IJBPAS, August, Special Issue, 2021, 10(8): 236-252

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

DNA SEQUENCING AND ITS BENEFITS - A REVIEW

M.VIGNESH[1], M.P.BRUNDHA[2*] AND ARCHANA SANTHANAM[3]

1: Department of Pathology, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai-77

2: Associate Professor, Department of Pathology, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Saveetha University,Chennai-77.

3: Assistant Professor, Department of Oral Pathology, Saveetha Dental College, Saveetha Institute Of Medical and Technical Sciences, Saveetha University, Chennai-77, India.

***Corresponding Author: E Mail: Dr. M.P.Brundha: brundha.sdc@saveetha.com**

Received 19th March 2021; Revised 20th April. 2021; Accepted 19th May 2021; Available online 1st Aug. 2021

https://doi.org/10.31032/IJBPAS/2021/10.8.1017

ABSTRACT

The speed, accuracy, efficiency, and cost-effectiveness of DNA sequencing have been improving continuously since the initial derivation of the technique in the mid-1970s.With the advent of massively parallel sequencing technologies, DNA sequencing costs have been dramatically reduced. No longer is it unimaginable to sequence hundreds or even thousands of genes in a single individual with a suspected genetic disease or complex disease predisposition. Along with the benefits offered by these technologies comea number of challenges that must be addressed before wide-scale sequencing becomes accepted medical

practice. Molecular diagnosticians will need to become comfortable with, and gain confidence in, these new platforms, which are based on radically different technologies compared to the standard DNA sequencers in routine use today, but despite these obstacles, and as a direct result of the promises these sequencing advances present, it will likely not be long before next-generation sequencing begins to make an impact in molecular medicine. The aim and importance

of this review is to provide DNA sequencing and its benefits and its recent advances to prevent and diagnose the defective gene and correct it.

Keywords: DNA Sequencing; Sanger; Molecular analysis; nucleotide base pairs

INTRODUCTION

DNA sequencing is defined as the sequencing of the specific site of the DNA to identify the particular sequence of the nucleotides or a defective sequence and the microbiology technique and determine precise order of nucleotides bases- adenine, guanine, cytosine, thymine in given template or fragment of DNA. Further advances in this technology will ensure the proliferation of new applications such as comparative sequencing and stimulate a paradigm shift in biology **[1]**. Technological advances like DNA sequencing over past 20yrs given rise to science of genomics enabled critical advances in other fields of epidemiology and forensics evolutionary biology and medical diagnostics **[2]**. Indeed, technical advances in sequencing have been compounding at such a pace that keeping up may be difficult even for those well-versed in molecular biology, let alone those who are the more clinically based end users of the technology. The incidence and prevalence of diabetes have doubled over the past two decades Genome-wide Association (GWA) studies test hundreds of thousands or even millions of common (Minor Allele Frequency [MAF] >5%) and low-frequency (MAF 1–5%) variants across both protein coding (exonic) and noncoding (intronic) regions of the genome. Large GWA studies have identified more than 50 genetic loci associated with various glycemic traits and at least 90 loci associated with type 2 diabetes in which DNA sequencing thriving to get the solution for the betterment of the general population for undergoing easy dental procedures and medical procedures without worrying about the diabetes **[3]**.

A 2015 study looked at mental illnesses and twins and found that the RBFOX1 gene may make someone more likely to develop generalized anxiety disorder by the way of DNA sequencing it can be corrected to undergo certain dental and medical procedures **[4]** and the DNA sequencing helps the abnormal people to thrive to undergo any checkup and procedures without worrying about the outcome and the genetic traits of the chia seeds know as ssp gene which gives the benefit to the humans is explored using DNA sequencing **[5]** and even the specific gene in probiotics had been identified using DNA sequencing which

helps to people in several ways **[6]** and the lunar effects on humans may cause some mutation changes which can be detected using DNA sequencing **[7]**. The ORMDL3 gene which is responsible for asthma and wheezing trait in humans are even detected using DNA sequencing **[8]** and some of the fluid spill can also be used for DNA sequencing **[8]** and heart rate and respiratory rate is determined by certain genetic traits which can also be determined by DNA sequencing **[9]**. Single nucleotide polymorphisms (snps) are responsible for the hypersensitive patients but full potential of the **[10]** gene expressions responsible for it is not known. The most common cause of hereditary breast cancer is an inherited mutation in the BRCA1 or BRCA2 gene which can be precisely detected using DNA sequencing **[11]**. The gene called TCF7L2, which affects insulin secretion and glucose production. ABCC8, which helps regulate insulin responsible for diabetes mellitus **[12]**. This review is intended as a current snapshot of the recent advances in gene sequencing with emphasis on which of the available "next-generation" technologies are most amenable and appropriate for clinical diagnostic use and it also helps in some histopathology **[13]**. GRS is associated with haemoglobin concentrations across the physiological range, including at haemoglobin levels that predict adverse outcomes in pregnancy, cardiovascular and neurologic disease, in addition to mortality in the elderly **[14]** and DNA sequencing also help to detect dental pulp stem cells **[15, 16]**.

