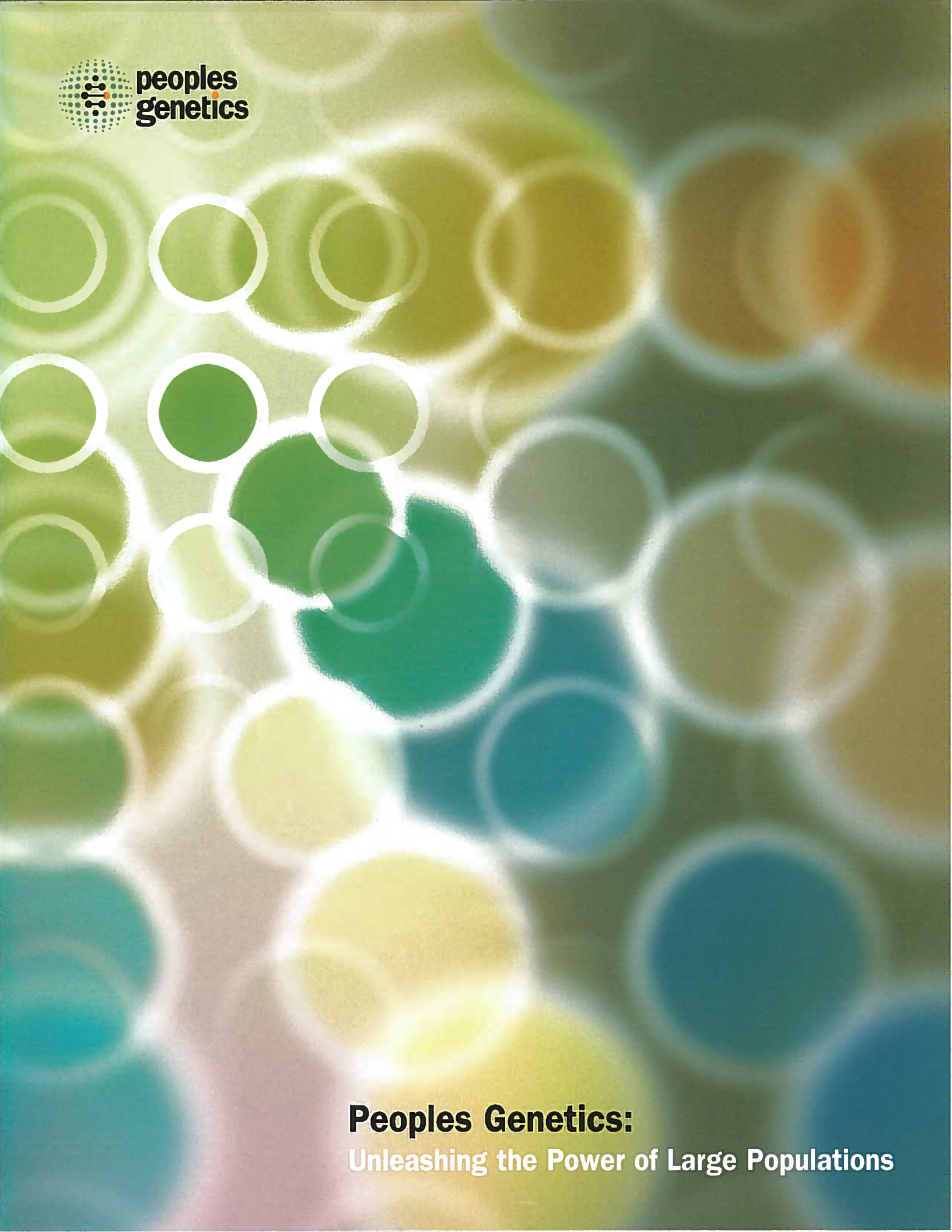
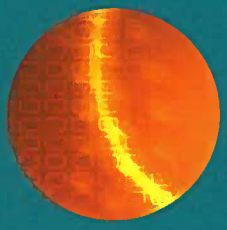
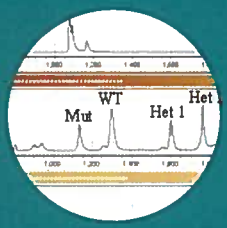

peoples
genetics
Peoples Genetics:
Unleashing the Power of Large Populations

Unleashing the Power of Large Populations

Now, for the first time, genomics researchers can analyze tens of thousands of DNA samples — reliably, quickly, and economically. The result is breakthrough statistical power and scientific insight, at a level never before possible.





