed.html](http://www.illinoiswildflowers.info/wetland/plants/gr_duckweed.html) and <https://www.google.com/url?sa=i&rct=j&q=&esrc=s&source=images&cd=&cad=rja&uact=8&ved=0ahUKEwjpoIus0crTAhUFuhQKHsAdYQjRwIBw&url=https%3A%2F%2Fwww.pinterest.com%2Ftgallatin%2Fgoatsli>

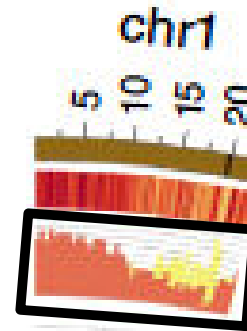
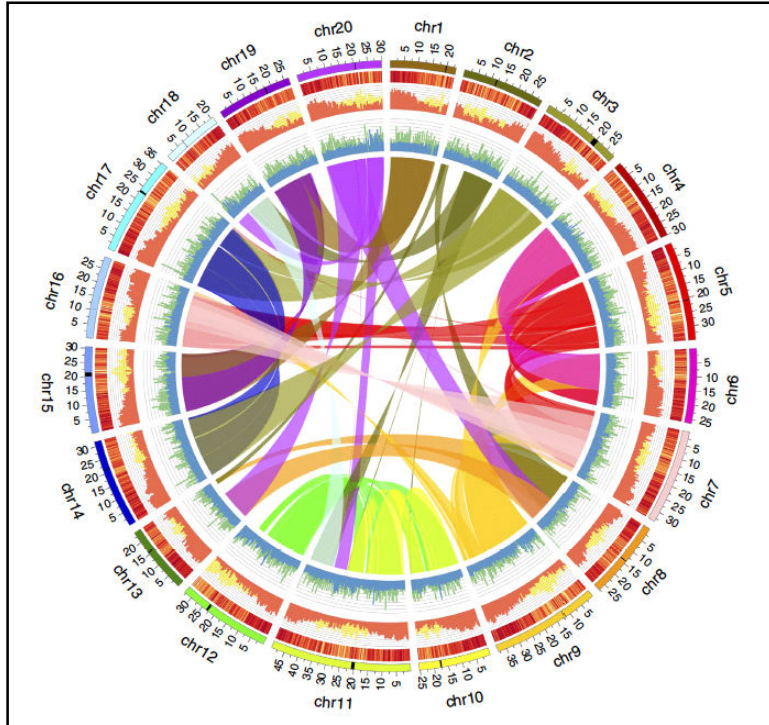
**36. The following is from:** Kagale, Koh, Nixon, et al. 2014. The emerging biofuel crop *Camelina sativa* retains a highly undifferentiated hexaploid genome structure. Nature Communications 5: Article 3706.

The sequencing and assembly of the *Camelina sativa* genome revealed chromosomal rearrangements when the ancestral crucifer karyotype (Fig. C) was compared to the ancestral *C. sativa* karyotype (Fig. D).



Sequentially name each structural rearrangement that needs to occur to create AK2/4 in figure D from chromosomes AK2 and AK4 in figure C. Drawing out a diagram might help envision the necessary steps.

Based on their sequencing data, the authors constructed a plot to highlight key features of chromosomes. Without having access to the figure legend, what do you think the orange and yellow bars stand for in third track that is highlighted by a black rectangle in the zoomed in version of chromosome 1. (Note the purple star signifies the putative location of the centromere.)



Based on the circos plot, the current karyotype for *C. sativa* is  $2n=$ \_\_\_\_ $X=$ \_\_\_\_\_

Based on Figure D above, the *ancestral* karyotype for *C. sativa* is  $2n=$ \_\_\_\_ $X=$ \_\_\_\_\_

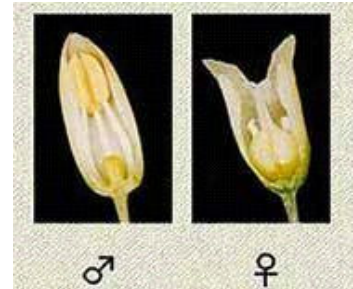
What feature of the circos plot indicates that WGDs took place in the past?

