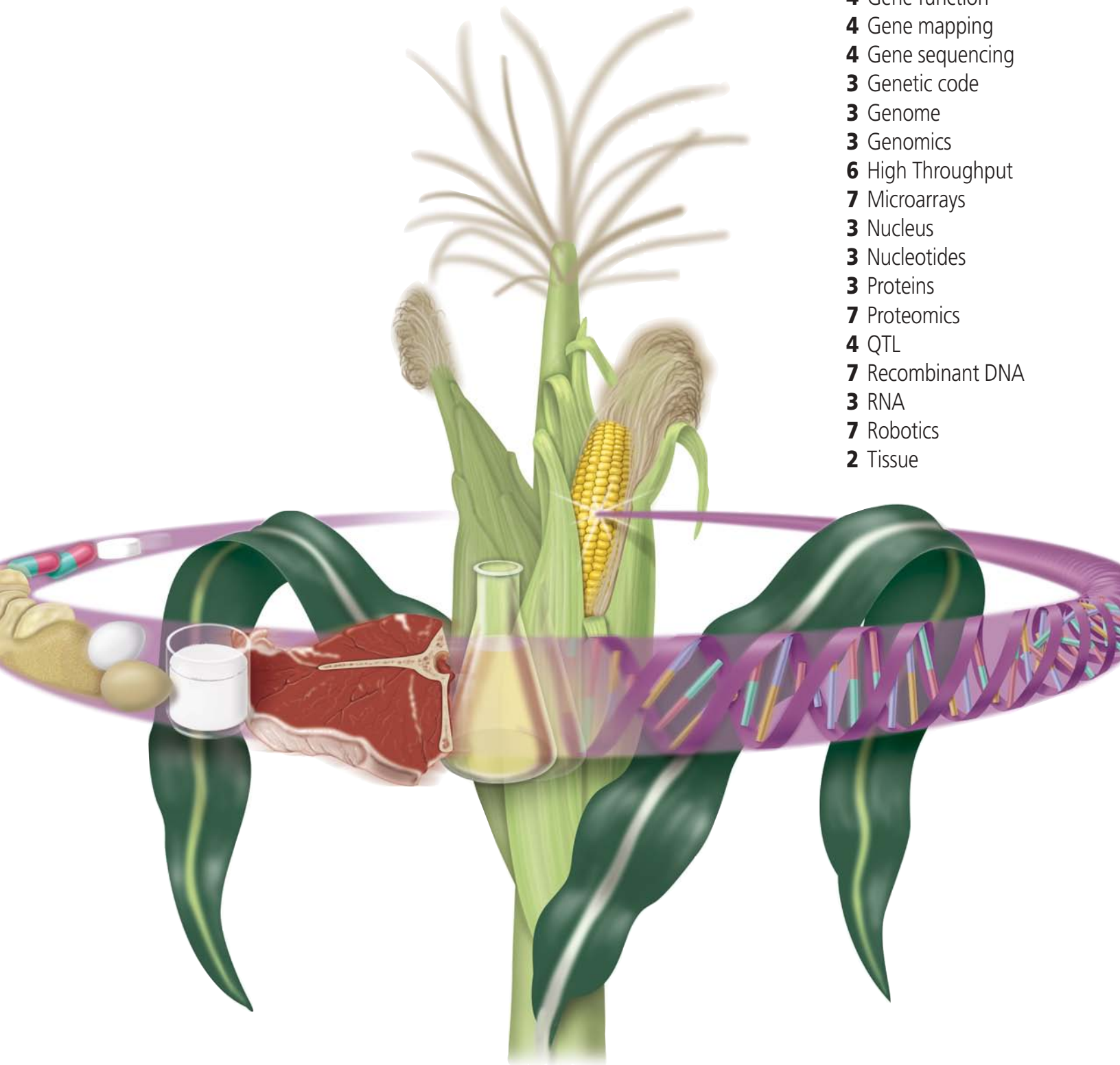


Using Genomics in Plant Genetics Research

Unlocking Genetic Potential for Increased Productivity

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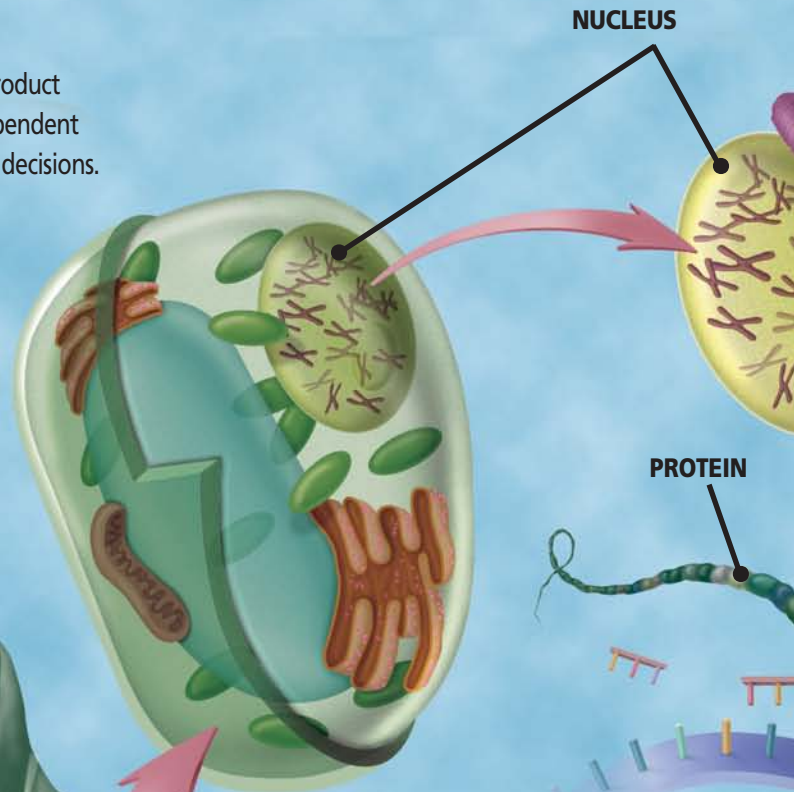


From the big picture to the most minute plant

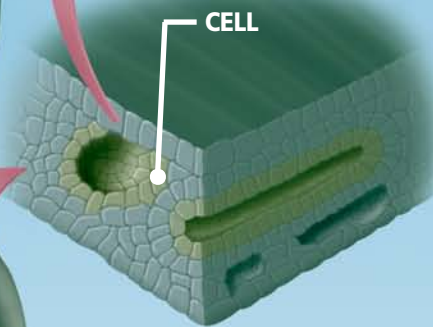
The knowledge of plant performance has mushroomed to bring more opportunities for researchers to increase plant productivity. Just changed from the telegraph to the telephone to the Internet, so has genomics helped plant genetic researchers work faster, smarter and

The structure of living things

From physical observation to molecular levels, researchers have turned the product development process inside out. It wasn't that long ago researchers were dependent on a plant's outward appearance and pedigree information to make genetic decisions. Today, they dig deep into the genetic makeup to determine gene function.



