

September 2021

PHARMA CHRONICLE

8th Edition



TABLE OF CONTENTS

ARTICLES

DMT: Getting High on Your Own Supply 4

Structural Comparison of SARS-CoV and SARS-CoV-2: Effects on Clinical Relevance 7

"Smart" Insulin - The Future of Pharmacotherapies for Diabetics 10

THC's Analgesic Effect Through Altering Neuronal Pain-Processing Pathways 13

A New Treatment for Tumor Ablation and Drug Delivery 16

RAS Modulators and the Future of PTSD Pharmacotherapy 19

Psychedelic Microdosing: Public Health Concern or Novel Therapeutic? 23

Plant-Based vs. Animal-Based Vaccines 27

The Consequential Diseases and Physiological Effects Associated with Vaping 30

Financial Incentives in Clinical Trials Come at a Price 34

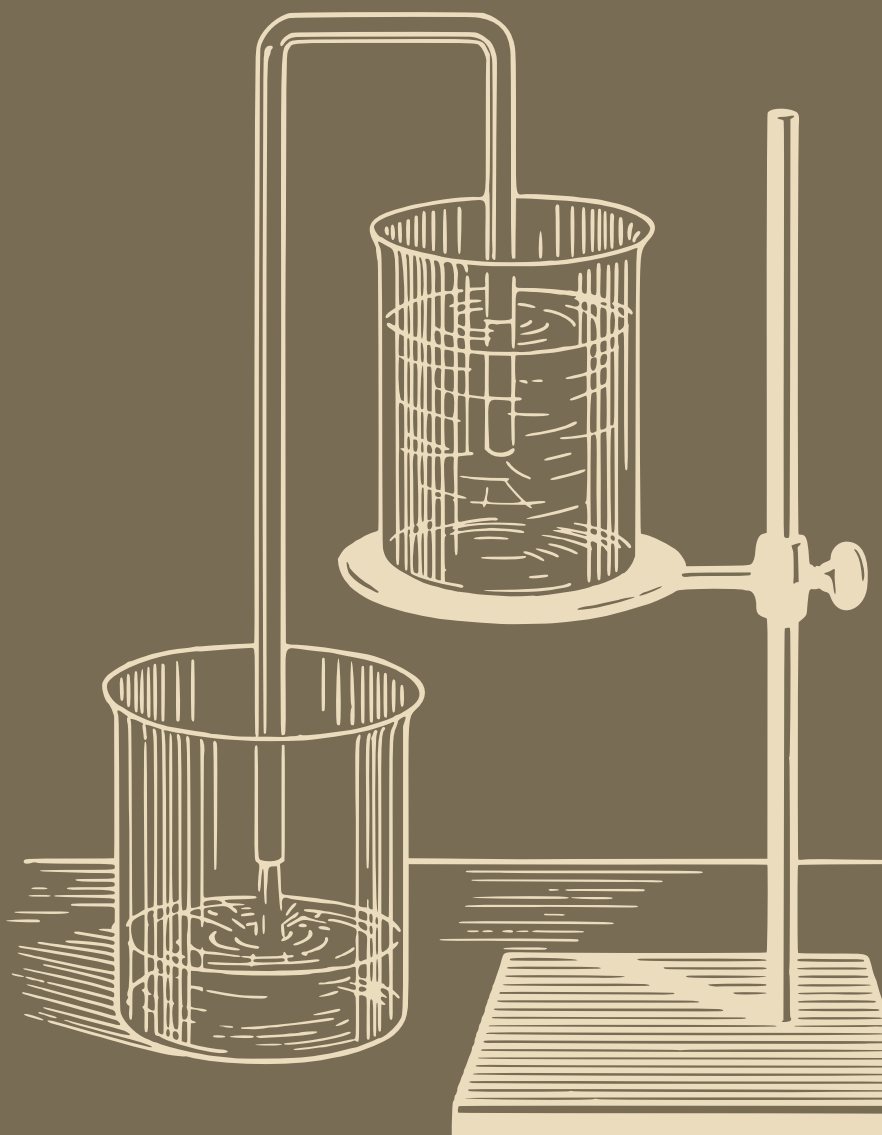
May Light Shed on Traditional Medicines 37

Novel Therapies for Acute Myeloid Leukemia 42

OTHER

Meet the Team 3

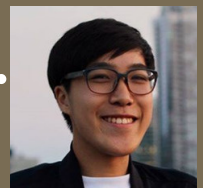
University of Toronto Undergraduate Computational Chemistry 45





MEET THE *Team*

- **Editor In Chief** Emily Mathers
- **Lead Writer** Emily Saso
- **Director Internal** Dinie Syahmi
- **Director External** Brian Chan



• **Lead Editor** Simon Qu



• **Lead Editor** Michelle Wang



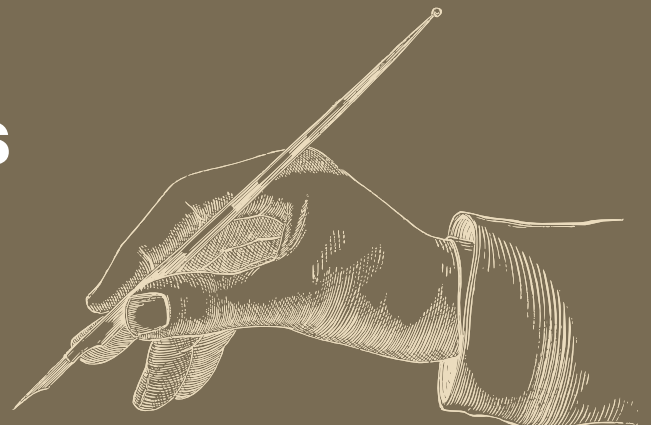
• **Senior Advisor** Daniel Li



• **Design** Prima Zhao

Writers

Gilad Yohananov / Ming Heng Wang /
Payton Midgley / Jia Yin Lu / Daphne Lin /
Karen Kuang / Rachel Kuah / Eric Kim /
Manu Kattimani / Carmen Chan / Joan
Zheng / Danlin Zeng



DMT: Getting High on Your Own Supply

By: Gilad Yohanonov

Intro: Hallucinations

A “hallucination” is a perception of something that is not literally real[1]. One experiences hallucinations when they perceive something in the absence of the supposed stimulus that induces such perception, which means that the hallucinatory experience is psychological[1]. Due to its psychological nature, only the one hallucinating can attempt to make sense of the hallucinatory experience[1]. Not only does this make studying hallucinations difficult, but conclusions based on the hallucination are solely based on personal experiences[1].

Hallucinations may occur in any perception of one's 5 senses; for instance, they are classified as visual, auditory, etc.[1]. Various stimuli can induce one or more sensory hallucinations which differ in intensity respective to the exposure to the stimulus, both in amount and duration[1]. Such stimuli include disease, mental health, lack of sleep, and medications[1]. Common medications that cause hallucinations include olanzapine, quetiapine, and haloperidol; nevertheless, these are also used to treat hallucinations[1]. What about a drug that is intended to cause hallucinations, and not just any hallucinations - but an intensive perception of dimension transitions (psychosis)?

What is DMT?

N,N- dimethyltryptamine, also simply known as DMT, is a hallucinogenic drug that elicits a vivid and powerful euphoria to consumers. DMT is a Schedule 1 controlled substance in the United States, making it a highly illegal narcotic, because of its high potential for abuse and lack of research claiming its safety and medical use[2]. Nevertheless, due to its relatively short duration of action and intense hallucinatory effects, DMT is a highly popular drug that is commonly consumed by spiritual activists[2]. DMT is most commonly inhaled or injected, which elicits a euphoric duration of action of 15 minutes[2].

The half-life of DMT is 12-19 minutes, with an apparent volume of distribution (Vd) of 36-55 L/kg; the Vd is high because the chemical structure of DMT is small and highly lipophilic[3]. Please note that further pharmacokinetic parameters will not be discussed in this article.

Users that consume this drug mention that the effects of DMT are the most intense and purest form of peace and happiness[4]. A ‘trip’ on DMT is unexplainable as users state they “break out of a simulation” and “meet aliens” in a superficial non-existent but existent fabric of space-time[4]. The user literally feels as if they are blasted off into a hyperspace that is filled with spirals and unimaginable geometric patterns that seem *familiar*[4]. Users state that due to this familiarity factor among intense hallucinations and mystic references, they feel as if this DMT hyperspace is the true and fundamental spiritual realm of life[4].

Biosynthesis of DMT

DMT is commonly extracted from *Psychotria Viridis* and *Banisteriopsis caapi*; however, as the article title suggests, DMT is endogenously found in mammalian brains[4]. The biosynthesis of DMT depends on two enzymes: aromatic-L-amino acid decarboxylase (AADC)

and indolethylamine-N-Methyltransferase (INMT)[5]. It is theorized that the dietary amino acid tryptophan is consumed and gets converted to tryptamine by AADC[5]. The product, tryptamine, then undergoes N,N-dimethylation which is facilitated by INMT to form DMT[5]. This is significant as a recent study[5] showed that the INMT & AADC are co-expressed in the cerebral cortex of a mammalian brain (rat). Specifically, Dean et al.[5] found a dominant overlapped expression of both enzymes in the mammalian brain compared to a non-overlapping expression of both enzymes in the rat peripheral organs such as the kidney, heart, and lung.

The same research group[5] observed a concentration of 1.02 nM of DMT in the mammalian brain (rat), which has a slightly smaller concentration of serotonin concentrated at 2nM. This shows that endogenous DMT synthesis solely occurs in the mammalian neocortex[5].

Moreover, Dean et al.[5] induced a cardiac arrest in rats in an attempt to mimic a near-death-experience. It was observed that baseline endogenous DMT in rats increased in response to a cardiac arrest, making it plausible that the brain itself synthesizes DMT in response to death[5]. It has been noted that the concentrations of DMT in the brain that were synthesized during cardiac arrest may not necessarily elicit the same effects as users feel[5]; however, a few questions arise. Does one really get high on their supply? Does one's brain respond to death by increasing his/her biosynthesis of DMT? Also, due to its out-of-body spiritual experiences, is DMT the link between the physical world & the spiritual world?

Past & Present Roles in Society (Historical perspective & Current role in society)

DMT has been utilized for many years, primarily due to its spiritual awakening characteristics[4]. Ayahuasca is a brew made by *Psychotria viridis* leaves and stalks of the *Banisteriopsis caapi* vine that was and still is utilized by ancient Amazonian tribes[6]. Needless to say, Ayahuasca is concentrated with DMT. Contrary to the "casual" 15-minute trip users usually feel from inhaling/injecting DMT, Ayahuasca is an orally ingested solution that takes 20-30 minutes to feel the effects which lasts 3-5 hours[6]. Prior to the ceremony, participants are recommended to undergo a dopamine cleanse, which sensitizes the brain to decrease its threshold for dopamine release[4,6]. This is thought of as a cleanse to the user and prepare them for the trip of their life. Traditionally, an Ayahuasca ceremony occurs at night and is monitored by an experienced healer, termed Shaman[6]. The Shaman leads the ceremony and splits up the ayahuasca to participants[6]. These ceremonies still take place and are in fact ever more popular now since DMT's usage has increased in popularity[6].

The number of people that use DMT is increasing, and it is mostly due to its supposed beneficial spiritual insight & cleanse. In fact, according to a study done by the American Journal of Addiction[7], the percentage of the population using DMT comparing 2008 to 2014 has increased by 0.5%. In practical terms, 0.5% of the population is a 40 million user increase. Perhaps the technological advancement and increased social media further exposes the population to DMT. In all actuality, are you, yourself, not tempted to witness what users describe as communicating with God in another dimension that is concentrated with intense and unimaginable geometric patterns? Taking that into consideration, most undergo the same dilemma with some pursuing the use of DMT.

Future Outlook

All in all, DMT is a chemical that is synthesized in rats[5], among additional potential organisms. This drug was historically and currently used to elicit intense hallucinations that allow the user to interpret a spiritual realm. Therefore, while the trip may hold a sort of significance to users, it is ultimately a hallucination. Because this hallucinatory state of mind is induced by the user, users are recommended to prepare for the trip; for instance, Ayahuasca ceremony participants undergo a dopamine cleanse. Modern psychological discoveries make sense of the fact that the brain is easily "tricked" and attempts to make sense of everything that is sensed. Consequently, a DMT user may not necessarily enter another dimension, but due to DMT's intense hallucination, they try to make sense of it by stating that it must have been a spiritual out of body experience. DMT itself does not have any proven benefit to users, but potentially this spirit drug can induce a belief in the user that there really is another realm filled with life. This could lead to them believing in God and maybe even a religion, which is proven to satisfy and increase one's well-being. Therefore, instead of focusing on the pharmacodynamic benefits of DMT, one could extrapolate the effects of this drug via psychological testing on users prior and post consumption. There is a lot of research needed to be done in order to accurately state DMT's role in society.





References

1. Chaudhury S. Hallucinations: Clinical aspects and management. *Ind Psychiatry J.* 2010;19(1):5-12. doi:10.4103/0972-6748.77625.
2. Timmermann C, Roseman L, Schartner M, et al. Neural correlates of the DMT experience assessed with multivariate EEG. *Sci Rep.* 2019;9(16324):1-13. doi:10.1038/s41598-019-51974-4.
3. Shen HW, Jiang XL, Winter JC, Yu AM. Psychedelic 5-Methoxy-N,N-dimethyltryptamine: Metabolism, Pharmacokinetics, Drug interactions, and Pharmacological Actions. *Curr Drug Metab.* 2010;11(8): 659- 666. doi:https://doi.org/10.2174/138920010794233495
4. Strassman R. DMT: The Spirit Molecule: A doctor's revolutionary research into the biology of near-death and mystical experiences. Park Street Press. 2010.
5. Dean JG. Indolethylamine-N-methyltransferase Polymorphisms: Genetic and Biochemical Approaches for Study of Endogenous N,N-dimethyltryptamine. *Front Neurosci.* 2018;12(232):1-16. doi:10.3389/fnins.2018.00232.
6. Frecska E, Bokor P, Winkelman M. The Therapeutic Potentials of Ayahuasca: Possible Effects against Various Disease of Civilization. *Front Pharmacol.* 2016;7(35):1-17. doi:10.3389/fphar.2016.00035.
7. Palamar JJ, Le A. Trends in DMT and other tryptamine use among young adults in the United States. *Am J Addict.* 2018;27(7):578-585. doi:10.1111/ajad.12803.

Structural Comparison of SARS-CoV and SARS-CoV-2: Effects on Clinical Relevance

By: Ming Heng Wang

Coronavirus 2019, also known as COVID-19 is a disease that emerged in Wuhan, China as of the end of 2019[1]. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a novel betacoronavirus that causes a respiratory disease that we all know as coronavirus disease 19 (COVID-19) and has had an immense impact on public health in the past year. At the end of January 2020, the World Health Organization (WHO) declared this a global health emergency. By the end of December 2020, there had been approximately 77,801,721 confirmed COVID-19 cases with 1,713,109 COVID-19 deaths reported worldwide[2].

Humans are at risk of infection by the virus when they are in close contact with infected people[1]. Research has shown that the pathophysiology of COVID-19 is involved most commonly with the virus entering through respiratory fluid into the respiratory system[1]. This is why many countries set up public health guidelines such as face mask protection in public and social distancing of 2m from others[1]. People who are infected often experience symptoms such as fever, coughing and fatigue. This virus can range from mild to severe[1]. Therefore, it is still important to adhere to public health guidelines that help prevent the spread of this virus[1].

SARS-CoV-2 is in the same family of betacoronavirus as severe acute respiratory syndrome coronavirus (SARS-CoV) and the Middle East respiratory syndrome coronavirus (MERS-CoV) which caused SARS and MERS, respectively[2]. Coronavirus is a broad term that encompasses a large group of positive-strand RNA viruses that inflict respiratory, enteric and neurologic dysfunctions in their host. Specifically, they are enveloped viruses, meaning they enter their host by inducing fusion between viral and cellular membranes[3].

SARS-CoV-2 is very similar to SARS-CoV in terms of their virology. In terms of transmission, both viruses possess the spike (S) glycoprotein that mediates membrane fusion with the target cell. The S protein is located on the capsid of the virus and binds to the angiotensin-converting enzyme-2 (ACE2) receptor on the host cell. The viral RNA genome enters the target cell via the mechanism of receptor-mediated endocytosis[4]. In addition, both viruses require S activation by host cell proteases, specifically TMPRSS2[2].

However, the 2 viruses differ in their biological behaviour. SARS-CoV causes a mortality rate of approximately 9.6% whereas SARS-CoV-2 has on average a mortality rate of less than 1%[2]. SARS-CoV could only spread by symptomatic individuals and is less effective at transmitting from human to human[2]. Whereas, SARS-CoV-2 can be spread by individuals who are asymptomatic during the incubation period or those who just don't exhibit any symptoms and is very effective at human-human transmission[2]. SARS-CoV infects the lower respiratory tract, whereas SARS-CoV-2 is present in the upper respiratory tract[2]. Research has been conducted to provide evidence that some of these differences could be due to genomic variation between SARS-CoV-2 and SARS-CoV, specifically differences in the S protein[2]. In this article, the content will be focused on the association between genomic variation and differences in transmissibility between the SARS-CoVs.