Peoples Genetics' Technology Benefits

Discovery, not scoring

- Peoples Genetics does not require *a priori* knowledge of SNPs.
- We simultaneously discover *and* quantify mutations.

Comprehensiveness

- We discover the identity and frequency of virtually all the alleles (down to a detection threshold of 5 X 10^{-4}) for any region scanned.

Statistical power

- Our combination of large sample sizes and a highly sensitive process ensures greater statistical power and less sample variance than alternative approaches.
- Our process enables comparisons of multiple (not just single) alleles, increasing statistical resolution.

Savings and speed

- The Peoples Genetics process for scanning large populations is faster, and far more economical than alternatives — by roughly two orders of magnitude.

Peoples Genetics conducts gene/disease causation experiments of unprecedented scale. We employ a process that allows us to pool and analyze samples containing DNA from up to 100,000 individuals, discovering the identity and frequency of nearly every mutation present in any gene studied. We deliver such data at less cost and in less time than any genomics organization in the world.

For over a decade, science has searched for the genetic causes of common diseases, in hope of designing more effective, customized therapies and diagnostic tools. Researchers now recognize that much larger population studies are needed to test the leading hypotheses of genetic causation. Peoples Genetics makes these large-scale tests feasible for the first time.

The Value of Our Approach

Until now, large-population genetic variation studies simply have not been feasible. The costs of alternative techniques, such as gene sequencing, grow linearly with the size of the sample populations. A single large experiment could cost millions of dollars and take months to complete.

Because Peoples Genetics' technology pools DNA samples, our experiments are virtually insensitive to the size of the population. Therefore, our process, from sample handling to scanning, is far less time-consuming and less expensive than other technologies — in fact, cheaper by roughly two orders of magnitude. This makes it practical, for the first time, to conduct genetic variation studies with tens of thousands of samples.

Large-population studies are essential to making further advances against common diseases, such as cancer and diabetes. Scientists exploring these areas have grown frustrated at the limited success of conventional studies, which typically involve only a few hundred patients and the results of which often cannot be replicated. Increasingly, it is thought that the genetic components of common maladies do not lie in single alleles or even single genes — hypotheses that require large populations to test. For example:

- Genetic risk for common diseases may be distributed over multiple alleles in risk-associated genes (multiallelic causation).
- Any of multiple genes may carry multiple alleles resulting in a disease with a single diagnosis (multigenic causation).
- Mutations in more than one gene, together, may be required to create risk for a disease in a single individual (polygenic causation).
- Environmental factors also may be necessary to influence an individual's risk, even when an independent, essential mutation is present (incomplete penetrance).

To test these hypotheses, researchers ultimately must detect all the alleles of any gene studied and measure fine differences between their frequencies in cases and controls. Only studies involving many thousands of DNA samples can provide the necessary comprehensiveness and statistical power.

Our Technology Platform and Sample Library

Peoples Genetics' technology allows us to analyze DNA samples from a population of up to 100,000 persons and detect all mutations present at a frequency of 1 part per 20,000.

The power of Peoples Genetics' approach begins with the pooling of DNA. Rather than sequencing each DNA sample from a population, we pool the samples and isolate sequences of interest. Our process then creates conditions that separate mutant and non-mutant DNA molecules, identifying specific gene variations present in the pool.

This process utilizes our patented form of capillary electrophoresis, Constant Denaturant Capillary Electrophoresis (CDCE), which we conduct at high-throughput levels. We also employ high-fidelity PCR and other sample preparation procedures.

In addition, Peoples Genetics draws upon its own DNA library, representing a wide range of cohorts. These include disease-affected populations — for diabetes, cancers, hyperlipidemia, and other maladies — as well as a young-adult reference population of 100,000.