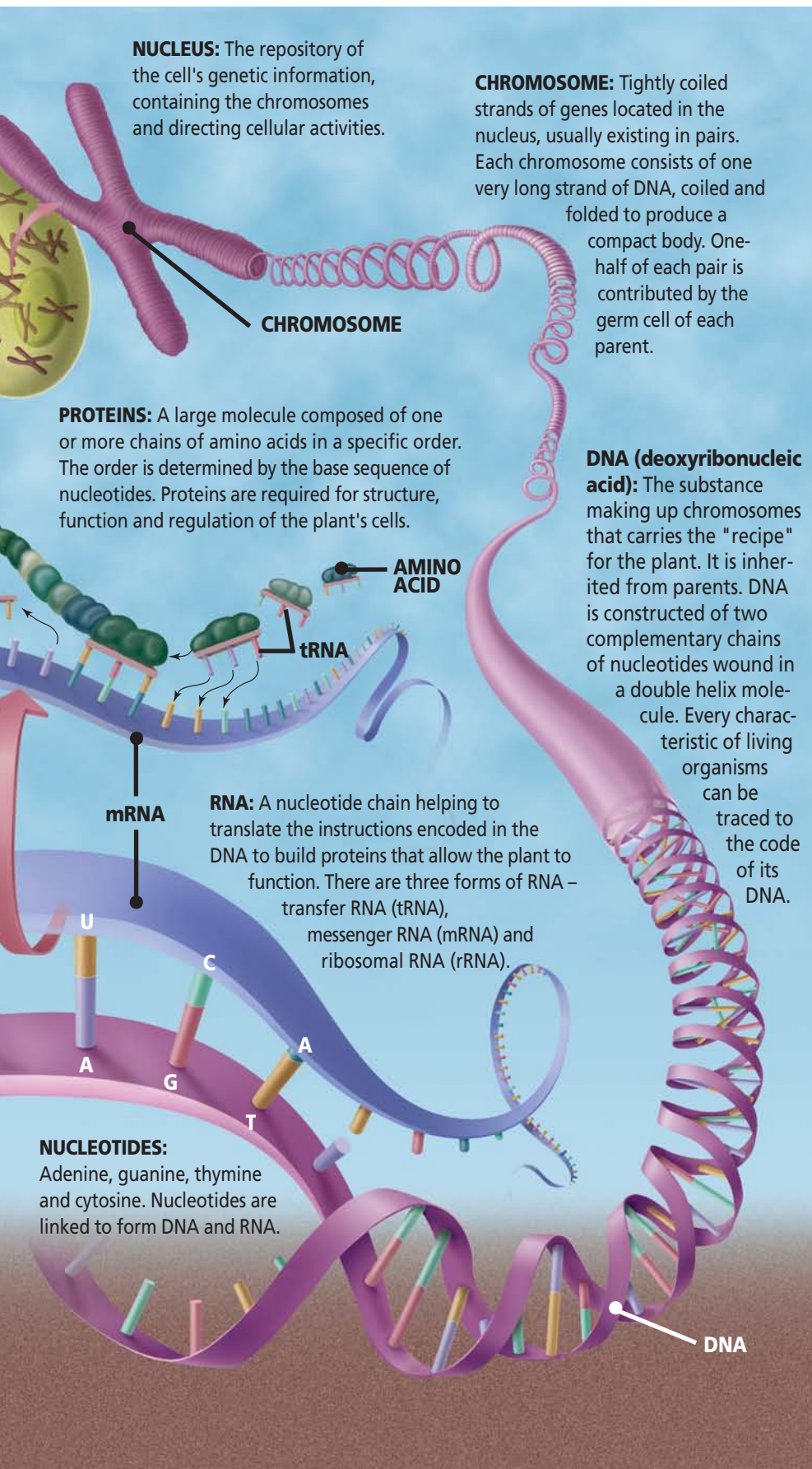
CELL: The basic unit of living systems.



TISSUE: A group of cells, often with a similar and unique structure that makes up part of a plant.

function

as communications have more precisely.



Genome: The full set of genes carried on chromosomes of an individual plant and thus all the genetic material of the plant. It's the "recipe" for the plant to function and is found in every cell of that organism.

Regardless of the plant's "recipe," one key ingredient is essential for success in product development with genomics. It is a germplasm pool. Germplasm is the living substance of the cell that encodes the hereditary properties of the organism and transmits these properties to the next generation. Pioneer Hi-Bred's germplasm pool consists of virtually thousands of parent lines containing genes from diverse global environments. It is expansive, well-documented, valuable and continually improved.

Gene: The basic physical and functional unit of heredity. Genes are composed of DNA.

Through proprietary technologies and accessed technologies from research partners, Pioneer has become gene rich. Pioneer researchers have confirmed and will continue to confirm gene function to develop improved products. Pioneer estimates corn plants contain between 80,000 and 100,000 genes, and Pioneer researchers have identified 80 percent of these genes.

Genetic Code: The ordered sequence of nucleotides, coded in triplets, along the DNA that determines the sequences of amino acids for protein synthesis. Protein synthesis builds the plant's structure and makes the plant function.

With proteomic technologies in-house, Pioneer researchers now look beyond DNA and RNA to truly understand the genetic code. Proteomics adds another dynamic to understanding plant function.