The increase in  $1x$  from a low number to a high number is called:

Briefly describe and/or diagram 3 ways through which  $1x$  can go from a low number to high number, while remaining  $1x$ . Use the back of this page.

**37. The following is inspired by :** Harkess et al., 2016. Retrotransposon proliferation coincident with the evolution of dioecy in *Asparagus* G3 6(9): 2679-2685.

The authors looked the relative abundance of transposons in the genomes of both hermaphroditic and dioecious species of *Asparagus*. They concluded that transposons, and not whole genome duplication, was responsible for genome size variation in this group.



- A. An LTR element was deemed responsible for an increase in genome size. What does LTR stand for?
- B. How do LTR elements increase genome size?
- C. How could an LTR element decrease genome size?
- D. Diagram out the mechanism mentioned in your answer to question C?

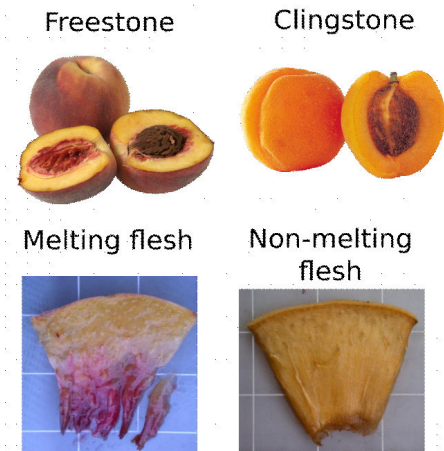
*Asparagus* flowers, a dioecious species. <http://squarefoot.creatingforum.com/t14386p180-asparagus>

**38. The following is from:** Gu et al., 2016. Copy number variation of a gene cluster encoding endopolygalacturonase mediates flesh texture and stone adhesion in peach. *Journal of Experimental Botany* 67(6): 1993-2005

Peaches are classified as 'freestone' or 'clingstone' depending on how tightly the seed is attached to the flesh. In turn, the flesh is classified as 'melting' or 'non-melting'.

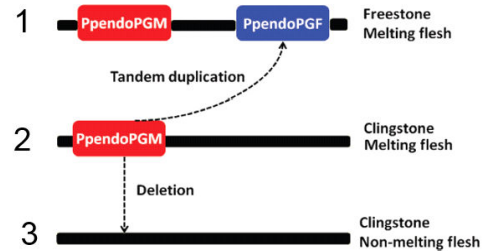
Researchers identified a particular genomic region in peach that controls both flesh texture and stone adhesion through the action of an endopolygalacturonase, ie, an enzyme that helps breakdown cell walls.

They conclude that three haplotypes exist in peach germplasm that correspond to the below figure.



A. Sequence based analysis lead the team to believe that having a single copy of this locus was the ancestral state of this genomic region. Name the mechanism that can account for the creation of a tandem duplicate.

B. Diagram out how the above mechanism can create a tandem duplicate.



C. One could imagine over time that haplotype 1 (above) could result in a change in copy number. What are two possible mechanisms that could alter the copy number from two copies to one copy or from two copies to three copies?

<https://www.rosbreed.org/sites/default/files/peach%20texture.png>



**39. The following is from:**

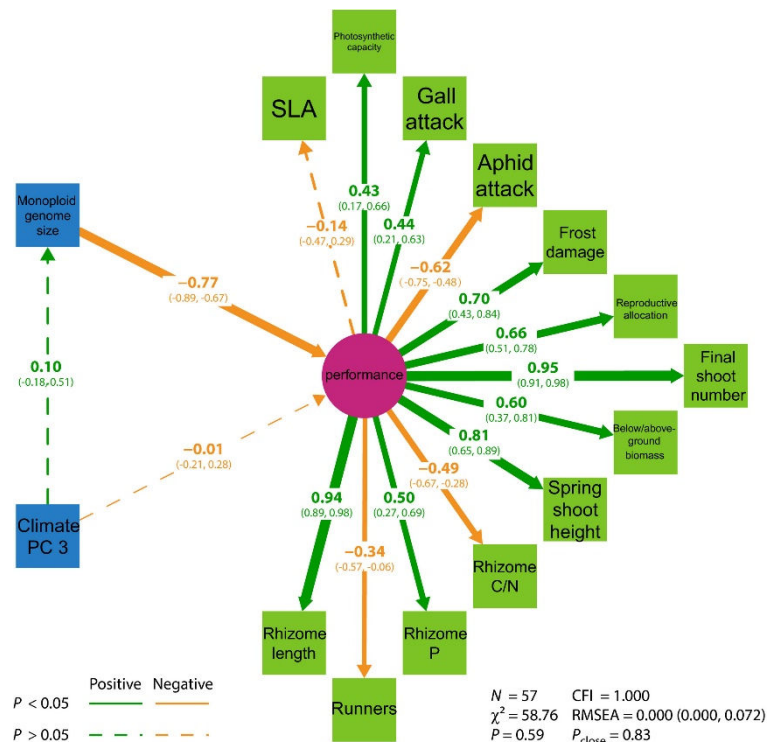
Pysek et al. 2018. -----native and invasive populations in an ecologically important cosmopolitan grass. Ecology 99(1): 79-90.