Peoples Genetics' technology was developed by its founding scientists, William Thilly, Sc.D., and Barry Karger, Ph.D., in a collaboration between the Massachusetts Institute of Technology and Northeastern University.

A Complement to Current Research

The Peoples Genetics approach complements other genomics technologies by picking up where they leave off. While current techniques work to identify areas of the genome that contain disease-causing alleles, we identify the causal alleles themselves. For gene expression studies, SNP association studies, and other discovery efforts, we act as a "finishing shop," pinpointing mutations of interest within any set of nominated genes and, critically, eliminating false positives.

Biopharmaceutical companies, research institutions, and other organizations can add value to their own studies by contracting with Peoples Genetics. Drawing on our high-throughput infrastructure and extensive sample library, we provide a turnkey solution for clients who require in-depth genetic analysis.

In addition, we can customize our analysis to the level of sensitivity a client requires. Our Deep Intelligence Scan allows the discovery of all alleles at a frequency as low as 1 part per 20,000 and permits researchers to make a clear determination of gene/disease causation. Our Reconnaissance Scan, which targets alleles at or above a 1% frequency, is ideal for researchers who require a cost-effective interrogation of larger areas of the genome. In both scans, we apply our analysis to pools of up to 100,000 samples, delivering unparalleled statistical power.

About Peoples Genetics

Peoples Genetics, Inc., is a privately held genomics company that is discovering the genetic causes of common diseases through experiments of unprecedented scale. The company provides services for biopharmaceutical companies and research institutions, while also pursuing its own genetic discoveries for future development of proprietary molecular diagnostics. Peoples Genetics holds exclusive licenses from the Massachusetts Institute of Technology and Northeastern University, and was founded in November 2000 by Joseph P. Kennedy II, William Thilly, Sc.D., and Barry Karger, Ph.D. The company is headquartered in Woburn, Massachusetts.

For more information about Peoples Genetics and its services, please contact us by phone at (781) 933-6068 or by e-mail at **info@peoplesgenetics.com.**

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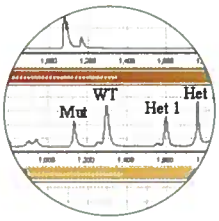
Services Available From Peoples Genetics

Peoples Genetics offers its deep genetic analysis on a contract basis to biopharmaceutical companies, academic research institutions, and other members of the life sciences community. Clients find that our approach is a logical complement to other genetic variation discovery techniques. Our massive, pooled experiments make definitive determinations about candidate genes that have been implicated through methods such as family studies, mRNA expression analysis, and work with model systems. In effect, Peoples Genetics functions as a "finishing shop," pinpointing causal mutations within any set of candidate genes.



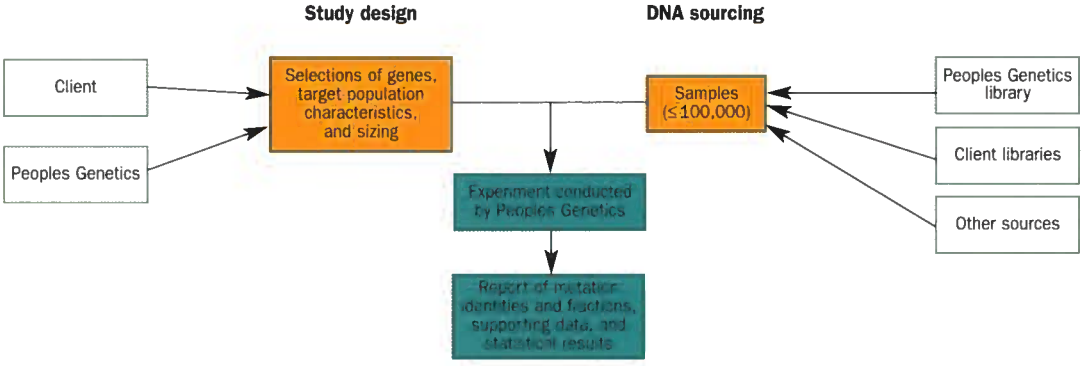
A Turnkey Solution

Drawing on our high-throughput infrastructure and a library of more than 100,000 DNA samples, we provide a complete, turnkey solution for candidate gene experimentation.

