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Preventing Coronavirus Spread

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From the Publisher's Desk

Welcome to Biotechnology Kiosk!

We are ready with another issue of Biotechnology Kiosk (BK) for our readers with the regular features that include high-end editorials by experts, biotechnology advances around the world and industry news from pharma and biotech sectors.

This issue contains a number of scholarly editorials and news and views on the current cutting edge topics including antiviral drug and vaccine developments for the novel Coronavirus (COVID-19), big data in healthcare, reproductive biology, cellular engineering, plant biotechnology and stem cells and bone regeneration among other topics and many more reporting on research breakthroughs from around the world.

We are happy to announce that we have now launched a dedicated channel on YouTube for Biotechnology Kiosk. Please check out the YouTube channel of BK and subscribe to it. The magazine is now a member of CrossRef. BK recently added two new distinguished editorial board members, Prof. H. Khoshbouei and Prof. A. Sfera to the board. We are also now open to consider manuscripts in all areas of biotechnology for

publication in BK. Please go to the sections 'Aims and Scope' to submit your manuscripts to BK.

We are once again proud to announce partnering with a very well-known trade magazine in the United States, Vacuum Technology & Coating (https://www.vtcmag.com/) to launch a special edition of Vacuum Advances in Biotechnology. Please check out the call for papers that is posted to BK's website. We look forward to receiving your contributions. We do hope that you will enjoy reading this issue of Biotechnology Kiosk. Please do write to us with your comments and feedback. Your suggestions are always appreciated.

Dr. Megha Agrawal and Dr. Shyamasri Biswas

Co Editors-in-Chief, Biotechnology Kiosk

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Antiviral Drugs

By Megha Agrawal, PhD

Co Editor-in-Chief

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Can Antiviral Drugs Stop Coronavirus (COVID-19)?

Preventing Coronavirus Spread

The recent emergence of a novel, pathogenic coronavirus known as Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2, officially COVID-19) in China and its subsequent rapid spread across the world has put global health at risk. Some early research data suggest that the cell entry of coronaviruses depends on the process of binding of the viral spike (S) proteins to cellular receptors along with S protein priming by host cell proteases as depicted in Figure 1 [1]. Further, the latest understanding tells us that angiotensin-converting enzyme 2 (ACE2) is the cellular receptor for SARS coronavirus and the new coronavirus, SARS-CoV-2, which is causing the epidemic COVID-19 [2]. Current research has indicated the need to focus more on understanding the cellular factors that are used by SARS-CoV-2 for entry that could provide vital insights into viral transmission and subsequently lead to reveal therapeutic targets [1].

Previous studies have suggested that members of the family Coronaviridae can circulate in the human population that results in mild respiratory disease [3]. However, the serious forms of respiratory disease such as the SARS-CoV and the Middle East respiratory syndrome coronavirus (MERS-CoV) occur via transmission from animals to humans and that can cause severe respiratory diseases [1]. In view of the rapid spread of coronavirus, a global effort is currently initiated that is focused on developing vaccines and also identifying other effective treatment approaches that employ drug agents with demonstrated antiviral activity against SARSCoV and MERS-CoV or related positive-sense RNA viruses [4]. To this end, researchers demonstrated the effectiveness of Camostat mesilate for its protection mechanisms against COVID-19 [1]. Figure 1 illustrates how SARS-CoV-2 uses the SARS-CoV receptor ACE2 for entry and the serine protease TMPRSS2 for S protein priming [1, 4]. Recent data using drug agents demonstrated that a TMPRSS2 inhibitor which is approved for clinical use can block entry. This might pave the way for an effective treatment option (Figure 1) [1].

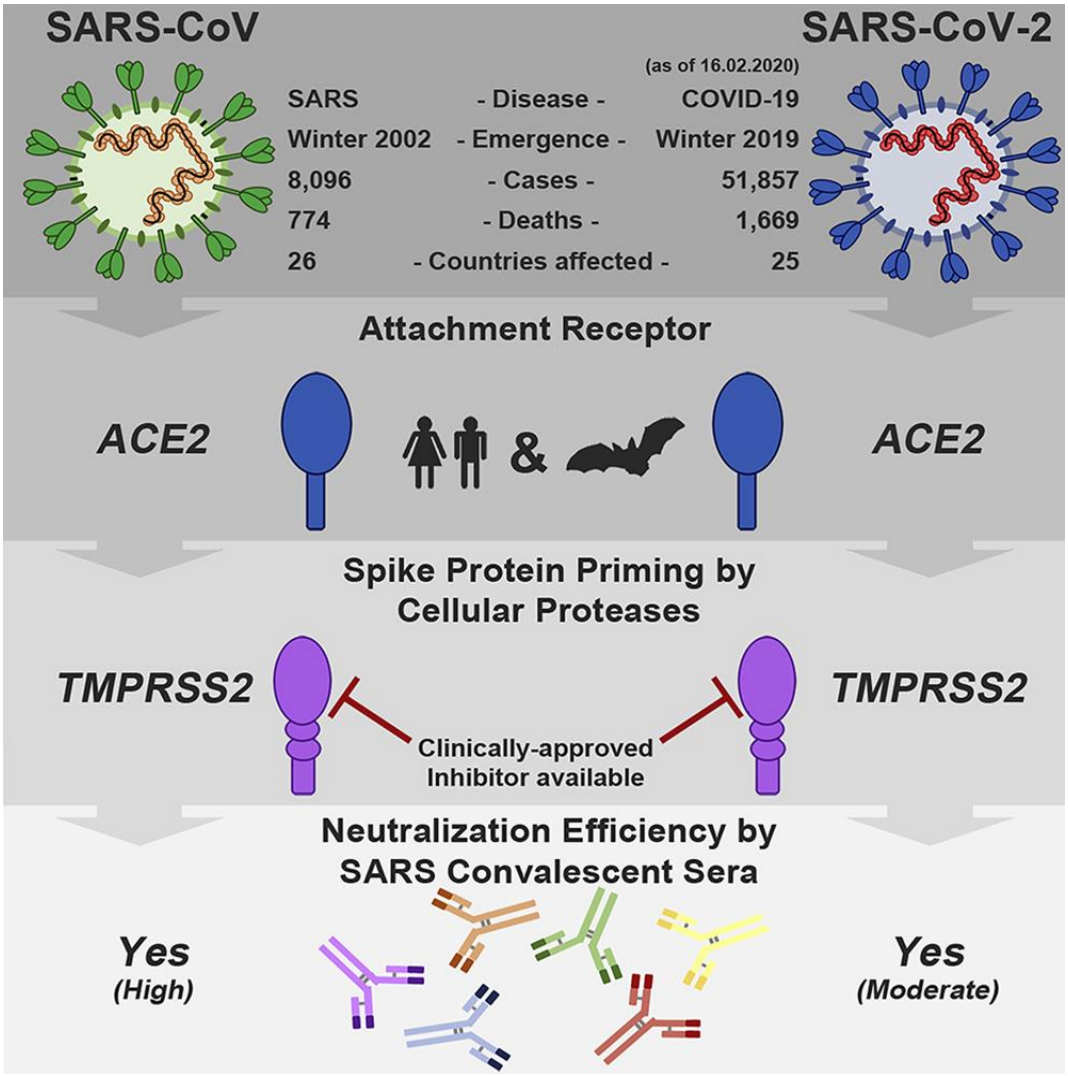

Figure 1: A schematic illustration showing possible mechanisms of cell entry of coronaviruses and the therapeutic actions of inhibitor [Source: Cell (2020)].

Repurposing Drugs to Battle Coronavirus

The latest preclinical data shown by researchers that uses the nucleotide analogue Remdesivir (RDV) are shown to be promising to combat coronavirus [4]. It has been shown that the RDV compound exhibit a broad range of antiviral activities against several RNA viruses [5-7], and also in battling SARS-CoV and MERS-CoV [4]. Some other studies have shed light on the effectiveness of RDV for its active actions against coronaviruses with divergent RNA-dependent RNA polymerases (RdRp) [4, 8]. The optimistic view of the drug remdesivir indicates that this compound might work for the pathogen responsible for COVID-19. However, there are not enough biochemical data available yet that can support these findings and also provide a possible mechanism of action [4].

This pre-clinical study done on the compound RDV has suggested that the

compound works by inhibiting an enzyme known as an RNA-dependent RNA polymerase. This polymerase is used by many RNA viruses including coronaviruses to replicate themselves [4]. In contrast, other RNA viruses that are known as retroviruses (HIV) use an enzyme called reverse transcriptase. This subsequently creates DNA from an RNA blueprint. This implies that the antiviral drug compound RDV has significant potential to be effective since coronaviruses also use RNA-dependent enzymes [4].

In this study, researchers co-expressed the MERS-CoV nonstructural proteins nsp5, nsp7, nsp8, and nsp12 (RdRp) in insect cells. This was chosen as a part of a polyprotein to study the mechanism of inhibition of MERS-CoV RdRp by RDV [4]. The triphosphate form of the inhibitor (RDV-TP) was shown to be competing with its natural counterpart ATP [4]. Based on the data obtained, researchers explained the high potency of RDV against RNA viruses in cell-based assays [4]. Researchers also showed that the sera from patients having convalescent SARS could be used to cross-neutralize SARS-2-S-driven entry. It was also shown that once the drug was incorporated into the growing RNA chain, the virus no longer replicated. These early results are promising that reveal key similarities between SARS-CoV-2 and SARS-CoV infection and that can be leveraged to identify a potential target for antiviral intervention [4].

Concluding Remarks

The early pre-clinical trials using antiviral drugs have shown promise in blocking COVID-19. Further, latest breakthroughs on structure-based rational design of binders with enhanced affinities to either ACE2 or the S protein of the coronaviruses can be leveraged for the development of vaccines. All these steps are believed to lead to the suppression of viral infection of coronavirus.

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Structural Biology & Vaccines

By Shyamasri Biswas, PhD

Co Editor-in-Chief

Biotechnology Kiosk, **2**, 3 (2020)

DOI: https://doi.org/10.37756/bk.20.2.3.2

Structural Pathways to Developing Vaccines against Novel Coronavirus (SARS-CoV-2)

The Emergence of Novel Coronavirus SARS-CoV-2

Coronaviruses belong to the family of RNA viruses that are known to enter human cells when their spike glycoprotein binds cell surface receptors. The recently emerged novel severe acute respiratory syndrome coronavirus (SARS-CoV-2; officially called COVID-19) can bind the angiotensin-converting enzyme 2 (ACE2) on human cells with much higher affinity than the virus that caused SARS in 2003 [1]. The current worldwide pandemic caused by COVID-19 was first identified and confirmed on December 29, 2019 that was traced to the Huanan Seafood Wholesale Market, Wuhan city, Hubei Province in China [1]. Studies have shown that COVID-19 spreads rapidly by human-to-human transmission with a median incubation period of 3.0 days (range, 0 to 24.0), and the time from symptom onset to developing pneumonia is about 4.0 days (range, 2.0 to 7.0) [2]. The conventional routes of transmission of SARS-CoV-2 are believed to be through respiratory droplets and direct contact along with the less known route of fecal-to-oral transmission as well [1, 2].