Genomics: The study of all components of genetic material in a chromosome set.

As the earliest entrant in the plant genomics research arena, Pioneer has a solid lead on other companies in the quest to understand corn and soybean genomes. Genomic research is not a linear process or an individual department at Pioneer. Once genes or new knowledge is discovered, Pioneer has the system in place to take advantage of the discovery and quickly develop elite products that will help increase productivity.

Uncovering what genes do

Using genomic tools in seed product development is not a linear process. Efforts don't necessarily move clearly from step one to steps two and three. Genomic knowledge is very powerful. Using this new field of science, researchers can discover a gene or acquire knowledge that could change the type of product delivered to the market.

Expressed Sequence Tag (EST): The tag of an expressed gene derived from the mRNA. ESTs provide a snapshot of the population of genes expressed from a specific tissue, such as a leaf or root. ESTs are useful for identifying landmarks on the genetic map. Researchers use ESTs from many genomic databases to compare information to gain further clues about gene function.

Pioneer researchers have ESTs to 80 percent of the genes in corn. With this information in hand, Pioneer researchers use other tools to confirm function before reincorporating valued genes into elite germplasm for further testing.

Gene expression: The process of turning the genetic code into traits, such as drought tolerance in corn or blue eyes in humans. Some genes are expressed only at certain times during a plant's life and not at others.

Understanding gene expression has been a goal of Pioneer researchers since the company was founded. Until recently, tracking gene expression meant following the plant's growth cycle and recording physical changes as environmental pressures took place. Today, Pioneer researchers are working at the RNA and protein levels to understand gene expression. By understanding gene expression, Pioneer researchers can track economically important genes for inclusion in seed products.

Gene function: The normal or characteristic action of a specific gene.

For more than 75 years, Pioneer researchers have been cataloguing and characterizing the corn plant's functions. This vast database and knowledge gives Pioneer researchers a significant advantage in developing seed products with the new tools of genomics and proteomics research. Once a gene function or knowledge is uncovered, Pioneer researchers can compare this information with information in a database of physical plant characteristics built from evaluations taken in millions of environments across multiple years.

Gene mapping: The location of a gene on a specific chromosome or genetic map location.

The physical location of genes on chromosomes lends more information to determine gene function. Together with DuPont, Pioneer has made significant progress in mapping the corn genome. Researchers can use this to increase breeding efficiencies and provide more precise data on the heritability of certain plant characteristics. From this information, Pioneer researchers develop experimental products for in-field evaluations across varied environments.

Gene sequencing: The determination of base sequences in DNA or RNA molecules that make up specific genes or the order of amino acids in a protein. Collecting sequence information is the basis for genomics research. It is a prerequisite for all attempts to determine gene function.

Pioneer worked through a research agreement with Human Genome Sciences (HGS), Rockville, Maryland, to gain access to high throughput technologies that helped Pioneer collect tags to most genes in corn and develop a robust sequence database. Pioneer continues this work with DuPont. Together, the companies have captured nearly 350,000 corn sequences.

Quantitative Trait Loci (QTL): The position on a chromosome occupied by a particular gene that determines a quantitative trait expression. QTL are identified via statistical procedures that integrate genotype (molecular markers) and phenotype (physical observation) data. QTL are assigned chromosome locations based on the positions of markers on linkage maps.

QTL lend more information to determining gene function. Pioneer and DuPont have gathered significant QTL information on the corn and soybean genomes. QTL and molecular marker data continue to help populate the genomic map for corn and soybeans.

