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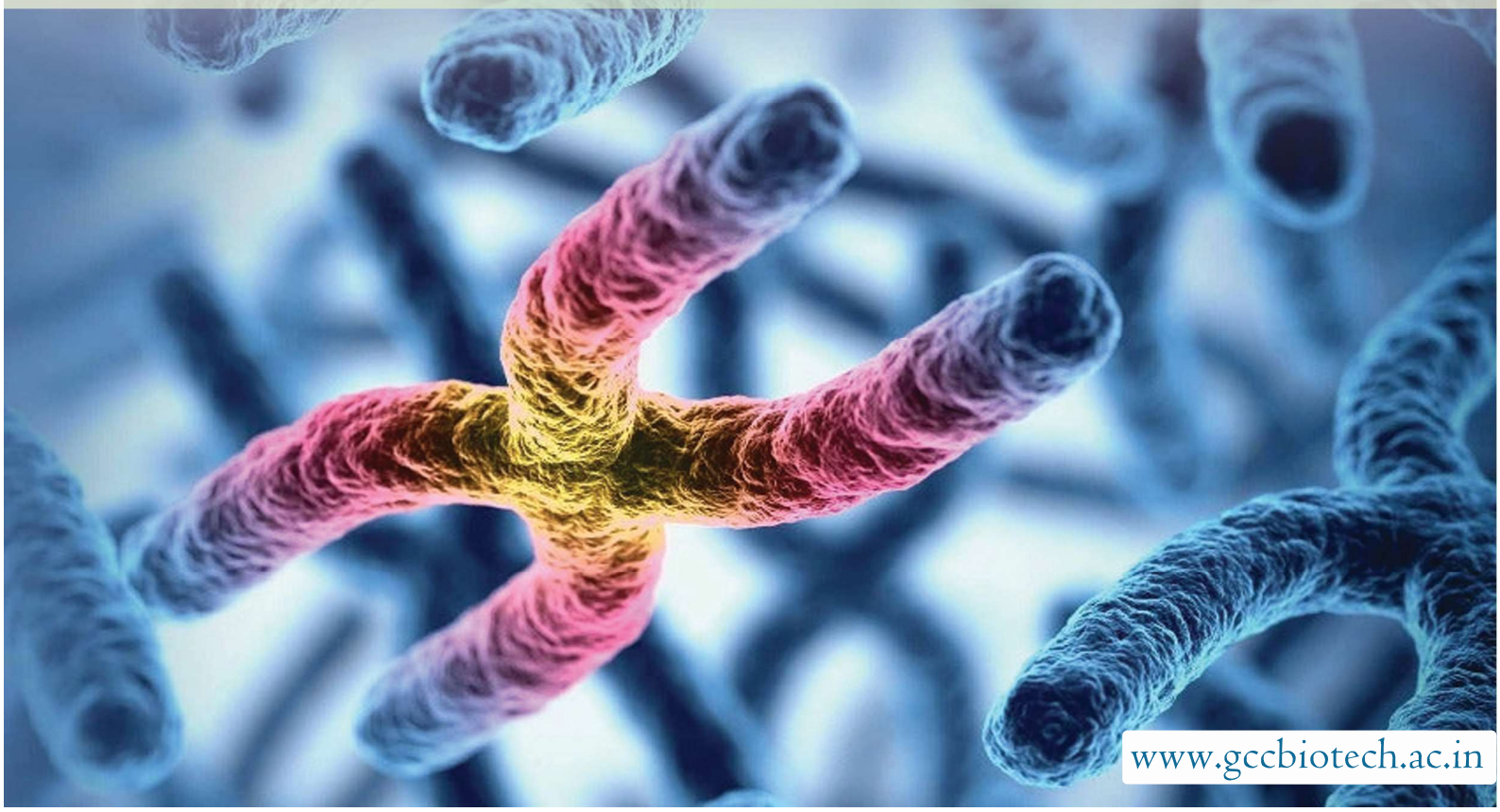

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REVOLUTIONIZING THE MODERN THERAPEUTIC RESEARCH THROUGH TARGETED GENETIC MODIFICATION

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ABSTRACT

The ability to engineer biological systems and organisms holds enormous potential for applications across basic science, medicine and biotechnology. Programmable sequence-specific endonucleases that facilitate precise editing of endogenous genomic loci are now enabling genetic variations creating new phenotypes. With the development of CRISPR/Cas9, the DNA sequence of any gene in the human genome can be modified in ways that can knock out its function completely, introduce or correct a very specific mutation, or add an extra sequence. However, the treatment of human diseases need to be tissue-specific, it is essential to efficiently delivery the CRISPR/Cas9 cargo into target tissue. The issues related to off-target effect of CRISPR/Cas9 should be investigated thoroughly before any clinical application.

Keywords: CRISPR/Cas9, human genome, mutation, tissue-specific, off-target

INTRODUCTION

In recent times, the breakthrough in genome modification has revolutionized the modern therapeutic research. The targeted genetic modification may result in the up regulation or down regulation of the desired gene expression. Previous genome editing tools such as zinc-finger nucleases (ZFN) and transcription-activator-like effector nucleases (TALEN) highlighted the potential of targeted manipulation of genes, but limited targeting capacity and complicated design hampered the usability of these technologies. Clustered regulatory interspaced short palindromic repeats (CRISPR)/CRISPR associated protein 9 (Cas9) is a novel and competent RNA-guided endonuclease-based genome editing technique, that is adapted from naturally occurring bacterial immune system (Sharma et al., 2021). Emmanuelle Charpentier and Jennifer A. Doudna have been awarded the 2020 Nobel Prize in Chemistry for their work on CRISPR/Cas9. They discovered a microbial immune mechanism can be transformed into a tool that can simply and cheaply edit genomes with high precision (Doudna & Charpentier, 2014).

TARGETING GENOME THOUGH CRISPR/CAS9 TECHNIQUE

CRISPR/Cas9 technology utilizes single guide RNA (sgRNA) sequence and Cas9 endonuclease. sgRNA is a combination of CRISPR RNA (crRNA) and trans-activating crRNA (tracrRNA), that identify and attach the sgRNA:Cas9 ribonucleoprotein complex to the target DNA (Chylinski et al., 2014). After DNA targeting, Cas9 generates double-stranded breaks (DSBs) at the site of target DNA (Brouns et al., 2008). These DSBs are further repaired by insertions, deletions, substitution or inversions. The DNA repair mechanism can be performed by either host's natural repair machinery or by using, customized DNA sequences (Porteus, 2019). Therefore, CRISPR/Cas9 technology is a promising genome-editing tool with therapeutic potential against incurable genetic disorders by modifying their DNA sequences. As compared with other genome-editing techniques, CRISPR-Cas9 is simple, efficient, and very specific (Sharma et al., 2021).

EX VIVO AND IN VIVO GENOME EDITING

Targeted gene modification via genome editing tools (CRISPR/Cas9) is a powerful method to precisely manipulate cellular behaviour and function (Li et al., 2020). The two most commonly used approaches are ex vivo and in vivo gene editing therapy (Fig. 1). For ex vivo editing therapy, cells are isolated from a patient to be treated, edited and then re-engrafted back to the patient. To achieve therapeutic success, the target cells must be able to survive in vitro and return to the target tissue after transplantation. In case of in vivo editing therapy, engineered nucleases are delivered by viral or non-viral approaches and directly injected into the patient for systemic or targeted tissue (such as the eye, brain, or muscle) to achieve therapeutic effect.

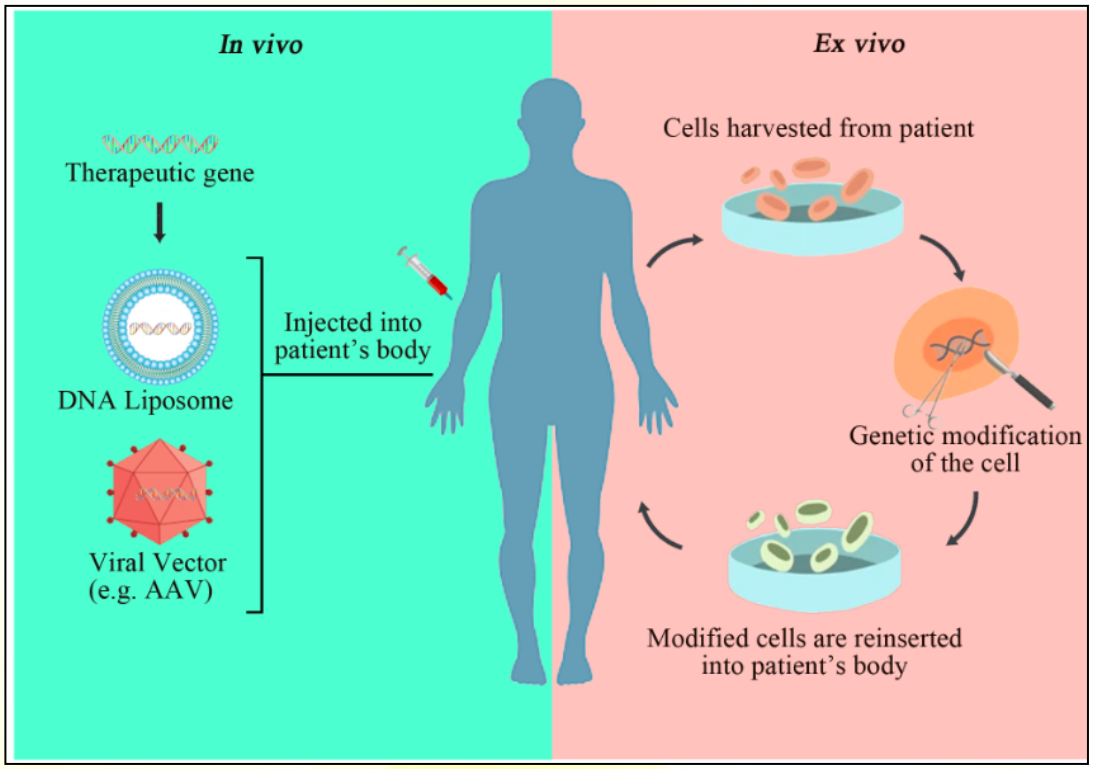

Fig. 1 : Basic concepts of ex vivo and in vivo genome editing therapy

CLINICAL TRIALS

China performed first human *ex vivo* clinical trial employing gene editing using CRISPR/Cas9 technique in a patient with aggressive lung cancer (Cyranoski, 2016). They targeted PD-1 gene in T cells from peripheral blood of the patient using electroporation of sgRNA and Cas9 plasmid, and infused them back into the patient. Later, they reported the presence of edited T cells in the peripheral blood of all patients who received infusion (Lu et al., 2020). The study concluded that, this method is feasible and safe; however, future trials should use superior gene editing approaches to improve therapeutic efficacy. The findings of CRISPR/Cas9 clinical trials offer more and more prospects and widen the therapeutic treatment opportunities. In 2019, successful treatment of sickle-cell disease and β-thalassemia was reported, which was conducted by CRISPR/Cas9-mediated disruption of BCL11A gene in the stem cells that were isolated from patient's peripheral blood with hemoglobinopathies (Haematology, 2019). In another *in vivo* clinical trial, CRISPR/Cas9 gene therapy-based drug AGN-151587 was given directly into the eye, via subretinal injection, to cure a rare blindness condition called Leber's congenital amaurosis 10 (LCA10) that is caused by mutations in the gene CEP290 (Ledford, 2020). In the past few years, a number of clinical trials on model animals and humans have been conducted to determine its therapeutic potential against various genetic disorders, including

Cancer (Zhou et al., 2015), Allergy (Chu et al., 2015), Duchenne muscular dystrophy (Long et al., 2014), Cardiovascular diseases (Seidah, 2013), Huntington disease (Yang et al., 2017), Alzheimer's disease (Rohn et al., 2018), Metabolic liver disease (Villiger et al., 2018), Fanconi anaemia (Osborn et al., 2016), Hereditary tyrosinemia (Wang et al., 2018), Sickle cell anemia (Wen et al., 2017), β-Thalassemia (Song et al., 2015), Cystic fibrosis (Schwank et al., 2013), Retinitis pigmentosa (Yu et al., 2017), Cataract (Yuan et al., 2016), Human immunodeficiency virus (Liao et al., 2015), Hepatitis B virus (Ramanan et al., 2015), Human papilloma virus (Yoshiba et al., 2019), Epstein–Barr virus (Chen et al., 2018) etc.

GENE DELIVERY

As the treatment of human diseases need to be tissue-specific, it is essential to efficiently delivery the CRISPR/Cas9 cargo into target tissue. Various physical, viral and non-viral systems have been used as a vector for the delivery of CRISPR/Cas9. The most commonly used viral vectors for gene editing are derived from adenovirus, adeno-associated virus (AAV), retrovirus, lentivirus, and herpes simplex virus (HSV). Adeno-associated virus and lentivirus are the most commonly used vectors, which have the ability to transduce both dividing and non-dividing cells (Worgall, 2005). However, the best vectors suffer the drawback of a relatively small gene-carrying capacity, which limits the inclusion of all regulatory elements. In some cases, these viral vectors may trigger the host immune response and cause an adverse effect. Non-viral vector systems include DNA plasmids or plasmids combined with chemically synthesized vehicles such as cationic liposomes (Alton et al., 1993). The advantage of non-viral vector systems is that they do not activate the host inflammatory and immune response; however, the efficiency of gene transfer for these vector systems is very low.

BIOETHICAL ISSUES IN HUMAN GENETIC MODIFICATION

Human genetic modification is a breakthrough in the treatment of genetic diseases; however, bioethical concerns have been raised by many experts that may impact the human race (Khan, 2019; Li et al., 2020). Any undesirable changes to a genome may lead to an unknown, long-term safety issues. Any error in the germ-line editing will become a permanent part of a child's genetic legacy and might affect generations to come. Moreover, the ability to modify the genes of children to produce "designer" babies may create a radical change in society. The children will not be embraced as precious gifts, but rather will become commodity that can be made at parent's choice. The cost of gene therapies for rare diseases is often targeted for certain population groups (wealthy), who possess the ability to pay. As a result, the rich will live a longer and healthier life, while the poor will lack basic healthcare, and this may create a societal discontent. It raises concerns on inequality in health outcomes between rich and poor nations. Religious thinkers believe that editing baby's genome is against nature's rule of inheritance. Genetic engineering may weaken the value of the parent-child relationship. Public opinion will evolve with regard to the application of human genome editing for enhancement in body mass, athletics, intelligence etc. Creating individuals with improved performance will have an unfair advantage in social and economic competition. Another concern in human genetic modification is the risk of bio-attack and crime. Unfair application and genetic modification may create humanized weapon that may create havoc in society.

CONCLUSION

CRISPR/Cas9 genome editing tool showed promising advancements in clinical trials. However, the results of CRISPR/Cas9-based genome editing is unpredictable and raises safety concerns. The safety of gene editing must be determined in well-controlled and well-monitored clinical trials before

being popularized for therapeutic purpose. Taking bioethical concern into consideration, it is suggested that genome-editing techniques must be utilized for therapeutic purpose but not for reproductive purpose.

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WHOLE GENOME SEQUENCING FOR INFECTION DIAGNOSTICS

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ABSTRACT

Next generation sequencing is a deep sequencing method which has revolutionised genomic research. NGS technology can likely replace conventional diagnostic methods in the near future. Conventional methods such as Sanger Sequencing takes over decade to decipher the human genome whereas in NGS whole genome can be sequenced in a single day. The invention of techniques like Illumina technology, Ion torrent technology (ITT) and Nanopore technology has paved way for utilization of whole genome sequencing (WGS) for infection diagnostics. Using NGS it is possible to sequence a large number of bacterial genomes and deliver information of the sequence in real-time but there are a few hurdles for its implementation in clinics and hospitals in the present setting. In this article, the potential application of WGS as a routine technique is discussed.

Keywords: NGS, WGS, Illumina Technology, ITT, Nanopore Technology.

