

The Population Explosion

An invitation: The activities for explorations in population density and nutrition in this document are based on some ideas and protocols that we have been developing and evaluating over the past two years. Teachers at workshops have been enthusiastic about the investigations and so we offer this version for your experimentation. Please let us hear from you, how you would improve these materials.

As Earth's human population density grows, there is increasing competition for resources, including food, space and energy. Can we assume that humans will always find ways to manage the limited resources on earth so that a high quality of life will be maintained regardless of the number of people to be accommodated? Can scientists continue to improve crop productivity so that all people will be adequately fed? What are the essential criteria to be considered that relate to the survival and continuance of all species, including Homo sapiens? What are the factors that sustain and limit human population on earth? This exploration, and the many activities that derive from it, address issues similar to those surrounding the existence and survival of the human species.

Ideally, any class entering into any of the following investigations will have already grown a complete life cycle of Fast Plants, or Rapidcycling *Brassica rapa* (Rbr). They will understand plant growth, development and reproduction, and will have observed and experimented with variation in a population by measuring height and other characteristics of Fast Plants at various days of the life cycle. They will have collected their own data and combined it as part of a class population data set.

The Exploration

The "Population Explosion" explores the effects of increasing plant population density on growth, development and reproduction. This highly visual hands-on activity illustrates various environmental, ecological and evolutionary concepts from which students should be able to deduce various implications of increasing population growth. Math and language are integral to all the investigations. Students observe, discuss, measure, and record. They organize their data and learn to draw inferences from what the data represent. They will discuss ideas and outcomes and persuade their peers of their perspective.

Graphing represents one way of organizing the data. Looking for patterns in the organization of the data leads to insight about the experiment and to new approaches for testing the model. Students should be able to explain orally and to write about what they have done, so that it is understandable to others. Can they explain what they are doing in mathematical symbols?

Design of the Exploration

The population explorations are presented as a set of standard models that represent three levels of increasing conceptualization, experimental execution and analysis. In the first level, one variable, the number of plants in each pot (density) is increased through five successive doublings.

In the second level, two variables, plant density and added nutrients are doubled successively to produce a grid of 25 pots.



At the third level are longer term activities that investigate the evolutionary concepts of selection and adaptation, creating new populations derived from the experimental mating of "selected" individuals from either the Plant Density or the Density X Nutrition populations.

A fourth option with a <u>storyline</u> offers students the chance to design their own experiment in which they distribute populations on five continents and study the consequences of population density.

Each of the investigations can be completed within 18 days unless the group decides to complete the life cycle for the purposes of counting the seeds produced or make selections of progeny (seed) for the next generation.



Model 1 – Plant Population Density: A Single Variable

Experienced Fast Plants students are aware that Rapid *Brassica rapa* (Rbr) is inherently variable within the population, even though the environment is relatively uniform. This variation is due largely to genetic (genotypic) differences among individuals, although small, uncontrolled differences in the environment of each plant contribute to some of the observed variation. This exploration introduces one *experimental variable*, that of change in the number of plants in each wick pot. The resources available (nutrients, light, water, etc.) for plant growth in each pot are held *constant*

From an ecological perspective, each pot could be viewed as representing a microhabitat defined by the pot's environment that supports the specified number of plants. At a certain population density and/or at a certain stage in their growth, the plants in each pot will begin to compete for the resources available in the pot. The consequences of *density dependent competition* can be observed, measured and recorded.

During density dependent competition one or more of the environmental resources become *limiting* to plant growth. Students should brainstorm and list the various physical (temperature, light, etc.), chemical (air, water, nutrients, etc.) and biological (microbes, other plants, etc.) factors that comprise the environment of the Fast Plants. They should speculate on which of these factors held constant from pot to pot are likely to be having a major affect on growth as plant density increases.

This single variable experiment can be the basis for further experiments in which other factors are varied and plant density is held constant. *Single variable experiments* generally provide the simplest experimental designs that can be most easily repeated.