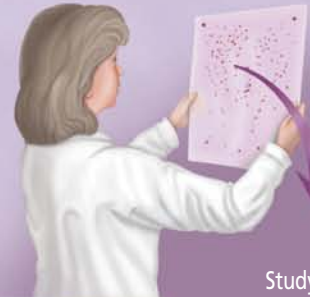
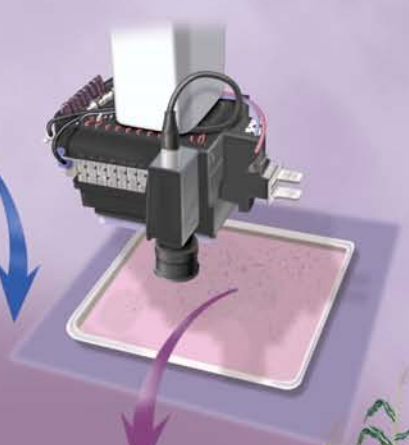
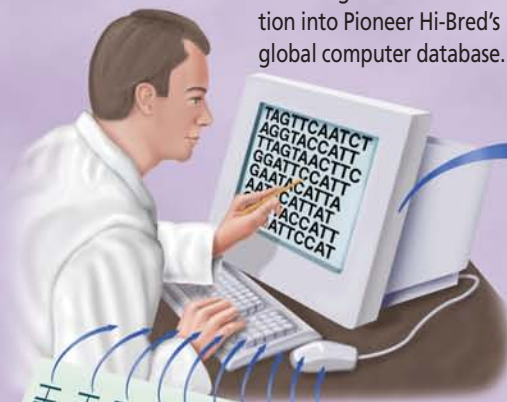
Identify plants with valued traits for study. Collect samples from plants with different performance to research the variation.



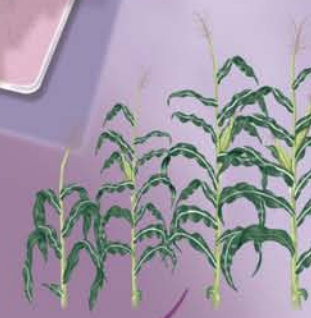
Take multiple samples throughout the growth cycle and from different plants. Prepare for laboratory analysis.



Transfer genetic code information into Pioneer Hi-Bred's global computer database.



Study genetic code using multiple tools (some proprietary to Pioneer) to determine gene function or to develop a better understanding of plant performance. During this process, researchers and geneticists are like detectives looking for multiple clues from various sources to confirm a theory. Researchers are looking for the relative differences of expression between specific genes. To identify which genes deserve further investigation.

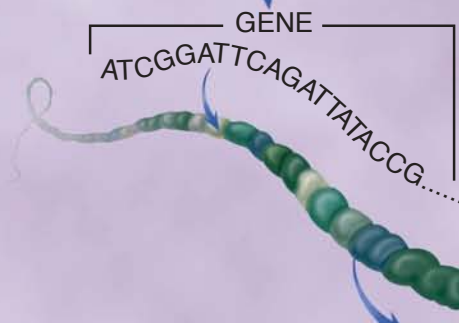


Collect genetic code.



Samples are ground. RNA extracted. Then, complement DNA is synthesized.

Identify valued gene(s) or difference(s) in performance.



Integrate valued gene(s) into elite germplasm through transformation technologies. Verify gene(s) is present. Then, develop products through plant breeding and other technologies. Perform extensive evaluation to confirm performance before commercial product release.

Tools for work at the molecular level

The genomic tools used to discover new drugs and medical therapies in the early 90s were applied to plants in early 1996. Although the subject of research is different, the power for discoveries of genes and knowledge has the same excitement and value.

Bioinformatics: This newly developed field combines information science with biology to accommodate the storage, orientation and indexing of genetic sequencing information. Popular sequence data banks such as GenBank and EMBL have been growing at exponential rates.

Although the term bioinformatics is relatively new, the information management group at Pioneer has been working with computer technologies to track plant expressions and profiles since computers were developed. Today, Pioneer researchers digitally capture and analyze more than 11 million data points during a typical six-to-eight-week harvest season from 2 million research plots. Besides this database of phenotypic and pedigree information, Pioneer has built a tremendous EST database. If EST sequences were laid end to end, they would be equivalent to a DNA molecule about 2 inches long. If a DNA molecule were the diameter of standard copper wire, the EST database would stretch 31 miles.

Contig: Groups of clones representing overlapping regions of a genome.