How to Block the Spread of SARS-CoV-2

To control or block the spread of SARS-CoV-2, there are no specific antiviral treatments or vaccines available yet primarily due to the reason that it is a new emerging viral disease. To mitigate the global health risk posed by the rapid spread of COVID-19, the current research has focused on developing safe and effective vaccines for the treatment of SARS-CoV-2 [1, 2]. To this end, researchers are assuming that virus-based vaccines would prove to be valuable in combatting COVID-19. In this regard, the CoV spike (S) glycoprotein is considered to be a key target for vaccines, therapeutic antibodies, and diagnostics [3].

Besides focusing on the entire virus particle-associated inactivated or attenuated viral vaccines, researchers are also employing the subunit candidates, such as S1 protein and/or the receptor-binding domain (RBD) element of SARS-CoV-2 that are considered as valuable targets for vaccine design [3]. It is believed that a deeper

understanding of the atomic-level structure of the COVID-19 spike protein will allow for additional protein engineering efforts that could be leveraged to improve antigenicity and protein expression for vaccine development. This information could pave the way for precision vaccine design and ultimately discovery of anti-viral therapeutics that would accelerate medical countermeasure (MCM) development [3].

As for the expected timeline of vaccine development, it is anticipated that a new SARS-CoV-2-based vaccine from gene sequence to clinical testing should happen in approximately 6-8 months [1].

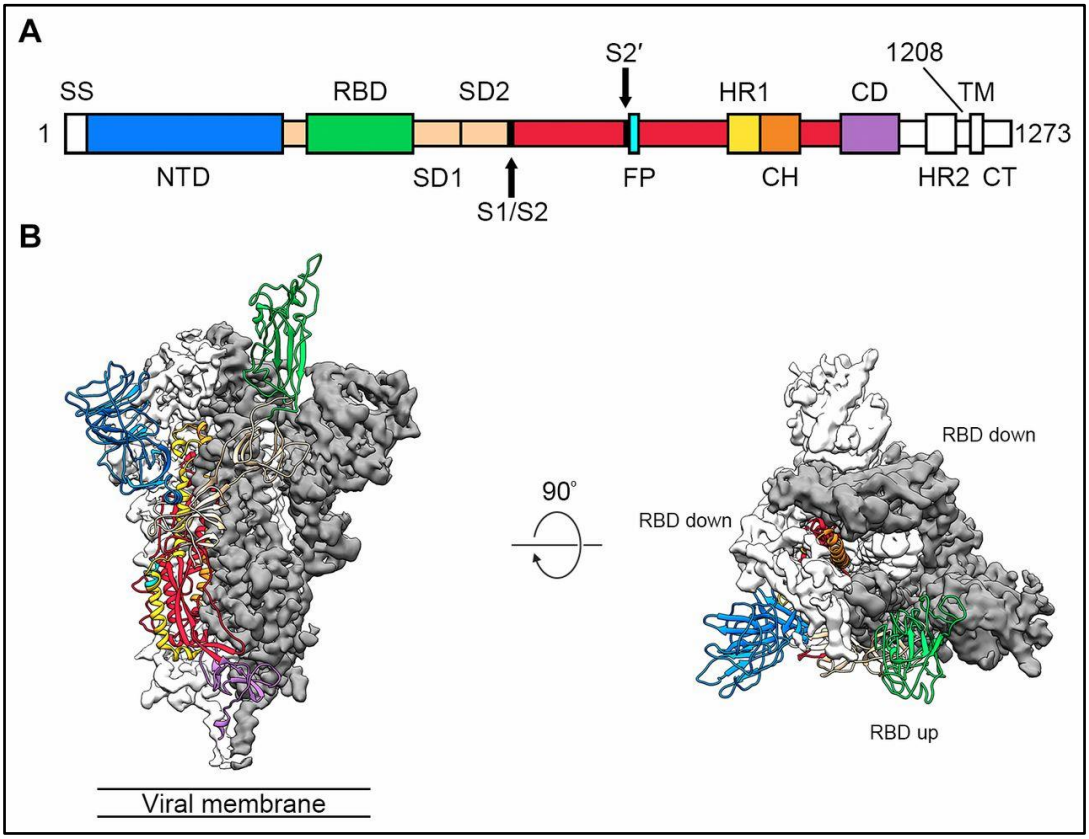

Figure 1: Structure of SARS-CoV-2 S in the prefusion conformation obtained by Cryo-EM. (A) Schematic representation of SARS-CoV-2 S primary structure that is colored by domain (the ectodomain expression construct are colored white). SS represents signal sequence and NTD represents N-terminal domain. RBD is receptor-binding domain and SD1 represents subdomain 1, whereas SD2 corresponds to subdomain 2, S1/S2= S1/S2 belong to protease cleavage site and S2' is the S2' protease cleavage site. FP corresponds to fusion peptide and HR1 is heptad repeat 1. Further, CH is the central helix and CD: connector domain. HR2 is heptad repeat 2 and TM is transmembrane domain, whereas CT corresponds to cytoplasmic tail. (B) Side and top views of the prefusion structure of the SARS-CoV-2 S protein with a single RBD in the up conformation. The two RBD-down protomers are shown as cryo-EM density in either white or gray. The RBD-up protomer is shown in ribbons which is colored corresponding to the schematic shown in (A) [Source: Science (2020)].

Solving Structure of Key Coronavirus Protein: A Significant Step Forward to Developing Vaccines

As we described in the preceding section, COVID-19 virus makes use of a densely glycosylated S protein to gain entry into host cells. The S protein is known to be a trimeric class I fusion protein, which exists in a metastable prefusion conformation state [3, 4]. This conformation subsequently undergoes a dramatic structural rearrangement that helps fuse the viral membrane with the host-cell membrane, which is triggered by the binding of S1 subunit to a host-cell receptor (Figure 1) [3]. This process is then followed by destabilizing of the prefusion trimer by the receptor binding that results in shedding of the S1 subunit and transition of the S2 subunit to a stable post fusion conformation (Figure 1) [3].

Further, studies have shown that the RBD of S1 undergoes hinge-like conformational movements, which transiently hide or expose the determinants of receptor binding in order to engage a host-cell receptor [3]. Since the S protein offers indispensable function, it represents a target for antibody-mediated neutralization. Simultaneously, the characterization of the prefusion S structure can provide atomic-level information to guide vaccine design and development [3]. To this end, to rapidly facilitate MCM development, researchers in a recent research of major significance, determined a 3.5 Angstrom resolution structure of the SARS-CoV-2 S trimer by employing a cryo-EM in the prefusion conformation (Figure 1) [3].

Researchers expressed ectodomain residues 1−1208 of SARS-CoV-2 S and added two stabilizing proline mutations in the C-terminal S2 fusion machinery based on a previous stabilization strategy that produced the domain organization of the expression construct and figure S1 shows the cryo-EM structure of the S-domain(Figure 1) [3]. This strategy proved effective for other betacoronavirus S proteins [3].

This study on solving the key protein structure of SARS-CoV-2 virus is considered a major breakthrough that results in a significant step toward developing a vaccine against the virus as well as treatments for COVID-19 [3].

Concluding Remarks

Solving the key protein structure of SARS-CoV-2 virus is a major step forward to rapidly develop vaccines and also therapeutics. We anticipate a rapid progress in vaccine development happening in the near future.

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Big Data

By Peeyush Prasad, MSc

Contributing Editor

How We Can Better Understand Biological Data

The Promise of Big Data

One of the problems with big data is making sense out of it. Technology such as proteomics, genomics, transcriptomics and metabolomics has generated large data set. Understanding these dataset and reaching to important and significant conclusion is very difficult. We often see how we arrive at insignificant drug target and biomarker after producing proteomics or genomics data. One of the reason for this is we lack proper understanding of molecular dynamics and our biased approach towards highly upregulated and downregulated molecules. This comes from lack of proper training and in depth knowledge towards cellular behavior, several negative and positive feedback loop and inability to capture the interaction between extra and intracellular environment. Therefore, it is important for students, researchers and scientists to have proper understanding of cellular behavior and parameters it depends on. Story of biological data does not stop at up or downregulated molecules and it goes much deeper than this. If there are 25000-30000 genes and if on an average there are two splice variants per gene then one can imagine the number of proteins coming from a genome. Further, think about different post translational modifications, peptides, miRNAs and metabolites that are oscillating or moving among different bioenergetic pathways. So complexity of biological data is often underestimated by researchers and we are in a loop of producing data after data without understanding it. From economical point of view also it is not fruitful. LC-MS based analysis of one peptide fraction cost around 300-400 dollar and similar is the case with genomics and transcriptomics experiments. On the other hand there are several questions also which need to be answered such as quality and significance of data coming from pooled sample. Age, gender and stage of any disease affect the quality of data and whether that data can act as representative for a particular disease. Disease-disease relationship has been established among various diseases by looking at common and unique factors deregulated in different disease conditions. Therefore, there is the need for proper

knowledge of different tools that can be employed for understanding large biological dataset. Here, specifically we will see about different network and pathway analysis tools along with platforms where one can look for specific gene or protein information. One of the key of working with these databases or tools is to have a proper understanding of cellular behavior.

Table 1. Important Systems Biology Tool for biological data analysis

Systems Biology Tool	**Function**	**Link**
3Omics	Tool for visualizing and integrating transcriptomic, proteomic and metabolomic human data.	https://3omics.cmdm.tw/
Phenotimer	Visualization tool for time-resolved biological data. It shows the time dependent phenotypic connections where network highlight the genes or functions corresponding to a particular time.	https://phenotimer.org/
GESTALT Workbench	This tool does the genomic sequence analysis, comparison and annotation along with visualization.	http://db.systemsbiology.net/gestalt/
Cell Designer	It performs the task of biochemical gene regulatory networks modeling.	http://www.celldesigner.org/
Morpheus	It does the modeling and simulation of environment for multiscale and multicellular system.	https://imc.zih.tu-dresden.de/wiki/morpheus/doku.php
Network portal	Analysis and visualization tool for gene regulatory networks.	http://networks.systemsbiology.net/
Cytoscape	It performs the network data integration, analysis and visualization	https://cytoscape.org/
AltAnalyze	Intuitive graphical user interface. Performs the transcriptome analysis	http://www.altanalyze.org/

Network and Pathway Analysis Tools

One of the very well-known tool for protein-protein interaction is STRING database where one can give query for single protein and multiple proteins. STRING database gives the idea about the protein-protein interaction but key is to look for gene ontology analysis it performs.