MATERIALS AND METHODS

Relevant articles are searched in the database like PMC database, medline, embrace, cohrance library using keywords and selected around 15 articles. Inclusion criteria selected for the review are molecular pathological studies and genetic disorders related studies. Exclusion criteria selected for this review are other than molecular pathological investigations.

DNA SEQUENCE INDICATIONS:

Next Generation Sequencing (NGS) made a possible for better understanding of genetic diseases and became a significant technological advance in the practice of diagnostic and clinical medicine **[16]** Ngs allows the analysis of multiple regions of the genome in one single reaction and has been shown to be a cost-effective and efficient tool in investigating patients with genetic diseases. The genetic material produced by Ngs offers significant benefits in clinical practice including accurate identification of the disease biomarkers, detecting inherited

disorders and identification of genes that can help predict responses to therapies **[14]**.

SAMPLE COLLECTION FROM DNA SEQUENCING:

Samples like saliva, blood, tissue culture cells are most commonly used for DNA sequencing.Other than these according to the illness the samples with any nucleated cells including inflammatory cells **[17]** may be collected and processed. Other cases of collection may also benefit from providing auxiliary or supervised samples with proper personal protective equipment **[18]**. Saliva is a proven method for easily collecting large amounts of DNA in case of lesions in the oral cavity.

Accessible donation maximizes recruitment:

For a large sample, hiring is important. The easiest way to get great prices is to collect where your donors are - at a clinic, in a remote area of the garden, at an event or in the comfort of their home. The sample collection process must be optimized to reach your number of contributors in their preparation. For example, sample resources may require custom adjustments (e.g. family and individual collection, child vs. adult), assembly, follow-up, distribution and / or storage capacity. To avoid the cost of a housekeeping service, consider sending a company to handle these meeting requirements.

PROCEDURES FOR DNA SEQUENCING:

SYSTEMS REQUIREMENTS FOR SEARCHING DNA:

Since there are many sequences such as sanger sequences in our viewing article we will tackle next-generation sequencing methods and their processes

The next generation sequences:

The next-generation sequencing platform is different from the Sanger process or the DNA sequencing method. Overall, it proposes millions of copies of a particular piece in a way that is highly compatible and "readable" for computational analysis.

The NGS process **(Figure 1)** is a bit complicated, However, it can be divided into 4 distinct steps:

1) Preparing for the library, 2) Collection generation, 3) DNA sequences and 4) Data analysis

1. Preparing for the library:

The preparation of the library is a combination of two response modes, clips and a backpack. CDNA fragmentation or fragmentation of DNA is accomplished by grinding the margin. Subsequently, small pieces of DNA are wrapped in the known DNA sequence.Known DNA sequences are

called adapters and the process is called adapter ligation. When the adapters are used, a small library of DNA is extracted. The fragmentary DNA fragments are washed by the dishwasher, the process of preparing the library is called tagmentation.

2. Collection generation:

A short oligonucleotide sequence is inserted into the mutant body corresponding to our adapter sequence. When the library of our fragmented DNA is loaded into the cell, it is bound by weak oligos in the solid phase and by the delivery of the bridge, the emergence of a set of DNA sequences. Here, at the extension of the bridge, the DNA fragments bend and bind the next oligo that forms the bridge. The primer binds to this DNA sequence and multiplies directly.

3. Sequencing:

As the polymerase adds the nucleotide into the bridge amplification **(Figure 2)**, the amplification signals are recorded each time. This will generate multiple sequencing databases for the DNA sequence.

4. Data analysis:

The read generated by the sequencing can be aligned to the reference genome sequence and by doing this we can identify any addition, deletion or variation into the sequence.

ADVANTAGES:

High standard gold sensitivity is a complete sequence with many sensitivity and provides% wild-type DNA regeneration; works with a split DVD and high sensitivity used for medical tests of oncogene mutations in CRC (ColoRectal Cancer) and NSCLC (Non–Small-Cell Lung Cancer) and high sensitivity provides a percentage of DNA-type mutations.

DISADVANTAGES

A tracking platform is a computer-based supplement that relies on data processing. In that case, a large, high-speed supercomputer is needed. Also, several sequences such as tandem duplication, DNA replication, sequenced species, and other replication regions cannot be properly studied. The chance of errors in the analysis of the original sample may result in significant economic losses, the latter being the two major disadvantages of DNA sequencing methods.