In this study, the authors looked at various factors that could distinguish between invasive and non-invasive populations of common reed, *Phragmites australis*.



[www.invasivespeciesinfo.gov/profile/common-reed](http://www.invasivespeciesinfo.gov/profile/common-reed)

- A) The figure generally shows that genome size (blue) is correlated with physiological traits (green). Explain why genome size can affect traits that contribute to invasiveness, using this figure to support your arguments, keeping in mind that some correlations are positive (green) and others negative (orange).



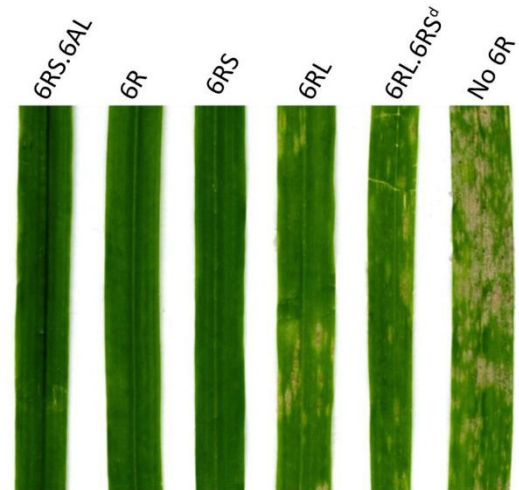
- B) Name and draw/describe/explain two phenomena that can *increase* the DNA content of an X genome without resorting to polyploidy. Use the back side of THIS page if need be.



**40. The following is from:**

Hao et al., 2018. Introgression of powdery mildew resistance gene Pm56 on rye chromosome arm 6RS into wheat. *Front. Plant Sci.*, 9: 1040

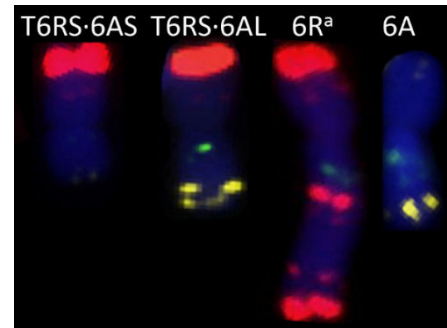
Here, the authors moved the Pm56 gene from chromosome 6R of rye cv 'Qinling' to Chinese Spring wheat. First they developed a 6R(6A) substitution line. This line was crossed with euploid wheat to get the double monosomic. Upon selfing, several chromosomal rearrangements took place, from which they were able to identify a Robertsonian fusion event between wheat and rye that contained the resistance.



- A) As a first step, the authors needed to determine if the resistance gene was on the long or short arm of 6R. The figure above illustrates the results obtained using telosomes, a substitution for 6R and a 6R with a partial deletion of 6RS, indicated by the d superscript. Where is the gene located? Explain the basis for your answer.
- B) Next, they obtained a substitution line. In this case, the substitution line was fortuitous, having been obtained by crossing rye x wheat, and selfing for 11 generations while selecting for resistance. Diagram a faster way to substitute 6R for 6A.

- C) Next, the substitution line was crossed with a euploid wheat to get the double monosomic. Give the expected meiotic (ie, chromosome pairing) configuration for the double monosomic. Which chromosomes will be monosomes? In other words, what will one see at Met I?