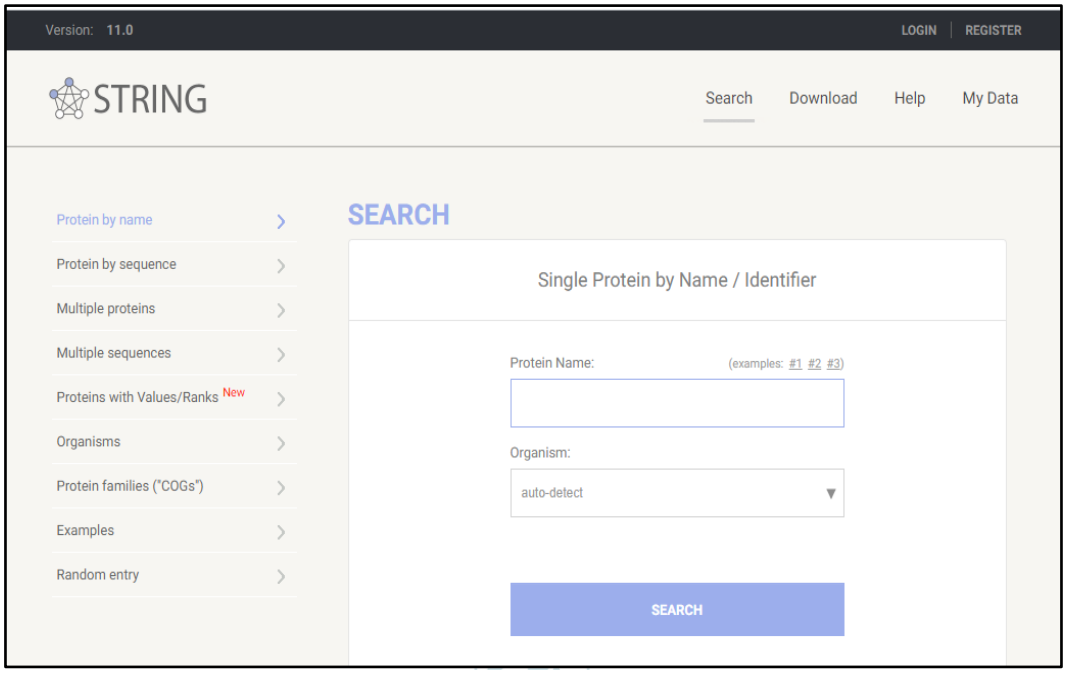

Figure 1. STRING database [Source: https://string-db.org/].

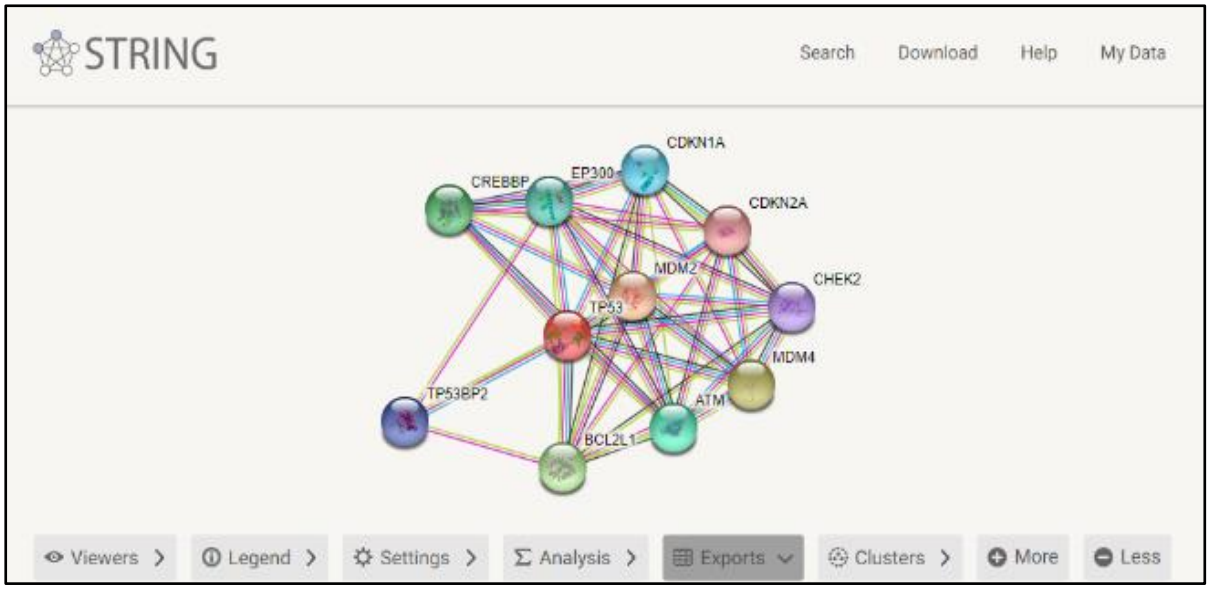

Figure 2. Interactors of TP53 protein [Source: https://string-db.org/cgi/network.pl?taskId= Led99vlDTilR

Network Stats

number of nodes:	11	expected number of edges:	22
number of edges:	46	PPI enrichment p-value:	4.66e-06
average node degree:	8.36	*your network has significantly more interactions than expected (what does that mean?)*	
avg. local clustering coefficient:	0.91		

Functional enrichments in your network *Note: some enrichments may be expected here (why?)*

Biological Process (GO)

GO-term	*description*	*count in gene set*	*false discovery rate*
GO:0071158	positive regulation of cell cycle arrest	8 of 85	3.93e-14
GO:2000045	regulation of G1/S transition of mitotic cell cycle	8 of 148	3.01e-13
GO:1901796	regulation of signal transduction by p53 class mediator	8 of 129	3.01e-13
GO:0072331	signal transduction by p53 class mediator	8 of 128	3.01e-13
GO:0071478	cellular response to radiation	8 of 153	3.01e-13
			(more ...)

Molecular Function (GO)

GO-term	*description*	*count in gene set*	*false discovery rate*
GO:0002039	p53 binding	6 of 73	2.72e-10
GO:0097718	disordered domain specific binding	3 of 33	7.84e-05
GO:0016740	transferase activity	8 of 2250	0.00021
GO:0005515	protein binding	11 of 6605	0.00026
GO:0003684	damaged DNA binding	3 of 64	0.00026
			(more ...)

Cellular Component (GO)

Figure 3. Gene Ontology (GO) analysis performed by STRING when TP53 was put as query protein [Source: https://string-db.org/cgi/network.pl?taskId=Led99vlDTilR].

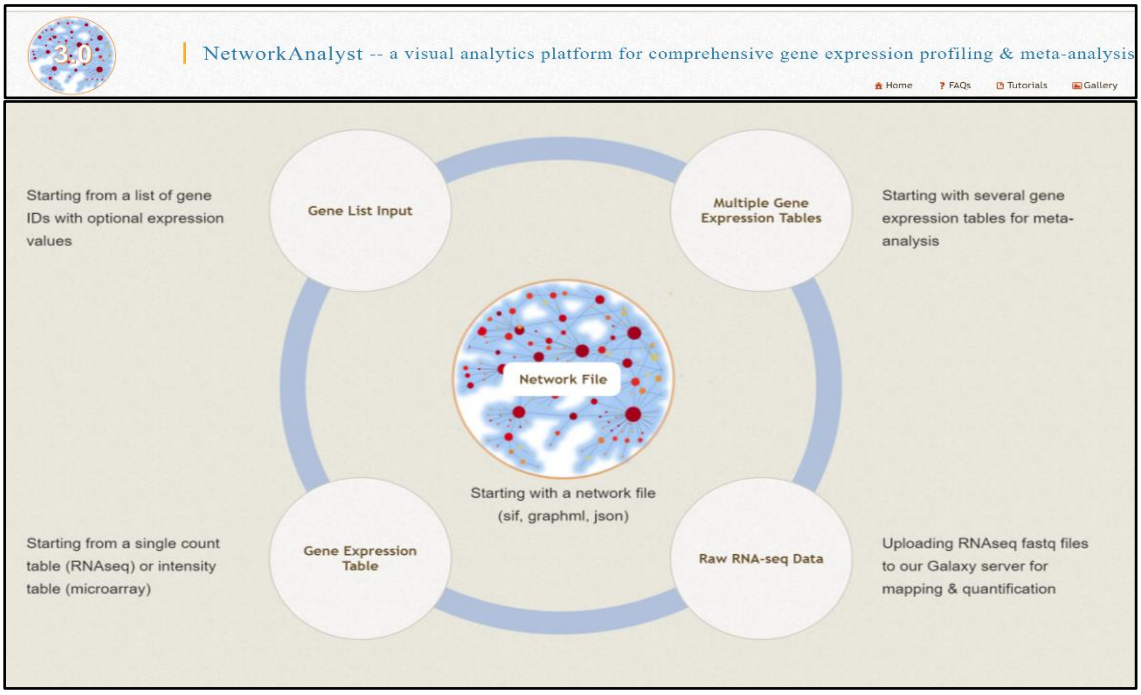

Figure 4. Network Analyst tool [Source: https://www.networkanalyst.ca/]

Another database for network analysis is Network Analyst which gives very insightful information regarding node and hub.