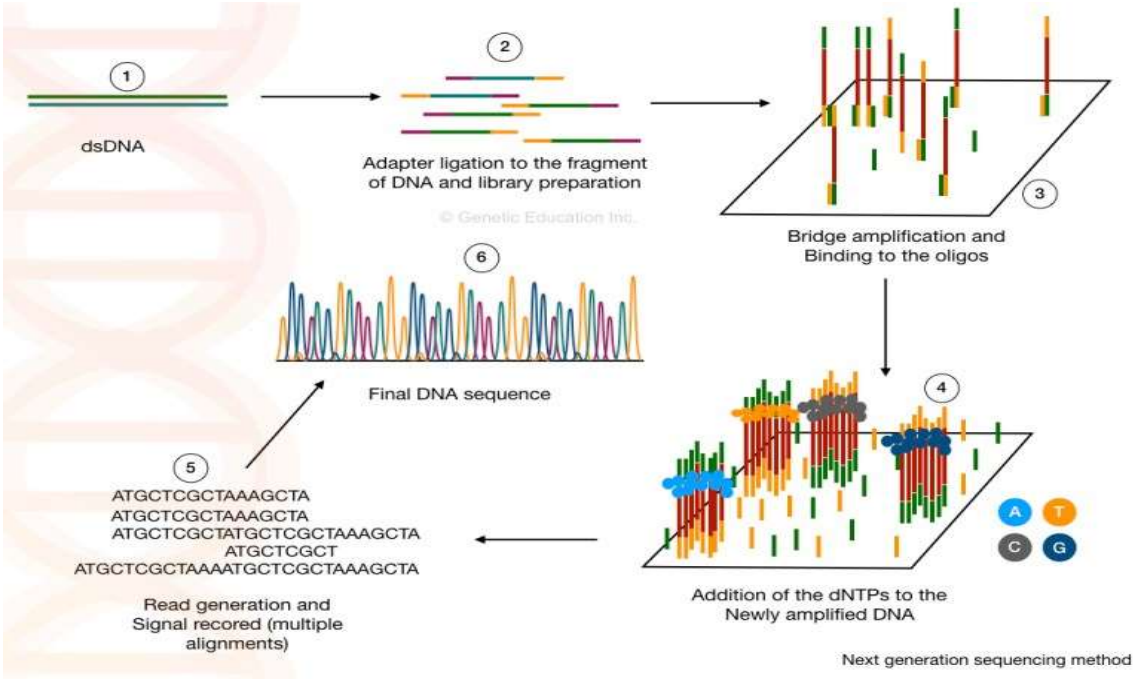

Figure 1: Schematic Diagram Showing Next Generation Sequencing Methods [19]