Bojkova et al. used differentially conserved amino acid positions (DCPS) to discover genetic differences that could explain the phenotypic differences between the SARS-CoVs 1 and 2[2]. Abundance of DCPs provides strong support for the differences in clinical behaviour of SARS-CoV-2 and SARS-CoV.



The S protein of SARS-CoV and SARS-CoV-2 are 77.36% identical in sequences while the remaining amino acids positions are DCPs (186 residues)[2].

There is an abundance of DCPs present in the surface loops of the S protein that interact with the ACE2 receptor[2]. This creates different conformations between the S proteins of the SARS-CoVs[2]. Consequently, SARS-CoV-2 has greater binding affinity to the ACE2 receptor than SARS-CoV, explaining the better transmission capability of SARS-CoV-2 than SARS-CoV[2].

Structurally speaking, hydrogen bonding is exhibited between Y41, H34, Y83 and K353 of the ACE2 receptor and T500, Y453, N487 and G502 on the S protein of SARS-CoV-2[5]. Hydrophobic interactions are also found between K31 and K353 of ACE2 and Y489 and Y505 (residues on the RBD) of the S protein[5]. These non-covalent interactions regulate and provide specificity for S protein binding to ACE2[6]. In comparison, the SARS-CoV S protein has a receptor binding loop (residues 424-494) that directly interacts with ACE2, commonly known as the receptor binding motif[7]. The motif has many tyrosine residues; out of the 14 residues in the motif that interact with ACE2, 6 are tyrosine. Du et al have identified that any mutations that result in amino acid changes at positions 479 and 487 will increase binding affinity of the S protein to the ACE2, thereby increasing human to human transmission[7].

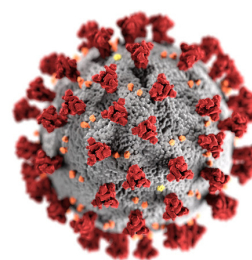
In the S protein of SARS-CoV, residue 487 is a threonine[7] while in SARS-CoV-2 S protein, the corresponding residue 501 is an asparagine[8]. The difference between T487 and N501, from threonine to asparagine, ultimately gave SARS-CoV-2 a better capability to bind to ACE2[8]. This is likely due to the additional support that this asparagine brings to stabilize the RBD[8]. In addition, there is a 4-residue motif (residues 482-485) that exists on the SARS-CoV-2 S protein's RBD[5]. This motif contains glycine, valine and glutamic acid that forms a more compact shape that can bind better to the N-terminal helix of ACE2[5].

In addition, F486 in the S protein's RBD binds better to the hydrophobic pocket of ACE2 due to its large side chain[5]. ACE2 contains viral hotspots that have to be stabilized in order to have effective binding between the S protein and the ACE2[5]. This is only a subset of information that explains why SARS-CoV2 can better transmit from one individual to another than SARS-CoV.

Aside from binding affinity, S proteins have to be cleaved using the host cell's proteases, specifically TMPRSS2[2]. S cleavage sites are conserved between the SARS-CoVs[2]. Research has provided strong evidence indicating that DCPs found near the S cleavage sites affect their sensitivity to TMPRSS2[2]. It is found that serine protease inhibitors, which ultimately inhibit S cleavage, have more of an effect on SARS-CoV-2 than on SARS-CoV[2]. Thus, DCPs are extremely important and bring great implications on the difference of clinical behaviour between SARS-CoVs[2].

However, it might be the case that the S protein of SARS-CoV-2 does not need host cell proteases in order to be activated and mediate membrane fusion[9]. Another finding that could explain SARS-CoV-2's efficient transmission capability[9]. Lastly, there is a cleavage motif that exists exclusively in the S protein of SARS-CoV-2 but not in SARS-CoV and MERS-CoV. This cleavage motif allows for cleavage by furin and phopho-regulation and has also been a prime suspect explaining the high infectivity rate of SARS-CoV-2[10].

Overall, structural changes between SARS-CoV and SARS-CoV-2 help to explain the differences we see in their clinical behaviour and can have great implications in how researchers move forward and make new advances in the medical field.



References

1. Giannis D, Ziogas IA, Gianni P. Coagulation disorders in coronavirus infected patients: COVID-19, SARS-CoV-1, MERS-CoV and lessons from the past. *Journal of Clinical Virology*. 2020;127:104362. doi:10.1016/j.jcv.2020.104362
2. Bojkova D, McGreig JE, McLaughlin K-M, et al. Differentially conserved amino acid positions may reflect differences in sars-cov-2 and sars-cov behaviour. *Bioinformatics*. 2021. doi:10.1093/bioinformatics/btab094
3. Simmons G, Reeves JD, Rennekamp AJ, Amberg SM, Piefer AJ, Bates P. Characterization of severe acute respiratory syndrome-associated coronavirus (SARS-CoV) spike glycoprotein-mediated viral entry. *Proceedings of the National Academy of Sciences*. 2004;101(12):4240-4245. doi:10.1073/pnas.0306446101
4. Choudhury A, Mukherjee S. In silico studies on the comparative characterization of the interactions of SARS-CoV-2 spike glycoprotein with ACE-2 receptor homologs and human TLRs. *Journal of Medical Virology*. 2020;92(10):2105-2113. doi:10.1002/jmv.25987
5. Shang J, Ye G, Shi K, et al. Structural basis of receptor recognition by SARS-CoV-2. *Nature*. 2020;581(7807):221-224. doi:10.1038/s41586-020-2179-y
6. Hussain M, Jabeen N, Raza F, et al. Structural variations in human ACE2 may influence its binding with SARS-CoV-2 spike protein. *Journal of Medical Virology*. 2020;92(9):1580-1586. doi:10.1002/jmv.25832
7. Du L, He Y, Zhou Y, Liu S, Zheng B-J, Jiang S. The spike protein of SARS-CoV — a target for vaccine and therapeutic development. *Nature Reviews Microbiology*. 2009;7(3):226-236. doi:10.1038/nrmicro2090
8. Yi C, Sun X, Ye J, Ding L, Liu M, Yang Z, Lu X, Zhang Y, Ma L, Gu W, Qu A, Xu J, Shi Z, Ling Z, & Sun B. Key residues of the receptor Binding motif in the spike protein OF SARS-COV-2 that interact With ACE2 and neutralizing antibodies. *Cellular & Molecular Immunology*. 2020;17(6):621-630. doi:10.1038/s41423-020-0458-z
9. Ou X, Liu Y, Lei X, et al. Characterization of spike glycoprotein of 2019-nCoV on virus entry and its immune cross-reactivity with spike glycoprotein of SARS-CoV. *Nature Communications*. 2020;11(1620):1-12. doi:10.1038/s41467-020-15562-9
10. Örd M, Faustova I, Loog M. Biochemical evidence of furin specificity and potential for phospho-regulation at Spike protein S1/S2 cleavage site in SARS-CoV2 but not in SARS-CoV1 or MERS-CoV. *Scientific Reports*. 2020;10(16944):1-10. doi:10.1038/s41598-020-74101-0



"Smart" Insulin - The Future of Pharmacotherapies for Diabetics

By: Payton Midgley

Introduction

Diabetes is a chronic disease where the body is either ineffective at producing enough insulin (type 1 diabetes, T1D) or is unable to effectively respond to insulin (type 2 diabetes, T2D), and is characterized by chronic hyperglycemia[1]. In 2019, it was estimated that over 463 million people (20-79 years) world-wide have diabetes[2], with over 1.5 million deaths being a direct result of diabetes[3]. About 3.2 million (8.8%) of Canadians were reported to have diabetes, with approximately 550 new cases every day[4]. The cost of treating diabetes was \$1.5 billion in Ontario alone, and just under \$30 billion nationally[5].

T1D requires insulin replacement therapy, whereas T2D may be managed through lifestyle changes; however, pharmacological adjunct therapy may be added[6]. Insulin replacement therapy is typically composed of a long-acting or ultra-long-acting insulin combined with a short-acting insulin[6,7]. Non-insulin replacement therapies include biguanides, sulfonylureas, thiazolidinediones, dipeptidyl peptidase-4 inhibitors, glucose-like peptide-1 receptor agonists, sodium-glucose cotransporter-2 inhibitors, and amylin analogs[6-8]. Traditionally, these therapies require multiple administrations throughout the day, and a common risk between both insulin and non-insulin treatment is hypoglycemia[1]. "Smart" insulin, or glucose-responsive insulin (GRI), are next generation insulin replacement therapies for T1D and T2D that was first proposed in 1979 by Michael Brownlee and Anthony Cerami[9]. Two "smart" insulin technologies, SmartInsulin and the smart patch, have gained popularity in the past few years as they have been reported to decrease the risk of hypoglycemia and improve the management of diabetes[10-13].

SmartCells and SmartInsulin

SmartCells was a start-up company founded by MIT graduate student Todd Zion in 2003 to engineer SmartInsulin (renamed MK-2640)[10,14]. MK-2640 is a glycosylated insulin that binds both the insulin receptor and endogenous mannose receptor C type 1 (MRC1) lectin protein in a manner that is dependent on glucose concentrations[10]. A rodent study by Yang et al. reported a dose-dependent decrease in glucose levels using mannosylated insulin without hypoglycemic levels, compared to recombinant insulin; this effect was greater in diabetic rodents compared to non-diabetic ones[11]. Mannosylated insulin was cleared at a faster rate by MRC1 than non-mannosylated and degraded by lysosomes at euglycemic or hypoglycemic states[11]. When glucose levels were elevated, mannosylated insulin was cleared at a slower rate, resulting in its accumulation and subsequent binding to insulin receptors to decrease glucose levels[11].

Similarly, in a phase I clinical trial by Krug et al., a 6% decrease in glucose levels following MK-2640 administration to patients with T1D was observed; however, this difference was not statistically significant, nor was as great as the 30% decrease observed in previous animal studies[10,15]. Furthermore, Krug et al. reported that hyperglycemia did not significantly decrease the systemic clearance rate of MK-2640[10], which is opposite compared to that observed by Yang et al. Interestingly, an increase in the quantity of MK-2640 was stimulated when there was a greater amount of glucose present[10], suggesting that MK-2640 is mimicking the endogenous insulin response as an increase in glucose results in the release of insulin from pancreatic β cells[1]. The significant pharmacodynamic changes and minimal pharmacokinetic changes observed in this phase I clinical trial presented great challenges to researchers that have yet to be further investigated.

Overall, the clinical trial was terminated due to limited efficacy[16]. Both studies point out two limitations that researchers are currently facing: GRI was less potent than other forms of insulin and had an increased systemic clearance rate[10,11]. Although glycosylated insulin had promising effects in animal studies as it simultaneously decreased the glucose levels without hypoglycemia, it has yet to be efficacious in humans.

Smart Patch

A smart patch is a removable coin-sized glucose-responsive micro-needle (GR-MN) transdermal patch, with the needles coated in an insulin-phenylboronic acid (PBA) complex mixture[12]. At physiological pH, PBA is positively charged and forms a complex with negatively charged insulin[12,13]. PBA also reversibly binds to diol substituents on compounds such as those found on glucose, therefore enabling PBA to serve as a glucose-response element[17]. Therefore, at hyperglycemic levels, PBA will bind to glucose which will decrease its electrostatic interaction with insulin[12,13]. The free insulin then binds to the insulin receptor to stimulate the uptake of glucose[1]. At euglycemic levels, this process is inhibited, which decreases the risk of hypoglycemia[12,13].

Yu *et al.* designed a microneedle that is synthesized based on *in situ* photopolymerization, which avoids the addition of extra organic solvents and elevated temperatures to maintain insulin integrity[13]. In streptozotocin (STZ)-induced diabetic mice, glucose levels returned to normal within 40 minutes post-glucose exposure in mice wearing the GR-MN patch, compared to controls[13]. The patch was able to regulate glucose levels for up to 10 h post-glucose administration[13]. In STZ-induced diabetic minipigs (which have comparable skin-type to humans) wearing the GR-MN patch, glucose levels returned to normal within 80 minutes post-feeding and remained at the reduced state overnight until the following meal[13]; however, it is unclear what the feeding schedule was.

In both animals, glucose levels were contained within normal ranges or 48 h and hypoglycemia levels and/or symptoms were not reported[13]. Yu *et al.* also reported that their patch can be personalized to patients to accommodate various weights and insulin sensitivity and was non-degradable[13]. The fact that the patch is non-degradable means that there is minimal risk of residual microneedles causing adverse effects; however, it may lead to long-term environmental problems.

Furthermore, a 2007 phase I clinical trial conducted by Altea Therapeutics (ClinicalTrials.gov identifier: NCT-00519623) investigated the pharmacokinetics and dynamics of the transdermal patch PassPort™ system on 9 participants with T1D[18,19]. Researchers found that insulin levels rose to therapeutic levels over a period of 4 hours following application of the patch and maintained steady state levels until removal[19]. PassPort™ is a novel diabetic therapeutic technology that provides a non-invasive alternative to the current injection therapies and has a better pharmacoeconomic profile[20]. Researchers are actively progressing through the drug development pipeline to design a therapeutic transdermal patch[20]. Future clinical trials have yet to be conducted to further investigate the efficacy of the patch in both T1D and T2D.

Conclusion

The future of diabetic pharmacotherapies is rapidly evolving with new innovative technologies such as SmartInsulin (MK-2640) and the smart patch. Although MK-2640 was unsuccessful in clinical trials, it still holds great potential to undergo modifications that make it more robust than current therapies. Furthermore, the transdermal patch provides a simple and convenient insulin supplementation therapy that could improve the management of diabetes, especially T1D in children since it is easier to teach a child how to put on a sticker than use an insulin pen. Both technologies offer the potential to reduce health care costs, improve the ease of the management of diabetes, and decrease the risk of hypoglycemia.



References

1. Guthrie RA, Guthrie DW. Pathophysiology of Diabetes Mellitus. *Crit Care Nurse Q.* 2004;27(2):113-125.
2. International Diabetes Federation. *IDF Diabetes Atlas.* 9th ed. https://www.diabetesatlas.org/upload/resources/material/20200302_133351_IDFATLAS9e-final-web.pdf. Accessed April 24, 2021.
3. Diabetes. World Health Organization. <https://www.who.int/news-room/fact-sheets/detail/diabetes>. Published April 13, 2021. Accessed April 24, 2021.
4. LeBlanc AG, Gao YJ, McRae L, Pelletier C. At-a-glance - Twenty years of diabetes surveillance using the Canadian Chronic Disease Surveillance System. *Health Promot Chron.* 2019;39(11):306-309.
5. New Data Shows Diabetes Rates And Economic Burden On Families Continue To Rise In Ontario. Diabetes Canada. <https://www.diabetes.ca/media-room/press-releases/new-data-shows-diabetes-rates-and-economic-burden-on-families-continue-to-rise-in-ontario-->. Published February 12, 2019. Accessed April 24, 2021.
6. Chatterjee S, Davies MJ. Current management of diabetes mellitus and future directions in care. *Postgrad Med J.* 2015;91(1081):612-621.
7. Ludvigsson J. The latest pharmacotherapy options for type 1 diabetes. *Expert Opin Pharmaco.* 2014;15(1):37-49.
8. Upadhyay J, Polyzos SA, Perakakis N, et al. Pharmacotherapy of type 2 diabetes: An update. *Metabolis.* 2018;78:13-42.
9. Brownlee M, Cerami A. A Glucose-Controlled Insulin-Delivery System: Semisynthetic Insulin Bound to Lectin. *Science.* 1979;206(4423):1190-1191.
10. Krug AW, Visser SAG, Tsai K, et al. Clinical Evaluation of MK-2640: An Insulin Analog With Glucose-Responsive Properties. *Clin Pharmacol Ther.* 2019;105(2):417-425.
11. Yang R, Wu M, Lin S, et al. A glucose-responsive insulin therapy protects animals against hypoglycemia. *JCI Insight.* 2018;3(1).
12. Yu J, Wang J, Zhang Y, et al. Glucose-responsive insulin patch for the regulation of blood glucose in mice and minipigs. *Nat Biomed Eng.* 2020;4(5):499-506.
13. Wang J, Yu J, Zhang Y, et al. Charge-switchable polymeric complex for glucose-responsive insulin delivery in mice and pigs. *Sci Adv.* 2019;5(7).
14. Matheson R. The half-billion-dollar idea. *MIT News | Massachusetts Institute of Technology.* <https://news.mit.edu/2013/todd-zion-smartcells-0408>. Published April 8, 2013. Accessed April 24, 2021.
15. Kaarsholm NC, Lin S, Yan L, et al. Engineering Glucose Responsiveness Into Insulin. *Diabetes.* 2018;67(2):299-308.
16. Visser SAG, Kandala B, Fancourt C, Krug AW, Cho CR. A Model-Informed Drug Discovery and Development Strategy for the Novel Glucose-Responsive Insulin MK-2640 Enabled Rapid Decision Making. *Clin Pharmacol Ther.* 2020;107(6):1296-1311.
17. Matsumoto A, Yoshida R, Kataoka K. Glucose-Responsive Polymer Gel Bearing Phenylborate Derivative as a Glucose-Sensing Moiety Operating at the Physiological pH. *Biomacromolecules.* 2004;5(3):1038-1045.
18. Altea Therapeutics. Transdermal Basal Insulin Patch Study in Type 1 Diabetes. *ClinicalTrials.gov.* <https://clinicaltrials.gov/ct2/show/results/NCT00519623?term=transdermal+patch&cond=Diabetes+Mellitus&draw=2&rank=4>. Published August 22, 2007. Updated December 30, 2010. Accessed April 27, 2021.
19. Smith A. Transdermal Delivery of Insulin in Patients with Type 1 Diabetes. American Diabetes Association. <https://professional.diabetes.org/abstract/transdermal-delivery-insulin-patients-type-1-diabetes>. Published 2008. Accessed April 27, 2021.
20. PassPort® Novel Transdermal Drug Delivery System. PassPort Technologies. <https://passport-tech.com/>. Updated 2021. Accessed April 27, 2021.