We work closely with our clients to design each study, selecting genes of interest and populations of appropriate characteristics and size. We source DNA from our own library and also coordinate with clients and third parties to obtain necessary samples. Once the samples are in place, a typical experiment (scanning 1kb of sequence in two populations of up to 100,000 persons each) takes less than six weeks. At that point, the client receives:

- Mutation identities and frequencies in each population
- Statistical results
- Data confirming process validity

Throughout its work, Peoples Genetics employs processes and technology to ensure the security of its clients' proprietary data.



Services to Meet Each Research Need

Peoples Genetics can customize its analysis to the level of sensitivity a client requires. Our **Deep Intelligence Scan** allows the discovery of all alleles at a frequency as low as 1 part per 20,000 and permits researchers to make a clear determination of gene/disease causation. Our **Reconnaissance Scan,** which targets alleles at or above a 1% frequency, is ideal for researchers who require a cost-effective interrogation of larger areas of the genome. In both scans, we apply our analysis to pools of up to 100,000 samples, delivering unparalleled statistical power.

Different scans, different needs	**Deep Intelligence Scan** (= 0.005% detection threshold)	**Reconnaissance Scan** (= 1.0% detection threshold)
Benefits	Discovery of all alleles; statistical comparison of multiple alleles	Cost-effective analysis of low-effect, higher-frequency alleles
Applications	Definitive causation testing for candidate genes selected through other techniques	Retesting of prior small-scale SNP associations in much larger populations
Constraints	Economics limit these studies to smaller regions of the genome	Lower-frequency alleles left undiscovered

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Our Sample Library: A Storehouse for Knowledge

Clients of Peoples Genetics can draw upon our internal library of more than 100,000 DNA samples. The library represents a wide range of cohorts, including disease-affected populations — such as those with diabetes, cancers, hyperlipidemia, and other maladies — as well as a young-adult (<35 years) reference population.

Our reference population represents a rich cross-section of the United States, drawing from populations in New England, the Mid-Atlantic, the Midwest, and the Pacific Northwest. The ethnic composition of our reference population is within ± 1% of the distribution determined by the 2000 U.S. Census. The size and diversity of this cohort enable the discovery and quantification of molecular diagnostic markers that are useful for the heterogeneous U.S. population.

Peoples Genetics will work with clients to customize sample pools from other populations, as needed. We have established relationships with several commercial diagnostic laboratories to obtain whole blood for use in our studies. And because we analyze such large populations, we do not require the costly, highly detailed phenotypic characterizations of samples that other technologies make necessary.

The power of the Peoples Genetics reference cohort	**Large, conventional study example**	**Study with Peoples Genetics reference cohort**
Afflicted population size	1,000 alleles	1,000 alleles
Tested allele frequency: afflicted population	0.8% (8 copies observed)	0.8% (8 copies observed)
Control population size	1,000 alleles	**200,000 alleles**
Tested allele frequency: control population	0.2% (2 copies observed)	0.2% **(400 copies observed)**
Results	Difference is **not** statistically **significant**	Difference is statistically **significant**

Cost-Efficiency

Since our technology pools DNA samples, our experiments are virtually insensitive to population size. So our services are far less expensive than others — in fact, cheaper by roughly two orders of magnitude. The exact fees for an experiment are based on the size of the sequences examined, the sourcing of DNA samples, the level of sensitivity required, and the number of pools scanned.

Here's what our cost-efficiency means:

- To study a population of 10,000 afflicted individuals vs. a reference population of 100,000, cost for our **Deep Intelligence Scan** typically would represent **only 1% of the cost of gene sequencing.**
- To analyze SNPs in populations of these sizes, our **Reconnaissance Scan** would cost only **5% of what SNP scoring** would cost, making the Peoples Genetics scan **20 times more cost-efficient.**

For More Information

To learn more about our services, please contact Peoples Genetics by phone at (781) 933-6068 or by e-mail at **info@peoplesgenetics.com**

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The Peoples Genetics Technology Platform

Peoples Genetics is able to analyze pooled DNA with up to 100,000 samples per pool by employing a highly sensitive process that separates, identifies, and quantifies every mutation present at a frequency as low as 1 part per 20,000.