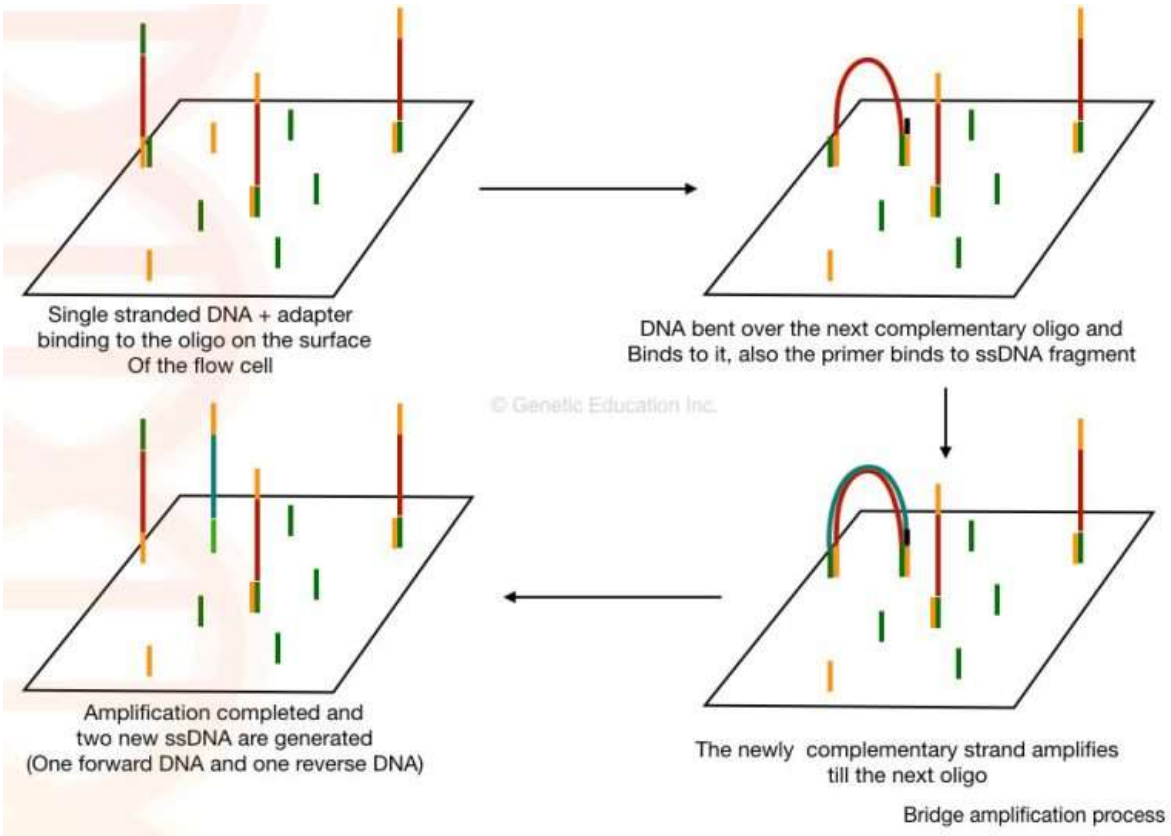

Figure 2: Procedure for Bridge Amplification [20]

DNA SEQUENCING IN DIFFERENT CANCER CELLS:

Cases found in breast cancer are associated with mutations in two genes: BRCA1 (BReast CAncer gene one) and BRCA2 (BReast CAncer second gene). Everyone has Brca1 and Brca2 variants. The function of the BRCA (BReast CAncer gene) species is to repair cell damage and to maintain breast, ovarian, and other normal growth cells. But if these genes contain mutations that are passed on from generation to generation, the types of event are less common and the breast, ovarian, and other cancer risks increase. Brca1 and Brca2 mutations can occur in up to 10% of all breast cancers, or 1 in 10 diseases. Having a Brca1 or Brca2 mutation does not mean you will get breast cancer. Researchers are learning that some mutations in chromosomes - called SNPs (single nucleotide polymorphisms) - can be linked to increased risk of breast cancer in women with Brca1 mutation and women who have not inherited a mutation in breast cancer. Women diagnosed with breast cancer and having a Brca1 or Brca2 mutation often have a family history of breast cancer, ovary cancer, and other cancers. However, the vast majority of breast cancer survivors have not inherited genes linked to breast cancer and have no family history of the disease **[19]**.

HEMATOLOGICAL TEST:

Our understanding of the predictive or prognostic value of various genetic parameters is changing rapidly. The collection of data on the long-term outcome of a large number of patients in the future will help us to confirm new genetic abnormalities. As of today, most genetic tests are being used to determine disease severity **[21]** in a patient who has been given a hematologic malady. In the future the genetic information will be included in treatment decisions on a regular basis and that will be another step towards personalized medicine in the modern era. A wide range of available markers including, clinical, radiological, cytochemical, immunophenotypic and genetic alterations are available in hematologic neoplasms. In this review, we focus only on the currently available genetic tests in clinical practice and their interpretation **[20]**. There are many types of hematological diseases that come out to see acute lymphoblastic leukemia.

ACUTE LYMPHOBLASTIC LEUKEMIA (ALL):

In every case the karyotype has been ordered to detect the level of ploidy in leukemic cells as it is now known that hyperdiploidy carries better prognosis than hypodiploid. Translocation t (12; 21) (p13: q22) is the

most common genetic malignancy in cases of B cell acute lymphoblastic leukemia (All). It is seen in about 25% of cases. This transfer leads to the integration of the 5 ′ gene TEL (ETV6) region, to the AML1 (Acute Myeloid Leukemia 1 Protein) (RUNX1(Runt-related transcription factor 1) locus and is associated with positive prognosis. In addition to providing important details of a particular migration they are considered as predictive markers. Translocation t (12; 21) involving TEL (translocation-Ets-leukemia virus) / Aml1 variants is associated with a favorable outcome and these patients show high cellular sensitivity to L asparaginase **[22]**. Similarly patients with t (1; 19) have a better reaction with high-dose methotrexate containing a regimen. However conventional karyotyping has limitations that it cannot detect many transfers everywhere because they are cryptic in nature and therefore FISH is the best recruitment tool in cytogenetic research for ALL (Acute Lymphoblastic Leukemia) patients.

Recent advances in the field of ALL using left-sided disease MRD (Minimal Residual Disease) after cognitive therapy and treatment planning. MRD is defined by leukosis cells that remain following the achievement of complete hematologic remission, but are subject to detection limits by microscopic examination of blood and bone marrow. The diagnosis of the remaining disease is usually based on the molecular or physical-phenotypic markers that are present in the malignant cells. Various techniques can be used to detect MRD, including immunophenotyping, cytogenetics, FISH, Southern blotting, spectral karyotyping, PCR(Polymerase Chain Reaction), and in-depth sequencing **[23-25]**.