Natural phenomena rarely result solely from the influence of a single environmental variable, though single variable experiments are usually the best way to begin understanding complex phenomena. Students (and scientists) need to learn to deal accurately (critical thinking and technique) with single variable experiments before they can design or interpret more complex, multiple variable (*multivariate*) investigations.

Preparation

By dividing the class into groups of five or fewer students each, the teams can replicate the experiment and compare their data.

- At least 50 seeds will be needed per group.
- Each group will plant five film can wick pots.
- The number of plants grown in each succeeding wick pot will double.

During the investigation, students will:

- observe details of growth and development of plants within and among the pots;
- measure and record height of the plants every two or three days; and
- take any other desired data, both numerical and written, e.g., numbers of leaves, number of flowers.

Materials

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- five film cans per group
- Peters[®] fertilizer
- thick, unpolished cotton string
- Fast Plants water reservoirs & mats
- recommended soil and vermiculite
- Fast Plants seeds (basic or dwarf stock)
- a light bank with six, or preferably eight cool white fluorescent bulbs

Procedure/Growing Instructions

- 1. Each group of students should prepare five labeled wick pots for planting.
- 2. Drill or melt an Figure 1 approximately 5 mm hole in cut here the bottom of height: 3 cm the film can. With a scissors, cut Pot 4 the height of label the film cans film can pots down to 3 cm high so that the volume is approximately 20 cc (Figure 1).
- 3. Use a 5 cm. length of unpolished cotton string or other wicking material to wick each film can pot (see Figure 2 as an example).

Note: Put the string in your mouth and chew it for a moment to moisten it and break the surface tension before positioning it in the wick pot.

- 4. Fill the wick pots with slightly moistened commercial peat/vermiculite soil such as Jiffy Mix[®] or Terralite Redi-earth[®]. Do not push down or pack the soil mixture.
- 5. Gently water from above until the wick drips. The soil will recede about 5 mm.
- 6. Plant seeds as shown in Figure 2. The recommended number of seeds for the various wick pots will be enough to yield the desired number of mature plants.

Cover the seeds with a layer of vermiculite, water from the top again and place wick pots on the water mat of the Fast Plants reservoir.

- 7. Plants should be grown under standard Fast Plants lighting 24 hr/day with tips of plants 5 to10 cm from lights at all stages in the life cycle.
- 8. Thin plants to the desired number on Day 3 or 4 by snipping out extra seedlings with a small scissors (see Figure 3). Notice that the number of plants doubles each time, so that pot #1 has one Fast Plant and pot #5 has sixteen plants.
- 9. Add nutrient. Mix a liquid fertilizer solution by adding one level soda bottle capful of Peters[®] (N-P-K: 20-20-20) fertilizer crystals to one liter (soda bottle) of water. (Other brands of soluble or liquid fertilizers can be used; use the recommended formulation on the product label.)

Apply 2 ml of the solution to each wick pot on Days 3, 7 and 14 (and Day 21 if you are not harvesting the plants until the end of the normal life cycle).

- 10. Pollinate whenever flowers open (usually Days 13 to 18).
- 11. Harvest on Day 18 (for Models 1 & 2).
- 12. Alternatively, your students can complete the entire life cycle. Twenty days after the last pollination withhold water and let the plants dry for 3-5 days and then harvest the pods and seeds (approximately 42 days after sowing).

Figure 2:

Seeds to be sown per pot in Model 1 investigation.











2 seeds

4 seeds

8 seeds

12 seeds

24 seeds

Prepare the plant presses

Before Day 18, each group of students can make a plant press (Figure 4). The press should be 8 1/2 by 11 inches. The outside layers can be made of heavy corrugated cardboard. Make filler pages from pages of telephone books, newspaper, or blotting paper. At least 40 pages of an old telephone book or newspaper or six pages of blotting paper are needed for each press.