THC's Analgesic Effect Through Altering Neuronal Pain-Processing Pathways

By: Jia Yin Lu

Prevalence of Chronic Pain

Chronic pain affects 20% of adults globally, with 10% of adults newly diagnosed with it each year[1]. Though there are several highly effective pain treatments including non-steroid anti-inflammatory drugs (NSAIDs) and opioids, prolonged use of these drugs carry multiple risks, such as cardiac failure or severe gastrointestinal bleeding[2]. Additionally, certain chronic conditions such as neuropathic pain are more resistant to opioids, rendering these drugs ineffective[3]. Neurological pain results from diseases or lesions in the somatosensory pathways[4], and an increasing number of studies are finding cannabinoid extracts useful in reducing both acute and neuropathic pain while having fewer side effects than NSAIDs and opioids[5].

Specifically, a recent study found THC to reduce experimentally induced pain in healthy participants[6]. The subjects' reduction in perceived pain correlated with lower activity levels in their Anterior Cingulate Cortex (ACC), an area that processes the affective components of pain[7]. This prompted Weizman et al.[8] to further investigate the role of the ACC and other neuronal networks in modulating cannabinoid-induced analgesia. They found that THC reduced the functional connectivity of major pain pathways, including the ACC and the dorsolateral prefrontal cortex (DLPFC), which correlated with a reduction in the patients' perceived pain. This reveals a promising mechanism to target when studying new treatments for pain reduction.

Downregulation of ACC-SII Functional Connectivity

Weizman et al.[8] conducted a counter-balanced study composed of seventeen male participants ages 27-40 with chronic neuropathic pain and no other comorbidities.

The participants received either 0.2mg/kg of THC or placebo oil in the first round and vice-versa in the second round after a one-week washout period. Each participants' baseline subjective pain ratings and resting-fMRIs were recorded and collected again 2 hours after oil administration. THC oil was found to significantly reduce the patients' self-reported chronic pain. Both groups had statistically similar baseline values, but only the THC group reported a significant decrease in perceived pain after treatment.

Resting-state fMRI scans were also performed on the subjects both pre- and post-treatment to measure the activity between the ACC and the rest of the brain. After comparing the two resting-state scans, only the THC group showed a reduction in functional connectivity between the ACC and the Somatosensory Cortex (SII). The ACC is involved in the emotional evaluation of pain while the SII processes sensory information[7]; the reduction in functional connectivity covaried with lower pain ratings after THC administration, while no significant differences were found in the fMRI scans of the placebo group. This correlation illuminates a potential target when treating patients with chronic pain: future studies can examine other methods and/or medications that disrupt the integration of these pathways in hopes of providing neuropathic pain relief.

Mechanistic Background of Pain Processing Pathways

Previous literature has revealed that the ACC regulates the cognitive evaluation of pain and that its pathways are heavily interconnected with the SII, which is involved in the sensory process of pain[7,9]. Thus, having lower ACC-SII functional integration corresponding to pain-reduction is consistent based on published research and builds on our existing knowledge of how these systems modulate pain.



Similarly, THC was found to lower network interconnectivity of the DLPFC to several brain regions, which again correlated with reduced pain ratings from patients[8]. The DLPFC is another brain region majorly involved in pain perception through active control of downstream cortical pathways[10]. Individuals with neuropathic pain have been found to have higher baseline connectivity of the DLPFC[11] – further implicating that disrupting the pain-affect pathways like the ACC and DLPFC may be key to mitigating chronic pain.

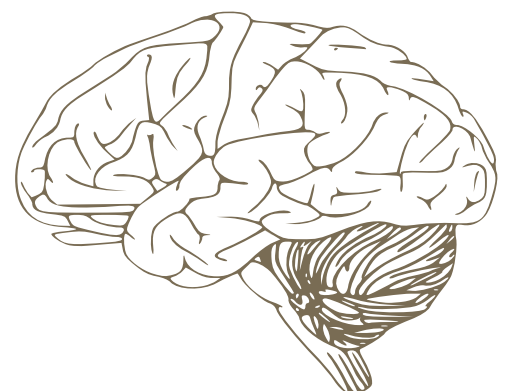
Potential Risks of THC in Chronic Pain Treatment

Weizman et al.[8] went beyond examining THC's effects on pain – they also compared pre- and post-treatment data on anxiety, blood pressure, and heart rate. The researchers found no changes in any of these values when compared to the subjects' baseline records. This is reassuring as THC has been shown to induce anxiety and paranoia in both healthy subjects and vulnerable individuals who occasionally experience paranoia[12,13]. However, existing evidence claims that THC-induced psychosis risk is strongly dose-dependent and tends to increase with prolonged use[14,15]. Therefore, the lack of changes in anxiety and cardiac measures in Weizman et al's study may be due to the low and infrequent THC administration. Thus, these side effects should be further investigated to determine the optimal dosage and frequency of THC use to maximize analgesic effects while minimizing psychiatric risks.

Future Directions

Weizman et al.[8] established an enlightening mechanism in how THC works to modulate chronic pain. Their finding opens the discussion for scientists to develop ways to maximize THC's efficacy in disrupting the ACC and DLPFC functional networks. However, THC is not the only therapeutic agent of cannabis. Many studies have shown that CBD not only provides analgesic relief but can also combat psychotic symptoms induced by THC[16].

Thus, to expand on the findings by Weizman et al., scientists could replicate this study by investigating the results of treatment groups that receive pure CBD and a THC/CBD mix. Furthermore, this can be performed as a long-term experiment where each of the groups are given a regular dosage of their respective treatment for an extended period of time. Although there is plentiful data to support THC's effectiveness in reducing the physiological aspects of chronic pain[5], there is a lack of studies looking at the long-term functional or structural changes in the pain pathways, as done by Weizman et al. Thus, repetition of their experiment in a longer duration study may be used to examine potential THC-induced neuroplasticity of these pain pathways. A continual design would also allow researchers to assess any potential side effects that manifest over prolonged use. Overall, there is emerging mechanistic support of using THC as chronic pain treatment, though further research on the safeness of this ingredient is necessary to enhance efficacy and minimize adverse effects.



References

1. Goldberg DS, McGee SJ. Pain as a global public health priority. *BMC Public Health*. 2011;11(1):770. doi:10.1186/1471-2458-11-770
2. Russo EB. Cannabinoids in the management of difficult to treat pain. *Ther Clin Risk Manag*. 2008;4(1):245-259. doi:10.2147/tcrm.s1928
3. Hamon M, Kayser V, Bourgoin S. [Neuropathic pain. Physiopathological mechanisms and therapeutic perspectives]. *Bull Academie Natl Med*. 2008;192(5):921-926.
4. Nascimento OJM, Pessoa BL, Orsini M, et al. Neuropathic pain treatment: still a challenge. *Neurol Int*. 2016;8(2). doi:10.4081/ni.2016.6322
5. Reiman A, Welty M, Solomon P. Cannabis as a Substitute for Opioid-Based Pain Medication: Patient Self-Report. *Cannabis Cannabinoid Res*. 2017;2(1):160-166. doi:10.1089/can.2017.0012
6. Lee MC, Ploner M, Wiech K, et al. Amygdala activity contributes to the dissociative effect of cannabis on pain perception. *Pain*. 2013;154(1):124-134. doi:https://doi.org/10.1016/j.pain.2012.09.017
7. Rainville P, Duncan G, Price D, Carrier B, Bushnell M. Pain Affect Encoded in Human Anterior Cingulate But Not Somatosensory Cortex. *Science (New York, NY)*. 1997;277:968-971. doi:10.1126/science.277.5328.968
8. Weizman L, Dayan L, Brill S, et al. Cannabis analgesia in chronic neuropathic pain is associated with altered brain connectivity. *Neurology*. 2018;91:1285-1294. doi:10.1212/WNL.0000000000006293
9. Price DD. Psychological and Neural Mechanisms of the Affective Dimension of Pain. *Science*. 2000;288(5472):1769-1772. doi:10.1126/science.288.5472.1769
10. Lorenz J, Minoshima S, Casey KL. Keeping pain out of mind: the role of the dorsolateral prefrontal cortex in pain modulation. *Brain*. 2003;126(5):1079-1091. doi:10.1093/brain/awg102
11. Cauda F, Sacco K, Duca S, et al. Altered Resting State in Diabetic Neuropathic Pain. *Plos One*. 2009;4(2):4542. doi: 10.1371/journal.pone.0004542
12. Morrison PD, Zois V, McKeown DA, et al. The acute effects of synthetic intravenous Delta9-tetrahydrocannabinol on psychosis, mood and cognitive functioning. *Psychol Med*. 2009;39(10):1607-1616. doi:10.1017/S0033291709005522
13. Freeman D, Dunn G, Murray RM, et al. How Cannabis Causes Paranoia: Using the Intravenous Administration of Δ 9-Tetrahydrocannabinol (THC) to Identify Key Cognitive Mechanisms Leading to Paranoia. *Schizophr Bull*. 2015;41(2):391-399. doi:10.1093/schbul/sbu098
14. Henquet C & Kuepper R. Does cannabidiol protect against the negative effects of THC? *The Br J Psychiatry*. 2010;197(4):259-260. doi:10.1192/bjp.bp.110.081380
15. Ware MA, Wang T, Shapiro S, et al. Smoked cannabis for chronic neuropathic pain: a randomized control trial. *CMAJ*. 2015;182(14):694-701. doi:10.1503/cmaj.091414
16. Bhattacharyya S, Morrison P, Fusar-Poli P, et al. Opposite Effects of delta-9-Tetrahydrocannabinol and Cannabidiol on Human Brain Function and Psychopathology. *NPP*. 2010;35(3):764-774. doi:10.1038/npp.2009.184



A New Treatment for Tumor Ablation and Drug Delivery

By: Daphne Lin

Hepatocellular carcinoma (HCC), is the most common type of liver cancer[1]. This neoplasia can develop due to viral hepatitis, excessive alcohol intake, or metabolic steatohepatitis, and is related to chronic liver diseases such as cirrhosis[2]. The 5-year survival rate after diagnoses for regional or metastatic liver cancer is under 10%. HCC treatment is difficult due to the high rate of late diagnosis as well as limited therapeutic options[2]. Treatment can be carried out by systemic chemotherapy; however, poor uniformity of drug delivery, toxicity to noncancerous tissue, and various side effects are all issues with this type of therapy[1]. In 1996, a novel study established liver transplantation as being the best treatment option for patients with HCC as it removes the known cancer as well as other undetected precancerous lesions[3]. With a 5-year survival rate sitting near 80%, liver transplantation is the best course of action for early-stage HCC[1]; however, due to limited organ availability, selection and management of patients who may have the best survival benefits can be difficult[3]. As only some stages of the disease are amenable for this option[3], the goal in clinical practice today is to maintain patients within transplant criteria until a liver is available[1].

In order to limit the stay on waitlist, percutaneous loco-regional therapies (LRTs) are performed[1]. HCC lesions can be resected in surgery; however, in most cases, this is not possible due to conditions such as cirrhosis. Additionally, surgical removal, even with favourable survival, has quite a high rate of recurrence[4]. LRTs such as thermal ablation, transarterial chemoembolization (TACE), and radiofrequency ablation are more widely used as they have better efficacy and safety[4].

The prevention of HCC progression through use of LRTs is favorable, but due to physiological challenges, such as tumor location and size, existence of multiple lesions, and concerns for injury to adjacent tissues, can all affect the use of these therapies before liver transplantation[1]. Liquid ablation agents such as percutaneous ethanol (EtOH) injection are used as an LRT currently as it causes dehydration and necrosis of tumor cells[5]. The use of this therapy can be attributed to its low cost and wide availability, however, it can lead to many complications such as portal vein thrombosis, hemoperitoneum, and liver failure, due to inefficient intratumoral diffusion[1].

Intravenous drug delivery within tumors is especially challenging in oncology as it is difficult to achieve uniform distribution[1]. Additionally, as the dose of drug is increased in order to penetrate the tumor, drug toxicity can become a barrier to this type of therapy[1]. This is because intravenous administration is not directly administered to the target tissues which can lead to nonspecific accumulation in body organs instead[1]. Direct intratumoral injection is an approach that can produce high local concentrations of therapy while being able to avoid systemic toxicity[1]. Although there is an abundance of clinical trials investigating intratumoral injection of varying therapeutics, including chemotherapy and immunotherapy, there is not yet a method that maximizes drug distribution and retention[1].

A novel ionic liquid-based agent termed, Locally-active Agent for Tumor Treatment and Eradication (LATTE), was developed to function as a drug carrier and percutaneous tumor ablation agent in a recent study[1]. The scientists hypothesized that ionic liquids could be able to induce local tumor ablation and deliver chemotherapeutic drugs. The ionic composition of the liquid can alter the local osmotic condition allowing for the ablation of tumor cells [1].

Additionally, hydrophobic anions in the ionic liquid may interact with the lipids in the cell membranes and enhance diffusion through the tissue [1]. Through intratumoral injection, this liquid would be able to deliver chemotherapy drugs directly to the tumor and avoid systemic toxicity[1].

LATTE was injected into the tissue under image guidance and was able to ablate tumors as well as maintain anticancer therapy for prolonged periods [1]. It was also found that LATTE was able to provoke a local immuno-stimulatory response suggesting the possible promotion and enhancement of immunotherapy[1]. The preparation of this novel ionic liquid-based composition was done using choline-geranate ionic liquid as the base ingredient. LATTE was then delivered intratumorally by needle-based injection into rat, rabbit, or pig liver tissues in order to assess distribution and monitor diffusion capability using ultrasound, microcomputed tomography imaging, and real-time magnetic resonance imaging [1]. LATTE was found to have spread rapidly within the liver and remained visible at 24 hours[1]. The injection into the tissue produced uniform, reproducible ablation zones in the rat, rabbit, and pig livers as well as distributed fluorescent dye indocyanine green (ICG), the x-ray contrast agent ExiTron and doxorubicin (Dox), surrogates for a drug, evenly and uniformly throughout the ablation zone, outperforming the commonly used EtOH injection[1]. Additionally, experiments indicated that LATTE had synergistic effects with anticancer drugs such as Dox, which could enhance its ablation capabilities[1]. LATTE at a low cost is able to successfully ablate tumors and hopefully become a more appealing LRT, bridging patients with HCC waiting for liver transplants.

The team that developed LATTE is now planning on investigating the mechanism in which it is able to induce cancer cell death as well as investigate the delivery of other types of anticancer therapies[1].

In addition to demonstrating activity in liver tumors, LATTE was also effective in 12 human tumors of varying types suggesting its efficacy against a wide range of solid tumors in other organs[1]. This means that LATTE may also be able to help treat residual tumors after resection, metastatic lesions, or nonresectable tumors[1]. Dox was the only chemotherapy drug used in the experiments, so looking into the delivery and functionality of other anticancer therapies, such as protein-based drugs, and the effects of the ionic properties of LATTE on their activity could further demonstrate this new LRT's capabilities[1].

The demonstration that LATTE is able to successfully ablate tumors in rat, rabbit, and pig livers as well as its efficacy as a drug carrier, leads to the idea that it can also be used as a drug carrier solvent or even within nanoparticles delivering anticancer therapeutics in the future[1]. The use of nanoparticles to deliver drugs is a relatively new and rapidly developing area where materials in the nanoscale range are able to deliver therapeutic agents to targeted sites[6]. Large sized materials in drug delivery lead to many challenges including in vivo instability, poor bioavailability, poor solubility, and poor absorption[6]. By using nanotechnology, therapeutic agents can deliver drugs to specific tissues and provide controlled release therapy enhancing the persistence of drugs within an ablation zone for an extended period of time[7]. This would decrease drug related toxicity and require less frequent dosing[7]. LATTE represents a new class of anticancer therapeutics that is able to efficiently ablate tissue alone[1]. If it can be used within nanoparticles as a drug carrier, the ability in which it is able to deliver drugs to affected areas may be enhanced leading to the improvement of survival outcomes in patients with cancer.