INTRODUCTION

Next generation sequencing (NGS) is a revolutionary technique for diagnostics of infectious diseases that can transform the conventional practices in microbiology and could be a useful technology in diagnostics in future (Köser et al., 2012). The classic approach on infection diagnosis have been the detection of presence of a pathogen in a clinical sample and culture assay. Finding selective media for pathogen isolation and maintaining appropriate culture conditions remains a challenge and it also requires a specially dedicated labour. This conventional method, although extensively used is ineffective with respect to major pathogens like *Mycobacterium leprae*, *Treponema palidum*,A, B, C, E viruses and fastidious bacteria (Lecuit & Eloit, 2014). This is progressively being replaced by nucleic acid-based testing methods like Nucleic Acid Sequence Base Amplification (NASBA) or Polymerase Chain Reaction (PCR). PCR is advantageous on speed, sensitivity and specificity but can only identify predefined targets. For pathogens with highly variable makeup, like DNA viruses such as adenovirus, papilloma virus and RNA virus- enterovirus, the method lacks discrimination between genotypes. To overcome the hurdle, several approaches have been made on development stage, aiming at broadening the detection range. One approach, which is utilization of direct hybridisation method of random amplified nucleic acids is unsatisfactory due to its reduced sensitivity (Dunne et al., 2012). On the other hand, the utilization of NGS paves way for increasing the depth of sequencing, increased sensitivity and detection of rare organisms as well. Using NGS, it is possible to sequence a large number of bacterial genomes and deliver information of the sequence in real-time, forming the basis of the technological advancement in the field. Large sequences of RNA/DNA can be combined to form complete genomes in prokaryotes or eukaryotes (Shendure & Ji, 2008). The reduction of costs over time coupled with high speed and accuracy aids the utilization of WGS in health and clinical sectors effectively (Loman & Pallen, 2008). WGS has deepened the capacity of sequencing exhibiting the ability to detect rare species. Tools of metagenomics are also employed for discovering new infectious agents (De Vlaminck et al., 2013) This approach involves De Novo assembly of genomes of pathogens and utilization of molecular

microbiological tools. These studies aid identification of novel phage or viruses and hence it is very crucial in infection diagnostics (Barzon et al., 2013).

WGS FOR ROUTINE PATHOGEN- ARE WE THERE YET?

For increased cost effectivity, higher batch sizes are preferable, contrasting to the demands of clinical practice for diagnosis where smaller batch sizes are required. To overcome this obstacle, sample preparation should be simplified. WGS must be sensitive for sequencing DNA directly from a single colony to avoid subculturing thereby drastically reducing the time consumption. Considering these, multiple approaches for designing tools are being considered. The major tool platforms include Oxford Nanopore 454, Illumina MiSeq and Ion Torrent (Bentley et al., 2008). A significant advantage is that the analysis can be carried out in a single step removing complications and multiple subtests.

***Illumina technology*:** A second-generation sequencing technique developed by Solexa and Lynx therapeutics is based on bridge amplification where DNA with desired adapters are used as a substrate for amplification followed by enzymatic steps and imaging which occur in a flow cell. The diverse platforms include MiSeq, HiSeq2000 or HiSeq2500 (Bentley et al., 2008). This method drastically reduces the time and skilled labour necessities. This technology can also be used for many types of detection and the sample size requirement is low. The issue with this technique is the lack of sync in synthesis process among members of cluster that may interfere with accurate consensus sequence. This can also affect the number of cycles that can be run (Buermans & Den Dunnen, 2014).

Ion Torrent Technology (ITT): Emulsion PCR forms the basis for ITT. Two primers complementary to sequence adapters are used, one in solution and another in base. During the process, molecules amplify to million identical copies bound to beads allowing signal detection (Barzon et al., 2013). Since the emergence, the technique has evolved rapidly and has chips with larger surface area and higher sensor well density. The average read length is high and have a low run time. The output is easily interpretable.

***Nanopore technology*:** Hundreds of GB of DNA sequence can be obtained at low cost by threading them through a nanopore. Two major systems developed are solid-state sensor technology and biological membrane systems. Two proteins used for pore generation are *Mycobacterium smegmatis* porin A (MspA) and alpha hemolysin (Liu et al., 2016). Oxford Nanopore Technologies with a MinION that is portable is the recent advancement of this technology. They use protein nanopores in polymer membrane in which current changes while nucleotide passes through the detector. The miniature size of MinION accounts for its applications. It can identify base modifications aiding epigenetic identification (Cao et al., 2016). Direct DNA/ RNA sequencing at lower cost remains the advantage. Currently the rate of errors is not negligible but with the development of sequencing methods, this can be overcome.

APPLICATION OF WGS

Real time tracking of Zika Virus (ZIKV): ZIKV is a mosquito-borne virus from *Flaviviridae* of viruses that is phylogenetically and antigenically related to Spondweni virus. Although ZIKV was identified years ago, a very little information on its genome is available because of its unique characteristics. In animals, this virus reaches its peak load in 1-2 days of infection and in humans, within a week (Faria, Sabino, et al., 2016). The characteristic early viral load peak and late onset of clinical symptoms reduces the chance of enough virus to be left for sequencing from the serum of patients with ZIKV infection. Also, the clinical enrichment for cell culture is laborious and may also lead to some genetic changes. The Zika in Brazil Real-time Analysis (ZiBRA) (Quick et al., 2016) was initiated for improving ZIKV sequencing in Brazil. This project aimed at extraction of information for vaccine design and to improve

the conventional techniques of molecular diagnostics. For the study, MinION, a portable genome sequencing device that works in real time was utilized for WGS after tiling PCR. The project helped for the generation of viral genomes with 50% coverage. In future, ZiBRA project is proposed to help detect and characterize arboviruses (Faria, Azevedo, et al., 2016).

***Epidemiological typing*:** WGS provides high resolution results in epidemiological typing proved by studies on cholera outbreaks. Transmission pathways of the organism can be reconstructed especially when the organism has a high rate of genomic change for infection control (Schürch & Siezen, 2010).

Viral drug resistance of HIV 1: WGS studies on HIV 1 helps predict treatment outcomes and to overcome virologic failure. Novel tools in bioinformatics have been used to process WGS data to reduce error rates and have higher performance (Cortez & Maldarelli, 2011). DEEPGEN HIV, the HIV-1 CCR5 Tropism Test (V3) and HIV-1 Co-receptor Tropism are the assays currently under research (Swenson et al., 2011).

Virus identification in clinical specimens: WGS is advantageous in viral identification where a viral agent is suspected but cannot be identified by commonly used techniques. Many viruses cannot be easily cultured or detected, wherein NGS aids an efficient, sensitive, and accurate detection strategy (Radford et al., 2012).

WGS for Covid variants: The outbreak of coronavirus 2 (SARS-CoV-2) has created a pandemic with over 5 million deaths worldwide. The approach of utilization of viral RNA isolation, quantification of viral load, next generation sequencing, and analysis was employed (Nazario-Toole et al., 2022). The identification of 109 nucleotide changes in the coding region genome of the SARS-CoV-2 and mutations in untranslated regions have helped in vaccine design. The shotgun transcriptome sequencing has also aided the tracking of viral origin by comparison with available sequences (Harilal et al., 2020).

CHALLENGES

Any new advancement in technology comes along with challenges. The biggest hurdle is the development of highly reproducible version of NGS that can be used for clinical identification of pathogens. The critical factor of development and maintenance of reference databases remains primitive. The correct mapping of contiguous sequence, sequencing of isolates where only short reads is obtained remain the most pressing challenges (Hasman et al., 2014). There is always a requirement for skilled workforce for collection, management and analysis of the data obtained. Random amplifications are unavoidable for all available technologies wherein they also amplify host nucleic acid and are not cost-effective. In order for the process to be effective, the microbe vs. host nucleic acid ratio must be high. A high genome coverage is mandatory for prediction of virulence and success in getting necessary information is not predictable when partial sequences are obtained. The analytical sensitivity that depends on genome length is not easy for evaluation and are influenced by matrix properties. The affordability remains a big concern for clinical applications of these technologies. The low costs of conventional techniques like NASBA and PCR makes them the go-to methods of infection diagnostics (Oniciuc et al., 2018).

CONCLUSION

WGS is an upcoming technology capable of replacing conventional techniques of infection diagnostics. Since the advancement of technologies, the goal is to design a system with higher cost efficiency and accuracy. Platforms that are easily operatable and requires lesser maintenance are preferred. Amalgamation of Artificial intelligence and WGS are currently under research. It has always exhibited

potential for outbreak detection and epidemiological surveillance. they can thus be used for assisting the track of AMR genes and mobile genetic elements. A comparison of Moore's law for sequencing equipment and computing suggests that WGS is developing faster. Thus, it may cause a bottle neck effect which needs to be sorted before large scale clinical application. With the advancements in nanopore technologies and cloud computing, the affordability for a number of clinical applications is expected in the near future.

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NANOGEL- A MINI-REVIEW

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ABSTRACT

Nanogels have been used as carriers for innovative drug delivery systems. Immunological drug delivery via self-nanogels with the ability to encapsulate large biomolecules is one of the most commonly studied and is majorly applied in the field of cancer vaccines, cytokine delivery, nasal vaccines, and nucleic acid delivery including several clinical trials. Nanogels show a high degree of drug loading capacity and can be produced by emulsion polymerization, free radical crosslinking polymerization technique, reverse microemulsion polymerization, photolithographic, and micro molding methods. Nanogels are innovative drug delivery systems that are currently one of the most unique and attractive immunological drug delivery systems and play an integral part in correlation with primitive and recent causes of therapy such as poor stability and nonspecific effect.

Keywords: Drug loading, polymerization, drug delivery

INTRODUCTION

Nanogels are composed of highly cross-linked nano sized hydrogel that can be ionic or non-ionic in nature. Nanogels are generally formed by physically or chemically cross-linked polymeric networks. Nanogels (nanometer-sized gel) are considered as an effective immunological drug-device of innovative drug delivery systems (Tahara & Akiyoshi, 2015). The nanogel size ranges from 20-200 nm and is spherical in shape with the final size of typically 1-1000 nm (Yadav et al., 2017). It is a hydrophilic network of three-dimensional structures which has the ability to hold a large amount of physiological fluid or bioactive drugs without affecting its internal structure (Sultana et al., 2013). There are various types of nanocarriers that are considered as a drug delivery system that includes carriers prepared from polymers such as hydrogel, micelles, and nanospheres, carriers of lipid choice such as liposomes, inorganic carriers include gold, silica, or magnetic nanoparticles which have been used for immunological delivery. Carriers in nano-size have been developed to deliver bioactive drugs, antigens, adjuvants, etc. effectively to certain immune cells such as lymph nodes or antigen-presenting cells which will further promote or suppress the immune response in the body. Biologically active peptides have a short half-life span to be used for therapeutics because it requires a controlled release system which is not as easy as that of conventional drugs because their large molecular size hinders the diffusion (Eckmann et al., 2014; Raemdonck et al., 2009). Nanogels are considered to be a promising carrier in the uptake of peptide, protein, drug and other compounds because of their property in inertness, high affinity to aqueous solution and stability. Nanogel also possesses the potential advantage of nanoscale formulation and their property in high drug loading capacity, hydrophobicity, biocompatibility, thermodynamic stability, and low viscosity (Yadav et al., 2017). It can also escape the renal clearance and extend the serum half-life period with the help of their size and that makes them considered as a best carrier (Arnfast et al., 2014). Nanogel as a carrier device has an advantage in preventing the drug from proteolytic or enzymatic degradation, intensify the absorption by cells and tissue which induces the immunostimulatory cascade and controls the pharmacokinetics (Tahara & Akiyoshi, 2015). Peptide or

protein-based nanoparticles are currently taking attention to be designed due to the minimal toxicity, biodegradability, and bio-compatibility. These protein based nanoparticles are mostly naturally occurring proteins with slight modification like albumin, casein, collagen and fibrin (Neamtu et al., 2017). In this review, a novel strategy of encapsulation of peptides into the nanogel using various techniques results in the use of nanogel as a novel therapeutic system.

WORKING OF NANOGEL

Nanogels are formed by physically or chemically cross-linked polymer networks, which has high specificity, low toxicity can be targeted in the area of interest and are widely used due to their property of drug encapsulation, uniformity, stability, minimal toxicity, responsiveness to external stimuli (Neamtu et al., 2017). The encapsulation of peptides in the natural polymer can be achieved by ionic gelation, polyelectrolyte complexation, and coacervation whereas, in the synthetic polymer can be achieved by interfacial polymerization, encapsulation-polymerization salting out, and nanoprecipitation (Patel et al., 2014). The release of the peptide into the targeted site can be achieved by simple diffusion, degradation of nanogel, change in pH, and temperature leading to release of the peptide from the nanogel (Yadav et al., 2017).

pH and Temperature responsive mechanism: Different pH values can be acquired by different physiological environments that induce transformation in the nanogel structure such as disassembly, rearrangement, or release of the peptide from the carrier. The change in the critical pH (pHc) level reflects the pH of the target in the body. Change in the degree of ionization is due to the changes in the osmotic pressure, this degree of ionization change induces the swelling or deswelling whereas, the cross-linked nanogel swell or deswell in response to increase in pH. Also, nanogel possess positive and negative charge along with the polymer chain which comprises amphoteric polyelectrolyte. The presence of this charge involves in the swelling and release of peptide from the nanogel structure. Release of peptide into an area largely depends on the pH possess by the area of the body (Eckmann et al., 2014).

Photo-responsive mechanism: Photo isomerization involves a number of the light-responsive group present in the structure of nanogel which undergoes changes in the size or shape due to light exposure. The release of peptide into the cytoplasm can be mediated by nanogel, which when they get excited produce singlet oxygen results in the oxidation in cellular compartment walls. Drug release from the nanogel to the target site is achieved by specific wavelength of light being irradiated. This method has been formulated for the cancer therapy (Wani et al., 2014).

CLASSIFICATION

The nanogels are classified based on different types such as response and the different types of polymers.

i) Type of response consists of stimuli and non-responsive response where former consist of response to many external factors such as pH, light, temperature, etc. Nanogels when exposed to these factors either swell or de-swell. Whereas latter are non-reactive to any environmental stimulus. And they react only when they react with water and generally swell when they come in contact with water (Sultana et al., 2013).

ii) There are different types of polymers used for the preparation of nanogels that should be biodegradable, non-toxic, non-antigenic, and biocompatible (Rangari & Ravikumar, 2015).

Polymers can be classified in two ways, ***Natural polymers*** such as Gelatin, Chitosan, Dextran, Heparin, Alginate are derived from a biodegradable and biocompatible component such as collagen, which is basically produced by partial hydrolysis of various animal body parts such as skin, connective tissue, etc. It can be used because of its non-toxicity, easy crosslinking, and most importantly its ability in the preparation of nanoparticles (Vandervoort & Ludwig, 2004).

Chitosan is a cationic polymer composed of mainly glucosamine units and is an N-deacetylated derivative of chitin. Because of its anti-inflammatory, anti-microbial and, anti-oxidant properties, this makes it a suitable vehicle for delivering biopharmaceutical drugs to treat dermatological disorders (Nagpal et al., 2010).

Dextran is one of the most promising macromolecular carrier candidates for a wide variety of therapeutic agents which is because of its excellent physicochemical properties and physiological acceptance (Dhaneshwar et al., 2006).

Heparin is a linear polysaccharide that consists of repeating units of uronic acid and glucosamine residues. The abundant functional groups present in heparin can be used to improve biocompatibility and therapeutic efficacy, cell adhesion, and controlled delivery of growth factors (Liang & Kiick, 2014).

Alginate is a polysaccharide found in the cell walls of brown algae. Alginate hydrogels have been used in wound healing, drug delivery, and tissue engineering and could be used as a carrier agent for drugs or any hormone (Lee & Mooney, 2012).

Synthetic polymers such as Polylactic acid, Polystyrene, Polyglycolic acid (PGA), Poly methyl methacrylate (PMMA) are biodegradable and bioactive polyesters. It can be used in the medical field because of its ability to degrade into non-toxic lactic acid. It is one of the plastics used in 3D-bioprinting. Polystyrene is biocompatible and is expected not to affect interactions of nanoparticles with drugs or biological systems (Loos et al., 2014). PGA is a suitable polymer for carrying drugs because of their form varying from highly amorphous to crystalline state. Polymethyl methacrylate (PMMA) is an amorphous polymer and is generally used because of some important properties such as low cost, non-toxic, easy process ability, compatibility and minimal inflammatory reactions with tissues (Ali et al., 2015).