Harvesting and pressing plants

- 1. Take final measurements Day 18.
- 2. Carefully harvest plants, one wick pot at a time, by cutting them off at soil level. Transfer plants from each wick pot to the plant press. There will be six or more filler sheets of telephone book paper or newsprint or one sheet of blotting paper between each group (pot) of plants.
 - First place the bottom cardboard cover on your work surface. Put six filler sheets (or one of blotting paper) on the cardboard.
 - Begin by harvesting the single plant from wick pot #1. Lay it carefully on the filler sheets. Attach a Post-it[™] note to the page, writing down the harvest date, the number of plants, and the level of nutrient.
 - Carefully spread the leaves and seed pods, pressing them firmly with a finger. Then cover with six or more filler sheets. This will complete the first layer of the plant press.

- 4. Harvest wick pot #2 and lay both plants on top of the filler sheets of the first layer. Make appropriate notations and cover with six or more filler sheets to complete the second layer of the press.
- 5. Continue the process until all wick pots have been harvested and pressed.
- 6. Place the other cardboard layer on top and secure the whole press with large (no. 64) rubber bands or clips (see Figure 5). Place three or four large books on the press to flatten the plants.
 Figure 5
- 7. Let the plants dry for a week.
- 8. One layer at a time, remove the dry plants from the plant press, being careful not to break off any parts.
- 9. Arrange the plants from this layer on a sheet of white 8 1/2" X 11" paper. There may be ways to arrange the pressed plants on the sheet that provides insights into "patterns" and numbers of plants.

Figure 6:* Appearance of 18 day old plant, grown at density of one plant per pot, at recommended concentration of fertilizer (in small grid, see page 9 for full-size grid and description).



- 10. Once plants are arranged, fix them to the sheet with small strips of clear tape.
- 11. Transfer the information about the density, pot number and harvest date to the bottom of the paper.
- 12. Continue until all pressed plants have been transferred to the white paper, secured with tape and with information noted.
- 13. Mounted plants can be covered with clear protective plastic wrap (e.g., Saran Wrap[®]), or put between clear sheets of plastic, e.g., in clear page mounts for 3-ring binders.
- 14. The mounted, pressed plants can then be xeroxed and the photocopies of the plants given to the students for observation and extraction of data. See Figures 6 and 7 below as examples.

Figure 7:* Appearance of 18 day old plants, grown at density of 16 plants per pot, at recommended concentration of fertilizer (\blacksquare in small grid).



* These plants (and those shown in Figures 9 and 10 on page 9) have the genotype *dwf1/dwf1*, and are shorter than the basic stock. Either stock can be used for your experiment. These figures were produced by xeroxing the pressed pages, electronically scanning the xeroxes, and then reducing the size of the image by half.

Organizing the data

Students within groups should brainstorm as to what to measure. That is, what are fair or reasonable indicators of plant growth? What about height, # of leaves, # of flowers, etc.? Groups should then come to class consensus on what and how to measure on the copied images of the pressed plants to create a class data set.

- Estimates of observations can be obtained by measuring individual plants in each population and presenting data as averages for each pot.
- For additional population data, calculate the range and standard deviation for each population.
- Estimates of growth, e.g., plant height, leaf length, should be either in mm (dry weight in mg) or in direct counts of leaves, open flowers, pods or seeds.
- Questions will arise as to the role of genotypic variation and the reliability of measurements, particularly at the lower plant densities of 1, 2 and 4 plants/pot. How can the reliability of these growth measures be improved? (Hint: replications.)

The data generated provide a rich resource for discussion of the experimental design and of the role that math, graphing and statistics play in developing an understanding of the natural phenomena exhibited in the experiment.

• Can students describe and write down in words how the experiment was designed?



Pot #	Plant Density (# plants/pot) = x	Average Plant Height (mm) = y					
1	1	22.8					
2	2	18.6					
3	4	15.3					
4	8	9.4					
5	16	7.2					

** hypothetical data

- Do they understand what it means to "hold experimental elements constant?"
- In organizing their data and graphing, do they understand the notion of variables?
- Can they develop a mathematical notation of the design?

In this exploration the variable of plant density is predetermined by the design of the experiment and can be referred to as the *experimental*, or *independent*, *variable*. The design of the experiment is to start with one plant in a pot and to double the number of plants in succeeding pots (the independent variable) for up to five pots. From among those observed, students will determine which indicators of growth they will record to analyze the consequences of crowding.

