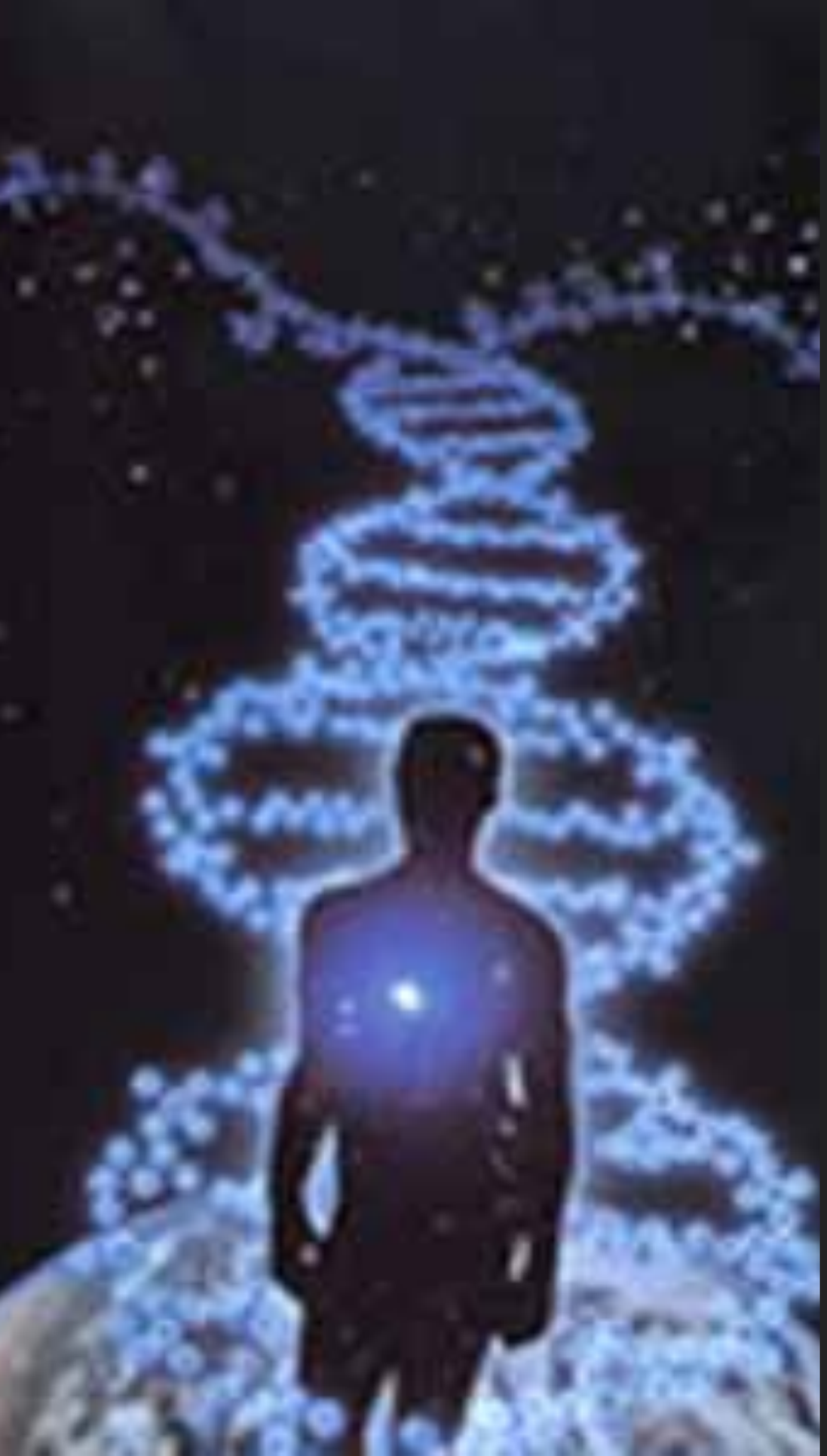
S.S. DISCOVERIES

"LET'S MAP A NEW PATH TO HEALTH"



ABOUT S.S. DISCOVERIES

S.S. Discoveries was founded by Madison Tammen and Rachael Murdoch in the hopes that one day the Dideoxy chain termination method or Sanger Sequencing will help millions of families and people around the world by discovering cures to tons of disorders.