APPLICATIONS

Nanoparticles are involved in providing the adjuvant activity by enhancing the innate immune response which includes virus-like particles (VLPs) which is being used in vaccines against hypertension, cancer, rheumatoid arthritis, a wide range of infectious disease, and Alzheimer's disease (Smith et al., 2013). Nano-emulsions is another application of nanotechnology which targets self-antigens to suppress the immune response (Wang et al., 2012). Nanogels is widely used as dispersed drug carriers which are hydrophilic in nature having the property of encapsulating small biologically active agents particularly biopharmaceuticals as active drug compounds and biomacromolecules which include their ease of formulation, stability of the resulting dispersion, and high loading capacity and are being used in the treatment of diseases like diabetes, neurodegenerative diseases (Kabanov & Vinogradov, 2009). Nanoparticles which include polymeric nanoparticle, nanogels, liposomes, and polymersomes are indicative drug delivery carriers mainly in the treatment of diseases like cancer immunotherapy (Sahdev et al., 2014). PEGylated nanoparticles mainly PLGA [Poly lactic-co-glycolic acid] are commonly employed in nano carrier-based immunological drug delivery which has the potential to undergo sustained drug release to elevate the T-cell response for long term memory (Demento et al., 2012).

Cancer vaccines that can be either prophylactic or therapeutic delivered using polysaccharide self-nanogels are considered to be essential immunotherapy for the treatment of diseases like cancer and to enhance the antigen-specific humoral and cellular immune response (Lesterhuis et al., 2011).

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POTENTIAL OF ETHNIC FERMENTED FOOD AS A PROMISING FUNCTIONAL FOOD INGREDIENT

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ABSTRACT

The functional aspects of fermented foods have been mostly related to probiotic bacteria or any other target microbial generation of fermented molecules, such as bioactive peptides during food fermentation. Many of the ethnic tribal groups produce fermented food products, which are animal or plant-based products that influence their social, cultural and economic life and most importantly their health. The fermentation process destroys many harmful microorganisms and chemicals in foods and adds beneficial bacteria. For e.g., Indian ethnic fermented food such as fermented soyabean (kinema), fish (gnari), rice (idli) etc. provides numerous benefits such as, they can make poorly digested and reactive foods into health-giving foods, enhancing the bioavailability of nutrients, reduces the symptoms of lactose intolerance, decreasing the prevalence of allergy in susceptible individuals and also reducing the risk of certain cancers and many more therapeutic aspects thereby improving overall health and wellbeing.

Keywords: Probiotic, ethnic fermented food, functional foods.

INTRODUCTION

The diversity of ethnic fermented foods in India is an unparalleled food culture of each community. Fermentation is the oldest food preservation method in the world, and this food processing technique reduces or even eliminate toxic compounds present in food (Soemarie et al., 2021). Many fermented foods like fermented soyabean (kinema and hawaijar), fermented fish (gnari), curd, fermented vegetables (gundruk, sinki, khalpi and soijin) are a great source of probiotics and nutrients. Various functional microorganisms play important roles in the ethnic fermentation processes by their functional properties enhancing several health-promoting benefits to the consumers such as bio-enrichment of nutritional value, protective properties, production of antioxidants, bioavailability of minerals, antimicrobial activities, non-production of biogenic amines, and probiotics properties (Tamang, 2020). Lactic acid bacteria (LAB) isolated from fermented foods are one of the popular probiotics found in ethnic fermented foods, which have considered as an important functional food group (Satish Kumar et al., 2013). Consumption of probiotics is useful for maintaining sound health against pathogenic bacteria in the gut microbiota, maintaining the normal balance of gut microbiota, improving digestive health as well as the immune system (Bansal et al., 2013). This review describes the probiotic potential of ethnic fermented food as a promising functional food ingredient.

HEALTH BENEFITS AND THERAPEUTIC EFFECTS FROM ETHNIC FERMENTED FOOD

Improvement of immunity system

Probiotics inhibit the growth of pathogenic bacteria in the body. They compete for nutrients for growth and proliferation, which would otherwise be utilized by the pathogens. Several studies have demonstrated that probiotics such as *Lactobacillus rhamnosus*GG and. plantarum could inhibit the attachment of enteropathogenic coli the GI tract (Galdeano et al., 2019). Probiotics could also strengthen the intestinal barrier by increasing the number of Goblet cells that reinforce the mucus layer (Galdeano

et al., 2019). *L. acidophilus* cell extract could increase the MUC2 expression in HT29 cells, and this effect was independent of probiotic adhesion to the cell monolayer (Kim et al., 2008). Probiotic bacteria also regulate the mucosal immune responses through the induction of different cytokines. After oral probiotic administration, cytokines produced by T cells in the lamina propria of the small intestine are secreted in slightly higher levels than those observed in the presence of commensal bacteria, specifically IFN-γ and TNF-α cytokines (Galdeano et al., 2019). Probiotics are also efficient in decreasing IgE immunoglobulin, as well as in alleviating symptoms in allergic reactions. Clinical trials have also confirmed that probiotics reduced blood glucose and insulin levels in diabetic patients.

Increasing the bioavailability of nutrients

Lactic acid bacteria (LAB) releases various enzymes and vitamins into the intestinal lumen which aids in digestion by alleviating symptoms of intestinal malabsorption, and also produces lactic acid, which lowers the pH of the intestinal content and helps to inhibit the development of pathogens such as. or. coli (Parvez et al., 2006). Bacterial enzymatic hydrolysis could enhance the bioavailability of protein and fat and also increase the production of free amino acids, propionic acid, short-chain fatty acids (SCFA), lactic acid, and butyric acid are also produced by LAB. SCFAs when absorbed contributes to the available energy pool of the host and protects the body against pathological changes in the colonic mucosa (Parvez et al., 2006).

Lowering of serum cholesterol

Fermented food products produced hypocholesteraemic effects (Grunewald,1982). This property therefore the potential healthful aspects of dairy products fermented with *L. acidophilus* since hypercholestermia has been considered as one of the major factors contributing to cardiovascular disease (Hasan et al., 2014).

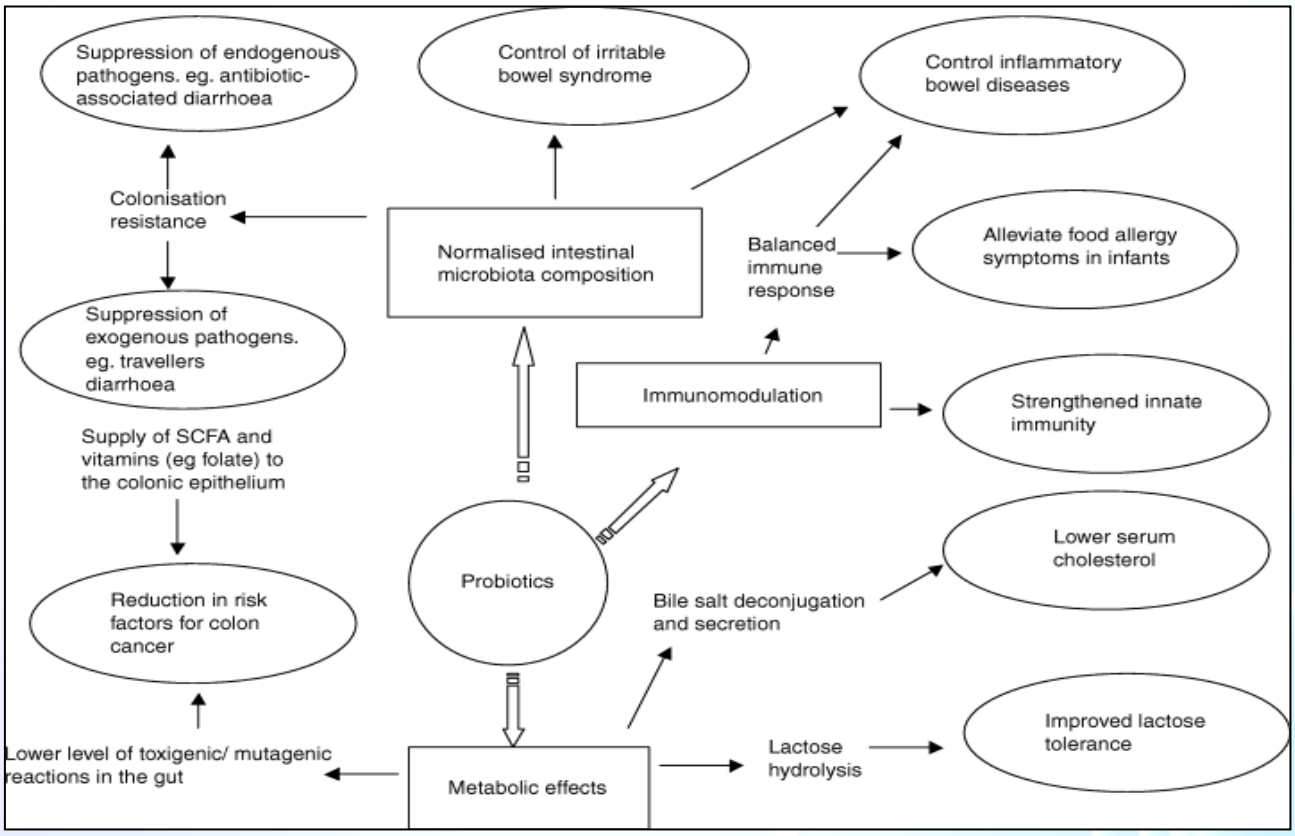

Fig. 1 - Various health benefits of consuming probiotics found in ethnic fermented foods (Parvez et al., 2006).

Anticarcinogenic properties

Fermented food products could work against certain types of cancers. In animal studies, it has been observed that lactic acid bacteria (LAB) exert an anticarcinogenic effect either by preventing cancer initiation or suppressing the initiated cancer (Hasan et al., 2014). Anticarcinogenic effects of products fermented with *L. acidophilus* have been reported. The potential mechanisms by which LAB exert antitumor effects have been suggested. These include changes in faecal enzymes that are thought to be involved in colon carcinogenesis, reducing the mutagenicity of chemical mutagens, cellular uptake of mutagenic compounds and suppression of tumors by improving immune response (Hosono et al., 1986).

Fig. 1 shows various the beneficial effects of consuming ethnic fermented foods such as - suppression of endogenous pathogens, controlling bowel diseases, reducing the risk of colon cancer, strengthening of innate immunity, improving lactose tolerance, lowering serum cholesterol etc

CONCLUSION

In India, almost all communities in Indian cuisine revolve around fermented foods, which is considered one of the good mechanisms of introducing the probiotic in human beings. Fermented soybeans (kinema), fish (gnari), rice (pakhalabhat) etc. are some common among Indian regional cuisine as well as the tribal cuisine. Different tribes make fermented beverages in their own way using different food items, be it plant or animal-based. These ethnic fermented foods not only help them in terms of nutrition but also act as preventive measures for lifestyle diseases such as hypertension, diabetes, cardiovascular diseases associated with the hypercholesterolemia etc. and also protects against various other pathogenic infections. Therefore, its high time to link the people knowledge of preparation and the traditional food commodities having numerous health benefits with different food and beverage manufacturing agencies, which will ultimately help in bringing innovative products into the market, thereby creating a win-win situation.

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THE ROLE OF BLOOD-BASED BIOMARKERS IN THE DIAGNOSIS OF INTRACEREBRAL HEMORRHAGE: A MINI-REVIEW

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ABSTRACT

Time is the brain when it comes to early detection of stroke. Diagnosis, differentiation, and characterization of stroke and its subtypes are crucial elements of treatment and the prognosis of the disease. Early diagnosis of ischemic stroke can lead to administration of tissue plasminogen activator (tPA) followed by thrombectomy in some instances that could significantly improve the outcome of the patient. Moreover, the early treatment ensures that the patient will suffer from a lesser disability and might also recover some functions that could be lost if interventions do not happen on time. Thus, it can be easily understood that early diagnosis is the key to increasing the chances of a patient's recovery and reducing mortality and permanent disability. Biomarkers pose as an effective alternative to complex procedures that consume much time and are not economically feasible for a vast population. This review discusses some of the more researched biomarkers for diagnosing and differentiating intracerebral hemorrhage.

Keywords: Stroke, tPA, thrombectomy, Biomarkers, intracerebral hemorrhage

INTRODUCTION

Approximately 5.5 million people die due to stroke every year. Strokes can be classified primarily into two types: Ischemic with approximately 87% of patients and hemorrhages with nearly 13%, including subarachnoid hemorrhage (Senn et al., 2014). Intracerebral hemorrhage (ICH) is one of the most critical diseases, with the second most common stroke with a high fatality rate. Long-term functional independence can be achieved by only 12 to 39% of the patient and, approximately 54% of the patients die within a year (An et al., 2017). Therefore, acute identification of the cause of ICH is required for proper treatment of the mechanism of hemorrhage, even though chronic hypertension and cerebral amyloid angiopathy are some of the major reasons for ICH (Schrag & Kirshner, 2020). Apart from hypertension and accumulation of amyloid, spontaneous ICH also comes under the etiological subcategory of ICH.

PATHOPHYSIOLOGY OF ICH

With the help of the study that focuses majorly on the new therapies and biomarkers to detect and cure ICH faster, we are now explaining the pathophysiology of ICH in a much better way. Presently the most accepted two-phase theory in which the mechanical injury that is the primary brain injury is followed by the edema formation and other inflammation (Keep et al., 2012).

Primary Brain injury

The initial brain injury leads to the disruption of the brain's architecture. This leads to the release of blood, thus increasing the intracranial pressure, which ultimately causes brain herniation. To lessen the morbidity after this stage, clot removal was also considered an option. The study was not successful due to the post effects of the surgery, which was worse than the primary stage. The secondary injury can be

prevented by the prevention of hematoma expansion which takes place on the first day after the ictus (Keep et al., 2012).

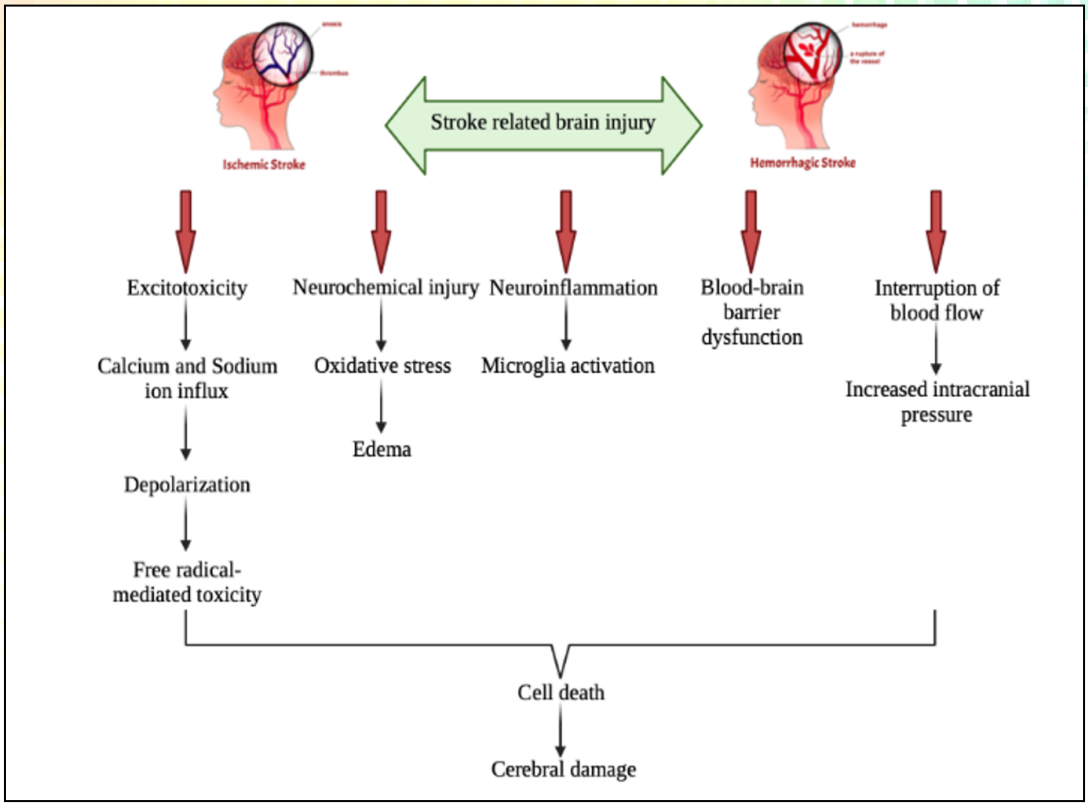

Fig 1. Molecular mechanism of stroke.