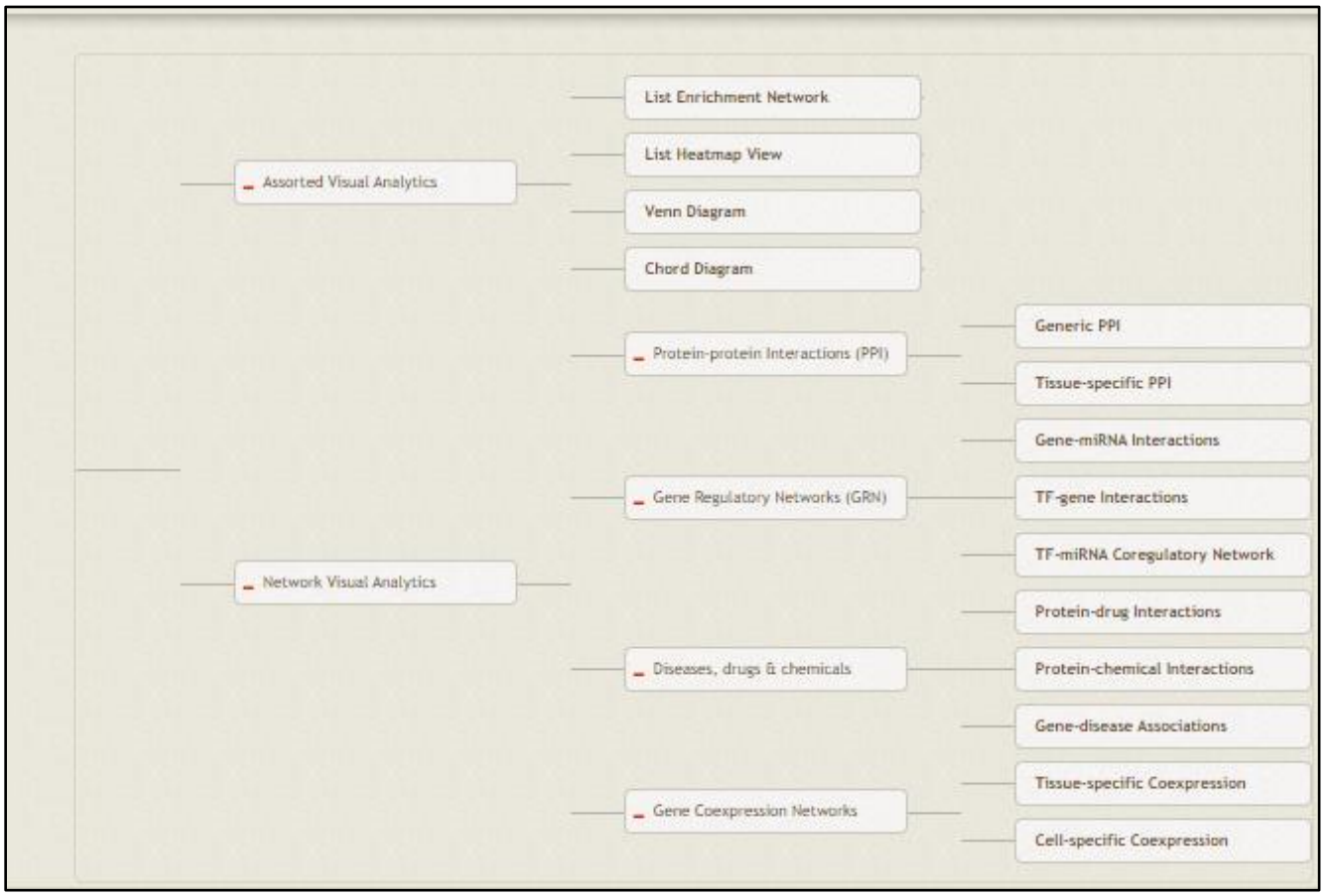

Figure 5. Different types of information such as gene regulatory network and TF-miRNA coregulatory network can be explored by using this tool [Source: https://www.networkanalyst.ca/NetworkAnalyst/Secure/network/ListAnalOverview.xhtmL]

List of important databases or tools for exploring information about single entity or set of large data.

1. NetPath (http://www.netpath.org/)
2. HPA (https://www.proteinatlas.org/)
3. Reactome (https://reactome.org/)
4. Ingenuity Pathway Analysis (https://www.qiagen.com/id/products/discovery-and-translational-research/next-generation-sequencing/informatics-and-data/interpretation-content-databases/ingenuity-pathway-analysis/?clear=true#orderinginformation)
5. GeneSpring (https://www.agilent.com/en/products/software-informatics/life-sciences-informatics/genespring-gx)
6. DisGeNet (https://www.disgenet.org/)
7. LocDB (https://www.rostlab.org/services/locDB/

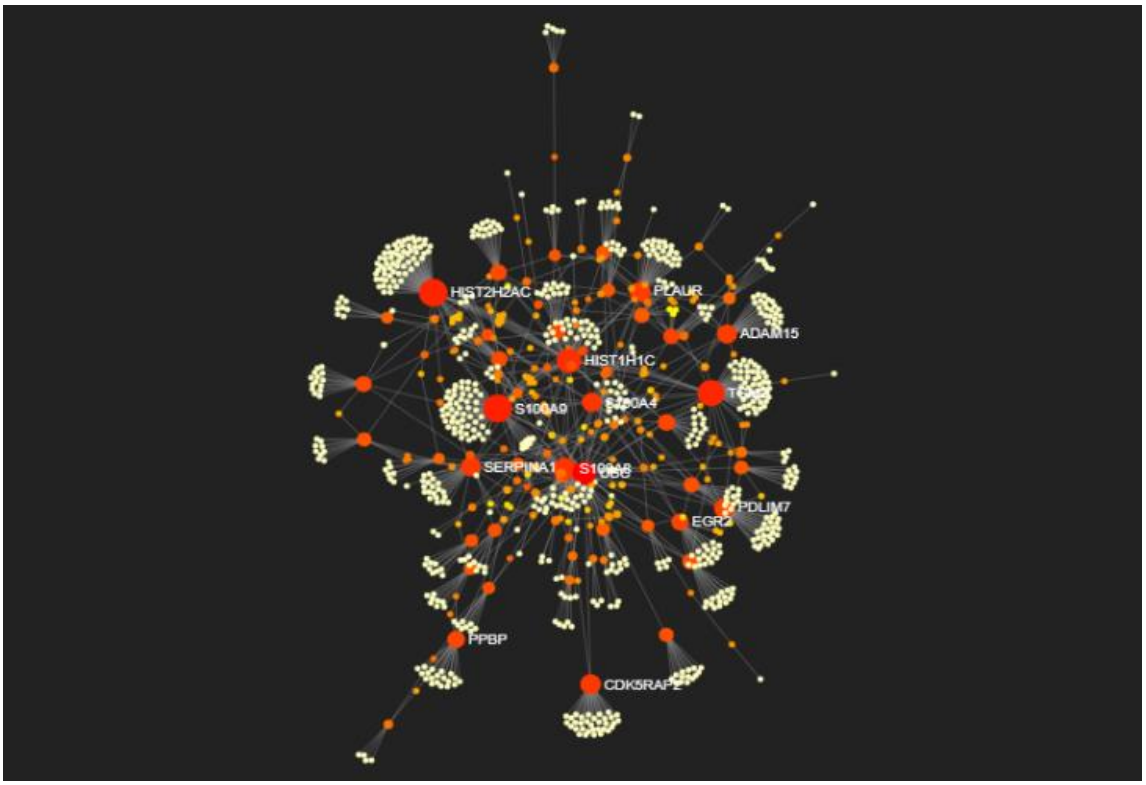

Figure 6. Protein-Protein interaction (PPI) network [Source: https://www.networkanalyst.ca/NetworkAnalyst/Secure/vis/NetworkView.xhtml].

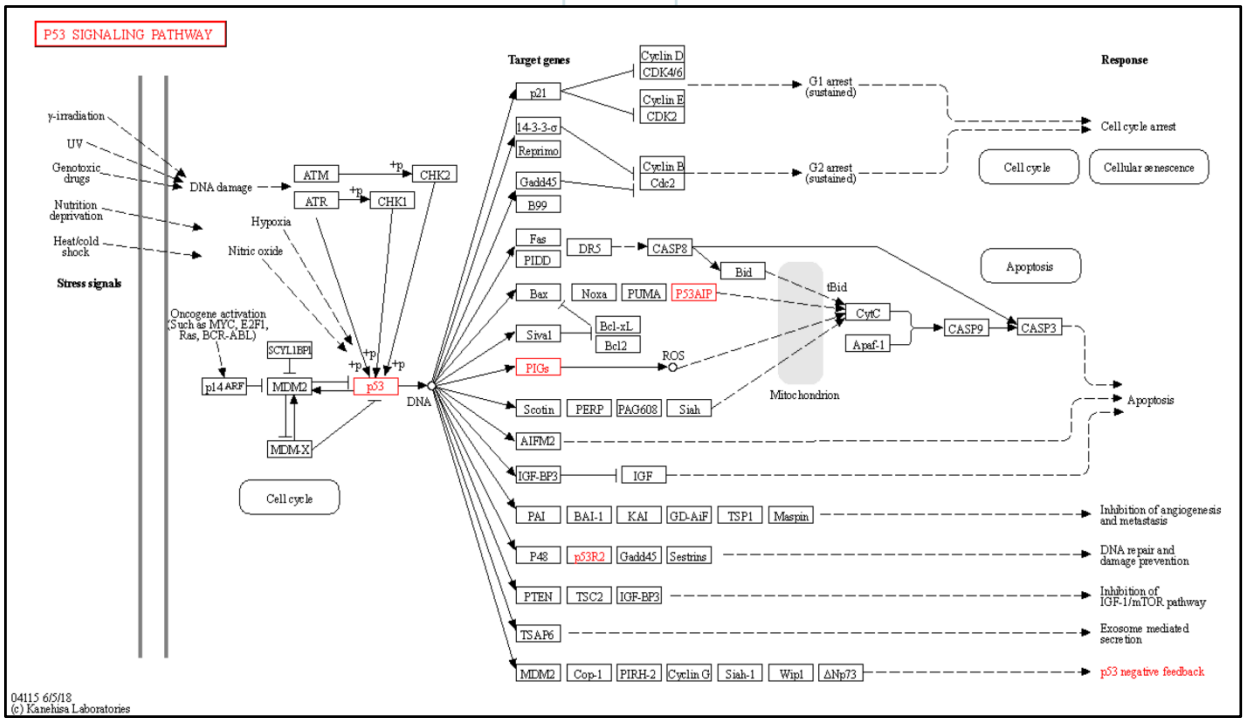

Figure7. KEGG pathway of P53 signaling [Source: https://www.kegg.jp/kegg-bin/highlight_pathway?scale=1.0&map=map04115&keyword=P53].

Along with network analysis tool there are tools such as KEGG and reactome where one can find out the information about each gene or protein. KEGG pathway gives not only the idea about the pathway but also the information about the associated phenotype.

Further databases such as DisGeNet, HPRD, HPA, miRNet provide the information about gene-disease association, protein function and post translational modification and miRNA network respectively.