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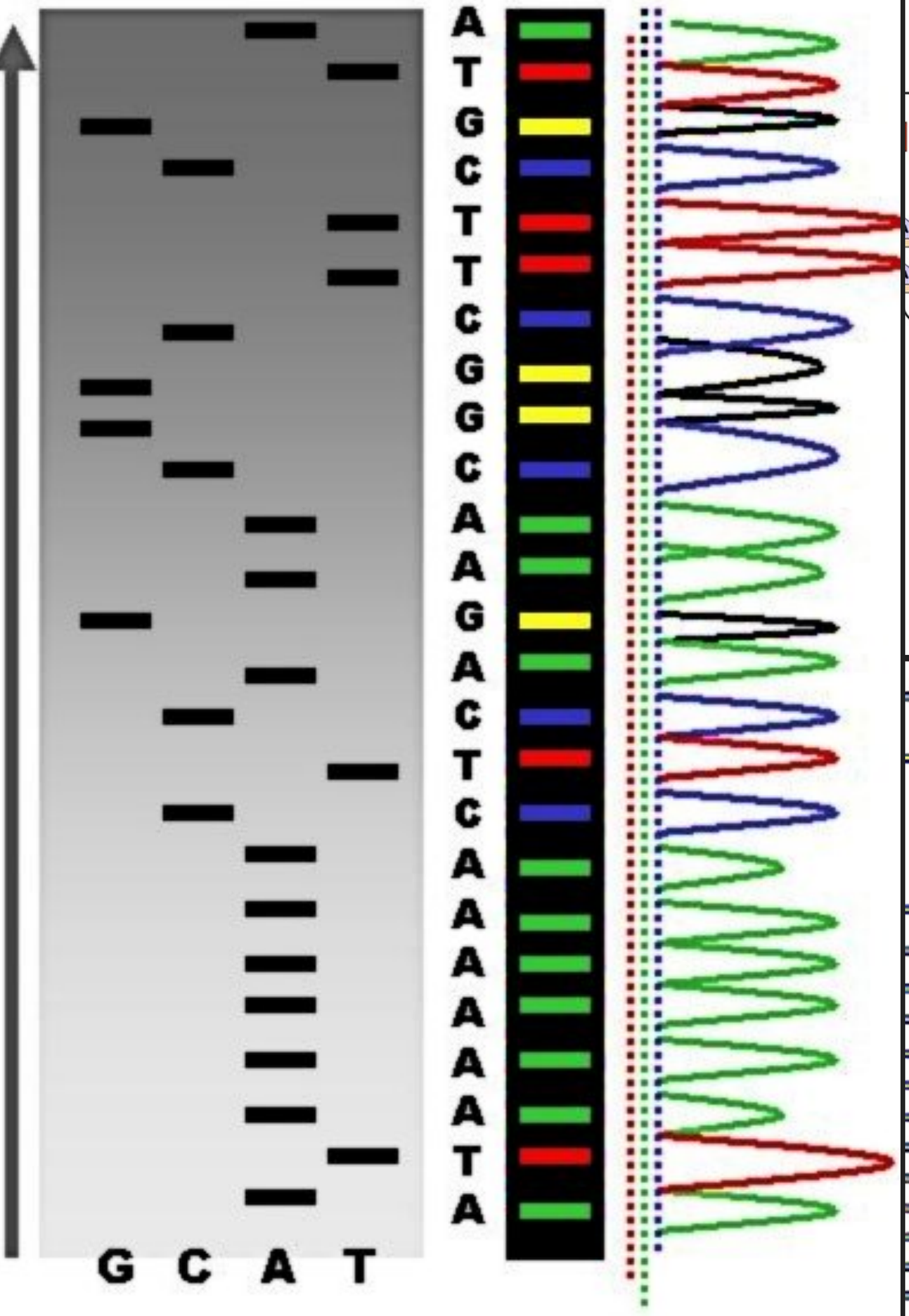
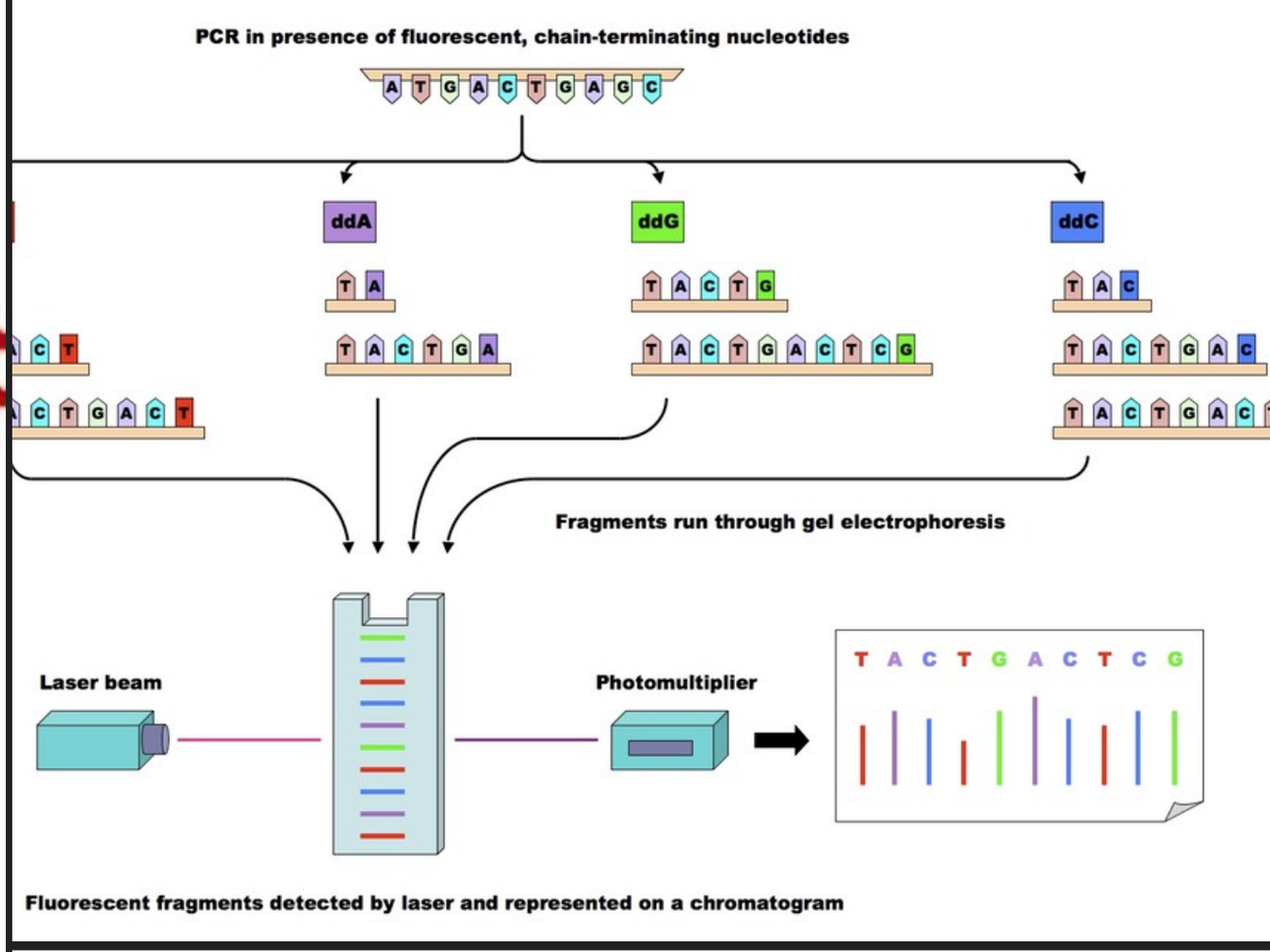