Secondary Response

The body reacts to the rise in intracranial pressure and bleeding, thus increasing the chances of morbidity. The roles of the clot component were also studied as they play an important role in secondary injury. Thrombin is a significant component released during the initial days of the primary injury and plays a role in hemostatic**.**This leads to the activation of microglia cells and forms scar tissues. The activation of the complement system is the last mechanism that leads to cell lysis and inflammation and thus reducing the injury (Hua et al., 2000).

With the help of neuroimaging, management of ICH is conducted. This includes the knowledge of the progression of the injury so that effective treatment can be done and thus fatality can be reduced. Biomarkers for diagnostic purposes are the major study attraction and still a challenge. The biomarkers can distinguish the type of strokes to be an effective treatment. Studies have been conducted by analyzing the biomarker at different intervals after the onset of the primary injury. This review has collected information about the different types of biomarkers and their role in diagnosing ICH.

BIOMARKERS OF ICH

Bilirubin

Bilirubin, the end product of the heme metabolism, can be toxic for the body if present in huge concentrations. But the byproduct has some properties that can be used as a defense mechanism for ICH. These properties are anti-inflammatory, antioxidant, and cytoprotective properties. The antioxidant property increased significantly after decreasing oxygen concentration in the body to 2% (Stocker et al., 1987). The drop of oxygen in the brain is due to ischemia which activates antioxidant activity to counter the harmful effects due to stress. The significant damage is due to the increased concentration of ROS

during ICH, which can be reduced by bilirubin, thus preventing further injuries (Thakkar et al., 2019)**.**The rise in stroke can be observed if proinflammatory cytokines are combined with dyslipidemia and promotes thrombosis. Bilirubin plays a role here by reducing the production of the cytokines, thus inhibiting the further mechanism. The ability of the cells to respond to the environment is called intracellular communication. Cellular adhesion molecules play an essential role in this process. But the increased expression of these molecules can cause the release of inflammatory cells into the arterial walls of vessels. Bilirubin can prevent this by impeding ICAM1, thus reducing inflammatory response (Wagner et al., 2015). All these properties play an essential role in lowering ICH damage.

Glutamate

The level of glutamate in the plasma is the primary source of study for the effect of glutamate in ICH patients. This will tell us the amount of glutamate that has been released from the brain. The glutamate level is high in the perihematomal area, with deep ICH patients (Wu et al., 2013)**.** Excitotoxicity will contribute to the secondary damages caused by ICH. The effect of this as a biomarker can only be confirmed after further clinical trial and majorly by inhibiting the excitotoxicity pathway and examining the different effects (Senn et al., 2014). It has a role in the formation of edema within five days of ICH, and its concentration in the plasma depends on the severity of the ICH. The glutamate concentration was four times 30 min after ICH and was continuously high for the next five hours (Rendevski et al., 2018).

Glial fibrillary acidic protein (GFAP)

Star-shaped glial cell intermediate filament protein is commonly found in the brain and spinal cord and central nervous system (Eng et al., 2000; Middeldorp & Hol, 2011). The main aim of these intermediary filaments is maintaining the cell shape, structure, and motility of astrocytes which contributes to the integrity between the blood and brain, myelination, and coloration of the white matter. Concerning the expansion of hematoma and destruction of the glial protein, then the GFAP is significantly released from the brain into the blood (Brunkhorst et al., 2010).In approximately 80% of the patients with ICH, GFAP was predominantly associated with the GFAP serum concentrations (Tichy et al., 2016), GFAP localization and time, and the quantity of blood released or the bleed size. A quantitative determination of GFAP concentrations in vitro was performed by electro chemiluminometric immunoassay. Also, These GFAP concentrations were proportionately higher in patients with Intra Cerebral Hemorrhage when compared to Ischemic stroke. Often, the severity of the stroke coincides with the GFAP levels and intensity of the brain injury. The major reason for the rapid GFAP release is hematoma expansion because of the destruction of glial cells. It is a structural protein. An increase in the concentrations of GFAP in the serum can result in a brain injury called Glioblastoma. In addition, bleeding in subarachnoid space may lead to more and more release of GFAP into the blood. The utmost destruction in the posterior fossa of the brain can lead to increased GFAP concentration (Torres-Platas et al., 2016). ICH influences the GFAP concentration. Moreover, GFAP expression is not distributed evenly throughout the brain. Excessive parenchymal bleeding in the brain lands in cell destruction; necrosis and cellular disintegration usually do not occur early. Some factors that contribute to the release of the GFAP into the blood include the disintegration between the blood-brain barrier and the mechanical disruption and necrosis resulting in the loss of the structural integrity of astrocytes (Brunkhorst et al., 2010). A process called reactive astrogliosis (Sofroniew, 2009) often lead to the release of a significant amount of GFAP, further increase in the concentrations of GFAP in the blood (Metting et al., 2012; Nylen et al., 2006; Pelinka et al., 2004).

Cellular fibronectin

Cellular fibronectin is a vital biomarker for predicting hemorrhage transformation of the ischemic lesion. It is a biomarker of the extracellular matrix and the microvascular basal lamina. According to the previous studies, a relationship has been found between secondary bleeding and the increased concentration of this biomarker in the blood. The biomarker level is high after the proteolysis of the extracellular matrix and the injury caused in the basal lamina. During the retrospective study, it was found that the level of c-Fn in the plasma was =3.6 µg/mL in the patients of acute ischemic stroke when examined after the Tissue-type plasminogen activator (tPA) administration and was independent of the hemorrhagic infarction type 2 (Castellanos et al., 2004). The high level of the circulation c-Fn can be due to the three outcomes that are edema in the brain, poor functional outcome, and also can be due to hemorrhagic transformation (Wang et al., 2020).

High mobility group box 1

High mobility group box 1 is a potential indicator for inflammation to analyze its concentration dependence on ICH. It is a DNA binding protein released by the microglia (Senn et al., 2014). Its concentration can also give an idea about the severity of the case. It was observed that the level of HMGB1 is directly proportional to the volume of hematoma (Lei et al., 2020) as it is related to the rupture of the vessel and necrosis. This is the reason it can be used as a prognostic biomarker. This is now a significant area of research so that prior knowledge of ICH can be predicted. After applying the translational approach, which will be helpful for stroke recovery (Singh et al., 2016).

MMP 9

The inflammatory response in the perihematomal region that accompanies the mechanical disruption of the expanding hematoma results in the ICH (Qureshi et al., 2009; Xi et al., 2006). Responses usually occur with activation of the microglia and astrocytes, which account for the blood resident glial cell, leukocyte recruitment to the site of injury, inflammation, edema formation, induction of some proinflammatory mediators and neurotoxic mediators, and the mechanical disruption of the blood-brain barrier (Barone & Feuerstein, 1999; Panickar & Norenberg, 2005; Power et al., 2003; Wang & Tsirka, 2005).inflammation leads to neuronal death and neuronal damage in the stroke. Mechanical disruption of the blood-brain barrier results in the activation of the matrix metalloproteinases (MMPs), also known as gelatinase B and perihematomal edema formation (Candelario-Jalil et al., 2009; Rosenberg et al., 1998; Tejima et al., 2007). Toll-like receptor 2 (TLR 2) is a transmembrane receptor that recognizes the peptidoglycan of the bacterial cell wall and lipoteichoic acid. And, thereupon, activate inflammatory signals. Aggregation of amyloid-β (Aβ) protein in the cerebral blood vessel wall results in cerebral amyloid angiopathy (CAA). It most commonly occurs in small and medium-sized arteries in the leptomeninges and cerebral cortex (Yamada, 2015). Some results suggest that the matrix metalloproteinases play an efficient role in CAA and CAA-related ICH. Zinc dependent endopeptidases which involve the MMPs help in the degradation of the extracellular matrix, including laminin, collagen, and fibronectin (Malla et al., 2008; Nagase et al., 2006; Page-McCaw et al., 2007).

Fas Receptor

The main form of cell death in consideration of the brain perihematomal region is Fas-mediated apoptosis. There were studies related to the Fas level in the plasma after an acute ICH. In just a day after ICH, the level of Fas was decreased, and the relation between s-Fas and PE enlargement was also observed (Delgado et al., 2008). The role of Fas was to act as an inhibitor of Fas interaction with its

ligand (Cascino et al., 1995). In another study carried out by Tarkowski, the level of s-Fas was low in the cerebrospinal fluid in patients suffering from Ischemic stroke. The relation between the s-Fas level and brain infarct volume was inversely related to neurological deficit. The study was done three weeks and three months after the stroke (Tarkowski et al., 1999). The level of s-Fas related to edema formation is still a research subject. The level of s-Fas comes back to normal, indicating that it can be a transient phenomenon after 24 hours. The overexpression of the s-Fas receptor, mainly the Fas-L, is a sign that the activation of Fas/Fas-L is the main contributor to brain damage (Delgado et al., 2008). Inhibition of Fas-L is proved to help reduce secondary brain damage in models of brain ischemia (Ackery et al., 2006). Researchers are still curious about the inhibition of this system and how it can be helpful in other CNS diseases.

Fibrinogen

The studies have shown that the rise in the fibrinogen level is a significant factor in increasing the risk of stroke. It was also observed that race plays another role in the risk of ischemic stroke; fibrinogen level in black people was considered high compared to other races. The increased level of fibrinogen can also be attributed to activities like smoking, diabetes, arterial pressure, which leads to stroke (Ropper et al., 2014).When this was compared to the patients with ICH, the fibrinogen level was low, but there was no significant difference between the two types of strokes.The risk of stroke of ICH was higher in men than in females, but the prevalence of ischemic stroke was reverse (Chitsaz et al., 2012). Studies were made regarding the protection of neurons; it was observed that the low concentration of thrombin outside the vascular could play a role as a protective factor against cell death by ischemic hemorrhage. A high concentration of the same can lead to seizure and cerebral edema due to losing the blood-brain barrier (Xi et al., 2003).

BNP

Intra cerebral hemorrhage is one of the most common devastating diseases (Broderick et al., 2007). Increased levels of S100b in the serum after the intracerebral hemorrhage in the subarachnoid region suggested a correlation between a long-term functional outcome and a complex nature in relation to the clinical use of the serological markers (Sanchez-Peña et al., 2008). S100b is a calcium-binding protein that belongs to the family of microglial proteins. It exists as the dimers of alpha and beta subunits, with alpha-beta and beta-beta. BNP is a hormone produced by neuroendocrine cells which acts as a potent vasorelaxant, diuretic, natriuretic peptide which is relatively released and stored in ventricular myocardium (Pandey, 2005) and produced as a pro-hormone called pro- BNP, which is formerly made up of 108 amino acids (Valli et al., 1999). BNP is cleaved by the enzymes that physiologically activate the BNP by getting rid of the amino-terminal portion of the pro-hormone after being released due to an increase in the wall tension from the cardiac ventricle (Mukoyama et al., 1991). BNP levels are increased in patients with heart failure. Because of its diuretic and natriuretic properties, this hormone produces innumerable biological effects including, fluid balances (Epstein et al., 1987), vasodilation (Koren et al., 1991), changes in the electrolyte and sympathetic nervous system (Floras, 1990).

HSP70

HSP70 is a good indicator of stress and cellular damage. Therefore, it can be used as a marker for the early detection of ICH. When the level of HSP70 was studied in patients suffering from intracranial hemorrhage, its concentration was observed to be associated positively with the bleeding volume and negatively with the Glasgow Coma Scale (Alatas et al., 2015). It is also a factor in the neurological deficit during the delayed phase of ICH (Manaenko et al., 2010). Therefore, the severity of the case can be analyzed by looking at the concentration level.

CONCLUSION

The potential of biomarkers in the rapid diagnosis and detection of stroke is a significantly exciting avenue for dealing with stroke and its subtypes. Intracerebral hemorrhage accounts for a substantial percentage of mortality in stroke patients and poor morbidity along with poor long-term outcomes. Thus, biomarker-based diagnosis has the long-term benefit of working in combination with current clinical standards of on-time stroke diagnosis and differentiation.

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GUT MICROBIOTA- AN AIL TO DEPRESSION?

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ABSTRACT

The human microbiome consists of microorganisms ranging from bacteria, fungi, archeae, to even viruses, forming different microbiota. The gut microbiota serves a vast range of biological functions. There are various research studies that indicate the change in dietary habits leads to gut microbiota alteration. It ultimately results in hiked health risks, one of which includes the mental health problems, such as depression. Depression is a common but serious mental illness which is shown to have prevalent increase globally. The food and other dietary sources are seen to have enhanced quality of gut microbiome. This paper would review some significant findings and studies done on interrelationship between gastrointestinal microbiota and brain.

Keywords: Microbiome, dietary habits, mental health, depression

INTRODUCTION

The gut microbiome is a home to nearly 10^{14} microorganisms including bacteria, fungi, archae, and viruses that colonize on the surface of body or within the body tissues (Gill et al., 2006). It includes dermis, oral, lung, oral mucosa, gut, mammary glands, seminal fluid, uterus, etc. The gut microbiota leads with the highest number of microorganisms. The relationship between a host and its gut microbiota is commensal as well as mutualistic. The host is responsible for providing nutrients and reliable niche to gut microbiota, where in return the microorganisms benefit the host by supporting intestinal immune function, digesting dietary digestion, defending against colonization by opportunistic pathogens, and involving themselves with enzyme production and synthesizing essential nutrients (Cresci & Teitelbaum, 2021). Reportedly, these microorganisms have benefits on mental health. It is established that food and other dietary sources enhance quality of microbiota in our gastrointestinal system which serves a significant role in individuals' day to day lives. Alterations of gut microflora also called gut dysbiosis are shown to be linked with various diseases and disorders such as type 2 diabetes, cardiovascular disease, depression (Zalar et al., 2018). Falling in line the mental illness, depression has been reported to increase in its prevalence worldwide.

THE GUT MICROBIOTA

The human gastrointestinal tract is colonized by 10^{14} microorganisms, exceeding 10 times the number of human cells in the body. It contains 150 times as many genes as our genome (Gill et al., 2006; Qin et al., 2010). In addition, gut microbiota has a crucial role in developing and maintaining an immunological response and in regulating various physiological processes. Over the past 5 years, advancements in the field of assessing the composition of gut microbiota have greatly impacted our understanding of host-microorganism interactions (Round et al., 2010).

It is estimated that the adult microbiota consists of more than 1,000 species (Qin et al., 2010) and 7,000 strains (Ley et al., 2006). Predominantly, the two bacterial phylotypes found in the human microbiota are Bacteriodetes and Firmicutes. Although, phyla of Proteobacteria, Actinobacteria, Fusobacteria and Verrucomicrobia are present in relatively low abundance (Eckburg et al., 2005). The bacterial communities vary greatly from individuals to individuals and their composition is believed to be at least partially genetically determined (Gulati et al., 2012). Diet is one of the key factors that is believed to have substantial effect on the balanced compositional signature of gut microbiota which on disruption confers disease susceptibility. For instance, the enterotype of *Bacteroids* spp. and *Prevotella* spp. are associated with diets which are high in fats or protein and high carbohydrate diets, respectively (Wu & Hui, 2011). Factors other than diet which include infection, disease and antibiotics can temporarily alter the stability of the gut microbiota composition and thereby they can lead to loss of stable well-being of the host (Forsythe et al., 2010). It has been reported that the core microbiota of elderly was different when compared with the younger.