It has been agreed that to be effective, the MRD detection process should be able to detect a single malignant cell per 1000 cells. Currently, FISH(Fluorescence in situ hybridization), spectral karyotyping, PCR and gene sequencing are complementing these processes. All is a disruption of the environment and malignant cells have a replication of immunoglobulin genes in ALL B-lineage and a rearrangement of T cells in T All. In the study of MRD by the process of molecules, specific rearrangements should be planned for each condition during the diagnosis. The presence of the same sequence was evaluated after completing a specific treatment phase to evaluate the value of MRD.

In some cases All cells carry a direct genetic imbalance such as transmission that leads to genetic congestion. The most common example of such type combinations is All

over BCR(Breakpoint Cluster Region Protein)-ABL1(Tyrosine-protein kinase).This finding makes MRD measurement easier as PCR-directed gene fusions require complex sequencing or patient-specific studies required for antigen-receptor replication studies because unchanged primers were used in all cases with a given fusion Bcr -abl1 **[26]**.

NEOPLASTIC HEREDITARY DISORDER:

DNA SEQUENCING IN ASTHMA:

Susceptibility and expression (severity) of common diseases such as asthma and allergy. Although current scientific findings will be discussed, one must realize that this is a rapidly evolving field of investigation and realize that new developments are likely (so one should always check sources such as PubMed for the latest results). It is extremely important to understand the basic principles of genetic approaches because the results of these studies will affect everyone both professionally and personally. Although most of us in the field feel it is still premature, there are multiple companies already offering genetic susceptibility testing for a wide range of common diseases including asthma.

Emphasis will be placed on the results from genome wide association studies (GWAS) using case control or case only approaches. Gwas approaches are based on the ability to rapidly analyze genetic variants (mainly SNPs, (single nucleotide polymorphisms), usually with a high degree of heterozygosity) across the whole genome to determine which genetic variants are associated with disease susceptibility (case-control studies) or which are associated with measures of disease severity or response to treatment, pharmacogenetics (case only studies). Gwas studies are also performed using families, especially trios, defined as an affected child with genotyping from both parents (for example: NHLBI CAMP study identified PDE4 as an asthma susceptibility gene, 1), but it is generally easily to ascertain and characterize a large number of unrelated cases and controls than to study multiple family members.

RHEUMATOID ARTHRITIS:

Whole-genome sequencing allows for the potential identification of every variant in the genome. It is the most straightforward of the Ngs methodologies since the entire genome is prepared and placed onto the sequencer with minimal processing. However, due to the large number of sequencing reads necessary to cover the entire genome, let alone the appropriate amount of coverage necessary to generate good quality variant calls, it remains the most expensive. For this

reason very few rheumatic diseases including eye disorders. studies have yet undertaken whole-genome sequencing. However, we anticipate that this will not be the case for much longer since the cost for whole-genome sequencing continues to decrease.

DENTAL DISORDERS:

The role of Ngs in general microbiology is to obtain a genomic definition of pathogens which may harbor information about drug sensitivity and the inter-relationship of the various pathogens which can be used to detect infection outbreaks **[22]**. The oral microflora is composed of numerous microorganisms which are normal commensals of the oral cavity. While some of them are harmless, certain microorganisms are known to be pathogenic and responsible for commonly occurring oral infections. Usually, the focus of dental research was restricted to a small fraction of oral microbes especially the opportunistic pathogens. The advent of sequencing methods like next-generation sequencing has enabled newer avenues in microbiome studies thereby providing information on the broad diversity of microbial taxa regardless of their cultivability **[23]**.

The use of Ngs has made tremendous progress in identification of genetic variation in diseases with underlying genetic disorder. Prior to the use of Ngs, it was not possible to identify the entire sequence of genetic alterations. Ngs has enabled the identification of the complete complement of DNA variants, de novo mutations and the genes underlying Mendelian forms of disease and characterization of important structural variation that may contribute to diseases like cleft lip and palate **[24]**.

Oral squamous cell carcinoma is a common epithelial malignancy known for its heterogeneous nature. The complexity of the lesion has resulted in the inability to accurately diagnose and manage them thereby resulting in poor prognosis. The use of Ngs has enabled researchers to identify the genomic alterations evident in oral squamous cell carcinoma (OSCC).

RECENT ADVANCES IN DNA SEQUENCING:

PCR:

For standard PCR, all you need is a DNA polymerase, magnesium, nucleotides, primers, the DNA template to be amplified and a thermocycler. The PCR mechanism is as simple as its purpose: 1) double-stranded DNA (dsDNA) is heat denatured, 2) primers align to the single DNA strands and 3) the primers are extended by DNA polymerase, resulting in two copies of the original DNA strand. The denaturation, annealing, and

elongation process over a series of temperatures and times is known as one cycle of amplification. Each step of the cycle should be optimized for the template and primer set used. This cycle is repeated approximately 20-40 times and the amplified product can then be analyzed. PCR is widely used to amplify DNA for subsequent experimental use. PCR also has applications in genetic testing or for the detection of pathogenic DNA.