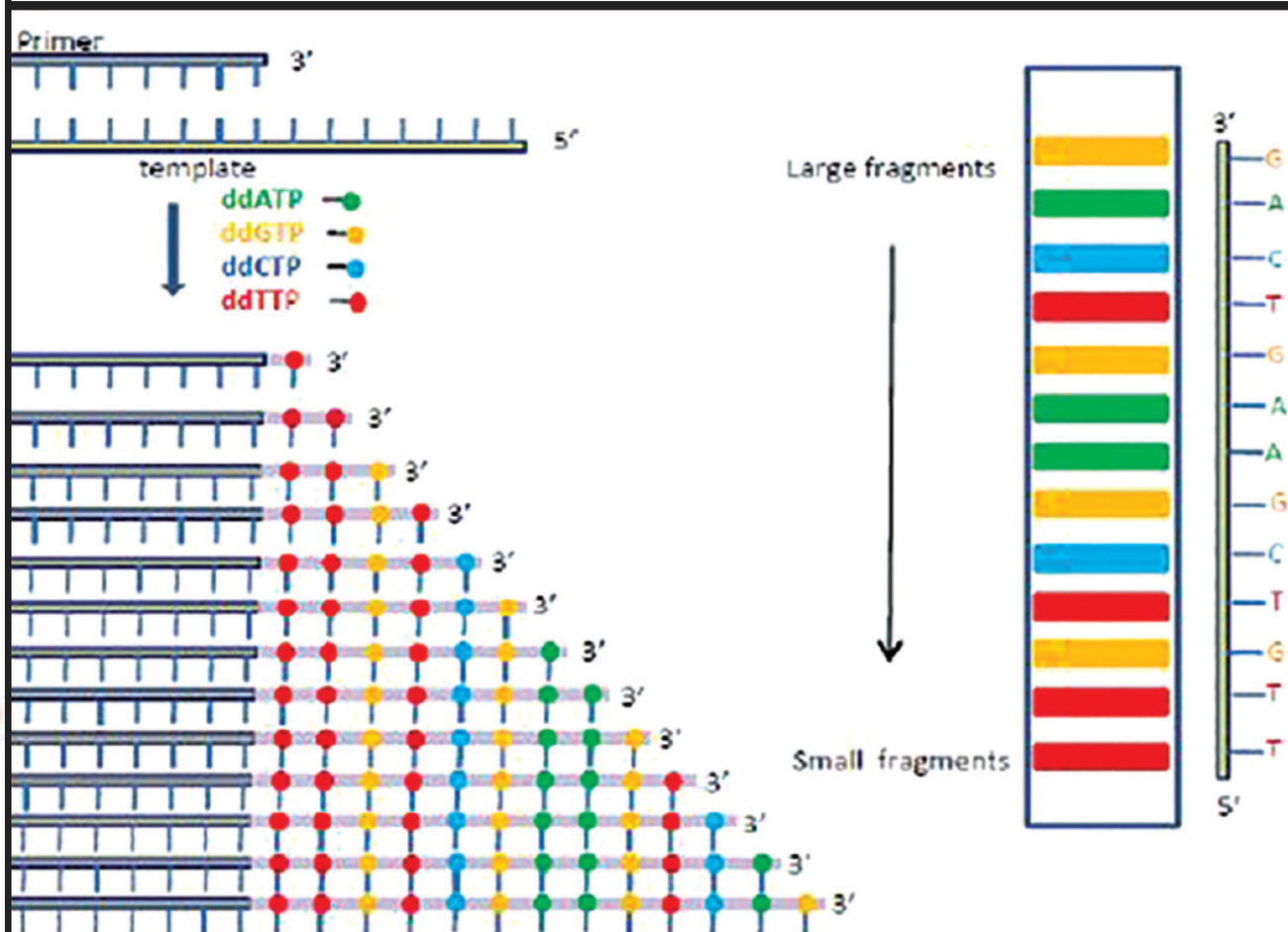
VIDEO CLIP