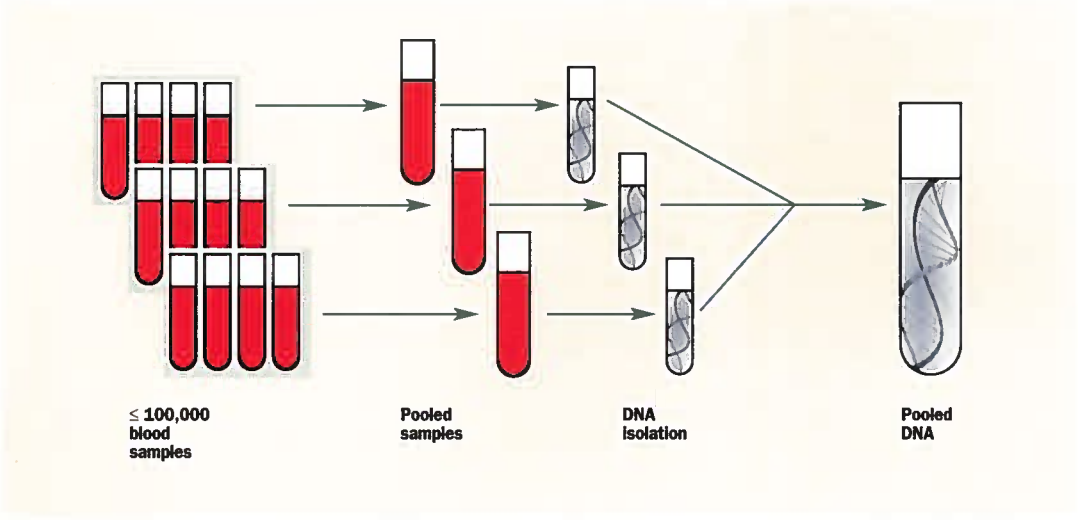
This process was developed by our founding scientists, William Thilly, Sc.D., and Barry Karger, Ph.D., in a collaboration between the Massachusetts Institute of Technology and Northeastern University. Our core technology is covered under U.S. Patent #5,633,129, as well as additional patents and patent applications now pending. The process involves five stages, which are outlined here.

1. Isolation of Genomic DNA

Typically, Peoples Genetics starts with up to 100,000 whole blood samples and pools these in groups of 96. DNA is isolated from these small pools using standard chemistry. After we perform quality control steps, we create larger pools of DNA.

Why this is important:

- *Pooling allows Peoples Genetics to isolate DNA with tremendous cost-efficiency.*

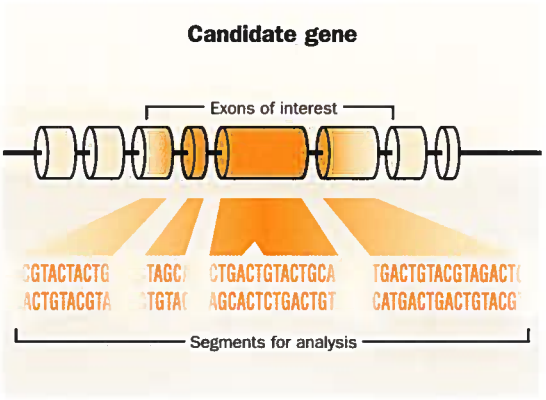


2. Isolation of Target Sequences

We break down the selected genes into a series of target sequences (average length of 80bp) using a computational assessment of their temperature melting domains. The genomic DNA is cut with restriction enzymes to isolate exons, and then we hybridize two 5' biotinylated probes, complementary to the Watson and Crick strands of each target fragment. After hybridization, we isolate the hybrid molecules from the genomic DNA on streptavidin-coated beads. We wash the captured fragments and elute them by heating.