As PCR is a highly sensitive method and very small volumes are required for single reactions, preparation of a master mix for several reactions is recommended. The master mix must be well mixed and then split by the number of reactions, ensuring that each reaction will contain the same amount of enzyme, dNTPs(Deoxynucleoside triphosphates) and primers. Many suppliers, such as Enzo Life Sciences, also offer PCR mixes that already contain everything except primers and the DNA template.

Guanine/Cytosine-rich (GC-rich) regions represent a challenge in standard PCR techniques. Gc-rich sequences are more stable than sequences with lower GC content. Furthermore, Gc-rich sequences tend to form secondary structures, such as hairpin loops. As a result, Gc-rich double strands are difficult to completely separate during the denaturation phase. Consequently, DNA polymerase cannot synthesize the new strand without hindrance. A higher denaturation temperature can improve this and adjustments towards a higher annealing temperature and shorter annealing time can prevent nonspecific binding of Gc-rich primers. Additional reagents can improve the amplification of Gc-rich sequences. DMSO(Dimethyl sulfoxide), glycerol and betaine help to disrupt the secondary structures that are caused by GC(guanine-cytosine content) interactions and thereby facilitate separation of the double strands.

Hot Start PCR:

Unspecific amplification is a problem that can occur during PCR. Most DNA polymerases that are used for PCR, work best at 68 - 72°C. Therefore, the chosen extension temperature should be in this range. The enzyme can, however, also be active to a lesser degree, at lower temperatures. At temperatures that are far below the annealing temperature, primers tend to bind non-specifically, which can lead to non-specific amplification, even if the reaction is set up on ice. This can be prevented by using polymerase inhibitors that dissociate from the DNA polymerase only once a certain temperature is reached. The inhibitor can be an antibody that binds the polymerase and

denatures at the initial denaturation temperature

RT-PCR:

Reverse transcription PCR, or RT-PCR, allows the use of RNA as a template. An additional step allows the detection and amplification of RNA. The RNA is reverse transcribed into complementary DNA (cDNA), using reverse transcriptase. The quality and purity of the RNA template is essential for the success of RT-PCR. The first step of RT-PCR is the synthesis of a DNA/RNA hybrid. Reverse transcriptase also has an RNase H function, which degrades the RNA portion of the hybrid. The single stranded DNA molecule is then completed by the DNA-dependent DNA polymerase activity of the reverse transcriptase into cDNA. The efficiency of the first-strand reaction can affect the amplification process. From here on, the standard PCR procedure is used to amplify the cDNA. The possibility to revert RNA into cDNA by Rt-PCR has many advantages. RNA is single-stranded and very unstable, which makes it difficult to work with. Most commonly, it serves as a first step in QPCR, which quantifies RNA transcripts in a biological sample.

qPCR and RT-qPCR:

Quantitative PCR (qPCR) is used to detect, characterize and quantify nucleic acids for numerous applications. Commonly, in RT-qPCR, RNA transcripts are quantified by reverse transcribing them into cDNA first, as described above and then qPCR is subsequently carried out. As in standard PCR, DNA is amplified by 3 repeating steps: denaturation, annealing and elongation. However, in qPCR, fluorescent labeling enables the collection of data as PCR progresses. This technique has many benefits due to a range of methods and chemistries available.

In dye-based qPCR (typically green), fluorescent labeling allows the quantification of the amplified DNA molecules by employing the use of a dsDNA binding dye. During each cycle, the fluorescence is measured. The fluorescence signal increases proportionally to the amount of replicated DNA and hence the DNA is quantified in "real time". The disadvantages to dye-based qPCR are that only one target can be examined at a time and that the dye will bind to any ds-DNA present in the sample.

In probe-based qPCR, many targets can be detected simultaneously in each sample but this requires optimization and design of a target specific probe(s), used in addition to primers. There are several types of probe designs available, but the most common type is a hydrolysis probe, which incorporates the

use of a fluorophore and quencher. Fluorescence resonance energy transfer (FRET) prevents the emission of the fluorophore via the quencher while the probe is intact. However, during the PCR reaction, the probe is hydrolyzed during primer extension and amplification of the specific sequence it is bound to. The cleavage of the probe separates the fluorophore from the quencher and results in an amplification-dependent increase in fluorescence. Thus, the fluorescence signal from a probe-based qPCR reaction is proportional to the amount of the probe target sequence present in the sample. Because probe-based qPCR is more specific than dye-based qPCR, it is often the technology used in qPCR diagnostic assays.