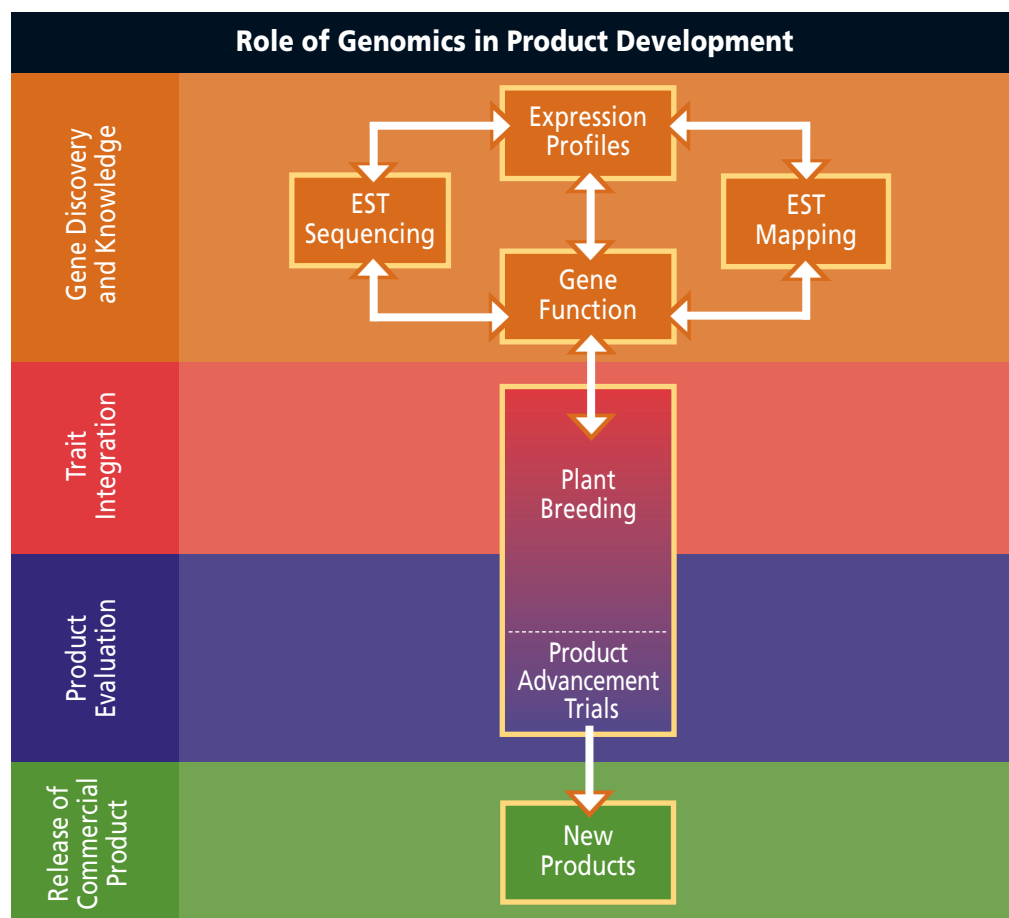
By sampling many parts of the plant at various growth stages to develop the sequencing database, Pioneer has captured a multitude of genomic informa-

tion. With this approach, researchers may capture the same genetic information in many different ways. As more is known within the genomic database, these samples are compared to identify overlapping information. This process helps confirm expression and function of these samples to find like sequences.

DNA Chips: A type of microarray designed to help analyze gene expression. To use DNA chips, researchers must know something about the gene of interest. This technology allows researchers to determine the interaction of thousands of gene combinations at once.

High Throughput: Genomic tools marry biochemistry and computer technologies into a high volume system to study genes at the molecular level. This high volume of analyzed samples is referred to as high throughput.

Just as plant breeding is a numbers game, genomics research is the same. Being able to sequence and identify 100,000 genes within the corn plant involves technologies with high sample throughput. These technologies have greatly impacted genomics research efforts at Pioneer, and allowed the company to reap the benefits to improve plant performance.



Microarrays: A closed system developed to study gene expression. As a closed system, something must be known about the gene of interest in a given condition to use microarray technologies. From a microarray, researchers receive "relative" information, such as a positive or negative correlation to the desired expression.

Proteomics: The study, separation, identification and characterization of proteins in organisms. Proteins provide the basis for plants to function. Proteomics is considered the next frontier of genomics research. Genes ultimately are expressed as proteins, so the application of protein technologies to aid in the discovery of genes is very powerful.