A T G C T T C G G C A G
A C T C A A A A A A T A
G C A T


PCR in presence of fluorescent, chain-terminating nucleotides
A T G A C T G A G C
ddA
ddG
ddC
Fragments run through gel electrophoresis
Laser beam
Photomultiplier
T A C T G A C T C G
Fluorescent fragments detected by laser and represented on a chromatogram


Primer
3′
5′
template
ddATP
ddGTP
ddCTP
ddTTP
Large fragments
Small fragments
3′
5′

HOW OUR TECHNOLOGY WORKS

- Dideoxy Chain termination is a method of DNA sequencing, or determining the order of nucleotides in a gene or DNA molecules.
- First, the DNA is denatured by heat into two single-stranded structures: the template strand and the complimentary strand.
- The template strand is copied through a Polymerase chain reaction and the complimentary strands are discarded. A primer is attached to one end of each DNA strand to stabilize it and allow nucleotides to be added later.
- The template strands are divided into four reaction mixtures containing DNA polymerase, free nucleotides, and one of four modified nucleotides: Dideoxy ATP (adenine), GTP (guanine), CTP (cytosine), and TTP (thymine).
- The reaction mixtures sit and the modified nucleotides bring out patterns of their specific nucleotide.
- A sample from each reaction mixture is centrifuged out and put into a gel electrophoresis.
- The combined results of the gel electrophoresis can tell us the sequence of bases in the original strand of DNA.

HISTORY

- Began in the 1970s
- Originally developed by Frederick Sanger in 1975
- In 1990 it began the human genome project using Sanger Sequencing
- In 2003 the human genome project was completed

HUMAN GENOME PROJECT

- An international scientific research project with those of determining the sequence of chemical base pairs which make up human DNA and identifying and mapping all of the genes of the human genome
- Use the Sanger Sequencing Method
- Lead to the discoveries in health
- Scientists can now compare DNA to ancestors

CURRENT USES

- The Human Genome Project is an effort to map out every gene that makes up human DNA.
- It started in 1990 and was completed in 2003.
- The Human Genome Project used the Dideoxy Chain termination method as one of the ways to map out the DNA sequences.
- The project completely mapped out the human genome , opening many more medical and genetic research opportunities.
- This method could be applied to map out the genome of any species.

CONS OF SANGER SEQUENCING

- Poor quality in the first 15-40 bases of the sequence due to primer binding and deteriorating quality of sequencing traces after 700-900 bases
- (Software provides an estimate of quality to aid)
- In cases where DNA fragments are cloned before sequencing, the resulting sequence may contain parts of the cloning vector
- Can only directly sequence about 300-1000 nucleotides in a single reaction. The main obstacle to sequencing DNA fragments above this size limit is insufficient power of separation for resolving large DNA fragments that differ in length by only 1 nucleotide
- There are occasional artifacts probably due to contaminant fragments
- Band pile ups can occur due to loop formation under the gel conditions, and is usually depicted as numerous bands in the same position or very close together on

PROS OF SANGER SEQUENCING

- Microfluidic Sanger sequencing fixes the majority of cons
- No preliminary extension is required therefore avoiding incubation and purification
- Requires commercially available DNA polymerase
- The results are better and with fewer artifacts and a larger sequence can be read
- Intermediate nucleotides in runs show up on bands which avoids estimating the number of nucleotides in each runs . the first nucleotide is usually the strongest and in one can estimate the number of nucleotides by the strength and width of each band
- Works on small scales and large scales due to better incorporation of 32p and longer incubation period resulting in longer extension
- 15-200 nucleotides can be read from primer site and can be determined with accuracy. Up to 300 is possible
- Resulted in the human genome project which lead to many genetic discoveries

BIOETHICAL ISSUES

- Some people argue that messing with DNA is against religious beliefs
- The costs of doing this method to test DNA can be expensive
- Some people argue that the outcome is not worth the trouble

CASE STUDY

- The Dideoxy chain termination method is a great method of sequencing DNA, but it doesn't have direct benefits of its own.
- It has brought new treatments by powering the development of personalized medicine, which is based on an individual's genome.
- In 2010 a man named Victor was diagnosed with with lung cancer and he was not able to undergo chemotherapy treatment. It was found that he had an ALK mutation that caused the disease.
- His gene was sequenced and a medicine was personalized to his genome. Eventually, the treatment shrunk the tumor.