Why this is important:

- *Analysis of the separate Watson and Crick strands provides a powerful control for DNA artefacts.*
- *Peoples Genetics' approach to sequence isolation minimizes DNA stock depletion.*



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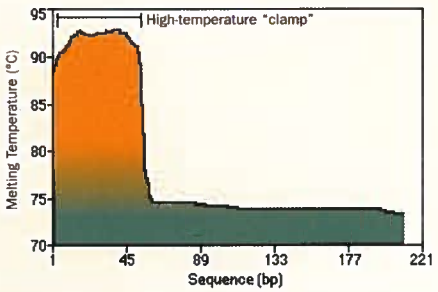
3. High-Fidelity PCR

We amplify target sequences through PCR, using a high-fidelity thermostable DNA polymerase and fluorescently labeled primers. Typically, a GC-rich clamp is used to create an artificial, high-melting domain next to each target, making it suitable for separation with our proprietary form of capillary electrophoresis. Before conducting PCR, we also introduce artificial mutants into the target-enriched sample to create internal standards for accurate quantification of mutant alleles.

Melting map



Why this is important:

- *Our process minimizes polymerase errors by limiting the number of doublings and using high-fidelity enzymes.*
- *Polymerase controls and the comparison of the Watson and Crick strands allow us to characterize any process-induced mutants.*

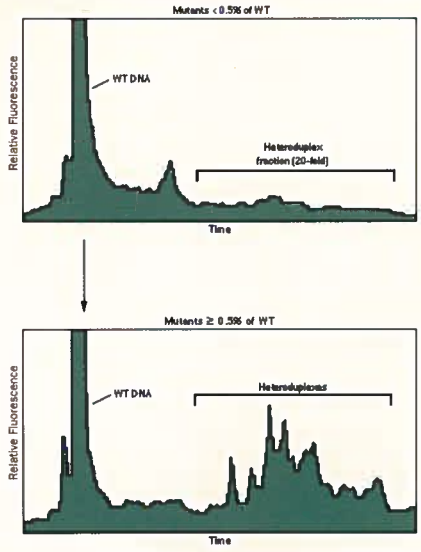
The Right Instrument for Sensitivity, Precision, and Throughput

To conduct CDCE, Peoples Genetics has developed a unique instrument, the High-Throughput Mutational Spectrometer (HTMS). This automated, multicapillary instrument features temperature control to ±0.01°C, fraction collection integrated with replaceable matrix, and an optical system with a 1 pM fluorescein detection limit and a dynamic range of 3 Decades. The HTMS provides our process with the high resolution necessary to detect mutants at a frequency of 1 part per 20,000 — and at a discovery rate of more than 1 million "person-genes" per year.

4. Separation Through CDCE

Peoples Genetics then separates wild-type homoduplexes and wild-type/mutant heteroduplexes in the PCR product based on their unique melting temperatures. To accomplish this, we perform Constant Denaturant Capillary Electrophoresis (CDCE). This process utilizes our High-Throughput Mutational Spectrometer (HTMS) and its laser-induced fluorescence detection system. Following CDCE, the heteroduplex fraction, enriched in mutant sequences, is gathered in our agarose gel fraction collector. We then amplify the fraction through additional high-fidelity PCR; mutants can be enriched further through another round of CDCE and heteroduplex collection.

Collecting heteroduplex fraction and PCR



Why this is important:

- *With high resolution, CDCE separates double-stranded DNA — differing by even a single base pair — because of the DNA cooperative melting equilibrium.*
- *The addition to a DNA fragment of a GC-rich clamp — an artificial high-melting domain — permits the analysis of almost any sequence.*

5. Identification and Quantification of Mutants

To create mutant homoduplexes, the fraction containing mutant heteroduplexes is subjected to a limited number of PCR cycles. We separate the mutant homoduplexes through CDCE, and individual mutants are isolated and sequenced. Typically, Peoples Genetics performs two or three rounds of CDCE collection and high-fidelity PCR to purify a mutant for sequencing. We determine the original mutant fraction of each mutant in the pooled sample by comparing the area under the mutant peak with those of internal standards.

Why this is important:

- *Comparison of mutant peak areas with those of internal standards permits precise quantification of allele frequencies.*

For More Information

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