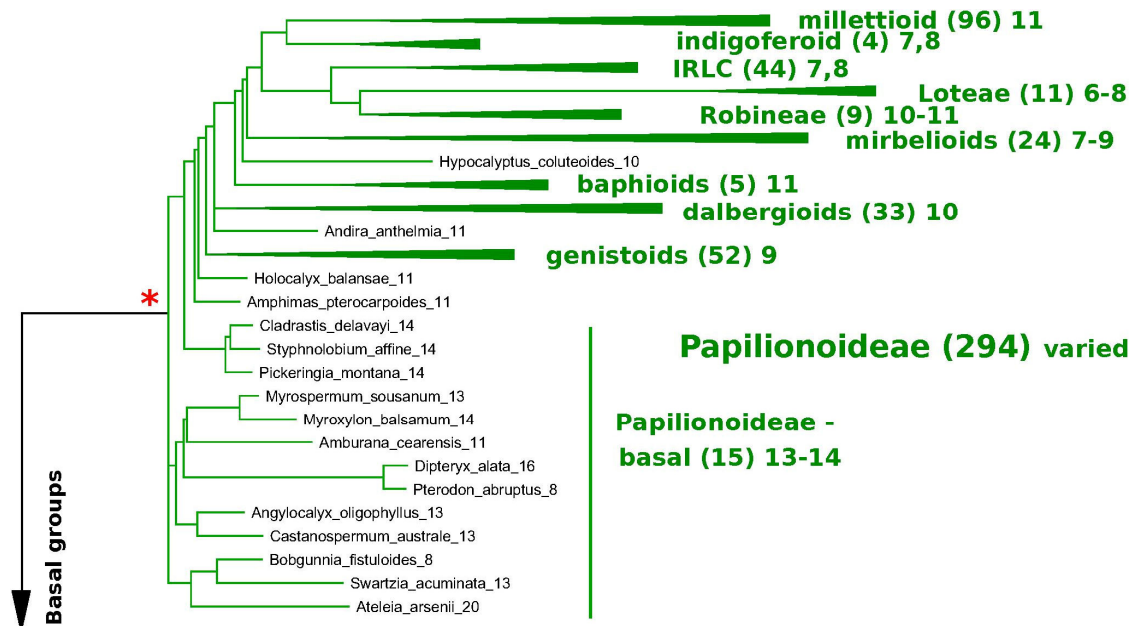
- D) Upon selfing the double monosomic, a Robertsonian fusion took place between the two monosomics, as shown at right. What is a Robertsonian fusion? Diagram the process.



- E) From a breeding perspective, are there any downsides to replacing 6AS with 6ARS? Explain your answer.

## 41. The following is from:

Stai et al, 2019. A non-polyploid genomic relic within the generally polyploid legume family. *Frontiers in Plant Science* 10: 345



(Numbers in parentheses) indicate the sample size, and the last number refers to the monoploid number. So, the basal chromosome number in legumes has gone from  $x = 13$  or  $14$  to  $11$ ,  $10$ ,  $9$ ,  $8$ ,  $7$ , and  $6$ .

- A) What is the name for this phenomenon whereby basal chromosome number decreases?
- B) Diagram and explain two mechanisms by which this happens. Use the back side of this page if need be.

## 42. The following is abstracted from:

GMOWatch: <https://www.gmwatch.org/en/news/latest-news/18731-new-study-claimed-to-show-safety-of-crispr-shows-the-opposite>

*Confirms process-based regulation<sup>2</sup> is necessary.*

A new study by Chinese researchers has been hailed in a lobbying post as showing that plant gene editing via CRISPR is precise, predictable, and controllable. In fact, however, it shows the opposite – that the CRISPR process, taken as a whole, causes large numbers of off-target mutations.

The researchers found that seed saved from wild type non-GM rice plants had 30 to 50 spontaneous mutations per plant.

They found few off-target effects from the CRISPR editing tools – only 2 out of 49 plants had them.

However – and this is the crucial aspect – they found a vast number of mutations from the tissue culture (200 per rice plant).

This is not to suggest that the number of mutations is all-important – it's what the mutations do that matters. That can only be established by further experiments – "omics" analyses to establish compositional changes and then long-term animal feeding studies to establish biological effects. These could include unexpected toxicity or allergenicity.