Up to this point, discovery work at Pioneer has been done at the RNA level which doesn't always give researchers the full story. Being able to study proteins will advance research knowledge of plant functions to further Pioneer researchers' abilities to create products with increased productivity. Pioneer recently entered into an agreement with Oxford Glyco-sciences to further develop proteomic technologies.

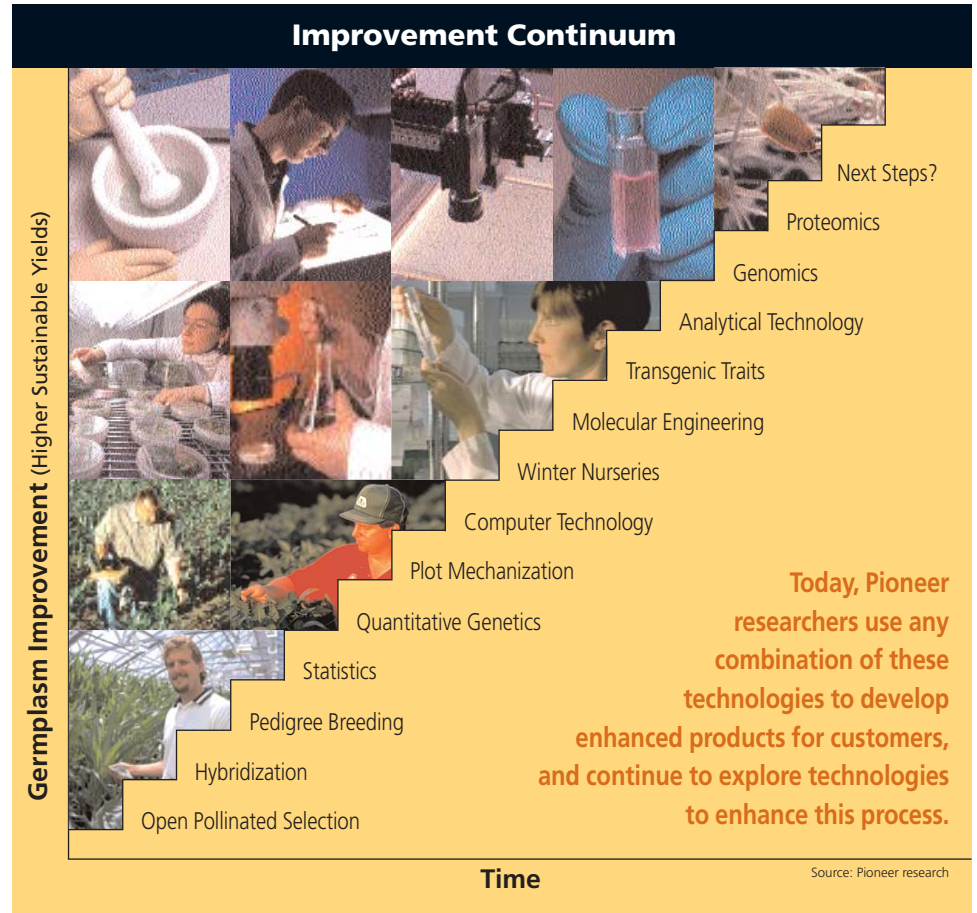
Recombinant DNA: A hybrid DNA produced by joining pieces of DNA from different sources into new combinations. This product is then compared to DNA with a known function or expression.

Robotics: A computer-controlled machine that can be programmed to carry out specific work. Robots often are used in research to

perform minute or repetitive tasks. This allows a significantly higher volume of research work to be completed more precisely.

Pioneer researchers lead the plant genetics industry in the use of robotics in research applications. Automation technologies such as robotics, image analysis and computer data acquisition help

expedite sample collection, handling and processing for analytical testing. It enables Pioneer researchers to increase sample analysis from hundreds of samples per day to thousands and even tens of thousands per day. The ability to analyze greater numbers of samples enables Pioneer to develop better products and to bring them to customers faster.



Notes:

For more information about genomics
or to use the photos or illustrations from
this brochure, please contact:

webeditor@phibred.com



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