> Any particular indicator of growth chosen, e.g., height, # leaves, dry weight, etc., is known as a response, or dependent variable, because it is assumed that this indicator has a dependent relationship to the independent or experimental variable. Normally, in organizing data by tabulating (Table 1) and graphing (Figure 8), the independent variable is assigned the symbol "x" and the dependent variable the symbol "y". In two dimensional graphing, as illustrated in Figure 8, the independent variable, x-axis, is normally portrayed in the horizontal direction and the yaxis vertically. Then create a class data set and discuss.

 Table 1: Relationship of plant density (x) to height (mm; y)

 on Day 18.**



Model 2 — Plant Population Density X Nutrition: Two Variables

This set of experiments builds upon the Plant Population Density experiment in which students will have observed the dramatic effects on plant growth as the environment becomes limiting under density-dependent competition for growth resources. Students will understand the effects of varying one factor (plant density, the independent variable) while holding all other experimental factors (potential variables) constant.

In the Plant Population Density X Nutrition experiment, the independent variable of doubling plant density is repeated, while at the same time a second independent variable, that of doubling the amount of the major element nutrients (nitrogen [N], phosphorous [P] and potassium [K]), is added at each of the plant densities (Figures 9 and 10).³

The interaction between the two independent variables may give insights into whether

Figure 9: Appearance of 18 day old plants, grown at density of eight plants per pot, at 16 times the recommended concentration of fertilizer (\square in small grid).

nutrition was a major limiting factor for Fast Plants' growth and reproduction. The Plant Population Density experiment may generate many other questions and hypotheses that could be tested with single variable experiments.

In moving from a single variable experiment, Population Density, into an experiment with two variables, plant population density and nutrition, students will begin to understand the difficulties facing agricultural scientists, ecologists, epidemiologists and nutritionists as they try to explain complex interactions relating to crop productivity, disease spread, the maintenance of ecological balance and the conservation of species, all of which are influenced by the exploding human population.

Procedure

Use the same overall procedureas in the Plant Density model *except* for the following changes and the further suggestions in the Mathematical Connections section.

Figure 10: Appearance of 18 day old plants, grown at density of eight plants per pot, at the recommended concentration of fertilizer (■ in small grid).



- 1. The class needs to be divided into five teams of students. Each team will plant and be responsible for one replicate of the Plant Density experiment (five wick pots, with seeds doubling each time). It would be ideal for each team to plant two or three replicates.
- Five different solutions of liquid fertilizer will be prepared at concentrations of 16X, 8X, 4X, 2X and 1X. To mix this series of dilutions you will need five empty 1-liter soda bottles.
 - Using a felt tipped pen, mark each bottle at the one liter and half liter levels. (Note, a 1-liter bottle has a 1050 ml volume)
 - Label the bottles #1 through #5.
 - Mix four level soda bottle capfuls of Peters[®] fertilizer crystals into Bottle #1 and fill to the one liter mark with water. Shake gently to dissolve and mix. Label it 16X.
 - Pour half the contents (500 ml) of Bottle 1 into Bottle 2. Set Bottle 1 aside. Add more water to Bottle 2, up to the one liter mark. Mix gently. Label it 8X.
 - Pour half the contents (500 ml) of Bottle 2 into Bottle 3. Set Bottle 2 aside. Add water to Bottle 3, up to the one liter mark. Mix gently. Label it 4X.
 - Continue in the same way for Bottles 4 & 5, labeling Bottle 4 "2X" and Bottle 5 "1X."
 - Peters[®] fertilizer is blue. Therefore, the solution in each succeeding bottle is lighter colored.

Can the students explain in words and mathematical notation how they made the dilution series of N-P-K solutions? How does diluting the fertilizer differ in effect from the doubling of plant number in the Plant Density experiment?

- 3. Each team will be assigned one of the nutrient solution levels to add to their plant density series of wick pots (Figure 11).
- 4. Each team should place their wick pots on a class reservoir, e.g., 2.4 liter square Rubbermaid[™] container, to form a grid of 25 wick pots as illustrated in Figure 11. As the plants grow they will display a three-dimensional "graph-like" landscape of plant growth which will raise many questions.