Dr Janet Cotter of the consultancy Logos Environmental commented on the new study findings, "The researchers found no off-target effects in the majority, but not all, cases. That is why regulation is needed – to ensure there are no potentially harmful effects from the genome-editing process."

Dr Cotter explained: "Off target effects are not the only genetic errors that CRISPR can cause. There may also be unexpected "on-target" effects – for example, deletions and re-arrangements of the host DNA, or interference with gene regulation.

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Using the back of this page, write a letter (not to exceed ⅓ to ½ page) to the editor about the article above. You must cite clear examples from class to support your arguments in the letter. It does not matter if you agree or disagree with the above letter. What matters is how you use the examples from class to support the points you make. Use at least 2 lines of evidence in your answer, and please write neatly!

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<sup>2</sup> Process-based regulation refers to the legal concept that whether something or not is regulated depends on how you make it, not what you make. Thus, you would regulate a variety made using CRISPR, but not one made using X-ray mutagenesis. The opposite legal concept is product-based regulation, where the product is regulated, regardless of the way it was made. The European regulatory system is process based, while the Canadian is product based. The US has a hybrid, whereas the process triggers regulation, but actual assessment is product based.

**43. The following is inspired by:**

Owens et al, 2018. Genomic sequence and copy number evolution during hybrid crop development in sunflowers. *Evolutionary Applications* 12(1): 54-65.

This paper centers on PAV in the sunflower genome. The traditional perspective is that plant breeders have found desirable alleles in wild germplasm, old varieties, landraces, etc. Through backcrossing, they replaced a chromosomal segment bearing the undesirable allele in the recurrent parent with a chromosomal segment bearing the desired allele from the donor parent. The chromosomal segment could also have undesirable alleles, leading to linkage drag.

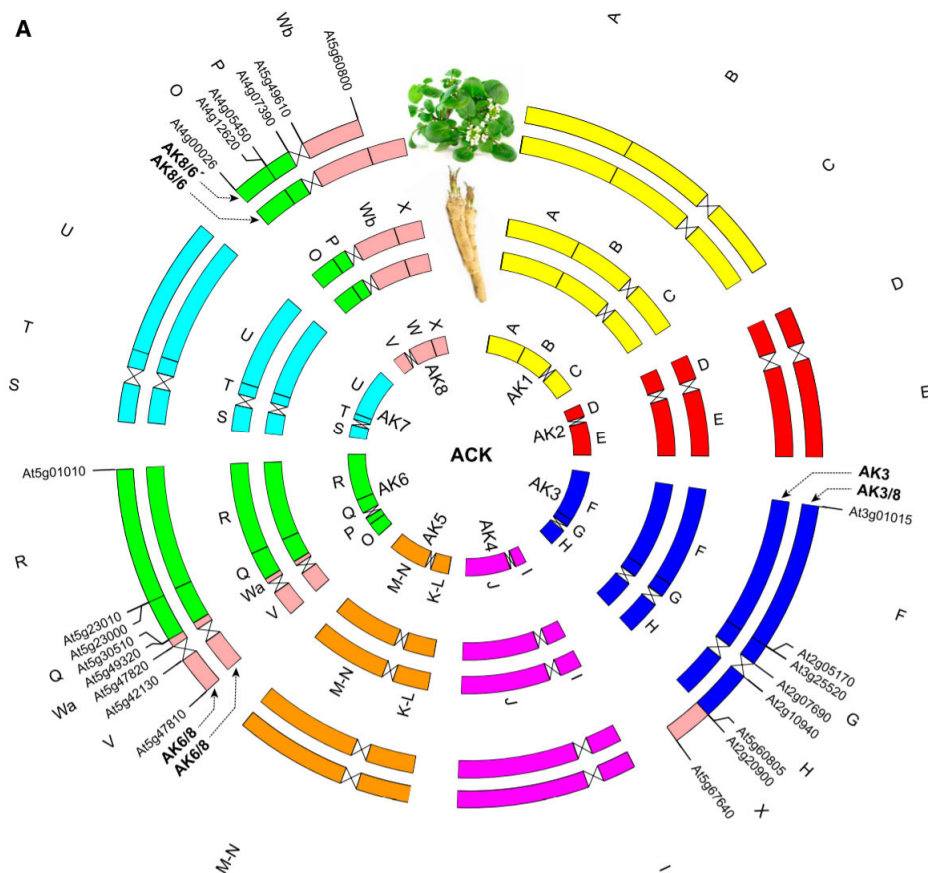
This traditional concept needs to be updated and expanded based upon current knowledge of the plant genome. What else can happen during backcrossing, besides allele replacement?