MICROBIOTA-GUT-BRAIN AXIS: GUT MICROBIOTA TOWARDS MODIFICATION OF THE BRAIN

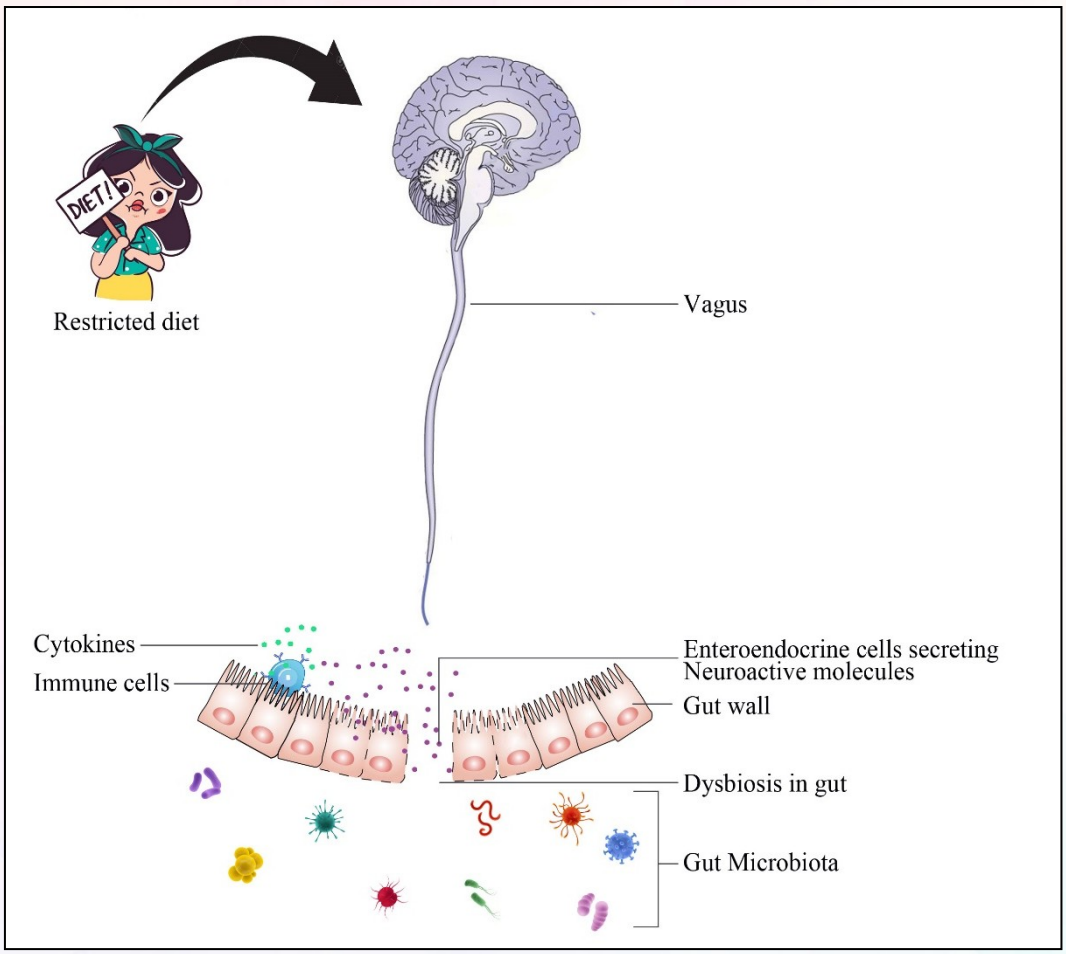
In a review article by Pacheco-Lopez and Gonzalez-Cervantes (2013), the mechanisms connecting the gut and brain are mentioned, however the stated facts are still unclear. According to the article, microbes in the gut interact with immune cells inducing the cells to produce cytokines which eventually circulate from blood to brain, leading to interaction between microbes and enteroendocrine cells that produce neuroactive molecules and peptides. These molecules interact with the vagus nerve, responsible for sending signals to the brain. Neurotransmitters and metabolites, such as butyrate, are produced by microbes in the gut, circulate to the brain. Some of them are small enough to pass through blood-brain barrier, where some alter cell activity at the barrier itself. Researchers at University of Alabama, Birmingham in 2018 reported to have found gut bacteria in human brain tissue, however the study has not been published and the full-fledged details are still unclear. The extensive bidirectional interactions between the gut microbiome and the central nervous system are maintained by multiple direct and indirect pathways which involves endocrine, immune and neural pathways (Grenham et al., 2011). This forms the basis of Microbiota-gut-brain axis. As observed in hypothalamus-pituitary-adrenal (HPA) axis, the brain is under stress, which can influence the composition of the gut microbiota.

GUT MICROBIOME AND DEPRESSION

Depression also known as major depressive disorder is a serious mental illness that affects negatively the way we feel, think, or even how we act. It is characterized by feelings of sadness, loss of interest in activities, which might lead to various emotional and physical problems decreasing your ability to function at work and home. The activation of inflammatory responses is switched on due to alterations in the composition of the gut microbiome triggering the production of microbial lipopolysaccharides. Cytokines send signals to the vagus nerve linking the process to the HPA axis which consequently causes behavioral effects. All these processes induce depression (Simkin, 2019). HPA is a neuro endocrine stress response system, which is important in both mood disorders and functional diseases. HPA system when altered are found to have various mental states including posttraumatic stress disorder (PTSD), social anxiety, schizophrenia, and depression (Naseribafrouei et al., 2014). The HPA axis helps in metabolizing microbiome diversity and its nutrient ability (Petra et al., 2015).

The gut microflora sends signals to the CNS via different pathways viz. neurotransmission, neurogenesis, etc. when it is under stable and stressful conditions. The gut microbiome dysbiosis has

been observed in patients diagnosed with mental conditions such as depression (Limbana et al., 2020). The healthy and depressed patients showed significant difference in the composition of bacteria. The depressed group showed mostly Firmicutes, Bacteroides, and Actinobacteria in the gut. This supports how the content of gut microbiota dysbiosis changes the behavior of the host (Limbana et al., 2020). Neurobiological modifications are seen to be related to the development of depression, one of which is the connection with inflammation. There were high levels of concentration of neurotransmitters in inflammation. According to Koopman and El Aidy (2017) as gut microbiomes help in controlling in curbing depression, the host must keep a stable microbial community.



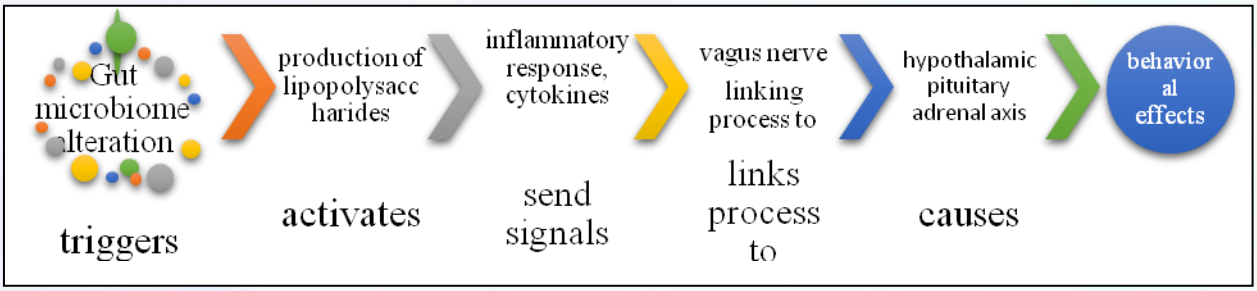


Fig. Relationship between gut microbiota and brain

YOU ARE WHAT YOU EAT- DIET IN COMPELLING HEALTHY GUT MICROBIOME

There are certain types of diet which are linked with improving mental health. Once such example includes Mediterranean diet which helps in promoting healthy food habits in contrast to the western counterparts. The type of diet choices has a significant effect on body systems, including endocrine, immune, and gastrointestinal system (Godlee, 2020). Data from animal models shows that diet-driven alterations in gut microbiota can lead to behavioral changes which shows symptoms of common mental

disorders such as anxiety and depression. There was increased Firmicutes/Bacteroidetes ratio, reduce in exploratory behavior, increased anxiety-like behavior as well as decreased memory in rodent models because of high-fat, Western- style diet (Jørgensen et al., 2015; Ohland et al., 2013), contributing to the dysbiotic conditions that send brain signals to alter diet intake behavior. High-fat diets not only associate with obesity but also cause widespread inflammation of body systems. A study on a few human data have discussed that when a diet high in inulin-rich vegetables were consumed there was an increase in *Bifidobacterium* which led to satiety improvements (Hiel et al., 2019). A recent study demonstrated that the elderly participants when exposed to 1-year Mediterranean dietary intervention showed improved cognitive function as well as reduced inflammatory markers C- protein and interleukin-17 (Ghosh et al., 2020).

Dietary supplements (probiotics and prebiotics) and fermented foods such as kimchi, sauerkraut and yogurt modulate the microbiota-gut-brain axis manipulating the gut microbiota (Long-Smith et al., 2020). There may be improvement in depression and anxiety by the introduction of living microorgansims; *Lactobacillus spp.* alone or when combined with *Bifidobacterium spp.* (Liu et al., 2019). Studies have shown promising outcomes in improving mood following fermented foods consumption.

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ANTIMICROBIAL PEPTIDES AS POTENTIAL THERAPEUTIC AGENTS TO COMBAT BACTERIAL DRUG RESISTANCE: A MINI-REVIEW

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ABSTRACT

Indiscriminate and irrational usage of antibiotics has caused the emergence of antimicrobial resistance (AMR) worldwide. Moreover, with the advent of extensively drug resistance (XDR) in clinically relevant pathogens, newer treatment regimens such as novel combinations of antibiotics and drug repurposing are displaying escalating ineffectiveness. To evade this problem, antimicrobial peptides (AMPs) can be a boon for therapeutic intervention due to their rapid killing effect, immune response modulation, antibiofilm potential, inflammation regulation and the slow emergence of resistance. The present review summarizes the biochemical nature of AMPs, their classification, mechanism of action and the recent *in-silico* advancements made in AMP research.

Keywords: Antibiotics, resistance, AMPs, antibiofilm

INTRODUCTION

AMPs, being integral in host innate immune system, are amphipathic oligopeptides ranging from 12-50 amino-acid residues and are mostly cationic (net charge~3.32) in nature (Heimlich et al., 2014; Huan et al., 2020). However, peptides like dermcidin from human sweat gland tissues and maximin-H5 from frog skin are anionic (Bahar & Ren, 2013). Since the isolation of magainin's from frog skin in 1987, several thousand AMPs have been discovered, among which 60 therapeutic AMPs are approved by the US Food and Drug Administration (FDA) (Zouhir et al., 2021).

CLASSIFICATION AND CHARACTERISTICS OF AMPS

Peptides like lysozyme imparting antimicrobial activities through enzymatic mechanisms are not considered AMPs due to their large size. The diverse nature of AMPs makes them challenging to classify; however, a broad classification is depicted in **Fig. 1.** Several physicochemical properties like peptide length, hydrophobicity, amphipathicity, net charge and solubility are crucial for AMP designing. AMPs with high structural similarity may vary in their mechanism of action. A minimum of 22 and 8 amino-acids residues are required to form α-helical and β-sheet AMPs respectively. In most cases, increased hydrophobicity, amphipathicity and solubility in aqueous solvents display better antimicrobial activity.

MECHANISM OF ACTION

AMPs are dynamic molecules and their mechanism of action can be broadly classified into membrane and non-membrane targeting mechanisms. Membrane targeting mechanisms like 'Carpet-like model' damage the cell membrane in a detergent-like manner as observed for Human cathelicidin LL-37 and β-sheet-rich AMPs. Upon reaching a concentration threshold, the hydrophobic end of AMP creates pores upon cleaving the phospholipid bilayer. In the Barrel-Stave model, peptides like alamethicin and

protegrin-1 aggregate to form multimers that penetrate the membrane bilayer causing cytoplasmic outflow and cell lysis; while in Toroidal Pore model followed by cationic peptides TC84, TC19, Lacticin Q and arenicin, AMPs accumulate on the cell membrane and form a ring hole of diameter 1-2nm causing cell lysis. Non-membrane targeting mechanisms by Bac7 and Tur1A involve inhibiting protein biosynthesis in *Escherichia coli*. Indolicidin exerts both protease and nucleic acid biosynthesis inhibition and has antibacterial (*E. coli*), antiviral (HIV) and antifungal potential, while tissue factor pathway inhibitor-1-1TC24 has the potential to degrade both DNA and RNA (Bahar & Ren, 2013; Huan et al., 2020).

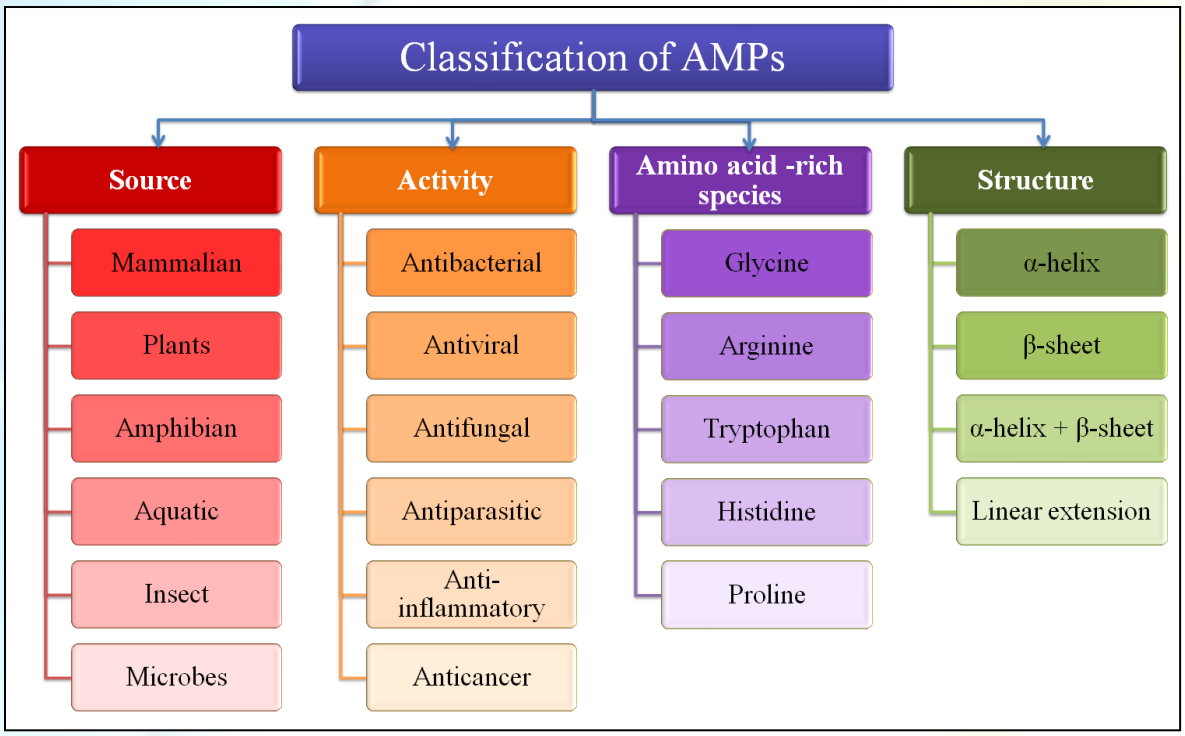

Fig. 1: Classification of antimicrobial peptides

IN-SILICO APPROACHES FOR DESIGNING NOVEL THERAPEUTIC AMPS

Notable challenges in AMP research are assessing human toxicity, selectivity, sensitivity towards extreme pH and protease, folding issues of large AMPs, AMP-resistance and production cost. These shifted the AMP research regimen towards computational screening and validation techniques. Web-based tools/servers and databases like Antimicrobial Peptide Database (APD3) (https://aps.unmc.edu/), and CPPsite 2.0 (http://crdd.osdd.net/raghava/cppsite/) are designed specifically for AMP research. Physicochemical properties from ProtParam (https://web.expasy.org/protparam/) and HLP (https://webs.iiitd.edu.in/raghava/hlp/interactive.htm) servers provide crucial insights into AMP stability. Peptide penetration ability and solubility could be screened from CellPPD (https://webs.iiitd.edu.in/raghava/cellppdmod/), CPPred-RF (http://server.malab.cn/CPPred-RF/) and ccSol (http://s.tartaglialab.com/page/ccsol_group) servers. 3D modelling of AMPs can be performed combining homology modelling, threading and molecular dynamics using I-Tasser (https://zhanggroup.org/I-TASSER/), RaptorX (http://raptorx.uchicago.edu/) and MODELLER 10.0. Peptide structures could be further refined using GalaxyRefine (http://galaxy.seoklab.org/index.html) server and validated from MolProbity (http://molprobity.biochem.duke.edu/index.php) and ProTSAV (http://www.scfbio-iitd.res.in/software/proteomics/protsav.jsp) servers. Thermodynamic parameters estimation [SCOOP: (http://babylone.ulb.ac.be/SCooP/k_query.php)] and aggregation possibility [Aggrescan: (http://bioinf.uab.es/aggrescan/)] are importance factor for AMP designing.