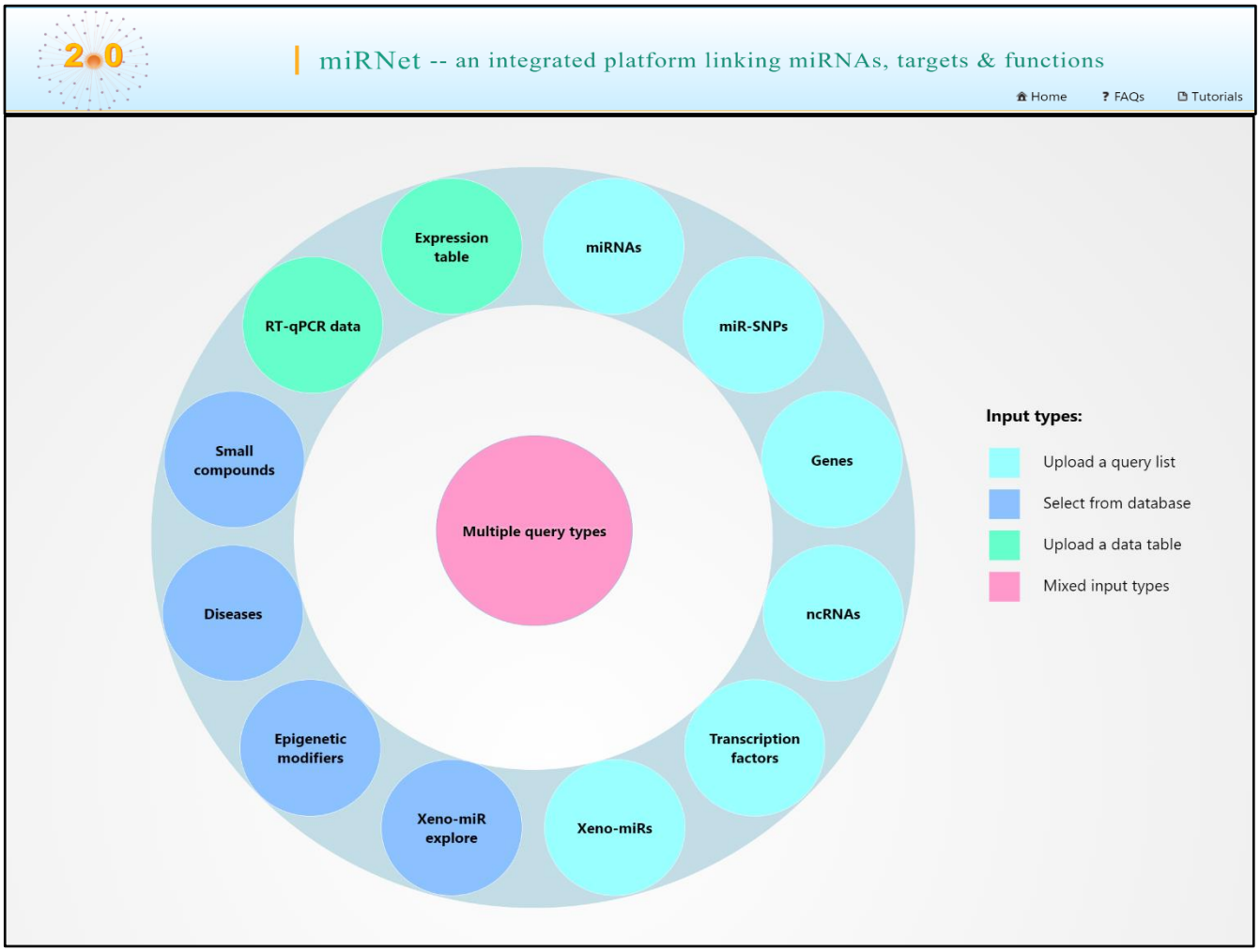

Figure 8. miRNet database where information such as disease associated with miRNA and epigenetic modifiers can be explored [Source: https://www.mirnet.ca/].

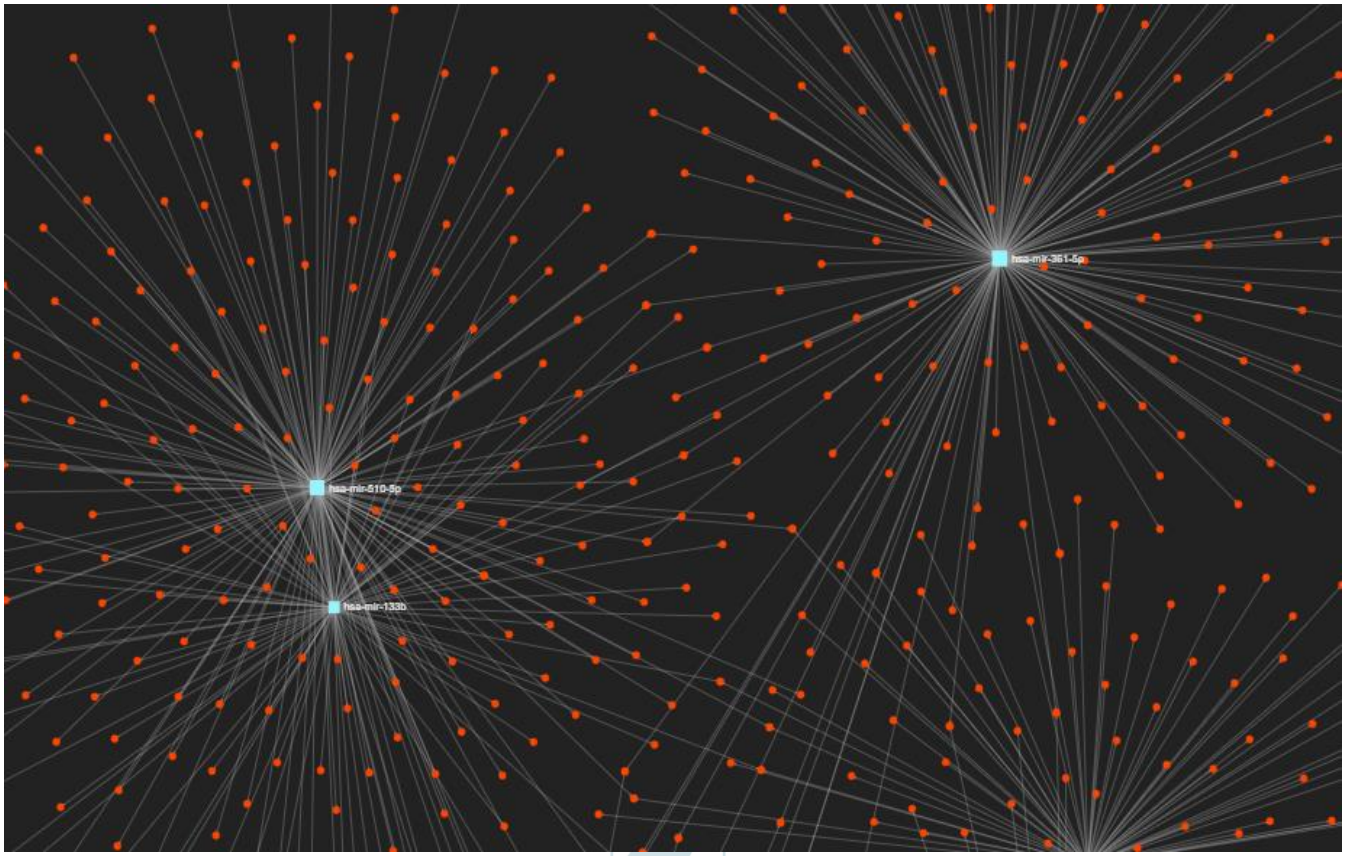

Figure 9. miRNet network of deregulated miRNA [Source: https://www.mirnet.ca/].

Figure 10. Human Protein Reference Database (HPRD) [Source: https://hprd.org/]. In depth information about protein can be exploredby using this database.

Future perspective

Despite of development and advancements in these databases we cannot deny the fact that these are computational tools and some of the result output is prediction based. There is the need for continuous improvement in biological databases where more and more experimental data can be added for increasing accuracy of analysis. With the help of systems biology one can hope to have better understanding of high-throughput data. Whether the information is updated or not in a database is another problem. Therefore, we need to look into these aspects more and more. Further, there is gap between people who generate data and people who curate it. Another big problem which researchers do not understand properly is concept of fold change. Whether the fold change value is coming from pooled sample or from individual sample can make huge difference in final conclusion. Therefore, in future integration and cross-talk between different bioinformatics tool is required.

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Reproductive Biology

By Shripriya Singh, PhD

Contributing Editor

Oral metabolic supplementation can reverse the reproductive clock in mice: A potential remedy to restore reproductive aging?

Introduction

It is popularly said that 'change is the only constant' and rightly so because it is the only means of survival and progress for any living species on earth. Homo sapiens are significantly unique because despite being a single species they have evolved into an indefinite number of communities, tribes, societies and nations with even more diverse religious, social and cultural practices. Depending upon the different beliefs and social mindset several topics were kept hush-hush and were seldom discussed publicly. One such crucial topic was that of the reproductive health in women, which is pivotal for the existence and survival of the entire human race but was still rarely spoken about or discussed. However, we are gradually heading towards an era of societal and mental evolution where orthodox mindsets are being challenged and age old norms are being broken to bring about healthy reforms in the society. Science has played the most important role in this because it is universal, univocal and unbiased. Women reproductive health has not only become a crucial health debate but also the focus of extensive scientific and medical research.

Changing lifestyles, stressful careers, struggle for financial stability and personal choices are often responsible for couples delaying child birth. It is perfectly acceptable for couples to choose the right stage for starting a family but the fact that several factors can negatively impact fertility is causing alarm. Diet, unhealthy lifestyles, smoking, alcohol addictions, hormonal changes, genetic and medical conditions are the important factors that can affect fertility in women. Further age is one of the most crucial factors that can negatively impact the fertility in women. The reproductive system ages faster than one realizes. A woman is born with her entire supply of eggs but as aging progresses the eggs become less viable and their quality deteriorates. There are several causes for female infertility such as ovulation problems, polycystic ovary syndrome, endometriosis, problems with the fallopian

tubes, problems with the uterus etc. Aging is a natural gradual process but its effects start manifesting earlier under conditions of stress, unhealthy diets, hormonal irregularities and disturbed lifestyles. The reproductive potential of a woman begins to decline after the age of 35-40 years and gradually diminishes up till menopause. Other difficulties for the older woman include increased risk of miscarriage and genetic abnormalities in the unborn baby.