## 44. The following is derived from:

Mandáková T & MA Lysek. 2019. Healthy roots and leaves: comparative genome structure of horseradish and watercress. *Plant Physiology* 179: 66-73.

The innermost circle shows the Ancestral Cruciferous Karyotype, while the outer circles show the chromosomes for 2 economically important crucifers. The middle pair are for horseradish, and the outer set are for watercress. Both are  $2n = 4x = 32$ . Horseradish can be grown from seed, but is usually vegetatively propagated. Watercress is grown from seeds.

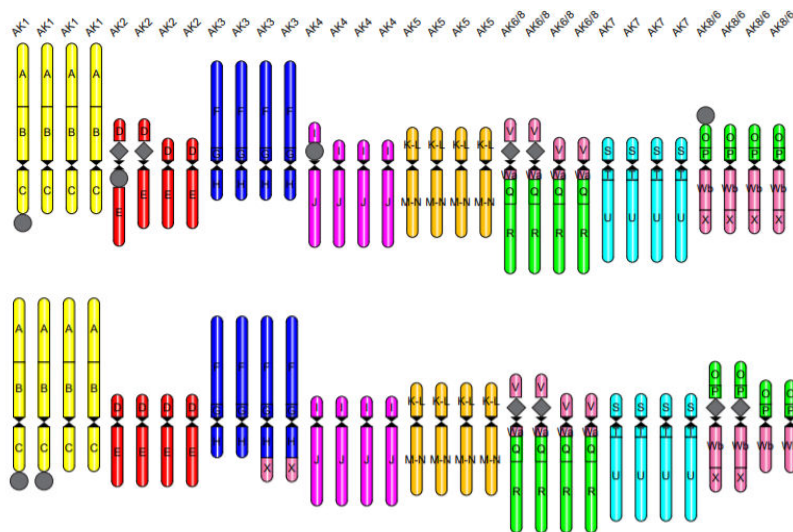
In horseradish, the two sets of chromosomes appear to be identical. In watercress, there is a chromosome 3/8 translocation in one of the sets. Both species have an 8/6 translocation that occurred in an ancestral species.



- a) Is watercress an auto or allopolyploid? Explain the chromosomal feature(s) that allow you make that determination.

- b) What about horseradish? Describe 3 lines of evidence and expected results that you would like to see before deciding if it is an auto or an allopolyploid. Any ancestral species are extinct, so cannot be used.

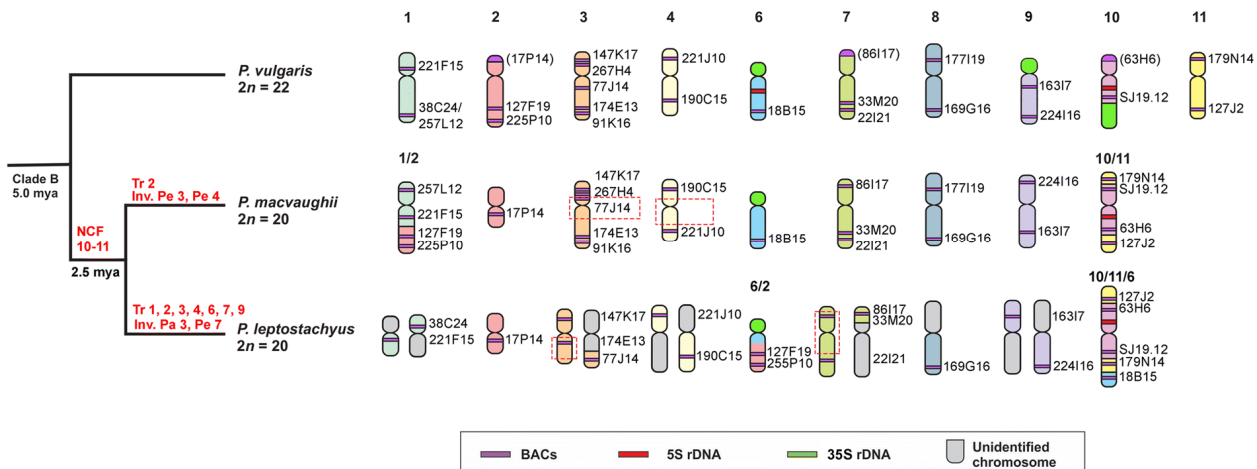
Upon further comparison of the horseradish (*top*) and watercress genomes (*bottom*), it is apparent that NOR regions in horseradish have moved. In the figure below, squares: 5S rDNA loci, grey circles: 35S rDNA loci.