Further, *in-vivo* characteristics could be estimated through allergenicity [AllergenFP)], immunogenicity [VaxiJen: (http://www.ddg-pharmfac.net/vaxijen/VaxiJen/VaxiJen.html)], proinflammatory response [ProInflam: (http://metagenomics.iiserb.ac.in/proinflam/index.html)], RBC lysis effect [HemoPI: (http://crdd.osdd.net/raghava/hemopi/)] and toxicity predictions [ToxinPred: (http://crdd.osdd.net/raghava/toxinpred/multi_submit.php)]. The resultant AMPs screened could be subsequently considered for protein-peptide docking to study the intermolecular interaction patterns through ClusPro2.0 (https://cluspro.bu.edu/login.php) and HPEPDOCK (http://huanglab.phys.hust.edu.cn/hpepdock/) servers. Finally, the stability of the interactions could be validated upon performing coarse and molecular dynamics simulation respectively using CABSflex (http://biocomp.chem.uw.edu.pl/CABSflex2/) and MDWebServer (https://mmb.irbbarcelona.org/MDWeb/index.php) (Behzadipour & Hemmati, 2019; Naha et al., 2021).

CLINICALLY PROVEN AMPS AND THEIR COMBINATION THERAPY - RECENT UPDATES

Although a handful of clinically approved AMPs have been identified, malacidin and teixobactin targeting lipid biosynthesis were found active against *Staphylococcus aureus* and *Mycobacterium tuberculosis* while lactocillin targets *S. aureus* and *Gardnerella vaginalis* (Magana et al., 2020). Human lactoferrin derivative hLF1-11 was proven effective against methicillin-resistant *S. aureus* (MRSA), *Acinetobacter baumannii* and fluconazole-resistant *Candida albicans* (Seo et al., 2012). AMP-antibiotic inhibitor combination of nisin Z-ampicillin and pediocin-penicillin lowered MICs in *Pseudomonas fluorescens* while nisin-vancomycin showed inhibitory action against MRSA by blocking cell wall synthesis (Bahar & Ren, 2013). Another study highlighted the antibacterial potency of Flowlicidin-2, Ostricacin-2 and Ostricacin-3 against MurE of *S. aureus* (Zouhir et al., 2021).

CONCLUSION

In the fight against AMR development, alternate therapeutics and drug repurposing have become pivotal in establishing a successful treatment regimen against MDR pathogens. In this aspect, AMPs serve as better alternatives in antimicrobial therapy and can be very lucrative for pharmaceutical companies. In the recent years, though FDA has approved a few novel and potent AMPs, still extensive *in-vitro/in-vivo* validations must be incorporated in the current treatment pipelines.

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SCOPE OF MACHINE LEARNING IN THE SPACE OF HEALTH CARE SYSTEM – A BRIEF STUDY ON THE CROSSOVER OF SCIENCE AND TECHNOLOGIES

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ABSTRACT

The complex technology of Machine Learning (ML) and Artificial Intelligence (AI) has impacted a great revolution in the space of different industries in the last decade where Health Care System is no different which includes not only the frontend operations but also the backend Research and Development system. The deployment of Machine Learning techniques in the space of Health Care System depends vastly on the huge volume of data collected and provided by the Internet of Things (IoT) devices in order to improve and optimize the delivery system of patient conclusions. The scope of application of the Machine Learning (ML) and Artificial Intelligence (AI) techniques has a great advantage in the area of medical genetic studies and processing of medical documents. Majority of the applications are found to be focused on the diagnosis, detection and prediction models in the Health Care System. In a broader understanding, there are two verticals of data in the Health Care System- the huge volume of data which is generated by the advanced medical devices (some of which are equipped with IoT systems) and on the other hand the system or infrastructure which processes, analyses and interprets the generated data. Due to lack of proper or advanced data processing infrastructure the medical data which are generated often gets misinterpreted or interpreted incompletely. With the induction of ML and AI technology the data processing, analysis and interpretation of the medical data can be optimized, improved to a near accurate level and interpreted without noises. In this study, we would examine a brief history of Machine Learning technology and the current state of its application in the Health Care System.

Keywords: Machine learning, artificial intelligence, health care system, genetic studies

INTRODUCTION

The rise and development of digital transformation in the space of Health Care System is always attributed by continual improvisation in the aspects of both application and practical implications. An underlying challenge which is prevalent in the field of Health Care System is the vast diversity of different sub-systems and that a proper unification and adoption of fully integrated or centralized system could not be accomplished in major part of the world yet. Understanding the complexity involved in the inherent nature of the complicated human biology along with the wide variation between any two individual patients has shown the importance of the inclusion of human element in the diagnosis and treatment of various diseases however, the advancement in the spectrum of digital technologies is in no doubts becoming the indispensable tools involved in providing the best health care ecosystem.

The recent developments with data technologies have enabled the widespread acceptance and adoption of Machine Learning in various industries including the Health Care Industry in order to facilitate pristine quality of health care services. The goal here is to use the large volume of healthcare data to find and analyze diagnostic decisions and build prediction models to help the physicians to make better decisions at individual patient level. This complex ecosystem of Machine Learning uses data like genetic

information, medical imaging data, drug combination and interaction data to enhance the outcomes. This also includes Natural Language Processing (NLP) of existing medical records.

In this study we would be focusing predominantly on two of the largest applications of Machine Learning (ML) technology in the space of medical and biomedical arena. As being one of the most prominent emerging technology, Machine Learning has found a vast range of applications which adds value in the space of healthcare system. From the wide variety of applications, we would specifically study two most pivotal applications in this study. The first in the list is the application of Machine Learning technology in processing and interpreting the medical images such as Magnetic Resonance Imaging (MRI), Ultrasound (USG) Imaging, Computerized Axial Tomography (CAT or CT) Scan Imaging and Positron Emission Tomography (PET) Scan Imaging. The reports of these imaging system are ideally a series of images which traditionally demands a radiologist to analyze and interpret and then make a diagnostic decision. Machine Learning technology is spontaneously progressing in this area to find, analyze and predict the image data to indicate a disease state or seriousness level.

The second in the application of Machine Learning surrounds the area of human genetics with the interest of finding and predicting diseases and its causes. With the recent development of Next Generation Sequencing (NGS) techniques and the evolution of genetic data which includes huge databases of genetic information, the approach to conclude meaningful and useful interpretation of how genetics affect human health is now at the warfront of many researches. Understanding how any complex diseases sprouts and how genetics may be involved in increasing or decreasing an individual person's risk, a predictive model developed with Machine Learning architecture can practically aid in preventative healthcare ecosystem. Such predictive models can provide the physicians with more precise and tailored information for a specific patient in order to reduce the risk of incorrect treatment approach or acquiring more complex diseases. The common challenge which persists in all the two discussed topics is how to translate the health data which are acquired from the sophisticated medical devices and medical IoT systems into a structured, understandable, useful, trustworthy information for the patients and the physicians.

ARTIFICIAL INTELLIGENCE AND THE EVOLUTION OF MACHINE LEARNING

The history of Artificial Intelligence (AI) goes back to the era of World War II. This technology was fathered by John McCarthy sharing the credit with Alan Turing. Johan McCarthy was one of the greatest computer scientists while being an eminent cognitive scientist as well. Alan Turing's work in disrupting the German Enigma machine during the world war became the basis many of the recent developments in the scope of computer science (Copeland, 2000).

Machine Learning has got its root firmly planted to Artificial Intelligence. Machine Learning is basically a subset of Artificial Intelligence and was coined by Arthur Samuel. His work on training computers to play checkers was published in the late 1950s while he was working with IBM (Arthur, 1959). Machine Learning, being the sub-set of AI and computer science, which focuses on the use of data and algorithms to imitate the way that human brain learns and while gradually improving its accuracy. Machine Learning directly mimics the decision-making ability and processing capacity of the human conscience, it gives machine the ability to learn and develop in an automated way without any intervention of human. An important part of Machine Learning is Artificial Neural Networks (ANNs) (Fukushima, 1975; Fukushima, 1980) which is based on the theoretical structure of human neuron connection and interaction. It is also important to note that computing or artificial technologies have not yet been advanced enough to take over human intelligence but it do aim for reducing the computational time or

any kind of turn-around-time.

With the introduction of Deep Blue by IBM and AlphaGo by Google, we have witnessed several leaps in the development of Artificial Intelligence which has proved the capabilities of Machine Learning to solve real world and complex problems (David Silver, 2016; Huang et al., 2018). The exponential widespread of implementation of the Machine Learning technology is mostly attributed to the availability of huge quantity of datasets and the fine-tuning of the computational techniques which in turn reduces the over fitting and improves the trained models. The main driving force to the wide adoption of Machine Learning techniques are these two factors. Machine Learning models when coupled with the network of interconnected devices or the IoT systems creates a rich and efficient infrastructure to build a predictive and automated systems. Machine learning has become a primary method to understand the massive influx of health data in today's advanced infrastructure. Many used cases have already proved the promising and effective results of Machine Learning.

APPLICATION OF MACHINE LEARNING IN MEDICAL IMAGES

In today's medical practice the images from USG, CT scan, PET Scan or X-Ray are digital in nature. This becomes advantageous when it comes to effectively utilize such image data. To do so, there are several challenges which needs to be addressed. The medical imaging is a collection of techniques to create visual representations of the interior parts of the human body for diagnosis, analysis and appropriate medical intervention. The imaging techniques in healthcare system is preferred as the initial tool for clinical diagnosis in order to understand the internal conditions of a human body while also avoiding the risks like infections, strokes and other complications associated with surgical approaches.

The current standard of clinical practice depends on assessment of the medical images by trained physicians, pathologists or radiologists whose responsibility is to examine the images and conclude the root cause of a clinical ailment or patient's complaint. This standard of operation however does carry the risk of human error along with variations also incur recurrent costs and often demands years of expertise in determining the root causes. By the demonstration portrayed by Andrew Ng where he used images pulled from YouTube videos, it proves why medical image processing was one of the first to be taken up during the initial adoption of Machine Learning techniques in the healthcare system (Le, 2013).

The importance of accuracy in the diagnosis is crucial as because any misinterpretation or human error might lead to severe consequences, sometimes fatal. With the development of image processing, most of the architectures now depends fundamentally on Deep Learning (DL) and more specifically Artificial Neural Networks (ANNs). In the recent research and developments, the approach was to improvise the ANNs and utilize it in the form of Convolutional Neural Networks (CNNs) in order to enhance and boost the performance to optimal level when classifying the medical images. CNNs strongly holds the mast when it comes to object detection in the medical images (Greenspan et al., 2016) [8]. Acceleration of Graphic Processing Unit (GPU) became a concrete base for the deep development of CNNs towards efficiency.

However, a prominent challenge in establishing a competent model still persists. The biggest challenge is the need for a large quantity of annotated medical image data, the cost to collate and create such a database is often difficult as it demands the time of trained physicians to annotate the medical images. Along with this the patient's right to privacy intervenes negatively in the possibility to make such databases open-sourced. This bottleneck increases the risk of overfitting and also results in depletion of accuracy of the prediction models (Giger, 2018). Objections against the implementation of Machine Learning in the clinical diagnosis system have been desked on the basis of proper validation of the

prediction and analytical models. Validating the results with other datasets could be difficult due to the lack of a large enough reference datasets for a particular disease. And the effort needed to aggregate these data can take more time than actually training the model. The medical imaging data is naturally more difficult to collect and even more difficult to store and process.

Detection of Lesion and Computer Automated Detection (CAD) techniques

Currently Machine Learning has got the most common usage in medical healthcare system specifically for Computer Automated Detection (CAD) in the detection of lesions which are found in Brain scans, mammograms and other body scans (Lundervold & Lundervold, 2019). These techniques make use of CNNs to conclude down at the probability if a patient's lesion is in fact a lesion or not. It often uses several 2D slices of 3D rotational scans of either CAT or MRI or even USG in training the system. A variety of methods such as randomized rotation of the images or aligning the lesions in centre of the images. Considering the area of mammography, CAD technique have especially reached a state where it is being used as a "second opinion" for the clinical radiologists which is immensely helping in improving the accuracy of screenings without incurring additional costs associated with using a human as the "second opinion" (Fig. 1).

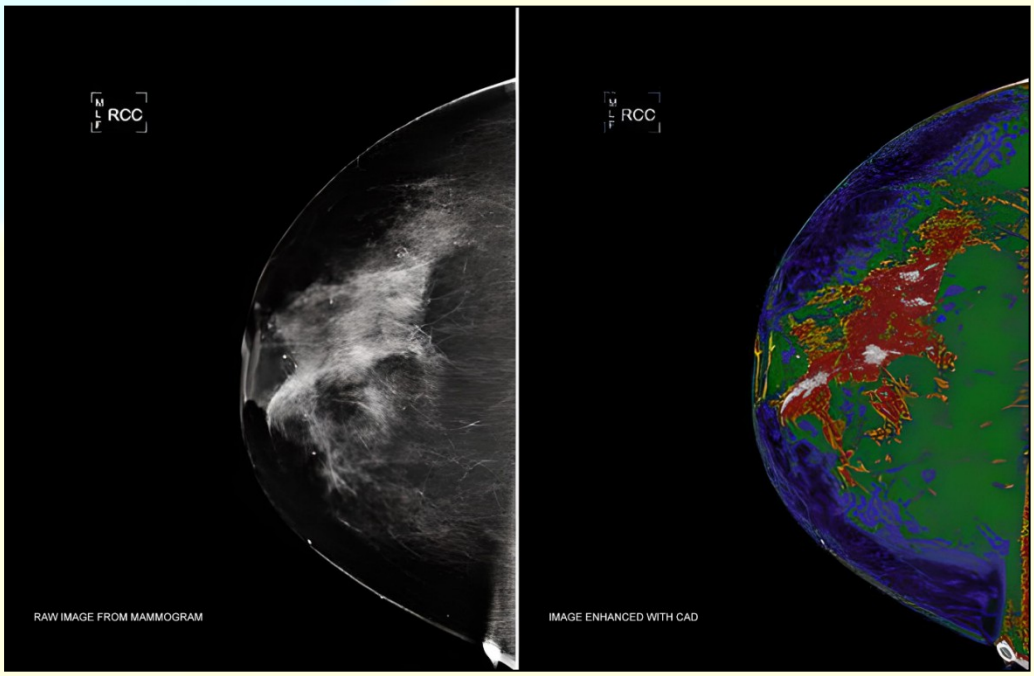

Fig. 1: Example of a mammogram where the left image being the raw mammogram while the right image is CAD enhanced using a NASA software which was originally used to enhance earth imagery. This picture is credited to Bartron Medical Imaging and is sourced from a NASA press release (Toh, 2021).

CAD is currently being used in the detection and diagnosis predominantly. While a lesion can be categorized being either benign or malignant using the knowledge of the physician and assessment however, the actual detection is crucial in the process of treating a patient. CAD helps with the actual recognition of potential lesions in a medical image. As an example, detection and segmentation of glioblastoma is quite a difficult task and due to the invasive and wide variety of these tumours. Deep Learning technology has aided by helping in automating the assessment of glioblastoma MRIs (Havaei et al., 2017).

Computer aided diagnosis system describes if a lesion is malignant in nature or not and is used to

improve the accuracy of diagnosis and also aid the process of early diagnosis in the clinical practice. This technological integration is already in use predominantly in brain related ailments due to the complex nature of assessing the brain health.

Application of machine learning in genetics for the prediction and analysis of complex diseases

Genetic engineering and study have seen a leap since around 2008 which carried huge volume of genetic information and datasets. This has created a pile of difficult challenges in the aspects of how to handle the exponentially growing volume of those data. The advancements in genetic sequencing speed, namely NGS technologies have fueled the increasing speed at which the whole human genome is sequenced. From the basic level of understanding, we know that the human genome is a complex structure that is responsible for all the information of human development, evolution and characteristics. The genomic structure is highly interconnected and decoding most of these is still a mystery to us. The diversity of the genomic structure between people adds on to the complexity of understanding the genetic interactions.

A lot of health care approaches have focused on acquiring large samples of human genomes in order to identify and help in understanding the statistical relevance of trends among the different population of human race. As we know already there are 23 chromosomes of the human genome which contains around 20,000 genes which have been identified to be the primary coding sequences which are responsible for the proteins necessary in building the biological components of the cellular structure (International Human Genome Sequencing, 2004). This count is an approximate estimate and some of the studies also estimates that there may be as many as 25,000 genes or as few as 19,000 as well (Ezkurdia et al., 2014). There is a large pool of genetic information in the human body that does not code for any proteins, which are not included in these estimates. A growing scale of research literature indicates that there are certain sections of what has been termed as genetic dark matter or missing heritability do exist (Insel, 2014). These terms refer to the portions of DNA which have no direct intervention in the protein coding process but may be relevant to the level of gene expression in a person's genetic code (Gibson & Dworkin, 2004). The levels of gene expression may cause difference in protein synthesis, may result into overload or even deficiency. Along with this, any structural differences in the physical arrangement of how the DNA is bound into chromosomes and then subsequently gets unwrapped during both the duplication process and translation process can also impact the level of gene expression.