References

1. Albadawi H, Zhang Z, Altun I, et al. Percutaneous liquid ablation agent for tumor treatment and drug delivery. *Science Translational Medicine*. 2021;13(580). doi:10.1126/scitranslmed.abe3889
2. Le Grazie M, Biagini MR, Tarocchi M, Polvani S, Galli A. Chemotherapy for hepatocellular carcinoma: The present and the future. *World Journal of Hepatology*. 2017;9(21):907. doi:10.4254/wjh.v9.i217.90
3. Santopaolo F, Lenci I, Milana M, Manzia TM, Baiocchi L. Liver transplantation for hepatocellular carcinoma: Where do we stand? *World Journal of Gastroenterology*. 2019;25(21):2591-2602. doi:10.3748/wjg.v25.i21.2591
4. Byrne TJ, Rakela J. Loco-regional therapies for patients with hepatocellular carcinoma awaiting liver transplantation: Selecting an optimal therapy. *World Journal of Transplantation*. 2016;6(2):306. doi:10.5500/wjt.v6.i2.306
5. Swierz MJ, Storman D, Riemsma RP, et al. Percutaneous ethanol injection for liver metastases. *Cochrane Database of Systematic Reviews*. 2020. doi:10.1002/14651858.cd008717.pub3
6. Patra JK, Das G, Fraceto LF, et al. Nano based drug delivery systems: recent developments and future prospects. *Journal of Nanobiotechnology*. 2018;16(1). doi:10.1186/s12951-018-0392-8
7. Rizvi SAA, Saleh AM. Applications of nanoparticle systems in drug delivery technology. *Saudi Pharmaceutical Journal*. 2018;26(1):64-70. doi:10.1016/j.jsps.2017.10.012

RAS Modulators and the Future of PTSD

Pharmacotherapy

By: Karen Kuang

Experiencing stress in the wake of and immediately following a traumatic event is a universal experience. It is well known that a sense of danger triggers the fight-or-flight response resulting in a whole myriad of physiological changes including an increased heart rate, perspiration, etc. Learning to fear danger is a survival mechanism and the stress response is meant to aid in coping with trauma; both are temporary measures that fade away as the dangerous situation becomes removed from the individual. Psychopathology emerges when the individual fails to properly recover from this stress response and chronically suffers from re-experiencing the trauma, negative cognitive/mood symptoms, hyperarousal symptoms, and avoidance symptoms; this is post-traumatic stress disorder (PTSD)[1].

As treatment for a psychological disorder, the focus of PTSD pharmacotherapy has largely been on neuropharmacological interventions. First-line drugs for PTSD include selective serotonin reuptake inhibitors (SSRIs) and serotonin-norepinephrine reuptake inhibitors (SNRIs)[1]. Both classes of antidepressants are effective in managing PTSD's hyperarousal and mood symptoms[1].

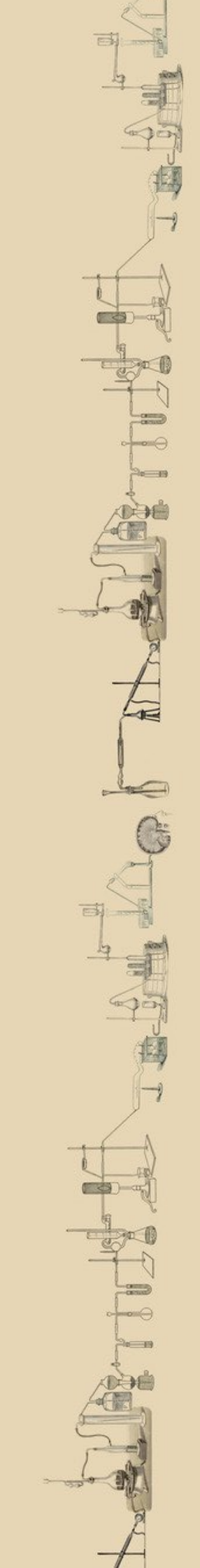
The relationship between the renin-angiotensin system and PTSD

More recently, researchers have begun exploring the possibility of treating PTSD symptoms with angiotensin-converting-enzyme (ACE) inhibitors (ACE-Is) and angiotensin receptor blockers (ARBs). Both classes of drugs are typically used as antihypertensives and target the renin-angiotensin system (RAS), regulating blood pressure through inhibiting angiotensin II-induced blood pressure raising activities like vasoconstriction and vasopressin release[2].

The interest in repurposing ACE-Is and ARBs as PTSD medications would be surprising if it were not for the numerous observational studies that found an association between PTSD and increased risk for cardiovascular disease[3-5]. Hypertension is a common comorbidity with this psychological disorder and many PTSD patients are also on blood pressure medications, among them ACE-Is and ARBs. Given the common comorbidity, Khoury et al. evaluated the PTSD diagnosis, symptom severity and use of blood pressure medication in a traumatized population[6]. This team observed an association between ACE-Is/ARB use and PTSD symptom levels, namely that the use of ACE-Is/ARBs was significantly associated with decreased PTSD symptoms[6]. This study was successfully replicated in 2021 by Seligowski et al., confirming that future exploration of ACE-I/ARB use in the treatment of PTSD is worthwhile. It is also worth noting that this study team investigated the relationship between other classes of antihypertensives (beta blockers, calcium channel blockers, diuretics) and PTSD; they found none, providing further evidence that the renin-angiotensin system plays a particular role in this debilitating psychopathology[7].

As of right now, these observational studies cannot definitively prove that there is a causal relationship between the variables in question, but previous research has shown that type 1 angiotensin (AT1) receptors, the target receptor of angiotensin II, are highly expressed throughout the HPA axis as well as in stress-regulating areas of the brain: the hippocampus, septum, and amygdala[8]. Additionally, the activation of beta-adrenergic receptors in the sympathetic stress response leads to increased renin levels, thus also increasing circulating amounts of angiotensin II which then bind to the AT1 receptors in the nervous system causing downstream enhancement of stress hormone release[9].





Thus, it is speculated that ACE-Is, in inhibiting the formation of angiotensin II, and ARBs, in antagonizing the AT1 receptor, modulate stress by disrupting the above chain of events[10]. The story may not be so simple since adrenomedullary tissue expresses both AT1 and AT2 receptors and it is actually the second variety that is more highly expressed; potential crosstalk between the two receptor types may add another layer of complexity to the hypothesized mechanism of action[9].

Other mechanisms by which RAS-modulating antihypertensives potentially decrease PTSD symptoms are also possible. A 2014 study conducted by Marinzalda et al. looked at the effect of blocking AT1 receptors in the amygdala on fear-potentiated behaviour in a rat model[11]. Fear conditioning was done by treating rats with footshocks and behaviour was analyzed using the elevated plus maze assay. Marinzalda et al. found that blocking AT1 receptors in the amygdala with losartan, an ARB, had an anxiolytic effect, preventing the decrease in time spent in the open arms of the maze that occurred in the control group[12]. The study team showed that the AT1 receptors in the amygdala contribute to the fear conditioning process and it would thus be interesting to explore whether or not ACE-Is and ARBs could be used as PTSD prophylaxis. Interestingly enough, Khoury et al. and Seligowski et al. both found that among those who experienced trauma, these two classes of antihypertensives were associated with lower rates of PTSD diagnosis[6,7].

However, as a disease characterized by the failure to recover, there is also the potential that ACE-Is/ARBs could play a role in healing the PTSD patient's pathological fear extinction, the process by which one unlearns a conditioned fear after non-harmful exposure to fear-inducing stimuli. In a 2014 study, Marvar et al. used a mouse model of PTSD and treated the mice with losartan[12].

They found that treatment with this ARB enhanced the fear extinction process in the mice; the group treated with losartan were better able to unlearn fear compared to the control group[13]. While clinical experimentation will be needed to confirm the effect in humans, these promising results indicate that AT1 receptor blockade could potentially be of great benefit in treating patients whose symptoms include the re-experiencing of trauma.

Pharmacogenetic considerations

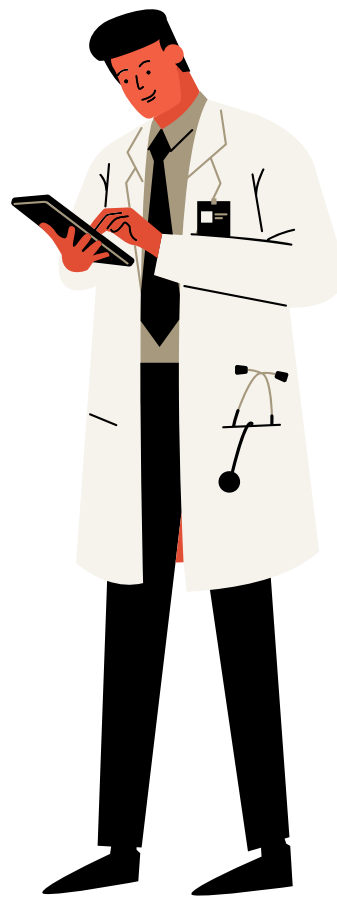
There is a growing body of evidence indicating neuroprotective benefits of ACE-Is and ARBs, justifying further investigation into the use of these drugs as PTSD pharmacotherapy, but there has been little research done on potential pharmacodynamic variation among different populations and individuals. One of the first to conduct this research was a 2015 study by Nylocks et al. which sought to elucidate the potential effect of genetic variation in the ACE gene on ACE-I/ARB treatment for PTSD[10]. One such source of genetic variation is the rs4311 single nucleotide polymorphism (SNP) which is associated with panic attacks[14]. Nylocks et al. recruited patients from a traumatized population and then assessed their rs4311 genotype, their ACE-I/ARB use, as well as their PTSD diagnosis and symptom levels. Not only was the rs4311 SNP found to be associated with PTSD symptoms and diagnosis, but the effect of ACE-I/ARB on symptoms and diagnosis was found to be different for those with the CC rs4311 genotype compared to T-carriers (CT or TT genotype) at the rs4311 SNP location on the ACE gene. From the analysis of Nylocks et al., T-carriers had a significantly greater risk of PTSD diagnosis and also did not reap any benefits from ACE-Is/ARBs with regards to PTSD symptoms—in fact, while the presence of RAS-modulating antihypertensives was associated with lower PTSD symptoms in those with the CC genotype, in T-carriers, the medications were associated with greater PTSD symptoms[10].

Given this result, PTSD treatment with ACE Is/ARBs would be beneficial to individuals with the CC genotype, but likely would not aid in remedying PTSD symptoms in individuals carrying a T allele; these treatments may even be harmful—this finding is of great clinical significance and must be considered in future investigations.

Conclusion

As future studies continue, it will be important to investigate the effects of race, sex, and genetics on the impact of ACE-Is/ARBs on PTSD. Clinical trials will also be necessary to determine if there is any causal relationship in the associations observed between these drugs and this disease.

PTSD patients often experience comorbid hypertension, so the possibility of using RAS modulating pharmacotherapy to treat PTSD symptoms in addition to high blood pressure is quite exciting—these drugs already have an established safety profile that involves relatively little risk[15]; this situation could be one in which medicine can achieve the proverbial killing of two birds with one stone. And as there are few pharmacotherapeutic options for PTSD patients in existence, adding this psychopathology to the indication of ACE-Is/ARBs would be of great benefit to those suffering from this disease.



References

1. Kirkpatrick H, Heller G. Post-Traumatic Stress Disorder: Theory and Treatment Update. *The International Journal of Psychiatry in Medicine*. 2014;47(4):337-346. doi:10.2190/pm.47.4.h
2. Barreras A, Gurk-Turner C. Angiotensin II Receptor Blockers. *Baylor University Medical Center Proceedings*. 2003;16(1):123-126. doi:10.1080/08998280.2003.11927893
3. Gupta M. Review of somatic symptoms in post-traumatic stress disorder. *International Review of Psychiatry*. 2013;25(1):86-99. doi:10.3109/09540261.2012.736367
4. Mcfarlane A, Atchison M, Rafalowicz E, Papay P. Physical symptoms in post-traumatic stress disorder. *J Psychosom Res*. 1994;38(7):715-726. doi:10.1016/0022-3999(94)90024-8
5. Kibler J, Joshi K, Ma M. Hypertension in Relation to Posttraumatic Stress Disorder and Depression in the US National Comorbidity Survey. *Behavioral Medicine*. 2009;34(4):125-132. doi:10.3200/bmed.34.4.125-132
6. Khoury N, Marvar P, Gillespie C, et al. The Renin-Angiotensin Pathway in Posttraumatic Stress Disorder. *J Clin Psychiatry*. 2012;73(06):849-855. doi:10.4088/jcp.11m07316
7. Seligowski A, Duffy L, Merker J, et al. The renin-angiotensin system in PTSD: a replication and extension. *Neuropsychopharmacology*. 2021;46(4):750-755. doi:10.1038/s41386-020-00923-1
8. Saavedra J, Sánchez-Lemus E, Benicky J. Blockade of brain angiotensin II AT1 receptors ameliorates stress, anxiety, brain inflammation and ischemia: Therapeutic implications. *Psychoneuroendocrinology*. 2011;36(1):1-18. doi:10.1016/j.psyneuen.2010.10.001
9. Jezova M, Armando I, Bregonzio C, et al. Angiotensin II AT1 and AT2 Receptors Contribute to Maintain Basal Adrenomedullary Norepinephrine Synthesis and Tyrosine Hydroxylase Transcription. *Endocrinology*. 2003;144(5):2092-2101. doi:10.1210/en.2002-0019
10. Nylocks K, Michopoulos V, Rothbaum A, et al. An angiotensin-converting enzyme (ACE) polymorphism may mitigate the effects of angiotensin-pathway medications on posttraumatic stress symptoms. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*. 2015;168(4):307-315. doi:10.1002/ajmg.b.32313
11. Marinzalda M, Pérez P, Gargiulo P, Casarsa B, Bregonzio C, Baiardi G. Fear-Potentiated Behaviour Is Modulated by Central Amygdala Angiotensin IIAT1Receptors Stimulation. *Biomed Res Int*. 2014;2014:1-7. doi:10.1155/2014/183248
12. Marvar P, Goodman J, Fuchs S, Choi D, Banerjee S, Ressler K. Angiotensin Type 1 Receptor Inhibition Enhances the Extinction of Fear Memory. *Biol Psychiatry*. 2014;75(11):864-872. doi:10.1016/j.biopsych.2013.08.024
13. Erhardt A, Lucae S, Kern N, et al. Association of polymorphisms in the angiotensin converting enzyme gene with syndromal panic attacks. *Mol Psychiatry*. 2008;13(3):242- 243. doi:10.1038/sj.mp.4002094
14. Conlin P. Efficacy and safety of angiotensin receptor blockers: a review of losartan in essential hypertension. *Current Therapeutic Research*. 2001;62(2):79-91. doi:10.1016/s0011-393x(01)80019-9

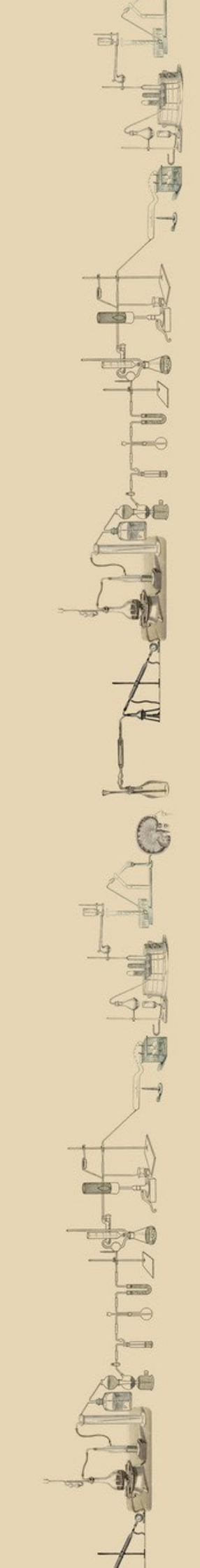
Psychedelic Microdosing: Public Health Concern or Novel Therapeutic?

By: Rachel Kuah

The potential for psychedelic microdosing to revolutionize the treatment of psychiatric disorders has recently become a popular topic of discussion within the scientific community. Psychedelic microdosing involves small doses of substances like lysergic acid diethylamide (LSD) or psilocybin taken every few days, with the intention of improving one's mental health, well-being, or cognition without eliciting a perception-altering experience[1,2]. Interest in psychedelic microdosing has been largely motivated by a plethora of anecdotal evidence recently provided by the public sphere and predominantly found in online forums like Reddit. Many users claim that psychedelic microdosing is an effective treatment for various psychiatric illnesses and mood disorders, while healthy individuals claim they can also be used more generally to improve creativity, productivity, emotional well-being and cognition[1,3-6]. The most commonly discussed psychedelics are LSD and dried psilocybin-containing mushrooms. This is not the first time scientists have investigated the potential therapeutic properties of psychedelics, as scientists were previously making ground-breaking discoveries on full doses as early as the 1940's, before government regulations prohibited their use in the 1970's[2]. With more countries now legalizing psychedelics, and influential figures openly sharing their experience with these drugs, the public's interest in psychedelic microdosing has grown faster than scientific research has been able to fully determine their short- and long-term effects[2]. Further, with psychedelic microdosing gaining attention from celebrities and various social media platforms, the need for scientific evidence to inform the public of the true effects of psychedelic microdosing is imperative to public health and safety.