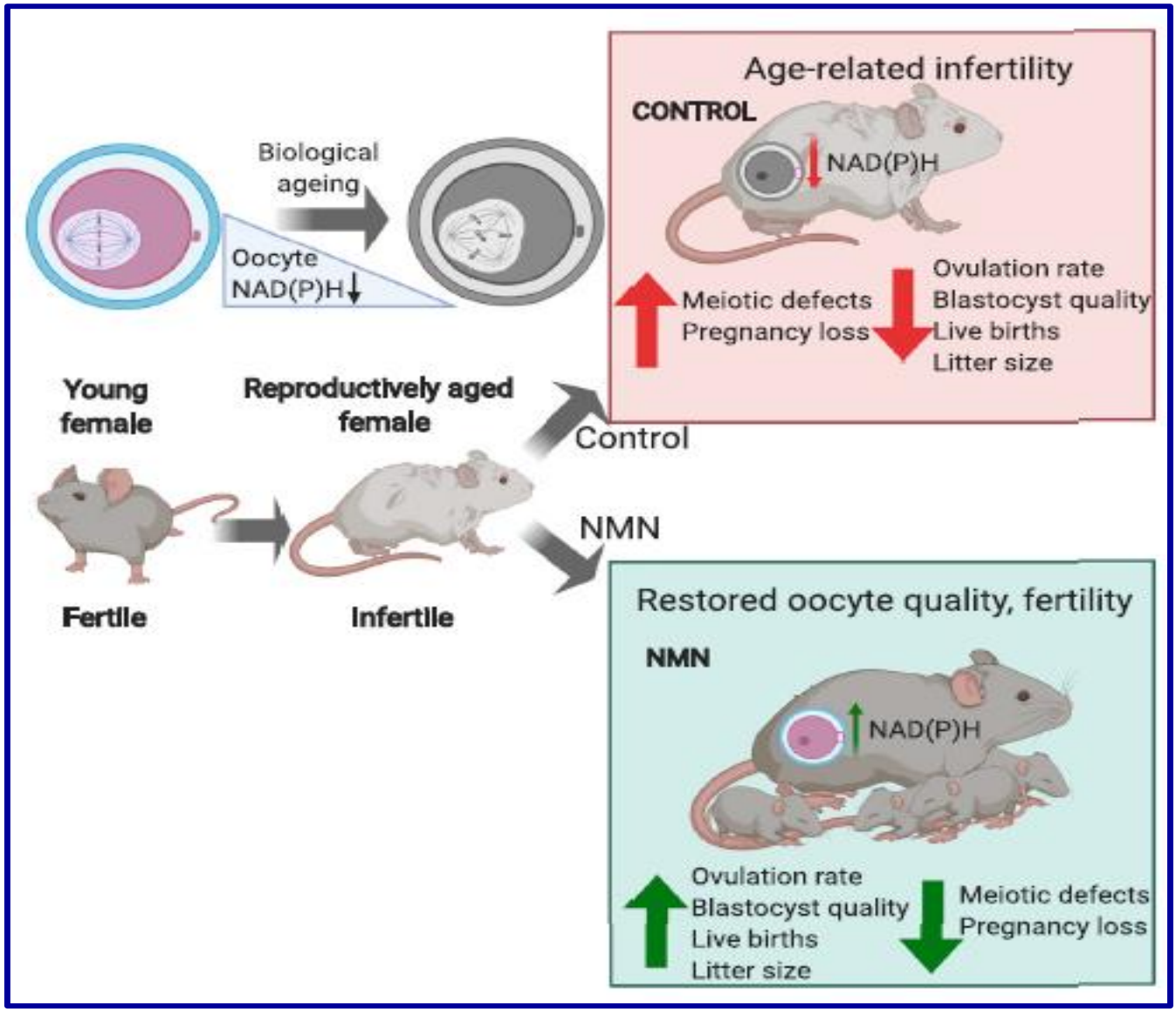

Figure1: Graphical summary of the study published. The oral administration of NMN restores reproductive aging in oocytes. Source: Cell Reports 2020 (1).

In our previous article "Have humans finally discovered path to reverse aging: Synergistic interaction between cellular pathways is the new hope in combating old age" we had highlighted the significance of aging and elaborated the research attempts made to

unravel the molecular cascades. In the current article we shall cite and discuss the latest research endeavor made to improve the fertility rates in female mice and reverse the aging process in the eggs. This study was carried out by a team of researchers at the University of Queensland and the promising results hold relevance for human beings which have made it feature as popular news (1).

The facts and relevance of the study

Reproductive aging in mammalian females is a gradual and an irreversible process which is associated with declining oocyte (egg) quality. The quality and viability of oocytes is the rate-limiting factor to fertility. The oocytes in the ovary of humans are laid down during in utero development, where they form a finite pool that does not undergo self-renewal. Therefore, oocytes are highly prone to age-related dysfunction. The molecular basis for the age related decline in oocyte quality is attributed to increased reactive oxygen species (ROS), reduced mitochondrial bioenergetics, genome instability and disturbances during meiotic chromosome segregation(2, 3). The molecular basis of this age related chromosome mis-segregation in oocytes is still unknown and thus this area remains beyond therapeutic correction. The metabolite nicotinamide adenine dinucleotide (NAD+/NADH) is a prominent enzyme substrate and redox cofactor that is necessary for energy metabolism, epigenetic homeostasis and DNA repair. As age progresses levels of this essential cofactor decline in somatic tissues and reversing this decline through treatment with metabolic precursors for NAD+ has gained interest as a treatment for improving health in aged individuals. The current study demonstrates that deteriorating oocyte quality is also associated with declining levels of nicotinamide adenine dinucleotide (NAD+) (4). Figure 1 summarizes the crux of the study via a graphical abstract.

Mouse was chosen as the experimental model organism because it shows similar reproductive aging that occurs in humans. In mice the fertility starts to decline around 8 months of age due to oocyte defects. Hyperspectral microscopy imaging techniques that exploit the autofluorescence of NADH and NADPH were used to detect and measure the levels of the metabolic cofactor. Twelve-month-old females were treated with NMN in drinking water (2 g/L) for 4 weeks, following which mature metaphase-II (MII) oocytes were recovered and subjected to multispectral microscopy imaging of autofluorescence to determine the relative abundances of native fluorophores. It was clearly observed that NAD(P)H levels declined in oocytes from aged animals as compared to young (4- to 5-week-old) animals. Further NMN treatment increased and replenished NAD(P)H levels in oocytes from aged animals (1) . The experiments were extended to ovarian tissues to determine whether this trend occurred across the entire ovary or was it confined to oocytes alone. Mass spectrometry results showed that there was no decline in whole ovary NAD(H) levels with age, further confirming that it the oocytes which are specially subject to an age-related decline in NAD+. Experiments further confirmed that the quality of oocytes could be improved by

treatment with the NAD+ metabolic precursor nicotinamide mononucleotide (NMN). Mice were fed with low doses of NMN in their drinking water over four weeks which rejuvenated the oocyte quality in aged animals, leading to restoration in fertility and significant increase in live births during a breeding trial. This effect was recapitulated by transgenic over expression of the NAD+-dependent deacylase SIRT2, though deletion of this enzyme does not impair oocyte quality (1). NMN supplementation can reverse the adverse effect of maternal age on developing embryos representing an opportunity to rescue female reproductive function in mammals. Figure 2 shows the imaging data depicting the NMN content in oocytes of mice.

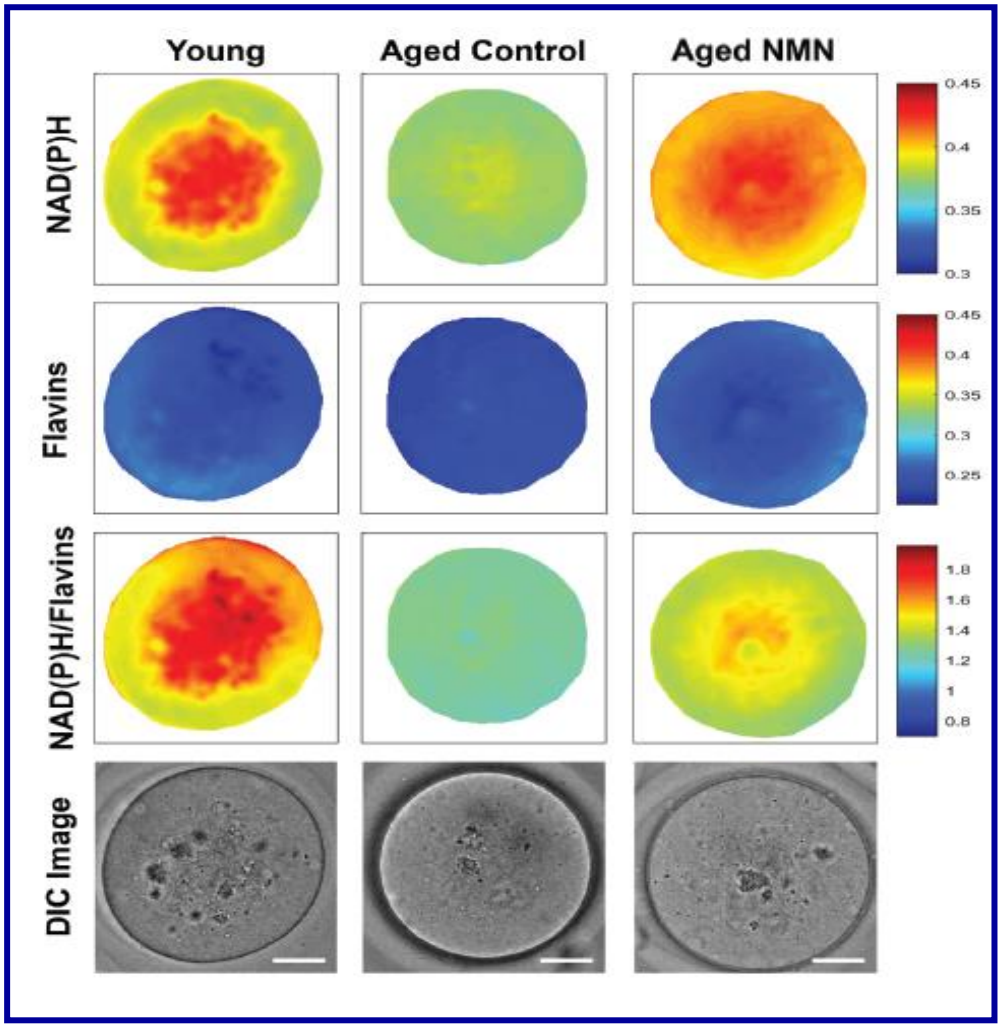

Figure 2: Multispectral imaging data showing NAD(P)H content in oocytes from young (4- to 5-week-old) and aged (12-month-old) mice treated with low doses of NMN in drinking water for 4 weeks. Source: Cell Reports 2020 (1).

Current and future prospects

Increasing maternal age is the biggest barrier in family planning and reproductive aging in

oocytes is a major hurdle to pregnancy for older women. Despite the enormous demand, there are yet no clinically viable strategies to either preserve or rejuvenate oocyte quality during aging. The oocyte quality is defined by the capacity of the oocyte to support meiotic maturation, fertilization, and subsequent embryonic development. Poor egg quality has become the single biggest challenge facing human fertility in developed countries. As more women are embarking on pregnancy later in life there is an increasing demand for assisted reproduction technologies (ARTs) such as in vitro fertilization (IVF), which is invasive, is expensive, carries health risks and has a limited success rate. Further IVF cannot improve egg quality, so the only alternative for older women at present is to use eggs donated by younger women. The results of the present study report a novel non-invasive treatment using oral administration of NAD-boosting agents which could maintain or restore the quality and number of eggs and restore reproductive function. This novel approach would be far less invasive than IVF and alleviate the biggest barrier to pregnancy for older women. The study shows promising potential and its credibility shall be further validated once the clinical trial testing is accomplished in humans. Till then we believe and hope that 'age will continue to remain just a number' and in times to come women will have better reproductive health and the right to choose motherhood even later in life. On that note we sign off by saying that maternal age and reproductive health in women are no longer hushed topics instead proving to become focal points of biological research, open discussions and medical interventions.

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Biotechnology Advances around the World

Editor's Picks

Every issue of Biotechnology Kiosk presents select latest research news picked by the editors-in-chief on significant research breakthroughs in different areas of biotechnology around the world. The aim is to promote further R&D in all of these cutting edge areas of biotechnology. The editors have compiled and included the following innovations and breakthroughs to highlight the recent biotechnology advances.