As an example, to understand, methylation or acetylation of the DNA backbone can make it difficult (methylation) or easier (acetylation) to unwind the DNA strand during normal cell processes like replication or protein synthesis. Understanding this highly interconnected, complex and nonlinear network between all the different areas of the human genome structure is pretty difficult.

With the help of Machine Learning, researchers have taken a step towards finding the patterns and trends that can be modelled into a predictable manner. With the use of the exponentially growing volume of genetic data, machine learning has got the potential to predict accurately who might be at risk of getting certain diseases such as cancers and Alzheimer's disease.

Prediction of cancer by germline copy number variations

One of the exciting areas we would discuss in this chapter, is the utilization of the germline copy number variations in the prediction of different types of cancer. We can make use of Machine Learning models specifically the Gradient Boosting Machines (GBM), which is a form of Decision Trees (DT), to predict if a person has a particular type of cancer. The testing models built were found to be able to predict cancers such as Ovarian Cancer (OV) and Glioblastoma multiforme with an AUC (are under the ROC

curve, where ROC stands for Receiver Operating Characteristic) of 0.89 and 0.86 respectively (Toh & Brody, 2020) [18], using the copy number variation data taken from germline blood samples. The results indicated a significant inherited portion contributing to cancer risk in many. And since these CNV (copy number variation) data is taken from germline DNA, the probability of continued inheritance to future generations is high. This method does not only depend solely on Single Nucleotide Polymorphisms unlike other methods (Lello et al., 2019). This method takes the approach of a whole genome by averaging the copy numbers of an individual's entire genome as the basis of predicting carcinoma.

Future studies and researches are expected to improve the performance of these models and could possibly be used as a standalone tool to assess an individual's risk towards any diseases, understanding the fact that these models can be designed and generalized for predicting any fatal diseases. The progressive work encompasses other potentially complicated diseases which might have inherited trace or components responsible.

CONCLUSION

Integration and implementation of digital technologies like Machine Learning in the space of healthcare is approaching towards a revolutionary era. The amalgamation of bio-informatics, molecular biology, genetic engineering and computer science is taking the shape of such an infrastructure which would change the way traditional medical diagnosis, treatment, research and development is operating till now. Not only this but also it would facilitate our knowledge about heredity and environmental factors which are still unfurled and might be responsible for many of the complex diseases. The capability of making use of the copy number variations (CNVs) in the prediction of carcinoma and its diagnosis can be a breakthrough development in coming phase. Understanding how the genomic landscape works and how machine learning can be used to develop an interpretable method, interlinks across genes to decode the inherited carcinoma risks could potentially improve the healthcare system on an individual level of study. As medical imaging system is a non-invasive method of looking inside the human body, any prospect of improvements in this space would always be beneficial while reducing the need for risky surgical approaches.

A vast variety of IoT devices and storage infrastructure need to be upgraded and standardized in order to ensure a seamless exchange of data and processing taking into consideration the exponentially increasing volume of data being generated and collected. The fact about how widely the human genetic variation could contribute in predicting an individual's risk facilitates the patients and the doctors to make effective lifestyle changes in order to take a preventative approach. Similarly, the predictive models can also be an informative system for the physicians to help them take proper types of prognostics and diagnostics decisions which becomes specifically relevant to an individual patient while saving both time and money.

The field of studies like bioinformatics, genetic engineering, forensic science and radiology can be upgraded and made highly effective and beneficial with the integration of the emerging digital technologies like Artificial Intelligence, Machine Learning, NLP etc. These digital technologies not only make lengthy analysis easier but also provide wide exposure of improvement and development while reducing the cost, time and variations. Looking into the industrial developments happening at a blazing speed, it is high time that we invest into the futuristic approaches of studies to match up the market and quality demands.

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PALMYRA PALM: A GIFT OF NATURE

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INTRODUCTION

Palmyra tree is a tall and swaying tree well known as "*Borassus flabellifer*", simply called as 'Palmyra'. The word "Borassus" was derived from a Greek word and it means the leathery covering of the fruit and "flabellifer", means fan-bearer. Palmyra palm tree belongs to the 'palme' family, termed as 'Tala' in Odia, 'Tar' in Hindi, and Palmyra Palm in English. The tree was named originally for the resemblance of its leaf to the palm of the human hand. The tree is mentioned as used for multiple purposes in the ancient epic 'Ramayana', 'Srimad Bhagabat Purana' and in holy 'Ramcharita Manasa'. It is also mentioned in the writing of Pannini (400BC), the ancient grammarian of Sanskrit as well as in 'Pali' Buddhist Canon (5th century BC).

SYSTEMATIC POSITION

Kingdom	-	Plantae
(Unranked)	-	Angiosperms
(Unranked)	-	Monocots
(Unranked)	-	Commelinids
Order	-	Arecales
Family	-	Arecaceae
Sub-family	-	Coryphoideae
Tribe	-	Borasseae
Sub-tribe	-	Lataniinae
Genus	-	*Borassus*
Species	-	*Flabellifer*

COMMON NAME

Some common name of Palmyra tree are Fan palm, Asian Palmyra palm, Toddy palm, Sugar palm, Cambodian palm, Kerigi, Mak tan kok, Panna-maram, Taan, Than, Doub palm, Tala palm, Wine palm, Borassus palm, Great fan palm, African fan palm etc.

AREAS OF CULTIVATION:

Palmyra palms are native to tropical regions of Africa, Asia and New Guinea. They are economically useful and widely cultivated, especially in South–East Asia. The Palmyra palm tree has long been one of the most important trees of Cambodia and India. It is native to South and South-east Asia, in the Indo-Malaya eco-zone.

MORPHOLOGY

Size

Borassus flabellifer is a robust tree (Fig. 1a) and can reach a height of 30-35 m, trunk is mostly grey in colour, robust and ringed with leaf scars; old leaves remain attached to the trunk for several years before falling cleanly. The leaves are fan-shaped and 3-3.5 m long, with robust black teeth on the petiole margins. *Borassus* species are dioecious with male and female flowers on separate plants.

The male flowers are less than 1.5 cm long and form semi-circular clusters, which are hidden beneath scale-like bracts within the catkin-like inflorescences. In contrast, the female flowers are golf ball-sized and solitary, sitting upon the surface of the inflorescence axis. After pollination, these blooms develop into fleshy fruits 15-25 cm wide, each containing 1-3 seeds. The fruits are black to brown with sweet, fibrous pulp and each seed is enclosed within a woody endocarp. Young palmyra seedlings grow slowly, producing only a few leaves each year, but at an as yet undetermined time, they grow rapidly, producing a substantial stem.

Leaves

The *Borassus flabellifer* leaves are fan-shaped and 3-3.5m long, with robust black teeth on the petiole margins used for thatching, mats, baskets, fans, hats, umbrellas, and as writing material. All the literature of the old Tamil was written in preserved Palm leaves also known as Palm- leaf manuscript. It was written with a sharpened iron piece. Most of the ancient literature in Telugu were written on palm leaves. In Indonesia the leaves were used in the ancient culture as paper, known as “lontar”. In the eastern part of India, the leaves are used to make hand fans. These are mostly used during the summer in parts of Assam and West Bengal.

Trunk

The trunk is grey, robust and ringed with leaf scars; old leaves remain attached to the trunk for several years used to make fences and also produce a strong, wiry fibre suitable for cordage and brushes. The black timber is hard, heavy, and durable and is highly valued for construction.

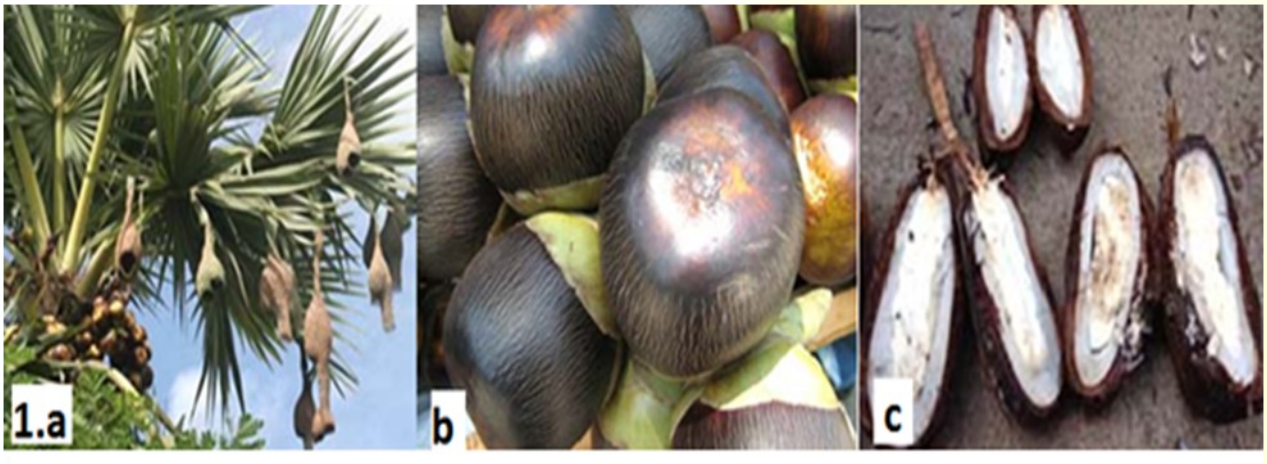
Fruit

The fruit measures 10-18 cm in diameter, has a black husk, and is borne in clusters (Fig. 1b). The top portion of the fruit must be cut off to reveal the sweet jelly seed sockets, translucent pale-white, similar to that of the lychee but with a milder flavour and no pit. The sweet jelly seed sockets occur in combinations of two, three or four seeds inside the fruit. The jelly part of the fruit is covered with a thin, yellowish-brown skin. Fruits contain watery fluid inside the fleshy white body. Conventionally, fruits are eaten unripe, but if the entire fruit is left to ripen, the fibrous outer layer of the palm fruits can also be eaten raw, boiled, or roasted. Bengali people have perfected to making various sweet dishes with the yellowish viscous jelly like substance obtained from a ripe palm fruit. These include Mustard oil fried Taler Bora, alternately fried in Sunflower oil, or mixed with thickened milk to form Taal-khee.

Sap

Sap traditionally is obtained after tapping the top shoots and the dripping juice is collected in hanging earthen pots. The juice so collected before morning is refreshing and light drink is extremely cool in sensation, and has a sugary sweet taste. The juice collected in evening or after fermentation becomes sour, and is called Tadi in Marathi. Tadi is consumed mostly by coastal villagers of Maharashtra as a raw alcoholic beverage. A sugary sap called Toddy, can be obtained from the young inflorescence.

Toddy is fermented to make a beverage called Arrack, or it is concentrated to a crude sugar called Jaggery or Taal Patali in Bengali. It is called *Gula Jawa* (Javanese sugar) in Indonesia, and is widely used in Javanese cuisine.



Sprouts

In Tamil Nadu, Andhra Pradesh, Telangana and Bihar and North eastern states of India, the seeds are planted and made to germinate and the fleshy stems (below the surface) are boiled or roasted and eaten. It is very fibrous and nutritious. The germinated seed's hard shell is also cut open to take out the crunchy kernel, which tastes like a sweeter water chestnut (Fig. 1c).

Fig.1 (a) A top portion of Palmyra pal, with fruits in clusters containing weaver bird's (Babui Pakhi) nests, (b) Fruits, (c) The germinated seed's hard shell is cut open to take out the crunchy kernel

IMPORTANCE

The white kernel of the ripe palm fruit after being left for a few months is used as an offering in Lakshmi Puja in various parts of Bengal and is also eaten raw. The plant is planted as a windbreak on the plains. It is also used as a natural shelter by birds, bats and wild animals. The palmyra tree is the official tree of Tamil Nadu. Highly respected in Tamil culture, it is called as "karpaha Veruksham" ("celestial tree") because all its parts have a use. Palmyra trees dotted the wilderness and banks of water bodies in Tamil Nadu. Products from the trees were used for food, wood, shelter and even as the source of toddy. So much was the prominence of the tree in Tamil Nadu that, palmyra was declared as the state tree of Tamil Nadu. The Asian palmyra palm is a symbol of Cambodia where it is a very common palm, found all over the country. It also grows near the Angkor Wat temple. In Indonesia the Palmyra tree is the symbol of South Sulawesi province. There is no need to water the seeds at regular intervals. A tree is capable of growing up to 100 feet in 10 years. A tree can last 100 years.

MEDICINAL USES

There are numerous medicinal uses for all parts of the Palmyra palm. The young plant is said to relieve biliousness, dysentery, and gonorrhoea. Young roots are diuretic and anthelmintic, and a decoction is given in certain respiratory diseases. The ash of the spadix is taken to relieve heartburn and enlarged spleen and liver. Sap from the flower stalk is prized as a tonic, diuretic, stimulant, laxative and anti-phlegmatic and amebicide. Sugar made from this sap is said to counteract poisoning, and it is prescribed in liver disorders. Candied, it is a remedy for coughs and various pulmonary complaints. Fresh toddy, heated to promote fermentation, is bandaged onto all kinds of ulcers. The pulp of the

mature fruit relieves dermatitis. Being rich in minerals and vitamins, sugar palm fruits are a healthy option for people on diet or suffering from diabetes. It is a rich source of vitamins B and C, iron, zinc, potassium, calcium, phosphorus, thiamine, and riboflavin. Palmyra fruit is an excellent home remedy for prickly heat during summer season. A thin layer of sugar palm fruit jelly applied on the affected area has a soothing effect and immediately alleviates the itchiness associated with prickly heat. Sugar palm fruits are also effective in reducing the symptoms of chicken pox and enhance the rate of healing. Sugar palm fruit is also beneficial in treating inflammatory skin problems such as redness due to intense heat. A face pack made from sugar palm fruit is excellent for the skin even for people with sensitive skin. It prevents prickly heat, boils and redness of-the-face. The palm fruit is ideal for treating burning sensation in the stomach. During summers use palm fruit, to keep body hydrated. It also replenishes the lost minerals and nutrients of the body and prevents painful urination and tiredness. Sugar palm fruit is a good option for those who are on a diet. It also prevents malnutrition in children and adults. Palm fruit has anti-inflammatory and anti-oxidant properties. The fruit pulp helps to cure skin inflammations. It is used to treat nausea and vomiting as well as worm infestation. It is used as an expectorant and also as a liver tonic. Palm jaggery is also known as Palmyra palm sugar and is a natural sweetener. It is used extensively in cooking and has many health benefits; (a) Low glycaemic index sugar helpful in reducing obesity and diabetes, (b) Provides a sustained and uniform energy supply to the body, (c) It is rich in many nutrients including vitamins B1, B2, B3, B6 and C. Pure sandalwood powder is mixed with ground fresh sugar palm fruit with a little coconut water. This pack is applied in a thin coat on the face and left to dry after which it is washed off with cold water. This is a good remedy for treating and preventing boils, prickly heat and redness of the face.

“The conservation of natural resources is the fundamental problem. Unless we solve that problem, it will avail us little to solve all others”. - *Theodore Roosevelt*.

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ANTIBIOTIC RESISTANCE: A WORLDWIDE CHALLENGE WITH HIGH MORBIDITY AND MORTALITY

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ABSTRACT

Antibiotic Resistance affects people of any age, in any country, making it one of the world's most urgent public health problems. In Gram-positive and Gram-negative bacteria, multidrug resistance patterns can be seen, which maybe untreatable with conventional antibiotics. As antibiotics become less effective, it is becoming hard to treat infections such as pneumonia, tuberculosis, gonorrhoea and salmonellosis, which are a result of chromosomal changes or the exchange of genetic material via plasmids and transposons in bacteria. This crisis is fuelled by the extensive use of antibiotics in the community and hospitals. In order to limit bacterial resistance, mechanisms such as antibiotic control programs, better hygiene, and synthesis of agents with improved antimicrobial activity need to be adopted.