Microdosing refers to the use of minute amounts of a substance and is commonly used in pharmacology to study the pharmacokinetics of a drug[2]. The generally accepted range for psychedelic microdosing is between 5% to 10% of a full dose[2]. Thus, LSD microdoses range from 5-20 micrograms while psilocybin microdoses from mushrooms range from 0.1-0.5 grams[1]. Psychedelics are potent agonists for the serotonergic receptor, 5-HT_{2A}, expressed highly in the brain, however it can activate other subtypes like the 5-HT_{1A} receptor, or receptors belonging to other neurotransmitter systems such as dopamine and adrenergic receptors[2,7]. Currently, more research has been conducted for the potential therapeutic effects of full dosing with psychedelics rather than microdosing. This research focuses on the potential of larger doses to treat mental illnesses such as depression, anxiety in end-of-life care or terminal cancer, and addiction to alcohol or tobacco[6]. However, the growing use of psychedelic microdosing by both clinical and non-clinical populations, often unsupervised by or unknown to users' healthcare providers, obligates scientists to rigorously investigate the short- and long-term effects of microdosing. Although psychedelics selectively target the 5-HT_{2A} receptor, microdosing research should not be completely informed by findings from full dose studies since microdosing does not evoke perception-altering experiences and other intense phenomena associated with full dosing[1,2]. As microdosing elicits a low receptor occupancy rate around 2%, it could stimulate modified 5-HT_{2A} receptor signaling that differs from full dosing on both the behavioral and therapeutic level[8]. Research on psychedelic microdosing is strongly justified as full dosing experiences can be traumatizing to users, subsequently doing more harm than good to their psychological state[6].





If further research finds psychedelic microdosing effective at treating psychiatric disorders, it could become a better use of psychedelics than full dosing, as well as an eventual replacement for current psychiatric drugs, like antidepressants, known to have low adherence and efficacy.

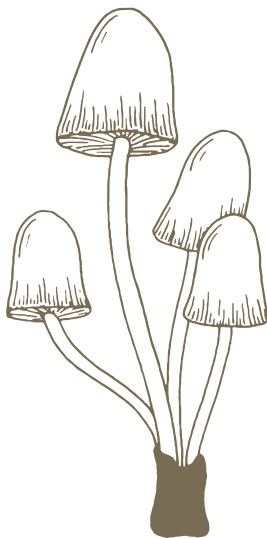
As of yet, research on psychedelic microdosing is in the preliminary stages, with many of the studies being observational and open label. Participants in these studies are commonly recruited from online forums like Reddit, and answer questionnaires about their experiences with psychedelic microdosing. A study by Lea et al. investigated the subjective experience of participants that self-administered psychedelic microdoses[3]. The sample consisted of non-clinical and clinical populations, with the latter being diagnosed with mental illnesses such as depression, anxiety, or alcohol addiction. They found that 44% of participants reported an improvement in their mental health after microdosing. In addition, many participants reportedly discontinued their psychiatric medications, however the percentage of participants was unclear[3]. Similarly designed studies have many participants reporting improvements in their mood and mental health that do not interfere with their daily activities, reductions in dysfunctional attitudes and negative emotions, as well as increased open-mindedness and creativity when compared to controls [1,4-6]. Even self-reports claiming alleviation of pain associated with migraines, pre-menstruation, traumatic brain injury, and shingles, have been collected from smaller samples[5]. Although the wide range of outcomes and therapeutic uses of psychedelic microdosing are generally positive, the observational and open label design of these studies leave many questions regarding the therapeutic potential of psychedelic microdosing unanswered. For instance, placebo effects and residual confounding cannot be ruled out from these observational studies. Around 40% of participants in the study by Lea et al. indicated that their aim was to improve their mental health through psychedelic microdosing[3].

Additionally, Kaertner et al. noted significant positive expectancy scores prior to commencing microdosing regimens, indicating the likely presence of a placebo effect due to participants believing they would experience enhancements in their well-being[1]. The nature of these study designs also indicate the presence of other biases such as confirmation bias and self-selection when recruiting participants[1,3]. Additional limitations include confounding effects from factors such as concomitant reductions in alcohol and other drug use during microdosing, lifestyle changes, and influence from the media or online forums[3].

A review article on fourteen experimentally controlled studies by Kuypers found that microdoses within the range of 10-20 micrograms for LSD and 1-3 milligrams for psilocybin have only slight effects on cognition and mental health[8]. Interestingly, positive outcomes were noted alongside negative changes in mood like enhanced anxiety, psychological distress and cycling between depressive and euphoric emotions[8]. However, findings between observational and experimental studies are not entirely inconsistent. Experimentally controlled trials on psychedelic microdosing support the safe dosing ranges determined by observational studies, as multiple experimental studies report no significant short-term changes to physiological measures like heart rate, blood pressure and basal body temperature[8,9].

With the claim that psychedelic microdosing can improve cognitive function, research has also investigated the potential for microdosing to alter brain function through measures like functional connectivity and blood flow through the brain. Functional connectivity is defined as the amount of interaction and co-activation of separate brain regions[11]. Brain imaging has shown that LSD microdosing can increase functional connectivity between the amygdala, right angular gyrus, right middle frontal gyrus and cerebellum[12]. Specifically, positive changes in mood were correlated with increased connectivity between the amygdala and middle frontal gyrus[12].

Decreased functional connectivity between the amygdala, postcentral gyrus and superior temporal gyrus was also observed[12]. Despite these changes in functional connectivity between brain structures of the limbic system, the effects of psychedelic microdosing on mood were subtle and ultimately considered negligible[12]. The authors noted that future studies should investigate the effects of repeated microdoses, as only a single psychedelic microdose was used in this study. The interest in the potential for psychedelic microdosing to treat Alzheimer's disease (AD) dementia is also growing due to 5HT_{2A} receptors being highly expressed in brain regions susceptible to dementia like the prefrontal cortex and hippocampus[7]. Symptoms of AD dementia include decline in cognition and memory due to decreased 5-HT_{2A} density in these brain regions[7]. It is hypothesized that psychedelic microdosing could reduce or treat AD symptoms by serotonergic agonism, increased neuroplasticity and neurogenesis[7]. In fact, enhanced functional connectivity is thought to be a mechanism by which symptomatic AD can be prevented[11], and fMRI studies have reported increased functional connectivity due to psychedelic microdosing that lasted five weeks in patients diagnosed with depression[7]. Thus, psychedelic microdosing has considerable potential as a novel treatment for AD dementia due to its selective agonism of 5-HT_{2A} receptors and its safer psychological effects relative to full doses of psychedelics.



References

1. Kaertner LS, Steinborn MB, Kettner H, et al. Positive expectations predict improved mental-health outcomes linked to psychedelic microdosing. *Sci Rep.* 2021;11(1): 1-11. doi:10.1038/s41598-021-81446-7
2. Kuypers KPC, Ng L, Erritzoe D, et al. Microdosing psychedelics: More questions than answers? An overview and suggestions for future research. *J Psychopharmacol.* 2019;33(9):1039-1056. doi:10.1177/0269881119857204
3. Lea T, Amada N, Jungaberle H, Shecke H, Scherbaum N, Klein M. Perceived outcomes of psychedelic microdosing as self-managed therapies for mental and substance use disorders. *Psychopharmacology (Berl).* 2020;237(5):1521-1532. doi:10.1007/s00213-020-05477-0
4. Johnstad PG. Powerful substances in tiny amounts. *Nord Stud Alcohol Drugs.* 2018;35(1):39-51. doi:10.1177/1455072517753339
5. Fadiman J, Korb S. Might Microdosing Psychedelics Be Safe and Beneficial? An Initial Exploration. *J Psychoactive Drugs.* 2019;51(2):118-122. doi:10.1080/02791072.2019.1593561
6. Anderson T, Petranker R, Rosenbaum D, et al. Microdosing psychedelics: personality, mental health, and creativity differences in microdosers. *Psychopharmacology (Berl).* 2019;236(2):731-740. doi:10.1007/s00213-018-5106-2
7. Vann Jones SA, O'Kelly A. Psychedelics as a Treatment for Alzheimer's Disease Dementia. *Front Synaptic Neurosci.* 2020;12(34):1-6. doi:10.3389/fnsyn.2020.00034
8. Kuypers KPC. The therapeutic potential of microdosing psychedelics in depression. *Ther Adv Psychopharmacol.* 2020;10:1-25. doi:10.1177/2045125320950567
9. Bershad AK, Schepers ST, Bremmer MP, Lee R, de Wit H. Acute Subjective and Behavioral Effects of Microdoses of Lysergic Acid Diethylamide in Healthy Human Volunteers. *Biol Psychiatry.* 2019;86(10):792-800. doi:10.1016/j.biopsych.2019.05.019
10. Rogers BP, Morgan VL, Newton AT, Gore JC. Assessing Functional Connectivity in the Human Brain by fMRI. *Magn Reson Imaging.* 2007;25(10):1347-1357. doi:10.1016/j.mri.2007.03.007
11. Kim S, Jeon SG, Nam Y, Kim H, Yoo DH, Moon M. Bilingualism for Dementia: Neurological Mechanisms Associated With Functional and Structural Changes in the Brain. *Front Neurosci.* 2019;13:1224. doi:10.3389/fnins.2019.01224
12. Bershad AK, Preller KH, Lee R, et al. Preliminary report on the effects of a low dose of LSD on resting state amygdalar functional connectivity. *Biol Psychiatry Cogn Neurosci Neuroimaging.* 2019;5(4):461-467. doi:10.1016/j.bpsc.2019.12.007

Plant-Based vs. Animal-Based Vaccines

By: Eric Kim

As the COVID-19 pandemic is ongoing, interests and concerns regarding vaccines for COVID-19 are growing as several types of vaccines from different companies are being administered after authorization for emergency use. This article will briefly introduce different sources of vaccines that are being produced as well as their advantages and challenges.

One of the most common methods of producing a vaccine is animal-based, especially egg-based production that has been used for about 70 years[1,2,3]. This method involves injection of viruses into fertilized eggs to allow for replication as the virus requires a host cell to replicate[1,2,3]. The fluid is then extracted, followed by inactivation of the virus and purification of viral antigens, in case of inactivated vaccines[1]. Some examples of vaccines that are produced this way include influenza, measles-mumps-rubella (MMR), rabies, and yellow fever vaccines[4]. This method, despite being widely used, also involves several limitations including sorting of appropriate virus strains to be replicated, difficulty in purification of antigens, and requirement for large numbers of eggs that extends the time for production[1,2,3]. In addition, people sensitive to eggs might develop allergic reactions to egg proteins included in the vaccine, notably ovalbumin[5].

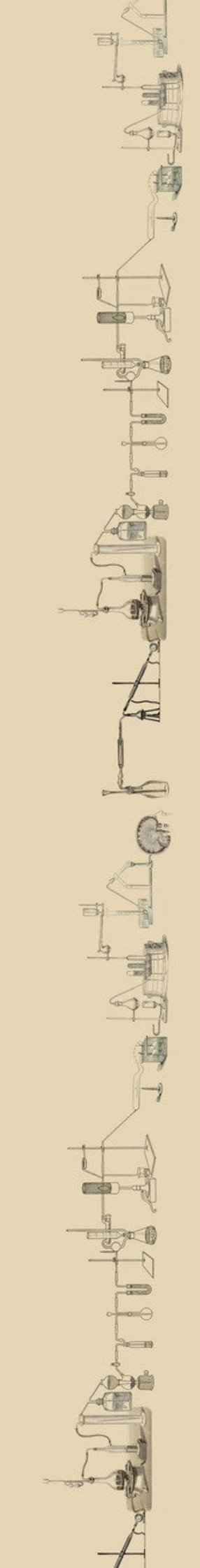
One alternative to egg-based vaccines is cell-based vaccines. In an example of flu vaccine, candidate viruses are made in the laboratory from influenza viruses which are then injected into mammalian cell lines cultured in a stainless-steel bucket[6,7]. The fluid containing the virus can then be extracted from the cells for antigen purification[6,7]. Since this method does not require egg supply, vaccines can be made more rapidly that could be especially helpful during the rapid spread of the virus it protects from[6,7].

In an example of cell-based flu vaccine, the cells for incubating the

vaccine can be kept frozen in a storage cell bank to ensure mass storage[6,7]. Also, there is no concern of egg-adapted changes to the virus when it is injected into eggs that could potentially induce changes in viral structure and production of antibodies that are less focused on the original viral strain that are circulating[6]. Therefore, we can expect higher protection from the infection, as shown by several clinical studies comparing the relative vaccines effectiveness including hospitalizations/ER visits due to influenza, asthma, chronic obstructive pulmonary disease, etc[8,9]. Some of the vaccines in this category include Flucelvax Quad-valent licensed as an inactivated flu vaccine, as well as vaccines for polio, rotavirus, hepatitis, chickenpox, smallpox, and rubella[6,7]. However, this vaccine type also entails some limitations such as the need for fermentation facilities and animal nutrients such as cow-derived amino acids, glycerol, detergents, gelatin, enzymes, and blood, for replication of virus in living cells since viruses use host metabolism for replication and nutrition requirement increases [2,3,10,11].

Instead of egg- or cell-based vaccines, using transgenic plants expressing viral antigens to produce plant-based vaccines is in the spotlight[2,3]. This method is expected to be especially cost-effective as it does not involve cold-chain storage and could have mass production as several candidate plants including tobacco, tomato, and corn are easier to grow[3]. Plant-based vaccine production process involves integration of viral antigen into the vector that will then be transformed into plant cells for expression through either a stable or transient transformation system[3]. In case of stable transformation, the plasmid of *Agrobacterium tumefaciens* strain can be used to accept the viral antigen transgene and form a recombinant vector to achieve permanent transformation by integration of the transgene into the plant genome[3].





Several studies have used this method to express subunit proteins for cholera toxin B or VP60 protein for rabbit hemorrhagic disease virus in potato leaf explants, hepatitis B surface antigens in tomato plants, and so on[3]. However, the plant species that *A. tumefaciens* can naturally infect are limited and the amount of antigen production is generally low, being limitations of stable transformation[3]. Instead, transient transformation where the transformed gene temporally resides in the host cell to be expressed into antigens but not integrated into the plant genome is thought to have higher efficiency[3]. This involves the genetic modification of a recombinant plant virus that infects plant cells, which happens through creation of chimeric genes for viral coat protein[3]. The plant virus can also be modified at capsid proteins which will then infect the plant cells and express viral antigens during viral replication, overall resulting in transient expression of the antigens in plant cells[3]. Several examples of this method include tobacco mosaic virus containing epitopes of murine hepatitis virus that can infect tobacco plants and cowpea mosaic virus that can infect *Vigna unguiculata* to produce VP2 capsid of mink enteritis virus[3]. Overall, some limitations to plant-based vaccines include selection of compatible antigens with the plant, variability in antigen production between different transgenic plants and difficulty of choosing dosage in patients based on level of immune response, age, and weight, and ensuring quality assurance of the vaccine by meeting good manufacturing practices in terms of growing plants and producing the vaccine[3]. Also, plant-based vaccines have not been licensed by FDA for human use due to being classified as genetically modified crop[3].

Recent mRNA vaccines for COVID-19 produced from Pfizer or Moderna do not seem to involve viral replication, but instead synthesis of mRNA in synthesizer followed by mixing with lipid droplets, potentially implying that problems arising with viral replication process in cells in animal- and plant-based vaccines might no longer be an issue once this type of vaccine becomes common[12].

Additionally, a noticeable number of vaccines for COVID-19 are known to incorporate adjuvants such as squalene[13]. Squalene is a type of lipid that enhances immune response to the vaccine by promoting uptake of viral antigen by cell, assisting with immune cell recruitment to the immunization site, and stabilizing the protein or virus in the vaccine[13]. Shark liver is a common source of high-purity squalene, which is used in flu vaccines as well as part of AS03 adjuvant for COVID-19 vaccines produced by GlaxoSmithKline, a pharmaceutical company[13]. However, a growing concern about massive decreases in shark populations followed by the negative impact on the ecosystem has sparked the need for squalene from other sources. For example, a biotechnology company named Amyris has been looking for squalene from non-shark sources, such as refined olive oil distillate and a semisynthetic method that synthesizes squalene from farnesene made from sugar fermentation by microbes[13]. Also, there are other vaccines that do not use squalene for adjuvants such as Pfizer and Moderna that use other lipid nanoparticles for carrying mRNA or Sinovac vaccines that use aluminum hydroxide instead[13].

As for now, there seems to be no absolute answer as to use which source for producing vaccines. However, it does seem clear that recent studies regarding vaccine development are changing focus from animal to plant-based production due to cost, safety, and environmental issues. Thus, it will be more important in the future to conduct more studies to improve effectiveness and solve limitations regarding the plant vaccines, should it serve as an alternative to conventional animal-based vaccines. Also, the vaccine manufacturers will also need to further investigate the potential of recent vaccine formulations, such as mRNA vaccines, to assess the pros and cons for continued development of plant-based vaccines.