- c) It is not known if the NOR changes were in the parent genomes of horseradish, or if they happened after tetraploidization. Regardless, explain the properties of NOR regions that permits them to move around the genome. Draw and upload a diagram as you wish.

**45. The following is from:**

Ferraz ME, A Fonsêca & A Pedrosa-Harand. 2020. Multiple and independent rearrangements revealed by comparative cytogenetic mapping in the dysploid *Leptostachyus* group (*Phaseolus* L., Leguminosae). *Chromosome Research* 28: 395-405



Ideotypes for common bean (*Phaseolus vulgaris*) and 2 other related species.

- Describe and/or add a diagram explaining how chromosomes 10 and 11 became one in *P. macvaughii*.
- Based on the karyotype, is *P. Leptostachyus* an allopolyploid, mesopolyploid, or diploid? Describe its karyotypic features that lead to that conclusion.



46. Is from:

<https://www.ams.usda.gov/sites/default/files/media/MSEExcludedMethodsApril2019ProposalWebApril2019.pdf> and

<https://www.ams.usda.gov/sites/default/files/media/MSEExcludedMethodsProposaFall2019.pdf>

Organic agriculture is defined according to a set of production standards. These tend to have a philosophical basis, and need not be science-based.

In general, the use of natural substances is allowed, unless explicitly prohibited; synthetic substances are forbidden unless explicitly allowed. These prohibited and allowed substances may be viewed on the [National List of Allowed and Prohibited Substances](#). Items are added/removed to the list by the National Organic Program (NOP) through a rule-making process as described earlier for the EPA.

Gene editing, in its current incarnation, is seen by breeders as a way to replace mutagenesis. The DNA lesion from editing is frequently indistinguishable from one created by mutagenesis. However, mutagenesis is random, while editing allows the breeder/geneticist to target the gene to be mutated.

The use of gene-editing is not allowed for organic agriculture. In addition there is a proposal before the NOP to exclude the use of conventionally mutagenized plants in organic production. The stated rationale for banning editing and perhaps mutagenesis is:

“The genome is respected as an indivisible entity, and technical/physical insertion, deletions, or rearrangements in the genome is refrained from (e.g. through transmission of isolated DNA, RNA, or proteins). In vitro nucleic acid techniques are considered to be invasion into the plant genome.”