5. Each team will harvest plants grown at a single nutritional level. The class can compile team data and pressed plant xeroxes to complete the total picture of the effects of nutrition and density on Fast Plants.

Mathematical Connections

1. As the plants grow in their 5 by 5 configuration, students should refer to the two-dimensional, density X nutrition, planting grid (Figure 11) and note the various numerical relationships among the pots in the grid. Note the fractional relations between plant density and nutrient concentration and the patterns of numbers they form in the grid.

The grid portrays a series of five single variable plant

density (x-axis) experiments, each at a different (constant) nutrient level (zaxis), while it also illustrates a series of five single variable nutrient experiments on



the z-axis, each at a different constant plant density represented by the x-axis.

These single variable sets can be measured and graphed by each team using height or other indicators of growth as the dependent variable on the y-axis. As in the single variable experiment, it is important to measure each plant in a given pot and calculate average estimates of the growth indicators used. These numbers can be entered in the open squares on the grid.

2. The interaction of density and nutrition becomes evident when numbers in diagonal positions to each other on the grid are compared. Fractional relationships can be investigated and discussed prior to actually measuring plant growth. Look at the fractions on various diagonals. Have students describe what the fractions represent in terms of plant density and nutrition. Then have them raise questions relating to plant growth and the interaction of nutrition and plant density (see Figures 9 and 10).

Team number z = concentration of NPK/ml (multiples of 1X solution; nutrition)

Figure 11: Plant Population Density x Nutrition, experimental design and data grid.

x = number of plants per pot (plant density)

- 3. Before harvesting, photograph (or illustrate) the experiment. After harvesting, pressing and mounting the plants, students should measure the plants for growth indicators agreed upon and calculate means and other useful statistics. They can fill in the rest of the grid with the growth indicators, e.g., height, width of largest cotyledons, etc.
- 4. Students might:
 - plot a three-dimensional graph and compare the graph with the photo or drawings of the living experiment prior to harvest.

- discuss other questions and experiments that could be run to examine the interactions of environment and plant growth.
- develop notations describing the algebraic relationships among the three variables.
- use calculators or computers to graph their data.
- think of other major variables that should be placed into the experimental design. The possibilities are virtually unlimited!



Model 3 – Population Density and Selection: Do the "Fit" Survive?

If you continue the Plant Population Density or Plant Population Density X Nutrition plantings as reproducing populations and collect seed from plants in the individual pots, then you have the possibility of extending these activities into experiments that can explore *selection* and *adaptation*, important concepts in evolution.

The three dimensional (xyz) "landscape" of Fast Plants' growth reflects the two variables of plant density and nutrition and represents a grid of varying environmental stress over a population of 155 plants (Figure 12).

From an ecological perspective pots on the grid may be viewed as environmentally defined habitats, each differing from the other in the quantities of nutrients available and the number of plants present. Plant growth is influenced by numerous factors, among which nutrition and density are limiting contributors. Some pots of plants in the grid may be sufficiently stressed as to be incapable of producing viable seed. Plants in other pots will vary in their capacity to produce viable offspring (*reproductive fitness*).

Preliminary Activity:

Characterizing the Reproductive Landscape

Carry out a preliminary Plant Population Density X Nutrition experiment as determined in Model 2 by interpollinating all the plants to determine the reproductive capability of plants in the various pots to produce offspring. This may refer to their relative fitness. Are pots anywhere in the grid incapable of reproduction? By plotting the number of seeds produced from each of the 25 pots on the grid, a landscape of reproductive potential (fitness) can be graphed threedimensionally.

- What is the reproductive output for each pot on the grid?
- Are there particular subpopulations (one or more pots, defined by particular nutrient X density combinations) on the grid that students would like to investigate with respect to their potential to grow and reproduce (adapt) in the defined environment?