References

1. How Influenza (Flu) Vaccines Are Made. Centers for Disease Control and Prevention. <https://www.cdc.gov/flu/prevent/how-fluvaccine-made.htm>. Accessed April 28, 2021.
2. Plant-based Vaccines. Creative Biolabs. <https://www.creative-biolabs.com/vaccine/plant-based-vaccines.htm#:~:text=The%20advantages%20of%20Plant%2Dbased%20Vaccines&text=Economically%20effective%20for%20they%20are,Easy%20to%20expand%20productiop%20scale>. Accessed April 28, 2021.
3. Laere E, Ling APK, Wong YP, Koh RY, Mohd Lila MA, Hussein S. Plant-Based Vaccines: Production and Challenges. *J Bot.* 2016;2016:4928637. doi:10.1155/2016/4928637.
4. Cox JE. Egg-based vaccines. *Pediatr Rev.* 2006;27(3):118-119. doi:10.1542/pir.27-3-118.
5. Flu Vaccine and People With Egg Allergies. Centers for Disease Control and Prevention. <https://www.cdc.gov/flu/prevent/egg-allergies.htm>. Accessed April 29, 2021.
6. Cell-Based Flu Vaccines. Centers for Disease Control and Prevention. <https://www.cdc.gov/flu/prevent/cell-based.htm>. Accessed April 29, 2021.
7. Cell Based Vaccines. Creative Biolabs. <https://www.creative-biolabs.com/vaccine/cell-based-vaccines.htm>. Accessed April 29, 2021.
8. Izurieta HS, Chillarige Y, Kelman J, et al. Relative Effectiveness of Influenza Vaccines Among the United States Elderly, 2018-2019. *J Infect Dis.* 2020;222(2):278-287. doi:10.1093/infdis/jiaa080.
9. Divino V, Krishnarajah G, Pelton SI, et al. A real-world study evaluating the relative vaccine effectiveness of a cell-based quadrivalent influenza vaccine compared to egg-based quadrivalent influenza vaccine in the US during the 2017-18 influenza season. *Vaccine.* 2020;38(40):6334-6343. doi:10.1016/j.vaccine.2020.07.023.
10. Bovine Derived Materials Used in Vaccine Manufacturing Questions and Answers. U.S. Food & Drug Administration. <https://www.fda.gov/vaccines-blood-biologics/questions-about-vaccines/bovine-derived-materials-used-vaccine-manufacturing-questions-and-answers>. Updated March 23, 2018. Accessed April 30, 2021.
11. Liu XP, Huang D, Tan WS, Luo J, Chen Z. Overcoming nutrient limitations for cell-based production of influenza vaccine. *Hum Vaccin Immunother.* 2015;11(7):1685-1688. doi:10.1080/21645515.2015.1044182.
12. Moore A. How are COVID-19 vaccines made? An expert explains. *The Conversation.* <https://theconversation.com/how-are-covid-19-vaccines-made-an-expert-explains-155430>. Published February 18, 2021. Accessed April 30, 2021.
13. Bomgardner MM. On the hunt for alternatives to shark squalene for vaccines. *Chemical & Engineering News.* <https://cen.acs.org/pharmaceuticals/vaccines/hunt-alternatives-shark-squalene-vaccines/98/i47>. Published December 6, 2020. Updated December 8, 2020. Accessed April 30, 2021.



The Consequential Diseases and Physiological Effects Associated with Vaping

By: Manu Kattimani

Introduction:

When the use of vape first started to merge with mainstream culture in 2003, it was heralded as a healthy alternative compared to smoking and an excellent mechanism for nicotine addiction cessation[1]. Similar to a nebulizer, which is used by asthma patients or patients who suffer from other lung conditions, vape devices turn liquid aerosol into a mist which the consumer can breathe into their lungs[2]. These devices became popular very quickly, particularly among young people, when compared to cigarettes in a historical context[3]. Medical professionals and regulatory bodies have been issuing warnings surrounding the health risks with vaping, particularly for the younger generation mainly due to the lack of information with the long term physiological effects and health risks associated with vaping and the emergence of disparate lung injuries[1]. This article will look to synthesize the information provided by various scientific literature inspecting physiological responses and the accompanying diseases associated with vaping.

Physiological Effects of Vape Ingredients:

Propylene glycol and glycerol are associated solvents that are used in conjugation with nicotine in vape liquid. Exposure to propylene glycol and glycerol in the form of vapor affects the expression of pulmonary circadian molecular clock genes including *arntl*, *npas2*, *nr1d1*, *nr1d2*, *per1*, *per2* and *per3*[4]. The altered expression of these genes occurred without a change to lung histology, numbers of mononuclear or neutrophil cells or bronchoalveolar lavage total cells and in the absence of an inflammatory response[4]. These circadian molecular clock genes have an effect on the brain, liver, kidney and skeletal muscle leading to a widespread systematic impact throughout the body[4].

The alteration in circadian rhythms throughout the body can lead to biological alterations and can amplify the rate of intensity for various pathologies[4]. Disrupted circadian rhythms in the lungs can lead to modulation in amplitude of response to endogenous cortisol, which in turn leads to an amplified response to bacterial agents[4]. In the digestive tract, mice which were deficient of molecular clock genes displayed weight gain and contained an abnormal amount of cholesterol and fat within the bloodstream[4]. Mice which contained deleted molecular clock genes displayed impaired glucose tolerance and decreased insulin secretion which often lead to the onset of diabetes mellitus[5].

The exposure to propylene glycol and glycerol also affected the expression of *hspal* and *hspalb* genes, which are rhythmic heat shock proteins. Mice exposed to the solvents displayed downregulation of genes within the liver, kidney, skeletal muscle and lungs[6]. Heat shock proteins play several roles within the immune system ranging from antigen presentation to tumor immunosurveillance and autoimmunity[6]. Alterations within the expression of heat shock proteins can lead to lung damage. *Hsp27*, *Hsp32*, *Hsp60* and *Hsp70* demonstrate an important role in cytoprotection during the phase of lung inflammation and injury[7]. *Hsp27* inhibits apoptosis after phases of oxidative stress, improves mortality after endotoxic shock and modulates cytoskeletal arrangement of actin filaments in lung epithelial cells upon phosphorylation[7]. The upregulation of *Hsp27* through phosphorylation by MAPKAPK-2 is correlated to lung exposure to endotoxins and correlated lung injuries. *Hsp32* or heme oxygenase is found in 3 forms (-1, -2, -3); it plays an important role in cytoprotective properties for fibroblasts, fetal lung cells, respiratory epithelial cells and coronary endothelial cells against oxygen toxicity and heme and hemoglobin toxicity, respectively[7].

Cytosolic Hsp60 stabilizes actin and tubulin and is expressed during cell stress providing stress tolerance in the lung[7]. Hsp70 is expressed with lung and specific lung cells in response to various types of stresses such as LPS-mediated apoptosis, thermal stress and hyperoxia[7]. Nicotine affects the lung by increasing inflammation and destroying cilia through heating of the lungs; propylene glycol and glycerol may exacerbate these effects through downregulation of heat shock proteins, which may be the reason why users of vape display worse reactions to endotoxins such as E. coli, Salmonella and V. cholerae[5].


Propylene glycol and glycerol may drive inflammation in the GI tract specifically in the colon while reducing the expression of genes that play a role in barrier function[8]. Occluding junctions, zonula occludens and claudin-2 which serve as markers of gut epithelial tight junctions have significantly reduced gene expression in mice that were exposed to nicotine-free aerosols[8]. The effects of associated solvents may be specific for the gut since they affect the tight junctions of the gut but do not affect the tight junctions of the skin. Inflammation is triggered in the gut once the mice were exposed to constant exposure to vape aerosols rather than acute exposure to aerosols. Associated solvents within vape disturb the integrity of the gut barrier since paracellular permeability is increased upon exposure to associated solvents while trans-epithelial electrical resistance is lowered[8]. Constant exposure to associated solvents causes inflammation in the colon through the presence of MCP1, IL-8 and TNF- α as pro-inflammatory cytokines such as il31ra, il1r2m, ccl8, ear2[8]. Ilk which participate in cytokine signaling are upregulated whereas trim30a which participates in downregulation of inflammatory cytokines are down regulated[8]. These various effects co-interact to compromise the health of the body broadly leaving it more susceptible to infections and inflammation particularly in the gut epithelium and the colon.

Diseases Observed in Vape Users:

Bronchiolitis obliterans, otherwise more commonly known as popcorn lung, has been observed in the youth demographic of vape users[9,10]. Popcorn lung describes the reduction in bronchioles within the lungs airway due to scarring tissue formed through lung damage. A patient study shows a healthy 17 year old with no prior health conditions and no nicotine cigarette usage had developed “popcorn lung” after daily use of vaping for 5 months. Many other cases such as this have been observed in North American youth with research linking bronchiolitis to vaping[9]. Primary spontaneous pneumothorax, otherwise known as a collapsed lung, has been observed in tall, thin individuals who are undergoing a period of growth during adolescence[10]. Weak points in air blisters on the top of the lungs develop due to the accelerated growth with smoking and vaping presenting an increased risk of bursting the blisters which leads to the collapse of the lungs[11]. There has also been an increase in product use-associated lung injuries linked to the detection of vitamin E acetate present in bronchoalveolar-lavage fluid originating from vape ingredients[10]. Vitamin E acetate when it is heated produces ketene which acts as an irritant to airways and propagates an inflammatory cascade[10,11]. It reduces surfactant function leading to atelectasis by collapsing alveoli. This can lead to damage within the lung due to collapsing alveoli, inflammation, and upregulated immune system[10,11].

Different variations of pneumonia have been observed in vape users. Hypersensitivity pneumonitis and the association with vape users was noted due to the cessation of immune response to organic mold and dust after stopping the use of vape within two patients[12]. Hypersensitivity pneumonitis occurs as an immune system disorder where the lungs are inflamed reacting to inhaled dust, microorganisms, plant and dirt type materials in an anaphylactic mechanism within the lung[12]. It is caused by the repeated exposure and sensitization to antigens which leads to inflammation[12,13].





Common antigens which are present in vape liquid are formaldehyde that is in the form of propylene glycol and terpene and benzene that is in the form of butane hash oil[13]. Lipoid pneumonia occurs as an inflammatory response due to exogenous or endogenous lipids which have been inhaled or aspirated[10,12]. These lipids can be found in laxatives, petroleum-based lubricants, mineral oils, or hydrocarbons. This association was made after observing 3 patients with the presence of lipid-laden macrophages in lavage fluid which can be attributed to lipid materials which are present in the flavoring agents[12]. Though there are no explicit oils present in vaping solution, it is possible that glycerin or other associated solvent can produce lipoid pneumonia through endogenous phospholipids. Heavy metals such as nickel, lead, tin and cadmium are found in many common vaping solutions[2,11]. Giant cell interstitial pneumonia is caused by the heavy metals interacting with interstitial tissues within the lungs leading to inflammation and fibrosis[12]. Organizing pneumonia is categorized as interstitial pneumonia caused by lung inflammation and scarring that leads to obstruction of small airways and alveoli within the lungs[11,12]. The exact drug reaction or inhalation exposure from vaping juice which causes organizing pneumonia is unknown. These various cases of pneumonia and the increased incidence of product associated lung injuries places a new focus on the pathophysiology and health effects of vaping.

Gastrointestinal disorders can be driven by the inflammation that occurs after the exposure to associated solvents within vaping solutions. Especially within the youth demographic, gastrointestinal irritation is just as prevalent as any respiratory based disorder or injury[8]. Vaping can have negative changes on the microbiome which is present within the gut[8]. As mentioned in the physiologic effects of vaping ingredients section, chronic gut inflammation can lead to inflammatory bowel disease including ulcerative colitis that encompasses inflammation and ulcers that are in the lining of the colon and rectum[8,14].

Chronic gut inflammation is also the primary risk factor for the development of cancer within the gastrointestinal system. Cancers that can result from inflammation within the gastrointestinal tract includes colon cancer, intestinal lymphoma, anal cancer, small bowel adenocarcinoma and cholangiocarcinoma[14].

Conclusion:

Vaping has various physiological effects on the body that are not observed with smokers due to associated solvents of vaping solutions, mainly propylene glycol and glycerol. This includes alteration in expression of pulmonary circadian molecular clock genes, down regulation in heat shock protein expression and inflammation in the GI tract. Bronchiolitis obliterans, primary spontaneous pneumothorax and product associated lung injuries from vitamin E acetate can injure the lungs of vape users. Different variations of pneumonia including hypersensitivity pneumonitis, lipoid pneumonia, giant cell interstitial pneumonia and organizing pneumonia is observed in chronic vape users due to the various toxic chemicals present in vaping solutions. Medical professionals should continue to raise urgent concerns with the use of vape in the youth and adult demographic due to the unknown short term and long term effects being discovered within vape users.



References

1. Outbreak of Lung Injury Associated with the Use of E-Cigarette, or Vaping, Products. Centers for Disease Control and Prevention. https://www.cdc.gov/tobacco/basic_information/e-cigarettes/severe-lung-disease.html. Updated November 27, 2020. Accessed April 28, 2021.
2. The Impact of E-Cigarettes on the Lung. American Lung Association. <https://www.lung.org/quit-smoking/e-cigarettes-vaping/impact-of-e-cigarettes-on-lung>. Updated July 13, 2020. Accessed April 28, 2021.
3. Vaping-Associated Lung Illness. Public Health Agency of Canada. <https://www.canada.ca/en/public-health/services/diseases/vaping-pulmonary-illness.html>. Updated August 20, 2020. Accessed April 28, 2021.
4. Lechasseur A, Jubinville E, Routhier J, et al. Exposure to electronic cigarette vapors affects pulmonary and systemic expression of circadian molecular clock genes. *Physiological Reports*. 2017;5(19):e14330. doi:10.14814/phy2.13440
5. Madison MC, Landers CT, Gu B, et al. Electronic cigarettes disrupt lung lipid homeostasis and innate immunity independent of nicotine. *Journal of Clinical Investigation*. 2019;129(10): 4290–4304. doi:10.1172/jci128531
6. Binder RJ. Functions of Heat Shock Proteins in Pathways of the Innate and Adaptive Immune System. *The Journal of Immunology*. 2014;193(12): 5765–5771. doi:10.4049/jimmunol.1401417
7. Wheeler DS, Wong HR. Heat shock response and acute lung injury. *Free Radical Biology and Medicine*. 2007;42(1):1–14. doi:10.1016/j.freeradbiomed.2006.08.028
8. Sharma A, Lee J, Fonseca AG, et al. E-cigarettes compromise the gut barrier and trigger inflammation. *iScience*. 2021;24(2):102035. doi:10.1016/j.isci.2021.102035
9. Landman ST, Dhaliwal I, Mackenzie CA, Martinu T, Steele A, Bosma KJ. Life-threatening bronchiolitis related to electronic cigarette use in a Canadian youth. *Canadian Medical Association Journal*. 2019; 191(48). doi:10.1503/cmaj.191402
10. Broderick ST. What Does Vaping Do to Your Lungs? Johns Hopkins Medicine. <https://www.hopkinsmedicine.org/health/wellness-and-prevention/what-does-vaping-do-to-your-lungs>. Accessed April 28, 2021.
11. Cherian SV, Kumar A, Estrada-Y-Martin RM. E-Cigarette or Vaping Product-Associated Lung Injury: A Review. *The American Journal of Medicine*. 2020;133(6):657–663. doi:10.1016/j.amjmed.2020.02.004
12. Henry TS, Kligerman SJ, Raptis CA, Mann H, Sechrist JW, Kanne JP. Imaging Findings of Vaping-Associated Lung Injury. *American Journal of Roentgenology*. 2020;214(3):498–505. doi:10.2214/AJR.19.22251
13. Medina-Pena A, Shiu D, Banks Paulino A. Vaping into acute hypoxic respiratory failure and hypersensitivity pneumonitis: A Case Report. *Chest* 2020;158(4). doi:10.1016/j.chest.2020.08.2006
14. Axelrad JE, Lichtiger S, Yajnik V. Inflammatory bowel disease and cancer: The role of inflammation, immunosuppression, and cancer treatment. *World Journal of Gastroenterology*. 2016;22(20):4794. doi:10.3748/wjg.v22.i20.4794



Financial Incentives in Clinical Trials Come at a Price

By: Carmen Chan

Financial Incentives

Clinical trials are conducted to assess the safety and efficacy of novel therapeutics in human subjects. Despite their importance in bringing the drug to market, participant enrolment has proven to be difficult. As reviewed in Hamel et al., approximately 22% to 50% of oncology trials in the United States (US) failed to achieve scientific objectives due to insufficient enrolment rates, resulting in wasted research efforts and a lengthened drug development process[1].