Dr. Megha Agrawal
Co Editor-in-Chief

Dr. Shyamasri Biswas
Co Editor-in-Chief

Comprehensive mapping of thymic cells across human life could lead to new immune therapies

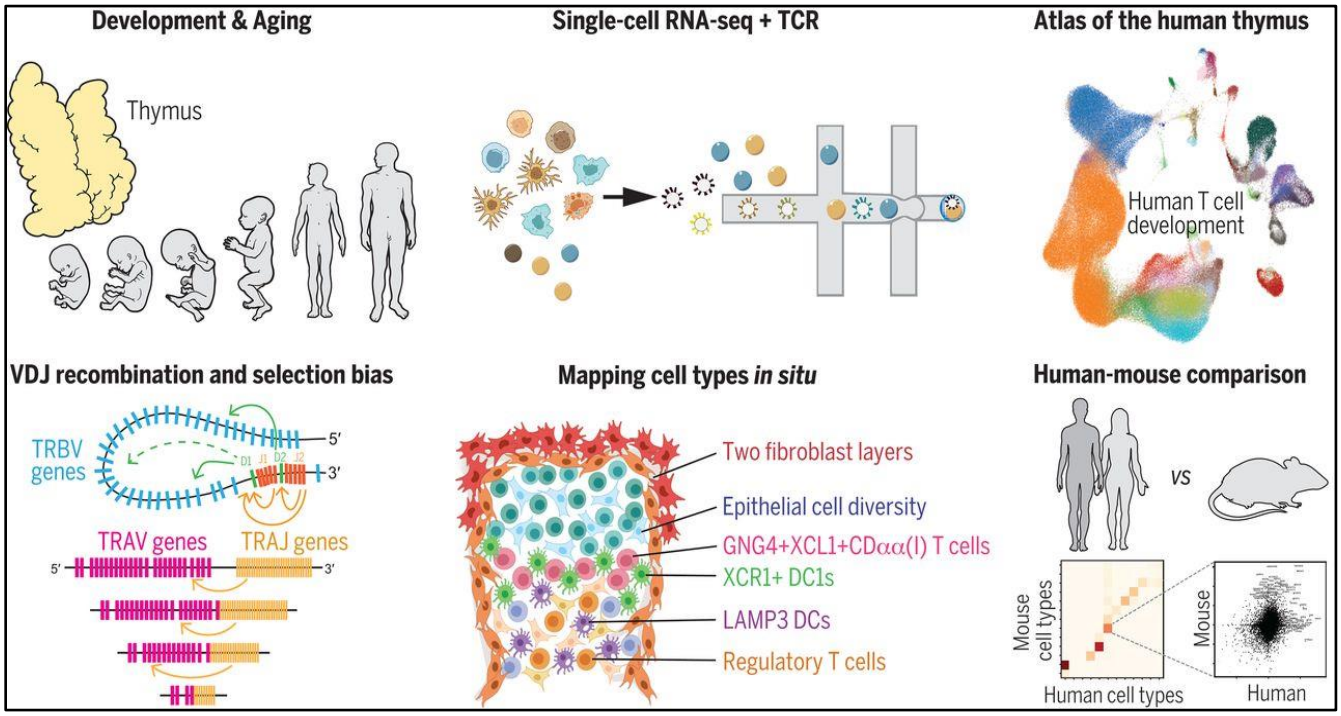
The thymus is a very important organ that is located in the chest. This organ is the largest and known to be most active in childhood that shrinks after attaining puberty. It is believed that a better understanding of the development of thymus and its subsequent withering could help shed light on aging process and how immune system changes through life. The significance of thymus organ or gland lies in the fact that it produces T cells and helps develop T cell receptor repertoire formation. T cells are immune cells and the functions of these cells include producing key white blood cells that fight infection and disease that build the adaptive immunity in human body. T cells can mature further by leaving the thymus and entering the blood stream and other parts of the body. Besides seeking out and destroying invading bacteria and viruses, T cells can also recognize cancer cells and destroy them.

Previous studies have revealed that disorders in thymus development can cause generation of defective T cells that results in severe immune deficiencies making people susceptible to infections. The regulation of T cells can also be affected by disordered thymus development, which results in autoimmune diseases such as Type 1 diabetes.

Mature T cells have been well studied. However, the missing link so far is the lack of study on the development of the human thymus and T cells within thymus. It is believed that human immunity cannot be understood fully without developing a detailed atlas of the human thymus across human life.

In a latest study of major significance, researchers performed for the first time, single-cell RNA sequencing on more than 250,000 cells and studied the changes that occur in the thymus over the standard course of a human life. Researchers from the U.K. and Belgium published their research in Science (A cell atlas of human thymic development defines T cell repertoire formation, Science, 2020; 367 (6480): eaay3224 DOI: 10.1126/science.aay3224), and reported about mapped thymus tissue through the human lifespan to understand how it develops and makes vital immune cells called T cells. The researchers envisioned that the atlas could help understand diseases that primarily affect T cell development such as severe combined immunodeficiency (SCID).

The Figure below illustrates the construction pathways of the human thymus atlas that has revealed new cell types and identified signals that tell immature immune cells how to develop into T cells. Researchers employed single cell technology for isolating and analyzing more than 250,000 individual cells from the developing thymus including child and adult thymus tissue. They looked at which genes were active in each individual cell to identify the cells, discovering new cell types, and used those genes as tags to map each cell to its exact location in the thymus.



Illustrations show the pathways for construction of human thymas cell atlas [Source: Jong-Eun Park et. al., Science (2020].

This map of the thymus producing human Cell Atlas can potentially chart every cell type in the human body. It can be employed to learn developmental pathways along with the age-associated decline of the immune system with potential applications in cellular engineering for creating an artificial thymus for regenerative medicine. Further, with this thymus cell atlas, researchers showed the possibility of using cellular signals of the developing thymus to reveal which genes can be switched on to convert early immune precursor cells into specific T cells.

This comprehensive study on mapping the origins of immune system could open the doors for new cancer immunotherapies for treating cancer and autoimmune diseases. Researchers envision that such cell atlas of human thymus could pave the way for breakthroughs in engineering improved therapeutic T cells. Therapeutic T cells are currently considered clinically important to treat B-cell lymphoma and leukaemia cancers. However, creating the right subtype of T cells is a major challenge in these T-cells based treatments. It is believed that this breakthrough study on producing human cell atlas could eventually lead to an artificial thymus and the ability to engineer improved therapeutic T cells.

Stem Cells & Bone Regeneration

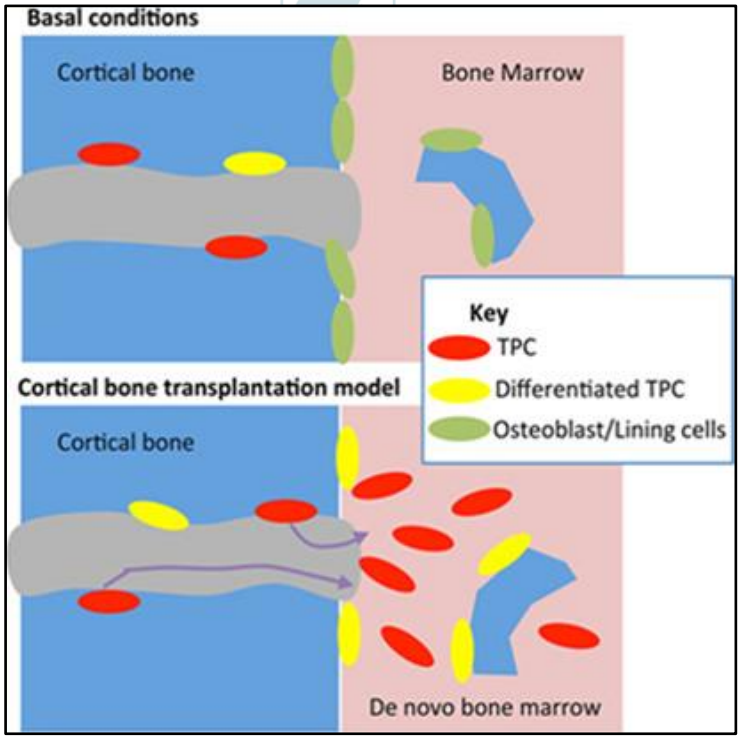
Population of new stem cells have the ability to generate bones

It has long been thought that stem cells for bone reside within bone marrow and the outer surface of bone. These cells are

believed to serve as reserve cells that are constantly used to generate new bone or even to repair damaged bone. Previous studies indicated the existence of a network of vascular channels that helped distribute blood cells out of the bone marrow. However, researchers did not prove the existence of cells within these channels that can form new bones.

A recent discovery made by a team of researchers in the United States and New Zealand that was published in the journal Stem Cells (Perivascular osteoprogenitors are associated with transcortical channels of long bones. STEM CELLS, 2020; DOI: 10.1002/stem.3159) demonstrated a population of stem cells with the ability to generate new bone. In this research of major significance, they revealed a new population of perivascular cells residing along the vascular channels that stretch across the bone and connect the inner and outer parts of the bone.

This study reported for the first time the existence of these progenitor cells within cortical bone that can generate new bone-forming cells known as osteoblasts (see the Figure below). This is quite novel because it can help remodel a bone. Researchers studied bone remodeling and regeneration and showed the dependence on resident stem/progenitor cells with the capability to replenish mature osteoblasts and repair the skeleton. They observed the stem cells within an ex-vivo bone transplantation model. These cells were then found to migrate out of the transplant, and subsequently began to reconstruct the bone marrow cavity that formed new bone.



Channels of long bones using transcortical perivascular cells (TPCs) [Source: Stem Cells (2020)]

This new discovery of population of perivascular cells residing within the bone itself is quite exciting due to the fact that they can generate new bone forming cells. It is believed that these cells can likely regulate bone formation and also participate in bone mass maintenance and repair.

The Figure shown here schematically describes the process of the formation of channels of long bones using transcortical perivascular cells (TPCs). This study suggests the real possibility of an osteoprogenitor specific to the cortical bone serving as a source of osteoblasts for intracortical remodeling.