Tagging Methods:

While next-generation sequencing platforms can easily sequence thousands of targets isolated from one sample, these instruments are unable to differentiate matching targets isolated from multiple samples. However, there are a number of work-around methods that address this predicament to different degrees. One solution is to mix separate tests with one another. For example, if genes A, B, and C are sequenced in patient #1, genes D, E, and F patient #2, and genes G, H, and I in patient #3, the patient samples may all be combined because the target genes to be sequenced are unique and known for each patient. Another alternative is to rely on physical divisions built into the sequencers themselves that provide sequence data independent of each other. Illumina's Genome Analyzer has eight such independent channels,while the ABI SOLiD system has eight channels per slide and can run two slides per run. The Heliscope Sequencer also runs two slides per run, but with each flow cell containing 25 independent channels. The 454 Genome Sequencer FLX PicoTiterPlate plate can be partitioned into two, four, or 16 regions, but doing so reduces overall coverage. However, the long sequencing reads produced by the 454 instrument allows for efficient addition of "DNA barcode tags," unique nucleotide signatures that allow one to mark and track individual samples. Barcodes can be added to the ends of fragments by either tagging during PCR **[25]** or following the isolation of targeted sequences **[26]**. Although barcodes can be used in a similar fashion with the higher throughput sequencers, the short read lengths produced by these instruments require that the tags be kept short or else the resulting genomic sequence may be difficult to align. Of course, this is considerably less of a problem if the reference sequence is confined to only a small portion of the

genome or if paired-end sequencing is used to, in effect, increase the sequencing read length. Paired-end sequencing is a strategy whereby both ends of immobilized fragments are sequenced, effectively doubling the read length **[27]**. An added benefit of paired-end sequencing is that it can allow for the identification of translocations in fragments containing breakpoints **[28]**. Mate-pair sequencing is a modification of the pair-end strategy whereby mate pairs are generated by ligating the ends of size-selected genomic fragments to a common linker. Each fragment end is then cleaved at a known distance from the linker and paired-end sequenced. Since the expected distance between paired ends is known, deviations from this value allows for the identification of sequence copy number variations in the region **[29]**.

FUTURE SCOPE:

There is more molecular pathology techniques and there is also high level of highly equipped instruments which are underdeveloped and recent advances are still progressing in research of hereditary disorders but our study does not extend upto to the ongoing research.In my study I Just plotted the procedures which are basic recent advances in DNA sequencing but there is more to invent in these field to correct the several hereditary outbreak

LIMITATION:

High technological and DNA sequencing methodology is not discussed and the karyotyping, genetics and molecular pathology were not elaborated in the study and even bioinformatics is also not included in this study which are still the highly advanced things that are to be improved to give support for DNA sequencing

CONCLUSION

DNA sequencing enables scientists to determine the genome sequence. Genome sequence- figuring out the order of DNA nucleotides and Scientists can now identify the genes responsible for genetic diseases. Many types of acquired diseases can also be detected by observing genes and serving a boon to mankind to determine the disease of genetically inherited and acquired diseases.

REFERENCES

[1] Holt RA, Jones SJM. The new paradigm of flow cell sequencing [Internet]. Vol. 18, Genome Research. 2008. p. 839–46. Available from: http://dx.doi.org/10.1101/gr.073262.107

[2] Doggett NA, Grady DL, Longmire JL, Deaven LL. Generation and

Mapping of Chromosome Specific Sequence-tagged Sites [Internet]. Automated DNA Sequencing and Analysis. 1994. p. 96–101. Available from: http://dx.doi.org/10.1016/b978-0-08-092639-1.50018-6

[3] Mp B, Brundha MP, Nallaswamy D. Hide and seek in pathology- A research on game-based histopathology learning [Internet]. Vol. 10, International Journal of Research in Pharmaceutical Sciences. 2019. p. 1410–4. Available from: http://dx.doi.org/10.26452/ijrps.v10i2.606

[4] Brundha MP, Pathmashri VP. Quantitative Changes of Red Blood cells in Cancer Patients under Palliative Radiotherapy-A Retrospective Study. Research Journal of Pharmacy and Technology. 2019;12(2):687–92.

[5] Prashaanthi N, Brundha MP. A Comparative Study between Popplet Notes and Conventional Notes for Learning Pathology. Research Journal of Pharmacy and Technology. 2018;11(1):175–8.

[6] Kumar MD, Brundha MP. Awareness about nocturia-A questionnaire survey. Research Journal of Pharmacy and Technology. 2016;9(10):1707–9.

[7] Hannah R, Ramani P, Brundha MP, Sherlin HJ, Ranjith G, Ramasubramanian A, et al. Liquid Paraffin as a Rehydrant for Air Dried Buccal Smear. Research Journal of Pharmacy and Technology. 2019;12(3):1197–200.

[8] Preethikaa S, Brundha MP. Awareness of diabetes mellitus among general population. Research Journal of Pharmacy and Technology. 2018;11(5):1825–9.