As a partial solution, incentives have been employed to optimize participant enrolment. In particular, financial incentives serve as compensation for the participant's time and travel (e.g., bus fare, parking passes), in addition to being a token of appreciation for their contribution to biomedical research[2,3]. The amount varies depending on multiple factors, such as the type of research, the time spent, and the procedures involved[2]. To illustrate, financial incentives likely play a more substantial role in optimizing enrolment for Phase I clinical trials, involving healthy participants, as there are no expected medical benefits but rather personal benefits (e.g. financial)[2,4]. In contrast, financial incentives likely play a smaller role in Phase III clinical trials, involving the intended patient population, as medical benefits are expected from the interventions employed[2,4]. In addition to this, a discrete choice experiment involving hypothetical clinical trials of varying risk and benefits in university students by Vellinga et al[3]. demonstrated that incentives of higher value and provided in cash were motivators for participation relative to no value and gift cards or vouchers. Together, this supports the notion that financial incentives serve as a motivator for trial participation, and therefore, improve enrolment rates.

Ethical Concerns

In spite of this, the use of financial incentives raises various ethical concerns, including financial inducement and biased enrolment[2,3,5]. According to the Nuremberg Code, experiments involving human participants must ensure voluntary consent is obtained[6]. In order for consent to be voluntary, it must be obtained in the absence of external influences, such that the individual can freely choose or refuse to participate in the absence of inducement or coercion. Considering this, financial incentives in clinical trials can be defined as a form of financial inducement because an individual's decision to participate may become compromised by the offer of payment[2,5]. In particular, it may encourage concealing information that does not meet clinical trial eligibility criteria and clouding of the capacity to properly assess the risks associated with participation[2,5]. In fact, a study by Bentley and Thacker found that financial incentives had a positive impact on students' willingness to participate in hypothetical clinical trials, regardless of the risk level associated with the trial, supporting the notion that financial incentives do indeed improve enrolment rates (*intention* to enrol, in this case) but may compromise judgment to the risks associated with participation[5]. As such, it has been suggested that the amount of money should not be so large that it compromises the individual's judgement to freely make a decision about their participation[2].

However, inducement does not solely stem from the value of the financial incentive, but also the individual's demographic information[5]. In particular, the use of financial incentives may disproportionately target individuals with lower socioeconomic status (SES), who may be more susceptible to financial inducement, resulting in biased enrolment[2,7].

This may be especially evident in Phase I clinical trials due to the nature of the motivations to participate, as previously discussed. Despite this sound reasoning, the literature suggests otherwise. A study by Unger et al[8]. found that individuals with lower income were less likely to participate in oncology trials. Consistent with this finding, El-Rayes et al. found that participation in pancreatic cancer Phase II clinical trials was associated with higher SES[7]. Together, these findings suggest that SES may be a barrier to trial participation, potentially due to increased susceptibility to indirect costs (e.g. lost wages, transportation), limited access to healthcare, increased comorbidities, and inability to meet stringent eligibility criteria as a result[7,8]. Historically, clinical trials generally report characteristics relevant to the scientific objectives, and rarely report demographic information, such as SES[2,9]. As SES is strongly associated with health and disease prognosis[2,9], it is important to improve reporting of demographic information to form a better understanding of the relationship between SES and trial participation, as well as the impact of SES on susceptibility to financial inducement in trial participation.

Nevertheless, it is important to note that academic research on this topic, which is already sparse, has largely focused on later phase clinical trials (i.e. Phase II and III)[4], where motivations to participate include medical benefit and may be viewed as more altruistic in nature. As such, although data suggests that lower SES acts as a barrier to participation, it is inappropriate to generalize this to Phase I clinical trials, where financial incentives are the main benefit to participants. Considering this, it is still reasonable to think that the use of financial incentives in Phase I clinical trials bias enrolment toward individuals with lower SES through increased susceptibility to financial inducement and the nature of the motivations to participation.

A clear example of the importance of this distinction can be observed in trial participation of racial and ethnic minorities.

Despite the calls for increased trial diversity in general, a review by Fisher and Kalbaugh[4] suggested that racial and ethnic minorities are over-represented in Phase I clinical trials, but under-represented in Phase III clinical trials despite higher cancer burden in the US[1]. As racial and ethnic inequalities are associated with lower SES[10], this may indicate that minorities are more susceptible to financial inducement, assuming greater risk (i.e. Phase I clinical trials), while receiving fewer benefits (e.g. medical benefits from Phase III clinical trials)[4]. Altogether, these findings indicate that SES and susceptibility to financial inducement is highly dependent on the type of clinical trial. As such, future studies should not only improve the reporting of SES and other demographic information, but also consider the differences between Phase I and later phase clinical trials when developing enrolment strategies. Gaining an understanding of these factors will prevent biased enrolment, improving the generalizability of results, and improve equal access to novel therapeutics[2,4].

Concluding Remarks

Although widespread use of financial incentives as a strategy to optimize participant enrolment in clinical trials seem promising in theory, the ethical concerns discussed suggest that employing this strategy will come at a price, literally and figuratively. As such, a more dimensional approach toward incentivizing trial participation should be adopted. The use of financial incentives must strike a balance: providing appropriate compensation for the time, travel and risks involved in participation, while avoiding undue inducement and biased enrolment. Additionally, various factors such as the type of clinical trial (e.g. Phase I vs. Phase III) and demographic factors (e.g. SES) should also be considered. Gaining a better understanding of how these factors play a role in susceptibility to financial inducement will not only aid in the development of better enrolment strategies, but also improve the generalizability of research and improve access to novel therapeutics in underrepresented populations.





References

1. Hamel LM, Penner LA, Albrecht TL, Heath E, Gwede CK, Eggly S. Barriers to Clinical Trial Enrollment in Racial and Ethnic Minority Patients with Cancer. *Cancer Control*. 2016;23(4):327-337. doi:10.1177/107327481602300404
2. Resnik DB. Bioethical Issues in Providing Financial Incentives to Research Participants. *Medicoleg Bioeth*. 2015;5:35-41. doi:10.2147/MB.S70416
3. Vellinga A, Devine C, Ho MY, et al. What do patients value as incentives for participation in clinical trials? A pilot discrete choice experiment. *Res Ethics*. 2020;16(1-2):1-12. doi:10.1177/1747016119898669
4. Fisher JA, Kalbaugh CA. Challenging assumptions about minority participation in US clinical research. *Am J Public Health*. 2011;101(12):2217-2222. doi:10.2105/AJPH.2011.300279
5. Bentley JP, Thacker PG. The influence of risk and monetary payment on the research participation decision making process. *J Med Ethics*. 2004;30:293-298. doi:10.1136/jme.2002.001594
6. Moreno JD, Schmidt U, Joffe S. The Nuremberg Code 70 Years Later. *JAMA*. 2017;318(9):795. doi:10.1001/jama.2017.10265
7. El-Rayes BF, Jasti P, Severson RK, et al. Impact of Race, Age, and Socioeconomic Status on Participation in Pancreatic Cancer Clinical Trials. *Pancreas*. 2010;39(7):967-971. doi:10.1097/MPA.0b013e3181da91dd
8. Unger JM, Hershman DL, Albain KS, et al. Patient Income Level and Cancer Clinical Trial Participation. *J Clin Oncol*. 2013;31(5):536-542. doi:10.1200/JCO.2012.45.4553
9. Lee SJ, Kavanaugh A. A need for greater reporting of socioeconomic status and race in clinical trials. *Ann Rheum Dis*. 2004;63(12):1700-1701. doi:10.1136/ard.2003.019588
10. Ethnic and Racial Minorities & Socioeconomic Status. American Psychological Association. <https://www.apa.org/pi/ses/resources/publications/minorities>. Published July 2017. Accessed April 28, 2021.

May Light Shed on Traditional Medicines

By: Joan Zheng

Today, there is no doubt that the use of health supplements sold on isles or any other forms of traditional/alternative medicines should be paid adequate attention and careful analysis when patients are prescribed drugs at professional clinics. However, it is tragic that even though massive unfortunate cases have occurred, commercialization dominates modern society and merchants never further clarify instructions and cautions on the package. On the other side, customers seldom pay attention to those small fonts concealed in the back of colorful patterns which address the hopeful outcomes of taking such supplements or medications. Aside from the discussion of human enzymes with respect to pharmacokinetics, one example I would like to bring up is the great variety of treatments for irregular menstruation periods. In my personal experience, people in Eastern Asia believe brown sugar, red dates, glue derived from donkeys' skin and some other herbs are the ultimate measure to improve the issue. Nonetheless, those with some intermediate knowledge on nutrition, chemistry and human biology will deny this opinion.

Would you not wonder what on earth was there when seeing patches of dry herbs rolling between bubbles in a pot, turning the colourless water into dark brown? Another thing regarding alternative medications is that patients who take those are usually also prescribed with modern drugs, and hence it is especially hard to determine the real originations of the beneficial or negative effects, and with the fact that alternative medications are often suggested as daily supplements to be taken over a long-term, the influence on the human body could be significant concerning the metabolism and absorption of other drugs while many patients do not report them as any medication history. The rates of revisits and emergency hospitalization due to St. John's Wort taken as an over-the-counter antidepressant could well demonstrate the significance of the problem[1].

Notwithstanding, anything is double-sided. Learning about antibiotics are mostly derived from microbial species and other medical compounds, which are important in clinical use with recognizable efficacies, reminds us that we should further explore the field with an intention of promoting modern medicines while advocating revisions of traditional medicines through the scope of modern medical sciences[2]. Primarily, we have little knowledge of traditional medicines today[3]. In general, professionals in traditional medicines advocate the minimal side effects of the medications, yet we should consider the efficacy and try to decipher the "recipe"[4]. Specifically, some side effects could be detrimental. Alternative medicines including Co-enzyme Q10 supplements, garlic and Xin Boa are commonly used for achieving antiarrhythmic effects, and patients usually consume in a daily manner. In consequence, many people do not inform physicians of modern medicines about the intake of those drugs: they do not regard them as drugs[4,5]. Nonetheless, long-term consumption may generate unintended effects when new drugs are prescribed by physicians[4]. In addition, since herbs are often unpurified, contamination has been a serious concern; some minor components may negatively affect patients' health as a whole[2]. Related to toxicology, research has proved the possibility of *Ma Huang*, a traditional Chinese herb relieving cardiovascular disorders, to cause cardiac infarction and sudden deaths[3].

At this point, we could notice the significance of misuse of traditional medications from the perspective of health. However, the effects of the lack of regulations on traditional medications could be equally concerning from the perspectives of economics and bioethics. As to the classification of traditional medication, what has been discussed above is "herbal medications"[6]. One popular herbal product in international markets is Ginseng, of which the pricing procedure could be critically varied based on the species and year of growth[7].





Specifically, as long as some foods and drinks are labeled “containing Ginseng”, the price could be much higher than “normal” ones since they are seemingly healthy, whereas the Ginseng component is minimal and the actual beneficial influence for consumers is hard to measure[8]. During the past decades, many companies may have generated unjust profits based on the missing clarification[9]. Other than herbal medications, folk medications which evolve from historical experience are also a major group of all traditional medications[10]. While those sedentary-style physical practices, such as meditation and yoga, may help with patients’ issues as a whole through a mental aspect, folk traditions involving animal products urgently require scientific elucidations[10]. Shark fin consumption could be a spectacular illustration for this topic. For many years, people exploited shark populations for the fins, and sharks were usually released back to the sea after their fins were cut[11]. The number of sharks worldwide has drastically decreased, and eventually scientists revealed the truth: shark fin has no distinct value and may even harm humans due to the high metal concentration resulting from bioamplification[12]. Folk medications, in fact, have placed many animal species in an endangered situation so far, and modern medical science would be the best media for persuading the public with suggestive substitutions[13].

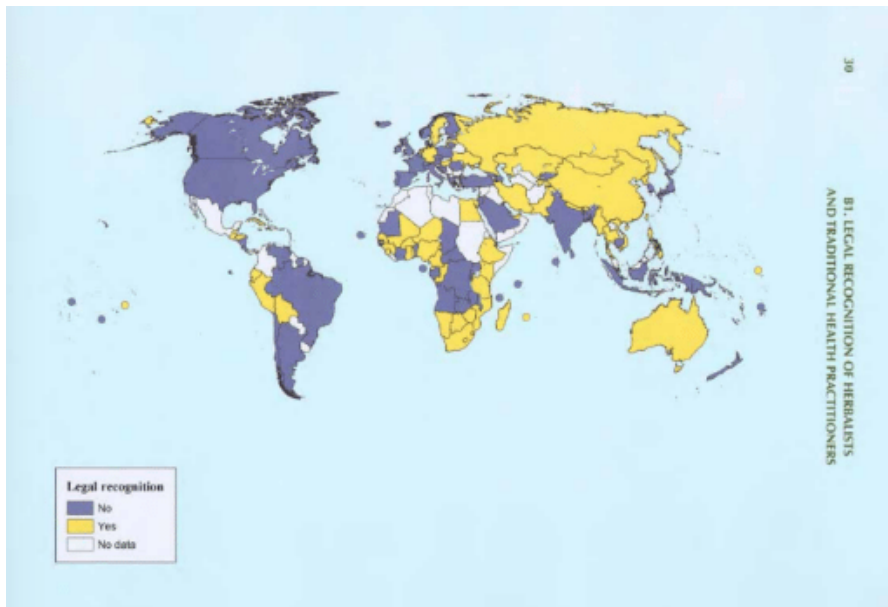
Coming back to the biggest topic of the year 2020: the coronavirus. Professionals in the field of pharmacology and medicines assumed that the likelihood of inventing new drugs in response to an emergent health crisis would be small, and such an assumption led scientists to step back, looking into traditional Chinese medicines with hopes[14]. Fortunately, a Chinese patent medicine called “Lianhua Qingwen capsule/granule” was used as a complementary treatment of Covid-19 shortly after the outbreak as it was regarded by the public as a common anti-inflammatory OTC drug[15]. Due to the observed benefits of the application, research done on the influence in clinical trials has proved a positive relationship

between the use of this medicine and the relief of symptoms, with an approximately 10% higher relieving rate in the treated group than the compared group[15]. Additionally, the influence imposed by Lianhua Qingwen was studied in vitro, and the experimental results proved that the medication could effectively generate morphological alternations of the virus, leading the virus to die[16]. The most optimal part comes from the variety of components of the drug which enabled simultaneous ameliorations of multiple symptoms; two key components *Lonicera japonica* and *Forsythia suspense* could block enzymes for the angiotensin conversion[17], while *pogostemon cablin*[18] and *Rodiola rosea*[19] could relieve GI conditions such as diarrhea and lung damage respectively. It is cheerful that scientists could determine the exact effective elements, and this case appeals to future research in traditional medicines, enhancing human health from a diversified, macroscopic view.

As Yoyo Tu said, “Artemisinin is a gift from nature to human beings”[20]. It is estimated that approximately 25% of modern medications evolve from traditional medications, which mostly consist of plants and animals[21]. We might consider incorporating traditional medicines into pharmaceutical innovation for its moderate effects and reduced interference of the host[2]. While many scientists focus on anti-cancer treatments, gynecological illnesses have received insufficient attention so far[22]. Current modern medications for treating bacterial and fungal vaginitis could significantly harm the native microbiota residing in the vaginal environment, leading to frequent relapse of the condition and diminished the protective mechanism belonging to the host[23]. Considering the moderate influence, we may incorporate traditional medicines to vaginitis treatments through both internal and external uses as a daily health care practice[2]. Advil is one of the most popular OTC drugs since its major component ibuprofen could ameliorate various conditions systematically while its effective component ibuprofen is a NSAID reversibly binding to both COX1 and COX2 and the dose depends on patients’ conditions[24].

In consequence, the lack of specificity could delay the recovery and generate adverse effects[24]. Some traditional medications may not be so effective in relieving discomforts, yet the specificity of some traditional drugs may be beneficial[3]. Furthermore, traditional herbal medications often involve specific methods for drug preparation, such as drying, powdering, boiling, and so on[25]. To fully understand the mechanism of certain drugs, we should examine the procedures carefully with modern scientific equipment and methods[25]. For instance, we should question if toxic substances are depleted during the boiling process? Any conversion reactions occur during the drying process?

Overall, future research is essential for launching regulations to exemplify misuse of traditional medicines while the potential in the field could be optimal. As to the quote of the English poet William Coawper employed at the beginning of the WHO final report on traditional medicines, “knowledge is proud that she knows so much; wisdom is humble that she knows no more”[26], for promoting human health, we pharmacology students are responsible for discovering more gifts from nature and historical intelligence. More excitement could be made to inspire the next generation, and medical sciences and civilisation will complement each other.



WHO global atlas of traditional, complementary and alternative medicines (2005)

While herbal medications could have significant threats on patients' health, many countries still lack of laws/regulations herb prescriptions.