Figure 12: Three dimensional graph showing 3 variables: x-axis = plant density (plants/pot); y-axis = plant height; and z-axis = nutrient concentration.



• What is the average fitness (seeds per plant) for the total population?

To investigate these questions, many variations in the design of this investigation are possible. See Figure 13, in which plants growing in two areas of the grid in Figure 11 defined by specific environmental parameters are each intermated as a "selected" subpopulation and their progeny repeatedly selected over a number of generations using the same criteria. The measure of the reproductive success of the original and subsequent generations could be studied as indicators of genetic change within the subpopulations.

Generation 1:

Repeat the experiment (plant the grid again), but this time students can select specified areas on the grid and intermate the plants within those areas.

- Intermate the plants growing in a stressed area of the grid in nutrient X density (N/D) ratios of 1/4 or less (see Figure 13).
- Similarly intermate plants in the area defined by a N/D ratio of 4/1 or greater.
- What are the reproductive capacities of these areas (seeds per pot or per plant)?
- What would be a *control* in this experiment?

Generations 2 & 3:

Replant the seed from each of the newly selected populations and repeat the experiment, but in this second and subsequent generations intermate only those plants from the nutrient stressed ($\leq 1/4$) environments.

Similarly, plants from the nutrient rich environments ($\geq 4/1$) should only be derived from those growing in that environment. A design of this model is in Figure 13.

- Keep careful data on growth and seed production for each generation's subpopulation, including the control population.
- This investigation could be continued through a number of generations of selection and populations grown to determine whether Fast Plants are adapting by exhibiting increased reproductive fitness within the experimental environments defined by nutrition and plant density.

- Produce as much seed as possible from each of the selected populations. The stressed populations will have to be sufficiently large, particularly in the early generations, to sow a subsequent population. Replication of student experiments will increase seed numbers.
- There are many other variables that can be taken into consideration in the experimental design of these investigations, e.g., actual number of plants in each selected population contributing to the subsequent generation. Should the numbers of plants in each generation, the subpopulation, be kept equal?, held constant for each generation?, allowed to vary?

Generation 4:

Replant each population as in the original grid, e.g., Figure 11. After four generations, has there been any evidence of gain (in fitness, etc.) from selection? Any evidence of evolution?





Student Designed Model: A Storyline

This "Brains-on" population density activity in experimental design is appropriate for middle school through college. The students become the "master planners" of a civilization of Fast Plants that is to exist in a world that has five similar sized continents in a global ocean.

- 1. Divide the class into teams of three or four students.
- 2. Give 50 Fast Plants seeds to each student team.
- 3. Each team is responsible for a world of five continents. Ask the teams to distribute, plant and grow the seeds in some manner within their continents.
- 4. Given the way the seeds are distributed and planted, the students will be expected to deduce something about the effects of population density on the growth, development and reproduction of the individuals (Fast Plants) on their continents.
- 5. Give the students sufficient instructions so that they understand what needs to be done without telling them what numbers of seeds to plant in each film can pot (continent). They'll come up with different designs for addressing the problem. They do not have to use all their seeds, but they may use no more than 50.

Students should develop a design:

- for deliberate distribution of the seed to the five continents.
- that allows them to make predictions about the populations on their continents based on the distribution of the seed.
- in which the distribution plan for seed should be clearly able to be repeated by the group and by other groups.

Students should be able to:

• present their model and persuade the other teams that their design is more informative than others and that it provides a model that leads to more reliable predictions.



Measuring height, development, reproductive capacity:

- Teacher and class should come to consensus on what growth and developmental variables will be measured by all groups. Individuals may measure other variables as they wish.
- If plants are grown only for observation and measurements, various "destructive" methods may be used for data taking, e.g., plant pressing, seed harvest.
- If plants are going to be measured, selected and inter-mated for seed harvest (density selection) they need to be measured "nondestructively."

Scientific process in experimental design:

- Setting up experiments.
- Keeping all inherent variables within the environment constant and varying one.
- Getting a reliable model; what is the control?
- Observation, measuring, replication, precision, accuracy?
- Questions coming from the observations and insights of students lead to other investigations.