[9] Kalaiselvi R, Brundha MP. Prevalence of hysterectomy in South Indian population. Research Journal of Pharmacy and Technology. 2016;9(11):1941–4.

[10] Harsha L, Brundha MP. Prevalence of dental developmental anomalies among men and women and its psychological effect in a given population. Journal of Pharmaceutical Sciences and Research. 2017;9(6):869–73.

[11] Balaji S, Brundha MP, Path DNB. Awareness of About Breast Cancer among Dental Surgeons. Res J Pharm Biol Chem Sci. 2016;8(8):797.

[12] Shenoy PB, Brundha MP. Awareness of polycystic ovarian disease among females of age group 18-30 years. Journal of Pharmaceutical Sciences and Research. 2016;8(8):813–6.

[13] Jamuar SS, Tan E-C. Clinical application of next-generation sequencing for Mendelian diseases. Hum Genomics. 2015 Jun 16;9:10.

[14] Rabbani B, Tekin M, Mahdieh N. The promise of whole-exome sequencing in medical genetics. J Hum Genet. 2014 Jan;59(1):5–15.

[15] Timothy CN, Samyuktha PS, Brundha MP. Dental pulp Stem Cells in Regenerative Medicine – A Literature Review [Internet]. Vol. 12, Research Journal of Pharmacy and Technology. 2019. p. 4052. Available from: http://dx.doi.org/10.5958/0974-360x.2019.00698.x

[16] Shreya S, Brundha MP. Alteration of Haemoglobin Value in Relation to Age, Sex and Dental Diseases-A Retrospective Correlation Study. Research Journal of Pharmacy and Technology. 2017;10(5):1363–6.

[17] Ferdioz J, Brundha MP. Awareness of stye. International Journal of Pharmaceutical Sciences Review and Research. 2016 Jan 1;40(1):30–2.

[18] Ravichandran H, Brundha MP. Awareness about personal protective equipments in hospital workers (sweepers and cleaners). International Journal of Pharmaceutical Sciences Review and Research. 2016;40(1):28–9.

[19] Shah M, Shanks M, Packham E, Williams J, Haysmoore J, MacLaren RE, et al. Next generation sequencing using phenotype-based panels for genetic testing in inherited retinal diseases. Ophthalmic Genet. 2020 Jun 16;1–7.

[20] Tanaka H, Watanabe T. Mechanisms Underlying Recurrent Genomic Amplification in Human Cancers. Trends Cancer Res. 2020 Jun;6(6):462–77.

[21] Brundha MP. A Comparative Study-The Role of Skin and Nerve Biopsy in Hansen's Disease. Journal of Pharmaceutical science and Research. 2015;7(10):837.

[22] Farndon S, Behjati S, Jonas N, Messahel B. How to use… lymph node biopsy in paediatrics [Internet].

Vol. 102, Archives of disease in childhood - Education & practice edition. 2017. p. 244–8. Available from: http://dx.doi.org/10.1136/archdischild-2015-309634

[23] Zaura E. Next-generation Sequencing Approaches to Understanding the Oral Microbiome [Internet]. Vol. 24, Advances in Dental Research. 2012. p. 81–5. Available from: http://dx.doi.org/10.1177/0022034512449466

[24] Glick AB, Van Waes C. Signaling Pathways in Squamous Cancer. Springer Science & Business Media; 2010. 462 p.

[25] Binladen J, Gilbert MTP, Bollback JP, Panitz F, Bendixen C, Nielsen R, et al. The use of coded PCR primers enables high-throughput sequencing of multiple homolog amplification products by 454 parallel sequencing. PLoS One. 2007 Feb 14;2(2):e197.

[26] Meyer M, Stenzel U, Myles S, Prüfer K, Hofreiter M. Targeted high-throughput sequencing of tagged nucleic acid samples [Internet]. Vol. 35, Nucleic Acids Research. 2007. p. e97. Available from: http://dx.doi.org/10.1093/nar/gkm566

[27] Dunn JJ, McCorkle SR, Everett L, Anderson CW. Paired-End Genomic Signature Tags: A Method for the Functional Analysis of Genomes and Epigenomes [Internet]. Genetic Engineering. 2007. p. 159–73. Available from: http://dx.doi.org/10.1007/978-0-387-34504-8_9

[28] Hadamard J. Lectures on Cauchy's Problem in Linear Partial Differential Equations. Courier Corporation; 2014. 320 p.

[29] Kamada M, Hase S, Sato K, Toyoda A, Fujiyama A, Sakakibara Y. Whole Genome Complete Resequencing of Bacillus subtilis Natto by Combining Long Reads with High-Quality Short Reads [Internet]. Vol. 9, PLoS ONE. 2014. p. e109999. Available from: http://dx.doi.org/10.1371/journal.pone.0109999