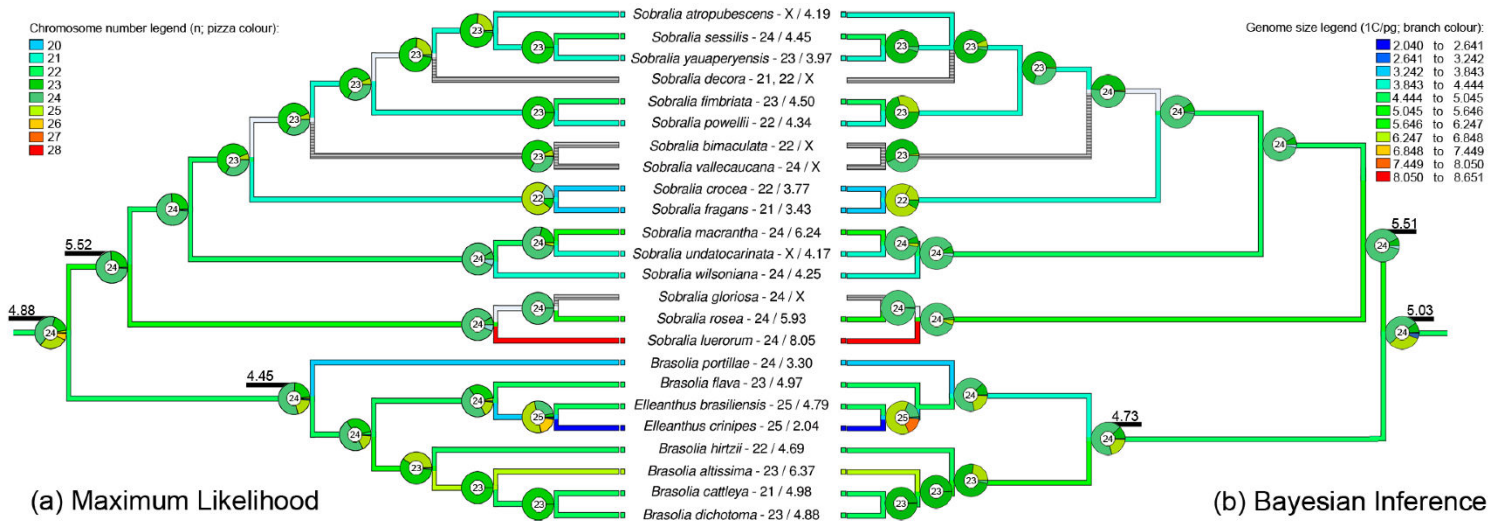
“We are aware that specific laboratory tests are not currently available to detect the use of several new excluded genetic modification technologies in organisms. However, we still believe that the technology should be listed as an excluded method, when appropriate, and anticipate tests or other methods will be developed over time to detect the presence of these technologies.”

Sooner or later, this proposed exclusion will go out for public comment. Go ahead and prepare your comments to support or not support the rationale being used to exclude gene editing and mutagenesis.

Limit your comments to a **maximum** of 1 page + additional pages for the references. Use *Plant Physiology* format for your references.

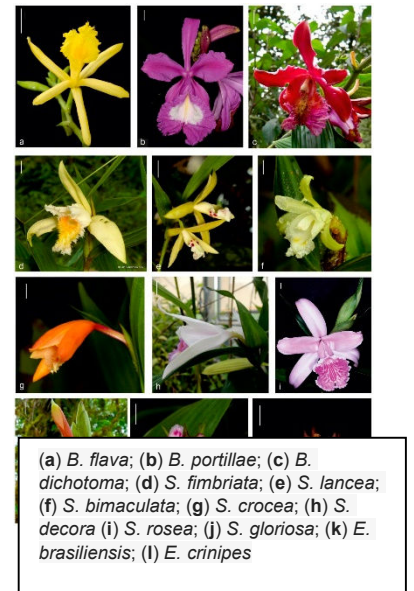
**47. The following is from:**

Baranow P, J Rojek, M Dudek, D Szalchetko, J Bohdanowicz, M Kapusta, I Jedrzejczyk, M Rewers & P Moraes. 2022. Chromosome number and genome size evolution in *Brasolia* and *Sobralia* (Sobralieae, Orchidaceae). International Journal of Molecular Sciences, 23(7): 3948.



Answer the following questions based on the phylogeny provided:

The ancestral chromosome number for this group of orchids is  $n = 24$ . Explain or diagram two ways that would explain how some *Sobralia* spp. came to have  $n = 23$



Based on the same phylogeny and set of spp., *Elleanthus crinepes* came to gain a chromosome. Explain/diagram one way by which this may happen.

Based on the same phylogeny and set of spp., the ancestral species is estimated to have had approximately 5 pg DNA per 1C.

- a) Explain/diagram how *Brasolia portillae* would have lost DNA to have just 3.3 pg/1C.
  
  
  
  
  
  
  
  
  
  
- b) At the same time, explain and/or diagram how *Sobralia luelrorum* would have gained DNA without increasing chromosome number

Based on the same phylogeny and set of spp., the ancestral species is estimated to have had approximately 5 pg DNA per 1C.

- c) Explain/diagram how *Brasolia portillae* would have lost DNA to have just 3.3 pg/1C.