Plant Biotechnology

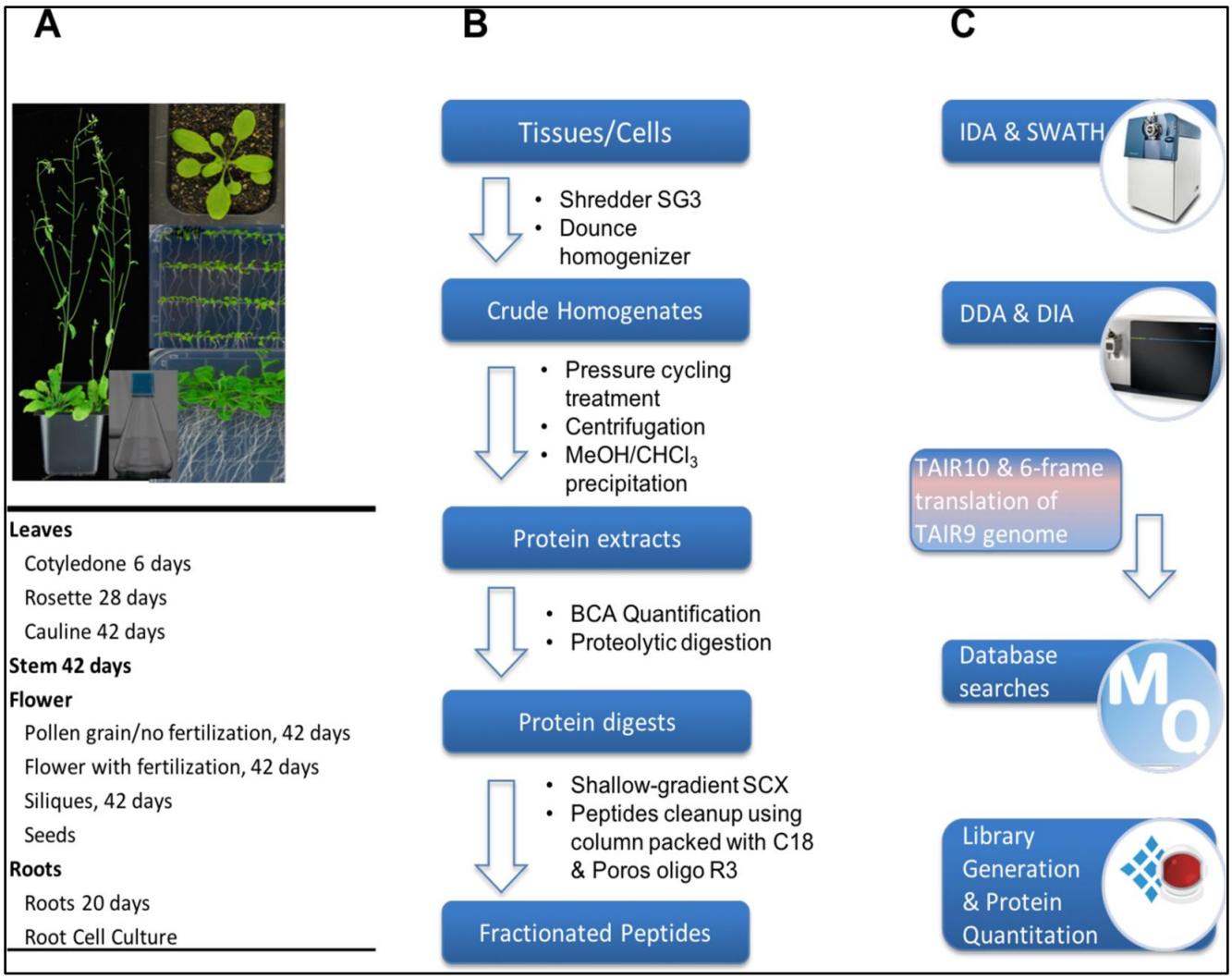
Comprehensive molecular map of the proteome of plant

Plants are essential for life on earth by providing food for almost all organisms and oxygen for breathing. Plant's functions are also to regulate the climate of the planet. To understand how plant creates tissues as diverse as a leaf that converts light into chemical energy and also how they produce oxygen, or a root that absorbs nutrients from the soil plants, we need to understand the protein pattern of the cells of the respective tissue. Proteins are known to play a critical in controlling all aspects of life including plants. They are considered molecular players in every cell. They are biocatalysts and transmit signals inside and between cells and form the structure of a cell.

Studying the protein pattern requires which proteins are present in a tissue, as well as in what quantities. This can be understood from an example of proteins of the photosynthesis machinery that are found primarily in leaves. They are also found in seeds. But quantitatively, they are found at a thousand times lower levels in seeds.

For the first time, a team of scientists in Germany has mapped around 18,000 of all the proteins found in the model plant Arabidopsis thaliana. Their research was recently published in Nature (Mass-spectrometry-based draft of the Arabidopsis proteome, Nature, 2020; DOI: 10.1038/s41586-020-2094-2). They comprehensively mapped the proteome, including proteins from the tissues of the model plant Arabidopsis, which allowed new insights into the complex biology of plants (see the Figure). This study presents insights from basic research on Arabidopsis that can often be transferred to crop plants, which makes makes Arabidopsis interesting for plant breeding research.

Researchers used a method called liquid chromatography-tandem mass spectrometry, to generate most of the data. This enabled the analysis of thousands of proteins in parallel in one experiment and bioinformatics methods helped analyze the huge amounts of data simultaneously. They summarized all results in a virtual atlas that addressed important questions such as how many of the approximately 27,000 genes exist in the plant as proteins (> 18,000) and also the location within the organism and in what approximate quantities they occur. These results form the basis for future analysis of crop plants.



The process of comprehensive molecular mapping of model plant Arabidopsis

Compiled and Edited by

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Biotech and Pharma Industry Roundup

Regeneron to begin COVID-19 clinical trials for newly identified antibodies

Drug and pharmaceutical companies continue to advance their forces against COVID-19 to combat the illness caused by the novel coronavirus. The New York-based Regeneron recently announced that its antibody program aimed at the virus is poised to enter the clinic by early summer. The company has identified hundreds of virus-neutralizing antibodies and intends to begin large-scale manufacturing by mid-April. Regeneron plans to administer the antibody cocktail therapy in the form of prophylaxis before exposure to the SARS-CoV-2 virus or as a treatment for those already infected [Source: https://www.biospace.com/].

mRNA Vaccine for COVID-19 to be developed by Pfizer and BioNTech

Germany's BioNTech announced a plan to develop an mRNA vaccine candidate in China for the prevention of COVID-19 with Fosun Pharma. The company also entered into an agreement with Pfizer to develop a vaccine for other areas in the world. Under the agreement, Pfizer and BioNTech SE will be engaged in the co-development and distribution of a potential mRNA-based coronavirus vaccine. This agreement allows the two companies to focus on BioNTech's BNT162, an mRNA-based vaccine candidate. The vaccine candidate, currently in the preclinical stage, comes from BioNTech's proprietary mRNA platforms for infectious diseases. BNT162 is expected to enter the clinic in April. It is considered the first product candidate from Project Lightspeed, the company's accelerated development program for COVID-19 [Source: https://www.biospace.com/].

Meningococcal Vaccines expected to touch US$ 9 billion by 2026

Bacteria Neisseria meningitides causes meningococcal disease. This disease occurs throughout the world especially in the developing world. Typical symptoms include inflammation in the membranes of the brain and spinal cord which is characteristic of meningitis. The meningococcal vaccines market is pretty vast with a lot of potential to make its footprints even larger in the future that is expected to reach $9 billion by 2026. This vaccine exists in the market and doing a great business due to its utility and need for preventing the incidence of this deadly disease [Source: https://www.biospace.com/].

AstraZeneca's checkpoint inhibitor Imfinzi shows promise in lung cancer

The company recently reported that high-level data analysis from its Phase III CASPIAN trial demonstrated that Imfinzi (durvalumab) in combination with standard-of-care chemotherapies suggested a clinically meaningful and sustained overall survival benefit in patients with extensive-stage small cell lung cancer treated in the

first-line setting [Source: https://www.biospace.com/].

The market of dental membrane and bone graft substitutes to reach US$ 1.1 Billion by 2027

Current dental R&D has extensively focused on the developments of quality oral health care including new dental membrane and bone graft substitutes. This is one area in dental research that has seen extensive developments and innovation in recent years. This is primarily due to increasing number of accidents and oral health issues. Initially, the global dental membrane and bone graft substitutes market was valued at ~US$ 620 Million in the year 2018. Due to the vastly improved market scenario, and an ever increasing demand for quality dental care, the dental membrane and bone graft market is projected to steadily increase and expand at a rate from 2019 to 2027 and reach US$1.1 Billion by 2027. The growth of the market can be attributed to rise in the dental disorders, worldwide aging population, growing medical tourism for dental procedures, and increase in the number of dental implant procedures around the world [Source: https://www.biospace.com/].

SMA drug produced by Roche gets a boost with new study data

Roche recently announced that its experimental drug risdiplam helped babies with the severest form of the rare disorder spinal muscular atrophy sit unassisted one year after beginning treatment. This finding should help Roche's case for approval now before the FDA. It is expected that if this drug is finally approved by the FDA, risidiplam will enter a highly competitive field already served by two injectable drugs, Biogen's Spinraza and Novartis' gene therapy Zolgensma [Source: https://www.biopharmadive.com/).

Lilly to start coronavirus program

Eli Lilly announced to join the pharmaceutical industry's push to develop treatments for COVID-19, the illness linked to the new coronavirus in a partnership with the private biotech AbCellera. The two companies will search for immune proteins that can neutralize the virus screen antibodies. These were isolated from one of the first U.S. patients who was infected and later recovered [Source: https://www.biopharmadive.com/)].

Gates, Wellcome donate $125M to speed coronavirus treatments

Three philanthropic foundations The Bill and Melinda Gates Foundation, Wellcome and the Mastercard Impact announced that they would spend up to $125 million to help advance treatments for the new coronavirus in a collaborative effort that would engage regulators, non-governmental organizations and the biopharma industry. The joint effort known as the COVID-19 Therapeutics Accelerator is designed to encourage research and development of existing drugs that can be repurposed for the coronavirus as well as new agents not yet authorized for use in humans [Source: https://www.biopharmadive.com/)].

Investors pouring cash to counter cancer genes

It is known for years that a family of genes that, when mutated, turn regular cells cancerous. There has been recent progress in developing drugs that can effectively silence those genes. This has fueled investor optimism in companies going after these genes that are known as RAS. The most recent example of investments comes from a California based biotech company named Revolution Medicines that has raised $238 million through an initial public offering. RAS genes are known to make special proteins that regulate how a cell grows, specializes and divides. One of these proteins, K-ras, has drawn a particularly large amount of attention for R&D. and Amgen, the country's largest biotech has released data showing an experimental drug appeared to stabilize disease in a small group of lung cancer patients with KRAS gene mutations. This has further fueled extensive R&D in this area [Source: https://www.biopharmadive.com/)].

Roche presents biomarker for checkpoint block blockade

Researchers at Swiss pharma giant Roche AG have demonstrated a subpopulation of dendritic cells that determines how cancer patients respond to immune checkpoint inhibitors [Source: https://european-biotechnology.com/].

French company Stilla raises €20m in series B financing round

French biotech company Stilla Technologies SA raised €20million in a Series B funding to boost digital PCR-based genetic testing [Source: https://european-biotechnology.com/].

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