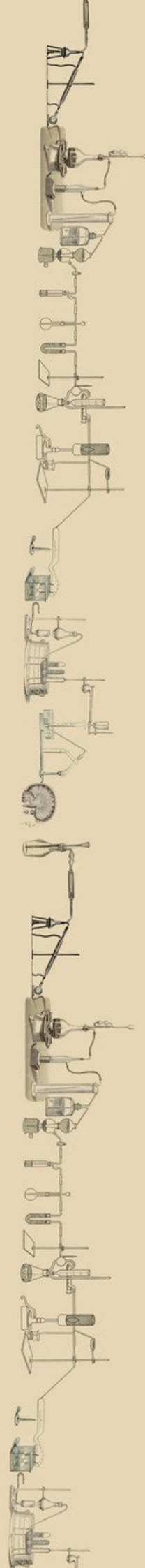




References

1. Nicolussi S, Drewe J, Butterweck V, & Meyer Zu Schwabedissen HE. Clinical relevance of St. John's wort drug interactions revisited. *British Journal of Pharmacology*. 2020;177(6):1212–1226. doi:10.1111/bph.14936
2. Lemonnier N, Zhou GB, Prasher B, et al. Traditional Knowledge-based Medicine: A Review of History, Principles, and Relevance in the Present Context of P4 Systems Medicine. *Progress in Preventive Medicine*. 2017;2(7): e0011. doi:10.1097/pp9.0000000000000011
3. Pan, SY, Litscher G, Gao SH, et al. Historical Perspective of Traditional Indigenous Medical Practices: The Current Renaissance and Conservation of Herbal Resources. *Evidence-Based Complementary and Alternative Medicine*. 2014;2014(525340):1–20. doi:10.1155/2014/525340
4. Pitkälä KH, Suominen MH, Bell JS, Strandberg TE. Herbal medications and other dietary supplements. A clinical review for physicians caring for older people. *Annals of medicine*. 2016;48(8):586–602. doi:10.1080/07853890.2016.1197414
5. Eisenberg DM, Kessler RC, Foster C, Norlock FE, Calkins DR, Delbanco TL. Unconventional Medicine in the United States -- Prevalence, Costs, and Patterns of Use. *New England Journal of Medicine*. 1993;328(4):246-252. doi:10.1056/nejm199301283280406
6. Alostad AH, Steinke DT, Schafheutle EI. Herbal Medicine Classification: Policy Recommendations. *Frontiers in Medicine*. 2020;7:31. doi:10.3389/fmed.2020.00031
7. Baeg IH, So SH. The world ginseng market and the ginseng (Korea). *Journal of Ginseng Research*. 2013;37(1):1-7. doi:10.5142/jgr.2013.37.1
8. Harkey MR, Henderson GL, Gershwin ME, Stern JS, Hackman RM. Variability in commercial ginseng products: an analysis of 25 preparations. *The American Journal of Clinical Nutrition*. 2001;73(6):1101–1106. doi:10.1093/ajcn/73.6.1101
9. Jia L, Zhao Y. Current Evaluation of the Millennium Phytomedicine- Ginseng (I): Etymology, Pharmacognosy, Phytochemistry, Market and Regulations. *Current Medicinal Chemistry*. 2009;16(19):2475-2484. doi:10.2174/092986709788682146
10. WHO global report on traditional and complementary medicine 2019. World Health Organization. <https://www.who.int/traditional-complementary-integrative-medicine/WhoGlobalReportOnTraditionalAndComplementaryMedicine2019.pdf>. Published 2019.
11. Martins APB, Feitosa LM, Lessa RP, et al. Analysis of the supply chain and conservation status of sharks (Elasmobranchii: Superorder Selachimorpha) based on fisher knowledge. *Plos One*. 2018;13(3). doi:10.1371/journal.pone.0193969
12. Ferretti F, Jacoby DMP, Pflieger MO, et al. Shark fin trade bans and sustainable shark fisheries. *Conservation Letters*. 2020;13(3). doi:10.1111/conl.12708
13. Vats R, Thomas S. A study on use of animals as traditional medicine by Sukuma Tribe of Busega District in North-western Tanzania. *Journal of ethnobiology and ethnomedicine*. 2015;11(38):1-11. doi:10.1186/s13002-015-0001-y

14. Xiong X, Wang P, Su K, Cho WC, Xing Y. Chinese herbal medicine for coronavirus disease 2019: A systematic review and meta-analysis. *Pharmacol Res.* 2020;160(105056):1-17. doi:10.1016/j.phrs.2020.105056
15. Hu K, Guan WJ, Bi Y, et al. Efficacy and safety of Lianhuaqingwen capsules, a repurposed Chinese herb, in patients with coronavirus disease 2019: A multicenter, prospective, randomized controlled trial. *Phytomedicine.* 2020;85(153242):1-9. doi:10.1016/j.phymed.2020.153242
16. Runfeng L, Yunlong H, Jicheng H, et al. Lianhuaqingwen exerts anti-viral and anti-inflammatory activity against novel coronavirus (SARS-CoV-2). *Pharmacological Research.* 2020;156(104761):1-6. doi:10.1016/j.phrs.2020.104761
17. Gu S, Yin N, Pei J, Lai L. Understanding traditional Chinese medicine anti-inflammatory herbal formulae by simulating their regulatory functions in the human arachidonic acid metabolic network. *Molecular BioSystems.* 2013;9(7):1931-1938. doi:10.1039/c3mb25605g
18. Zhou TR, Huang JJ, Huang ZT, Cao HY, Tan B. Inhibitory effects of patchouli alcohol on stress-induced diarrhea-predominant irritable bowel syndrome. *World Journal of Gastroenterology.* 2018;24(6):693-705. doi:10.3748/wjg.v24.i6.693
19. Li L, Yang Y, Ma X, Wei Y, Qin Y. 0004 Salidroside Ameliorates Chronic Intermittent Hypoxia-induced Endothelial Insulin Resistance via Suppression of ERK1/2 Activation. *Sleep.* 2018;41 Suppl 1:A120. doi:10.1093/sleep/zsy061.003
20. Tu Y. Artemisinin-A Gift from Traditional Chinese Medicine to the World (Nobel Lecture). *Angewandte Chemie International Edition.* 2016;55(35):10210–10226. doi:10.1002/anie.201601967
21. Zhang J, Onakpoya IJ, Posadzki P, Eddouks M. The Safety of Herbal Medicine: From Prejudice to Evidence. *Evidence-Based Complementary and Alternative Medicine.* 2015;2015(316706):1-3. doi:10.1155/2015/316706
22. Billock J. Pain bias: The health inequality rarely discussed. *BBC Future.* <https://www.bbc.com/future/article/20180518-the-inequality-in-how-women-are-treated-for-pain>. Published May 22, 2018.
23. Menard JP. Antibacterial treatment of bacterial vaginosis: current and emerging therapies. *International Journal of Women's Health.* 2011;3:295-305.. doi:10.2147/ijwh.s23814
24. Rainsford KD. Ibuprofen: pharmacology, efficacy and safety. *Inflammopharmacology.* 2009;17(6):275-342. doi:10.1007/s10787-009-0016-x
25. Zhao Z, Liang Z, Chan K, et al. A Unique Issue in the Standardization of Chinese Materia Medica: Processing. *Planta Medica.* 2010;76(17):1975–1986. doi:10.1055/s-0030-1250522
26. Patwardhan B. Traditional Medicine: Modern Approach For Affordable Global Health. World Health Organization. <https://www.who.int/intellectualproperty/studies/B.Patwardhan2.pdf>. Published March 25, 2005. Accessed December 28, 2020.



Novel Therapies for Acute Myeloid Leukemia

By: Danlin Zeng

Acute Myeloid Leukemia (AML) is a heterogeneous disease with an aggressive clinical course, involving rapid cellular proliferation and differentiation arrest of myeloid progenitor cells[1]. AML primarily affects older adults, as 68 years is the median age of diagnosis for AML[2]. In adults, the 5-year net survival rate for AML is merely 24%[2]. There is a lack of therapy success for AML over the last few decades, partially due to the heterogeneity of AML, causing considerable variability both among and within patients. There has been no significant improvement in the overall survival rate of AML patients since 1968[3]. Better tolerated and more effective therapies are currently in development to improve the prognosis of this deadly disease.

AML possesses a subpopulation of cells that are responsible for the initiation and maintenance of the cancer, also known as cancer stem cells[1,2]. The presence of cancer stem cells explains why conventional chemotherapy is rarely curative for AML, as the limitless self-renewal of the cancer stem cells could regenerate the cancer[2]. Thus, eradication of cancer stem cells is crucial for preventing cancer relapse after treatment[2]. As emerging research reaffirms the heterogeneity of AML, "one size fits all" chemotherapy regimens, which includes cytarabine and anthracycline drugs followed by consolidation chemotherapy, are being replaced by more targeted individualized therapies[4]. In the last few years, there have been nine new drugs approved by the US Food and Drug Administration for the treatment of AML[4]. In this new era of precision treatment, inhibitors of FMS-like tyrosine kinase 3 (FLT3), isocitrate dehydrogenase (IDH), B-cell lymphoma 2 (BCL-2), and the hedgehog signaling pathway are being used to treat AML[5,6]. Moreover, there are liposomal formulations of a fixed combination of daunorubicin and cytarabine as well as an anti-CD33 monoclonal antibody included in the newly approved therapies for AML[5].

More therapies are also being developed that target the differences between cancer stem cells and normal tissue stem cells, more effectively preventing AML relapse[5].

Approximately one-third of AML patients have mutations in their FLT3 gene[6]. FLT3 mutations have historically been associated with inferior prognosis[6]. Midostaurin and gilteritinib are FDA-approved FLT3 inhibitors that have shown to improve remission rates compared to conventional chemotherapies in clinical trials involving FLT3-mutated AML[6]. Currently, several other FLT3 inhibitors are in development, including quizartinib and crenolanib[6].

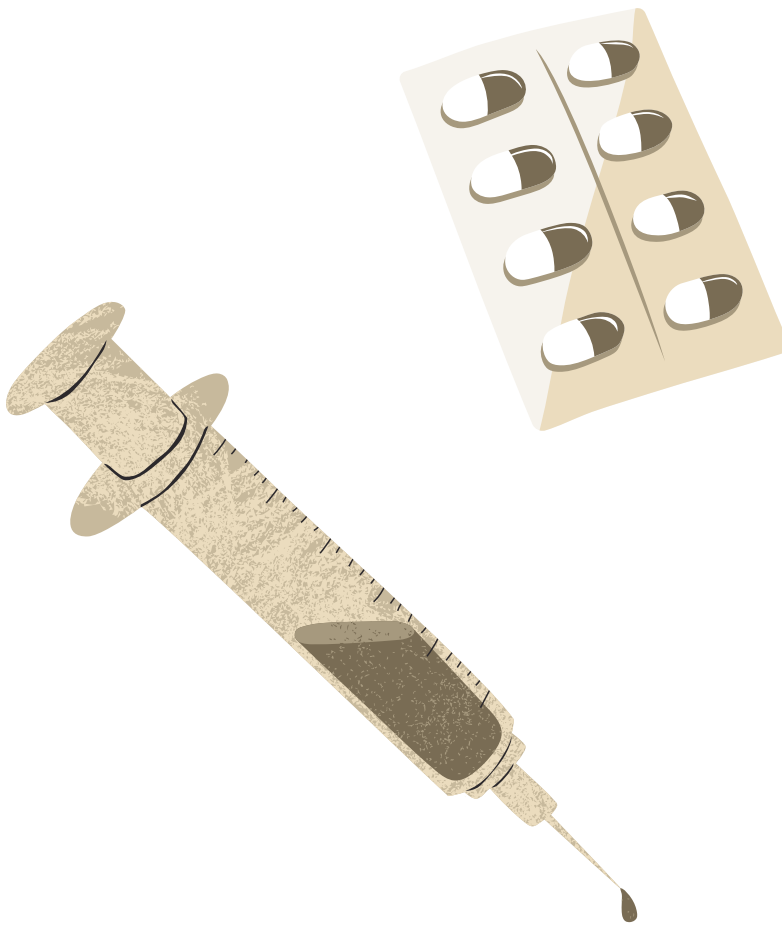
IDH1 and IDH2 mutations, found in 20% of AML patients result in the aberrant production of 2-hydroxyglutarate, leading to DNA and histone hypermethylation and impaired myeloid differentiation, promoting oncogenesis in AML[4,6]. IDH inhibitors have been evaluated in clinical trials of patients with AML for decades, but only recently, have two of the inhibitors been approved by the FDA for the treatment of patients with IDH1- or IDH2-mutant AML. Ivosidenib is indicated for patients with IDH1 mutations, while enasidenib is indicated for patients with IDH2 mutations[7]. Recent data from preclinical studies also suggest a synergistic effect when the BCL-2 inhibitor venetoclax is co-administered with FLT3- and IDH- inhibitors. IDH1- and IDH2-mutant cells are more sensitive to apoptosis following exposure to venetoclax relative to wildtype AML cells[6].

BCL-2, an antiapoptotic protein, is often overexpressed in AML patients[3]. Venetoclax is a BH3 mimetic that inhibits BCL-2 selectively and was approved by the FDA in 2016[3]. For elderly patients that cannot tolerate conventional chemotherapy due to its intensity, venetoclax can be administered in combination with low-dose cytarabine to improve survival rates[3,7].

However, while this remains promising, the median overall survival was only lengthened to about 14 to 15 months, leaving substantial room for improvement[7].

Several novel strategies are also being investigated for the treatment of patients without targetable mutations. Another one of the newly approved drugs by the FDA, CPX-351, which a liposomal formulation of daunorubin and cytarabine which releases the drugs in fixed a 5:1 ratio, was evaluated in a randomized phase II and III clinical trials[8]. Compared to standard chemotherapy, the use of CPX-351 resulted in a superior overall remission rate of 47.7% as opposed to just 33.3%[8]. It also had an improved safety profile and was associated with higher overall survival of 9.56 months compared to 5.95 months[8].

Although these emerging therapies for AML address numerous unmet needs in patients with AML, much clinical research remains to be done to determine the optimal dosage and combinations of these agents. In the recent decades, there has been a shift towards the use of targeted therapies in AML due to the heterogeneity of the disease. For instance, mutations in FLT3 and IDH can be targeted, especially in elderly patients that cannot tolerate intensive standard chemotherapy[4-7]. Thus, knowing the molecular characteristics of patients with AML is of utmost importance to plan the best course of treatment.

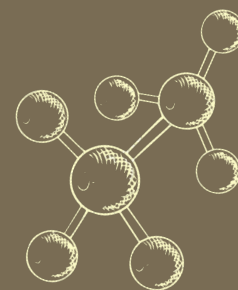




References

1. Bulaeva E, Pellacani D, Nakamichi N, et al. MYC-induced human acute myeloid leukemia requires a continuing IL-3/GM-CSF costimulus. *Blood* 2020;136(24):2764-2773. doi:10.1182/blood.2020006374
2. Nair R, Salinas-Illarena A, Baldauf HM. New strategies to treat AML: novel insights into AML survival pathways and combination therapies. *Leukemia* 2021; 35:299–311. doi:10.1038/s41375-020-01069-1
3. Shallis RM, Wang R, Davidoff A, Ma X, Zeidan AM. Epidemiology of acute myeloid leukemia: Recent progress and enduring challenges. *Blood Rev.* 2019;36:70-87. doi:10.1016/j.blre.2019.04.005
4. Daver N, Wei AH, Pollyea DA, et al. New directions for emerging therapies in acute myeloid leukemia: the next chapter. *Blood Cancer J* 2020; 10(107):1-12. doi:10.1038/s41408-020-00376-1
5. Smith CC. FLT3 Inhibition in Acute Myeloid Leukemia. *Clin Lymphoma Myeloma Leuk.* 2020; 20 Suppl 1:S5-S6. doi:10.1016/S2152-2650(20)30441-9
6. Kayser S, Levis MJ. Advances in targeted therapy for acute myeloid leukaemia. *Br J Haematol.* 2018;180(4):484-500. doi:10.1111/bjh.15032
7. Swaminathan M, Wang ES. Novel therapies for AML: a round-up for clinicians. *Expert Rev Clin Pharmacol.* 2020;13(12):1389-1400. doi:10.1080/17512433.2020.1850255
8. Wei AH, Strickland SA Jr, Hou JZ, et al. Venetoclax Combined With Low-Dose Cytarabine for Previously Untreated Patients With Acute Myeloid Leukemia: Results From a Phase Ib/II Study. *J Clin Oncol.* 2019;37(15):1277-1284. doi:10.1200/JCO.18.01600

UNIVERSITY OF TORONTO



UNDERGRADUATE COMPUTATIONAL CHEMISTRY

Check out recent PCL472/474 student projects from the lab of Pharmacology and Toxicology Associate Professor, Dr. Matthieu Schapira, at the SGC! Including visualizations of protein structures and binding sites, and descriptions of individual student projects.



[HTTPS://WWW.YOUTUBE.COM/CHANNEL/UCeZ7KLN6PB2WMF42OLASTNG](https://www.youtube.com/channel/UCeZ7KLN6PB2WMF42OLASTNG)

PROJECTS

ANGELA (CHAE JIN) KWAK

In silico analysis of Wee1 kinase and it's small molecule inhibitors

CARMEN CHAN

Evaluating structure-based and ligand-based virtual screening methods for the design of Bcl-xL inhibitors

JUSTIN LAM

In silico virtual screening and drug design for the androgen receptor

MARC DENZEL M. NUNEZ

Utilization of in silico techniques to examine cathepsin L inhibition and identify novel inhibitors

VERONICA CHIANG

Analyzing in silico structure-based and ligand-based screening approaches to optimize drug design on the glucocorticoid nuclear hormone receptor

Thank You

FOR READING THE
PHARMACHRONICLE
8TH EDITION