Keywords: Antibiotic resistance; bacteria; public health; infections

INTRODUCTION

Historically a major cause of mortality was infectious diseases. During the 20th century, developments within medicine and public health helped to reduce the deaths associated with infectious diseases. The "golden era" of antibiotic development was heralded by the discovery of penicillin by Alexander Fleming, and it led to the introduction of many new antibiotics, causing many to believe that infectious diseases would soon be conquered. However, infectious diseases are again causing numerous deaths, especially those infectious diseases which can no longer be treated using the previously discovered antibiotics. Due to the ability to evolve, infectious pathogens have developed resistance to the currently prescribed and newly-developed antibiotics. *Streptococcus pyogenes*, *S. pneumoniae* and *Staphylococci* members of the Enterobacteriaceae family, which cause respiratory and cutaneous infections and members of the *Pseudomonas* family that cause urinary tract infection, diarrhea etc. are now resistant to virtually all of the older antibiotics.

As the early identification of causative microorganisms and their antimicrobial susceptibility patterns in patients with bacteria and other serious infections is lacking in many healthcare settings, broad-spectrum antibiotics are liberally and mostly unnecessarily used. There is a dramatic increase in emerging antibiotic resistance, and when it is coupled with poor infection control practices in the society, resistant bacteria can easily spread to other patients and finally to the environment. Most infections caused by resistant microorganisms fail to respond to conventional treatment, and even last-resort antibiotics have lost their power. A large number of studies are present which document levels of resistance and its clinical impact and there are concerns that both will rise in the foreseeable future.

ANTIBIOTIC RESISTANCE

The resistance towards antibiotic continues to increase while the pipeline is drying up for new antibiotic development. After the use of antibiotics for only seven decades, bacterial infections which were easily treated earlier are becoming untreatable. There are distinct factors that lead to the rise in antibiotic resistance globally. In developing nations, key contributors identified included: (1) Lack of surveillance of resistance development, (2) poor quality control of available antibiotics, (3) clinical misuse, and (4) ease of availability (Chokshi et al., 2019). There are a number of factors that contribute to antibiotic resistance in developed countries, which include poor hospital-level regulation and overuse of antibiotics in food-producing animals (Chokshi et al., 2019).

Antibiotic resistance in humans is also caused by antibiotic misuse, which occurs due to the use of antibiotics when not clinically indicated. When treating infections, proper diagnostic methods are often not utilized, and therefore, antibiotics are prescribed when not necessary. A study in a Chinese University hospital highlighted that of 1025 cases where antibiotics are prescribed, only 39 had a microbiological examination done to identify the source of the infection and nearly 77.8% of patients have prescribed one or more antibiotics (Hu et al., 2003). During a 2010 study from Mumbai, India, it was found that when 106 private practitioners were asked to treat a patient with pulmonary TB, 63 different drug regimens were prescribed, and only 6 of those were appropriate (Ab Rahman et al., 2016).

The use of antibiotics in food-producing animals is considered as being one of the major contributors to antibiotic-resistant infections. The significant use of antibiotics in food-producing animals and the transmission of drug-resistant species to humans is a cause for concern. In Turkey, a study was done on the presence of antibiotic-resistant *S. aureus* on meat and chicken samples and it shows that out of 80 *S. aureus* strains isolated and identified, 67.5% were found to be resistant to methicillin, and 87.5% were resistant to bacitracin (Gundogan et al., 2005). In fact, many studies that have been done globally all confirmed that the association between use of antibiotics in food-producing animals and the presence of antibiotic-resistant bacterial species that cause human infections come from the meat products of those animals (Control & Prevention, 2014). Moreover, the significant lack of research on new antibiotics affects both the developing and developed nations. A report published by WHO shows that the current antibiotics in clinical development are not sufficient to control the rising antimicrobial resistance, particularly the pathogens which are the greatest threat to human life (WHO, 2017). So, to keep the rising infectious diseases in check, new and more efficient antibiotics are a huge need in today's society.

CHALLENGE ASSOCIATED WITH ANTIBIOTIC RESISTANCE

Antimicrobial resistance in bacterial pathogens is a worldwide challenge associated with high morbidity and mortality. The organisms which are antibiotic resistant are known as superbugs. Over recent decades, multidrug-resistant (MDR) bacterial strains have been assumed to cause thousands of human deaths every year all around the world, which made antibiotic resistance the new worldwide fear (Abat et al., 2017). Antibiotic resistance affects both developed and developing countries equally so it is important to analyze antibiotic resistance development globally. Global consumption of more than 70 billion doses per annum of antibiotics has recently been estimated (Van Boeckel et al., 2014). Every year the cause of death of nearly 50,000 individuals is Methicillin-resistant *Staphylococcus aureus* (MRSA) in the United States and Europe alone along with many more dying from it in other settings. In developing countries antibiotic-resistant diseases such as tuberculosis (TB) have significant impacts. In 2013, there were approximately 480,000 cases of multidrug-resistant TB. It is predicted that if appropriate and necessary measures are not taken against antibiotic-resistant infections, then it can cause nearly 10 million deaths per year by 2050 (O'Neill, 2014). Over the last few decades penicillin has been used to treat pneumococcal infections which resulted in the decrease of worldwide mortality from pneumococcal pneumonia, dropping from 20–40% to only 5% which contributed to the decrease in number of deaths due to lower respiratory infections over the years but in the current years only a very small proportion of the pneumococcal strains that are currently isolated worldwide are resistant to penicillin (Rolain et al., 2016).

PREVENTION OF ANTIBIOTIC RESISTANCE

In order to combat antibiotic resistance, it is essential to have two key elements: (a) improving the regulatory framework to control antibiotic use globally, and (b) promote research on novel antibiotics. Globally it is essential to maintain proper surveillance of antibiotic resistance development. In some developing countries, it is necessary to further control the manufacturing process, quality, availability and use of antibiotics. Whereas in developed nations, hospital-based interventions and use of antibiotics in food-producing animals needs to be properly regulated. In addition, further research should be promoted in order to develop new antibiotics which can be used in the future.

In 2014, the WHO released the first-ever report, which mentioned antimicrobial resistance and it showed the national data on nine bacterial infections and antibiotic combinations (Reardon, 2014). The report WHO (2014) illustrated a comprehensive analysis of the worldwide antimicrobial resistance and showed the lack of coordination and significant gaps in surveillance concerning antibiotics, especially in many of the developing countries from where no national data was obtained but already having a high burden of antibiotic resistance (Reardon, 2014). The WHO has launched the Global AMR surveillance system (GLASS) to narrow the gaps in surveillance, and the surveillance system has enrolled more than one-fourth of the WHO member states until December 2017 (WHO, 2018). The WHO also organized World Health Day with the theme "Combat drug resistance: no action today means no cure tomorrow", resulting in an increased focus on antimicrobial research and the development of promising strategies to restore treatment options against infections by resistant resistance bacterial pathogens (Chellat et al., 2016).

CONCLUSION

As infectious pathogens are constantly evolving and developing antibiotic resistance, it is necessary to examine the key socioeconomic and political factors that contribute to the increase in antibiotic resistance in both developing and developed nations. Measures need to be taken at both national and international levels so that unnecessary use of antibiotics can be stopped and research work into developing more effective antibiotics as well as augment research into novel therapies need to be taken. Improving the situation concerning antibiotic resistance will require considerable changes to the ways in which global health statistics are collected as the approaches present now cannot live up to the task. But in future, if the situation improves the primary benefit will be a more accurate assessment of the global disease burden due to antibiotic resistance and its forward trajectory, helping make a case for investment in combating the problem.

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TOXIC CHEMICALS PRESENT IN COSMETICS, WITH SPECIAL REFERENCE TO SUNSCREEN CREAM

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ABSTRACT

Food, water, and various other ordinary products include chemicals and pollutants associated with skin and tissue damage. Many individuals are unaware that many common cosmetics contain elements that are detrimental to those who use them. Metals such as lead, arsenic, mercury, aluminium, zinc, calcium, and iron are found in many personal care items and cosmetics. These metals can be found in everyday items. Lipstick, whitening toothpaste, eyeliner, eye drops, foundations, sunscreens, nail color, and other cosmetics contain them. Some of these metals are added on purpose, while others are pollutants. Metal exposure has been related to toxicity in the reproductive, immunological, and neurological systems. A carcinogen is a phrase that refers to compounds that cause cancer. A common example of a carcinogen in a cosmetic product is formaldehyde. The possible side effects of exposure to these compounds include cancer, endocrine disruption, developmental and reproductive toxicity, bioaccumulation (accumulation on the skin) and ecotoxicity.

Keywords: Tissue damage; cosmetics; metals; sunscreens

INTRODUCTION

Everything that isn't a red flag ingredient is considered a non-toxic skincare ingredient. Hormone disruptors, carcinogens, and allergies are not present in these components. They are non-toxic and have not been classified as such by environmental agencies. The top dangerous cosmetics and skincare ingredients are listed below:

Sulfates: Sulfates are salts that result from the reaction of sulphuric acid (H_2SO_4) with another chemical. They are made from various sources, including petroleum and plant-based oils like coconut and palm oil. They function as surfactants and are used to lather. Sulfates have the potential to irritate your eyes and skin. They have the potential to prematurely remove the colour from your hair. The degradation of rainforests is caused by obtaining them from natural sources such as palm oil. They can be poisonous to aquatic organisms if they are washed away.

Phthalates: Phthalates are salts or plasticizing compounds used to improve a product's spread ability. Nail paints, fragrances, and lotions, to mention a few, contain them. They are also used as shampoo softeners and are reproductive and developmental poisons.

Parabens: Preservatives such as parabens are used to keep your skincare and makeup fresh and free of germs. They can be found in a wide range of items, including soaps, lotions, and cosmetics. Parabens enter the body through the skin and act like oestrogen, causing excessive cell division in the breast. Breast cancer develops as a result of this. They boost oestrogen (female sex hormone) production and interfere with reproductive and cognitive function.

Triclosan: Triclosan is a chemical found in toothpaste tubes, antibacterial soaps, and deodorants. It is antibacterial and efficient against germs, but it is also an endocrine disruptor and a skin irritant. In mammals, triclosan can cause intestinal inflammation and tumour formation.

Synthetic colors: Petroleum or coal tar are used to make synthetic colours. Hydrocarbons, carbon, and water make up coal tar. It is a viscous, thick, dark liquid with a distinct odour. Increased coal tar percentages are more likely when the pigment is heavier. Synthetic colours have been linked to skin irritations, cancer, acne breakouts, and attention deficit hyperactivity disorder (ADHD) (attention deficit hyperactivity disorder).

Fragrance: Perfumes, moisturizers, shampoos, cleansers, and conditioners are all examples of skincare products that contain fragrances. They're produced using chemicals that have been linked to respiratory problems, skin allergies, dermatitis, and reproductive system adverse effects. Fragrances are carcinogens (agents that cause cancer), irritants, and endocrine disruptors.

Formaldehyde: In skincare, formaldehyde is a common preservative. It's a colourless gas that stops bacteria from growing. Nail polishes, hair straightening treatments, hair gels, nail hardeners, shampoos, deodorants, lotions, and cosmetics all contain formaldehyde. Hair loss, scalp burns, asthma, and neurotoxicity have all been linked to it. Formaldehyde can make you dizzy and make you suffocate if you breathe it in.

Chemicals in sunscreens: Sunscreens contain chemicals such as PABA, benzophenone, oxybenzone, ethoxycinnmate, and homosalate. They are thought to absorb light, but instead of protecting the body, they harm it. These compounds in sunscreen are endocrine disruptors.

Talc: Talc is the softest mineral on the planet. Talc is commonly used in baby powders, eye shadows, blush, deodorants, and some soaps and is used to absorb moisture. Talc has a direct link to ovarian cancer. Talc can induce lung cancers when breathed.

Lead: Lipsticks, eyeliners, foundation, and whitening toothpastes all have lead in them. Cosmetics are regulated by the United States Food and Drug Administration (FDA), which allows lead in concentrations ranging from 0 to 20 parts per million (ppm).

Polyethylene glycol (PEG): PEG is a thickening ingredient found in cosmetics such as lotions, sunscreens, and shampoos. It has the potential to cause cancer and respiratory problems. It can also deplete your skin of natural oils (sebum) and cause your sebaceous glands (oil-producing glands) to produce more sebum, making your skin greasy.

Diethanolamine: It's a foamy agent found in body washes, shampoos, cleansers, and bubble bath. It's a cancer-causing substance as well as a respiratory toxin.

Alcohol: In skincare, alcohol (drying alcohol) can make the skin dry and flaky. It interferes with the skin's natural renewal process. Fatty alcohols, which are made from natural fats and oils, are excellent moisturizers.

Hydroquinone: Hydroquinone is a skin lightening agent used in cosmetics. Acne scars, freckles, melasma, age spots, and post-inflammatory hyperpigmentation are all treated with this product. Hydroquinone works by lowering the number of cells that produce melanin pigment (melanocytes). Because it lowers melanocytes to a large extent, prolonged use can result in skin whitening.

Petrolatum: Petrolatum is a softening agent that is ideal for dry skin. Petrolatum is found in lip balms and moisturizers. It provides a barrier that keeps water from escaping while also preventing moisture from the air from being absorbed.

Nowadays, there are many cosmetic products like eyeliners, lipsticks, hair products, makeup, sunscreen, deodorant, perfumes, and creams. We will mainly deal with sunscreen cream in detail.

SUNSCREEN

Sunscreen lotions contain a variety of characteristics that protect our skin from harmful ultraviolet (UV) radiation and prevent the damage caused by sun exposure. The higher the potential for light radiation to cause biological damage, the shorter the wavelength. UVA1, UVA2, and UVB rays are all blocked by sunscreen filters. Chemical filters absorb high-intensity UV radiation, causing higher-energy states to be excited. When these molecules return to their ground states, the absorbed energy is converted into lower-energy wavelengths. BHA (butylated hydroxyanisole) and BHT (butylated hydroxytoluene) are two more preservatives that act as endocrine disruptors when consumed or absorbed. Chemical sunscreens absorb the sun's rays like a sponge. Harmful chemicals, namely, oxybenzone, avobenzone, octisalate, octocrylene, homosalate or octinoxate (harmful) as one or more active components. Endometriosis has been connected to female exposure to oxybenzone and other similar substances. These sunscreens are easier to apply and don't leave a white film on your skin. Physical sunscreens (also known as mineral sunscreens) operate as a barrier between you and the sun. They deflect the sun's rays by sitting on the surface of your skin. Safer chemicals include titanium dioxide, zinc oxide, or both as active components. Sunscreen agents should be safe, chemically inert, non-irritating, non-toxic, photostable, and capable of providing total protection against solar radiation damage to the skin. The substances should stay on the skin's upper layers even after sweating and swimming. It should be able to scavenge singlet oxygen and other reactive oxygen species effectively. They should also effectively filter both UVB and UVA rays, which should be attainable with an SPF of 30 or above. This type of ingredients of sunscreen is safer to use if you have sensitive skin. Anything that is "designed to diagnose, cure, mitigate, treat, or prevent disease" is classified as a drug by the FDA. Sunscreen is considered a medicine because it has the ability to prevent sunburn, reduce the risk of skin cancer, and reduce early skin ageing. Titanium dioxide absorbs UVB and some UVA rays but does not always give complete UVA protection. When titanium dioxide is non-nanoparticle, it is harmless for people and the environment. Zinc oxide is a UV absorber that is found in nature. Zinc oxide protects against UVA and UVB radiation, providing broad-spectrum protection. Zinc is harmless for humans and environment when it is not a nanoparticle. These are less harmful sunscreen compounds. The SPF, or Sun Protection Factor, is a number that is assigned to your sunscreen or other sun cream to signify the level of sun protection it gives. Higher SPF products are more powerful and provide a higher proportion of protection. It's a frequent fallacy that increasing the SPF by two will provide double the protection. The SPF value indicates how long a sunscreen can protect your skin from sun damage.

CONCLUSION

Humans are protected from two types of UV rays by using sunscreen. UVA light has a longer wavelength and is the type of ray linked to skin ageing prematurely. UVB radiation has a shorter wavelength, and it is the ray that causes sunburn and skin damage. It's critical to be protected from both. Avoid the sun's harshest rays. If possible, avoid the sun between the hours of 10 a.m. and 2 p.m. The sun protection factor (SPF) measures how well your skin protects you from the sun. A higher SPF may not imply a considerable increase in coverage. UVB rays are blocked by SPF 15 in 93% of cases. UVB rays are blocked by SPF 30 in 97% of cases. UVB rays are blocked by SPF 50 in 98 percent of cases. UVB rays are blocked by SPF 100 for 99 percent of the time. SPF is a measurement of UVB ray protection only. It's also crucial to protect yourself from UVA radiation, so look for a "wide spectrum" sunscreen that shields you from both UVA and UVB rays. So, never purchase any cosmetic because of its brand value, which might affect your skin.

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